Attachment 1.

Date June 22, 1994
July 19, 1994

(COVER SHEET) ETHICAL REVIEW COMMITTEE, ICDDR,B

Principal Investigator Dr. S. K. Roy

Trainee Investigator (if any) _____

Application No. 94-011

Supporting Agency (if Non-ICDDR,B) _____

Title of Study: Zinc balance and bioavailability from two different dietary regimes in children with acute and persistent diarrhoea syndrome in Bangladesh using stable isotope.

Project status:
( ) New Study
( ) Continuation with change
( ) No change (do not fill out rest of form)

Circle the appropriate answer to each of the following (If Not Applicable write NA).

Source of Population:

(a)	Ill subjects	(Yes) No
(b)	Non-ill subjects	Yes No
(c)	Minors or persons under guardianship	Yes No

Does the study involve:

(a)	Physical risks to the subjects	Yes (No)
(b)	Social Risks	Yes (No)
(c)	Psychological risks to subjects	Yes (No)
(d)	Discomfort to subjects	Yes (No)
(e)	Invasion of privacy	Yes (No)
(f)	Disclosure of information damaging to subject or others	Yes (No)

Does the study involve:

(a)	Use of records, (hospital, medical, death, birth or other)	(Yes) No
(b)	Use of fetal tissue or abortus	Yes (No)
(c)	Use of organs or body fluids	(Yes) No

Are subjects clearly informed about:

(a)	Nature and purposes of study	(Yes) No
(b)	Procedures to be followed including alternatives used	(Yes) No
(c)	Physical risks	(Yes) No
(d)	Sensitive questions	(Yes) No
(e)	Benefits to be derived	(Yes) No
(f)	Right to refuse to participate or to withdraw from study	(Yes) No
(g)	Confidential handling of data	(Yes) No
(h)	Compensation &/or treatment where there are risks or privacy is involved in any particular procedure	(Yes) No

5. Will signed consent form be required:
   (a) From subjects Yes (No)
   (b) From parent or guardian (if subjects are minors) (Yes) No
6. Will precautions be taken to protect anonymity of subjects (Yes) No
7. Check documents being submitted herewith to Committee:

___ Umbrella proposal - Initially submit an overview (all other requirements will be submitted with individual studies).
✓ Protocol (Required)
✓ Abstract Summary (Required)
✓ Statement given or read to subjects on nature of study, risks, types of questions to be asked, and right to refuse to participate or withdraw (Required)
___ Informed consent form for subjects
✓ Informed consent form for parent or guardian
✓ Procedure for maintaining confidentiality
___ Questionnaire or interview schedule *

* If the final instrument is not completed prior to review, the following information should be included in the abstract summary:
1. A description of the areas to be covered in the questionnaire or interview which could be considered either sensitive or which would constitute an invasion of privacy.
2. Examples of the type of specific questions to be asked in the sensitive areas.
3. An indication as to when the questionnaire will be presented to the Ctte. for review.

I agree to obtain approval of the Ethical Review Committee for any changes involving the rights and welfare of subjects before making such change.

S.K. Roy

Principal Investigator

Trainee

A-031975

SECTION 1: RESEARCH PROTOCOL

1. Title: **Zinc balance and bioavailability from two different dietary regimes for children with acute and persistent diarrhoea syndrome in Bangladesh using stable isotope..**

2.	Principal Investigator Co Principal Investigator	: :	Dr. S. K. Roy Prof. Andrew Tomkins
3.	Collaborative Investigators	:	Dr. Susan Fairweather Tait ARFC Laboratory, Norwich, UK
3.	Co-investigators	:	Dr. Dilip Mahalanabis Dr.S.M.Akramuzzaman Dr. Nahrina Dewan
4.	Starting date	:	As soon as possible.
5.	Completion date	:	1 year from the beginning of project
6.	Total direct cost: US$	:	US$ 110,725
7.	Source of fund:		
8.	Scientific programme	:	This protocol has been approved by the Clinical Sciences Division.

