Effect of crude extract of *Origanum vulgare* on the inhibition of some pathogenic bacteria and causing spoilage of food

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Abstract

Aromatic and medicinal plants are widespread throughout the world. This study was conducted to investigate the inhibitory role of crude extract of *Origanum vulgare* plant on some pathogenic bacterial species: *Pseudomonous aerogenosa*, *Staphylococcus aureus*, *Salmonella typhimurium*, *E. coli*, *Listeria monocytogenus*, *Streptococcus pyogenus*, *Brucella abortus* and *Bacillus anthracis*. These strains were cultured on nutrient agar then each Petri dish contain one of the pathogenic bacteria has been injected into the three holes by extract concentration 50 mg/ml and 100 mg/ml and ethanol alcohol 70% as control, the inhibitory zone were measured. The results revealed that the inhibitory effect of alcoholic extract was more effective against *Listeria monocytogenus* in inhibition zone diameter (31-32 mm), followed by *Staphylococcus aureus* in inhibition zone diameter (27-32 mm) then *Salmonella typhimurium*, *E. coli* and *Brucella abortus* in inhibition zone diameter (26-30 mm, 25-29 mm, 25-30 mm) respectively and moderate activity on *Streptococcus pyogenus*, *Pseudomonous aerogenosa* in inhibition zone diameter (19-20 mm, 19-28 mm) then *Bacillus anthracis* in inhibition zone diameter (17-25 mm) respectively.

Keywords: *Origanum vulgare*, inhibitory effect, pathogenic bacteria.

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Introduction

Traditional folk medicine obtained from plant resources still uses from major part of the total population in developing countries (1). Origanum vulgare, Lamiaceae family, is being applied in traditional medicine systems in many countries and widely known as a very versatile plant with many therapeutic properties (diaphoretic, carminative, antispasmodic, antiseptic, tonic) (2, 3). Origanum or Oregano often called locally joy of mountains, the name is derived from two Greek words, oros (mountain) and ganos (joy) during the middle ages in Poland Oregano was used against number of diseases. Oregano used more for medicinal purposes than culinary uses By Greeks and Romans (4). In latest years, increased attention of the oregano biological activities of researchers and industry, as well as consumers (5). It is an important culinary herb. It is mostly used for flavouring meat, especially for mutton and lamb, it can be usually found on table, together with paprika salt and pepper, In barbecue and kebab restaurants (6). Oregano (Origanum vulgare) is an important Mediterranean herb in literature there are many articles concerned with the antimicrobial activity of essential oils, it is rich in phenolic compounds with antioxidant and antimicrobial activity against a wide range of microorganisms (7, 8), and particularly against common food pathogens. These natural products do not enhance antibiotic resistance as with the long-term use of synthetic antibiotics and that is the main advantage (9). The ability of essential oils plant to protect foods against pathogenic and spoilage microorganism with the benefit of being natural products and thus more acceptable by the consumers, has been reported by several researchers (10). The aim of this study was to investigate in vitro the antimicrobial activity of crude extract of Origanum vulgare on some pathogenic bacterial species.

Materials and Methods

- **Bacterial culture:**
  a. Bacterial isolates serotypes that were used in vitro study were Pseudomonous aerogenosa, Staphylococcus aureus, Salmonella typhimurium, E.coli, Listeria monocytogenus, Streptococcus pyogenus, Brucella abortus and Bacillus anthracis were obtained from Zoonoses Unit/ Veterinary Medicine/ Baghdad University, and the biochemical properties were tested depending on the method of (11).
  b. The bacterial suspension (1 ×10^8) cfu of the pathogenic bacteria were prepared as the method in (11).
- **Plant material and Extraction:** Origanum vulgare Linn flowers were purchased from the local market of Baghdad, Preparation of Origanum vulgare extract according to (12).
  a. Ethanolic extract of plant was weighting 50g of powder and dissolved in 500 ml of 70% ethanolic alcohol extract in Erlyn Myer flask and freezing in deep freeze for 9 days then on magnetic stirrer for 2 hours at room temperature.
  b. The sediments were filtered by gauze and then by filter paper. The mixture was filtered by using filter paper (Wattman No.1).
  c. The supernatant was evaporated to dryness (45°C) under reduced pressure in a rotary evaporator.
  d. The weight of crude extract resulted from that amount of powdered plant was measured.
  e. The crude extract then was kept at -20°C until the time of use.
- **Sensitivity test:** as the following:
  a. Were taken 24 Petri dishes of agar-type Nutrient agar, which wiped every threedishes for one type of bacteria four drops of bacterial suspension that prepared
and calculated manner according to McFarland tube (first tube), and after that dried the dishes, punctured dishes by using the drilling cork (three holes/one dish).
b. Each Petri dish contain one of the pathogenic bacteria has been injected into the three holes by extract concentration 50 and 100 mg, and ethanol alcohol 70% as control respectively. Then all the dishes were incubated at 37°C for 24 hours After incubation inhibition zone diameters were measured to the nearest millimeter (mm) and recorded of the results.

