

A Comparative Between Influence of Antibiotics and Extracts from *Myrtus Communis* and *Allium Sativum* Against *S. Aureus* Isolated from Some Pathogenic States

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Abstract Aim of the study included isolating *staphylococcus aureus* bacteria for several different medical conditions and revealing the most important virulence factors it possesses. Determination of the presence of staphylococcus aureus bacteria that are resistant to antibiotics and isolated from this conditions. comparative between the effect of antibiotics used in this study on *S.aureus* with the effect of alcoholic and aqueous extracts of the *Myrtus communis* and *Allium sativum*, 125 samples were collected on different clinical cases (wounds, acne, tonsils, ear, eye) The results showed that the highest rate of isolation of *Staphylococcus aureus* bacteria is from samples taken from wounds, Until it reached its lowest sensitivity to the anti-G Penicillin direction, as it reached 5%, the alcoholic extract of garlic was the first in its inhibitory effect on *S.aureus* isolates for all concentrations used under study, so it showed high moral inhibition and overcome the standard drug (Control) Vancomycin Alcoholic extract of the ace plant followed, as the concentration appeared between (100,75) mg/ml, The aqueous extract of the base plant came last. The standard drug Vancomycin showed a significant moral superiority over the extract and for all the concentrations used under study.

Introduction

Although *Staphylococcus aureus* is part of the normal flora of the skin, nose, pharynx and gastrointestinal tract and reproductive tract of the human being, they are important nurses for the human being as they cause numerous injuries ranging from difficult to possible treatment due to their ability to penetrate the body's defenses and invade tissues by possessing various ferocity factors as well in multiplicity Opportunistic Pathogens It causes minor skin infections to life-threatening systemic diseases, as it has a high ability to resist antibiotics [1-2], as the most serious infections are caused by multiple strains resistant to antibiotics oxacillin resistant staphylococcus aureus. Which became a serious health problem [3]. These bacteria infect different areas of the body such as the eye, throat, pharynx, urethra, vagina, and stomach, but they become more dangerous when they enter the bloodstream, causing diseases such as pneumonia, urinary tract infection, tonsillitis and nasal sinuses [4-5] *Staphylococcus aureus* (*Staphylococcus aureus*) has the ability to produce hemolysin called staphylolysines, as well as staphylokinase, to help spread bacteria, cerebellum formation, leucocidin, and hyaluronidase, as well as its ability to cause endocarditis [6-8]. Medium diseases and toxins such as Toxic shock syndrome and food poisoning [9] are also the most important causes of skin infection such as boils, Pimples, Abscesses, Impetigo, and wound infection. Scalded Skin Syndrome [10] has a role in causing impetigo injuries that multiply into acute inflammation, such as Acne [9-11]. This is why bacteria have an important role in Osteomyelitis [12], so efforts are now being replaced to use a therapeutic side. Another safer antibiotic is a good plant extract that has the inhibiting effect of different types of microorganisms

2 Materials and working methods:

130 samples of samples (wounds, acne, tonsils, ear, eye) were collected from patients attending the General Teaching Hospital in Al-Kirkuk city at the age of (10-60) years for the period (1/12/2018 - 1/4/2019). Cultivation on the agricultural media on the solid salt mannitol medium and the blood agar medium and the solid nutrient medium and then diagnose the staphylococci bacteria by electing the colonies growing on the mannitol medium and fermenting it, and then conducting diagnostic tests for all isolates according to what is mentioned in [13-15].

The ability of bacteria to produce coagulase was determined in two ways:

1. Piping: To extend the free enzyme, as 1 ml of plasma was taken and placed in a sterile tube, and the vaccine and bacterium were added to it by the carrier and incubated at a temperature of 37 C for a period of four hours. If the clotting is an indication that the bacteria produce the clotting enzyme if the plasma remains as it demonstrates the inability of bacteria to produce a plasma coagulation enzyme.

