Antimicrobial activity of extracs from *Ageratina neriifolia* (Asteraceae)

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Abstract

The antimicrobian activity of ethanol, acetone and aqueous extracts of the aereal parts of Ageratina neriifolia (Asteraceae) were evaluated againts Staphylococcus aureus (ATCC 25923), Enterococcus faecalis (ATCC 29212), Escherichia coli (ATCC 25992), Pseudomonas aeruginosa (ATCC 27853) and Candida albicans (ATCC 90028), using the disc diffusion method. Only the acetonic and ethanolic extracts showed activity against S. aureus, E. faecalis, proving that the compounds present in A. neriifolia are active againts Gram positive bacterias. This is the first time antimicrobian activity has been reported for this species.

Key words: Ageratina neriifolia; antimicrobial activity; asteraceae.

Actividad antimicrobiana de los extractos de *Ageratina neriifolia* (Asteraceae)

Resumen

The antimicrobian activity of ethanol, acetone and aqueous extracts of the aereal parts of Ageratina neriifolia (Asteraceae) were evaluated againts Staphylococcus aureus (ATCC 25923), Enterococcus faecalis (ATCC 29212), Escherichia coli (ATCC 25992), Pseudomonas aeruginosa (ATCC 27853) and Candida albicans (ATCC 90028), using the disc diffusion method. Only the acetonic and ethanolic extracts showed activity against S. aureus, E. faecalis, proving that the compounds present in A. neriifolia are active againts Gram positive bacterias. This is the first time antimicrobian activity has been reported for this species.

Palabras clve: Ageratina neriifolia; antimicrobial activity; asteraceae.

1. Introduction

Investigations on the family Asteraceae have demostrated that a variety of species belonging to it have strong antimicrobial activity against commonly microorganisms related to different infections (1, 2).

The *Ageratina* Spach genus belongs to the family Asteraceae (tribe Eupatoriae), it compri-

ses around 248 species divided in to 5 subgenera (3). Extracts of the leaves of the *A. gracilis* (*Eupatorium gracile*) and *A. viscosa*, have been used in folk medicine as tonic, antipyretic, emmenagogue, diaphoretical and the roots to treat syphilis (4).

A variety of compounds such as benzofurans, cromens, flavonoids, poliacetilens, sesquiterpens, triterpens, diterpens and steroids has been isolated of *Ageratina* genus (5-10). Kaurene

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diterpenoids have been reported for *A. neriifolia* (11); this type of components have also been isolated from *Espeletia barclaya* (Asteraceae) showing antimicrobial activity (12).

In this investigations the antimicrobial activity of different crude extracts from the aereal parts of *Ageratina neriifolia* were tested against *Staphylococcus aureus* (ATCC 25923), *Enterococcus faecalis* (ATCC 29212), *Escherichia coli* (ATCC 25992), *Pseudomonas aeruginosa* (ATCC 27853) and *Candida albicans* (ATCC 90028) by using the agar diffusion disc method.

2. Materials and Methods

2.1. Plant material

The aereal parts of *A. neriifolia* were colleted from El Valle (Mérida state -Venezuela). The plant material was identified in the herbarium MERF of the Facultad de Farmacia y Bioanálisis, Universidad de los Andes (ULA), Mérida-Venezuela. A herbarium voucher was lodged in the herbarium MERF (DB003).

2.2. Extraccion of plant material

Plant material dried and grounded (25 μ g) was extracted separately with ethanol, acetone and distilled water by steeping at room temperature for 12 h. All the extracts were stirred up for 30 minutes using an automatic rotator at 200 rpm over a steans bath at 60°C. These extract were filtrated and concentrated under reduced pressure at 40°C of temperature. Every dried extract was redissolved in 10 ml of solvent (ethanol, acetone and distilled water, respectively) and from these concentrated extracts, dilutions (1:10) were prepared.

2.3. Evaluation of antimicrobial activity

The microorganisms used in the present investigation were *Staphylococcus aureus* (ATCC 25923), *Enterococcus faecalis* (ATCC 29212), *Escherichia coli* (ATCC 25992), *Pseudomonas aeruginosa* (ATCC 27853) and *Candida albicans* (ATCC 90028). The antimicrobial activity was carried out according to the disc difussion method described by Ramírez *et al.*, 2000, and partially modified in the laboratory.

