RESEARCH PAPER

Zinc Supplementation for Prevention of Acute Respiratory Infections in Infants: *A Randomized Controlled Trial*

AKASH MALIK, DAVENDRA K TANEJA, #NIVEDHITA DEVASENAPATHY AND *K RAJESHWARI

*From Departments of Community Medicine and *Pediatrics, Maulana Azad Medical College, New Delhi, India; and #Indian Institute of Public Health, Gurgaon, Haryana, India.*

Correspondence to: Dr Akash Malik, National RMNCH+A Unit, Room No. 107 D, Ministry of Health and Family Welfare, Government of India, Nirman Bhawan, New Delhi 110 001, India. drakashmalik28@gmail.com

Received: May 27, 2014; Initial review: June 13, 2014; Accepted: August 12, 2014.

Objective: To study the effect of 2 weeks of prophylactic zinc supplementation on incidence and duration of acute respiratory infections.

Design: Randomized double blind controlled trial.

Setting: Community based; urban resettlement area in North-East Delhi, India.

Participants: 272 children aged 6-11 months with acute respiratory infections. Children receiving zinc supplement within the past 3 months, severely malnourished, immuno-deficient, on steroid therapy, with severe illness requiring hospitalization, or children of families likely to migrate from the study area were excluded.

Intervention: Placebo (syrup base) or zinc (20 mg/5 mL elemental zinc as zinc sulfate) orally given for a period of 2 weeks.

Main outcome measure(s): Incidence, type and duration of acute respiratory infections, and adverse effects.

Results: No effect on incidence of acute respiratory infections was noted. A decrease of 15% (0.78-0.94) in days and 12% (0.78-0.94) in duration of episode in acute respiratory infections was observed. Incidence of acute lower respiratory infections decreased by 62% (0.26-0.36) and the effect remained for full five months of follow up. There were no drop outs due to side effects.

Conclusions: Prophylactic zinc supplementation for two weeks may reduce the morbidity due to acute lower respiratory infections but not overall rate of acute respiratory infections in infants aged 6-11 months in similar populations.

Keywords: *Micronutrient, Pneumonia, Public health, Prophylaxis.*

Trial Registration No. CTRI/2010/091/001417

Zinc is a vital micronutrient in humans and is essential for protein synthesis, cell growth, and differentiation and thus is important for functioning of the immune system [1]. Mild to moderate zinc deficiency is common in several developing countries, including India, because the commonly consumed staple foods have low zinc contents and are rich in phytates, which inhibit the absorption and utilization of zinc [2]. Strong evidence for a causal relationship between zinc deficiency and childhood infections has come from randomized controlled trials of zinc supplementation [3].

Acute respiratory infections (ARIs) especially Acute lower respiratory infections (ALRIs), are among the leading causes of death in children under the age of 5 years [4-6]. Zinc deficiency is projected to be responsible for 118,000 thousand deaths in children less than 5 years in developing countries [7]. Recent trials and meta-analyses have demonstrated that zinc supplementation both therapeutic and prophylactic reduces the duration, severity and incidence of ARIs [8,9]. However, most of these trials have used continuous supplementation, in a wide age group and produced variable results. The current study aimed to evaluate whether zinc prophylaxis for a short duration has any role in reducing the morbidity due to ARIs in apparently healthy infants of 6-11 months of age.

Accompanying Editorials: Pages 775-78.

METHODS

This was a community-based, randomized, double-blind, parallel-arm placebo-controlled trial, conducted from 1st January 2011 to 15th January 2012. We included all infants 6-11 months of age residing in Gokulpuri, an urban resettlement colony in North East District of Delhi, India, who were likely to stay till the completion of the study. Gokulpuri has a predominantly migrant population of about 23000, the majority belonging to the middle and lower socioeconomic strata. To achieve the final sample size, additional children were recruited from adjacent area of Gangavihar which has a similar population.

The study was approved by the Institutional Ethical Committee of Maulana Azad Medical College and Associated Hospitals, New Delhi.

