Chapter 1

Overview of Zinc Nutrition

1.1 Biological functions of zinc

Zinc is ubiquitously present throughout all biologic systems and has abundant and varied functions within these systems. These characteristics owe to its unusually versatile physicochemical properties. For example, zinc is able to assume a number of coordination numbers and geometries, which make it steriochemically adaptable to the functional needs of various ligands [1]. Moreover, under physiologic conditions, zinc is not subject to oxido-reductive reactions, so it is relatively non-toxic. These properties make zinc an ideal element to participate in catalytic, structural, and cellular regulatory functions [2].

More than 100 specific enzymes require zinc for their catalytic function [2]. If zinc is removed from the catalytic site, activity is lost; replacement of zinc restores activity. Unlike any other metal, examples of zinc-requiring enzymes are found in all six enzyme-classes (oxidoreductases, transferases, hydrolases, lysases, isomerases, and ligases) and include RNA polymerase, alcohol dehydrogenase, carbonic anhydrase, and alkaline phosphatase [1]. Zinc may provide activity to these enzymes by serving as an electron acceptor. In its structural role, zinc facilitates the folding of proteins into three-dimensional configurations, thus enabling their biologic activity. This folding often involves chelation of zinc with the amino acids cysteine and histidine and the formation of finger-like motifs, referred to as 'zinc fingers.' Metal transcription factor 1, retinoic acid receptor, and enzymes (such as copper-zinc superoxide dismutase) are examples of proteins that require zinc in a structural role. Processes regulated by zinc include expression of the metallothionein gene, apoptosis (or programmed cell death) and synaptic signaling.

In summary, zinc is the most ubiquitous of all trace elements involved in human metabolism. Zinc participates in all major biochemical pathways and plays multiple roles in the perpetuation of genetic material, including transcription of DNA, translation of RNA, and ultimately cellular division.

1.2 Tissue zinc distribution and reserves

The zinc content of the adult human body 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 in the body. However, most zinc is located in the fat-free mass, with about 30 mg zinc/kg tissue, almost all of which (> 95%) is intracellular. Due to the bulk of skeletal muscle and bone in the body, zinc in these tissues accounts for the majority (83%) of whole body zinc [3]. The concentration and total zinc content of various tissues, and the proportion contributed to total body zinc, are shown in table 1.1.

When total body zinc content is reduced during depletion, the loss of zinc is not uniform across all tissues. Skeletal muscle, skin, and heart zinc are maintained, while zinc levels decline in bone, liver, testes, and plasma [4]. It is not known what signals certain tissues to continue to release zinc during depletion while others retain zinc.

There are no conventional tissue reserves of zinc that can be released or sequestered quickly in response to variations in dietary supply. Nevertheless, it has been proposed that bone may serve as a passive reserve because some zinc may become available during normal turnover of osseous tissue. Thus, although the release of zinc from bone does not increase during deficiency [5], less of the zinc that is released during normal remodeling of bone may be re-deposited in the skeleton when the dietary supply is very low [6]. This passive reserve of zinc may be even more important in growing individuals, as bone turnover is more active. For example, young rats fed zinc-deficient diets for 24 days had nearly 50% less bone zinc content than did rats in a control group [7]. Interestingly, chicks fed higher zinc-containing diets during a baseline study period accumulated more skeletal zinc and were more resistant to growth failure during a subsequent period of very low zinc intake than were comparison animals fed a diet that was marginally adequate for zinc during the baseline period [8]. This suggests that intermittent zinc supplementation may be able to reduce the risk of

TABLE 1.1. Zinc content of major organs and tissues in an adult (70 kg) man[a]

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
Cartilage	34	30	1
Periarticular tissue	11	11	< 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/tissues	Variable	50	2
Total		2,240	100

a. adapted from Iyengar [3]

symptomatic zinc deficiency among people with poor dietary intakes.

1.3 Zinc metabolism

Zinc is released from food as free ions during digestion. These liberated ions may then bind to endogenously secreted ligands or to exogenous material in the intestinal lumen before their transcellular uptake in the distal duodenum and proximal jejunum [2]. Zinc transport into the enterocyte demonstrates saturable kinetics, suggesting involvement of a specific carrier mechanism. With high intakes, zinc is also absorbed through a passive, paracellular route. Other specific transporters, such as zinc transporter protein–1 (ZnTP-1) may facilitate passage of zinc across the basolateral membrane of the enterocyte into the portal circulation [9].

The portal system carries absorbed zinc directly to the liver, where it is taken up rapidly and released into the systemic circulation for delivery to other tissues [2]. About 70% of zinc in circulation is bound to albumin, and any conditions that alter serum albumin concentration have a secondary effect on serum zinc levels. For example, serum zinc concentration declines in concert with serum albumin during pregnancy, due to expansion in plasma volume. Serum zinc concentration also falls with the hypoalbuminemia that accompanies aging and protein-energy malnutrition. The concentration of circulating zinc is also altered by conditions that affect its uptake by tissues. For example, infections, acute trauma, and other stresses that induce increased secretion of cortisol and cytokines (such as interleukin 6) also augment tissue zinc uptake and thereby reduce serum zinc concentrations. During fasting, serum zinc concentrations rise due to release by muscle during catabolism; following meals, serum zinc levels decline progressively in association with hormonal changes and tissue uptake of circulating nutrients induced by fuel metabolism. Although serum zinc represents only 0.1% of the whole body zinc, the circulating zinc turns over rapidly (~ 150 times per day) to meet tissue needs. Notably, during the course of 24 hours, the equivalent of approximately one-fourth to one-third (~ 450 mg) of total body zinc exchanges between the bloodstream and other tissues [5].

Loss of zinc through the gastrointestinal tract accounts for approximately half of all zinc eliminated from the body. Considerable amounts of zinc (~3–5 mg) are secreted into the intestine from the pancreas following each meal, and biliary and intestinal secretions also contain sizeable amounts of zinc [10]. The total endogenous gastrointestinal zinc secretion may well exceed the amount consumed in the diet. However, much of the zinc that is secreted into the intestine is subsequently reabsorbed, and this process serves as an important point of regulation of zinc balance. Other routes of zinc excretion include the urine, which accounts for approximately 15% of total zinc losses, and epithelial cell desquamation, sweat, semen, hair, and menstrual blood, which together account for approximately 17% of total zinc losses [11]. The fecal loss of endogenous zinc from the body is less than 1 mg/d when a virtually zinc-free diet is consumed by healthy individuals studied under experimental conditions [10]. Under these conditions, urinary zinc losses decline by about 95%, largely due to the effects of glucagon and renal zinc transporters [2, 10].

In general, the amount of endogenous zinc excreted in the feces goes up as the total absorbed zinc increases; fecal excretion of endogenous zinc declines when either dietary zinc intake is reduced or zinc needs are increased due to growth or lactation [12–14]. When dietary zinc is decreased, the individual goes into negative zinc balance for a period of time before zinc balance is re-established at the lower level of intake [5]. This transient negative zinc balance results in a small loss of zinc from the exchangeable zinc pool; the amount lost depends on the length of time required to achieve zinc balance. This small loss of exchangeable zinc could have a subtle effect on zinc function, for example a reduction in immune function. However, functional consequences are generally not apparent until the capacity of these

adaptive mechanisms is exceeded.

1.4 Importance of zinc for human health

Given the diverse array of biologic functions of zinc, it is not surprising that multiple physiologic and metabolic functions, such as physical growth, immuno-competence, reproductive function, and neuro-behavioral development are all affected by zinc status. When the supply of dietary zinc is insufficient to support these functions, biochemical abnormalities and clinical signs may develop. Evidence regarding the effects of zinc status on physiologic function has been derived from three types of studies in human subjects: (1) evaluations of individuals with acrodermatitis enteropathica; (2) studies of the association between markers of zinc status and specific functions; and (3) clinical or community-based intervention trials. In some cases, studies in experimental animals also provide insights into the functional consequences of zinc deficiency.

Acrodermatitis enteropathica is a rare autosomal recessive genetic disorder that results in zinc malabsorption [15]. The classification of clinical signs associated with this disease has provided much insight into the functional outcomes of zinc deficiency, and therefore the physiologic roles of zinc. The clinical manifestations of acrodermatitis enteropathica and the frequency with which they have been observed are listed in table 1.2 [16]. Impairments of the dermal, gastrointestinal, neurologic, and immunologic systems predominate. Iatrogenic causes of zinc deficiency—such as prolonged administration of total parenteral nutrition with inadequate zinc content [17], long-term penicillamine therapy for Wilson's disease, which results in chelation of circulating zinc [18], and chlorthiazide administration [19], which increases urinary losses of zinc—have produced similar clinical signs as those described for acrodermatitis enteropathica.

TABLE 1.2. Clinical manifestations of acrodermatitis enteropathica[a]

Symptom	Frequency (%)
Dermatitis	84
Intermittent diarrhea	54
Alopecia	48
Growth retardation (stunting)	46
Weight loss (wasting)	43
Mood changes	39
Birth defects	31
Recurrent infections	30
Nail deformation	25
Miscarriage	23
Blepharoconjunctivitis	22
Death	20
Anorexia/hypogeusia	15
Photophobia	14
Pale skin	8
Neurological defects	3

a. adapted from Van Wouwe [16]

The severity and manifestations of frank zinc deficiency may vary at different ages [20]. In infants up to 2 months of age, diarrhea is a prominent symptom. Early zinc deficiency leads to cognitive function impairment, behavioral problems, mood changes, memory impairment, problems with spatial learning, and neuronal atrophy (optic and cerebellar) [21]. Skin problems become more frequent and gastrointestinal problems, anorexia, and mood changes less frequent as the child grows older [16]. Alopecia (hair loss), growth retardation, blepharoconjuctivitis (inflammation of eyelids and conjuctiva), and recurrent infections are common findings in school-aged children. Chronic non-healing leg ulcers and recurrent infections occur among the elderly [22].

Of the aforementioned types of study designs, placebo-controlled intervention trials provide the strongest inferences regarding the functional importance of zinc nutriture and the expected impact of interventions designed to enhance zinc status. When measurable functional changes occur in response to supplemental zinc in adequately controlled trials, preexisting zinc deficiency can be inferred, and the specific functions that are responsive to zinc can be identified. Unlike the situation with acrodermatitis enteropathica described above, where the myriad of clinical signs are likely to reflect severe deficiency, functional impairments identified in community-based trials may be more representative of mild or moderate deficiency. The range of functional impairments reported from these trials is described in the following sections.

It is important to recognize that the results of these studies may be affected by concurrent deficiency of other nutrients. Among the set of other nutrients that are commonly consumed in inadequate quantities, several also have demonstrated effects on growth [23], immune function [24], or neurologic or behavioral function [25]. While several of the zinc supplementation trials cited in this report have included other nutrients in the supplement formulation (e.g., iron plus folate, multi-micronutrients), zinc was the only factor differing between control and treatment groups in the results reported herein. In cases where zinc was given alone and compared with a placebo, the effect of zinc treatment may have been reduced or absent, either because zinc deficiency was not prevalent or because other concurrent nutrient deficiencies limited the impact of zinc [26, 27]. Considerations for the inclusion of zinc with other nutrition programs are discussed later in section 3.5 of chapter 3.

Immune function and risk of infection

The role of zinc in immune function has been reviewed in detail elsewhere [28, 29]. Zinc affects both nonspecific and specific immune function at a variety of levels. At least some effects of zinc on immune function are mediated via release of glucocorticoids, decreased thymulin activity, and possibly antioxidant properties. In terms of nonspecific immunity, zinc affects the integrity of epithelial barrier, and function of neutrophils, natural killer cells, monocytes, and macrophages. With regard to specific immunity, lymphopoenia and declined lymphocyte function occur, as do alterations in the balance of T helper cell populations (TH1 and TH2) and cytokine production.

Although most knowledge of the effects of zinc on immune function has been derived from experimental animals or in vitro models, several studies have shown that perturbations of zinc status can affect immune competence in adult human subjects [30–37]. For example, elderly subjects in higher-income countries who received supplemental zinc demonstrated improvements in delayed cutaneous hypersensitivity [34, 36], the number of circulating T cells, and serum IgG antibody response to tetanus toxoid [36]. In other studies of experimentally induced mild zinc deficiency among adults, reduced serum thymulin and IL-2 activity, and reductions in specific subpopulations of lymphocytes occurred during zinc depletion, and these returned to normal levels following zinc repletion [30, 37]. The specific links between zinc-related aspects of immune function and the incidence and severity of different infections are not well understood. Nevertheless, it can be assumed that the reported changes in immune function are clinically important because decreased rates of infection have been observed following zinc supplementation in population-based studies, as described below.

Diarrhea

Several studies have demonstrated reductions in the incidence and duration of acute and persistent diarrhea in zinc-supplemented children compared with their placebo-treated counterparts [38–41]. In two cases [42, 43], beneficial effects of supplemental zinc on the incidence of diarrhea extended beyond the actual period of zinc administration. Recently, a pooled analysis of randomized, controlled trials of zinc supplementation performed in nine lower-income countries in Latin America and the Caribbean, south and southeast Asia, and the western Pacific, indicated that supplemental zinc led to an 18% reduction in the incidence of diarrhea and a 25% reduction in diarrheal prevalence [44]. Notably, this analysis did not find differences in the effect of zinc by age, baseline serum zinc status, presence of wasting, or sex, suggesting that the benefits of zinc supplementation are likely to occur in all subgroups of children living in areas where there is an elevated risk of zinc deficiency (see section 2.2 in chapter 2). Since the publication of the pooled analysis just cited, results from two additional zinc supplementation trials have been reported from Africa [45, 46]. Both of these trials demonstrated significant reductions in the incidence or number of days with diarrhea, thus confirming that the preventative effect of zinc on diarrheal infection is consistent across a wide range of geographic regions. It is noteworthy that the impact of supplemental zinc on reducing diarrheal morbidity is comparable to that observed in programs to improve water quality, water availability, and excreta disposal [47].

Respiratory infections

Reductions in the incidence of acute lower respiratory infections in response to zinc supplementation have also been documented [48, 49]. The recent pooled analysis of trials conducted in India, Jamaica, Peru, and Vietnam indicated an overall 41% reduction in the incidence of pneumonia among zinc-supplemented children [44].

Malaria

Randomized, placebo-controlled studies in Gambia [50] and Papua New Guinea [51] suggest that zinc may play a role in morbidity reduction related to *Plasmodium falciparum* infections. The trial conducted in Gambia demonstrated a 32% reduction in clinic visits due to *P. falciparum* infections among those given 70 mg zinc twice weekly for 18 months. Similarly, the trial in Papua New Guinea showed a 38% reduction in clinic visits attributable to *P. falciparum* parasitemia among pre-school children provided with 10 mg zinc daily [51]. In the latter study, zinc supplementation resulted in an even greater reduction (69%) in clinic-based malarial episodes with high densities of *P. falciparum* parasites in the blood (i.e., > 100,000/μl). On the other hand, one recent trial conducted in Burkina Faso did not find any reduction in episodes of falciparum malaria among children who received daily supplementation with 10 mg zinc for 6 months [45]. However, malaria episodes were identified using daily household visits in this study. The contrast in results may occur because zinc has an ameliorating effect on the severity of falciparum-related morbidity, possibly resulting in fewer clinic visits, but not necessarily fewer infections.

Mortality

Only limited information is available concerning the impact of zinc supplementation on child mortality, although several large-scale studies are currently in progress. In one study of full-term, small-for-gestational-age infants in north India, daily supplementation

of zinc (1–5 mg/day, from 15–30 days of age, followed by 5 mg/day, from 30 days of age and continued until reaching 269 days of age) significantly reduced mortality by 67% compared with a control group that did not receive zinc supplements [52]. Another, smaller trial of older children in Burkina Faso also found that mortality from all causes was reduced by more than 50% among those who received zinc supplements, although this difference was not statistically significant [45].

Growth and development

Given the multiple roles of zinc in DNA replication, RNA transcription, endocrine function and metabolic pathways, it is not surprising that the state of zinc nutriture affects growth and development. Although the primary mechanism(s) whereby zinc influences growth is uncertain, there is a large body of literature indicating that zinc depletion limits growth and development, as summarized in the following paragraphs.

Low-birthweight infants

Low-birthweight infants (i.e., those < 2500 g at birth) may be especially vulnerable to zinc deficiency. Two studies of low-birthweight infants have been undertaken in lower-income countries, and in both cases weight gain increased among those who received the zinc supplement [43, 53]. Responses in linear growth have been less consistent. Castillo-Duran et al. [53] reported increased growth among low-birthweight Chilean infants, but this was not seen in the study in Brazil [43], possibly because the zinc supplement (5 mg zinc/day) was given for only a short period (i.e., 8 weeks) in the latter study. In a study of very low-birthweight, premature infants in Canada [54], a significant increase in linear growth was reported, but only in the zinc-supplemented female infants.

Severely malnourished infants and children

Some of the earliest studies of zinc supplementation in severely malnourished children were carried out in Jamaica, where zinc supplementation (1.6–9.8 mg/kg body weight/day, for 2 weeks, starting between the 4th and 12th week after hospital admission) was found to increase weight gain and lean tissue synthesis compared to unsupplemented children [55, 56]. Subsequent trials in Bangladesh [57, 58] have likewise found greater weight gain among severely malnourished inpatients who received supplemental zinc (10 mg/kg body weight/day up to a maximum of 50 mg/day, for 3 weeks) during the course of nutritional rehabilitation. However, one group of investigators [59] who provided either 1.5 or 6 mg zinc/kg body weight/day for 15 or 30 days starting immediately after hospital admission, found that severely malnourished inpatients who received the higher dose of supplemental zinc had increased mortality compared with similar patients who received 1.5 mg zinc/kg body weight/day, suggesting that excessive zinc supplementation may increase the risk of severe complications.

There have been fewer reports of a positive effect of zinc supplementation on children s linear growth during recovery from severe malnutrition, perhaps because most of the available studies were too brief to detect significant changes in linear growth, which generally occurs only after recovery of weight deficits in these patients [60].

