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Antibacterial Effect of Some Asteraceae of Southern Algeria on Nosocomial Strains of the Genus *Staphylococcus*

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Abstract

The study presents an interest in the antibacterial effect of some Asteraceae of southern Algeria, on nosocomial strains of the genus *Staphylococcus* precisely strains of two species *Staphylococcus aureus* and *Staphylococcus epidermidis*. Both species of *Staphylococcus* were omnipresent in nosocomial environment investigated according a to previous study. Plants tested were all shown an antimicrobial effect against the tested strains with maximum inhibition zone of 21 mm for *Staphylococcus epidermidis* for *Cotula cinerea*, *Staphylococcus aureus* shown maximum inhibition zones of 22 mm also for *Cotula cinerea* and *bubonium graveolens* and those for the reference strain, *Staphylococcus aureus* ATCC 25923, were 32 mm for *Bubonium graveolens*. Plants tested shown minimal inhibitory concentration ranging from 1×10^{-3} g to 5×10^{-4} g. *Staphylococcus* strains tested were totally resistant to β lactams with rate of 51.84% whereas their rate of resistance for cotula cinerea was only 07.21%.

Keywords: Asteraceae; *Staphylococcus aureus*; *Staphylococcus epidermidis*; Nosocomial bacteria; Inhibition zone; Résistance; Antibiotics

Introduction

WHO estimates that an average of 190 million people are hospitalized each year worldwide and 9 million of them contract an infection at that time [1-6]. In Algeria, according to a national prevalence survey on nosocomial infections, led by the ministry of Health, population and hospital reform; at the hospital mustapha bacha of Algier, 8% of hospitalized patients contract a nosocomial infection [7]. Bacteria are the most common pathogens responsible for nosocomial infections [7] and major pathogens implicated are Gram-negative bacilli: 60% Cocci Gram positive 30% (*Staphylococcus aureus*) 15% [8-10]. According to a study done in 2004 on Algerian monitoring the resistance of nosocomial germs; of a total of 14400 bacterial strains isolated from hospitalized patients, 3246 are multi-resistant bacteria (22.5%), MRSA are responsible for 38.6% of nosocomial infections contracted [11]. In the present study, we are in search for medicinal plants with active antimicrobial effect on nosocomial bacteria represented by strains of the genus *Staphylococcus*. Plants studied were chosen from those used by the local population against infectious Diseases, supposed to have an antibacterial effect. The choice will be made after ethnobotanical survey conducted among the population of south-western Algeria on the basis of a questionnaire drawn up for the research of plants with an antibacterial effect (Table 1 and Figure 1).

Rate of resistance to families of antibiotics tested

Families of antibiotics	*S. aureus* ATCC 25923	Rate of Resistance of strains of *Staphylococcus aureus*		Rate of resistance of strains of *Staphylococcus épidermidis*		Rate of resistance of all strains tested	
	[*]Diameters of internal control (mm)	[*]resistance with Critical Diameters	[**]total Résistance	[*]resistance with Critical Diameters	[**]total Résistance	[*]resistance with Critical Diameters	[**]total Résistance
β lactams	24- 36	88.88	88.88	66.66	44.43	77.77	51.84
Aminosides	21-29	25	25	33.33	00	28.57	04.07
Macrolides-Lincosamides-Streptogramin (MLS)	24-29	100	50	00	00	50	12.5
Tetracyclines	25-28	100	100	100	100	100	100

Sulfamides	27-30	68.75	68.75	66.66	66.66	67.70	67.70
Polyptides	17-20	46.33	27.96	30.33	3.26	38.33	13.60
Phenicol	19-26	00	00	00	00	00	00
Quinolones	17-21	00	00	00	00	00	00
Fosfomycine	27-31	00	00	00	00	00	00
% Résistance Globale	00	47.66	40.06	32.99	23.81	40.26	27.74

*calculation of resistance rate considering critical diameters specified for *Staphylococcus aureus* [11].

**calculation of resistance rate for zone inhibition equal to zero (oo mm)

Table 1: Resistance to families of antibiotics tested.

