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Antimicrobial Activity Of Propolis Against Some Bacteria And Fungi

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ABSTRACT

Propolis is a natural, low cost, non toxic bee product. The study was aimed to evaluate the antimicrobial activities of crude ethanolic extract of Egyptian propolis (EEEP) against two multidrug resistant bacteria and twelve veterinary medical important fungi.

Antibacterial and antifungal activity was determined by diffusion (disc and agar well diffusion) and dilution methods, compared with other antimicrobial agents .

Results revealed that *S.aureus* showed higher sensitivity to EEEP than other *E.coli*. The effect of EEP was elevated when the concentration increased to 80 and 160 mg/ml. Considering the diameter of the inhibitory zones and MIC values, *S.aureus* and *E.coli* showed susceptibility to propolis with inhibition zone diameters of 23 and 17 mm,respectively by disc diffusion test, while they reach 24 and 23 mm, respectively, by agar well diffusion test. Propolis showed similar antibacterial activity as the other tested agents (Oxacillin, Nalidixic acid, Neomycin, Ciprofloxacin, E-moxclav, Doxycycline, Erythromycin, Gentamicin and Sulphamethoxazole Trimethoprim) ,MIC value was 6.4 mg/ml for both bacterial spp.

EEP at concentration, 20 mg/ml was effective against all examined five yeast spcies by well diffusion as well as dilution method and can be arranged as *Trichosporon cutaneum* > *Cryptococcus neoformans* and *Rhodotorula rubra* > *Candida albicans* > *Geotrichum candidum* . Moulds species tested by the dilution (media incorporated with propolis and other antimycotic drugs) method ,indicated that, propolis showed important antifungal activity against tested dermatophytes at 40 mg/l concentration while non dermatophytes at 160 mg/l.

The obtained results emphasized that ,the propolis can be used as alternative treatment in some bacterial and fungal infections, but more research should be carried out to standardize their active ingredients and action.

INTRODUCTION

Since thousands of years, natural products have been used in folk medicine to treat several diseases. Among them, propolis has got an increased interest because of its antimicrobial activity spectra against wide range of pathogenic microorganisms *(1)*. Propolis is a complex resinous mixture collected by honey bees (*Apis melifera)* from buds and extract of certain plants.*(1,2)*. Its colour may be brownish green, chestnut or even black; it tastes bitter but has a sweet and pleasant odour. Colour and composition largely depend on its botanical origin and the type of bee that produce it *(3)*.

The chemical composition of propolis is quite complicated. Its compounds and biological activities depend on many different factors such as the geographical region, collecting time, and plant source *(4-6)*.

Propolis constituents are phenolic acid esters (72.7 %) , phenolic acids (1.1%), aliphatic acids (2.4%), dihydrochalcones (6.5 %),Chalcones (1.7 %); flavanones (1.9 %); flavones (4.6 %) and tetrahydrofuran

derivatives (0.7 %) *(7)*. Propolis contains some minerals such as Mg, Ca, I, K, Na, Cu, Zn, Mn and Fe, vitamins like B_1, B_2, B_6, C and E, a number of fatty acids and some enzymes as glucose -6- phosphatase dehydrogenase , adenosine triphosphatase and acid phosphatase *(8)*.

Many biological properties, including antibacterial, antifungal, antiviral, local anaesthetic, anti-inflammatory, antioxidant, antitumor , anti trypanosomal, scar forming and tissue regeneration, hepatoprotective, immunostimulating, and cytostatic activities have been ascribed to propolis *(3,9-11)*. Propolis is available commercially in different formulated forms such as tablets, capsules, tooth paste, mouth wash preparations, face creams, ointments, lotions, and solutions *(12-16)* . Egyptians ,Greeks and Romans used propolis to cure some skin lesions, additionally, it was used by Cameroon population to treat wounds, burns ,stomach ulcer , respiratory and dental infections *(17)* .

Resistance to antibiotics of frequently isolated pathogen has jeopardized the clinical usefulness of several types of important antimicrobial compounds . As resistance to fluoroquinolones of critical Gram positive pathogens such as *Staphylococci* and Gram negative bacilli such as *Enterobacteriaceae* or *Pseudomonas* spp *(18)* , has rendered the search for new antibacterial agents.The antibacterial properties of propolis, have been investigated against different pathogens. MICs and MBCs were determined on 320 bacterial strains showed good antimicrobial activity against most of the isolates, particularly S. *pneumoniae, H. influenzae* and *M. catarrhalis*, but not against *Enterobacteriaceae (19, 20)* .

The incidence of yeast infections in human and animals has increased significantly in recent years, because there are relatively few antifungal agents available for its treatment, resistance among individual yeast species or strains has been a serious problem. Candida, Trichosporon, and Rhodotorula have certain species resistant to standard antifungal drugs *(21)* so research for newer less toxic antifungal drugs are usually under trials.The published data *(22,23)* evaluated the antifungal activity of six commercial propolis extracts against *Candida* spp. that was isolated from the oral cavity of removable dentures users and superficial mycoses.The *in vitro* activities of propolis against dermatophytes (*Trichophyton rubrum and T. mentagrophytes*) were compared with those of Terbinafine, Itraconazole, Ketoconazole, and Fluconazole. Among the systemic antifungals tested, Terbinafine was the most potent ,and Propolis showed important antifungal activity as a potentially useful agent for the treatment of dermatophytosis *(24)* .