Infants and children

The effects of zinc supplementation on children's growth were examined in a recently completed meta-analysis of 33 randomized intervention trials that were conducted in pre-pubertal children [61]. Zinc supplementation produced highly significant positive responses in linear growth and weight gain (mean effect sizes of 0.30–0.35 SD units), but no effect on weight-for-height indices. Growth responses were greater in children with low initial weight-for-age or height-for-age Z-scores. Thus, the beneficial effect of zinc on children's growth may be limited to populations with evidence of pre-existing growth failure. The magnitude of the zinc-induced growth impact tended to be greater in studies that enrolled younger children, but these age-related differences were not statistically significant, possibly because of the limited number of studies available for analysis.

In some studies, zinc supplementation had a greater impact in males than in females [62–64], but this finding was not consistent in all of the trials that identified a significant impact of zinc supplementation. Males have a higher percentage of total body weight comprised of muscle, which in turn contains a higher content of zinc than fat. Additionally, the growth rate of males is generally higher than females, so their zinc requirements are probably greater.

Adolescents

The first cases of human zinc deficiency described in the 1960s were reported in male adolescents from the Middle East [65, 66]. In this group, zinc deficiency was characterized by delayed sexual development, short stature, anemia, enlargement of the liver and spleen, and abnormalities in skeletal maturation. Zinc supplementation resulted in significantly increased height, weight, bone development, and sexual maturation [26, 67]. Decreased sperm counts and testosterone levels were also observed during experimental zinc depletion among adolescent males [68]. Since these early studies, very few zinc supplementation trials have been performed in this age group.

In a study of Chilean adolescents with idiopathic short stature, zinc supplementation (10 mg zinc/day as zinc sulfate) for 12 months significantly increased height-for-age Z-scores in boys, but not in girls, com-

pared with their unsupplemented counterparts [69].

Maternal health and pregnancy outcome

Zinc supplementation trials during pregnancy have been reviewed extensively by Tamura and Goldenberg [70], Caulfield et al. [71], and King [72]. Adverse outcomes associated with zinc status in at least some of these trials include intrauterine growth retardation, low-birthweight, poor fetal neurobehavioral development, and increased neonatal morbidity. Adverse maternal outcomes include preterm delivery and pregnancy-induced hypertension. Other possible consequences of maternal zinc deficiency on pregnancy outcome have been suggested from clinical observations of women with acrodermatitis enteropathica and cross-sectional studies of maternal zinc status. Notable associative outcomes not observed in controlled trials include fetal congenital anomalies, intra- or postpartum hemorrhage, and prolonged labor.

Figure 1.1 summarizes the consequences of maternal zinc deficiency, as derived from the controlled trials (shaded), and other possible consequences determined from observational studies, with indication of possible relationships among the various outcomes.

Most of the zinc supplementation trials have measured only a subset of the possible outcomes described above. Of the 13 published, randomized, placebo-controlled zinc supplementation trials identified for this report, only four have been conducted in lower-income countries: South Africa [73], India [74], Peru [75], and Bangladesh [76]. Six of the 13 trials reported no effect on pregnancy outcomes measured. Two studies found significantly improved fetal growth, as measured by birth weight, one of which was carried out in the US among African American women with below average plasma zinc concentrations [77], and the other among poor urban Indian women [74]. Significant reductions in preterm deliveries were reported in three of the zinc supplementation trials [74, 77, 78], although only in women of normal body weight in the study of Cherry et al. [78]. Two studies observed reductions in the incidence of maternal complications [79, 80], although only one of these studies statistically analyzed the outcome data [79]; this study of adult Hispanic Californian women showed a significant reduction in pregnancy-induced hypertension. However, a similar study conducted among Hispanic adolescents in California did not show any effect of zinc supplementation on blood pressure [81].

Fetal and infant health have also been assessed as pregnancy outcome variables. In the Peruvian trial, inclusion of zinc with iron and folate in maternal

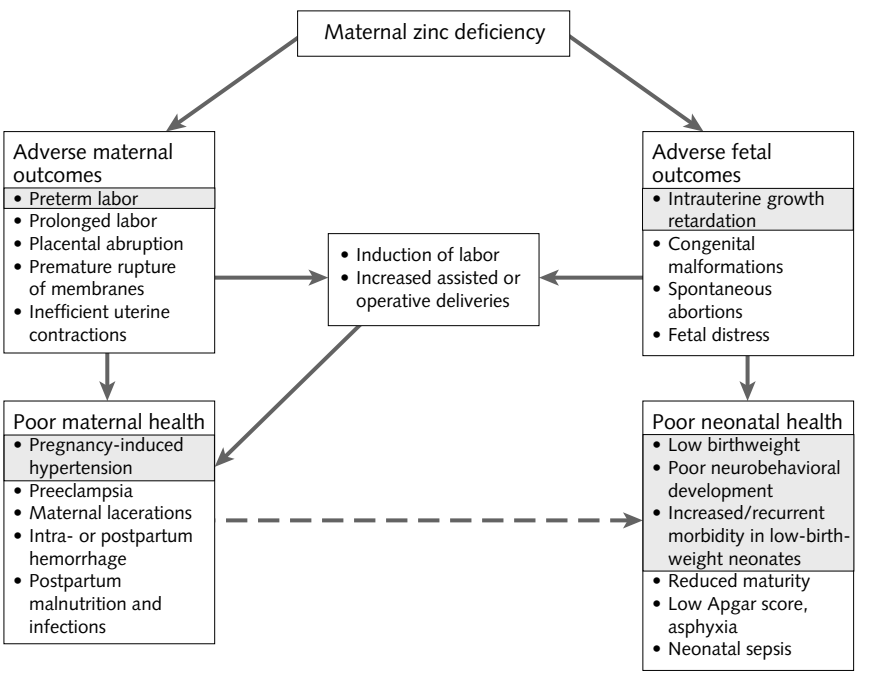

FIG. 1.1. Possible consequences of maternal zinc deficiency on birth outcomes and maternal and perinatal health. Outcomes observed in randomized, controlled zinc supplementation trials are shaded, indicating greater confidence in their association with zinc deficiency. Unshaded outcomes are those derived from observational studies of human maternal zinc status and pregnancy outcome, and their association with zinc deficiency can be considered only tentative.

supplements resulted in greater fetal heart rates and fetal movements, indicators of fetal neurobehavioral development, compared with those receiving only iron and folate [82]. Although no effects on birth weight were observed in the Bangladeshi trial, a follow-up study found that the risks of acute diarrhea, dysentery, and impetigo at 6 months were lower among infants whose mothers had received zinc supplements during pregnancy [83]. No effects on mental development or behavior, as assessed by the Bayley and modified Wolke's scales, respectively, were identified during later follow-up on the same infants at 13 months of age [84].

In summary, the results of zinc supplementation trials during pregnancy have been inconsistent. Several reasons may account for these discrepancies, including small sample sizes, varying degrees of underlying zinc deficiency, differing levels and periods of supplementation and measures of pregnancy outcome, and failure to account for confounding factors, including concurrent nutrient deficiencies. Clearly, more double-blind, placebo-controlled trials among pregnant women are needed in those lower-income countries where there is an elevated risk of zinc deficiency and where poor fetal growth is prevalent. Further, it is important to look beyond poor fetal growth alone as a possible outcome of maternal zinc deficiency, and to consider that suboptimal maternal zinc status may also manifest during postnatal development as poor infant growth and increased risk of infant morbidity and mortality.

Degenerative changes among the elderly

Several of the degenerative changes associated with aging may be due in part to zinc deficiency. These include a decline in immunocompetence [85], a decrease in taste acuity (hypoguesia) [86–88], delayed wound healing [89, 90], certain limitations of neurologic and psychologic function [91, 92], and deterioration of glucose tolerance [93, 94].

Neurobehavioral function

Some studies provide evidence that zinc deficiency contributes to compromised neuro-behavioral function among infants and children. One study among very low-birthweight infants showed improved developmental scores among those receiving supplemental zinc in formula [54]. In a zinc supplementation trial among infants in rural Guatemala, attainment of motor milestones was not affected, but activity patterns were improved with zinc supplementation [95]. A study among one-year-old Indian children also indicated that supplemental zinc plus vitamins resulted in higher activity levels than when vitamins alone were given [96]. School-aged children in China demonstrated improved neuropsychologic test performance with supplemental zinc, with the greatest improvements observed when other micronutrients were also given [27].

Zinc and appetite

Zinc deficiency has also been associated with reductions in appetite and may thereby contribute to deficiencies of other nutrients. Decreased food intake is observed early during the course of zinc depletion in animal models [97], and anorexia is a symptom of clinical zinc deficiency in humans [16]. Zinc-responsive anorexia has been demonstrated in population studies as well. A controlled zinc supplementation trial among low-income US children with evidence of mild zinc deficiency resulted in increased dietary intakes (137% energy intake of control group) after one-year of zinc supplement use (~ 4.2 mg zinc/day) [98]. A significant reduction in reported anorexia was also observed following zinc supplementation among stunted Ethiopian children [46]. However, because the high rates of morbidity were also responsive to zinc in the latter study, it is not possible to determine to what extent anorexia resulted primarily from zinc deficiency or was associated with morbidity. The mechanisms that link zinc status to appetite control are not well understood, and it is unclear whether appetite reduction precedes growth retardation or vice versa [99]. Nonetheless, the effects of zinc status on growth and appetite may be integrally related and both outcomes would likely be corrected simultaneously through improved zinc intakes.

1.5 Human zinc requirements

Since the mid 1990s, the World Health Organization/Food and Agriculture Organization/International Atomic Energy Association (WHO/FAO/IAEA) and the Food and Nutrition Board (FNB) of the US Institute of Medicine (IOM) have convened expert committees to develop estimates of human zinc requirements and to propose the corresponding dietary intakes that are needed to satisfy these requirements [11, 100, 101]. For most age and physiologic groups, the committees used a factorial method to estimate the average physiologic zinc requirement, which is defined as the amount of zinc that must be absorbed to offset the amount of endogenous zinc lost from both intestinal and non-intestinal sites. Non-intestinal sources of zinc loss include the urine, "surface losses" (desquamated skin, hair, nails, sweat), and, in adolescents and adults, semen or menstrual flow. In growing children and pregnant women, the amount of zinc retained in newly accrued tissue is also factored into total physiologic needs, and in lactating women the zinc transferred in breastmilk is added to the requirements.

When applying this conceptual framework, the committees considered the amounts of zinc lost from non-intestinal sites to be fixed, because these losses are generally constant across a wide range of zinc intakes [11]. On the other hand, intestinal excretion of endogenous zinc varies considerably in relation to the amount of absorbed zinc. Thus, the figure used for intestinal loss of endogenous zinc is appropriately estimated as the level that occurs when total absorbed zinc is just adequate to meet the theoretical physiologic needs. In the following paragraphs, we will describe in greater detail the conceptual frameworks and specific sources of data used by the various expert committees to estimate the physiologic requirements for absorbed zinc in different age and physiologic groups. In section 1.6, we will then discuss several issues concerning the absorption of dietary zinc and provide estimates of dietary zinc requirements and recommended dietary intakes.

1.5.1 Adult men

As indicated above, the physiologic requirement for zinc can be defined as the amount of zinc that must be absorbed to counterbalance the sum of endogenous zinc lost through all routes of excretion plus the amount of zinc retained in newly accrued tissue. The FNB/IOM estimated mean urinary zinc excretion by adult males to be 0.63 mg/day, based on the amounts reported from 17 previously published studies of individuals whose zinc intakes (4 to 25 mg/day) were within the range at which urinary concentrations are not influenced by zinc intake [11]. The corresponding figure published by WHO (0.3 mg/day) was based on just two studies [102, 103], which were conducted in men who had very low zinc intakes (0.8 to 3.6 mg/day); the observed amounts of urinary zinc were then inflated by 40% to account for the degree of reduction in urinary zinc excretion that occurred in response to the low intakes that were provided in those same studies. The authors of the present document conclude that the information derived by the FNB/IOM committee is more reliable because that report did the following: (1) reviewed a larger number of studies; (2) included only studies in which zinc intakes fell within the range in which urinary excretion is constant and which is likely to include the true physiologic requirement; and (3) provided more extensive documentation of the analytic process.

The FNB/IOM report considered just one study of integumental and sweat losses of zinc [104], which was carried out in 11 adult males whose mean zinc losses of 0.54 mg/day did not change in response to different levels of dietary zinc intakes ranging from 1.4 to 10.3 mg/day during periods of 28–35 days. Hence, this single figure was applied for surface losses of endogenous zinc. The WHO reports referred to a single earlier study of eight adult male volunteers [102] in whom surface losses of zinc declined from 0.49 mg/day when they consumed a diet containing 8.3 mg zinc per day to 0.28 mg/day when they consumed only 3.6 mg zinc per day. The IZiNCG SC concluded that although surface losses of zinc may decline with very restricted zinc intakes, it is preferable to estimate endogenous losses through this route when intakes are sufficient to meet physiologic needs. Thus, the IZiNCG SC decided that until more information becomes available it would be appropriate to use the study results applied by the FNB/IOM committee. However, as it is desirable to adjust zinc losses by this route for body size, as discussed in further detail below, the IZiNCG SC applied the figure per kg body weight (i.e., 6.5 μg/kg) as derived from Johnson et al [104]. Thus for a 65-kg adult man, the amount of zinc lost via the integument is 0.42 mg/day.

Unlike the WHO committees, which did not include an estimate of zinc loss in semen, the FNB/IOM committee considered information provided in two papers [104, 105] on the zinc concentration of semen and ejaculate volume of the same 11 volunteers for whom surface losses were reported above. The men's semen zinc concentrations (0.11 mg/ml) did not change with restricted dietary zinc intakes, and the ejaculate volume decreased significantly only at the lowest level of zinc intake (1.4 mg/day). Thus, the FNB/IOM committee decided to use a single figure of 0.10 mg zinc loss per day in semen, considering a mean ejaculate volume of 2.8 ml and a mean number of 2.45 ejaculations per week. The IZiNCG committee agrees with the general approach used by FNB/IOM, although more information is needed from a greater range of individuals, particularly on the mean daily volume of semen. Pending the availability of additional information, IZiNCG accepts the figure of 0.10 mg/day for average zinc loss in semen.

To estimate the intestinal loss of endogenous zinc, the WHO committees used the results from one study that reported a total fecal zinc excretion of 0.5 mg/day in six young adult males who received 0.28 mg zinc per day for 4–9 weeks [106]. This level of fecal zinc output was felt to represent the minimal amount that might be excreted after adaptation to a severely restricted diet. The WHO committee then inflated the figure for fecal losses by 40%, although the basis for this adjustment was not well substantiated in the WHO reports. Thus, the derivation of the figure of 0.8 mg/day used by WHO to indicate endogenous fecal zinc losses in adult men not adapted to low intakes of zinc is questionable.

The FNB/IOM committee considered a larger number of studies for their analyses and applied a somewhat different conceptual approach to estimate intestinal losses of endogenous zinc. In particular, the FNB/IOM committee identified 10 studies from 7 published articles that measured total absorbed zinc and intestinal excretion of endogenous zinc

using radio- or stable-isotope techniques, where the absorbed zinc was estimated from a whole day's dietary intake. Only studies that were conducted in North American or European men 19–50 years of age were accepted for inclusion in the analysis. After examining this information, the FNB/IOM committee concluded that, "excretion of endogenous zinc via the intestine is a major variable in the maintenance of zinc homeostasis and is strongly correlated with absorbed zinc." Therefore, to estimate the physiologic requirement for absorbed zinc, they decided that it would be necessary to consider the amount of intestinal losses of endogenous zinc that would occur when the absorbed zinc is just sufficient to offset the sum of all sources of endogenous zinc lost from both intestinal and non-intestinal sites.

Using this analytic approach, the FNB/IOM committee concluded that 2.57 mg/day of endogenous zinc would be excreted in feces when the amount of absorbed zinc is equivalent to the total losses of endogenous zinc from all sources, and the physiologic requirement for absorbed zinc in adult men is therefore 3.84 mg/day (i.e., with endogenous losses 0.63 mg/day from urine, 0.54 mg/day from integument and sweat, 0.10 mg/day from semen, and 2.57 mg/day in feces, it would be necessary to replace a total of 3.84 mg/day of endogenous losses of zinc). The estimate of the physiologic requirement for absorbed zinc derived from these data is illustrated graphically in figure 1.2, which is excerpted from the FNB/IOM report. The ten data points that the FNB/IOM committee used for these analyses are summarized in table 1.3 [13, 14, 103, 107–115]. As shown in the table, the studies employed diets that had a fairly low range of phytate:zinc ratios, including several that were formula diets prepared from purified ingredients.

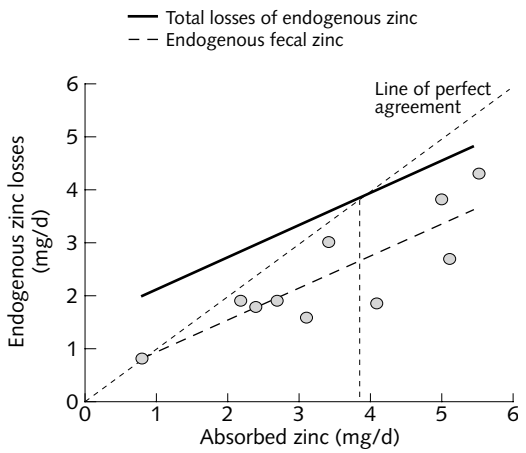

FIG. 1.2. Graphical representation of the conceptual model used by FNB/IOM [11] to estimate intestinal endogenous losses of zinc and total endogenous losses of zinc, when the amount of absorbed zinc is sufficient to offset all losses. The 10 data points represent mean data from 7 published studies of zinc absorption and intestinal losses of endogenous zinc in adult American or Western European men (19–50 years of age). The regression line (- - -) of the data points represents the relationship between total absorbed zinc and intestinal losses of endogenous zinc. The parallel line above (——) represents the total endogenous losses of zinc after adding the static losses through urine, integument, and semen. The line of perfect agreement (-----) indicates where total endogenous losses would be equal to the amount of absorbed zinc. The vertical line (⋮) is derived from the point where the total endogenous losses of zinc crosses with the line of perfect agreement, thus representing the physiologic requirement for absorbed zinc for North American adult men (i.e., 3.84 mg/day).

