

International Centre for Diarrhoeal Disease Research, Bangladesh

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Protocol No: 98-020

Date:

RESEARCH PROTOCOL RRC Approval: Yes/ No Date:

ERC Approval: Yes/No Date:

1. Title of Project (Do not exceed 60 characters including spaces and punctuations)
Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Below 2 Years of Age

2a. Name of the Principal Investigator(s) : 1) W. Abdullah Brooks, MD, MPH; 2) : ASG Faruque, MB,BS, MPH; 3) MA, Wahed, BSc; 4) George Fuchs, MD; 5) Robert Black, MD, MPH; 6) Mathuram Santosham, MD, MPH; 7) Abdullah H. Baqui, MBBS, Ph.D. **2b.** 1) Health & Child Survival Advisor; 2) Scientist; 3) Medical Biochemist 4) Director of Clinical Sciences Division. 5) Prof. & Chair Int'l Health, JHU. 6) Prof. Int'l Health, JHU; 7) Scientist **2c. Qualifications**

3. Name of the Division/ Branch / Programme of ICDDR,B under which the study will be carried out. Clinical Sciences Division/Laboratory Sciences Division

• **Contact Address of the Principal Investigator**

4a. Office Location:

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4b. Fax No: 883.116**4c. E-mail:** abrooks@citechco.net**4d. Phone / Ext:** 871.751 X 2327**5. Use of Human Subjects**Yes ☒ YesNo ☐**5a. Use of Live Animal**☐No ☐**5b. If Yes, Specify Animal Species****1. Dates of Proposed Period of Support**

Two years, to begin as soon as funding is available.

(\$):77,842 3rd Year: N/A**7. Cost Required for the Budget Period****7a. 1st Year (\$):152,278 2nd Year****7b. Direct Cost (\$):184,097 Total Cost (\$):230,120**

\$50,000 is being sought from SDC. Co-funding CHR Targeted Research and USAID is sought for the remainder.

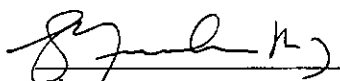
8. Approval of the Project by the Division Director of the Applicant

The above-mentioned project has been discussed and reviewed at the Division level as well by the external reviewers.

The protocol has been revised according to the reviewer's comments and is approved.

George Fuchs, MD

Name of the Division Director



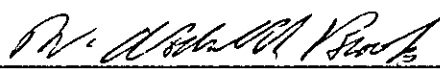
Signature

Date of Approval

1. Certification by the Principal Investigator

I certify that the statements herein are true, complete and accurate to the best of my knowledge. I am aware that any false, fictitious, or fraudulent statements or claims may subject me to criminal, civil, or administrative penalties. I agree to accept responsibility for the scientific conduct of the project and to provide the required progress reports if a grant is awarded as a result of this application.

10. Signature of PI



Date: 02.08.1998

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Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Below 2 Years of Age

Principal Investigator: W. Abdullah Brooks, MD, MPH

Co-principal Investigator: ASG Faruque, MBBS, MPH

Co-investigators: George Fuchs, MD; MA Wahed, BSc; Robert Black, MD, MPH; Mathuram Santosham, MD, MPH; Abdullah Baqui, MBBS, Ph.D.

ABSTRACT SUMMARY

The World Health Organization has identified diarrhoea and acute lower respiratory tract infection (ALRI) as high priority areas for intervention in order to reduce childhood morbidity and mortality. Prevention would address both of these objectives, and would have high cost benefit. Presently, there is no general prophylactic agent with demonstrated efficacy against these two diseases in this age group. The most vulnerable age group to ALRI and diarrhoea is that below two years of age. Limited data from community-based studies have reported preventive benefit from daily doses of zinc in children against both diarrhoeal and, more recently, acute lower respiratory tract diseases. Previous studies have not concentrated on the under two-year age group, nor has this group been shown to derive a clear benefit from zinc. We hypothesise that a relative zinc deficiency exists in the vast majority of infants below 2 years of age in developing countries and results in impaired cellular immunity needed to resist ALRI and diarrhoeal infections. We propose to supplement infants below two years of age with zinc once-weekly to determine whether or not zinc supplementation in young infants can lessen the incidence of ALRI and diarrhoeal disease. In so doing, we will get an estimate of the community incidence of ALRI in the under-two year old population, where disease-specific morbidity and mortality is highest. The results of this study will have major implications for the control of diarrhoeal disease and ALRI around the world.

1. Children less than two years of age are the most vulnerable to ALRI and diarrhoeal disease incidence and adverse outcome. Any study that seeks to measure the effect size of an intervention for this age group must be done in this age group. See Rationale in Design section, item 1.
2. Risks are minimal. There is some report of gastrointestinal upset in an extremely small number of infants, but this is of the same prevalence as has stomach upset from multivitamins. In any case it is self-limited. Higher doses of zinc have been used in this age group with no adverse effects whatever. In fact, no adverse effects have ever been reported from the doses of zinc proposed in this study in this age group.
3. Careful monitoring of patients by trained staff will continue throughout the study. The greater risks are from the diseases we aim to prevent. Zinc will not interfere with any standard treatment regimen, but may improve a child's response.

4. Confidentiality will be maintained by:
 - 4.1. Keeping all records in a secure place (field office) under lock and key and;
 - 4.2. Not sharing information with any one, or reporting information in a way that identifies any single patient.
5. Signed consent is to be obtained by trained personnel using a standardised form (see enclosed) at the time of the first interview of the child's caretakers (see Methods).
 - 5.1. Consent is signed.
 - 5.2. No information is withheld.
 - 5.3. No risk or invasion of privacy is involved, other than to take a detailed medical history of the child, which will be kept confidential as above, and to ask basic demographic data from the family (education, income).
6. Initial interview will take place at the home of the individual. This will determine eligibility for the study and obtain basic demographic and a brief medical history and physical exam. All subsequent interviews will be for surveillance for pneumonia and diarrhoea and will also take place at the child's home with the caretaker. Each interview will require 10 minutes.
7. Risks and Benefits of This Project. The potential benefits from this trial of oral zinc supplementation against two WHO-priority infectious diseases in young infants are practically unrivalled. These benefits extend to both the child (and its family) and the Centre (the health provider community). To the child and family, the intervention: may be efficacious as a prophylactic against a broad spectrum of infectious agents, possibly effective in the most vulnerable period of the infant's life, and may reduce the proportion of reactive airways disease that is triggered by respiratory infection. Furthermore, there may be an interaction with other primary prevention measures; cellular immunity may improve, for example, enhancing the child's immunological response to vaccines. The benefits may extend beyond immunity to other parameters of the child's wellbeing, such as improved nutritional status and growth - effects that other disease prevention measures like vaccines cannot offer. The intervention will not interfere with present therapeutic regimens for either ALRI or diarrhoea. The risks from this trial include: possible mild gastrointestinal irritation from the zinc gluconate(65); depression of copper status, which to date has not been reported, and; the risk of infection from non-sterile blood drawing technique.

The potential benefits to the health provider community include that it:

1. Addresses two WHO-priority diseases (ALRI, Diarrhoea) simultaneously;
2. Addresses two expert panel-identified issues in zinc:
 - Dosing;
 - Effect on copper status;
3. Seeks to intervene in the most vulnerable age group, < 2 year olds, hence could result in high cost benefit as a preventive measure;
4. Is safe (insofar as available data suggest);

5. Would have benefits other than immunological, i.e., nutritional and growth effects;
6. Intersects with new and emerging areas of interest such as IMCI, and will deepen our expertise in the area of ALRI, which is a major IMCI component;
7. Will provide data on Bangladeshi infants that no one presently has and which are relevant, if not vital, to our new and emerging areas of interest. These data include ALRI incidence in urban infants, and (new data on) diarrhoea incidence;
 - Provides capacity building for national staff in an emerging and important area, ALRI, which will continue to have benefits to the Centre after the project is completed.
8. The study requires no use of records, tissues, organs, or parts or foetal tissues., but it will use body fluids (blood for CBC and serum zinc and copper).

Consent Forms in English**Consent Forms in Bangla**☒

Check here if appendix is included

PROJECT SUMMARY: Describe in concise terms, the hypothesis, objectives, and the relevant background of the project. Describe concisely the experimental design and research methods for achieving the objectives. This description will serve as a succinct and precise and accurate description of the proposed research is required. This summary must be understandable and interpretable when removed from the main application. (TYPE TEXT WITHIN THE SPACE PROVIDED).

Principal Investigator **W. Abdullah Brooks, MD, MPH**

Project Name **Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Less Than 2 Years of Age**

Total Budget \$230,120
available, for two years.

Beginning Date As soon as funding is
Ending Date

ABSTRACT

The World Health Organization has identified diarrhoea and acute lower respiratory tract infection (ALRI) as high priority areas for intervention in order to reduce childhood morbidity and mortality. Prevention would address both of these objectives, and would have high cost benefit. Presently, there is no general prophylactic agent with demonstrated efficacy against these two diseases in this age group. The most vulnerable age group to ALRI and diarrhoea is that below two years of age. Limited data from community-based studies have reported preventive benefit from daily doses of zinc in children against both diarrhoeal and, more recently, acute lower respiratory tract diseases. Previous studies have not concentrated on the under two-year age group, nor has this group been shown to derive a clear benefit from zinc. We hypothesise that a relative zinc deficiency exists in the vast majority of infants below 2 years of age in developing countries and results in impaired cellular immunity needed to resist ALRI and diarrhoeal infections, and that by giving them zinc supplements, we could lower their incidence of ALRI and diarrhoea. We propose to supplement infants below two years of age once-weekly with zinc in a placebo controlled community trial to determine whether or not zinc supplementation in young infants can lessen the incidence of ALRI and diarrhoeal disease, and to assess the efficacy of a once-weekly regimen in preventing disease. In so doing, we will get an estimate of the community incidence of ALRI in the under-two year old population, where disease-specific morbidity and mortality is highest. The results of this study will have major implications for the control of ALRI and diarrhoeal disease around the world.

KEY PERSONNEL (List names of all investigators including PI and their respective specialties)

Name	Professional Discipline/ Specialty
Role in the Project	
1. W. Abdullah Brooks, MD, MPH	Preventive Medicine, Paediatrics
2. ASG Faruque, MB,BS; MPH	Scientist, Epidemiologist
3. MA Wahed	Laboratory Scientist
4. George Fuchs, MD	Gastroenterologist, Paediatrics
5. Robert E. Black, MD, MPH	Professor & Chair, Int'l Health, JHU
6. Mathuram Santosham, MD, MPH	Professor, Int'l Health, JHU
7. Abdullah H. Baqui, MBBS, Ph.D	Scientist, Epidemiologist

DESCRIPTION OF THE RESEARCH PROJECT**HYPOTHESIS**

1. Once-weekly zinc supplements will lower the incidence of ALRI and diarrhoeal diseases compared to placebo.
2. The effect size for lowering the incidence of diarrhoea is greater in children ≥ 12 months of age.

Specific Aims:**OBJECTIVES**

1. To determine if weekly zinc supplementation in zinc deficient young infants reduces the incidence of ALRI or diarrhoeal disease.
2. To determine if the effect size for diarrhoeal incidence reduction is affected by age.

SPECIFIC AIMS

1. To determine if 70 mg /week of zinc supplementation is effective in reducing the incidence of pneumonia and diarrhoea in infants 2 months – 24 months of age.
2. To determine if a once-weekly zinc supplementation regimen is as effective as the published effects of a daily regimen in preventing ALRI and diarrhoea.
3. To compare the effect on diarrhoea prevention in children less than 12 months to that in children 12 months of age or older.

SECONDARY AIMS

1. To estimate the effect of 12 mo Zn supplementation on growth.
2. To measure the effect of 12 mo Zn supplementation on Cu status.
3. To measure the effect of 12 mo Zn supplementation on Zn status.

Rationale:

SEE DESIGN, RATIONALE IS ITEMISED BY SPECIFIC AIM.

Background of the Project including Preliminary Observations

BACKGROUND

Acute lower respiratory infection (ALRI) is a leading cause of morbidity and mortality in children below five years of age (1). Pneumonia accounts for most of these deaths and is our principal area of interest. In fact, 25-33% of the deaths among children under 5 years are attributable to pneumonia (4 million deaths/year world-wide); two thirds of which occur during infancy and over 90% of which take place in developing countries (1,2). Acute respiratory illness (ARI), overall, ranks as one of the two leading causes of hospitalisation of children in Bangladesh (3). **It has been reported that the incidence of ALRI is 8-9 episodes/child/yr during the first 2 years of life, dropping to 3-4 episodes/yr in 2-4 year olds (7). Thus, the susceptibility to ALRI in children under two years is more than twice that of older children.** While overall child mortality in some developing countries has declined among younger age groups, the number of deaths attributable to ALRI has remained unchanged, suggesting an increasing importance of ALRI as a cause of death among young children (8). A pooled estimate from a recent meta-analysis concluded that a case management approach to children with rapid breathing could reduce total infant mortality by 20% and mortality for all children under five years by 25% (1). It further argued that a case management approach that relies principally on early diagnosis and appropriate antibiotic therapy could reduce pneumonia case fatality by as much as 50% (1). The most recent figures for ALRI incidence in Bangladesh are from a rural setting (79). Incidence for children below age 24 months ranged 0.2 – 0.5 episodes/child/year, averaging 0.3 episodes/child/year.

Diarrhoeal disease is also a worldwide problem of major importance in developing countries. **One fourth of avoidable deaths in hospitalised children is a result of diarrhoea, and 80% of these deaths occur in children <2 years of age (9).** The numbers of children dying each year from diarrhoea has decreased over the last decade from five to three million, in large part due to the widespread use of oral rehydration solution (ORS) (9). Despite the overall reduction in mortality, the persistently high mortality rate emphasises the need to develop additional strategies. The use of ORS to correct fluid and electrolyte abnormalities and antimicrobials to treat selected diarrhoeal diseases, including cholera and Shigella, remain central to the management of childhood diarrhoea in developing countries.

Despite advances in diagnosis and treatment, the total number of deaths attributable to both ALRI (pneumonia) and diarrhoea in children below five years of age is over 6 million, or 38% of all deaths (29). Recent data from Bangladesh indicate that ALRI is the most important cause of death in children under age 5 years, accounting for about 18% of all deaths (75). The effect is greater for younger age groups, where 30% of deaths in the one to 11 month age group is attributed to ALRI, while it accounts for 11% of deaths in the neonatal period, second only to neonatal tetanus. In the post-neonatal young infant age group, an additional 8.6% of young infant deaths were attributable to ALRI in combination with some form of diarrhoeal disease, while a further 10.6% of deaths were possibly due to ALRI, with or without concomitant infections. Thus, in Bangladesh as much as 50% of mortality among young infants is possibly attributable to ALRI (75). Furthermore, the under-five mortality trend has increased slightly from 18% in the period 1989-91 to 21% in the 1992-94. Understandably, WHO identifies ALRI and diarrhoea among the five priority areas in childhood illness for intervention as a means to decrease overall child morbidity and mortality (29).

There are established treatments for both ALRI (antibiotics, supportive care) and diarrhoea (oral rehydration therapy) (30,31,32). Their specific purpose has been to reduce mortality. Still, case fatality for both diseases remains high. Even if they can reduce mortality these treatments will have little impact on morbidity. Prevention alone reduces the incidence of morbidity, and may reduce mortality. Therefore, prevention must be the cornerstone of further efforts against these two priority diseases.

Vaccine-based prevention has had an impact on many but not all causes of acute pneumonia. At present, only a small proportion of all pneumonia is effectively prevented (35,36). Clearly, vaccine utilisation in the global community could be vastly improved, but cost and logistics are obstacles (34). Pneumococcal, RSV and rotaviral vaccines promise to make further reductions in the incidence of ALRI and diarrhoea. However, these interventions will face the same obstacles of cost and maintenance of cold chain integrity that have hampered existing vaccination programmes. Assuming that these vaccines are 100% efficacious and effective, a rotaviral vaccine could reduce diarrhoeal morbidity by no more than 46% (19). However, the currently licensed vaccine is only 80% efficacious. Similarly, an optimal RSV vaccine could reduce ALRI morbidity by no more than 40% (37). The efficacy of present pneumococcal vaccines in Asia, including the Subcontinent, is uncertain at this time (38,39) and trials are underway to address this issue. Clearly, there is a role for additional safe, relatively inexpensive, and easily administered preventive measures that can be integrated into existing childhood illness management strategies, and that can be applied to the most vulnerable age groups, irrespective of disease aetiology.

Cellular Immunity and Malnutrition

The link between nutrition and immunity is now well-known. One study stated that 56% of all deaths among children 6 to 59 months old had malnutrition as a potentiating factor, 83% of which was attributable to the mild-moderate form (74). Recent studies have reported a link between even sub-clinical malnutrition and impaired cellular immunity, and indicate that impaired cellular immunity is a risk factor for development of ALRI (7) and diarrhoeal disease (40,41). Several studies have demonstrated an association between malnutrition and the incidence and severity of pneumonia (7,42,43). Malnutrition is also believed to increase the risk of pneumonia-related mortality (1,44). One study reported that the risk of ALRI-related death for first, second and third degree malnutrition was 4.4, 11.3 and 27.0, respectively, compared to children without malnutrition (45). Other studies have reported similar increases in mortality risk for malnourished children (46,47).