2. Slide: It is conducted to determine the binding enzyme and is done by taking a drop of plasma and putting it on sterile slides, then pollinated by the transferor with the bacterial vaccine and mixed well with it. After ten minutes, a clotting presence and a lack of it is observed as the plasma clotting is a positive result

As for determining the production of the hemolysin enzyme, it was done in the following way:- The dishes containing the blood acids medium were stalked with bacteria of staphylococcus and incubated with a temperature of 37 mm for 24 hours that the emergence of a transparent decomposition area around the colonies is a positive result and vice versa. 14) Hence, a staphylococcus aureus isolates sensitivity test is performed

For antibiotics according to [16]. As 4-5 pure colonies were transferred to test tubes containing 5 ml each of the broth medium and incubated tubes at a temperature of 37 mm 24 hours after which the growth in the tubes was compared with the McFarland tube and equal to approximately 1.5×10^8 cells/ml and using Sterile cotton swab spreading bacteria to test their sensitivity on the Muller Hinton steel medium evenly and ready-made antibiotic tablets were placed on the surface of sterile dishes by sterile forceps and gently pressed and then brooding at a temperature of 37 C⁵ for 24 hours then determine the resistant and sensitive bacteria of life antibodies by measuring the diameter of the inhibition region from the second side of the plate using a transparent ruler, after which it was compared with the records fixed in the standard table [17]. National Committee of Clinical Laboratory standers.

Yace leaf was obtained from the college garden and garlic plant from the local market. Then the Yace leaves and garlic cloves were washed with plain water and then distilled water then left to dry at room temperature then crushed dry leaves of Yace with an electric mill and kept the powder with a dry bottle until use, then the aqueous extract of a plant is prepared yace and garlic, as stated in [18], which include mixing a quantity of

Dry powder of yace plant with a quantity of garlic crushed with an electric mill with a quantity of distilled water in the proportion of 1 g: 2 ml distilled water using an electric mixer Blender at room temperature Leave the solution for 24 hours and then filter the resulting mixture by several layers of sterile medical gauze then distribute the suspended substance from the filtration In test tubes in the centrifuge at 3500 rpm

The filtrate was taken for the purpose of drying with an electric oven with a temperature of (35 ° C) equipped with an air fan to prepare a hot air stream in drying on the one hand and to reduce the effect of heat on the components of the extract on the other hand in order to prevent any changes that might lead to a decrease in the biological effectiveness of the extracts after the weight of the dry powder and keep clean glass bottles tightly closed and placed in the refrigerator until use. As for the method of preparing alcoholic extracts for pomegranate peel plant, it was done according to [19].

As 15 g of garlic or yace powder were taken and 200 ml of ethyl alcohol was added to it at a concentration of 80% and put in Thumblc in a continuous extract device and the extract was left for 7 hours at a temperature of 60 C⁵ then filtered with a filter type 1. Watt man No The filtrate was then exposed to evaporation using the Rotary vacuum evaporator until a thick liquid was obtained, after which the liquid was evaporated using the 45 ° C

electric oven to obtain the dry powder. Stock Solution prepared by taking 2gm aqueous extract was dissolved and dissolved in (10) ml of sterile distilled water, so its concentration became 200 mg/ml, and ascending concentrations were prepared (25, 50, 75, 100) mg/ml. As for the control treatment, it was represented by distilled water only.

As for alcoholic extracts, the standard dilutions were prepared using ethylene glycol at a concentration of 100% as a diluent and a good solvent and ineffective against microorganisms if they were used as a control treatment as well and the diffusion method in agar was used by drilling [12].

This method included making five holes with a diameter of 6 mm by a cork purer in equal dimensions, then spreading 0.1 ml of the bacterial trap by the publisher (Spreader) homogeneously over the Muller Hinton medium (after comparison with the standard McFarland tube), then the prepared concentrations were added to each extract (25, 50, 100, 200) mg/ml by 0.1 ml per hole with control remained. Then the dishes were incubated at 37 ° C for 24 hours after which it was possible to know the efficacy of the extract by measuring the inhibition zone by ruler around each hole.

