THE ANTIBACTERIAL ACTIVITY OF AQUEOUS EXTRACT OF CINNAMON AND CLOVE AGAINST *STAPHYLOCOCCUS AUREUS*.

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Abstract

The antibacterial activity of aqueous extracts of cinnamon and clove against *Staphylococcus aureus* was investigated. Agar diffusion technique was applied in the trial. Both cinnamon and clove were found to have inhibitory effect against *Staphylococcus aureus* and the mean of the diameter of inhibition zones ranges from (7-17) mm for cinnamon and (7.5-19) mm for clove and statistical analysis using t-test demonstrated that there were significant differences between cinnamon and clove at each concentration used in this study in order that clove was found to have greater effects against *Staphylococcus aureus.* The minimum bactericidal concentration (MBC) was also determined and it was shown that 30% was the MBC for cinnamon and 15% was the MBC for clove.

Key words: - antibacterial activity, cinnamon, clove, *Staphylococcus aureus*.

Introduction

Staphylococcus aureus is a very common bacterium that lives on our skin and generally causes no trouble. But if it gets into the blood stream, through a cut or surgery. *Staphylococcus aureus* can very quickly damage the heart, lungs, brain or poison the entire system [(1)] and it is an important cause of food poisoning [(2)].

Microbial resistance to antibiotics especially among Staphylococcal strains is a major threat to public health [(3)].

Therefore plant extract are suitable alternatives now to treat resistant organisms. Down the ages plants extracts have evoked interest as sources of natural products.

They have been screened for their potential uses as alternative remedies for the treatment of many infections diseases [(4)]. Plants extracts have been shown to posse's antibacterial, antifungal, antiviral, insecticidal and antioxidant properties [(5), (6)]. Some plants extracts are used in food preservation [(7)] as they are a rich source of biologically active compounds [(8)]. Plants extracts such as cinnamon and clove have been traditionally used by people for various purposes in different parts of the world.

Cinnamon and clove had shown antibacterial and antifungal activity cinnamon also posse's antidiabetic property [(9)].

The present study was undertaken in order to evaluate the in-vitro antibacterial activity of cinnamon and clove aqueous extract against *Staphylococcus aureus*.

Materials and methods

Evaluation of antibacterial effects of two aqueous extracts (cinnamon, clove) against *Staphylococcus aureus* was done in this study.

The two aqueous extracts were prepared according to Cowans [(10)].

Five diagnosed isolates of *Staphylococcus aureus* obtained from Baghdad teaching laboratories were used in the present study.

Staphylococcus aureus cultures of 18 hours at 37 ^{o}C in Mueller Hinton broth were used. The cultures were adjusted to approximately 10^5 CFU/ml with sterile saline solution.

Agar diffusion technique was applied to study the antibacterial effect of the previously mentioned extracts.

Final concentration of 2.5%, 5%, 10%, 15%, 20%, 30%, 40%, 50%, 60% and 70% of the aqueous extract of cinnamon and clove in Mueller agar were obtained for each extract separately.

Mueller Hinton agar plates were swabbed with a suspension of Staphylococcus *aureus* prepared as mentioned earlier, using sterile cotton swab. Plugs were removed from each agar plate producing holes. To each hole 100 µl from different concentration of each extract was added and allowed to diffuse at room temperature for 20 minutes, the plates were then incubated aerobically overnight at

37 ^{o}C. Determination of the minimum bactericidal concentration (MBC), the lowest concentration of the antimicrobial agent causing negative growth (fewer than three colonies) was done by serial dilution of Mueller Hinton broth and lineation of Mueller Hinton agar [(11), (12)].

Results and Discussions:-

Each extract was tested against the five isolates of *Staphylococcus aureus.* The antibacterial activity of the extracts were recorded as the mean diameter of the resulting inhibition zones of growth measured in (millimeters). The antibacterial activity of cinnamon aqueous extract is summarized in Table (1).

The results revealed that cinnamon exhibited antibacterial activity against *Staphylococcus aureus*. As the mean of the diameter of inhibition zone were (7mm), (8mm), (11mm), (13mm), (15mm), (15.5mm), (17mm) for the concentrations of (15%), (20%), (30%), (40%), (50%), (60%) and (70%) respectively while the concentrations of (2.5%), (5%), (10%) did not give any inhibition zone.

The antimicrobial effect of medicinal plants is well documented [(13)] and the results of different studies provide evidence that some medicinal plants might indeed be potential sources of new antibacterial agent even against some antibiotic–resistant strains of bacteria [(14)].

In this current study using the agar diffusion technique it was observed that extract of cinnamon produce antibacterial activity against *Staphylococcus aureus* these results confirmed the observation of earlier studies of Fan *et.al*. [(15)], Yuste *et.al.* [(16)]. Seeniva San *et.al*. [(17)] also reported that cinnamon showed potent and maximum activity among some plants extracts against *Staphylococcus aureus*.

The antibacterial activity of cinnamon has been attributed to the presence of some active constituents [(18), (19)]. Cinnamaldehyde is the active compound in cinnamon and the earlier studies suggested that the antibacterial activity of cinnamon was probably due to this compound found in cinnamon and cinnamaldehyde is also a natural antioxidant[(20)].