We hypothesized that zinc supplementation for 2 weeks will reduce the incidence of ARIs in subsequent months. We excluded childred receiving zinc supplement in the past 3 months, those who were severely malnourished, known immuno-deficient or on steroid therapy, severely ill children requiring hospitalization, and children of families likely to migrate from the study area. A house-to-house survey was done at the beginning of the study to identify and recruit the eligible infants. The study purpose was explained and an informed consent was obtained from parents of all infants before they were recruited. The recruitment was done during first two weeks of January and July followed by subsequent five months of follow-up.

Intervention: The liquid preparations were prepared by Abyss Pharma, Delhi. Each 5 mL of the preparation contained placebo (syrup base) or zinc (20 mg elemental zinc as zinc sulfate). The syrups were of similar color (orange), taste (orange flavored), and consistency, and were packaged in similar bottles. We randomized the treatment allocation by simple randomization using computer generated random numbers (Excel 2010). The bottles were labeled with serial numbers after randomization in the Department of Community Medicine, MAMC, without the knowledge of the field investigator. The field investigator and parents were blinded to the treatment allocation till the end of follow-up period. The mothers received the bottles with labeled serial numbers and names. The field investigator administered the first dose of the intervention at the time of recruitment and advised the mother to give 5 mL of syrup (using standard 5 mL plastic spoon) daily to the infant for the remaining 13 days. Subsequently visits were made on the 7th and the 14th day to ensure compliance. In case the syrup had not been given regularly, a maximum of one week was given to complete the dosages. We collected data for any possible side effects as reported by the caregivers during these visits. To ensure that the child did not receive additional doses of zinc, we provided mothers with identity cards indicating the study title and that the infant was participating in the study. These cards were to be produced whenever the child was taken to any medical practitioner.

Outcomes and Follow up: The primary outcome was the incidence of ARIs per child-year. Secondary outcomes included incidence of Acute Upper Respiratory Infections (AURI) and ALRI per child-year, duration of ARIs, and side effects. AURI was diagnosed if the child had cough or cold with or without fever. ALRI was diagnosed if the child had symptoms of cough with difficult and/or rapid breathing or chest indrawing as informed by the caregiver [10]. Duration was assessed as the number of days with ARIs and as mean number of days an ARI episode lasted. A baseline assessment was done at the time of recruitment which included weight and length measurements using a Salter weighing Scale (up to 100 g) and an infantometer (up to 1 mm), respectively. All the outcomes were assessed by a trained field investigator based on history by caregiver.

Follow-up for ARIs began at the 15th day post-intervention. Each child was followed up fortnightly (± 3) days and the follow-up continued till 5 months after completion of zinc/placebo supplementation. At each follow-up, mother/caregiver was asked about history of ARIs during the previous 15 days. Recovery from an ARI episode was considered when the last day of ARIs was followed by a 72-hour ARI-free period [10]. Subsequent episodes were considered to be new ARIs episodes.

Sample Size: For sample size calculation, incidence of ARI was taken as 5.5 episodes (SD = 3.15) per child-year as per previous studies [11]. Thus, for a 20% reduction in the incidence of ARI (α 0.05 and power 80%), we required 258 infants (129 in each group). Taking into account possible 5% attrition, the final sample size was 272.

Statistical Analysis: The data were collected and checked for accuracy on a daily basis and entered in SPSS version 16. The incidence was expressed as episodes per child per year. The counts were expressed by means and standard deviation. Difference between means was tested using t-test, for normally distributed data or Mann Whitney U test, for skewed data.

Generalized Estimating Equations (GEE) were used to obtain an incident rate ratio with 95% confidence intervals, in order to compare month-wise number of episodes and duration of ARIs using Poisson log linear distribution, by intention to treat analysis. The exchangeable working correlation matrix was selected for all the outcomes. We included all children who had taken at least two doses of the intervention for the analyses. The follow-up visits for which the infant outcomes were not available were imputed using the worst case (2 episodes of ARI) and best case scenarios (no episodes). As it did not change the study results, the missing data were excluded from the final analysis. We decided to adjust the incident rate ratio for covariates which appeared to be different at baseline in the two groups. We also decided to compare the month-wise mean episodes of ARIs in the two groups.