Results

Results of the inhibitory activity of *Orginum vulgare* crude extract on the growth of different Gram-postive and Gram-negative pathogenic bacterial species are summarized on below Table (1). The results revealed variation in the antimicrobial properties of selected Crud extract at concentration 50 mg and 100 mg showed strong activity on *Listeria monocytogenus* in inhibition zone diameter (31-32 mm) was showed in Fig. (1), followed by *Staphylococcus aureus* in inhibition zone diameter (27-32 mm) was showed in Fig. (2) then *Salmonella typhimurium*, *E.coli* and *Brucella abortus* in inhibition zone diameter (26-30 mm, 25-29 mm, 25-30 mm) respectively were showed in Fig. (2, 3, 4) and moderate activity on *Streptococcus pyogenus*, *Pseudomonous aerogenosa* in inhibition zone diameter (19-20 mm, 19-28 mm) were showed in Fig. (1, 4) then *Bacillus anthracis* in inhibition zone diameter (17-25 mm) was showed in Fig. (3).

Table (1) Shows Inhibitory properties (inhibition zone diameter in mm) of plant extracts on some pathogenic bacteria

<table>
<thead>
<tr>
<th>Type of bacteria</th>
<th>Mean zone inhibition in(mm)</th>
<th>Dose 50 mg/ml</th>
<th>Dose 100 mg/ml</th>
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<tbody>
<tr>
<td>Gram positive Bacteria</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>27</td>
<td>32</td>
<td></td>
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<tr>
<td><em>Streptococcus pyogenus</em></td>
<td>19</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td><em>Listeria monocytogenus</em></td>
<td>31</td>
<td>32</td>
<td></td>
</tr>
<tr>
<td><em>Bacillus anthracis</em></td>
<td>17</td>
<td>25</td>
<td></td>
</tr>
<tr>
<td>Gram negative Bacteria</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>E.coli</em></td>
<td>25</td>
<td>29</td>
<td></td>
</tr>
<tr>
<td><em>Salmonella typhimurium</em></td>
<td>26</td>
<td>30</td>
<td></td>
</tr>
<tr>
<td><em>Brucella abortus</em></td>
<td>25</td>
<td>30</td>
<td></td>
</tr>
<tr>
<td><em>Pseudomonous aerogenosa</em></td>
<td>19</td>
<td>28</td>
<td></td>
</tr>
</tbody>
</table>

(-)the inhibition zone diameter less than (6) millimeter (Resistant)
(+*) the inhibition zone diameter equal or more than (6) millimeter (Sensitive)
Fig. (2) Inhibition zone diameter in millimeter of crude extract against *Staphylococcus aureus* and *Brucella abortus*

Fig. (3) Inhibition zone diameter in millimeter of crude extract against *Salmonella typhimurium* and *Bacillus anthracis*

Fig. (4) Inhibition zone diameter in millimeter of crude extract against *E.coli* and *Pseudomonous aerogenosa*
Discussion

Plant essential oils and extracts have been used for many thousands of years, in food preservation, pharmaceuticals, alternative medicine and natural therapies. Plant extracts are potential sources of novel antimicrobial compounds especially against bacterial pathogens (13). The results indicated that the crude extracts of oregano studied showed antibacterial activities towards the gram positive and gram negative bacteria these results are consistent with previous reports found oregano extract was effective at inhibiting the growth of Listeria monocytogenes (14). Listeria monocytogenes is found in many food products (6). The oregano oil exhibited significant inhibitory activity against Citrobacter spp., Salmonella typhii and Escherichia coli (15) the essential oil of oregano (Origanum vulgare) exhibited antibacterial activity against Staphylococcus aureus (16), thymol and carvacol are the main components of the oregano essential oil which are responsible for its antioxidative, antimicrobial and antifungal effects (17). Infections caused by P. aeruginosa, especially those with multi-drug resistance, are among the most difficult to treat with conventional antibiotics (18) in our study, the growth of P. aeruginosa was remarkably inhibited by oregano extract and these results was not agreed with (19).

Conclusion

The main conclusion drawn from this data is that the crude extracts of oregano studied showed antibacterial activities towards some gram positive and gram negative bacteria at different rates and that oregano represents an economic source of natural mixtures of antibacterial compounds that can be as effective as modern medicine to combat pathogenic microorganisms and safe alternative to treat infectious diseases.

References


