Recibo: Agosto 2002
Received: August 2002

NUCLEAR METHODOLOGY FOR STUDYING BIOLOGICAL FUNCTIONS OF MAMMALIANS SUBMITTED TO URANIUM INGESTION

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ABSTRACT

To investigate the biological behavior of mammalians that has been fed with chow doped by natural uranium, for a long period, neutron activation analysis has been used to determine the concentration of the elements in the biological samples of blood and urine. According to this investigation we could point some advantages of using nuclear analysis in comparison with the conventional clinical analysis for diagnose anomalies in organs.

Keywords: blood, urine, neutron activation, clinical analysis, mammalians.

I. INTRODUCTION

The present study is part of the large one: "Study of uranium biodistribution in the food chain" [1] , with the aim of investigating the uranium transfer from soil to the human beings. Particularly, in the present experiment, the main idea is to investigate physiological alterations induced by intake of natural uranium in animals. In the health area it is usual to perform a lot of clinical examination, presented in Table 1 to identify anomalies in the human organs but, to perform all these conventional analysis it is very expensive and it needs a lot of biological material. The purpose of this study is to investigate the biological behavior of animals (Wistars, chickens and Beagles) that has been fed with chow doped by natural uranium for a long period using neutron activation analysis to determine the concentration of the elements in the biological materials.

TABLE 1. Clinical Examinations to Investigate the Biological Functions of Human Being Organs.

Conventional Analysis	Biological material/ Quantities	(*)Cost R$
Ionograma	Urine / 5.0 ml	57,60
Natremia	Blood / 3.0 ml	33,70
Na urinary	Urine / 10.0 ml	37,07
Calemia	Blood / 1.0 ml	33,70
K urinary	Urine / 5.0 ml	37,07
Cloremia	Blood / 0.5 ml	30,33
Calcemia	Blood/ 0.5 ml	40,44
Calciúria	Urine 24h/20.0 ml	57,29
Zinc	Blood / 2.0 ml	33,70
Zinc	Urine 24h / 7.0 ml	161,76
Chromium	Blood / 2.0 ml	252,75
Aluminum	Blood / 2.0 ml	33,70
Aluminum	Urine 24h	279,71
Iron	Blood / 1.0 ml	43,81
Iron	Urine 24h / 7.0 ml	242,64

(*) several laboratories have been investigated and these values are the medium cost.

According to Table 1 we can notice that this procedure involves: high cost, large quantities of biological material to perform all analysis and the use of different techniques to perform them. Using the NAA it is possible to identify and to quantify the elements (related in Table 2) present in the biological samples and so to compare the results of the control animal with the doped. For all elements it is necessary to verify if there is changes in the value (concentration measured) along of the time. In this way it is possible to notice any biological anomalies in these animals.

TABLE 2. Nuclear Parameters of the Maim Elements to be Quantify in the Biological Material of Mammalians .

Element/ Biological material	*Nuclear Parameters [2]: Radioisotope ($T_{1/2}$) ; E_γ(keV)*
Aluminum/ Blood	^{28}Al (2.24min); 1779
Aluminum/ Urine	
Bromine/ Blood	^{82}Br (1.47d); 554
Calcium/ Blood	^{49}Ca (8.7min); 3084
Calcium/ Urine	
Chlorine/ Blood	^{38}Cl (37min); 1642
Chlorine/ Urine	
Iron/ Urine	^{59}Fe (44d); 1099
Iron/ Blood	
Zinc/ Blood	^{69} Zn (14 h); 438
Potassium/ Blood	^{42} K (12.2 h); 1525
Potassium/ Urine	
Sodium/ Blood	^{24} Na (15 h); 1368
Sodium/ Urine	
Manganese/ Blood	^{56} Mn (2.58h); 847
Magnesium/ Blood	^{27} Mg (9.5m); 844

II. EXPERIMENTAL PROCEDURE

The present investigation involves rats, dogs and chickens. The biological samples of urine and blood come from experiments which were performed at the facilities of the UNITOX laboratory (protocol: oed 409, mod.180 days - cf. GLP) from the University Santo Amaro (UNISA). In these experiments the animals were housed in bails at controlled room temperature and fed daily with chow doped with uranyl nitrate at different concentrations (from 5 to 100 ppm), except the control animal. For dogs this procedure was performed during 5 months: the uranium ingestion started after weaning (~60 days) and continued in the animal maturity. After the 5Th month these animals were sacrificed. For chickens and rats two months and in the end of the experiments the animals were sacrificed.

