

The *in vitro* Antibacterial activity of *Citrus aurantifolia* var *acidica* fruits

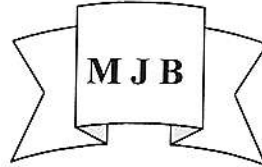
A. R. Abdul – Latif , Mohammed A. K. Al- Saadi

B. A. Al- Khafaji , E. A. S. Al-Janabi *

Babylon University , College of Medicine Dept. of Pharmacology

,Hilla,Iraq

* Babylon University , College of Medicine, Dept. of Microbiology



Abstract

The *in vitro* antibacterial activity of *Citrus aurantifolia* fruits (CA) is being reported . Three extraction methods were tried as : Hot water (HWCA) cold water (CWCA) , and alcoholic (ACA) .

The screening of antibacterial activity (AA) was done by agar diffusion technique (ADT) . Minimum Inhibitory Concentration (MIC) studies were made by agar dilution technique (ADT) . the AA and MIC studies were conducted on Muller – Hinton agar (MHA) . The *in vitro* AA from highest to the lowest effective were as follows, ACA , CWCA , and HWCA respectively . The MICs were 3.125 , 6.25 , 6.25 and 12.5 mg/ml for *S. aureus* , *Proteus vulgaris* , *Pseudomonas aeruginosa* and *E. coli* respectively . *K. pneumoniae* was unaffected by ACA, CWCA and HWCA . Such finding may be due to capsule permeability masking effect.

ACA can be recommended on an *in vitro* basis as antibacterial agent in topical pharmaceutical preparations and as canned food preservative .

الخلاصة

تم في هذه الدراسة تسجيل لنشاط خلاصات ثمر نومي البصرة *Citrus aurantifolia* في الزجاج ، اذ جرى تحضير ثلاثة انواع من الخلاصات ، خلاصة الماء الحار ، خلاصة الماء البارد وخلاصة الكحول . تم التحري عن الفعل المضاد للبكتريا بطريقة الانتشار في الاكار وتحديد التركيز المثبط الادنى (MIC) بطريقة التخفيف في الاكار باستخدام وسط اكار مولر هنتن . وترتب نشاط الخلاصات المضاد للبكتريا من الاعلى حتى الادنى تأثيراً : خلاصة الكحول وخلاصة الماء البارد ثم خلاصة الماء الحار وتبين بان التركيز المثبط الادنى (MIC) هو 3.125 ، 6.25 ، 6.25 ، 12.5 ملغم /مل لكل من *S. aureus* ، *Proteus vulgaris* ، *Pseudomonas aeruginosa* ، *E. coli* على التوالي . واتضح بان *K. pneumoniae* لا تتأثر بالخلاصات الثلاث المدروسة ومثل هذه النتيجة توحى الى فعل حجب التناضح من قبل المحفظة البكتيرية وبناءً على نتائج هذا العمل ربما من المعقول التوصية باستخدامه في التحضيرات الصيدلانية ذات الاستعمال الخارجي او كمادة حافظة للاغذية المعلبة.

Introduction

Botanical world consist of an array of feed , poisonous , economic and medicinal plants (1-3). Among these plants *Citrus aurantifolia* var *acidica* (

Nomi Busrah) was a subject of several investigations to uncover its biological potentials . Watery extracts of this fruit showed a prostogen like effect (4) and

Table 1 Judgement of the *in vitro* antibacterial activity of *C. aurantifolia* var *acidica* (Nomi Busrah).

Parameters	
Inhibition zones (mm)	Conclusion
0 – 9	resistant
10 –30	sensitive
0-2	Control saline and alcoholic

Table 2 The *in vitro* antibacterial screening Millimeter inhibition zones (IZ) for different extracts of *Citrus aurantifolia* var *acidica* (150 mg/ml) against five types of bacteria. Control saline and alcoholic

Bacteria	IZ		
	Hot water Extract (HWCA)	Cold water extract (CWCA)	Alcoholic extract (ACA)
<i>S. aureus</i>	10	13	15
<i>E. coli</i>	3	3	7
<i>K. pneumonia</i>	4	3	10
<i>P. vulgaris</i>	6	6	12
<i>P. aeruginosa</i>	6	8	10

Table 3 Minimum inhibitory concentrations (MICs) mg / ml for different extracts of *Citrus aurantifolia* var *acidica* against five types of bacteria:

Bacteria	(HWCA)	(CWCA)	(ACA)
<i>S. aureus</i>	50	25	3.125
<i>E. coli</i>	75	50	12.5
<i>K. pneumoniae</i>	200	125	100
<i>P. vulgaris</i>	100	50	6.25
<i>P. aeruginosa</i>	125	75	6.25

the present study was at reporting its *in vitro* antibacterial effect .

