

# NAA TECHNIQUE FOR CLINICAL INVESTIGATION OF MICE IMMUNIZED WITH *BOTHROP* VENOM

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**Abstract.** In the present study Neutron Activation Analysis (NAA) technique was used to determine sodium concentration in whole blood of mice immunized with *Bothrops* venom. With this value it was possible to perform clinical investigation in this animal model using whole blood.

**Keywords:** mice, blood, *bothrop* venom, NAA  
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## INTRODUCTION

To produce several types of antivenom including anti-*bothrops* serum, mice are currently used as an animal model due to the low cost and also to the easiness related to medico-legal implications. Considering that Na is the major component in the *Bothrops* venom (mix of snake venom that has high prevalence in Brazil) and also majority in blood, in this study Neutron Activation Analysis technique was used to determine its concentration in whole blood of mice not immunized (control group) and in mice immunized with *Bothrops* venom Atrox Rio Negro (Amazonas) from north region of Brazil. With these data it was possible to check the clinical status of this laboratory animal using whole blood during immunization process.

## EXPERIMENTAL PROCEDURE

The samples came from Instituto Butantan at São Paulo city. To perform these measurements the blood was collected by the retro-orbital venous plexus from six adult mice of each group. Several biological samples were sealed into a polyethylene bag and irradiated for 2 minutes in the IEA-R1, 2-4MW nuclear reactor at IPEN (in a thermal neutron flux of  $\sim 10^{11}$  n/cm<sup>2</sup>.s<sup>1</sup>). After the irradiation, each sample was gamma-counted for 10 minutes using an HPGe Spectrometer (FWHM = 1.87keV). The concentration of sodium was obtained by using in-house software. Following this experimental procedure (the irradiation time of 2 minutes, counting time of 30 seconds for the Au activation detector and 10 minutes for the biological sample and

background radiation) it was possible to identify  $^{24}\text{Na}$  ( $T_{1/2}=15\text{h}$ ,  $E_{\gamma}=1368\text{ keV}$ ) in several samples (at least six) by one irradiation proving that this nuclear procedure is very agile.

## RESULTS AND DISCUSSION

The concentration of Na in whole blood samples of mice immunized (IG) are shown in Table I. All the results are a mean of duplicate analyses. The reference interval, the minimum and maximum values were also presented. The data for the control group (CG) was included for comparison.

Table I. Sodium concentration in whole blood of mice

Na, $\text{gL}^{-1}$ ( <i>n</i> )	Mean	$\pm 1\text{SD}$	Minimum Value	Maximum Value	Reference Interval $\pm 1\text{SD}$
IG (6)	2.17	0.33	1.79	2.42	1.84 – 2.50
CG (9)	1.74	0.19	1.63	1.96	1.55 – 1.93

(*n*): number of samples

Considering a confidence interval of 95%, usually adopted as references for clinical practice, the mean value for Na in the immunized group ( $1.51\text{gL}^{-1}$ –  $2.83\text{gL}^{-1}$ ) are in agreement with the control ( $1.36\text{gL}^{-1}$ – $2.12\text{gL}^{-1}$ ), suggesting no physiologic differences. However the high Na levels in the immunized group suggest that this element must be constantly evaluated during immunological investigations for sera production using this experimental model.

## CONCLUSION

The biochemical values in blood obtained could be used for clinical practice, for checking the health status, of this laboratory animal. The NAA technique can be used for chemical clinical analysis with advantages [1] against the conventional methods because it uses smaller quantities of biological material (whole blood) something not always possible in the conventional clinical analysis [2].

## REFERENCE

1. L. C. Oliveira, C. B. Zamboni, F. A. Genezini, A. M. G. Figueiredo, G. S. Zahn, J. Radioanal. Nucl. Chem., 263 (2005) 783.
2. J. P. Fréjaville, P. Kamoun, Guide des examens de laboratoire, Flammarion, Paris 1981.

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