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Intake of iron, zinc and iodine in 28 Ethiopian children living in Wonji Shoa Sugar Estate, assessed by duplicate portion technique

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In the present study, duplicate diets (four-day survey) of 28 Ethiopian children were analysed for iron, zinc and iodine. The mothers of the children filled out a food frequency questionnaire describing general intake of local food ingredients. The average daily intake of zinc was found to be 5.7 ± 5.2 mg, while the daily intake of iron and iodine was 52 ± 43 mg and 0.13 ± 0.10 mg, respectively. Most of the investigated children had an adequate nutrient intake of zinc and iodine, while the intake of iron was high compared to international recommendations. There was a great individual variation in intake of these trace elements, and even though the mean and median values were above or in the range of the requirement, some children had a too low intake of zinc and iodine. The iron intake varied the most, and was generally high. The diet was cereal based and contained little fruit and food of animal origin which may reduce the availability of iron and zinc.

Key words: Trace elements, nutrition, low-income, Africa, enjera, teff, cereals, iron, zinc, iodine intake, Ethiopian children.

INTRODUCTION

It was until the early 1990's that micronutrient deficiencies, referred to as the "hidden hunger", was given worldwide attention (FAO/WHO, 1992). Special emphasis was put on iodine-, iron- and vitamin A deficiencies. Iron deficiency is the most pervasive nutritional problem in the world (Yip, 2001) and is associated with a variety of non-haematological symptoms affecting learning ability, growth, immunity, and work capacity (Cook and Skinke, 1989). Iodine deficiency is a major public health problem in Ethiopia (Abuye and Berhane, 2007). It is the primary cause of preventable mental retardation and brain damage, and increase the chance of infant mortality, miscarriage, and stillbirth (Semba, 2001).

The detrimental effects of zinc deficiency on the infant child, and the maternal health were not recognised by the United Nations until 1997. Zinc was then included among the micronutrient deficiencies listed as a priority in the Third Report on the World Nutrition Situation (ACC/SCN, 1997). Iron and zinc deficiencies are common in children from developing countries due to absence of animal food, a high dietary content of phytic acid and tannins, as well as an inadequate food intake (Bhatnagar and Natchu, 2004; Umeta et al., 2005). In infants, both the quantity and type of food, and the frequency of feeding are important factors related e.g. to stunting (Umeta et al., 2003). The iron and zinc contents of food vary greatly from sample to sample, reflecting a variety of factors, such as differences in soil and climatic conditions. Thus, the use of food composition tables across ecological zones and between regions might threaten the validity and reliability of a database (Barikmo et al., 2007). Local data are important in order to do nutritional assessments,

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plan and implement food and/or supplements, and to be able to plan specific nutrition and educational programs.

In Ethiopia, there are data on iron and zinc both in the Food Composition Table for use in Ethiopia (EHNRI, 1998), and in recent published reports (Umeta et al., 2005; Abebe et al., 2007; Nurfeta et al., 2008). Contrary to zinc and iron, there are few reports on iodine content of Ethiopian food. lodine is not included in the Composition Table for use in Ethiopia (EHNRI, 1998). Fish and other marine foods are regarded as the most important natural sources of dietary iodine (Clugston and Hetzel, 1993). In Ethiopia, fresh water fish from rivers and inland lakes contributes to the iodine intake and may be of importance (Eckhoff and Maage, 1997). Also, water from the local lakes (Lake Abase and Lake Zwai) contained significant concentrations of iodine (Eckhoff and Maage, 1997). In the present study, iron, zinc and iodine intake were assessed in Ethiopian children using the duplicate diet approach. Of all the dietary survey methods, this method is regarded as the most accurate (Margetts and Nelson, 1997). When analysing a duplicate diet, the nutritional content of the diet reflects variables such as the chemical composition of the ingredients, the water used for preparation, contamination from the environment, and leach from cooking utensils. Thus, this technique gives valuable additional information to analyses of ingredients especially in communities where these factors may be of major importance. The method has also the advantage of being independent of errors associated with the use of food composition tables (Garrow et al., 2000) and it is not necessary to have access to food composition table. A disadvantage by use of the method is that subjects may not have the extra food required to make a second portion, resulting in a smaller duplicate portion, or that expensive food ingredients (e.g meat or oil) is not included in the duplicate portion (Garrow et al., 2000).