Associate Director
Clinical Sciences Division

Date: 20/6/94

9. Abstract summary

Children with acute and persistent diarrhoea aged between 3 and 24 months will be studied at ICDDR,B for micronutrient balance, in response to zinc supplementation. 15 patients in each diarrhoea group will be studied for metabolic balance of zinc, copper, iron and magnesium in a 72h balance study. Zinc absorption will be measured and endogenous zinc secretion in diarrhoeal stool will be quantitated using stable isotope ^{67}Zn infusion. Mucosal integrity will be estimated using lactulose/mannitol permeability test. Zinc supplementation will be given in doses of 20 mg per day for 2 weeks. Patients will be with two dietary regimens e.g. chicken meat based diet and rice based diets and will have repeat absorption study 4 weeks after clinical recovery as each patient will be its own control. The study will help to estimate net excretion of zinc and other essential trace metals during diarrhoea and will quantitate the exact balance of trace metals during diarrhoea and at recovery. The overall benefit of the study will be to ascertain the appropriate dose of zinc, copper or magnesium in malnourished children during and after diarrhoea.

10. Reviews

a. Ethical Review Committee: _____

b. Research Review Committee: _____

c. Director: _____

SECTION II – RESEARCH PLAN

A. INTRODUCTION

1. Objectives:

A. To estimate the bioavailability of zinc from two different diets with acute and persistent diarrhoea, using stable isotope ^{67}Zn and apparent absorption of iron; copper and magnesium by 72 h balance study.

B. Effect of zinc supplementation on diarrhoeal duration, fluid loss and mucosal permeability during diarrhoea.

2. Background:

Severe zinc deficiency in man is a well recognized disease characterized with diarrhoea and skin lesions known as acrodermatitis enteropathica (AE) (Moynahan 1974). Higher concentration of zinc loss has been detected in stool during Crohn's disease (Beeken et al 1976) and severe protracted diarrhoea (Rothbaum et al 1982). Until now only one study in Chilean children has shown by conventional balance technique that zinc loss in acute diarrhoea was 159 µg/kg/day (Castillo-Duran et al 1988). The patients remained in a negative zinc balance of 180 µg/kg/day. In chronic diarrhoea patients, stool zinc loss was recorded to be 300 µg/kg/day where 200 to 300 µg zinc/kg/day was required intervenously to maintain a normal serum alkaline phosphatase level and to resolve skin rash due to zinc deficiency (Rathbaum et al. 1982).

In Bangladesh, diarrhoeal morbidity is a significant burden to majority of the malnourished young children. These malnourished children have been identified as zinc deficient in Bangladesh (Khanum et al. 1988) and other countries like Jamaica (Golden et al. 1979), Thailand. A recent study in Bangladesh showed that zinc deficiency limits linear growth after diarrhoea (Behrens, Tomkins and Roy 1990) suggesting a hypothesis that diarrhoea causes a zinc deprivation state leading to a diarrhoea-malnutrition cycle. Until now there is no information on the absorption and secretion of zinc in children with acute and persistent diarrhoea specially with reliable methods such as stable isotope. Zinc absorption is more interfared in the malnourished children with various grades of mucosal integrity during and after diarrhoeal attacks.

Jackson et al (1984) have shown that thermal ionization mass spectrometry can be used to measure stable isotope which is entirely safe and relatively cheap. Stable isotope ^{67}Zn can be used to study endogenous zinc secretion in diarrhoeal stool. The technique involves the enrichment of the ex changeable body pool by infusion of isotopic ^{67}Zn and comparison of the enrichment in plasma over a period of time when the intake levels of zinc are known.