The IZiNCG SC concluded that the conceptual approach used by the FNB/IOM committee to estimate intestinal excretion of endogenous zinc is more appropriate than that used by WHO. However, the IZiNCG SC felt that, for the development of internationally relevant estimates of zinc requirements, it is appropriate and desirable to use the same methodologic types of studies as the FNB/IOM, but to expand the database used in this analysis to include all available studies of apparently healthy men and women, regardless of their age and nationality. Further, the IZiNCG SC initially restricted its analysis to those studies that used mixed diets prepared from common foods (i.e., studies in which supplemental zinc salts or exogenous phytate were added to the diet were excluded). Studies that manipulated several nutrients or isolated food components simultaneously were also excluded. Using these criteria, nine new studies were identified. For this new set of studies, the relationship between total absorbed zinc and fecal endogenous losses was examined by linear regression analysis, weighting by the sample size of the respective studies.

The relationship between total absorbed zinc and fecal losses of endogenous zinc was then explored to determine whether this relationship was inherently different between the new set of studies added and those used previously by the FNB/IOM. Notably, there were no significant differences in the slopes or intercepts of the respective best-fit lines, as shown in figure 1.3. Further, when all studies that employed diets prepared with common foods were then compared with those that employed semi-purified, formula diets, there were no significant differences in the relationship between absorbed zinc and endogenous fecal losses of zinc. Moreover, there were no significant differences in this relationship when studies conducted in men or women were compared. It may thus be interpreted that, once absorbed, the relationship between the amount of absorbed zinc and the amount of endogenous zinc excreted via the intestine is not dependent on the source of zinc ingested (i.e., zinc salts vs dietary zinc).

Because of the limited number of available studies in

TABLE 1.3. Summary of studies used by the FNB/IOM committee [11], the IZiNCG SC, or both committees, to estimate the relationship between total absorbed zinc and intestinal losses of endogenous zinc.

Source of data	Diet description	*n*	Total zinc intake (mg/day)	Phytate: zinc molar ratio	Intestinal losses of endogenous zinc (mg/day)	Total absorbed zinc (mg/day)
FNB/IOM only						
Lee et al. [13]	Soy protein-based, 6 months	8	4.1	21	1.8	2.4
Taylor et al. [103]	Semi-purified formula	5	5.6	0	1.9	2.2
Taylor et al. [103]	Semi-purified formula	5	0.9	0	0.8	0.8
Turnlund et al. [107]	Purified formula diet, young men	6	15.0	0	3.8	5.0
Turnlund et al. [108]	Semi-purified formula	4	15.0	0	2.7	5.1
FNB/IOM and IZiNCG						
Lee et al. [13]	Hospital	8	12.6	*	4.3	5.5
Jackson et al. [109]	Mixed	1	7.2	*	3.0	3.4
Hunt et al. [110]	Mixed	14	14.0	5	1.6	3.1
Wada et al. [111]	Mixed	6	16.5	4	1.9	4.1
Wada et al. [111]	Mixed	6	5.5	12	1.9	2.7
IZiNCG only						
Knudsen et al. [112]	High-fiber, mixed	8	10.7	6	2.6	3.1
Hunt et al. [113]	Lacto-ovo-vegetarian	21	9.1	18	0.8	2.4
Hunt et al. [113]	Mixed	21	11.1	5	1.4	3.7
Hunt et al. [114]	Low meat content	14	6.7	15	0.4	2.0
Hunt et al. [114]	High meat content	14	13.0	8	0.9	3.6
Sian et al. [14]	Plant-based	10	5.2	11	1.3	1.6
Sian et al. [14]	Mixed	10	8.1	10	2.3	2.8
Hunt et al. [110]	Mixed	14	7.8	5	2.0	2.3
Lowe et al. [115]	Mixed	6	7.0	8	2.0	2.0

* = Not available.

each of the foregoing subsets analyzed by the FNB/IOM and IZiNCG committees, respectively, the IZiNCG SC decided that the most reliable estimate of the relationship between total absorbed zinc and endogenous fecal zinc should be based on the full set of available information from all 19 studies (including the ten studies in the original FNB/IOM analysis and the nine additional studies identified by the IZiNCG SC). Figure 1.4 shows the relationship between total absorbed zinc and fecal endogenous zinc for the combined data set. Based on this combined set of information, 1.54 mg/day of endogenous zinc would be excreted in feces when the amount of absorbed zinc is equivalent to the total losses of endogenous zinc from all sources, and the physiologic requirement for absorbed zinc in adult men is therefore 2.69 mg/day. That is, with endogenous losses of 0.63 mg/day from urine, 0.42 mg/day from integument and sweat, 0.10 mg/day from semen, and 1.54 mg/day in feces, it would be necessary to replace a total of 2.69 mg/day of endogenous losses of zinc. Estimates for the various sources of endogenous losses of zinc for 65-kg adult men derived by the IZiNCG SC are summarized in table 1.4. For the purpose of comparison, the estimates derived by the WHO and FNB/IOM committees are also shown.

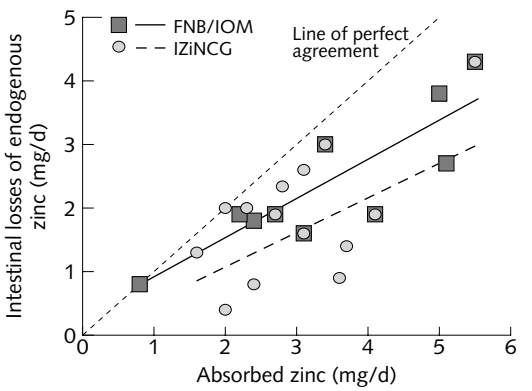

FIG. 1.3. Comparison of regression lines of total absorbed zinc and intestinal losses of endogenous zinc used by the FNB/IOM committee [11] and the IZiNCG SC for the estimation of physiologic zinc requirements.

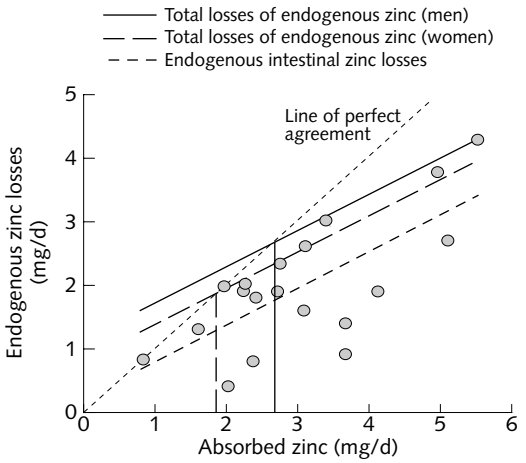

FIG. 1.4. Graphical representation of the model used by the IZiNCG SC in the present report to estimate intestinal endogenous losses of zinc and total endogenous losses of zinc, when the amount of absorbed zinc is sufficient to offset all losses. The 19 data points represent mean data from 12 published studies of zinc absorption and intestinal losses of endogenous zinc in adult men and women (19–50 years of age). The regression line (- - -) of the data points represents the relationship between total absorbed zinc and intestinal losses of endogenous zinc. The parallel lines above (—— men, – – women) represent the total endogenous losses of zinc after adding the static losses through urine, integument, and semen. The line of perfect agreement (-----) indicates where total endogenous losses would be equal to the amount of absorbed zinc. The vertical lines (| men, ¦ women) are derived from the point where the total endogenous losses of zinc crosses with the line of perfect agreement, thus representing the physiologic requirement for absorbed zinc for 65 kg adult men (i.e., 2.69 mg/day) and 55 kg adult women (i.e., 1.86 mg/day).

One final consideration for the derivation of physiologic zinc requirement estimates that are internationally applicable is that of reference body weights. The FNB/IOM Dietary Reference Intake committee applied reference body weights that are suitable for North American populations. However, the IZiNCG SC felt that the reference body weights adopted by the WHO, and based on the NCHS/CDC 1977 growth reference data, are more suitable for the present report. Based on the available data for body weight of subjects from 17 of the 19 studies used in the analysis of endogenous losses of intestinal zinc, the average body weights of the adult subjects (men, 71 kg; women, 61 kg) were intermediate to the reference body weights used by FNB/IOM (men, 75 kg; women, 65 kg) and those used by WHO (men, 65 kg; women, 55 kg). The IZiNCG SC thus felt it was unnecessary to correct intestinal losses according to different body weights among adult men. Similarly, it is unlikely that urinary losses of zinc would vary predictably according to different body weights among adults of different sizes. On the other hand, as integumental zinc losses may be more directly associated with body surface area and hence body size, it was felt appropriate to apply these losses to both adult men and women according to reference body weight.

TABLE 1.4. Estimated physiological requirements for absorbed zinc in adult men and women, as developed by expert committees of the WHO [99, 100], the US FNB/IOM [11], and as reviewed by IZiNCG

Endogenous zinc losses (mg/day) in adult men and women, by source of loss	Source of estimated physiological requirements		
	WHO	IOM	Revisions suggested by IZiNCG
Men			
Reference body weight (kg)	65	75	65
Urinary excretion	0.30	0.63	0.63
Integument	0.30	0.54	0.42
Semen	—	0.10	0.10
Total non-intestinal endogenous losses	0.60	1.27	1.15
Intestinal excretion of endogenous zinc	0.80	2.57	1.54
Total endogenous losses	1.40	3.84	2.69
Women			
Reference body weight (kg)	55	65	55
Urinary excretion	0.30	0.44	0.44
Integument	0.20	0.46	0.36
Menstrual blood	—	0.10	0
Total non-intestinal endogenous losses	0.50	1.00	0.80
Intestinal excretion of endogenous zinc	0.50	2.30	1.06
Total endogenous losses	1.00	3.30	1.86
Additional requirements for pregnancy (first, second, third trimesters)	0.1, 0.3, 0.7	0.16, 0.39, 0.63	0.70[a]
Additional requirements for lactation (0–3 months, 3–6 months, > 6 months)	1.4, 0.8, 0.5	1.35[b]	1.0[b]

a. A single estimate for additional zinc requirements is applied throughout pregnancy.
b. A single estimate for additional zinc requirements is applied throughout lactation.

1.5.2 Adult women

The foregoing conceptual issues concerning zinc

requirements generally apply to women as well as men. However, for several reasons, the specific figures used for endogenous zinc losses differ by sex, as described in the following paragraphs. The FNB/IOM committee examined the results of 10 published studies to calculate a mean urinary zinc excretion of 0.44 mg/day for adult females. The WHO committee relied on the results of just one study of women who were receiving very restricted zinc intakes, and then inflated the results by 40%, as described previously for men. For the same reasons described above, the IZiNCG SC has more confidence in the figure proposed by the FNB/IOM committee for urinary losses of zinc in women.

Each of the former expert committees estimated women's surface losses of zinc by adjusting for differences in body surface area to extrapolate from the data available from men. Because the IZiNCG SC preferred the original estimates for adult males proposed by the FNB/IOM committee, these same figures (adjusted for body size) were adopted for adult females (i.e., 0.0065 mg zinc/kg body weight/day × 55 kg = 0.36 mg zinc/day). There is little information on endogenous zinc losses in menstrual fluid. In one study [116], the average excretion of menstrual fluid during a single period was 60 g, and the mean zinc content was approximately 2.8 μg/g menstrual fluid or 154 μg during each menstrual period. This resulted in an average daily zinc loss of about 5 μg/day, considering the average cycle length of the women in the study. The WHO committees did not account for menstrual zinc losses, but the FNB/IOM committee estimated average menstrual losses to be 0.1 mg/day. However, this estimate was based on erroneous interpretation of data from the aforementioned study [116]. Because loss of zinc by this route is negligible, the IZiNCG SC concluded that it can be ignored in estimates of zinc requirements.

As discussed above, the IZiNCG SC prefers the conceptual approach used by the FNB/IOM committee to estimate fecal losses of endogenous zinc. Using this approach, the amount of intestinal losses of endogenous zinc that would occur in adult women is 1.06 mg/day when the amount of absorbed zinc is just sufficient to offset the sum of all sources of endogenous zinc loss (1.86 mg/day) from both intestinal and non-intestinal sites, assuming a reference body weight of 55 kg, as shown in figure 1.4. The endogenous losses of zinc by intestinal and non-intestinal routes for a 55-kg adult woman are summarized in table 1.4.

1.5.3 Children

Infants 0-6 months

Very little empirical information is available on zinc homeostasis, and, therefore, little information is available on physiologic requirements for absorbed zinc in infants less than 6 months of age. Moreover, there is some evidence that young infants may be able to acquire a portion of their zinc needs by mobilizing hepatic reserves accumulated during gestation [117], thereby possibly modifying their need for absorbed zinc from the diet. Studies of exclusively breastfed, healthy, term infants in the United States found no differences in the growth patterns of those who received zinc supplements or placebo from 4–6 months of age, suggesting that their zinc intake from breastmilk, along with any additional zinc contributed from pre-existing stores, was adequate to maintain normal growth [118]. On the other hand, European investigators found that infants 4–9 months of age of immigrant populations had increased rates of growth when supplemented with zinc (5 mg/day for three months) [119]. However, these latter infants were not exclusively breastfed, so it is conceivable that foods with lower zinc density were displacing breastmilk and/or these foods interfered with zinc absorption from breastmilk. Although more information is needed, it appears that zinc transfer from breastmilk is adequate for full-term, normal-birthweight, exclusively breastfed infants until about 6 months of age.

The FNB/IOM committee did not attempt to estimate physiologic requirements of zinc for young infants. Instead, the committee described presumably adequate intakes (AIs), based on the content of zinc in breastmilk at different ages and the average amount of milk consumed. It is important to note that because the zinc concentration of human milk declines sharply during the first few months post-partum, the total zinc transferred through milk falls from about 2.5 mg/day at one month to approximately 0.8 mg/day at six months [11]. Based on average figures for zinc transfer in breastmilk from 0–5 months post-partum, the FNB/IOM proposed 2.0 mg/day as the AI for infants in this age range. By contrast, the WHO committees developed estimates of physiologic zinc requirements of young infants by extrapolating from data for adults in relation to metabolic rate and then adding the amount of zinc incorporated into newly deposited tissue. Using this approach, the WHO committees suggested that the requirement for absorbed zinc from 0–5 months of age ranges from 0.7–1.3 mg/day, depending on age and sex. This compares with the estimate for absorbed zinc of ~ 0.7 mg/day developed by Krebs and Hambidge [120]. Considering the available information, IZiNCG concludes that breastmilk is a sufficient source of zinc for exclusively breastfed, normal-birthweight term infants until about 6 months of age. Non-exclusively breastfed infants need to absorb approximately 1.3 mg/day during the first three months of life and 0.7 mg/day during months 3–5.

Even less information is available on the zinc requirements of infants with low birth weight due to prematurity and/or intra-uterine growth retardation. Low-birthweight infants may have greater needs

for absorbed zinc than normal-birthweight infants because of limited hepatic reserves at birth and higher rates of postnatal growth. Notably, several researchers have found that low-birthweight infants had increased growth rates following zinc supplementation, which ranged from 2 to 5 mg/day of supplemental zinc in the available studies [43, 53, 54]. More research is needed to establish the physiologic requirements for zinc among low-birthweight infants.

Children 6 months to 18 years

The FNB/IOM used a factorial method to estimate physiologic zinc requirements of older infants and children. Losses of endogenous zinc from non-intestinal sites (i.e., urinary and surface losses) were estimated to be 0.014 mg/kg/day on the basis of extrapolations from adults per unit body weight. Fecal excretion of endogenous zinc was estimated to be 0.050 mg/kg/day for infants 6–11 months of age, based on empirical data obtained from breastfed infants, and 0.034 mg/kg/day for older children, as extrapolated from adult data. To these figures for endogenous losses were added the amount of zinc required for growth, which is estimated to be 0.020 mg/g of tissue gained. The figures for endogenous losses and zinc content of accrued tissue were then multiplied, respectively, by the reference body weight and the expected rate of weight gain at different ages. For male adolescents 14–18 years of age, an additional 0.1 mg/day was included in the estimated physiologic requirements to account for losses in semen. The WHO committees estimated physiologic zinc requirements throughout childhood by extrapolating from the data used to estimate endogenous losses in adults.

For the sake of consistency with the information discussed above for adults, the IZiNCG SC prefers to follow the factorial approach used by the FNB/IOM, but to base intestinal losses of endogenous zinc on the estimates derived by IZiNCG and to use the NCHS/CDC/WHO reference body weights, as summarized in table 1.5. Total endogenous zinc losses are calculated as 0.064 mg/kg/day for infants 6–11 months of age and 0.034 mg/kg/day for children one year of age and older (i.e., urinary losses 0.0075 mg/kg/day, surface losses 0.0065 mg/kg/day, and intestinal losses of 0.05 mg/kg/day for infants 6–11 months or 0.02 mg/kg/day for children 1 year and older). For example, children 6–11 months of age who have a reference body weight of 9 kg and expected weight gain of 13 g/day, need 0.576 mg/day (i.e., 9 kg × 0.064 mg/kg) to replace endogenous losses and 0.260 mg/day (13 g/day × 0.020 mg/g) for tissue accrual, resulting in a total physiologic requirement of 0.836 mg/day. The same procedure was used in children 1–3, 4–8, 9–13, and 14–18 years of age, using their respective reference body weights and rates of weight gain (table 1.5).