Socioeconomic status was assessed using the Modified Kuppuswamy Scale (based on education and occupation of family head and total family income)

modified for Consumer Price Index for industrial workers of India for 2011 [12]. Z-scores for length and weight were calculated using WHO reference tables for length and weight [13,14].

RESULTS

From a total of 3155 households identified during the house to house survey, we assessed 272 infants for eligibility and all were recruited (***Fig.* 1**). Infants in both the groups shared similar baseline characteristics (***Table* I**). Seven families which migrated during the study period also shared similar baseline characteristics. Final analyses included 134 infants in the zinc group and 124 in the placebo group, who had completed the study. The mean number of follow-ups was 10 in each group. A total of 19 infants (13.5%) in zinc group and 26 infants (20%) in placebo group were given additional one week to complete the intervention as they were found to be initially non-compliant. The parents of infants who refused to be the part of the study even after this period were excluded from the analysis as refusals.

Out of the total 862 episodes observed, 424 episodes occurred in the zinc group and 438 in the placebo group, accounting for an incidence of 7.84 and 8.70 per child-year, respectively, at the end of 5 months (***Table* II**). GEE regression model showed that there was a non-significant reduction of 9% (Adjusted IRR 0.91, 95% CI 0.81-1.02) in episodes of ARIs in the zinc group as compared to the placebo group.

When types of ARIs were analyzed separately (***Table* II**), we found a non-significant increase of 1% in the episodes of AURIs (Adjusted IRR 1.01, 95% CI: 0.89-1.14). However a significant decrease of 62% in the episodes of ALRIs (Adjusted IRR 0.38, 95% CI: 0.26-0.36) was observed in the zinc group.

Zinc supplementation led to a significant reduction of 15% (Adjusted RR 0.85, 95% CI: 0.78-0.94) in days with ARIs. There was also a significant reduction of 12% in duration per episode of ARIs (Adjusted RR 0.88, 95% CI: 0.78-0.94) observed in the zinc group (***Table* II**). A subgroup analysis on wasted and stunted infants showed similar effects of prophylactic zinc (***Web Table* I**).

After the second month the episodes were almost similar in the two groups. Zinc prophylaxis significantly reduced the incidence of ALRI for all months of follow-up (***Web Table* II**).

Reported side effects were diarrhea, vomiting and constipation. The percentage of children reporting these were 9%, 10.4% and 1.5%, respectively in the zinc group and 7.3%, 4.8% and 0%, respectively in the placebo group; the difference was non-significant. A death due to diarrhea was reported in the zinc group three months after recruitment. Verbal autopsy revealed severe dehydration due to non-administration of oral rehydration solution or home available fluids.