There are concern about interaction of one trace elements with the other in relation to competition in absorption and balance in the body. The effects of zinc on iron and copper in food and vice versa are particularly important. High dose of zinc supplementation has resulted in reduction of tissue calcium and fetal iron accretion in rats (Southon et al 1981). High calcium in diet (2g/d) had reduced zinc absorption whereas high phosphorus did not change the absorption of zinc (Spencer et al 1984). Die tary Fe/Zn ratio of 1:1 slightly inhibited zinc absorption while higher iron ratios 2:1 and 3:1 inhibited zinc uptake (Solomons et al 1981). Animals fed high zinc level have reduced copper absorption by retaining more copper in mucosal cells (Fisher et al 1981). As mentioned earlier, supplementation of copper during diarrhoea led to excess (159µg/kg/day) zinc loss in stool (Castillo-Duran et al 1990). To plan correct micronutrient supplementation, it is essential to know the extent of micronutrient interac tion in terms of absorption and balance.

Our previous study showed that zinc supplementation during diarrhoea had beneficial effects in terms of earlier recovery (Roy et al. 1990, 1991) but the magnitude of loss of zinc in stool and the level of absorption were not known. It was also not known whether absorption or balance of other micronutrients were altered by zinc supllementation at the dose 20 mg elemental zinc/child/day equivalent to twice the RDA of US Food and Nutrition Board (1980) for better nourished children. In view of higher amount of zinc loss in chronic diarrhoea due to mucosal injury and reduced absorption, the oral dose of zinc used in our previous study might be inadequate. It is not known whether high purging rate is also related with zinc loss in stool. It may be possible that excess of micronutrient may be lost similar to loss of macronutrients in patients with persistent diarrhoea and malnutrition (Roy et al. 1992).

The clinical benefit of zinc supplementation is well known in severe chronic diarrhoea syndrome such as acrodermatitis enteropathica (Moynahan 1974). But only recently , it has been found that zinc enhances clinical recovery, reduces diarrhoeal stool volume in undernourished children with acute or persistent diarrhoea (Roy 1991). The earlier recovery and reduced stool weight may be due to better repair of mucosal integrity (Roy et al 1992) or intraluminal reduction of pathogens (Fenwick et al 1990) or improved electro lyte transport with zinc supplementation (Roy et al 1986, Patrick et al 1980).

In our previous study it was noticed that improvement in mucosal integrity was significant in children with acute and persistent diarrhoea with zinc supplementation (Roy 1992). Intestinal permeability was measured with urinary excretion of lactulose and mannitol after an oral dose. It is not known whether mucosal damage is an important determinant of micronutrient absorption or endogenous secretion during diarrhoea. To answer this question, a quantitative study on absorbtion of zinc and other micronutrients in relation to mucosal damage is indicated.

3. **Hypothesis:**

With the above state of knowledge we hypothesize that

1. Zinc absorption during acute or persistent diarrhoea can maintain positive zinc balance in children who are malnourished.

2. Balance of other micronutrients such as copper, iron, magnesium are not offset in malnourished patient during diarrhoea.

3. Zinc supplementation during diarrhoea does not increase net loss of other micronutrients.

4. **Rationale:**

Zinc deficiency is associated with growth retardation, severe diarrhoea and increased morbidity. Supplementation of zinc has shown significant benefit to the malnourished children in diarhoeal severity and subsequent morbidity and growth as well as reduced risk of mortality. These findings have not been substantiated with estimation of micronutrient absorption and balance during or in absence of diarrhoea. The study is expected to find benefit or adverse effect of zinc on micronutrient balance during supplementation in diarrhoeal patients with malnutrition. The above information would significantly increase understanding on micronutrient balance and will help formulate appropriate dose of micronutrients during and after diarrhoea.

5. **Specific aims:**

a. Estimation of the loss of zinc and other micronutrients from comminuted chicken diet and rice based diet in children with persitent diarrhoea and milk based diet in acute diarrhoea patients.

b. Absorption of zinc and net balance of iron, copper and magnesium will be studied using stable isotope ^{67}Zinc in a 72 h balance study.

c. The effect of mucosal damage on absorption of zinc.

