Use of serum zinc concentration as an indicator of population zinc status

Sonja Y. Hess, Janet M. Peerson, Janet C. King, and Kenneth H. Brown

Abstract

Assessing the prevalence and severity of zinc deficiency in populations is critical to determine the need for and appropriate targeting of zinc intervention programs and to assess their effectiveness for improving the health and well-being of high-risk populations. However, there is very little information on the zinc status of populations worldwide due to the lack of consensus on appropriate biochemical indicators of zinc status. The objective of this review was to evaluate the use of serum zinc concentration as an indicator of population zinc status.

We have reviewed the response of serum zinc concentration to dietary zinc restriction and zinc supplementation. In addition, we completed pooled analyses of nine zinc intervention trials in young children to assess the relations between serum zinc concentration of individuals before treatment and their responses to zinc supplementation. Also, in updated combined analyses of previously published data, we investigated the relation between the mean initial serum zinc concentration of a study population and their mean growth responses to zinc supplementation in randomized intervention trials among children.

The results from depletion/repletion studies indicate that serum zinc concentrations respond appreciably to severe dietary zinc restriction, although there is considerable interindividual variation in these responses. There is also clear evidence that both individual and population mean serum zinc concentrations increase consistently during zinc supplementation, regardless of the initial level of serum zinc concentration. By contrast, an individual's serum zinc concentration does not reliably predict that person's response to zinc supplementation.

Serum zinc concentration can be considered a useful biomarker of a population's risk of zinc deficiency and response to zinc interventions, although it may not be a reliable indicator of individual zinc status.

Key words: Serum zinc, plasma zinc, zinc status, indicator, zinc deficiency, assessment

Introduction

Need for indicators to assess zinc status and to evaluate zinc intervention programs

Adequate zinc nutrition is necessary for normal child growth, protection from infection, and satisfactory outcomes of pregnancy [1]. Despite the serious adverse consequences of zinc deficiency for human health, and the availability of considerable evidence to suggest that this condition is common in many lower-income countries [1], there have been few attempts to implement large-scale intervention programs to prevent the problem. One possible explanation for this continuing failure to confront zinc deficiency more aggressively is the lack of specific data on the zinc status of populations worldwide. Thus, there is an urgent need to obtain better information on the true prevalence of this condition, both to determine the presence and magnitude of the problem in particular settings and to identify specific population segments at elevated risk. This information will enable health planners to determine the need for zinc intervention activities and to estimate the likely health benefits that might be achieved following successful execution of programs to increase zinc intake. Moreover, once control strategies are introduced, periodic monitoring of population zinc status will be necessary to assess the effectiveness and safety of these interventions.

In summary, assessing the prevalence and severity

Sonja Y. Hess, Janet M. Peerson, and Kenneth H. Brown are affiliated with the Program in International and Community Nutrition and the Department of Nutrition, University of California, Davis, California, USA; Janet C. King is affiliated with the Children's Hospital Oakland Research Institute, Oakland, California, USA.

Please direct queries to the corresponding author: Kenneth H. Brown, Department of Nutrition, University of California, One Shields Ave., Davis, CA 95616, USA; e-mail: khbrown@ucdavis.edu.

of zinc deficiency in populations is critical to determine the need for and appropriate targeting of zinc intervention programs and to assess their effectiveness for improving the health and well-being of high-risk populations. For these reasons, the World Health Organization (WHO), UNICEF, the International Atomic Energy Agency (IAEA), and the International Zinc Nutrition Consultative Group (IZiNCG) organized a Working Group Meeting to develop consensus on appropriate indicators of zinc status at the population level. In preparation for this meeting, we compiled the present background paper on the use of serum zinc concentration, focusing on three main questions: 1) Does the serum zinc concentration of individuals or populations reflect their recent or longer-term zinc intake? 2) Does the serum zinc concentration of individuals or populations respond consistently to interventions designed to increase zinc intake? 3) Can the serum zinc concentration of individuals or populations be used to predict functional responses to zinc intervention programs? Before addressing each of these questions, several relevant background issues will be reviewed.

Biochemical indicators of zinc status

Previous publications have described the range of techniques that have been proposed to assess zinc status or the risk of inadequate zinc intake [1–3]. The assessment techniques that seem most promising for field application include the measurement of dietary zinc intake (specifically, dietary intake of absorbable zinc) and/or the measurement of serum (or plasma) zinc concentration in a representative sample of the population of interest [1]. Zinc concentration has also been measured in a number of other tissues, such as different blood cell types, hair, and nails, and a variety of a zinc-binding proteins, such as metallothionein and other zinc metalloenzymes, have been investigated as possible indicators of zinc status [1, 3]. However, these approaches do not seem to provide any greater sensitivity or analytic convenience than using serum zinc concentration, so this paper will focus only on the latter biomarker. More recently, several groups of investigators have applied tracer methods and kinetic modeling to study the mass of different internal zinc pools and zinc flux among these metabolic compartments. However, the significant demands on study subjects and the labor intensity, expense, and complexity of this methodology preclude its usefulness as a routine tool for assessing zinc status [4]. Therefore, despite several limitations of serum zinc concentration, which will be discussed below in more detail, this indicator remains the most appropriate biochemical marker for assessing population zinc status.

Individual versus population assessment

Nutritional assessment methods may be applied to evaluate the status of either individuals or populations, and it is important to distinguish between these two applications when exploring the appropriateness and validity of different assessment techniques. Assessment of individuals is used to guide case-specific treatment or counseling, so the results of the assessment must be valid for the particular individuals in question. By contrast, assessment of populations is used to plan and evaluate larger-scale, population-based interventions, so the results of the assessment do not need to provide certainty with regard to any particular individual's true zinc status.

Because of efficient homeostatic mechanisms, an individual's serum zinc concentration is maintained within a fairly narrow span of values, even when a broad range of zinc intakes below and above the theoretical requirements are consumed. Thus, decreases in an individual's serum zinc concentration are detectable only when zinc depletion is severe or prolonged [3, 5]. Nevertheless, there is considerable evidence that serum zinc concentration is a useful indicator of population zinc status, as described below in more detail. In other words, when a higher than expected percentage of individuals in a population have low serum zinc concentrations, it can be inferred that there is an elevated risk of zinc deficiency occurring within the population [1, 3, 6, 7].

Serum versus plasma zinc concentration

Serum and plasma zinc concentrations are the most widely used biochemical indicators of zinc status. Because of the different procedures used for collecting blood and separating serum or plasma, their respective zinc concentrations are not necessarily identical. For example, in two studies in which blood samples were collected simultaneously from the same individuals and separated as either serum or plasma, the zinc concentrations were greater in serum than in plasma [8, 9]. One set of investigators speculated that the observed differences could be explained by the fact that the serum samples were separated from blood cells after a longer delay than the plasma samples, so more zinc exited from the cells into serum than into plasma. Notably, a follow-up study that systematically controlled both the amount of blood collected and the time of cell separation found no difference in the zinc concentrations of serum and plasma [9], thereby supporting the foregoing hypothesis. For the sake of simplicity, this paper will generally refer to "serum zinc" for both types of specimens, unless it is important to distinguish between the two types of specimens in a particular situation.

Physiology of plasma zinc concentration

Distribution of tissue zinc

The total body zinc content of human adults ranges from 1.5 to 2.5 g, with higher average contents in men than in women. Zinc is present in all organs, tissues, fluids, and secretions, but most (> 95%) is located in the fat-free mass, mainly in intracellular compartments. Due to the bulk of skeletal muscle and bone in the body, the zinc contents of these tissues account for the majority (~83%) of whole-body zinc [10]. The concentration and total zinc content of various tissues, and the proportions contributed by each to total body zinc, are shown in **table 1**. Less than 0.2% of the total body zinc (i.e., ~3.5 mg of zinc) circulates in plasma, although the plasma zinc pool turns over ~125 times per day [11]. When total body zinc content is depleted during periods of low intake, proportionately more zinc is lost from bone, liver, testes, and plasma, whereas the zinc contents of skeletal muscle, skin, and heart are better preserved [12]. There are no conventional tissue reserves of zinc that can be released or sequestered quickly in response to variations in dietary zinc supply; the average mass of the so-called "rapidly exchangeable" metabolic pool of zinc is only about 140 to 175 mg in adults [11, 13, 14].

TABLE 1. Zinc content of major organs and tissues in adult (70-kg) man

Organ or tissue	Zinc concentration (mg/kg wet weight)	Total zinc content (mg)	Proportion of total body zinc (%)
Skeletal muscle	50	1,400	63
Skeleton			
Bone	90	450	20
Marrow	20	60	3
Periarticular tissue	11	11	<1
Cartilage	34	30	1
Liver	40	72	3
Lung	40	40	2
Skin	15	39	2
Whole blood	6	33	1
Kidney	50	15	1
Brain	10	14	1
Teeth	250	11.5	1
Hair	200	4	< 1
Spleen	20	3.6	< 1
Lymph nodes	14	3.5	< 1
Gastrointestinal tract	15	1.8	< 1
Prostate	100	1.6	< 1
Other organs or tissues	Variable	50	2
Total		2,240	100

Source: Adapted and reprinted with the permission of the publisher, from Iyengar [10].

Regulation of plasma zinc concentration

In healthy individuals, fasting plasma zinc concentrations are maintained homeostatically within fairly narrow limits (about 80 to 100 μg/dL [12 to 15 μmol/L]). Nearly all of the plasma zinc is associated with one of two major proteins: albumin (~70%) and α_2-macroglobulin (the remainder). A very small amount of the plasma zinc, ~0.01%, is ultrafilterable and complexed with amino acids, primarily cysteine and histidine [15]. Albumin-bound zinc responds to hormones, such as insulin and leptin [16], and cytokines; hormonally mediated metabolic shifts related to fasting, meals, and acute stress can influence plasma zinc concentrations.

The mechanism whereby hormones and cytokines affect plasma zinc concentrations has not been identified, but it likely involves the zinc transporter proteins. Two families of zinc transporter proteins have been defined. The ZIP family of transporters increases cytoplasmic zinc concentration by increasing cellular zinc uptake, thereby decreasing plasma zinc concentrations, and prompting the release of zinc from cellular vesicules. By contrast, the ZnT family of transporters reduces cytoplasmic zinc concentration by exporting cellular zinc, thereby increasing plasma zinc concentrations, and facilitating the movement of cytoplasmic zinc into intracellular organelles [17]. These transporters help maintain plasma zinc concentrations within a narrow range while zinc intake varies widely.

Kinetic modeling (flux to/from plasma pool)

Kinetic data from both radioactive and stable isotopic zinc tracer studies provide important information on the movement, or flux, of zinc from the plasma to various body pools with changes in zinc status. The total body zinc can be divided into two metabolic pools: a rapidly exchangeable pool with a turnover of about 12.5 days, and a slow pool with a turnover of about 300 days [15]. The rapid pool, composed of the most metabolically active forms of zinc, includes zinc in plasma and extracellular fluid and in liver, pancreatic, kidney, and intestinal tissue. Skeletal muscle and bone are the primary components of the slow pool, which represents nearly 90% of the whole-body zinc.

Severe dietary zinc restriction (< 1 mg/day for 4 to 5 weeks) causes a marked drop (about 35%) of zinc in the rapidly exchangeable zinc pool, but has little or no measurable effect on the size of the slow zinc pool, which is maintained at the expense of the rapid pool [18]. The shift in tissue zinc distribution occurs relatively rapidly and is unlikely to be due to cellular

zinc deficiencies. Instead, the signaling pathways of endocrine receptors may be altered early in severe zinc depletion, thereby causing shifts in tissue zinc distribution. For example, cessation of growth in zinc-deficient rats was related to an early reduction in the expression of hepatic insulin-like growth factor-1 and growth hormone receptor genes [19]. In contrast to the response to severe restriction of zinc intake, the size of the rapidly exchangeable zinc pool does not change with a marginal zinc intake (4.6 mg/day) [11]. Presumably, adjustments in the expression of the zinc transporters can maintain normal levels of plasma zinc concentration with low, but not severe, levels of zinc depletion.