Every bacteria culture was incubated in 2.5 mL of Mueller Hinton broth at 37°C of temperatu-

re for 18 h, except for C. albicans that was incubated in Sabouraud Dextrosa agar at 25°C. Mueller Hinton agar and Sabouraud Dextrosa 4% agar were placed in to Petri dishes for bacterias and the yeast (C. albicans), respectively. Once solified, a suspension of each microorganism was adjusted with SSF to the MacFarland N° 0.5 turbiness patron. This suspensions was then streaked evenly on the agar surface with a sterile swab. Filter paper discs (12 mm) satured with 50 μL of every extract were placed on the inoculated agar surface and different negative controls (ethanol 95%, acetone and distilled water) according to each case were also tested. A positive control was also assayed to check the sensitivity of the tested organisms using the following antibiotics: Ampicillin-Sulbactam® (10/10 μg), Vancomycin® (30 μg), Streptomycine® (10 μg), Cefoperazone® (75 μg) and Fluconazole (25 μg/mL). The plates were pre incubated at 4°C for 18 h and then incubated at 37°C for 24 h. The inhibition zone readings were carried out at 24 and 48 h (13). The diameter of the inhibition zone was expressed in millimeters (mm). The test was considered negative whenever microbial growth was observed around the disc as well as negative controls. The experiments were repeated at least twice (14).

3. Results and Discussion

Leaf and steam extracts once concentrated showed the following yieldings 5.89~g~(23.56%) and 7.65~g~(30.60%) in distilled water, 3.11~g~(12.44%) and 1.67~g~(6.68%) in acetone and 5.72~g~(22.88%) y 3.27~g~(13.08%) in etanol 95%. These concentrated extracts were then redissolved in 10 ml of each solvent.

The results of the antibacterial activity carried out with different extracts of *A. neriifolia* indicates that the acetonic and ethanolic extracts (concentrated and diluited) of leaves and stems are active against Gram positive bacterias (*S. aureus* ATCC 25923 y *E. faecalis* ATCC 29212). None of the extracts tested showed activity against Gram negative bacterias or the yeast *Candida albicans* ATCC 90028 (Table 1).

Antibacterial activity of *A. neriifolia* was evaluated against Gram positive and Gram negative bacteria. These microorganisms are both

Tabla 1 Antimicrobial activity of Ageratina neriifolia

			Leaves	es					Stems	ms			Ref	erenc	ce Co	Reference Compounds	qs
Concentrated	ntrate	G	q	Dilu	Dilution 1:10	:10	Cone	Concentrated	ited	Dilu	Dilution 1:10	:10					
E A		-	W	臼	A	W	臼	A	W	臼	A	M	SAM VA	VA	S	CEF	FL
17* 18*	*		0	15*	14*	0	19*	18*	0	14*	14*	0	20*	1	1	1	1
16* 16* (•	0	14*	13*	0	16*	17*	0	14*	14*	0	1	27*	1	1	1
0 0		0	0	0	0	0	0	0	0	0	0	0	1	1	22*	1	l
0 0		0	0	0	0	0	0	0	0	0	0	0	1	1	I	27*	I
0 0			0	0	0	0	0	0	0	0	0	0	1	1	1	I	25*

E: Ethanol extract; A: Acetone extract; W. Aqueous extract. SAM: Ampicillin – Sulbactam; VA: Vancomycin; S: Streptomycin; CEF: Cefoperazone; FL: Fluconazole. *: Values (Inhibition zone in mm, disc diameter 6 mm, average of two consecutive assays); 0: No inhibition zone was observed.

morphologically and physiologically different, thus the results obtained are representative of the antibacterial activity of these plant extracts. In the present investigation it was demonstrated the potencial activity of this species against some human pathogens microorganisms such as *S. aureus* y *E. faecalis*.

Antibacterial activity have been reported for a other genera of the family Asteraceae (14-17). Such activity has been atributed to the presence of flavonoids and kaurene diterpenoids present in the genera (2, 12, 13, 18).

4. Conclusions

Further investigations would be of interest due to the potential antimicrobial activity of *A. neriifolia* against Gram positive bacterias in order to identify the substance(s) responsible for this activity. This is the time antimicrobial activity has been reported for *A. neriifolia*.

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