

Assessment of dietary zinc in a population^{1,2}

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ABSTRACT Assessment of dietary zinc status in a population requires several steps, consisting of the measurement of food intake distributions in the population; the analysis of local staple foods, from which zinc intake distributions can be determined, and the comparison of zinc intakes with requirement estimates to determine the risk of inadequate intakes. In low-income countries, these steps may be complicated by the lack of preexisting food-composition data, variations in food preparation methods, inhibition of absorption by other compounds in the diet, and variations in intake among seasons, individuals, and populations. Different techniques for determining the adequacy of zinc intake are compared. Whereas the techniques described in this paper allow for the determination of probability estimates for risk of zinc inadequacy, they do not allow for the identification of actual individuals in a population who are zinc deficient, or define the severity of zinc inadequacy. This information is vital, especially in areas where zinc deficiency is but one of many health problems, and can be obtained only from more detailed biochemical and physiologic studies of zinc status. *Am J Clin Nutr* 1998;68(suppl):430S–4S.

KEY WORDS Dietary zinc intakes, interactive 24-h recall, food-composition table, bioavailability, probability approach

INTRODUCTION

It is well recognized that zinc intake data alone do not provide information on zinc nutritional status. Such information can only be obtained when dietary data are combined with biochemical, anthropometric, and clinical information. Nevertheless, dietary data can be used to determine the distribution of usual zinc intakes within a population. This distribution can then be used to derive an estimate of the prevalence of inadequate intakes by comparison with the distribution of zinc requirements, and thus the proportion of the population at risk for suboptimal zinc status. Nevertheless, there are uncertainties in this prevalence estimate arising from the method of determining usual zinc intakes and the distribution of individual zinc requirements within the population. Sources of these uncertainties and their effect on the prevalence estimates are discussed below.

MEASUREMENT OF DISTRIBUTION OF USUAL FOOD INTAKES IN A POPULATION

When the objective of a dietary survey is to characterize the distribution of usual zinc intakes of a population, quantitative

dietary assessment methods, based on recalls or records measuring the quantity of food consumed by an individual over > 1 d, are required. A detailed description of these methods can be found in Gibson (1).

The number, selection, and spacing of days of dietary assessment required per person depend on the day-to-day variation in food intake within one individual (ie, intrasubject variation). This variation is affected by the nutrient being studied, study population, dietary survey method, and seasonal variations of intake. Nonadjacent days representative of the population should be selected, when possible, to enhance statistical information. In studies of rural areas in less-industrialized countries, market days as well as weekend and weekdays should be proportionally included because the foods consumed can vary between market and nonmarket days (2). Other factors that must be taken into account include characteristics of the subjects within the study population, the respondent burden of the method, and the available resources. Generally, the more accurate assessment methods are associated with higher costs, greater respondent burdens, and lower response rates.

Both random and systematic errors may occur when measuring zinc intakes. Random errors affect the precision of the methods. Such errors can be reduced by increasing the number of measurement days, but cannot be entirely eliminated. In contrast, systematic measurement errors cannot be minimized by extending the number of measurement days. These systematic measurement errors are important because they can introduce a significant bias into the results that cannot be removed by subsequent statistical analysis. Procedures for reducing measurement errors in the dietary assessment protocol include training and retraining sessions for the interviewers and coders, standardizing interviewing techniques and questionnaires, and pretesting and administering a pilot survey before conducting the survey (1).

In less-industrialized countries, weighed food records completed by trained research assistants have often been the method of choice for providing quantitative dietary data because of difficulties intrinsic in collecting recall data from illiterate populations. However, this method is time consuming, expensive, and invasive and may enhance the likelihood that the respondents change their dietary intake during the recording period. Hence, we have developed a modified interactive 24-h recall for illiter-

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ate populations that can be used to provide reliable prevalence estimates for inadequate intakes of dietary zinc.