Effects of age, sex, meals, time of day, infection/inflammation/stress, pregnancy, and genetics on serum zinc concentration

Serum zinc concentrations vary according to the time of day, proximity of previously consumed meals, and occurrence of recent exercise or other forms of stress, fluctuating by as much as 20% during a 24-hour period [20]. The diurnal variation in circulating zinc concentration is largely a result of metabolic changes after meal consumption, although some variation may occur as a result of normal circadian variation in metabolism [21, 22]. Meal consumption results in a decrease in serum zinc concentrations, which is cumulative following repeated meals [22, 23], whereas overnight and daytime fasting result in increased circulating zinc concentrations [22]. Hotz et al. [6] reanalyzed the data for serum zinc concentrations that were obtained during the second US National Health and Nutrition Examination Survey (NHANES II) and found significant differences according to fasting state (morning fasting compared with morning nonfasting specimens) and time of day (morning compared with afternoon specimens), averaging 7.3% and 9.5%, respectively.

Serum zinc concentration also varied with age and sex, although the differences introduced by these characteristics were of a somewhat smaller magnitude than those due to the time of day and fasting status [6]. For both males and females, serum zinc values were lower in childhood, peaked during adolescence and young adulthood, and declined slightly with age thereafter. From adolescence onwards, men had higher serum zinc concentration than women, and these differences were greatest for the 20- to 40-year age group.

Infection and inflammation can decrease serum zinc values, with the magnitude of change depending on the severity and stage of infection [24]. In community-based surveys, the reductions in serum zinc concentration due to infection average ~10% to 12% compared with uninfected reference groups [25]. Several other factors, such as low serum albumin, elevated white blood cell counts, pregnancy, lactation, and use of oral contraceptives or other hormones, can also affect serum zinc levels and must be considered in the interpretation of laboratory results [1].

The extent to which genetic factors influence an individual's serum zinc concentration is unknown. Cousins et al. [26] identified 104 genes that responded positively to zinc supplementation (i.e., gene expression increased as cellular zinc concentration rose) and 86 genes that responded negatively (i.e., gene expression decreased as cellular zinc climbed). The authors concluded that the spectrum of zinc responsiveness exhibited by genes coding for zinc transporters demonstrates both their potential collective importance in coordinating cellular zinc trafficking and their integrating influence on zinc homeostasis. Genetic polymorphisms that affect gene expression may alter zinc metabolism and homeostasis, as occurs, for example, with a specific mutation of the SLC39A4 gene, which affects the synthesis of the ZIP4 zinc transporter, resulting in intestinal malabsorption of zinc and the clinical syndrome of acrodermatitis enteropathica [27].

Response of plasma zinc concentration to dietary modification and zinc supplementation

As described above, zinc metabolism and serum zinc concentration are homeostatically controlled. Nevertheless, there are limits to homeostasis when dietary intakes are consistently inadequate or excessive, so serum zinc concentrations change under these circumstances. In the following section, we review studies that examined the relationships between serum zinc concentration and either dietary zinc intake or zinc supplementation. Three types of studies will be considered: experimental studies of controlled dietary zinc restriction and repletion, controlled intervention trials of zinc supplementation, and observational studies of population dietary zinc intake and serum zinc concentration.

Depletion/repletion studies

The recommended dietary zinc intakes proposed by the US Institute of Medicine [28] are 11 mg/day for men and 8 mg/day for women. Several groups of investigators have monitored serum zinc concentrations during experimental studies in which healthy adult volunteers were exposed to controlled diets, both during baseline periods when zinc intakes were adequate and following periods during which dietary zinc intakes were restricted to varying degrees (**table 2**). Baseline mean zinc intakes ranged from 8.2 to 16.5 mg/day for the 13 available studies included in this review. In six of these studies [13, 18, 29–32], zinc intakes were severely restricted during the depletion phase by using synthetic

TABLE 2. Response of serum zinc concentration during dietary zinc restriction and repletion studies in healthy adult men and women

Reference	*N*	Baseline period		Restriction period		Repletion period		Mean ± SD serum zinc (μg/dL)[1]		
		Duration (days)	Zinc intake (mg/day)	Duration (day)	Zinc intake (mg/day)	Duration (days)	Zinc intake (mg/day)	Baseline	End of restriction	End of repletion
Hess et al. [29]	10 women	Usual diet	~10	24	0.17	NA	NA	83 ± 12^a	54 ± 20^b	NA
Baer and King [30][2]	6 men	7	15.7	28–63	0.28	14–35	6.0–46.3	89 ± 17^a	50 ± 20^b	94 ± 10^a
Sutherland [31][3]	8 men	16	12	40	< 0.5	30	12	72 ± 10^a	28 ± 14^b	61 ± 9^a
Van Loan et al. [32][2,4]	8 men	17	12	33–41	0.3	27–34	12	72 ± 4^a	24 ± 5^b	66 ± 2^a
King et al. [18]	5 men	16	12.2	41	0.23	NA	NA	71 ± 8^a	25 ± 7^b	NA
Lowe et al. [13][4]	5 men	16	12.2	41	0.23	29	12.2	76 ± 10^a	19 ± 7^b	76 ± 8^a
Hunt et al. [39][5]	11 men	28	10.4	4 × 35	1.4	35	10.4	83 ± 8^a	73 ± 18^b	90 ± 6^a
					2.5				89 ± 11^a	
					3.4				94 ± 13^a	
					4.4				96 ± 10^a	
Milne et al. [33][6]	3 men	31–42	8.2	108–126	3.4	18–24	32.5	86	84	101
Lukaski et al. [41][7]	5 men	30	8.6	120	3.6	30	33.6	88^a	77^b	89^a
Wada et al. [34][7]	6 men	12	16.5	54	5.5	9	16.5	114	108	113
Ruz et al. [38][8]	15 men	7	15	7	0.6	14	35	97 ± 11^a	80 ± 13^b	101 ± 14^a
				42	then 4					
Allan et al. [36]	7 men	35	13.7	70	4.6	35	13.7	75 ± 4	74 ± 3	69 ± 4
Pinna et al. [37]	8 men	35	13.7	70	4.6	35	13.7	73 ± 9	75 ± 7	71 ± 12
Chung et al., unpublished[9]	9 men	13	11	7	0.6	28	11	81 ± 7	79 ± 9	86 ± 10
				33	then 4					

NA, not available

[1] Different letters indicate significant differences within studies ($p < .05$).

[2] The restriction period was continued to allow individual response of plasma zinc concentration.

[3] One subgroup received two intravenous infusions of 50 mg zinc overnight during the repletion phase. Combined results of subgroups are shown.

[4] One subgroup received two intravenous infusions of 66 mg zinc overnight during the repletion phase. Combined results of subgroups are shown.

[5] Four different restriction diets (1.4, 2.5, 3.4, and 4.4 mg/day) were provided in random order for 35 days each.

[6] The mean serum zinc concentration was estimated from a graph that showed results from three volunteers only. No information on significant differences was provided.

[7] The mean serum zinc concentration was estimated from a graph.

[8] During the restriction period, subjects received 0.6 mg zinc/day for 7 days and 4 mg zinc/day for 6 weeks. Both diets contained high levels of phytic acid.

[9] Chung CS, Dare D, Peerson JM, King JC, Brown KH, unpublished data. Subjects received 0.6 mg zinc/day for 7 days and 4 mg zinc/day for 33 days, and 1.3 g/day of phytic acid was given for the first 21 days of the restriction period.

formula diets that provided zinc intakes of less than 1 mg/day, and in five studies [33–37] zinc restriction was moderate, with zinc intakes ranging from 3.4 to 5.5 mg/day. In two of the remaining studies ([38] and Chung CS, Dare D, Peerson JM, King JC, Brown KH, unpublished data), the subjects received 0.6 mg zinc/day for 1 week, then 4 mg zinc/day for 5 to 6 weeks. In the other study [39], zinc intakes were varied monthly, in random order, from 1.4 to 4.4 mg/day.

In all of the studies of severe zinc restriction (including the multilevel restriction study when the lowest level of zinc was provided), the mean serum zinc concentrations decreased significantly, sometimes dramatically, during the depletion period, with final mean serum zinc concentrations ranging from 25% to 88% of the initial ones. Interestingly, there was considerable variability in the magnitude and velocity of change, both across studies and among subjects within a particular study. **Figure 1**, for example, shows individual changes according to the duration of dietary depletion for the subjects in the study by Sutherland [31]. In some studies [32], changes were evident within 2 weeks of introduction of the restricted diet, whereas in other studies, changes were detectable only after 5 to 9 weeks [30, 40]. The reasons for these variable responses across studies are unknown, but they may relate to other components of the diet or to the subjects' preexisting zinc status or genetic factors controlling zinc homeostasis. In all cases, the serum concentrations returned rapidly to near baseline levels following repletion with adequate zinc diets for periods of 9 to 35 days.

The mean serum zinc concentrations decreased significantly in only two of the studies in which dietary zinc restriction was moderate. In one of these studies, the changes in mean serum zinc concentration occurred only after 90 days of exposure to reduced zinc intakes [41]. In the other study, serum zinc concentration fell within 7 days; but, as described above, in this study there was a 1-week period of severe zinc restriction and increased phytate intake preceding the period of moderate zinc restriction [38]. In the two studies in which the mean serum zinc concentrations decreased following introduction of moderately restricted intakes, the final mean concentrations ranged from 82% to 86% of the initial values, and they returned to baseline levels within 2 weeks after initiation of dietary repletion.

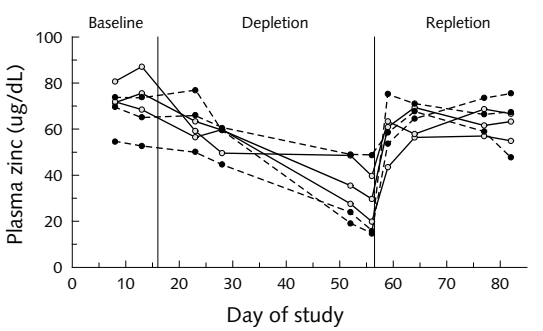

FIG. 1. Fasting plasma zinc concentration from six individual male subjects (aged 20 to 40 years) undergoing three metabolic periods: a 16-day baseline period (12 mg zinc/day), a 40-day depletion period (< 0.5 mg zinc/day) , and a 30-day repletion. During repletion period, the subjects were divided into two groups. Three subjects (broken lines) received an intravenous infusion of 50 mg zinc overnight on day 57 and continued on the diet providing less than 0.5 mg zinc/day until day 67 when they received another 50 mg zinc infusion. On day 69 of the repletion period, they started the 12 mg zinc/day diet and continued it for the remainder of repletion. Three subjects (solid lines) received 12 mg zinc/day throughout repletion [31]

Lowe et al. [13] measured changes in total body zinc content using metabolic balance techniques during their depletion study, and they found a strong correlation between the change in total body zinc content and change in changes in serum zinc concentration (r^2 = 0.826, p < .001). They concluded that during short-term, acute perturbations in intake, and in the absence of confounding factors, changes in serum zinc concentration accurately reflect changes in whole-body zinc status.

In summary, serum zinc concentration responds appreciably and fairly rapidly (within less than 2 weeks in some cases) to severe dietary zinc restriction, although there is considerable inter-individual variation in these responses. Thus, there is convincing evidence of a strong relationship between dietary zinc intake and serum zinc concentration under these experimental conditions. When zinc intake is moderately restricted (~3 to 5 mg/day) among individuals who were previously adequately nourished, serum zinc concentrations either remain unchanged or decrease just slightly, and only after more prolonged periods of exposure to the restricted diets. Similar experimental data are lacking for individuals who had been chronically exposed to marginal zinc intakes before any dietary interventions. Therefore, it is not known whether the relatively small responses that were observed in the aforementioned studies of moderate dietary restriction of previously well-nourished adults can be extrapolated to marginally nourished populations.

