

# Antibacterial *in vitro* of effect *urtica dioica* and *piper angustifolium* in alpacas (*vicugna pacus*) with diarrheal enteropathies

## Abstract

The study was antibacterial to the effect of *Urtica dioica* and *Piper angustifolium* *in vitro* in alpacas with enteropathy. 120 faecal samples from 90 calves born 15 days alpacas were taken. *Escherichia coli*, *Clostridium sp* were obtained through biochemical tests. The antibacterial susceptibility was evaluated in three groups: Matico G1, G2 and G3- Nettle Witness-Enrofloxacin concentrations 10 and 5% by disk diffusion method and method Optical turbidimetry. The MIC and MBC was significant ( $P < 0.01$ ) *Escherichia coli* in groups G1 (Sensitive= $29.3 \pm 0.2a$ , Intermediate= $15.2 \pm 0.3a$ , resistance= $0.0$ ) and G2 (Sensitive= $26, 1 \pm 0.4ab$ , Intermediate= $16.2 \pm 0.2a$  resistance= $0.0$ ) and CMB G1 ( $0.1a \pm 22.0, 21.0 \pm 1.3a$ ), G2 ( $0.5ab \pm 37.0, 39.0 \pm 0.2ab$ ) against group G3 ( $=20.2 \pm 0.2ac$  Sensitive, Intermediate = $15.1 \pm 0.1ac$ , Resistance= $10.4 \pm 0.3c$ ) and CMB ( $0.4ac \pm 420, 460 \pm 1.6c$ ), *Urtica dioica* and *Piper angustifolium* demonstrate antibacterial effectiveness in alpacas of diarrheal enteropathies.

**Keywords:** alpacas, enteropathies, *piper angustifolium*, *urtica dioica*

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## Introduction

The domestic South American Camelids, the alpaca and llama represent a strategic natural resource, of great social, economic and ecological importance in the life of the high Andean population of Peru and Bolivia and to a lesser degree Argentina, Ecuador and Chile, mainly associated with the production of fibre and meat.<sup>1</sup> About 90% of the alpacas are in the hands of small producers and peasant communities.<sup>2,3</sup> In these places, the exploitation is carried out following traditional production systems, lacking adequate technologies and having high mortality rates of alpaca neonates, reaching up to 70%,<sup>4</sup> the *Clostridium perfringens* and enteropathogenic *Escherichia coli* are of greater sanitary and economic impact in the alpaca production<sup>5</sup> because they are causal agents of pathologies that present with intestinal dysfunction, generating the neonatal diarrheal complex in the alpacas<sup>6</sup> and there are no veterinary or related products to date that could be effective for the treatment of the neonatal diarrheal complex in South American camelids,<sup>1</sup> this led us to study the antibacterial effect of *Urtica dioica* and *Piper angustifolium* *in vitro* on alpacas with diarrheal enteropathies. Ethanolic del extract of *Urtica dioica* and *Piper angustifolium* *in vivo* as antibacterial This would be an alternative to reduce the high mortality levels of alpaca neonates because they are excellent antibacterials for the neonatal diarrhoea complexes of alpacas and stimulators to reduce the immunological resistance of the causative agents of diarrheal bacterial pathologies.

## Materials and methods

The study was carried out in the Central Animal Health Area Research Laboratory of the National University of Huancavelica-Peru, the extraction of the extractosetanolics was carried out according to the protocol dissertated by Hernández<sup>7</sup> which consisted of: collection, selection and drying, pulverization, weighing, addition of solvents,

stirring, 2nd addition of solvent, casting, filtration, solvent removal, resuspension and sterilization. 120 stool samples were collected from 90 alpaca pups with enteropathies. The samples were taken by the rectal swab method and suspended in Stuart transport medium. For the isolation and identification of the microorganisms, biochemical tests were used using selective media of Macconkey agar and TNS agar, the study groups were: G1-Matico, G2-Ortigay G3 Control-Enrofloxacin at a concentration of 5 and 10%. The antibacterial active ingredient was determined by the disk diffusion method [Minimum Inhibitory Concentration (MIC)] and optical method of Turbidimetry (McFarland and absorbance by spectrophotometry) described by Aldana<sup>8</sup> [Minimum Bacterial Concentration (CMB)]. Analysis of variance and the Tukey test ( $P < 0.01$ ) using the SPSS v. twenty.