A detailed discussion of the relative validity of a modified interactive 24-h recall developed to assess the adequacy of trace mineral intakes in illiterate populations is given in Ferguson et al (2). The method involves training the respondents before the recall and incorporates the use of plates, food models, and picture charts to aid respondents in portion size estimation and recall. Each respondent is given a plate and asked to eat their food from that plate to aid visual recall of portion size. Picture charts depicting commonly consumed staple food items are also given to the respondent for use as a checklist on the day the food is actually consumed and for comparison with the recall on the following day. Any discrepancies are queried and resolved. Also, to aid in the estimation of portion size, food samples of commonly consumed foods are prepared daily using local cooking methods. Respondents indicate how much of these prepared food items they consumed during the previous 24 h using their own plates and utensils. The research assistant then weighs and records the amount. Questions on supplement usage must also be included where appropriate. The modifications used aim to reduce systematic and random measurement errors by enhancing recall of foods consumed, reducing the number of memory lapses, and improving the portion size estimates.

COLLECTION, PREPARATION, AND ANALYSIS OF LOCAL STAPLE FOODS FOR COMPILATION OF A LOCAL FOOD-COMPOSITION TABLE

In industrialized countries, nutrient and antinutrient intakes can generally be calculated from food-composition data by using food-composition tables or nutrient databases. Unfortunately, in less-industrialized countries the nutrient and antinutrient contents of local staple foods are often not available. Substitution of values for staple foods grown in Western countries is not advisable because the mineral and phytate contents of plant-based foods tends to reflect local soil mineral levels and food-preparation and processing techniques (3, 4). Hence, in less-industrialized countries, samples of staple food items must be collected, prepared "as eaten," and chemically analyzed for zinc and the promoters and antagonists of zinc absorption before the adequacy of zinc intakes can be determined. Care must also be taken to ensure that there are no missing food-composition values in the database. If necessary, missing values should be computed by using the nutrient and antinutrient contents of a similar food and the procedure used documented.

When collecting food samples for analysis, a standardized sampling protocol must be established to avoid adventitious contamination and to take into account, where appropriate, such factors as seasonal, regional, and genetic differences in the composition of the food; use of fertilizer; stage of ripeness; and method of food preparation (5). In this way, the average nutrient and antinutrient contents of the local staple foods on a year-round nationwide or regional basis can be obtained.

The sampling protocol should involve collecting a few subsamples of each food item, the number selected depending on the importance of the food item in the diet and its expected variation in zinc and antinutrient contents. The subsamples are then combined into a single composite sample for analysis by appropriate analytic methods (5). In some circumstances, subsamples can be analyzed separately to derive an estimate of the natural variation

in the zinc and antinutrient contents of each food item. During the analysis, appropriate analytic quality-control procedures should be used (6).

Raw foods are analyzed most frequently. Nutrient and antinutrient values for cooked composite dishes derived from specific recipes can be calculated from these raw food values by using standardized calculation procedures, which take into account loss in weight during cooking and losses into the cooking water where appropriate (7).

The analytic method most commonly used to analyze the mineral and trace mineral content of staple foods is atomic-absorption spectrophotometry, as outlined in Ferguson et al (4). Until recently, food-composition values for phytic acid (myo-inositol hexaphosphate), the most potent inhibitor of zinc absorption (8), were often analyzed by using the anion-exchange column separation method (9). This method, however, does not differentiate between the higher inositol phosphates (IPs) (IP-6 and IP-5) and the lower IPs (IP-4, IP-3, IP-2, IP-1): only the higher IPs compromise zinc absorption (10). As a result, an HPLC method that can separately identify and quantify both the higher and lower IPs is preferred (11). Use of the HPLC method is especially important for certain prepared foods that have undergone soaking, germination, or fermentation, when some enzymatic and nonenzymatic hydrolysis of hexa- and penta-IPs to lower IPs may occur (12).

High concentrations of calcium exacerbate the inhibitory effect of phytate on zinc absorption in humans by forming a calcium-zinc-phytate complex in the intestine that is even less soluble than phytate complexes formed by either ion alone (13). In general, the calcium content of most plant-based diets in less-industrialized countries is too low to potentiate the negative effect of phytate on zinc bioavailability. Notable exceptions include diets of lactoovovegetarians (14), diets based on tortillas prepared with lime-soaked maize (15), and possibly diets of persons who chew betel nut with lime (16).

