Assessment of Dietary Zinc Ingestion in Pakistan

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Zinc concentration was determined in the Pakistani diet with the use of instrumental neutron activation analysis. It varied from 12.94 to 37.77 μg/g, with an average value of 22.94 ± 5.54 μg/g, indicating a daily intake of 13.59 ± 3.31 mg/d for a reference Pakistani man weighing 63.9 kg. These values are comparable to those recommended by the International Commission of Radiological Protection for a man weighing 70 kg. We also found that the Pakistani diet contains zinc concentrations recommended by the U.S. Food and Nutrition Board and the International Commission of Radiological Protection. *Nutrition* 2002;18:274–278. ©Elsevier Science Inc. 2002

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INTRODUCTION

Human growth metabolism depends on a balanced diet containing protein, lipids, and carbohydrates. It also relies on the supply of optimum quantities of inorganic micronutrients. These micronutrients constitute a small fraction of the entire diet but play important roles in different metabolic processes. Their excess or deficiency may disturb normal biochemical functions of the body.

The reference values for trace elements provided by the International Commission on Radiological Protection (ICRP) are for a Caucasian man (Western European or North American who is 170 cm tall and weighs 70 kg).[1] These values did not provide a realistic estimation for the native populations of Asia, who are smaller (height = 167 cm, weight = 58 kg)[2] and constitute more than 50% of the world's population. The recommended daily dietary allowance of different micronutrients may not be same for these populations because of their body mass indexes. For these reasons, we measured trace elements in daily food and assessed the adequacy and safety of the human diet in Pakistan.

Zinc is an essential element that is found in every cell of the human body. It has diverse roles in the development and healthy growth of the human body:

- enables the activity of more than 200 biological enzymes
- assists in the manufacture of proteins and genetic material
- enables normal growth and skeletal development
- stimulates hair growth
- enables development of taste perception
- assists in hormonal activity, reproduction, and lactation
- lets the body carry out immune functions such as protecting against infection and cancer

Zinc deficiency can result in poor growth, difficulty in wound healing, loss of appetite, undesirable skin changes, and adverse effects on the immune system. The recommended daily allowance for zinc is 15 mg/d.[3] Overload of zinc (>100 mg/d) also can be dangerous. It can depress the immune system, cause anemia and copper deficiency, and decrease high-density lipoprotein cholesterol in blood. The main sources of zinc are animals: meat, liver, eggs, seafood, etc. It is also present in legumes, whole grain cereals, wheat germ, and nuts. Zinc from vegetable sources generally is less absorbed as compared with zinc from animal sources.

Data on zinc concentration and other essential elements in the Pakistani diet were not available. Therefore, a coordinated research program, Ingestion and Organ Content of Trace Elements of Importance in Radiological Protection, Reference Asian Man (phase 2), was carried out with the collaboration of the International Atomic Energy Authority (IAEA) and eight Asian countries, namely Bangladesh, China, India, Japan, Korea, Philippines, Pakistan, and Vietnam. The results on zinc levels in the Pakistani diet are presented in this article.

MATERIAL AND METHODS

Pakistan is basically an agricultural country located between the latitudes of 22′ 30″ and 36′ 31″. It has four provinces: Punjab, Sind, NWFP, and Baluchistan. FATA, the states of Azad Jamu and Kashmir, and northern areas are additional areas administratively attached to Pakistan. Most of the diet consumed by its population is home grown. The typical Pakistani diet is presented in Table I.

Sample Collection and Preparation

To prepare typical Pakistani diets (Table I), 23 different foods were collected according to the Market Basket Method* from 23 major districts or cities in Pakistan (Table II). The collected food samples (e.g., rice, pulses, meat, vegetables, and fruits) were washed with tap water, weighed, oven dried, and pulverized separately in a commercially available grinder with steel blades. (Braun GmbH, Kronberg, Germany). An integrated diet was prepared by mixing the 23 foods according to the criteria of the National Institute of Health in Pakistan.[4] Water, tea, fish, and spices (e.g. garlic pepper, salt, and turmeric) were not added. Dietary samples were prepared with a technique slightly different from the one used by the IAEA.[5] Integrated diets were prepared by

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* In this technique, a typical diet of a particular country, area, or population is constructed, and foods are purchased and prepared for consumption before analysis. The construction of the diet is often based on household food surveys and identical to those consumed by individuals or groups of individuals over a certain period. This method does not assess the variability of individual intakes or reveal extreme intakes but does provide useful basis for the comparison of individual studies. If continued over an extended period, it can expose trends in trace element intake.