Host factors are also major determinants of diarrhoea-associated morbidity and mortality (20). The interaction between diarrhoeal disease and malnutrition, which coexist in children from the developing world, is particularly significant (21,22). We now believe that deficiencies of several specific nutrients, particularly zinc, increase the risk of diarrhoea. Ironically, zinc deficiency is exacerbated by diarrhoea, which is often associated with gross increases in faecal zinc loss (23).

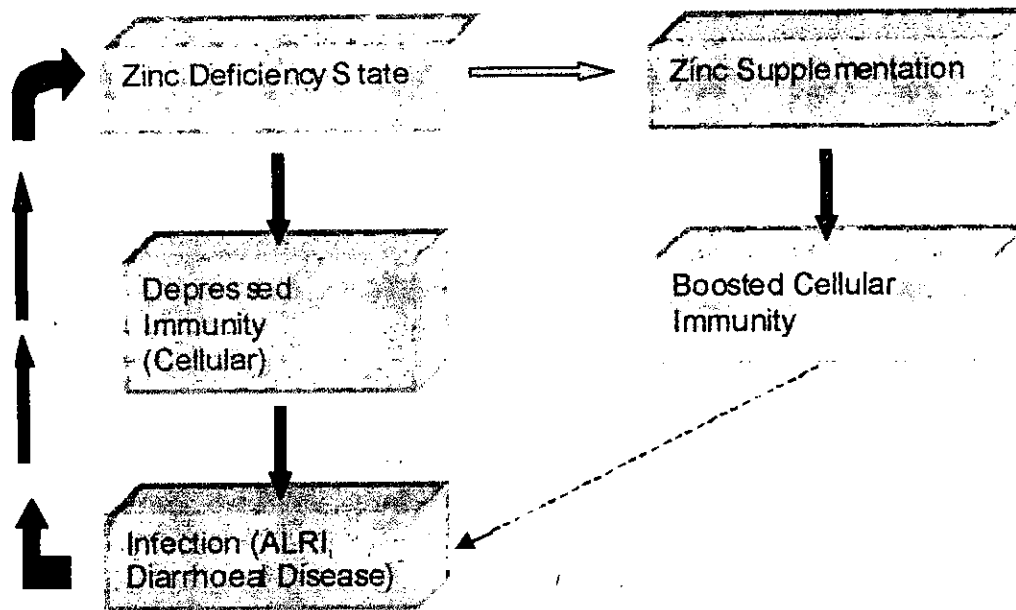
Zinc, Immunity and Morbidity

Current evidence on the interaction between nutritional status, immunity and disease has underscored the importance of cellular immunity (7), which has been documented in malnourished children (7,43,48). Evidence suggests that the impaired cellular immunity in malnourished children may be due, in part, to zinc deficiency since the latter is associated with impaired cellular immunity (7,42) and correction of zinc deficiency is associated with a reduction in the incidence and prevalence of diarrhoeal disease (24,25,26,27,48,49), and a

significant reduction in the incidence of respiratory infection (42,43). Indeed, there is evidence of a correlation between the severity of malnutrition and the magnitude of the effect on persistent diarrhoea from zinc supplementation (50). Figure 1 provides an overview of the interaction between the zinc deficiency state and the incidence of ALRI and diarrhoeal infection. In the infected state food, and therefore micronutrient intake, is less. In the case of diarrhoea, total body zinc loss is accelerated, possibly rendering the child more prone to future infections.

Figure 1 (Below).

Overview of ALRI/Diarrhoea Prevention Trial Hypothesis



Zinc Deficiency Linked to Increased Incidence ALRI and Diarrhoea

Zinc deficiency has been reported among young children in many developing countries (26,43,49) with prevalence as high as 38 - 50% (27,28). Both diarrhoea and ALRI have been associated with hypozincaemia, some of which may be attributable to accelerated zinc loss with diarrhoea (23,51,52). This sequence could create a loop or cycle between zinc deficiency, impaired cellular immunity and infection, with significant deleterious effects on the growth and survival of young infants.

Efficacy of Zinc as a Preventive Agent in Zinc Deficient Children

It is clear that host factors are important determinants of ALRI and diarrhoea-associated morbidity and mortality, and that nutritional status is one of the most critical host factors, affecting both immunity and resistance to infection.

Community-based controlled trials of zinc supplementation have reported reductions in the incidence of both ALRI and diarrhoeal disease in children (24,25,26,27, 28,42,43,48).

ALRI. A recent study reported a 45% (95% CI = 10-67%) reduction in the incidence of pneumonia among children aged 6 to 35 months old given zinc supplements over a six month period (42). Another recent study reported reductions in pneumonia incidence among

growth-retarded children aged 4 - 36 months given zinc supplements over 5 a month period (43). Two studies found no effect on ALRI incidence (26,49).

Diarrhoea One study reported a 26% lower incidence and a 35% lower prevalence of diarrhoea for children > 11 months old in a six month zinc supplementation trial (27). Two other recent studies reported diarrhoeal incidence reduction rates between 36% and 71% (43, 49).

Only one of these trials included children below six months of age (43), but the sample size was too small to draw any conclusions on this sub-group. All studies included children in older age groups, for whom ALRI and diarrhoeal disease incidence and mortality is substantially lower. Another of these studies only observed significant reduction in diarrhoea incidence among children one year of age and above (27, Black personal communication). Based on available epidemiology, reductions of incidence among the under two-year age group, the most vulnerable population, would have the biggest effect on ALRI and diarrhoeal disease. Mortality data on children presenting to our Centre indicate that 57% of deaths occur in children below one year of age (Figures 2 and 3).

Mortality Data for Oct96-Sep97

Figure 2.

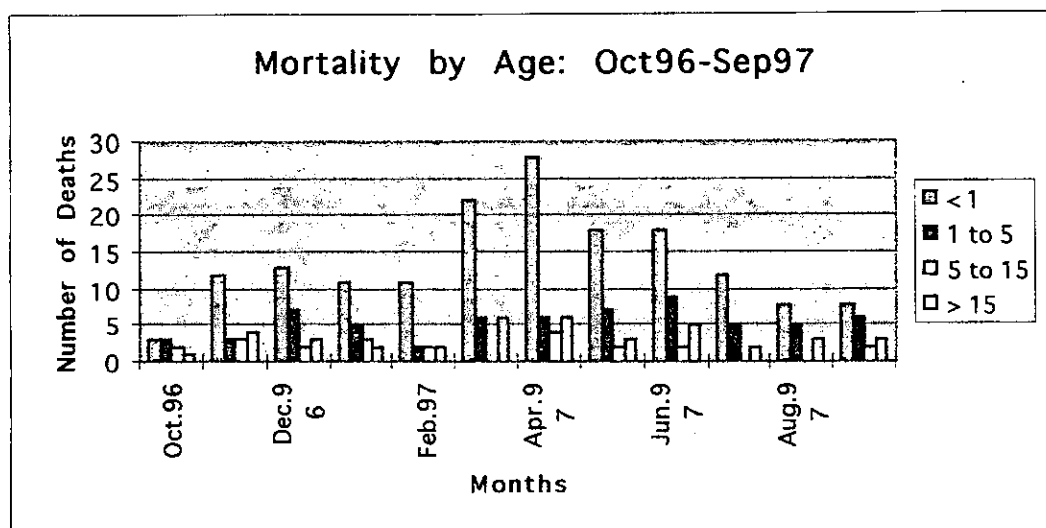
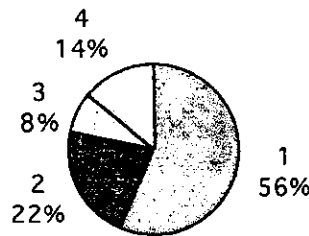


Figure 3.

Mortality Distribution for ICDDR,B Oct/96-Sep/97



*LEGEND

- Group 1 = < 1 year old
- Group 2 = 1 - 5 year old
- Group 3 = 5 - 15 year old
- Group 4 = > 15 year old

Mortality among children under two years comprises the majority of the remaining deaths of the under five-year age group (data not shown). **These findings are consistent with other data on this age group and indicate that the children under age two years are at the greatest risk for adverse outcome and most in need of an effective intervention (7,9).** Data on the interaction between zinc and disease incidence in young infants are inadequate. Most studies of zinc supplementation, to date, were not designed to test the effect of zinc specifically in the under two-year age group. Since older children do derive a benefit from zinc supplements, it is important to know whether or not the younger, more vulnerable age groups derive the same benefit. Finally, since there are data to suggest that many children do experience preventive benefits from zinc, studies that explore the efficacy of less frequent dosing strategies are needed. This study will provide the necessary data to assist national and international ALRI and diarrhoeal disease control programmes to set policy and devise programmatic strategies concerning the routine use of zinc in infants below 2 years of age who are zinc deficient.

Zinc Dosing, Toxicity and Effects on Other Micronutrients

The amount of zinc needed in young infants to restore and maintain positive balance, in areas with a high prevalence of zinc deficiency, is unknown. We could find no data from infants in our age range, however metabolic balance data from healthy adults indicate that turnover of body zinc is about 6 mg/day and that positive balance is maintained with an intake of 12.5 mg/day (56). The bioavailability of zinc ranges with the intake from 11% - 30% at < 7 mg/day in preadolescent girls (80) to 40% at 15 mg/day in adults (56). Thus 15 mg/day has been recommended for adults and 3 - 10 mg/day in children (55, 56). Most studies on ALRI have used 10 mg/day of elemental zinc, while many diarrhoeal disease studies, particularly on treatment effects, have used approximately 20 mg or more of elemental zinc per day. This latter dose is twice the US recommended daily allowance (RDA) for children one to eleven years of age, and four times the RDA for children under one year (55).

While an inadequate dose might fail to produce a preventive effect against disease, there is concern that excessive amounts of zinc can be harmful (56,57). The question is, what is

excessive in the young, zinc deficient infant. There are two categories of adverse effects, direct effects of zinc toxicity, and effects of zinc on other micronutrients, specifically copper. Dose-related adverse effects of zinc can be divided into three dosing categories: acutely toxic intakes, pharmacological intakes and moderately excessive intakes (62). Table 1 in the Addendum shows a breakdown of several studies that have reported zinc toxicity. *Acutely toxic* effects include nausea, vomiting, abdominal pain and cramping, and diarrhoea (62). These data are largely from case reports on food poisoning. This is uncommon and has consistently involved doses in the extreme range e.g. 1 - 6 g/day. *Pharmacological intakes* involve doses between 100 - 300 mg/day i.e. 10 - 20 times the recommended daily dose. Most of the adverse effects on **copper metabolism** have been reported in this dosing range. These adverse effects, including hypocupraemia, anaemia, leukopaenia and neutropaenia (62, 66, 67), arise from copper deficiency as a result of excessive zinc intake, and are reversed with cessation of zinc administration with or without copper supplementation (62, 81). Moreover, these effects on copper have been reported from studies utilising prolonged supplementation of ≥ 10 months, whereas studies utilising the same dose for shorter periods have not reported copper disturbances (62). A second category of adverse effect in this dosing range is a disturbance in **immunological function**. One study on 11 healthy adult males reported a depression in lymphocyte stimulation index, chemotactic migration and ingestion of bacteria after six weeks of supplementation at 300 mg/day (56). However, this study reported that none of the volunteers showed any clinical signs of illness. Another study reported depressed fungicidal activity of monocytes, but no differences in systemic illness between study subjects (58). A case report on a 13 month old female who received 120 mg/day for 6 months followed by 180 mg/day for one month reported listlessness, anaemia, neutropaenia, poor weight gain, abnormal sparse hair and bone changes consistent with copper deficiency (81). These effects were completely resolved one month after cessation of zinc and a short course of copper supplementation. Data on *moderately excessive* intakes of zinc (15 - 100 mg/day) have focussed on copper balance, and have attempted to assess the need for an increase in copper intake (61, 62, 82, 83, 84). One reported that as the amount of zinc in the diet increased, so too did the amount of copper required to maintain positive balance (83), while another found no effect on copper balance at a dose of 16 mg/day in adult females (84). Two others recorded an increase in faecal loss of copper (61, 82). One reported that the increase in faecal copper loss was statistically significant (61) and the other that it was not (82).

The results of these studies and case reports are mixed. Haematological changes have only been reported from studies using high doses for at least 10 months, and changes in immunological indices have occurred in shorter periods, but without clinically apparent disease. Excluding the reports of isolated cases of poisoning, the majority of these studies at the pharmacological or moderately excessive range are small studies with very low power, as shown in the addendum (56, 58, 62). The only study with standard power did not report clinically apparent ill effects (61). Perhaps more importantly, in a study in the Gambia on 110 infants aged 0.57 - 2.30 years, there were no clinically apparent differences between those supplemented with 70 mg *twice weekly* and the placebo group. Moreover, there were no significant haematological or immunological derangements in the supplemented group (78). On balance, there is *no* evidence that zinc in the range of 10 - 20 mg/day causes any toxic effects or clinically significant copper deficiency in infants.

A more practical question related to dose is its frequency. If the same dose that has been associated with preventive benefit on a daily basis can be given less frequently, while retaining the same efficacy, this would have logistical benefits, cost savings and might

enhance compliance. This alone warrants an investigation of the efficacy of less frequent dosing.

At two recent zinc symposia, one at Johns Hopkins University in Baltimore, Maryland during November, 1996 and another at the International Centre for Diarrhoeal Disease Research, Bangladesh in Dhaka on 15-16 May, 1997, an international expert panel agreed that further research on the efficacy of zinc and its dose is needed in the management of childhood disease before universal recommendations can be made (73). It was also agreed that assessment of copper status should be included in new zinc supplementation trials. Only by the further accumulation of quality data can questions on the dose, the frequency of delivery and the effects on copper status be clearly answered. This study can make a significant contribution to all of the above.

Risks and Benefits of This Project

The potential benefits from this trial are substantial. There are benefits to both the lay and health provider communities. To the lay community, the study may provide an efficacious prophylactic against a broad spectrum of infectious agents, at a stage in young infants' lives when they are most vulnerable to infection and death. Furthermore, there may be an interaction with other primary prevention measures; cellular immunity may improve, for example, enhancing the child's immunological response to vaccines. The benefits may extend beyond immunity to other parameters of the child's wellbeing, such as improved nutritional status and growth - effects that other disease prevention measures like vaccines cannot offer. The intervention will not interfere with existing prevention or therapeutic regimens for either ALRI or diarrhoea. The risks from this trial are minimal and include: possible mild gastrointestinal irritation from the zinc gluconate (65), discomfort from phlebotomy and the risk of infection from non-sterile blood drawing technique.

The potential benefits to the health provider community are equally significant. The study:

1. Addresses two WHO-priority diseases;
2. Addresses two expert panel-identified issues in zinc;
3. Dosing;
4. Effect on copper status;
5. May provide an effective intervention for the most vulnerable age group, which could result in high cost benefit as a preventive measure;
6. Provides data of direct programmatic relevance on the efficacy of less frequent dosing;
7. May have benefits other than immunological, i.e., nutritional and growth effects;
8. Will deepen our expertise in ALRI, which is a major component of the strategic plans of several organisations, including WHO and USAID;
9. Will provide important data, presently not available, on the incidence of pneumonia among urban Bangladeshi infants;
10. Provides capacity building for national staff in an emerging and important area.

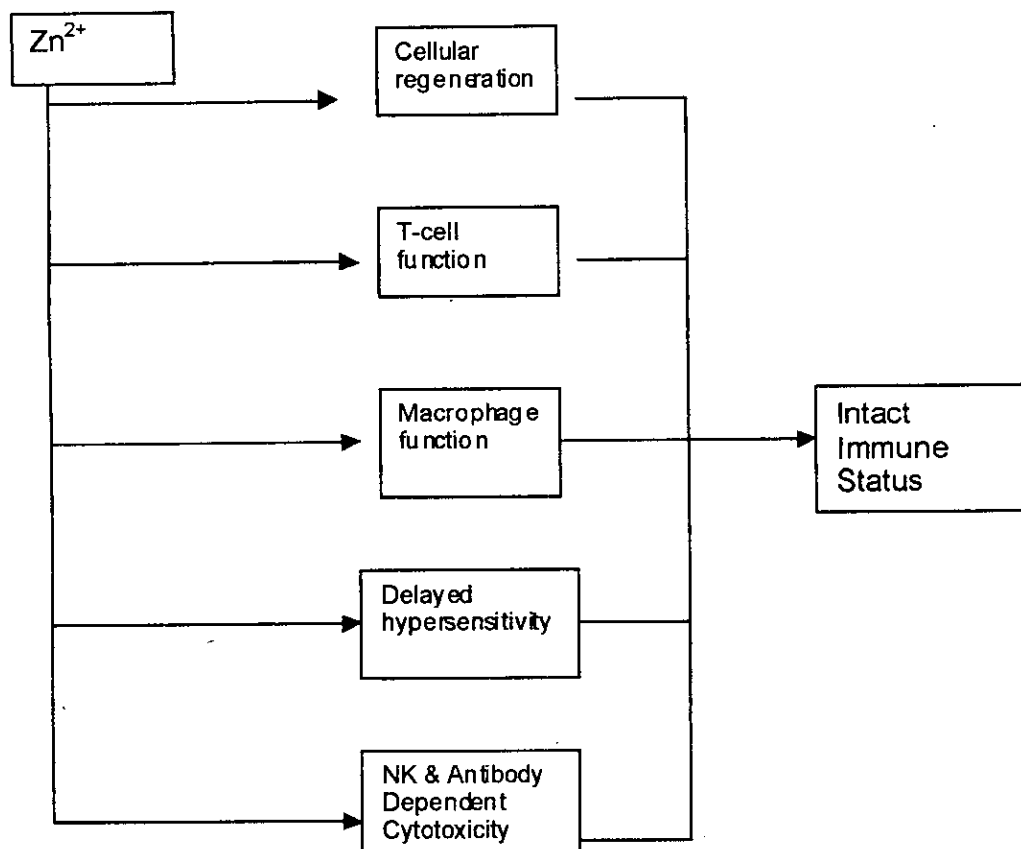
General Goal and Significance

ALRI is the leading cause of infection in infants and, taken together with diarrhoeal disease, accounts for about 40% of all infant mortality. Our long-term goal is to reduce the incidence of community-acquired pneumonia and diarrhoea with a safe preventive agent that will not impair the effectiveness of other prevention strategies.