The aim of the present study was to evaluate and compare the antimicrobial activity spectra of propolis with some antimicrobial agents through its effect against some important bacteria and fungi in order to confirm the quality of their actions as an alternative antimicrobial agents.

MATERIAL AND METHOD

Propolis

Crude propolis sample was collected from hive of honeybees of El Sharkia, Egypt. Propolis sample was cleaned free of wax, paint wood, cut into small pieces and placed in clean container. EEP was prepared as previously described *(25,26)* .

Tested microorganisms (Bacterial and fungal spp)

Two multidrug resistant bacterial spp (*Staph aureus* and *Escherichia coli*) ,five yeast spp (*Candida albicans Cryptococcus neoformans, Rhodotorula rubra, Trichosporon cutaneum and Geotrichum candidum*) and mould species, dermatophytes (*Trichophyton rubrum, Trichophyton mentagrophytes, Microsporum canis and Microsporum gypseum) and non* dermatophytes (*Penicillium species, Fusarium moniliforme and Alternaria alternate*) were identified and supplied by Department of Bacteriology, Mycology and Immunology, Faculty of Veterinary Medicine, Zagazig University.

Antibacterial discs (Oxoid) and propolis discs

As shown in Table 1, four antibacterial discs (OX, CIP, CN and AMC) were used for *S.aureus* and six (NA, E, N, SXT, DO and CN) for *E. coli* . Propolis discs were prepared by plain filter paper discs (6 mm) , sterilized by autoclaving and soaked in different concentrations of propolis (20,40,80 and 160 mg /ml) *(27)*.

Table 1. Antibacterial discs

Antibacterial disc	Symbol	Conc./Disc
Oxacillin	OX	30µg
Nalidixic acid	NA	30µg
Neomycin	N	30µg
Ciprofloxacin	CIP	5µg
E-moxclav	AMC	30µg
Doxycycline	DO	30µg
Erythromycin	E	15µg
Gentamicin	CN	10µg
SulphamethoxazoleTrimethoprim	SXT	25µg
Propolis 20 mg/ml	PE	200 µg
Propolis 40 mg/ml	PE	400 µg
Propolis 80 mg/ml	PE	800 µg
Propolis 160 mg/ml	PE	1600 µg

Antifungal drugs used in comparison with propolis

Fluconazole (Pfizer), Itraconazole (Janssen-Cliag) and Terbinafine (Novartis) were used to compare their antifungal activity with propolis.

In vitro antibacterial susceptibility tests of propolis extract

Disc diffusion method

The test was carried according to the published paper *(28)* and its modification using discs of propolis extract (PE) in comparison to discs of other antimicrobial agents as in Table 1,to test their antibacterial activities against the tested bacteria using Muller Hinton agar medium (MHA) (Oxoid) , and triple plates were used for each concentration .

The bacterial suspension was prepared from each strain of Gram-positive and Gram-negative bacteria (*S. aureus* and *E. coli respectively*) and adjusted by comparison against 0.5 McFarland turbidity standards (1.5 $\times 10^8$ CFU/ ml). After inoculation of the plates and application the discs, the agar plates maintained at room temperature for half hour allowing for diffusion of the solution . All plates were inverted then incubated at 37°C for 16- 18 hours and the zones of inhibition around the disc were subsequently measured in millimeter according to recommendations of NCCLS*(29)*.

Agar well diffusion test - Cup plate method

Plates of Muller- Hinton agar were used *(30)*. Agar media were seeded separately with a suspension of microorganisms that adjusted by comparison against 0.5 McFarland turbidity standards as mentioned in disc diffusion method . Four equidistant wells (6mm in diameter) were cut into the agar by sterile cork borer in case of *staph aureus* while six wells in case of *E. coli*. One drop of the agar was used to seal the bottom of the bored hole so that the antibiotics and the PE will not sip beneath the agar, 0.1ml of PE with different concentrations

(20, 40, 80 and 160 mg/ml) were added separately to fill the middle well, also, 0.1ml of the tested antibiotics (as in Table 1 except OX and NA) were added at wells in the periphery, incubation was performed at 37°C for 16- 18 hours.