Materials & Methods

One , gram positive and four , gram negative clinical isolates were identified as in Baron *et al* (6) . Watery hot and cold as well as alcoholic extracts were performed according to (7) . Stock solutions of 150 mg /m l and their serial double dilutions were done as in (8) .

Agar diffusion technique (ADT) was used for screening antibacterial activity . Agar dilution technique (ADT) was conducted using Müller – Hinton agar plates in accordance with (8) to determine minimum inhibitory concentration (MIC) .

Results & Discussion

Alcoholic control showed *invitro* antibacterial effect of up to 2mm inhibition zones (IZ) While saline control revealed nill activity (table 1) .

The hot water extract (HWCA) gave inhibition zones IZ ranges from 2 to 10 mm , cold water (CWCA) extracts showed IZ ranges of 3 to 13 mm and alcoholic extracts (ACA) presents IZ ranges of 7 to 15 mm using ADT (table 2). Thus the antibacterial activities of different extracts showed different IZ sizes which may be an indication for different active compo-nent

The minimum inhibitory concentration MIC ranges were ; 50 -

Thus, on summing up one may state :

1-The *in vitro* antibacterial activity from highest effective were as :ACA , CWCA then HWCA .

2-The ACA was effective against *S. aureus* , *P. vulgaris* , *P. aeruginosa* and *E. coli* with MIC of 3.125 , 6.25 , 6.25 & 12.5mg/ml respectively .

200 for HWCA , 25-75 for CWCA and 3.125- 100 mg/ml for ACA (table 3) *S. aureus* were sensitive to all test extracts . *P. vulgaris* and *P. aeruginosa* as well as *E. coli* were sensitive to ACA alone . *K. pneumoniae* was resistant to all test extracts . ACA was the most effective among the others . ACA can be an antibacterial agent (AA) *in vitro* effective against gram positive and gram negative bacteria (table 1,2,3) . ACA on *in vitro* basis may be recommended so for as food preservative mouth wash and an ingridient in skin topical preparation.(9)

3- *K. pneumonia* was resistant all test extract may be due to capsule .

4- ACA may be recomonded on an *in vitro* basis in topical pharmaceutical preparations and as food preservative.

References

- 1-Lawrence,G.H.M. 1951. Taxonomy of vascular plants. MacCmillan company : New York .
- 2-Al- Rawi, A. and Chakravarty, H.L. (1988). Medicinal plants of Iraq. 2nd ed. Baghdad .
- 3-Alyounish,A.A.A.,and AL-Karkuchy,A.A.A.1977.The Cultivation of Industrial plants in Iraq . Mosul University .Iraq .
- 4-Shihata, I.M.; Abdul Latif A.R. and Aziz, H.A. ,1978, J. Egypt . Vet. Med. Assa. 4 , 87.
- 5-Shihata, I.M. ; Abdul Latif A.R. and Al-Alousi T. I. (1980) J. Coll. Vet. Med. Mosul . ,1,1, 17.
- 6.Baron,E.J.;Peterson,L.R.&Finegold,S.M .,1994.,Diagnostic Microbiology.9th ed.Mosby.St.Louis.
- 7-Tyler, V.E. J. Am . Pharm . Assoc .,1996, 36 ,1, 29 .
- 8-Harborne, J. B., 1984 , . Phytochemical methods : A guide to modern technique of plant analysis , 2nd . ed. Chapman , Hall , London .
- 9-Woedltke , V. T. ; Schluter , B . P.;legel , P. Lindequist ,U . and Juice , W.D. ,1999, . Aspects of antimicrobial efficacy, 452.