Several other dietary components inhibit zinc bioavailability. Of these, the amount and type of dietary fiber may be an additional inhibiting factor in diets of less-industrialized countries. The relative importance of dietary fiber compared with phytate in compromising zinc absorption is controversial, in part because these two antinutrients generally coexist in plant-based diets (17), making it difficult to establish independent effects. Notable exceptions are diets based on sweet potatoes, taros, bananas, and sago in Papua New Guinea and diets based on cassavas and plantains in the forest regions of Ghana. Such diets have a relatively low phytate but high dietary fiber content (18). It appears that in rural diets of less-industrialized countries where protein intakes are generally low, insoluble cereal and vegetable fibers (ie, cellulose and lignin) probably exacerbate the adverse effect of phytate on zinc absorption to some degree (19).

Numerous methods exist for the analysis of dietary fiber in foods, some of which are unsatisfactory. Supplements to the fifth edition of *McCance and Widdowson's The Composition of Foods* (20) present values for cellulose, lignin, and the soluble and insoluble noncellulosic polysaccharides. These components can be analyzed by gas chromatography followed by fractionation (21)

CALCULATION OF THE DISTRIBUTION OF USUAL ZINC INTAKES IN POPULATIONS

Food-composition data, in conjunction with food intake data, can be used to calculate nutrient and antinutrient intakes, prefer-

ably with use of a computer program. A compilation of data on intakes of calcium, zinc, and phytic acid, and the molar ratios of phytate zinc in children from several less-industrialized countries as well as from Canada is presented in **Table 1**.

Intakes of zinc, like those of all nutrients, vary from day to day both among individuals (between or intersubject variation) and within one individual over time (within or intrasubject variation). These sources of variation contribute to errors in the estimations of usual zinc intakes. However, unlike with measurement errors, no attempt should be made to minimize inter- and intrasubject variations because they characterize the true usual intake. Instead, the dietary method should be designed in such a way that these two sources of variability can be separated and estimated statistically by using analysis of variance. This can be achieved provided food intakes have been measured for each individual for > 1 d (1). If it is not feasible to carry out repeated observations on all the subjects, the intra- and intersubject variations can be calculated on repeated intakes obtained from a subsample of the subjects. The number and distribution of intakes to be repeated should be determined before the survey in consultation with a statistician.

The ratio of intra- to intersubject variation is known as the variance ratio. The estimate of intrasubject variation that is obtained from the analysis of variance represents the sum of true variation in intake from day to day within the same person, plus the remaining sources of random measurement errors. Intrasubject variation cannot be distinguished statistically from the random measurement errors that may occur when assessing zinc intakes. Nevertheless, by using appropriate quality control procedures to minimize measurement errors, some of the confounding effects of measurement errors on intrasubject variability can be reduced (28).

Coefficients of intra- and intersubject variance (as percentages) and the variance ratios for zinc intakes in less-industrialized countries are summarized in **Table 2**. Variance ratios depend critically on sample size, number of days per subject over which the food intake was measured, age, sex, the dietary assessment methodology used, the nutrient of interest, and the sociocultural group. A ratio of 1.0 indicates that the intrasubject and intersubject variances are equal whereas a ratio > 1.0 indicates that the intrasubject variance is greater than the intersub-

ject variance (29). Note that in Ecuadorian men, all of the variance is intraindividual (30) and even in Malawi, the intrasubject variation is larger than the intersubject variation, despite the consumption of relatively monotonous diets. As well, the variance ratios for repeated 24-h recall data for Malawian women are markedly higher than those for weighed records completed over the same 2 recall days (2). This discrepancy might be attributed to the technical errors inherent in the recall technique compared with the weighed record. Sources of these errors are summarized in Gibson (1).

High variance ratios from 24-h recall data indicate that a larger number of recalls than weighed records per subject are required to achieve a similar level of measurement precision for assessing usual zinc intakes. Seasonal variations in zinc intakes may be more marked in less-industrialized countries than in industrialized countries, and should be taken into account in the data collection protocol.

When the objective is to estimate the prevalence of inadequate zinc intakes in the study population by comparison with the recommended nutrient intakes, it is essential that the zinc intake data used refer to the same time frame as the data used to formulate the recommended nutrient intakes for zinc. Hence, because the latter are based on intakes over a moderate period of time rather than on 1 particular day, data on the distribution of usual zinc intakes are required. The degree of uncertainty in estimating the distribution of usual zinc intakes in a population depends on the magnitude of the intra- and intersubject variability in zinc intakes. However, by calculating both the intra- and interindividual variances for each age-sex category by using analysis of variance and following statistical procedures outlined by the National Research Council (29), the observed distribution of zinc intakes can be adjusted to derive a distribution of usual zinc intakes. A comparison of observed and adjusted distributions of usual intakes for zinc for 60 pregnant women, calculated by using these statistical procedures, is given in **Figure 1**. The dietary intakes are based on 2 repeated interactive 24-h recalls. Note that the adjustment process yields a distribution with reduced variability that preserves the shape of the original observed distribution.

