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REVIEW BOARD ON THE USE OF HUMAN VOLUNTEERS Date_____
CRC

Principal Investigator Zahid Mozaffar and Dr. Ayesha Molla Trainee investigator(if any) X 26

Application No 80-017 (Revised) Supporting Agency(if Non-CRL)_____

Title of study Malabsorption tests in apparently healthy Bangladeshi children

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):

1. Source of population:
   a) Ill subjects Yes (No)
   b) Non-ill subjects (Yes) No
   c) Minors or persons under guardianship (Yes) No
2. 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 possibly damaging to subject or others Yes (No)
3. 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
4. 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
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 Board for review.

We agree to obtain approval of the Review Board on Use of Human Volunteers for any changes involving the rights and welfare of subjects before making such change.

Zahid Mozaffar. Ayesha Molla
Principal Investigator

Trainee

...return 2 copies of review protocol to Chairman, Review Board ...

80-017 (Revised
Rec'd 27/5/80

1. 50 apparently healthy children age between 2 to 5 years will be selected for the study. The rational of taking children in this study is that, these group of population of our society mostly are the sufferers of malabsorption which becomes even worst after recurrent infection of intestinal parasites, bacterial and viral diarrhoeas.

2. No potential risks

3. None

4. The subjects will be kept anonymous and data will be kept under strict confidentially.

5. Informed consent will be obtained from the study patients. In case of minor groups, guardians will be asked for the consent. Description of the consent form is described and attached in separate sheet.

   (a) Not applicable

   (b) Not applicable

6. Only clinical history will be necessary.

7. This study will be helpful to assess the degree of malabsorption of foods due to various reasons and eventually the patients will be benefitted in getting proper management and advice for the recovery of the syndromes.

8. On admission a blood sample will be taken for biochemical tests. For xylose test five hour urine sample and for lactose tolerance test finger prick blood sample will be taken for research purpose. Stool samples will also be used for research purpose. Patients clinical history and birth data will be used for age determination.

SECTION I - RESEARCH PROTOCOL

1. TITLE : Malabsorption tests in apparently healthy Bangladeshi children

2. PRINCIPLE INVESTIGATOR : Zahid Mozaffar, Fellow-in-Nutrition
   Dr. Ayesha Molla, Supervisor

3. STARTING DATE : July 1, 1980

4. COMPLETION DATE : December 31, 1980

5. TOTAL DIRECT COST : $ 136,134.53 and incremental cost $ 4,441.67

6. SCIENTIFIC PROGRAMME HEAD

   This protocol has been approved by the Working Group. _____

   Signature of Scientific Programme Head _____

   Date _____

7. ABSTRACT SUMMARY

Malabsorption syndrome is one of the most common problem of our community. The children particularly those under 5 years of age are the worst sufferers. The purpose of the present study is to investigate the criteria of malabsorption in paediatric age group. A total of 50 apparently healthy children (age between 2 to 5 years) will be selected for the study. On first day, nutritional status will be determined by measuring height, weight, head and mid-arm circumference and biochemical tests like serum total protein, albumin, globumin, haematocrit, vitamin A, TWBC, and differential count values. Stool microscopic examination and stool culture will also be done. On the second day of hospitalization D-xylose absorption test will be performed after an overnight fast. On the third day after an overnight fast lactose tolerance test will be carried out by feeding a dose of 2g lactose/kg body weight. Finger prick capillary blood samples (50 µl) at 0 and 30 minutes after lactose feeding, will be taken and blood glucose will be analysed. After the completion of above tests fat balance study will be done and in which only stool sample will be collected for fat estimation.

8. REVIEWS

   (a) Ethical Review Committee _____

   (b) Research Review Committee _____

   (c) Director _____

   (d) BMRC _____

SECTION II - RESEARCH PLAN

A. INTRODUCTION

1. Objectives

The objective of the present study is to determine the baseline value of xylose absorption test, lactose tolerance test and fat balance study in apparently healthy Bangladeshi children aged between 2 to 5 years. This would allow us to understand the value of these tests performed in children to assess the malabsorption due to diarrhoea of various aetiology.