1.5.4 Pregnancy

Accrual of zinc in newly synthesized fetal and maternal

TABLE 1.5. Estimated physiologic requirements for absorbed zinc during childhood by age group and sex, and during pregnancy and lactation, as developed by expert committees of the WHO [100, 101], the US FNB/IOM [11], and as reviewed by IZiNCG

WHO			FNB/IOM			Revisions suggested by IZiNCG		
Age, sex	Reference weight (kg)	Physiologic requirement (mg/day)	Age, sex	Reference weight (kg)	Physiologic requirement (mg/day)	Age, sex	Reference weight (kg)	Physiologic requirement (mg/day)
6–12 mo	9	0.84	7–12 mo	9	0.84	6–11 mo	9	0.84
1–3 yrs	12	0.83	1–3 yrs	13	0.74	1–3 yrs	12	0.53
3–6 yrs	17	0.97	4–8 yrs	22	1.20	4–8 yrs	21	0.83
6–10 yrs	25	1.12						
10–12 yrs, M	35	1.40	9–13 yrs	40	2.12	9–13 yrs	38	1.53
10–12 yrs, F	37	1.26						
12–15 yrs, M	48	1.82						
12–15 yrs, F	48	1.55						
15–18 yrs, M	64	1.97	14–18 yrs, M	64	3.37	14–18 yrs, M	64	2.52
15–18 yrs, F	55	1.54	14–18 yrs, F	57	3.02	14–18 yrs, F	56	1.98
Pregnancy	—	2.27	Pregnancy (1st, 2nd, 3rd trimester)	—	4.12, 4.42, 5.02	Pregnancy	—	2.68
Lactation	—	2.89	Lactation (0–3, 3–6, 6–12 mo)	—	4.92, 3.82, 4.52	Lactation	—	2.98

tissue during pregnancy imposes an additional physiologic requirement for zinc. Both the FNB/IOM and WHO committees based these requirement estimates on data derived from Swanson and King [121]. The FNB/IOM estimated these additional zinc needs as 0.16 mg/day during the first trimester of pregnancy, 0.39 mg/day during the second trimester, and 0.63 mg/day during the third trimester; and WHO provided similar estimates of the respective amounts needed in each trimester, as follows: 0.1 mg/day during the first trimester, 0.3 mg/day during the second, and 0.7 mg/day during the third. To provide a single figure for the amount of additional zinc that needs to be absorbed during pregnancy, IZiNCG proposes using the figure of 0.7 mg/day, which covers the amount needed in the third trimester. It should be recognized, however, that this single figure overestimates the average requirements for absorbed zinc in the first and second trimesters. This additional amount of zinc needed during pregnancy should be added to the usual age-specific physiologic requirements for absorbed zinc of adolescent or adult women.

1.5.5 Lactation

The amount of zinc transferred from mother to infant in breastmilk must be added to lactating women's physiologic requirements for absorbed zinc. This amount is calculated by multiplying the average volume of milk transferred to the infant by the zinc concentration of human milk at different post-partum periods. To complete these calculations, the FNB/IOM committee applied milk volumes that were measured in US women during the first year post-partum (0.78 L/day). The FNB/IOM also summarized the results of 12 studies to provide age-specific information on the zinc concentration of human milk (2.75 mg/L at 4 weeks, 2.0 mg/L at 8 weeks, 1.5 mg/L at 12 weeks, and 1.2 mg/L at 24 weeks). Using these two sets of information, the FNB/IOM committee produced a single estimate of 1.35 mg/day for the average additional amount of absorbed zinc needed to support lactation, after discounting an assumed 1 mg/day of endogenous zinc that may become available during the first month post-partum because of involution of reproductive tissue. The WHO committees used data from just 3 of the 12 studies cited by FNB/IOM to estimate the zinc content of human milk (2.5 mg/L at 1 month, 0.9 mg/L at 3 months, and 0.7 mg/L at 4 months). WHO estimated that an additional 1.4 mg zinc/day is needed from 0–3 months post-partum, 0.8 mg/day from 3–6 months, and 0.5 mg/day thereafter.

Because women from developing countries typically breast feed for longer periods of time than do US women, and because breastmilk volume changes with infant age, the IZiNCG SC felt it would be desirable to derive estimates of zinc transfer in breastmilk using data on milk output from women in developing countries. Because the zinc concentration of human milk seems to be affected minimally, if at all, by maternal zinc status, it seems reasonable to use the more extensive set of data on milk zinc concentrations that was summarized by the FNB/IOM. Table 1.6 shows the mean amount of milk consumed by infants in developing countries at different ages, as published in a recent review [122]. These figures were multiplied by the mean zinc concentration of human milk for the same postpartum periods as reported by FNB/IOM [11] to estimate the total amount of zinc excreted in breastmilk. As indicated in table 1.6, the additional zinc needs imposed by lactation are considerable, especially during the early months of breastfeeding. On average, about 1 mg of additional zinc must be absorbed during lactation. Although more than this amount might be needed during the initial months, it is likely that this is partially offset by zinc released during involution of reproductive tissue. Thus, the figure of 1 mg/day seems to be a reasonable estimate of the additional amount of absorbed zinc needed throughout lactation. This additional amount should be added to the usual age-specific physiologic requirements for absorbed zinc of adolescent or adult women.

1.6 Dietary sources of zinc and suggested revisions of Recommended Daily Intakes

To translate physiologic requirements for absorbed zinc into recommendations for daily dietary zinc intakes, it is necessary to take into account the proportion of zinc in the diet that is absorbed by the intestine. In this section, we present the following: (1) a review of the dietary factors that affect zinc absorption; (2) estimates of zinc absorption from different diets; and (3) the derivation of dietary requirements, which incorporate information on the physiologic requirements for absorbed zinc (as described in section 1.5) and the estimated average zinc absorption.

TABLE 1.6. Amount of zinc transferred from mother to child in human milk, by infant age

Age range (months)	Milk volume (ml/day)[a]	Zinc concentration (mg/100 ml)[b]	Zinc amount (mg/day)
0–2	714	0.230	1.64
3–5	784	0.135	1.06
6–8	776	0.120	0.93
9–11	616	0.120	0.74
12–23	549	0.120	0.66

a. Data from Brown et al. [122]
b. Data from FNB/IOM [11]

1.6.1 Dietary sources of zinc and factors affecting the proportion of zinc available for absorption

Zinc occurs in a wide variety of food sources, but is found in highest concentrations in animal-source foods, particularly in the organs and/or flesh of beef, pork, poultry, fish and shellfish, and with lesser amounts in eggs and dairy products. Zinc content is relatively high in nuts, seeds, legumes, and whole-grain cereals, and is lower in tubers, refined cereals, fruits, and vegetables. Average ranges of zinc content (mg/100 g fresh weight) and zinc density (mg/100 kcal) in a variety of food sources are summarized in table 1.7, using information provided in the International MiniList (WorldFood Dietary Assessment System, 2.0, University of California, Berkeley; www.fao.org/infoods/software/worldfood.html).

As a result of physico-chemical interactions, dietary factors can alter the proportion of zinc that is available for absorption in the intestine by as much as 10-fold. Most of the available information on the effect of specific dietary factors on zinc absorption has been derived from studies measuring absorption from single test meals. However, it is questionable whether zinc absorption determined from single meal studies reflects the true proportion of zinc that would be absorbed from meals over the course of a whole day, as discussed below. Nonetheless, the large amount of data from these single meal studies is useful to identify the factors that do affect zinc absorption and to indicate their relative impact. Based on these single meal studies, the dietary components that demonstrate a substantial impact on the absorption of zinc are phytate and dietary calcium, which inhibit zinc absorption, and protein, which enhances absorption [123]. The total zinc content of a meal also influences the absorption of zinc; specifically, the percent absorption decreases with increasing intake of zinc [124], although the absolute amount of zinc absorbed increases.

Myo-inositol hexaphosphate (phytic acid) consists of a ring of six phosphate ester groups. Phytate is the magnesium, calcium, or potassium salt of phytic acid; the term "phytate" is used generically in this document to refer to the phytic acid molecule, as well as its salt forms. Phytate is a phosphorus storage molecule with a high natural content in seeds, including cereal grains, nuts, and legumes, and a lower content in other plant foods, such as fruits, leaves, and other vegetables. In legumes, phytate is uniformly distributed and associated with protein, whereas in cereal grains it is generally concentrated in the bran; in maize, the majority of phytate exists in the germ. Phytate is a strong chelator of minerals, including zinc. Because phytate cannot be digested or absorbed in the human intestinal tract, minerals bound to phytate also pass through the intestine unabsorbed. The inhibitory effect of phytate on zinc absorption appears to follow a dose-dependent response, and the phytate:zinc molar ratio of the diet has been used to estimate the proportion of absorbable zinc. The phytate:zinc molar ratio of foods or diets is calculated as follows:

$$\frac{\text{mg phytate}/660}{\text{mg zinc}/65.4},$$

TABLE 1.7. Zinc content, zinc density, phytate content, and phytate-to-zinc molar ratios of commonly consumed foods; data derived from the International MiniList[a]

	Zinc content		Phytate content	
Food groups	mg/100 g	mg/100 kcal	mg/100 g	Phytate:zinc molar ratio
Liver, kidney (beef, poultry)	4.2–6.1	2.7–3.8	0	0
Meat (beef, pork)	2.9–4.7	1.1–2.8	0	0
Poultry (chicken, duck, etc.)	1.8–3.0	0.6–1.4	0	0
Seafood (fish, etc.)	0.5–5.2	0.3–1.7	0	0
Eggs (chicken, duck)	1.1–1.4	0.7–0.8	0	0
Dairy (milk, cheese)	0.4–3.1	0.3–1.0	0	0
Seeds, nuts (sesame, pumpkin, almond, etc.)	2.9–7.8	0.5–1.4	1,760–4,710	22–88
Beans, lentils (soy, kidney bean, chickpea, etc.)	1.0–2.0	0.9–1.2	110–617	19–56
Whole-grain cereal (wheat, maize, brown rice, etc.)	0.5–3.2	0.4–0.9	211–618	22–53
Refined cereal grains (white flour, white rice, etc.)	0.4–0.8	0.2–0.4	30–439	16–54
Bread (white flour, yeast)	0.9	0.3	30	3
Fermented cassava root	0.7	0.2	70	10
Tubers	0.3–0.5	0.2–0.5	93–131	26–31
Vegetables	0.1–0.8	0.3–3.5	0–116	0–42
Fruits	0–0.2	0–0.6	0–63	0–31

a. WorldFood Dietary Assessment Program, 2.0, University of California, Berkeley

where 660 = the molecular weight of phytate, and
65.4 = the molecular weight of zinc.

The phytate content, and the phytate:zinc molar ratio of some commonly consumed foods are shown in table 1.7. In general, seeds, nuts, legumes, and unrefined cereal grains have the highest phytate:zinc molar ratios, which range from 22–88, while other plant foods have phytate:zinc molar ratios in the range of 0–42. Animal source foods do not contain phytate and therefore have a phytate:zinc molar ratio equivalent to zero.

Calcium also has an inhibitory effect on zinc absorption, although this may only occur when phytate is also present in the diet [123]. The inhibitory effect of calcium may result from the formation of insoluble calcium-zinc-phytate complexes in the intestinal tract. Both the total amount and type of protein in the diet influence zinc absorption. Increasing protein content results in a greater percent absorption of dietary zinc [124]. Animal protein, such as from meat and eggs, including whey protein, appear to have further enhancing effects on zinc absorption, although casein may be inhibitory [123].

1.6.2 Revised estimates of dietary requirements and recommended intakes for zinc

Two committees charged with developing dietary reference values, the FAO/WHO/IAEA Expert Consultation [100, 101] and the US Food and Nutrition Board/Institute of Medicine Standing Committee on the Evaluation of Dietary Reference Intakes [11], have estimated the percent absorption of dietary zinc. Both committees used a similar conceptual approach to develop these estimates, although the types of studies used in their analyses differ markedly. Each committee extracted data from studies of zinc absorption and plotted the mean amount of absorbed zinc against the total zinc ingested from the meal or diet being tested. A regression equation was then derived from the data and used to determine the amount of total zinc that would need to be ingested such that the amount of absorbed zinc would be equivalent to the physiologic requirement. This amount of total zinc ingested represents the daily "estimated average requirement" from the diet, or the EAR. The physiologic requirement for absorbed zinc divided by the associated total zinc intake ($\times$ 100%) represents the "critical" average zinc absorption, i.e., the percent of dietary zinc that is absorbed when the level of intake is just adequate to satisfy the physiologic requirement.

The IZiNCG SC reviewed the methods used by the WHO and FNB/IOM committees to estimate zinc absorption, taking into consideration the methodology used to measure absorption, the types of diets and subjects from which data were derived, as well as the models used to summarize these data.

Two general types of study designs have been used most commonly to estimate dietary zinc absorption: single-meal studies and total-diet studies. Single-meal studies are those that measure absorption from a single test meal, whereas total-diet studies measure zinc absorption from multiple meals consumed over 1 or more days. Zinc absorption data derived from total-diet studies have two main advantages. First, these studies label meals with either radioisotopes or stable-isotopes of zinc and, using fecal monitoring techniques, are able to estimate true zinc absorption for each individual by correcting for the simultaneous intestinal losses of endogenous zinc that occur during digestion. On the contrary, most single-meal studies have used radioisotope tracers and whole-body counting to measure zinc retention. This method can only estimate true absorption of zinc by applying an average correction factor for intestinal losses of endogenous zinc, derived from a separate study. Second, there is also evidence from studies of iron absorption that the percent iron absorption measured from a single test meal differs significantly from that measured from a total diet of similar composition as the single test meal [125, 126]. One of these studies [125] suggested that the enhancing and inhibiting effects of dietary factors (e.g., ascorbic acid, phytate) on iron absorption are exaggerated when measured from single test meals, whereas another suggested that iron absorption is much higher when measured from a breakfast meal than when the same test meal is consumed at later times during the day. It is possible that the same situation holds for zinc, although this has not been studied directly. For these reasons, the IZiNCG SC considered the total-diet study methodology as the reference method for measuring zinc absorption from the diet, particularly where the goal is to calculate dietary zinc requirements.

In determining its zinc absorption estimates, the WHO committee used data from a combination of single-meal studies and total-diet studies, although the specific studies used were not referenced [100]. Given the availability of zinc absorption studies at the time these estimates were made, it is likely that most of the data used were derived from single-meal studies. The WHO committee divided the available data into three categories according to the phytate:zinc molar ratio of the test meal or diet; where ratios of < 5, 5–15, and > 15 were considered to represent diets of relatively high, moderate, and low absorption levels, respectively. The process described above was applied to each of the three sets of data to derive zinc absorption estimates. A description of the diet types for each of the three categories and the associated absorption estimates are summarized in table 1.8. Because the specific studies used in these estimates were not reported, it is not possible to describe the specific meals or diets studied, nor the sex or geographic origin of the participants.

The FNB/IOM committee selected 10 data points

TABLE 1.8. Estimates of dietary zinc absorption, as developed by WHO [100, 101], FNB/IOM [11], and IZiNCG, and summaries of the data used to derive them

	WHO			IOM	IZiNCG	
Diet types represented	Highly refined[a]	Mixed/ refined vegetarian[b]	Unrefined[c]	Mixed, n = 5 Semi-purified, n = 4 EDTA-washed soy protein, n = 1	Mixed, n = 11 Refined vegetarian, n = 3	Unrefined, cereal-based, n = 1
Study type	Single meal & total diet			Total diet	Total diet	
Subjects	NA[d]	NA	NA	Men 19–50 yrs	Men & women 20+ yrs	
Phytate:zinc molar ratio	< 5	5–15	> 15	NA	4–18	> 18
Zinc absorption[e]	50%	30%	15%	41%	26% men 34% women	18% men 25% women

a. Refined diets low in cereal fiber, and where animal foods provide the principal source of protein. Includes semi-purified formula diets.
b. Mixed diets, and lacto-ovo-vegetarian diets that are not based on unrefined cereal grains or high extraction rate (> 90%) flours.
c. Cereal-based diets, with > 50% of energy intake from unrefined cereal grains or legumes and negligible intake of animal protein.
d. NA = not available
e. These figures represent the "critical" level of zinc absorption, or that which occurs when zinc intakes are just sufficient to meet physiologic requirements for absorbed zinc.

from 7 published studies of zinc absorption, using only total-diet studies, likely for the reasons described above [11]. The studies used by the FNB/IOM committee included only those of North American or Western European adult male subjects (19–50 years) and the diet types represented both mixed diets and semi-purified formula diets; all data points were considered in a single diet category, regardless of the composition of the diet. The same regression line relating zinc intake and total absorbed zinc that was derived from the studies of men was used to derive a zinc absorption estimate for women, based on their physiologic requirement for absorbed zinc. The studies included in the analysis and the absorption estimates derived are summarized in table 1.8.

For the reasons described above, the IZiNCG SC concurred with the FNB/IOM that total diet studies of zinc absorption provide the most valid estimates of dietary zinc absorption. Further, the IZiNCG SC felt it was important to consider differences in zinc absorption based on diet type and dietary content of known enhancers and inhibitors of zinc absorption, as done by the WHO, because—on the basis of their higher content of inhibitors of zinc absorption [127]—diets consumed habitually by a large proportion of the global population would be expected to have a lower fractional absorption of zinc than the diets considered by the FNB/IOM committee to establish their estimates. As a further step to ensure the appropriateness of the absorption estimates for internationally representative diet types, the IZiNCG SC rejected absorption studies that included semi-purified diets or other diets that included exogenous sources of zinc in the form of zinc salts. These types of diets do not represent typical diets consumed by populations, and the zinc absorption is expected to be higher from liquid formulas than from solid food matrices [128], and possibly higher from soluble zinc salts added exogenously than from an equivalent amount of zinc endogenous to the food. It also appeared unnecessary to exclude zinc absorption data derived from studies of women because the same regression curve is used to derive the zinc absorption estimate for both men and women. Finally, as the absorption estimates are intended for international use, geographic restriction on the origin of studies is unnecessary. Therefore, the selection criteria for the present IZiNCG analysis included the following: (1) radio- or stable-isotope studies that estimated true zinc absorption from total diets by correcting for intestinal losses of endogenous zinc; (2) studies of typical mixed, refined vegetarian, or unrefined, cereal-based diets, but not those that used semi-purified formula diets or diets with exogenous zinc salts added; and (3) studies with male or female adults, with no geographic limitations.

Initially, 17 data points from 11 published articles meeting the above criteria were identified. Information on the content of zinc, phytate, protein, and calcium of the study diets was derived either from the published article, by estimation from the published food composition of the study diet using a dietary assessment program, and/or from unpublished information obtained from the authors. Zinc and phytate contents of the study diets were available for 15 studies, and, calcium and protein contents were also available for 12 of these studies. The 15 data points, derived from 9 separate published articles, for which at least zinc and phytate contents were available were used in the final analyses [14, 110–115, 129, 130]. The diet types represented by these studies are summarized in table 1.8.

