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EFFECT OF PLANT EXTRACTED *SALVADORA PERSICA* L.ON SOME ISOLATED PATHOGENS FROM MOUTH AND TEETH

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ABSTRACT

This study aimed on using stem of plant Salvadora persica L.which it commonly used in cleaning teeth and has highly efficiency as Antioxident Anti-inflammatoryand Antimutageneic against many pathogens. Therefore the current study included isolation and identification some pathogens from mouth and teeth and included the Staphyllococcus aureus, *Streptococcus sanguis*, *Streptococcus salivarius* and *Candida albicans* and test the effect of each of the aqueous and ethyl acetate extract of *Salvadora persica* L.in different concentrations (1000, 750, 500, 250, 125, 62.6) µg/ml on these pathogen, in addition to detect of The minimum inhibitory concentration (MIC) and the minimum bacteriosidal concentration (MBC) . The results showed that aqueous extract had antimicrobial against these isolates higher than ethyl acetate extract in all concentrations specially at 1000 µg /ml with diameter means of inhibition zones (26.5, 18.2, 11.4, 13.7) mmeter for *Staphylococcus aureus, Streptococcus sanguis, Streptococcus salivarius* and *Candida albicans* respectively compared with ethyl acetate extract (17.4, 12.1, 9.1, 8.3) mm respectively with significant differences at $p< 0.05$. The isolates of *Staphylococcus aureus* were more sensitive among tested isolates.

KEYWORDS: antimicrobial, *Streptococcus salivarius* and *Candida albicans.*

INTRODUCTION

Salvadora persica L. (Arak) tree and locally called Miswak belongs to Salvadoracea family which grows in different area of the world including the Middle east and Africa, it is one of the most commonly used medicinal plants for oral hygiene among global Muslim community.[1] It contains important phyto-constituents such as Vitamin C, Salvadorine, Salvadorurea, alkaloids, Trimethylamine, Cyanogenic glucosides, Tannins, Saponions and

salts mostly as chlorides.[2] The history and the use of Miswak (tooth sticks) as an oral tooth as well as the biological effects of *Salvadora persica* extracts are reviewed.

Variety of studies have been performed on the antimicrobial effect of these sticks of *Salvadora persica* it has been shown various biological properties including significant antibacterial.[3] Antifungal[4] through suppress or destroy microbial growth, thus susceptibility of the microorganisms , penetration of antimicrobial agent to the infected site.[5] Others[7] found that alcoholic and water extracts of *Salvadora persica* inhibited the growth of *Streptococcus pyogens*, *Staphylococcus aureus*, *E. coli* and *Pseudomons aeruginosa*. Allafi and ababaneh[6] found that benzyl thiocyanate present in *Salvadora persica* inhibited the growth of *Streptococcus mulans* and suggested that *Salvadora persica* decreses the incidence of dental caries. While others[7,8] found the active component of *Salvadora persica* has a great antimicrobial activity against various Gram positive and Gram negative bacteria.

The aim of the present study was to evaluate the antimicrobial activity of aqueous and ethyl acetate extracts of *Salvadora persica* (Miswak) after isolatation and identification of the microbes from oral cavity of patients.

MATERIAL AND METHODS

Plant material

Dried stems of *Salvadora persica* were purchased from a local market in Baghdad city, and identified by specialized plant classification / Baghdad university according to their color and scent.

Extract Preparation

Aqueous Extraction

Forty grams of the powdered stems were transferred to sterile screw – capped bottle and 200 ml of sterile distilled water was added to the powder samples which were allowed to soak for 24 hours at 4ºC The mixtures were then centrifuged at 2000 rpm for 10 minutes, the supernatants were filtered through Millipore filters diameter (0.22) µm and freeze drie. , the final dried material was stored in labeled sterile bottles and kept in a freezer at -20 ºC.[9]

Ethyl Acetate Extraction

Two- hundred grams of the powder was exhaustively extracted with alcohol (ethanol 70%) and concentrated under reduced pressure using rotary evaporator at 60 ºC to give a

concentrated extract 50 ml. The crude aqueous ethanolic extract was fractionated using gradient solvents petroleum ether, chloroform and ethyl acetate then the ethyl acetate extract was frozen and freeze dried until use.[10]

Microorganism

Twenty samples was collected from oral cavity of patients suffering from denture stomatitis by using swabbing method and all isolates were identified according to Bergeys Manual.[11]

Antimicrobial Activity Assay of Salvadora Persica Extracts

Agar well method according to[12] was used to detect antimicrobial activities of aqueous and ethyl acetate extracts against some species of bacteria and yeasts. The concentrations of extracts prepared at (100 , 750 , 500 , 250 , 125 , 62.5) µg /ml .The minimum inhibitory concentrations (MSC) and Minimum bactericidal concentration (MBC) assay were performed in concentrations (1.24, 512 , 256, 128 , 46 , 32, 16 , 8 , 4 , 2 , 1) µg / ml as described by.[12]