2. Background

Abnormal net transfer of nutrients from the intestinal lumen into the body results in the malabsorption. Malabsorption can also occur during bacterial and viral diarrhoea, parasitic infectation and due to protein energy malnutrition, which is most commonly seen in this country. To investigate malabsorption, lactose tolerance test, faecal fat study, vitamin B_{12} absorption studies were performed by various workers (1,2,3,4). Besides, peroral biopsy of small intestine were also used as an important tool to identify intestinal mucosal lessions in different malabsorption syndromes. Measurement of D-xylose absorption is a standard procedure used by many workers (5,6). Urinary xylose excretion is sometimes misleading if there is delayed gastric emptying, decreased circulating blood, incomplete urine collection and impaired renal function.

An alternative method for xylose estimation is to measure the blood xylose level after one hour ingestion of a definite dose (5gm) of xylose (7). Ordinarily, adults excrete 2 to 5mg of pentose sugars per kg of body weight per 24 hour and none is normally detectable in serum (8). Lactose malabsorption due to lactase deficiency is very common in diseased as well as many healthy population of the different parts of the world (9). Several studies in children indicated that primary lactose malabsorption is rare in infants (4,10,11) and increases with age (12,13). In individuals with lactose malabsorption, intake of milk products may induce diarrhoea. In case of children it was also observed that children fed normal milk were found to excrete significantly more fat in 24 hour than those fed lactose hydrolysed milk (14). In our population, because of scarcity and high cost of milk, its intake is automatically less than other developed nations. Measurement of faecal fat excretion over a definite period of time were used by many workers (15,16) as a good index of fat absorption. In patients with sprue, the available evidence shows that the daily quantity of faecal fat is largely dependant on the amount ingested (17,18). In normal adults it has been shown that the reduced but significant fat excretion could be obtained after receiving fat free diets (19). The endogenous

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component of faecal fat may derive in varying amounts from lipid excretion bacterial synthesis, desquamated mucosal cells and bile (24). All these parameters of malabsorption have been tested in developed countries in different types of population and their validity or significance among the local paediatric population of Bangladesh have not been studied.

Considering the above background this study is undertaken to assess the degree of malabsorption and simultaneously nutritional status of the paediatric age group. Three different criteria namely xylose test, lactose tolerance test and fat balance study will be performed and attempt will be made to correlate the results with anthropometrical and biochemical parameters of the children. This study will provide us valuable data for assessment of malabsorption in apparently healthy children.

3. Rationale

Although malabsorption is a common condition in Bangladesh, there has not been any study to define the index of absorption and criteria to define malabsorption in apparently healthy children. This study will help to establish the criteria to define the degree of malabsorption in paediatric age group.

B. SPECIFIC AIMS

To standardise the different tests like xylose absorption, lactose tolerance and faecal fat estimation to study malabsorption in apparently healthy children.

C. METHODS OF PROCEDURE

About 50 apparently healthy children of 2 to 5 years of age will be chosen for this study. "Apparently healthy child" can be defined as those having nutritional status within 10-20% of normal of the international standard without having any history of diarrhoea for last one month. Subjects with other complicated diseases will not be selected for the study.

On admission, nutritional status will be assessed by measurement of body weight, height, head and mid-arm circumference and a venous blood sample will be drawn to determine total protein, albumin, globulin, vitamin A, haematocrit, TWBC and differential count. Stool microscopic examinations and stool culture will also be done on the same day. Children will be selected from a neighbouring village near Narayanganj.

Besides anthropometric and biochemical assessment the following absorption tests will be performed.

Xylose absorption test

Xylose test will be performed according to Roe & Rice (20). Following day of admission after an overnight fast the subjects will be fed 5g of xylose in 25ml water followed by a drink of a 250ml water. After xylose feeding the subjects will be encouraged to drink water and five hour urine sample will be collected for xylose measurement. To prevent bacterial growth 2 or 3 drops of toluene will be added.

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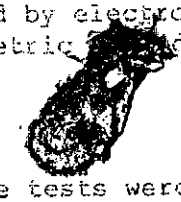
Lactose tolerance test

Lactose tolerance test will be done according to Woteki et.al. (21). On the 3rd day of admission after an overnight fast lactose tolerance test will be performed using a dose of 2g lactose/kg body weight. Lemon juice can be added as flavour in the solution. Finger prick capillary blood samples (50 µl) at fasting and at 30 minutes after lactose ingestion will be taken. Blood glucose will be analysed by Nelson and Somogyii method. A blood glucose rise over of 25mg/100ml or more than fasting will be interpreted as lactose absorption.

