

Research Journal of Pharmaceutical, Biological and Chemical Sciences

A Comparative Study of the Antibacterial Activity of Two Chenopodiaceae:

Haloxylon scoparium (Pomel) and Traganum nudatum Del.

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ABSTRACT

The present work is aimed mainly to investigate and compare the antibacterial activities of some extracts of the aerial parts of two Chenopodiaceae species : *Haloxylon scoparium* and *Traganum nudatum* against seven bacteria strains: three gram negative (*Serratia marcescens ATCC 13880, Pseudomonas aeruginosa ATCC 10145* and *Escherichia coli ATCC 25922*) and four gram positive (*Bacillus subtilis ATCC 6051, Enterococcus faecalis ATCC 29212, Staphylococcus aureus ATCC 25923* and *Bacillus cereus ATCC 11778*) using well diffusion method. The results revealed that both extracts exhibited a certain bioactivity against gram positive bacteria. *H. scoparium* extracts showed higher activity compared to *T. nudatum*. Moreover the butanol extract of *H. scoparium* and the methylene chloride extract of *T. nudatum* proved to be more noteworthy against *Staphylococcus aureus* with a maximum inhibition zone diameter of 22 mm and 19 mm respectively at 1000 µg/ml.

Keywords: Haloxylon scoparium; Traganum nudatum; antibacterial activity.

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INTRODUCTION

The medicinal plants have been used for ages as remedies for human diseases. Plant derived compounds are getting more and more interest owing to their adaptable applications. Today there is an incessant and imperative necessity to find out new antibacterial compounds with various chemical structures and new mechanisms of action because there has been a shocking increase in the prevalence of new and reemerging contagious syndromes. Another big concern is the development of resistance to the antibiotics in current clinical uses [Anubhuti*et al*, 2011; Bouzid *et al*, 2011; Ali-Emmanuel *et al*, 2002; Millogo-Kone *et al*, 2006]. An increasing number of reports dealing with the assessment of antimicrobial effects of different extracts of various medicinal plants are frequently available [Ashokkumar&Ramaswamy, 2013; Bindu *et al*, 2011; Mukundam *et al*, 2012; Khazaei *et al*, 2011]. The aim of this work is to screen the antimicrobial activity of three different extracts of two selected medicinal plants.

Haloxylon scopariun known locally as 'Remth' is used in local folk medicine to cure stomachache, scorpion bites, and wounds infertility and bone pain. In Tunisia and Morocco it is used to treat eye disorders. Aqueous extracts of this plant have also been reported to show anti-cancer, antiplasmodial and larvicidal activity [Ben Salah *et al*, 2002]. Infusion and powder infusion of aerial part of *H. scoparium* are sometimes used for their antidiabetic effects [Bnouham *et al*, 2002].

Traganum nudatum known locally as '*Damran*' is used in traditional medicine to cure some diseases such as Diarrhea, wounds, rheumatism, dermatosis , and others [OuldElhadj *et al*, 2003].

EXPERIMENTAL

Preparation of Extract

The aerial parts of *H. scoparium* were collected from Ghardaia (Berienne region) in November 2012. The aerial parts of *T. nudatum* were collected from Touggourt (gamaa region) in April 2013. The plants were identified by Pr. Abdelmadjid Chehma from Ouargla University and voucher specimens (MA4 and MA5), were deposited at the Chemistry Department, University of Ouargla. The plant materials were dried under dark and then ground and stored in closed container away from light and moisture.

The extracts were prepared by soaking 200 g of the plant powder in a mixture of $EtOH/H_2O$ [70/30] for 24 hours. The procedure was repeated three times and the filtrates were combined before being evaporated under reduced pressure. The resulting extracts were diluted with distilled water and left overnight. The filtrates were subjected to extraction by various solvents with increasing polarity (petroleum ether, dichloromethane, ethyl acetate, and butanol). The organic phases were separated and evaporated.

Microorganisms

All bacterial standard strains: Serratia marcescens ATCC 13880, Pseudomonas aeruginosa ATCC 10145, Bacillus subtilis ATCC 6051, Bacillus cereus ATCC 11778, Escherichia coli ATCC 25922, Enterococcus faecalis ATCC 29212 and Staphylococcus aureus ATCC 25923 were obtained from the National Reference Center for Streptococci at the University Hospital of Aachen in Germany.



Preparation of the bacterial culture media

3.7 % of Mueller Hinton agar was mixed with hot distilled water and autoclaved at 121°C and 2 atm for 15 minutes. After autoclaving, it was allowed to cool to 45°C in a water bath. Then the medium was poured into sterilized petri dishes with a uniform depth of approximately 5 mm [Cappuccino and Sherman, 1999; Swarnamoni *et al*, 2013].