Data for these four dietary factors were log trans-

formed and a logit regression model was used to describe their relationship with the percentage of zinc intake that was absorbed. The logit model was used because, unlike the logarithmic transformation, it constrains predicted zinc absorption to between 0 and 100%. Neither protein nor calcium added significant predictive power, so the final model ($r^2 = 0.413$, $p < 0.001$) included only zinc and the phytate:zinc molar ratio, both of which were highly significant predictors of percent zinc absorption. Therefore, it appears to be valid to continue to use the phytate:zinc molar ratio to define diet types, with respect to zinc absorption. The prediction equation for proportion of absorbed zinc given as a fraction, using the dietary phytate:zinc molar ratio and zinc content derived from this model is*:

$$\text{Logit (fraction of absorbed zinc)} = 1.1365 - 0.6129 \times \ln(\text{mg zinc}) - 0.3164 \times \ln\left(\frac{\text{phytate:zinc}}{\text{molar ratio}}\right)$$

and

$$\text{Fraction of absorbed zinc} = \frac{\exp(\text{logit(fraction of absorbed zinc)})}{1 + \exp(\text{logit(fraction of absorbed zinc)})}$$

The range of phytate:zinc molar ratios was divided into two categories: (1) 4–18, which represents mixed or refined vegetarian diets, and (2) 18–30, representing unrefined, cereal-based diets (table 1.8). Using the prediction equation above, the phytate:zinc molar ratio was set at the midpoint of the range for the first diet category (i.e., 11) and then at the midpoint for the second diet category (i.e., 24), and the percent absorption of zinc associated with each level of total zinc intake between 4.2 and 16.5 mg (i.e., the range of values for zinc in the diets of the studies included in the analysis) was used to calculate the associated amounts of absorbed zinc. Curves were then generated showing the relationship between total zinc intake and absorbed zinc for the two diet categories (figure 1.5). Using these curves and the physiologic requirement for absorbed zinc of adult men (2.69 mg zinc/day) and women (1.86 mg zinc/day), as described in section 1.5, the amount of total zinc intake needed to meet this requirement was determined for each diet type, as shown in figure 1.5; this amount represents the dietary requirement. The percent zinc absorption represented

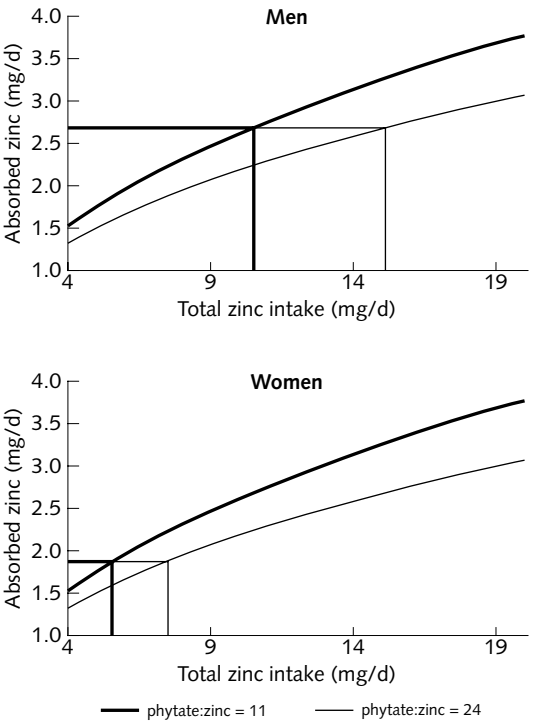

FIG. 1.5. Derivation of the estimated average requirement for men and women and the critical level of zinc absorption for mixed/refined vegetarian diets (P:Z = 11)and unrefined cereal-based diets (P:Z = 24), using the association between total zinc intake and absorbed zinc for each diet type and the physiologic requirement. Top panel is for men. Bottom panel is for women.

at this level of intake is referred to as the "critical" level of absorption. For example, the critical absorption level of an adult man consuming a mixed diet is calculated as 1.86 mg zinc/day (physiologic requirement) ÷ 10.4 mg zinc/day (dietary requirement) × 100% = 26%. Following this example, the calculated critical levels of zinc absorption were 26% for men and 34% for women for mixed/refined vegetarian diets, and 18% for men and 25% for women for unrefined, cereal-based diets.**

These estimates of zinc absorption should be considered as tentative until further data are available from a wider range of diet types, particularly from unrefined diets with a high phytate:zinc molar ratio (i.e., > 18), for which we identified only one data point from total diet studies of zinc absorption in adults. Notably, the estimate for zinc absorption from mixed or refined

* This prediction equation may be used to estimate fractional zinc absorption from adult diets. However, because the equation is dependent on total zinc intake it may not provide accurate estimates of fractional absorption from children's diets given that their dietary requirements and usual intakes are lower than for adults.

** The figures for zinc absorption which correspond to the amount of ingested zinc needed to meet the physiologic requirements of adult men and women with the higher reference body weights assumed by the FNB/IOM committee (i.e., men 75 kg and women 65 kg) are 24% for men and 31% for women consuming mixed/refined vegetarian diets and 16% for men and 22% for women consuming unrefined, cereal-based diets.

vegetarian diets in the present report is not as high as the estimate made by the FNB/IOM committee (41%) for North American diets or the upper estimate made by the WHO committee (50%) for highly refined diets, likely due to the IZiNCG SC's exclusion of data from semi-purified formula diets.

The IZiNCG SC felt that there was no justification at this time for assuming different levels of zinc absorption for different age groups, and therefore the mean of the absorption figures for adult men and women from each diet type were applied to children 1–18 years of age (i.e., 31% absorption from mixed/refined vegetarian diets and 23% from unrefined, cereal-based diets). The FNB/IOM committee applied lower figures for the critical absorption level for children than they used for adults. However, these data for children were based on results from only two studies of zinc absorption from single meals, and the average absorption from these two studies was 30%, similar to the absorption level that the IZiNCG SC derived for adults consuming mixed or refined vegetarian diets. The WHO committee did not propose different levels of zinc absorption for pregnant or lactating women. The FNB/IOM committee concluded that zinc absorption is not increased significantly during pregnancy. This conclusion was based on the results of one study in which zinc absorption was measured in women prior to conception and at 24–26 weeks and 34–36 weeks of gestation [12], with average zinc intakes of 15 mg/day throughout the study. In that study, the slight increase from 15% absorption at preconception to 19% absorption during pregnancy was statistically insignificant. Although these results do not exclude the possibility that zinc absorption is increased during pregnancy in women with lower zinc intakes, the IZiNCG SC concurs with the FNB/IOM committee that there is insufficient evidence to suggest a higher level of zinc absorption for pregnancy. Therefore, the IZiNCG SC also felt it was reasonable to apply the same zinc absorption estimates for pregnant as well as non-pregnant women, for each diet type. On the other hand, it does appear that zinc absorption increases significantly during lactation [12, 131–133]. In the study by Fung et al. [12], for example, absolute zinc absorption was increased by 10% among healthy, North American, lactating women who ingested 15 mg zinc per day from a combination of diet and supplements, compared to a non-pregnant, non-lactating, control group. The FNB/IOM committee suggested a level of absorption for lactating women that was 10% greater than that determined for pregnant women above (i.e., 27% + 10%), on the basis of the study by Fung et al. [12]. The study by Moser-Veillon et al. [131] reported a mean zinc intake of 8.0 mg/day by lactating women with 15% higher zinc absorption than measured in non-lactating women, and the study by Sian et al. [133] reported a mean zinc intake of 7.6 mg/day by lactating women, and a 19% greater absolute zinc absorption than determined in a separate study of non-lactating women. As there is insufficient information to determine whether these lactating women were meeting their zinc requirements, it is preferable to assume the figure of 10% increased absorption, as determined by Fung et al. [12]. Therefore, the IZiNCG SC estimated zinc absorption during lactation as 44% ($\geq$ 19 years of age) and 40% (14–18 years of age) for those consuming mixed or refined vegetarian diets and 35% ($\geq$ 19 years of age) and 32% (14–18 years of age) for those consuming unrefined, cereal-based diets.

Recommended derivation of the estimated average requirements

The estimates for absorption can now be applied to the physiologic requirements for absorbed zinc to derive EARs and Recommended Daily Allowances (RDA) for dietary intakes of zinc. The derivation of these Dietary Reference Intakes and their uses are described in the following paragraphs.

Different types of dietary reference intakes are derived depending on whether they are being used to assess the intakes of individuals or populations. Methods for calculating these reference intake values and their uses have been described previously by the FNB/IOM Dietary Reference Intake Committees [11], and the same terminology and methods are applied here. The EAR and the RDA for zinc developed by the IZiNCG SC for the purpose of international application are presented below. The upper limits for zinc intakes will be discussed in section 1.7.

The EAR represents the mean dietary requirement, or the dietary intake level at which 50% of individu-

TABLE 1.9. Revised estimated average requirement (EAR) for zinc by life stage and diet type, as suggested by IZiNCG

Age	Sex	Reference body weight (kg)	Revisions suggested by IZiNCG for EAR for zinc (mg/d)	
			Mixed or refined vegetarian diets	Unrefined, cereal-based diets
6–11 mo	M + F	9	3	4
1–3 yr	M + F	12	2	2
4–8 yr	M + F	21	3	4
9–13 yr	M + F	38	5	7
14–18 yr	M	64	8	11
14–18 yr	F	56	7	9
Pregnancy	F	—	9	12
Lactation	F	—	8	9
≥ 19 yr	M	65	10	15
≥ 19 yr	F	55	6	7
Pregnancy	F	—	8	10
Lactation	F	—	7	8

als would meet their physiologic requirement. The EAR is thus derived by dividing the mean physiologic requirement for absorbed zinc by the estimated average absorption of zinc. For example, the EAR for adult women (55 kg) consuming unrefined, cereal-based diets would be calculated as: 1.86 mg absorbed zinc/day ÷ 0.25 = 7.4 mg zinc/day, and rounded to 7 mg/day. The EAR for all age, sex, and life stage groups is given in table 1.9, for both mixed/refined vegetarian diets and for unrefined, cereal-based diets.

For consideration of breastfeeding infants 6–12 months of age, the FNB/IOM committee assumed that 50% of zinc in breastmilk is available for absorption [134] and that the average breastmilk consumption is 0.76 L/day. The amount of zinc required from complementary foods was then determined by difference. The EAR for breastfed children was calculated as the amount of zinc acquired from breastmilk plus the amount of zinc required from complementary foods, assuming 30% zinc absorption from the complementary diet. The WHO committee assumed that the absorption of zinc from breastmilk was 80%, although this estimate was not based on direct measures of absorption, and estimates of zinc intake from breastmilk in exclusively breastfed infants were derived from a single study of infants 1–3 months of age [135]. The IZiNCG committee used a similar approach as the FNB/IOM committee and also assumed 50% absorption of zinc from breastmilk. However, different estimates for average breastmilk consumption and milk zinc concentrations were used in each age group, as described in section 1.5 (table 1.6). Using these figures, the calculated total zinc requirements are somewhat lower than those that are derived when it is assumed that all dietary zinc is derived from complementary foods. The IZiNCG SC felt that it is unnecessary to provide two different sets of EARs for breastfed or non-breastfed children. Therefore, the slightly higher EARs, calculated assuming that all dietary zinc is derived from non-breastmilk sources, is provided in table 1.9.

The EAR has two primary uses, both of which apply to assessing the adequacy of dietary zinc intake by populations. First, the EAR can be used to evaluate the risk of inadequate intakes by a population by determining the proportion of the population whose intakes fall below the EAR. Second, the EAR may also be used for setting a *recommended mean intake for a population* (considering the observed variation in intakes of the population), such that only a small proportion of the population's intakes fall below the EAR. The application of the EAR in assessment of adequacy of zinc intakes by populations will be discussed in further detail in section 2.3.1, which describes methods to estimate the risk of inadequate zinc intakes.

Recommended derivation of the recommended daily allowances

It is not possible to know the true nutrient requirements of a particular individual, as these vary among individuals. However, when the normal variation of a physiologic nutrient requirement is known, the recommended intake level of that nutrient can be set at two standard deviations (SD) above the EAR. When calculated as such, almost all individuals (97.5%) whose intakes meet or exceed the recommended amount for any given nutrient will theoretically meet their physiologic requirement. This recommended intake level used for the purpose of individual assessments is commonly referred to as the RDA. The FNB/IOM committee has set RDAs for zinc. As for all nutrients for which adequate information on requirement distributions does not exist, zinc requirements were assumed to vary by ± 10% (i.e., the coefficient of variation (CV) of the requirement distribution is 10%) and the RDA was thus set as 120% (mean + 2 SD) of the EAR. The 1996 WHO report did not attempt to estimate the variability in zinc requirements. However, the more recent 2002 report assumed that the CV for zinc requirements was 25%, although the rationale for this assumption can be challenged. This estimate was based on two components. First of all, it was assumed that variation in the physiologic requirement for zinc is similar to that for protein (i.e., 12.5%) because both are related to tissue turnover and growth. An additional 12.5% was then added to account for variation in zinc absorption, resulting in an assumed total variability of ±25%, and the RDA was set at 150% of the EAR. Nevertheless it could be argued that the variation in protein digestibility is incorporated in the figure for variability in protein requirements, which was used as the basis for the estimate of the variability in zinc requirements, so it may not be justifiable to increase the estimate further to account for variation in zinc absorption. For this reason, the IZiNCG SC concluded that an estimate of ±12.5% for the variability in the zinc requirement might be more appropriate, and that is the figure that is adopted for the present report. The assumptions of each respective committee regarding the inter-individual variation in zinc requirements for women are the same as those for men.

The RDA for dietary zinc intakes was thus calculated as the EAR plus two times the CV (2 × 12.5), and is equivalent to 125% of the EAR. The RDAs for dietary zinc intakes derived by the IZiNCG SC are presented in table 1.10 for each sex and life-stage group. It is noteworthy that there is a negligible difference in the resultant RDAs when the CV of the physiologic requirement is assumed to be 10% vs. 12.5%.

TABLE 1.10. Revised recommended dietary allowances (RDAs) for zinc, by life stage and diet type, as suggested by IZiNCG

Age	Sex	Reference body weight (kg)	Revisions suggested by IZiNCG for RDA for zinc (mg/d)	
			Mixed or refined vegetarian diets	Unrefined, cereal-based diets
6–11 mo	M + F	9	4	5
1–3 yr	M + F	12	3	3
4–8 yr	M + F	21	4	5
9–13 yr	M + F	38	6	9
14–18 yr	M	64	10	14
14–18 yr	F	56	9	11
Pregnancy	F	—	11	15
Lactation	F	—	10	11
≥ 19 yr	M	65	13	19
≥ 19 yr	F	55	8	9
Pregnancy	F	—	10	13
Lactation	F	—	9	10

1.7 Zinc toxicity

Individuals may be exposed to high intakes of zinc, either through supplemental zinc or by contact with environmental zinc. Overt toxicity symptoms, such as nausea, vomiting, epigastric pain, diarrhea, lethargy and fatigue, may occur with acute, high zinc intakes [136]. Approximately 225–450 mg zinc is known to produce immediate vomiting in adults. Short-term exposure to very high levels of contaminant zinc (> 300 ppm) from the improper storage of food or beverages in galvanized vessels has caused acute gastroenteritis [136]. After receiving 150 mg zinc/day for six weeks, 26/47 human subjects reported gastrointestinal disturbances (abdominal cramps, nausea, and vomiting) [138].

Chronic overdosage of zinc, in the range of 100–300 mg zinc/day for adults, may induce copper deficiency [139] and alterations in the immune response and serum lipoprotein levels [140]. Some of these disturbances may also occur at lower doses (i.e., 50 mg zinc/day), although the data are conflicting and require confirmation [141–144]. Doses of 25–35 mg zinc/day in adults do not appear to pose a health hazard [145]. Intakes as low as 50 mg supplemental zinc/day affected copper metabolism, as measured by a decrease in erythrocyte copper-zinc SOD activity [144, 146]. However, the clinical significance of the depressed erythrocyte SOD activity is unknown. This same level of intake also resulted in a decline in serum ferritin concentration, which did not occur when 50 mg of iron was included with the daily zinc supplement [144]. Doses between 50 and 160 mg/day lowered the levels of high-density lipoprotein cholesterol in some, but not all studies [11, 147]. Among female subjects in one study who received 100 mg zinc/day, there was a significant reduction in high-density lipoprotein cholesterol levels after four weeks [142]. However, these levels returned to normal after eight weeks, suggesting that this effect may only be transient. A single case of a 13-month old child who received 16 mg zinc/day for six months and 24 mg zinc/day for one month was associated with copper deficiency attributed to excessive zinc intake [148].

The WHO/FAO/IAEA Expert Consultation derived upper limits for zinc intakes [100, 101]. These were based on the observation that 60 mg of supplemental zinc/day resulted in adverse interactions with other nutrients, although the source of these data was not provided, and it was considered that intakes should not exceed this amount. After accounting for a 25% possible variation in population intakes, the upper limit for males was set at 45 mg/day. Due to the limited availability of studies that looked at possible adverse effects of supplemental zinc, the upper limit for adult males was extrapolated to other age and sex groups based on differences in metabolic rates. These upper limits are shown in table 1.11. Although the case report of the 1-year-old child cited above was not included in the derivation of these upper limits, the 16 mg/day intake, after accounting for a 25% possible variation in intakes, is consistent with the 13 mg/day upper limit set by WHO [100].

