Trace Elements at Whole Blood of Distinct Mouse Lines by Using NAA

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Keywords: NAA, trace elements, whole blood, mice
PACS: 82.80.Jp

INTRODUCTION

Animal experimentation is an important and useful tool in many research areas mainly those related to life sciences. Particularly, in the veterinary medicine, small-sized animals are currently used, such as mice, rabbits, guinea-pigs and others on investigations for new medicines and vaccines, as well as for medical diagnostic studies, before to be tested in human being. One of the most used animals is the mouse [*Mus musculus*] due to the low cost, easily handling and also to the facilities related to medico-legal implications. In this study the Absolute Neutron Activation Analysis technique (ANAA) was used to determine the element concentrations, such as, Cl, K, Mg and Na, in whole blood samples of distinct mouse lines. The basic principle of this technique is the irradiation of the biological material with neutrons followed by the measurement of the γ-ray activities induced in the biological sample, where the elements can be identified by the characteristic γ-rays. As this nuclear methodology has been successfully used in the public health field for the investigation of elements in urine, bones and organs of small and medium-sized animals [1, 2, 3] now, we want to extend its application to analyze whole blood samples of eight distinct mouse lines.

EXPERIMENTAL PROCEDURE

To determine the concentration of the elements in the biological samples the Cd Ratio Technique was used for the measurement of thermal flux distribution [3]. In this technique, Au foils ($\sim$1mg), both bare and Cd covered (1mm thick), are irradiated together with the biological sample in the IEA-R1 nuclear reactor at IPEN/SP (IEA-R1, 3MW, pool type), for few minutes, allowing the simultaneous activation of these materials under the exact same irradiation conditions. Using this procedure the γ-ray activities induced in the Au foils by both the thermal and epithermal neutrons were obtained as well as the activation of biological samples. A γ-spectrometer system with a semiconductor detector connected to an ADCAM multichannel analyzer and to a PC computer were then used to measure the induced gamma-ray activity. The detector was a HGPe of high resolution (FWHM=1.85 keV) calibrated for energy and efficiency through the measure-

CP884, *VI Latin American Symposium on Nuclear Physics and Applications*,
edited by O. Civitarese, C. Dorso, G. García Bermúdez, A. J. Kreiner, A. J. Pacheco, and N. N. Scoccola

ments of standard sources of ^{56}Co and ^{152}Eu. For this study was collected whole blood samples from 8 adult females Mice (*Mus musculus*). The biological samples were from the Butantan Institute at São Paulo, Brazil. Samples were obtained from 2 to 3 months old adult female NZB, B_{10}, BALB/c and A/J homozygous isogenic lines and from the genetically selected high antibody responder H_{III} and the low responder L_{III} lines, as well as the lines selected according to the maximal [AIR_{MAX}] or the minimal [AIR_{MIN}] acute inflammatory reactivity. About 0.2 ml of whole blood was collect of each specimen and aliquots of 100 μl was then transferred to the filter paper and dried for few minutes using an infrared lamp. Each sample was sealed into an individual polyethylene bag and irradiated together with the Au foils at the nuclear reactor. Using this procedure it is possible to identify the following radioactive nuclides: ^{38}Cl ($T_{1/2}$=37 min, E_γ =1642 keV), ^{42}K ($T_{1/2}$=12h, E_γ =1525 keV), ^{56}Mn ($T_{1/2}$=2h, E_γ =846 keV) and ^{24}Na ($T_{1/2}$=15h, E_γ =1368 keV). The concentration of each element was then obtained by using a software *Ativação* [4].

RESULTS

The concentration of Cl, K, Mg and Na in whole blood samples of Mice are shown in Table 1. All the results are a mean of triplicate analyses. Theses results were correlated with the indicative interval values for this elements in human being whole blood [5].

TABLE 1. The Concentration of Cl, K, Mn and Na in whole blood samples of Mice

Element	Mean (gl^{-1})	SD (gl^{-1})	Minimum Value (gl^{-1})	Maximum Value (gl^{-1})	Indicative Interval for Humans (gl^{-1}) [5]
Cl	2.54	0.27	2.28	2.90	2.41-3.33
K	2.06	0.40	1.91	2.25	1.30-1.84
Mn	0.0012	0.0001	0.0011	0.0016	0.0010-0.0022
Na	1.66	0.14	1.54	1.83	1.43-1.85

DISCUSSION

The relevancy of this study is that blood represents the most important biological referential to the circulatory system and a great number of general anomalies. Thus, the knowledge of its elemental composition in each strain may reveals physiologic difference among them, providing general notions correlated with genetic and environmental factors that participate determining these expression of these ion in the blood content. Besides, comparing these results with human being whole blood estimation, for Cl, K, Mn and Na, it will be possible to select the similar convenient line or species as reference and for experiments.

ACKNOWLEDGMENTS

The authors would like to thank R. A Aguiar for helping in the blood collection.

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