Determination of Sodium in Blood Samples by Instrumental Neutron Activation Analysis Using ²⁴¹Am-Be Neutron Source[†]

D L SAMUDRALWAR & A N GARG*

Department of Chemistry, Nagpur University, Nagpur 440 010 Received 21 July 1986; revised and accepted 1 December 1986

An instrumental neutron activation analysis (INAA) method has been proposed for the determination of sodium in goat, cow, chicken, horse and human blood samples. The method involves irradiation of 1 g dry whole blood sample for 45 hr with thermal neutrons from 241 Am³Be source and counting 1.37 MeV gamma-ray activity of 24 Na on NaI(Tl) detector and a single channel analyzer. The method is simple and accurate.

Low flux neutron sources have been widely used for activation analysis of a number of elements in a variety of matrices.¹⁻⁸ The availability of low flux puts a limitation on their use to a few elements of high natural abundance and thermal neutron cross section along with a product nuclide of short half life. There are only a few reports where these sources have been exploited for the analysis of sodium in plant leaves.⁴ Sodium is one of the most abundant and essential constituent of blood having physiological significance in maintaining pH and osmotic pressure. In spite of its being a major and essential constituent, the determination of sodium in blood samples has posed problems mainly due to non-availability of suitable reagents or complicated chemical procedures.9 Though flame photometric and recently developed ion selective electrode¹⁰ methods are quite common for sodium determination in serum or plasma, these methods require sample dissolution and other chemical manipulations.

In this note, we report an instrumental neutron activation (INAA) method for the determination of sodium in whole blood samples of human and some animals like goat, chicken, horse and cow. The method is simple, accurate, and may also be used for monitoring sodium levels in dry blood samples stored in blood banks.

Sample preparation

Venous blood (20-25 ml) samples, drawn from various species, were mixed with ~ 200 mg citric acid (to avoid coagulation) and dried under an IR lamp at $\sim 150^{\circ}$ C. The powdered sample was passed through 50-100 mesh sieve to obtain even particle size. Weighed samples of 0.5-1.0 g were packed in polythene packets ($3 \text{ cm} \times 1 \text{ cm}$). NBS standard, Bovine Liver SRM-1577,¹¹ and another from IAEA, Animal Muscle, H-4¹², in powder form were used as standards as well as for intercomparison of results.

Irradiation and counting

The samples and standards were irradiated with thermal neutrons using a 5 Ci ²⁴¹Am-Be source for ~ 45 hr. After a delay of 20 min, 1.37 MeV γ -rays of ²⁴Na were counted for 30 min, on a well-type (1.75" × 2")NaI(Tl) detector coupled with a single channel analyzer (ECIL, Hyderabad).²⁴Na was identified by following its half life at the photopeak position. Concentration of sodium was calculated using NBS and IA-EA standards.

Sodium abundances in blood

The mean values of at least four replicate analyses of sodium along with range in percentage dry weight are given in Table 1. Where two or more different samples were analyzed, average of all these values was taken. Also given are the computed concentrations in liquid whole blood (in mg/100 ml) for comparison with the literature values. Sodium concentrations were also calculated in NBS and IAEA standards using each other as a standard. The mean values compare well with the reported values (Table 1), but the observed ranges in both standards as well as samples are quite large. It may be due to low peak to background ratio, varying self absorption effects or the fact that method is not so precise, though it gives accurate results. For IAEA standard, H-4, a range of 0.144-0.27% has been observed by different laboratories¹² and our values are well within this range. Parr¹² has stated an acceptable range of 0.193 to 0.218% with 95% confidence and other values as outliers. Thus, our method yields good results only when a large number of measurements are made and a mean is taken. Perhaps this situation may be improved if a more intense source and a more efficient large crystal detector are used.