METHODS OF PROCEDURE

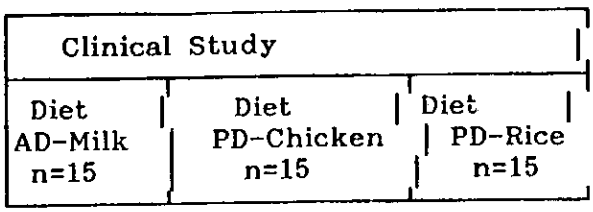
Children who come to the diarrhoea treatment centre for acute and persistent diarrhoea will be recruited for the study. Children will be studied for zinc balance and apparent absorption of copper, iron calcium and magnesium after recovery. All patients will receive standard treatment according to ICDDR,B practice.

* Exclusion Criteria:

1. Children with systemic infection, T.B., temperature >38^0 C.
2. Children with Kwashiorkor.
3. Children with xeropthalmia, nightblindness, bitot's spot.

* Inclusion/selection criteria

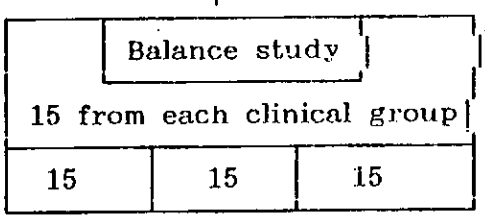
1. Age: 6-24 months.
2. Sex: Male children for metabolic studies and both sexes for clinical studies.
3. Diarrhoea for less than 3 days for acute diarrhoea groups.
4. Diarrhoea for >2 weeks and <2 months for persistent diarrhoea group.
5. Nutritional status: ≤75% wt/age.
6. Agreement by the parents for balance studies.



1. Stool weight
2. Duration of diar
3. Stool frequency
4. Body Wt,Ht,MUAC

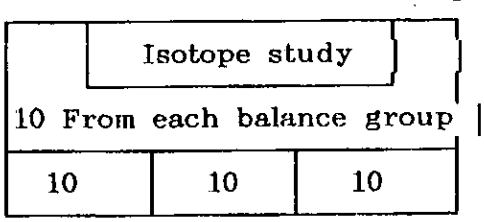
For 7 days
1. Morbidity of diarrhoea & ARI
2. Measure weight, Height & MUAC

5. Intestinal permeability test — on D1, D5 and D15
6. Blood sample for Zn,Fe,Cu,Mg, on D1 - and - D15



Measure:

1. Stool
2. Urine — For 24 hr prebalance and 72 hr balance period
3. Dietary intake in acute and recovery stage
4. ORS intake
5. I.V. input
6. Estimate Zn,Fe,Cu,Mg & Energy from 72 hr stool & food samples



Follow methods & procedures for 24 hr prebalance & 72 hr balance

1. 0.5 mg ^{67}Zn in isotonic saline at 0 min.
2. Blood samples at 0,30,60 & 240 min. for estimation of ^{67}Zn
3. 72 hr stool for estimation of ^{67}Zn

* Working Definitions:

1. Acute Diarrhoea: 3 or more loose stools per day diarrhoea less than 7 days.

2. Persistent diarrhoea: Diarrhoea of acute origin that continues for more than 14 days.

3. Malnutrition:
   According to National Centre for Health Statistics (NCHS) standard, nutritional status will be assessed. Children <75% wt./age will be selected for the study.

4. Systemic Infection

   These will include septicaemia, bronchopneumia and meningitis.

5. Recovery from Diarrhoea:

   Passage of soft stool and absence of diarrhoea for 3 days in both acute and persistent diarrheoa.

Outcome variables:

1. Daily stool output in gramme per kg body weight per day over the treatment period of 7 days.
2. Period of recovery.
3. Macronutrient intake in diet and loss in stool.
4. Micronutrient intake in and loss.
5. Stable isotope in blood and stool.
6. Intestinal permeability, lactulose/mannitol.
7. Anthropometric measurement:
   Weight in kg (precision up to 10g),
   length measurement in cm (precision 1mm),
   mid-upper arm circumference in cm (precision upto 1 mm).