Fat balance study

From the 3rd day of hospitalization fat balance study will be carried out. In this study patients will be given a diet containing 40gm of fat for 72 consecutive hours. Food and a charcoal marker will be fed in the beginning of the study and faeces collection will start soon after excretion of the marker. A second marker will be fed at the end of 72 hour and stool collection will be continued till the marker comes out with it. Stool samples will be refrigerated from the beginning of the study and after the collection of last stool all the samples will be homogenized in a blender, weighed and an aliquote will be saved for analysis of fat according to the method of Van de Kamer (22).

Total protein, albumin and globulin will be determined by electrophoresis, vitamin A will be determined by the fluorometric method of Hansen and Warwich (23).



D. SIGNIFICANCE

D-xylose absorption, fat balance and lactose tolerance tests were done before in sick children by many workers to assess the degree of malabsorption. In Bangladesh apparently healthy population probably suffer from various subclinical infection altering the intestinal mucosa. Therefore, to get the base line data of these control group of children, this study will be extremely helpful for evaluating the importance of these tests.

E. FACILITIES REQUIRED

1.	Office space	:	None
2.	Laboratory space	:	Biochemistry Laboratory
3.	Hospital resources	:	ICDDR,B ward facilities is required
4.	Logistic support	:	ICDDR,B transport facilities is required
5.	Equipment	:	Equipments in Biochemistry and Pathology Branch will be used.
6.	Extra personnel	:	No such extra personnel is absolutely required. But assistance from the existing Biochemistry technician may be required.

"Apparently healthy children" and their expected health problems. Out of eight subgroups only group-1 for this study will be healthy by our defination though we might not always be able to pick this group of children from a village based society.