TABLE 1

Dietary intakes of zinc, calcium, and phytate; phytate-to-zinc molar ratios; and percentage of children at risk for inadequate zinc intakes¹

Country (reference)	Age y	Zinc mg/d ²	Calcium mg/d ²	Phytic acid mg/d ²	Phytate:zinc	
					ratio	SAR %
Papua New Guinea (16), n = 67	6–10	4.4 ± 1.3	359 ± 160	646 ± 663	12	76 ³
Ghana (22), n = 148	3–6	4.7 ± 1.1	344 ± 145	591 ± 153	13	72 ⁴
Malawi (22), n = 67	4–6	6.6 ± 1.7	413 ± 189	1899 ± 590	25	94 ⁴
Canada (23), n = 106	4–6	6.9 ± 2.3	702 ± 249	300 ⁵ (NA)	5	2 ³
Egypt (24), n = 96	1.5–2.5	5.2 ± 1.6	218 ± 89	796 ± 249	16	36 ⁴
Kenya (24), n = 100	1.5–2.5	3.7 ± 0.9	210 ± 99	1066 ± 324	28	90 ⁴
Mexico (24), n = 59	1.5–2.5	5.3 ± 1.3	735 ± 199	1666 ± 650	30	68 ⁴
Nigeria (25), n = NA	1–10	3.6 ± 0.9	—	—	—	—
Guatemala (26), n = 136	6–8	9.0 ± 2.7	621 (442–819) ⁵	962 (576–1427) ⁵	11	—
Gambia (27), n = 99	1.2–1.5	4 (NA)	284 (NA)	—	—	—

¹ SAR, subjects at risk for inadequate zinc intake; NA, not available.

² $\bar{x} \pm$ SD.

³ Based on less than two-thirds recommended nutrient intake.

⁴ Based on probability approach.

⁵ Median (first–third quartile).

TABLE 2

Observed coefficients of intra- and intersubject variation (as percentages) and ratios of intra- to intersubject variances for zinc intakes¹

	Mean intake	Intrasubject variation	Intersubject variation	IA:IR
	mg/d	%	%	
Two 24-h recalls of rural Malawian women, <i>n</i> = 60	6.2	34	21	2.6
Two 1-d weighed records of Malawian women, <i>n</i> = 60	6.8	44	23	3.7
Four 24-h recalls of Ecuadorian women (10–20 y of age), <i>n</i> = 13	6.3	37	18	4.4
Four 24-h recalls of Ecuadorian men (10–20 y of age), <i>n</i> = 15	6.9	58	0	—

¹ IA:IR, ratio of intra- to intersubject variation.

ADEQUACY OF ZINC INTAKES IN RELATION TO REQUIREMENT ESTIMATES

The final stage in this process is to evaluate the adequacy of the distribution of usual zinc intakes in the population, after the variance adjustment, by comparison with an appropriate set of dietary reference values, taking into account the estimated dietary zinc bioavailability. Several tables of dietary reference values are available; they are discussed in detail in Gibson (1). For studies in less-industrialized countries, the newly revised requirement estimates for zinc set by the World Health Organization (6) should be used. They include estimates for both basal and normative zinc requirements. The basal requirement is the amount needed to prevent clinically detectable signs of functional impairment whereas the normative requirement is the amount needed to maintain tissue stores or reserve capacity.