Increase in length will be measured with the help of an electronic digital knee-heel vernier for accurate measurement which has been used successfully in Denmark.

Sample Size Calculation

1. Metabolic balance study on Zn, Cu, Fe, Mg, will be done in Acute and Persistent Diarrhoea and at convalescence. There will be 15 children in each sub group giving 45 study children for balance study.
   (15 X 3)= 45 X 2 = 90 metabolic balance study for other micronutients
   (10 X 3) =30 X 2= 60 ^{67}zn stable isotope study

The calculation of sample size are based on data from the following:

1. For zinc absorption: Castillo-Duran et al (1988), Solomons et al. (1981).

The formula used for calculation of sample size:

Assumption was made that the zinc supplementation will compensate zinc loss by 30% in diarrhoea patients compared to that after recovery. Taking the probability at 5% level and power 80%, calculation was done with following formula according to Kirkwood (1988).

$$n \geq \frac{(u+v)2\ (S1^2 + S2^2)}{(x_1 - x_2)^2}$$

when u=power, v=significance level
S_1 and S_2=standard deviations
x_1 and x_2 = group means. n = 15.
This leads to a total number as following:
in 1) Acute diarrhoea= 15
2) Persistent diarrhoea= 15 X 2=30 (groups= 2 diets).
Total 45

Treatment allocation:

After selection the patients will be randomly allocated to either diets. Groups will receive 20 mg zinc per day in three divided doses for 2 weeks.

Trial groups:

1. Persistent diarrhoea :
   a) comminuted chicken
   b) rice suzi
2. Acute diarrhoea :
   c) milk suzi

Complete formula is given in the appendix

1. Balance and Metabolic study:

24 hour - prebalance period with milk-based diet for acute diarrhoea and with rice-based diet and comminuted chicken diet for persistent diarrhoea patients will be allowed.

A 72 hour balance study with the above diets will be carried out using standard procedure.

Patients will receive diets and 20 mg elemental zinc with vitamins per day.

Rehydration during diarrhoea will be maintained with intravenous polyelectrolyte solution.

Measure: Dietary energy and Zn, Fe, Cu, Mg, Ca intake.

Stool loss of energy, Zn, Fe, Cu, Mg,Ca.

2. Stable isotope study

Zinc absorption can be measured by stable isotope fecal monitoring following an oral dose of enriched ^{67}zn. This is the simplest method of assessing zinc absorption rates by which the subject receives an oral dose of enriched ^{67}zn. All stool collections over a period of 7 days are then analysed for isotopic enrichment. The absorption of zinc is presumed to be represented by that proportion of the oral dose of isotope which does not appear in the faeces. The dose of ^{67}zinc is about 5 mg. The advantages of this test are that it requires relatively few analyses of isotope enrichment, although for good precision, these need to be undertaken by thermal ionisation-mass spectrometry. In a group of normal adults consuming similar diets the precision would be expected to be about 20-25% (c.v%). A recent work has proven this efficacy (Mason et al Br. J. Nutr. 63: 597-611; 1990, Fairweather Tait et al. 1989, Fairweather Tait et al. 1991).

Clinical procedure:

Patient fulfilling the inclusion criteria will be selected for the study. Informed and written consent will be obtained from the parents. Patients will be clinically examined thoroughly and initial observation in regards to stool output for 4 hours in acute diarrhoea group. Patients having stool output of 250 gm/kg of body weight in 4 hours will be selected for metabolic study.

Patients with persistent diarrhoea will enter into metabolic balance study.

Correction of initial dehydration and maintenance of hydration of the patients undergoing balance study will be done by the intravenous cholera fluid (Poly-electrolyte solution).

Metabolic balance study:

Stable isotope method for zinc absorption (Jackson et al 1984) will be used. Conventional metabolic balance studies will be carried out in children using carmine marker. Milk based diet will be given to children in acute diarrhoea and rice based diet will be given to the patients with persistent diarrhoea. Basal zinc intake will be estimated. Zinc supplementation will be given in doses of 20 mg.