This survey was part of the project "Fluoride in Food and Water: a Health Problem in East Africa" funded by the Norwegian Universities' Committee for Research and Education (NUFU). The present study, which assesses the nutritional intake of the micronutrients iron, zinc and iodine in Ethiopian children by use of the duplicate portion technique, is the third paper published from this food survey.

SUBJECTS, MATERIALS AND METHODS

Study area and population

The material needed for the study was collected during four consecutive days in November 1997 (from Sunday 17th to Wednesday 20th), at the Wonji Shoa Sugar Estate (WSSE). The WSSE is located in a rural area in the Ethiopian Rift Valley (110 km South-East of Addis Ababa), bordering the Awash River. Thirty families were selected from two factory villages, labelled A and K. The criteria for inclusion in the project were the presence of at least one child in the household, fully weaned and below the age of five.

Only one child per household was studied.

The families who volunteered to participate in the survey were compensated with a sack of teff, which is the locally preferred staple food. Information concerning the subject, material and method part has been described previously (Malde et al., 2003; Malde et al., 2004). Wonji Shoa Sugar Estate is run by a state-owned company, the Wonji-Shoa Sugar Factory (WSSF). The estate includes two factory villages and 14 plantation villages. The total population is approximately 30,000 people. As WSSF provides free residential facilities including housing, water supply and electricity, schools, and free medical service to its employees and their immediate families, the standard of living is more or less the same in all the families involved in the study.

Ethical considerations

Ethical approval for this study was obtained from Health and Public authorities (Oromia Region, Ethiopia). Informed consent was obtained from all families participating in the study. The procedures followed were in accordance with the ethical standards of the responsible committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 1983.

Food survey

A duplicate portion technique was used for the collection of food samples for element analysis, that is, the mothers prepared a second portion of food and beverages, identical in content and volume to the portion taken by the child. The duplicate portions were stored in plastic containers with cover, the number of containers corresponding to the number of meals and the composition of the diet during the 24-h period. E.g. sauce and enjera were stored separately and were weighed separately. After weighing (precision of scale 0.1 g) at site, all the food were mixed in one plastic container with cover, and brought to the local hospital for homogenisation. Containers with cover were used in order to avoid contamination of the samples during storage and transport. After homogenisation, approximately 50 g of each 24-h duplicate portion was transferred to plastic containers, dried in an oven (50ºC) and thereafter, kept in a refrigerator. After one week, the samples were transported to the National Institute of Nutrition and Seafood Research (NIFES), Bergen, Norway. Twenty-four-hour duplicate portions were collected over four consecutive days (Sunday through Wednesday), including one of the fasting days of the Orthodox Church.

A native nurse from the local hospital instructed the mothers in the sampling procedure. On the first day of the survey, all families were visited to ensure correct sampling. The mothers gave a description of the ingredients used for preparing the food, and explained which food was given to the different meals during the 24-h period. Once, during the study period, the mothers provided relevant background information concerning the general use of various food ingredients through answering a food frequency questionnaire. The questionnaire listed 44 food ingredients and beverages, and the frequency alternatives were; "more than once a day", "once a day", "3 - 6 times per week", "1 - 2 times per week", "1 timer per month or less" and "never". A local nurse filled out the questionnaire based on a face-to-face interview of the mother. The same nurse completed all the interviews.

