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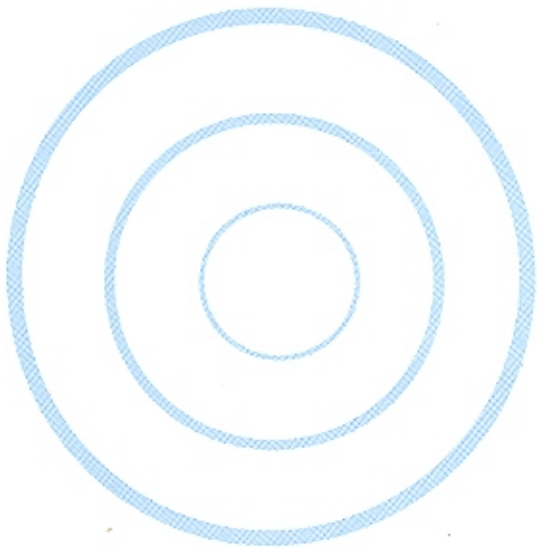

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JOURNAL OF DIARRHOEAL DISEASES RESEARCH

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CONTENTS

Acceptability of Rice-based and Flavoured Glucose-based Oral Rehydration Solutions: a Randomized Controlled Trial

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ABSTRACT

The acceptability of prepackaged rice-based (Oresol-R) and flavoured (Oresol-F) glucose-based oral rehydration salts (ORS) solutions was compared with that of standard glucose-based ORS (Oresol-G) in a randomized field trial. Additionally, it is determined if presenting rice-based ORS as a solution that would help stop diarrhoea (Oresol-K) enhanced its acceptability. A total of 437 non-dehydrated children aged less than five years presenting to health centres with acute diarrhoea were randomly assigned to the three ORS groups. Acceptability was determined by the amounts of ORS consumed at home by children still with diarrhoea on 24- or 48-hour follow-up. The amounts of ORS consumed by children given Oresol-R (54 [95% CL 38-70] mL/kg/24 h) and Oresol-F (47 [24-70]) were similar to the amount of Oresol-G (44 [32-56]). ORS consumption was not affected by the child's age, nutritional status, feeding before the episode, duration of diarrhoea at health centre visit, maternal education and previous ORS use. Informing the caretaker that rice-based ORS would help stop diarrhoea did not lead to increased consumption of the solution (Oresol-R 54 [38-70] mL/kg/24 h; Oresol-K 50 [32-68]). Solution preparation was likewise similar among the treatment groups. Reactions to the different ORS types were generally favourable but did not differ between the groups.

Key words: Oral rehydration solutions; Randomised controlled trials; Rice; Glucose; Diarrhoea

INTRODUCTION

The low proportion of episodes for which oral rehydration salts (ORS) solution is used (1,2) and the inadequate quantities administered for both rehydration and maintenance therapy of diarrhoea (2,3) have been attributed to problems of acceptability of the solution. Frequently reported reasons for dissatisfaction are the failure of ORS to diminish the frequency, volume and duration of diarrhoea and its bad taste (4,5). Two alternatives to the standard formulation, flavoured ORS and one in which pre-cooked rice powder is substituted for glucose address the perceived problem of palatability. In addition, rice-based ORS offers the potential of addressing the desire for an antidiarrhoeal effect (6). The efficiency of rice-based ORS in promoting fluid and salt absorption is attributed to its capacity to release more glucose from rice starch than is present in glucose-based ORS while maintaining low osmolality.

Thus, we compared the acceptability and usage of rice-based (Oresol-R), flavoured (Oresol-F), and standard glucose ORS (Oresol-G) in a randomized field trial. In a second phase, we examined whether the acceptability would be improved when rice-based ORS was presented as a solution which would 'help stop diarrhoea' (Oresol-K).

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MATERIALS AND METHODS

The study was carried out in six government health centres in Metropolitan Manila and conducted during a ten-week period. The study protocol was approved by the Institutional Review Board of the Research Institute for Tropical Medicine.

All ORS formulations used contained 3.5 g/L of sodium chloride, 2.9 g/L of sodium citrate dihydrate, and 1.5 g/L of potassium chloride. Oresol-G and Oresol-F contained 20 g/L of glucose, while the rice-based preparations contained 50 g/L of cooked rice powder. The pineapple flavoured ORS used was selected from three different flavoured solutions in a pre-trial test using the members of the health centre staff as subjects. No chemical clour was added to the solution.

The six study health centres were randomly assigned to give out one type of ORS each week using a computer-generated block randomization schedule. During the first 8 weeks (first phase), randomization was limited to Oresol-G, Oresol-F, and Oresol-R. All six health centres gave out only Oresol-K during the last 2 weeks of the study (second phase).

To ensure that acceptability would not be affected by packaging, all types of ORS were repackaged in aluminum foil in the Philippines. Labels for all solutions were designed to resemble as closely as possible the labeling used on the standard government packet. For rice-based solutions, packet instructions were modified to indicate that the solution should be discarded after 12 instead of 24 hours. On the packet for Oresol-K, the message *'This solution will help stop diarrhoea*' was given in English and in Tagalog.

The study was designed to minimize any interference with the health centre routine. Each week during the first phase, health centres were supplied with packets of the ORS type to be used that week along with information sheets about that particular type of ORS, and all unused packets from the previous week were collected. The designated ORS was provided to all patients consulting for diarrhoea that week whether they were eligible for the study or not. Because rice-based ORS should be discarded after 12 hours, during the weeks when rice-based ORS was the type provided, health workers gave patients four packets instead of two. Caretakers were asked to dissolve ORS in one litre of drinking water as per packet instructions. Caretakers receiving rice-based ORS were reminded to discard the solution after 12 hours and stir it each time it was administered. During the second phase, when Oresol-K was given out at all health centres, health workers were asked to inform caretakers that Oresol-K would help stop the child's diarrhoea. Otherwise, the usual diarrhoea case management procedures were observed following the WHO guidelines and operational procedures as practised by the health workers.

Subjects were non-dehydrated children aged less than five years with acute watery diarrhoea (<7 days) and no previous consultation for the episode. Patients were recruited at the time of discharge, after they had been seen by the health centre staff. A brief questionnaire and checklist was used by a trained field worker to determine the child's eligibility and other symptoms presented in addition to diarrhoea. Patients were visited at home either the next day (24-hour follow-up) or the day after (48-hour follow-up) with alternate patients assigned to each time of follow-up.

During the follow-up visit, information was obtained about the household's socioeconomic and demographic status; the child's current status, including hydration and weight (to the nearest 0.1 kg using a Salter scale); intake of food and liquids, including ORS for the past 24 hours; drugs taken; preparation of Oresol, including the time each batch was prepared and consumed or discarded in the past 24 hours, the amount of water used for preparing the last batch, the number of packets the mother still had, and the caretaker's spontaneous and elicited attitudes about the solution used, other ORS solutions, home-prepared rice water and antidiarrhoeals. In obtaining liquid intake, the caretaker was asked to demonstrate with the household container used the approximate amount of each type of liquid taken by the child. This amount was subsequently transferred to a measuring cup for conversion into mL. For each food type the caretaker gave the amounts consumed per feeding in household measures (e.g. teaspoon, cup). These were then converted to mg using a food conversion table adapted from the WHO guidelines (7).

The following warranted withdrawal of the subjects from the study: 1) development of complications which prevented administration of ORS, and 2) consultation at another health facility after recruitment.

Analysis focused on comparisons of pairs of treatments. The main outcome measure of solution acceptability was the amount of ORS consumed in the 24 hours before the interview. For comparison of intake of ORS, other fluids and solids, only those who continued to have diarrhoea on follow-up (i.e. last abnormal stool passed within 24 hours before interview) were included in the analysis.

Differences between treatments were tested for significance by the chi-square test or the Fisher's exact test for categorical variables, the Student's *t*-test for comparing two group means, and the analysis of variance when more than two group means were involved. Bivariate analysis was employed to determine the factors affecting ORS intake.

RESULTS

Of the 437 children enrolled in the study, 352 were followed up and their caretakers interviewed. No data were collected from those whose house could not be located (58), caretaker was not available (25), or refused interview (2).

There were no differences between treatment groups in baseline characteristics of children and caretakers except for less-educated mothers in the Oresol-F group (Table I). Eighty-seven per cent of the children were already on solids and about one-third were still being breastfed. Twenty-five percent were classified as moderately to severely malnourished (weight-for-age 75% of National Center for Health Statistics median). Close to one-half presented at the health centre with concomitant illness, predominantly respiratory in nature.

Table I. Characteristics of 352 children and caretakers enrolled in the study

Characteristic	Oresol-G (n=105)	Oresol-F (n=76)	Oresol-R (n=94)	Oresol-K (n=77)
Children				
Age (months)	15±13	15±10	14±12	16±13
M/F	59/46	44/32	47/47	41/36
Weight (kg)	8.5±2.5	8.4±2.2	7.8±2.1	8.4±2.7
Malnourished - moderate/severe (%)	23	29	28	20
Feeding before episode (%)				
Breastfed	33	30	29	35
Given milk formula	70	67	77	68
Given solids	91	90	86	83
Other illness (%)	48	49	41	54
Diarrhoea duration at health centre visit(h)	48±39	46±38	45±36	44±35
No diarrhoea on follow-up (%)	29	21	24	36
Caretakers				
Age (year)	29±10	30±10	29±9	29±7
% working	18	18	18	16
% less than high school education	50	68	47	52
Household monthly income (x 1,000 pesos)	2.0±1.2	1.8±1.3	1.8±0.9	1.8±1.4
% previous ORS use	53	59	55	55

Plus-minus values are means ?1 S.D. Except for the proportion of caretakers with less than a high school education in the flavoured ORS group ($p<0.03$ by chi-square), there were no significant differences among groups

Two hundred eighteen children still had diarrhoea on follow-up. No child was dehydrated, had developed complications or consulted at another health facility. Table II summarizes the 24-hour intake of foods and fluids during diarrhoea. Total intake of ORS as well as total fluids and solids was comparable between treatment groups. The child's age, nutritional status, feeding before the episode, duration of diarrhoea at health centre visit, concomitant illness, and maternal characteristics (including education and previous ORS use) did not significantly affect ORS consumption.

Table II. Fluid intake and consumption of rice during diarrhoea

Variable	Oresol-G	Oresol-F	Oresol-R	Oresol-K
Fluids				
No. with diarrhoea during 24 h prior to home visit	67	49	60	42
Total fluid intake (mL/kg/24 h)*	190(161-219)	172(132-212)	177(141-213)	165(133-197)
ORS intake (mL/kg/24 h)*	44(32-56)	47(24-70)	54(38-70)	50(32-68)
% with no ORS intake	13	20	13	21
Rice				
No. on solids before episode	65	43	51	35
Rice gruel consumed (g/kg/24 h)*	11(5-17)	13(5-21)	11(6-16)	9(0-18)
% with no intake	58	60	61	57
Boiled rice consumed (g/kg/24 h)*	6(3-9)	5(2-8)	7(4-10)	5(3-7)
% with no intake	40	38	46	37

*Mean (95% CL); values include those with no intake. These were no differences in amounts of fluid or rice consumed among groups (p>0.05, by t-test for two independent samples: G vs. F, R or K; R vs. K).

Table III. Solution preparation in each treatment group*

	Oresol-G (n=94)	Oresol-F (n=71)	Oresol-R (n=86)	Oresol-K (n=72)
% added sugar	3.2	2.8	0	2.8
% added salt	0	0	0	1.4
% used other liquid	2.2	0	0	0
% used <1 packet	4.3	0	11.6	5.6
Volume of water used (mL)				
for those who used 1 packet	984±189	974±183	1032±164	990±155
Minimum	530	440	500	660
Maximum	1700	1540	1700	1660

* There were no significant differences between groups. Plus minus values are means ± s.d.

ORS solution was prepared by 323 (92%) of the 352 caretakers followed up. No significant differences were observed in the amounts of water used (mean 959 ±233 mL) and in the number of caretakers who added sugar to the solutions among treatment groups (Table III). Although more caretakers used less than 1 packet in the Oresol-R and Oresol-K groups, this difference was not significant. Only one (in the Oresol-K group) added salt to the solution.

Although few children (3.4%) took antidiarrhoeals, antimicrobials and other drugs were frequently prescribed at the health centre. Forty-three per cent received no medicines at home with no difference observed among treatment groups. Children with diarrhoea only were less likely to be given drugs than those presenting with concomitant illness (46% vs. 69%; p<0.0001).

Caretakers' reactions to the different types of ORS given were similar and were generally favourable. About half of the 322 mothers who prepared the solution perceived it to taste good (49%), lessen weakness (59%), and stop diarrhoea (51%); more than two-thirds would use it again. The lack of difference in attitudes was maintained whether or not the caretaker had previously used ORS.

DISCUSSION

Our findings indicate that acceptability, as measured by the child's intake and the caretaker's reports, was not significantly greater for rice-based or flavoured ORS than for standard glucose ORS. Moreover, telling mothers that rice-based solution would help stop diarrhoea did not enhance acceptability. In children with mild non-dehydrating diarrhoea, such as those enrolled in this study, this benefit would be unlikely to be noticeable. Clinical trials have shown that the effect of rice-based ORS in lowering stool volume and shortening the duration of diarrhoea is greatest in rapidly purging patients, such as those with cholera (6,8).

What is interesting is that mothers did not perceive Oresol-K (rice-based ORS presented as a solution that would 'help stop diarrhoea') as having such an effect, a characteristic readily attributed to proprietary antidiarrhoeals with no objectively noticeable effect (9). While health workers making a special effort to emphasize this characteristic might have predisposed caretakers to perceive it, that would have violated the study requirement of realistic clinic conditions.

It is also important to note that this study measured the relative acceptability of different types of ORS given to mothers in the context of a consultation at a clinic. For mothers buying ORS over-the-counter, a product that would 'help stop diarrhoea' or that was flavoured could be more attractive than standard ORS. That possibility was not tested in this study.

Only two other studies, both conducted in Bangladesh, have examined the acceptability of packaged rice-based ORS as home treatment of diarrhoea (10,11). Those studies differed from the one reported here in several important ways: the packets of both rice-based and standard ORS were distributed to mothers in the village; the packets used required cooking of the powdered rice; and acceptability was inferred from simple usage rates and number of ORS packets used without reporting estimates of ORS intake. Usage rate was higher for rice-ORS (71%) than for glucose-ORS (60%) in the first study, but the second more recent trial showed that glucose-ORS was preferred (10,11).

One of the concerns of promoting rice-based ORS, particularly in areas where rice is a weaning food, is that mothers may view the solution as food and, consequently, reduce feeding during diarrhoea (12). This was not observed in the study. Likewise, the potential problem of using more water in preparing rice ORS, which has a greater bulk of dry ingredients than glucose ORS, was not encountered. Flavoured ORS was not perceived to be more palatable; flavouring did not lead to over-consumption of the solution and its dreaded complications. This may partly be due to the finding that comparable numbers regarded the various ORS types as tasty, i.e. Oresol-F was not necessarily perceived to be superior in terms of palatability. Moreover, among those with no prior history of ORS use, flavoured ORS was least perceived to stop/help cure diarrhoea. Only one study has documented the increased consumption of flavoured (vs. non-flavoured) ORS leading to hypernatermia and overhydration, but this involved sicker children in a hospital setting (13).

While mothers did not experience difficulties in preparing and administering the solutions nor substitute rice-based ORS for food, our measure of acceptability – home consumption of solutions received at the clinic – shows no clear advantage of rice-based and flavoured solutions over standard glucose ORS. This observation lends support to the continued recommendation of glucose ORS as standard therapy.

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Human Colostrum IgA Antibodies Reacting to Enteropathogenic *Escherichia coli* Antigens and Their Persistence in the Faeces of a Breastfed Infant

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ABSTRACT

IgA antibodies reacting to enteropathogenic *Escherichia coli* (EPEC) antigens in human colostrum and their role in the inhibition of EPEC adherence to HEp-2 cells were studied. Colostrum IgA was isolated with a Sepharose anti-IgA column. IgA-depleted colostrum lost its inhibitory effect on EPEC adhesion, while the IgA-enriched eluate was a potent adherence inhibitor. The same eluate showed a significant loss of inhibitory activity after absorption with an EPEC strain showing localised adherence (LA+), but no alteration after absorption with an LA- strain. No bands were observed in Western blot analysis with LA+ absorbed eluate and with a crude extract of the EPEC strain, but the eluate absorbed with LA- showed a strong recognition of a 94-kDa band, a molecular weight equivalent to that of intimin. Colostrum antibodies reacting to non-protein antigens were not detected by Western blot analysis. The persistence of anti-EPEC IgA in the gastrointestinal tract was shown by the strong reactivity to the 94-kDa band in Western blot analysis of one mother's colostrum and her infant's faeces. These data confirm the role of colostrum antibodies in protecting the neonate against infections due to EPEC.

***Key words*:** IgA; Bacterial adhesion; Breast feeding; *Escherichia coli,* Enteropathogenic; Diarrhoea, Infantile; Colostrum; Antibodies, Bacterial; Antigens, Bacterial; Immunity

INTRODUCTION

Enteropathogenic *Escherichia coli* (EPEC) is one of the principal agents of acute diarrhoea in infants aged up to one year in developing countries (1, 2). A three-stage model has been proposed to explain the pathogenesis of EPEC infections (3): 1. Formation of bacterial clusters on the epithelial cell surface; 2. Transduction of a signal to the cell resulting in the activation of its tyrosine kinase activity and increased intracellular calcium concentrations; 3. Close attachment of the bacteria to the membrane of the epithelial cell, with damage to the microvilli and accumulation of cytoskeletal proteins beneath the adhering bacterial cells. The second and the third stages are referred to as attaching and effacing (A/E). Subsequently, some of the adhering bacteria may invade the intestinal cell (4, 5), a 32 kDa protein being involved in this process (5). The initial, or localised adherence (LA) of the organism to the epithelial cell depends on a 50 to 70 kb plasmid (6,7). The adhesive property encoded in this plasmid has been designated the EPEC adhesive factor (EAF) (8). A 94-kDa outer-membrane protein, referred to as intimin, is involved in the A/E effect (9). Intimin is coded by chromosomal genes *(eaeA)* controlled by plasmid genes. Surface structures of 18 kDa in EPEC strains, named bundle-forming pilus (BFP), mediate LA (I0).

During the first six months of life, breastfeeding protects infants against the most important enteric pathogens (11,12), including EPEC (13,14). Both secretory IgA (15,16) and oligosaccharides (15) present in breast milk inhibit EPEC adherence to HeLa and HEp-2 cells. Cravioto *et al*. (15) and Camara *et al*. (16) have described colostrum antibodies reacting to the EPEC 94-kDa outer-membrane protein.

According to Davidson and Lonnerdal (17), human milk proteins are present in the faeces of breastfed infants. These infants excrete significant amounts of secretory IgA, suggesting that it is protected against degradation during its passage through the gut.

The aim of this study was to analyse the presence in human colostrum of IgA antibodies reacting to EPEC antigens, their role in the inhibition of EPEC adherence to HEp-2 cells and their persistence in the faeces of breastfed infants.

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MATERIALS AND METHODS

Colostrum collection and preparation of IgA-enriched eluate

After informed consent, colostrum specimens were collected from 46 women who had given birth to normal term babies at the University Hospital, São Paulo. Colostrum was collected up to 72 hours postpartum and skimmed by centrifugation at 2,000 G. The aqueous phase was stored at –20 ºC until assayed. Equal volumes (1 mL) of colostrum samples were pooled and used for all procedures. An 8-mL aliquot of the pooled colostrum was decaseinated with acetic acid, centrifuged for 10 minutes at 3,000 G, and then depleted of IgA in an anti-IgA Sepharose column. CNBr-activated Sepharose (Sigma) was linked to purified anti-human IgA (a chain specific) (18). The pooled colostrum was applied to the anti-human IgA Sepharose column to obtain IgA-depleted colostrum. IgA was eluted from the column with glycine-HCI buffer. The initial volume of all materials was restored after affinity chromatography to maintain the original concentration.

Collection of one infant's faeces, serum and saliva.

With the agreement of one of the participating mothers, faeces from her full-term, appropriate-for-gestational age infant were collected 72 hours after the child started to breastfeed. The faeces were soaked in phosphate-buffered saline (PBS) to extract soluble proteins. The extract was then centrifuged and the supernatant was inactivated at 56 ºC for 15 minutes and stored at –70 ºC until analysis. Samples of serum, saliva and faeces of another infant, with acute diarrhoea due to EPEC, as well as pooled normal adult sera, were included as controls for Western blot analysis. Saliva was centrifuged and stored at –70 ºC; serum samples were maintained at –20 ºC until use.

Absorption of column eluate with EPEC strains

The role of specific antibodies against EPEC adhesins was studied by the absorption of column eluate with an adherent (LA+) EPEC strain serotype 0111:H^- 0041-1/85 (LA+, *eaeA*+, EAF+, *bfp*+) as described by Pál *et al.* (19). The control experiment was the absorption of column eluate with a non-adherent (LA-) EPEC strain 606/54 (LA-, *bfp*-) of the same serotype.

Adhesion assays

Original, decaseinated and IgA-depleted colostrum, as well as the eluate before and after absorption with LA+ and LA- bacterial strains were submitted to adherence assays as described by Silva and Giampaglia (20). The adherence-inhibiting capacity of colostrum samples was evaluated using *E. coli* strain 0041-1/85. Briefly, HEp-2 cells (ATCC-CCL 23) were grown on Lab-Tek eight-chambered tissue culture slides

(Nunc, Inc, Naperville, ILL.) in Dulbecco's modified Eagle medium (DMEM) with 10% fetal calf serum for 48 hours at 37 ºC. For the test, each chamber was inoculated in duplicate with a 400 µL aliquot of the following suspension: 300 µL colostrum or derivatives, 200 µL bacterial exponential culture and 320 µL culture medium (DMEM with 2% fetal calf serum and 1% D-mannose). After 30 minutes incubation at 37 ºC, monolayers were washed 6 times in PBS and fresh DMEM was added, with a subsequent 3 hours incubation at 37 ºC. Cells were then washed 4 times in PBS, fixed with methanol, stained for 5 minutes with May-Grünwald and 20 minutes with Giemsa (12), mounted with coverslips and Entellan (Merck), and observed under the light microscope at magnification 1,000. At least 300 cells were observed in each experiment.

The colostrum inhibitory effect was determined by calculating the percentage of HEp-2 cells with bacterial clusters on their surface as compared to the controls.

Total IgA determination

All materials were submitted to total IgA determination by the quantitative radial immunodiffusion assay (21) using anti-human a chain antibody on agarose plates and standard colostrum, kindly provided by Professor L.A. Hanson, University of Göteborg, Sweden.

Western blot analysis

The specificity of IgA antibodies reactive to EPEC adhesion-related structures was studied by Western blot analysis (22). EPEC strains 0041-1/85 (LA+, *eaeA+, EAF+, bfp+)* and 606/54 (LA-, *bfp*-) were grown on blood agar plates to provide BFP expression (10). The overnight growth was collected from each agar plate in 5 mL saline, centrifuged for 10 minutes at 3,500 G and resuspended with saline to an optical density of 0.5 at 420 nm. This suspension was centrifuged in Eppendorf tubes at 11,700 G in a Spin 1 centrifuge for 5 minutes. The pellet was resuspended in 50 µL of sample buffer with 2% SDS, 3 µL b - mercaptoethanol and 2 µL bromophenol blue, and boiled for 10 minutes. Bacterial components were separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) in 10% polyacrylamide gels. Molecular weight standards (Pharmacia, Upsala, Sweden) were run in each gel. After transferring to nitro-cellulose membranes, strips were incubated overnight with one of these materials: 1:100 dilution of colostrum samples, 1:10 dilution of faeces extracts, 1:5 dilution of saliva or 1:50 dilution of sera, and afterwards for 2 hours with a 1:500 dilution of anti-human IgA peroxidase conjugate for colostrum, saliva or faeces and anti-immunoglobulins polyvalent peroxidase conjugate for sera. The reaction was developed with the substrate in 3,3'-diaminobenzidine (DAB) (Sigma).

Proteinase K treatment

To investigate the nature of the bacterial antigens recognised by the colostrum antibodies, a crude extract of *E. coli* 0041-1/85 was submitted to proteinase K treatment to confirm the protein constitution of bacterial antigens observed in SDS-PAGE. This method, described by Hitchcock and Brown (23), eliminates proteins while leaving intact lipopolysaccharide or polysaccharide components such as O antigens. Bacterial extracts were run in duplicate for protein identification with Coomassie blue or silver staining.

RESULTS

Figure 1 shows the results obtained in adherence assays carried out in the presence of the pooled colostrum samples, decaseinated and IgA-depleted colostrum and eluate, before and after absorption with LA+ and LA- EPEC strains. IgA-depleted colostrum lost its inhibitory effect on EPEC adherence to HEp-2 cells, while the IgA-enriched eluate was a potent adherence inhibitor. After absorption with LA+ strain, the eluate showed a significant loss of inhibitory activity, while absorption with the LA- strain did not significantly alter its inhibitory effect.