Enrichment of body pools with ^{67}Zn:

^{67}Zn will be dissolved in hydrochloric acid and will be neutralized with sodium hydroxide. The solution will be diluted in 0.9% saline before use. Each subject will receive a single intravenous injection of 2 mg enriched ^{70}Zn in sterile saline and the 1st carmine fecal marker will be given orally before injection and urine and stool will be collected. A second carmine marker will be given orally after 72 hours and total stool will be collected between the two markers. Blood (5ml) will be taken for ^{70}Zn analysis at 0, 30, 60 and 240 minutes followed by samples at intervals of day 1, 2, and 3. Thermal isonization mass spectrometry will be used to analyse ^{70}Zn (Fairweather-Tait et al. 1989). A logarithmic plot of plasma enrichment of zinc vs time after zinc infusion will be plotted and from this plasma enrichment at the mid-point of balance period will be derived. Comparison of this value with the ^{67}Zn enrichment of the feces from that period will allow the proportion of the total fecal zinc derived from gastrointestinal secretion will be calculated.

Since the total fecal output (F) is equal to the dietary intake (D) minus the zinc absorbed (A) plus that secreted into the gut (S) (i.e F = D - A + S), a knowledge of the gastrointestinal zinc secretion, the total fecal zinc and the dietary intake allowed the zinc absorbed to be calculated (i.e A = D - F + S) (Fairweather Tait et al. 1991).

Permeability test:

Children with persistent diarrhoea will receive a 20 ml oral dose of 5 g lactulose and 1 g mannitol and subsequent urine will be collected for a 5 hour period. The percent recovery of the dose will be calculated from urinary concentration and total urine volume. The Permeability test with disaccharide lactulose and monosaccharide mannitol probes will be able to identify small intestinal mucosal integrity without using invasive procedure like biopsy.

LABORATORY INVESTIGATIONS:

Stool M/E and routine pathogen isolation e.g.cholera, shigella, rotavirus, campylobacter, ETEC.

Blood: TC, DC, Zinc, Cu, Mg on admission and at recovry.

Sample for stable isotope ^{67}Zn:

0, 30, 60, 240 minutes. ^{67}Zn will be assayed by one of the investigators Dr. M.J. Jackson of the university of Liverpool.

Permeability test:

Before and after balance, 5th day, 8th day and on 15th Lactulose will be measured by and automated enzyme assay using oxidation of sugar which will be measured in Cobas-bio. Mannitol will be analyzed with oxidation of sugar by mannitol dehydrogenase as described by Yamanaka (1965) using Cobas-bio (Beherens et al 1984).

Stool: 8 hourly stool collection throughout the metabolic period and measurement up to 7 days.

Urine: Complete collection during balance period and permeability tests.

Micronutrient estimation of food and excreta:

Zinc, Iron, Copper and magnesium content of diet and all output during balance will be measured by atomic absorption spectrometry. Trace elements will be assayed in the biochemistry and nutrition laboratory in I.C.D.D.R,B.

Energy estimation of food and excreta:

Energy in diet and stool will be estimated using adiabatic bomb calorimeter in the biochemistry laboratory of ICDDR,B.

Data analysis plan:

(1) Descriptive statistics will be done for baseline information on micronutrient loss in diarrhoea. There relationships will be seen with extent of mucosal damage by results of permeability test.

(2) Comparison of apparent absorption of micronutrients and true absorption of zinc between diarrhoeal phase and convalescence within the same groups of children will be performed by paired t-test.

(3) Comparison of apparent absorption of micronutrients and true absorption of zinc between two dietary groups of persistent diarrhoea and between acute diarrhoea and individual persistent diarrhoea groups will be performed by t-test.

(4) The relationship of apparent absorption of micronutrients and true absorption of zinc with acute and persistent diarrhoea, dietary groups and different grades of mucosal damage as measured by permeability test will be assessed.