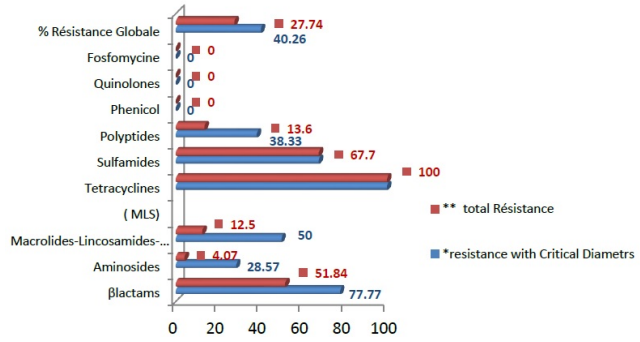

Figure 1: Resistance to families of antibiotics tested.

Materials and Methods

- Strains tested were collected from surfaces and air of five hospitals located in south west of Algeria in totally 100 strains.
- Sampling of surfaces: We adopted the method of wet swab on a surface of 25 cm^2 [12,13].
- Sampling of air: collect of air.
- The culture medium used: blood agar and Chapman agar.
- Incubation: 37 °C for 18 hours.
- Identification: Phenotypic of colonies, Microscopic identification: Gram stain.
- Biochemical Identification: Test Staphylocoagulase and Gallery biochemical Api for Staphylococci.
- Antibiotic resistance: We tested a range of antibiotics belonging to different antibiotics families.
- Plants tested: *Cotula cinerea*, *Warionia saharae*, *Bubonium graveolens*, *Launaea nudicaulis* and *Launaea arboresens*.
- Plant Extracts tested: We conducted a total of 40 extracts of five plants selected from locale practitioners typically use the aerial part of the plant without flowers and leaves separated; The number of solvents used was eight divided into two aqueous solvents: Distilled water and the hydrochloric acid, and six organic solvents: methylene chloride, chloroform, methyl alcohol, ethyl alcohol, petroleum ether and acetone.

We put in a 250 ml flask 15 g of plant material to which is added 100 ml of solvent, the mixture is refluxed for 2 hours at a constant temperature.

The extract is filtered and the filtrate is totally dried white rotavapor.

The quantities used to impregnate the discs are drawn from the dried extract.

For the study of activity, résistance and limits of inhibition zone we used different quantities/ Disc which are: 2 mg, 4 mg.

For the study of minimal inhibitory concentration we used different quantities which are: 100 mg, 10 mg, 4 mg, 2 mg, 1 mg, 900 µg, 800 µg 700 µg, 600 µg, 500 µg and 400 µg from extract of distilled water.

Plants were identified at the LPSO (Phytochemistry and Organic Synthesis Laboratory University of Bechar).

*Technical Antibiogram: Concerning the technique of antibiogram, we used the method of agar diffusion standardized by the National Committee for Clinical Laboratory Standards (NCCLS) [11-14] which is an agar diffusion techniques which consists of seeding by tight grooves with a swab from a pure bacterial solution adjusted to 0.5 McFarland. and incubated at 35 °C for 18 hours.

Discussion

It is noted that the reference strain showed no resistance to the antibiotics tested and zones shown were within in the specified limits recommended by Rahal et al. [11] whereas nosocomial strains were considerably resistant to tested antibiotics. Strains of *Staphylococcus aureus* were resistant with a rate of 47.66% considering critical diameters but only 40.06% shown a total resistance with negative inhibition zone (00 mm). Strains of *Staphylococcus epidermidis* were resistant to 32.99% considering critical diameters and only 23.81% shown a total resistance. Strains of both species of genus Staphylococcus tested were resistant to 40.26% considering critical diameters and 27.74% shown a total resistance. *Staphylococcus aureus* showed resistance to most classes of antibiotics tested. These results were consistent with those of Borg et al. [12,13], *Staphylococcus epidermidis* has shown a marked resistance to β lactams. These results correlate with those of Fass et al. [14,15].

Antibacterial effect of plant studied

To interpret the results, we have taken for comparison four parameters that are:

- Rate of activity of Plants extracts: Represented by the number of positive tests (presence of inhibition zones).
- Rate of resistance of Staphylococcus strains.
- Limits of Inhibition zone.
- Minimal Inhibitory Concentration.

We perform a comparison between the antibacterial effect of the plants studied with the effects of all solvents confounded for each plant (Figure 2).