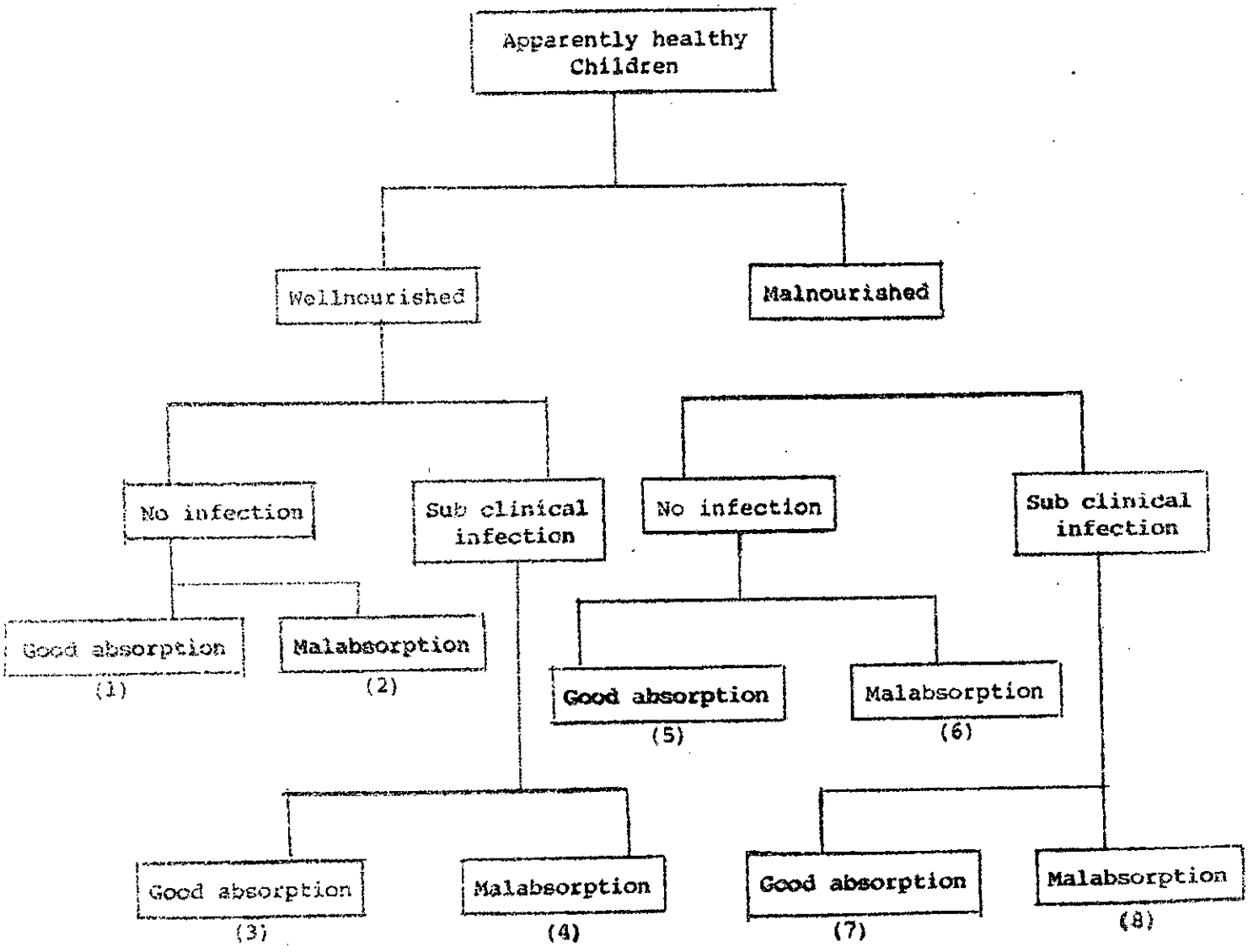


Figure 1. Schematic diagramme shows the criteria of "Apparently healthy children"

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SECTION III - BUDGET

A. DETAILED BUDGET

1. PERSONNEL SERVICES

N A M E	POSITION	% OF EFFORT	ANNUAL SALARY	PROJECT REQUIREMENT	US $
Zahid Mozaffar	Fellow-in-Nutrition	100	Tk. 24,840.00	Tk.24,840.00	1,656.00
Dr. Ayesha Molla	Supervisor	20	Tk.1,15,925.11	Tk.23,185.02	1,545.67
Dr. A.M. Molla	Scientist	10	Tk.1,32,211.76	Tk.13,221.18	881.41
Study Nurse	---	20	Tk. 23,316.66	Tk. 4,663.33	310.89
Physician	---	10	Tk. 36,000.00	Tk. 3,600.00	240.00
				Tk.69,509.53	4,633.97

2. SUPPLIES AND MATERIALS

T E S T	COST PER TEST	TOTAL TAKA	US $
Total Protein Albumin Globulin Electrophoresis	36.00	3,600.00	240.00
Vitamin A	8.00	800.00	53.30
Hematocrit, TWBC & Differential count	4.20	420.00	28.00
Glucose	1.50	150.00	15.00
Stool M/E	2.05	205.00	13.67
Stool culture	15.00	750.00	50.00
Fat	36.00	3,600.00	240.00
		9,525.00	639.97

ITEMS	AMOUNT REQUIRED	TOTAL TAKA	US $
Xylose	1 kg	2,000.00	133.33
Lactose	5 kg	2,000.00	133.33
Tab. Ultra Carbon	500 tab.	500.00	33.33
		14,025.00	939.96

3. EQUIPMENT — Nil

4. PATIENT HOSPITALIZATION — 150 x 6 x 50 = 45,000.00

5. OUTPATIENT CARE — Nil

6. ICDDR,B TRANSPORT — 600 miles, 3 x 600 = Tk.1,800.00

7. TRAVEL AND TRANSPORTATION OF PERSONS — 3 x 600 = Tk.1,800.00

8. TRANSPORTATION OF THINGS — Nil

9. PRINTING AND REPRODUCTION
   - Mimeograph — Tk.500.00
   - Xeroxing — Tk.500.00

10. OTHER CONTRACTUAL SERVICES — Tk.1,500/- for remuneration
    Tk.1,500/- for local transport

11. CONSTRUCTION, RENNOVATION, ALTERATIONS — Nil

12. RENT / COMMUNICATION — Nil

BUDGET SUMMARY

	CATEGORY	TAKA	DOLLAR
1.	Personnel	69,509.53	4,636.97
2.	Supplies	14,025.00	939.96
3.	Equipment	--	--
4.	Patient Hospitalization	45,000.00	3,000.00
5.	Out patient care	--	--
6.	Travel and Transportation of persons	1,800.00	120.00
7.	ICDDR,B Transport	1,800.00	120.00
8.	Transportation of things	Nil	Nil
9.	Printing and Production	1,000.00	66.67
10.	Contractual services	3,000.00	200.00
11.	Construction	--	--
12.	Rent / Communication	--	--
		136,134.53	9,080.60