Colostrum lost 99.2% of its original IgA concentration (from 17.13 g/L to 0.13 g/L in IgA-depleted colostrum), showing that the affinity column efficiently adsorbs IgA from colostrum.

In Western blot analysis, original colostrum pool and IgA-enriched eluate showed several bands of different molecular weight. A strong reaction was observed with a 94-kDa band, a molecular weight equivalent to that of intimin, a protein related to the A/E effect. A strong reaction was also observed with a ~35-kDa band. IgA depleted colostrum did not show the 94-kDa band, but the ~35-kDa band was still present. No bands were observed in LA+ absorbed eluate, while the eluate absorbed with LA- strain showed a good recognition of the 94-kDa band (Fig. 2).

Colostrum and pooled normal adult sera showed a similar reaction pattern in Western blot analysis (Fig. 3). Many bands between <20 kDa and 94 kDa were observed in both materials. No reaction with the 94-kDa band was observed when serum, saliva, and faeces from an infant with diarrhoea due to EPEC were assayed. Saliva and faeces from the same infant showed a poor reaction, with bands of ~20 and ~30 kDa. The 94-kDa band was recognised by IgA antibodies in the faeces extract of the healthy neonate and in his mother's colostrum. Antibodies reactive to non-protein antigens were not observed in Western blot analysis performed with the same materials and proteinase K-treated antigens (Fig. 3).

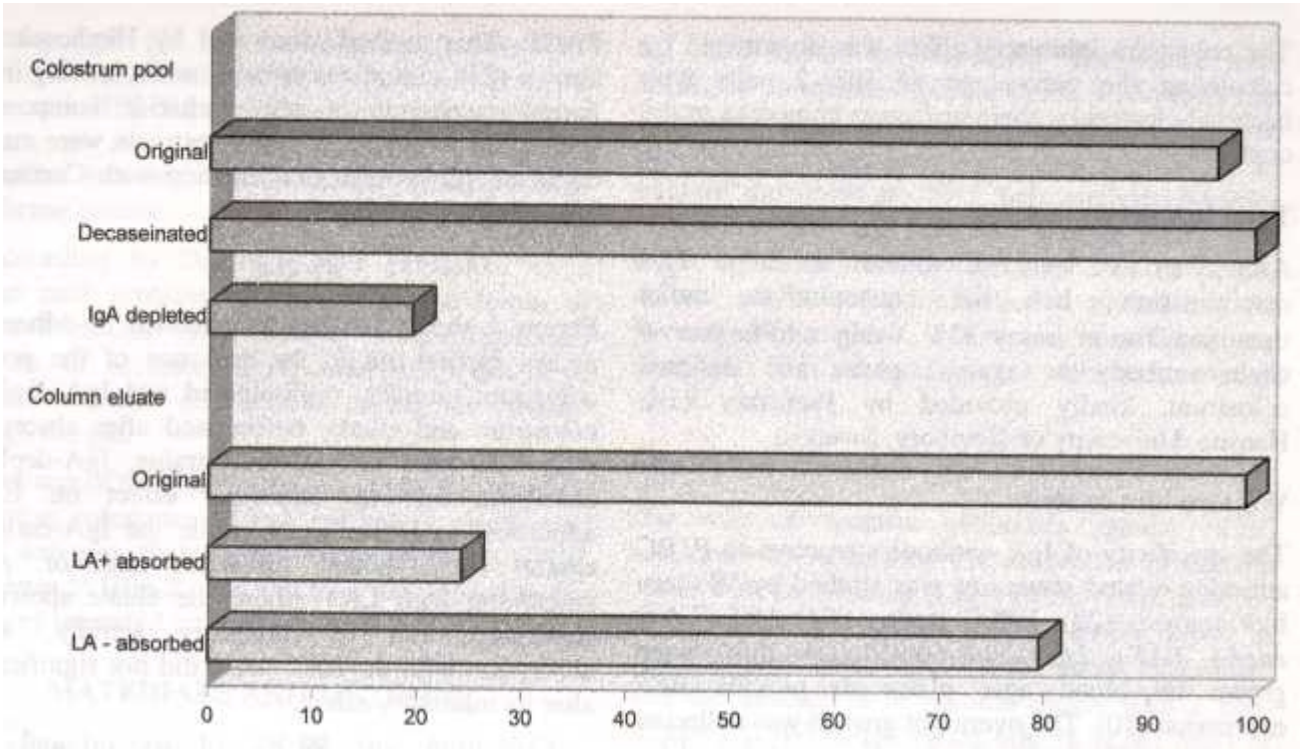

Fig. 1: Adherence inhibition of EPEC in assays carried out in the presence of a pool of colostrum samples, decaseinated and IgA-depleted colostrum, and column eluate before and after absorption with EPEC strains showing localised adherence (LA+) or not (LA-).

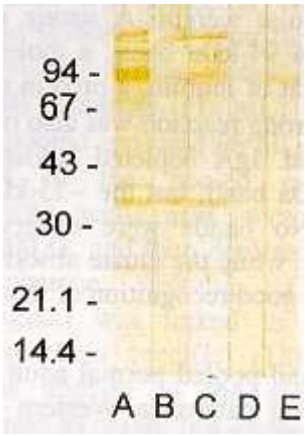

Fig. 2: Western blot of pooled colostrum samples (lane A), IgA-depleted colostrum (lane B), Sepharose anti-IgA eluate (lane C), eluate absorbed with E. coli showing localised adherence (LA+) (lane D), and eluate absorbed with E. coli LA- (lane E). The reaction was revealed by an anti-human (? chain)-peroxidase conjugate. The molecular-weight scale is given on the left.

adherence to HEp-2 cells (15,16). This is confirmed in the present study:IgA-depleted colostrum had no inhibitory effect on EPEC adhesion, while the LgA-enriched eluate was a potent adherence inhibitor as the orginal colostrum.

Fig. 3: Western blot of pooled colostrum samples (lanes A and a), pooled normal adult sera (lanes B and b), serum, saliva and faeces extract of a sick child (lanes C, D and E respectively and c, d and e), faeces extract of a healthy neonate (lanes F and f) and his mother's colostrum (lanes G and g). Antigen used is a crude extract of Escherichia coli showing localised adherence (lanes A to G) and the same materials after treatment with proteinase K (lanes a to g). The molecular-weight scale is given on the left.

DISCUSSION

Colostrum antibodies have been described as important factors acting on the inhibition of EPEC adherence to HEp-2 cells (15,16). This is confirmed in the present study: IgA-depleted colostrum had no inhibitory effect on EPEC adhesion, while the IgA-enriched eluate was a potent adherence inhibitor as the original colostrum.

Milk oligosaccharides are known to participate in the protection against infections due to *Streptococcus pneumoniae* and *Haemophilus influenzae* (24). The role of oligosaccharides in EPEC adherence inhibition was not observed in this work: IgA-depleted colostrum containing low molecular weight components did not show this effect. Previous laboratory experiments (16) showed no EPEC adherence inhibition by a low molecular weight colostrum fraction. However, Cravioto *et al*. (15) described such an effect when using an enriched fraction of milk oligosaccharides.

Colostrum and milk anti-intimin antibodies are involved in EPEC adherence inhibition (15,16). In the search for antibodies against other bacterial proteins involved in EPEC adhesion, IgA-enriched eluate was absorbed with EPEC strains whether presenting localised adherence or not. Adherence assays with IgA-enriched eluate absorbed with LA+ strain presented a significant decrease in the original inhibitory activity, showing that antibodies related to EPEC adhesion were withdrawn by this process.

Antibodies related to EPEC adhesion were distinguished after absorption of IgA-enriched eluate with the LA- EPEC strain. No decrease in the inhibitory levels was observed, indicating that antibodies against EPEC adhesins were present in this preparation. Western blot analysis of this absorbed eluate showed a strong 94-kDa band, a molecular weight equivalent to that of intimin (9), the protein involved in EPEC adherence.

Western blot analysis of the original colostrum and the IgA-enriched eluate showed reactivity to ~20 to ~94-kDa bands, confirming that several EPEC proteins are recognised by colostrum antibodies. The ~35-kDa band recognised in all materials except absorbed eluates could be the 32-kDa protein recently

described as related to EPEC cell invasion (5), or the product of the espB gene, a secreted protein involved in the signal transduction occurring in A/E lesions on the epithelial cells (25). Nevertheless, antibodies binding to this protein were absorbed with the LA- strain, showing that this antigen is not exclusively present in pathogenic strains.

Two faint bands of <20 kDa were recognised in some of the materials analysed, such as colostrum and serum samples. One of them could be related to BFP subunits. To confirm this hypothesis, it would be necessary to improve the Western blot's ability to detect low molecular weight proteins. To verify if any of these bands were due to reactivity to non-protein antigens, the same assay was performed with proteinase K-treated antigen. No antibodies reacting to non-protein antigens were detected.

Serum from the infant with diarrhoea showed a different Western blot pattern compared to normal adult pooled sera, with no reaction to the 94-kDa protein. These results are in keeping with those of Levine *et al*. (8) in adult volunteers who ingested a suspension of *E coli* O127:H6. One volunteer who failed to develop diarrhoea had detectable serum antibodies to the protein before the challenge. Faeces from the same child did not present the 94-kDa band. This infant was not receiving breastfeeding, and so anti-intimin antibodies were not detected in Western blot analysis. Laboratory studies are underway comparing the antibody repertoire in healthy and sick children with EPEC diarrhoea.

Western blot of a mother's colostrum and the PBS extract of her infant's faeces also recognised the 94-kDa band. Davidson and Lönnerdal already found some human milk proteins passing intact through the gastrointestinal tract of the breastfed infant (17). In this work, we describe for the first time the presence of anti-intimin immunoglobulins in an infant’s faeces and their effect on EPEC adhesion to cultured cells.

The present study suggests that colostrum antibodies against EPEC adhesins go through the neonate’s gut maintaining their antigenic reactivity and perhaps their inhibitory capacity of bacterial adherence. This would explain the protective role of colostrum antibodies against infections.

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Sequential Changes in Gut Mucosa of Rabbits Infected with Vibrio cholerae O139 Bengal: an Ultrastructural Study

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ABSTRACT

Adhesion and subsequent colonisation are important events in the infection by *Vibrio cholerae* O139 Bengal. To determine in details the pathological changes in the gut mucosa, an epidemic strain of O139 Bengal was inoculated in a rabbit ileal loop model. Electron microscopic studies were done at different time intervals after inoculation of the strain to see the histological changes at the ultrastructural level. From 10 hours onwards, cellular invasive processes with presence of bacteria in the lamina propria and other associated inflammatory changes were revealed.

Key words*:* Cholera; *Vibrio cholerae;* Virulence; Disease models, Animal

INTRODUCTION

In 1992, a new serogroup of Vibrio cholerae emerged in the Indian subcontinent. It had epidemic as well as pandemic potentials and was classified as V. cholerae O139 Bengal (1). Although V. cholerae O139 has remarkable phenotypic and genotypic similarities with V. cholerae O1 of the El Tor biotype, the O139 differs from the O1 serogroup in important ways (2). These include (i) non-agglutinability of the O139 serogroup with O1 antisera, (ii) the O antigen of the O139 serogroup contains colitose, a sugar not associated with the lipopolysaccharides of other vibrios and (iii) the presence of a capsular polysaccharide among the O139 strains.

V. cholerae O1, the causative agent of cholera, after ingestion, reaches the small intestine, penetrates the mucus layer and adheres to the surface of the epithelial cells (3). The mechanism of the attachment of the vibrios to intestinal cells includes bacterial motility, chemotaxis, elaboration of fimbrial or pilus structures to initiate the intestinal attachment and subsequent colonisation process (4-7).

Little is known about how the V. cholerae non-O1, especially the virulent epidemic O139 strains, interact with the intestinal mucosa, how far they invade it, and the histopathological alterations they produce. To have a clear idea of these, sequential ultrastructural changes were studied by electron microscopy at given time periods after introduction of the novel epidemic strain O139 Bengal in a rabbit ileal loop model.

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MATERIALS & METHODS

Bacterial strain

V. cholerae O139 Bengal SG24 strain, isolated from a human patient in December 1992, was used throughout the study.

Experimental animals

New Zealand white rabbits of either sex, 6 to 8 weeks of age weighing 1.75 to 2 kg were used. They were treated with metronidazole (125mg/day) and sulphaquinoxaline (464 mg/day) for 5 days, the course being repeated three times with an interval of 2 days to clear the animals of Giardia and Coccidia.

Culture conditions

The SG24 strain was inoculated in 2 mL of tryptic soy broth (TSB; Difco, USA), and incubated at 37 ° C in a shaker bath (100 rpm) for 4 hours. An aliquot of the starter culture was added to 25 mL of TSB in a 500 mL Erlenmeyer flask. After 18 hours of growth, the culture was harvested by centrifugation at 8000 rpm for 15 minutes. The pellet was suspended in sterile phosphate buffered saline (PBS), pH 7.4. The bacterial density was estimated at 540 nm and diluted in PBS to attain an optical density of approximately 109 cells/mL for use as inoculum. The final inoculum was kept on ice until inoculated into the experimental rabbits. Serial 10-fold dilution of the inocula was plated before and after challenge for quantitative counts to confirm the challenge dose.

Experimental animals

Rabbits were fasted for 24 hours prior to surgery, except that they received water ad libitum. Anaesthesia was maintained with ketamine (35 mg/kg body weight.). After 6 minutes, 5 mL of 2% xylocaine was given subcutaneously along the linea alba. A 4 to 5 cm mid line incision was made observing proper aseptic precautions. The ileocaecal junction was identified and 3 blind loops were made, approximately 3 cm long and 1 cm apart. Two loops were inoculated with 0.1 mL of PBS containing 109 log-phase broth culture of V. cholerae O139 while the third loop, which served as a control, received 0.1 mL of sterile PBS. The abdominal cavity was closed by suturing. The animals were then kept in warm cages. At 2, 6, 10, 14 and 18 hours' intervals, the rabbits were sacrificed by injecting intravenously 2 mL of Euthansia 6 solution. For each time interval three rabbits were used.

Fixation and processing of intestinal tissue for electron microscopy

After sacrificing the rabbits at the required time interval, the intestinal loops were removed. The loops were opened longitudinally, washed in cold PBS and fixed in 3% chilled cacodylate buffered glutaraldehyde, post-fixed in 1% osmium tetroxide, treated with graded series of ethanol and propylene oxide and finally embedded in Agar 100 resin (Agar Aids, UK). Ultrathin sections were made using a LKB ultramicrotome Nova and stained with uranyl acetate and lead citrate. Sections were examined with Philips EM 201C and 420 T transmission electron microscopes.

Fig. 1: Transmission electron micrograph showing normal villus architecture with uniform microvillus brush border. No inflammatory changes were noted in a control loop; 6 hours after challenge. Bar = 2 µm

RESULTS

At all time intervals, the control loops showed normal villus architecture with uniform microvillus brush border and no inflammatory changes (Fig. 1). As to the loops inoculated with V. cholerae, at 2 hours after inoculation, bacteria were seen in the lumen (Fig. 2). No junctional changes were observed but it appeared that the bacteria were attempting to invade the intercellular junctions. At 6 hours after inoculation, desquamated epithelial cells were noted in the intestinal lumen along with bacteria, with many vibrios being phagocytosed by neutrophils and macrophages (Fig. 3). Many polymorphonuclear leukocytes were seen in the lamina propria. After 10 hours, bacteria were seen along with inflammatory cells in the lamina propria (Fig. 4) and after 14 and 18 hours, there was shortening and denudation of the villi, bacteria were seen in the crypts and inside the capillary and the lamina propria (Fig. 5 & 6), with considerable crypt degeneration and vascular congestion.

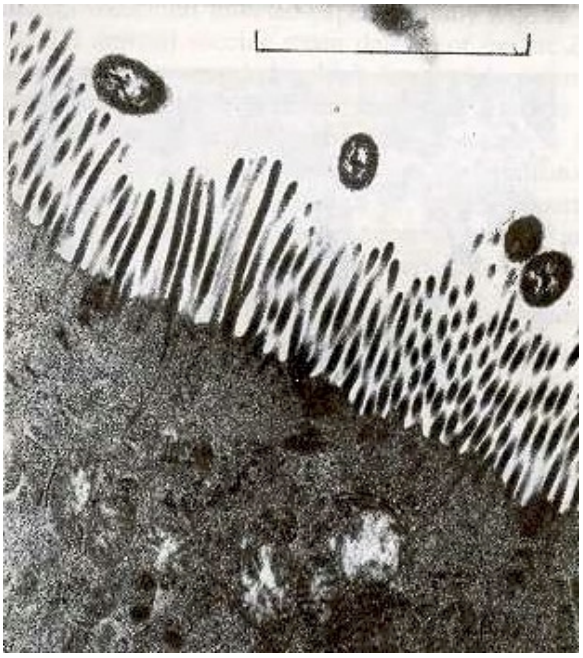

Fig. 2: Transmission electron micrograph showing O139 Bengal (strain SG24) on the ileal epithelial surface; 2 hours after challenge. Bar = 2 µm

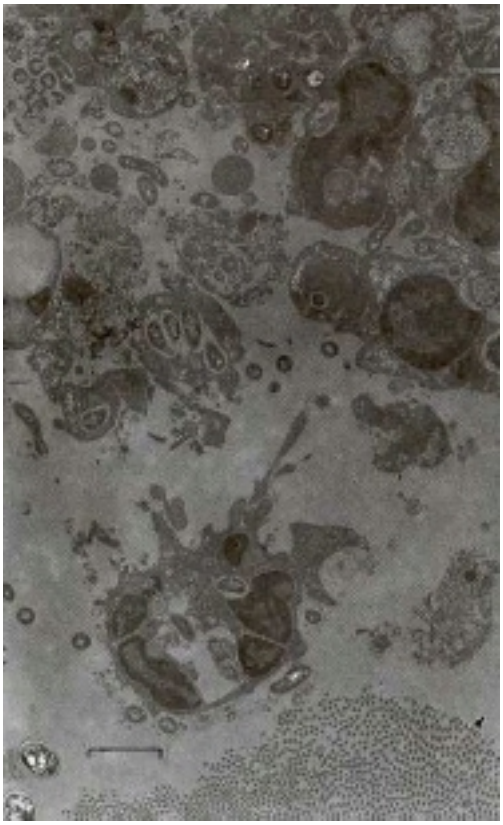

Fig. 3: Polymorphous cholera vibrios (O139 strain SG24) on the microvillus surface and in the lumen. Degenerated cells along with bacteria phagocytosed by neutrophils are seen 6 hours after challenge Bar = 2 µm

DISCUSSION

The strain used in this study was the novel epidemic serogroup classified as V. cholerae O139 Bengal in which the virulence genes comprising the virulence cassette of V. cholerae were intact. Intestinal adhesion and colonisation are important steps in the infection of the gut by V. cholerae O139 (9,10).

Previous reports on rabbit ileal loop experiment by Polotsky et al. in 1977 (11), have documented that V. cholerae O1 El Tor vibrios showed attachment onto intestinal enterocytes, but without destruction of the epithelium or marked inflammatory changes. The study of experimental canine cholera by Elliott et al. (12) also showed absence of acute inflammation in the production of cholera fluid by the gut. Acute inflammatory cells were reported to be either lacking in rabbits with vibrio-induced or toxin-induced cholera, or present only transiently in small numbers. Also, mucosal capillaries showed no damage when examined under the electron microscope (13-16).

Fewer studies have been done so far about the pathogenic effects of virulent strains of V. cholerae O139. In 1990, Panigrahi et al., using an in vitro model with Caco-2 cells infected with V. cholerae non-O1 NRT 36S (serogroup O31), demonstrated adherence of the bacteria to Caco-2 cells and damage to the microvilli (17). Russell et al, in 1992 documented epithelial damage and presence of inflammatory cells in the lamina propria in rabbits infected with V. cholerae non-O1 NRT 36S (serogroup 031) in the RITARD model (18). Another study by the oral colonisation model of rabbits with O139 Bengal (19) demonstrated proliferation of the bacteria in the small intestine and colonisation of its mucosal surface, along with disruption of the epithelial cells' apical membranes. The present study agrees with all these findings.

Presence of bacteria on the brush border, or located close to them in the lumen, bacterial polymorphism, oedema of lamina propria, as identified by Polotsky et al. with V. cholerae El Tor corroborates our results.

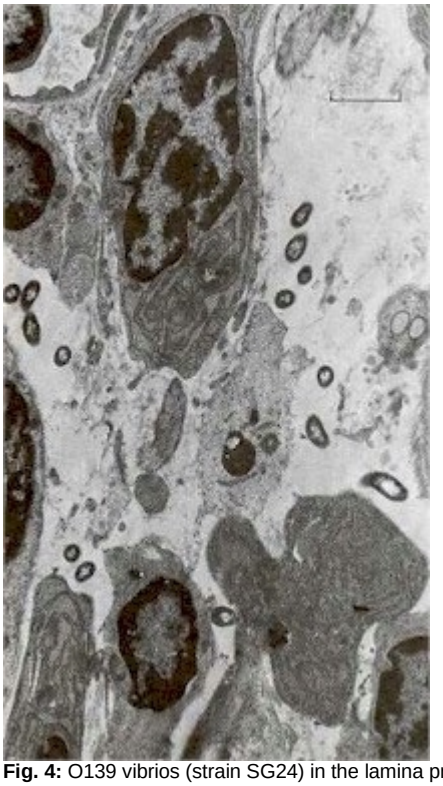

Fig. 4: O139 vibrios (strain SG24) in the lamina propria along with few plasma cells; 10 hours after challenge. Bar = 2µm

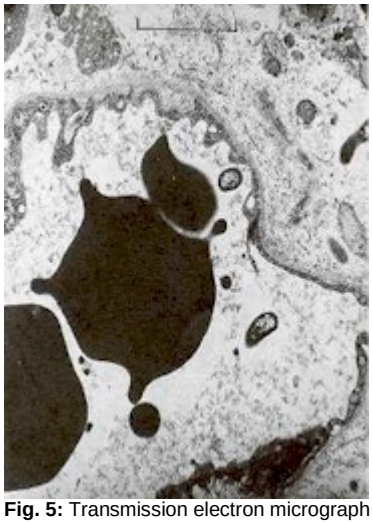

Fig. 5: Transmission electron micrograph showing bacterial strain SG24 inside a distended capillary lumen; 14 hours after challenge. Bar = 2 µm

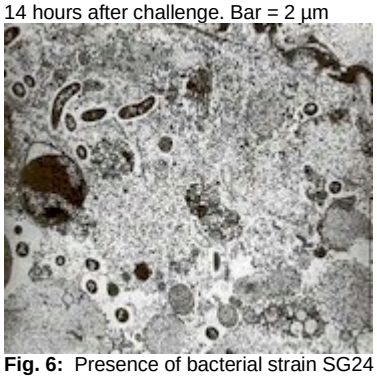

Fig. 6: Presence of bacterial strain SG24 in the lamina propria; 14 hours after challenge. Bar = 2 ?m

In addition, our virulent strain of V. cholerae O139 caused some more changes. Indeed, we found as early as 6 hours after challenge desquamated epithelial cells in the intestinal lumen, along with bacteria and polymorphonuclear neutrophils. There also were many neutrophils in the lamina propria. Bacteria in the oedematous lamina propria were seen 10 hours after inoculation. Later, along with these changes, there was marked spotty denudation of the villi, congested blood vessels, degenerative changes in the cells, with bacteria in the lamina propria and inside the capillary lumina.

The secretory response to bacterial toxin is a complicated event. Recent evidence suggests that cholera toxin may stimulate epithelial cells to secrete IL-6, a pro-inflammatory cytokine. This cytokine in turn, may stimulate the submucosa, leading to the production of inflammatory cells with local production of mediators, such as prostaglandin and leukotrienes. These, in turn, stimulate the fluid secretion by the epithelial cells (20,21). Duodenal biopsies from patients infected with V. cholerae O1 revealed only modest inflammation (22). Stools of human volunteers infected experimentally with an El Tor O1 derived vaccine strain deleted of the ctx Zot and ace genes revealed a high level of lactoferrin (24) that is normally found in secondary granules of polymorphonuclear leukocytes. The mechanism by which this strain stimulates an inflammatory response is unknown, though it may be partly responsible for the residual reactogenicity and diarrhoea observed in these volunteers (25).

Gastrointestinal symptoms and diarrhoea caused by V. cholerae O139 are very similar to that of cholera caused by the O1 serogroup, the exception being that O139 infected patients appeared to complain of abdominal pain more frequently than O1 infected patients. Our present study with O139 Bengal in a rabbit ileal loop model shows clear-cut histological and ultrastructural changes in the intestinal mucosa, with cell invasion and presence of bacteria and inflammatory cells in the lamina propria. Further studies with several other mutants having defined altered virulence properties are likely to help us to better understand the complex pathophysiology of choleric diarrhoea and improve our knowledge regarding the management of cholera-like diseases.