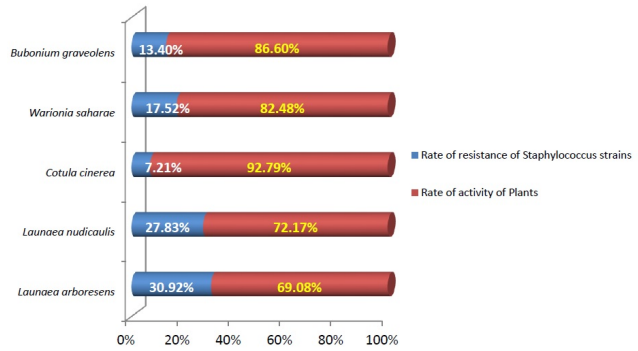

Figure 2: Activity rate of plants extracts.

All of the plants tested shown an antibacterial effect against nosocomial strains of genus *staphylococcus* tested but with variable rate of activity.

It is noted that *Cotula cinerea* has shown against the nosocomial strains the highest activity rate of 92.79% followed by *Bubonium graveolens* with a rate of 86.6%; *Warionia saharae*: 82.48%; *Launea nudicaulis*: 72.17% and *Launeae arboresens*: 69.08% (Figure 3) [15,16].

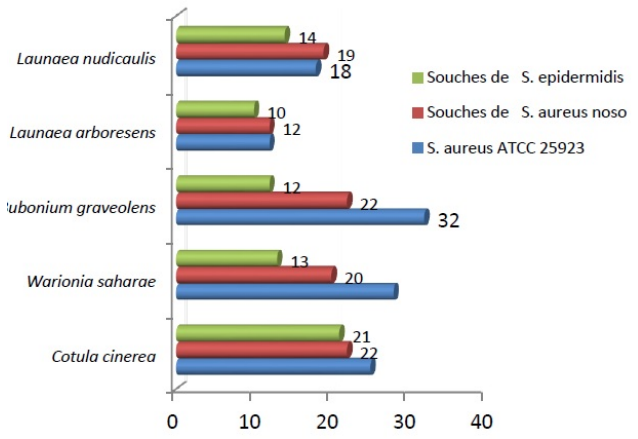

Figure 3: Limits of Inhibition zone of plants studied on the strains tested.

The maximum inhibition zones were shown by *Cotula cinerea* with areas of 22 mm to 21 mm for *S. aureus* and *Staphylococcus epidermidis*, followed by *Bubonium graveolens* with 22 mm for *S. aureus* but only 12 mm for *Staphylococcus epidermidis*. *Launaea arboresens* showed the smaller maximum inhibition zones for all strains tested. The other plants showed maximum inhibition zones relatively closely spaced (Figure 4) [17-20].

Concerning the reference strain maximum zones of inhibition were significantly greater for most of the plants tested (Figure 5).

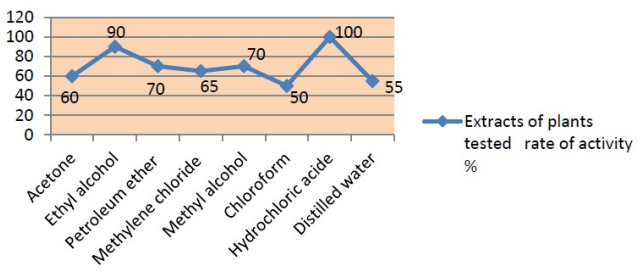

Figure 4: Extracts of plants tested.

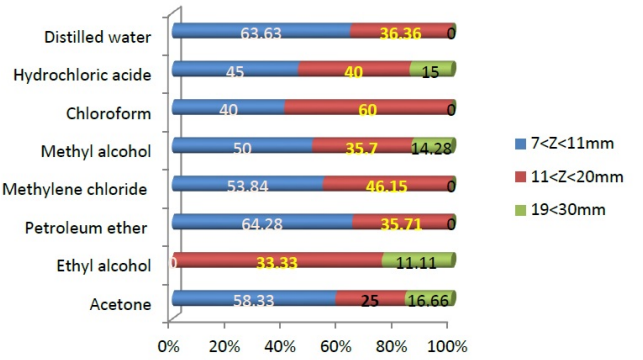

Figure 5: Rates of activity of different plants extracts against Nosocomial strains of genus Staphylococus.