The FNB/IOM committee also established upper tolerable limits for zinc [11]. This committee also based the upper limits on the studies that measured the effect of supplemental zinc intakes on measures of copper status, including erythrocyte superoxide dismutase (SOD) activity or the concentration of copper or ceruloplasmin in serum, but estimated the upper tolerable limits based on the Lowest Observed Adverse Effect Level (LOAEL) and the No Observed Adverse Effect Level (NOAEL). For adults, a LOAEL of 60 mg zinc/day (50 mg zinc/day from the supplement and an estimated 10 mg/day from the diet) was derived from the results of the study by Yadrick et al [144], as described above, and were supported by the data from Fischer et al. [146]. To take into account intra-individual variation in this response, an uncertainty factor of 1.5 was assumed, and this factor was used to extrapolate the LOAEL (60 mg zinc/day) to the NOAEL (40 mg zinc/day) for both male and female adults. For children, the upper limit was based on the results of just one available study [149]. This study measured serum copper and cholesterol concentrations in newborn infants receiving a formula that provided 5.8 mg zinc/L, compared with a formula with 1.8 mg zinc/L. No changes in measures of copper status were found in either group after 6 months. Based on an estimated consumption of 0.78 L formula/day, the formula with

TABLE 1.11. Upper limits or no observed adverse effects levels (NOAEL) for zinc intake by life stage, as developed by expert committees of the WHO [100, 101], the US FNB/IOM [11], and as reviewed by IZiNCG

WHO		FNB/IOM		Revisions suggested by IZiNCG	
Age/sex	Upper limit (mg/d)	Age/sex	Upper limit (mg/d)	Age/sex	No observed adverse effect level (mg/d)
0–6 mo	—	0–6 mo	4	0–5 mo	—
7–12 mo	13	7–12 mo	5	6–11 mo	6
1–3 yr	23	1–3 yr	7	1–3 yr	8
3–6 yr	23	4–8 yr	12	4–8 yr	14
6–10 yr	28				
10–12 yr, M	34	9–13 yr	23	9–13 yr	26
10–12 yr, F	32				
12–15 yr, M	40				
12–15 yr, F	36				
15–18 yr, M	48	14–18 yr, M	34	14–18 yr, M	44
15–18 yr, F	38	14–18 yr, F	34	14–18 yr, F	39
18–60+ yr, M	45	≥ 19 yr, M	40	≥ 19 yr, M	40[a]
18–60+ yr, F	35	≥ 19 yr, F	40	≥ 19 yr, F	40[a]

a. Represent upper limits.

5.8 mg zinc/L was estimated to provide an average intake of 4.5 mg zinc/day, and this figure was used as the NOAEL for infants 0–6 months of age. This was rounded down to 4 mg zinc/day for the upper limit; an uncertainty factor of 1 was applied. This upper limit was then extrapolated to older children based on differences in reference body weights (section 1.5). The upper limits derived by the FNB/IOM committee are presented in table 1.11.

The IZiNCG SC concurs with the upper limit of 40 mg zinc/day set for adults by the FNB/IOM, as derived from the LOAEL of 60 mg zinc/day; no further data on the effects of supplemental zinc on copper status in adults were found since the publication of the FNB/IOM report. Unfortunately, there is a lack of adequate data to better define the upper limits for children. By definition, the level of zinc intake described by the NOAEL does not exclude the possibility that chronic intakes of higher levels of zinc would also not cause an adverse effect. It is appears likely that the upper limits proposed by the FNB/IOM for young children (< 3 years of age) may be inappropriately low. This presents concern for the development of interventions to improve zinc intakes among this age group, because the margin between the RDA for zinc and the upper limit is rather narrow (only 0–5 mg zinc/day, depending on age and diet type). Further, it is apparent that a large proportion of US children have usual zinc intakes greater than the IOM/FNB upper limits. For example, the median intake of zinc by presumably healthy US infants 2–11 months of age from the diet is 5.5 mg/day (with or without consideration of supplement use) (NHANES III [150], whereas the upper limit for zinc for children in this age range set by FNB/IOM was 5 mg/day. The median zinc intake by children 1–3 years of age in the United States is 6.3 mg/day from the diet alone, and 6.4 mg/day when zinc supplements are also included, and the FNB/IOM upper limit for zinc for this age group is 7 mg/day. Although the proportion of children with intakes above the upper limit was not reported, it is likely that about half of children 2–11 months of age exceeded the upper limit of 5 mg/day and many children 1–3 years old would have exceeded the upper limit of 7 mg zinc/day. Given the unlikelihood that the described toxic effects of excessive zinc intakes occur in such a large proportion of children from this relatively healthy, US population, the degree of confidence in the upper limit is relatively low. Therefore, the IZiNCG SC is unsatisfied with the upper limits presented by the FNB/IOM for children, as these may have important implications on recommendations for the design of intervention strategies to improve zinc status among young children, particularly where supplements are used (section 3.1.2). For the reasons described above, IZiNCG will report only a NOAEL for children to indicate that insufficient data exists to set an upper limit with confidence.

Results from two community-based zinc supplementation studies in children have become available recently, and the IZiNCG SC felt it was important to take these into consideration in the calculation of the NOAEL. One study was conducted in India among children 6–30 months of age [151]. For a period of

4 months, children between 6 and 12 months of age received 10 mg zinc/day and those 1–2.5 years of age received 20 mg zinc/day. Plasma copper concentration was reported to be lower in the group that received supplemental zinc compared to the placebo group. The results for plasma copper were not presented according to age group, however, so it cannot be distinguished as to what dosage level and which age group was associated with changes in plasma copper concentration. The second study was conducted in Indonesia among children 6 months of age [152], who were provided with either 10 mg zinc or a placebo for 6 months. In this study, plasma copper concentration did not differ between the zinc supplemented group and the control group at the end of the supplementation period. The results from the Indonesian study may thus be used to set the NOAEL and derive upper limits for infants and older children. Considering unconsumed portions of the supplement monitored in the Indonesian study, the estimated intake of zinc from the supplement was 8.2 mg/day. However, as information was not available on the usual dietary zinc intake by the infants in the Indonesian study, the total zinc intake applied to the NOAEL will be somewhat underestimated. On the other hand, it was noted that the zinc supplement was provided apart from meals, therefore possibly avoiding direct interference between the supplemental zinc and the absorption of dietary copper or endogenous copper secreted in the intestine postprandially. Given that some further interference of the supplement with copper absorption may occur if supplements are consumed with a meal, an uncertainty factor of 1.5 was applied. Based on the average zinc intake from the supplement of 8.2 mg/day, and the mean body weight between baseline and the end of the study of 7.9 kg, the zinc intake was equivalent to 1.0 mg/kg body weight/day, or 0.7 mg/kg/d when considering the uncertainty factor of 1.5. This figure is then applied to the reference body weights for children to derive the NOAEL (table 1.11).

Reporting of the NOAELs for children does not preclude studies of possible adverse effects of higher intakes of zinc, with the caveat that appropriate monitoring is included. Indeed, further prospective studies of the possible adverse effects of varying levels of supplemental zinc are urgently required to improve the derivation of upper limits for total zinc intakes, particularly among children. Several issues, however, must be considered and controlled for in the design of such studies to facilitate the interpretation of results, including: (1) the proportion of zinc acquired from the diet versus zinc derived from pharmacologic supplements; (2) the comparative effects of zinc on copper status when supplemental zinc is taken with copper-containing meals, or between meals; (3) if supplemental zinc is taken with meals, the estimated bioavailability of zinc based on the phytate:zinc molar ratio of the usual diet of the subjects involved; and (4) the baseline copper status and copper intake of individuals and the possible influence of infections on biochemical indicators of copper status.

1.8 Causes of zinc deficiency and groups at high risk

Development of zinc deficiency can be attributed to at least five general causes occurring either in isolation or in combination. These include inadequate intake, increased requirements, malabsorption, increased losses, and impaired utilization [153]. The conditions or circumstances underlying these mechanisms are described below.

Inadequate dietary intake of absorbable zinc is likely to be the primary cause of zinc deficiency in most situations. This may result from a combination of low total dietary intake, heavy reliance on foods with a low zinc content and/or with zinc that is poorly absorbable. Several estimates of dietary zinc intakes indicate that inadequacy of intakes is widespread, occurring across a wide variety of geographical areas and dietary patterns [154, 155]. Low intakes of absorbable zinc are further exacerbated by physiologic or pathological conditions that lead to greater requirements for zinc (per kg body weight). The physiologic and pathologic conditions associated with elevated zinc requirements place individuals in these subgroups at an increased risk of zinc deficiency; these subgroups are described in further detail below.

Malabsorption of zinc may result from a number of different conditions. For example, acrodermatitis enteropathica is a rare genetic defect that specifically affects zinc absorption (section 1.4). Certain disease states, such as malabsorption syndromes and inflammatory diseases of the bowel, may result in poor absorption and/or losses of zinc from the body. Hence, these conditions may precipitate secondary zinc deficiency states, particularly in the presence of marginal dietary zinc intakes [156]. Certain drugs, such as phenytoin and tetracycline, are also noted to reduce the absorption of zinc [157]. Several studies suggest that zinc absorption is antagonized by pharmacologic doses of iron which would result from competitive interaction between these elements [12, 158–160].

Impaired utilization of zinc may occur as a result of administration of certain drugs (e.g., ethambutol, halogenated 8-hydroxyquinolines, penacillamine) that chelate zinc systemically and make it less available for use by tissues [156]. Presence of infection in general results in sequestration of zinc in the liver [161], and decreased circulating levels of zinc, which will reduce the availability of zinc to other tissues. In response to infection-induced secretion of cytokines, such as interleukin-1 and tumor necrosis factor–alpha,

by monocytes and activated macrophages, there is increased hepatic synthesis of metallothionein (MT), an intracellular metal-binding protein [162], and subsequently increased hepatic uptake of zinc and reduction in serum zinc concentration. It is not known whether these alterations in zinc metabolism may benefit the host by making more zinc available for particular processes in the liver or by reducing zinc availability in the peripheral blood.

Certain disease states or conditions that result in increased losses of endogenous zinc from the body include chronic renal disease, trauma, prolonged bed rest, and other conditions associated with bone or muscle atrophy. As the secretion and re-absorption of endogenous zinc in the intestine are key mechanisms in maintaining zinc homeostasis, conditions that perturb intestinal function or the integrity of the intestinal mucosa may have profound effects on the body's ability to maintain zinc status. For example, endogenous zinc losses are increased in infants with cystic fibrosis [163], and fecal zinc excretion is elevated during acute diarrhea [164]. However, it is unclear to what extent the increased fecal zinc represents unabsorbed dietary zinc or zinc of endogenous origin. Because diarrheal disease is a common infection in many lower-income countries, the possible effects of diarrhea on endogenous zinc depletion merit further study and quantification. Not only does zinc deficiency appear to augment the susceptibility to, and severity of, childhood diarrhea, but increased losses of endogenous zinc that occur during diarrhea may further deplete body zinc and propagate a cycle of diarrhea and further zinc depletion. Two studies of zinc homeostasis in Malawian children have demonstrated unusually high intestinal losses of endogenous zinc, even among apparently healthy children [165, 166]. While the causes of these increased losses were not identified, it was speculated that this may be attributed in part to poor intestinal health and may parallel the occurrence of poor intestinal permeability, which is consistently observed in lower-income populations [167], including infants [168]. Further measures of intestinal losses of endogenous zinc among populations living in areas with high exposure to environmental pathogens are needed to determine whether this phenomenon is widespread and to what extent it may contribute to zinc deficiency.

Population subgroups at increased risk of zinc deficiency

Population subgroups with particularly high risks of zinc deficiency can be identified on the basis of their age and physiologic status or the presence of particular pathologic conditions, as described in the following sections.

Infants and young children

Theoretical estimates of zinc requirements suggest that exclusively breastfed infants of mothers with adequate zinc nutriture can satisfy their zinc requirements for the first 5–6 months of life [120]. This is well supported by experimental evidence [169–172]. However, after approximately six months of age, it is unlikely that breastmilk alone can supply sufficient zinc to meet infants' needs [120, 173]. Therefore, if the introduction of complementary foods to breastfed infants is delayed until after six months of age, or if the complementary foods introduced contain inadequate amounts of absorbable zinc, infants will be at increased risk of zinc deficiency. In many lower-income countries, cereals or starchy roots or tubers are used as the basis for complementary foods and these foods often have a low content of total or absorbable zinc. Thus, the complementary diet fails to meet the estimated needs for zinc [174].

Conversely, the premature introduction of other food sources will reduce net zinc absorption if these foods displace breastmilk, have a lower concentration of absorbable zinc than breastmilk, and/or contain substances like phytate, which may interfere with absorption of zinc from breastmilk [175]. Notably, one zinc supplementation trial of non-exclusively breastfed infants of African immigrants to France found that those infants who received supplemental zinc for 3 months beginning at 4–9 months of age had increased weight gain and linear growth, possibly because the foods that had been introduced in addition to breastmilk had an adverse effect on total zinc intake and/or absorption [119]. This combined set of results suggests that premature introduction of complementary foods may impose an increased risk of poor zinc status in early infancy.

Adolescents

Physiologic requirements for zinc peak during adolescence at the time of the pubertal growth spurt, which generally occurs in girls between 10–15 years, and in boys between 12–15 years. Even when the growth spurt has ceased, adolescents may require additional zinc to replete tissue zinc pools depleted during puberty [176].

Pregnant and lactating women

Increased nutritional demands during pregnancy and lactation predispose women to developing zinc deficiency. These demands are greater for lactation than for pregnancy, although physiologic adjustments in zinc absorption help to meet the needs for lactation [72]. Smoking and alcohol abuse during pregnancy may also reduce the amount of zinc available for fetal development by compromising blood flow, and therefore transfer of zinc, to the placenta. As noted above, several studies have indicated that

iron supplements reduce the absorption of zinc [12, 158–160]. All of these studies were conducted in pregnant and/or lactating women and included either prophylactic or therapeutic doses of iron ranging from 60 to > 200 mg/day. Where dietary intakes of zinc are low, supplemental iron, in dosages as low as 60 mg/day, may prevent women from meeting their increased needs for zinc during pregnancy and lactation [160].

Elderly

Dietary surveys indicate that zinc intakes in the elderly, even in higher-income countries, are often inadequate [177]. Zinc deficiency among the elderly has been reported in various countries, and senior citizens living in nursing homes appear to be at increased risk [178]. A number of factors may contribute to the risk of poor zinc nutrition among the elderly, including reductions in total food intake due to reduced mobility, decreased energy needs, and possibly depression, and low intakes of zinc-rich foods, such as meat, poultry, or fish due to poverty or physical disabilities (e.g., swallowing and dental problems). Low zinc intakes may be compounded if efficiency of zinc absorption decreases with age, as has been suggested by some [179– 181], but not all [182], investigators.

Low-birthweight infants

Low-birthweight infants have a reduced size at birth, and hence a smaller content of hepatic zinc metallothionein, which reportedly acts as a zinc reserve in young infants [117]. For low-birthweight infants born prematurely, their body zinc content at birth will be further compromised because more than two-thirds of the zinc is transferred during the last trimester of pregnancy [183]. Moreover, preterm infants may have reduced absorption because of their immature gastrointestinal tract. These impairments result in elevated zinc requirements during the neonatal period, although specific requirements for these infants have not been established.

Malnourished infants and children

The dietary requirements for zinc in malnourished children are estimated to be between 2 and 4 mg/kg/day, depending on the volume of food intake and rate of growth [184]. These zinc requirements are markedly higher than those estimated for healthy children (e.g., 0.17 mg zinc/kg/day for children 1–3 years of age; table 1.9), presumably due to prior zinc depletion, the need for zinc for tissue synthesis, problems of malabsorption due to changes in the intestinal tract, and possibly increased losses due to diarrhea.

1.9 Summary

Due to the multiple biologic functions of zinc and its ubiquitous distribution in human tissues, there is a broad range of physiologic signs of zinc deficiency, which may vary depending on the affected individuals' sex and stage of the lifecycle. The functional effects of zinc deficiency have been determined primarily through community-based zinc supplementation trials and clinical studies of individuals with acrodermatitis enteropathica, children with severe malnutrition, and the elderly. The adverse consequences of zinc deficiency include the following: (1) impaired immunocompetence and increased prevalence and incidence of childhood infections, such as diarrhea and pneumonia, which may result in increased rates of mortality; (2) impaired growth and development of infants, children and adolescents; and (3) impaired maternal health and pregnancy outcomes. These complications of zinc deficiency may be better defined when the specific biochemical mechanisms that link zinc status to these outcomes are elucidated.

Although other factors may contribute to the development of zinc deficiency, inadequate dietary intake of absorbable zinc is likely to be the most common cause. The adequacy of zinc intake is affected by the presence of dietary factors that inhibit zinc absorption, primarily the phytate:zinc molar ratio. Diets based largely on unprocessed cereals or tubers and negligible amounts of animal source foods increase the dietary requirements for zinc, and therefore heighten the challenge of acquiring an adequate amount of zinc from the diet. It is also recognized that zinc deficiency in many populations may be attributable to underlying social and economic problems, such as poverty, poor quality food supply, lack of nutrition education, and elevated exposure to pathogens because of poor environmental sanitation and/or personal hygiene. The identification of high-risk groups within populations on the basis of socio-demographic variables is covered in section 2.5.

Groups at increased risk of zinc deficiency include those with high requirements for zinc and those for whom other factors make it difficult to acquire diets with adequate zinc content. These high-risk groups include pre-term infants, small-for-gestational-age term infants, young children after the period of exclusive breastfeeding, children presenting with and recovering from malnutrition, adolescents, pregnant and lactating women, and the elderly. Based on the large body of evidence for positive effects of supplemental zinc on multiple outcomes of concern to public health, it is evident that similar benefits would be realized in programs designed to improve zinc intakes among those at high risk for zinc deficiency. Identification of nutritional zinc deficiency and its specific causes has therefore become a growing concern for public health planners. Methods to estimate the risk of zinc deficiency in populations and strategies for improving zinc status are considered in the following chapters.

There are still a number of zinc-related issues for which additional research is needed: (1) the full range of functional consequences of zinc deficiency, (2) zinc requirements and safe upper limits of zinc intake, and (3) zinc absorption from mixed diets. New information on the functional consequences of zinc deficiency would be useful to motivate greater interest in the likely benefits of zinc intervention programs and to define the full range of conditions for which such interventions might be helpful. Additional research on zinc requirements is needed to provide relevant information for different population subgroups, as defined by age, sex, and physiologic status. For example, more information is needed on the quantitative losses of endogenous zinc from different sites to define physiologic requirements more precisely for these subgroups. In addition, research is needed on exogenous factors, such as infections or pre-existing malnutrition, that might modify these estimates of physiologic requirements. Studies are also needed to define more precisely excessive levels of zinc intake. Finally, additional studies are needed on zinc absorption from a broad range of mixed diets with varying levels of factors that are known or believed to modify zinc absorption (e.g., levels of zinc, phytate, protein from different sources, and calcium and other minerals), and on the effects of commonly occurring diseases, such as tropical enteropathy, acute and persistent diarrhea, and intestinal helminthic infections, on zinc absorption. A more detailed discussion of these research priorities is presented in chapter 4.

References

1. Vallee BL, Falchuk KH. The biochemical basis of zinc physiology. Physiol Rev 1993;73:79–118.
2. Cousins RJ. Zinc. In: Ziegler EE, Filer LJ Jr, eds. Present Knowledge in Nutrition, 7th ed. Washington, DC: ILSI Press, 1996.
3. Iyengar GV. Reevaluation of the trace element content in reference man. Radiat Phys Chem 1998;51:545–60.
4. Jackson MJ, Jones DA, Edwards RH. Tissue zinc levels as an index of body zinc status. Clin Physiol 1982;2:333–43.
5. King JC, Shames DM, Woodhouse LR. Zinc homeostasis in humans. J Nutr 2000;130:1360S–6S.
6. Zhou JR, Canar MM, Erdman JW Jr. Bone zinc is poorly released in young, growing rats fed marginally zinc-restricted diet. J Nutr 1993;123:1383–8.
7. Giugliano R, Millward DJ. Growth and zinc homeostasis in the severely Zn-deficient rat. Br J Nutr 1984;52:545–60.
8. Emmert JL, Baker DH. Zinc stores in chickens delay the onset of zinc deficiency symptoms. Poult Sci 1995;74: 1011–21.
9. McMahon RJ, Cousins RJ. Mammalian zinc transporters. J Nutr 1998;128:667–70.
10. King JC, Keen CL. Zinc. In: Shils ME, Olson JA, Shike M, Ross AC, eds. Modern Nutrition in Health and Disease, 9th ed. Philadelphia: Lea & Febiger, 1999.
11. Food and Nutrition Board, Institute of Medicine. Dietary reference intakes of vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. Washington, DC: National Academy Press, 2002.
12. Fung EB, Ritchie LD, Woodhouse LR, Roehl R, King JC. Zinc absorption in women during pregnancy and lactation: a longitudinal study. Am J Clin Nutr 1997;66: 80–8.
13. Lee DY, Prasad AS, Hydrick-Adair C, Brewer G, Johnson PE. Homeostasis of zinc in marginal human zinc deficiency: role of absorption and endogenous excretion of zinc. J Lab Clin Med 1993;122:549–56.
14. Sian L, Mingyan X, Miller LV, Tong L, Krebs NF, Hambidge KM. Zinc absorption and intestinal losses of endogenous zinc in young Chinese women with marginal zinc intakes. Am J Clin Nutr 1996;63:348–53.
15. McKusick VA. The Mendelian inheritance in man. Baltimore, MD: The John Hopkins University Press, 1985.
16. Van Wouwe JP. Clinical and laboratory diagnosis of acrodermatitis enteropathica. Eur J Pediatr 1989;149:2–8.
17. Kay RG, Tasman-Jones C. Letter: Zinc deficiency and intravenous feeding. Lancet 1975;2:605–6.
18. Smolarek C, Stremmel W. Therapy of Wilson disease. Z Gastroenterol 1999;37:293–300.
19. Klingberg WG, Prasad AS, Oberleas D. Trace elements in human health and disease. New York: Academic Press, 1976.
20. Hambidge KM. Zinc deficiency in the weanling—how important? Acta Paediatr Scand Suppl 1986;323:52–8.
21. Penland JG. Behavioral data and methodology issues in studies of zinc nutrition in humans. J Nutr 2000;130: 361S–4S.
22. Kohn S, Kohn D, Schiller D. Effect of zinc supplementation on epidermal Langerhans' cells of elderly patients with decubital ulcers. J Dermatol 2000;27:258–63.
23. Gibson RS, Hotz C. Nutritional causes of linear growth faltering in infants during the complementary feeding period. In: Martorell R, Haschke F, eds. Nutrition and growth. 47th Nestle Nutrition Workshop Series, Pediatric Program, Santiago de Chile, April 2–6, 2000. Philadelphia: Lippincott, Williams & Wilkins, 2001; 159–92.
24. Chandra S, Chandra RK. Nutrition, immune response, and outcome. Prog Food Nutr Sci 1986;10:1–65.
25. Grantham-McGregor SM, Fernald LC, Sethuraman K. Effects of health and nutrition on cognitive and behavioural development in children in the first three years of life. Part 2: Infections and micronutrient deficiencies: iodine, iron, and zinc. Food Nutr Bull 1999;20:76–99.
26. Ronaghy HA, Reinhold JG, Mahloudji M, Ghavami P, Fox MR, Halsted JA. Zinc supplementation of malnourished schoolboys in Iran: increased growth and other effects. Am J Clin Nutr 1974;27:112–21.
27. Sandstead HH, Penland JG, Alcock NW, Dayal HH, Chen XC, Li JS, Zhao F, Yang JJ. Effects of repletion with zinc and other micronutrients on neuropsychologic performance and growth of Chinese children. Am J Clin Nutr 1998;68:470S–5S.

28. Shankar AH, Prasad AS. Zinc and immune function: the biological basis of altered resistance to infection. Am J Clin Nutr 1998;68:447S–63S.
29. Fraker PJ, King LE, Laakko T, Vollmer TL. The dynamic link between the integrity of the immune system and zinc status. J Nutr 2000;130:1399S–406S.
30. Beck FW, Prasad AS, Kaplan J, Fitzgerald JT, Brewer GJ. Changes in cytokine production and T cell subpopulations in experimentally induced zinc-deficient humans. Am J Physiol 1997;272:E1002–7.
31. Bogden JD, Oleske JM, Munves EM, Lavenhar MA, Bruening KS, Kemp FW, Holding KJ, Denny TN, Louria DB. Zinc and immunocompetence in the elderly: baseline data on zinc nutriture and immunity in unsupplemented subjects. Am J Clin Nutr 1987;46: 101–9.
32. Bogden JD, Oleske JM, Lavenhar MA, Munves EM, Kemp FW, Bruening KS, Holding KJ, Denny TN, Guarino MA, Krieger LM, Holland BK. Zinc and immunocompetence in elderly people: effects of zinc supplementation for 3 months. Am J Clin Nutr 1988;48:655–63.
33. Bogden JD, Oleske JM, Lavenhar MA, Munves EM, Kemp FW, Bruening KS, Holding KJ, Denny TN, Guarino MA, Holland BK. Effects of one year of supplementation with zinc and other micronutrients on cellular immunity in the elderly. J Am Coll Nutr 1990;9:214–25.
34. Bogden J, Bendich A, Kemp F, Bruening K, Shurnick J, Denny T, Baker H, Louria D. Daily micronutrient supplements enhance delayed-hypersensitivity skin test responses in older people. Am J Clin Nutr 1994;60: 437–47.
35. Chandra R. Nutrition and immunity in the elderly: clinical significance. Nutr Rev 1995;53:S80–5.
36. Duchateau J, Delepesse G, Vrijens R, Collet H. Beneficial effects of oral zinc supplementation on the immune response of old people. Am J Med 1981;70:1001–4.
37. Prasad AS, Meftah S, Abdallah J, Kaplan J, Brewer GJ, Bach JF, Dardenne M. Serum thymulin in human zinc deficiency. J Clin Invest 1988;82:1202–10.
38. Ruel MT, Rivera JA, Santizo MC, Lonnerdal B, Brown KH. Impact of zinc supplementation on morbidity from diarrhea and respiratory infections among rural Guatemalan children. Pediatrics 1997;99:808–13.
39. Sazawal S, Black R, Bhan M, Bhandari N, Sinha A, Jalla S. Zinc supplementation in young children with acute diarrhea in India. N Engl J Med 1995;333:839–44.
40. Sazawal S, Black RE, Bhan MK, Jalla S, Bhandari N, Sinha A, Majumdar S. Zinc supplementation reduces the incidence of persistent diarrhea and dysentery among low socioeconomic children in India. J Nutr 1996;126: 443–50.
41. Sazawal S, Black RE, Bhan MK, Jalla S, Sinha A, Bhandari N. Efficacy of zinc supplementation in reducing the incidence and prevalence of acute diarrhea—a community-based, double-blind, controlled trial. Am J Clin Nutr 1997;66:413–418.
42. Behrens RH, Tomkins AM, Roy SK. Zinc supplementation during diarrhoea, a fortification against malnutrition? Lancet 1990;336:442–3.
43. Lira PI, Ashworth A, Morris SS. Effect of zinc supplementation on the morbidity, immune function, and growth of low-birth-weight, full-term infants in northeast Brazil. Am J Clin Nutr 1998;68:418S–24S.
44. Bhutta ZA, Black RE, Brown KH, Gardner JM, Gore S, Hidayat A, Khatun F, Martorell R, Ninh NX, Penny ME, Rosado JL, Roy SK, Ruel M, Sazawal S, Shankar A (Zinc Investigators' Collaborative Group). Prevention of diarrhea and pneumonia by zinc supplementation in children in developing countries: pooled analysis of randomized controlled trials. J Pediatr 1999;135: 689–97.
45. Müller O, Becher H, van Zweeden AB, Ye Y, Diallo DA, Konate AT, Gbangou A, Kouyate B, Garenne M. Effect of zinc supplementation on malaria and other causes of morbidity in west African children: randomised double blind placebo controlled trial. BMJ 2001;322:1567.
46. Umeta M, West CE, Haidar J, Deurenberg P, Hautvast JGAJ. Zinc supplementation and stunted infants in Ethiopia: a randomised controlled trial. Lancet 2000;355: 2021–6.
47. Esrey SA, Feachem RG, Hughes JM. Interventions for the control of diarrhoeal diseases among young children: improving water supplies and excreta disposal facilities. Bull World Health Organ 1985;63:757–72.
48. Ninh NX, Thissen JP, Collette L, Gerard G, Khoi HH, Ketelslegers JM. Zinc supplementation increases growth and circulating insulin-like growth factor I (IGF-I) in growth-retarded Vietnamese children. Am J Clin Nutr 1996;63:514–9.
49. Sazawal S, Black RE, Jalla S, Mazumdar S, Sinha A, Bhan MK. Zinc supplementation reduces the incidence of acute lower respiratory infections in infants and preschool children: a double-blind, controlled trial. Pediatrics 1998;102:1–5.
50. Bates CJ, Evans PH, Dardenne M, Prentice A, Lunn PG, Northrop-Clewes CA, Hoare S, Cole TJ, Horan SJ, Longman SC, Stirling D, Aggett PJ. A trial of zinc supplementation in young rural Gambian children. Br J Nutr 1993;69:243–55.
51. Shankar AH, Genton B, Baisor M, Paino J, Tamja S, Adiguma T, Wu L, Rare L, Bannon D, Tielsch JM, West KP Jr, Alpers MP. The influence of zinc supplementation on morbidity due to Plasmodium falciparum: a randomized trial in preschool children in Papua New Guinea. Am J Trop Med Hyg 2000;62:663–9.
52. Sazawal S, Black RE, Menon VP, Dinghra P, Caulfield LE, Dhingra U, Bagati A. Zinc supplementation in infants born small for gestational age reduces mortality: a prospective, randomized, controlled trial. Pediatrics 2001;108:1280–6.
53. Castillo-Duran C, Rodriguez A, Venegas G, Alvarez P, Icaza G. Zinc supplementation and growth of infants born small for gestational age. J Pediatr 1995;127: 206–11.
54. Friel JK, Andrews WL, Matthew JD, Long DR, Cornel AM, Cox M, McKim E, Zerbe GO. Zinc supplementation in very-low-birth-weight infants. J Pediatr Gastroenterol Nutr 1993;17:97–104.
55. Golden BE, Golden MHN. Plasma zinc, rate of weight gain, and the energy cost of tissue deposition in children recovering from severe malnutrition on a cow's milk or soya protein based diet. Am J Clin Nutr 1981;34:892–9.
56. Golden MHN, Golden BE. Effect of zinc supplementation on the dietary intake, rate of weight gain, and energy cost of tissue deposition in children recovering from severe malnutrition. Am J Clin Nutr 1981;34:900–8.

57. Khanum S, Alam AN, Anwar I, Akbar Ali M, Mujibur Rahaman M. Effect of zinc supplementation on the dietary intake and weight gain of Bangladeshi children recovering from protein-energy malnutrition. Eur J Clin Nutr 1988;42:709–14.
58. Simmer K, Khanum S, Carlsson L, Thompson RP. Nutritional rehabilitation in Bangladesh—the importance of zinc. Am J Clin Nutr 1988;47:1036–40.
59. Doherty CP, Sarkar MA, Shakur MS, Ling SC, Elton RA, Cutting WA. Zinc and rehabilitation from severe protein-energy malnutrition: higher-dose regimens are associated with increased mortality. Am J Clin Nutr 1998;68:742–8.
60. Waterlow JC. Relationship of gain in height to gain in weight. Eur J Clin Nutr 1994;48(Suppl)1:S72–3; discussion S73–4.
61. Brown KH, Peerson JM, Rivera J, Allen LH. Effect of supplemental zinc on the growth and serum zinc concentrations of prepubertal children: a meta-analysis of randomized controlled trials. Am J Clin Nutr 2002;75: 1062–71.
62. Hambidge KM, Hambidge C, Jacobs M, Baum JD. Low levels of zinc in hair, anorexia, poor growth, and hypogeusia in children. Pediatr Res 1972;6:868–74.
63. Hambidge KM, Krebs NF, Walravens PA. Growth velocity of young children receiving a dietary zinc supplement. Nutr Res 1985;1S:306–16.
64. Ruz M, Castillo-Duran C, Lara X, Codoceo J, Rebolledo A, Atalah E. A 14-mo zinc-supplementation trial in apparently healthy Chilean preschool children. Am J Clin Nutr 1997;66:1406–13.
65. Prasad AS, Miale A Jr, Farid Z, Sandstead HH, Schulert AR. Zinc metabolism in patients with the syndrome of iron-deficiency anemia, hepatosplenomegaly, dwarfism, and hypogonadism. 1963. J Lab Clin Med 1990;116: 737–49.
66. Halsted JA, Ronaghy HA, Abadi P, Haghshenass M, Amirhakemi GH, Barakat RM, Reinhold JG. Zinc deficiency in man: the Shiraz experiment. Am J Med 1972; 53:277–84.
67. Ronaghy H, Fox MR, Garnsm, Israel H, Harp A, Moe PG, Halsted JA. Controlled zinc supplementation for malnourished school boys: a pilot experiment. Am J Clin Nutr 1969;22:1279–89.
68. Abbasi AA, Prasad AS, Rabbani P, DuMouchelle E. Experimental zinc deficiency in man: effect on testicular function. J Lab Clin Med 1980;96:544–50.
69. Castillo-Duran C, Garcia H, Venegas P, Torrealba I, Panteon E, Concha N, Perez P. Zinc supplementation increases growth velocity of male children and adolescents with short stature. Acta Paediatr 1994;83:833–7.
70. Tamura T, Goldenberg RL. Zinc nutriture and pregnancy outcome. Nutr Res 1996;16:139–81.
71. Caulfield LE, Zavaleta N, Shankar AH, Merialdi M. Potential contribution of maternal zinc supplementation during pregnancy to maternal and child survival. Am J Clin Nutr 1998;68:499S–508S.
72. King JC. Determinants of maternal zinc status during pregnancy. Am J Clin Nutr 2000;71:1334S–43S.
73. Ross SM, Nel E, Naeye RL. Differing effects of low and high bulk maternal dietary supplements during pregnancy. Early Hum Dev 1985;10:295–302.
74. Garg HK, Singhal KC, Arshad Z. A study of the effect of oral zinc supplemetation during pregnancy on pregnancy outcome. Indian J Physiol Pharmacol 1993;37:276–84.
75. Caulfield LE, Zavaleta N, Figueroa A, Leon Z. Maternal zinc supplementation does not affect size at birth or pregnancy duration in Peru. J Nutr 1999;129:1563–8.
76. Osendarp SJ, van Raaij JM, Arifeen SE, Wahed M, Baqui AH, Fuchs GJ. A randomized, placebo-controlled trial of the effect of zinc supplementation during pregnancy on pregnancy outcome in Bangladeshi urban poor. Am J Clin Nutr 2000;71:114–9.
77. Goldenberg RL, Tamura T, Neggers Y, Copper RL, Johnston KE, DuBard MB, Hauth JC. The effect of zinc supplementation on pregnancy outcome. JAMA 1995;274:463–8.
78. Cherry FF, Sandstead HH, Rojas P, Johnson LK, Batson HK, Wang XB. Adolescent pregnancy: associations among body weight, zinc nutriture, and pregnancy outcome. Am J Clin Nutr 1989;50:945–54.
79. Hunt IF, Murphy NJ, Cleaver AE, Faraji B, Swendseid ME, Coulson AH, Clark VA, Browdy BL, Cabalum T, Smith JC Jr. Zinc supplementation during pregnancy: effects on selected blood constituents and on progress and outcome of pregnancy in low-income women of Mexican descent. Am J Clin Nutr 1984;40:508–21.
80. Jameson S. Zinc status in pregnancy: the effect of zinc therapy on perinatal mortality, prematurity, and placental ablation. Ann N Y Acad Sci 1993;678:178–92.
81. Hunt IF, Murphy NJ, Cleaver AE, Faraji B, Swendseid ME, Browdy BL, Coulson AH, Clark VA, Settlage RH, Smith JC Jr. Zinc supplementation during pregnancy in low-income teenagers of Mexican descent: effects on selected blood constituents and on progress and outcome of pregnancy. Am J Clin Nutr 1985;42:815–28.
82. Merialdi M, Caulfield LE, Zavaleta N, Figueroa A, DiPietro JA. Adding zinc to prenatal iron and folate tablets improves fetal neurobehavioral development. Am J Obstet Gynecol 1999;180:483–90.
83. Osendarp SJM, van Raaij JMA, Darmstadt GL, Baqui AH, Hautvast JG, Fuchs GJ. Zinc supplementation during pregnancy and effects on growth and morbidity in low birthweight infants: a randomised placebo controlled trial. Lancet 2001;357:1080–5.
84. Hamadani JD, Fuchs GJ, Osendarp SJ, Huda SN, Grantham-McGregor SM. Zinc supplementation during pregnancy and effects on mental development and behaviour of infants: a follow-up study. Lancet 2002;360:290–4.
85. Chandra RK, Joshi P, Au B, Woodford G, Chandra S. Nutrition and immuno-competence of the elderly: effect of short-term nutritional supplementation on cell-mediated immunity and lymphocyte subsets. Nutr Res 1982;2:223–32.
86. Greger JL, Geissler AH. Effect of zinc supplementation on taste acuity of the aged. Am J Clin Nutr 1978;31: 633–7.
87. Henkin RI, Schecter PJ, Friedewald WT, Demets DL, Raff M. A double blind study of the effects of zinc sulfate on taste and smell dysfunction. Am J Med Sci 1976;272: 285–99.
88. Wright AL, King JC, Baer MT, Citron LJ. Experimental zinc depletion and altered taste perception for NaCl in young adult males. Am J Clin Nutr 1981;34:848–52.
89. Husain SL, Fell GS, Scott R, Trethewie ER. Zinc and