Detemination of MIC of propolis (dilution method)

MIC was determined by the macrodilution method *(31)*. Serial five fold EPE concentrations 32, 6.4, 1.28, 0.256, 0.0512 and 0.01024 mg/ ml of saline were prepared in 6 tubes, each containing 0.8 ml of sterile saline by adding 0.2 ml of PE (160mg/ ml) . Then 0.8 ml of freshly standardized Muller Hinton broth cultures (1.5×10^8 CFU/ml) of selected bacteria (*Staph aureus and Escherichia coli*) were separately inoculated into 6 sterile tubes contaning 0.2 ml of each above mentioned dilution series of PE, so that the final concentrations of propolis after 1/25 fold dilution were 6.4, 1.28, 0.256, 0.0512, 0.01024 and 0.002048 mg/ml .For each batch of MIC determinations, a blank tube (i.e 1ml non inoculated Muller Hinton broth) as control negative, In addition to, positive control (by mixing of 0.8ml adjusted broth culture with 0.2ml sterile saline) was used for each strain . After 18-24 hours, incubation at 37°C ,MIC was visually recorded, as the lowest concentration (highest dilution) of the propolis that inhibited the visible growth (no turbidity) of tested bacteria compared to the control .

In vitro antifungal susceptibility tests of propolis extract

Agar well diffution test

The method was done on Potato dextrose agar with chloramphenicol (PDA) (Himedia) *(32)* .The medium was seeded separately with a suspension of each yeast. Four equidistant wells (6mm in diameter) were cut into the agar by sterile cork borer . As done in bacterial isolates, then 40 µl of each concentrations of PE (2.5,5,10 and 20 mg/ml) were pipetted into one plate wells for each species . All tests were duplicated for confirmation , and incubated at 30 °C for 72 hours. The degree of antifungal activities was based on measurement of the diameter of the growth inhibition zone in mm formed around the well.

Media incorporated with propolis and other antimycotic drugs (dilution test)

Broth media incorporated with 20 and 40 mg /l propolis and other antimycotic drugs

This method was applied on five species of yeasts and three non dermatophyte moulds, using Sabouraud's dextrose broth (SDB) with chloramphenicol *(33,34)*. After dissolving of the antifungal drugs in DMSO (20mg/ml),the media was divided in to 5 partitions ,each one equals 200ml,then the antifungal drugs and propolis were added to each partition (20 and 40 mg/l). Each partition of media was dispensed in 24 test tubes (i.e. the test was Triplicate for confirmation). After sterilization the media left to reach 40-45°C and inoculated with the eight fungal species then incubated at 30°C/10 days. After incubation the antifungal activity of propolis was determined by detection of the decrease in growth rate when compared with control group (by weighting all tubes before and after incubation).

Agar media incorporated with 40, 80 and 160 mg/l propolis and other antimycotic drugs

This test was applied on seven mould species (four dermatophytes and three non dermatophytes moulds) by Potato dextrose agar (PDA) with chloramphenicol (Himedia) *(35)* . After dissolving of the antifungal drugs and dividing the medium into 5 partitions , the antifungal drugs and propolis were added in concentration 400, 800 and 1600 µl/200 ml (each partition) of medium .Each partition of the medium was sterilized and then dispensed in 14 test tubes (the test was duplicated for confirmation). After dispensing the medium in tubes ,left to solidify in slopes and then inoculated with the tested moulds and incubated at 30°C/10 days. After incubation the antifungal activity of propolis and other drugs was determined as mentioned above .

RESULTS

Antibacteria activites of propolis

Disc diffusion method

Staphylococcus aureus was sensitive to CIP and CN with inhibition zone 28 and 22 mm diameter, respectively and by increasing the concentration of propolis extract (20, 40, 80 and 160 mg/ml) the inhibition zone was increased (7, 8, 16 and 23 mm), respectively (Fig 1). while *E. coli*, was resistant to all used antibiotics except CN with inhibition zone of 17 mm diameter and the diameter of the inhibition zone against propolis extract was increased by increasing the concentration (0, 7, 11 and 17mm) (Fig1).

Agar well diffusion test-Cup plate method

As in disc diffusion test , *Staphylococcus aureus* was resistant to AMC and sensitive to CIP and CN with inhibition zone 27and 21 mm diameter, respectively. By increasing the concentration of propolis extract (20, 40, 80 and 160 mg/ml) the inhibition zone was increased (7, 15, 21 and 24 mm), respectively (Fig 2). While *E. coli* was resistant to all used antibiotics (N, E, DO, SXT) except CN with inhibition zone of 19 mm and the diameter of the inhibition zone against propolis extract was increased by increasing the concentration (0, 10, 19, and 23mm) (Fig 2).

MIC of propolis on the selected bacterial species (dilution method)

MIC value of propolis against *S. aureus* and *E. coli* strains was 6.4 mg/ml for both species.

Antifungal activites of propolis

Agar well diffusion test

As shown in Table 2 ,Fig 3, the obtained data of antifungal activity of propolis at different concentrations (2.5, 5, 10 and 20 mg/l) revealed, that propolis showed significant antifungal effect against all tested yeasts at all concentrations. At 20 mg/ml was more effective, showed inhibition zones of 19, 20, 18, 20 and 28 mm for *Candida albicans ,Rhodotorula rubra, Geotrichum candidum, Cryptococcus neoformans and Trichosporon cutaneum* respectively.

Media incorporated with propolis and other antimycotic drugs

Broth media (SDB) incorporated with 20 and 40 mg/l propolis and other antimycotic drugs

The antifungal activity of propolis and other drugs was indicated by the decrease in growth rates of tested fungi when compared with control group and the activities increased as their concentration increased as shown inTable 3.