Theoretical Model (Figure 4.)

Zinc deficiency: (i) impairs cell regeneration (due to its role in metalloenzymes, and other metabolic pathways); (ii) impairs immunocompetence with reduced cell mediated immune response, decreased T- lymphocytes, and abnormal T-Helper and/or suppresser cell functions(11), (iii) impairs macrophage function (11), thus depressing the delayed hypersensitivity reaction; (vi) reduces the natural killer cells and antibody dependent cytotoxicity (12), (v) impairs epithelial barrier functions. We believe that the combined effect increases the risk of infection in an infant exposed to ALRI and diarrhoeal pathogens.

Figure 4.



Research Design and Methods

DESIGN

A community-based double-blind placebo controlled preventive trial.

1. To test Specific Aim 1. *Rationale.* The under-two year old population represents the most vulnerable to ALRI and diarrhoea, has been shown to be zinc deficient in the developing world, including Bangladesh, and zinc may boost immune function and thereby reduce the incidence of these two diseases. *Protocol.* Infants aged 2 to 11 months old will be recruited from two adjacent communities (see Population, below) with a combined total population of 44,000. A request to participate will be made at the first home visit during the mapping phase. An intake interview and baseline physical examination will be made during the first interview on all willing participants. Infants will then be randomised to one of two groups (TABLE 1) and followed in the community for twelve months. Group A will

receive 70 mg of elemental zinc once weekly and Group B will receive once weekly placebo. A local pharmaceutical company (e.g. ACME, Opsonin) will prepare these according to the enclosed specifications (see **Methods**).

TABLE 1.

Age	A	B
	Zinc 70 mg/week	Placebo/week
2 – 11 mo	338	338
12-23 mo	338	338
Total	776	776

Community Follow-up. Enrolled infants will be followed in the community once weekly for 12 months. The weekly visits will have two objectives: 1) to administer the weekly zinc supplements; 2) to gather data on intercurrent ALRI or diarrhoeal episodes. Parents will be asked about signs of ALRI: rapid or difficult breathing, cough with or without danger signs (inability to feed, lethargy); and diarrhoeal illness: (three or more loose stools), by day for each of the days since the previous visit. The health worker will use a calendar to assist in data collection. Each infant's respiratory rate will then be counted for one full minute irrespective of reported illness. If it is elevated, the rate will be counted twice and the mean recorded by the health worker. The medical officer will visit any infant noted by the health worker to have a respiratory rate > 50/minunte or any danger signs, as defined in Methods. The medical officer will auscultate the chest and make a clinical diagnosis, based both on auscultation and clinical observation of danger signs. **Thus, diagnosis of illness, and therefore determination of incidence, will be clinical.** The severity of illness and its management will be made according the Methods section. Infants with pneumonia (see Definitions in Methods) will be managed as outpatients with amoxycillin (see Methods). Infants with severe pneumonia will be referred to ICCDR,B hospital, Dhaka for treatment. Infants with diarrhoea but some or less dehydration (see Methods) will be given ORS packets and instructions for use and the criteria for seeking medical attention. Infants with signs of severe dehydration will be referred to ICDDR,B hospital.

Data. Demographic data will be *categorical*. Univariate analysis using two by two tables and Chi Square tests will be performed between outcome (ALRI and diarrhoeal illness) and each covariate to assess interaction. Multivariate analysis will be done to control for confounding. A final model of high risk will be determined by logistic regression. Morbidity data will be *continuous*. *ALRI-associated*; episodes of age-specific tachypnoea, chest indrawing, fever. *Diarrhoea-associated*; episodes of three or more loose watery stools per day or bloody mucoid stools. These will be calculated as **incidence** (mean frequency of occurrence during the study period per group), and **prevalence** (mean duration of symptoms by treatment group). Analysis type: ALRI incidence is likely to be infrequent and have a small mean. This will result in a Poisson distribution and be analysed accordingly. Longitudinal surveillance data on incidence for each child will be divided into child-periods of 30 days each and analysed by Generalised Estimating Equations (GEE). The advantage of this method of analysis is that it does not assume independence of morbid events, and so prevents overestimation of significance from correlation of morbidity within children. Thus, we will derive the most conservative estimate of effect from zinc supplementation. A model will be created using GEE with logit link, binomial variance and exchangeable

correlation. Incidence will be calculated by modelling each child period as a bivariate dependant variable with treatment group status as the covariate. Interactions between the dependant variable and other covariates, such as age, sociodemographic variables, nutritional status, zinc and copper status, will be similarly analysed. Interaction within the model will be assessed by the Wald test.

Difficulties and Alternatives. 1) Missed visits. Infants may not be present at the time of the health worker visit. Any infant not present will be re-visited by the healthworker within 24 hours of that scheduled visit. A tally of missed visits will be kept and included in the analysis. If that is not successful, then the information will be obtained on the next scheduled visit. No recalled data greater than seven days old (i.e. collected after the next scheduled visit from the original date) will be accepted. Missing days will be subtracted from the denominator during the analysis. We will use intention to treat analysis on all children unless they have < 30 days of observation, in which case they will be dropped from the analysis. 2) Missing information. Certain data, such as past medical history, immunisation history, may be difficult to ascertain. Information that can be verified by records will be (such as immunisation cards). Further, where there is record-report discordance, the written record will be entered into the data set.

2. To test Specific Aim 2. *Rationale.* To assess the relative efficacy of a less intensive, therefore easier to administer dosing regimen, which would have significant programmatic implications. *Protocol.* Infants will be randomised to one of two groups. Group A will receive 70 mg once per week. Group B will receive a once weekly dose of placebo, as described above. The healthworker will give the dose of supplement at each visit (once-weekly). In order to detect treatment group differences, power is set at 90% in the sample size calculations, and determines the sample size. The magnitude of effect for ALRI and diarrhoea are the conservative estimates published from previous studies and cited in the Background section. *Data.* The outcome will be morbidity data and will be analysed as described above. A comparison of our effect magnitude will be made against those published for daily doses in children of comparable ages. *Difficulties and Alternatives.* 1) Concern over single dose. We have discussed the perception that 70 mg once weekly may be a large amount of zinc at once, since the RDA for infants less than 12 months is 5 mg of elemental zinc. However, Bates *et al.* supplemented young Gambian children from 0.5 to 2.3 years of age with 70 mg elemental zinc *twice* weekly over a 1.25 year period with no adverse effects, as discussed above (78). None are anticipated here. The data suggest no risk of either toxicity or significant copper deficiency. 3) Masking. Zinc is bitter. Flavouring will be added to disguise the taste. Infants in Group A may react to the taste in such a way as to provoke suspicion in the caretaker and healthworkers. Every attempt will be made to ensure the tastes are comparable. 3) The less intense regimen could affect compliance but there will be no way to directly measure this effect since there will be no comparison daily dose. We will attempt to make comparisons against daily doses by comparing our rate of compliance with that found in comparable studies using a daily regimen. 4) As there are no published studies to date that have documented a significant positive effect in children in the under two year age group, effect size comparisons for prevention of ALRI and diarrhoea between our studies and others will not be exact. This will be a particular problem if we detect no effect. On the other hand, if our effect size against ALRI and diarrhoea for the older age group (12-23 mo) is comparable to that observed in daily dose studies, we will be able to estimate the comparative efficacy against daily dosing for the younger age group.

3. To test Specific Aim 3. *Rationale.* Studies have documented a greater preventive effect in children above 12 months of age than in younger infants. More data are needed to better define these effect sizes. *Protocol.* Infants will be post-stratified into two age groups in the analysis of the effect on diarrhoea; 2 to 11 months, 12 to 23 months. Stratified randomisation will not be used since the treatment groups are large (76). *Data.* Morbidity data will be analysed as outlined above. *Difficulties and Alternatives.* Minimal. Post-stratification will not affect randomisation, thus will not affect recruitment time or introduce bias. We took stratification into account in the sample size calculation, so power will not be diminished. However, gains in statistical precision will be small to nil if age is not related to outcome.

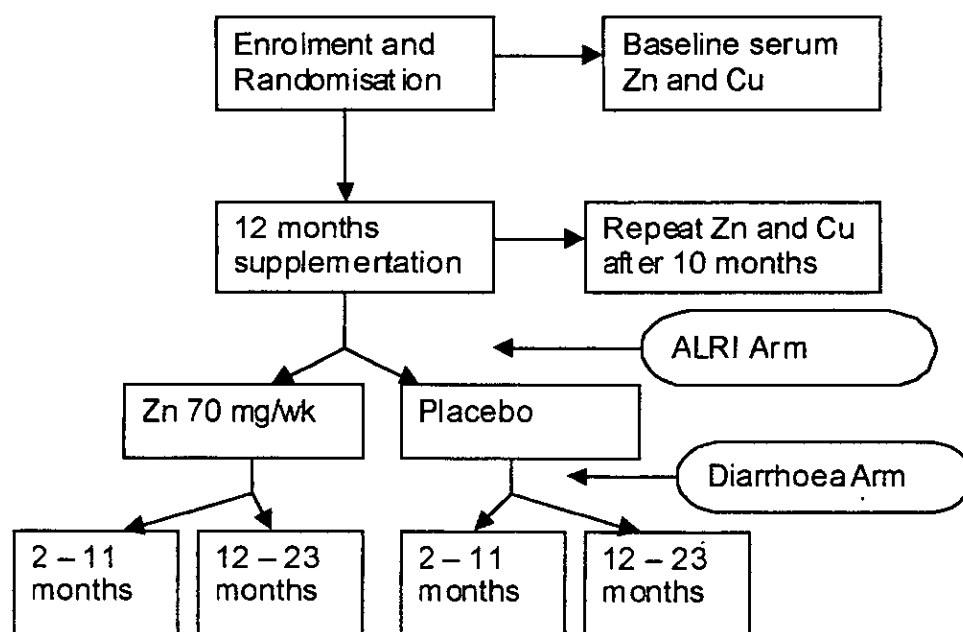
4. To test Secondary Aim 1. *Rationale.* To document whether or not improved isolated zinc status enhances growth in young infants. *Protocol.* Anthropometric measurements of weight and height will be collected at baseline and at three-monthly intervals on each infant (every twelfth visit). Weight will be measured twice to the nearest 1 gram and height will be measured twice to the nearest 0.1 cm. The health worker will record the mean of both measurements. The anthropometric measurements will be numbered 0 - 4, beginning with the baseline measurement at the time of enrolment. The research assistant and the principal investigators will check to ensure that each patient has a total of seven measurements. *Data.* Growth: Height, weight. Calculated as z-scores for weight for age (WAZ), height for age (HAZ) and weight for height (WHZ). We will calculate geometric means and standard deviations by treatment group, and measure inter-group variation by ANOVA or ANCOVA. *Difficulties and Alternatives.* Anthropometric measurements in the field are susceptible to imprecision if health workers are hurried. Second, inter-observer variation can affect overall growth outcome. Two countermeasures will be used. First, we will hire adequate numbers of staff so that a health worker sees 20 infants per day. Second, we will test and quantify the degree of inter-observer variation as part of the health worker training. A 10% sub-sample of measurements will be screened to monitor gross error. If variance remains high, a 20% sub-sample will be brought to clinic for anthropometry. These data will be used in the analysis.

5. To test Secondary Aim 2. *Rationale.* Infants with marginal copper status may be pushed into frank copper deficiency by an antagonistic divalent ion, such as zinc. Clinical signs of copper deficiency should be correlated with haematological parameters. *Protocol.* At baseline a 5 ml blood sample will be obtained for measurement of copper, haemoglobin and white blood cell count (WBC). Copper will be analysed by atomic absorption spectrophotometry. On the first visit after 300 days of supplementation, 5 ml of blood will be drawn from each patient and stored in serum isolator tubes for later transport back to the Centre. Serum will be analysed for copper. Serum copper levels will be correlated with clinical signs of copper deficiency, such as anaemia, leukopaenia, and neutropaenia, osteoporosis and pathological fractures resembling scurvy, as well as growth during data analysis. *Data.* Continuous numerical data will be summarised by treatment group, and the mean and standard deviations will be compared by ANCOVA. These can be directly regressed on values for haemoglobin, WBC and neutrophil count. Transformation into categorical data will allow for modelling of high-risk infants. *Difficulties and Alternatives.* 1) Sample collection. The community has a perception that health workers may collect blood for later sale to local hospitals for profit (private communication to the PI from community leaders). Caretakers have expressed distress about the discomfort caused to the infant during phlebotomy. Finally, prolonged transport times can affect the preservation of the serum samples. To circumvent these difficulties, we will bring infants to a field office for

blood drawing. Blood samples will then be labelled and stored for transportation later that day to the hospital laboratory. The cost of patient transportation, if needed, will be provided by the project. If drawing blood from the entire population is impracticable, a 20% sub-sample will be randomly selected and used for the analysis.

6. To test Secondary Aim 3. *Rationale.* Baseline zinc status is central to the hypothesis of a high prevalence of zinc deficiency. As this has been reported in previous studies, its demonstration is not a primary objective here. *Protocol.* Baseline and post 300 days supplementation blood samples will be drawn, as discussed above. *Data.* The data will be the same as for copper status, and will be handled as discussed above. *Difficulties and Alternatives.* These will be the same as for copper, and will be handled in the manner discussed above.

Protocol summary. Figure 5.



Timeline. The entire study is projected to last for two years, as outlined below.

Figure 6.

Task	Time	In	Months						
Mapping	X								
Training	X								
Recruitment	X	X							
Zn Supplmnt		X	X	X	X	X	X		
Blood work		X				X			
Lab analysis							X		
Data entry		X	X	X	X	X	X	X	
Data analysis							X	X	
Writeup							X	X	X
	3	6	9	12	15	18	21	24	

METHODS

List of Contents

1. Definitions (Diagnoses)
2. Inclusion, exclusion criteria
3. Sample size calculation
4. Preparation of zinc and placebo supplements
5. Population and study site mapping
6. Randomisation
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8. Clinical management
9. Anthropometry
10. Laboratory
11. Quality assurance
12. Consent procedures

Definitions

ALRI: Acute lower respiratory tract infection defined by respiratory rate (RR) alone has a low predictive value (42,69). In a recent community-based trial in India (42), when fever or lower chest indrawing was added to elevated respiratory rate, it increased the specificity from 58% to 82% and the positive predictive value (PPV) from 57.5% to 77.4%. We will use chest indrawing as described below.

Severe Pneumonia: RR > 50/min for infants with *any* of the following: chest indrawing, central cyanosis, inability to feed, lethargy. No differentiation will be made between severe and very severe pneumonia in this trial, as this distinction will not affect treatment.

Pneumonia: RR > 50/min but *none* of signs for severe pneumonia.

A clinical exam with a finding of rales (crepitations) is necessary for diagnosis of severe pneumonia or pneumonia.

Severe dehydration: will be defined according to the Dhaka method (See Appendix).

Duration of illness. Seven or more symptom-free days (i.e., one intervening symptom-free visit) differentiate episodes of pneumonia, and three or more symptom-free days differentiate episodes of diarrhoea. The diagnosis and category of treatment recommended (outpatient or inpatient) will be recorded on the surveillance form, irrespective of whether or not the patient complies with the recommendation.

De jure population comprises the normal residents of a community, including those who may be absent during the enumeration.

De facto population consists of those who slept in the locality the night before the census was taken. Most national censuses record the *de facto* population, whereas in an intervention trial the *de jure* is the more appropriate (77).

Inclusion Criteria

Infants between 2 and 11 months, inclusively, at the time of enrolment, who comprise the *de jure* population of the target communities of Gulapbagh or Dholpur and whose parents give consent for their participation in the study.

