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Trace element contents in serum of healthy elderly population of metropolitan São Paulo area in Brazil

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Abstract

In this study, the elements Br, Ca, Cl, Fe, Na, Rb, Se and Zn were determined in serum of a healthy elderly population residing in the São Paulo Metropolitan area, using instrumental neutron activation analysis. Comparison studies indicated that only Se concentration was significantly lower in the elderly group aged 75–91 years than those obtained for the group aged 60–74 years. Between genders, males presented lower Br concentrations and, the females lower Fe. Mean concentrations obtained for most elements were within the values reported in the literature.

Keywords: Trace element; Blood serum; Neutron activation analysis; Elderly

Introduction

Trace element determinations in blood serum have become important to investigate their vital role in human metabolism, as well as to obtain information regarding the health status of individuals. Thus, trace elements in serum have been determined to see whether or not there is a correlation between trace element content and diseases.

The health of an individual is generally evaluated by interpretation of laboratory test results that requires normal or reference values. Although it is recognized that age and several other parameters influence the concentrations of certain elements in human organisms, the reference values are established on individuals of an adult population which usually does not include the elderly. So, the evaluation of trace element levels in serum samples of an elderly population is of particular interest.

The objective of this study was to evaluate the influence of gender and age in the serum element concentrations in addition to comparing the baseline data obtained for healthy elderly with those values published in the literature.

Materials and method

Sampling and sample preparation

The selection of individuals and the control of sample contaminations were performed according to

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the protocols established in our previous publication [1]. The population consisted of elderly subjects considered healthy and who attended at the Hospital das Clínicas of the São Paulo University Medical School. The participants are of a "Successful Ageing" program and the following exclusion parameters were used to select the individuals: alcoholism and smoking; hepatitis, human immunodeficiency virus, chronic disease, diabetes, hypertension, atypical dietary habits, blood transfusion within the previous 6 months, anemia, supplement intakes and mental disorders.

Blood samples were collected from 68 subjects (18 males and 50 females) aged 60–91 years, (mean age = 72 ± 7 years) using sterile metallic needles. The blood was collected in heparin-free BD Vacutainer™ tubes, royal blue Hemogard™ closure, without additive (Becton-Dickson and Company, USA) for trace element determinations. The blood was centrifuged after it completely clotted and the serum obtained was freeze - dried. The weight loss during this freeze-drying process was about 90.9%.

Analytical procedure

The analytical methodology used in this study was instrumental neutron activation analysis (INAA). Aliquots of about 180 mg of dried serum weighed in polyethylene involucres were irradiated in the IEA-R1 nuclear reactor along with the synthetic standards of the elements. In these standards the quantities of each element, in μg (in parentheses) were the following: Br (5.0), Ca (500.1), Cl (200.0), Fe (350), Na (100.0), Rb (10.0), Se (8.0) and Zn (35.0). Short irradiations of 15 s under a thermal neutron flux of 1.4×10^{12} n cm^{-2} s^{-1} were carried out for Cl and Na determinations. Samples and standards submitted to these short irradiations were measured after about 30 min of decay time. Longer irradiations of 16 h under a thermal neutron flux of about 5×10^{12} n cm^{-2} s^{-1} were performed for elements Br, Ca, Fe, Rb, Se and Zn determinations. Additional elements were not determined due to its low concentrations in the serum or due to the interference of high activities of ^{24}Na and ^{32}P. Samples and standards submitted to these long-period irradiations were measured twice, after about 4 and 10 days of decay times. The irradiated samples and standards were measured by an HGe detector coupled to a gamma ray spectrometer. The radioisotopes were identified according to their half-lives and gamma-ray energies and the element concentrations were calculated by comparative method.

To evaluate the precision and accuracy of the results, certified reference materials NIST 1566b oyster tissue

Table 1. Concentrations of elements in certified reference materials NIST 1566b oyster tissue and IAEA-A-13 animal blood

Elements mg kg^{-1}	NIST 1566b Oyster tissue				
	This work				Certified values [2]
	Mean ± SD (a)	RSD (b), %	Er (c), %	Zscore	
Br	50.9 ± 3.8	7.5	–	–	–
Ca	839 ± 37	0.4	0.1	0.02	838 ± 20
Cl	4999 ± 106	2.1	2.7	−0.97	5140 ± 100
Fe	196.4 ± 7,1	3.6	4.6	−0.97	205.8 ± 6.8
Na	3164 ± 146	4.6	4.0	−0.86	3297 ± 53
Rb	3.151 ± 0.132	4.2	3.4	−0.56	3.262 ± 0.145
Se	2.08 ± 0.02	0.1	0.1	0.13	2.06 ± 0.15
Zn	1373 ± 41	3.0	3,6	−0.83	1424 ± 46
Elements mg kg^{-1}	IAEA-A-13 Animal blood				
	This work				Recommended values [3]
	Mean ± SD	RSD, %	Er (c) %	Z score	
Br	22.23 ± 0.65	2.9	1.0	0.04	22 (19–24)
Ca	266 ± 27	10.1	7.0	−0.16	286 (226–332)
Cl	16297 ± 483	3.0	–	–	–
Fe	2261 ± 76	3.4	5.8	−0.24	2400 (2200–2500)
Na	12742 ± 117	0.9	1.2	0.07	12600 (11600–13500)
Rb	2.53 ± 0.16	6.3	10.0	0.20	2.3 (1.7–3.1)
Se	0.246 ± 0.025	10.2	2.5	0.06	0.24 (0.15–0.31)
Zn	13.2 ± 0.4	3.0	1.5	0.07	13 (12–14)

(a) Mean ± SD = Arithmetic mean and standard deviation; (b) RSD = Relative standard deviation; (c) Er = Relative error.

and IAEA-A-13 animal blood were analyzed. Data obtained in these analyses presented in Table 1 indicate good precision and good agreement with the certified values. The relative standard deviations of the results were lower than 10% and relative errors varied from 0.1 to 10%. The standardized difference or Zscore values [4] obtained for the elements analyzed were $|Zscore| < 1$, indicating that our results are satisfactory and are within the ranges of certified data at the 95% confidence interval level.