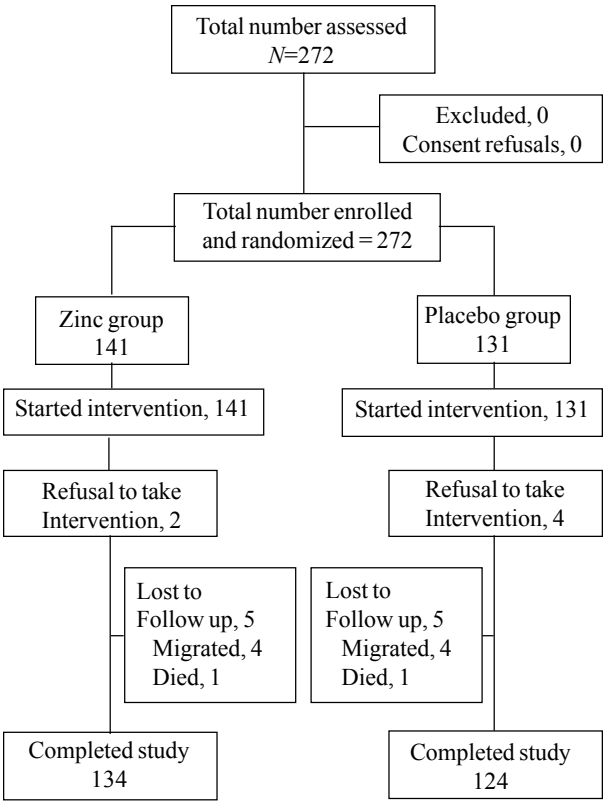

FIG. 1 *Trial flow.*

TABLE I BASELINE CHARACTERISTICS OF THE STUDY PARTICIPANTS

Characteristic	*Zinc (n=141)*	*Placebo (n=131)*
Male gender, *n* (%)	67 (47.5%)	68 (51.9%)
Mean age, mo; mean (SD)	8.77 (1.73)	8.76 (1.86)
Socioeconomic status		
Upper & upper middle	43 (30.5%)	34 (26%)
Lower middle and lower	98 (69.5%)	97 (74.1%)
Length for age, Z score*	-1.76 (1.46)	-1.69 (1.48)
Stunted, *n* (%)	51(36.0%)	54 (41.0%)
Weight-for-age Z score*	-1.50 (1.15)	-1.58 (1.21)
Wasted; *n* (%)	43(30.0%)	53 (41.0%)

**Mean (SD)*

DISCUSSION

We observed a significant reduction in duration of all

forms of ARIs after 14 days of zinc supplementation (20 mg/day). A significant decrease of 62% in the episodes of ALRIs was observed.

The major limitation of this study is that serum zinc levels were not done to assess the deficiency and the subsequent effect on serum zinc levels. Nevertheless, previous studies in similar populations of Delhi have shown that zinc deficiency is prevalent to the extent of 73.3% for values less than 10.4 μmol/L and 33.8% for values less than 9 μmol/L [15]. Moreover, in our study the proportion of stunted infants was more than 20%, which suggests an elevated risk of zinc deficiency since stunting is a proxy indicator of zinc deficiency in population studies [16]. Thus the results of this study may be extrapolated to similar zinc deficient populations only.

Previous studies done in healthy infants also observed a non-significant or no reduction in the total number of episodes of respiratory illness in the zinc group [17-21]. A meta-analysis [22] showed that, the children receiving zinc had fewer attacks of ARI (RR-0.92, 95% CI: 0.85-0.99), and fewer days with all ARI (RR-0.95, 95% CI: 0.84-1.07). However, the authors excluded studies with short-course prophylaxis in this meta-analysis.

Osendarp, *et al*. [19] reported 70% fewer episodes of ALRIs with zinc prophylaxis in healthy infants with low serum zinc levels at baseline. Though serum zinc levels were not measured in the current trial, the study population is expected to have low serum zinc levels [15,23]. However, in the above study, continuous zinc prophylaxis was given, ranging from 5 to 12 months which is in contrast to current trial. With the similar study setting, sample size and short course zinc prophylaxis of zinc (20 mg/d for 2 weeks), Rahman, *et al*. [10] reported that the incidence and prevalence of ALRI were significantly higher in the zinc group than in the placebo group after 6 months of follow-up. Other trials which had given zinc prophylxis for continuously long durations reported either a non-significant or no reduction in incidence of ALRI [24-26]. The meta-analyses of continuous and short course zinc proplylaxis on the other hand have concluded that zinc prophylaxis significantly reduces the incidence of ALRI [8,9,26-28]. Bhutta, *et al*. [9] also showed that point estimates of effects were not different in continuous and short-course zinc prophylaxis trials [9]. Despite different duration of zinc prophylaxis used in the above studies, the effect on ALRI was either similar or better in the current trial.

