

ETHICAL REVIEW COMMITTEE, ICDDR,B.

Principal Investigator Dr Nigar Shahn
Application No. 83-037(P)
Title of Study Cryptosporidium as a pathogen for diarrhoea in Bangladesh

Trainee Investigator (if any) _____
Supporting Agency (if Non-ICDDR,B) _____
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 Cttee. for review.

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

Nigar S Shahn
Principal Investigator

Trainee

SECTION I - PILOT PROTOCOL

1. Title : Cryptosporidium as a pathogen for diarrhoea in Bangladesh
2. Principal Investigator : Dr. Nigar S. Shahid
- Co-Investigator : Dr. Hamidur Rahman,
Dr. K.A. Mahmud
- Consultant : Dr. Bruce A. Anderson
3. Starting Date : October 1, 1983
4. Completion Date : March 31, 1984
5. Total Direct Cost : US \$ 2,795.00
6. Scientific Program Head :

This protocol has been approved by the Disease Transmission Working Group.

Signature of the Program Head :

Date :

W.B. [Signature]
3.10.83

7. Abstract Summary :

Stool of diarrhoeal patients coming under the surveillance system (4% systematic sample) in Dhaka hospital will be stained with Giemsa stain and examined under oil emersion in light microscope. In the surveillance system the samples are regularly being examined for Salmonella, Shigella, V. cholerae O1 and V. cholerae non O1, Campylobacter, ETEC, and Rotavirus. Stool microscopy is also performed. Clinical and social characteristic of the patients are also recorded. Being attached to the surveillance system we should be able to obtain a clear account of the diarrhoea produced by this protozoa.

8. Reviews :

- a. Ethical Review Committee : _____
- b. Research Review Committee : _____
- c. Director : _____
- d. BMRC : _____

SECTION II - RESEARCH PLAN

Introduction:

1. Objective:

The objective of this protocol will be to establish cryptosporidium as a cause of diarrhoea in Bangladesh.

2. Background:

Despite the large number of enteropathogens identified in recent years, the cause of many diarrhoeal episodes remain undetermined.¹ The ICDDR,B. surveillance system in 1982 could identify a cause of diarrhoea for 60% of diarrhoeal cases coming under surveillance. Of these 1.6 were Entamoeba histolytica and 2.7 Giardia lamblia. The bacterial pathogens looked for were ETEC, Campylobacter, V. cholerae O1 and non O1, Salmonella, Shigella, and rotavirus which is performed by ELISA method.

The protozoan cryptosporidium was first described by Tyzzer² in 1907 and has recently been shown to infect and cause enteritis in a number of animal species by adhering to the microvillus border of intestine epithelial cells.^{3,4,5,6} Cryptosporidia was first described in humans in 1976⁷ and a number of cases of cryptosporidiasis in humans have subsequently been reported.⁸⁻¹⁷ Six reports involved immunologically compromised patients^{8-11,13,17} and five cases have been described in immunologically normal patients.^{7,12,14-16} The infections were determined either in the faeces^{12,14,16} or in intestinal biopsies.^{7-11,13,15,17} Diarrhoea usually lasted for 1-3 weeks and was involved with vomiting, abdominal pain and fever. In an urban hospital patients with gastroenteritis in Australia showed that 7% of patients with diarrhoea during summer were excreting cryptosporidium OO cysts in the feces most of which showed no evidence of any other enteropathogen.

Since a majority of these patients were city dwellers they had little or no contact with animals.¹⁸

We have found cryptosporidium in calves and calve handlers in Bangladesh (Dr. Hamidur Rahman, personal communication) and this shows that this protozoan may be an important pathogen for diarrhoea here.

Rationale. In view of the fact that the cause of large number of diarrhoeal episodes remain unanswered it is necessary to look for newer agents of diarrhoea and to establish it as a cause of diarrhoea in Bangladesh.

Materials and Methods:

Stool samples coming from the patients in the surveillance study will be examined for cryptosporidium in Giemsa stained faecal smears observed under oil-emersion in light microscope and under phase contrast microscope. Medical histories, clinical features, socio-demographic information and microbiological information will already be available through the surveillance study.

Method for preparation of Geimsa stained faecal smears

A thin smear will be passed from fresh stool and quickly dried in air and then fixed with methyl alcohol. The solution for staining will be prepared by adding 1 part of stock Giemsa stain to approximatley 50 parts of buffered water (pH 7.0). The slide will be placed in the staining solution for 45 minutes. The slide will then be removed and dipped into buffer solution for 3 minutes and allowed to dry. It will then be observed wtih oil immersion lens under light microscopy.¹⁹

Significance

This protocol will help up establish a new agent for diarrhoea in Bangladeshi population.

Specific Aims:

1. To establish cryptosporidium as a diarrhoeal pathogen in Bangladesh.
2. To define the age specific incidence of diarrhoea by cryptosporidium.
3. To define the clinical features of cyptosporidium diarrhoea.

Facilities Required

No new physical facility will be required.

Dr. Bruce C. Anderson of the University of Idaho, USA, has been requested to be consultant to this project. He is also a consultant in Dr. Hamidur Rahman's protocol on Cryptosporidium.