Supplementation trials

Three different sets of studies are available to examine the relations between controlled zinc supplementation and serum zinc concentrations of free-living individuals. One set of relatively short-term studies (1 to 4 weeks) was conducted among presumably well-nourished adults in the United States, and the other sets of longer-term studies (2 to 15 months) were carried out among either children or pregnant women in lower-income countries. The volunteers who participated in the five available studies of adults in the United States

TABLE 3. Serum zinc concentration in response to short-term zinc supplementation of healthy, presumably zinc-sufficient adults

Reference	*N*	Supplementation		Group	Serum zinc concentration (μg/dL)			
		Duration (days)	Zinc dose (mg/day)		Baseline	Supplementation		
					0 days	6–7 days	10–15 days	28 days
Sullivan and Cousins [42][1]	20 men	15	50	Zinc	75	140**	93*	NA
				Placebo		75	79	NA
Sullivan et al. [43]	25 men	18	50	Zinc	77 ± 3	140 ± 7**	95 ± 4*	NA
				Placebo		75 ± 3	85 ± 2	NA
Cragg et al. [45] [2]	6 men, 12 women	14	25	Zinc	112 ± 17	NA	131 ± 41	NA
				Placebo		NA	109 ± 12	NA
Cao and Cousins [44][1]	16 men	10	15	Zinc	98	105**	98	NA
				Placebo		92	92	NA
Morejohn and Brown, unpublished[3]	26 men	28	15	All	78 ± 10	83 ± 12†	86 ± 14†	86 ± 15†

NA, not available
[1]Mean serum zinc concentration was estimated from a graph.
[2] Additional data supplied by the authors.
[3] Morejohn B, Brown KH, unpublished data. No placebo group was included in the study.
* Significant difference between zinc-supplemented and control subjects ($p < .05$).
** Significant difference between zinc-supplemented and control subjects ($p < .01$).
† Significant difference vs. baseline serum zinc concentration ($p < .05$).

received 15 to 50 mg of elemental zinc per day as supplements to their usual diets ([42–45] and Morejohn B, Brown KH, unpublished data). In each of these studies there was a significant rise in the mean serum zinc concentrations within the first 6 days of supplementation (**table 3**). Although the mean serum zinc concentrations subsequently declined from the peak postsupplementation levels, they remained above the baseline values throughout the periods of observation in almost all of the available studies. We located just one longer-term supplementation trial of presumably well-nourished adults, which found a sustained increase in serum zinc concentrations following a 6-month period of zinc supplementation of middle-aged French men [46].

Longer-term trials among children in lower-income countries have also consistently found significantly greater serum zinc concentrations in the supplemented group than in the control group, and these differences persisted during the period of supplementation, which often lasted for a number of months [47–50]. In a published meta-analysis of zinc supplementation and child growth [7], 15 of the studies (comprising a total of 1,141 subjects) provided data on serum zinc concentration [51–65], as shown in **figure 2**. There was a consistent, moderately large increase in the mean serum zinc concentrations of the zinc-supplemented children after 2 to 15 months of supplementation, with an overall effect size of 0.820 SD units (95% confidence interval [CI], 0.499 to 1.14 SD). These relationships were explored further in a newly completed pooled analysis described below.

Serum zinc concentrations decline during the course

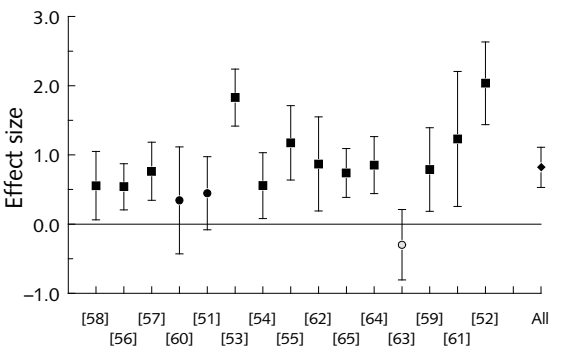

FIG. 2. Weighted mean effect size and 95% CI (in SD units) for the effect of zinc supplementation on children's serum zinc concentration: results for the individual studies and the meta-analysis of all studies combined. Open circle, study with a negative effect size and confidence limits that include zero; filled circles, studies with a positive effect size and confidence limits that include zero; filled squares, studies with a positive effect size and confidence limits that do not include zero [7]. Adapted and reproduced with permission from *The American Journal of Clinical Nutrition*. © American Society for Nutrition.

of pregnancy, so these values must be interpreted in relation to the stage of gestation. For the present review, we summarized the responses in serum zinc concentration that were observed during previously completed randomized trials of zinc supplementation among pregnant women. The effect sizes for change in serum zinc concentration were analyzed statistically by a random-effects meta-analysis. Five of the available nine studies did not provide explicit information on the

standard deviation of the change in plasma zinc concentration, so the standard deviations were estimated from the initial and final standard deviations, assuming no correlation between the two sets of values.

The studies ([66–73] and Dirren H, Gil J, unpublished data) included in this meta-analysis (comprising a total of 2,633 subjects) are listed in **table 4**. The women's mean serum zinc concentration at baseline ranged from 54 to 104 μg/dL. In eight of nine studies, zinc supplementation had a positive effect on change in serum zinc concentration, with effect sizes for all nine studies ranging from 0.0 to 0.55. Overall, there was a significant positive effect of zinc supplementation on the mean serum zinc concentrations of pregnant women, with an overall effect size of 0.200 SD units (95% CI, 0.051 to 0.348 SD) (**fig. 3**).

In conclusion, short-term zinc supplementation of presumably well nourished adults increases their mean serum zinc concentrations; and longer-term supplementation of children in lower-income countries results in consistent, positive responses in serum zinc concentration, which persist for as long as 15 months. Zinc supplementation also increases the serum zinc concentration among pregnant women, and partially mitigates the usual decline in serum zinc concentration that occurs over the course of pregnancy. Thus, collection of information on the mean serum zinc concentration of groups of individuals before and after zinc supplementation is a reliable method for assessing successful delivery of the intervention.

Descriptive studies of dietary intake and serum zinc concentration

A final approach that can be used to understand the relations between serum zinc concentration and usual dietary zinc intake is to examine descriptive studies of free-living subjects to determine whether the two sets of information are correlated. Such studies can be conducted with either individuals or populations as the unit of analysis. However, as already discussed, serum zinc concentration is unlikely to reflect an individual's

TABLE 4. Serum zinc concentration in response to zinc supplementation in pregnant women[1]

Reference	Country	*N*	Supplementation		Group[1]	Serum zinc concentration (μg/dL)		Time of follow-up
			Zinc dose (mg/day)	Duration		Baseline	Follow-up	
Hunt et al. [66]	USA[2]	213	20	Before 27 wk gestation until delivery	Zinc Control	65 ± 12 66 ± 11	64 ± 10† 63 ± 10†	7–8 mo gestation
Hunt et al. [67]	USA[2]	138	20	Before 27 wk gestation until delivery	Zinc Control	69 ± 11 69 ± 12	63 ± 9† 60 ± 8†	31 or 36 wk gestation
Neggers et al. [68]	USA[2]	493	25	19 wk gestation until delivery	Zinc Control	63 ± 10 63 ± 10	59 ± 10** 56 ± 10	38 wk gestation
Caulfield et al. [69]	Peru	538	15	10–24 wk gestation until delivery	Zinc Control	69 ± 14 68 ± 14	56 ± 10* 55 ± 9	37–38 wk gestation
Osendarp et al. [70]	Bangladesh	446	30	12–16 wk gestation until delivery	Zinc Control	100 ± 27 104 ± 29	101 ± 29 99 ± 28	7 mo gestation
Christian et al. [71]	Nepal	202	25	3 wk	Zinc Control	56 ± 18 54 ± 12	63[3]* 50[3]	End of intervention
Castillo-Duran et al. [72]	Chile	507	20	Before 20 wk gestation until delivery	Zinc Control	78 ± 10 77 ± 9	69 ± 9 67 ± 9	36–38 wk gestation
Hafeez et al. [73]	Pakistan	128	20	10–16 wk gestation until delivery	Zinc Control	73 75	88† 72	Delivery
Dirren and Gil, unpublished[4]	Ecuador	198	30	8–14 wk gestation until delivery	Zinc Control	83 ± 10 82 ± 7	74 ± 10** 66 ± 7	8 mo gestation

[1] Studies are presented here only if they had a zinc-only treatment group or if zinc was given together with other nutrients and the same nutrients except for zinc were supplied to the control group.
[2] Low-income women.
[3] The value was calculated from serum zinc concentration at baseline and change in serum zinc over time.
[4] Dirren H, Gil J, unpublished data.
* Significant difference between zinc-supplemented and control subjects ($p < .05$).
** Significant difference between zinc-supplemented and control subjects ($p < .01$).
† Significant change from baseline ($p < .05$).

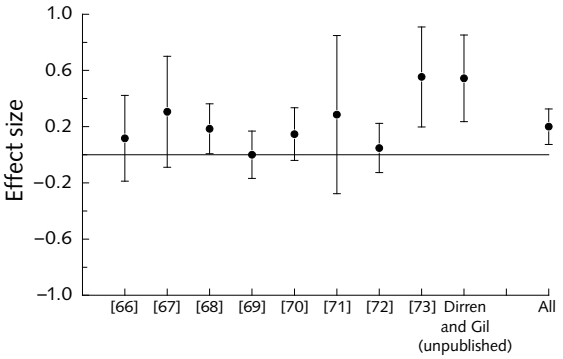

FIG. 3. Weighted mean effect size and 95% CI (in SD units) for the effect of zinc supplementation on serum zinc concentration in pregnant women: results of the individual studies and the meta-analysis of nine zinc supplementation trials ([66–73] and Dirren H, Gil J, unpublished data). See **table 4** for details of study characteristics

true zinc status, except in the cases of either relatively severe zinc deficiency or continuous consumption of zinc supplements, so misclassification of individual zinc status is possible. Moreover, multiple days of observation are necessary to characterize an individual's usual dietary zinc intake with reasonably good precision, so misclassification of individual dietary intake may also occur unless appropriate care is taken in the study design. Thus, it is unlikely that strong correlations between individual dietary zinc intake and serum zinc concentration would be detectable under most study conditions.

Despite these methodological conundrums, it is reasonable to expect that there should be a positive relationship between zinc intake and serum zinc concentration at the population level, because usual dietary zinc intake is undoubtedly a major determinant of zinc status. Thus, populations that are exposed to chronically low dietary zinc intakes should have a greater proportion of individuals with zinc deficiency and hence low serum zinc concentrations, and vice versa. However, before exploring these relationships, it is worth considering several additional methodological issues.

There are two possible approaches to explore the relationship between dietary zinc intake and serum zinc concentrations of different population groups: 1) to compare the percentage of individuals in different populations whose usual zinc intakes are classified as inadequate in relation to theoretical zinc requirements and the percentage of individuals in the same populations who have low serum zinc concentrations, or 2) to compare the mean dietary zinc intakes of different populations and the mean serum zinc concentrations in the same populations. The former analysis requires the collection of at least 2 days of dietary observations to estimate the distribution of usual zinc intakes in the population, hence the percentage of individuals with inadequate intakes. Moreover, because the biological relationship between dietary zinc intake and zinc status depends on the amount of dietary zinc that is absorbed, for both types of comparisons zinc absorption from the diet must be estimated, and individuals known to have intestinal malabsorption should be excluded from the analysis. For each of these analytic approaches, the unit of analysis for correlational studies would be the population, so multiple populations would need to be included in the study. After consideration of all of these caveats, there are very few studies that have used appropriate methods to compare dietary zinc intakes and serum zinc concentrations in different population groups.