During the experiments the daily control and measurements of the ingested food had been carried out as well as the daily control and measurements of the animal weight. The collection of biological materials (urine and blood) had been performed weekly for rats and chickens and fortnightly for dogs.

To determine the concentration of the elements (Table 2) present in the biological samples aliquots of 200 µl of blood were pipetted into 1cm^2 pieces of Whatman N° 40 filter paper, that were sealed in polyethylene bags. Ge detector (FWHM<1.9 keV at 1.32 MeV of ^{60}Co) connected to Adcam multichannel and to a PC computer was used to measure the induced γ-ray activity. The energy calibration and the efficiency of the detector were done using standard gamma rays of ^{60}Co, ^{109}Cd, ^{133}Ba, ^{60}Cs and ^{152}Eu. The efficiency of the detector is shown in Figure 1.

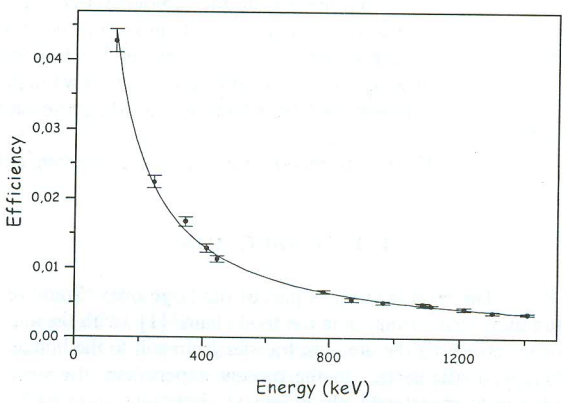

Figure 1. The Fit for the Efficiency of Ge Detector.

The biological samples were irradiated for a period (few minutes to hours) in the IEA – R1m reactor of IPEN/SP, in a thermal neutron flux . After irradiation, the samples were gamma-counted and the area of the select gamma-ray peaks were obtained by using the IDF program [3] . The concentration of the elements was obtained using the absolute method. The same procedure was taken with urine samples.

The thermal neutron flux is measured by gold activation foils (bare and covered with cadmium).

III. RESULTS

To illustrate the procedure of analysis the results involving a blood sample of Beagles will be presented. The γ-ray spectrum obtained for blood sample (dog control) are shown in figures 2 and 3.