Agar media (PDA) incorporated with 40, 80 and 160 mg/l propolis and other antimycotic drugs

At 40 mg /l concentration, Propolis and all the drugs completely inhibit the growth of dermatophytes (Table 4,Fig 4),while in case of non dermatophytes , only Terbinafine (the most effective) cause complete inhibition of all non dermatophytes, followed by Itraconazole and Fluconazole . While propolis permited moderate growth (++) equivalent to other antimycotic drugs as shown inTable 4.

At 80 mg/l concentration, Terbinafine was excluded (as it caused complete growth inhibition with all dermatophyte and non dermatophyte moulds at 40 mg/l concentration). Propolis,Itraconazole and Fluconazole were applied only on non dermatophyte moulds, the most efffective drug was Itraconazole followed by Fluconazole and propolis , as shown in Table 5 .

At 160 mg/l concentration, propolis revealed complete inhibition for all used non dermatophyte moulds as depicated in Table 6 , Fig 5.

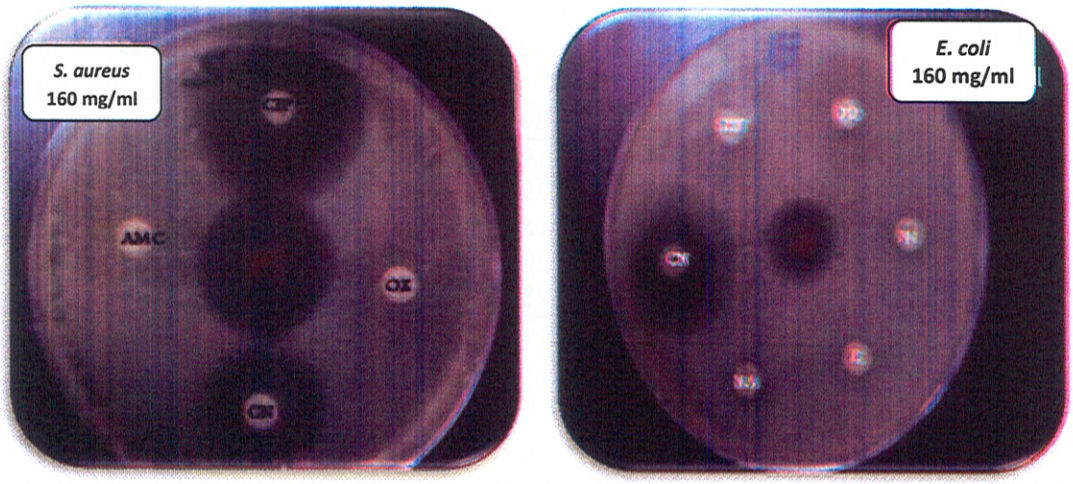

Fig. 1. Disc diffusion susceptibility test of tested bacteria against propolis and antibacterial agents

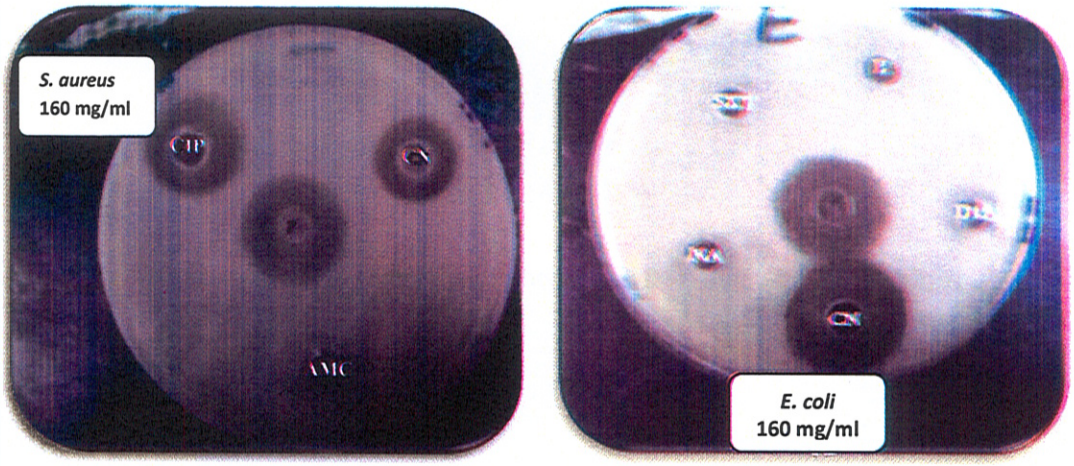

Fig 2. Agar well diffusion susceptibility test of tested bacteria against propolis and antibacterial agents

Table 2. Agar well diffusion test for yeast species against different concentrations of propolis after 72h

Strain	Diameter of inhibition zone in mm with different propolis concentrations (mg/ml)			
	20	**10**	**5**	**2.5**
Candida albicans	19	16	15	13
Rhodotorula rubra	20	16	13	-
Geotrichum candidum	18	17	14	11
Cryptococcus neoformans	20	18	14	11
Trichosporon cutaneum	28	20	18	16