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TABLE I.

TYPICAL DIET OF A COMMON PAKISTANI MAN

Food	Amount (g/d)	Food types
Cereals	500	Wheat (roti, paratha, weaning food, rusk, biscuits), rice, corn.
Milk	161	Cow, buffalo, commercial milk products, lassi, yogurt
Roots	40	Potatoes, other root vegetables
Pulses	43	Pulses, gram
Meat	39	Beef, mutton, poultry
Fish	06	Any kind
Egg	7	Any kind
Oils	33	Ghee and other vegetable oils
Vegetables	101	All vegetables except roots
Fruit	8	Any kind
Sugar	39	Any kind

mixing dried rather than wet foods because meat and animal fat are difficult to blend homogeneously.

After mixing, the preparation was cooked for 3 min in microwave oven, ground, and oven dried. The homogenized dry samples were stored in air-tight jars for analysis.

Thirty-one integrated diets were prepared: 19 representative diets according to established Pakistani criteria[4] and 12 diets representative of three socioeconomic strata (wealthy, middle class, poor) in four districts (Karachi, Sialkot, Mianwali, and Muzafarabad).

Analytical Methods

Trace elements by definition occur in very small amounts and their measurement requires reliable and sensitive analytical techniques. The concentration of ^{65}Zn was determined with instrumental neutron activation analysis. The Standard Reference Material (SRM; 1571, i.e., Orchard leaves, obtained from NBS, U.S. Department of Commerce and 1548a, typical diet, NIST, USA) and samples encapsulated in silicon vials were packed in an aluminum container, cold welded, and irradiated at approximately 10 MW (thermal neutron flux $= 7 \times 10^{13}$ n/cm^2/s) in Pakistan Research Reactor 1 for 24 h. After appropriate cooling, the samples were transferred to preweighed polythene capsules and radiometric analysis was performed with a high-purity germanium based γ-ray spectrometer. Calculations were made by using the comparative method.

TABLE III.

CONCENTRATION OF ZINC IN PAKISTANI DIETS

Sample no.	Sample code	Altitude of sample collection: Latitude	Longitude	Concentration (μg/g)
1	PK-DT-01	32′ 30″	74′ 32″	18.47
2	PK-DT-02	32′ 30″	74′ 32″	25.13
3	PK-DT-03	32′ 30″	74′ 32″	28.00
4	PK-DT-04	32′ 34″	71′ 32″	28.93
5	PK-DT-05	32′ 34″	71′ 32″	25.16
6	PK-DT-06	32′ 34″	71′ 32″	25.29
7	PK-DT-07	24′ 53″	67′ 02″	26.79
8	PK-DT-08	24′ 53″	67′ 02″	18.32
9	PK-DT-09	24′ 53″	67′ 02″	20.03
10	PK-DT-10	34′ 24″	73′ 29″	17.20
11	PK-DT-11	34′ 24″	73′ 29″	17.94
12	PK-DT-12	34′ 24″	73′ 29″	14.00
13	PK-DT-13	34′ 12″	72′ 03″	27.85
14	PK-DT-14	34′ 10″	71′ 32″	12.94
15	PK-DT-15	25′ 23″	68′ 21″	20.52
16	PK-DT-16	33′ 55″	73′ 24″	31.87
17	PK-DT-17	33′ 43″	73′ 05″	24.88
18	PK-DT-18	32′ 60″	72′ 40″	18.32
19	PK-DT-19	31′ 34″	74′ 19″	20.48
20	PK-DT-20	31′ 25″	73′ 06″	26.10
21	PK-DT-21	31′ 42″	73′ 59″	18.07
22	PK-DT-22	34′ 08″	73′ 12″	24.75
23	PK-DT-23	29′ 24″	71′ 42″	21.03
24	PK-DT-24	30′ 12″	67′ 01″	19.82
25	PK-DT-25	30′ 12″	71′ 28″	15.74
26	PK-DT-26	27′ 42″	68′ 52″	28.12
27	PK-DT-28	30′ 57″	72′ 28″	19.82
28	PK-DT-29	30′ 44″	70′ 50″	29.02
29	PK-DT-30	30′ 49″	73′ 26″	37.77
30	PK-DT-31	34′ 09″	71′ 46″	24.07
31	PK-DT-32	26′ 15″	68′ 26″	24.76