Total direct cost = Tk.136,134.53

Incremental cost = Tk.66,625.00 $ 4,441.67

Tk.15.00 = US $ 1.00

সম্মতি পত্র

আই, সি, ডি, ডি, আর, বি, ঢাকা কেন্দ্র মোটামুটি স্বাস্থ্যবান শিশুর শরীরে খাদ্য ঠিকমত ব্যাবহৃত হচ্ছে কি না সে সর্ম্পকে গবেষণা চালানোর সিদ্ধান্ত নিয়াছে। এই পরীক্ষা দ্বারা আমরা বলতে সক্ষম হব যে আপনার সন্তান যে খাবার গ্রহণ করছে তা তার শরীরে কতটুকু ব্যবহৃত হচ্ছে।

যদি আপনি চান যে আপনার সন্তান এই গবেষণায় অংশগ্রহণ করুক তবে নিম্নলিখিত পরীক্ষাগুলো করা হবে এবং এই গবেষণায় যদি কোন রোগ পাওয়া যায় তবে তার উপযুক্ত চিকিৎসা করা হবে।

(১) প্রথমদিন আপনার বাচ্চার শরীরের ওজন এবং উচ্চতা মাপা হবে এবং তার রক্ত ও পায়ুখানা পরীক্ষা করা হবে।

(২) সেই দিন রাত দশটায় আপনার বাচ্চাকে শেষ খাবার দেওয়া হবে এবং পরদিন সকালে জাইলোজ (চিনি জাতীয় খাবার) এর সরবত খাওয়ানো হবে এবং ৫ ঘন্টা পর্য্যন্ত তার প্রস্রাব সংগ্রহ করা হবে। তারপর আপনার বাচ্চাকে খাবার খেতে দেওয়া হবে।

(৩) দ্বিতীয় দিন আপনার সন্তানকে রাত দশটায় শেষ খাবার দেওয়া হবে এবং পরদিন সকালে লেকটোজ (চিনি) এর সরবত খাওয়ানো হবে এবং তার আংগুল হতে ০ এবং ৩০ মিঃ পর অতি সামান্য (৫০ মাঃ লিঃ) পরিমান রক্ত নেওয়া হবে।

(৪) চতুর্থদিন হতে দুই চারকোল মার্কারের মাঝের ৭২ ঘন্টায় পায়ুখানা সংগ্রহ করা হবে। এই মার্কার শরীরের কোন ক্ষতি করে না।

আপনি যে কোন সময় গবেষণা হতে আপনার বাচ্চাকে প্রত্যাহার করে নিতে পারেন।

যদি আপনি এই গবেষণায় অংশ নিতে চান তাহলে দয়া করে এই সম্মতি পত্রে স্বাক্ষর করুন।

অভিভাবকের স্বাক্ষরঃ---------- তারিখ----------

গবেষকের স্বাক্ষরঃ---------- তারিখ----------

CONSENT FORM

ICDDR,B, Dacca centre has undertaken a research programme to determine the malabsorption tests in apparently healthy children. From this study we would be able to tell you whether the food normally taken by your child is properly absorbed in his/her body or not.

If you would like your child to take part in this study the following procedures will be performed. Out of this investigation if your child is found to be malabsorber he/she will be treated with proper medication.

(1) On first day we shall take the weight and height of your children. Stool and blood samples will also be examined.

(2) On the same day the child will be kept N.P.O. from 10.00 pm and next morning they will be given a drink of xylose (a harmless, inert sugar) and five hour urine sample will be collected. The child will be allowed to take food afterwards.

(3) On the second day the child will be kept N.P.O. from 10.00 pm and on the following morning he/she will be given a drink of lactose (sugar) and finger prick blood sample (50 μl) will be taken at 0 and 30 minutes afyer lactose ingestion.

(4) From fourth day onwards only stool samples for 72 hours will be collected between two charcoal markers. The marker is a harmless substance.

You have every right to withdraw your child from taking part in the study at any time you may wish.

If you agree to take part in this examination, please sign the concent form.

Your signature

_____ Date _____

_____ Date _____

Investigator's signature