EVALUATION OF ANTIBACTERIAL ACTIVITY OF CALLISTEMON VIMINALIS IN VITRO [+]

تقييم فعالية نبات فرشاة البُطل المضادة للجراثيم في الزجاج

Jasim H. Taher [*] **Layla S. Hameed** [**]
Bushra A. Farhan ** **Zainab A. Talib****

Key words: Plant, extract, bacteria and antibacterial.

Abstract:

The ethanolic extract of *Callistemon viminalis* (Sol. ex. Gaertn) G. Don. (the fruits), was screened for it's antibacterial activity against *Pseudomonas aeruginosa* and *Staphylococcus aureus*, isolated from some clinical specimens. Using agar diffusion technique, it was found that the extract posses antibacterial effect.

المستخلص:

درست فعالية تأثير المستخلص الكحولي لثمار نبات فرشاة البطل ضد جراثيم الزائفة الزنجارية والعنقودية الذهبية المعزولة من عينات سريرية، حيث وجد ان لهذا المستخلص فعالية ضد هذه الجراثيم بأستخدام تقنية أنتشار الآكار.

Introduction :

The development of microbial resistance toward substances which have selective lethal or inhibitory action upon them makes the use of some of the antibacterial of little or no use in therapeutics. The problem has prompted continual search for a new source of antimicrobial agents. The antibiotics, at the present time, are produced either synthetically or through microbial fermentation. Plants, however may provide an additional source for antimicrobial substances [1]. Since the Iraqi flora is rich plants which have not been studies yet[2], the possibility of finding new antimicrobial agent (s) from plants is existed[3]. The project of our present investigation is to screen the *Callistemon viminalis* (Sol. ex. Gaertn) G. Don. For possible antibacterial effect upon *P. aeruginosa* and *S. aureus*, isolated from the patients complaining from different pathological conditions in Al-Sadder Teaching Hospital, Najaf – Iraq, in 1994. *Callistemon viminalis* is a tree, attaining a height of 2-4cm., leaves lanceolate, 4-8cm long. Spikes 5-6 cm long brightred. Fruit ovoid, contracted at the summit [4]. The leaves and twigs contains 0.06 – 0.22 % of essential oil rich in cineole. The oils contains traces of dipenten, sesquiterpentene, limonene α – terpinol and phenols. The

[+]**Received on 6/1/1997 , Accepted on 4/6/1997 .**
[*]**Lech . Technical Institute/ Kufa**
[**] **Ass . Lech . Technical Institute/ Kufa**

berries, on examination gives 51.49% moisture, 5.39% fat, 2.97% nitrogenous matter, 14.63 crude fibre and and 5.58% ash. The oil contains steoric, Oleic, palmitic, and linoleic acid [5].

Materials & Methodes:

P. aeruginosa and *S.aureus* were isolated from culture commonly associated with different pathological conditions in Al-Sadder Teaching Hospital, Najaf-Iraq. The cultures were maintained in nutrient agar slops following overnight incubation at 30 ℃ and were subcultred at two weeks intervals.

Culture media:

Nutrient broth and nutrient agar (Difco) prepared by solidification of nutrient broth with 2 % bacteriological agar were used. The ovoid fruits of the plant were collected in june, 1994 from the gardens of Technical Institute/ Kufa. The plant was identified in College Science Herbarium, Baghdad University by prof. Ali. AL-Mosawi.

Plant extraction

The dried fruit of plant were ground and extracted 3 times at room temperature with 80% ethanol. The three portions were then combined and evaporated under vacuum at 40 ℃ to dryness. The dried extracts were then tested for their antibacterial activity [6]. Preliminary phytochemical studies revealed the presence of several constituents with possible medical, biological activities like tannins, glycosides, saponine and resins [7]. The PH value of the aqueous extract was acidic (PH :4.4) and measured by the PH. Meter 7020 PYE U.K.

Antibacterial assay

Erlenmeyer flasks containing 250 ml of sterilized nutrient broth were inoculated with desired test organisms and incubation, a required volume of the culture was added to 200 ml nutrient agar portions to give a final concentration of 10 cells/ml [8]. The agar then poured on a glass plate (35×25cm) and after solidification, holes (10 mm) were made. To facilitate the removal of the agar pelletes a cork borer connected to a vaccum pump was used. 0.2 ml aliquote of different aqueous concentration (0.5, 1, 5 and 25 mg/ ml) of the plant extract were placed in each well. The plates were incubated at 30℃ and examined after 16 hr. to detect the presence of the inhibition zones [9].