Exclusion Criteria

Children with known reactive airways disease (i.e. have been placed on standing or PRN albuterol or other β_2 - agonist), tuberculosis, severe malnutrition that requires hospitalisation, congenital abnormalities, or who do not normally reside in the community.

Sample Size Calculation

Diarrhoea Range is 4.6 - 5.0 episodes/child/year for Dhaka urban incidence of diarrhoea (72). In order to detect a 20% reduction in the incidence of diarrhoea, using a SE of 2.5 and calculating 95% confidence and 90% power: we will need 163 infants per group. Allowing for 20% withdrawal gives 196/group for a total of 784 infants.

Pneumonia In a recent study of zinc supplementation in urban children 6 - 36 months old in the Subcontinent (42), *pneumonia incidence* among controls was calculated as 0.35 episodes/child/year. There are no recent published data on pneumonia incidence in this age group for Dhaka. The incidence of true pneumonia in Matlab for the 2 month to 23 month age group is 0.2 episodes/child/year (Abdullah Baqui, personal communication). In order to detect a 30% decrease in the incidence of ALRI, (0.2 episodes/child/year), with a probability of a Type 1 error of 5% and a Type 2 error of 20% we will need 647 infants per group. Allowing for 20% withdrawal from the study, we will recruit 776 infants per group for a total population of **1,552**.

Preparation of Zinc Supplements

Design of treatment groups

Infants will be randomised to one of two groups (TABLE 1). Group A will receive 70 mg of elemental zinc and Group B will receive placebo, both once weekly. The health workers will dispense the dose at the time of the weekly visit.

Requirements for zinc and placebo

1. Zinc gluconate for Group B (70 mg/week):
Concentration 14 mg/ml. Dose is 70 mg/5 ml. Total = 52 doses per infant. Volume = 260 ml.
2. Placebo for Group B 5 ml per dose for total of 52 doses. Volume = 260 ml.
3. Both groups will receive one 300 ml bottle that will be kept in the infant's home.

Special instructions:

1. Supplement and placebo have to be identical in taste, appearance, and smell. The taste of the zinc needs to be masked as well (being given to young infants 2 -24 months)
2. To dissolve the zinc gluconate, any base substance normally used for vitamin supplements can be used, but must be vitamin-free
3. Taste may be improved by non-nutritious substances only (flavours, sucrose)
4. Any preservative normally used in vitamin syrups may be used to maintain shelf-life up to 12 months
5. Bottles must have a measurement scale attached to show amount for each dose.

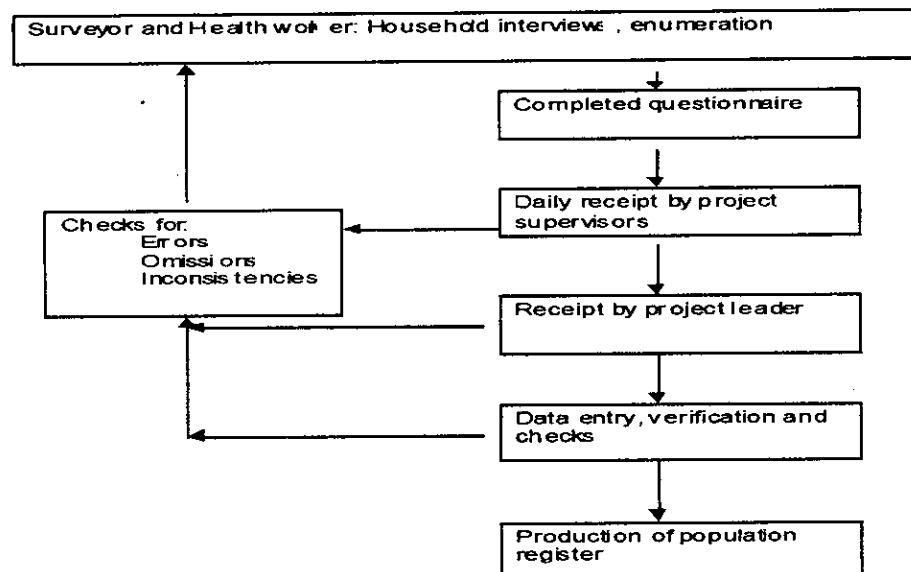
Population and Study Site Mapping

In order to find the requisite number of patients, assuming the less than two-year old population constitutes 4.5% of the total population, we would need a community of $2000/0.045 = 25,778$ or about 26,000 pop in the community. A community has been identified to serve as the study site, Dholpur, which has recently served as an intervention site for a community-based education intervention trial, and its adjacent community, Gulapbagh. Both are urban slum areas in the southwestern quarter of the city of Dhaka. Both have a population of 10,000 households, each with about 4 members per household. The under-five year population comprises about 20%. Taking the most conservative estimate of 4.5% for the under-two population as above allows us to utilise both areas for selecting treatment and placebo groups.

The area will need to be mapped. The average ground speed of a trained surveyor is 200 households/day in a densely populated urban area (77). Combined with other agenda, such as census taking, the rate may be halved. We will deploy 10 teams consisting of one mapping assistant and one health worker. They will perform mapping and listing among 20,000 households in the study site. At a rate of 100 households per day, working five days per week, it will take 10 teams 4 weeks to complete the task.

There are well-trained staff and a field research officer available to the project familiar with that area and its inhabitants. These can be paired with contracted surveyors (masters' students from graduate schools in surveying programmes).

Flow chart for census and data collection (Adapted from Smith and Morrow).



Randomisation

Effectively all eligible infants in the study site will be selected, thus avoiding the need for random selection. Infants recruited will be randomised at the time of consent to one of the three treatment groups. This will be *fixed randomisation*. Allocation of study subjects will be uniform, as the assignment probabilities for test treatments and control groups are equal, according to the formula:

$$P_1 = P_2 = \dots = P_i = \dots = P_{t+1}$$

Where P_i , $i = 1, \dots, t$, denote assignment probabilities for the t test treatment and P_{t+1} denotes the assignment probability for the control group, and where $(t+1) \sum_{i=1}^t P_i = 1$. Randomisation will be achieved by permuted blocks of variable length, where the smallest

block size will be equal to the sum of integers of the allocation ratio, according to the formula $(t+1)\sum_{i=1}^t r_i = B$, where B is the minimum block size. As there are three treatment groups, $B = 3$. Thus, the range of block sizes will be 3 to 12. The larger the block the greater the likelihood of departure from the allocation ratio, so blocks greater than 4 times the number of treatment cells will be avoided. The block sizes used for the construction of the allocation schedule will not be divulged to the project personnel until after the trial to avoid unmasking.

Data Collection

Background personal, demographic and medical information; including identification (name, age, date of birth), birth history, previous hospitalisations or treatments for respiratory complaints, and feeding history; will be gathered using a standardised form at the time of enrolment (see APPENDIX). Trained staff members only will collect data using the standardised forms.

Clinical Management

Treatment of Severe Infection: Infants observed by the visiting nurse or the caretaker to have severe pneumonia or diarrhoea with severe dehydration or bloody mucoid stools will be referred to ICDDR,B hospital for admission and treatment, according to existing hospital standards. Hospitalisation, treatment and transportation costs will be provided by the project.

Treatment of Non-severe Infection:

ALRI/ Infants with non-severe ALRI or diarrhoea will be treated in the community according to published WHO guidelines (70). This will be either Amoxycillin 40 mg/kg PO + TID, or Cotrimoxazole 10 mg TMP PO + BID. Both antibiotics will be continued for 5 days. Attempts will be made to visit every infant on outpatient management after 48 hours of therapy. Caretakers will be instructed to bring infants to the field clinic if they believe the infant's condition is worsening. Infants who fail to improve after 48 hours of antibiotics, or whose condition worsens on antibiotics in the judgement of the medical officer or the investigators will be referred to hospital for definitive care.

Diarrhoea Infants with diarrhoea and "some" or "no sign" of dehydration *and* no history of bloody mucoid stool will be offered ORS packets. Caretakers will be instructed to manage them according to published WHO guidelines. For "Some" dehydration this will be 50 – 100 ml after each loose stool, up to (approximately) 500 ml per day. The health team will reassess infants who require more for possible referral to hospital. If there is "No sign" of dehydration, caretakers will be advised to give extra fluid, continue feeding (particularly breastfeeding), and to notify the health team if any of the following develop: the infant starts drinking poorly or is unable to drink, becomes sicker, develops a fever, has blood in the stool or makes urine less than three times per day.

Medications for ALRI and dysentery, and ORS packets for diarrhoea will be provided by the project.

Anthropometry

Weights will be obtained to the nearest 1g using an electronic digital scale standardised with a one kilogram standard weight each morning and prior to weighing each subject. Supine length measurements will be made with a metal sliding board. The mean of two consecutive measurements to the nearest 0.1 cm for all indices will be recorded as the observed value.

Measurements will be compared to the standards of the National Center for Health Statistic (NCHS) data. Stunting will be defined as Z score of < -2 for length or height for age (> 2 standard deviations below the median for the National Centre for Health Statistics reference population); and wasting as a Z score of < -2 standard deviations for weight for length.

Laboratory Procedures

ICDDR,B Hospital, Dhaka, has a modern medical laboratory and routinely performs standard haematological tests, such as haemoglobin, haematocrit, white blood cell count and differential count according to internationally established methods. Samples will be collected in polypropylene tubes to avoid zinc contamination.

Zinc Assay. Samples (200 μ l) in polypropylene tubes are diluted 1:12 with HNO₃ and Briiz-35. Zinc is determined by flame atomic absorption spectrophotometry (AAS) using standard solutions prepared in the same matrix. AAS is equipped with nebulizer assembly (Shimadzu) and C₂H₂/air burner. Zinc hollow-cathode lamp is operated with background correction wavelength of 213 nm. Determination of zinc is done by aspiration of a series of working standards (at least 4) and a standard curve is prepared for each lot by plotting the concentration (mg/l) versus absorbance using software provided by the manufacturer. Two quality control sera (low and high range) are run with each lot and if any value of these quality control samples is not within the acceptable range, the result is ignored and the whole lot is repeated.

Copper Assay. Samples (400 μ l) in polypropylene tubes are diluted 1:12 with HNO₃ and Briiz-35. Copper is determined by flame atomic absorption spectrophotometry using standard solutions prepared in the same matrix. Copper Again, a C₂H₂/air burner is used. AAS wavelength is 324.7 nm.

Quality Assurance

A 10% sample of the visits will be randomly selected and repeated by the research assistant or the investigators within 24 hours of the visiting health team. Periodic review and testing of field staff will be done and the degree of inter-observer variation will be quantified. The percentage of agreement and kappa statistic will be calculated to assess inter-observer variation. Spot checks at infants' homes will be made by the staff to measure the residual amount of zinc syrup in the bottles in order to assess compliance.

Consent Procedures

Consent will be obtained using a standardised form prior to enrolment in the study. Parents will be advised that their children's participation in this experimental study is voluntary and that they may withdraw their children from the study at any time, and that as new information becomes available that might affect this study, it will be made available to them (SEE APPENDIX).

Facilities Available

Describe the availability of physical facilities at the place where the study will be carried out. For clinical and laboratory-based studies, indicate the provision of hospital and other types of patient's care facilities and adequate laboratory support. Point out the laboratory facilities and major equipments that will be required for the study. For field studies, describe the field area including its size, population, and means of communications. (TYPE WITHIN THE PROVIDED SPACE).

The hospital is an approximately 250 bed facility. Ill subjects who require hospitalisation will be brought back to hospital and treated in either the Short Stay Ward, a 120 bed facility that has an average stay of 20 hours, or the General Ward, a 60 bed facility. The existing hospital laboratory facilities will be used. The hospital laboratory processes approximately 18,000 serum samples per year for a wide range of tests. It possesses an atomic spectrophotometer that will be used to process our serum zinc and copper samples.

Data Analysis

Describe plans for data analysis. Indicate whether data will be analyzed by the investigators themselves or by other professionals. Specify what statistical softwares packages will be used and if the study is blinded, when the code will be opened. For clinical trials, indicate if interim data analysis will be required to monitor further progress of the study. **(TYPE WITHIN THE PROVIDED SPACE).**

X. Data analysis: SEE DESIGN ABOVE FOR DETAILS.

Ethical Assurance for Protection of Human Rights

Describe in the space provided the justifications for conducting this research in human subjects. If the study needs observations on sick individuals, provide sufficient reasons for using them. Indicate how subject's rights are protected and if there is any benefit or risk to each subject of the study.

XI. Consent: SEE METHODS ABOVE and ENCLOSED CONSENT FORM.

Use of Animals

Describe in the space provided the type and species of animal that will be used in the study. Justify with reasons the use of particular animal species in the experiment and the compliance of the animal ethical guidelines for conducting the proposed procedures.

N/A

Literature Cited

Identify all cited references to published literature in the text by number in parentheses. List all cited references sequentially as they appear in the text. For unpublished references, provide complete information in the text and do not include them in the list of Literature Cited. There is no page limit for this section, however exercise judgment in assessing the "standard" length.

REFERENCES

1. Sazawal S, Black RE. Meta-analysis of intervention trials on case-management of pneumonia in community settings. Lancet 1992;340:528-33
2. Bang A, Bang R, Tale O, Sontakke P, Solanki J, Wargantwar R, Kelzarkar P. Reduction in pneumonia mortality and total childhood mortality by means of community-based intervention trial in Gadchiroli, India. Lancet 1990;336:201-06

- and physical growth of children in rural Bangladesh I. Patterns of morbidity. *Am J Epidemiol* 1982; 115:305-14
4. Selwyn BJ. The epidemiology of acute respiratory tract infection in young children: comparison of findings from several developing countries. *Rev Infect Dis* 1990 (Suppl 8): S870-S888
 5. H Borrero I, P Fajardo L, M Bedoya A, Zea A, Carmona F, Borrero MF de. Acute respiratory tract infections among a birth cohort of children from Cali, Colombia, who were studied through 17 months of age. *Rev Infect Dis* 1990; 12(Suppl 8): S950-S956
 6. Wafula EM, Onyango FE, Mirza WM, Macharia WM, Wamola I, Ndinya-Achola JO, Agwanda R, Waigwa RN, Musia J. Epidemiology of acute respiratory tract infections among young children in Kenya. *Rev Infect Dis* 1990; 12(Suppl 8): S1035-S1038
 7. Zaman K, Baqui AH, Yunus M, Sack RB, Chowdhury HR, Black RE. Malnutrition, cell-mediated immune deficiency and acute upper respiratory infections in rural Bangladeshi children. *Acta Pædiatr* 1997; 86:923-7
 8. Hortal M, Benitez A, Contera M, Etorena P, Montano A, Meny M. A community-based study of acute respiratory tract infections in children in Uruguay. *Rev Infect Dis* 1990 (Suppl 8); 12: S966-S973
 9. WHO/CDD/SER/80.2: A manual for the treatment of acute diarrhoea. Programme for Control of Diarrheal Diseases, 1990. Geneva World health Organization, 1990.
 10. Institute of Medicine: Prospects for immunizing against rotavirus. *In* New vaccine development. Establishing priorities. Diseases of importance in developing countries. Vol. 2. National Academy Press, 1986, pp. 308-318.
 11. Birch CJ, Lewis FA, Kennet ML, et al.: A study of prevalence of rotavirus infection in children with gastroenteritis admitted to an infectious diseases hospital. *J Med Virol* 1977;1:69-77.
 12. Bryden AS, Davies HA, Hadley RE, et al.: Rotavirus enteritis in the West Midlands during 1974. *Lancet* 1974;2:241-243.
 13. Gurwith MJ, Williams TW. Gastroenteritis in children: A two-year review in Manitoba. I. Etiology. *J Infect Dis* 1977;136:239-247.
 14. Kapikian AZ, Kim HW, Wyatt RG, et al. Human reovirus-like agent as the major pathogen associated with "winter" gastroenteritis in hospitalized infants and young children. *N Engl J Med*. 1976;294:965-972.
 15. Ostravik I, Figenschau KJ, Haug KW, et al. A reovirus-like agent (rotavirus) in gastroenteritis of children. *Scand J Infect Dis* 1976;8:1-5.