Nutrient analysis of food

Prior to the element analyses, the food samples were freeze dried, pulverized and stored in closed bottles until analyses. For the

Age of children	n	Mean	Median	SD	Minimum	Maximum
(Months)						
12 - 24	1	66.5	-	-	-	-
25 - 36	11	67.4	48.1	64.2	24.1	255
37 - 48	13	47.3	45.4	12.8	30.7	71.2
49 - 59	3	99.4	101	3.4	95.4	102
Total	28	61.5	51.6	43.3	24.1	255

Table 1. Age, mean and median iron intake, standard deviation (SD) and range of duplicate portions sampled on four consecutive days (N=28).

Table 2. Age, mean and median food intake, standard deviation (SD) and range of duplicate portions sampled on four consecutive days (N=28).

Age of children	n	Mean	Median	SD	Minimum	Maximum		
(Months)	Food intake (g)							
1 - 24	1	529	-	-	-	-		
25 - 36	11	447	434	102	434	315		
37 - 48	13	422	375	118	285	694		
49 - 59	3	550	512	180	392	746		
Total	28	450	430	119	285	746		

determination of iron and zinc, sub-samples (approximately 0.2 g) were submitted to microwave-assisted wet digestion using 2.0 ml of concentrated nitric acid (Merck, Darmstadt, Germany) and 0.50 ml of 30% w/w H_2O_2 (Merck) in an Ethos Pro microwave system (Milestone, Holger Teknologi, Oslo, Norway) (Julshamn et al., 2000). An atomic absorption spectrometer (Perkin Elmer 3300 AAS Norwalk CT, USA) was used for the determination of iron and zinc. The instrument was equipped with an autosampler and specific hallow cathode lamps. The concentration of iron and zinc were calculated using a four point external calibration procedure.

The trueness and precision were assessed by concomitant analysis of standard reference materials (SRM) as well as participation in proficiency tests. The methods used are both accredited by the Norwegian Metrology and Accreditation Service. The results from the determinations of iron and zinc are presented as mean value of duplicate determinations of each sample. An ICP-MS (Perkin-Elmer ELAN 5000, SCIEX, Toronto, Canada) equipped with standard configuration was used as an iodine-specific detector for the determination of the iodine concentration of the samples. The sample solutions were pumped by a peristaltic pump from tubes arranged on a Perkin-Elmer AS-90 autosampler and aspirated into the argon plasma. The instrument was run at normal resolution and set to detect the iodine signal intensity at m/z 127 (Julshamn et al., 2001). Prior to the ICP-MS determinations, subsamples of 0.1 - 0.5 g (dry mass) were alkaline extracted using TMAH.

The method of standard addition calibration was used for quantification of iodine (Julshamn et al., 2001). The moisture content of the food samples was determined by drying the samples to constant weight: first by drying for 24-h (50° C) at the laboratory in Ethiopia, and thereafter, to constant weight in Norway (104° C) (NKML, 2002).

Data analysis

Descriptive statistics were performed by the use of STATISTICA version 9 (Statsoft Norway, Oslo, Norway).

RESULTS

The daily iron intake of the children studied varied, and the mean and median iron intake was high compared to the iron requirement of 8 mg (Nordic Council of Ministers, 2004) for this age group. Also, the children with the lowest iron intake were above the requirement (Table 1). The highest intake was registered in the oldest age group (49 - 59 months). This was also the group with the highest food intake (Table 2). The mean and median intake of zinc was in accordance with the average individual normative requirement of zinc (5.3 - 6.4 mg) from diets with low zinc availability (FAO/IAEA/WHO, 1996). However, the children being in the lower range had a zinc intake below the requirement (Table 3). Also for iodine, some children had a lower intake than the requirements of 0.09 mg iodine (FAO/IAEA/WHO, 1996).

The mean and median iodine intake was slightly above the recommended value except for the age group of 37 -48 months were the median value was 0.06 mg (Table 4). The duplicate diets sampled during the four days showed little variation in food ingredients. Food made from cereals was most commonly used. The locally grown teff was the only cereal used daily by all 28 families. Approximately, 40% (in grams) of the food sampled consisted of enjera, fried enjera pieces, or bread made from teff. The children ate little fruit or food of animal origin during the period of the food survey. Only three children were given fruit or meat, while one child was given fish at least once during the four day food survey.

