

ANTIBACTERIAL ACTIVITY OF PROPOLIS ON BACTERIA
ISOLATED FROM CHILDREN WITH IMPETIGO
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ABSTRACT

Background: Impetigo is a contagious superficial pyogenic infection of the skin , caused by *Staphylococcus aureus* or *Streptococcus pyogenes* and some times *proteus mirabilis* , different antibiotics was used for the treatment of impetigo , given topically or systemically .

Objectives: The aim of the present work is to estimate the effectiveness of propolis on different types of bacteria which was isolated from children suffering from impetigo skin infection .

Methods: The study was done in the College of Medicine / Diyala University. Forty two samples with positive bacterial cultures, were obtained from the skin lesions of children with impetigo infection , for the period of 2/Feb. to 30/May/2010 . They were tested by doing antibiotic sensitivity test of different antibiotics and the resistant strains of different types of bacteria , were tested by 10% of ethanolic extract of propolis and it's phenolic compounds .

Results: The study revealed that 10% of ethanolic extract of propolis and it's phenolic compounds was effective as antibacterial agents, against antibiotic's resistant strains of *Staph . aureus*, *Strep. Pyogenes* and *Proteus mirabilis*.

Keyword: Impetigo, *Staph. aureus* , *Strep. pyogenes* , *Protus mirabilis* , propolis .

نشاط العكبر المضاد للجراثيم المعزولة من الاطفال المصابين بالقوباء

بروج محمد رزوقي

كلية الطب / جامعة ديالى

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الخلاصة

تمهيد: القوباء هو التهاب الجلد الجرثومي المعدي السطحي تسببه جرثيم المكورات العنقودية الذهبية أو المكورات المسبحية وفي بعض الأحيان المتقلبات . يعالج بأنواع مختلفة من المضادات الحيوية بعضها تعطى موضعياً والأخرى جهازياً

الهدف: هذه الدراسة هو لدراسة تأثير المستخلص الكحولي للعكبر على انواع الجراثيم المعزولة من الاطفال المصابين بالقوباء .

الطرق: أخذت 42 عينة جرثومية من المكورات العنقودية الذهبية والمكورات المسبحية والمتقلبات لمرضى القوباء وتم إجراء اختبار الحساسية لسبعة من مضادات الحياة وتم انتقاء العزلات المقاومة لإجراء اختبار الحساسية للمستخلص الكحولي للعكبر ومركباته الفينولية عليها .

النتائج : أظهرت الدراسة بان المستخلص الكحولي للعكبر ومركباته الفينولية ذات تأثير كمضاد حيوي على المسببات الجرثومية المختلفة للقوباء والتي تشمل جراثيم المكورات العنقودية والمكورات المسبحية والمتقلبات.

الكلمات المفتاحية: القوباء , المكورات العنقودية , المكورات المسبحية , المتقلبات , العكبر .

INTRODUCTION

Impetigo is a contagious superficial pyogenic infection of the skin , it is of two main clinical forms : non – bullous impetigo (Impetigo Contagiosa of Tilbury Fox) and bullous impetigo⁽¹⁾. Contagiosa Bullous impetigo is accepted as a *Staphylococcal* disease , although *Streptococcal* bullous impetigo has been reported⁽²⁾ . The non – bullous form may be caused by *Staphylococcus aureus* , *Streptococcus pyogenes* , or by both organisms together , it accounts for more than 70 % of all cases , rarely caused by *Proteus mirabilis* (1,2,3,4,5) .

Diagnosis is made by clinical criteria followed by Gram stain and confirmed by culture of exudates from lesion⁽³⁾ .

Treatment : in mild and localized infection , a topical antibiotic alone may suffice e.g. mupirocin , fusidic acid , bacitracin for both *Staphylococcal* and *Streptococcal* impetigo , if the infection is wide – spread or sever or is accompanied by lymphadenopathy or if there is reason to suspect a nephritogenic *Streptococcus* , an oral antibiotic such as flucloxacillin , or erythromycin is indicated also azithromycin , cephalixin , cefaelor , cefprozil and

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clindamycin are alternative therapies ^(1,3) . Black tea (as 2% topical ointment) also give a good result in treatment ⁽⁶⁾ .

Propolis is one of the honey comb products which is produced by the bees and used to close the pores and opening of the honey comb to protect it from infectious agents ⁽⁷⁾ . Propolis is used in medicine since thousands of years , as antibacterial , antifungal (8,9,10) , antiviral (11) and antineoplastic agent (12) . Propolis has a complicated structure , which is composed of more than 200 substances include : 50%resins , 30%wax , 10%essential oils , 5%pollen and 5%organic compounds like sugars , vitamins and amino acid , also contain phenolic compounds (e.g. benzoic acids , cinnamic acid , aldehydes , steroidetc) (13) . Propolis was found to be effective as antibacterial agent against different microbes isolated from different systemic infection (14) .