RESULTS AND DISCUSSION

Zinc concentrations in 31 diet samples prepared from foods collected from 23 districts or cities in Pakistan (Table II) are presented in Table III. It varied from 12.94 to 37.77 μg/g and its average value was 22.94 $\pm$ 5.54 μg/g, where the error is the standard deviation of 31 values from the mean. Using Student's t distribution, the 95% confidence limit was defined as:

TABLE II.

SAMPLING SITES

Site no.	Sampling sites	Site no.	Sampling sites	Site no.	Sampling sites
1	Abbottabad	2	Bahawalpur	3	Charsada
4	Faisalabad	5	Hyderabad	6	Islamabad
7	Jhang	8	Karachi	9	Lahore
10	Mardan	11	Mianwali	12	Multan
13	Murree	14	Muzaffarabad	15	Nawabshah
16	Okara	17	Peshawer	18	Quetta
19	Sargodha	20	Sialkot	21	Sheikhupura
22	Sukkur	23	Toba Tek Singh		

TABLE IV.

CONCENTRATION OF ZINC IN PAKISTANI DIET AND ITS DAILY INTAKE

Specification	No. of Samples	Concentration (μg/g) Minimum	Maximum	Average	Daily intake (mg/d) Minimum	Maximum	Average
All diets	31	12.94	37.77	22.94 ± 5.54	7.62	23.14	13.59 ± 3.31
Diet R	19	12.94	37.77	23.47 ± 5.95	7.62	23.12	13.95 ± 3.60
Diet A	4	17.20	28.93	22.85 ± 5.88	10.32	17.17	13.53 ± 3.48
Diet B	4	17.94	25.16	21.64 ± 4.05	10.73	15.13	12.79 ± 2.30
Diet C	4	14.00	28.00	21.83 ± 6.18	8.36	15.80	12.71 ± 3.43

A, wealthy; B, middle class; C, Poor; R, representative sample

$$m \pm t_{0.975} \frac{\sigma}{\sqrt{n-1}}$$

where m is the mean, σ is the standard deviation, and n is the number of samples. For the present case of 31 samples, from tables of t distribution, $t_{0.975}$ ($\nu = 30$) is 2.04. Therefore, the 95% confidence interval for zinc concentration in the Pakistani diet is 22.94 ± 2.06 μg/g.

Daily intakes of zinc were calculated by multiplying zinc concentration with daily dietary zinc consumption for the different categories of the Pakistani population and are listed in Table IV. The minimum value of daily, zinc intake was 7.62 mg/d, which was about 49% smaller than the value recommended by the U.S. Food and Nutrition Board (FNB).[3] Maximum daily intake was 23.14 mg/d, which was 54% greater than that recommended by the U.S. FNB but very much less than the recommended upper limit of 100 mg/d. Average dietary intake of zinc for the Pakistani population was 13.59 ± 3.31 mg/d. The frequency distribution on the daily intake of zinc is shown in Figure 1 with a Gaussian fit to the data. This figure shows that the distribution is skewed (0.42) to the right.

Average daily intake estimated through the 19 representative diets and 12 dietary samples collected from Sialkot, Mianwali, Karachi, and Muzafarabad for the three population groups (i.e., rich, middle class, and poor classes) were 13.95 ± 3.60, 13.53 ± 3.48, 12.79 ± 2.30, and 12.71 ± 3.43 mg/d, respectively. Therefore, these values are not significantly different.

Measured daily dietary intake was 13.59 mg/d. The daily dietary allowance of the Pakistani man weighing 63.9 kg[2] was calculated as 13.69 mg/d when using the ICRP reference values of 15 mg for 70-kg man.[1] Therefore, there was no significant difference between measured value and desired value. This result shows that the Pakistani diet contains the recommended level of zinc.

Although the zinc concentration (13.59 ± 3.31 mg) in the Pakistani diet seems sufficient and comparable with the ICRP values, its bioavailability may be less due to dietary composition and type of food eaten. Cereals are our staple food, which are rich in phytate that inhibit zinc absorption. Consumption of animal food (a readily available source) is also very low. The deficiency due to high intake of cereals and low intake of animal food can be improved if we increase the quantity of readily absorbed food, i.e., animal food,[3] or fortified food.