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Detection of Non-culturable Shigella dysenteriae 1 from Artificially Contaminated Volunteers' Fingers Using Fluorescent Antibody and PCR Techniques

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SUMMARY

Epidemiological studies have demonstrated that hands may be an important vehicle for transmission of shigellosis. The present study was carried out to find out the survival potential of *Shigella dysenteriae* 1 on fingers of volunteers. Finger surface was inoculated with 10^5 cfu of *S. dysenteriae* 1 and then the bacteria were detected using conventional culture, PCR and fluorescent antibody (FA) techniques after different time intervals. It was found that *S. dysenteriae* 1 survived for up to one hour in culturable form but up to four hours in non-culturable form on human fingers. The non-culturable *S. dysenteriae* was detected by PCR and FA techniques. This study elaborates on the role that fingers have in the transmission of shigellae.

Key words: Dysentery, Bacillary; *Shigella dysenteriae;* Disease transmission; Polymerase chain reaction; Flourescent antibody technique

INTRODUCTION

Shigellosis is mainly associated with low levels of personal hygiene and sanitation (14). Shigellosis is hyperendemic in some parts of Bangladesh, and occasionally flares into epidemics. In routine admissions in a diarrhoeal disease hospital in Bangladesh, the isolation rate of shigellae is approximately 11 to 12% (7).

Shigellosis is mainly transmitted by the faecal-oral route. Several studies have demonstrated that hands may be an important vehicle for transmission of shigellae (5,15,24). Hardy and Watt (6) isolated shigellae from the fingers of mentally retarded individuals in a custodial institution.

The non-culturable phenomenon, a survival strategy of shigellae has been reported (1, 9). The potential health hazard presented by non-culturable shigella spp. may be significant, because of the possibility of conversion into culturable state in the appropriate milieu. It has recently been shown that non-culturable bacteria can revert to the culturable state (18,21,28).

However, one difficulty in elucidating the potential hazard of non-culturable pathogenic bacteria is the inability of routine bacteriological methods to detect such germs from environmental samples, specially when they are present in low numbers. Polymerase chain reaction (PCR) can detect low number of germs in a sample. This technique allows a specific segment of DNA to be amplified by a factor of 10^6 or more within hours (22). It depends only on the presence of target DNA. Thus, PCR is potentially able to detect the presence of non-culturable cells.

On the other hand, it has also been reported that fluorescent antibody (FA) technique is a highly selective and sensitive method for detection and identification of bacterial population in the environment (10, 11,

23, 26). Thus, this technique can also be employed for detection of low numbers of germs, including non-culturable shigellae from environmental samples (9).

Even though hands play an important role in the transmission of shigellae, there is little information about the persistence of shigellae on contaminated fingers. Hutchinson, using conventional cultural techniques (8), has shown that *Shigella sonnei* can survive for up to three hours on fingers in temperate climates, but there is no information on survival of *S. dysenteriae* 1 in a tropical climate. Moreover, when the study on *S. sonnei* was conducted there was no knowledge about non-culturable organisms which could potentially transmit the disease. We are also not aware of any study about the survival of *S. dysenteriae* 1 on fingers using culture, PCR and FA. Therefore, the present study was undertaken to find out the persistence of *S. dysenteriae* 1 on human fingers using culture, FA and PCR techniques.

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SUBJECTS AND METHODS

Volunteers and bacterial strain

Four male, healthy volunteers were selected for this study. One clinical strain of *S. dysenteriae* 1 was obtained from the Microbiology Laboratory of the International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B). This strain was reconfirmed by morphological, cultural, biochemical, and serological tests following standard procedures (13).

Preparation of inoculum

The strain was first inoculated onto MacConkey agar (MA) and incubated at 37 ^{o}C for 24 hours. A loopful of growth was suspended in 10 mL phosphate buffered saline (PBS, pH 7.3). The 90% transmittance of the suspension at A_{585} was measured with a spectrophotometer (Coleman Junior, IIA model 6/20A: Perkin Elmer corp.). Bacterial counts were assessed following standard procedures.

Inoculation of fingers

The finger surface of volunteers was swabbed with 70% alcohol and allowed to dry following washing with sterile distilled water. A drop of 20 m l inoculum (~10^5 cfu) was placed onto each finger tip of the left hand, except the thumb, and was spread over an area of 2 cm x 1 cm. The right hand fingers were not inoculated and used as negative control. The volunteers did not touch anything and were confined to the laboratory during the experiment.

Sampling

Each inoculated finger was washed once in 2 mL washing solution (0.25 Ringer solution containing 0.1% Tween 20) by rubbing with the thumb of the same hand. The thumb was then sterilised by alcohol and rinsed with sterile distilled water. Different fingers were washed at different time intervals (0, 5, 10, 20, 30, 60 minutes, 2, 3, 4, 5, and 8 hours), and shigellae were counted from the Ringer solution. Aliquots of the solution were stored at –20 ^{o}C for detection of shigellae by PCR and FA technique.

Culturable cell count

The viable germ numbers in the finger washes were enumerated on MacConkey agar (MA), Salmonella-Shigella agar (SS), xylose lysine deoxycholate agar (XLD), and Hektoen enteric agar (HEA) following standard procedure with a sensitivity of 1 cfu/cm^2 (9). Two to three colonies from the plates were tested with *S. dysenteriae* 1 antiserum (Difco) by slide agglutination for confirmation of the strain. The bacterial count was expressed as cfu/cm^2 of finger surface. When the count of culturable cells came down to less than 1 cfu/cm^2, then the culturability of the germs was tested by culturing on MA following enrichment with Gram-negative (GN) broth.

DNA extraction

When there were no culturable germs, one mL sample was put in an Eppendorf microfuge (model 5415C) and centrifuged at 3000 x G for three minutes to remove skin particles and debris. The supernatant was collected and DNA of bacterial cells was extracted by the method described by Islam *et al.* (9). The extracted DNA was stored at –20 ^oC if the subsequent experiment was not carried out immediately. The DNA was extracted from the non-culturable form of *S. dysenteriae* after 2, 3, 4, 5, and 8 hours of exposure on the fingers.

DNA amplification by PCR

The amplification of *ipa*H sequences was performed in 100 m l reaction mixture in polypropylene microfuge tubes (Perkin Elmer Cetus). The reaction mixture was prepared using 10 m l buffer solution, 2 m l of each deoxyribonucleoside triphosphate (dATP, dCTP, dGTP, and dTTP), 0.5 m l of Taq DNA polymerase, 1 m l of each primers (H8, 5'-GTTCCTTGACCGCCTTTCCGATAC-3' and H15, 5'-GCCGGTCAGCCACCCTC-3') which were derived from an invasion plasmid of *S. flexneri* M90T, (27) 8 m l of extracted DNA and sterile deionized water to a final volume of 100 m l. The reaction mixture was overlaid with 50 m l mineral oil. The PCR reaction was done for 35 cycles of one minute each at 94 ^oC (for denaturation), 1.5 minute each at 60 ^oC (for annealing of primers to single-stranded DNA), and 0.25 to one minute each at 72 ^oC (for DNA polymerase-mediated extension) according to the procedure described by Echeverria *et al.* (3).

Analysis of PCR product

The amplified DNA was analysed by agarose gel electrophoresis. The PCR product and molecular weight marker (HaeIII digest of f X174 replicative form DNA) were subjected to electrophoresis. The separated product was visualised by UV transilluminator (Ultraviolet products Inc. San Gabriel, California, USA) after staining with ethidium bromide.

Hybridisation of PCR product

The PCR product was also identified by hybridisation technique according to the ECL (enhanced chemiluminescence) direct nucleic acid labelling and detection systems protocol (Amersham, UK). After electrophoresis, the gel was processed with the denaturation solution (1.5 M NaCl, 0.5 M NaOH) and neutralisation solution (1.5 M NaCl, 0.5 M Tris HCl, pH 7.5), followed by washing with deionised water. The separated product was then transferred to a nylon membrane by capillary blotting technique (17). A single-stranded *ipa*H probe, described previously (27) was labelled with horseradish peroxidase (20). The hybridisation between PCR product fixed on the blot and the labelled probe was carried out under stringent conditions. The blot was removed immediately from the hybridisation medium and washed twice in primary wash buffer (0.4% SDS, 0.5 x SSC) for 10 minutes at 55 ^oC followed by another wash in secondary wash buffer (2 x SSC) for five minutes at room temperature. The occurrence of hybridisation was detected using ECL detection reagents (Amersham, UK). Autoradiography film was used for detecting hybridised product on the nylon membrane.

Fluorescent microscopy

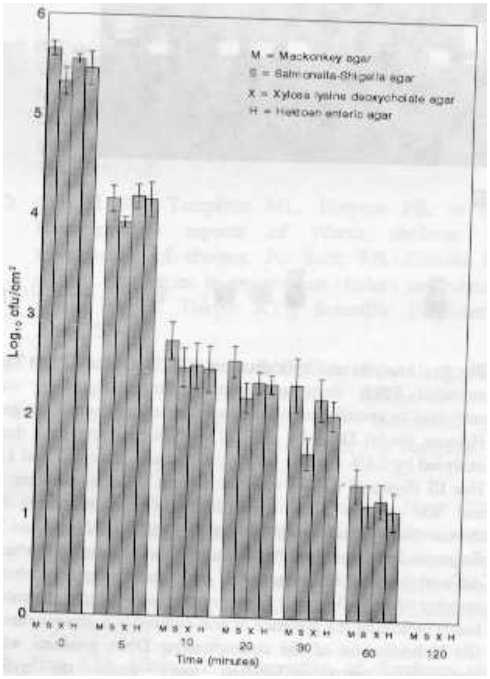
One mL of hand wash sample was put into an Eppendorf microfuge tube and centrifuged at 3000 x G for three minutes to remove the skin particles and debris. Supernatant was collected and was centrifuged at 15,000 x G for five minutes. The supernatant was discarded and the pellet was resuspended in 25 m l of PBS (pH 7.3). 5 m l of this suspension was applied to a microscopic slide. The germs were stained with *S. dysenteriae* 1 polyclonal antiserum (Wellcome Diagnostics, Dartford, United Kingdom) and fluorescein isothiocyanate-conjugated anti-rabbit goat serum. The slide was mounted under a coverslip with buffered glycerol (pH 8.3) and then examined under an epifluorescence microscope (model BH-2; Olympus). *S. dysenteriae* 1 was used as a positive control.

Data analysis

The experiment was carried out with four volunteers and was repeated four times with each individual. The count of the culturable *S. dysenteriae* 1 on the fingers was calculated at intervals of 0, 5, 10, 20, 30, 60 and 120 minutes. Firstly the average counts were obtained from the 4 readings of each individual; then the mean values of the counts were expressed with standard deviation (SD) from the average counts of the four individuals at each sampling interval.

RESULTS

Survival of *S. dysenteriae* 1 in the culturable state on the finger was considered as the ability of that organism to multiply and to form colonies on various selective media, e.g. MA, SS, XLD, and HEA. Fig. 1 shows the survival pattern of *S. dysenteriae* 1 on the fingers of four volunteers. The result demonstrated that the growth response of *S. dysenteriae* 1 was similar on all the media used. The initial count was over 10^5 cfu/cm^2 which gradually decreased with time and the cells were not culturable after 60 minutes on the fingers of all the four individuals.



Time (minutes)

Fig. 1: Culturability of S. dysenteriae 1 on different media after inoculation on volunteers' fingers. Log10 counts of S. dysenteriae type 1 on fingers ar different time intervals are shown with ? 1SD. Symbols: MacConkey agar (M), Salmonella-Shigella agar (S), xylose lysine deoxycholate agar (X) and Hektoen enteric agar (H)

The DNA was subjected to amplification by PCR and the PCR products were analysed by agarose gel electrophoresis (Fig. 2A). The analysed PCR products were then transferred to nylon membrane and were hybridised with the *ipa*H probe. Figure 2A shows that the finger washing samples collected at 2, 3, and 4 hours intervals generated the expected 700-bp fragment of the H locus which hybridised to the *ipa*H probe (Fig. 2B), indicating the presence of non-culturable *S. dysenteriae* 1 on the fingers for up to four hours.

The non-culturable *S. dysenteriae* (Fig. 3A) on the fingers were also detected up to four hours using FA technique and appeared as rounded bodies rather than rods, contrasting with the culturable form of *S. dysenteriae* 1 (Fig. 3B).

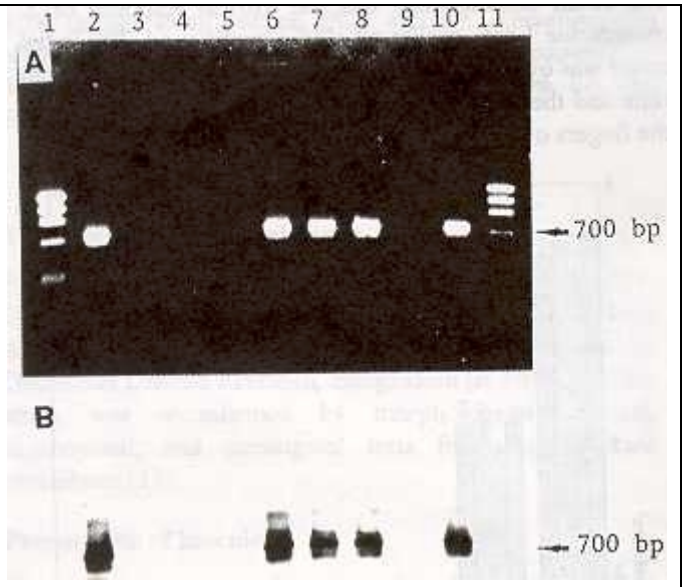


Fig. 2: Analysis and hybridisation of PCR products : (A) The extracted DNA from contaminated finger washings was subjected to specific amplification of invasion plasmid antigen H locus (ipaH) DNA by PCR. The PCR products were then analysed by 0.8% agarose gel electrophoresis. Lanes : 1 and 11, Hae III digest of ?X174 replicative-form DNA as markers; 2 and 10, positive controls with DNA from culturable S. dysenteriae 1; 3 and 9 negative controls without DNA; 4 and 5, finger washing samples after 8 and 6 hours respectively which did not generate any bands; and 6, 7 and 8, finger washing samples after 4, 3 and 2 hours respectively which generated bands indicating the presence of non-culturable S. dysenteriae 1 (B) Hybridisation of the corresponding DNA products with horseradish peroxidase-labelled ipaH probe on nylon membrane.

DISCUSSION

In the present study, *S. dysenteriae* 1 was shown to survive in a culturable state for up to one hour but persisted for up to four hours as non-culturable state on volunteers' fingers. The non-culturable stage in the life cycle of a bacterium appears to be a strategy for survival when the organism is exposed to conditions that are less than optimal for cell growth and division.

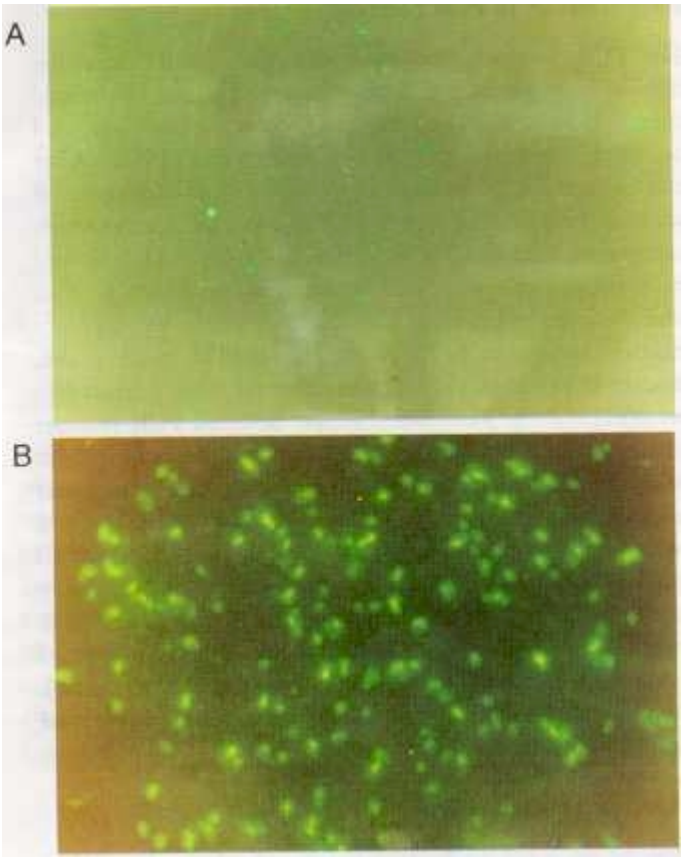


Fig. 3: Fluorescence photomicrograph of S. dysenteriae 1. (A) non-culturable S. dysenteriae 1 obtained from the finger washings of volunteers (B) culturable S. dysenteriea 1 as a positive control (magnification x 1000 for both)

There is evidence that *S. sonnei* can survive on human hands (8), and human hands have been implicated as sources of transmission in outbreaks of shigellosis (15). This has led us to explore the persistence of *S. dysenteriae* on human fingers. The non-culturable form of *S. dysenteriae* has been investigated, because this non-culturable form has been shown to occur in *V. cholerae*, *Salmonella enteritidis*, enteropathogenic *E. coli* and other pathogens (1, 21,29). It is, therefore, important that methods to detect such germs be developed. Moreover, a technique to detect small numbers of cells is essential, since many bacteria are present in the environment only at low densities. Such detection is especially important for bacteria, such as *Shigellae*, which can produce disease after ingestion of as few as 10 organisms (16). The PCR technique seems ideally suited for this goal, since it potentially allows amplification of the DNA obtained from only a few germs (25).

In the present study, the persistence of *S. dysenteriae* on volunteers' fingers has also been observed by fluorescence microscopy for up to four hours, despite the fact that after one hour the cells could not be cultured. This study shows that *S. dysenteriae* became non-culturable within one hour on finger surface, which may be due to dryness and the low pH of the skin (19). Moreover, antimicrobial substances (e.g. lysozyme, complex lipid products, antibodies, primarily IgA and IgG) excreted from the skin (12) may play a significant role in the quick transformation of culturable to non-culturable state of *S. dysenteriae* on human fingers. However, these non-culturable bacteria, if still viable, may revert to the culturable form in a favourable environment. Colwell *et al*. (2) observed that non-culturable *V. cholerae*, when ingested by volunteers, reverted to culturable form and were excreted in the stools.

The present study shows that the present methods of detecting shigellae by conventional culture techniques are not fully adequate. This is corroborated by outbreaks of shigellosis in which no organisms can be isolated from the suspected transmission vehicles by conventional culture techniques (8). However, the PCR and fluorescent antibody methods allow the detection of these non-culturable *S. dysenteriae* germs, even if present in very low numbers.

The non-culturable state reported here for *S. dysenteriae* 1 may be significant for understanding the epidemiology of shigellosis. Recently, a community-based study in an endemic area of Thailand has demonstrated that non-culturable shigellae detected by PCR can cause dysentery with similar clinical characteristics as when caused by shigellae (4). Therefore, it is possible that such non-culturable shigellae may pose health problems. The results of this study, therefore, demonstrate the significance of the persistence of non-culturable *S. dysenteriae* 1 on fingers, which may be important from a public health point of view.

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SHORT REPORT

Intestinal Parasitic Infections in Patients with Malignancy

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ABSTRACT

The frequency of intestinal parasitic infections was studied retrospectively in 1,029 cancer patients presenting with symptoms of diarrhoea. Intestinal parasites were diagnosed by stool examination, using both the direct and concentration techniques and also the modified acid fast stain. Parasitic infection was found in 16.5% of the cases. The majority of the patients with intestinal parasitosis had cancer of the haemopoietic system and were on anticancer chemotherapy. The most prevalent parasites were *Entamoeba histolytica*/*Entamoeba dispar* (8.5%) and *Giardia lamblia* (3.1%). Much more rare were *Strongyloides stercoralis* (0.6%), *Cryptosporidium parvum* (0.3%) and *Isospora belli* (0.1%). All the patients with intestinal parasites were negative for HIV antibodies.

Key words: Intestinal diseases, Parasitic; Diarrhoea; Parasites; Neoplasms; Retrospective studies; Malignant carcinoid syndrome

INTRODUCTION

Infectious complications have become frequent causes of morbidity and mortality in cancer patients, often replacing the primary disease as the leading cause of death (1). In the past 20 years, major advances in medicine have allowed cancer patients to live prolonged productive lives, in some cases free of malignant disease. The price we pay for these achievements is a major deficiency of the host defence mechanisms that place the patient at risk for infectious complications with high morbidity and mortality. In the early 1980s, it appeared that the importance of parasitic infections was declining in immunocompromised cancer patients. In recent years however, the frequency of these diseases has risen owing to the increased use of corticosteroids in chemotherapeutic regimens (1). With this background in mind, we at Kidwai Memorial Institute of Oncology, undertook this retrospective analysis to know the frequency of intestinal parasitic infections in cancer patients at our hospital over a 4-year period.

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PATIENTS AND METHODS

Patients

A retrospective analysis was undertaken of the results of stool examination done in cancer patients suffering from diarrhoea who either attended the outpatient department or were hospitalised in the Kidwai Memorial Institute of Oncology, Bangalore, India for the management of various malignancies.

Specimen processing

Stool specimens were examined microscopically by suspending an aliquot each, in a drop of physiological saline and of Lugol's iodine. This was followed by formol-ether concentration and microscopic examination of the sediment. In addition, sediment smears were also stained by the modified acid-fast stain for detecting Cryptosporidium oocysts (2). Blood was collected for HIV serology only from patients whose stools revealed parasites to rule out the possibility of immunosuppression due to HIV infection. Simple frequencies and chi-squared test were used for statistical analysis.

RESULTS

Over a 4-year period, 1029 faecal samples were screened for intestinal parasites. Of these, 326 (31.7%) were from patients having solid tumours and 703 (68.3%) from those having haematological malignancies. The male/female ratio in the patients was 1.5 (612/417). Their ages ranged from 0 to 87 years with a median of 17 years and an interquartile range of 5 to 43 years. A total of 170 patients (16.5%) showed evidence of intestinal parasitic infection. Their male/female ratio was 1.5 (101/69), and their ages ranged from 0 to 76 years with a median of 18 years and an interquartile range of 6 to 40 years. As shown in table I, 63.5% (108/170) of these patients with diarrhoea had a haematological malignancy compared to 36.5% (62/170) who had a solid tumour. Sixty-three per cent of the patients with intestinal parasites (106/170) were on anticancer chemotherapy; 14.9% (25/170) of them were receiving radiotherapy; only 1.2% (2/170) were on steroids and a small percentage were on combination therapy. In contrast, 18.2% (31/170) were not receiving any form of anticancer treatment. None of these 170 patients were positive for HIV antibodies, ruling out the possibility of opportunistic infections due to HIV infection.

Of the 170 patients, 160 (94.1%) had only 1 parasitic infection, whereas 10 (5.9%) were infected with 2 parasites concomitantly. Five of these 10 had Entamoeba histolytica/Entamoeba dispar and hookworms; 2 were infected with both E. histolytica/E. dispar and Giardia lamblia; and one patient each had E. histolytica/E. dispar and roundworm; hookworm and roundworm; and G. lamblia and Hymenolepis nana.

Table I. Treatment details of cancer patients with intestinal parasites

	Therapy			
Diagnosis	CT	RT	Others	NT
Haematological malignancies	79	4	7	18
(n=108)	(73)	(4)	(7)	(17)
Solid tumours	27	21	1	13
(n=62)	(44)	(34)	(2)	(21)
Total	**106**	**25**	**8**	**35**
(n=170)	(62)	(15)	(5)	(21)

CT = Chemotherapy; RT = Radiotherapy; Others = Combination treatments; NT = No treatment
Figures in parentheses indicate row percentages

Table II. Intestinal parasites in different malignancies

Diagnosis	Parasites									
	E.h	G.l	Hw	Rw	S.s	T.t	C.p	H.n	I.b	Total
Haematological malignancies (n=108)	52 (46)	27 (24)	10 (9)	11 (10)	5 (4)	3 (3)	3 (3)	2 (2)	1 (1)	114 (100)
Solid tumours (n=62)	35 (53)	5 (8)	14 (21)	9 (14)	1 (2)	2 (3)	0 -	0 -	0 -	66 (100)
Total (n=170)	**87** (48)	**32** (18)	**24** (13)	**20** (11)	**6** (3)	**5** (3)	**3** (2)	**2** (1)	**1** (0.6)	**180** (100)
%	(8.5)	(3.1)	(2.3)	(1.9)	(0.6)	(0.5)	(0.3)	(0.2)	(0.1)	(17.5)

E.h = *E. histolytica*; G.l = *G. lamblia*; Hw = Hookworm; Rw = Roundworm; S.s = *S. stercoralis*; T.t = *T. trichiura*; C.p = *C. parvum*; H.n = *H. nana*; I.b = *I. belli*
Figures in parentheses are row percentages; (%) = percentages of the total number of samples screened (n = 1,029)

E.h = E. histolytica; G.l = G. lamblia; Hw = Hookworm; Rw = Roundworm; S.s = S. stercoralis; T.t = T. trichiura; C.p = C. parvum; H.n = H. nana; I.b = I. belli
Figures in parentheses are row percentages; (%) = percentages of the total number of samples screened (n = 1,029)

Table II shows the parasites in decreasing order of frequency: E. histolytica/E. dispar (87, i.e. 8.5%), G. lamblia (32, i.e. 3.1%), hookworms (24, i.e. 2.3%), roundworms (20, i.e. 1.9%), Strongyloides stercoralis (6, i.e. 0.6%), Trichuris trichiura (5, i.e. 0.5%), Cryptosporidium parvum (3, i.e. 0.3%), H. nana (2, i.e. 0.2%) and Isospora belli (1, i.e. 0.1%).

