Effect of Pomegranate barks solution (*Punica granatum L*) On some Pathogenic bacteria in vitro

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Summary

Powder of pomegranate barks (Punica granatum L) was prepared. Different concentration of watery pomegranate solution contain, 5, 10, 20, and 40 mg / ml were prepared Six types of pathogenic bacteria were used. Suspension was made from those bacteria and each type of bacteria contain (10x1 - 10x2) / ml. Those bacteria include Escherichia coli, Salmonella, Staphylococcus, Streptococcus, Klebsiella and Corynebacterium The affect of the plant solution of pomegranate was tested on petri – dishes containing Trypticase Soya agar by diffuse agar method. Equal amount of each type of the tested bacteria was mixed with the varying concentration of plant of pomegranate barks solution. All petri – dishes of the tested bacteria with the pomegranate barks powder were incubated at 37 c for 1-2 days. The result showed that the pomegranate barks solution of 40 mg / ml had the strongest inhibiting zone on all tested types of bacteria, while solution of 20 mg/ml. showed weak inhibiting zone. But the lower concentration showed no inhibiting zone for any of the tested bacteria.

تأثير محلول قشور الرمان على بعض الجراثيم المرضية في الزجاج

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الخلاصة

حضر مسحوق قشور الرمان وحضر منه محاليل مائية مختلفة التركيز (5 ، 10، 40،20) ثم حضر معلق جرثومي لستة أنواع من الجراثيم تحتوي على (10x1-10x2) مل وشملت الجراثيم الاشريشيا القولونية ، السالمونيلا ، المكورات العنقودية ،المكورات المسبحيه الكلبسيه والوتديات . تم فحص تأثير المحلول المائي لنبات قشور الرمان على أطباق بتري تحتوي على أوساط زرعيه صلبه من التربتوكوكس صويا أكر باستخدام طريقة الانتشار الاكر . أخذت كميه متساوية من كل نوع من هذه الجراثيم ومزجت مع مختلف التراكيز من محلول قشور الرمان في أطباق الأوساط الزرعيه وحضنت الأطباق جميعها بدرجة حرارة 37 م لمدة 1-2 يوم . أظهرت النتائج أن المحلول المائي لقشور الرمان بتركيز (40 ملغم /مل) له تأثير مثبط قوي على جميع أنواع الجراثيم التي درست وأظهر المحلول المائي لقشور الرمان بتركيز (20 ملغم/مل) تأثير ضعيف في حين لم تظهر التراكيز الأخرى أي تأثير على جميع أنواع الجراثيم التي درست .

Introduction

Suggesting that pomegranate barks antidiarrheaic action, against bleeding and accelerate wound healing was reported by Matomed and David, incited by (16, 19) other reported its activity in treatment of tape worm, against aid virus and bacteria especially that causing food poisoning such as Salmonella (19). Many reports indicate that E. coli and Salmonella cause diarrhea, while Staphylococcus and streptococcus can cause mastitis and other diseases in human and animals and Corynebacterium can cause many diseases (1,3,4,5,6,7,8,9,12,13,14,20) the aim of the present research is to study the effect of hydrolic solution of pomegranate barks on some pathogenic bacteria.

Materials and methods

Pomegranate barks (Punica granatum -L) was dried at room temperature 25 Ċ, samples collected randomly and send to the Iraqi national herbarium for identification. Adry bomegrante barks was rounded by electrical blender and kept in plastic bottles . A total of (20) grams powder were suspended (50) ml of sterile distalled water ,mixed by sterile glass rod and left at room temperature until cooling, then refined by metal net funnel. The concentration would be 40 mg /ml and considered as NO1. Used as stock solution, The pH of the above solution was 3.1, adjusted by pH meter. Stock solution as 20 mg/ ml. 10 mg /ml and 5 mg /ml was made by diluting stock no. 1 as 40 mg / ml. by sterile distilled water and were given numbered as stock 2,3 and 4 respectively. Trypticase Soya agar media was prepared in petri- dishes, one plate was made for each type bacteria and designated as plates I, II, III, IV, V and VI, which was used for E. coli, Salmonella, Klebsiella, Staphylococcus, Streptococcus and Corynebacterium respectively, supplied by the Department of Microbiology, College of Veterinary Medicine Baghdad university. Each type of bacteria was inoculated in Trypticase Soya broth and incubated at 37 c for 24 hours. Serial dilution technique using sterile broth at 10x1 –10x2 bacteria/ml. suspension. In each plate of the media five holes were made by wide mouth of sterile pasture pipette and designated as beta 1, 2, 3, 4, 5, and 6. Diffusion agar technique was used (11,15) plate number I for E. coli drop of E.coli was put in every hole, varying concentration of pomegranate barks solution started from 40 mg /ml then 20, 10, 5 mg/ml was put to each hole ,sterilewater was in to the last hole . Plate no II. Was for Salmonella, the plate No. III for staphylococcus, plate IV for streptococcus, plate V for Klebsiella and plate VI for Corynebacterium, using the same procedure . Then all plates were be incubated at 37c for 1-2 days.

Results

The results in table (1) showed the concentration at 40 mg/ml of pomegranate barks solution was the most largest inhibition zone surrounding the hole containing Corynebacterium the affect was graded as (+++) . It was followed by grade (++) for each of E.coli Salmonella, and Klebsiella, while those as Staphylococcus and Streptococcus was less as (+±). For concentration at 20 mg / ml of pomegranate barks solution , the strongest effect was on Corynebacterium . and graded as (+±) and (+) for each of Staphylococcus and Streptococcus . No inhibition was noted at concentration of 10 mg / ml, 5 mg / ml and with sterile distilled water.

Table No.(1)

Name of bacteria	Inhibition zone at a concentration				Control
	40 mg /ml	20 mg /ml	10 mg/ml	5 mg /ml	
E. coli	++	+	-	-	-
Salmonella	++	+	-	-	-
Klebsiella	++	+	-	-	-
Staphylococcus	+±	±	-	-	-
Streptococcus	+±	±	-	-	-
Corynebacterium	+++	+±	-	-	-

+++ = Largest inhibition zone
++ = Less inhibition zone
+ = Small inhibition zone
- = no inhibition zone

Discussion

It appeared from the tested findings that pomegranate barks solution at high concentration of 40 and 20 mg/ ml., had the greatest bactericidal activity shown by the wideinhibition zone due to its contents of tanninus, collotninic acid and oxalic acid (16, 17).the bactericidal activity could be also due to the presence of specific substances in pomegranate barks solution, which affect the growth of bacteria, such as pelletiernin , which include isopelletierine, methypseudopelletierine resine , sugar , tannin and mucilage (10 , 18 , 19) .Many researches workers were reported that 19 substances in pomegranate barks solution (12, 17, and 18). The present paper showed that Corynebacterium was more sensitive to Ponegranate followed by E.coli, Salmonella, and Klebsiella then Staphylococcus and Streptococcus were the lesser sensitive, this might be due to variation in composition of bacterial structure. . Other research workers reported that pomegranate has an effect on bacteria that causing food poisoning such as Salmonella,(19). Further research needed to identify more ingredients in the pomegranate barks that has an effect on bacteria growth by using more modified technique .

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