

Reference values in blood from inhabitants of Brazil: Br, Cl, K and Na determination using NAA

L. C. Oliveira · C. B. Zamboni · S. Metairon

Received: 1 July 2009 / Published online: 28 July 2009
© Akadémiai Kiadó, Budapest, Hungary 2009

Abstract In this study, bromine (Br), chlorine (Cl), potassium (K) and sodium (Na) were measured in the blood of Brazilian inhabitants from Southeast (S) and Northeast (N) regions. A reference range was established as a function of sex and considering lifestyle factors (non-smokers and non-drinkers). For both regions lower values of K were found in females when compared to males and lower values of Na were found in males compared to females. Increasing trends for Na and Br were observed in the Northeast region.

Keywords NAA · Whole Blood · Reference Values · Biochemistry

Introduction

Knowledge of an element's concentration in body fluids, mainly blood, of persons without occupational exposure is a fundamental need to establish the reference values for evaluation of the intensity of exposure in human beings. The data also provide a basis for checking clinical disorders mainly by intoxication. In many countries, these data have been constantly evaluated but there is a lacuna for the Brazilian population. Based on these facts, in the last few years we have performed several investigations using NAA aiming to establish reference range data for some elements in urine, serum and whole blood in small groups of subjects living in different regions of Brazil [1–5]. These data have

shown the necessity to investigate each element as a function of geographic occupation and gender.

In this context, the elements Br, Cl, K and Na were evaluated in the blood of Brazilian inhabitants (male and female subjects) living in two distinct regions, Southeast (Sao Paulo city) and Northeast (Recife city). These regions were selected because they present some similarities, such as, high concentrations of people living in an industrialized center, suggesting that these inhabitants have a similar history of occupational exposure. However, the alimentary habits and the climate are quite different: while the diet of the subjects living in the Northeast region is rich in sea food, in the Southeast region it is diversified. While the temperature is constant and warm (28–30 °C) in the Northeast region, there are climatic changes, about 5 °C (winter) to 30 °C (summer), in the Southeast region.

Experimental

In this study, the biological samples came from several blood banks from two different regions of Brazil (Southeast and Northeast). Ethical approval for the study was obtained from the Ethics Committee authority. For this investigation a healthy group constituted by male ($n = 150$) and female ($n = 133$) blood donors, ages between 18 and 65 years and above 50 kg, were selected. This selection involved inhabitants with no history of toxicological exposure, no consumption of cigarettes, non-drinkers (never consumed alcohol), and drinkers (low consumption estimated for each individual considering the daily intake).

To perform the donor's selection the serum samples from the volunteers were submitted to the routine tests described in detail by Oliveira et al. [2]. For sample preparation about 2.0 mL of whole blood was collected in a

L. C. Oliveira (✉) · C. B. Zamboni · S. Metairon
Instituto de Pesquisas Energéticas e Nucleares, IPEN –
CNEN/SP, Av. Prof. Lineu Prestes 2242- Cidade Universitária,
São Paulo, SP 05508-000, Brazil
e-mail: laura@ipen.br

vacuum plastic tube (without anticoagulant) attached to the donor's arm. Immediately after the collection, before the blood coagulation, 100 μL of whole blood was transferred to the Whatman filter paper ($\sim 2.5 \text{ cm}^2$) and dried for few minutes using an infrared lamp. The biological material still in the plastic tube was then centrifuged and the serum obtained was used to perform screening tests for transmitted diseases as Hepatitis B and C, AIDS, Syphilis and Chagas.

For this investigation, the whole blood samples were prepared in duplicate. Aliquots of standard solutions of Br, Cl, K and Na were shaped in a same manner as the biological sample.

The sample and standard were irradiated for 3 min in a pneumatic station in the nuclear reactor (IEA-R1, 2–4 MW, pool type) at IPEN in a thermal neutron flux of $3.3 \times 10^{12} \text{ n cm}^{-2} \text{ s}^{-1}$. After the irradiation, the activated materials were gamma-counted for 10 min using a high resolution HPGe Spectrometer (FWHM = 1.87 keV) connected to an ADCAM multichannel analyzer (ORTEC 919E) and a PC computer. The concentration for each element was obtained using in-house software. For analytical quality control, IAEA A-13 animal blood was used. The results obtained for these reference materials were in good agreement with their respective nominal values: the relative errors were lower than 8.1% and relative standard deviations were lower than 6.3%.