A comparison of daily dietary estimates of the reference Pakistani man, reference Caucasian man, and other reported values[6–13] in the literature is presented in Table V. It shows that the concentration of zinc in the Pakistani diet is comparable to other reference values according to average body weight. For a reference Pakistani man weighing 63.9 kg, zinc concentration was 0.213 mg/kg, for the ICRP man weighing 70 kg, the concentration is 0.214 mg/kg.

Quality Control

Accurate measurement of trace elements at micro and sub-micro levels is a tedious and difficult job. The reliability of these values was ensured by internal and external quality control measures for meaningful results.

INTERNAL QUALITY CONTROL. For good quality assurance and measurement, the analysis of diet samples was compared with SRM 1548a. SRM was run with each batch during the study.

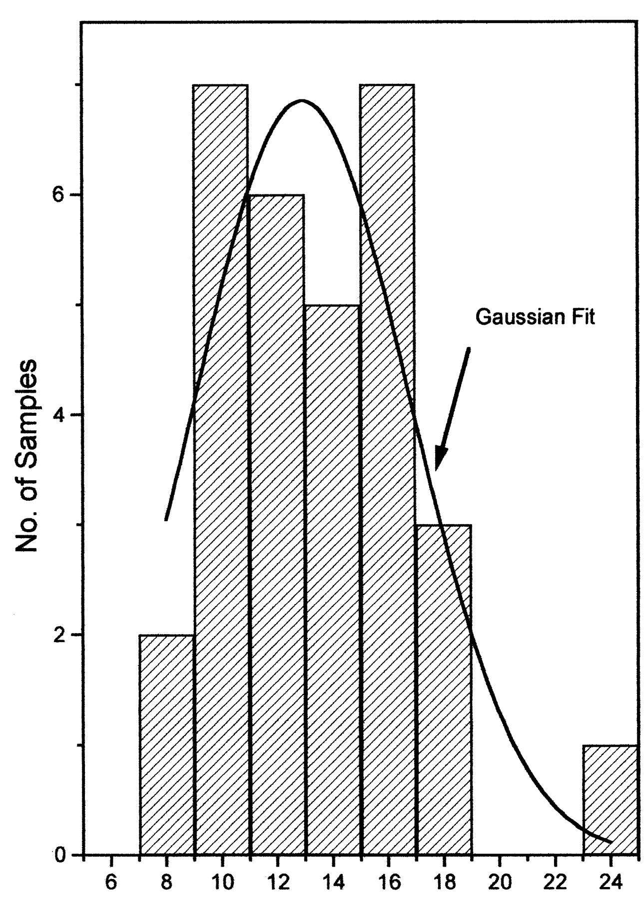

FIG. 1. Daily dietary intake of zinc in Pakistan.

TABLE V.

COMPARISON OF DAILY ZINC INTAKE BY PAKISTANS WITH DATA REPORTED IN THE LITERATURE

Countries	Food (mg/d)*	Sampling method	Reference
Australia	10.6 F, 14.7 M	DPS	6
Bangladesh	7.88	MBS	7
Brazil	8.8	MBS	6
Canada	15.6	MBS	6
China	12.45	MBS	8
Denmark	11 F, 15 M	—	6
Finland	14	DPS	6
France	10.12 F, 12.52 M	MBS	6
Germany	7.6 F, 9.8 M	DPS	6
IAEA study	10	—	6
ICRP value	15	—	1
ICRP revised value	16.3		9
India	9.1	MBS	10
Iran	11	DPS	6
Italy	15.2	MBS	6
Japan	7.5	DPS	11
Morocco	7.5	MBS	6
Myanmar	7.5 F, 9.8 M	DPS	6
Netherlands	8.4	DPS	6
New Zealand	10	DPS	6
Pakistan	13.59	MBS	Present work
Philippine	3.27	MBS	12
RDA	15	—	3
Spain	10	DPS	6
Sudan	10	DPS	6
Sweden	7.8	DPS	6
Thailand	13	DPS	6
Turkey	11	DPS	6
UK	12	DPS	6
USA	14	DPS	6
Vietnam	4.92	MBS, DPS	13