TABLE II EFFECT OF ZINC SUPPLEMENTATION ON INCIDENCE AND DURATION OF ARIS IN THE STUDY PARTICIPANTS

Outcome	*Zinc*	*Placebo*	*Adjusted IRR**
Child years observed	54.8	50.2	
ARI Incidence[$]	7.8	8.7	0.9 (0.81-1.02)
AURI Incidence[$]	7.2	7.2	1.0 (0.89-1.14)
ALRI Incidence[$]	0.5	1.5	0.3 (0.26-0.56)
Days[#] with ARI	11.4 (6.6)	14.7 (8.0)	0.8 (0.78-0.94)
Days[#] per episode ARI	3.6 (1.6)	3.8 (1.2)	0.8 (0.78-0.94)

**Incident rate ratio adjusted for wasting (95% CI); [#]Mean (SD); [$]episode/child/yr.*.

This trial on short course prophylactic zinc supplementation for 2 weeks in infants of 6-11 months has shown to cause a large decrease the incidence and duration of ALRIs in subsequent 5 months. Zinc prophylaxis in zinc deficient populations may significantly decrease morbidity due to ALRIs. The results of this study have important cost- and operational-implications as short-course prophylaxis of zinc in adequate dose might be more feasible than continuous therapy.

Contributors: AM: formulated the research question and contributed to study design, carried out the field investigations and initial data analyses, and wrote the initial draft; DKT: contributed to study design, and methodology and revised the manuscript; ND: supervised the data collection, analyzed the data, and interpreted the findings; KR; supervised the clinical data collection and management, and contributed to methodology. all authors approved the final manuscript.
Funding: Indian Council of Medical Research, Department of Health Research (Ministry of Health and Family Welfare) Government of India. Ref No. 3/2/2011/PG-thesis-MPD-10.
Competing interests: None stated.

REFERENCES

1. Biesel WR. Single nutrient and immunity. Am J Clin Nutr. 1982;35:417-68.
2. Black RE. Zinc deficiency, immune function, and morbidity and mortality from infectious disease among children in developing countries. Food Nutr Bull. 2001;22: 155-62.
3. Baqui AH, Black RE, Walker CLF, Shams A, Yunus M. Effect of zinc supplementation started during diarrhea on morbidity and mortality in Bangladeshi children: community randomized trial. BMJ. 2002;325:1059-65.
4. Pneumonia, the forgotten killer of children Geneva: UNICEF/ WHO; 2006. Available from: *http://whqlibdoc.who.int/publications/2006/9280640489_eng.pdf*. Accessed April 15, 2014.
5. Brian G. Global action plan for prevention and control of pneumonia (GAPP). Bull World Health Organ. 2008;86:322.
6. Walker CF, Rudan I, Liu L, Nair H, Theodoratou E, Bhutta ZA, *et al*. Global burden of childhood pneumonia and diarrhoea. Lancet. 2013;381:1405-16.

WHAT IS ALREADY KNOWN?

- Zinc supplementation for >3 months reduces the incidence and severity of acute lower respiratory tract infection but not overall acute respiratory tract infection in children aged 1-5 years.

WHAT THIS STUDY ADDS?

- Short-course (2 weeks) prophylactic zinc supplementation reduced acute lower respiratory tract infection morbidity in apparently healthy infants of 6 to 11 months over 5 months of follow-up.

