Evaluation of some essential and trace elements in diets from 3 nurseries from Juiz de Fora, M.G., Brazil, by neutron activation analysis

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A study was made in diets offered to a group of pre-school children, whose mean age was 67 months and remained the whole day in three day care centers from Juiz de Fora, M.G., Brazil. For sampling, the duplicate portion technique was used, and the diets were collected and analyzed separately each day in the 3 nurseries. Instrumental neutron activation analysis was applied to the determination of 16 elements. The daily dietary intake values were compared to the RDA (children 4–6 years old). Based on this reference, Ca, Fe, Se and Zn were found to be deficient, Mg and Mn were comparable to the RDA and the Cl and Na concentrations were higher compared to their RDA. For the other elements measured, there are no RDA's for children.

Introduction

Minerals and trace elements are very important components in diets. Both their deficiency and excess may cause serious health problems. The effects of trace element deficiency are most severe during development and growth and they are especially important for infants and children.

Recent publications have called attention to the fact that the inadequate ingestion of micronutrients is still a world-wide public health issue, affecting mainly children under age six, elderly people and nursing children.^{1–3} BLUM¹ emphasizes that although many efforts have been made to reduce poverty, improve nutritional education and to offer to the world population safer foodstuffs, over 2 billion people are ill or disabled and millions die prematurely each year as a result of macronutrient deficiencies.

Vitamin A, iron and iodine are three nutrients that are frequently noted to be deficient in specific populations. Other vitamins and minerals such as calcium, zinc and selenium, when marginally deficient have also caused serious health problems world-wide. On the other hand, it is well known that these essential elements cannot be considered individually, since to maintain a good health status, it is necessary to ensure adequate ingestion of all nutrients.

Moreover, several recent discoveries, and numerous refinements in analytical techniques, have substantially increased the knowledge of the role of trace elements in human health.⁴

As the diet is generally the single most important source of all elements for human beings in a natural environment, it is important to know the composition of the major, minor and trace element constituents in a subject's diet. Analyses of actual diets have been undertaken in many countries since the 80's and are still being carried out, in order to verify the intake data for essential and toxic elements.

In the last few years in Brazil, neutron activation analysis, because of its high precision, accuracy and sensitivity, has been used for this purpose in several studies, $^{5-8}$ at the Radiochemistry Division of IPEN-CNEN/SP.

In the present work, the macronutrient, mineral and trace element contents have been determined in diets consumed by pre-school children of three nurseries from the city of Juiz de Fora, MG, Brazil. This is an important contribution to the knowledge of the composition of these diets, so that actions can be taken in order to correct deficiencies in the future and try to improve the diets with the aim of a rehabilitation in the development of these children.

Experimental

Sample collection and preparation

The diets offered to the children have been collected during a period of one week using the duplicate portion method.

After collection, the foodstuffs were transferred to polyethylene flasks, previously demineralized in a 30% nitric acid bath, for at least 12 hours and then rinsed in deionized water, in order to minimize the elemental contamination.

The foodstuffs of each meal collected were weighed separately and then stored at -20 °C. Afterwards, the

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diets were transferred to a container in ice to the Laboratory of Experimental Nutrition of the University of São Paulo, where they were left to attain room temperature and were then mixed over stainless steel trays covered with Teflon film. The foodstuffs were dried in a ventilated oven at 60 °C and weighed, powdered and homogenized in a mill.

After this step, each sample was divided in two portions. One of the portions was retained at the Laboratory of Experimental Nutrition for analysis of macronutrients and the other was transported to the Radiochemistry Division of IPEN/CNEN-SP, for analysis of mineral and trace elements.

Determination of proximate composition

The diets were analyzed for their moisture content at 105 °C, fixed mineral residue (550 °C), protein (micro Kjeldahl method) and ether extract (Soxhlet method), according to the methodology of the AOAC (1990).⁹ The Nifext fraction or nitrogen free extract was obtained by difference. The total caloric content of each diet was obtained by utilizing the values of 4 kcal/g of protein and carbohydrate and 9 kcal/g of fat.

Methodology

Instrumental neutron activation analysis: Instrumental neutron activation analysis was used in this work to determine quantitatively the minor and trace elements in diet samples.