Preparations of plant extract impregnated discs

Whatman N°1 filter paper was used to prepare discs of 6 mm in diameter. They were sterilized by autoclaving and then dried during the autoclaving cycle. The discs were then impregnated with the extract of the plants [Swarnamoni *et al*, 2013].

Disc diffusion method

Disc diffusion method for antimicrobial susceptibility test was carried out according to the standard method by Kirby-Bauer [Bauer *et al*, 1966]to assess the presence of antimicrobial activities of plant extracts. A bacterial suspension adjusted to 0.5 McFarland standard (1.5×10 ⁸ CFU/ml) was used to inoculate Mueller Hinton agar plates evenly using a sterile swab. The discs impregnated with the plant extracts were placed individually on the Mueller Hinton agar surface with flamed forceps and gently pressed down to ensure contact with the agar surface. The discs were spaced far enough to avoid both reflection waves from the edges of the Petri dishes and overlapping rings of inhibition. The plate was then incubated at 37°c for 18 hours in inverted position to look for zones of inhibition. Zones of inhibitions produced by the sensitive organisms were demarcated by a circular area of clearing around the plant extract impregnated discs. The diameter of the zone of inhibition through the center of the disc was measured to the nearest millimeter.

RESULTS

	Diameter of Inhibition zone (mm)									
Bacteria strains		Methylene chloride			Ethyl acetate Extract			Butanol Extract		
		Extract (µg/ml)			(µg/ml)			(µg/ml)		
	300	500	100	300	500	1000	300	500	1000	
			0							
Serratia marcescens ATCC 13880	0	0	0	0	0	0	0	0	0	
Pseudomonas aeruginosa ATCC 10145	0	0	7	0	0	0	0	0	7	
Bacillus cereus ATCC 11778	0	13	16	0	10	16	6	12	16	
Bacillus subtilis ATCC 6051	7	13	14	0	9	11	0	9	13	
Escherichia coli ATCC 259220	0	0	8	0	0	0	0	7	9	
Enterococcus faecalis ATCC 29212	0	0	0	0	0	0	0	0	0	
Staphylococcus aureus ATCC 25923	8	12	15	8	11	13	14	17	22	

Table 1: Antibacterial activity of *Haloxylon scoparium*.

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Bacteria strains		Diameter of Inhibition zone (mm)									
		Methylene chloride			Ethyl acetate			Butanol Extract			
		Extract (µg/ml)			Extract (µg/ml)			(µg/ml)			
	300	500	1000	300	500	1000	300	500	1000		
Serratia marcescens ATCC 13880	0	0	7	0	0	0	0	0	0		
Pseudomonas aeruginosa ATCC 10145	0	5	7	6	7	9	0	0	7		
Bacillus cereus ATCC 11778	0	0	13	0	0	10	0	9	11		
Bacillus subtilis ATCC 6051	7	12	13	0	6	10	0	0	0		
Escherichia coli ATCC 259220	0	0	0	0	0	0	0	0	0		
Enterococcus faecalis ATCC 29212	0	0	0	0	5	8	0	0	7		
Staphylococcus aureus ATCC 25923	10	12	19	8	9	15	6	10	14		

Table 2: Antibacterial activity of Traganum nudatum .

DISCUSSION

Results for antibacterial activity as obtained with different solvent extracts of two plants revealed that the three extracts of *haloxylon scoparium* exhibited a positive effect against three types of gram positive bacteria *Bacillus cereus, Bacillus subtilis, Staphylococcus aureus* where the maximum activity was recorded against *Staphylococcus aureus* and a maximum inhibition diameter of 22 mm with the butanol extract. On the other hand the three extracts were ineffective against *Serratia marcescens* and *Enterococcus faecalis*. Moreover the ethyl acetate extract showed no effect against *Pseudomonas aeruginosa and Escherichia coli*. Weak inhibition was recorded with the dichloromethane and butanol extracts against *Pseudomonas aeruginosa* and *Escherichia coli*. As far as *Traganum nudatum* is concerned the three extracts exhibit no actions against *Escherichia coli*, and *Serratia marcescens*. However weak effects are recorded with the remaining bacteria strains with an important increase in favor of *Staphylococcus aureus*.

Generally, the extracts of two plants are more or less effective towards the tested bacteria and *haloxylon Scoparium* extracts are more potent compared to *Traganum nudatum* extracts.

CONCLUSION

This study underscored the antimicrobial activity of two chenopodiaceae species namely: *Haloxylon scoparium* and *Traganum nudatum* using three different solvents with increasing polarity against seven bacteria strains. The two plants averred to be effective against three types of Gram positive bacteria and *Haloxylon scoparium* extracts are more effective compared to *Traganum nudatum*. The results partially justify the claimed uses of the two selected plants in the traditional system of medicine to treat various infectious diseases caused by the microbes. Further chemical and pharmacological investigations may be carried out to isolate and identify the chemical constituents in the selected plants responsible for the antimicrobial activity.

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