Table(2) summarized the antibacterial activity of colve aqueous extract and the results showed that clove extract exhibited antibacterial activity against *Staphylococcus aureus* at nine of the ten concentrations used in this study and the mean of the diameter of inhibition zone were (7.5 mm), (8 mm), (10 mm), (11 mm), (13 mm), (15 mm), (16.5 mm), (17 mm), (19 mm) for the concentrations of (5%), (10%), (15%), (20%), (30%), (40%), (50%), (60%), (70%) of clove respectively.

These results revealed that clove extract also exhibit antibacterial activity against *Staphylococcus aureus*.

This was in agreement with the results of Seeniva San *et.al*. [(17)] who found that clove extract inhibit the growth of *Staphylococcus aureus* this was also reported by Agaoglu *et.al*. [(21)] Hitokoto *et.al*. [(22)]. In another hand Burt *et.al*. [(23)] reported that clove has antiseptic as well as bacteriostatic and bactericidal activity against *Staphylococcus aureus.*

The effect of clove may be explained by the action of eugenol and eugenol acetate contained in its volatile oils as many investigators have reported [(24) (25) (26)].

Sensitivity of *Staphylococcus aureus* to different concentrations of cinnamon and clove extracts were tested separetly. As mentioned earlier both of them have antibacterial activity against *Staphylococcus aureus*. These results were also reported by Barbosa- Kanovas *et.al*. [(27)] who showed that cinnamon and clove had a strong inhibitory activity against microorganisms.

Another study done by Fyfe *et.al.*[(28)] demonstrated that cinnamon and clove significantly decreased the production of enterotoxin A and enterotoxin B of *staphylococcus aureus*.

The results of the present study using student t-test demonstrated that there are significant differences at $P<0.05$ between cinnamon and clove for each concentration used in this study and this was demonstrated in Table (3). From these results we noticed that clove extract had greater effects than cinnamon extract at each concentration used on *Staphylococcus aureus*.

This may be explained by the fact that extracts that contain eugenol have been show to exhibit the strongest antimicrobial activity [29] while Seeniva San *et.al*.[17] Reported that cinnamon extract exhibit greater activity than clove on *Staphylococcus aureus* but both of them exhibit antibacterial activity against *Staphylococcus aureus*. The main factors that determine antibacterial activity are type, composition of the extract used, pH and temperature of the environment [30]. Table (4) showed the minimum bactericidal concentration (MBC) of cinnamon and clove aqueous extract.

The results of this study demonstrated that the minimum bactericidal concentration (MBC) of cinnamon aqueous extract for *Staphylococcus aureus* was 30% the majority of *Staphylococcus aureus* isolates were sensitive at this concentration it seems to be toxic to these bacteria.

For clove aqueous extract the concentration of 15% was able to make all isolates or strains of *Staphylococcus aureus* sensitive at this concentration and it was considered the MBC

In conclusion cinnamon and clove were found to have important antibacterial activity against *Staphylococcus aureus*. In this regard, the use of them as natural preservatives in food products (to prevent food poisoning by *Staphylococcus aureus*) may be alternative of chemical additives they can be also incorporated into creams, lotion to treat diseased caused by *Staphylococcus aureus*.

Table (1)
Sensitivity of *Staphylococcus aureus* to different concentrations of cinnamon aqueous extract.

Conce.of cinnamon	frequency	Mean diameter of inhibition zones (mm)
70%	5	17
60%	5	15.5
50%	5	15
40%	5	13
30%	5	11
20%	5	8
15%	5	7
10%	5	0
5%	5	0
2.50%	5	0

Table (2)
Sensitivity of *Staphylococcus aureus* to different concentrations of clove aqueous extract.

Conce. of clove	frequency	Mean diameter of inhibition zones (mm)
70%	5	19
60%	5	17
50%	5	16.5
40%	5	15
30%	5	13
20%	5	11
15%	5	10
10%	5	8
5%	5	7.5
2.50%	5	0

Table (3)
Comparison between cinnamon and clove aqueous extract for each test concentration in relation to the sensitivity of *Staphylococcus aureus*.

Percent concentration	extracts	Mean (mm)	Sig.
5%	cinnamon	0	S
	clove	7.5	
10%	cinnamon	0	S
	clove	8	
15%	cinnamon	7	S
	clove	10	
20%	cinnamon	8	S
	clove	11	
30%	cinnamon	11	S
	clove	13	
40%	cinnamon	13	S
	clove	15	
50%	cinnamon	15	S
	clove	16.5	
60%	cinnamon	15.5	S
	clove	17	
70%	cinnamon	17	S
	clove	19	

S: Significant difference at level P<0.05.
Mean: mean of diameter of inhibition zones.

Table (4)
Minimum bactericidal concentrations (MBC) of cinnamon and clove aqueous extract for *Staphylococcus aureus.*

type of extract	frequency	No. of isolates within the MBC of extract % concentrations							
		5.00%	10%	15%	20%	25%	30%	35%	40%
cinnamon extract	5	0	0	0	0	1	4	0	0
clove extract	5	0	0	5	0	0	0	0	0

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