

# Extraction, and Antibacterial activities of *Teucrium polium* (Algeada) growing in Iraq

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## Abstract

This study was designed to isolate the most important bioactive compounds from the Iraqi plant *Teucrium polium* using a liquid-liquid extraction method, detected on thin layer chromatography (TLC) plates, and examine the antimicrobial activities *Teucrium polium* extracts against five bacterial species by using disc-diffusion method. The extracts were identified via Thin Layer Chromatography (TLC) using a mixture of ethyl acetate : methanol : water (100 : 15 : 10). as chromatographic eluent .

## استخلاص و دراسة الفعالية الحيوية لنبات الجعدة العراقي

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## المستخلص:

في هذه الدراسة تم استخلاص نبات الجعدة العراقي بمذيبات قطبية ولا قطبية وتشخيص المستخلصات بوساطة تقنية الكروماتوغرافيا الطبقة الرقيقة، ومن ثم دراسة الفعالية الحيوية حيث اظهرت النتائج ان المستخلص الكحولي يملك اعلى فاعلية ضد انواع البكتريا قيد الدراسة وخصوصا السالبة منها .

## Introduction

Reactive Oxygen Species are various form of activated oxygen which include free radicals such as superoxide, hydroxyl radical, as well as hydrogen peroxide(1-2). Exogenous sources of free radicals include tobacco smoke, ionizing radiation, organic solvents and pesticides(3). The medicinal use of plants dates back to ancient times. *Teucrium polium* is a plant that has been used for over 2000 years in traditional medicine due to its diuretic, diaphoretic, tonic, antipyretic, antispasmodic and cholagogic properties (4–6). In addition, the plant possesses hypoglycemic, insulinotropic and anti-inflammatory activities (7–9), reduces body weight and lowers high blood pressure (10,11) and has hypolipidemic, antinociceptive and antioxidant properties (12–14).

### **Chemicals**

Ethyl acetate, methanol, ethanol, iodine, hydrochloric acid (HCl), and Dimethyl sulphoxide (DMSO) obtained from BDH company.

### **Plant**

*Teucrium polium* (Algeada) were purchased from a local market in Iraq, and identified at the College of Agriculture and Baghdad University - Iraq.

### **Preparation of Samples**

*Teucrium polium* leaves were washed with distilled water and dried at room temperature in the dark and then ground to powder using a blender. An accurately weighed amount of *Teucrium polium* powder (10 g each) was dissolved in 250 ml of (Hexane or Ethanol) . The solution was heated for 6 hour using a Soxhlet apparatus and the extracts were dried in vacuum by a rotary evaporator.

### **Microorganisms**

All microorganisms were obtained from the Department of Biology, College of Science, Al-Mustansiriyah University - Iraq. Four strains of Gram-negative bacteria; *Escherichia coli* , *Proteus mirabilis* , *Klebsiella pneumonia* , *Pseudomonas aeruginosa* and two strains of Gram-positive bacteria; *Staphylococcus aureus*, *Bacillus cereus* were used as the tested bacteria. The cultures of bacteria were maintained in their appropriate agar slants at 4°C throughout the study and used as stock cultures.

## Antibacterial Activity

### Disc diffusion assay

0.1mL of 108 cell/mL of each bacteria used in this experiments onto Nutrient agar. The inocula were spreaded using glass spreader or sterile cotton swab. To study the effect of the two extractants in microbes growth, we prepared filter paper disks saturated with different concentrations of extractants by adding 0.1mL for each concentration to a container contains 10 sterilized disks, then the cultures inoculated at 37°C for (24) h .The following antibiotic disks Cefalexin, and Ampicillin were utilized as a positive control to the microbes(15).

### Results and Discussion

The determination of the MIC (Minimum inhibition concentration) by means of the disc diffusion assay (Table 1) showed that 2 plant extracts tested exhibited an antimicrobial effect against Gram positive, and Gram negative bacteria.

**Table 1. The MIC values in mg/mL of *Teucrium polium* extracts in Disk diffusion assay.**

Plant ex.	<i>Bactreia</i>					
	<i>Staphylococcus aureus</i> (G +ve)	<i>Bacillus cereus</i> (G +ve)	<i>Escherichia coli</i> (G -ve)	<i>Proteus mirabilis</i> (G -ve)	<i>Klebsiella pneumonia</i> (G -ve)	<i>Pseudomonas aeruginosa</i> (G -ve)
By ethanol	4		3	4	2	6
By hexane	4		5	2	4	5

According to the results given in Table 2, the extracts of *Teucrium polium* had great antimicrobial activity against all investigated microorganism. The diameters of growth inhibition zone ranged from 13 to 24 mm (including the diameter of the disc—6 mm) with the highest inhibition zone values observed against *Escherichia coli* , and *Pseudomonas aeruginosa* (23-24 mm). The extracts showed greater activity on Gram-positive and Gram-negative bacteria.

**Table 1. Antibacterial activity of of extracts of *Teucrium polium*.**

Microorganism	Inhibition zone diameter (mm)			
	By ethanol	By hexane	Cefalexin,	Ampicillin
<b>Gram (+)</b>				
<i>Staphylococcus aureus</i>	20	15	31	28
<i>Bacillus cereus</i>	19	16	32	27
<b>Gram (-)</b>				
<i>Escherichia coli</i>	24	23	32	31
<i>Proteus mirabilis</i>	18	14	34	30
<i>Klebsiella pneumonia</i>	18	13	33	30
<i>Pseudomonas aeruginosa</i>	23	23	35	29

In general, the extract by ethanol showed better activity than the hexane extract. The hexane extract showed strong antibacterial activity against the bacteria *Escherichia coli*, inhibition zone is 23 mm. Although extracts had similar sizes of the zone of inhibition for *Escherichia coli* and *Pseudomonas aeruginosa* (23,24 and 23mm, respectively). On all other microorganism the hexane extract showed less activity than ehtanolic extract.

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