Several studies have compared serum zinc concentration in meat-eaters versus non-meat-eaters, so these could be considered as "population studies." Although not all of these studies found significant differences between the means of the different self-selected groups, as defined by the nature of their diets ([74–76] and Morejohn B, Brown KH, unpublished data), possibly because of their small sample sizes, most studies found trends in the expected direction. Morejohn and Brown, for example, found a mean ± SD serum zinc concentration of 76 ± 12 in young men selected for study because of reported low red meat consumption ($N = 13$) versus 80 ± 10 μg/dL in subjects with high red meat consumption ($N = 13$) ($p = .31$). Likewise, in a study of New Zealand women who were not using oral contraceptives, the mean serum zinc concentration was greater among the 149 carnivores than among the 48 women who did not eat red meat (80 versus 77 μg/dL, respectively; $p < .05$) [75]. In the same study, in a selected subgroup of 195 women who excluded red meat from their diet, 19% had serum zinc values less than 70 μg/dL, compared with only 9% of those who ate red meat ($p = .055$). Similarly, among Canadian women, 24% of the vegetarians ($N = 79$), 33% of the semivegetarians ($N = 16$), and 18% of the omnivores ($N = 29$) had serum zinc concentrations less than 70 μg/dL ($p = .52$) [74].

Flesh foods are rich sources of zinc, whereas plant-based diets often contain high levels of phytic acid and dietary fiber, components known to inhibit dietary zinc absorption [77]. A significant inverse relationship was found between serum zinc concentrations and the phytate:zinc molar ratios in the diets of women in New Zealand and Canada [74, 75] and of adult men in the United States (Morejohn B, Brown KH, unpublished data). Among women in New Zealand, the mean serum zinc concentration was greater among those with dietary phytate:zinc ratios less than 15 versus greater than 15 (i.e., 80 vs. 78 μg/dL, $p < .05$) [75]. In contrast, a cross-sectional study of 152 pregnant women in Malawi did not find a significant difference in plasma zinc concentration when subjects were classified into groups with low or high molar ratios of phytate:zinc

in their diets [78]. A possible explanation for the lack of association in the latter study could be that the phytate:zinc molar ratios in this study were very high (median > 17) and the mean plasma zinc concentration was very low, even lower than reported from other studies in developing countries [79].

Based on the methodological issues described above, studies should be reviewed critically before any conclusions are drawn regarding the usefulness of serum zinc concentration as an indicator of dietary zinc intake in free-living populations. Among the limited number of available studies, there is a trend for dietary intake of absorbable zinc or indicators of dietary quality that affect zinc absorption to be associated with the mean serum zinc concentration of the population, although the magnitude of the dietary effects is small and the associations fairly weak, possibly because of the methodological difficulties described. As more national surveys are completed to collect information on dietary intake and serum zinc concentration in representative samples of the population, it will be of interest to assess the correlation between the two sets of information across populations.

Usefulness of serum zinc concentration to predict functional responses to zinc interventions among individuals and populations

A potentially valuable application of nutritional assessment techniques is to predict beneficial or adverse responses to specific nutrition-related interventions. To determine whether either individual or population mean serum zinc concentrations can predict functional responses to zinc supplementation, we considered two sets of information. First, we conducted pooled analyses of individual data that were made available by the authors of previously completed supplementation trials. Second, we reviewed results of an updated version of a combined analysis of published studies of the mean effects of zinc supplementation on child growth. The purpose of these two sets of analyses was to determine whether individual children or groups of children were more or less likely to respond to the interventions in relation to their initial, pre-intervention serum zinc concentrations. The methods and results of the pooled analyses are described in the following section, and the results of the updated combined analyses are summarized subsequently.

Pooled analyses of the relations between baseline (preintervention) serum zinc concentrations of individuals and their responses to zinc supplementation

Data sets were identified in two ways for inclusion in the pooled analyses: by using a computerized bibliographic search (PubMed with key words: zinc supplementation; morbidity), and by reviewing the bibliographies of a previously published pooled analysis of zinc supplementation and risk of infection [80] and meta-analyses of zinc intervention trials and child growth [7, 81]. Double-blind, placebo-controlled intervention trials among prepubertal children were considered acceptable for inclusion in the current pooled analyses if they involved a zinc-only treatment group or if zinc was given together with other nutrients and the same nutrients except zinc were supplied to the control group. Other selection criteria were that the studies were published or conducted after 1990 (to facilitate access to the authors and their data sets), the studies were conducted in lower-income countries, the children were not intentionally selected because of the presence of infection at baseline (because infection might confound the interpretation of the baseline serum zinc concentration), and the total sample size was greater than 50. The outcomes that were considered for the pooled analyses were the incidence and prevalence of diarrhea, the incidence of acute lower respiratory infection, anthropometric evidence of physical growth, and change in serum zinc concentration.

A total of nine potentially acceptable studies were identified from the computerized search and nine from the review of other bibliographic sources. The first authors of these 18 studies were contacted, and 10 replied positively with complete data sets containing individual-level information. Of these, one study was excluded because baseline serum zinc concentrations were unavailable, resulting in a final set of nine studies for the analyses, comprising a total of 2,012 children, of whom 1,883 had data for initial serum zinc concentrations.

The nine studies [56–58, 63, 64, 82–85] included in the analyses are summarized in **table 5**. These nine studies were conducted in seven different countries: five in Latin America and two in Asia. The sample sizes of the individual studies ranged from 80 to 638 children. The time of blood collection and fasting state varied among the nine included studies. For three studies, no information on the time of blood sampling and fasting state was provided. In one trial, blood samples were collected in a nonfasting state, and in five trials, fasting blood samples were collected with the use of different definitions of fasting. Outlying values for anthropometric data were eliminated if the associated z-score for height-for-age ($N = 6$), weight-for-age ($N = 49$), or weight-for-height ($N = 10$) was less than −5 or greater than +5, and any values for change in z-score less than −3 or greater than +3 ($N = 53$) were coded as missing. Because the 99th percentile of the NHANES II reference data for children 3 to 10 years of age is 127 μg/dL, the 43 initial values (2.3%) for serum zinc concentration that were greater than this

TABLE 5. Selected characteristics of studies and study subjects included in the pooled analysis[1]

			Zinc supplementation			Initial characteristics of study subjects (mean ± SD)					
Reference	Country	N	Dose (mg)	Frequency	Duration (mo)	Age (mo)	Serum zinc (μg/dL)	Weight (kg)	HAZ	WAZ	WHZ
Cavan et al. [56]	Guatemala	163	10	5 days/wk	6	81.6 ± 7.0	92.5 ± 15.0	19.7 ± 2.5	–1.5 ± 0.9	–1.0 ± 0.8	0.06 ± 0.7
Dirren et al. [57]	Ecuador	96	10	6 days/wk	15	31.6 ± 11.7	74.3 ± 12.8	11.2 ± 2.0	–2.5 ± 0.8	–1.8 ± 0.9	–0.01 ± 0.9
Castillo-Duran et al. [58]	Chile	71	3	Daily	6	0.01 ± 0.0	79.2 ± 15.4	2.3 ± 0.2	–1.0 ± 0.8	–2.0 ± 0.3	–1.3 ± 0.9
Ruz et al. [63]	Chile	98	10	Daily	14	39.8 ± 6.2	109.5 ± 12.5	NA	NA	NA	NA
Rosado et al. [64]	Mexico	110	20	5 days/wk	12	28.9 ± 8.0	83.2 ± 23.4	11.1 ± 1.6	–1.7 ± 0.9	–1.6 ± 1.0	–0.5 ± 0.9
Osendarp et al. [82]	Bangladesh	301	5	Daily	5	0.9 ± 0.1	75.9 ± 17.0	3.4 ± 0.5	–1.0 ± 0.9	–1.3 ± 0.7	–0.6 ± 0.8
Lind et al. [83]	Indonesia	326	10	Daily	6	6.1 ± 0.4	60.4 ± 14.0	7.3 ± 0.9	–0.4 ± 1.0	–0.4 ± 1.0	–0.01 ± 1.1
Brooks et al. [84]	Bangladesh	638	70	Weekly	12	6.6 ± 2.8	64.5 ± 12.6	6.3 ± 1.3	–1.3 ± 1.1	–1.6 ± 1.3	–0.7 ± 1.2
Brown et al. [85]	Peru	200	3	Daily	6	7.5 ± 0.9	77.3 ± 14.4	7.6 ± 0.9	–1.2 ± 0.5	–0.8 ± 0.9	0.4 ± 1.0

HAZ, height-for-age z-score; WAZ, weight-for-age z-score; WHZ, weight-for-height z-score; NA, not available

[1]Data are presented only for subjects for whom data on initial serum zinc concentration were available.

cutoff were coded as missing. None of the 75 values of final serum zinc concentration that were greater than 127 μg/dL (4.7%) were eliminated from consideration, because these higher than expected results may have been due to the effect of zinc supplementation.

For each study and outcome variable, separate analyses of covariance were completed with the following possible explanatory variables: treatment group (zinc or control), initial serum zinc concentration of each individual, and the interaction of these two main effects. We also considered the square of initial serum zinc concentration and its interaction with treatment group, but these latter variables did not contribute to the understanding of these relationships, so they were excluded from the final models.

The mean ± SD initial serum zinc concentrations ranged from 60 ± 14 to 110 ± 12 μg/dL in the different studies. Eight of the nine study data sets provided information on changes in height and weight and in height-for-age, weight-for-age, and weight-for-height z-scores following the introduction of zinc supplementation or control intervention (**table 6**). The mean initial height-for-age z-scores ranged from –0.4 to –2.5 z across studies, and only two were less than –1.5 z. This is important to emphasize, because the previous meta-analyses of the effect of zinc supplementation on children's growth found positive responses to zinc only among those studies that enrolled children whose initial mean height-for-age or weight-for-age z-scores were less than approximately –1.5 z [7, 81]. Thus, the growth of children included in most of these currently available trials would not have been expected to respond to zinc supplementation. Indeed, as shown in **table 7**, there was a significant main effect of treatment group on change in height-for-age in only two of these studies [57, 58], and there were marginally significant ($p \sim .055$) interactions between initial serum zinc concentration and treatment group in just two other studies [82, 85].

The results of the four studies that found some effect of zinc supplementation on linear growth are shown in **figure 4**. In the studies by Dirren et al. [57] and Castillo-Duran et al. [58], there were no significant interactions between the individuals' initial serum zinc concentration and the magnitude of their response to supplementation. In the study by Osendarp et al. [82], children with lower initial serum zinc concentrations tended to have larger, positive growth responses to zinc supplementation and vice versa, but the opposite relationship between initial serum zinc concentration and treatment effect was observed in the study by Brown et al. [85]. Thus, the initial serum zinc concentrations of individual children included in these studies did not seem to predict consistently their growth responses to zinc supplementation. However, more information is needed from populations that have a greater initial degree of severity of growth stunting.