It is reported that drying of biological fluids concentrates the elemental abundances, mainly due to water loss. In case of whole blood this concentration is nearly five times.¹³ We have observed the factor, ratio of liquid volume to dry weight, to be least for human blood (4.1-4.5) and highest for calves (5.3-5.8). This obviously reflects the fluid to cellular ratio in the respective samples. Therefore, the concentration of sodium was calculated in mg/100 ml in liquid whole

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Blood species (Age group)	No. of samples analyzed	Conc. range in dry weight (%)	Dry weight† mean(%) ± std. Dev.	Conc. in whole blood‡ (mg/100 ml)	Lit. value (serum) (mg/100 ml)
CALF					
I. Kathiawad (1 yr)	2	0.614-0.654	0.633 ± 0.019	112-119	304-34914
II. Jersey (1 yr)	1	0.800-0.934	0.863 ± 0.069	160-187	304-34914
GOAT					
I. Haemolysed serum(3 yr)	1	0.797-0.936	0.866 ± 0.070	159-187	327-356 ¹⁴
II. Cells only (3 yr)	1	0.204-0.315	0.271 ± 0.049	41-63	25-35 ¹⁴
CHICKEN					
I. Broiler (mature)	2	0.740-0.760	0.750 ± 0.010	144-148	347-370 ¹⁴
II. Egg Laying (mature)	1	0.600-0.781	0.684 ± 0.074	120-156	330-36014
HORSE					
I. Male (4 vr)	1	0.411-0.499	0.441 ± 0.040	83-100	304-349 ¹⁴
II. Female (10 yr)	2	0.408-0.510	0.459 ± 0.051	82-102	304-349 ¹⁴
HUMAN					
I. Unmarried (26 yr)	3	0.452-0.547	0.499 ± 0.048	105-127	90-100 ¹⁵
II. Married (40 yr)	2	0.457-0.545	0.501 ± 0.044	107-128	304-349 ¹⁴
STANDARDS					
I. Muscle (NBS 1577)	1	0.165-0.345	0.258 ± 0.070	-	0.263% ^{11a}
. ,					0.240% ^{11b}
II. Muscle (IAEA H-4)	1	0.146-0.303	0.208 ± 0.062	-	0.207%14

Table 1-Concentration of Sodium in Blood Samples of Animals and Humans

†At least four or more replicate analyses were performed for each sample. †See equation No. 1.

blood based on the ratio of weight/volume and normalised to 100 ml of blood for each sample. Thus,

$$\frac{\text{mg of Na per}}{100 \text{ ml blood}} = \left\{ \frac{\text{Dry weight (g)}}{\text{Liquid volume(ml)}} \times 100 \text{ ml} \right\}$$
$$\times \text{ mg of Na in 1 g dry blood} \quad \dots (1)$$

In Fig. 1, we have plotted the activity vs weight for calf and human blood samples. In both cases a linear plot is obtained upto 1 g and after that a deviation is observed which may presumably be due to self absorption effects or geometric factors. Thus, an optimum sample size of 0.5 to 1.0 g should be used.

In Table 1 are also mentioned the literature values¹⁴ for sodium in serum of various animals. For whole blood no such values are available except for the human blood sample.¹⁵ Blood contains about 45% cells and 55% plasma. While two third of sodium is present in plasma, the rest is in cells.¹⁴ In case of human blood, the plasma value is in the range 300-355 mg and for whole blood 90-100 mg.¹⁵ Thus, our value of 118 mg is ingood agreement. Similarly our mean values of 115 mg for cows (Kathiawad) and 92 mg for horse also compare well with those derived from the literature values for plasma.¹⁴

Since our whole blood concentrations are comparable with the literature values for different species (Table 1), our values are quite acceptable as far as the level of sodium concentration is concerned. The pro-



Fig. 1-Variation of activity with weight of blood in two different samples

posed NAA method is fast, reasonably accurate, economic and non-destructive for the determination of sodium in biological tissue samples. It may be used as a guideline for prior removal over HAP or a resin before proceeding for NAA determination of trace elements. However, it can be used only for sodium content of 1000 ppm or more in a sample size of about 1 g since precision decreases for smaller amounts.

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