The individual or multielemental solutions were used as primary element standards. The synthetic standard preparation was described in a previous paper.⁶

Irradiation and counting: Aliquots of about 200 mg of diet samples and reference material Oyster Tissue (NIST SRM 1566^a) (weighed in pre-cleaned polyethylene bags) with the synthetic standard were placed into polyethylene or aluminum containers, and irradiated in the IEA-R1 research reactor. Short and long irradiations were carried out depending on the half-life of the induced radionuclides.

For determination of the short lived radioisotopes: $^{38}\text{Cl},~^{27}\text{Mg},~^{56}\text{Mn},~^{24}\text{Na}$ and ^{42}K , the samples and standard were irradiated for 2 minutes in a thermal neutron flux of $10^{11}~\text{n}\cdot\text{cm}^{-2}\cdot\text{s}^{-1}$.

For long irradiations, the samples and standards were irradiated for 8 hours under a thermal neutron flux of 10^{12} – 10^{13} n·cm^{-2·s⁻¹}. After different cooling times, the following radioisotopes were determined: ⁸²Br, ⁴⁷Ca, ⁶⁰Co, ⁵¹Cr, ⁵⁹Fe, ⁸⁶Rb, ⁷⁵Se, ⁴⁶Sc and ⁶⁵Zn. Two series of countings were made: the first, after a one week decay and the second, after 15–20 days. The counting time was about 2 hours for each sample and reference material and

half an hour for each synthetic standard in the first counting. In the second one, the counting time of 50,000 seconds for samples and reference materials and again half an hour for each synthetic standard were used.

The gamma-ray spectra were obtained in a counting system with an Ortec EG&G high resolution solid state Ge detector, POP TOP, Model 20190 with resolution of 1.9 keV for 1332 keV γ -ray peak of ⁶⁰Co. This detector was coupled to an EG&G Ortec ACE8K card and associated electronics. Spectrum analysis was performed using the VISPECT2 software¹⁰ to locate the gamma-ray peaks and ESPECTRO program to calculate the concentrations. The last program was developed at the Radiochemistry Division/IPEN.

Validation of the methodology: The reference material Oyster Tissue (NIST SRM 1566^a) was used for checking the precision and accuracy of the method.

Results and discussion

Table 1 presents the results of the analysis of the macronutrient contents in the diets consumed by the children of the three nurseries under study.

The children, in general, consumed 4 meals in the nursery: breakfast, lunch, snack and dinner. The most common food items consumed were: milk with chocolate, bread or biscuit, rice, beans, corn porridge, meat or chicken. The vegetables consumed were: beats, tomatoes, potatoes, zucchini and sweet potatoes. The only leafy vegetable consumed by the children was cabbage. The main fruits offered were: apples, bananas and oranges.

As can be observed in Table 1, the diets of the nurseries Benfica and Vila Ideal surpassed the recommendation of protein for the age range studied, although the caloric percentage adequacy for this nutrient was in the range considered as adequate (10 to 15% of the total caloric value) (RDA, 1989).¹¹

The lipid contents of the Nurseries Benfica and Santa Luzia contributed with low caloric percentages, which resulted in a caloric percentage adequacy below the recommended value of 30%.

As to the carbohydrates, it can be observed that the presented values can be considered as inadequate, following the current guidance of recommendation that the calories from these nutrients should complete the total caloric value of the diet.

The total caloric value of the diets of the three nurseries studied can be considered as quite low, especially for the nurseries Benfica and Santa Luzia. The average caloric intake from the diets of the three nurseries was far below the 1800 kcal/day recommendation for children of the age range studied (RDA, 1989).¹¹

	Benfica	Nurseries Vila Ideal	Santa Luzia	
Food consumption, g/day, wet weight	858.1 ± 151.0	944.3 ± 52.6	812.2 ± 82.5	
Food consumption, g/day, dry weight	204.6 ± 27.6	250.2 ± 20.3	197.7 ± 21.9	
Protein, g/day TCV, %	31.7 ± 9.2 15.1	37.9 ± 3.6 14.0	24.8 ± 3.2 12.5	
Lipids, g/day TCV, %	19.8 ± 7.6 21.5	$\begin{array}{r} 34.7 \pm 3.9 \\ 28.8 \end{array}$	15.9 ± 3.7 18.2	
Carbohydrate, g/day TCV, %	$130.6 \pm 19.2 \\ 63.4$	155.1 ± 14.9 57.2	$137.7 \pm 17.9 \\ 69.3$	
Energy, kcal	827.8 ± 106.9	1084.2 ± 86.5	793.6 ± 69.1	