16. Wyatt RG, Kapikian AZ, Thornhill TS, et al. In vitro cultivation in human fetal intestinal organ culture of a reovirus-like agent associated with nonbacterial gastroenteritis in infants and children. *J Infect Dis* 1974;130:523-528.
17. Mata L, Simhon A, Urrutia JJ, et al. Epidemiology of rotaviruses in a cohort of 45 Guatemalan Mayan Indian Children observed from birth to the age of three years. 1983;148:452-461.
18. Guerrant RL, Kirchoff LV, Schields DS, et al. Prospective study of diarrheal illnesses in northeastern Brazil: Patterns of disease, nutritional impact, etiologies, and risk factors. *J Infect Dis* 1983;148:986-997.
19. Black RE, Merson MH, Mizanur Rahman ASM, et al. A 2 year study of bacterial, viral and parasitic agents associated with diarrhea in rural Bangladesh. *J Infect Dis* 1980;142:660-664.
20. Pelletier DL. Potentiating effects of malnutrition on child mortality: epidemiologic evidence and policy implications. *Food Nutr Bull* 1995;16:206-13.
21. The World Health Report 1995: bridging the gaps. World Health Organization, 1995.
22. The progress of nations 1995. UNICEF, 1995.
23. Hambidge KM. Zinc and diarrhea. *Acta Paediatr* 1992, vol;81Suppl 381: 82-6.
24. Vallee BL Biochemistry, physiology and pathology of zinc. *Physiol Rev* 1959;39: 443-89.B
25. Rosado JL, Allen LH Lopez P et al. The effect of zinc and/or iron supplementation on morbidity: a double blind, randomized community trial in preschoolers. *The FASEB J*. 1995;9(3): abstracts part1 A157.
26. Ruel MT, Rivera JA, Santizo MC et al. Impact of zinc supplementation on morbidity from diarrhea and respiratory infections among rural Guatemalan children. *Pediatrics* 1997; 99:808-813
27. Sazawal S, Black RE, Bhan MK, et al. Zinc supplementation in young children with acute diarrhea in India. *N Engl. J Med* 1995;333:839-44.
28. Sachdev HPS, Mittal SK, Bhan MK, et al. A controlled trial on utility of oral zinc supplementation in acute dehydrating diarrhoea in infants. *J Pediatr Gastroenterol Nutr* 1988;7:877-81
29. WHO Division of Child Health Development. Integrated Management of Childhood Illness (IMCI) Information. September 1997
30. Rahman MM, Aziz KMS, Patwary W, et al. Diarrhoeal mortality in two Bangladeshi villages with and without community-based Oral Rehydration. *Lancet* 1979;2:809-812.

31. Pizzarro D. Oral rehydration in infants in developing countries. *Drug* 36 1988;(suppl. 4):39-47
32. UNICEF. A simple solution. UNICEF Special Report. 1987.
33. Salam AS. Antimicrobial Therapy for Shigellosis: Issues on Antimicrobial Resistance. Abstract. 1998.
34. Orenstein WA, Markowitz LE, Hinman AR. Diseases Controlled Primarily by Vaccination: Measles. Cited in Maxcy-Rosenau-Last. *Public Health & Preventive Medicine*, 13th Edition. Appleton & Lance. 1992;65-68.
35. Fauveau V, Stewart MK, Chakraborty J, Khan SA. Impact on mortality of a community-based programme to control acute lower respiratory infections. *Bull WHO* 1992;70:109-116.
36. Koenig MA, Khan MA, Wojtyniak B, et al. Impact of measles vaccination on childhood mortality in rural Bangladesh. *Bull WHO* 1990;68:441-47.
37. Parrot RH, Kim HW, Brandt CD, et al. Respiratory syncytial virus in infants and children. *Prev Med* 1974;3:473-480.
38. Saha SK; Rikitomi N; Biswas D; Watanabe K; Ruhulamin M; Ahmed K; Hanif M; Matsumoto K; Sack RB; Nagatake T. Serotypes of *Streptococcus pneumoniae* causing invasive childhood infections in Bangladesh, 1992 to 1995. *I J Clin Microbiol* 1997 Mar;35(3):785-7;
39. Black S, Shinefield H. Issues and challenges: pneumococcal vaccination in pediatrics. *Pediatr Ann* 1997 Jun;26(6):355-60
40. Baqui AH, Black RE, Sack RB, Chowdhury HR, Yunus M, Siddique AK. Malnutrition, cell-mediated immune deficiency and diarrhoea: A community-based longitudinal study in rural Bangladeshi children. *Am J Epidemiol* 1993;137:355-365.
41. Black RE, Lanata CF, Lazo F. Delayed cutaneous hypersensitivity: epidemiologic factors affecting usefulness in predicting diarrhoeal incidence in young Peruvian children. *Ped Infec Dis J*. 1989;8:210-215.
42. Sazawal S, Black RE, Jall S, Mazumdar S, Sinha A, Bhan MK. Zinc supplementation reduces the incidence of acute lower respiratory infection in infants and preschool children: A double-blind controlled trial. *Manuscript Pediatrics* 7/97.
43. 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.
44. Spooner V, Baker J, Tulloch S, et al. Clinical signs and risk factors associated with pneumonia in children admitted to Goroka Hospital, Papua New Guinea. *J Trop Pediatr* 1989;39:295-300.

45. Tupasi TE, Velmonte MA, Sanvictores MEG, Abraham L, Leon LED, Tan SA, Miguel CA, Saniel MC. Determinants of morbidity and mortality due to acute respiratory infections: implications for intervention. *J Infect Dis* 1988; 157: 615-623
46. Heywood PF. Nutritional status as a risk factor for mortality in children in the high of Papua New Guinea. In: Taylor TG, Jenkins NK, eds. *Proceedings of the Thirteenth International Congress of Nutrition*. London: John Libbey, 1986: 103-6
47. Escobar JA, Dover AS, Duenas A, Leal E, Medina P, Arguello A, de Gaiter M, Gracer DL, Spillman R, Reyes MA. Etiology of respiratory tract infections in children in Cali, Colombia. *Pediatrics* 1976; 57: 123-30.
48. Rosado JL, Lopez P, Munoz E, Martinez H, Allen LH. Zinc supplementation reduced morbidity, but neither zinc nor iron supplementation affected growth or body composition of Mexican preschoolers. *Am J Clin Nutr* 1997;65:13-9.
49. Schlesinger L, Arevalo M, Arredondo S, et al. Effect of zinc-fortified formula on immunocompetence and growth of malnourished infants. *Am J Clin Nutr*. 1992;56: 481-498.
50. Roy SK, Tomkins SM, Akramuzzaman SM, Behrens RH, Haider R, Mahalanabis D. Randomised controlled trial of zinc supplementation in malnourished Bangladeshi children with acute diarrhoea. *Arch Dis Childhood* 1997;77:196-200.
51. Castillo-Duran C, Vial P, Uauy R. Trace mineral balance during acute diarrhoea in infants. *J Pediatr* 1988;113:452-7.
52. Khatri PC, Gupta BD, Miglani N, Jain A. Serum Zinc in Pulmonary Infections. *Ind Pediatr* 1981;18:120-122.
53. Denny FW, Loda FA. Acute respiratory infections are the leading cause of death in children in developing countries. *Am J Trop Med Hyg* 1986; 35: 1-2.
54. Lewski J. Mortality from acute respiratory infections in children under 5 years of age: global estimates. *World Health Stat Q* 1986; 39: 138-44.
55. National Research Council. *Recommended dietary allowances*. 10th ed. Washington, DC: National Academy Press, 1989.
56. Chandra RK,. Excessive intake of zinc impairs immune responses. *JAMA* 1984;252:1443-46.
57. Chandra RK. Trace elements and immune response. *Clin Nutr* 1987;3:118-125.
58. Schlesinger L, Arevalo M, Arredondo S, Lönnnerdal B, Stekel A. Zinc supplementation impairs monocyte function. *Acta Paediatr* 1993;82:734-8.
59. Cousins RJ, Hempe JM. Zinc. In: Brown ML, ed. *Present knowledge in nutrition*, Ed 6. Washington, DC: International Life Sciences Institute, Nutrition Foundation, 1990:251-60.

60. Sokol RJ. Wilson's Disease and Indian Childhood Cirrhosis. In: Suchy FJ, ed. Liver disease in children. St. Louis: Mosby, 1994:747-72.
61. Festa MD, Anderson HL, Dowdy RP, Ellersieck MR. Effect of zinc intake on copper excretion and retention in men. *Am J Clin Nutr* 1985;41:258-92.
62. Fosmire GJ. Zinc toxicity. *Am J Clin Nutr* 1990;51:225-7.
63. Cordano A, Baertl JM, Graham GG. Copper deficiency in infancy. *Pediatrics* 1964;34:324-36.
64. Castillo-Duran C, Fisberg M, Valenzuela A, Egaña JI, Uauy R. Controlled trial of copper supplementation during the recovery from marasmus. *Am J Clin Nutr* 1983;37:898-903.
65. Castillo-Duran C, Uauy R. Copper deficiency impairs growth of infants recovering from malnutrition. *Am J Clin Nutr* 1988;47:710-4.
66. O'Dell BL. Roles for iron and copper in connective tissue biosynthesis. *Philosophical Trans Royal Soc London B* 1981;294:91-104.
67. Danks DM. Copper deficiency in humans. *Ann Rev Nutr* 1988;8:235-57.
68. Eggleston PA. Asthma. taken from *Principles and Practices of Pediatrics*. Oski F. JB Lippincott Company. 1990. 201-207.
69. Falade AG, Tschappeler H, Greenwood BM, Mulholland EK. Use of simple clinical signs to predict pneumonia in young Gambian children: the influence of malnutrition. *Bulletin of World Health Organization* 1995;73:299-304.
70. WHO. Management of the child with a serious infection or severe malnutrition: Guidelines for care at the first referral level in developing countries. Draft 19: October 1997.
71. Lopez-Alarcon M, Villalpando S, Fajardo A. Breast-feeding lowers the frequency and duration of acute respiratory infection and diarrhea in infants less than six months of age. *J Nutr*. 1997;127:436-443.
72. Baqui AH, Paljor N, Silimperi DR. The prevention of diarrhoea in Dhaka urban slums. Urban Health Extension Project: Urban FP/MCH working paper No. 4; May 1993:9-10.
73. *American Journal of Clinical Nutrition*, in press.
74. Pelletier DL, Frongillo EA, Schroeder DG, Habicht JP. The effects of malnutrition on child mortality in developing countries. *Bull World Health Organ* 1995;73:443-8.
75. Baqui AH, Black RE, El Arifeen S, Hill K, Mitra SN, Sabir AA. Causes of Childhood Deaths in Bangladesh: Results of a nation-wide verbal autopsy study. *WHO Bull* (in press).

76. Grizzle JE. A note on stratifying versus complete random assignment in clinical trial. *Controlled Clin Trials*. 1982;3:365-368.
77. Smith PG, Morrow RH. "Census and mapping"; taken from Methods for Field Trials of Interventions Against Tropical Diseases: A Toolbox. UNDP/World Bank/WHO Special Programme for Research and Training in Tropical Diseases. Oxford University Press. 1991;95-127.
78. Bates CJ; Evans PH; Dardenne M; Prentice A; Lunn PG; Northrop-Clewes CA; Hoare S; Cole TJ; Horan SJ; Longman SC; et al. A trial of zinc supplementation in young rural Gambian children. *Br J Nutr* 1993 Jan;69(1):243-55.
79. Zaman K, Baqui AH, Yunus M, Sack RB, Bateman OM, Chowdhury HR, Black RE. Acute respiratory infections in children: a community-based longitudinal study in rural Bangladesh. *J Trop Ped* 1997;43:133-137.
80. Price NO, Bunce GE, Engel RW. Copper, Manganese, and Zinc Balance in Preadolescent Girls. *Am J Clin Nutr* 1970;23:258-260.
81. Botash AS, Nasca J, Dubowy R, Weinberger HL, Oliphant M. Zinc-induced copper deficiency in an infant. *AJDC* 1992;146:709-711.
82. Gregor JL, Zaikis SC, Abernathy RP, Bennet OA, Huffman J. Zinc, nitrogen, copper, iron, and manganese balance in adolescent females fed two levels of zinc. *J Nutr* 1978;108:1449-1456.
83. Sandstead HH. Copper bioavailability and requirements. *Am J Clin Nutr* 1982;35:809-814.
84. Taper LJ, Hinnens ML, Ritchey SJ. Effects of zinc intake on copper balance in adult females. *Am J Clin Nutr* 1980;33:1077-1082.

Dissemination and Use of Findings

Describe explicitly the plans for disseminating the accomplished results. Describe what type of publication is anticipated: working papers, internal (institutional) publication, international publications, international conferences and agencies, workshops etc. Mention if the project is linked to the Government of Bangladesh through a training programme.

~~The results of this research will be organised into standard scientific journal format and submitted to peer reviewed journals for dissemination to the relevant academic and scientific communities.~~

Collaborative Arrangements

Describe briefly if this study involves any scientific, administrative, fiscal, or programmatic arrangements with other national or international organizations or individuals. Indicate the nature and extent of

collaboration and include a letter of agreement between the applicant or his/her organization and the collaborating organization. **(DO NOT EXCEED ONE PAGE)**

N/A

Biography of the Investigators PLEASE SEE ATTACHED CV's

Voluntary Consent Form PLEASE SEE APPENDIX

Detailed Budget for New Proposal

Project Title: Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Less Than 2 Years of Age

Name of PI: W. Abdullah Brooks, MD, MPH

Budget Justifications

Please provide one page statement justifying the budgeted amount for each major item. Justify use of man power, major equipment, and laboratory services.

Budget Justification

1. **ASG Faruqe.** Dr. Faruqe has considerable experience in field epidemiology and in ALRI and diarrhoeal disease. He heads the hospital surveillance system. He adds experience and a necessary familiarity with the culture, in addition to the time he will provide, which will be equivalent to one half day per five-day workweek.
2. **MA Wahed.** Mr. Wahed is a medical biochemist with over 20 years of experience. He will oversee and personally be involved with the processing of our serum zinc and copper samples, as well as ensuring the quality of adherence to the laboratory protocol. Assuming that he devotes 3 hours/day of a five day work week to the processing of samples over a two month period, he will work $3/8 \text{ hours} * 5 \text{ days} / 7 \text{ days} * 21.5 \text{ days/month} * 2 \text{ months} / 258 \text{ days per year} * 100 = 5\% \text{ of the work year}$ during both years.
3. **Research Investigator.** Since the PI will contribute no more than 50% time and the Co-PI 10%, we need a full-time project supervisor, qualified to oversee the entire project when no one else is available. This person must be experienced in the conduct of field studies, standardisation and quality assurance procedures, data collection methodologies, be able to conduct and supervise the clinical examination, and participate in the routine 10% sub-sample re-visits. Even when the PI is present, someone who understands both the aims and methods of the study, and who speaks the language, will be needed when there are difficulties. Since this is a large study, and the first field for the PI in Bangladesh, this investigator should be experienced and senior. At present there is one candidate with several years of field experience who has worked with other Centre personnel and has assisted in the background phase of this study.
4. **Research Medical Officer.** A full-time physician in the field is essential to this project, as the determination of disease incidence is clinical. This individual must be accessible at

the field office to accompany health workers to homes of children suspected to be ill, make an accurate diagnosis of the nature of the illness, assess the degree of severity and decide between outpatient management versus referral to hospital. The medical officer will participate in blood drawing. The medical officer will also spend at least 20% of their time in the field training the health workers to appropriately assess respiratory rate and dehydration, and will conduct periodic monitoring for quality assurance. Presently there are two candidates with field trial experience for the position.