The aim of the present study is to evaluate the antibacterial effects of ethanolic extract of propolis and it's phenolic compounds on different types of bacteria , isolated from children with impetigo .

METHODS

The study was done in Departments of Medicine (Unite of Dermatology) and of Microbiology / College of Medicine / Diyala University for the period 2-Feb. to 30-May 2012 . Forty two samples of cultured bacteria , isolated from children with impetigo , which were collected by using cotton swap and cultured on MacConky and Blood agar , biochemical tests were used for isolation and identification of different types of bacteria (15) . The samples were tested by antibiotic sensitivity test by using Kirby-bauer disk method , which include: methicillin , cefotaxime , cefalexin , piperacillin , gentamicin , erythromycin and ciprofloxacin , ⁽¹⁶⁾ . The resistant strains to the preceding antibiotics and to the 95% ethanol (selected according to the diameter of inhibition zone) , were selected and sensitivity testing to ethanolic extract of propolis and it's phenolic compounds (10mg/ml stock solution=10% concentration) was done by using agar-well diffusion(Iraqi honey comb) ⁽¹⁷⁾ . Different concentrations of propolis extract (1mg,2mg,3mg,4mg and 5mg/ ml) were obtained from the original stock solution of propolis and diluted by using 95% ethanol (to obtain a total 10ml for

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each concentration) and tested on the resistant strains of isolated bacteria , by measuring the diameter of inhibition zone .

Also the MIC and MBC of different concentration (1mg,2mg,3mg,4mg and5mg/ ml) of propolis and it's phenolic compound extract were measured by using Agar – dilution method⁽¹⁸⁾ .

RESULTS

Table-1 shows the antibiotic sensitivity tests of different types of antibiotic on different types of bacteria . The percentage of resistance of *Staph. aureus* to cephalixin , methicillin and erythromycin was (50 , 70.8 and 62.5%) , *Strep. pyogenes* resistance was (71.4 , 71.4 and 78.6%) and *Proteus mirabilis* resistance was (50 , 50 and 75%) respectively , while resistance of *Staph. aureus* to cefotaxim and gentamicin was (37.5 and 33.3%) , *Strep . pyogenes* resistance was 42.9% for both and *Proteus mirabilis* resistance was (25% for both antibiotics) . The resistance of *Staph .aureus* to ciprofloxacin and piperacillin was (33.3 and 41.7%) , *Strep. pyogenes* resistance was 35.7% for both and *Proteus mirabilis* has no resistance to both antibiotics .

The study shows that the propolis and it's phenolic compounds was effective against different types of resistant bacteria (*Staph aureus* , *Strep. pyogenes* and *Proteus mirabilis*) as in table-2 . For *Staph aureus* (Figure-1) the inhibition diameters was (30.12 , 31.81 , 38.32 , 39.21 , 41.20)mm for different concentrations of phenolic compounds , to *Strep pyogenes* (Figure-2) was (25.18 , 26.20 , 28.17 , 31.33 , 34.12) mm and for *Proteus mirabilis* (Figure-3) was (0,00 , 7.12 , 10.14 , 13.15 , 15.12)mm . Its antibacterial effect was directly proportional with concentration of propolis and it's phenolic compounds . According to the values of MIC & MBC as in table-3 , the results revealed that the phenolic compounds were more effective as antibacterial agent than the propolis against *Proteus mirabilis* , *Strep. Pyogenes* , than *Staph. aureus* which was due to the presence of oxidative compounds , which were highly toxic against bacteria⁽¹⁴⁾ .

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Table-1 : Percentage of resistant bacteria to different antibiotics .

Antibiotics	<i>Staph. aureus</i> -total 24samples	<i>Strep. pyogenes</i> – total 14samples	<i>Proteus mirabilis</i> - total 4samples
Cephalexin	Resistant-12 50%	10 71.4 %	2 50%
Cefotaxime	Resistant-9 37.5%	6 42.9%	1 25%
Methicillin	Resistant-17 70.8%	10 71.4%	2 50.%
Erythromycin	Resistant-15 62.5%	11 78.6%	3 75%
Gentamicin	Resistant-8 33.3%	6 42.9%	1 25%
Piperacillin(Resistant-10 41.7%	5 35.7%	0 0%
Ciprofloxacin	Resistant-8 33.3%	5 35.7%	0 0%

Table-2 : Inhibition zone (mm) of propolis and it's phenolic compounds on resistant bacteria .

Bacteria	Extract	Inhibition zone in different concentrations (mm)				
		1mg/ml	2mg/ml	3mg/ml	4mg/ml	5mg/ml
<i>Staph. aureus</i>	Phenolic C.	30.12	31.81	38.32	39.21	41.20
	Propolis	13.28	14.12	15.20	17.40	18.28
<i>Strep. pyogenes</i>	Phenolic C.	25.18	26.20	28.17	31.33	34.12
	Propolis	1.00	1.50	8.15	10.17	11.14
<i>Proteus mirabilis</i>	Phenolic C.	13.40	14.21	19.20	22.14	24.11
	Propolis	0.00	7.12	10.14	13.15	15.12

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Table-3 : Values of MIC and MBC of propolis and it's phenolic compounds(mg/ml).