TABLE 6. Changes in HAZ, WAZ, WHZ, and serum zinc concentration in zinc-supplemented and control groups over the whole study period in nine zinc intervention trials in children

Reference	*N*	Study duration (mo)	Group	HAZ change	WAZ change	WHZ change	Serum zinc change (μg/dL)
Cavan et al. [56]	163	6	Zinc	−0.06 ± 0.14	0.06 ± 0.23	0.12 ± 0.27	14.2 ± 16.9***
			Control	−0.03 ± 0.15	0.09 ± 0.31	0.15 ± 0.47	4.2 ± 17.4
Dirren et al. [57]	96	15	Zinc	0.15 ± 0.37*	0.20 ± 0.36	0.14 ± 0.48	17.7 ± 23.8***
			Control	−0.03 ± 0.36	0.20 ± 0.49	−0.04 ± 0.46	3.0 ± 12.6
Castillo-Duran et al. [58]	71	6	Zinc	0.56 ± 0.81**	1.66 ± 0.69***	1.20 ± 1.17	−10.3 ± 15.2***
			Control	−0.19 ± 1.06	0.79 ± 0.74	0.94 ± 1.12	−20.6 ± 21.5
Ruz et al. [63]	98	14	Zinc	NA	NA	NA	3.1 ± 15.3
			Control	NA	NA	NA	10.3 ± 14.9
Rosado et al. [64]	110	12	Zinc	0.23 ± 0.42	0.54 ± 0.52	0.25 ± 0.50	25.3 ± 30.6***
			Control	0.21 ± 0.42	0.39 ± 0.39	0.26 ± 0.56	4.5 ± 18.2
Osendarp et al. [82]	301	6	Zinc	0.05 ± 0.69	0.05 ± 0.84	0.09 ± 1.04	10.3 ± 30.0***
			Control	0.006 ± 0.80	−0.03 ± 0.86	−0.14 ± 1.03	−5.0 ± 24.1
Lind et al. [83]	326	6	Zinc	−0.40 ± 0.76	−1.10 ± 0.62*	−0.71 ± 0.99	20.3 ± 32.6***
			Control	−0.39 ± 0.73	−1.23 ± 0.50	−0.91 ± 0.81	−0.01 ± 21.1
Brooks et al. [84]	638	12	Zinc	−0.79 ± 0.69	−1.06 ± 0.89	−0.56 ± 0.92*	5.9 ± 19.6**
			Control	−0.86 ± 0.69	−1.17 ± 0.84	−0.73 ± 0.97	1.6 ± 16.0
Brown et al. [85]	200	6	Zinc	−0.16 ± 0.33	−0.57 ± 0.69	−0.56 ± 0.67	4.8 ± 14.8***
			Control	−0.16 ± 0.37	−0.69 ± 0.52	−0.49 ± 0.67	−3.9 ± 15.2

HAZ, height-for-age z-score; WAZ, weight-for-age z-score; WHZ, weight-for-height z-score; NA, not available
* Significant difference between zinc-supplemented and control group ($p < .05$).
** Significant difference between zinc-supplemented and control group ($p < .01$).
***Significant difference between zinc-supplemented and control group ($p < .001$).

TABLE 7. Statistical significance (p values) from analysis of covariance with main effects of treatment group, initial serum zinc concentration, and their interaction as predictors of changes in HAZ, WAZ, and WHZ in eight zinc intervention trials[1]

Dependent variable		Cavan et al. [56]	Dirren et al. [57]	Castillo-Duran et al. [58]	Rosado et al. [64]	Osendarp et al. [82]	Lind et al. [83]	Brooks et al. [84]	Brown et al. [85]
HAZ change	Group	.175	**.018**	**.002**	.818	.628	.915	.215	.928
	Initial zinc	.523	**.010**	.937	.820	**.001**	.247	.158	.627
	Group * initial zinc	.231	.782	.832	.260	**.054**	.150	.583	**.055**
WAZ change	Group	.422	.992	< **.0001**	.148	.399	**.040**	.148	.221
	Initial zinc	.822	.270	.210	.258	**.001**	.723	.906	.950
	Group * initial zinc	.834	.897	.391	.608	.859	**.077**	.360	**.013**
WHZ change	Group	.649	**.059**	.418	.922	**.066**	**.063**	**.033**	.513
	Initial zinc	.682	**.068**	.665	.833	.740	.521	.823	.351
	Group * initial zinc	.971	.965	.227	.556	.520	.787	.778	.235

HAZ, height-for-age z-score; WAZ, weight-for-age z-score; WHZ, weight-for-height z-score.
[1] p values < 0.1 are indicated in bold.

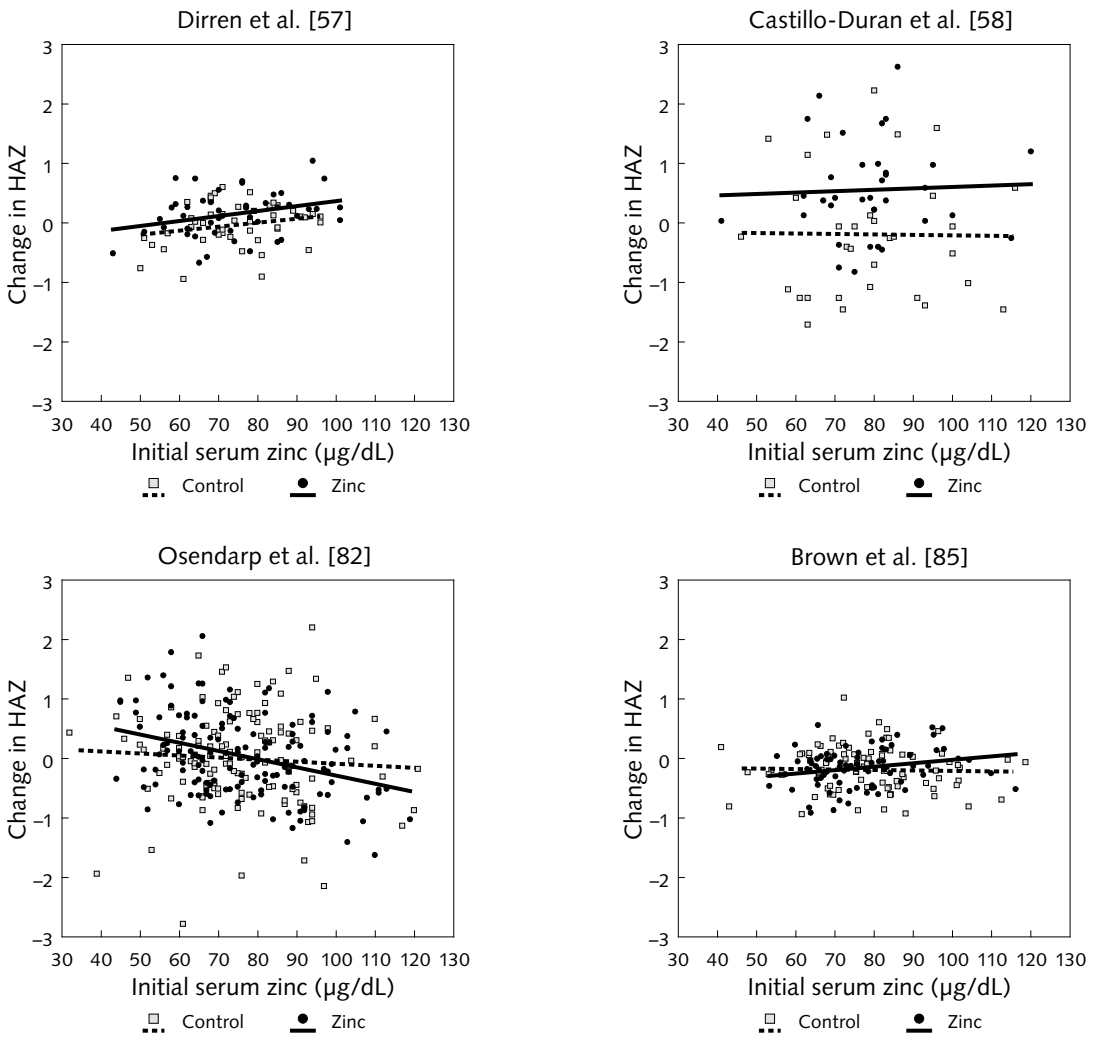

FIG. 4. Change in height-for-age z-scores (HAZ) of individuals in response to zinc supplementation according to initial serum zinc concentration in four zinc intervention trials [57, 58, 82, 85] with significant effect of treatment or significant or marginally significant interaction between initial serum zinc concentration and treatment. See **table 7** for results of statistical analyses

Data from all the available studies were also combined in a single model, using study, treatment group (zinc or control), initial serum zinc concentration, initial serum zinc concentration squared, and all possible interactions (except zinc by zinc-squared) as independent variables. Again, there was no significant interaction between initial serum zinc concentration and the effect of zinc supplementation, nor was there significant nonlinearity. Finally, because the studies that were available for the current pooled analyses did not include many subjects with moderately severe or severe stunting, we also completed a separate pooled analysis including only the subset of all children whose initial height-for-age z-scores were less than –1.5 z. Even in this subset of 628 children, there were no significant interactions between initial serum zinc concentration and linear growth response to supplementation.

The mean baseline weight-for-age z-scores ranged from –0.4 to –2.0 z, and there were significant main effects of zinc supplementation on change in these z-scores in just two of the trials [58, 83], as shown in **table 7**. In two studies there were significant or marginal interactions between initial serum zinc concentrations and treatment group with respect to change in weight-for-age z-scores [83, 85]. Both of these were in the direction of greater growth responses among supplemented children with higher initial serum zinc concentrations (**fig. 5**). These conclusions were not altered when the data from all of the studies were combined in a single model, nor when just those 716 children with initial weight-for-age z-scores less than –1.5 were included in the model.

There was a significant main effect of supplementation on change in weight-for-height z-scores in only

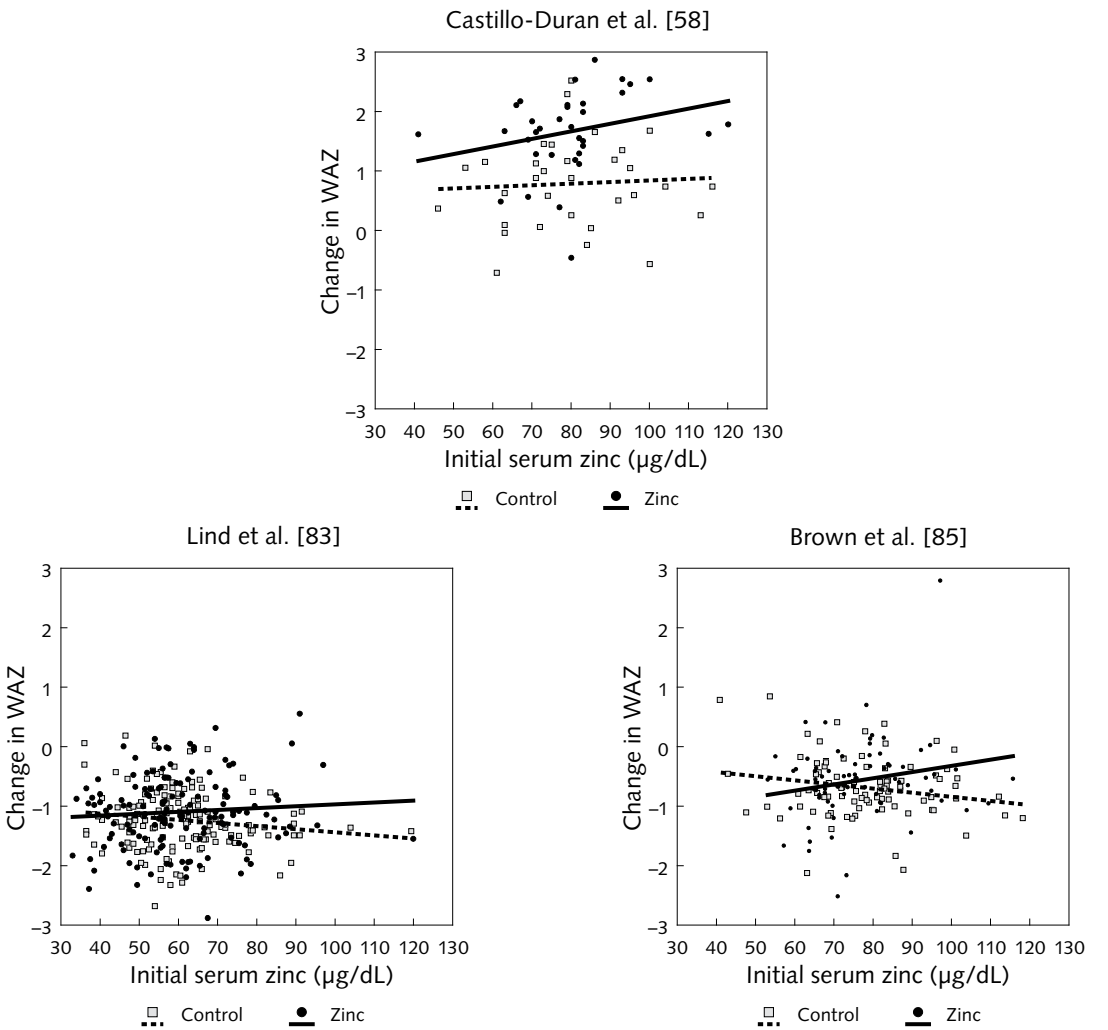

FIG. 5. Change in weight-for-age z-scores (WAZ) of individuals in response to zinc supplementation according to initial serum zinc concentration in three zinc intervention trials [58, 83, 85] with significant effect of treatment or significant or marginally significant interaction between initial serum zinc concentration and treatment. See **table 7** for results of statistical analyses

one study [84], and there were no significant interactions between initial serum zinc concentrations and treatment group in any of the trials (**table 7**).