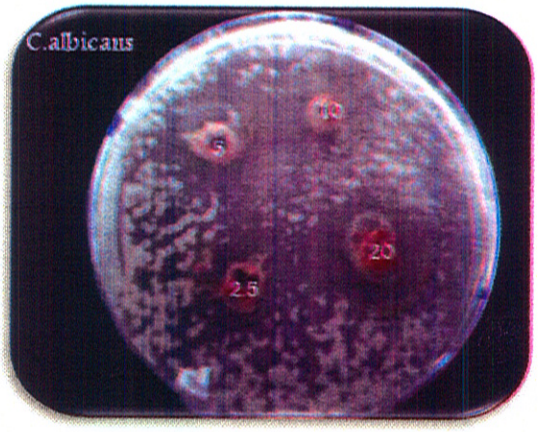

Fig. 3. Agar well diffusion test of *Candida albicans* against different concentrations of propolis after 72 h

Table 3. The mean values of fungal growth weigpht in the two concentrations 20 and 40 mg/l of propolis and other antimycotic drugs

Strain \ Tested material	Control	Propolis	Itraconazole	Fluconazole	Terbinafine
Candida albicans (1)	2080	1750	1780	1270	1460
Candida albicans (2)		1370	1610	1250	820
Rhodotorula rubra (1)	2410	1650	1730	690	2090
Rhodotorula rubra (2)		1070	750	80	1970
Cryptococcus neoformans (1)	1990	1520	1000	1550	1780
Cryptococcus neoformans (2)		1480	930	1340	1090
Geotrichum candidum (1)	2260	1570	1920	760	1730
Geotrichum candidum (2)		950	1050	1060	590
Trichosporon Cutaneum (1)	2230	1160	1730	400	1640
Trichosporon cutaneum (2)		1150	1700	150	1610
Fusarium moliniforme (1)	1500	1360	1470	860	1360
Fusarium moliniforme (2)		1320	520	660	1220
Alternaria alternate (1)	2250	2180	1570	1630	1690
Alternaria alternate (2)		1840	430	1310	750
Penicellium spp *(1)*	1480	1440	1280	1470	1440
Penicellium spp *(2)*		860	960	480	1100

(1) 20 mg/l concentration (2) 40 mg/l concentration

Table 4. Growth of the dermatophyte and non dermatophyte moulds on PDA incorporated with 40 mg/l propolis and other antimycotic drugs after 10 days

Strain \ Tested material	Control	Propolis	Itraconazole	Fluconazole	Terbinafine
Non dermatophytes					
1.Fusarium moliniforme					
(a)	++++	++	+	++	-
(b)	++++	++	+	++	-
2.Alternaria alternate					
(a)	++++	++	+	++	-
(b)	++++	++	+	++	-
3-penicellium spp.					
(a)	++++	++	+	+	-
(b)	++++	++	+	+	-
Dermatophytes					
1- Trichophyton rubrum					
(a)	+++	-	-	-	-
(b)	+++	-	-	-	-
2.Trichophyton mentagophytes					
(a)	+++	-	-	-	-
(b)	+++	-	-	-	-
3- Microsporum canis					
(a)	+++	-	-	-	-
(b)	+++	-	-	-	-
4- Microsporum gypseum					
(a)	+++	-	-	-	-
(b)	+++	-	-	-	-

++++ Very good growth. + Poor growth. +++ Good growth. -No growth.
++ Moderate growth. N.B: all tests were duplicated (a, b) for confirmation.