Extracts shown variables rates of activity among strains tested and the highest rate was shown by extracts of hydrochloric acid with 100% with a maximum inhibition zone of 22 mm followed by the ethyl alcohol 90% and inhibition zone of 16 mm methyl alcohol extracts of a rate of 70% and an inhibition zone of 21 mm; petroleum ether:70% and 18 mm; Dichlorométhane:65% and 21% Acetone:60% and 21 mm, Distilled water:55% and 14% Chloroform:50% and 19 mm (Figure 6).

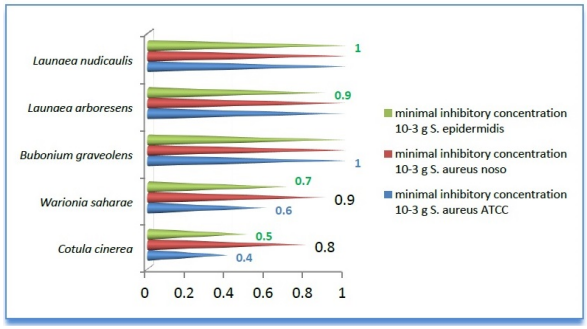

Figure 6: Minimal Inhibitory Concentration.

Plants tested shown minimal inhibitory concentration ranging from 1×10^{-3} g to 5×10^{-4} g. *Cotula cinerea* showed the smallest minimal inhibitory concentration of 5×10^{-4} g followed by *Warionia saharae* with a concentration of 7×10^{-4} g *Launaea arboresens* with a concentration of 9×10^{-4} g and the rest of plants with the concentration of 1×10^{-3} g (Figure 7).

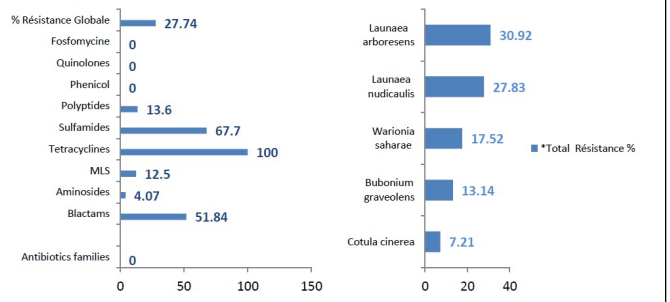

Figure 7: Antibactérial Activity of Plants extracts and Antibiotics families.

Staphylococcus strains tested were Resistant to tetracyclines, Sulfamides and β lactams with successive rates of 100%, 67.70% and 51.84% whereas their rates of resistance for plants tested were included between 30.92% concerning *Launaea arboreseens* and only a rate of 07.21% for *Cotula cinerea*.

Conclusion

Strains of both species of genus Staphylococcus tested were resistant to 40.26% considering critical diameters and 27.74% was totally resistant with negative inhibition zone. Whereas the reference strain showed no resistance to antibiotics tested.

All of the plants tested shown an antibacterial effect against nosocomial strains of genus staphylococcus tested but with variable rate of activity. *Cotula cinerea* has shown against the nosocomial strains the highest activity rate of 92.79%.

The maximum inhibition zones were shown by *Cotula cinerea* with areas of 22 mm to 21 mm for *S. aureus* and *Staphylococcus epidermidis*.

Extracts shown variables rates of activity against strains tested and the highest rate was shown by extracts of hydrochloric acid with 100% with a maximum inhibition zone of 22 mm.

Plants tested shown minimal inhibitory concentration ranging from 1×10^{-3} g to 5×10^{-4} g and *Cotula cinerea* showed the smallest minimal inhibitory concentration of 5×10^{-4} g.

Staphylococcus strains tested were Resistant to tetracyclines, Sulfamides and β lactams with successive rates of 100%, 67.70% and 51.84% whereas their rates of resistance for plants tested were included between 30.92% concerning *Launaea arboreseens* and only a rate of 07.21% for *Cotula cinerea*.

Cotula cinerea has shown the highest antibacterial activity against strains of staphylococcus tested.

Acknowledgments

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