Results and discussion

Comparison made between the results obtained in sera from male and of female groups indicated significant differences only for Br and Fe ($p = 0.05$). Bromine mean concentrations of $2.89 \pm 0.90\,mg\,L^{-1}$ and $3.66 \pm 0.75\,mg\,L^{-1}$ were obtained for male and female, respectively. Fe concentrations were higher for males ($1.35 \pm 0.36\,mg\,L^{-1}$) than those found for females ($1.11 \pm 0.27\,mg\,L^{-1}$). The influence of gender in the levels of Br and Fe may be attributed to the differences in physiological factors and metabolic pathways [5,6]. Differences in Fe concentrations between female and male sera have also been reported [7–9].

A comparative study based on two different age groups of healthy elderly population was also carried out. Results obtained for these two groups indicated that only Se concentrations from elderly group aged 60–74 years ($77.2 \pm 23.3\,\mu g\,L^{-1}$) are significantly higher than those found for the group of 75–91 years ($66.0 \pm 13.3\,\mu g\,L^{-1}$) ($p = 0.05$). Savarino et al. [10] also observed a decrease in Se concentrations in serum from a group aged 91–110 years when compared with those results obtained for an elderly group aged 60–90 years.

Table 2 shows the mean concentrations of trace elements determined in sera from a healthy elderly population together with the values used in clinical laboratories and those presented in literature. Published

Table 2. Concentration of trace elements in serum of healthy elderly group population of the São Paulo Metropolitan region

Elements	This work Mean ± SD	Range	Values used in clinical labs [7]	Literature values (remarks)
Br	43.4 ± 9.5	26.7–69.3	100–187	• 69.2 ± 8.39 (Mean age = 65.6; $n = 27$; PIXE) [11]
Ca	2.39 ± 0.20	1.92–2.98	2.20–2.55	• Range 2.20–2.60 (80 years old; $n = 211$) [12] • 2.25 ± 0.23 (W, 60's and over; $n = 17$; Korean; INAA) [13]
Cl	87.4 ± 5.4	71.7–103.0	98–108	• 99.2 ± 6.7 (W, 60's and over; $n = 17$; Korean; INAA) [13]
Fe	21.0 ± 5.4	10.8–39.7	11.6–31.3 (M) 9.0–30.4 (W)	• 21.5 ± 8.1 (Mean age = 65.6; $n = 27$; PIXE) [11] • Range 13–35 (M; $n = 211$); 10–28 (W; $n = 211$) [12] • 20.8 ± 8.4 (W, 60's and over; $n = 17$; Korean; INAA) [13]
Na	132.0 ± 8.7	113.7–158.8	136–145	• 138.7 ± 11.9 (W, 60's and over; $n = 17$; Korean; INAA) [13]
Rb	3.9 ± 0.7	2.3–5.4	0.9–6.5	• 1.80(1.00–2.60) (M; $n = 62$; Central Bohemia/ Czech; INAA) [14] • 1.2 ± 0.4 (Mean age = 65.6; $n = 27$; PIXE) [11]
Se	0.92 ± 0.26	0.51–2.35	0.58–1.81	• 1.14 ± 0.22 (Age 60–90; $n = 62$; ETAAS) [10] • 0.871 (0.623–1.119) (M; $n = 348$; Prague and Central Bohemia; ETAAS) [14] • 0.947 (0.749–1.145) (W; $n = 249$; Central Bohemia; ETAAS) [14]
Zn	14.9 ± 2.2	10.2–21.4	10.7–18.4	• 15.6 ± 2.6 (Mean age = 65.6; $n = 27$; PIXE) [11] • 14.51 ± 2.07 (M; $n = 62$; FAAS) [10] • 13.48(9.23–17.73) (W; $n = 118$; Central Bohemia /Czech) [14]

Units for Ca, Cl and Na are $mmol\,L^{-1}$ and, for the others are $\mu mol\,L^{-1}$. Number of samples analyzed was 68 for all elements excepting Fe. Five samples excluded for Fe determinations due to the hemolysis during serum preparationAbbreviations used: M = men; W = women; PIXE = Particle induced x-ray emission; INAA = Instrumental neutron activation Analysis; FAAS = Flame atomic absorption spectrometry; ETAAS = Electrothermal AAS.

data of serum analysis for elderly groups are very scarce. An evaluation of the obtained data indicated that the mean values found are within the values used in clinical laboratories or previous published data, with the exception of Br result. The mean value of $43.4 \pm 9\,\mu mol\,L^{-1}$ obtained for Br is slightly lower than reported values of $69.2 \pm 8.3\,\mu mol\,L^{-1}$ [11] and $79.6 \pm 14.8\,\mu mol\,L^{-1}$ [13].

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