(5) Clinical outcome as measured by stool output per kg per day and period of recovery will be determined for individual groups and will also be compared between the groups by t-test.

Facilities required:

Space:

Will be required to preserve and prepare samples for mass-spectrometer estimation. Computer facilities: To be increased. A 486 new computer with 2 high density drives and 80 MB HD and 25 MHZ speed would be required for this project.

Laboratory aspects:

ICDDR,B laboratories will be utilized.

Hospital resources:

Outdoor and indoor facilities will be used to get patients and continue metabolic, biochemical and clinical studies. Metabolic ward will be used.

Storage space will be required in the biochemistry department and hospital building.

Exchange programme:

A clinical research fellow of the department of international child health of Institute of child health will work in the project and participate in clinical research and training in the centre, salary will be included in this project. Principal investigators will supervise and work in the project in Dhaka and London. Personnel from ICDDRB, will be sent for training in stable isotope works.

Collaborative Arrangement

The project will be conducted in a collaborative arrangement among ICDDR,B, ICH (London), ARFC laboratories. There will be technology transfer and and mutual strengthening of research capacity.

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BUDGET

1 Year

A.	Manpower:		
1)	Dr. S. K. Roy (25%)	US$ 7325	
2)	Dr. S.M. Akramuzzaman	US$ 3500	
3)	(20%)	US$ 3500	
4)	Dr. Nahrina Dewan (20%)	US$ 2000	
5)	Ms. Makduma Khatoon (10%)	US$ 2000	
6)	Secretarial service	US$ 4000	
7)	Research Fellow (2)	US$ 3000	
8)	Health Assistants (3) Ward Attendants (3)	US$ 1400	US$ 26,725
B.	Travel		
1	Local	US$ 1000	
2	International	US$ 3500	US$ 4500
C.	Supplies and Materials		
1.	Office supplies	US$ 3000	
2.	Medical supplies	US$ 2000	
3.	Chemicals	US$ 1000	
5.	Isotope	US$ 6000	
6.	Airfreight	US$ 2000	US$ 14,000
D.	Interdependent patient	US$ 4000	US$ 4,000
E.	Interdepartmental Service		
1.	Lab	US$ 3000	
2.	Bio	US$ 4000	
3.	Zn	US$ 6000	
4.	NIE	US$ 5000	
5.	L/M	US$ 3000	
6.	Isotope	US$ 30000	US$ 51,000
F.	Capital Expenditure		
1.	Computer	US$ 4000	
2.	Furniture	US$ 1500	US$ 5,500
G.	Other contractual services		
1.	Postage	US$ 2000	
2.	Fax	US$ 2000	
3.	Maintenance	US$ 1000	US$ 5,000
		Total	US$ 110,725
	31% overhead cost		US$ 34,325
			==========
	GRAND TOTAL		US$ 145,050

Abstract Summary for Ethical Review Committee

1. This study aims to estimate the bioavailability of zinc in children with acute and persistent diarrhoea from some effective dietary regime. There has been evidence that micronutrient is lost more during diarrhoea. These diets have been shown to be effective in ICDDR,B for early recovery from diarrhoea. Since children progress into malnutrition rapidly with persistent diarrhoea and it is a risk condition for mortality, a thorough knowledge in this field will help better management with appropriate supplementation plan.

   The children will be of age 6 month to 24 months as this is the high risk age bracket for acute and persistent diarrhoea. Children will receive standard ICDDR,B diets and 20mg zinc per day.

2. There is no risk with stable isotope dose which naturally occur in body tissue.

3. The research team of the study will give all clinical care full time and patients will be in observation in the metabolic ward.

4. Mothers or guardians will be explained fully about the purpose of the study and benefit of the study, procedure of the study and informed written consent will be taken.

5. Confidentiality will be maintained by using identification number and keeping the patient records in filing cabinets.

6. Trained Health Assistants will take interview for ten minutes at a time. Stool and urine and 5 ml blood will be collected 4 times during the study period. Balance study will be carried out with stable isotape ^{67} zinc.