5. **Nurse.** One GS5-level nurse will be needed to help train and supervise the field research assistants to count respirations and assess dehydration. The nurse will also conduct home visits. Additionally, the nurse will draw blood for serum zinc and copper both at the time of enrolment and after 300 days of supplementation and perform anthropometric measurements.
6. **Field Research Assistants.** We will need 16 GS3-level field research assistants working 100% time to administer surveillance questionnaires and conduct exams (count respirations, assess the child for danger signs). There will be about 1552 infants visited once weekly. Assuming a rate of 4 interviews per hour, an actual work day of 5 hours for interviews and a five day work week, each worker will interview 20 infants per day. We need to provide salary support for 17 interviewers over a 17 month period to complete the work, according to the formula:
7. $1552 \text{ Interviews} / 7 \text{ days} * 1 \text{ day} / 20 \text{ interviews} * 1 \text{ worker} / 5 \text{ days/week} * 7 \text{ days} / 1 \text{ week} = 16 \text{ workers.}$
8. We will need them for 16 months accordingly: Training 1 month; Mapping 1 month; Recruitment 1 month = 3 months + 12 month supplementation + mop up 1 month = 16 months.
9. **Field Research Officer.** The study will require a research officer to oversee the work schedule of the research assistants, monitor the schedule for the monthly anthropometry, motivate the caretakers of the infants to comply with the provisions of the study (e.g., blood drawing, taking the supplements regularly, informing the staff of illness), ensure the coding of the questionnaire data, and the timely transport of data and materials (blood samples, questionnaires) to the Centre.
10. **Secretarial.** This is a requirement of the Centre.
11. **Data Entry Technician.** A full-time data entry technician is required. In order to ensure quality assurance and quality control, data must be checked, cleaned and entered from the beginning of the study. There will be 1552 intake forms, followed by 1552 surveillance forms per week. Assuming a rate of 2 minutes per short form, data entry by a trained technician takes 52 hours per week for 1552 forms. The technician will need to be assisted at least one day a week to keep up with the workload.
12. **Programmer.** In the second year, a statistical programmer will be required to assist with the running of our statistical analyses. This person will need to be familiar with SPSS and SAS software, and be able to make judgements about the appropriateness of statistical tests for a given type of data or question. We estimate that it will take about 40 hours to transform all of the data into appropriate format, followed by the equivalent of 35 hours per week for six to seven weeks for planning and conducting our analysis and reviewing our results.
13. **Mapping Assistants.** We will deploy 10 teams consisting of one mapping assistant and one health worker. They will perform mapping and listing among 20,000 households in the study site. At a rate of 100 households per day, working five days per week, it will take 10 teams 4 weeks to complete the task.
14. **Travel. International.** The PI will need to travel to the US the first year to meet with the international co-investigators and obtain consults from resident epidemiologists and

- biostatisticians. The second year he will need to travel to the US to present the findings of the study at Johns Hopkins (presentations are already being planned with different departments at the Schools of Public Health and Medicine). *Domestic*. We estimate a cost of \$3.00 per day per team of two for food and transportation, calculated against a 7-day workweek. There are 12 months of support in year 1 and 5 months in year 2.
15. **Drugs.** The majority of the zinc must be prepared during the first year. This represents the estimated annual cost of preparation and packaging per dose per patient based on historical experience at the Centre for both years.
 16. **Non stock drugs.** Year 1: ALRI antibiotics. Assuming an infection rate of 0.3 episodes/child/year = $0.3 * 1552 \text{ infants} * \$5.00 \text{ Amoxycillin/infant} * 9/12 \text{ year} = \$1,164.00$ for Amox. Diarrhoea rate = 4 episodes/child/year = $4 * 1552 \text{ infants} * \$0.25 \text{ per ORS treatment (at } \$0.05/\text{packet for 5 days)} * 9/12 \text{ year} = \$1,164.00$ for ORS. Luer Lock 3 cc Syringes 100/box at \$10/box = \$200. Set of 2000 Abbott 23 gauge butterfly infusion needles 40/box at \$33/box = \$1650. Gloves 40 boxes of 100 at \$4/box = \$160. Sharps containers, 8 X-large 5 gallon large funnel set of 8 at \$14/each = \$112. B-D 3 cc sterile red-top vacutainer tubes 20 boxes of 100 at \$18/box = \$360. Total for year 1 = \$5,719.00. Year 2: figures are the same, \$5,719.00.
 17. **Office supplies.** Assuming we rent a field office with 800 square feet of floor space at \$300/month for 12 months in year 1, we will spend \$3,600 for rent and utilities. Capital costs for furniture: (2 desks, 15 chairs, 1 small fridge, lamps, 2 ceiling fans, 4 4-drawer file cabinets) = \$1,500. Recurrent costs for paper, pencils, erasers, printer cartridges \$50/month = \$600. Total costs = \$5,700. Year two is the same minus the capital costs. The figure is calculated for 5 months.
 18. **Serum copper.** The cost is the actual cost of the analysis to our lab, \$3.00 per sample X 500 samples.
 19. **Serum zinc.** The cost is the actual cost of the analysis to our lab, \$3.00 per sample X 500 samples.
 20. **Photocopying.** Intake questionnaires: $\$0.05/\text{page} * 3 \text{ pages/copy} * 1552 \text{ copies} = \233 . Consent forms: $\$0.05/\text{page} * 2 \text{ pages/copy} * 1552 \text{ copies} = \155 . Surveillance forms: $\$0.05/\text{page} * 1 \text{ page/copy} * 1 \text{ copies/week} * 52 \text{ weeks} * 9/12\text{yr} = \$3,026/\text{yr} + 10\% = \$3,329$. Training manuals: $0.05 * 10 \text{ pages/copy} * 15 \text{ copies} = \7.50 . Total for first year is \$3,727.00. For year 2, minus the intake questionnaires and training manuals it is $0.05 * 1 \text{ page/copy} * 2 \text{ copies/week} * 52 \text{ weeks} * 6/12 \text{ year} = \$2,825$.
 21. **Hospitalisation.** The Centre requires this. We assume ALRI infection rate of 0.3 episodes/child/year * 1552 infants * 0.33 admission rate * \$6/day * 5 days * 9/12 year for year 1, and 4/12 year for year 2. For diarrhoea we assume a 10% rate for 1:4 episodes of diarrhoea at a rate of 4 episodes/child/year * \$6/day * 3 days * 9/12 year in year 1 and 4/12 in year 2. The figures are the respective sums.
 22. **Internet, fax, DHL, phone.** These are conservative estimates for the cost of these services/year of the study, taking into account overseas collaboration.
 23. **Library.** These reflect conservative estimates for the cost of retrieval and copying of articles, particularly in year 2 when we move towards publication.
 24. **Print and publication.** These are conservative estimates of the cost, which is present in year 2 only.
 25. **Miscellaneous.** This will cover sundry items, such as respiration counters from the GoB ARI Programme, glass dispensers and stainless steel spoons for sterile dispensing of zinc suspension in the field.
 26. **Indirect costs.** This is the limit of what the Swiss Development Corporation will pay to the Centre.

Other Support

Describe sources, amount, duration, and grant number of all other research funding currently granted to PI or under consideration. (DO NOT EXCEED ONE PAGE FOR EACH INVESTIGATOR)

NONE

Check List

After completing the protocol, please check that the following selected items have been included.

27. Face Sheet Included

☐

28. Approval of the Division Director on Face Sheet

☐

1) Certification and Signature of PI on Face Sheet, #9 and #10

1. Table on Contents

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5. Project Summary

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• Literature Cited

☐

7. Biography of Investigators

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1. Ethical Assurance

☐

1. Consent Forms

☐

rs, 1998 and 1999 Name	Pay Level	No. of Staff	% Effort	Mo/ly Rate	Man/ month	1998	1999
						Subtotal month	adj 5% Subtotal
3122 Dr. ASG Faruque	Co-PI	1	0.10	1,688	12.0	2,026	12 2,127
3122 MA Wahed	Co-Inv	1	0.05	1,400	12.0	840	12 882
3121 Research Investigator	NO-B	1	1.00	765	12.0	9,180	12 9,639
3121 Medical Officer	NO-A	1	1.00	632	12.0	7,584	6 3,982
3121 Nurse	GS5	1	1.00	347	12.0	4,164	6 2,186
3121 Field Research Assistant	GS3	16	1.00	224	12.0	43,008	6 22,579
3121 Data analysis - Programmer X 2 mo		1	1.00	832	0.5	416	1.5 1,310
3121 Field Research Officer	GS5	1	1.00	347	12.0	4,164	6 2,186
3121 Mappers		10	1.00	161	1.0	1,610	0 0
3121 Secretarial		1	0.20	510	12.0	1,224	12 1,285
3121 Data Entry Technician	GS3	1	1.00	224	12.0	2,688	8 1,882
3121 Health workers		4	1.00	60	12.0	2,880	6 1,512
Subtotal						79,784	49,570
Travel							
3600 International						3,500	2,500
3500 Domestic						8,450	3,560
Subtotal						11,950	6,060
Supplies and Materials							
3701 Drugs (Zinc)						1,100	0
3712 Non-stock drugs						3,815	4,000
3704 Office supplies						5,700	1,750
3 0300 Capital Exp/length boards (4)						200	0
Subtotal						10,815	5,750
Lab. test and other							
4808 Serum Cu						1,500	1,500
4809 Serum Zinc						1,500	1,500
4806 Photocopying						3,727	2,825
4813 Hospitalisation costs						4,397	1,954
4817 Internet						200	200
4821 Library Service Charge						100	150
Subtotal						11,424	8,129
Others							
3900 Fax, DHL, Phone						300	300
4000 Training						200	0
4300 Print & Publication						500	500
4500 Misc						250	250
1 0300 Capital Exp/computer/printer/UPS etc						7,000	0
2 0300 Capital Exp/4 wt scales/2 fridges etc						1,300	0
Subtotal						9,550	1,050
Total Direct						123,523	70,559
SDC Funds						50,000	0
Adjusted Direct						73,523	70,559
Total Indirect (25%)						18,381	17,640
Total Costs						91,903	88,199
Grand Total All Sources							230,102

S. Koi
Shamima Moln
31/10/98
Controller, Budget & Costing

ALRI/Diarrhoea Prevention with Zinc Trial in Infants < 2 Year Old Consent Form

1. **The activity involves an experiment.** We are asking you to allow your child to participate in an experiment. We want to know if zinc can prevent pneumonia and diarrhoea in infants. To find out, we will give some infants syrup with zinc and others syrup without zinc for 12 months. We will come to your home once a week to ask if your child has been sick since our last visit. Once a month, we will measure your child's height and weight at our office.
2. **The scope, aims and purpose of the research.** Pneumonia and diarrhoea cause much of the sickness and death in young infants. We have medicine to treat sick infants, but we want to prevent them from becoming sick. We know that zinc is important for health. Most children in Bangladesh do not get enough zinc in their food. We want to know if we can prevent children from getting pneumonia and diarrhoea by giving them zinc.
3. **Foreseeable harm and possible level of risk.** There have been many other studies in which children have been given zinc, and no children have been harmed by the doses we will use. A few children get a mild stomachache that lasts a short time. Most do not get any stomachache. We need a small sample of blood (5 ml) before your child starts taking the syrup, and again after 300 days. We will clean the skin and use a sterile needle to guard against infection.
4. **Likely or expected benefits.** Your child may have fewer episodes of pneumonia and diarrhoea by taking zinc. We will also check to see if zinc makes your child grow taller.
5. **Alternative procedures, treatments or preventive strategies.** Other than immunisations, including the ones that all infants should receive – including infants in this study - there are no treatments to prevent pneumonia and diarrhoea.
6. **Subjects will be notified of new information that might affect their participation.** We will be closely watching the progress of all the children in our study. We will notify you if we detect any problems, or if we get new information that gives us the answers we are looking for.
7. **Confidentiality will be maintained.** All the information you give us will be kept in a locked cabinet. Only our staff will have access to it. We will not give it to anyone.
8. **Explanation about medical treatment and care for research-related illness.** If your child gets sick with pneumonia or diarrhoea during this six-month period, we want you to notify us immediately so that we can give you medicine. Your infant should continue taking the syrup, unless we advise you to stop. If your child is very ill with pneumonia or diarrhoea during this twelve months, we will send you to ICDDR,B Hospital, where we will provide care.

9. **Participation is voluntary and refusal to participate will involve no penalty.** You may withdraw your child from this study at any time, and there will be no penalty. If you have any questions, please ask us.

_____ (Parent) _____ (Witness) __/__/__ (d/m/y)

Consent with Zinc Community Trial

9. **Participation is voluntary and refusal to participate will involve no penalty.** You may withdraw your child from this study at any time, and there will be no penalty. If you have any questions, please ask us.

_____ (Parent) _____ (Witness) __/__/__ (d/m/y)

6.2. Cu
6.3. CBC

Prevention of ALRI and Diarrhoea with Zinc Community Trial Field Questionnaire

1. Identifying Information

- 1.1. Date of enrolment
- 1.2. Date of visitation
- 1.3. Visitation number (circle 1 - 25)
- 1.4. Interviewer name
- 1.5. Interviewer code
- 1.6. Name of subject
- 1.7. Patient record number
- 1.8. Randomisation number
- 1.9. Age
- 1.10. DOB
- 1.11. Sex
- 1.12. Relation of caretaker (interviewee) to subject

2. Compliance with supplementation

- 2.1. Schedule of administration (QD vs QWk)
- 2.2. Caretaker report compliance (Y/N)
- 2.3. Medicine bottle present?
- 2.4. Quantity administered 7 days ago (mL)
- 2.5. Quantity remaining today, difference (mL)
- 2.6. Quantity administered today (mL)

3. Surveillance data - Recall

- 3.1. Cough during past week (Y/N)
- 3.2. Duration (days)
- 3.3. Tachypnoea during past week (Y/N)
- 3.4. Duration (days)
- 3.5. Chest indrawing during past week (Y/N)
- 3.6. Duration (days)
- 3.7. Feeding difficulties (Y/N)
- 3.8. Duration (days)
- 3.9. Convulsions (Y/N)
- 3.10. Duration (days)
- 3.11. Fever during past week (Y/N)
- 3.12. Duration (days)
- 3.13. Diarrhoea during past week (Y/N)
- 3.14. Duration (days)
- 3.15. Seen by doctor past week (Y/N)
- 3.16. Reason (categories)
- 3.17. Visit hospital past week (Y/N)
- 3.18. Reason (categories)

4. Surveillance data - Observed

- 4.1. Cough present (Y/N)
- 4.2. Duration (days)
- 4.3. Tachypnoea present (Y/N)
- 4.4. Duration (days)
- 4.5. Rate (R/min)
- 4.6. Chest indrawing (Y/N)
- 4.7. Duration (days)
- 4.8. Fever present (Y/N)
- 4.9. Value (° C)
- 4.10. Diarrhoea present (Y/N)
- 4.11. Duration (days)
- 4.12. Quality (watery/bloody-mucoid)
- 4.13. Frequency
- 4.14. Dehydration present (Y/N)
- 4.15. Some/Severe
5. Treatment (Y/N)
 - 5.1. Amox
 - 5.2. Other Abx
 - 5.3. ORS (No. of packets)
6. Referred to hospital (Y/N)
7. Anthropometric data (circle visits 4,8,12,16,20,24)
 - 7.1. Height (0.1cm)
 - 7.2. Weight (1gm)
 - 7.3. HAZ
 - 7.4. WAZ
 - 7.5. WHZ
8. Blood work [visit 34+] (Y/N)

	Age	Size	Power	Daily Dose mg	Time (weeks)	Signs	HypoCu	Haem	Immune	Faecal Cu
Fosmire		1		2000+		X				
Adult	11			100-300	6			X	X	
Adult				150	55		X			
Adult	1			110-165	42		X			
Adult				25-100	12					
Festa et al	Adult	9	1.00	1.8 to 21	9					X
Bates et al	6-28 mo	110		20	65					
Leuk			0.15							
Haem			0.13							
SOD			0.30							
Overall			0.19							
Inherent for 30% effect size			0.87							
Schlesinger	7 mo	39		2 mg/kg	15			X	X	
Leuk			0.39							
Ferritin			0.03							
Mono phag			0.41							
Mono fung			0.37							
Impetigo			0.60							
Overall power			0.36							
Inherent for 30% effect size			0.16							
Chandra et al	Adult	11		300	6				X	
Chemotactic			0.83							
Ingestion			0.98							
Bactericidal			0.10							
Leukocytes			0.07							
Inherent for 30% effect size										
Bactericidal			0.51							
Leukocyte count			0.60							
Botash et al	13 mo	1		120	6	X	X	X	X	
				180	1					

Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Below 2 Years of Age

Responses to Reviewers

Reviewer 1 (Mahalanabis)

1. The weekly dose has been changed to 70 mg QWk.
2. The intercurrent episodes of ALRI have been changed to 5 days (effectively two consecutive visits). Diarrhoeal episodes remain unchanged at 3 symptom-free days.
3. Double-blinding has been addressed in the design section, Specific Aim 3.
4. Surveillance is twice weekly.
5. Patients will be retained in the study, irrespective of compliance, so as not to bias the study.

Reviewer 2 (Ruel)

1. Design: the selection of subjects is changed and is now community-based.
2. The age-range for selection is 2 – 18 months.
3. The section on rationale has been modified and has been made included in the Design section and itemised by Specific-Aim (the format recommended by NIH reviewers, see
4. The statement "easy to administer" has been dropped. The adjective "inexpensive" is retained, however, as the reviewer's opinion is his own and does not reflect expert consensus. As routine administration of zinc is *not* yet policy, operational and distribution cost estimates would have to be based on clinical and field studies, which reflect a controlled (artificial) situation and *are* expensive. As the reviewer points out, zinc itself is inexpensive and distribution systems for mass consumption have not been developed.
5. The reviewer's comments are noted. In fact that study is cited as an example of one which did look at the age group of interest, but did not find an effect (Efficacy of Zinc as a Preventive Agent, paragraph 3).
6. Comments are included about the rationale for using zinc once weekly in both the Background (Efficacy of Zinc as a Preventive Agent, paragraph 6) and Design (Item 2 "Specific Aim 3" paragraph 1) sections. We could find no published studies done on a once weekly regimen, hence our proposal. We believe this to be an important piece of missing data in the growing literature of zinc supplementation as a preventive agent in children.
7. The layout of the proposal has been changed. The description of the study is in the Design section, which precedes Methods, where definitions and methodologies are given.
8. A discussion of the type of supplement, blinding etc is given in Design, Items 1 and 2. The actual amount (volume per dose) is under active discussion with the pharmaceutical company, but will be either 5 or 10 ml per dose.
9. The rationale for choosing partially breast-fed children was an attempt to standardise the diet, as most children in the age group above six months would be partially breast-fed. However, this has been dropped from the design. The type of diet will be one of the variables analysed, but is not a

- . specific aim of this study, as data on the protective effect of breastfeeding exist, thus diet did not factor into the sample size calculation.
- 10. The effect size was modified to fit the mean effect for ALRI based on meta-analyses (personal communication Robert E. Black, data not yet published). Sample size is driven by ALRI effect between treatment groups. Diarrhoea effect is 20%, which is more conservative than published effect sizes. Power is set at 90% for both, in order to detect treatment group differences.
- 11. The data analysis section has been changed and, as the reviewer appropriately observes, should be itemised by specific aim, which it is and is found in the Design section.