## Result and discussions

The significant antibacterial effect of CMB ( $P < 0.01$ ) showed the G1 groups ( $22.0 \pm 0.1a, 21.0 \pm 1.3a$ ), G2 ( $37.0 \pm 0.5ab, 39.0 \pm 0.2ab$ ) that the group G3 ( $42.0 \pm 0.4ac, 46.0 \pm 1.6c$ ), there are still significant differences in the groups G1 and G2 ( $P < 0.01$ ) in the concentrations of 5 and 10%, *Escherichia Coli* being very sensitive that the *Clostridium sp* (Table 1). The antibacterial effectiveness demonstrated by nettle and matico, is due to the fact that these plants contain components of fluoroquinolones and flavonoids,<sup>9</sup> which are based on their mechanism of action in inhibiting the enzyme DNA gyrase de enterobacteria. Thanks to these mechanisms, inflammatory inhibition occurs. Cell tissue regeneration.<sup>10</sup> A study conducted by Maravi<sup>11</sup> with samples of vocal shrouds of patients with conopharynx, reported the CMB ( $0.3562 \pm 1.6$ ) on *Streptococcus mutans* with Oregano and with *Lactobacillus acidophilus* ( $0, 4812 \pm 1.2$ ) and *Candida albicans* ( $0.3922 \pm 1.0$ ) with *Hierba Luisa*; Ocares<sup>12</sup> reports that the dichloromethane extract of *Rumex palustris* showed antimicrobial activity against *Staphylococcus aureus* and *Enterococcus faecalis*

at CMB of 1000µg/ml, being inactive against *Escherichia coli* and *Pseudomonas aeruginosa*, and there are no studies aimed at animal health the ethnopharmacology of there its importance of the study.

The CMI was significant ( $P < 0.01$ ) in the G1 group (Sensitive=29.3±0.2a, Intermediate=15.2±0.3a, Resistant=0.0) and G2 (Sensitive=26.1±0.4ab, Intermediate=16.2±0.2a Resistant=0.0,) than the group aG3 (Sensitive=20.2±0.2ac, Intermediate=15.1±0.1ac, Resistant=10.4±0.3c) and greater sensitivity showed in *Escherichia*

*Coli* than in *Clostridium sp* (Table 2), because the *Dioica* and *Piper angustifolium* have antibacterial components that inhibit cell wall synthesis and nucleic acids causing cell destruction and degradation.<sup>13</sup> Becerra<sup>14</sup> use ethanolic extract of *Eucalyptus globulus* at concentrations of: 100%, 50%, 25%, 12.5, 6.25% and 3.13% against *Lactobacillus sp*, finding the MIC at 3.13% and the 6.25% WBC and the aqueous extract did not present an antibacterial effect on the strain of *Lactobacillus sp*.

**Table 1** Measures and standard deviation of the minimum bacterial concentration (CMB) of *Escherichia coli* and *Clostridium sp* (650nm)

Groups	Concen	Doses	N	<i>Escherichia coli</i>		<i>Clostridium sp</i>	
				Pre absorbance (nm)	Post absorbance (nm)	Pre absorbance (nm)	Post absorbance (nm)
G1 EE Matico	10%	10ML	20	66,0±0,2a	22,0±0,1a	64,0±1,2a	21,0±1,3a
	5%	5ML	20	64,0±0,8b	28,0±0,3b	58,0±0,7b	28,0±0,5b
G2 EE Ortiga	10%	10ML	20	65,0±0,4ab	37,0±0,5ab	62,0±1,3ab	39,0±0,2ab
	5%	5ML	20	59,0±0,1b	34,0±0,2b	62,0±0,1ab	37,0±0,6ab
G3 Enrofloxacino	10%	10ML	20	61,0±0,5c	42,0±0,4ac	67,0±0,6c	46,0±1,6c
	5%	5ML	20	63,0±0,6c	45,0±0,1c	66,0±0,4ac	49,0±0,4ac

a≠b, b≠c to Tukey prueba ( $P < 0.01$ )

**Table 2** Measures and standard deviation of minimum inhibitory concentration (MIC) *Escherichia coli* and *Clostridium sp*

Treatments	Concen µL	N	<i>Escherichia coli</i> (R ≤14, I 14 ≥18, S ≥18)			<i>Clostridium sp</i> (R ≤14, I 14 ≥18, S ≥18)		
			120	S	I	R	S	I
G1 EE Matico	10	20	29,3±0.2a	15,2±0.3a	0,0	27,3±0.1a	15,2±0.4a	0,0
	5	20	28,6±0.2b	14,4±0.5b	0,0	26,6±0.2b	14,4± 0.5b	0,0
G2 EE Ortiga	10	20	26,1±0.4ab	16,2±0.2a	0,0	23,1±0.3ab	17,2±0.2a	0,0
	5	20	18,5±0.1b	14,1±0.6b	0,0	19,5±0.3b	16,1±0.6b	0,0
G3 Enrofloxacino	10	20	20,2±0.2ac	15,1±0.1ac	10,4±0.3c	22,3±0.5ac	18,1±0.1ac	13,4±0.6c
	5	20	19,0±0.4c	16,8±0.1ac	11,0±0.3ac	18,0±0.4c	14,8±0.3ac	11,2±0.2ac

a≠b, b≠c to the Tukey prueba ( $p < 0.01$ )

## Conclusion

*Urtica dioica* and *Piper angustifolium* showed antibacterial effectiveness *in vitro* in alpacas with diarrheal enteropathies.

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## Conflict of interest

The authors declare no conflict of interest.

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