The results from the food frequency questionnaire substantiate the lack of variation in food ingredients

Age of children	n	Mean	Median	SD	Minimum	Maximum		
(Months)	Zinc intake (mg)							
12 - 24	1	4.4	-	-	-	-		
25 - 36	11	8.5	5.8	7.3	3.2	23.4		
37 - 48	13	6.9	6.7	3.8	3.0	17.7		
49 - 59	3	6.2	5.0	2.2	4.8	8.8		
Total	28	7.3	5.7	5.2	3.0	23.4		

Table 3. Age, mean and median zinc intake, standard deviation (SD) and range of duplicate portions sampled on four consecutive days (N=28).

Table 4. Age, mean and median iodine intake, standard deviation (SD) and range of duplicate portions sampled on four consecutive days (N=28).

Age of children	n	Mean	Median	SD	Minimum	Maximum		
(Months)	lodine intake (mg)							
12 - 24	1	0.22	-	-	-	-		
25 - 36	11	0.16	0.14	0.09	0.04	0.31		
37 - 48	13	0.13	0.06	0.12	0.03	0.36		
49 - 59	3	0.15	0.16	0.03	0.12	0.17		
Total	28	0.15	0.13	0.10	0.03	0.34		

(Table 5). Beef, fish and chicken were rarely used (once per month or less) by most of the families, whereas onion, teff and beans were reported to be consumed daily by most of the families. Salt (NaCl) was used daily by all families. Twenty-three of the mothers answered that the season had little influence on the choice of food ingredients. The children were usually given the same food as the rest of the family, but with an extra meal during the day. Twenty-five of the children were served four meals daily, while two children were served three meals daily (one family did not answer).

Dietary habits and dietary intake of energy, fluoride, calcium and magnesium is previously reported (Malde et al., 2003; Malde et al., 2004). In the previous reports, the children were divided into two groups (Village A versus Village K) depending on the fluoride content of the drinking water. In the present report, the children are regarded one group.

Discussion

The present study shows that most of the investigated children had an adequate nutrient intake of zinc and iodine (Tables 3 and 4), while the intake of iron (Table 1) was high compared to international recommendations. There was a great individual variation in intake of these trace elements, and even though the mean and median values were above or in the range of the requirement, some children had a too low intake of zinc and iodine. The iron intake varied the most, and was generally high (Table 1). Teff, which is a staple food in this area, is known to have a high mineral content which varies with the quality of the teff flour.

According to the Ethiopian Food Composition Table (EHNRI, 1998), red teff flour contains 150 mg Fe/100 a while the more refined white version contain 23 mg Fe/100 g. A similar tendency has been reported for the iodine content: red teff containing seven times the iodine concentration in white teff (FAO, 1987). The difference in iodine content was, however, not confirmed by Eckhoff and Maage (1997) who found 70 and 72 µg iodine/kg d.w. in white and red teff, respectively. Selected food ingredients of fish and cereals, as well as samples of drinking water from the Ethiopian Rift Valley (Awasa) have been shown to contain detectable amounts of iodine (Eckhoff and Maage, 1997). The iodine concentrations reported for cereals like teff and maize can not alone explain the high iodine intake found in the present study. Salt, which was reported to be used once a day or more by all 28 households, may add to the local intake of iodine. On an average, however, less than 50% of the Ethiopian households consume adequately iodized salt (UNICEF, 2005). In a report regarding drinking water quality in the Ethiopian part of Rift Valley, 138 samples of drinking water were analysed for several elements. The iodine content of drinking water analyzed ranged from 0.31 - 961, the median being 11 µg/l (Reimann et al., 2002). The duplicate diet analyzed in the present study was prepared on local well water, and this may partly explain the high iodine content found in the children diet.