SECTION III

A. DETAILED BUDGET

1. PERSONNEL SERVICES

<u>Name</u>		<u>% Time</u>	<u>Annual Salary</u>	<u>Project-Requirement</u>	
				<u>Taka</u>	<u>Dollar</u>
Dr. Nigar Shahid	Principal Investigator	10%	\$ 5,730/-	-	286.50
Dr. Hamidur Rahman	Co-Investigator	10%	\$ 4,850/-	-	242.50
Dr. K.A. Mahmud	Co-Investigator	10%	\$ 7,560/-	-	378.00
To be named	2 Lab. Technician	10%	\$ 1,970/-	-	394.00
			Sub total	-	\$ 1,301.00
				=====	=====

2. SUPPLIES AND MATERIALS

Giemsa strain \$ 400.00
Buffer Solution
Methyl alcohol

3. EQUIPMENT - None

4. PATIENT HOSPITALIZATION - None

5. OUTPATIENT CARE - None

6. ICDDR,B TRANSPORT - None

7. TRAVEL AND TRANSPORTATION OF PERSONS

Air ticket of Dr. Bruce C. Anderson \$ 1,200/-
return trip from USA

8. TRANSPORTATION OF THINGS - None

9. RENT, COMMUNICATION AND UTILITIES

Stay at ICDDR,B Guest House - 14 days \$ 350/-
(25 x 14)

Taka US Dollar

10. PRINTING AND REPRODUCTION

\$ 200/-

11. OTHER CONTRACTUAL SERVICES - None

12. CONSTRUCTION, RENOVATION AND ALTERATION - None

13. MISCELLANEOUS - None

BUDGET SUMMARY

	<u>US \$</u>	
1. Personnel Services		
2. Supplies & Materials	400.00	
3. Equipment	-	
4. Patients Hospitalization	-	
5. Out-patient Care	-	
6. Transport	-	
7. Travel & Transportation	1,200.00	
8. Transportation of things	-	
9. Rent/Communication/Utility	350.00	
10. Printing & Reproduction	200.00	
11. Other contractual services	-	
12. Construction/Renovation/ Alteration	-	
	<hr/>	
Total :	\$ 2,150.00	
Incremental cost (30%):	645.00	
	<hr/>	
Grand Total	2,795.00	US \$
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Abstract Summary

1. Patients coming under the surveillance study (4%) sample and who provide stool will be taken into the study as to minimize cost for looking for other pathogens.
2. There are no risks involved.
3. N.A.
4. Confidentiality shall be maintained.
5. N.A.

The same consent form will be used as the Surveillance protocol.

References

1. Stoll BJ, Glass RI, Huq MI, Khan MU, Holt JE, Banu H. Surveillance of patients attending a diarrhoeal disease hospital in Bangladesh. *British Medical Journal*. 1982, 285, 1185-88.
2. Tyzzer CE. A sporozoon found in the peptic glands of the common mouse. *Proc. Soc. Exp. Biol. Med.* 1907, 5: 12-13.
3. Morin M, Lariviere S, Lallier R, Begin M, Roy R. and Ethier R. Neonatal calf diarrhoea : pathology and microbiology of spontaneous cases in dairy herds and incidence of enteropathogens implicated as aetiological agents. *Proc. 2nd Symp. Neonatal Diarrhoea (VID) Sadkatton.*, 1978, 347-369.
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19. Bruce C, Anderson. Pattern of shedding of Cryptosporidial oocysts in Idaho calves. *Journal American Vet. Med. Assoc.* 1981, 178(9) : 982-984.

স্বাস্থ্য তিথিকা

আন্তর্জাতিক উদযোগ্য জাতিসংঘ (কেন্দ্র) স্বাস্থ্যবিদগণে যে
সব যোগ্যী আমেরন আমের স্বাস্থ্য প্রতি ২০৩৫ যোগ্যী- বিস্তারিত
তথ্য সংগ্রহ করা হয়। উক্তসংগে পরীক্ষার সব আমেরন।
আমেরন বিতামিনাক। আমেরন অভিজ্ঞতাক কিছু প্রশ্ন করা
হবে। আমেরন ধুব যোগ্যী অসুস্থ্য থাকলে প্রয়োজনীয় চিকিৎসা
দ্বারা সুস্থ্য হওয়ার সব প্রশ্ন করা হবে। যোগ্যীকাল পরীক্ষার
জন্য কিছু পরিধান পরামর্শ নেওয়া হবে।

আমেরন সুবিধার জন্য উক্তসংগে নিশ্চিত ওয়ুর্ন আমেরন
সংস্কার করতে চেষ্টা করবে। আমেরন সকল ব্যক্তিগত
তথ্য গোপন রাখা হবে। এই জাতিসংঘ সংস্কার কালে আমেরন
কোন বিশেষ সম্ভাবনা নেই বরং আমেরন অনেক সুবিধা
ও যত্ন পাবে।

আমেরন এই স্বাস্থ্যযোগ্যী হওয়া কালে প্রয়োজন করতে
পারবে যে আমেরন চিকিৎসা কোন সর্টি হবে না। আমেরন
স্বাস্থ্যযোগ্যী করতে সর্টি থাকলে নীচে আমেরন স্বাস্থ্যকর সব
চিঠি সহি দিন।

স্বাস্থ্য/চিঠি সহি _____

যোগ্যী নাম _____

যোগ্যী নং _____

কর্মী স্বাস্থ্যকর 3
গণিকা _____