Reviewer 3 (Penny)

- 1. Hypothesis: The references to cellular immunity have been dropped and the wording modified to reflect measurement of clinical outcomes.
- 2. Background: A) The under-two year old population comprises the bulk of ALRI morbidity, and is worth studying. In order to have sufficient power to measure the effect of age on ALRI incidence within the same statistical parameters, we would have to recruit 3,386 infants. This would increase our cost from \$263K to about \$349K. Moreover, we would have to enlist an additional community in order to achieve the sample size. We believe that the cost would outweigh the benefits. However, we will stratify our analysis to examine the effect of age on diarrhoea incidence, which is an identified specific aim. B) The statement in "treatments" does not read "prevention is the only way to reduce morbidity", but "Prevention alone reduces the *incidence* of morbidity..." which is technically correct and reflects the PI's speciality training in preventive medicine. Perhaps the reviewer was not differentiating primary from secondary from tertiary prevention, which involve progressively increasing intervention by health providers. Indeed, primary prevention specifically reduces incidence, whereas the latter prevention methods involve effective case management and these primarily reduce disease prevalence and possibly mortality. (See Last J. Scope and Methods of Prevention in *Public Health and Preventive Medicine*. Appleton-Lange. 1996:3-10.) C) The wording of the proposal has been modified to emphasise that the intervention is correcting zinc deficiency and not that zinc is therapeutic, per se.
- 3. Zinc dosing. This section has been modified and does include a discussion of the benefits of cost and compliance for weekly dosing. The reference to toxicity states that it is "unlikely" in the doses being used, and all other references to toxicity have been dropped. The only toxicity issue remaining in the protocol has to do with the effect on copper status, and here haematological parameters i.e., serum Cu and leukopaenia, will be measured and correlated with clinical status.
- 4. Risks/benefits. A) We have made no claim that zinc reduces reactive airways disease (RAD). The benefits section reads, that supplements "may reduce the proportion of reactive airways disease that is *triggered by respiratory infection...*" which is a reasonable expectation. Studies have shown that a significant proportion of reactive airways disease is triggered by ALRI, particularly by respiratory syncytial virus (RSV) in young infants. (See Jeng MJ; Lemen RJ. Respiratory syncytial virus bronchiolitis. Am

- . Fam Physician 1997 Mar;55(4):1139-46.) If the incidence of ALRI is reduced, it stands to reason that the proportion of ALRI-mediated RAD exacerbations should also be reduced (see NIH Asthma Expert Panel Report 2, 1997). B) The reference to expense has been dropped.
5. Design. The design section has been significantly re-worked. The study is double blind.
6. Definitions. The field workers will visit twice weekly. Our definition of episode relies ultimately on clinical exam, not caretaker report of respiratory rate or cough, which has come under criticism for surveillance and reporting bias in other studies. Reports of illness will be included in the analysis of days with illness, as discussed in Methods. We believe the problems identified by the reviewer are correct, but are mitigated by twice weekly visits and corroboration of observed signs of respiratory distress with chest auscultation by a trained medical officer.
7. Anthropometry. Supine length is being measured, and the wording has been changed to emphasise this.
8. Laboratory. This section has been modified and indicates what precautions are being taken to avoid zinc contamination.
9. Randomisation. The study is double blind, and this is discussed in both Design and Methods. The individual bottles will have the child's code, not 'A', 'B', or 'C', which are instructions for the pharmaceutical company. The study does not rely on weekly recall. Visits are twice weekly but detection of disease (determination of incidence) is dependent on physical exam.
10. Compliance. The dose is 5 ml. Compliance will be measured twice weekly and on random spot checks (usually by the FRO and the investigators). The surveillance should be intense enough to discourage defaulting by caretakers who make a good-faith effort to comply. The only other option would be to have the field workers administer the syrup, which would be impractical for daily dosing in a study of this size. Indeed, it will provide useful insights if study subjects are non-compliant in significant proportion. We will not be able to assess the relative compliance between weekly vs daily doses, since the weekly dose will be masked as a daily dose. Children will not be withdrawn.
11. Treatment. Diagnosis of ALRI will neither depend on, nor include any references to, temperature since the presence or absence of temperature will not affect intention to treat. Numerous studies have shown fever to have too poor or inconsistent a correlation to warrant its use as a marker for ALRI, whereas chest indrawing, cyanosis, feeding, and abnormal auscultation findings consistently add to the predictive value of true ALRI when paired with tachypnoea (See Margolis et al.)
12. Inclusion criteria. The references to feeding status have been dropped.
13. Exclusion criteria. "Known" reactive airways disease has been defined. Rash was to be excluded to rule out measles, which increases the risk of subsequent infection. It has been dropped for concern over recall bias. The study site has not been used for other zinc trials. This was verified at a recent town meeting with community leaders, therefore we do not expect site contamination. An attempt will be made to limit the participants to one per household. If that is not possible, we will label the bottle for the mother to avoid confusion (e.g., a piece of tape with a circle vs an "X" to

- . distinguish two individuals). The actual dosing regimen will be same, irrespective of the true dose.
- 14. Quality assurance. Sample repeat visits are a valid method for assessing quality and variance of technique (see Smith P, Morrow R, Section 9.4 pp38), at least if the second party is blind to the findings of first. The reliability depends to an extent on the type and format of questions on the questionnaire. Ours will be categorical closed-type questions, which tend to show less variance than open questions. Additionally, we will conduct periodic reviews of interviewing technique, respiration and dehydration assessment among the staff.
- 15. Sample size. The effect size is 30% for both diseases, which is the mean effect noted from meta-analysis (Black, non-published data).
- 16. Data analysis. By "intention to treat", we presume this refers to compliance, which has been addressed above for both zinc supplementation and disease management.
- 17. Budget. The co-PI is presumed to work one-half day per five day workweek, or 10%.
- 18. The educational level of parents will be listed separately.
- 19. The finalised form of the questionnaire has not been completed.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH

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CONFIDENTIAL

February 18, 1998

Dr. Dilip Mahalanabis, — ICDDR,B
Calcutta

Dear Dr. Mahalanabis,

Scientists at the International Centre for Diarrhoeal Disease Research, Bangladesh conduct many specific research projects relevant to the overall aim and objectives of this Centre. Project proposals for research are developed by the Centre's scientists and sometimes in collaboration with scientists from other National and International Institutions.

As part of our review of such proposals we often seek the opinion of referees, known to be experts in their field, prior to consideration and decision on the proposals by the Research Review and Ethical Review Committees of ICDDR,B.

We would therefore be most grateful if you could kindly let us have your views on the enclosed proposal. For this proposal you may care to use the enclosed evaluation form or, if you prefer, a more general report will be satisfactory.

As the proposal is scheduled for consideration at the forthcoming meetings of the Research Review and Ethical Review Committees, we would be grateful if you could send us your comments, under confidential cover, to reach us as soon as possible.

Because the contents of the proposal must remain confidential, I would be grateful if, after review, it be destroyed, or returned to us along with your comments.

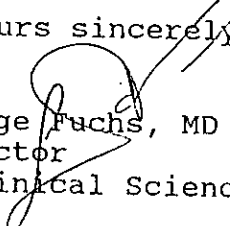
U

The ICDDR,B has a system of paying an honorarium of approximately US\$ 50 or equivalent for reviewing a proposal. May I request you to send your comments by the fastest available regular mail service.

We thank you for your cooperation.

Encl: a/a

Yours sincerely,


George Fuchs, MD
Director
Clinical Sciences Div.

Title: Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children below 2 years of age

Summary of Referee's Opinions: Please see the following table to evaluate the various aspects of the proposal by checking the appropriate boxes. Your detailed comments are sought on a separate, attached page.

Rank Score

	High	Medium	Low
Quality of Project	✓		
Adequacy of Project Design		✓	
Suitability of Methodology	✓		
Feasibility within time period	✓		
Appropriateness of budget	✓		
Potential value of field of knowledge	✓		

CONCLUSIONS

I support the application:

- a) without qualification ☐
- b) with qualification
 - on technical grounds ☒
 - on level of financial support ☐

I do not support the application ☐

Name of Referee:

Signature: *David Lewis*
Date: 19.12.1988

Position: *Director*

Institution: *Society for Applied Studies*

Detailed Comments

Please briefly provide your opinions of this proposal, giving special attention to the originality and feasibility of the project, its potential for providing new knowledge and the justification of financial support sought; include suggestions for modifications (scientific or financial) where you feel they are justified.

(Use additional pages if necessary)

Title:

PI:

Reviewer:

~~This research proposal is~~

~~In my opinion,~~

I give a very high priority to the topic of research in this proposal. The investigators are competent to do it and the institutional infrastructure is excellent. I have a few comments that follow.

Comments:

1. The weekly dose of 10mg appears rather low. What is the justification for it?
2. For ALAI, is ~~there~~ three + symptom free days enough to differentiate episodes?

3. How the double-blind status will be maintained when one group is getting a weekly dose? This should be described.
4. With once a week surveillance, how would they evaluate the respiratory rate and fever for attacks occurring between visits. The investigators should consider the feasibility of twice a week visit.
5. "Compliance Check" (dropping rule). The patients with inadequate compliance should remain in the study and be evaluated at the end of the study.

Title of proposal: Controlled Trial to Prevent Acute Lower Respiratory Tract Infection and Diarrhoea with Zinc Supplementation in Children Below 2 Years of Age

Autors: W. Abdullah Brooks, ASG Faruque, MA Wahed, George Fuchs

General Comments

The proposal is addressing an important question and the research has the potential to make an important contribution to the understanding of zinc deficiency and its consequences as well as its prevention in developing countries. The focus on children less than 2 years of age and on the two major causes of infant mortality in the developing world is extremely relevant.

My main concern with the proposal is the design, or more specifically the selection of subjects. The authors indicate that the study is a placebo controlled preventive trial in the community. It is not clear to me whether the authors are referring to a 'community trial'. In any case, the study

3. How the double-blind status will be maintained when one group is getting a weekly dose? This should be described.
4. With once a week surveillance, how would they evaluate the respiratory rate and fever for attacks occurring between visits. The investigators should consider the feasibility of twice a week visit.
5. "Compliance Check" (dropping rule). The patients with inadequate compliance should remain in the study and evaluated at the end of the study.

interest in this study. As indicated in the background section, 2/3 of all deaths due to pneumonia occur during infancy (the first year), so it would seem indicated to ensure that this age group is well represented.

Specific comments

Section on rationale (p. 5): This section is repeated further in the section on 'benefits of this project' (p. 9). The content is actually more appropriate for the section on benefits than for the section on rationale. The section on 'rationale' should include a discussion of why Bangladesh, why this age group, what makes us suspect that zinc deficiency is a problem in this population (e.g. diet, breast milk composition/quantity, complementary feeding, high rates of infection, etc.). In other words, why is this research justified in this particular context and what are the potential policy implications of the expected findings. Also, what is the rationale for the proposed treatment schedules (1/week vs 1/day).

Point 4 of the rationale: 'is inexpensive, easy to administer...': zinc supplementation, like iron supplementation, is not inexpensive from an operational and distributional point of view. The pills might be relatively cheap, but distribution systems that ensure high compliance have yet to be developed and are likely to be very costly.

Review of the literature (p. 7): the Ruel et al. study was done in children less than 2 years of age (age at recruitment was 6-9 months and the follow-up period was 7 months).

Some comments should be made about the rationale for using a once a week approach to treatment. Is there any literature, similar to that for iron, that would indicate that this could be equally effective as a daily dose and less likely to cause side effects?

Section on design (p. 10): Before giving the definitions and baseline data collection information, the description of the actual intervention should be given. One is interested in knowing what exactly will these children receive and how will the project be implemented before getting the other details.

The methods section should give more information on the type of supplement that will be provided: will a syrup-type supplement be used? What amount will be provided (total quantity of the supplement/day). Please provide information on preparation and also on how children who will receive the supplement once a week will be blinded to the intervention (i.e. will they be given a placebo 6 days a week and the supplement on one day to mimic the daily supplementation of the other group)? Please explain.

P. 13: why do children have to be partially breast-fed as opposed to exclusively? Couldn't they be exclusively breast-fed and just receive the supplement in addition?

Sample size: the magnitude of effects planned is large (45% for pneumonia incidence and 40% for

diarrhea). This means that the study will not have the power to detect differences of, say, 30 or 35%, which would still be biologically meaningful. Please provide information on sample sizes that would be needed for smaller effects to be detected (such as 30 or 35% for example).

The hypothesis of this study is that there will be no difference between the 2 treatments, but that both treatments will have a positive effect on morbidity reduction compared to the control group. When a study is interested in documenting the lack of difference between 2 treatments, sample size calculations are usually made based on a power of 90% as opposed to the usual 80%. This is because if you find no difference between the groups, you want to be sure that this is not because of lack of power, but rather, because there is really no difference between the groups (and in this case this is one of your main hypotheses).

The data analysis section (p. 14) should provide more details about which statistical analyses will be used to address each of the objectives of the study.

The budget for the study seems reasonable.

Date: Tue, 14 Apr 1998 14:07:20
From: mpenny@iin.sld.pe
To: gfuchs@icddr.org
Subject: protocol review

Dear George, I am sending this as a text file and as an annex, love,
Mary

INSTITUTO DE INVESTIGACION NUTRICIONAL
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Telfs. 3496023- 3496024 - 3496144 - 3496145 - 3496150
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Apartado 18-0191, Lima 18 - Peru

13th April 1998

Dear George,

How nice to hear from you. I receive "Glimpse" regularly so I keep in contact with ICDDR,B's activities. You seem to be very busy and productive.

Thank you for sending the proposal "Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children below 2 years of age" for my review. I enclose my comments. I will try to send this by email as the quickest and cheapest route, assuming I can find your email address.

Regards to all my friends in ICDDR,B.

Best wishes,

Mary Penny
Medical Investigator
Instituto de Investigacion Nutricional

"Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children below 2 years of age"

General comment

This is an interesting and important study and in general the protocol has been well thought out. The issues addressed are important to both local and international child health. The principal investigator has plenty of experience in clinical research and of course the centre has considerable experience so that I am confident that the study can be conducted as proposed. I support the project without qualification and my comments are intended to help ensure the success of the project..

If not already done, it would be worth discussing some of the aspects of the field work aspects with the investigators at ICDDR,B who have conducted similar community studies since it seems to me that details of the way that the supplement will be given and the collection of morbidity data are the weakest points in an otherwise

Christa
Reviewer-3
Send copy to PI (A. Brooks)
File original
Thanks,
87.

Hypotheses

The first and second hypotheses relate to cellular immunity needed to resist infections but in fact the study is not designed to test cellular immunity and the study outcomes relate to morbidity. The investigators may be able to speculate that any changes they observe are attributable to improved cell mediated immunity but they will have no evidence for this. The hypotheses should be change to reflect the question to be answered by the project. The third hypothesis states that the preventive effect will be "dose-dependent". I think that this gives the idea that two different daily doses of zinc are being used, it would be clearer here and throughout the project (including the "Objective") to clarify that this is weekly versus daily administration of zinc. The Specific aims are quite clear except that you could add incidence and prevalence as this is the intention.

Background

The subject has been well reviewed and the emphasis on the fact that infant mortality is highest in the under two year olds is pertinent. As the investigators note, the highest mortality is in those aged under one year and there is less evidence on the benefit of zinc supplementation in this group. The discussion almost convinces me that a study in the under 1 years would really be the most interesting but I presume that there would not be sufficient children to allow this. However, it would be useful to stratify by under/over 12 months at enrollment and include this subgroup analysis in the analysis plan and if possible allow for this in the sample size calculations.

I do not entirely agree with the paragraph on "treatments", the specific purpose of the mentioned treatments is not only to reduce mortality, and prevention is not the only way to reduce morbidity. There are lots of justifications for taking preventive measures as well as improving case management of illnesses, I don't think anyone would argue with the desirability of "prevention" regardless of the effectiveness of case management.

In order to put the potential benefit of the intervention into perspective I think it is important to mention that positive results of supplementation have been seen in zinc deficient populations, at the moment there is no evidence to suppose that zinc supplements will benefit children who are not zinc deficient, in other words we are talking about correcting a deficiency rather than a therapeutic or pharmacological effect. This does not weaken the potential importance of the intervention as zinc deficiency is likely to be common in nearly all populations with high infant morbidity and mortality. I think these issues should be mentioned in the proposal. If the investigator wants to argue for a pharmacological effect of zinc this could be discussed.

Zinc dosing

The investigators propose to compare once weekly with daily doses of ~~different they could have achieved this by giving a lower daily dose~~ in comparison with 10mg daily. There has been considerable discussion about the potential difficulty of having to give a daily zinc supplement and there is interest in weekly dosing since if this worked it might be easier to administer, possibly increase compliance and perhaps more importantly indicate that if, in practice, many daily doses were forgotten the intervention might still be efficacious. There are also cost considerations. Weekly dosing would reduce the cost of this potentially expensive intervention. I think these logistics issues are the real reason for testing weekly versus daily rather than the fear of toxicity and this should be discussed.

conscious. There are also cost considerations. Weekly dosing would reduce the cost of this potentially expensive intervention. I think these logistics issues are the real reason for testing weekly versus daily rather than the fear of toxicity and this should be discussed.