Only four of the available studies provided information on diarrhea incidence, and two provided information on diarrhea prevalence. Two studies also provided information on the incidence of acute lower respiratory tract infections (**table 8**). There were no significant main effects of zinc supplementation and no interactions between treatment group and initial serum zinc concentration in any of the studies.

In summary, the results of these pooled analyses are disappointing for several reasons. First, most of the available studies enrolled children who were not sufficiently growth retarded to respond to zinc supplementation, and very few of the studies found any positive main effects of zinc supplementation on growth. Second, very few studies were available that collected data on both initial serum zinc concentration and rates of morbidity following supplementation, and none of these studies found a beneficial impact of supplementation. Thus, the databases that were available for the pooled analyses were, for the most part, not adequate to test the hypotheses of interest. Finally, when we explored for interactions between initial serum zinc concentration and effect of treatment, the results were not consistent for the few studies that did show significant or marginally significant interactions, suggesting that these results may have been due to spurious associations. Thus, with the sets of information that are currently available, we conclude that individual serum zinc concentrations are not useful for identifying particular children who are more likely to respond to zinc supplementation with increased growth or reduced rates of infections. However, these types of pooled analyses should be repeated as more information becomes available from populations with higher rates of growth stunting and from studies that monitor morbidity outcomes.

Each of the studies included in the foregoing pooled analyses provided data on the children's final serum zinc concentrations, so we also explored the relations between initial serum zinc concentration and the change in this variable in response to the intervention (**fig. 6**, **table 6**). All but one of the studies found highly statistically significant positive effects of zinc supplementation on the change in serum zinc concentration (**table 9**). Almost all of the studies also found a significant negative relation between the initial serum zinc concentration and change in zinc concentration during the course of the study regardless of the assigned study group. However, there was only one significant interaction between treatment group and initial serum zinc concentration, indicating that the absolute effect of supplementation in each study was generally constant, regardless of the individuals' initial levels of serum zinc.

TABLE 8. Incidence and prevalence of diarrhea and incidence of lower respiratory infection in four zinc supplementation trials in children[1]

Reference	*N*	Group	Diarrhea incidence/100 days	Diarrhea prevalence/100 days	Lower respiratory infection incidence/100 days
Dirren et al. [57]	96	Zinc	2.0 ± 3.0	NA	NA
		Control	3.0 ± 4.2	NA	NA
Rosado et al. [64]	110	Zinc	0.2 ± 0.2	NA	NA
		Control	0.3 ± 0.4	NA	NA
Osendarp et al. [82]	301	Zinc	1.2 ± 1.2	5.9 ± 6.4	0.38 ± 0.67
		Control	1.1 ± 1.1	5.9 ± 6.6	0.34 ± 0.56
Brown et al. [85]	200	Zinc	3.5 ± 3.0	8.3 ± 8.7	0.25 ± 0.58
		Control	2.8 ± 2.0	5.3 ± 4.2	0.20 ± 0.42

NA, not available

[1] There were no significant differences ($p < .05$) between zinc-supplemented and control groups in any of the studies.

TABLE 9. Statistical significance (*p* values) from analysis of covariance with main effects of treatment group, initial serum zinc concentration, and their interaction as predictors of change in serum zinc concentration in nine zinc intervention trials

	Cavan et al. [56]	Dirren et al. [57]	Castillo-Duran et al. [58]	Ruz et al. [63]	Rosado et al. [64]	Osendarp et al. [82]	Lind et al. [83]	Brooks et al. [84]	Brown et al. [85]
Group	.0001	< .0001	.0003	.064	.0002	< .0001	< .0001	.0016	< .0001
Initial zinc	< .0001	< .0001	< .0001	.0006	.343	< .0001	< .0001	< .0001	< .0001
Group * initial zinc	.024	.391	.170	.677	.841	.954	.725	.212	.411

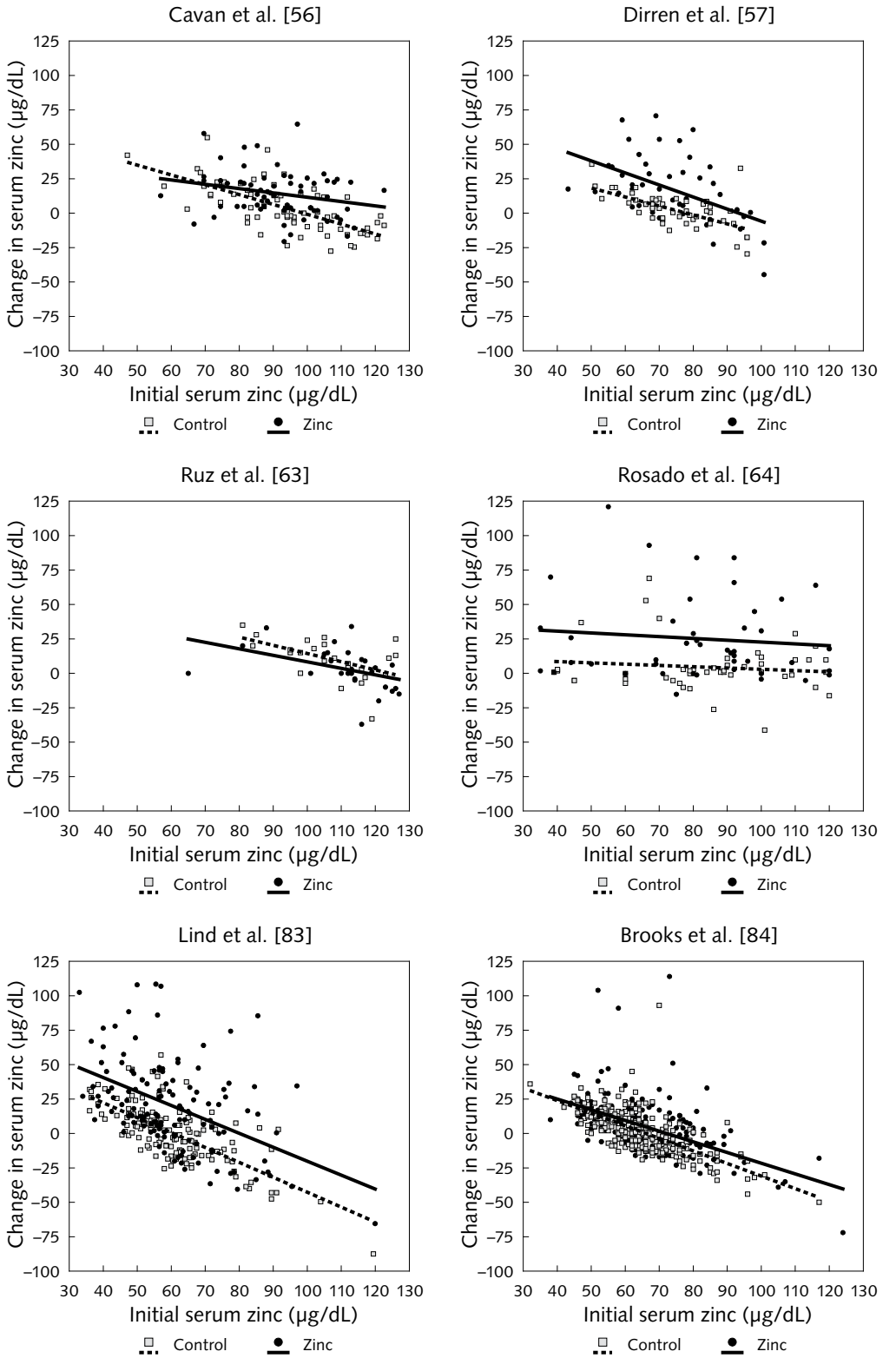

FIG. 6. Change in serum zinc concentration of individuals in response to zinc supplementation according to initial serum zinc concentration in nine zinc intervention trials [56–58, 63, 64, 82–85]. See **table 9** for results of statistical analyses

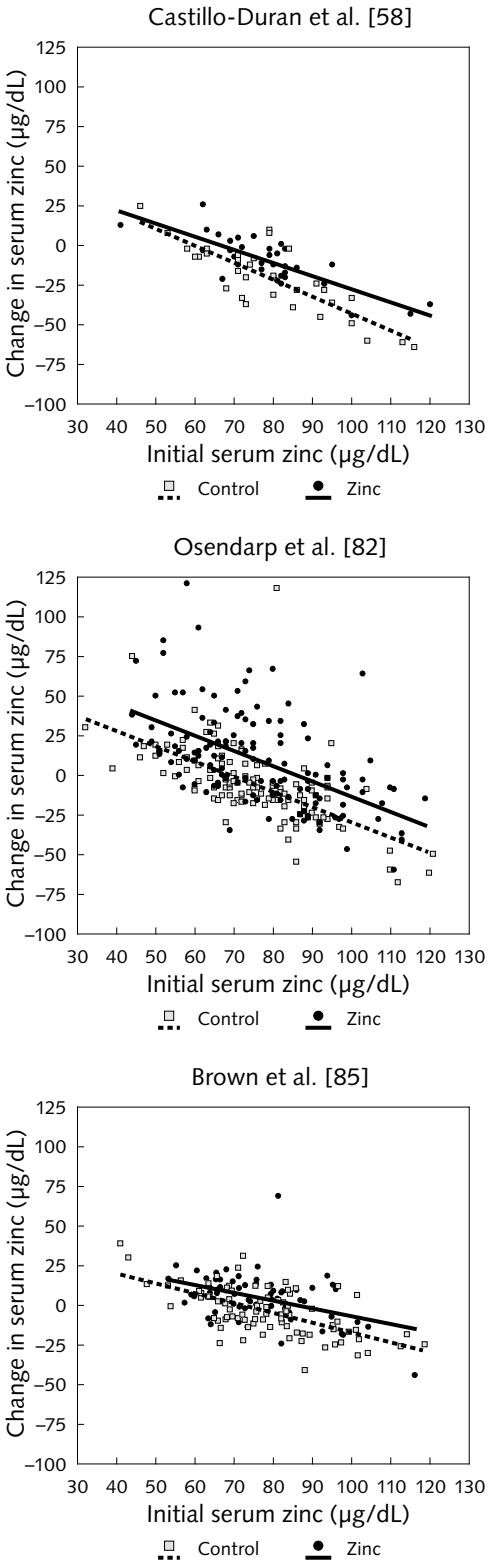

FIG. 6. *(continued)*

The negative relation between initial serum zinc concentration and change in concentration by time probably represents regression to the mean, because it occurred in both the intervention group and the control group.