Results

The Tables 1 and 2 show the results for Br and Cl, and K and Na, respectively, as a function of the sex for both regions (Southeast and Northeast). The concentrations for

Br for male and female, as a function of geographic occupation, is showed in Fig. 1.

Discussion

The reference ranges derived from the two groups (regions) and sub groups (gender) show small differences. According to Tables 1 and 2, for adopted significance level ($p < 0.05$), there is evidences to discard the equality hypothesis in these regions as a function of sex: while lower values of potassium were found in females (1.33–1.71 g L^{-1} at SR and 1.16–1.80 g L^{-1} at NR) when compared to males (1.37–1.91 g L^{-1} at SR and 1.35–1.91 g L^{-1} at NR), lower values of Na in males were found (1.45–1.89 g L^{-1} at SR and 1.49–2.09 g L^{-1} at NR) when compared to females (1.56–1.92 g L^{-1} at SR and 1.52–2.10 g L^{-1} at NR). Yet, according to Tables 1 and 2 an increase of Br (see Fig. 1 as an illustrative visualization) and Na was observed in the Northeast region. It could be related to the fact that Recife is a coastal city suggesting that the local population has a greater intake of sea food (rich in Br and Na) but studies related to nutrition habits must be performed in order to subsidize this discussion. Particularly for Br, it is important to extend its evaluation to others groups, mainly those that take certain medicines (such as antidepressant and somniferous drug that are rich in bromides [6] and consumed in significant quantities by the Brazilian population) due its toxicological significance. Yet, considering that the Brazilian population legislation [7] does not foresee allowable limit for Br in industrialized, generic or manipulated drugs ours data suggest the necessity of Br levels evaluation mainly in antidepressant and somniferous. Recently, a study performed by Leal et al. [8] for determining chemical elements in drugs

Table 1 Blood concentrations of Br and Cl of inhabitants at Brazil (Southeast and Northeast) by sex

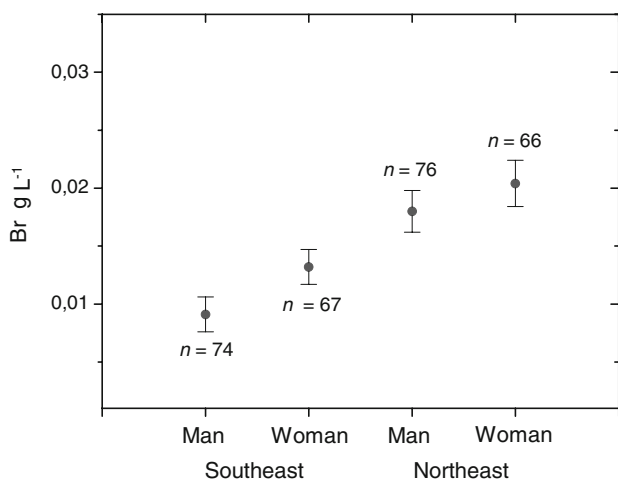
	Southeast MV \pm SD (range)	Northeast MV \pm SD (range)	Test <i>t</i>
Br, g L^{-1}			
Total	0.0102 \pm 0.0074 (0.0028–0.0176)	0.0183 \pm 0.0096 (0.0087–0.0279)	$t = 7.94 p < 0.05$
Man	0.0091 \pm 0.0075 (0.0016–0.0166)	0.0180 \pm 0.0091 (0.0089–0.0271)	$t = 6.53 p < 0.05$
Woman	0.0132 \pm 0.0065 (0.0067–0.0197)	0.0204 \pm 0.0121 (0.0083–0.0325)	$t = 4.28 p < 0.05$
Cl, g L^{-1}			
Total	2.99 \pm 0.44 (2.55–3.43)	3.03 \pm 0.50 (2.53–3.53)	$t = 0.71 p > 0.05$
Man	2.94 \pm 0.42 (2.52–3.36)	3.02 \pm 0.49 (2.53–3.51)	$t = 1.07 p > 0.05$
Woman	3.12 \pm 0.49 (2.63–3.61)	3.12 \pm 0.53 (2.59–3.65)	$t = 0 p > 0.05$