1 Results and Discussion

The highest incidence of *Staphylococcus aureus* isolation was recorded in samples taken from wounds 76%, followed by acne 56%, then ear 36%, tonsils 28%, and finally the eye 20%. It was found from the results of the study that most of the bacteria producing the coagulation enzyme are producing hemolysin which is the most important Firm factor of this bacterium are described. It is observed that there is a close correlation between the susceptibility of bacteria to hemolysin production and plasma coagulation.

Table. 1. Types of *Staphylococcus aureus* isolated from clinical samples understudy

Isolation of S.aureus	Total of isolation		NO. isolation product hemolysin		NO. isolation, not product hemolysin	
	NO	%	NO	%	NO	%
	32	59.2	29	91	3	9
	22	41	6	27	16	73

Table. 2. Percentage of *S.aureus* isolates produced and coagulase.

Samples Clinical	Number Samples	<i>S .aureus</i>		<i>Staphylococcus</i> spp.		No growth	
		NO	%	NO	%	NO	%
Wounds	25	19	76	4	16	2	8
Acne	25	14	56	8	32	3	12
Tonsils	25	7	28	14	56	4	16
The ear	25	9	36	10	40	6	24
Eye	25	5	20	15	60	5	20
Total	125	54		51		20	

The sensitivity of *S. aureus* isolates has been studied in the direction of several selected life antibiotics, as Table (3) shows that there is a clear variation in the resistance of *Staphylococcus aureus* isolates to these antagonists. It showed high resistance to Penicillin G, as it reached 100%. As the use of these antihistamines became limited due to their possession of a B-lactam ring in addition to the ability of these bacteria to resist these antihistamines to produce beta-lactamase enzymes encoded by plasmids or chromosomes [22] and there was high resistance to the anti-Amoxicillin and Ciprofloxacin by 96% for each of them then graded Resistance became close to Chloramphenicol, Nitrofuration, and Tetracycline, Rivampicin (56, 48, 44,41%), respectively, then resistance to Neomycin decreased to 37%, reaching Vancomycin as it reached 0%. Resistance to Tetracycline was encoded in *Staphylococcus aureus* by [15] whereas, the resistance of Rifampicin antigen

then changed the target site of this antagonist, which is the DNA-Depended RNA-Polymerase with chromosomal mutations [23,24]. The Vancomycin antagonist is considered to be a killer antagonist, inhibiting the synthesis of the cellular neighbor by preventing the binding of the molecules of the peptidoglycan polymers [25].

Table 3. percentages of isolates resistant to antibiotics for *Staphylococcus aureus*.

Antibiotic	The concentration of antibiotic in disc	Resistance isolation		Sensitive isolation	
		NO	%	NO	%
Neomycin	10 gm	20	37	34	63
Penicilling	10 gm	54	100	0	/
Amoxicillin	10 gm	52	96	2	4
Tetracyclin	30 gm	24	44	30	56
Chloramphenicol	30 gm	22	41	32	59
Vancomycin	5 gm	0	/	54	100
Cloxacillin	5 gm	47	87	7	31
Ciprofloxacin	30 gm	52	96	2	4
Nitrofurantion	300 gm	30	56	4	44
Rifampicin	5 gm	26	48	28	52

Table (4) shows that the alcoholic extract of the garlic plant came first in its effect on *staphylococcus aureus*. The standard drug Vancomycin did not show any significant superiority over the extract and all the concentrations used under study, but the extract showed all its concentrations the effect of high moral inhibition and overpowered the drug (Control) came second in the alcoholic extract of the acetate plant, As the standard drug Vancomycin showed significant superiority over the extract with a concentration of (22.50) mg/ml, as the damping diameter of the standard drug was 16.9 mm, while the two concentrations (75, 100) mg/ml showed a superior inhibitory effect. The aqueous extract of garlic ranked third, as the standard drug outperformed the extract by concentrations (25, 50, 75) mg/ml, while the concentration showed 100 mg/ml, a superior inhibitory effect. The water extract of the exponent plant came in the last place, as the standard showed significant superiority over the extract and all its concentrations. We conclude from all the above that the alcoholic extract has overcome the aqueous extract of the medicinal plants used under study to inhibit the isolates of *staphylococcus aureus*.