 Table 1. Food consumption, average nutrient ingestion, total caloric value and percentile of adequacy of the macronutrient and total caloric value of the diets consumed by children of three nurseries of the city of Juiz de Fora, MG, Brazil

RDA values:¹¹ Children from 4 to 6 years = 24 g protein/day; 1800 kcal/day.

Protein = 10 to 15% of the total caloric value.

Lipids = until 30% of the total caloric value.

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Carbohydrate = rest of the total caloric value.

TCV, % = % in relation to the total caloric value.

Element	Benfica Nursery		Santa Luzia Nursery		Vila Ideal Nursery	
	Range	Mean \pm sd	Range	Mean \pm sd	Range	Mean \pm sd
Ba	3.5 - 5.7	5 ± 1	4.5 - 6.3	5 ± 1	6.9 - 12	10 ± 2
Br	5.0 - 9.4	6.7 ± 1.9	5.6 - 8.9	7.5 ± 1.6	6.3 - 8.4	7.5 ± 0.9
Ca	1184 - 1691	1513 ± 202	455 - 1273	815 ± 372	1364 - 2129	1819 ± 333
Cl	9776 - 15024	13382 ± 2097	12838 - 17544	15035 ± 1772	12174 - 15863	13339 ± 1483
Co, ng/g	8.5 - 44	21 ± 14	17.9 – 41	29 ± 9	38 - 54	44 ± 6
Cr	0.1 - 0.23	0.2 ± 0.1	0.1 - 0.93	0.5 ± 0.4	0.11 - 0.44	0.3 ± 0.1
Cs, ng/g	39 - 109	73 ± 27	36 - 86	56 ± 21	54 - 58	56 ± 3
Fe	14.7 – 29	25 ± 6	23.7 - 42	36 ± 7	21 - 43	33 ± 9
K	4865 - 6867	6298 ± 834	5051 - 6172	5270 ± 644	4408 - 6603	5375 ± 811
Mg	473 - 782	574 ± 123	398 - 926	595 ± 196	446 - 539	$486~\pm~46$
Mn	4.6 - 11.2	6.7 ± 2.7	5.5 - 10.3	8 ± 2	4.8 - 6.5	5.5 ± 0.7
Na	5925 – 9927	8532 ± 1537	10020 - 11525	10578 ± 604	7729 – 8548	8558 ± 900
Rb	6.9 - 11.1	9.4 ± 1.7	6.9 - 8.1	7 ± 2	11.8 - 17	14 ± 2
Sc, ng/g	0.6 - 1.2	1.0 ± 0.3	1.0 - 2.7	1.8 ± 0.8	1.3 - 2.4	1.9 ± 0.4
Se, ng/g	39 - 56	44 ± 7	55 - 94	67 ± 32	52 - 89	71 ± 14
Zn	14.8 - 34	24 ± 9	15.9 – 29	22 ± 6	19.0 - 30	24 ± 4

Table 2. Results (in mg·kg⁻¹) of diet samples analyzed by INAA (dry weight)

Figure 1 shows the results obtained in the Oyster Tissue reference material analysis by INAA according to the *z*-score criteria. The calculation of the standardized difference or *z*-value was made according to BODE.¹²

If |z|<3 means that the individual result of the control sample (reference material) should be in the 99% confidence interval of the target value.