MV Mean value, SD Standard deviation

Table 2 Blood concentrations of K and Na of inhabitants at Brazil (Southeast and Northeast) by sex

	Southeast MV \pm SD (range)	Northeast MV \pm SD (range)	Test <i>t</i>
K, g L⁻¹			
Total	1.60 \pm 0.25 (1.35–1.85)	1.61 \pm 0.29 (1.32–1.90)	<i>t</i> = 0.31 <i>p</i> > 0.05
Man	1.64 \pm 0.27 (1.37–1.91)	1.63 \pm 0.28 (1.35–1.91)	<i>t</i> = 0.22 <i>p</i> > 0.05
Woman	1.52 \pm 0.19 (1.33–1.71)	1.48 \pm 0.32 (1.16–1.80)	<i>t</i> = 0.88 <i>p</i> > 0.05
Na, g L⁻¹			
Total	1.69 \pm 0.21 (1.48–1.90)	1.80 \pm 0.30 (1.50–2.10)	<i>t</i> = 3.57 <i>p</i> < 0.05
Man	1.67 \pm 0.22 (1.45–1.89)	1.79 \pm 0.30 (1.49–2.09)	<i>t</i> = 2.79 <i>p</i> < 0.05
Woman	1.74 \pm 0.18 (1.56–1.92)	1.84 \pm 0.32 (1.52–2.10)	<i>t</i> = 2.22 <i>p</i> < 0.05

MV Mean value, SD Standard deviation

**Fig. 1** Br concentration in blood as a function of sex and region

largely consumed by Brazilian population emphasizes the necessity of investigation of reference values in blood

in groups of subjects with dairy (chronic diseases) consumption of drugs.

More systematic studies are needed to establish reference values aiming its application, in the future, for studying in more details reference values of common deficiencies in Brazilian population helping their diagnostics.

Conclusions

The reference ranges for Br, Cl, K and Na in whole blood for the Brazilian population show small differences when a comparison is performed as a function of the individual characteristics (sex and geographic location). These data also provide scientific basis for more detailed research on common deficiencies in the Brazilian population, which could aid diagnoses and check the limits of exposure for public protection and occupational health. For Br, its toxicological significance suggests that it's important to extend its evaluation to other subgroups.

Considering that the determination of reference value of elements in human whole blood involves the analysis of hundreds of samples, the use of the NAA technique could be considered agile and precise.

Acknowledgments The authors thank the clinical staff at Blood Banks for technical assistance given during the blood collection and the financial support from CNPq and CAPES.

References

- Oliveira LC, Zamboni CB, Lins PS, Azevedo MRA (2005) *Braz J Phys* 35:793
- Zamboni CB, Oliveira LC, Mesa J (2006) *J Radioanal Nucl Chem* 269:541
- Kovacs L, Zamboni CB, Oliveira LC, Salvador VLR, Sato IM, Azevedo MRA (2008) *J Radioanal Nucl Chem* 278:543
- Oliveira LC, Kovacs L, Zamboni CB, Medeiros JAG, Azevedo MRA (2007) *Revista Brasileira de Hematologia e Hemoterapia* 29:607
- Santos NF, Vilela EC, Zamboni CB, Kovacs L (2006) *Fisioterapia Brasil, suppl. SPQV*, 42:42
- Canavese C, Constanzi ED, Stratta P (2006) *Am J Kidney* 48:1018
- ANVISA (National Health Surveillance Agency) (2006) <http://www.anvisa.gov.br>
- Leal AS, Menezes MABC, Andoine O, Vermaercke P, Sneyers L (2008) *Appl Radiat Isot* 66:1307