Resistant bacteria	Propolis		Phenolic compounds	
	MIC	MBC	MIC	MBC
<i>Staph. aureus</i>	2	3	1	2
<i>Strep. pyogenes</i>	2	4	1	2
<i>Proteus mirabilis</i>	3	5	2	3

MIC: Minimum inhibition concentration .

MBC: Minimum bactericidal concentration .

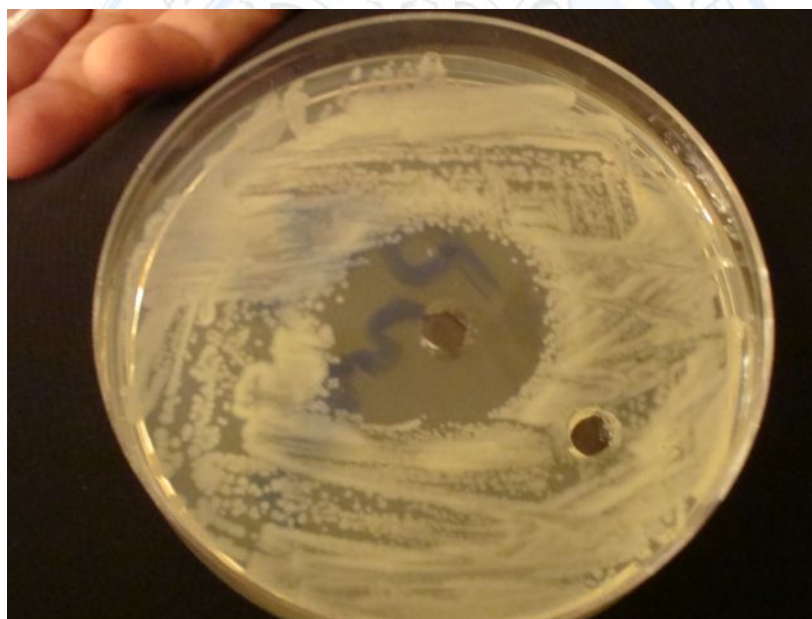


Figure -1. *Staph. aureus* inhibition zone

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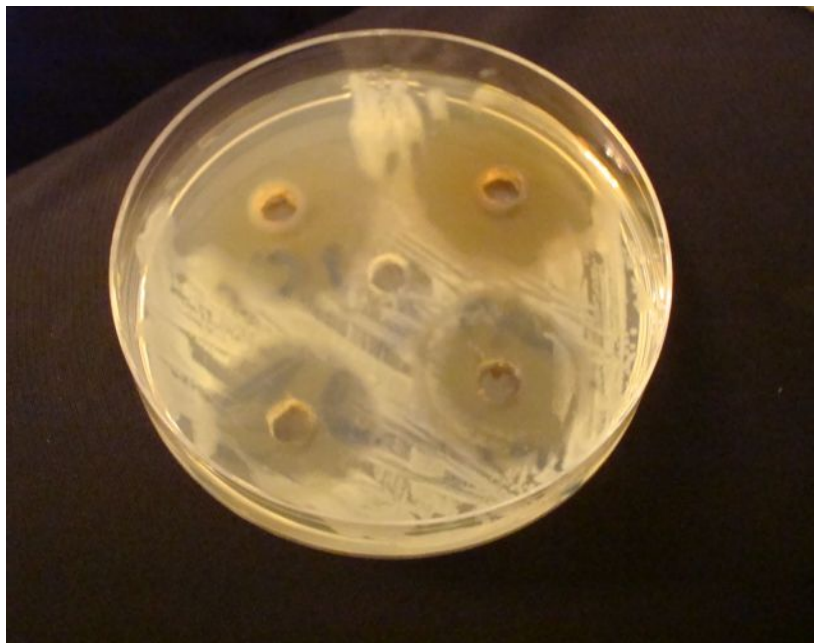


Figure -2. *Strep. pyogenes* inhibition zone



Figure-3. *Proteus mirabilis* inhibition zone

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DISCUSSION

The results of this study revealed that the propolis and its phenolic compounds were effective in vitro as an antibacterial agent and *Staph. aureus* was more sensitive than *Protues mirabilis* and *Strep. pyogenes*. The antibacterial activity of propolis was due to the presence of phenolic compounds e.g. diterpenic acid, which cause denaturation of protein of the cell membrane and kill the bacteria⁽¹⁹⁾. Also the study shows that the antibacterial activity of propolis and its phenolic compounds were more effective in vitro as antibacterial agent than different types of antibiotics, black and green tea^(1,3,6,10,14,18). In this study the propolis was more effective than green and black tea (4,6), also more effective than different types of antibiotics (1,3).

We concluded that the ethanolic extract of propolis was relatively effective as antibacterial agent in vitro and its effect was directly proportional with its concentration.

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