7. Subjects will be provided with standard management for diarrhoeal disease. This study will bring new knowledge on zinc balance during diarrhoea to help better management of children for diarrhoea.

^{67}ZINC BALANCE STUDY

Consent Form

The ICDDR,B is carrying out research for better management of malnourished children with acute and persistent diarrhoea. It has been known that essential trace element like zinc is excessively lost during diarrhoea. to give the appropriate amount of micronutrient for better recovery it is necessary to measure the amount of zinc lost in the stool of your child.

Your child will undergo following procedures during the study:

1. Your child will be given appropriate food.

2. Stool and urine and 5 ml of venous blood will be collected 4 times during balance study.

3. He will be given a dose of zinc stable isotope during diarrhoea and on recovery.

There is no risk in participating in this study. You will have the right to withdraw your child at any time during the study. Your child will receive appropriate medical care even if you withdraw your child from the study. We shall maintain confidentiality of your child.

If you wish to make your child participate in the study please sign or put your left thumb impression below:

Signature of the
Principal Investigator

Date:_____

Signature or left thumb
impression of patients or guardian

Date:_____

জিংক (দস্তা) এর পরিমান নির্ণয় গবেষনা

সম্মতি পত্র

অপুষ্টিজনিত কারনে রোগাক্রান্ত শিশু যারা সুদ্ধ ও দীর্ঘ মেয়াদী ডায়রিয়ায় ভুগছে তাদের সুচিকিৎসার জন্য আন্তর্জাতিক উদরাময় গবেষনা কেন্দ্র (আই, সি, ডি, ডি, আর, বি) একটি গবেষনা প্রকল্প হাতে নিয়েছে, যা প্রমানিত হয়েছে যে খনিজ পদার্থ যেমন জিংক (দস্তা) যা ডায়রিয়া হলে প্রচুর পরিমানে বের হয়ে যায় তা শিশুর অপুষ্টি ও দীর্ঘ রোগভোগের একটি কারন,

আপনার শিশুর পায়খানায় এর সঠিক পরিমান নির্ণয় করা প্রয়োজন, যাতে করে আপনার শিশুর দ্রুত আরোগ্যের জন্য এই খনিজ পদার্থ জিংক (দস্তা) সঠিক পরিমানে দেয়া যেতে পারে,

এই গবেষনা চলাকালীন সময়ে আপনার শিশুর নিম্নলিখিত ব্যবস্থা নেয়া হবে:

(১) শিশুকে প্রয়োজনীয় খাদ্য দেয়া হবে,

(২) চিকিৎসা কালীন সময়ে পায়খানা, প্রস্রাব এবং মোট চারবার আপনার শিশুর শিরা থেকে ৫ মি.লি (১ চা চামচে সমপরিমান) রক্ত নিয়ে পরীক্ষা করা হবে,

(৩) আপনার শিশুকে চিকিৎসার শুরুতে একবার ও শেষে একবার জিংকে আইসোটোপ দেয়া হবে,

এই গবেষনায় অংশগ্রহন করা আপনার শিশুর জন্য সম্পূর্ন নিরাপদ, আপনি যে কোন সময়ে এই গবেষনা থেকে আপনার শিশুকে প্রত্যাহার করে নিতে পারেন, প্রত্যাহার করার পরও আপনার শিশু এই হাসপাতালের প্রচলিত সুচিকিৎসা পাবে। আপনার শিশু সম্পর্কিত সমস্ত তথ্য গোপন রাখা হবে,

এই গবেষনায় আপনার শিশুকে অংশগ্রহন করাতে চাইলে নীচে স্বাক্ষর / টিপসই করুন,

গবেষকের স্বাক্ষর	সাক্ষীর স্বাক্ষর	অভিভাবকের স্বাক্ষর / বামহাতের বৃদ্ধাঙ্গুলির ছাপ,
তারিখ:	তারিখ:	তারিখ: