

Zinc deficiency, immune function, and morbidity and mortality from infectious disease among children in developing countries

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Abstract

Zinc deficiency is prevalent in developing countries and has adverse effects on child health. Decreased or abnormal immune function in children can occur as a consequence of zinc deficiency, either during gestation or after delivery, and may impair host defenses against infectious diseases. Controlled trials of therapy of acute and persistent diarrhea have consistently demonstrated that zinc-supplemented children have diarrheal episodes of shorter duration and reduced severity. Controlled trials of zinc supplementation in the prevention of infectious diseases have demonstrated reductions in the incidences of diarrhea, pneumonia, and malaria, the most common causes of death in children in developing countries. Preliminary evidence from one controlled trial in full-term small-for-gestational-age infants in India found a two-thirds reduction in mortality with zinc supplementation. In conclusion, zinc deficiency reduces immune function and increases the risk of morbidity and mortality from infectious disease in children in developing countries.

Introduction

Deficiency of the trace element zinc may be more widespread and have greater effects on the health of infants and children than previously realized. It has been well recognized that severe deficiency, as in acrodermatitis enteropathica, has many manifestations, including failure to thrive, thymic atrophy, severe depression of immunity, and diarrhea [1]. It has only been recognized more recently that mild to moderate zinc deficiency may present clinically as impaired growth, which may have previously been attributed to other factors [2]. Effects of mild to moderate zinc deficiency on immune function and the risk of infectious diseases in children in developing countries have

also been demonstrated [3]. These relationships are the subject of this review.

Gestational zinc deficiency and immune function after birth

Gestational zinc deficiency has been studied in a number of animal models, including mice and nonhuman primates. Even with mild levels of zinc deficiency during gestation, there is a reduction in lymphoid tissue, especially manifest as atrophy of the thymus, and a reduction in immunoglobulin concentrations in the blood of the newborn [4–7]. Additionally, there appears to be more specific suppression of particular immunoglobulins, such as IgM, IgA, and some subtypes of IgG, which might affect the immunologic protection in early infancy from systemic bacterial infections [8, 9]. There may also be alterations in the function of the cellular elements, such as lymphocytes and neutrophils [7]. Evidence from animal studies suggests that these abnormalities present at birth can be long-lasting or even permanent. In fact, some evidence suggests that this immunocompromise can persist even to second- or third-generation offspring of animals deprived of adequate zinc during gestation [10].

Studies in humans are limited but suggest similar immunologic consequences of zinc deficiency during gestation. In trials of zinc supplementation during pregnancy, children of zinc-supplemented mothers have greater immunoglobulin concentrations and reduced rates of infectious disease during infancy [3]. Additional studies are under way to determine whether zinc supplementation in pregnancy in a presumably zinc-deficient population enhances the immune response to vaccines in infancy.

Zinc deficiency and immune function

The defenses against infection are particularly sensitive to disturbances in zinc status. The barrier functions

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of the skin [1], as well as those of the pulmonary and gastrointestinal tracts [11], are damaged, and the development, function, or both of most immunologic cells suffer deleterious effects [3].

The human immune system is quite sensitive to zinc deficiency. Among the cellular elements that are affected, natural killer cells have reduced activity [12], polymorphonuclear leukocytes have impaired chemotactic responses and microbicidal function [13, 14], and macrophages or monocytes have impaired chemotactic responses, reduced phagocytosis, and reduced intracellular killing of microorganisms [15, 16].

The development of T lymphocytes is profoundly altered by zinc deficiency, with a reduction in the size and cellularity of the thymus and depletion of lymphocytes from the spleen, lymph nodes, and peripheral blood in animal models [12, 17]. Zinc-deficient humans have reduced total numbers of lymphocytes, especially T lymphocytes, in the blood and peripheral lymphoid tissue [18–20]. Decreased CD4+/CD8+ cell ratios are also seen [21]. These abnormalities in humans can be reversed by administration of supplemental zinc. T-lymphocyte function is also altered in zinc deficiency, with a reduction in delayed hypersensitivity responses and cytotoxic activity, which are reversed by zinc supplementation in humans [12, 18–22]. Other studies have shown that zinc deficiency alters T-lymphocyte activation and proliferation and may reduce the response to antigens (and potentially even to vaccines, especially those requiring T-cell-dependent antibody production) [23, 24].

The development of B lymphocytes in the bone marrow is also adversely affected by zinc deficiency, with a reduction in both number and function [25, 26]. Antibody responses are inhibited by zinc deficiency, with abnormalities in mitogenic and cytokine responses; however, T-cell-dependent antibody responses seem to be more affected than T-cell-independent responses [27, 28]. Little is known about the effects of zinc deficiency on secretory immunity or other local mucosal host defenses.