Perhaps this is due to several reasons, including the method used in the extraction, as well as the type of bacteria located under the extract stream and the nature of the substances present in the aqueous extract are somewhat different from that in the pseudo-extract liberating an enzyme as we note that the bacterial inhibition process is proportional to the increase in the concentration of the extract as it was mentioned [18] that the more concentrations of the inhibitory substance in the extract, the greater its concentrations, the greater its effective effect on inhibition of the bacterial species. We also deduced from this study the increased inhibitory efficacy of garlic on the wall to inhibit *staphylococcus aureus* isolates as garlic contains volatile oils consisting of volatile compounds consisting of sulfur compounds such as Diallyl trisulfide, Diallyl disulfide, Allicin, as Allicin oil is responsible for the therapeutic properties of garlic, as the Allinase enzyme is released and the Allin compound turns into the active allicin compound that has the Allicin compound found in garlic. It has a wide inhibiting activity against viruses, bacteria, and fungi, especially against *staphylococcus aureus* causing secondary infections such as deadly pneumonia especially in young children.

Table 4. Inhibitory effect of aqueous and alcoholic extracts of medicinal plants under study on *Staphylococcus aureus* isolates

Plant used	Commend name	Family name	Antibiotic	Type of extract	Size of Inhibition zone For 5 concentration extract			
					25	50	75	100

Allium sativum	Garlic (lopes)	Liliaceae	Inhibition zone Vancomycin 19.6mm cd	aqueous	17 a	17.4 b	19.1 c	22 d
			Inhibition zone Vancomycin 19.6mm a	alcoholic	21 ab	25 c	28.3 d	33.4 e
Myrtus communis	Myrtle leaves	Myrtaceae	Inhibition zone Vancomycin 19.6mm	Aqueous	19.3d e	18.3 c	16.3 b	15.6 a
			Inhibition zone Vancomycin 19.6mm bC	alcoholic	22.7 e	20.cd	19.3 b	15.6 a
				Athelinycol	0	0	0	0

And pomegranate peel contains other important compounds Allin (S-allyl-L-Cysteine sulfoxide (S-methyl-L-Cysteine, sulfoxide) and proteins, vitamins, and enzymes such as (Allinase, Peroxidase, Myrosinase) [26] Also, the sulfur bonds of the compound Allicin increase the effectiveness Antibiotics for this plant [27]. Also, the extract of the leaves and fruits of ace has a deadly and inhibitory effect of Staphylococcus aureus isolates, which contains volatile essential oils, resins, sugars and glycerides, and the oils contain many substances including Alpha Pinene, Camphene, Nerol, Geraniol, cineole, Myrtol, Dipinene [23]. Also, the good inhibitory activity of acetylcholine, which is positive for gram stain, even at low concentrations, is due to the presence of flavonoid glycosides which, when hydrolyzed, give the compounds Ellagic acid, Gallic acid, Myricetin is effective against bacteria that are positive for the dye of a particular gram Staphylococcus aureus [19]. It was also mentioned that two new compounds of Yace leaves, A-Myrtucommulone, and B-Myrtucommulone, were observed, as the effectiveness of the Myrtucommulone-A compound was observed against gram-positive bacteria [27].

Different letters within a single row mean that there is a significant difference at the level of (P <0.05).

Similar letters within a single row mean that there is no significant difference at the level of P <0.05))

LSD values for aqueous garlic extract 0.9

LSD values for Alcoholic Garlic Extract 1.67

LSD values for the aqueous extract were 1.055

LSD values for the alcoholic extract were 1.16

Conclusion

This paper studied the isolating staphylococcus aureus bacteria for several different medical conditions and revealing the most important virulence factors it possesses. Determination of the presence of staphylococcus aureus bacteria that are resistant to antibiotics and isolated from this conditions.comparative between the effect of antibiotics used in this study on S.aureus with the effect of alcoholic and aqueous extracts of the Myrtus communis and Allium sativum.

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