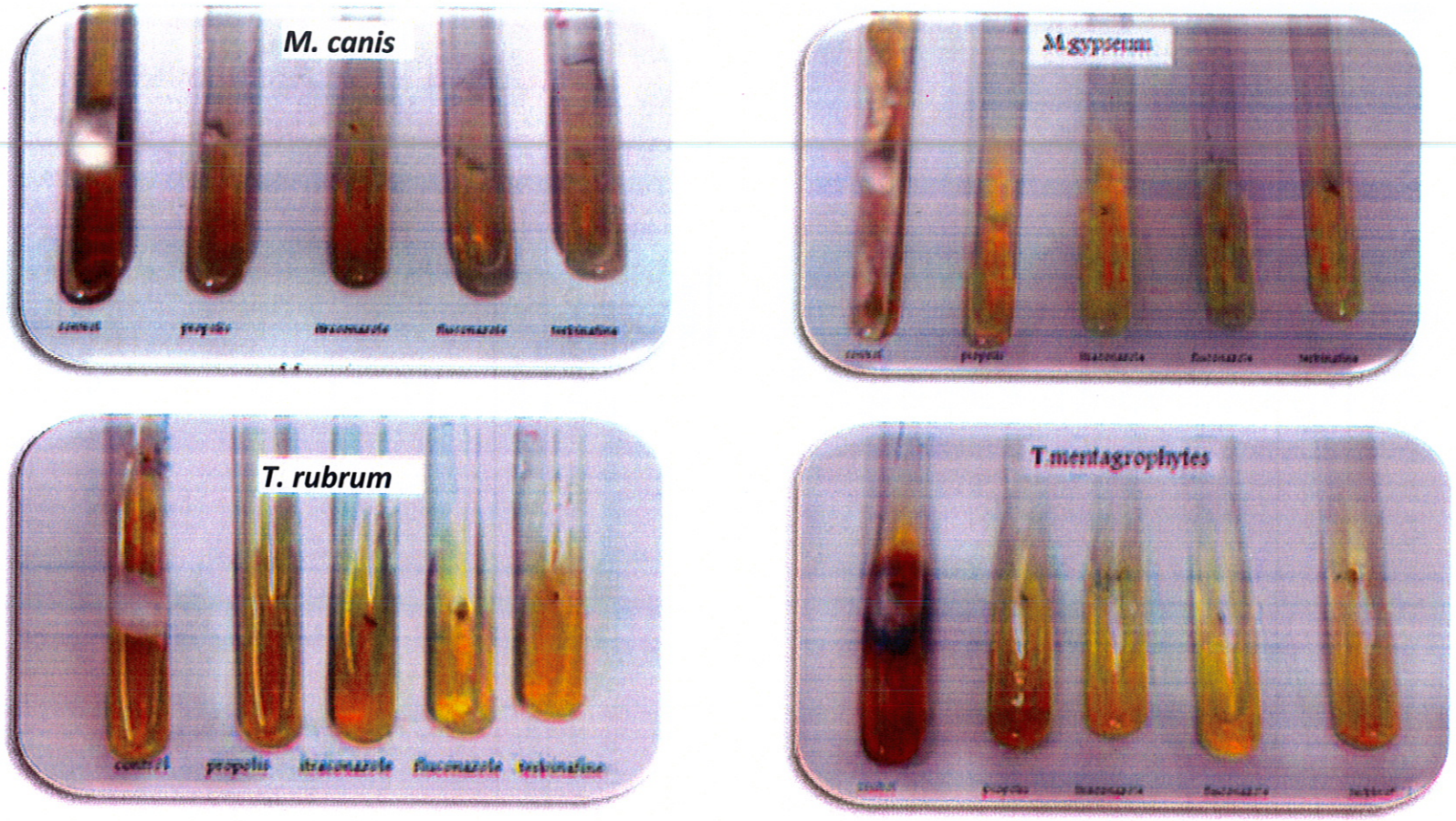

Fig. 4. Absence of growth of the tested dermatophyte moulds on PDA incorporated with 40 mg/l propolis and other antimycotic drugs after 10 day.

Table 5. Growth of tested non dermatophyte moulds on PDA incorporated with 80 mg/l propolis and other antimycotic drugs after 10 days

Strain \ Tested material	Control	Propolis	Itraconazole	Fluconazole
1.Fusarium moniliforme				
(a)	++++	++	±	±
(b)	++++	++	-	±
2.Alternaria alternate				
(a)	++++	+	±	±
(b)	++++	++	±	±
3-Penicellium spp.				
(a)	++++	+	-	±
(b)	++++	+	-	±

++++ Very good growth. ± Slight or no growth.
++ Moderate growth - No growth.
\+ Poor growth.
N.B: all tests were duplicated (a, b) for confirmation.

Table 6. Growth inhibition of tested non dermatophyte moulds on PDA incorporated with 160 mg/l propolis after 10 days

Strain \ Tested material	Control	Propolis
1.Fusarium moniliforme		
(a)	++++	-
(b)	++++	-
2.Alternaria alternate		
(a)	++++	-
(b)	++++	-
3-Penicellium spp.		
(a)	++++	-
(b)	++++	-

++++ Very good growth. - No growth.
N.B: all tests were duplicated (a, b) for confirmation.