Table III. Intestinal parasites according to different modes of treatment

Parasite	Therapy					
	CT	RT	ST	Other	NT	Total
E.h	58	12	0	3	14	87
G.l	23	1	1	1	6	32
H.w	9	9	0	0	6	24
R.w	11	2	1	0	6	20
S.s	6	0	0	0	0	6
T.t	1	2	0	0	2	5
C.p	2	0	0	1	0	3
H.n	1	0	0	0	1	2
I.b	1	0	0	0	0	1
Total	112	26	2	5	35	180

E.h = *E. histolytica*; G.l = *G. lamblia*; Hw = Hookworm; Rw = Roundworm; S.s = *S. stercoralis*; T.t = *T. trichiura*; C.p = *C. parvum*; H.n = *H. nana*; I.b = *I. belli*;
CT = Chemotherapy; RT = Radiotherapy; ST = Steroids; Other = Various combinations of the former; NT = No treatment

DISCUSSION

Infections have long been recognised as one of the most significant causes of death among patients with malignancy. Despite advances in antineoplastic therapy, infections still remain the most significant complication of therapy. Patients suffering from various malignancies present with varying degrees of immunodeficiency caused by their treatments or due to the disease process itself. Data from our laboratory were analysed to find out the frequency of different intestinal parasitic infections in different malignancies. In our study, parasitic infections were associated with diarrhoea in 16.5% of our patients. We had more cases with haematological malignancies than in those with solid tumours. This figure is lower than that reported by Abaza et al. (3), who found 23% of their patients to have intestinal parasitic infection, but higher than that reported by Rivera et al. (4), who found only 12.9% of children with acute

leukaemia to have intestinal parasitoses. These differences could be explained by the variation by region in the incidence of intestinal parasites. Chemotherapy other than corticosteroids appeared to predispose the patients towards getting parasitic infections more than any other form of anticancer treatment, which is contrary to what has been reported earlier (1). The most prevalent parasites in our study were E. histolytica/E. dispar and G. lamblia. A similar finding has been reported by Azab et al. (5), who have also used serology for diagnosing 8 parasitic infections. Opportunistic strongyloidosis including the syndrome of hyperinfection, occur most often in patients with T-lymphocyte impairment. Sporadic cases have been reported in patients with lymphoma and chronic lymphoid leukaemia (6). We found 0.5% of our patients with diarrhoea to have strongyloidosis, all of whom being on chemotherapy and the majority of them belonging to the haematological malignancies group. The reported incidence of strongyloidosis in cancer patients, however, varies from 0.7% (3) to 3.6% (7). This variation could be attributed to the degree of immunosuppression, since the parasite is usually known to exist in the gut without causing severe disability. However, in patients with impaired host defences or those receiving antitumour or immunosuppressive agents, a serious infection may develop (6). Cryptosporidiosis has been reported in cases of acute leukaemia (8-10), rhabdomyosarcoma (11), and as a complication of cancer chemotherapy (12). In a recent report, Abaza et al. (3) found the prevalence of cryptosporidiosis amongst cancer patients to be 6.3%. In contrast, in the present study we found only 0.3% of our patients harbouring this infection. These three patients had haematological malignancies and were either receiving anticancer chemotherapy alone or chemotherapy with steroids. In an earlier study however, we have reported the incidence of cryptosporidiosis among cancer patients to be 1.3% ($p < 0.05$) (13). Khalil et al. (7) and Makled et al. (14), on the other hand, did not find any case of cryptosporidiosis amongst cancer patients. This variation in the incidence of cryptosporidiosis among cancer patients could be explained by the extent of exposure to infected animals since cryptosporidiosis is basically a zoonosis. Isosporiasis is a common opportunistic infection in acute lymphoblastic leukaemia (15), adult T-cell leukaemia (16-18), and Hodgkin's disease. It also is often associated with HTLV-1 seropositivity. Reports on its incidence in cancer patients are lacking. Abaza et al. (3) did not find any case of isosporiasis among different groups of immunocompromised hosts. We found only 1 case of I. belli infection that was not associated with HTLV-1 infection (19). The low prevalence of this parasitic infection in our study, however, may not reflect the actual picture, since use of antibiotics both in prophylaxis and empirical treatment of infections is widespread in our country. A prospective case control study would perhaps help us in determining whether any modality of anticancer treatment does indeed predispose cancer patients to developing parasitic infections.

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LETTER TO THE EDITOR

Immature *Hymenolepis nana* Worms in the Stools of aPatient Treated for Acute Lymphoblastic Leukaemia:an Uncommon Observation

Sir,

We report here a case of Hymenolepis nana infestation in a patient with acute lymphoblastic leukaemia on anti-cancer treatment. A 25-year old female patient was admitted to our hospital for the management of acute lymphoblastic leukaemia. She completed her induction chemotherapy over a 4- week period. She first received vincristine, 2 mg intravenously once a day for 5 days; L-asparginase, 10,000 U subcutaneously once a day for 8 days; daunomycin, 50 mg intravenously once a day for 3 days; intrathecal methotrexate, 15 mg once a week for 4 weeks and oral prednisolone, 50 mg once a day for 28 days. The next phase of chemotherapy consisted of cyclophosphamide, 1200 mg once a day intravenously for 2 days; oral mercaptopurine, 125mg once a day for 14 days; intrathecal methotrexate 15 mg once a week for 4 weeks and cranial irradiation for 10 days. She completed this cycle on 19 July 1996. The same day the patient was discharged on personal request to go home for 2 days. A repeat induction chemotherapy (same regimen as above) was started on 30 July 1996. The following day, she developed loose stools and vomiting. The stools were tinged with blood and contained mucus. Microscopic examination of a faeces sample under low power objective (40x magnification) revealed mucosal fragments and 2 to 3 immature worms of H. nana per field. The worms had only the scolex and the neck (Fig. 1 and 2). The scolex contained 4 suckers and a rostellum bearing a circular row of 24 hooklets. Some of these immature worms were seen adherent to the mucosal fragments. Eggs or cysticercoid larvae were not seen. A diagnosis of severe infestation with H. nana was made. While anti-cancer drugs are known to cause severe mucositis (1), denudation of the intestinal mucosa is rare. In the case presented, we think chemotherapy produced denudation of the mucosa, resulting in the appearance of the immature H. nana worms in the stools. Treatment was instituted with albendazole 15 mg/kg/day in 3 doses for 8 days, as recommended for cysticercosis and taking into account that the patient could not afford praziquantel (2). The patient's condition and the very rare occurrence of numerous H. nana in a stool sample, rather than the characteristic eggs, made us fear dissemination of worms into extra-mucosal sites. Thus, we aimed specifically at attacking worms possibly escaping into body tissues. The patient's complaints disappeared in a few days. Repeat stool examinations did not reveal either worms or eggs of H. nana, though this worm can maintain itself in its human host by internal auto-infection.

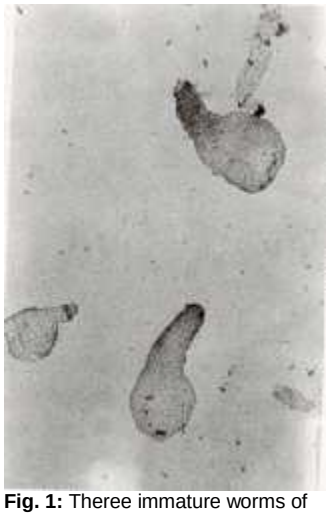

Fig. 1: Theree immature worms of H. nana (40x magnification)

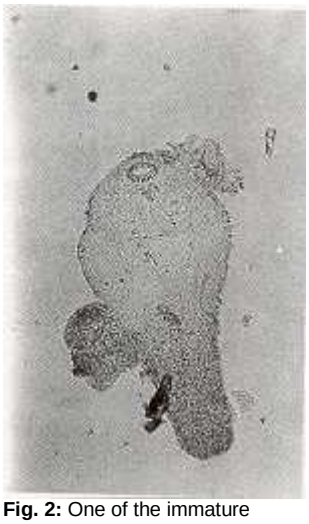

Fig. 2: One of the immature worms of H. nana (100x magnification)

Anti-cancer chemotherapy depresses the immune system. Little is known about how this affects man's defence against intestinal parasitoses. In this case, diagnosis of hymenolepiasis was based on the chance finding of H. nana worms rather than the characteristic eggs in the stools. It highlights that it is worthwhile to investigate cancer patients for intestinal parasites before and during chemotherapy.

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REPRODUCTIVE TRACT INFECTIONS: PUBLIC HEALTH AND EPIDEMIOLOGY-I

Management of Sexually Transmitted Diseases by Rural Practitioners

Mohsin U. Ahmed, Shameem Ahmed, Parveen A. Khanum, and Mobarak H. Khan

Objective: Identify the knowledge and perceptions of rural health practitioners regarding sexually transmitted diseases (STDs).

Methodology: To assess the type of management of reproductive tract infections (RTIs) in rural Bangladesh, 24 practitioners were interviewed randomly. Data collected in the structured questionnaire were analyzed. Two unions of Abhoynagar thana, Jessore, were included. This study was conducted in July 1996. Rural practitioners who have had no institutional training, but practise homeo and allopathic medicine in rural settings were surveyed.

Results: Findings indicate that although almost all of them are in confusion regarding the recognition and management of STDs/RTIs, most are aware of the modes of transmission and means of prevention of STDs/STIs. STD patients are found to visit and revisit the rural practitioners. Most practitioners prefer referral of the patients to government facilities at the union and thana level.

Conclusion: Knowledge of the rural practitioners regarding STDs/RTIs needs to be strengthened, and an effective referral system needs to be established for STD/RTI patients.

Comparison of the Laboratory and Clinical Diagnoses of Bacterial Vaginosis: Can Simple Clinical Criteria be Used at PHC Level?

Nazmul Alam, Shamim Sufia Islam, Kaniz Gausia, Farid Ahmed, Andres de Francisco, and Sarah Hawkes

Objective: Compare clinical criteria for diagnosing the presence of bacterial vaginosis against well-recognized laboratory criteria, and hence, to test the sensitivity and specificity of clinical criteria.

Methodology: As part of the RTI prevalence study in Matlab, all women in both population-based (n=804) and symptomatic (n=465) sub-groups had a physical examination (including genital examination) and microbiological samples were taken to screen for the presence of infection. Bacterial vaginosis was diagnosed using recognized laboratory criteria: the presence of epithelial cells with adherent Gram variable Coccobacilli and the absence of normal flora on vaginal swabs. Laboratory diagnoses were then tabulated against accepted criteria (presence of "abnormal" vaginal discharge; vaginal pH>4.7; KOH test positive) to see which test or combination of tests was the best predictor for the presence of BV.

Results: Seventeen percent of the women in the symptomatic group and 11.7% in the population-based group had Bacterial Vaginosis on laboratory diagnosis. Using Gram stain (clue cells) as the "gold standard" for diagnosis of BV, clinicians correctly identified only 35% of the symptomatic group and 29% of the population-based group of infected women. Analyzing diagnostic signs, pH>4.7 had the greatest sensitivity (87%), but a low specificity (48% in symptomatic women). However, specificity was greatly improved by using combinations of 2 or 3 diagnostic criteria.

Conclusion: To improve the diagnosis of bacterial vaginosis in the absence of accessible laboratory facilities, it is recommended that pH>4.7 can be used as a screening test, and then other clinical criteria (abnormal-looking discharge and whiff test positive) can be used for increasing the specificity of diagnosis.

A Cross-sectional Study on the Prevalence of Sexually Transmitted Infections among Dhaka Slum Dwellers

Keith Sabin, Mahbubur Rahman, Sarah Hawkes, Khaled Ahsan, Lutfa Begum, Shams El Arifeen, and Abdullah H. Baqui

Objective: Determine the prevalence of selected sexually transmitted infections in a Dhaka slum population.

Methodology: Responses to a questionnaire and blood and urine specimens were collected during a single visit to randomly selected Dhaka slum dwellers. Syphilis and hepatitis B serologies were performed, and urine was tested for gonorrhoea and chlamydia with polymerase chain reaction methods. Dhaka slums located in five thanas: Mohammadpur, Lalbagh, Kotwali, Demra, and Sutrapur, were the study sites. The study was conducted during 31 July-15 October 1996. Five hundred forty men and 993 women respondents were enrolled in the study.

Results: Hepatitis B surface antigen was present in 5.8% (31/530) men and 2.9% women (29/984). Serological evidence of current syphilis infection was found in 11.5% (62/530) men and in 5.4% (54/984) women. Gonorrhoea and chlamydia were diagnosed by polymerase chain reaction -- both had a prevalence of below 1%.

Conclusion: Prevalence of hepatitis B carriers and positive syphilis is high in the Dhaka slum population. Given the known adverse outcomes of both infections, two policy recommendations are suggested: (1) addition of the hepatitis B vaccine to EPI; (2) screening for women for syphilis during routine antenatal care**.**

Surveillance of HIV-1 Seropositivity in Bangladeshi Children with Persistent Diarrhoea and Malnutrition

G.J. Fuchs, M. Ali, A. Hossein, and S. Vermund

Objective: Children with persistent diarrhoea and malnutrition have up to a forty-fold increase in prevalence of HIV infection compared to the prevalence in the general population in regions endemic for HIV infection. Persistent diarrhoea with malnutrition is one of the more common severe conditions in children admitted to the ICDDR,B Dhaka treatment centre. The presence of HIV infection with persistent diarrhoea and malnutrition would affect the results and interpretation of epidemiological and intervention studies of persistent diarrhoea and malnutrition at ICDDR,B. Further, this population of patients would provide sentinel information about the prevalence of HIV in the general population. A delinked and anonymous surveillance system was initiated in November 1996 to determine the prevalence of HIV seropositivity in Bangladeshi children with persistent diarrhoea and malnutrition admitted to the Clinical Research and Service Centre ("ICDDR,B Hospital") in Dhaka.

Methodology: Two years old or younger children with persistent diarrhoea (lasting for 14 or more days within the previous 3 weeks) with average number of stools greater than 3 per day, and weight-for-height less than 90% of the median were included. HIV seropositivity was determined using a standard method.

Results: Approximately 10% of the initial 250 children tested were positive by ELISA. Ultimately, four of the 401 subjects were repeatedly positive by ELISA. Of these four repeatedly positive specimens, none was positive by Western Blot.

Conclusion: HIV infection is, to date, absent in our children with persistent diarrhoea and malnutrition; these children have a higher-than-predicted prevalence of a circulating cross-reacting substance with HIV antibody. The current WHO recommendation for the universal use of repeatedly positive ELISA as a specific test for diagnosis and for screening blood products for transfusion should be reconsidered.

Prevalence of Reproductive Tract Infections among Women Attending the BWHC Clinic in Mirpur, Dhaka

J. Bogaerts, J. Ahmed, N. Akhter, and N. Begum

Objective: Document the prevalence and aetiology of reproductive tract infections among women attending the BWHC clinic in Mirpur, Dhaka.

Methodology: This is a cross-sectional study among randomly selected married women. The women undergo a gynaecological examination and have laboratory tests for bacterial vaginosis (BV) and infection with N. gonorrhoeae, T. vaginalis, T. pallidum, C. trachomatis, and yeasts. The study was conducted at the Bangladesh Women's Health Coalition Project clinic in Mirpur, Dhaka, during July 1996-December 1997. The unit of analysis was married women who attended the antenatal and general health clinics, the expanded programme of immunization, the family planning and menstrual regulation units. Outcome measures are: BV; clue cells on Gram staining; N. gonorrhoeae: culture; T. vaginalis: fresh examination; T. pallidum: serum antibodies: ELISA; C. trachomatis; yeasts: fresh examination, Gram staining.

Results: Thirty percent of the women had vaginal discharge, and 30% had symptoms of cervicitis. Clue cells were observed among 14% of the women. Other laboratory findings were: N. gonorrhoeae 1%; T. vaginalis <1%; T. pallidum antibodies 1% C. trachomatis 1%; yeasts 30%.

Conclusion: RTIs are common among women attending the BWHC clinic. The prevalence of STI is lower than suggested by previous studies conducted in Bangladesh.

Using the Results of the Matlab RTI/STI Study to Develop Suitable STI Control Programmes in Bangladesh

Sarah Hawkes, Andres de Francisco, J. Chakraborty, Kaniz Gausia, Linda Williams, and David Mabey

Objective: Use the results of population-based and clinic-based RTI/STI studies to develop suitable controlled and management programmes.

Methodology: A large population-based study of the prevalence of RTIs and STIs was undertaken in the Matlab area over an 18-month period in 1995-1996. More than 4,500 people were included in five arms of the survey which looked into reported symptomatology, clinical presentation, treatment-seeking histories and laboratory diagnoses in both population- and clinic-based samples.

Results: The overall prevalence of STIs was low, but this was found equally among men and women. Endogenous infections (candida and bacterial vaginosis) in women were more common. Clinical overdiagnosis of both STIs and RTIs was common, and classical clinical signs were poor predictors of the presence of infection. Most people initially sought treatment outside the public sector services for RTI/STI symptoms.

Conclusion: The feasibility of introducing currently recommended syndromic management flow charts for the treatment of both STIs and RTIs at the PHC level has been evaluated in the population under study. Further development of the recommended algorithms for treating endogenous infections is needed. Suitable methods for diagnosis and management of STIs in a low-prevalence situation need careful consideration. Possible strategies at the public health level include: ophthalmia neonatorum prophylaxis; screening pregnant women for syphilis; inclusion of men in service provision; inclusion of the private sector in training and quality control; prevention and information campaigns to encourage effective health care-seeking behaviour strategies; and possible targeting of STI service provision in areas with higher prevalence.

REPRODUCTIVE TRACT INFECTIONS: SOCIAL AND BEHAVIOURAL, ASPECTS-I

AIDS Awareness in Rural Chittagong

Mizanur Rahman, Barkat-e-Khuda, and Masud Reza

Objective: Examine the level and pattern of awareness of AIDS among rural men and women.

Methodology: Two questions--"Have you heard about a disease called AIDS?" and "How is it transmitted?"--were asked in a survey on awareness of, accessibility to, and use of, health and family planning services in three rural thanas of Chittagong district during late 1996. Married women of reproductive age (n=1,876) and married men (n=1,506), whose wives were of reproductive age, were the respondents in the survey. Level of, and factors associated with, AIDS awareness were examined in bivariate and/or logistic regression analyses.

Results: Slightly over 20 percent of the females and about 40 percent of the males reported having heard about AIDS. Of those who had heard about AIDS, about 33 and 66 percent of the females and males, respectively, could specify probable transmission mechanisms. About 90 percent women reported that AIDS is transmitted through extramarital sex. Men reported that extramarital sex (48%), sex with commercial sex workers (16%), and transmission of the disease from AIDS patients through blood or sex (24%), were the major mechanisms of AIDS transmission. Among both men and women, awareness was higher among the educated than uneducated respondents. Middle-aged women or women with fewer children were more aware of AIDS than other women. Awareness among males was not associated with demographic variables.

Conclusion: The level of AIDS awareness is extremely low among rural women and relatively low among men. A comprehensive AIDS educational campaign would probably be the most effective approach toward preventing an AIDS epidemic in Bangladesh.

Risk Behaviour among Injection Drug Users in Bangladesh: Risk of Rapid Spread of HIV in the Expected Epidemic

Albert Felsenstein, Ashraful Alam Neeloy, and Bert Pelto

Objective: Provide preliminary information concerning behaviour patterns of injection drug users, and the relationship between intravenous (IV) drug use and high-risk behaviour in relation to possible HIV infections.

Methodology: This pilot study was conducted from August 1996 to January 1997 using the case study method. Ten in-depth case studies were completed and analyzed. All interviews were conducted in the Mukti Clinic in Gulshan, Dhaka. All respondents were relatively affluent patients under treatment for IV drug abuse. Descriptive data include: history (sequences) of drug adoptions, sharing of needles, sources of supply, sexual practices, and peer relations.

Results: All subjects used non-intravenously administered drugs before becoming IV drug users. All subjects admitted occasional sharing of needles, although most were aware of the health hazards resulting from this practice. Almost all the respondents belonged to a group of friends with strong peer pressure leading to a sequence of increasing drug abuse. Most admitted to have unsafe sexual contacts with sex workers and/or other partners. The data suggest that the IV drug users, though a few in number, could play a disproportionately large role in the spread of HIV infections, as is true, for example, in nearby Manipur in India.

Conclusion: The unsafe practices with sex workers and others reveal a possible two-way method for spread of HIV infection when the pre-epidemic stage for HIV reaches Bangladesh. Identification and knowledge of this drug injection sub-culture could enhance the success of preventive programmes related to HIV. The information gathered in this pilot study should be applied to plan and implement larger-scale studies, eventually with operational intervention aspects.

Impact of AIDS Awareness Activities in a Remote Rural Area of Bangladesh

M.A. Hanifi, Moazzem Hossain, Ayesha Aziz, and Abbas Bhuiya

Objective: See the impact of AIDS awareness programme on knowledge of AIDS.

Methodology: Surveys were carried out before and after the AIDS Awareness Campaign on the "World AIDS Day 1996." Community members, including teachers and students were involved in miking, rally, discussion, meeting, drama, etc. in community AIDS-related campaign. The study was conducted during November-December 1996 in three unions of Chakaria thana under Cox's Bazar district. Each survey covered 300 men and 300 women aged 18-60 years from a population of 65000.

Results: Thirty-eight percent of the respondents in the post-intervention survey reported to have heard about AIDS compared to 19% in the pre-intervention survey. The level of increase in knowledge was higher among males than females. Of those who heard about AIDS, the knowledge of prevention was 24% before the intervention compared to 36% after the intervention. A similar pattern was also observed for the consequences of AIDS.

Conclusion: AIDS awareness programme can be undertaken in the villages. If participatory research methods are applied, the villagers would participate enthusiastically in mass awareness campaigns for diseases, such as AIDS.

Sexual Behaviours among Cases with Sexually Transmitted Diseases in Dhaka Slums

Anjali Sharma, Papreen Nahar, Keith Sabin, Lutfa Begum, Khaled Ahsan, Shams El Arifeen, and Abdullah H. Baqui

Objective: Identify the behavioural basis for sexually transmitted diseases among cases in Dhaka slums.

Methodology: Thirty male cases and 30 female cases who met the WHO syndromic algorithm for STD and/or tested positive for STDs during an earlier survey, were revisited for follow-up in-depth interviews and treatment. The interviews were manually scoured for perception of causes of their STDs as well as actual reported behaviour which might have led to STD. The Urban Surveillance System clusters located in Mohammadpur, Lalbagh, and Sutrapur areas of Dhaka city were selected for the study conducted during 31 July -30 October 1996.

Results: Behaviours that put sexually active adults at risk for acquiring STD were common in this population. Age at first sexual intercourse ranged from 12 to 15 years for both men and women. However, while most men reported this as premarital sex for both partners, most women reported their first partners to be their husbands. The men reported as visiting commercial sex workers, women whom they pretend to love, women whom they do love, and in 8.5% cases having male sex partners, while unmarried. Some also reported visiting CSWs, and a few reported visiting women whom they loved after marriage. Approximately, half of the women suspect their husbands have had or are having extra-marital affairs. Nonetheless, the men seldom reported using a condom and women said that their husbands rarely used condom.

Conclusion: The Dhaka slum-dwelling population appears to have core groups of sexually active, non-monogamous adults who may transmit STD to one mother and non-core group members. Reproductive/sexual health clinics need to attract these adults for treatment and counselling on disease prevention.

Comparing the Cases of Men and Women Attending Sexual Health Clinics in Matlab

Sarah Hawkes, S.M. Nurul Alam, Hasanur Rahman, Habibur Rahman,
Ashraful Alam Neeloy, Tamanna Sharmin, James Ross, and Bert Pelto

Objective: Compare and contrast males and females in rural Matlab in terms of their patterns of symptom presentation, communications with health workers, as well as the styles of communication of health workers with the clients.

