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Antifungal Activity of Some Plant Extracts Against Two Yeasts Isolates *In Vitro*.

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ABSTRACT

During this study tested antifungal activity of six plant extracts which collected from Kurdistan region in North of Iraq and prepared three concentrations to ethanol extracts for each one (500 mg /ml ,250 mg /ml ,125 mg /ml). The results showed the extracts of *Qurecus aegilops* in concentration 0.5 gm /ml appeared highest activity against *Candida albicans* while the extract of *Elaegnus angustifolia* appeared lowest activity in the same concentration against *Cryptococcus neoformans*. Also tested specific chemical reagent for this extracts and appeared positive test for all reagents.

Keywords: activity antifungal, plant extract, North of Iraq

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INTRODUCTION

The most common mycoses are pityriasis versicolor,¹ the various clinical forms dermatophytosis of, cryptococcosis and candidiasis. [1] may be because of drug resistance, mainly in fungal isolates In-vitro susceptibility testing of anti-fungal agents is becoming increasingly important because of introduction of new anti-fungal agents and the recovery of clinical isolates that exhibit inherent or developed resistance to available anti-fungal[2] So went the attention of researchers around the plant kingdom, which is a treasure trove and inexhaustible from studies which exhibited antifungal activity in world. Refer[3] to highest activity antifungal of ethanolic extract of *Allium porrum* , *Hymenocrater longiflorus* against *Candida albicans* , *C.krusei* , *C. rugosa*, *Cryptococcus neoformans* , *C. laurentis* , *C. albidus* .He also noted[4] to the effectiveness of the tannins extracted from the plant *Quercus aegilops* in the inhibition of the *Candida albicans* , *Aspergillus niger*. In Sri Lanka, a survey conducted by[5] biological activity about the effectiveness of some plants, such as *Piper nigrum*, *Quercus infectoria* the study showed antifungal and anticancer activity as well as its active role in the treatment of dental pain. And [6] Effectiveness antibody antimicrobial activity of *Equisetum ramosissimum* against some fungal and bacterial isolates what content of the compounds effective phenolic and alkaloides and Flavonoides compounds. As mentioned[7] contain plant *Elaeagnus angustiflorus* on volatile oils known its effectiveness antifungal activity.

Aims of study

Test activity antifungal of plant extract against two clinical isolates to suggest as alternative drug.

MATERIAL AND METHODS

Classification of plants

Classify plants Prof. Dr. Abdul Redha Akber Alwan Al-Mayah in Biology Department College of Science / University of Basra

S	Scientific plant name	Use part
1	<i>Quercus infectoria</i>	Leaves
2	<i>Q. aegilops</i>	Leaves
3	<i>Equisetum ramosissimum</i>	Whole plant
4	<i>Allium porrum</i>	Leaves
5	<i>Elaeagnus angustifolia</i>	Leaves
6	<i>Hymenocrater longiflorus</i>	Flower

Prepare ethanolic extracts according to [8]

Specific tests

Conducted several tests to detect about active compounds in test plants under depending on the [9]

preliminary survey of the ethanol extracts

For the initial evaluation of the ethanolic extracts toward two of the yeasts that have been obtained from laboratory fungi / College of Science / University of Basra, used for test antifungal activity agar diffusion method and used concentrations 500 mg / ml, 250 mg / ml, 125 mg / ml and summarized methods as follows: -

- Used sabourauds dextrose agar medium (Himedia Company) For the purpose of activating fungal isolates so incubated these isolates at a temperature of 27 C° for three days .
- Taking 0.2 µl of yeasts suspension which compared with MacFarland tube 3 *10⁶ colony form unit / ml by sterile a pipette and spread on culture medium by sterilized L-Shap with alcohol and flame then left the culture medium for a period of one hour to dry suspension.
- Make three wells a diameter of 6 mm for each dish and the use of sterile cork borer.
- 100 µl has been added to each well of extract and using Micropippite with sterile lids and gingerly to avoid scattering extracts above the surface of the culture medium.
- Culture medium were incubated 27 C for 3 days in order to note the clear zone around the well, which represents growth inhibition zone diameter measured in mm[10]

RESULTS

inhibitory efficacy of the ethanolic plant extracts against isolates two yeasts isolates.