* Values are for men unless noted otherwise.
DPS, Duplicate Portion Study, F, female; IAEA, International Atomic Energy Authority; ICRP, International Commission on Radiological Protection; M, male; MBS, Market Basket Study; RDA, recommended daily allowance

Precision and accuracy were measured. The precision was determined in terms of the relative variation coefficient ($V_c = [\sigma/X] \times 100$), where σ is the standard deviation and X is the arithmetic mean. The accuracy was determined by comparing our mean value (X) with certified values (X_c). These parameters were measured for all sample batches analyzed in this study. The calculated values are presented in Table VI. Our value of zinc concentration in the SRM 1548a sample, using the comparative method with SRM 1571 as the standard was 24.89 ± 2.02 μg/g, whereas the certified value was 24.60 ± 1.7 μg/g. This indicates that analysis of zinc in the Pakistani diet was carried out with an overall average accuracy and precision of 1.20% and 8.11%, respectively.

TABLE VI.

DETERMINATION OF ZINC WITH DIFFERENT ANALYTICAL TECHNIQUES

	INAA		ICP-AES		
Sample code	Mean	σ	Mean	σ	*Z* score
PK-DT-14	12.94	0.19	11.1	0.4	1.08
SRM 1548a	24.89	2.02	23.5	1.4	0.41

ICP-AES, inductively coupled plasma plus atomic emission spectometry; INAA, instrumental neutron activation analysis; , Standard deviation; SRM, Standard Reference Material

TABLE VII.

ZINC CONCENTRATION IN SRM 1548A USING SRM 1571 AS A STANDARD*

Value in present study†	24.89 ± 2.02
Certified value†	24.60 ± 1.79
Accuracy (% difference)	1.20
Precision (%RSD)	8.11

* Micrograms of zinc per gram of food.
† Data are presented as mean ± standard deviation.
RSD, Relative standard deviation; SRM, Standard Reference Material

EXTERNAL QUALITY CONTROL. To check validity and authenticity of the analytical data, we compared our data with those from the Central Reference Laboratory, Japan, whose measurements were done using the ICP-AES technique. The results are presented in Table VII and Figure 2. The measured values by ICP-AES are 6% to 15% smaller than the INAA values. *Z* scores were also calculated and are listed in Table VII. The calculated values of *Z* scores lie well within the acceptable limit of −2 to +2, confirming the validity of our analytical technique.

CONCLUSION

Baseline analytical data for an essential micronutrient, zinc, has been determined in the Pakistani diet. The daily dietary intake of a Pakistani man (20 to 50 y) was 13.59 ± 3.31 mg/d, or 0.213

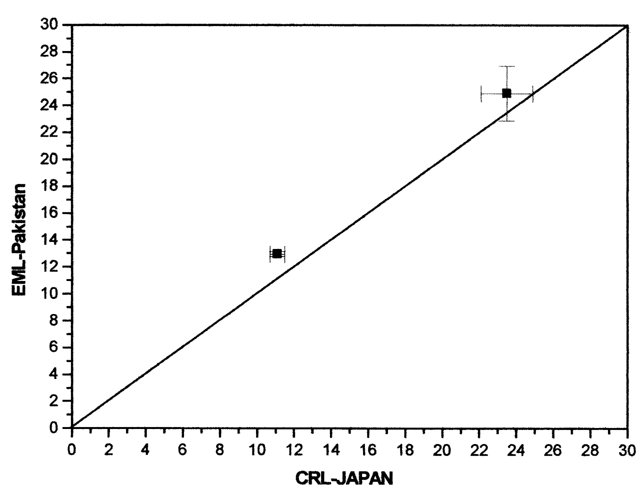

FIG. 2. Zinc concentration (mean ± standard deviation), expressed as micrograms per gram of body weight, in the Pakistani diet: instrumental neutron activation analysis (EML-Pakistan) versus inductively coupled plasma plus atomic emission spectrometry (CRL-Japan). CRL, Central Reference Laboratory, EML, Environmental Monitoring Laboratory.

mg/kg of body weight, which seems to be comparable with the ICRP recommended value of 0.214 mg/kg. The actual zinc concentration in the Pakistani diet might be slightly higher if contributions from water, tea, fish, and spices are also considered.

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