Table 1:Antibacterial activity of *Callistemon viminalis* extract

Mean of inhibition zone (mm)					
Bacterial isolation source	NO. of cases	Concentration of extract (mg/ ml)			
		0.5	1.0	5.0	25
P. aeruginosa		0.0	10.6 ± 0.52	16.2 ± 0.48	20.8 ± 0.66
Chronic external otitis	4				
Chronic otitis media	22				
Burn wound infections	28				
Urinary tract infections	16				
Eye infections	9	0.0	11.25 ± 0.39	17.32 ± 0.57	23.34 ± 0.64
S.aureus					
Aspiration pneumonia	3				
Septic arthritis	5				
Wound infections	15				

Streptomycin sulphate (0.5 mg/ ml) was used as standard growth inhibiter, while water was used as the experimental control. All determination were made in triplicate [8].

Results and discussion:

The present study reports the screening of the antibacterial activity of ethanolic extract representing *Callistemon viminalis*, of Iraqi higher plant against *P. aeroginosa* and *S. aureus*. From different methods available for testing antimicrobial effect, agar diffusion was selected. The main advantage for applying this method is that it doesn't require sterilizing the plant extract before test. The plant extract examined (tab. 1) shows its antibacterial effect as indicated by the formation of inhibition zone. Their activities varied according to the test organism. The extract was active against *S.aureus* and *P.aeroginosa*, one of them is gram positive and the other gram negative bacteria. The two species isolated from different pathological conditions, representing 102 samples (tab. 1), for the purpose of further identification and isolation of active gradients (s), the extract elicited a diameter of inhibition zone more than 15 mm at a concentration of 25 mg/ ml and considered highly active and, so further investigation are required. The activity of extract was concentration dependent and the antibacterial action may due to the prescence of tannin where chemically complex substances, they usually occure as mixture of phenols that are difficult to separate because they don't crystallized [10]. Phenols lead to irreveresable general coagulation of cytoplasmic

constituents e.g protein precipitation. Both positive and negative bacteria are sensitive to phenols [11].

References:

1-Aizeman, B. E. "Higher plants as a source for preparation of new antibiotics" *Microbiologichnyi Zurnal*.; **40**: 233 – 241, 1978.

2-Al-Rawi, A. & Chakravarty, H. L. *Medical plants of Iraq Techical pulletin No. 15, Ministry of Agriculture, Government press, Baghdad*, 1964.

3-Nickel, L. G. "Antimicrobial activity of vascular plants *Economic Botany*; **13**: 281 – 318, 1959.

4-Chakravarty, H. L. *Plant wealth of Iraq*. Vol. 1 Ministry of Agr. And Agrarian Reform. Baghdad – Iraq, 1976.

5-Low, D., Rawal, B. D. & Griffin, W. J. "Antibactacterial action of the essential oils of some Australian Myrataceae with special references to activity of chromatography fraction of oil of *Eucalyptus citriodora*." *Plant medica*. **26**:184 – 189, 1974.

6-Shugar, J. C., Ronald, A. S., Lawrance, B. & Rese, S. B. "*Chemical technicians ready reference handbook.*" 2nd Ed. Mc. Graw, Hill Book; company, Newyork, 1981.

7-Al-Zuhayri, A. M. *Some phytochemical and pharmacological studies on Myrtus communis*. MSC thesis University of Baghdad, 1982.

8-May, T. N. & Fadhil, M. S. "Evalution of antibacterial activity in some of the Iraqi plants." *J. Biol. Sci. Res*. Vol. **19** (3): 169 – 178, 1985.

9-Rawlins, E. A. *Bentlys Text Book of pharmaceutic*. 8th Ed. The English Language Book Society and Bailluere Tindall, pp. 750: 1977.

10-Varre, E. T., Lynn, R. B. & James, E. R. *pharmacognesy*. 9th Ed. Lea and febiger Philadelphia. pp. 77, 1988.

11-Bertram, G. Katzung. *Basic and clinical pharmacology* a lange medical book 4th Ed. Middle East Edition, 1989.