The present study demonstrated the effectiveness of inhibition of plant extracts six table (1) and three concentrations are: 500 mg / ml ,250 mg / ml ,125 mg / ml. Toward test yeasts isolates. Inhibitory effectiveness as varied by the type of plant and fungal isolation and in general had a higher rate of inhibition diameters (21) mm of *Candida albicans* at concentration of 500 mg / ml for *Qurecus aegilops*, while the lowest rate of inhibition diameters (7 mm) of *Cryptococcus neoformans* of the plant *Elaegnus angusifolia* at a concentration of 500 mg / ml plate (1).

DISCUSSION

The effectiveness of ethanolic plant extracts under study toward the tested isolates

The study results showed the current Table (2-7) the effectiveness of plant extracts under study toward the isolates tested and generally have attributed the cause to the use of ethanol as a solvent polar extraction plants study which serves to pull the polar active compounds As shown in Table (1) example (phenols, tannins and saponins) where these compounds operate on Union with protein deposition and cell altering the nature and function

as a good solvent for fatty substances, that is lead to analyzed the membranes of living cells and as a result he graduated cellular components inside to outside and die the yeast cell [11] But we noticed variation in the effect of plant extracts crude toward yeasts isolates Since the extraction method unified and solvent used one, so it may be cause of the discrepancy is due the lack of quantity of active substances in the extracts or to the weakness of their effectiveness and probably is attributable to the need to separate the active ingredients from plants study .Refer the [12] It sometimes leads the presence of active ingredients with each other in the crude extracts to a negative effect. May be cause loss flavonoid in *Elaeagnus angustifolia* but found it in *Quercus aegilops* lead to this result Table(1) this is consistent with the study of [13,14] Appeared the current study, the effectiveness of the antifungal activity of ethanolic extract of *Quercus aegilops*, which may be due to the effectiveness of this extract to contain tannin and flavonoid while refer[15] to low antifungal activity of *Elaeagnus angustifolia* And in general was a yeast *Candida albicans* More sensitive to all the extracts tested, compared with *Cryptococcus neoformans* The reason to own a yeast capsule surrounds cell membrane which prevents or hinders enter plant extracts compared to *candida albicans* [16] Many studies in different country test antifungal activity of *Hymenocrater* , *Allium* and *Equisetum* [17,18,19].

CONCLUSIONS

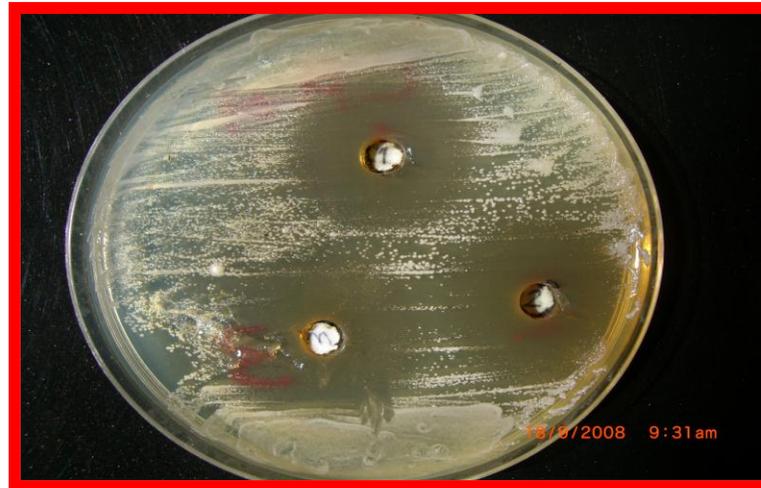
We conclude from the current study antifungal activity alcoholic extracts of six plants and contain more active compounds allowing Recommended therapeutic alternatives to antifungal chemical drugs

Table 1: The specific test of the ethanolic extracts of study plantscurrent study