healing. Lancet 1970;2:1361–2.
90. Pories WJ, Henzel JH, Rob CG, Strain WH. Acceleration of healing with zinc sulfate. Ann Surg 1967;165:432–6.
91. Penland JG. Cognitive performance affects low zinc (Zn) intakes in healthy adult men (abstract). FASEB J 1991;5: A938.
92. Sandstead HH. Zinc: essentiality for brain development and function. Nutr Rev 1985;43:129–37.
93. Hendricks DG, Mahoney AW. Glucose tolerance in zinc-deficient rats. J Nutr 1972;102:1079–84.
94. Quarterman J, Mills CF, Humphries WR. The reduced secretion of, and sensitivity to insulin in zinc-deficient rats. Biochem Biophys Res Commun 1966;25:354–8.
95. Bentley ME, Caulfield LE, Ram M, Santizo MC, Hurtado E, Rivera JA, Ruel MT, Brown KH. Zinc supplementation affects the activity patterns of rural Guatemalan infants. J Nutr 1997;127:1333–8.
96. Sazawal S, Bentley M, Black RE, Dhingra P, George S, Bhan MK. Effect of zinc supplementation on observed activity in low socioeconomic Indian preschool children. Pediatrics 1996;98:1132–7.
97. Chesters JK, Quarterman J. Effects of zinc deficiency on food intake and feeding patterns of rats. Br J Nutr 1970;24:1061–9.
98. Krebs NF, Hambidge KM, Walravens PA. Increased food intake of young children receiving a zinc supplement. Am J Dis Child 1984;138:270–3.
99. Shay NF, Mangian HF. Neurobiology of zinc-influenced eating behavior. J Nutr 2000;130:1493S–9S.
100. World Health Organization (WHO), Food and Agriculture Organization (FAO), International Atomic Energy Association (IAEA). Trace elements in human health and nutrition. Geneva: World Health Organization, 1996.
101. World Health Organization (WHO), Food and Agriculture Organization (FAO), International Atomic Energy Association (IAEA). Trace elements in human health and nutrition, 2nd ed. Geneva: World Health Organization, 2002.
102. Milne DB, Canfield WK, Mahalko JR, Sandstead HH. Effect of dietary zinc on whole body surface loss of zinc: impact on estimation of zinc retention by balance method. Am J Clin Nutr 1983;38:181–6.
103. Taylor CM, Bacon JR, Aggett PJ, Bremner I. Homeostatic regulation of zinc absorption and endogenous losses in zinc-deprived men. Am J Clin Nutr 1991;53:755–63. Erratum in: Am J Clin Nutr 1992 Aug;56(2):462.
104. Johnson PE, Hunt CD, Milne DB, Mullen LK. Homeostatic control of zinc metabolism in men: zinc excretion and balance in men fed diets low in zinc. Am J Clin Nutr 1993;57:557–65.
105. Hunt CD, Johnson PE, Herbel J, Mullen LK. Effects of dietary zinc depletion on seminal volume and zinc loss, serum testosterone concentrations, and sperm morphology in young men. Am J Clin Nutr 1992;56: 148–57.
106. Baer MT, King JC. Tissue zinc levels and zinc excretion during experimental zinc depletion in young men. Am J Clin Nutr 1984;39:556–70.
107. Turnlund JR, Durkin N, Costa F, Margen S. Stable isotope studies of zinc absorption and retention in young and elderly men. J Nutr 1986;116:1239–47.
108. Turnlund JR, King JC, Keyes WR, Gong B, Michel MC. A stable isotope study of zinc absorption in young men: effects of phytate and alpha-cellulose. Am J Clin Nutr 1984;40:1071–7.
109. Jackson MJ, Jones DA, Edwards RH, Swainbank IG, Coleman ML. Zinc homeostasis in man: studies using a new stable isotope-dilution technique. Br J Nutr 1984; 51:199–208.
110. Hunt JR, Mullen LK, Lykken GI. Zinc retention from an experimental diet based on the US FDA Total Diet Study. Nutr Res 1992;12:1335–44.
111. Wada L, Turnlund JR, King JC. Zinc utilization in young men fed adequate and low zinc intakes. J Nutr 1985;115: 1345–54.
112. Knudsen E, Sandström B, Solgaard P. Zinc, copper and magnesium absorption from a fibre-rich diet. J Trace Elem Med Biol 1996;10:68–76.
113. Hunt JR, Matthys LA, Johnson LK. Zinc absorption, mineral balance, and blood lipids in women consuming controlled lactoovovegetarian and omnivorous diets for 8 wk. Am J Clin Nutr 1998;67:421–30.
114. Hunt JR, Gallagher SK, Johnson LK, Lykken GI. High-versus low-meat diets: effects on zinc absorption, iron status, and calcium, copper, iron, magnesium, manganese, nitrogen, phosphorus, and zinc balance in postmenopausal women. Am J Clin Nutr 1995;62: 621–32.
115. Lowe NM, Shames DM, Woodhouse LR., Matel JS, Roehl R, Saccomani MP, Toffolo G, Cobelli C, King JC. A compartmental model of zinc metabolism in healthy women using oral and intravenous stable isotope tracers. Am J Clin Nutr 1997;65:1810–9.
116. Hess FM, King JC, Margen S. Zinc excretion in young women on low zinc intakes and oral contraceptive agents. J Nutr 1977;107:1610–20.
117. Zlotkin SH, Cherian MG. Hepatic metallothionein as a source of zinc and cysteine during the first year of life. Pediatr Res 1988;24:326–9.
118. Heinig MJ, Brown KH, Lönnerdal B, Dewey KG. Zinc supplementation does not affect growth, morbidity, or motor development of US breastfed infants at 4–10 mo. FASEB J 1998;12:5617 (A970).
119. Walravens PA, Chakar A, Mokni R, Denise J, Lemonnier D. Zinc supplements in breastfed infants. Lancet 1992;340:683–5.
120. Krebs NF, Hambidge KM. Zinc requirements and zinc intakes of breast-fed infants. Am J Clin Nutr 1998;43: 288–92.
121. Swanson CA, King JC. Zinc and pregnancy outcome. Am J Clin Nutr 1987;46:763–71.
122. Brown KH, Dewey KG, Allen LH. Complementary feeding of young children in developing countries: a review of current scientific knowledge, WHO/NUT/ 98.1. Geneva: World Health Organization, 1998.
123. Lönnerdal B. Dietary factors influencing zinc absorption. J Nutr 2000;130:1378S–83S.
124. Sandström B. Dose dependence of zinc and manganese absorption in man. Proc Nutr Soc 1992;51:211–8.
125. Cook JD, Dassenko SA, Lynch SR. Assessment of the role of nonheme-iron availability in iron balance. Am J Clin Nutr 1991;54:717–22.
126. Tidehag P, Hallmans G, Wing K, Sjöström R, Ågren G, Lundin E, Zhang JX. A comparison of iron absorption from single meals and daily diets using radioFe (^{55}Fe,

^{59}Fe). Br J Nutr 1996;75:281–9.
127. Brown KH, Wuehler SE, Peerson JM. The importance of zinc in human nutrition and estimation of the global prevalence of zinc deficiency. Food Nutr Bull 2001;22: 113–25.
128. Sandström B, Davidsson L, Cederblad A, Lonnerdal B. Oral iron, dietary ligands and zinc absorption. J Nutr 1985;115:411–4.
129. Adams CL, Hambidge M, Raboy V, Dorsch JA, Sian L, Westcott JL, Krebs NF. Zinc absorption from a low-phytic acid maize. Am J Clin Nutr 2002;76:556–9.
130. Pinna K. Effect of a low zinc intake on zinc homeostasis and immune function in adult men. Doctoral dissertation. Berkeley, California: University of California, 1999.
131. Moser-Veillon PB, Patterson KY, Veillon C. Zinc absorption is enhanced during lactation (abstract). FASEB J 1995;9:A729.
132. Woodhouse LR, Vargas Zapata CL, Donangelo CM, King JC. Fractional zinc absorption (FZA) in Brazilian women: a longitudinal study during pregnancy and lactation. In: Elmadfa I, Konig J, eds. 17th International Congress of Nutrition, August 27–31, 2001, Vienna, Austria. Ann Nutr Metab 2001;45(suppl 1):12.
133. Sian L, Krebs NF, Westcott JE, Fengliang L, Tong L, Miller LV, Sonko B, Hambidge M. Zinc homeostasis during lactation in a population with a low zinc intake. Am J Clin Nutr 2002;75:99–103.
134. Abrams SA, Wen J, Stuff JE. Absorption of calcium, zinc, and iron from breast milk by five- to seven-month-old infants. Pediatr Res 1997;41:384–90. Erratum in: Pediatr Res 1997 Jun;41(6):814.
135. Vuori E. Intake of copper, iron, manganese and zinc by healthy, exclusively breast-fed infants during the first 3 months of life. Br J Nutr 1979;42:407–11.
136. Fosmire GJ. Zinc toxicity. Am J Clin Nutr 1990;51: 225–7.
137. Brown MA, Thom JV, Orth GL, Cova P, Juarez J. Food poisoning involving zinc contamination. Arch Environ Health 1964;34:657–60.
138. Samman S, Roberts DCK. The effect of zinc supplements on plasma zinc and copper levels and the reported symptoms in healthy volunteers. Med J Aust 1987;146: 246–9.
139. Prasad AS, Brewer GJ, Schoomaker EB, Rabbani P. Hypocupremia induced by zinc therapy in adults. JAMA 1978;240:2166–8.
140. Chandra RK. Excessive intake of zinc impairs immune responses. JAMA 1984;252:1443–6.
141. Hooper P, Visconti L, Garry P, Johnson G. Zinc lowers high-density lipoprotein-cholesterol levels. JAMA 1980;244:1960–1.
142. Freeland-Graves J, Friedman B, Han W, Shorey R, Young R. Effect of zinc supplementation on plasma high-density lipoprotein cholesterol and zinc. Am J Clin Nutr 1982;35:988–92.
143. Black M, Medeiros D, Brunett E, Welke R. Zinc supplements and serum lipids in young adult white males. Am J Clin Nutr 1988;47:970–5.
144. Yadrick MK, Kenney MA, Winterfeldt EA. Iron, copper, and zinc status: response to supplementation with zinc or zinc and iron in adult females. Am J Clin Nutr 1989;49:145–50.
145. Smith CJJ. Comparison of reference dose with recommended dietary allowances for zinc: methodologies and levels. In: Mertz W, Abernathy CO, Olin SS, eds. Risk assessment of essential elements. Washington, DC: ILSI Press, 1994.
146. Fischer P, Giroux A, L'Abbe M. Effect of zinc supplementation on copper status in adult man. Am J Clin Nutr 1984;40:743–6.
147. Mertz W, Abernathy CO, Olin SS. Risk assessment of essential elements. Washington, DC: ILSI Press, 1994.
148. Botash AS, Nasca J, Dubowy R, Weinberger HL, Oliphant M. Zinc-induced copper deficiency in an infant. Am J Dis Child 1992;146:709–11.
149. Walravens PA, Hambidge KM. Growth of infants fed a zinc supplemented formula. Am J Clin Nutr 1976;29: 1114–21.
150. Briefel RR, Bialostosky K, Kennedy-Stephenson J, McDowell MA, Ervin RB, Wright JD. Zinc intake of the U.S. population: findings from the third National Health and Nutrition Examination Survey, 1988–1994. J Nutr 2000;130:1367S–73S.
151. Bhandari N, Bahl R, Taneja S, Strand T, Molbak K, Ulvik RJ, Sommerfelt H, Bhan MK. Substantial reduction in severe diarrheal morbidity by daily zinc supplementation in young north Indian children. Pediatrics 2002;109: e86.
152. Lind T, Lonnerdal B, Stenlund H, Ismail D, Seswandhana R, Ekstrom EC, Persson LA. A community-based randomized controlled trial of iron and zinc supplementation in Indonesian infants: interactions between iron and zinc. Am J Clin Nutr 2003;77:883-90.
153. Solomons NW, Cousins RJ. Zinc. In: Solomons NW, Rosenberg IH, eds. Absorption and malabsorption of mineral nutrients. New York: Alan R. Liss, 1984.
154. Gibson RS. Zinc nutrition in developing countries. Nutr Res Rev 1994;7:151–73.
155. Shrimpton R. Zinc deficiency: Is it widespread but under-recognized? ACC/SCN News 1993;9:24–7.
156. Aggett PJ, Harries JT. Current status of zinc in health and disease states. Arch Dis Child 1979;54:909–17.
157. Weismann K. Chelating drugs and zinc. Dan Med Bull 1986;33:208–11.
158. Hambidge KM, Krebs NF, Jacobs MA, Favier A, Guyette L, Ikle DN. Zinc nutritional status during pregnancy: a longitudinal study. Am J Clin Nutr 1983;37:429–42.
159. Hambidge KM, Krebs NF, Sibley L, English J. Acute effects of iron therapy on zinc status during pregnancy. Obstet Gynecol 1987;70:593–6.
160. O'Brien KO, Zavaleta N, Caulfield LE, Wen J, Abrams SA. Prenatal iron supplements impair zinc absorption in pregnant Peruvian women. J Nutr 2000;130:2251–5.
161. Cousins RJ, Leinart AS. Tissue-specific regulation of zinc metabolism and metallothionein genes by interleukin 1. FASEB J 1988;2:2884–90.
162. Schroeder JJ, Cousins RJ. Interleukin 6 regulates metallothionein gene experession and zinc metabolism in hepatocyte monolayer cultures. Proc Natl Acad Sci U S A 1990;87:3137–41.
163. Krebs NF, Westcott JE, Arnold TD, Kluger BM, Accurso FJ, Miller LV, Hambidge KM. Abnormalities in zinc homeostasis in young infants with cystic fibrosis. Pediatr Res 2000;48:256–61.
164. Castillo-Duran C, Vial P, Uauy R. Trace mineral balance

during acute diarrhea in infants. J Pediatr 1988;113: 452–7.
165. Manary MJ, Hotz C, Krebs NF, Gibson RS, Westcott JE, Arnold T, Broadhead RL, Hambidge KM. Dietary phytate reduction improves zinc absorption in Malawian children recovering from tuberculosis but not in well children. J Nutr 2000;130:2959–64.
166. Manary MJ, Hotz C, Krebs NF, Gibson RS, Westcott JE, Broadhead RL, Hambidge KM. Zinc homeostasis in Malawian children consuming a high-phytate, maize-based diet. Am J Clin Nutr 2002;75:1057–61.
167. Menzies IS, Zuckerman MJ, Nukajam WS, Somasundaram SG, Murphy B, Jenkins AP, Crane RS, Gregory GG. Geography of intestinal permeability and absorption. Gut 1999;44:483–9.
168. Goto K, Chew F, Torun B, Peerson JM, Brown KH. Epidemiology of altered intestinal permeability to lactulose and mannitol in Guatemalan infants. J Pediatr Gastroenterol Nutr 1999;28:282–90.
169. Cohen RJ, Brown KH, Canahuati J, Rivera LL, Dewey KG. Effects of age of introduction of complementary foods on infant breastmilk intake, total energy intake, and growth: a randomised intervention study in Honduras. Lancet 1994;344:288–93.
170. Dewey KG, Heinig MJ, Nommsen LA, Lonnerdal B. Zinc status in breastfed infants. Lancet 1992;340:1417.
171. Krebs NF, Reidinger CJ, Robertson AD, Hambidge KM. Growth and intakes of energy and zinc in infants fed human milk. J Pediatr 1994;124:32–9.
172. Michaelsen KF, Samuelson G, Graham TW, Lonnerdal B. Zinc intake, zinc status and growth in a longitudinal study of healthy Danish infants. Acta Paediatr 1994;83: 1115–21.
173. World Health Organization. Complementary feeding of young children in developing countries: a review of current scientific knowledge. Geneva: World Health Organization, 1998.
174. Gibson RS, Ferguson EL. Assessment of dietary zinc in a population. Am J Clin Nutr 1998;68:430S–4S.
175. Bell JG, Keen CL, Lonnerdal B. Effect of infant cereals on zinc and copper absorption during weaning. Am J Dis Child 1987;141:1128–32.
176. King JC. Does poor zinc nutriture retard skeletal growth and mineralization in adolescents? Am J Clin Nutr 1996;64:375–6.
177. Mares-Perlman JA, Subar AF, Block G, Greger JL, Luby MH. Zinc intake and sources in the US adult population: 1976–1980. J Am Coll Nutr 1995;14:349–57.
178. Worwag M, Classen HG, Schumacher E. Prevalence of magnesium and zinc deficiencies in nursing home residents in Germany. Magnes Res 1999;12:181–9.
179. August D, Janghorbani M, Young VR. Determination of zinc and copper absorption at three dietary Zn-Cu ratios by using stable isotope methods in young adult and elderly subjects. Am J Clin Nutr 1989;50:1457–63.
180. Bales CW, DiSilvestro RA, Currie KL, Plaisted CS, Joung H, Galanos AN, Lin PH. Marginal zinc deficiency in older adults: responsiveness of zinc status indicators. J Am Coll Nutr 1994;13:455–62.
181. Turnlund JR, Michel MC, Keyes WR, King JC, Margen S. Use of enriched stable isotopes to determine zinc and iron absorption in elderly men. Am J Clin Nutr 1982;35: 1033–40.
182. Couzy F, Kastenmayer P, Mansourian R, Guinchard S, Munoz-Box R, Dirren H. Zinc absorption in healthy elderly humans and the effect of diet. Am J Clin Nutr 1993;58:690–4.
183. Shaw JCL. Trace elements in the fetus and young infant. I. Zinc. Am J Dis Child 1979;133:1260–8.
184. Briend A, Golden MH. Treatment of severe child malnutrition in refugee camps. Eur J Clin Nutr 1993;47: 750–4.