



## Analysis of the coxal liquid of *O. brasiliensis* tick species by analytic techniques

C. B. Zamboni<sup>1</sup>, D. N. S. Giovanni<sup>1</sup> and S. M. Simons<sup>2</sup>

<sup>1</sup>czamboni@ipen.br, Instituto de Pesquisas  
Energéticas e Nucleares IPEN/CNEN-SP,  
Brasil

<sup>1</sup>daltongiovanni@alumni.usp.br, Instituto de  
Pesquisas Energéticas e Nucleares  
IPEN/CNEN-SP, Brasil

<sup>2</sup>simone.simons@fundacaobutantan.org.br,  
Instituto Butantan, IBu-SP, Brasil

### 1. Introduction

The *Ornithodoros brasiliensis* (*O. brasiliensis*) (*ACARI: ARGASIDAE*) (*ARAGÃO, 1923*) species belongs to the Argasidae familie, order Ixodida. It is found only in Brazil, with restricted distribution to Rio Grande do Sul (Southern region) [1]. These ticks are blood sucking parasites on wild and domestic animals and human beings and can trigger severe manifestations toxic (fever and toxic syndrome ticks) [2]. An important characteristic of this family is their rapid engorgement, which leads to the development of strategies for maintaining osmoregulation and the hydrostatic equilibrium, by secreting coxal liquid (excess body fluid secreted by glands coxais during and after feeding) [3]. Their body fluids secreted can act a vector of diseases causing clinical disturbances. This tick has been the target of several studies as a vector of diseases (parasitism and infectious diseases) and induction of slow-healing lesions caused by its salivary gland [4]. For decades there was no accident records caused by this tick, raising doubts about its extinction. However, in the last years, cases of parasitism have been reported mainly in Southern region [5]. Since, the number of accidents caused by this tick has increased. These facts have stimulated the investigation of their secretions for the understanding of the many physiological processes. In this investigation, coxal liquid (CL) samples were investigated by Instrumental Neutron Activation Analyses (INAA) and Energy Dispersive X-Ray Fluorescence (EDXRF) techniques aiming to get a detailed description of its elemental composition. This knowledge contributes for tick coxal liquid characterization contributes for interpretation of the metabolic processes as well as to evaluate its toxicological potential.

### 2. Methodology

The biological material came from Butantan Institute (IBu) at São Paulo city, Brazil. The *O. brasiliensis* species used in this study were collected in São Francisco de Paula (29 ° 26 '52 "S 50 ° 35' 02" W), state of Rio Grande do Sul (Brazil). The colony of ticks was kept in a controlled environment chamber (BOD) at the Parasitology Laboratory of the Butantan Institute (IBu, SP) at 20 ± 1 ° C and 90 ± 10 % of humidity. The collection (~2ml) was carried monthly, for 6 months, out in micropipette manual, frozen (in dry ice) and lyophilized. Considering that the INAA and EDXRF procedures are non-destructive, the same sample was used for both analyses. For INAA the samples were irradiated at IEA-R1 (4.5 MW, pool type) nuclear reactor at IPEN in a thermal neutron flux, for minutes to hours, and gamma counting using HPGe (GEM-60195) detector connected to a MCA ORTEC-919E. For EDXRF analysis, the samples were placed inside sample holder (with 6.3 mm in the diameter) and excited and measured using a portable X-Ray Spectrometer: Ag X-Ray target and Si Drift detector (25 mm2 x 500 µm / 12.5 µm Be window) and counting time of 200 to 600 s

measured with 30 kV and 5  $\mu$ A.

### 3. Results and Discussion

The elements concentrations determined in the coxal liquid samples are presented in Table 1 by the mean values with associated error, represented by one standard deviation (1SD).

Table I: Element concentrations in coxal liquid of *O. Brasiliensis* species by NAA and XRF technique

Elements	AAN	FRX
n=6	MV $\pm$ 1SD	
Br, mg/g	0.119 $\pm$ 0.022	0.136 $\pm$ 0.020
Mg, mg/g	1.55 $\pm$ 0.38	nd
Mn, $\mu$ g/L	2.23 $\pm$ 0.78	nd
Na, mg/g	293 $\pm$ 9	nd
K, mg/g	6.96 $\pm$ 1.02	8.5 $\pm$ 0.9
Cl, mg/g	349 $\pm$ 11	322 $\pm$ 30
P, mg/g	80 $\pm$ 9	77 $\pm$ 11
Ca, mg/g	2.7 $\pm$ 0.8	3.4 $\pm$ 0.7
S, mg/g	143 $\pm$ 40	115 $\pm$ 28
Fe, mg/g	201 $\pm$ 10	195 $\pm$ 12
Zn, mg/g	40.8 $\pm$ 0.9	46.1 $\pm$ 2.3

n: number of samples  
 MV: mean value  
 SD: Stander Deviation  
 nd: not determined

The student's *t-test* was applied for results comparison between EDXRF and INAA techniques. The results for Br, P, Cl, K, Ca, Fe, and S were considered statistically equal ( $p > 0.05$ ). Furthermore, according to table 1, the presence of heavy metals was not observed.

### 4. Conclusions

Neutron Activation Analyses (NAA) and X Ray Fluorescence (XRF) techniques presents get a detailed description of its elemental composition. The knowledge of the elemental composition contributes to the tick coxal liquid physical-chemical and biological characterization, to the interpretation of the metabolic processes, as well as to evaluate its toxicological potential.

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