7. Roth DE, Caulfield LE, Ezzati M, Balck RE. Acute lower respiratory infections in childhood: Opportunities for reducing the global burden through nutritional interventions. Bull World Health Organ. 2008;86:356-64.
8. Brown KH, Sonja YS, Peerson JM, Baker SK. Does preventive zinc supplementation of infants and young children affect their risk of selected illnesses, survival, and physical growth. Food Nutr Bull. 2009;30:S12-S40.
9. Zinc Investigators' Collaborative Group: Bhutta ZA, Black RE, Brown KH, Gardner JM, Gore S, *et al*. 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.
10. Rahman MM, Vermund SH, Wahed MA, Fuchs GJ, Baqui AH, Alvarez JO. Simultaneous zinc and vitamin A supplementation in Bangladeshi children: randomized double blind controlled trial. BMJ. 2001;323:314-8.
11. Zaman K, Baqui AH, Yunus M, Sack RB, Bateman OM, Chowdhury HR, *et al*. Acute respiratory infections in children: A community-based longitudinal study in rural Bangladesh. J Trop Pediatr. 1997;43:133-7.
12. Kuppuswamy B. Manual of Socioeconomic Status (Urban), Manasayan, Delhi, 1981.
13. World Health Organization. Child growth standards. Weight-for-age. Available from: *www.who.int/childgrowth/standards/weight_for_age/en/index.html*. Accessed April 14, 2014.
14. World Health Organization. Child growth standards. Height-for-age. Available at: *http://www.who.int/childgrowth/standards/height_for_age/en/index.html*. Accessed April 14, 2014.
15. Dhingra U, Hiremath G, Menon VP, Dhingra P, Sarkar A, Sazawal S. Zinc Deficiency: Descriptive epidemiology and morbidity among preschool children in peri-urban population in Delhi, India. J Health Popul Nutr. 2009;27:632-9.
16. de Benoist B, Darnton-Hill I, Davidsson L, Fontaine O, Hotz C. Conclusions of the joint WHO/UNICEF/IAEA/IZiNCG interagency meeting on zinc status indicators. Food Nutr Bull. 2007;28:S480-4.
17. Rosado JL, Lopez P, Mufioz E, Martinez H, Allen LH. Zinc supplementation reduced morbidity, but neither zinc nor iron supplementation affected growth or body composition of Mexican pre-schoolers. Am J Clin Nutr. 1997;65:13-9.
18. 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.
19. Osendarp JM, Santosham M, Black RE, Wahed MA, van Raaij JMA, Fuchs GJ. Effect of zinc supplementation between 1 and 6 mo of life on growth and morbidity of Bangladeshi infants in urban slums. Am J Clin Nutr. 2002;76:1401-8.
20. Long KZ, Montoya Y, Hertzmark E, Santos JI, Rosado JL. A double-blind, randomized, clinical trial of the effect of vitamin A and zinc supplementation on diarrheal disease and respiratory infections in children in Mexico City, Mexico. Am J Clin Nutr. 2006;83:693-700.
21. Heinig MJ, Brown KH, Lönnerdal B, Dewey KG. Zinc supplementation does not affect growth, morbidity, or motor development of US term breastfed infants at 4-10 mo of age. Am J Clin Nutr. 2006;84:594-601.
22. Aggarwal R, Sentz J, Miller MA. Role of zinc administration in prevention of childhood diarrhoea and respiratory illnesses: A meta-analysis. Paediatrics 2007;119:1120-30.
23. Bhandari N, Behl R, Taneja S, Strand T, Molbak K. Effect of routine zinc supplementation on pneumonia in children aged 6 months to 3 years: randomised controlled trial in an urban slum. BMJ. 2002;324:1358-63.
24. Baqui AH, Zaman K, Persson LA, Arifeen SE, Yunus M, Begum N, *et al*. Simultaneous weekly supplementation of iron and zinc is associated with lower morbidity due to diarrhoea and acute lower respiratory infection in Bangladeshi infants. J Nutr. 2003;133:4150-7.
25. Tielsch JM, Khatry SK, Stoltzfus RJ, Katz J, LeClerq SC, Adhikari R, *et al*. Effect of daily zinc supplementation on child mortality in southern Nepal: a community-based, cluster randomised, placebo controlled trial. Lancet. 2007;370:1230-9.
26. Roth DE, Richard SA, Black RE. Zinc supplementation for the prevention of acute lower respiratory infection in children in developing countries: meta-analysis and meta-regression of randomized trials. Int J Epidemiol. 2010;39:795-808.
27. Yakoob MY, Theodoratou E, Jabeen A, Imdad A, Eisele TP. Preventive zinc supplementation in developing countries: impact on mortality and morbidity due to diarrhoea, pneumonia and malaria. BMC Public Health 2011;11:S23.
28. Mathew JL, Patwari AK, Gupta P, Shah D, Gera T, Gogia S, *et al*. Acute respiratory infection and pneumonia in India: a systematic review of literature for advocacy and action: UNICEF-PHFI series on newborn and child health, India. Indian Pediatr. 2011;48:191-218.