RESULTS AND DISCUSSION

Bacterial isolates were obtained and identified as *Staphylococcus aureus*, *Streptococcus sunguis* , *Streptococcus salivarius* and yeast of *Candida albicans*, the reason for the spread of bacteria to posse a high mechanism resistance and ease of transmission of resistance determinants by conjugation and transformation,[13] with regard to the presence of *Candida albicans* that belongs to possessing opportunistic ability that relating to the patients' immune in order to growth and reproduction.[14]

Antibacterial activity assay of *Salvadora persica* aqueous and ethylacetate extracts was done on 3 isolates from all of (Staphylococcus aureus, *Streptococcus sunguis* *Stapylococcus salivarius* and *Candida albicans*). The results obtained demonstrated that Salvadora persica aqueous extract was the most effective against gram positive bacteria and *Candida albicans* than the ethyl acetate extract, the differences were significant ($p < o.o5$) in all concentrations Both of aqueous and ethyl acetate extractions.

At concentrations (1000, 750, 500) mg / ml showed closely related results and had effects on all isolates, as well as the most effective one was (1000) mg/ml as shown in (Table- 1), (Table -2). These results were in agreement with many studies that revealed the antimicrobial agent of *Salvadora persica* extract against many bacterial and fungal genera,[2] Other workers[15] found that alcoholic and water extracts of *Salvadora persica* inhibited the growth

of *streptococcus pyogens, staphylococcus aureus, E. coli and Pseudomonus aeruginosa* while other workers[16 ,17] showed various effectiveness of aqueous and alcoholic extracts of *Salvadora persica* against many bacterial genera and *Candida albicans* isolated from oral cavity of patients in Iraq.

Table-1 – Antimicrobial activity of aqueous extracts of *Salvadora persica.*

Microorganisms	**Mean ±sd of inhibition zone (mm) Concentrations µg/ml**						
Staph.aureus	0.0	1000	750	500	250	125	62.5
	0.0	26.2± 0.6*	25.2± 0.08*	20.7± 0.02*	14.2± 1.2*	8.3± 0.13*	6.4± 0.7*
Strep. sanguis	0.0	18.2± 1.3*	16.5± 1.9*	14.1± 0.14*	6.3± 1.9*	5.7± 0.02*	2.5± 1.4*
Strep. salivaris	0.0	11.4± 1.5*	10.5± 0.07*	8.11± 0.13*	5.5± 1.6*	4.6± 1.8*	4.1± 1.1*
C. albicans	0.0	13.7± 0.9*	12.1± 1.4*	10.8± 0.5*	7.4± 0.02*	5.9± 1.1*	3.8± 1.3*

* Significant difference at ($p < 0.05$).

Table-2 – Antimicrobial activity of ethyl acetate extracts of *Salvadora persica.*

Microorganisms	**Mean ±sd of inhibition zone (mm) Concentrations µg/ml**						
Staph.aureus	0.0	1000	750	500	250	125	62.5
	0.0	17.4± 1.2*	15.4± 0.1*	12.9± 0.02*	9.4± 0.05*	6.8± 1.9*	4.9± 0.15*
Strep. sanguis	0.0	12.1± 2.7*	11.3± 0.128*	9.8± 0.08*	7.1± 1.3*	4.5± 0.9*	2.1± 1.1*
Strep. salivaris	0.0	9.1± 2.3*	8.0± 1.02*	6.5± 0.4*	5.2± 0.05*	3.1± 1.03*	1.9± 0.9*
C. albicans	0.0	8.3± 1.8*	7.1± 0.08*	6.9± 1.5*	4.9± 0.06*	3.9± 0.6*	2.8± 0.03*

* Significant difference at ($p < 0.05$)

Table -3 showed MIC and MBC values of *Salvadora persica* extracts and the most affected isolates was *Stapylococcus aureus* no.1 which its MIC was 4 and MBC was 8 for aqueous extract but its MSC for ethyl acetate was 32 and MBC was 64 .That possess significant antimicrobial activity against all isolates in contrast with control ($p < 0.05$), that belong to its chemical components such as Vitamin C, Sulfure, Alkloids, falvenoids, Tannins, Saponins, sodium bicarbonate, silica, cloried and thiocyand mostly as chlorides and play an important role as antimicrobial agent.[18,19]

Table- 3 – MIC and MBC values of aqueous and ethyl acetate extracts against isolated microorganisms.

Microorganisms	(MSC) and (MBC) values mg/ml			
	Aqueous extract		Ethyl acetate extract	
	MSC MBC		MSC MBC	
Staph.aureus no.1	4	8	32	64
Staph.aureus no.2	16	32	128	256
Staph.aureus no.3	128	256	256	512
Strep. Sanguis no.1	64	128	256	512
Strep. Sanguis no.1	8	16	64	128
Strep. Sanguis no.1	256	512	1024	-
Strep. Saliarius no.1	128	256	512	1024
Strep. Saliarius no.1	32	64	64	128
Strep. Saliarius no.1	8	64	256	512
C. albicans no .1	16	32	128	256
C. albicans no .2	256	512	1024	-
C. albicans no .3	64	128	64	128

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