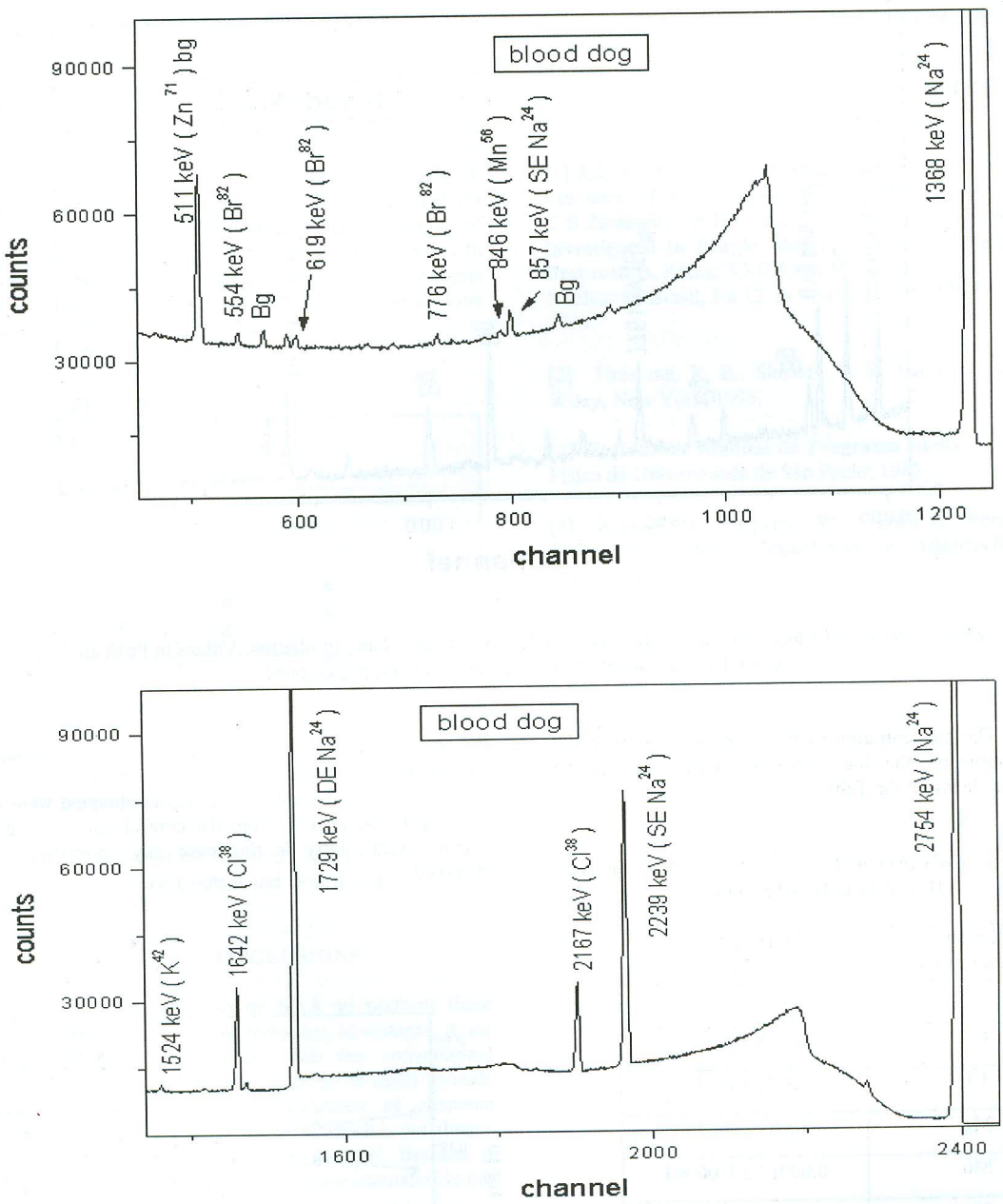

Figure 2. γ Ray Spectrum of Blood Sample of Dog (control), Taken at a Short Time Irradiation. Values in Peak are Energies (in keV). Bg Indicates Peaks Occurring in Natural Background.

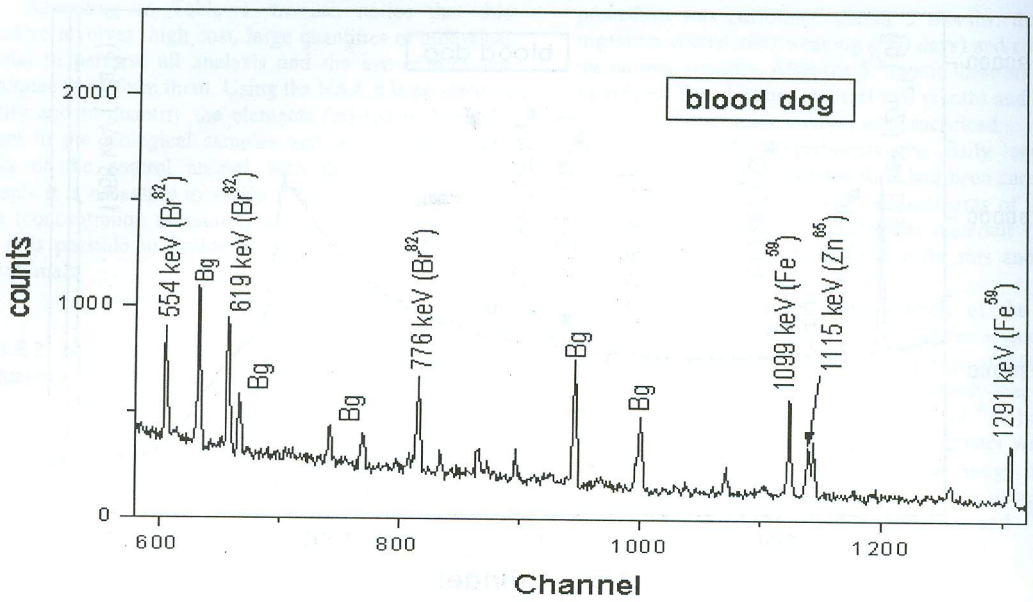

Figure 3. γ Ray Spectrum of Blood Sample of Dog (control) Taken at a Long Time Irradiation. Values in Peak are Energies (in keV). Bg Indicates Peaks Occurring in Natural Background.