Updated combined analyses of the relation between mean initial serum zinc concentration of the study population and mean growth responses to zinc supplementation in randomized intervention trials among children

Two meta-analyses have been published on the effect of zinc supplementation on children's growth [7, 81] The individual studies included in each of these meta-analyses differed somewhat, according to their availability at the time of the respective reviews and some of the study selection criteria. In particular, the earlier analysis included some studies of hospitalized, severely malnourished children and of premature infants, whereas the later one did not. For the present review, we reanalyzed the relation between the children's mean initial serum zinc concentration for individual studies and the effect sizes of their different growth outcomes for the studies that were incorporated in either of the two prior meta-analyses, along with the newly available studies that were included in the pooled analyses reported above. Only studies that were conducted in lower-income countries and provided data on mean initial serum zinc concentration were considered. Studies that only recruited children who were infected at baseline were eliminated, because their baseline serum zinc concentrations may have been altered by infection. Studies of severely malnourished children were not excluded, although these studies are indicated separately in the following figures. The relations between the mean serum zinc concentration and effect sizes for growth outcomes were analyzed statistically by using random-effects meta-regression analyses, which assume that the true effect size for each study is randomly distributed around a mean value that may or may not be a function of the initial serum zinc for the study.

The studies [52, 54–61, 63, 64, 82–94] included in this combined analysis of 24 zinc supplementation trials and children's growth are listed in **table 10**. The mean initial serum zinc concentrations of children enrolled in these studies ranged from 42 to 140 μg/dL. There was a slightly negative, but nonsignificant relation between the mean initial serum zinc concentrations and the effect sizes for change in height (N = 23 studies, r = –0.15, p = .67; **fig. 7**). Interestingly, there was a marginally significant, negative relation between the initial mean serum zinc concentration and the effect sizes for change in weight (N = 24 studies, r = –0.42, p = .071; **fig. 8**), and there was a significant negative relation between the initial serum values and the effect sizes for change in weight-for-height (N = 14 studies, r = –0.37, p = .023; **fig. 9**). Thus, despite the

TABLE 10. Characteristics of studies and study subjects included in a combined analysis of zinc supplementation trials and children's growth

Reference	Country	*N*	Zinc supplementation			Initial characteristics of study subjects (mean)					
			Dose (mg)	Frequency	Duration (mo)	Age (yr)	Serum zinc (μg/dL)	Weight (kg)	HAZ[1]	WAZ[1]	WHZ[1]
Studies in nonseverely malnourished children											
Ronaghy et al. [87]	Iran	39	32	6 days/wk	16	13	71	31	−2.8	−1.9	NA
Hong [54]	China	83	7.3	Daily	3	0.01	86	3.2	−0.3	−0.3	-0.4
Hong et al. [55]	China	65	7.6	Daily	6	0.01	86	3.3	−0.3	−0.02	−0.1
Udomkesmalee et al. [52]	Thailand	68	25	5 days/wk	6	9.3	86	23	−1.6	−1.5	−0.6
Cavan et al. [56]	Guatemala	162	10	5 days/wk	6	6.8	93	19.7	−1.5	−1.0	0.06
Bates et al. [93]	Gambia	109	70	2 days/wk	15	1.5	99	8.9	−1.5	−1.8	−1.4
Dirren et al. [57]	Ecuador	96	10	5 days/wk	15	2.6	74	11.2	−2.5	−1.8	−0.01
Castillo-Duran et al. [94]	Chile	42	10	Daily	12	8.7	114	NA	−2.5	NA	NA
Castillo-Duran et al. [58]	Chile	68	3	Daily	6	0.004	79	2.3	−1.0	−2.0	−1.3
Sempertegui et al. [59]	Ecuador	50	10	Daily	2	3.5	87	NA	−2.0	−1.4	−0.2
Friis et al. [60]	Zimbabwe	191	30	3–4 days/wk	3	10.1	79	26.6	−1.0	−1.2	−0.7
Ruz et al. [63]	Chile	98	10	Daily	14	3.3	110	15.5	−0.5	0.1	0.7
Rosado et al. [64]	Mexico	109	20	5 days/wk	12	2.4	83	11.1	−1.7	−1.6	−0.5
Smith et al. [61]	Belize	22	70	Weekly	6	4.0	75	14	−2.4	−1.3	0.10
Sayeg Porto et al. [92]	Brazil	21	—	Daily	6	9.9	101	21.2	−2.7	−2.1	−0.8
Osendarp et al. [82]	Bangladesh	301	5	Daily	5	0.08	76	3.5	−1.0	−1.3	−0.6
Lind et al. [83]	Indonesia	326	10	Daily	6	0.5	60	7.3	−0.4	−0.4	−0.01
Brooks et al. [84]	Bangladesh	638	70	Weekly	12	0.6	65	6.3	−1.3	−1.6	−0.7
Brown et al. [85]	Peru	200	3	Daily	6	0.6	77	7.6	−1.2	−0.8	0.4
Studies in severely malnourished children											
Gatheru et al. [90]	Kenya	82	40	Daily	0.3	1.7	42	8	NA	−2.9	NA
Khanum et al. [91]	Bangladesh	60	40	Daily	0.8	2.4	52	NA	−4.8	−4.4	−2.1
Simmer et al. [88]	Bangladesh	23	50	Daily	0.5	3.2	64	7.3	−4.7	−4.7	−3.1
Golden and Golden [89]	Jamaica	11	4.5	Daily	1.5	1.2	67	5.0	−3.8	−5.4	−4.1
Schlesinger et al. [86]	Chile	39	11	Daily	3.5	0.6	140	5.1	−3.2	−3.2	−1.0

HAZ, height-for-age z-score; WAZ, weight-for-age z-score; WHZ, weight-for-height z-score; NA, not available
[1]Actual results or estimated scores from median or mean height, weight, and age.

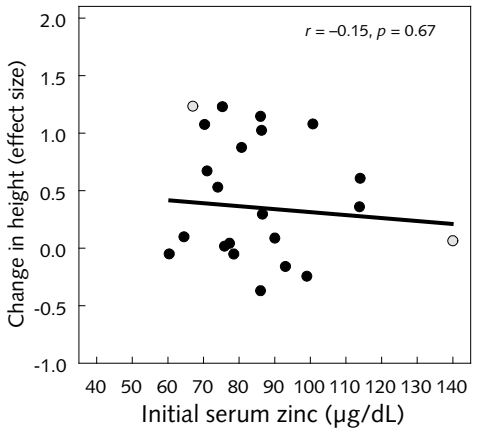

FIG. 7. Relation between mean initial serum zinc concentration and effect size for mean change in height following zinc supplementation in 22 intervention trials. Filled circles, studies in nonseverely malnourished children; open circles, studies in severely malnourished children. See **table 10** for details of study characteristics

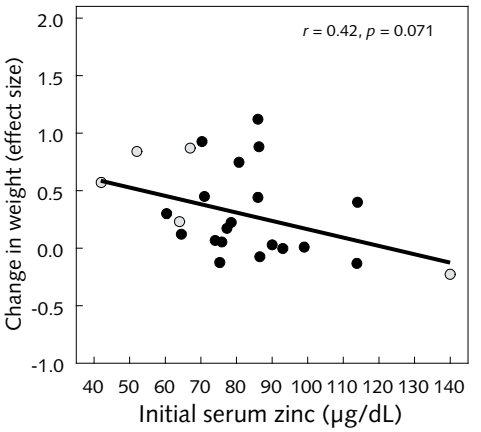

FIG. 8. Relation between mean initial serum zinc concentration and effect size for mean change in weight following zinc supplementation in 24 intervention trials. Filled circles, studies in nonseverely malnourished children; open circles, studies in severely malnourished children. See **table 10** for details of study characteristics

small sample sizes available for these analyses (in which the unit of analysis is the individual studies), the baseline (preintervention) mean serum zinc concentration of the study population does seem to indicate which groups of subjects are more likely to respond to supplementation with increased weight gain. These results do not seem to be unduly influenced by the inclusion of studies of severely malnourished children.

Reference data for serum zinc concentration

There are two possible strategies for defining normal ranges of serum zinc concentration. One approach is to use a statistical definition, based on the observed distribution of serum zinc concentrations among presumably healthy, well-nourished individuals to define upper and lower cutoffs, using either the 2.5th and 97.5th percentiles or the first and 99th percentiles of the observed reference population distributions. A second, more physiological approach is to determine a level (or levels) of serum zinc concentration below (and/or above) which some undesirable functional outcome occurs, such as impaired growth, increased morbidity, or unfavorable alterations of metabolism. To date, no studies have systematically examined possible relationships between serum zinc concentration and functional outcomes, so cutoffs for normal serum zinc concentration are currently based on statistical definitions derived from reference population data.

Reference cutoffs derived from NHANES II

As indicated above, information on normal serum zinc concentration can be drawn from surveys of representative samples of presumably well-nourished

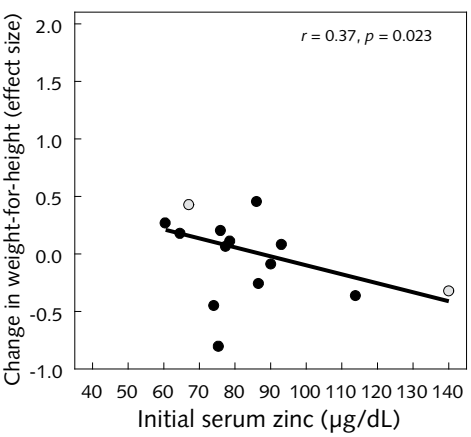

FIG. 9. Relation between mean initial serum zinc concentration and effect size for mean change in weight-for-height following zinc supplementation in 14 intervention trials. Filled circles, studies in nonseverely malnourished children; open circles, studies in severely malnourished children. See **table 10** for details of study characteristics

populations. There are few such databases available for analysis, but the NHANES II survey, which was conducted in the United States from 1976 to 1980, provides extensive information that has been used for this purpose. Specifically, information on serum zinc concentration was collected from a representative sample of persons 3 to 74 years of age, using standardized specimen collection and laboratory procedures, as previously reported [95, 96]. Hotz et al. [6] reanalyzed the results obtained from 14,770 of the NHANES II subjects for whom serum zinc data were available, taking into account several confounding factors, such as age and sex, pregnancy status and oral contracep-

TABLE 11. Suggested lower cutoffs (2.5% percentiles) for the assessment of serum zinc concentration in population studies, derived from NHANES II [6]

	Serum zinc (μg/dL [μmol/L])[1]		
		≥ 10 yr	
Time of measurement	< 10 yr	Females	Males
Morning fasting[2]	NA	70 (10.7)	74 (11.3)
Morning nonfasting	65 (9.9)	66 (10.1)	70 (10.7)
Afternoon	57 (8.7)	59 (9.0)	61 (9.3)

NA, not available
[1] Conversion factor: μmol/L = μg/dL ÷ 6.54.
[2] Based on data from subjects aged ≥ 20 years.

tive use, presence of infection, time of day of blood collection, and fasting status. Specific lower cutoffs for serum zinc concentration were developed for noninfected individuals using the 2.5th percentile based on sex, age, time of day of blood collection, and fasting status **(table 11)**, and these values have been adopted by IZiNCG [1]. The lower cutoffs for boys and girls aged 3 to 9 years were combined, since the difference between sexes was negligible [6].

Reference data for young children

Serum zinc concentration was not measured among children less than 3 years of age in the NHANES II survey, and currently there are no universally accepted reference values for early childhood. To determine whether different cutoffs should be applied for younger children, we reexamined the results of four studies that presented information for healthy children less than 5 years of age in Australia[97], Belgium [98], the United States [6], and Canada [99]. Two of these studies [97, 98] provided the results disaggregated by age subgroups (**table 12**). After reanalyzing the data from the study of Australian children, all of whom were less than 9 months of age, we found no significant differences in the mean concentrations (as estimated from the percentiles presented in the published paper) among the different age subgroups. By contrast, the study of Belgian children found significantly lower mean serum zinc concentrations among infants less than 12 months of age (77±19 μg/dL) than among older children (82±18 μg/dL), although there were no age-related differences among the subgroups of children less than1 year of age. Likewise, in the NHANES II data set, there were no significant differences in the mean values for children 36 to 47 months of age versus those 48 to 60 months of age. The study of Canadian children could not be used for this analysis, because the results were combined for all children 1 to 5 years of age. We found one longitudinal study of healthy Danish infants, which included only children less than 1 year of age. This study found no significant change in mean serum zinc concentration during the first 6 months of life (69 μg/dL at 6 months, $N = 50$), but a significant decrease from 6 to 9 months of age (55 μg/dL at 9 months of age; $N = 49$) [100]. These sets of results suggest that it should be possible to use a single reference value for all children greater than 1 year of age, although a different cutoff may be necessary for infants.