Methodology: Clinical interviews with 40 women and 20 men attending Primary Health Care (PHC) clinics in Matlab were recorded on tape by trained male and female health workers. Tapes were translated into English and entered into the computer for data analysis. Presentations of symptoms were treated as "free lists" and entered into the ANTHROPAC computer programmes for compilation of relative frequencies. The transcriptions of interviews were subjected to systematic content analysis of salient variables.

Results: The study shows that both women and men report RTI symptoms, including possible STDs quite freely, but both female health workers and client women are reluctant to discuss female sexual behaviours. Discussion on male sexual behaviours is somewhat more open. In contrast, male clients and male health workers discuss sexual topics more freely. Information given by health workers is not always factually correct, and often involves value judgements and declarations concerning sexual behaviours.

Conclusion: To assess risk factors and other aspects of possible STDs, questions about sexual behaviours should be asked in the clinic, but women particularly will rarely acknowledge sexual activities, or even implicate their husbands, while men are apparently more open in their discussions and admissions concerning sexual behaviour. Current interview practices by health care workers may discourage open communication (because of judgemental attitudes). So, further training of the health workers seems needed in the area of sensitive sexual behaviour information, to get more frank discussions, and provide opportunities for more appropriate primary and secondary interventions concerning sexually transmitted infections.

Determinants of AIDS Awareness and Knowledge among Rural Men and Women in Bangladesh

Elisabeth L. Fulton, Nashid Kamal, Syed Masud Ahmed, and Monirul Islam Khan

Objective: Analyze baseline AIDS awareness in rural Bangladesh as related to socio-economic, demographic, communication and development access variables.

Methodology: A large sample (3,687 women and 2,272 men) was interviewed for the first round survey of the joint BRAC-ICDDR,B Research Project on Socio-economic Development and Human Well-being. Their AIDS awareness was statistically related to socio-economic and demographic variables, as well as to variables related to family planning, communication and development access. Four research cells within Matlab thana were included: one with only ICDDR,B intervention, one with only BRAC intervention, one with both, and one with none. The interviews were carried out between April and August 1995, but the analysis was carried out in the fall and winter of 1996-1997. Fourteen villages were chosen from the study area, and every household was represented by one woman for the women's questionnaire and one man for the men's questionnaire. Variables that had been found significant by preliminary analysis were regressed against AIDS awareness using logistic regression techniques.

Results: Seven percent of the female population and 16% of the male population were found to have heard of AIDS but 80% of these AIDS-aware women and 70% of these AIDS-aware men did not know how one gets AIDS or how to prevent it. Literacy was the most significant predictor of AIDS awareness, but communication with neighbours about family planning and a woman's technical understanding of the mechanism behind the oral contraceptive use also increased one's chances of being AIDS-aware. Being a member of an NGO was insignificant as a predictor of AIDS awareness for both groups.

Conclusion: The knowledge required for AIDS prevention was clearly inadequate in rural Bangladesh at the time of the survey, and there was a strong need for BRAC and ICDDR,B to increase their AIDS education efforts. A special emphasis on educating the illiterate population is recommended.

REPRODUCTIVE TRACT INFECTIONS: SOCIAL AND BEHAVIOURAL ASPECTS-II

Are We Intentionally Spreading STDs and HIV/AIDS?: A Lesson Learned from Professional Blood Donors

Sharif Md. Ismail Hossain, Ismat Bhuiya, and Kim Streatfield

Objective: Explore the high-risk behaviours of male professional blood donors (PBDs) for sexually transmitted diseases (STDs) and HIV acquisition, and the possibility of the transmission and spread of these diseases to blood recipients .

Methodology: Both qualitative and quantitative approaches were used for data collection. One-on-one in-depth interviews, observation on participant, and unobtrusive observation were the methods of data collection. A checklist and a semi-structured questionnaire were the data collection tools. The study used purposive sampling for selection of respondents and sites. In total, 288 respondents were interviewed. The study area comprised blood banks of different hospitals and clinics in four divisions (Dhaka, Chittagong, Rajshahi, and Khulna). This study was conducted during 1995-1996.

Results: Over three-quarters of the blood donors had multiple sex partners, and most of the partners were commercial sex workers (CSWs). Nearly three-quarters of the PBDs had engaged in premarital sex, over half of them with known individuals. About one-third of the blood donors had engaged in extramarital sex, mostly with floating and brothel sex workers. Over ten percent of PBDs had engaged in homosexual sex (men having sex with men), oral sex, anal sex with women or had a bisexual relationship. Surprisingly, one-seventh and over one-fifth of PBDs had been selling blood, knowing that their blood was positive for syphilis (VDRL) and hepatitis B (HBsAg) respectively.

Conclusion: Given their sexual behaviours, unsafe sex practices and the unreliability of the care sought for STD, this study concluded that there is a high chance of PBDs for acquiring STDs and HIV/AIDS. As blood is not screened, recipients of blood are, thus, at high risk for acquiring transfusion transmissible infections. This dangerous situation needs urgent intervention, with particular emphasis given on screening donated blood before transfusion, and information and education on safe-sex practices provided to donors.

The Population Council, Dhaka

Sexuality and Sexual Behaviours of Male STD Patients in Dhaka City

Md. Sharful Islam Khan[1] and Abu Jamil Faisel[2]

Objective: Explore in-depth sexuality and sexual behaviours of male STD patients in the context of STD/HIV/AIDS transmission in Bangladesh.

Methodology: Qualitative exploratory research design was employed. In-depth interviews were made, with a non-probability purposive sample of 15 male STD patients, using open-ended questions with an interview guideline. The Skin/VD Outpatient Department of Dhaka Medical College Hospital was selected for collecting the respondents who were already diagnosed and treated as STD patients. This study was conducted during August-October 1996. Content and contextual qualitative data analysis methodology was used. The variables from the 'Health Belief Model' and Dixon-Mueller's analytical framework for sexuality and sexual behaviours were measured along with the background characteristics.

Results: Sexual relationships were established with multiple partners. Mainly commercial sex workers and also friends of both sexes, were described as situational and opportunistic, occurring both before and after marriage with high frequency of contact. Evidence of practising vaginal, anal, and oral sex was stated (sometimes in group) under the influence of pornofilms mostly not using condoms was stated. They did not follow personal preventive and curative measures learnt from peers which were biomedically ineffective. There was a massive gap of knowledge about sexual and reproductive capacities, disease transmission, and safe sex practices. These were constructed by cultural, religious values and beliefs, influenced by peer pressure, gender role and power relations in the societal context.

Conclusion: All future strategies and interventions for prevention of STD/HIV/AIDS should be based on factual information regarding the dynamics of sexuality and sexual behaviours which are constructed in local socio-cultural, socio-economic, psycho-social and spiritual context. Sexuality education should be developed on a life-cycle basis and such education should start from a very young age both at institutional and non-institutional (community) level.

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Risky Sexual Behaviour in a Conservative Society

Ruchira Tabassum Naved

Objective: Explore risky sexual behaviour in a conservative society.

Methodology: Qualitative research methods, such as key informant interviews, focus group discussions, and case studies were applied. Social practices, particularly individual practices were explored through triangulation with different data collecting techniques. Risky sexual practices were identified through these exercises. The study was conducted in a rural area, north-east of Dhaka, during January 1995-September 1996. The study included female and male respondents who provided information on themselves as well as serving as key informants.

Results: Risky sexual behaviour is widespread in this otherwise conservative rural setting. Multiple-partner sexual practices are quite common among men. Their sexual partners are from both within and outside the villages. Within the villages, the partners are usually extremely poor, and destitute women turned into commercial sex workers, poor and abandoned women subject to forced sex, and a few women practising multiple partnership for the sake of pleasure. Commercial sex workers are the usual partners outside the villages. Though the formal attitude toward these practices is negative, the affluent were reported to cater to these practices. Homosexuality is prevalent in the area. It is not considered a risky behaviour as it does not lead to pregnancy.

Conclusion: In the context of a rapid spread of STD/HIV in the Indian subcontinent, mass awareness raising programmes on the effect of STD/HIV and mode of transmission of these diseases along with women's empowerment programmes might be a solution for changing the scenario.

Save the Children (USA), Dhaka, Bangladesh

Knowledge, Attitudes, and Practices (KAP) Survey among Commercial Sex Workers Residing in Taan Bazar Brothel

Julia Ahmed[*] and Sarah Hawkes

Objective: Identify the needs of an intervention programme, using data on socio-demographic characteristics, clinical histories, patterns of health care-seeking behaviour, sources of health care, knowledge about STIs/HIV and infection prevention among commercial sex workers residing in Taan Bazar brothel.

Methodology: The survey was conducted following random sampling techniques with a coverage of 346 respondents of the 2800 CSWs, using a pre tested and semi-precoded questionnaire by trained interviewers. Data were analyzed using SPSS software. The study was conducted in Taan Bazar brothel, Narayangonj, the largest brothel in Bangladesh, located 25 km from Dhaka city. The total brothel area consists of 18 buildings built on either side of a narrow lane, approximately a quarter mile long. The study was conducted during September 1995-June 1996.

Results: Although the women generally understand the nature of risk of RTI/STI/HIV/AIDS, their reported rate of primary prevention is surprisingly low. Rates of longer-duration abortion care and health care from local quacks are high. The study highlighted a high-level of unmet need for contraceptive, menstrual regulation services and access to high quality comprehensive reproductive health services within the close proximity of the brothel community. It further highlighted the urgent need for an education programme to change behaviour to reduce susceptibility to RTIs/STIs.

Conclusion: Research on various aspects of RTIs/STIs in Bangladesh is still in a very early stage. Interventions in relation to the spread of RTIs/STIs must be based on relevant information, including sexual behaviour and susceptibility to RTI/STI transmission. In the recent past, the Bangladesh Women's Health Coalition (BWHC) has developed a distinct programme to address the need of women at higher risk of getting RTI/STI and HIV. The above findings will help BWHC implement an effective programme aiming at exploring and managing the multifaceted context and consequences of RTI/STIs.

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A Community-based Survey of Commercial Sex Workers (CSWs) in a Brothel of Bangladesh on Knowledge, Intent, Trial, and Practice for Use of Condom

Swarup Sarkar[1], Florence Durandin[1], Smarajit Jana[2], Rezaul Hassan[1], Enamul Hoque[1], M.A. Quddus[1], and Nazrul Islam[3]

Objective: Find out the knowledge on STD/HIV, intent, trial, and actual use of condom among Commercial Sex Workers (CSWs) in a brothel setting.

Methodology: In a cross-sectional study of 300 CSWs (selected out of 600 women), each from alternative rooms of every house of the brothel, each woman was interviewed using an anonymous questionnaire for knowledge on STD/AIDS, risk perception for STD, intent, self-reported behaviour for trial and practice for using condom during the last 24 hours. In a semi-urban brothel, selected women who consented to participate were interviewed by project staff in a confidential setting in the room of the respondent. The study was conducted during June-July 1996. Participation rate was more than 90%. Knowledge on STD/AIDS and their mode on transmission and prevention, intent to use condom, self-reported use of condom during the last 24 hours were recorded.

Results: The results show that 66.9% and 87.5% of CSWs knew respectively about gonorrhoea and syphilis as sexually transmitted diseases. AIDS spread from sexual route, and healthy persons were known to 36.5% of the respondents. Thirty-six percent women knew about the protective role of condom in the prevention of STDs; 28% had intent to use condom while 11.8% women had tried condom; and 3.4% actually used condom in more than 50% occasions during the last 24 hours. Result of multivariate analysis for condom use will be presented.

Conclusion: Comprehensive education and culture-appropriate intervention with enabling strategies are required to ensure use of condom among CSWs.

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Health Care-seeking Behaviour of Floating Sex Workers in Dhaka

Kazi Golam Rasul and Yasmin H. Ahmed

Objective: Understand the health care needs of floating sex workers and use of available health care services.

Methodology: Floating sex workers were identified and interviewed through a structured questionnaire, administered by a trained interviewer. Three hundred nineteen sex workers were identified and interviewed in seven locations of Dhaka city. The interviews were conducted during 26 May-10 July 1996. Percentage distribution of various responses was studied.

Results: About 80% of the respondents had experienced some kind of reproductive health problem, with lower abdominal pain and discharge being most frequently reported. More than two-thirds of them had consulted someone for the problem; medical doctors were consulted by half of them followed by pharmacists (24%). About 25% of the respondents had spent between Tk 51 to 200 for treatment/services.

Conclusion: There is a high prevalence of RTI/STD problems among women under study. Interventions to reduce the problem need to be low-cost and accessible.

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Marie Stopes Clinic Society, Dhaka, Bangladesh

REPRODUCTIVE TRACT INFECTIONS: PUBLIC HEALTH AND EPIDEMIOLOGY-II

High STD and Low HIV Prevalence among Commercial Sex Workers (CSWs) in a Brothel of Bangladesh: Scope for Prevention

Swarup Sarkar[1], Florence Durandin[2], Debashis Mandal[3], G. Corbitt[3], and Nazrul Islam[4]

Objective: Assess the prevalence of certain selected STDs like chlamydia, gonorrhoea, syphilis, and HIV among commercial sex workers (CSWs) in a brothel setting.

Methodology: In a cross-sectional study of 300 CSWs (selected out of 600 women), each from alternative rooms of every house of the brothel, each woman was examined for chlamydia and gonorrhoea; and 150 consecutive blood samples were analyzed (for syphilis and HIV). Samples for HIV were unlinked immediately after collection of blood. Women recruited from the brothel were examined clinically at the brothel-based project clinic. Samples for TPHA/VDRL and HIV by ELISA were tested at IPGMR Hospital, Dhaka. Chlamydia and gonorrhoea were examined by PCR at UK from endocervical swab preserved and transported at -20° C at the end of the study. This study was conducted during June-July 1996. Selected subjects who consented were recruited. Participation rate was more than 90%.

Results: The results show that 60% of the women were TPHA-positive. VDRL (more than 1:8 dilution) was positive among 6.8% of the women. Twenty-seven percent of the women were positive for either chlamydia or gonorrhoea. None was found to be positive for HIV by ELISA.

Conclusion: Low prevalence of HIV and high prevalence of STD suggested potential rapid spread of HIV. Opportunity for prevention of HIV should not be missed.

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Antibiotic Sensitivity Pattern of Neisseria gonorrhoeae spp. among Females of High-risk Behaviour in Dhaka City

Yasmin Jahan[1], Mahbub Murshed[1], Md. Ahsanul Kabir[2], and Khurshida Begum[2]

Objective: Detect the prevalence of Neisseria gonorrhoeae itself among high-risk female population; augment, control strategies in Bangladesh against sexually transmitted diseases and strengthen existing syndromic case management (SMx) approach.

Methodology: A cross-sectional study was conducted among females of high-risk behaviour. Forty endocervical swabs were collected from females of high-risk behaviour irrespective of signs and symptoms of gonorrhoea. The subjects of the survey were inmates of vagrant homes aged 13-40 years, Mirpur.

Results: Among the 40 specimens, 8 were found positive for N. gonorrhoeae species. The organisms isolated were confirmed biochemically. Eight antimicrobial agents were used for in vitro antibiotic sensitivity testing. Eight resistance profiles were found among these 8 positive strains. None of these strains was found to be sensitive to all of the antimicrobial. These were also resistant to 2-7 drugs used in the study; 8 (100%) of the strains were resistant to cephradine; 5(62.5%) to penicillin, amoxycillin, and trimethoprim- sulphamethoxazole (SXT); 3 (37.5%) to erythromycin and pefloxacin; 2 (25%) to doxycycline; and 1 (12.5%) to ciprofloxacin. This shows that no single drug can be effective against all the strains.

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A Study on Vaginal Discharge in 150 Cases

A.Z.M. Maidul, A. Momin, A. Nargis, S. Tauhida, H. Tahmida, and A. Zubaida

Objective: Observe the prevalence of gonococcal infection.

Methodology: A prospective randomized study among patients seeking medical care for vaginal discharge was conducted in the Skin and VD Outpatient Department and Gynae & Obstetrics Outpatient Department in IPGMR, Dhaka during September-February 1993, 1994 and 1995 consecutively. One hundred fifty patients of childbearing age were included in the study. High vaginal swab (HVS) was collected and examined in wetfilm followed by Gram stain and culture.

Results: N. gonorrhoeae was found in 4.7% cases. Other isolates included: Gram-negative bacilli 66%, Monilia 48%, Gram-positive cocci 24%, Trichomonas vaginalis 13%, and Clue cell 4% . Many cases were found infected with multiple organism.

Conclusion: Organisms other than N. gonorrhoeae are found to be predominent in reproductive tract infections. Due importance is to be given in policy formulation to prevent and control reproductive tract infection and its consequences.

Skin and VD Department, IPGMR, Dhaka 1000, Bangladesh

Patterns of RTI/STD Diseases in Two Urban Clinics of Dhaka

Hashrat Ara Begum and Rukhsana Reza

Objective: Understand the distribution of various RTI/STD cases coming to the two clinics

Methodology: The medical records of all clients who presented to the Marie Stopes Clinics (located at Elephant Road and Mohakhali) during November-December 1996 were scanned. Clients with RTI/STD were identified, and the information contained in their forms--630 RTI/STD clients in the Elephant Road clinic and 230 clients from the Mohakhali clinic--were analyzed. Proportion of RTI/STD clients among all clients and distribution of various RTI/STD problems were the main outcome measures.

Results: About 23% of the clients presenting to the clinic had RTI/STD. Of them, about 21% of the cases were self-reported; the rest were diagnosed after screening. Vaginal discharge was the most common syndromic diagnosis (about 46% of the positive cases), followed by cervicitis and PID. Among the three major groups of clients i.e. sex workers, garments factory workers and general clients, to whom the clinics provided services, the prevalance of vaginal discharge was the highest among the garments workers, the highest cervicits among the general clients and PID among the sex workers. The five cases of genital lesions were all found among sex workers.

Conclusion: Awareness about RTI/STD problems needs to be created to encourage women to self-report symptoms and get access to treatment. Interventions must address not only identified groups in risk (e.g. sex workers), but also the community as a whole.

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Marie Stopes Clinic Society, Dhaka, Bangladesh

Prevalence of RTI/STDs in a Rural Area of Bangladesh

Md. Afzal Hussain, N.G. Banik, and G.S. Rahman

Objective: Assess the point prevalence of reproductive tract infections/sexually transmitted diseases (RTI/STDs) among the married women of reproductive age (MWRA) in a rural area of Bangladesh and their socio-economic characteristics and treatment-seeking behaviour.

Methodology: Thirty subjects were selected by cluster sampling technique. Data on knowledge, attitude, and practices (KAP), and the socio-demography were obtained by questionnaire survey at household level, and were brought to a clinical setting in the field for clinical examination. The laboratory tests of the materials collected from the subjects were done at Dhaka by the DIPHAM Research and Service Centre. The study was conducted in a remote rural setting in six unions of Nasirnagar thana under Brahmanbaria district of Bangladesh during 1 August 1995-12 October 1995. In total, 613 MWRAs who were not pregnant or taking allopathic medicines or menstruating at the time of clinical examination were studied. Laboratory confirmation of six selected RTI/STDs and correlation with the clinical assessment, reported consultation patterns and sources of treatment as used by them were the main outcome measures.

Results: Prevalence of RTI/STD was 56.1%, being almost uniform across the age range; 23.2% of these were STDs. The prevalence showed a strong positive association with mobility ($p<0.05$), economic conditions and occupation of the husbands. More than one-third of MWRAs with RTI complaints did not seek any treatments. Most of those who sought treatment used indigenous/traditional healers as source of treatment. Husband and other family members' advice, perceived cause and seriousness, confidence on and confidentiality of the service/service-provider, economic involvement, etc. influenced the treatment-seeking behaviour.

Conclusion: Mass awareness building and services for reproductive health care need to go beyond the traditional "high-risk" group and should include the general rural population. Socio-economic upliftment activities need to actively incorporate reproductive health care aspects. Otherwise, these effects may result in the increased prevalence of RTI/STDs.

Save the Children (USA), Dhaka, Bangladesh

Post-partum Genital Tract Infection among Mothers in Rural Bangladesh

Elizabeth Goodburn, A.M.R. Chowdhury, Rukhsana Gazi, Tom Marshal, and Wendy Graham

Objective: Identify the influence of maternity care on outcome in the post-partum period.

Methodology: The main part of the study was a prospective survey, with retrospective collection of data on pregnancy, delivery and post-partum period. The pregnant mothers were identified in their first trimester and were being followed up till the third month after child birth. BRAC and the London School of Hygiene & Tropical Medicine (LSHTM) jointly carried out a study on maternal morbidity in three unions of Manikganj district. A total of 2099 pregnant women in three unions of Manikganj district were studied. Post-partum genital tract infection was studied in relation to TBA training and clean delivery practice.

Results: It was found that 26% of the women had evidence of infection during the first two weeks after delivery and 14.7% during the 2-6 weeks after delivery. A significant association was found between the duration of the second stage of labor and the development of infection, and also between the birth attendant having put her hands in the vagina and the development of infection. Women who had a lower MUAC during pregnancy were more likely to develop infection in the post-partum period. If 'clean' is defined by the "three cleans", i.e. clean hands, clean cord, clean surface, then TBAs with training were significantly more likely to perform a clean delivery than an untrained TBA. However, training status of the birth attendant had no significant association with development of post-partum infection. Trained TBAs were found to be 20% more likely to insert their hands during labor than were untrained TBAs. It seems that insertion of hands, possibly associated with delivery complications, may be a potent factor for introducing or exacerbating infection. Some predisposing factors, such as maternal nutritional status, pre-existing reproductive tract infection, could be acting as a potent precursor of post-partum infection.

Conclusion: The study suggests that training of TBAs using the protocols currently existing in Bangladesh has no preventive effect for post-partum infection. A far more rigorous approach to the evaluation of TBA training programmes needs to be adopted for firm conclusions.

Research and Education Department, BRAC

REPRODUCTIVE AND CHILD HEALTH-I

Participation in Socio-economic Development Programme and Acceptance of Family Planning Methods

M Mustafa, Abbas Bhuiya, and Mushtaque Chowdhury

Objective: Examine how far BRAC's membership induce favourable attitudes toward family planning programmes and practice of birth control methods.

Methodology: A quantitative survey technique was used in data collection. Data were analyzed by using cross-tabulation and multivariate statistical techniques. Data were collected during April-August 1995 in 14 villages located in both ICDDR,B and BRAC's intervention areas in Matlab. All ever-married women of reproductive age in the study villages were included. Attitude toward family planning and family planning practices were the main outcome variables.

Results: The proportion of the respondents approving FP was higher among the BRAC members than the non-members. This was more prominent among the husbands. BRAC membership also induced more communication between husbands and wives about FP. Twenty percent of the BRAC members had discussion with their husbands compared to 15% among the non-members during the 30 days preceding the survey. The practice of FP was also higher among the BRAC members than the non-members in the area with only government family planning programme. The contraceptive prevalence rate was 53% and 36% among the BRAC members and non-members respectively in the area.

Conclusion: Participation in development activities induces favourable attitude to FP adoption. Higher practice in the absence of an intensive FP programme indicated that socio-economic development programme might increase FP practice substantially even at a lower level of FP service delivery.

Bangladesh Rural Advancement Committee (BRAC)

Management of Abortion in a Rural Area of Bangladesh

Abbas Bhuiya, A.M.R. Chowdhury, and Ayesha Aziz

Objective: Find out the nature of complications relating to abortion and management in a rural area of Bangladesh.

Methodology: In a retrospective study, all 91 induced and 77 spontaneous abortion cases that took place in the study area were included. Physical examination by a medical professional was carried out for 20 cases. Data were collected from 144 villages of Matlab thana during July-October 1995.

Results: Forty-three percent of the induced abortions were performed under the care of health professionals. Of them, 35% were done by an MBBS doctor and 8% by Family Welfare Visitor. The remaining abortions were performed by using indigenous techniques, such as inserting creepers, sticks, etc. Fifty-eight percent of the induced abortion cases and 61% of the spontaneous abortion cases had bleeding beyond 7 days after the abortion. Sixty-six percent of the induced abortion cases and 47% of the spontaneous abortion cases had fever after the abortion. Various other post-abortion complications were also reported. Eighty percent of the induced and 77% of the spontaneous abortion cases consulted with a health care provider for the treatment of the complications. In almost all abortion cases, the abortion management was hazardous.

Conclusion: Improper abortion technique and poor abortion management practices are highly prevalent. These may put women in life-threatening condition and in severe health consequences. Adequate attention should be given to abortion and its management for improving women's reproductive health.

Improvement in Female Survival: A Silent Revolution in Bangladesh

Ashish K. Datta and Radheshyam Bairagi

Objective: Investigate the trends in mortality and compare the improvement in survival between males and females in Matlab, Bangladesh.

Methodology: Data were taken from the Matlab Demographic Surveillance System for the period 1970-95. Mortality rates and sex ratios of mortality in different years for different age groups were examined graphically, and the findings were compared with the results from the model life tables to estimate excess female mortality.

Results: Mortality levels have improved in both MCH-FP and the comparison areas; however, the improvement was greater in the MCH-FP area. Life expectancy at birth was lower for females until 1985, when the life expectancy of females exceeded male in the MCH-FP area. This crossing similarly took place in the comparison area in 1992. The overall gain in life expectancy, has mainly been due to improvements in infant and child survival, and not specifically due to a decrease in the maternal mortality rate. It is estimated that female mortality could be reduced by 10% in infancy and by 35% in childhood if there was no discrimination against female children in Matlab.

Conclusion: Improvement in mortality was relatively greater for females and for children aged less than five years of age. The MCH-FP project interventions had a positive impact on this improvement, while a change in the societal attitude toward female child may have further contributed to this improvement. The study suggests that the MCH-FP programme is helpful in reducing mortality and the number of excess female deaths. Changes in the societal outlook toward these issues, particularly gender preference, are also essential for further improvement in these matters.

Childlessness as a Risk Factor of Divorce among Young Married Women in Bangladesh

Lutfun Nahar and Jeroen K. van Ginneken

Objective: Inability to bear a child within a few years after being married may lead to marital dissolution in Bangladesh. The objective of the paper is to test this hypothesis more specifically. The study was undertaken to find out whether delay in producing a child leads to marital dissolution after 3 years of marriage.

Methodology: Data from the Matlab Demographic Surveillance system were used for studying 1,405 girls who were married for the first time during the calendar years 1982-1985. Events like first birth after marriage, migration, marital dissolution (divorced or widow) and death were recorded during the three years after marriage. The likelihood of divorce was oriented to discrete-time hazard models where having a child is the single most important variable. The effect of having a child is evaluated in the model taking into account the other demographic and socio-economic variables.

Results: It was found that childbearing strongly and significantly reduced the risk of divorce. Other important findings are that the couple's socio-economic characteristics like education of both partners and male partners' previous marital status; and parents' characteristics like possession of land, occupation, and education influence the chance of divorce.

Conclusion: The finding that early childbearing after marriage of a girl prevents the marriage from being breaking down has an important policy implication for the family planning programme in Bangladesh. Contraceptive use among the newly-married couples is extremely low. Any attempts by the programme managers to motivate the newly-wed couples to adopt a contraceptive method to delay first birth may be counter-productive.

Risk Factors of Death due to Violence among Women of Reproductive Age in Rural Bangladesh

M. Kapil Ahmed, M.K. Barua, Jeroen K. van Ginneken, and Abdur Razzaque

Objective: Probe into the risk factors of death of women due to violence.

Methodology: The study has used longitudinal data from the Demographic Surveillance System (DSS) of Matlab, a rural area of Bangladesh, to examine some risk factors of deaths (suicide, homicide, and accidents), due to violence among women of reproductive age (15-44 years). A case-control study was designed to examine the risk factors of deaths due to violence. Deaths from violence and all other causes were compared with survivors. All deaths from violence (n=159) and other causes (n=1041) during 1982-1995 were included as cases. Same number of surviving women aged 15-44 years were selected randomly as the comparison group in July 1995.

Results: DSS data show that 13 percent of all deaths among women aged 15-44 years were due to violence. Risk of death from violence increases with age. Childlessness seems to be a risk factor for violence. Women (15-44 years) who did not have any children had an odds ratio of 4.79 of dying from violence compared to women who had 1-3 children. For other natural deaths, childlessness is not a risk factor. It was also found that unmarried (never married, widow and divorced) women had substantially higher risk of death from natural causes than married women, which is in the expected direction. The same is not true for deaths due to violence.

Conclusion: The findings corroborate with the observation and indicate that childlessness is a high risk factor of violence. Further qualitative studies should be undertaken to understand the situation why violence occurs among women. Maternal and child health and family planning programmes should have services for childless women so that they receive counselling and treatment for childbearing.

Do Female Education and Mobility Lead to Less Intra-family Abuse?

Mohammad Ahsanul Amin, Barkat-e-Khuda, Ariful Islam, and Ann Levin

Objective: Assess the impact of female education and mobility in determining the extent of female abuse in rural areas of Bangladesh.

Methodology: Data were obtained from an in-depth survey conducted in 1993 in six areas i.e., Sirajganj, Gopalpur, Abhoynagar, Fultala, Bagherpara and Keshobpur thanas in rural Bangladesh. In total, 10,368 eligible currently married women of 15-49 years age group residing in these areas have been taken as sample in the study. Both bivariate and multivariate analyses were done. Different kinds of abuse i.e., verbal abuse, physical violence, and threat of divorce, were the main outcome variables.

Results: Seventy-eight percent of the total sample had encountered verbal abuse, 42% had experienced physical violence and 18% had received threat of divorce from their husbands. Women were more likely to be abused if they were older, not educated or less mobile and if their husbands were less educated and less wealthy. Women's ownership of land and husband's occupation were also statistically significant predictors of abuse.

Conclusion: The study suggests that female education in Bangladesh should be further strengthened, and various IEC messages about the problems of abuse should be used for supplementing the effect of education. More activities that promote social mobility of rural women should also be undertaken.

FINANCING AND DELIVERY OF HEALTH SERVICES

Toward Sustainable MCH-FP Service Delivery System: Use of CBD and Outreach Service Deliveries

Yousuf Hasan, Ali Ashraf, Barkat-e-Khuda, and Bruce Caldwell

Objective: Examine health service use by rural women at CBD and its alternative fixed sites, such as cluster and satellite clinics over time during the intervention period.

Methodology: Under the cluster point approach, services are provided by a field worker to a group of about 50 women at a centrally located house in the neighborhood, rather than at the homes of individual clients. Satellite clinics and EPI (SC+EPI) sites were combined to offer services from one site with the consideration that it could be a viable facility with a wide range of MCH-FP services, if it is closer to women's residence. One such site was established for every 1,000 population or 200 MWRAs. Abhoynagar thana of Jessore district and Mirsarai thana of Chittagong district were selected for the study which was conducted during January 1995-June 1996. The methodology included interviews of currently married women in 11,016 households by using a structured questionnaire through longitudinal surveillance system. Method-specific trend of contraceptive prevalence from different service delivery points was examined in the study.

Results: Contrary to the initial fears that contraceptive use may decline in the cluster areas as opposed to CBD, it has stayed at about the same level in the intervention union in the high-performing area, and has increased in the intervention union in the low-performing area. SC+EPI as a source of family planning methods has increased in the area where CBD is weak. SC+EPI is the major source of clinical methods, particularly injectables, which from a programme perspective is positive because of longer term effects and being more reliable. While trials on CBD injectable delivery have proved successful, there are advantages in SC+EPI delivery as the female paramedic—rather than the field worker.

Conclusion: To attract more women to clusters, some additional services may be needed, and the service-providing capacity of the field workers should be increased with an appropriate training. Delivery of family planning from SC+EPI sites should also have benefits in side-effect management, and in the long-term, in reducing drop outs. This is not possible at doorstep and difficult at this stage at cluster.

Cost of MCH-FP Service Delivery: An Analysis of Concerned Women for Family Planning Branches

Aye Aye Thwin, Subrata Routh, Ann Levin, Md. Abu Yousuf, Anwara Begum, Zahidul Quayyum, and Nadia Barb

Objective: Determine the unit cost of MCH-FP services provided by the Concerned Women for Family Planning (CWFP), and identify areas for cost reduction and reallocation of resources for better efficiency

Methodology: A tool was developed to assist the analysis of production costs for a comprehensive set of MCH-FP services in CWFP. The field-level programme managers collected information on recurrent costs of labour, drugs, vaccines, other supplies, rent and utilities from six service delivery sites from routine service records. The costs of different inputs were further apportioned into cost centres to determine unit cost per service (in takas) based on personnel time allocation and service volume. The study was conducted in April-September 1996. Six branch units of CWFP, namely Rajshahi, Dhaka, Chittagong, Khulna, Tangail, and Magura, were included in the study.

Results: The analysis showed that the costs for recruiting new acceptors of family planning methods were much higher i.e. almost 2-3 times more than resupply costs. Personnel costs account for 65-70 percent of the production costs. For services produced at low volume, the personnel costs for administration and management outweighed those for direct service delivery. Differences in catchment areas and managerial practices resulted in variations in costs among the service delivery sites.

Conclusion: The information highlights the need for alternative service delivery strategies that improve cost-effectiveness. The analysis informs that it is possible to conduct cost analyses through routine service records data. Further work is necessary to develop inexpensive techniques for estimating personnel time allocation accurately. Cost analyses provide a useful basis for budgetary planning and the development of strategies for improving programme sustainability.

Coordination and Monitoring of Urban Health Services: A Needs Assessment of the Health Department of Dhaka City Corporation

M.A. Bhuiyan, S.U. Alamgir, C. Tunon, J. Uddin, and S. Nasreen

Objective: Assess the capacity of the Health Department of DCC to plan, monitor, implement and coordinate maternal and child health activities, including family planning.

Methodology: The study was conducted as part of the technical assistance provided by the MCH-FP Extension Project (Urban) to Dhaka City Corporation (DCC). The study used data from various sources. Inventories of staff and resources were conducted in zonal offices of DCC. Workshops, jointly organized by the Project and Dhaka City Corporation, were held with various categories of staff. During these workshops, special methodologies were used for identifying and prioritizing problems, analyzing causes and exploring solutions. The participants also completed a questionnaire which inquired into departmental practices concerning coordination, planning, job descriptions, human resources development, information systems, and logistics. In addition to the above sources of data, the project staff also made observations during meetings of DCC officials with representatives from other service delivery agencies at the zone level.

Results: The current focus of DCC in the area of MCH-FP is almost entirely circumscribed to the provision of immunization services. The study revealed major problems limiting the capacity of the Health Department to fulfill its role in monitoring and coordination of urban health services at the primary care level. Among others, the problems relating to unclear roles, responsibilities and lines of authority; lack of effective planning, monitoring and supervision systems; shortage of skilled staff and inadequate logistics support to the zone level.

Conclusion: The Health Department of DCC needs to be restructured if it is decided that it will assume a more active role in monitoring and coordinating urban primary health care activities. This new role would represent an expansion on the current focus of the Department on the delivery of immunization services and would require more consistent coordination with providers in the non-government voluntary and commercial sectors. Above all, decisions on any expansion on the role of the municipality need to be accompanied with clearly defined policies and support to DCC from relevant agencies in the health and in the local government sectors.

Effect of the Introduction of Contraceptive Pricing on its Use

Ann Levin, Mohammad Ahsanul Amin, Khorshed Mozumder, and Rumana Saifi

Objective: A contraceptive pricing experiment is underway in two unions: Rajghat Union of Abhoynagar and Dhum Union of Mirsarai. This study investigates the effects of this cost recovery on the use of contraception in these areas.

Methodology: Data were taken from two sources: (1) a baseline survey of the use pattern of eligible couples in the intervention unions, and (2) SRS data that were collected from a sample of contracepting couples every two months on expenditure, source of method, and client perceptions on the user fees. Both univariate and bivariate methods of analysis were used.

Results: The results indicated that the majority of users perceived the fees for contraception to be justified. While the introduction of fees did not affect the contraceptive use levels, it had an effect on contraceptive-seeking behaviour. The percent of users that obtained their methods from static clinics rather than at the home increased.

Conclusion: Nominal contraceptive pricing does not have any significant effects on use, although it does affect the choice of provider to obtain contraceptives. The study has policy implications for the introduction of user fees, suggesting that the judicious application of fees by the GoB for contraceptives can be tried at the union level.

Willingness and Ability to Pay for MCH-FP Services in Urban Bangladesh

Zahidul Quayyum, Aye Aye Thwin, Abdullah H. Baqui, Shams El Arifeen, and M.A. Majumder

Objective: Examine the willingness and ability to pay for selected MCH-FP services among urban dwellers in Bangladesh. The information would be useful to determine user fees for services without adversely affecting service use.

Methodology: Data were collected through household surveys of clients and field workers of an NGO: Concerned Women for Family Planning (CWFP). The respondents were randomly selected from client registers, and non-users from the neighbouring households of clients. The study was conducted in the CWFP service delivery sites in urban Dhaka, Chittagong, and Rajshahi in October-November 1996. Data were collected on users of pills, condoms, and injectable services, antenatal care, child health care, and other services of CWFP. Willingness and ability to pay for different bids of prices for MCH-FP services provided by CWFP were examined.

Results: In general, a substantial increase in service charges for pills and condoms under present conditions at CWFP facilities was acceptable to more than 80% of the clients, most clients were ready for an increase in service charges from Taka 20 to Taka 30 for antenatal care. More than 80% expressed the willingness to pay a nominal charge of Taka 5 for child immunization. Clients from Rajshahi were of the lowest socio-economic status, who nonetheless, expressed a higher willingness to pay for most services than in Dhaka and Chittagong.

Conclusion: Pricing mechanisms that consider demand-related aspects of willingness to pay can facilitate setting prices that would protect, rather than deter, the poor from services. This, in turn, would provide a sound basis toward improvement in programme sustainability.

Sharing of Needles and Syringes in the Expanded Programme on Immunization (EPI) in Bangladesh: Potential Threats, Opportunities, Costs and Benefits

Ali Ashraf, Mohsin U. Ahmed, Mizanur Rahman, and Barkat-e-Khuda

Objective: Assess field workers' observance of the recommended protocol for sterilizing reusable needles and syringes during the immunization of infants and mothers at the EPI sites in rural Bangladesh. In addition, the study discusses the management problem with regard to logistics for sterilization in the implementation process and recommends a solution.

Methodology: Observation on participants at the immunization sites of 12 rural sub-districts in south-eastern Bangladesh was carried out during July 1995-June 1996. The activities of the 156 male Health Assistants (HA) and 64 female Family Welfare Assistants (FWA) at the immunization sites were observed.

Results: Observation on the 172 immunization sites revealed that sterilizers for needles and syringes were not carried to 21% of the sites. In addition, stoves were not available at 68% of the sites. While the workers at 71% of the sites claimed that they sterilized the needles and syringes at their homes, they were not following the recommended protocol.

Conclusion: This issue has a tremendous public health significance since Bangladesh is located in the epicentre of an impending AIDS epidemic in Asia. Studies have shown that reusable equipment is four times as expensive as disposables. The potential for using disposables instead of reusable needles and syringes merits attention.

RECENT DEVELOPMENTS IN DIARRHOEAL DISEASE RESEARCH

Monitoring of Diarrhoeal Diseases by Spatial Analysis and Exploration of Environmental Factors

J. Myaux, J. Chakraborty, and M. Ali

Objective: Explore the potential of Geographic Information System (GIS) for monitoring of diarrhoeal diseases in a community.

Methodology: Areas with high prevalence of diarrhoea were mapped and compared visually with various thematic maps of the study area. To assess the geographic variation of acute watery diarrhoea in children aged 0 to 5 years in rural Bangladesh, all cases of 'cholera-like' diarrhoea in 1989 were plotted on the map.

Results: A clustering pattern was noticed and validated by a non-parametric clustering test for non-homogeneous population. Several areas with risk for the disease were identified. In these areas, the point prevalence (8.7 per 1,000) was notably higher than other areas (0.41 per 1,000; 95% confidence interval, 15.55-29.30). The persistence of the areas with risk and the dynamics of the spatial distribution over the years were explored by superimposing those areas from 1989 on the dot maps of diarrhoea cases for 1990 and 1991. By visual impression, it was observed that several areas would remain at higher risk over time. The same technique was used for correlating the location of diarrhoea cases to the presence of the flood control embankment, proximity to the bazaars and concentration of the Hindu communities. The areas with risk were matched with the distribution of the catchment areas of the community workers to look at possible reporting bias from the workers. The density of population is not clearly related to the presence of acute watery diarrhoea in the community.

Conclusion: The study shows the potential of GIS in communicable disease control. The technique of overlaying thematic maps is a useful complementary approach for a rapid assessment of particular health situations in communities.

Treatment of Rotavirus Diarrhoea in Children with Immunoglobulins from Immunized Bovine Colostrum

S.A. Sarkar, T.H. Casswall[1], N.H. Alam, J. Albert, G.J. Fuchs, D. Mahalanabis[2], and L. Hammarströ m

Objective: Evaluate therapeutic use of immunoglobulin from immunized bovine colostrum (HBC) containing a high titre of antibody against all rotavirus serotypes in the treatment of rotavirus diarrhoea in children.

Methodology: This is a randomized double-blind, placebo-controlled clinical trial on children with diarrhoea due to rotavirus. The children were randomly assigned to treatment with either 10g of HBC containing 2g of anti-rotavirus immunoglobulins, dissolved in water, divided into 4 doses daily during 4 days or the same amount of a placebo preparation. The study was conducted in the Clinical Research and Service Centre of the International Centre for Diarrhoeal Disease Research, Bangladesh during April 1995-December 1996. Boys aged 4-24 months with a history of acute diarrhoea of less than 48 hours were selected for the study. The daily stool rate (ml/kg), intake of oral rehydration solution (ml/kg.d), stool frequency (number/day) and intestinal pathogens were monitored daily for 4 days.

Results: The children who received HBC required significantly less oral rehydration solution ($p=<0.05$) and had less daily and total stool output compared to those of the placebo group ($p=<0.05$). The clearance of rotavirus from the stool was also observed earlier in the treatment group (mean day 1.5 vs 2.9, $p=<0.001$).

Conclusion: Anti-rotavirus immunoglobulins from hyperimmunized cows is effective in the treatment of children with acute rotaviral diarrhoea. The results suggest that passive immunotherapy is useful not only prophylactically but also therapeutically.

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Intra- and Extra-cellular Water Distributions during Rehydration in Cholera and Non-cholera Patients

I. Kabir, M.I. Hossain, G.J. Fuchs, M.J. McCutcheon, J.O. Alvarez, and M.A. Khaled

Objective: Estimate the intra- and extra-cellular body water compartments during rehydration of patients with cholera and non-cholera diarrhoea by bioimpedance analyzer.

Methodology: The study was done in the study ward of the Clinical Research and Service Centre of ICDDR,B. Total body water (TBW), intra-cellular water (ICW), and extra-cellular water (ECW) of severely dehydrated adult patients were measured with a dual frequency bioimpedance analyzer at different phases of rehydration. Fluid compartments between cholera and non-cholera patients were compared.

Results: Cholera patients gained more TBW than non-cholera patients during recovery. Unlike patients with non-cholera diarrhoea, the gain in cholera patients was mainly contributed by the intra-cellular water compartments (ICW = 1.5± 1.6 L vs. 3.0± 1.2 L respectively, $p<0.01$). It was also observed that the recovery of intra-cellular water compartment in cholera patients occurred rapidly within the first two hours after starting infusion.

Conclusion: Different dynamics of body water compartments in cholera compared to non-cholera patients as observed in this study may contribute further to understand the mechanism of dehydration in diarrhoeal disease which might be helpful for improved case management.

Cow's Milk Allergy in Children: Association with IgG and IgE Antibodies to Milk-Protein and Presentation of a Case

Tahmeed Ahmed, R. Sumazaki*, M. Shibasaki, and H. Takita*

Objective: Investigate the association between cow's milk-specific IgG and IgE antibodies and immediate and delayed types of cow's milk allergy (CMA), and the selectivity of the IgG and IgE antibodies in reacting with different protein components of cow's milk in the two types of CMA. A case of Heiner syndrome, characterized by CMA, hemoptysis and pulmonary hemosiderosis is also presented.

Methodology: The study was conducted in the Department of Paediatrics, University of Tsukuba, Japan. IgG and IgE antibodies to cow's milk and its component proteins were investigated in patients with delayed type CMA (n=6), immediate type CMA (n=25) and normal subjects (n=90) by ELISA and Sepharose-RAST respectively. Reactivity of serum IgG and IgE antibodies to the protein components of cow's milk was examined by Western blotting.

Results: Delayed and immediate type of CMA are associated with high titres of IgG and IgE antibodies to cow's milk-proteins respectively. Six different proteins in cow's milk react with serum IgG and IgE. b -Lactoglobulin, a -casein and b -casein are the most antigenic proteins in cow's milk. IgG and IgE antibodies did not show any selectivity in reacting with the component proteins of cow's milk in the two types of CMA.

Conclusion: Delayed and immediate type of CMA are associated with high titres of IgG and IgE antibodies to cow's milk-proteins.

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In situ Characterization of Inflammatory Responses in the Gut Mucosa in Shigellosis

Dilara Islam, Bela Veress[*]*, Pradip Kumar Bardhan, Alf A. Lindberg*[*]*, and Birger Christensson*[*]

Objective: Analyze the local gut inflammation, morphological changes and mediators that may be involved in regulatory mechanisms of cell activation and cell proliferation.

Methodology: The above analyses were done immunohistochemically in rectal mucosal biopsies at the acute phase and at convalescence in shigellosis. Rectal biopsies from 25 Shigella dysenteriae 1 and 10 S. flexneri-infected patients and 40 controls were studied. Patients were recruited from the outpatient section at the Clinical Research and Service Centre of ICDDR,B during January 1992-July 1993. The frequencies of proliferative cells ($Ki67^+$ cells) and p53 immunostaining cells were analyzed. Inducible nitric oxide synthase (iNOS) and endothelial NOS were assessed by immunostaining. In addition, the frequencies of apoptotic cells and $CD68^+$ cells that engulf apoptotic cells or those that may themselves become apoptotic were assessed.

Results: Extensive inflammation was observed in the acute phase biopsies. In spite of clinically resolved disease, a significant proportion of the patients still had morphological signs of mucosal inflammation compared to controls. Data suggested that in acute shigellosis activation of p53 and iNOS may contribute to the local inflammatory responses, and the major mode of death is apoptosis.

Conclusion: The results indicate that Shigella-induced inflammation is associated with a complex series of cellular reactions in the rectal gut mucosa persisting long after clinical symptoms have resolved.

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Impacts of an Integrated Water-Sanitation Programme on Hygiene and Health: A Case Study from Bangladesh

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Objective: Bangladesh has achieved remarkable success in developing safe water supply. Over 95% of the rural population now has a safe drinking water source within the convenient distance. However, the country lags far behind in sanitation with only 44% of the rural households having sanitary latrines. To improve this situation, the Department of Public Health Engineering (DPHE), in collaboration with UNICEF, undertook an integrated programme in Barisal district during 1989-1991. In 1994, the Environmental Health Programme (EHP) of ICDDR,B studied the impacts of that programme on hygiene and health. The main objectives of the study were to examine the use of water and sanitation provisions, the performance of home-made latrines, the role of various partners in the programme and to assess health impacts.

Methodology: The study was conducted on randomly selected 900 families with children aged less than five years. Conducted over a one-year period, the study included three thanas of Barisal district. Data were collected by interviews and observation. A small number of environmental sampling (hand and water contamination) was included.

Results: More than 80% of the adult members of the studied families claimed they used some kind of sanitary latrine. Children's faeces, however, were mostly disposed of indiscriminately. The single most important stated reason for installing and using latrine was to control scattering of faeces by poultry. Almost all the families used tubewell water for drinking, but the use of tubewell water for other domestic purposes was quite limited. Bacteriological tests on stored drinking water and handwash samples showed high contamination. About 70% of the latrines were in good condition. Most respondents (over 50%) reported that government workers from the Ministry of Health and Family Welfare and DPHE were the main promoters.

Conclusion: Government workers from the Ministry of Health and Family Welfare and DPHE were reported to be the main promoters in water and sanitation issues. Poor handwashing practices showed a significant association with diarrhoeal disease prevalence.

REPRODUCTIVE AND CHILD HEALTH-II

Maternal Health Care Practice in Rural Bangladesh: Determinants of Antenatal Care-seeking Behaviour

Md. Mafizur Rahman, Barkat-e-Khuda, Thomas T. Kane, Khorshed A. Mozumder, and Md. Masud Reza

Objective: Investigate the maternal health-care practices in rural Bangladesh, and examine the association of antenatal care-seeking behaviour of rural women with their socio-economic and demographic characteristics.

Methodology: The data were drawn from a cross-sectional survey. In analyzing data, descriptive statistics, as well as multivariate regression methods were used. The survey was conducted in four rural thanas of Bangladesh during September 1993 to March 1994. The sample comprised 10,368 rural women of reproductive age. The main outcome measures include: (1) percent women seeking antenatal care during their last pregnancy; and (2) odds ratios of logistic regression measuring association of seeking antenatal care with socio-economic and demographic variables.

Results: The data show that 41% of the rural women did not seek antenatal care during their last pregnancies. Among those who did, only 14% consulted qualified professionals, such as doctors, nurses, and midwives. Almost all deliveries took place in homes, and most of these were conducted by untrained traditional birth attendants or relatives in the most unsafe way. The young, educated, and employed women are more likely to seek antenatal care than the older, uneducated, and unemployed women. Poor women are also less likely to seek antenatal care.

Conclusion: Emphasis should be given on IEC activities to educate the community, particularly the uneducated and poor women, on the need for regular antenatal check-ups and safe delivery by qualified health personnel. To ensure better use of trained TBAs, they should be linked with the health and family planning service delivery system. Female education and employment should also be promoted.

Socio-cultural Factors Influencing Early Age at Marriage and Early Childbearing in Rural Bangladesh: A Qualitative Study

Fazilatun Nessa

Objective: Assess the socio-cultural factors influencing early age at marriage, timing of first child birth and non-use of contraceptives before first birth.

Methodology: Data were collected on 3247 women aged less than10 years of age from the Sample Registration System and 12 Focus Group sessions. Abhoynagar thana where the MCH-FP Extension Project has been functioning, was the study site. The study period was December 1995-March 1996. Husbands and fathers of the sample women were also included in the study. Age at first marriage, age at first birth, and contraceptive use before first birth were the main outcome measures.

Results: Differences between the knowledge about age at first marriage and age at first child birth in comparison with the actual age at first marriage and age at first child birth were observed. Important reasons for an early marriage were: prevailing socio-cultural and religious values, criticism of community about grown-up unmarried girls, fear that girls may be victims of unsocial elements, like elopement and rape, lack of knowledge about legal age at marriage, demand for dowry for older girls and poverty of father. Important reasons for early childbearing were: husband's and in-laws' desire, religion, lack of knowledge about different aspects of contraceptives and infrequent visits by health and family planning workers.

Conclusion: Although most respondents viewed that age at first marriage and first childbirth should be increased, this was not seen in practice. The study suggests that efforts should be made in the community to delay age at first marriage and encourage the use of contraceptives among the newly weds.

Childhood Mortality in Dhaka

Henry Perry, Suraiya Begum, Rafique-ul Islam, and Abdullah H. Baqui

Objective: Assess the levels of and influences on mortality in a representative urban population.

Methodology: Field interviewers collected vital events and sociodemographic information on 3,889 children through quarterly home visits. Mortality rates were calculated for population subgroups according to age, sex, and socioeconomic status. Data were obtained from a stratified multistage cluster sample of households from the Zone 3 population of approximately 400,000 persons living in the Bakshi Bazar, Lalbagh, and Rayer Bazar areas of the city. Data were collected between November 1994 and March 1996. Sociodemographic information was collected for all households. Mortality rates were calculated for specific age groups by sex and socioeconomic status. The main outcome measures were infants aged 1-4 year(s), and under-five mortality rates for 1995.

Results: The infant mortality rate was 95.9 per 1000 live births. Eighty-two percent of all deaths among children aged less than five years occurred during the first six months of life. Children living in the poorest households of Zone 3 are almost three times more likely to die before the age of five than are other children. One-third of the under-five mortality in Zone 3 can be attributed to severe impoverishment.

Conclusion: Pregnant women and children aged less than six months living in the poorest households in Zone 3 constitute a target group for mortality prevention efforts. This group constitutes only 1% of the Zone 3 population. Hence, ensuring that this target group receives a basic package of services aimed at reducing the risk of early childhood mortality seems appropriate and feasible.

Neonatal and Early Post-neonatal Morbidity and Mortality in Mirzapur, Bangladesh

Kh.Zahid Hasan,K.M.A.ZIZ,A.K.Siddique,B.P.Pati,E.Roy,M.N.Rahaman,M.Ali, F.Haque,and R.B.Sack

Objective: Study the pregnancy outcome and neonatal and post-neonatal morbidity and mortality in Mirzapur, Bangladesh.

Methods: A census was conducted in ten villages of Mirzapur in Tangail district during August-September 1993 to study ALRI and diarrhoea in a cohort of newborns. Pregnant women were identified and followed longitudinally till child birth. A detailed delivery history and status of child at birth, including birth weight, was recorded. Twice a week household surveillance provided information on morbidity. The causes of death were established by a physician.

Results: During a 12-month period, 312 pregnant women were identified, of whom 288 (92.3%) had a live-birth and 24 pregnancies were wasted. Five newborns had congenital abnormalities and two were twin-births. Neonatal morbidity was largely due to infections (85%), such as URTI, ALRI, skin infection, cord infection, and conjunctivitis. Thirteen percent birth was associated with birth asphyxia, jaundice, and hypothermia. Morbidity during early post-neonatal period was due to infectious diseases. Of the 288 live-births, 22 died during first 3 months (73 % during neonatal period). Birth asphyxia (50%), prematurity (19%), and pneumonia (13%) were the major causes of death among the neonates. During the first 90 days, 25 children had ALRI needing hospitalization, and of them, 4 died. Low birth-weight was significantly associated with mortality ($p<0.012$) during the first three months of age.

Conclusion: Birth injury, infectious diseases, and low birth weight are associated with morbidity and mortality during neonatal and early post-neonatal period. The issues for improvement of delivery, child care and low birth-weight need to be adequately addressed to reduce their ill effects.

Current Infant Feeding Practices in Rural Bangladesh

Shameem Ahmed, Sadia D. Parveen, Ariful Islam, and Farzana Sobhan

Objective: Assess the current knowledge and practices of rural mothers regarding infant feeding in Bangladesh.

Methodology: Data for this study were obtained from a special survey of 1,919 women in two rural thanas: Abhoynagar and Mirsarai, field sites of the MCH-FP Extension Project (Rural). The study was conducted during June-September 1996. All married women who had delivered within a year of the study period were included. Data were recorded on women's knowledge about infant feeding and their breast feeding practices.

Results: Of the women interviewed, 12 percent stated that newborns should be given colostrum first, and only about 27 percent knew that exclusive breast feeding should be given for at least five months. Of the 1,699 women who had live-births, only 10 percent actually gave colostrum, in practice, as the first food to their newborns; most others gave prelacteal feeds. Only 19 percent had initiated breast feeding immediately after birth, 35 percent on the first day and about 45 percent on the second day or later; one percent did not breastfeed at all. Only 16 percent of the children were exclusively breastfed at five months of age.

Conclusion: The study suggests that the on-going national breast feeding campaign, which started five years ago, needs to be further strengthened in rural Bangladesh.

MCH-FP Programme Performance in Rural Chittagong: A Comparison Between Areas Covered by Government and NGO Workers

Mizanur Rahman and Barkat-e-Khuda

Objective: Examine differences in female family planning (FP) behaviour and use of MCH-FP facilities between areas served by government and NGO workers.

Methodology: More than 10% of the positions of Family Welfare Assistant (government-employed workers who deliver FP services at the doorstep) are vacant in Chittagong district. NGO workers provide services in these areas. This study is based on information collected during 1995-1996 on about 6,000 married women of reproductive age in nine thanas of Chittagong district. Desired fertility, contraceptive use, and use of Satellite Clinic (SC) and Health and Family Welfare Centre (H&FWC) were modelled in logistic regressions to find the net difference between government and NGO areas after controlling for the effects of socio-demographic and programmatic variables. Areas where there are both types of workers were excluded.

Results: Desired fertility was significantly higher in NGO than the government areas. Contraceptive use was 28% lower after controlling the effects of all control variables, including that of desired fertility. The use of SC was about 50% lower in NGO than the government areas, while use of H&FWC was similar.

Conclusion: The findings do not support the popular belief that the FP performance of the NGO workers is better than that of the government workers. Lower SC use in NGO areas probably indicates a lack of adequate coordination between the government and NGO field activities. Improved mechanisms of reviewing NGO field activities and government-NGO coordination at the field level will enhance use of MCH-FP services.

POSTER PRESENTATIONS

Assessment of People's Knowledge on HIV/AIDS in Matlab

Hashima-e-Nasreen, Abbas Bhuiya, Mushtaque Chowdhury, A.K.M. Masud Rana, and Sayed Masud Ahmed

Objective: Assess the level of knowledge of rural people about HIV/AIDS regarding what AIDS is, its transmission, how it is not spread, high-risk behaviour and high-risk population before and after giving a short, simple and effective training on HIV/AIDS; explore the level of dissemination of knowledge among the BRAC members and neighbours, and husbands and wives.

Methodology: About 800 households of the BRAC members and neighbours were surveyed randomly using a pretested structured questionnaire covering socio-demographic information, respondents' knowledge of HIV/AIDS, such as what AIDS is, its transmission, how it is not transmitted, prevention, high-risk behaviour and high risk population in 14 selected villages of Matlab-DSS area. During analysis, comparison was made between the respondents before and after the training. Appropriate statistical tests were done where needed. SPSS for Windows statistical package was used for data analysis.

Results: The analysis showed that overall knowledge of the BRAC members on AIDS had increased, but the knowledge of the neighbours and husbands had increased very little in some criteria. From the findings it is understood that dissemination of knowledge among the BRAC members, neighbours and husbands was very poor**.**

Conclusion: Information concerning the local people's knowledge of AIDS would be helpful for the policy makers in formulating new policies to launch IEC campaigns in a more specific way.

Male Sexual Illness in the Culture of Rural Bangladesh

Hasanur Rahman, Bert Pelto, Habibur Rahman, S. M. Nurul Alam, Sarah Hawkes, Ashraful Alam Neeloy, Jim Ross, and G. N. I. Faisal

Objective: Develop a deeper understanding of how rural males in Bangladesh describe and define sexual illness (including STDs): their patterns of communication about sexual illness, concepts of causation, and treatment-seeking behaviours.

Methodology: In-depth interviews were conducted with 60 males who came for consultation concerning their sexual health problems to male clinics in the Matlab area. In addition, several group discussions and follow-up interviews were also conducted, as well as key informant interviews with different kinds of health care providers. Data were gathered during August 1996-January 1997.

Results: Men presented a wide range of problems: nocturnal emissions and other semen emission, abnormal (including small) penis, impotence, pain in urination, as well as boils and lesions and other symptoms that suggest sexually transmitted infections. A number of men recognized contact with sex workers as a potential cause of their symptoms; but many men did not identify any specific cause of their problems. Masturbation was also frequently mentioned as a cause of sexual health problems. Concerning treatment-seeking behaviours, a very "pluralistic" pattern was reported, including many individuals resorting to home remedies, treatment by *kabiraj*, spiritual healers, and other "traditional" practitioners, but also many contacts with both qualified and less qualified allopathic health services, as well as homeopathic practitioners.

Conclusion: The data suggest that knowledge of sexual transmission of infections is common among these rural males, but many of their sexual problems and anxieties (concerning impotence, excessive semen-loss, and "weakness") are not necessarily connected with STDs. Those psycho-social anxieties about sex (including "masturbation anxiety") as well as the general lack of condom use and lack of knowledge concerning appropriate treatment options, point to a need for much more dissemination of reproductive health information in this population.

State of RTI/STD Research in Bangladesh: Current Issues and Future Needs

S.M. Nurul Alam and Hasanur Rahman

Objective: Highlight the general lack (until very recently) of effective research on RTIs/STDs in Bangladesh, and describe current developments in this field. The review focuses on the main lines of current research and provides ideas regarding future needs and priorities in relation to the apparent increases in STDs in Bangladesh, as well as the threats posed by the HIV epidemic in South and Southeast Asia.

Methodology: An extensive search of published and unpublished literature on RTI/STD research has been conducted. Contacts and interviews with researchers in NGOs, government agencies, and other organizations in the area of reproductive health have been made. Discussions with resource persons in various sectors of research in Bangladesh have provided useful feedback for developing ideas concerning future research directions and priorities in the field of RTIs/STDs.

Results: A review of the past and current research shows that there is a recent growth of new data, as well as some increased sophistication in research in the complex, sensitive area of RTIs/STDs. The data show a need for integration, compilation, and rapid dissemination as the new materials become available, because some relevant materials are not available in published form. Many gaps in the available data call for immediate attention.

Conclusion: The identification of major gaps in knowledge of the prevalence, cultural belief patterns and treatment-seeking behaviours concerning RTI/STDs in various sectors of the Bangladesh population will be useful as a guide to future research. Also, methodological approaches and guidelines will be suggested for consideration by researchers interested in contributing to this area of study.

Cultural Model of White Discharge (Sada Srab) among Rural Matlab Women

Tamanna Sharmin, Kamrun Nahar, S. M. Nurul Alam, Bert Pelto, and James Ross

Objective: Gain a deeper understanding of how rural women describe and define reproductive tract infections (including STDs): their patterns of communication about these illnesses, explanations of causes, and varieties of treatment-seeking behaviours for these problems.

Methodology: In-depth interviews were conducted with an opportunistic sample of approximately 50 women in Matlab: 40 of them were "cases" examined at the sexual health clinic of Dr. Sarah Hawkes, and 14 were selected from the same villages as the non-cases. The in-depth interviews probed their vocabularies of symptoms, explanations of possible causal factors, and full history of treatment-seeking behaviours.

Results: Women identify reproductive tract infections, particularly "white discharge" (leucorrhoea) as a very common health problem. A large number of different labels are used in speaking of the problem, but *sada srab* (white discharge) is the most common vocabulary used when presenting with symptoms at the clinic. Women often discuss the problem with their husbands, as well as some of their female in-laws. A large number of women link their RTIs to family planning methods and to a variety of other causes unrelated to sexual transmission. On the other hand, many women are aware of possible sexual transmission, but are very reluctant to admit that route of cause; very few women reported that their husbands might be involved with extramarital sexual contacts. Treatment priorities begin with home remedies, beyond which various traditional and modern providers are used, though women try hard to avoid physical examination.

Conclusion: The reproductive tract infections, particularly white discharge, seriously affect women's daily activities, including sexual relations with their husbands. The lack of programmatic attention, until recently, to women's reproductive health problems has contributed to a very large unmet need for health care, as well as a large gap in reproductive health information that should be met through greatly expanded service delivery as well as new approaches to information dissemination.

Management of Reproductive Tract Infections

M. Tariq Azim, Shams El Arifeen, Selina Amin, Anwara Begum, and Tahmina Sarker

Objective: Strengthen the quality of RTI case management through standardized service delivery protocols.

Methodology: Two tools--the RTI protocol and the RTI checklist -- have been developed as part of Basic Service Package (BSP) and Clinic Information System (CIS) interventions of the MCH-FP Extension Project (Urban) of ICDDR,B respectively. The RTI protocol has been adopted from the WHO's protocol on syndromic management of RTI/STD cases and was tested in three NGO clinics and three government dispensaries in Dhaka city. In two of the NGO clinics where BSP is implemented, the other intervention, CIS is also being tested. In these two clinics, the RTI checklist has been incorporated in the client-oriented, card-based record-keeping system. This checklist was developed based on the RTI protocol of BSP intervention. The two tools are, therefore, inter-related.

The study is based on a pret-est and post-test design: O_1 X_1 O_2 X_2 O_3, where X_1 represents the introduction of and training on RTI protocol, and X_2 represents the introduction of the RTI checklist. The study evaluates changes in RTI case management practices of NGO paramedics after training on the protocol and then after introduction of the checklist. Data have been collected from the client records in the clinics, the CIS cards.

Results: With the use of RTI checklist as part of the clinic record-keeping system, the syndromic case management of RTI according to the protocol has improved.

Conclusion: The RTI checklist supports the use of RTI protocol and reinforces the training provided to the paramedics on the syndromic management of RTI. Together with protocols and training on the syndromic management of RTI, service providers require a user-friendly information system that supports good RTI management practices.

Addressing Partner Management as a Primary Element of STD/RTI Services: A Male Involvement Approach

Syeda Nahid Mukith Chowdhury and Joachim Victor Gomes

Objective: While RTIs have infected individuals for centuries, it is only with the shift to a reproductive health agendum that their diagnosis and treatment has gained attention at the national levels. To reduce the spread of sexually transmitted RTIs, it is essential that individuals at risk understand the modes of transmission, the signs and symptoms, and the consequences of untreated infection. Identifying and treating partners of infected individuals is an absolutely essential aspect of appropriate clinical services.

Methodology: Male patients attending the two STD clinics in Dhaka are the subjects of the research. The health record of every patient includes nature of complaints, previous medical history, clinical diagnosis, sexual history, and partner management. The study encompasses awareness building and education on STDs/RTIs in the field, health education and counseling at the clinic, and diagnosis and treatment. From the ongoing study, the case history of the first 900 male patients who attended the clinic was examined. The study period is July 1996 through June 1997 in two branches of Marie Stopes clinics in Dhaka.

Results: The outcome of the study will identify factors essential for effective partner management through men. Along with clinical diagnosis, the information includes the results of in-depth interviews of the clients' sexual history: partners, condom use, and treatment patterns.

Conclusion: The results would help define an integrated approach toward STD services. It will provide further knowledge on clients' perceptions of STDs, the language they use to describe conditions and the importance they ascribe to appropriate treatment and behaviour modification. Finally, IEC developed for this male STD intervention will be evaluated to determine whether it did raise awareness of sexual responsibility for oneself, partners and children.

The Population Council, Dhaka, Bangladesh

Breaking the Silence

Sadia A. Chowdhury, Zia Uddin, and Disha Ali

Objectives: Get an insight into what rural women think about RTI/ STD and the conventional attitude of the rural society toward this very common problem of women.

Methodology: A series of six focus group discussions was conducted among general mass of women, volunteers (Sasthya Shebikas) and traditional birth attendants. Mymensingh (near Dhaka) and Dinajpur (far from Dhaka)--two of the BRAC's working area--were chosen for discussions. The discussion was conducted in Mymensingh between 11 and 13 February 1995 and in Dinajpur between 6 March 1995 and 8 March 1995. Forty-eight respondents from two areas comprising general group of women, volunteers (Sasthya Shebika) and traditional birth attendants of that community. Perceptions of the rural women regarding their own health problems were recorded.

Results: Most respondents (90%) stated vaginal discharge mainly whitish discharge to be the commonest problem of a woman in those communities. The frequently used local terminologies are more or less same in both areas which indicate some sort of universality within the country. Women of Mymensingh are more familiar with sexually transmitted diseases, either by name or by symptoms. Treatment-seeking pattern is dominated by financial condition, accessibility to and availability of female service providers, and their first choice of service provider is local *kabiraj* who prescribe herbal remedies.

Conclusion: Involving the volunteers (Sasthya Shebika) in raising awareness and integrating an intervention in the existing health programme that would better address the problems relating to RTI/ STD.

Bangladesh Rural Advancement Committee (BRAC)

Barriers to Seeking Treatment for Sexually Transmitted Diseases among the Dhaka Slum Dwellers

Keith Sabin, Sarah Hawkes, Mahbubur Rahman, Khaled Ahsan, Lutfa Begum, Shams El Arifeen, and Abdullah H. Baqui

Objective: Quantify barriers to seeking treatment for sexually transmitted diseases among the Dhaka slum dwellers.

Methodology: Slum households were selected by cluster sampling for questionnaire and specimen collection. Respondents were asked to report reproductive health problems and whether they sought treatment for those problems. Blood specimens were collected from all participants and tested for syphilis and hepatitis B. The Urban Surveillance System clusters located in Mohammadpur, Lalbagh, Sutrapur, Kotwali, and Demra thanas were selected for the study during 31 July 1996-30 October 1996. Five hundred and forty men and 993 women completed the questionnaire.

Results: The most common barriers to treatment-seeking for men, regardless of complaint, were cost and lack of severity of symptoms. Women cited cost and shyness as the most important barriers to treatment-seeking. 68.5% (370/540) men reported dysuria, 34% percent of whom did not seek treatment. Primary reasons for not seeking treatment were: lack of severity (48.5% of responses) and expense (32%). 50% men reported urethral discharge, and most (61%) had not sought treatment. Women reported not seeking treatment for vaginal discharge (58%; 373/645), genital itching (53%; 183/389), lower abdominal pain (71%; 275/390), dyspareunia (95%; 225/238), dysuria (70%; 259/371), genital prolapse (75%; 76/101), and genital ulcers (31%; 15/49). 40% of the responses noted expense while 29% referred to shyness as the main barriers to treatment seeking. 92% of the TPHA-positive GUD patients (33/36) sought treatment, 94% (31/33) reported that they were cured after treatment. However, 77% (24/31; 16 of the pharmacy patients and 5 of the MBBS patients) tested positive for primary syphilis. Those who did not seek treatment reported it was too expensive to afford.

Conclusion: Important barriers to treatment-seeking for STDs exist in the population under study. Clinics must offer inexpensive, adequate treatment for STDs. Treatment for genital ulcer disease is probably inadequate in these communities. Slum residents need education to recognize symptoms requiring treatment, and women's concerns for privacy must be met.

Comparing Physicians', Health-care Workers' and Laboratory Diagnoses of RTIs/STIs in Matlab

Kaniz Gausia, Shamim Sufia Islam, Nazmul Alam, Farid Ahmed, J. Chakraborty, Andres de Francisco, and Sarah Hawkes

Objective: Compare diagnoses made by physicians and other health-care workers against laboratory findings in women symptomatic for possible RTI/STI infections.

Methodology: Women complaining of possible RTI/STI symptoms reporting to subcentres in Matlab were included in this study carried out in the last three months of 1996. Results from a total of 94 non-pregnant women aged 15-49 years are available. Each woman was seen by a physician and a trained senior health assistant or nurse-midwife. The physician and paramedic recorded their diagnoses separately and in confidence of each other. Clinical diagnoses were then compared to laboratory-based aetiological results.

Results: Of the 94 women seen, 22% had bacterial vaginosis (BV), 15% had Candida and none had cervical infections (*N. gonorrhoeae* or *C. trachomatis*) diagnosed according to laboratory criteria. Diagnostic rates for these infections were: 17%, 13%, and 3% respectively for the physician and 22%, 14%, and 2% respectively for the paramedic staff. Cross-tabulations of diagnoses with laboratory results showed that in all cases the sensitivity of the clinical diagnosis was low, but specificity was very high. Over-diagnosis is common, except in the case of bacterial vaginosis which was diagnosed more correctly by the physician.

Conclusion: These results show that even in the case of highly trained staff, the accuracy of clinical diagnosis is low. Over-diagnosis of both endogenous infections and sexually transmitted cervical infections was common, although in all cases the specificity of diagnosis was high. To improve the sensitivity and accuracy of diagnosis, improved clinical training and simple diagnostics may be needed.

Introduction of Safe Blood Transfusion in Rural Hospitals of Bangladesh

Mohsin U. Ahmed, Shameem Ahmed, and Mobarak H. Khan

Objective: Introduce safe blood transfusion procedures at the thana level to prevent haemorrhage-related maternal mortality.

Methodology: As a part of the Emergency Obstetric Care (EOC) intervention of the MCH-FP Extension Project (Rural), providers at the Thana Health Complex (THC) were trained on blood transfusion, and necessary logistics were supplied. Data were collected from hospital registers, regarding the indications of blood transfusion, general characteristics of blood recipients and donors, and the results of blood screening tests of the donors. This study was conducted in the Thana Health Complex, Abhoynagar, Jessore during June 1995-December 1996. Patients who received emergency transfusion on admission, and persons who donated (or intended to donate) blood for the said recipients were studied. The number of haemorrhage-related cases referred to higher health centres, through introduction of safe transfusion procedures at the THC. Providers can motivate the relatives of the patients to donate their blood. Screening of donors for STDs will help ensure a quality transfusion.

Results: Seventy-six transfusions have been given at THC, of which 29 were maternity-related. All of the donors were volunteers. Each was screened for syphilis and hepatitis B, although none was found positive. Referral to the higher health centre has dropped. A cost-recovery initiative has been implemented.

Conclusion: Safe blood transfusion made available at THC, and the first referral rural hospital will go a long way in reducing the country's maternal mortality. However, with the rising risks of AIDS, it is also worthwhile to do HIV screening for all blood transfusions, even at the thana level.

Determinants of Reproductive Tract Infections among IUD Users

Mohsin U. Ahmed, Mizanur Rahman, Shameem Ahmed, Yousuf Hasan, Mehrab A. Khan, and Indrani Haque

Objective: Determine whether the complications of IUDs are suggestive of reproductive tract infections (RTI).

Methodology: In-depth and baseline survey data of the MCH-FP Extension Project (Rural) were collected from more than 500 married women of reproductive age who were currently using an IUD. Side-effects and their management were analyzed. The quality of the government and MCH-FP IUD services was reviewed through the use of several sources. The study was conducted in Abhoynagar, Sirajganj, Mirsarai, Satkania, and other rural thanas of Chittagong district, the field sites of the MCH-FP Extension Project (Rural). The data sources include in-depth survey 1993 and 1994; Needs Assessment Survey, 1996 and Baseline Survey of Basic Service Package 1996. The unit of analysis was married women of reproductive age currently using an IUD, and the outcome measures were provider-induced complications of IUD.

Results: About half of the women currently using an IUD had complications. There was evidence suggestive of RTIs among a large percentage of the IUD clients. Hence, it was shown that providers of MCH-FP services had not maintained a consistent level of quality care.

Conclusion: There is a need to improve the quality of IUD services, which, in turn, would reduce complications.

Validity of Syndromic Management in Diagnosis of Chlamydia and Gonorrhoea in Brothel Setting of Bangladesh

Swarup Sarkar[1], Florence Durandin[1], Nahid Siddiqui[1], Debashis Mandal[2],G. Corbitt[3], Fazlul Karim Chowdhury[1], Samiran Panda[4], and Nazrul Islam[5]

Objective: Find out the sensitivity and specificity of complaints of vaginal discharge to detect chlamydia and gonorrhoea infections among commercial sex workers (CSWs) in a brothel setting in Bangladesh.