The concentration of the elements present in the blood sample of the dog control was calculated and the results are shown in the Table 3.

TABLE 3. The Concentration of Al, Ca, Cl, Mg, Mn, Na, Br and Fe in Blood Sample .

Element / Blood sample	Concentration μg/μl
Al	0.11 ± 0.03
Ca	0.128 ± 0.002
Cl	2.24 ± 0.34
Mg	0.087 ± 0.013
Mn	0.00017 ± 0.00003
Na	1.26 ± 0.19
Br	0.074 ± 0.06
Fe	0.29 ± 0.11

To investigate the biological behavior of animals the elements concentration were calculated in function of the time for each biological sample . For example, the Aluminum concentration in blood dog in function of days are shown in the Figure 4. The results obtained were used to compare the results from the control animal with the doped. Particularly in this case any anomalies where observed.

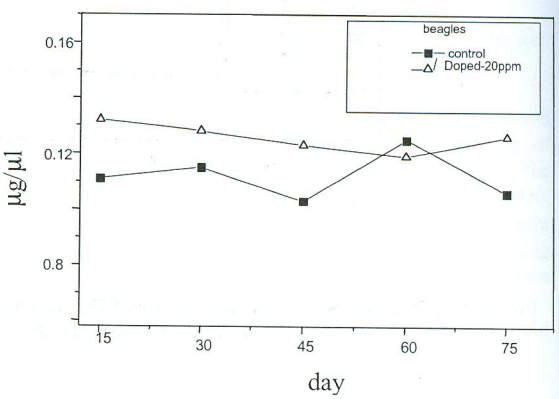

Figure 4. Aluminum Concentration in Blood Dog in Function of the Days.

In a recent study performed by Randal *et al* [4] the normal value for Ca, Mn, K, and Na in serum for mammalians have been established but no values are proposed for blood .

Particularly, the investigation using Beagles are interesting because 90% of their physiological characteristics are similar to humans [4]. In this way the normal value established for human been can be used for comparison. Using this condition the urine sample of Beagles were analyzed and the results were compared with the normal range. To illustrate this procedure of analysis the results involving the behavior of the Chlorine in urine samples are presented in figure 5.

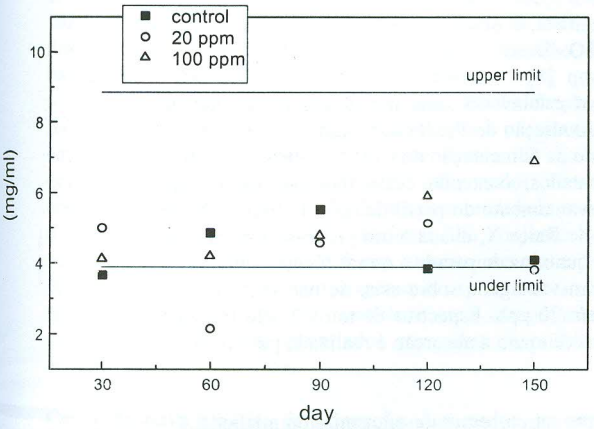

Figure 5. Behavior of the Element Chlorine in the Urine Sample of the Control and Doped Dogs .

IV. CONCLUSIONS

Considering the use of NAA to perform these analysis we could point out the following advantages: it use small quantities in comparison with the conventional analysis that permits a clinical evaluation of small animals It also permits simultaneous evaluation of elements concentràtion in the biological samples, which is not always possible in the conventional clinical analysis. Besides, it can be one alternative method for diagnose anomalies in the biological functions.

Another important advantage is the use of absolute method to calculated the concentration of the elements in the biological samples using neutron activation. Of course this procedure give much more work: it is necessary to performed the measure of the thermal neutron flux and also to obtain the efficiency of the gamma detector. But, considering the fact we need to analyze hundreds samples of biological material, from the three experiments with different mammalians (rats, chickens and dogs), the absolute method became agile (because it is possible to obtain the concentration of activated elements in each irradiation) and economic (because it is not necessary to use standard) .

Regarding the disadvantage of using this method is the necessity of a nuclear reactor.

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