	S.	S. Luzia		V. Ideal		Benfica	
Element	Average	Range	Average	Range	Average	Range	
Ba. µg	970	797 – 1429	2550	1712 - 3143	973	674 – 1197	
Br, mg	1.47	1.04 - 1.79	1.87	1.5 - 2.2	1.38	0.9 - 2.1	
Ca, mg	162	84.4 - 263	458.1	322 - 596	311	228 - 391	800 ^a
Cl, mg	2947	2787 - 3219	3329	2944 - 3795	2742	1883 - 3480	500-600 ^c
Co, µg	5.8	3.2 - 8.8	10.9	9.4 - 13.4	4.3	1.4 - 8.5	
Cr, µg	105	25 - 194	79	26 - 105	32	19 - 45	30-120 ^b
Cs, µg	11.0	7.8 - 16.0	13.3	12.8 - 13.9	15.2	7.9 - 26.1	
Fe, mg	7.0	5.4 - 8.2	8.3	5.0 - 12.3	5.0	3.3 - 6.7	10 ^a
K, mg	1045	836 - 1292	1336	1199 - 1580	1279	1079 – 1499	1400-1600 ^c
Mg, mg	119	86 - 210	122.0	107 - 152	117	86 - 151	120
Mn, mg	1.6	1.1 - 2.3	1.4	1.2 - 1.6	1.4	0.9 - 2.3	$1.5 - 2.0^{b}$
Na, mg	2083	1925 - 2274	2137	1869 - 2410	1746	1142 - 2212	300–400 ^c
Rb, mg	1.38	0.64 - 1.84	3.4	2.9 - 4.1	1.9	1.4 - 2.6	
Sc, ng	345	227 - 495	474	314 - 567	196	133 - 270	
Se, µg	12.9	5.6 - 17.2	17.9	12.4 - 21.0	9.1	7.0 - 13.4	20 ^a
Zn, mg	4.4	2.9 - 6.0	6.0	4.5 - 7.3	5.0	3.3 - 7.7	10 ^a

Table 3. Daily intakes of the essential elements analyzed in the diet samples and RI)A ¹¹
Recommendations	

^a Recommended dietary allowance.

^b Estimated safe and adequate daily dietary intakes.

^c Estimated minimum requirements for children from 2 to 9 years.



Fig. 1. Control chart (z-values) for inspection of the normalized concentrations of some elements in Oyster Tissue reference material sample (NIST SRM 1566^a)

Table 2 shows the means and standard deviations, the range values for each essential and trace element analyzed by INAA. The daily intakes of elements were obtained by multiplying the concentrations in the diet samples by the total weight of food consumed per day. The amount of food consumed daily in the nurseries varied from 167.07 to 239.33 g/person, from 177.43 to 226.90 g/person and from 236.23 to 285.77 g/person for Benfica, Santa Luzia and Vila Ideal nurseries, respectively. The average daily intake of essential and trace elements are presented in Table 3.

From Table 3 it can be observed that the diets of children show deficiency for the essential elements Ca, Fe, Se and Zn, when their daily intake are compared with recommended values of RDA.¹¹

The Ca, Fe and Zn deficiencies in children can impair their growth and development. Studies have also shown that the cognitive development and immune system can be impaired by Zn and Fe deficiency. The iron deficiency also is considered a problem of public health in Brazil, because of its low intake in the diet and the low bioavailability, so anaemia is common in the children populations.

Concerning the other elements such as Ba, Br, Co, Cs, Rb and Sc there are no recommendations for children, but their determination is an important contribution for the knowledge of level of these trace elements in the children's diets.

Conclusions

The results obtained show that the diets of the three nurseries are not compatible with the children's needs, presenting an energetic value far below the recommended levels. The adequacy percentage was around 46%, 60% and 44% for the nurseries Benfica, Vila Ideal and Santa Luzia, respectively. Therefore, these children have significant dietary deficiencies, including deficiencies in elemental nutrients.

When we analyze the macronutrients distribution in these diets we can observe that what is really missing is an increase of food intake as a whole. For example, the lipids are far below the recommendation, contributing to the low energetic values in that each lipid gram supplies 9 kcal which is twice as much as any other diet source. The results obtained for the essential elements Ca, Fe, Se and Zn are far below the recommended values. The high intake of Na and Cl, on the other hand, may favor the Ca elimination. Cr, K, Mg and Mn intakes were in good agreement with the recommendations.

The consequences of these deficiencies are already well known, affecting the children's development as a whole, mainly for the immune system which will bring infections more often, contributing for more morbidity and mortality. In addition, this will also affect mental and intellectual development of the children.

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