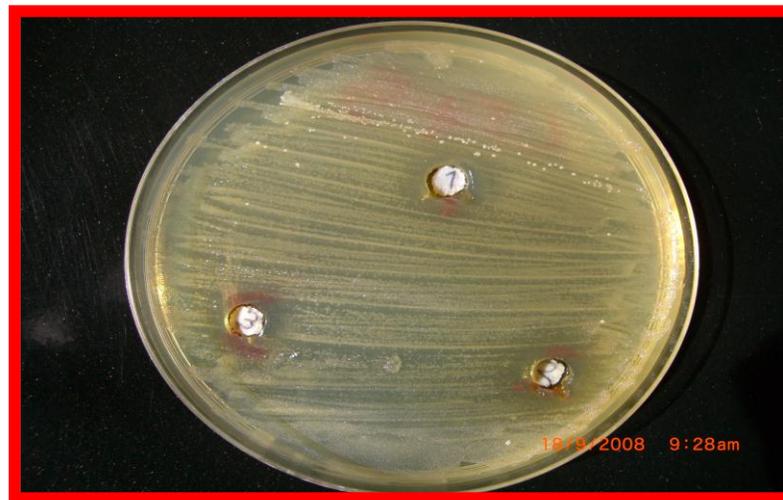
s.	Plants used	Active compounds that have been detected					
		Amino acids	Phenols	Alkaloids	Flavonoids	Tannins	resene
1	<i>Quercus infectoria</i>	-	+	+	+	+	+
2	<i>Q. aegilops</i>	-	+	+	+	+	+
3	<i>Equisetum ramosissimum</i>	-	+	+	+	+	+
4	<i>Allium porrum</i>	-	+	+	+	+	+
5	<i>Elaeagnus angustifolia</i>	-	+	+	-	+	+
6	<i>Hymenorater longiflorus</i>	-	+	+	+	+	+

(-) = Negative detection

(+) = Positive detection



a. *Candida albicans*



b. *Cryptococcus neoformans*

Plate 1 :a- highest inhibition zone rate for ethanolic extract of *Qurecus aegilops* against *Candida albicans* at concentration 500 mg/ml

b- lowest inhibition zone rate for ethanolic extract of *Elaegnus angustifolia* against *Cryptococcus neoformans* at concentration 500 mg/ml

Table 2: Inhibition zone rate (mm) for Ethanolic extract of *Elaegnus angustifolia*

concentration yeasts isolates	Ethanolic extract of <i>Elaegnus angustifolia</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	10	10	8
<i>Cryptocoues neoformans</i>	7	6	6

Table 3: Inhibition zone rate (mm) for Ethanolic extract of *Quercus infectoria*

concentration yeasts isolates	Ethanolic extract of <i>Quercus infectoria</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	20	17	15
<i>Cryptocoues neoformans</i>	9	7	6

Table 4: Inhibition zone rate (mm) for Ethanolic extract of *Quercus aegilops*

concentration yeasts isolates	Ethanolic extract of <i>Quercus aegilops</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	21	16	14
<i>Cryptocoues neoformans</i>	13	9	6

Table 5: Inhibition zone rate (mm) for Ethanolic extract of *Equisetum ramosissimum*

concentration yeasts isolates	Ethanolic extract of <i>Equisetum ramosissimum</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	10	10	8
<i>Cryptocoues neoformans</i>	9	7	7

Table 6: Inhibition zone rate (mm) for Ethanolic extract of *Allium porrum*

concentration yeasts isolates	Ethanolic extract of <i>Allium porrum</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	12	8	6
<i>Cryptocoues neoformans</i>	9	7	6

Table 7: Inhibition zone rate (mm) for Ethanolic extract of *Hymenocrater longiflorus*

concentration yeasts isolates	Ethanolic extract of <i>Hymenocrater longiflorus</i>		
	Inhibition zone rat (mm)		
	500 mg/ml	250 mg /ml	125 mg/ml
<i>Candida albicans</i>	10	10	8
<i>Cryptocoues neoformans</i>	10	6	0

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