In animal models, there is a reduction in the activity of thymulin and altered release of cytokines, and an imbalance of cell-mediated and antibody-mediated immunity may occur, even with mild levels of zinc deficiency [29–31].

The applicability of all of the studies of zinc deficiency in animal models to human health is unclear, but immunocompromise in humans with mild zinc deficiency has been demonstrated, with the abnormalities reversing with small quantities of additional zinc [17, 32]. The rapidity of response in these functions to repletion of zinc varies, but some functions, such as intracellular killing by macrophages, are restored very rapidly with supplementation [15]. Additional studies are needed on the level of functional abnormalities with differing degrees of zinc deficiency and the cor-

relation of these abnormalities with susceptibility to specific infectious diseases.

Effects of zinc supplementation on therapy of diarrhea

The therapeutic effects of zinc supplementation during diarrhea have been investigated in five trials of acute diarrhea and five trials of persistent diarrhea [33–38]. These studies show consistent benefits of zinc supplementation. In the studies of acute diarrhea (duration on enrollment in these studies ranged from less than three days to more than seven days) (table 1), zinc supplementation generally resulted in shorter diarrheal episodes and a reduced likelihood that the episode would continue for more than seven days after enrollment in the study. With the small sample size in some of these studies, not all of the apparent effects were statistically significant. A pooled analysis of zinc supplementation in acute diarrhea trials for which original data could be obtained (three trials) revealed an overall 15% (95% confidence interval, 5% to 24%) reduced probability of continuing diarrhea in the supplemented group [33]. A meta-analysis of all five trials found a statistically significant effect size of 0.162 (table 2) [33]. Several of the studies also reported reductions in episode severity, as measured by frequency of watery stools or measured stool output (table 1).

The trials of treatment of persistent diarrhea (episodes lasting for more than 14 days) likewise showed benefits with zinc supplementation (table 3) [33, 39–43]. Generally, the studies found that supplementation was associated with a shorter episode duration. The pooled analysis involving four of these trials indicated that children had a 24% (95% confidence interval, 9% to 37%) reduced probability of continuing diarrhea if they received the zinc supplement [33]. A meta-analysis of all five trials found a statistically significant summary effect size of 0.293 (table 4) [33]. There was also a suggestion of reduced episode severity in one trial, but not in a second trial (table 3). Importantly, though, most trials reported large reductions in the rate of treatment failure or death (table 3). Overall, in the pooled analysis, there was a 42% (95% confidence interval, 10% to 63%) reduced rate of treatment failure or death in children given zinc supplements [33].

Subgroup analyses were performed by age (< 12 months vs ≥ 12 mo), wasting (< -2 Z vs ≥ -2 Z weight-for-height), and sex [33]. For acute diarrhea, each of the subgroups had statistically significant benefits of zinc supplementation. For persistent diarrhea, the subgroups of < 12 months wasted and male had statistically significant effects; their corresponding alternatives had smaller beneficial effects that were not statistically significant.

TABLE 1. Trials evaluating the therapeutic effects of zinc supplementation on acute diarrhea

Country	No. in Zn/control groups	Enrollment		Zn supplement	Control supplement	Effect of Zn on episode duration	Effect of Zn on no. of episodes > 7 days	Effect of Zn on episode severity
		Age (mo)	Nutritional criteria					
India [34]	25/25	6–18	Excluded moderate to severe malnutrition	20 mg as sulfate	Placebo	9.4% shorter episodes	—	18.2% lower stool frequency
India [35]	456/481	6–35	Excluded severe malnutrition	20 mg as gluconate; vitamins A,B,D,E	Vitamins A, B, D, E	21.3% reduced probability of continuing diarrhea ^a	15% fewer	39% fewer watery stools ^a
Bangladesh [36]	57/54	3–24	Included if weight-for-age < 76th percentile	20 mg as acetate; vitamins A,B,D,E	Vitamins A,B,D,E	14.5% reduced probability of continuing diarrhea	23% fewer ^a	28% lower stool output
Bangladesh [37]	343/341	6–23	Excluded severe malnutrition	14 or 40 mg as acetate; vitamin C; half with vitamin A	Vitamin C; half with vitamin A	20% reduced probability of continuing diarrhea ^a	43% fewer ^a	—
Indonesia [38]	739/659	3–25	None	4–5 mg/kg	Placebo as acetate	11%	28% fewer shorter episodes ^a	—

a. Statistically significant ($p < .05$) effect.

TABLE 2. Meta-analysis of the therapeutic effects of zinc supplementation on the mean duration of acute diarrhea

	Zinc group	Control group	Effect size (95% confidence interval)	<i>p</i>
Trial	Mean ± SD			
India [34]	3.4 ± 1.8	3.8 ± 1.7	0.199 (−0.357, 0.755)	0.50
India [35]	4.5 ± 3.6	5.4 ± 3.4	0.238 (0.109, 0.367)	< 0.01
Bangladesh [36]	5.1 ± 2.5	5.5 ± 2.7	0.122 (−0.269, 0.513)	0.54
Bangladesh [37]	6.1 ± 5.1	7.1 ± 5.1	0.178 (0.028, 0.329)	0.03
Indonesia [38]	3.5 ± 2.4	3.8 ± 2.6	0.096 (−0.010, 0.201)	0.08
Summary estimate			0.162 (0.068, 0.2560)	

Effects of zinc supplementation on prevention of infectious diseases

Zinc deficiency has been associated with higher rates of infectious diseases, including skin infections, diarrhea, respiratory infections, and malaria, as well as with delayed wound healing. With regard to effects in developing countries, the most studied have been diarrhea and lower respiratory infections, although limited information on malaria is also available [44–50]. Because of the difficulty in assessing the zinc status of children in a population, most information on the effect of zinc deficiency on the risk of infectious disease comes from randomized, controlled trials of

zinc supplementation. In these trials, when zinc is the only experimental variable, there is direct causal evidence that additional zinc can result in lower rates of infectious diseases.

Seven trials of zinc supplementation provide information on outcomes of diarrhea, and four of these provide information on pneumonia (table 5). Most trials were performed with preschool children who were typical of poor developing-country populations, but two studies selected more poorly nourished children. These trials were done in seven different countries, representing a wide range of social and economic development and nutritional status.

The results with regard to diarrheal incidence are

TABLE 3. Trials evaluating the therapeutic effects of zinc supplementation on persistent diarrhea

Country	No. in Zn/control groups	Enrollment		Zn supplement	Control supplement	Effect of Zn on episode duration	Effect of Zn on episode severity	Effect of Zn on treatment failure or death
		Age (mo)	Nutritional criteria					
India [39]	20/20	6–18	Excluded moderate to severe malnutrition	20 mg as acetate	Placebo	18.9% shorter episodes	21.4% lower stool frequency	
Bangladesh [40]	95/95	3–24	None	20 mg as acetate	Vitamins A, B, D	15.3% reduced probability of continuing diarrhea	—	63% reduction ^a
Peru [41]	139/136	6–35	None	20 mg as gluconate	Placebo	18% reduced probability of continuing diarrhea	—	19% reduction
Pakistan [42]	43/44	6–36	Weight-for-age $\leq -2Z$	3 mg/kg as sulfate; vitamins A, B, D, E	Vitamins A, B, D, E	2.0% reduced probability of continuing diarrhea	No effect	58% increase
Bangladesh [43]	44/44	6–24	Weight-for-age < 76th percentile	20 mg as acetate; vitamins B, C, D; half with vitamin A	Vitamins B, C, D; half with vitamin A	55% reduced probability of continuing diarrhea ^a	—	75% reduction ^a

a. Statistically significant ($p < .05$) effect.

TABLE 4. Meta-analysis of the therapeutic effects of zinc supplementation on the mean duration of persistent diarrhea

	Zinc group	Control group		
Trial	Mean ± SD		Effect size (95% confidence interval)	<i>p</i>
India [39]	3.7 ± 1.1	3.8 ± 2.6	0.530 (−0.101, 1.160)	0.20
Bangladesh [40]	6.5 ± 3.7	7.0 ± 3.8	0.135 (−0.199, 0.470)	0.42
Peru [41]	2.2 ± 1.7	3.0 ± 2.5	0.360 (0.051, 0.670)	< 0.03
Pakistan [42]	5.1 ± 3.3	5.5 ± 2.7	0.122 (0.408, 0.652)	0.66
Bangladesh [43]	2.9 ± 1.4	3.5 ± 1.4	0.421 (−0.059, 0.901)	0.13
Summary estimate			0.293 (0.060, 0.525)	

consistent in showing that zinc-supplemented children have lower rates of diarrhea than control children. Most of these studies individually found statistically significant differences in diarrheal incidence, and a pooled analysis showed that overall the incidence of diarrhea in zinc-supplemented children was 18% (95% confidence interval, 7% to 28%) less than in unsupplemented children [51]. Since it also appears that zinc supplements reduce the duration of diarrhea, it is not surprising that the overall effect in the pooled analysis on the prevalence of diarrhea was greater than the effect on incidence, i.e., a 25% (95% confidence interval, 12% to 37%) lower prevalence of diarrhea in the zinc-supplemented children.