I do not agree that a larger preventive effect of the weekly supplementation would necessarily suggest that the daily doses were toxic.

Risks and benefits

I think that there is not enough evidence to suggest that the zinc supplements will reduce reactive airways disease, the promise of reductions in pneumonia are sufficient to make this an attractive intervention.

I don't think that this intervention could be regarded as "inexpensive", it will not be compared with therapeutic treatments but with public health preventive measures. The investigator might explain how this intervention will fit in with IMCI.

Research design and methods

The design would be greatly strengthened if the study was double blind, as I understand it the weekly dose group will be distinguishable, could you consider giving everyone daily doses with the weekly doses group 6 placebos to one zinc dose?

Definitions

No problem with diarrhea. The fieldworker will only visit once a week but the definition of ALRI requires raised RR and one other sign or symptom of which fever ≥ 37.9 , chest indrawing and cyanosis which presumably depend on the fieldworker observation. If you are not visiting daily how will you be able to measure duration of the episode and how will you define the end of the episode in order to differentiate episodes? If the mother says that the child had fast breathing, cough, fever 6 days ago but is normal on the fieldworker exam this episode would be missed. This is why many community studies end up defining episodes of ALRI as cough with raised respiratory rate (+/- other symptoms) at any time during the episode and do not attempt to define duration except in rather imprecise ways. Instead they use prevalence of symptoms which the mother can reliably report, i.e. cough. This aspect does need a bit more thought.

Anthropometry

At this age you will measure supine length rather than height.

Laboratory investigations

The precautions to be taken to avoid zinc contamination should be explained, especially as the blood samples are to be collected in the homes.

Randomization

It is not clear to me if it is really double-blind or whether the weekly and daily dosing schedules will be evident. What dosing regime will the placebo group follow? It seems that the supplements will be divided into "A", "B", and "C". This makes it very easy for the codes to be broken especially as the study will be doing laboratory analyses. The study would be much stronger if the medicine bottles could be individually relabelled with the code of the child so that the danger that fieldstaff and investigators can know which

It seems a pity to rely on weekly recall data when we know that after 48 hours the reliability of recall goes down. If I remember correctly investigators at ICDDR,B are among those who have demonstrated this. Could you not do twice weekly visits and only accept recall over the preceding week? If you visit once a week and the mother is not present then more than 7 days will pass before the next visit and recall for over this period really should not be considered reliable and you will then have gaps in your information about the child. If you schedule visits twice a week if the mother is absent for one visit you still have a chance of recuperating the information at the next visit.

Compliance

This is a weak aspect of the study and needs more discussion of practical details. How big is the daily dose (5ml?) how will the residual amount be measured, with what precision? How will you know whether the mother has given the dose daily or if she realizes she has forgotten a dose she may give a larger dose so that the residual amount will be the same? This is always a problem when dosing cannot be observed and does weaken the study since if you have negative results your only evidence that this is not due to non compliance will be the biochemical results. At the very least you could make additional spot checks between the healthworker visits in order to double check the residual in the bottle. These issues should be discussed more fully in the proposal. If a mother is not compliant but wants to remain in the study to "remind her of the study agreement" seems to be to be an invitation to make sure that she appears to be compliant next time by ensuring that the medicine has disappeared even though she may not necessarily have given it to the child. Children who are not compliant should not be withdrawn from the study as this may well introduce a bias, mothers of children who do not seem to be responding because they have more illness, for instance, may be less compliant and would end up being withdrawn. If the children are not followed up until the end you will not be able to do intention to treat analysis. These issues are especially important if there is a possibility that the codes may be inadvertently broken. You could set a priori definitions for compliance and apply these in your analysis.

Treatment for non-severe infection.

Will you only take temperature if the child has raised RR?

Inclusion criteria

Why only include partially breastfed children? How will this be defined? What effect will this inclusion criterion have on the number of children available and how will this effect the age distribution?

Exclusion criteria

In these very young children how will you define reactive airways disease? Why exclude children with rash? What sort of rash? How and who will make the individual decisions on rashes? Mention the justification for excluding exclusively breast or bottle fed babies. I imagine that children who have previously received supplements of zinc, including children who have already participated in the study, will be excluded. What will the policy be for children who live in the same household as a study participant, for instance siblings or cousins? . In these cases there is the risk of confusion between the medicines.

Quality assurance

I am not sure how useful repeat visits are to calculate and assess

quality assurance

I am not sure how useful repeat visits are to calculate and assess

inter-observer variation. Mothers may well change their report especially when asked to report over a long period and an exam conducted up to 24 hours after the first visit may be completely different without being inaccurate. Since RR are such an important study outcome it would be better to do some simultaneous visits with both supervisor/research assistant/investigator and health worker counting the RR at the same time. Regular standardization exercises against a "gold standard" are also helpful and the results can be reported.

Outcomes - see earlier discussion

Sample size

Considering that not all studies have found an effect on ALRI incidence, a 45% reduction may be optimistic in the sample size calculation. A 40% reduction in diarrhea also seems high but as usual these figures may be dictated more by the budget than other considerations.

Data analysis

Mention whether intention to treat or not, if not explain selection.

Budget

Under manpower the co-principal is said to contribute one hour per week, a imagine this is one day per week as in the budget calculation!

The budget seems reasonable, although it is difficult to know without inside knowledge on how many visits the field team can be expected to do per day and how many repeat visits they would have to make.

Appendices

Appendix 2

II D. Ask education level of mother and father separately

Appendix 3

III How will the form be used to determine duration of an episode. For instance a child who is reported to have had 10 days of diarrhea in the last week might have had these two days at the beginning of the week and be symptomfree for three days or may have started two days ago and so the episode might be going to last longer. Under G you mention diarrhea but the definition to be used is Three or more liquid stools so the question should be asked in terms of "how many stools did the child have each day" and "how many of these were loos or watery" ?

I hope that these comments are useful, good luck with the study!

APPENDIX

Clinical Assessment of Dehydration (Dhaka Method)

CONDITION*	(1=normal, 2=irritable/less active, 3=lethargic/comatose)
EYES	(1=normal, 2=sunken)
MUCOSA	(1=normal, 2=dry)
THIRST*	(1=normal, 2=thirsty, 3=unable to drink)
SKIN TURGOR*	(1=normal, 2=reduced)
PULSE*	(1=normal, 3=feeble/absent)

** Denotes key signs*

DEHYDRATION STATUS:

1 = no dehydration

2 = some dehydration (two signs coded 2 with at least one key sign*)

3 = severe dehydration (some dehydration, plus one key sign coded 3)

- * This method was recently validated by a group of international experts in oral rehydration therapy who were invited to ICDDR,B and is currently being used in a large WHO sponsored multi-centre trial for the evaluation of low osmolarity ORS. All the study investigators and staff have been trained in this method.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH

Mail : ICDDR,B, GPO Box 128, Dhaka-1000, Bangladesh

Phone: 871751-60, Telex : 675612 ICDD BJ

Fax : 880-2-883116, 886050, 871568, 871686, Cable : Cholera Dhaka

>Memorandum<

To : Dr. Abdullah Brooks

July 21, 1998

From : Prof. Farida Huq
Chairperson, ERC

Subject : **Your protocol # 98-011**

Thank you for your memo of July 19 containing your responses to the observations made by the Committee on your protocol # 98-011 entitled "Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children less than 2 years of age".

On the basis of the reviewer's satisfaction with your responses, I am pleased to **approve** the protocol.

Thank you.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH

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Memorandum

DATE: 19.07.1998

TO: Farida Huq, Chairperson ERC

FROM: W. Abdullah Brooks, MD, MPH

RE: Protocol 98-011 "Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children less than 2 years of age"

cc: George Fuchs, Director CSD

Per the Committee's request, the following steps regarding the above protocol have been taken.

1. We agree to the selection of an independent monitor for the first four months. At present Dr's Black and Santosham are looking for someone to fill this role presently. The monitor's report will be sent to the Committee at the end of the observation period.
2. The instructions to the Bangladesh pharmaceutical company(s), i.e. ACME or Opsonin, that will be contracted at the lowest bid to prepare the supplements and placebo have been attached to the protocol as an addendum. These specify the composition of the placebo.
3. The amount of blood to be drawn for zinc monitoring (5 ml) has been added to the consent form.
4. The face sheet 2(d) has been modified according to the request of the Committee i.e. marked "Yes".
5. The face sheet 3© has been modified according to the request of the Committee; i.e. marked "Yes".

On behalf of my co-investigators, I thank the Committee for the opportunity to further improve our protocol, and I pray these modifications are satisfactory and will expedite the protocol's approval. Thank you.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH

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Memorandum

To : Dr. Abdullah Brooks

June 25, 1998

From : Prof. Farida Huq
Chairperson, ERC

Farida Huq

Subject: **Approval of your protocol # 98-011 entitled "Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children under two years of age"**

The Committee met on the 24th of June 1998 and considered your above-mentioned protocol for review and discussion. After a thorough review and discussion, the Committee made the following observations to be addressed in your protocol:

- (1) An independent monitor should be appointed to observe the adverse effect, if any, during the first 4 months of administration of zinc and placebo to the subjects. The report of the monitor should be submitted to the Committee.
- (2) The composition of the placebo and the source of the drug should be indicated in the protocol.
- (3) The amount of blood to be drawn should be mentioned in the consent form.
- (4) Serial # 2(d) (Discomfort to subjects) of the face sheet of the protocol should be marked with "Yes".
- (5) Serial # 3© (Use of organs or body fluids) of the face sheet of the protocol should be marked with "Yes".

I would appreciate if you please modify your protocol incorporating the above observations in it and resubmit a copy of the same to the Chair of the Committee for further necessary action.

Thank you.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH
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Memorandum

Date : June 16, 1998

To : Dr. Abdullah Brooks

From : Chair, RRC

Subject : Approval of your protocol # 98-011

The Committee met on 15 June 1998 and considered your protocol # 98-011 entitled "Controlled trial to prevent Acute Lower Respiratory Tract Infection and Diarrhoea with zinc supplementation in children below 2 years of age." After a thorough review and discussion on your protocol, the Committee found the following observations, which should be addressed for further consideration:

- (1) All diarrhoeal episodes should be recorded and further be subdivided into watery diarrhoea and dysentery.
- (2) Instead of giving two separate bottles to the mothers to administer, 1 bottle (10 gm zinc or placebo) be given to them to administer and the health visitors should give a weekly dose (70 mg zinc or placebo) during their visit.

You are requested to please address the above observations into your protocol and resubmit a copy of the same to the Chair of the Committee for further necessary action.

Thank you.



INTERNATIONAL CENTRE FOR DIARRHOEAL DISEASE RESEARCH, BANGLADESH
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Memorandum

DATE: 16.06.1998

TO: Prof VI Mathan, Chairman RRC, Director
LSD

FROM: W. Abdullah Brooks, MD, MPH *WAB*

RE: Protocol modifications for Community-
based ALRI/diarrhoea prevention trial
with zinc

cc: George Fuchs, MD, Director CSD *G.F.*

Per the request of the RRC to modify the protocol "Controlled trial to prevent acute lower respiratory tract infection and diarrhoea with zinc supplementation in children under two years of age", I am pleased to report the following:

1. The request to modify the dispensing of the 70 mg once-weekly dose of zinc has been done (testing specific aim 3, protocol page 14, paragraph 3). The health workers will now give one dose each week to each child, which will coincide with the 70 mg dose for Group B.
2. I was mistaken in my statement that we were surveilling acute watery diarrhoea only. In fact the protocol states that we are surveilling and treating "diarrhoea with severe dehydration or bloody mucoid stools" and the sample size was calculated accordingly (page 20, paragraph 1). Therefore, the protocol is already designed to record the incidence of all acute diarrhoea, watery or dysenteric. The questionnaire will allow us to distinguish which type of diarrhoea a child has, and these can be analysed together or separately.

On behalf of my co-investigators and myself, I thank you for the opportunity to make these improvements in the protocol and hope that these modifications will hasten its approval.

CURRICULUM VITAE

W. Abdullah Brooks, MD, MPH.

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Baltimore, MD, 21202
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(410) 283-5686 (Pager)

Education

1994-Present	Johns Hopkins School of Hygiene and Public Health, Baltimore, MD Preventive Medicine Residency
1994-1995	Johns Hopkins School of Hygiene and Public Health, Baltimore, MD Master of Public Health (MPH)
1991-1994	The New York Hospital/Cornell Medical Ctr. New York, NY Residency - Pediatrics
1985-1991	Stanford University School of Medicine, Stanford CA Medical Doctorate (MD)
1983-85	American River College Sacramento, CA Post-baccalaureate sciences
1976-1978	University of California, Los Angeles Bachelor of Arts (BA) International Relations
1974-1976	University of California, San Diego Political Science

Research Activities

July-October 1995	International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B) Dhaka, Bangladesh Multicentre Study on Lower Osmolar oral rehydration solution (ORS). Clinical trial to assess the efficacy of lower osmolarity ORS relative to standard ORS (WHO formula) in adults with cholera and infants under two years of age with acute watery diarrhea.
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W. Abdullah Brooks, MD, MPH. CV97

- July-October 1995 ICDDR,B Dhaka, Bangladesh
Analysis of surveillance data on risk factors for Cryptosporidiosis among hospitalised and clinic patients at the Diarrhoea Hospital.
- July-October 1992 University Hospital, Salvador
Bahia, Brazil.
Field study on risk factors for invasive diarrhoeal disease among slum children in Salvador. Included collection of epidemiological data as well as isolation of pathogens and testing with DNA probes.
- Oct-December 1990 The Center for Disease Control and Prevention
Atlanta, GA
Epidemiology Intelligence Service Clerkship for Medical Students - Enteric Branch. Included outbreak investigation of E. coli 0157-H7 in Helena MT among children at an elementary school, and surveillance system analysis for Salmonella typhi carriers among state health departments in all 50 states and territories.
- July-Sept 1986 The National Institutes of Health (NIH)
Bethesda, MD
Division of Parasitology. Isolation of Leishmania major attachment proteins to the macrophage, as a possible means of prevention of parasitic infection.
- 1985-1986 Stanford University School of Medicine
Stanford, CA
Identification of heat-shock proteins in Fasciola hepatica morphogenesis, as a model for morphological regulation in Schistosoma species.

Professional

- 1996-1997 Chief Resident, Johns Hopkins Preventive Medicine Pgm
Baltimore, MD
- 1996 Consultant for PAHO, EPI/SVI
Georgetown, Guyana
Planned and implemented mass vaccination campaign against

W. Abdullah Brooks, MD, MPH. CV97

- 1995+ St. Joseph's Hospital Pediatric Express
Baltimore, MD
Emergency room physician for pediatric patients
- 1995+ Sinai Hospital Emergency Room
Baltimore, MD
Emergency room physician for pediatric patients
- 1995+ Baltimore County Health Department
Baltimore, MD
Physician for School-based clinics in Baltimore County School District
- 1994+ Kennedy Krieger Institute
Baltimore, MD
Pediatric physician for inpatients

Grants

- 1996 Small study on serological-based evaluation of silicate exposure in patients with scleroderma: NIH; Lawless D, Brooks W, Lamm S, Teed K. Not Funded.

Presentations

- April 1993 Department of Pediatrics Grand Rounds: Original research results on risk factors for invasive diarrhoeal disease among slum children in Salvador, Bahia, Brazil
The New York Hospital/Cornell Medical Ctr, NY, NY
- Oct 1990 Genetic evidence of the common origin and essential oneness of the human race: Association for Bahá'í Studies, Atlanta, GA

Awards and Honours

- Jul-Oct 1995 International Fellow, The International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B) Clinical Sciences Division
Dhaka, Bangladesh
- 1994-1995 Health and Child Survival Fellows Scholarship for Master of Public

W. Abdullah Brooks, MD, MPH. CV97

- | | |
|-----------|---|
| 1986 | Ciba-Geigy Summer Internship Award for medical student research
National Institutes of Health, Bethesda MD |
| 1974-1975 | Pacific Palisades Chamber of Commerce Scholarship for Academic
Achievement and Student Leadership |

Teaching Experiences

- | | |
|------|---|
| 1995 | Johns Hopkins University Intersession Course on Public Health
Career Options |
| 1993 | Methodist Community Hospital, Brooklyn NY
Chief Resident: Clinical training of pediatric residents as part of that
hospital's accreditation process |

Memberships and Activities

- | | |
|-------|---|
| 1994+ | American Public Health Association |
| 1993+ | American Academy of Pediatrics |
| 1995+ | Association of Preventive Medicine Residents |
| 1992+ | American Academy for the Advancement of Science |
| 1990+ | Bahá'í International Health Agency |
| 1977+ | Member of Bahá'í Faith, Bahá'í Community of the United States of
America |

Professional Licensure and Certification

- Board Certified, New York Board of Medical Examiners
- Board Certified, Maryland Board of Medical Examiners

W. Abdullah Brooks, MD, MPH CV97

Languages

French - Speaking, reading, writing proficiency

Portuguese - Speaking, reading, writing proficiency

Arabic - Reading, writing comprehension (with difficulty)

Swahili - Conversant (with difficulty)