Unfortunately, the time of day of blood sampling, fasting state, and other possible confounders were not considered for sample collection or analysis in the foregoing studies from Australia, Belgium, and Canada. However, the mean serum zinc concentrations

TABLE 12. Mean serum zinc concentrations among healthy young children from Australia [97], Belgium [98], Canada [99], and the United States (NHANES II) [6][1]

Age group (mo)	Karr et al. [97][2]		Van Biervliet et al. [98][3]		Lockitch et al. [99][2]		Hotz et al. [6][3]	
	N	Zinc (μg/dL)	*N*	Zinc (μg/dL)	*N*	Zinc (μg/dL)	*N*	Zinc (μg/dL)
< 12		NA	135	77 ± 19		NA		NA
12–23	132	92 ± 16[4]	80	83 ± 18		NA		NA
24–35	109	88 ± 18	52	83 ± 20		NA		NA
36–47	99	88 ± 15	51	82 ± 19		NA	333	80 ± 13
48–62	127	92 ± 16	39	80 ± 18		NA	408	79 ± 15
12–62	467	91 ± 17[4]	222	82 ± 18	77	93 ± 13		NA

NA, not available
[1] The results of the statistical comparison are described in the text under Reference data for young children
[2] Data derived from 2.5th and 97.5th percentiles assuming normal distribution.
[3] Additional data supplied by the authors.
[4] This cell also includes data for children 9 to 11 months of age.

in these studies are intermediate to those found in the NHANES II survey for blood samples collected in the morning and afternoon from young children 3 to 4 years of age (**table 12**). Therefore, it seems reasonable to use the same lower cutoffs that were established for children less than 10 years, based on the NHANES II data (as presented in **table 11**), until more appropriate reference values are established.

Reference data for pregnancy and lactation

Several studies have concluded that serum zinc concentrations decrease during the course of pregnancy [6, 78, 101]. For example, in a large cross-sectional study of 3,448 pregnant women, Tamura et al. [101] found that serum zinc concentrations were progressively lower from early gestation until week 22, declining at a rate of ≈1.3 µg/dL (0.2 µmol/L) per week in all subjects combined, after which these concentrations remained constant. This decline may occur in response to hormonal changes and/or hemodilution, or may be a normal physiologic adjustment to pregnancy [101–103].

At present, there are no reference values available for serum zinc concentration during pregnancy. Only 61 pregnant women were available in the NHANES II survey. Because the mean serum zinc concentrations of pregnant women in this survey did not vary significantly according to fasting status and time of day of blood collection, Hotz et al. [6] combined data from all the women to maximize the sample size for further analysis. Lower cutoffs were derived by using the 2.5th percentile (**table 13**) [1, 6]. Because of the small sample size available for this analysis, more information is still needed to develop more reliable cutoffs for serum zinc concentration during pregnancy.

It was not possible to derive a reliable estimate of the 2.5th percentile for lactating women in the NHANES II data set due to the even more limited available sample size ($N = 23$). Nonetheless, the mean serum zinc concentration of lactating women was not as low as during pregnancy [6]. Until further reference data are available for this subgroup, it seems appropriate to apply the lower cutoffs derived from nonpregnant, nonlactating women (**table 11**) [1].

TABLE 13. Suggested lower cutoffs (2.5% percentiles) for the assessment of serum zinc concentration during pregnancy in population studies, derived from NHANES II [1, 6]

Trimester	Serum zinc (µg/dL [µmol/L])[1]
1	56 (8.6)
2	50 (7.6)
3	50 (7.6)

[1] Conversion factor: µmol/L = µg/dL ÷ 6.54.

Methodological issues in measuring serum zinc concentration

The collection and preparation of biological materials for zinc analysis should be performed in a controlled environment to ensure accurate assessment. Several precautions are needed to avoid contamination of samples, which can lead to false and inconsistent results for serum zinc concentration. IZiNCG [1, 104] recently published detailed recommendations for the collection, preparation, and analysis of serum zinc samples, so only a brief summary of these recommended procedures will be provided here. Variation in serum zinc results also may be caused by changes in intravascular pressure at the time of blood drawing, which varies with stress levels, position, and venous occlusion due to the use of tourniquet. The raised intravascular pressure causes the outward movement of fluid into the interstitial space, thereby increasing the concentration of serum proteins and zinc. To minimize this artifact, it is recommended that the subject should remain in a seated position for the blood-drawing procedure, and the tourniquet should be placed for a standardized length of time (preferably less than 1 minute).

Collection of morning fasting samples has been proposed previously as a standardized approach. However, this may be difficult in a large population-based survey, particularly in infants and young children. Therefore, it is simply recommended that the time of blood collection and the fasting status of all subjects be recorded. (Fasting is considered to be more than 8 hours since the last meal.) Serum zinc concentrations should be compared with the appropriate reference cutoff values, as given in **table 11**. When serum zinc concentration is assessed to evaluate the impact of zinc intervention programs, final blood collection has to occur before the end of the intervention, as studies in volunteers have shown a quick decrease in serum zinc concentration after cessation of zinc supplementation [43, 44].

Either serum or plasma can be used for analysis of circulating zinc concentrations. As discussed above, differences between zinc concentrations in plasma and serum appear to be partly dependent on the time between collection and separation. Plasma is commonly separated shortly after collection, but for serum adequate time is needed to allow samples to clot prior to separation. During this clotting time, zinc may be released from platelets and blood cells, leading to slightly increased serum zinc concentration. To avoid the time-related increase in serum or plasma zinc concentration prior to removing the cells, samples should be refrigerated or placed in a cold box prior to separation [105]. Anticoagulants, such as heparin or EDTA (ethylenediaminetetraacetic acid), which are required for separation of plasma, are potential sources of zinc contamination [106] and have been shown to vary in zinc binding selectivity [107]. Therefore, to facilitate

comparison of results from different studies or surveys, it is recommended that zinc-free heparin be chosen as the standard anticoagulant for use in plasma zinc analysis [1]. Regardless of the final choice of anticoagulant, the material should be analyzed prior to use to identify any possible zinc contamination. Where zinc contamination by anticoagulants is a concern, serum should be used instead of plasma.

A standardized clotting time of 30 to 40 minutes is recommended for serum samples, with specimens kept under refrigeration during this time [1]. For plasma samples, centrifugation procedures should be adequate (e.g., 2,000 to 3,000 × g for 10 to 15 minutes) to remove all blood cells, as these contain higher concentrations of zinc and create a risk of contamination [10]. Samples that are obviously hemolyzed should be discarded, as the zinc released from erythrocytes into the serum/plasma will falsely increase zinc concentration. For longer storage periods, serum and plasma samples should be kept frozen at –25°C or lower. In general, zinc will be stable in frozen samples for prolonged periods if dehydration of the samples is prevented by storing them in heat-sealed plastic bags [1]. For short-term storage prior to analysis (i.e., 2 to 3 weeks), refrigeration (4°C) of plasma or serum samples is acceptable.

Conclusions

An individual's serum zinc concentration is generally maintained within a fairly narrow range by homeostatic mechanisms. Therefore, this biomarker appears to be a useful indicator of individual zinc status only under more extreme dietary conditions. Nevertheless, studies show that changes in serum zinc concentration do occur, both following severe dietary restriction and during zinc supplementation. Moreover, careful metabolic studies show that there is a strong relationship between changes in total body zinc and changes in individual serum zinc concentration [13]. Thus, there is little doubt that serum zinc concentration does reflect an individual's zinc status, although abnormally low serum zinc concentrations in relation to population reference values may be detectable only after fairly large perturbations of zinc intake or zinc balance. Also, because many factors can affect serum zinc concentration independently of true zinc status, samples must be collected under carefully standardized conditions and interpreted appropriately.

Figure 10 illustrates the likely relationship between usual dietary intake and serum zinc concentration of adults based on the data compiled herein from multiple studies of dietary zinc restriction (as shown in **table 2**), studies of short-term and longer-term zinc supplementation (as shown in **table 3** and reported in the text), and data on the usual zinc intakes of a representative sample of US adult males (CSFII) [108] and the observed range of serum zinc concentrations among another representative sample of US adult males (NHANES II) [6]. This compilation of studies indicates that serum zinc concentrations fall abruptly when dietary zinc intakes are less than ~2 to 3 mg/day. With greater dietary zinc intakes, it appears that serum zinc concentration may rise continuously, but only slightly, reaching a plateau when intakes approach ~25 to 30 mg/day. This observation is consistent with other studies that show that the fractional absorption of zinc decreases logarithmically with greater doses of zinc supplementation, such that the net zinc absorption from supplements reaches a plateau with doses of around 20 mg/day [109].

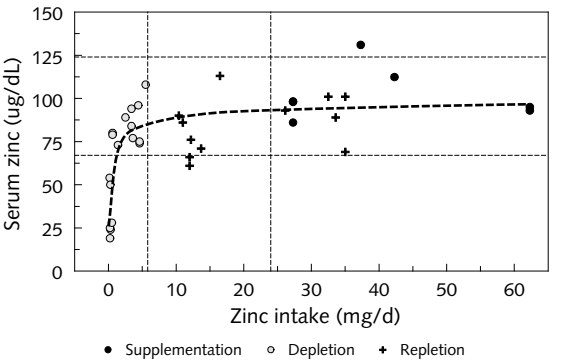

FIG. 10. Relation between mean dietary zinc intake and mean serum zinc concentration among adults in studies compiled for this review. Data compiled from multiple studies of dietary zinc restriction (open circles) and repletion (+) (as in **table 2**), studies of short-term and longer-term supplementation (filled circles, as in **table 3** and reported in text), and data on the usual zinc intakes of a representative sample of US adult men (CSFII) [108] and the observed range of serum zinc concentration among another representative sample of US adult men (NHANES II) [6]

Based on these observations, it is reasonable to conclude that the distribution of serum zinc concentrations in a population would reflect the range of usual dietary zinc intakes of the individuals who make up that population. Thus, in settings where more individuals are consuming diets that are inadequate to maintain zinc homeostasis, the distribution of serum zinc concentrations would shift to the left, and the percentage of individuals with low serum zinc concentrations would increase. Thus, the mean serum zinc concentration of the population or the percentage of individuals with low serum zinc concentration (using a statistical cutoff based on the observed distribution in a presumably well-nourished population) can be used as an indicator of the population's risk of zinc deficiency. Appropriate reference data from a presumably well-nourished population are available for this purpose [6], as summarized in **table 11**.

Individual serum zinc concentration does not seem

to predict an individual's ability to respond to a zinc intervention with improved functional outcomes, although the availability of relevant information is still very limited. On the other hand, there is some evidence to suggest that the mean serum zinc concentration of a population may foretell the population's growth-responsiveness to supplementation.

There is clear evidence that both individual and population mean serum zinc concentrations increase consistently during zinc supplementation, regardless of the initial level of serum zinc concentration. Thus, comparing baseline and postintervention serum zinc concentrations is a useful way of confirming whether the intervention is reaching the intended beneficiaries. Because serum zinc concentration increases during supplementation irrespective of the baseline preintervention values, it appears that these increases cannot be used as an indicator of prior zinc deficiency, as has been claimed by some authors.

In summary, serum zinc concentration is a useful biomarker of a population's risk of zinc deficiency, and this indicator can be used both to determine whether interventions are needed to enhance zinc status, and, once implemented, whether these interventions have been delivered successfully.

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