Direct measurements of the bioavailability of zinc in plant-based diets consumed in many less-industrialized countries are limited; some have been made in men and nonpregnant women with use of isotopic methods (6). Interpretation of the isotopic data are difficult, however, because zinc bioavailability is affected by the isotopic labeling technique used, and the absorption of zinc from a meal is not simply the sum of the zinc contents of individual foods (31). Nevertheless, these isotopic zinc absorption data have been used to develop a model for classifying diets as having high (ie, 50%), moderate (ie, 30%), and low (ie, 15%) zinc bioavailability (6). This model is based on the dietary content of animal and fish protein, calcium (< or > 1 g Ca/d), and daily molar ratios of phytate to zinc (< 5, 5–15, and > 15). The last mentioned are said to predict the inhibitory effect of phytate on zinc absorption when the dietary zinc intake is close to the requirement (8, 31). Ratios above 15 have been associated with suboptimal zinc status (6, 14, 32).

Based on this model (6), estimates of zinc absorption have ranged from 15% for diets in Malawi, Kenya, and Guatemala to 30% for diets in Ghana, Guatemala, Egypt, Papua New Guinea, and Ghana (15, 22, 24, 26). Zinc absorption is probably higher in Ghanaian and Egyptian diets because their plant-based staples generally have lower molar ratios of phytate to zinc: fermented maize and cassava products—kenkey, banku, and gari—in Ghana (22) and yeast-leavened wheat-based bread in Egypt (24). In Papua New Guinean diets, zinc absorption is also estimated to be 30% because sago is the staple, which has a low phytate content (16).

Conventionally, a fixed cutoff approach has been used to estimate the prevalence of inadequate zinc intakes, based on either the dietary reference value or two-thirds of this value (1). However, this approach does not take into account the variability in requirements among individuals. Moreover, it may over- or underestimate the proportion of the population at risk for dietary zinc inadequacy, depending on the position of the zinc require-

ment distribution in relation to the distribution of zinc intakes for the population. Recently, a probability approach was recommended by the National Research Council (29). The method predicts the number of persons within a group with zinc intakes below their own requirements and hence provides an estimate of the proportion of the population at risk of inadequate zinc intakes, provided reliable data on the distribution of usual zinc intakes based on repeated observations on individual intakes are available. The approach assumes that the distribution of zinc requirements is Gaussian with a CV of 15%, and that the correlation between intake and requirement is very low.

When this approach is used, risk of dietary zinc inadequacy is notably higher in children from Malawi and Kenya than in children from Ghana and Egypt (22, 24). Such inadequacies in zinc intakes in plant-based diets of less-industrialized countries are likely to occur even when predicted energy requirements are met (24). Effects of systematic under- or overreporting across population subgroups on these prevalence estimates has not yet been established. Efforts should always be made to minimize reporting bias by improving the methods for collecting food intake data, thereby minimizing the effect on the prevalence estimates.

It is noteworthy that these comparisons of zinc intakes with estimated requirements do not take into account the possibility that humans can adapt to chronically low zinc intakes and achieve zinc balance by increasing zinc absorption (33) or reducing urinary and fecal zinc excretion (33, 34). Whether such adjustments occur with very high-phytate diets is unknown. Brune et al (35) reported that adult long-term vegetarians did not adapt to high phytate intakes by increased absorption of ⁵⁹Fe.

In summary, probability estimates for risk of zinc deficiency can be calculated from dietary zinc intake alone, provided reliable data on the distribution of usual zinc intakes are obtained. However, probability estimates do not identify actual individuals

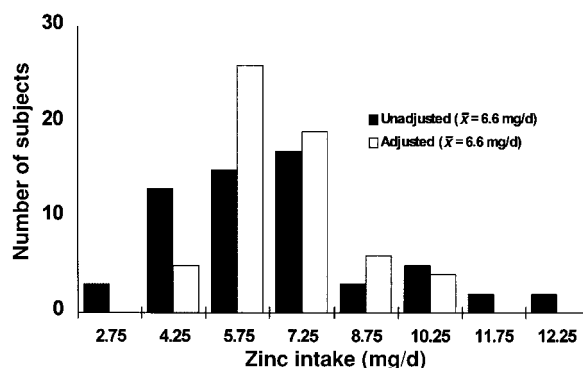



Figure 1. Unadjusted and adjusted zinc intakes of some rural Malawian pregnant women.

in the population who are deficient or define the severity of the zinc inadequacy. Such information can only be obtained when the dietary intake data are combined with biochemical and functional physiologic indexes of zinc status. This is especially important in developing countries, where the coexistence of many other multifaceted health problems often confounds the diagnosis of zinc deficiency. 

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