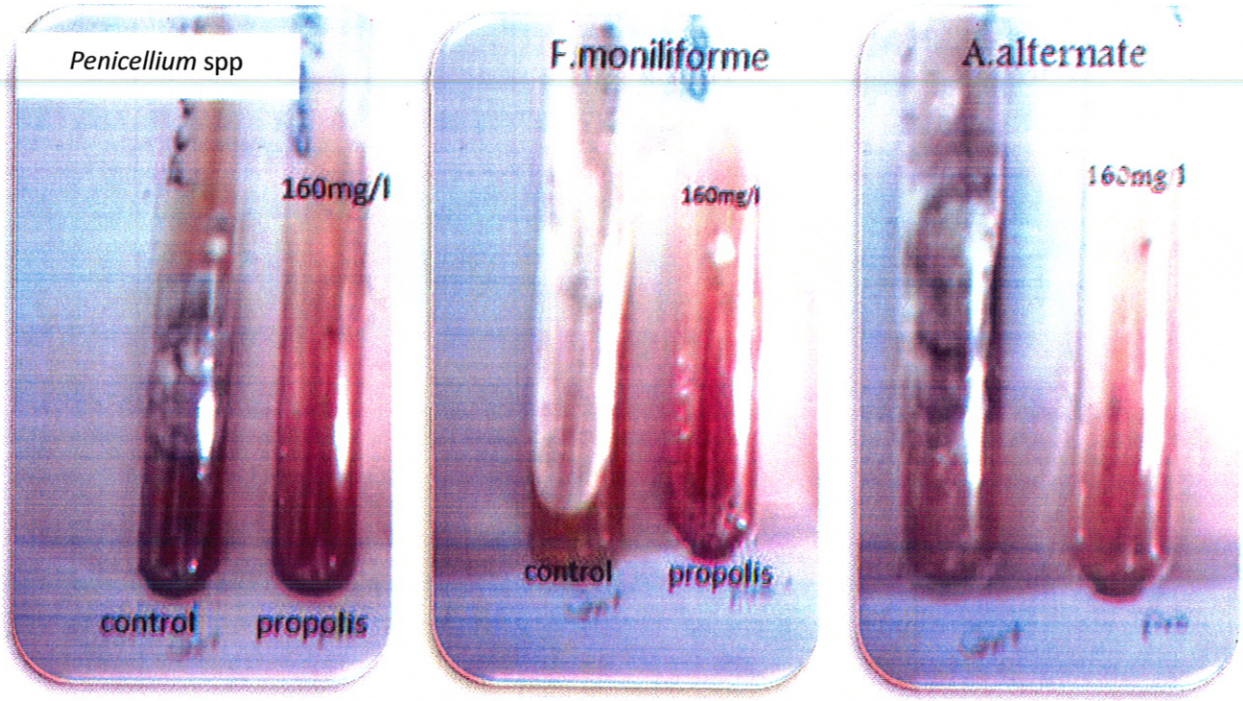

Fig. 5. Growth of the tested non dermatophyte moulds on PDA incorporated with 160 mg/l propolis after 10 day

DISCUSSION

The antimicrobial activity of propolis against a wide range of bacteria, fungi, yeasts and viruses has been investigated since the late 1940s and it showed variable activity against different microorganisms. Furthermore recent studies have shown that propolis has appreciable antibacterial and antifungal as well as antiviral actions . As a general role, propolis extract considered active against both bacteria and fungi if the zone of inhibition is greater than 6 mm *(27)*.

The procedures for detection of the antibacterial and antifungal activity of propolis in this study were based on several antimicrobial tests.

The antibacterial activity of propolis using diffusion assay (disc diffusion and agar well diffusion) and dilution assay (MIC determination) was done.

On the light of the diffusion assay in the present study, the antibacterial activity of different concentrations of propolis, 20, 40, 80 and 160 mg/ml, confirmed an inhibitory activity on the growth of tested bacterial species, moreover Gram +ve , *S.aureus*, has larger inhibition zone (7,8,16 and 23 mm) (7,15,21 and 24mm) than Gram –ve ,*E.coli*,(0, 7, 11 and 17 mm) (0, 10, 19 and 23 mm) by employing both disc and well diffusion tests respectively,which confirm previous studies in different Countries (Brazil,Kenya and Iraq) *(36-40)* and which verified that *S.aureus* is more sensitive to different types of propolis with 15-22 mm inhibition zones than *E.coli* which has lower inhibition zone (7-8.8 mm). Another study differs from our results and demonstrated that EPE inhibited the growth of *S.aureus* but do not affect *E.coli (41)*.

This investigation revealed that antibacterial activity of propolis was better than the activity of some tested antimicrobial agents (AMC, SXT, OX, NA, E and N) , but it varied in its potency according to its concentration . Similarly *S.aureus* was susceptible to all six propolis samples with inhibition zones of 15, 18, 19, 19, 20 and 22 mm and so it was more active than some used antibiotics as ampicillin, amoxicillin, garamycin, nalidixic acid, streptomycin, tetracycline, ciprofloxacin, erythromycin and cephalexin with inhibition zones ranging from 8 to 12 mm , *(39)*.

Regarding to the dilution assay in this study, the MIC of EPE that inhibited both S.*aureus* and *E.coli* growth, was the same (6.4 mg/ml). And this was confirmed by previous study which showed that propolis has antibacterial activities against *S.aureus* and *E.coli* with equal and higher MIC value (14 µg/ml) *(42)*. Different results reported the antibacterial activity of propolis by dilution assay against *S.aureus* with MIC value 0.175-0.7 mg/ml and showed weak inhibitory action with *E.coli* (28 mg/ml) *(39)* or without any action against *E.coli (43)*.

For the antifungal activity, diffusion (agar well diffusion test) and dilution assay (SDB and PDA incorporated with propolis and other antimycotic drugs) were used.

The agar well diffusion test was carried out in the present study against five yeast species with different propolis concentrations (2.5, 5, 10, 20 mg/ml). Propolis showed a significant antifungal activity against all tested yeasts at all concentrations and the zones of inhibition ranged from 11 to 28 mm and was increased by increasing the concentration except *R.rubra* which gave no inhibition zone at 2.5 mg/ml. The *in vitro* activity of Itraconazole and propolis against *C.albicans, C.glabrata, Trichosporon* spp and *Rhodotoruola* spp was recorded *(22)* . Meanwhile other auther *(44)* recorded that EPE showed moderate activity against *C.albicans* with 13.2 mm inhibition zone diameter at 200 mg/ml concentration by agar well diffusion test.

