**Prunus mahaleb Inhibits The Gram-negative Bacterium Escherichia coli**

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**Abstract**

Foodborne infections caused by gram-negative pathogens reach in millions of illnesses worldwide. Transmissible of foodborne infection by raw fruits and vegetables is increasing. A preventative treatment of produce that can kill gram-negative bacteria prior to consumption can reduce the risk of infections. Species have been used as an effective preventative against foodborne infections in many cultures for centuries, thus these plants are possible sources of antimicrobials. In this study, *Prunus mahaleb* was screened for antimicrobial compounds because it has long been used as a spice in the Middle East. *P. mahaleb* seeds were ground in ethanol or isopropyl alcohol to a final concentration of 33-67 mg/mL. These extracts were tested for antimicrobial activity against *Escherichia coli*. *Saccharomyces cervisiae* and *Lactobacillus acidophilus* in agar diffusion assays. The ethanolic extract (33 mg/mL) inhibited *E. coli* with a 2 mm zone of inhibition. The maximum bacterial concentration is 62.5 mg/mL, and the maximum bacterial concentration is 80 mg/mL. The effect of the least-extract on bacterial growth rate is being determined. The effectiveness of this extract pecting *E. coli* survival and growth is to be presented. These results suggest that *P. mahaleb* has the potential to prevent survival and growth of gram-negative bacteria in foods.

**Hypothesis**

*Prunus mahaleb* ethanolic seed extract has antimicrobial activity that will inhibit growth of gram-negative *Escherichia coli* bacteria.

**Methods**

**Ethanolic Extract of Prunus mahaleb**

1. *P. mahaleb* seeds were purchased from a Middle Eastern store or from Zaitouni Spices.
2. Seeds were crushed and mixed in 95% ethanol alcohol to make the concentrations of 33, 50, and 100 mg/mL.
3. **Dilution Disk Assay**
   - Nutrient agar was inoculated with *Escherichia coli* (ATCC 17775).
   - Filter paper disks saturated with each ethanolic extract were placed on the nutrient agar.
4. **Bacterial survival in bean sprouts**
   - Any commercially purchased bean sprouts were treated with 33, 50, and 100 mg/mL ethanolic extract were inoculated with a 24-hr *E. coli* culture.
5. **Absorbance was recorded** at 540 nm for 25 hr.
6. **E. coli survival in bean sprouts**
   - After 24 hr, ethanolic extract was filtered with 3 mm g/mL. *E. coli* extract was inoculated with 1 mL of *E. coli* and incubated at 35°C.
7. **Bacterial survival was determined using heterotrophic plate counts.**
8. **Chromatography**
   - Paper chromatography in isopropanol-ammonium hydroxide solvent was used to isolate the antibacterial compound(s) in *P. mahaleb*.
9. **The dried chromatogram was cut into 5-cm pieces,** which were used in a disk diffusion assay.

**Results**

- 2 mg/mL ethanolic *P. mahaleb* seed extract was most effective in the disk diffusion assay, with a zone of inhibition of 2 mm. The ethanolic solution had a zone of inhibition of 0.5 mm.
- The MIC is 62.5 mg/mL, and the MBC is 50 mg/mL.
- *E. coli* grew better in nutrient broth with mahaleb extract than in the control (Figure 2).
- Mahaleb extract did not affect *E. coli* survival in bean sprouts (Figure 4).
- The antibacterial compounds were found at Rf values between 0.45 and 0.64. The inhibition zone was about 1 mm (Figure 2).

**Discussion & Conclusion**

- Foodborne pathogens affect approximately 76 million people or 75% of the United States population annually (2). It is long been used as a spice in the Middle East.
- This study was done in a disk diffusion assay. *P. mahaleb* seeds were ground in ethanol or isopropyl alcohol to a final concentration of 33-67 mg/mL. These extracts were tested for antimicrobial activity against *Escherichia coli*. *Saccharomyces cervisiae* and *Lactobacillus acidophilus* in agar diffusion assays.
- The ethanolic extract (33 mg/mL) inhibited *E. coli* with a 2 mm zone of inhibition. The maximum bacterial concentration is 62.5 mg/mL, and the maximum bacterial concentration is 80 mg/mL. The effect of the least extract on bacterial growth rate is being determined. The effectiveness of this extract pecting *E. coli* survival and growth is to be presented. These results suggest that *P. mahaleb* has the potential to prevent survival and growth of gram-negative bacteria in foods.

**Literature Cited**


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