Enjera made from teff flour is reported to have an iron content of 41 mg/100 g and a zinc content of 1.8 mg/100 g (EHNRI, 1998), Enjera is unleavened bread prepared

Table 5. Food frequency questionnaire answered by the mothers (N=28).

Food item		4 7 4	0	
Cereals/cereal based food items	Once/more than once per day	1-7 times per week	Once per month or less	
Teff	28	0	0	
Sorghum	18	0	10	
Villet	13	0	15	
Berbere	26	2	0	
Maize*	5	18	4	
Wheat	4	20	4	
Barley	1	3	24	
Macaroni	0	6	22	
Spaghetti	0	4	24	
Faffa	0	2	26	
/egetables				
Dnion	28	0	0	
Garlic	9	17	2	
Fomato	2	20	6	
Potatoes	1	21	6	
Beetroot	0	25	3	
Cabbage	0	25	3	
Carrot	0	13	15	
_ettuce	0	5	23	
Sweet potatoes	0	3	25	
Ensete	0	1	27	
Rice	0	1	27	
Pulses				
Beans	27	0	1	
_entils	9	19	0	
Chickpeas	4	23	1	
Haricot beans*	1	18	8	
Fruits				
Drange	0	4	24	
Banana	1	2	25	
Vlango	1	1	26	
Guava	1	0	27	
Animal products				
Egg	0	5	23	
Beef	0	1	27	
Fish	0	1	27	
Chicken	0	0	28	

* One person did not answer.

by fermentation of teff, wheat, barley, maize or sorghum, or from a mixture of these (Stewart and Getachew, 1962). Enjera and other products made from teff constituted about 40% of the food sampled during the four study days, and may, therefore, be regarded a main staple in

the area. Previous studies have estimated the daily intake of iron in parts of Ethiopia to be between 180 and 500 mg/day, which is 10 - 20 times the suggested daily requirement (Hofvander, 1968). However, even though the population using teff as a staple food may have a high iron intake, iron deficiency anaemia is a common problem in Ethiopia. In a study investigating iron deficiency anaemia in preschool children in northern Ethiopia, the lack of iron absorbing enhancers in the children's' diet was suggested to be a main risk factor (Adish et al., 1999).

Interestingly, iron intake proved to be the least important factor. Iron from the cereal-based staple food has low availability, due to absorption inhibitors like tannins, phytic acid, phenols and fibre. The high and variable iron and zinc content of the food analyzed in the present study may also be due to contamination. The duplicate diets analyzed were prepared by the mothers, and we had no control on possible contamination from soil or dust. Soil may have a high mineral content, but the bioavailability is low (Tatala et al. 1998; Hooda et al. 2004).Thus, a high intake may not necessarily mean that the children had an adequate intake of these nutrients.

The children's diet contained little food rich in vitamin C or other iron enhancers (Table 2). Fruits were for instance only consumed by three children during the four day survey. The low fruit intake was confirmed also in the food frequency questionnaire (Table 2). Most mothers indicated that fruits like orange, banana, mango and guava were consumed only once per month or less.

Stunting is common in Ethiopian children and a prevalence of 51% stunting and 8% wasting has been reported (de Onis et al. 1993; UNICEF, 2000). Both a low protein and low mineral intake can affect linear growth (Waterlow 1992; Prentice and Bates, 1994). It has been suggested that, also zinc deficiency may have a role in stunting, especially in developing countries (de Onis et al., 1993). Zinc supplementation has been shown to halt the stunting process in stunted infants in rural Ethiopia (Umeta et al., 2000). Unrefined cereals and legumes contain high levels of phytic acid which form insoluble Zn-phytic acid complexes in the intestine (Lönnerdal, 2000; Nerdal, 2000). The children studied had a cereal based diet with few dietary zinc enhancers, e.g. protein from animal sources.

Most of the children had an adequate intake of iodine, and a variable, but generally high intake of iron and zinc. However, the diet was cereal based and contained little fruit and food of animal origin, which may reduce the availability of iron and zinc. The high variation in iron and zinc content of the food may be due to contamination from the soil.

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