Concerning with the dillution assay , yeast spp, were sensitive to propolis and this result is in agreement with previous study *(45-47)*. While dermatophyte moulds were inhibited by propolis at 40 mg/l .The susceptibility of *T. rubrum* and *T. mentagrophtes* to green and red Brazilian propolis was demonstrated by using broth micro dilution method *(48)*. Also

several authers *(47,49)* agreed with our study and clarified that PE showed strong inhibitory action against *T.mentagropytes, T. rubrum* and *M. gypseum.* From the afore-mentioned data, EPE with other used antimycotic drugs (itraconazole, fluconazole and terbinafine) showed the same inhibitory action against all dermatophyte moulds at the concentration of 40 mg/l, this result differ from the previous investigations *(50)* which showed that Terbinafine, Itraconazole and propolis were the most active agents against *T. rubrum* and Ketoconazole and Fluconazole were the least active. Whereas the non dermatophyte moulds, in present work ,revealed that it needs a higher concentration of propolis (160 mg/l) more than dermatophytes and yeasts to be inhibited and this differ from the available data *(51)* which has been demonstrated that pure PE of a concentration of 15-30 mg/ml was needed to inhibit the growth of *Penicillium viridicatum* and *Penicillium notatum.*

Finally we can say that propolis merits further investigation as potentially useful agent for treatment of fungal and bacterial infections.

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الملخص العربي

نشاط صمغ النحل المقاوم للميكروبات ضد بعض البكتريا والفطريات

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ان الآثار الجانبية لمضادات الميكروبات وزيادة مقاومة الميكروبات للعديد منها جعلت فائدتها العلاجية مهددة مما جعل البحث عن عقارات جديدة شيء ضروري وحتمي.

وقد تم أجراء هذا البحث للكشف عن نشاط صمغ النحل المضاد للبكتيريا والفطريات ولإلقاء الضوء على امكانية استخدامه كدواء بديل أقل في التكلفة و السمية.

ولتحقيق هذا الهدف تم إجراء الآتي :

اختبار حساسية بعض العترات الفطرية والبكتيرية الاتيه لصمغ النحل:

الفطريات الجلديه واللاجلديه (*بنيسيلليوم, فيوزاريوم مولينيفورمي , ألترناريا ألترناتا , تريكوفيتون روبرم , تريكوفيتون منتاجروفيت , ميكروسبورم كانز و ميكروسبورم جيبسيوم*) بالاضافه الى خمس اجناس من الخمائر (*كانديدا ألبيكانز, كريبتو كوكاس نيو فورمانس , رودوتوريولا روبرا , تريكوسبورون كيوتانيوم و جيوتريكم كانديدم*) . كذلك عترتين من البكتيريا (*ستافيلوكوكس أوريس و إيشيريشيا كولاي*).

ولقد أوضحت الإختبارات الآتي:

- تأثرت العترات البكتيرية المستخدمة (*ستافيلوكوكس أوريس و إيشيريشيا كولاي*) باستخدام تقنية انتشار المضاد عبر القرص وتراوح قطر المنطقة التي انعدم فيها النموبين ٧مم إلى ٢٣مم لميكروب *ستافيلوكوكس أوريس* وتراوح بين صفر مم إلى ١٧ مم لميكروب *إيشيريشيا كولاي*.
- وكذلك باستخدام تقنية انتشار المضاد عبر البئر وتراوح قطر المنطقة التي انعدم فيها النموبين ٧مم إلى٢٤مم لميكروب *ستافيلوكوكس أوريس* وتراوح بين صفر مم إلى٢٣ مم لميكروب *إيشيريشيا كولاي*.
- و بتحديد أقل تركيز مثبط كانت النتيجة ٦,٤ مج/مل لكلا من(*ستافيلوكوكس أوريس و إيشيريشيا كولاي*) .
- كما اوضحت الاختبارات ان كل اجناس الخمائر كانت حساسة لصمغ العسل باستخدم تقنية إنتشار صمغ العسل عبر البئر وكذلك تقنية التخفيف ورتبت الاجناس على لنحو *التالي* : *تريكوسبورون كيوتانيوم* > *كريبتو كوكاس- نيو فورمانس و رودوتوريولا روبرا* > *كانديدا ألبيكانز*> *جيوتريكم كانديدم*.
- اما بالنسبه لاجناس الفطريات:فقد ظهرتاثير صمغ النحل باستخدام تقنية التخفيف عبر المستنبتات الشامله عند التركيز ٤٠ مج/مل للفطريات الجلديه وهو مساوى للمضادات الاخرى وعند ١٦٠ مج/مل للفطريات اللاجلديه وكان اعلى من تركيزات المضادات الفطريه المستخدمه .