In subgroup analyses, there was a statistically significant pooled effect of zinc on diarrheal incidence among children 12 months of age or older (fig. 1), but no significant effect in the younger children (fig. 2). With regard to nutritional status, both subgroups ($< -2Z$ vs $\geq -2Z$ weight-for-age) had statistically significant pooled effects of zinc on diarrheal incidence (figs. 3 and 4). The effects were similar for boys and girls, although that for girls was statistically significant and that for boys was of borderline significance. Only two studies contributed to the subgroup analyses by plasma zinc status (< 60 vs ≥ 60 $\mu\text{g/dl}$), so the data were limited. There was a trend toward a benefit of zinc supplementation on the incidence of diarrhea in

TABLE 5. Trials evaluating the therapeutic effects of zinc supplementation on prevention of diarrhea or pneumonia

Country	No. in Zn/control groups	Child-years		Enrollment		Zn supplement	Control supplement	Effect of Zn on incidence of diarrhea	Effect of Zn on incidence of pneumonia
		Zn	Control	Age (mo)	Other criteria				
Vietnam [44]	73/73	30.8	30.8	4–36	Weight-for-age and height-for-age < -2Z	10 mg as sulfate	Placebo	44% less ^a	44% less ^a
India [45, 46]	286/293	122.9	124.8	6–35	Recovered from acute diarrhea	10 mg as gluconate; vitamins A, B, D, E	Vitamins A,B,D,E	8% less	43% less ^a
Mexico [47]	97/97	116.0	117.1	18–36	—	20 mg as methionate; half with iron	Placebo; half with iron	37% less ^a	—
Guatemala [48]	45/44	23.2	22.9	6–9	—	10 mg as sulfate	Placebo	18% less ^a	—
Jamaica [49]	31/30	7.1	6.2	6–24	Weight-for-age < -2Z	5 mg as sulfate; vitamins A, B, C, D	Vitamins A,B,C,D	8% less	88% less
Peru [41]	80/79	36.1	37.4	6–35	Recovered from persistent from diarrhea	10 mg as gluconate	Placebo	12% less ^a	15% less
Papua New Guinea [50]	136/138	75.3	80.7	6–60	—	10 mg as gluconate	Placebo	12% less	—

a. Statistically significant ($p < .05$) effect.

both subgroups, which was larger in the group with lower plasma zinc, but the effect was not statistically significant in either group [51].

Four studies provide information on the effects of zinc supplementation on the incidence of pneumonia [41, 44, 46, 49]. These studies consistently showed that zinc-supplemented children had lower rates of pneumonia, and two of these studies showed sizable and statistically significant effects individually [44, 46]. The other two studies had smaller numbers, and the differences did not reach statistical significance [41, 49]. Overall, in the pooled analysis, there was a 41% (95% confidence interval, 17% to 59%) lower rate of pneumonia in zinc-supplemented children [51].

Only two randomized, controlled trials provide information on the effects of zinc supplementation on clinical attacks of malaria. In the Gambia, with a twice-weekly 70-mg zinc supplement, there was a 46% reduction in clinic visits due to malaria, which was of only borderline statistical significance in this small study [52]. In a larger trial with daily zinc supplementation using 10 mg of elemental zinc as gluconate in Papua New Guinea, there was a statistically significant reduc-

tion of 40% in *Plasmodium falciparum* malaria clinic-based attack rates, and a higher efficacy (70%) for clinical attacks with parasite densities of more than 100,000/ μ l of blood [50].

Effects of zinc supplementation on child mortality

Pneumonia and diarrhea are the two most common causes of death in children in developing countries, and malaria also contributes substantially to mortality in many settings. With the large and consistent effects of zinc supplementation on the incidence and, in some cases, severity of these infectious diseases, one might hypothesize an effect also on child mortality. None of the studies of zinc supplementation done to date have been of sufficient size to fully address the effects on mortality. One recent study in India provides preliminary evidence that zinc-supplemented infants have a lower rate of overall mortality [53]. In this trial, among 1,250 small-for-gestational-age infants studied from one to nine months of age, there was

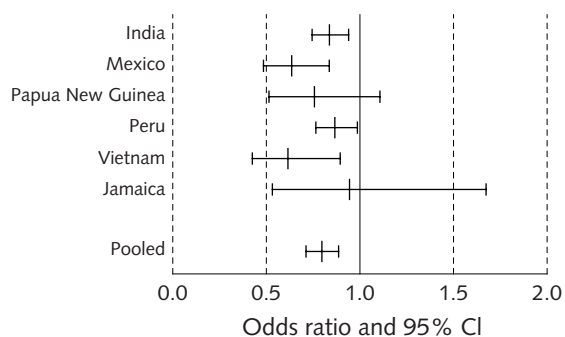


FIG. 1. Preventive effect of zinc supplementation on diarrheal incidence in continuous supplementation trials in children 12 months or more of age [51]

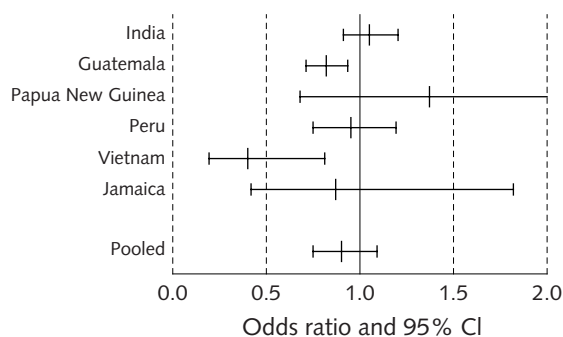


FIG. 2. Preventive effect of zinc supplementation on diarrheal incidence in continuous supplementation trials in children less than 12 months of age [51].

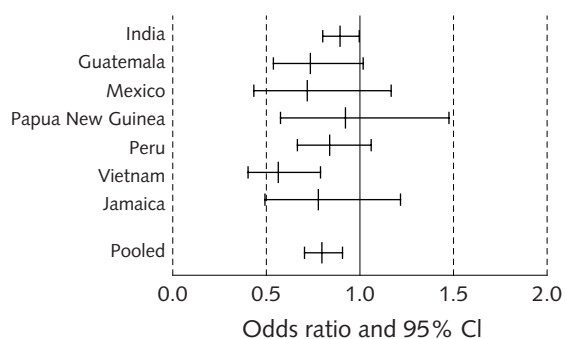


FIG. 3. Preventive effect of zinc supplementation on diarrheal incidence in continuous supplementation trials in children with weight-for-age Z-score < -2 [51].

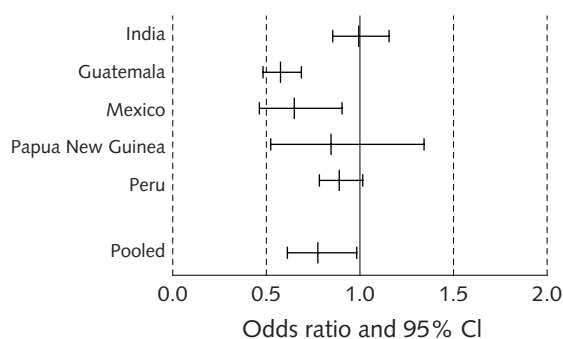


FIG. 4. Preventive effect of zinc supplementation on diarrheal incidence in continuous supplementation trials in children with weight-for-age Z-score ≥ -2 [51].

a 67% reduction in mortality in zinc-supplemented infants. Supplementation with selected other vitamins and minerals, including iron, was not associated with a significant reduction in mortality in this four-cell factorial design trial.

Conclusions

It is clear that zinc deficiency compromises many immune functions, although the direct relationship of these abnormalities to recovery from or risk of infectious diseases is not clear. However, there is now evidence that zinc supplementation can improve the outcomes of acute and persistent diarrhea and can prevent attacks of diarrhea and pneumonia, as well as possibly malaria. It is important to confirm these findings, especially those for pneumonia and malaria, for which results from diverse settings are limited.

Additionally, it is important to evaluate the effects of zinc supplementation on child mortality in potentially vulnerable developing-country populations. Plans are under way to conduct such trials in three settings—Zanzibar, India, and Nepal—and these studies should start within the next year. In the meantime, it appears that there is ample reason to consider public health applications using zinc either therapeutically or preventively. The therapeutic uses of zinc during diarrhea need further evaluation, particularly from the standpoint of testing feasible modes of delivery. For the preventive uses of zinc, there is need to explore various alternative ways to improve zinc nutriture of children in developing countries. These include improving the dietary quantity and bioavailability of zinc, fortifying foods with zinc, and providing supplements, perhaps in combination with iron and other micronutrients.

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