Methodology: In a cross-sectional study of 300 CSWs, each from alternative rooms of every house of the brothel selected out of 600 women, was examined, clinically interviewed and examined using speculum and was validated against chlamydia or gonorrhoea detected by PCR from endocervical swab. Women recruited from a brothel were examined clinically at a brothel-based project clinic. Women with an evidence of vaginal and/or endocervical discharge and/or history of vaginal discharge were clinically detected. Samples of endocervical swab were preserved and transported at -20° C to laboratory of the UK for PCR. The study was conducted during June-July 1996. Selected subjects who consented were recruited. Participation rate was more than 90% outcomes. Evidence of vaginal discharge by history, endocervical discharge by clinical examination with or without history of vaginal discharge and chlamydia or gonorrhoea by PCR were the outcome measures.

Results: Less than half of the women (sensitivity 40%) having chlamydia or gonorrhoea were detected by history of discharge. Nearly one-third of patients (specificity 70%) would be wrongly detected and treated when they are actually not infected by chlamydia or gonorrhoea.

Conclusion: Further studies are required to evolve a suitable diagnostic strategy for STDs among high-risk population groups.

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Experiences of Integrating RTI/STI Case Management with MCH-FP Services

Sukanta Sarker and Abu Jamil Faisel

Objective: Examine the feasibility of integrating RTI/STI case management with MCH-FP services.

Methodology: A simple descriptive analysis of the data obtained from urban MCH-FP clinics in Bangladesh has been used. This study was conducted during January-December 1996. All clients (both male and female) who attended the clinics with RTI/STI problems were studied. Five variables have been looked into, namely: why clients came to the clinics, provisional diagnosis made **using the syndromic approach, completion/compliance of treatment, partner management, and willingness to pay.**

Results: Data analysis is yet to be completed.

Conclusion: On preliminary review of the data it can be said that the integration of RTI/STI case management with MCH-FP services needs some more work on IEC, community education, counseling of clients and follow-up.

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An Explanatory Model of Infertility among the Urban Slum Population in Dhaka City

Papreen Nahar, Anjali Sharma, Keith Sabin, Lutfa Begum, Khaled Ahsan, and Abdullah H. Baqui

Objectives: Provide preliminary information on the people's concepts and vocabulary regarding infertility. Perceived causes and subsequent treatment-seeking behaviour among adults in urban Dhaka slums are of particular interest.

Methodology: Freelisting of vocabulary with 60 male STD cases and 60 female STD cases as well as 15 case studies were conducted by trained interviewers. The freelisting data were analyzed on ANTHROPAC to draw a composite list of local terms, and the case studies were manually scoured for perception of causes, social consequences and resort to treatment. The study was conducted in the Urban Surveillance System clusters located in Mohammadpur, Lalbagh, and Sutrapur areas of Dhaka city during 31 July- 30 October 1996.

Results: There is limited vocabulary for infertility in this population. However, there are terms that label both men and women infertile. The causes people refer to range from the supernatural to physiological and psychosexual disorders. Women are more frequently blamed for the lack of children in the family. Though largely believed to be incurable, the *kobiraj* and religious healers are often approached for treatment.

Conclusion: Preliminary analysis suggests that women bear the largest burden for infertility and suffer serious emotional and social consequences. There should be health care facilities that address their concerns about infertility, counsel on the biological and pathological basis of infertility as well as awareness of STDs as one of the causes of infertility.

Reproductive and Sexual Health Problems as Perceived by Women and Men in a Rural Area of Bangladesh

Ayesha Aziz, S.M.A. Hanifi, and Abbas Bhuiya

Objective: Obtain a list of the local names of the reproductive/sexual health problems and to reveal the perception of similarity and severity of the diseases.

Methodology: Freelisting and pilesorting techniques were used in data collection. Average salience and severity was calculated to assess the importance of the diseases. Multidimensional scaling technique was used for studying the perceived similarity of the diseases. Data were collected in June 1996 from the three unions of Chakaria thana in Cox's Bazar district. Men and women of reproductive age were selected as key informants.

Results: Respondents were aware of various types of reproductive/sexual health problems in the study area, and they considered that the number of sexual/reproductive health problems is smaller among men than women. The total number of male and female diseases mentioned by the informants were 14 and 42 respectively. Among the male diseases, *dhatu(sta)* (spermatorrhoea) scored the highest salience index followed by *gonorrhoea*, *akkham*, *akshira/bakshira/* (hernia/hydrocele) and *purushangey gaa* (ulcer in the genitalia). Among the female diseases, *sada srab* (white discharge) scored the highest followed by *mashikey gulmal* (menstrual problem), *sutika* (post-partum diarrhoea and burning in the hand and feet), *sharam jaigaigaa* (ulcer in the area of shame), *shawpna dush* (emission of vaginal fluid at night in dream), and *adinna nasta howa* (abortion). There were indications of variation in male and female perception about the importance and severity of the male and female diseases.

Conclusion: The local names of the sexual/reproductive health problems can be used in further studies. Intervention priorities can be determined on the basis of the perceived importance and severity of the diseases. Both men's and women's perceptions about the similarity of the diseases can be used in designing health education curricula and material.

Environmental Determinants for Sexually Transmitted Diseases (STD) in a Rural Setting of Bangladesh

J. Myaux, M. Ali, and Sarah Hawkes

Objective: Examine the dynamics of the location of STD cases in the community and possible environmental risk factors.

Methodology: Data from a community-based survey in Matlab in 1995/6 were aggregated at the level of the *bari* and linked to georeferenced files from the Geographic Information Sytem (GIS). STD cases were plotted on the map against the location of *bazaars* and highly populated areas, identified by using isoline technique. A spatial analysis was performed on the cases based on a nearest neighbour test (Cuzick, 1992).

Results: Of the study sample of 1,929 *baris*, 98 presented a case of STD; 43 chlamydia, 16 *N. gonorrhoeae,* and 10 *Trichomonas vaginalis*. The spatial distribution presented no pattern except for *Chlamydia* cases (Cuzick test: k=3; $p<0.02$). The location of the cases was neither significantly related to the proximity of a *bazaar* nor to a crowded environment ($p=0.17$).

Conclusion: Very little is known on environmental determinants of STD, particularly in the rural community. Surprisingly, the prevalent cases were quite homogenously distributed across the study area, with only a slight clustering in case of chlamydia infection. Neither the proximity to the Gumti river nor the presence of a bazaar, communication indicators, determined the location of the cases. There is no evidence that cases would be more frequent in highly-populated neighbourhoods.

Evaluation of Field Workers Accuracy in the Estimation of Fluid Volumes

K. Zaman, J. Chakraborty, Md. Yunus, Andres de Francisco, D.S. Alam, and K.M.A. Aziz

Objective: Determine the accuracy of direct observation in estimating fluid volumes by the field workers.

Methodology: Eleven female field workers who had at least 12th grade of education were recruited for a study to observe children's fluid intake during diarrhoea at their homes. As part of the study, they were trained at Matlab between August and September 1996 in observational techniques to estimate the quantity of fluid in different containers commonly used by the rural mothers at Matlab. The test was conducted in October 1996. Measured quantities of water were placed in ten containers of varying shapes and sizes. The field workers were then asked to estimate the quantities of water in each container. Measured volumes of water were then removed from each container, and the workers were again asked to estimate the quantity of water remaining in each container. For each field worker, the absolute error, percentage of error for each estimate, and Pearson's correlation coefficients were calculated between estimated and actual fluid volumes.

Results: The mean volume for the actual mass of water was 80 ml and the workers' mean estimated volumes ranged between 78 ml and 82 ml. The mean absolute error ranged from 4.4 ml to 11.4 ml. In percentage, the field workers' mean errors ranged between 8% and 18%.The field workers' estimate of fluid volumes were highly correlated with actual volumes (r = 0.97-0.99). There was a slight tendency of the field workers to underestimate the difference between the two volumes, but the degree of error was only a few ml.

Conclusion: The field workers showed a high level of accuracy in their estimations of fluid volumes. Following successful training, direct observation can be used as an alternative to actual measurement in estimating children's fluid intake, particularly during a diarrhoeal episode.

Case Studies of Abortion: Who, Why and How

Shameem Ahmed, Ariful Islam, Sadia D. Parveen, and Parveen A. Khanum

Objective: Assess the characteristics of rural Bangladeshi women who have had abortions, their care-seeking behaviour, reasons for their abortion, and their contraceptive use.

Methodology: The analyses were done using a dataset of 127 exit interviews. The women were first interviewed immediately after they had received services and then followed up at home. The study was conducted at Abhoynagar and Mirsarai thanas in the intervention areas of the MCH-FP Extension Project (Rural) of ICDDR,B during June-December 1996. Women who had attended the Thana Health Complex in the study areas for abortion/MR services, or for management of complications were studied. The care-seeking behaviour of women having abortions, the reasons for induced abortion, and the contraceptive use of the women were the main outcome measures.

Results: Two-thirds of the abortions were induced, and among them, the woman was the decision-maker in 35% of the cases. Forty-three percent of them were performed by women themselves or by untrained persons in the village. Sixty percent developed complications, like fever, haemorrhage, and sepsis. Eighty percent of the women were not using contraceptives at the time of conception, and more than half had repeated abortions. Seventy-three percent had at least one male child, and a majority stated that they did not want any more children.

Conclusion: Appropriate programmes need to be designed for the unmet need for contraceptives in rural Bangladesh.

A Methodology for Planning the Distribution of Primary Health Care (PHC) Facilities to Improve Access to Basic Health Services in Urban Areas

S.U. Alamgir, Shams El Arifeen, C. Tunon, and J. Uddin

Objective: The urban Primary Health Care (PHC) facilities are managed by multiple organizations belonging to Dhaka City Corporation (DCC), Directorate of Health Services DHS), Directorate of Family Planning (DFP), non-government organization (NGOs) and a large for-profit private sector. There are gaps and overlaps, limiting availability and access to basic services due to improper locations. Most facilities provide only one or two services. Cross-referral among these facilities is weak. The MCH-FP Extension Project (Urban) of ICDDR,B developed a methodology to assist local managers and decision makers in developing a reorganization plan of facilities to improve the availability of, and access to basic services, particularly in poor areas. The objective of this study is to describe this methodology.

Methodology: In two zones of DCC, a wardwise detailed inventory was done of all government and NGO facilities. The inventory included mapping, staffing pattern, service availability and use of the facilities. Estimations on what should be the requirement of provider/clinic based on population size, proportion of poor and assumptions on current and expected clinic use were done with the help of a matrix. The calculated needs of each ward was compared with the actual availability of services, and shortages or excesses of resources were identified. The findings were discussed in a day-long participatory workshop of decision makers and local managers of the organizations of Zone 3 of DCC. As a consequence, a reorganization plan with recommendations for changes was developed.

Results: Although there were sufficient number of facilities, these were poorly distributed. Recommendations included relocation of EPI and Family Planning staff from wards with surplus to wards with inadequate services, assignment of full-time medical officers in DCC dispensaries to strengthen curative services and measures to strengthen cross-referral.

Conclusions: Considerable willingness and potential exist for more effective use of present resources to improve the availability of, and access to basic services.

Diarrhoea Case Management in Urban PHC Clinics Before and After the Implementation of the Diarrhoea Management

Selina Amin, Abdullah H. Baqui, Rasheda Khanam, Shams El Arifeen, and C. Tunon

Objective: Measure the effect of implementing diarrhoea management programme in the urban primary health care clinics.

Methodology: To expand the range and to strengthen the quality of service in the urban PHC clinics, the MCH-FP Extension Project (Urban) of ICDDR,B provided training to health service providers of three (CWFP) clinics and three government dispensaries on the use of standardized protocols which include providing training on an adapted diarrhoea protocol. Two government dispensaries and two CWFP clinics have been selected as the comparison site. The intervention clinics are being monitored by a physician by visiting the clinics once every 15 days to see whether the providers are following the protocols. Data are collected on diarrhoea services from both intervention and comparison clinics.

Results: More specific and correct diagnosis was made, and appropriate treatment was more often given in the intervention clinics after introducing the protocol. Use of metronidazole for the treatment of diarrhoea has been decreased from about 41% as baseline to about 2% in a government dispensary and from about 8% to nil in a CWFP clinic. Providers more often give advice regarding taking more fluid and food in intervention clinics. However, it is difficult to change providers' practice regarding the use of antibiotics, especially the use of metronidazole for mucoid diarrhoea.

Conclusion: The preliminary findings suggest that management of diarrhoea can be successful with the provision of other services in the urban primary health care clinics, following a standardized protocol provided proper supervision and monitoring systems are in place. Use of protocol guides the providers to give an appropriate treatment to the patients, and thus, reducing drug misuse.

Distribution of Basic Services for Child and Reproductive Health in Dhaka

C. Tunon, M.A. Mazumder, Abdullah H. Baqui, M.A. Bhuiyan, and Shams El Arifeen

Objective: Describe the distribution of essential services for child and reproductive health in Dhaka City Corporation area and to analyze the current patterns of distribution of services in relation to the population.

Methodology: The data used in this analysis came from an inventory of major providers of health and family planning services carried out by the MCH-FP Extension Project (Urban) of ICDDR,B from April to June 1995 in Dhaka City Corporation area. Trained interviewers collected data from facilities in all zones of DCC. The collected information included the name and address of the facility, the clinic and field services available, the staff and the managing organization. Information on private practitioners and pharmacies was only collected in Zone 3 of DCC. Findings on the location of services and facilities were compared with estimates for the population in different zones of the city, including the estimated slum population.

Results: The inventory was one of the first attempts to identify health and family planning facilities in the entire area administered by the municipality. The facilities identified in the inventory were managed by a variety of government agencies and private concerns. The managing organizations include the Directorate of Health Services, the Directorate of Family Planning, Dhaka City Corporation, various non-government organizations working in health and family planning, and a growing commercial sector. Most facilities were managed by NGOs and the commercial sector. The inventory revealed an unplanned distribution of services that may tend to generate gaps in the coverage and inefficient use of resources. There is a large commercial sector providing services at all levels, including primary care, but with a mainly curative focus. NGOs and government agencies, on the other hand, tend to provide more preventive services. Facilities managed by NGOs are also more likely to provide a basic package of MCH-FP services. The inventory confirms the existence of many government clinics dedicated to the provision of one or two services, though their staff is trained to provide more comprehensive care.

Conclusion: This exercise helped highlight policy issues and research areas that need to be addressed toward the establishment of a more cost-effective system for delivering quality basic services in urban areas. The findings suggest that it is necessary to improve the physical distribution of the facilities to increase access for the urban poor to essential clinical services for child and reproductive health. Above all, it is critical to look into the factors limiting the availability of a package of basic services from government facilities. More research needs to be conducted on the range of services provided by the commercial sector.

Determinants of Reported Morbidity among Married Women of Reproductive Age in Rural Bangladesh

Yousuf Hasan, Mizanur Rahman, Masud Reza, Barkat-e-Khuda, and Mohsin U. Ahmed

Objective: Examine the pattern and determinants of reported morbidity.

Methodology: A survey instrument was used for collecting data from the married women of reproductive age in two rural thanas. Data were analyzed using the bivariate and multivariate statistical technique The study was conducted in Abhoynagar thana of Jessore district and Patiya thana of Chittagong district during June-August 1996. 2624 married women of reproductive age group were asked about their sufferings from any illnesses during the last 15 days as well as 90 days. Reported illness during the last three months and within two weeks, type of treatment providers, socio-economic characterstics of women were the main outcome variables.

Results: About 10% of the women reported illness while about 26% reported illness during the last 90 days. About two-thirds of the morbid cases were due to any infections. About 80%, of those who reported illness during the last 15 days received treatment from unqualified allopaths and non-allopath practitioners. Educated, Muslim, and landless women whose husbands stay abroad and those who are using injectable contraceptives and IUD reported more morbidity compared to others. Abhoynagar reported higher morbidity than Patiya.

Conclusion: Health-conscious people seem to report higher morbidity. Poor women and women residing without their husbands probably feel unhealthy and report higher illness. Having higher illness among injectables and IUD users reflect side-effects of these contraceptives. Treatment by mostly unqualified doctors necessitates strategies to make public health service deliveries more effective.

Alternative Service Delivery Strategies for MCH-FP Services in the Urban Areas: Preliminary Findings

Subrata Routh, Shamim Ara Jahan, Shams El Arifeen, Anwara Begum, and Aye Aye Thwin

Objective: Develop alternative strategies (to door-step delivery) for cost-effective delivery of MCH-FP services.

Methodology: This is an operations research intervention of ICDDR,B's MCH-FP Extension Project (Urban) in partnership with the Concerned Women for Family Planning. The effect of the intervention was evaluated through quasi-experimental and non-experimental research designs. Two alternative strategies are currently being field-tested in Dhaka city -- one at Hazaribag (services delivered from a static Primary Health Care Clinic-PHCC), and the other at Gandaria (with a transitional arrangement of distributing pills and condoms from Community Service Points-CSPs). The study is still under experimentation since January 1996, and is planned to continue till mid-1998. The current analyses are based on an evaluation conducted in October 1996 using data from clinic and field service records and population-based sample surveys.

Results: Use of the primary health care clinic is increasing over time. Thirty-five clients were served daily in October compared to around 19 in March 1996. PHCC-based strategy shows an increase in clinical contraceptive use: from 32% to 35% during the reporting period. Contraceptive Prevalence Rate increased from 63% to 67% at Hazaribag, and decreased from 55% to 53% at Gandaria. Labour costs and travel time were reduced by 18%.

Conclusion: In urban Dhaka, door-step distribution of contraceptives can be replaced by clinic-based service delivery strategies with targeted home visits to provide information and motivation. The clinic-based system enables a holistic approach toward addressing client needs through a package of services.

Acinetobacter Bacteraemia in Patients with Diarrhoeal Disease

M.I. Hossain, I. Kabir, W.A. Khan, and G.J. Fuchs

Objective: Determine the incidence and significance of bacteraemia by *Acinetobacter* spp. in hospitalized diarrhoeal patients and antimicrobial susceptibility.

Methodology: In a descriptive study, records of all patients positive for *Acinetobacter* spp. in blood culture were reviewed. Data analyzed included: age, sex, weight, season, clinical and laboratory information, final diagnosis, and outcomes. The study was conducted at the Clinical Research and Service Centre (CRSC) of ICDDR,B from 1 January through 31 December 1994. All admitted patients of CRSC with *Acinetobacter* bacteraemia were studied. Main outcome measures were: clinical pattern, morbidity and mortality of patients with *Acinetobacter bacteraemia*.

Results: Of all positive blood cultures, 171 (27%) were due to *Acinetobacter* spp., of which 138 were significant bacteraemia (91 community acquired and 47 nosocomial) constituting 2.2% of all admitted patients. Acinetobacteraemic children were severely malnourished. Incidence was the lowest during post-monsoon to early winter months. *Acinetobacter* bacteraemia-associated mortality was twice (16%) than that in all other admitted patients (7.7%) ($p<0.005$) and accounted for 4.7% of all hospital deaths during the study period.

Conclusion: Bacteraemia caused by *Acinetobacter* spp. is an important and potentially preventable cause of death among patients with diarrhoeal disease in Bangladesh. Earlier identification of these patients and prompt institution of effective antibiotic therapy could save many of these lives.

Folic Acid in Children with Acute Watery Diarrhoea: A Randomized Double-blind Controlled Trial

*H. Ashraf, M.M. Rahman, G.J. Fuchs, and D. Mahalanabis**

Objective: Evaluate the clinical efficacy of folic acid in acute watery diarrhoea in children.

Methodology: In a double-blind randomized, placebo-controlled clinical trial, 106 boys received either folic acid in a dose of 5 mg at 8-hour intervals or a placebo for 5 days. Rehydration was achieved by rice-based oral rehydration solution (ORS). In addition, the children were given breast milk, a milk cereal mixture, or rice-lentil mixture. The study was conducted in the Clinical Research and Service Centre of ICDDR,B during November 1992-June 1994. Boys aged 6-23 months with a history of acute watery diarrhoea of less than 72 hours were selected for the study. Main outcome variables were: daily stool output (g/kg), intake of oral rehydration solution (g/kg), and duration of diarrhoea (h).

Results: There were 54 children in the folic acid group and 52 in the placebo group. The admission characteristics including age, breast feeding and nutritional status, the duration and frequency of diarrhoea, and vomiting, were comparable between the two groups. There were no significant differences in intake of ORS or stool output between the groups. The mean ± SD of total stool output (g/kg) in the folic acid vs placebo group was 532± 476 vs 479± 354 respectively, and the duration (h) of diarrhoea was 108± 68 and 103± 53 respectively. In more than 40% of the children in either group, diarrhoea continued beyond 5 days.

Conclusion: In contrast to an earlier report indicating efficacy, the results of this randomized, double-masked, placebo-controlled trial demonstrate that folic acid as an adjunct therapy does not provide clinical benefit to children with acute diarrhoea. Our findings, therefore, should have a positive influence on preventing further inappropriate use of folic acid in diarrhoea.

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Relationship Between Specific Enteropathogens and Nutritional Status of Children Attending a Diarrhoea Treatment Centre in Bangladesh

Nahrina Dewan, A.S.G. Faruque, and G.J. Fuchs

Objective: Examine the distribution of diarrhoeal pathogens according to nutritional status.

Methodology: In an analytical study conducted in ICDDR,B's Dhaka hospital during 1990-1994, children aged less than 5 years were enrolled in the Hospital Surveillance System. Outcome measures were: association between nutritional status and specific enteric pathogens.

Results. The mean ± SD age of the children was 15± 12 months, and 63% were males. Overall, 12% of the children were severely underweight, 4% were severely wasted, and 7% were severely stunted. A specific pathogen was isolated from 5,914 (60%) of the children. Children with *Shigellae* and *Vibrio cholerae* O1 were significantly more malnourished than those with rotavirus diarrhoea ($p<.000001$). The rate of isolation of rotavirus increased from 10% among the severely malnourished children to 30% among well-nourished children ($p<0.0001$). No relationship was detected between nutritional status and diarrhoea due to *Campylobacter or Salmonella.*

Conclusion: The results indicate that an effective nutrition programme for young children might have greater impact on diarrhoeal illness due to *Shigella* and *V. cholerae* than on most other common enteric pathogens.

Determinants of Use of Services of the Health and Family Welfare Centres in Bangladesh

Md. Mafizur Rahman, Barkat-e-Khuda, Yousuf Hasan, and Md. Masud Reza

Objectives: Determine the level of use of Health and Family Welfare Centres (HFWCs), and examine the association of various socio-economic and demographic factors with the use of services.

Methodology: Data for this study were drawn from a cross-sectional survey. In analyzing data, descriptive statistics, as well as logistic regression method were used. The survey was conducted in four rural thanas during September 1993-March 1994. The sample comprised 10,368 rural women of reproductive age. Visit of rural women to HFWC, and effects of socio-economic and demographic factors on rural women's visit to HFWC were examined.

Results: Use of services of HFWCs is very poor. Only one half of the women reported to have ever visited HFWCs. Among them, for more than half of the cases, the visit took place before the previous 6 months. The older, educated, and high-parity women are more likely to visit HFWCs. The women who work for income, and adopt contraception have also higher probability of visiting HFWCs. However, landholding and income appear to have negative association with visit to HFWCs. Also, travel time to HFWC is negatively associated with visit to HFWC.

Conclusion: The results suggest the need for strategic measures to be undertaken to increase the use of services of the HFWCs. Female education and employment should also be promoted.

Incidence of Pneumococcal Bacteraemia in a Diarrhoea Treatment Centre

U. Dhar, C. Seas, W.A. Khan, M.A. Salam, and M.L. Bennish

Objective: Determine the epidemiological and clinical features of Pneumococcal bacteraemia occurring in patients admitted in a diarrhoea treatment unit.

Methodology: Retrospective analysis of patients' records admitted in the inpatient ward of the Clinical Research and Service Centre (CRSC) between January 1990 and December 1993 was done. All patients who had *Strep. pneumoniae* from blood culture were included. A form was developed to record inpatient information obtained from records of individual patient.

Results: Fifty-seven percent of the episodes occurred during the five months of winter and early spring. Ten (6%) patients had a second pathogen identified in the blood culture. Nine percent of the bacteraemia episodes were nosocomially acquired. Patients with bacteraemia had a median age of 10 m (range 1 m - 60y); 60% of the patients were <1y. On admission, 151 (97%) patients had diarrhoea; 129 (83%) fever, 72 (46%) respiratory distress, and 76 (49%) a history of cough. Sixty percent of the 124 bacteraemic children aged five years or less were severely malnourished. *S. pneumoniae* was isolated from 8 (47%) of the 17 patients from whom spinal fluid was obtained. All but 9(6%) patients received an antimicrobial agent effective against *S. pneumoniae* on admission. Forty-nine (31%) of the patients with pneumococcal bacteraemia died, accounting for 1.8% of all hospital deaths during the study. In a multiple regression analysis, factors predictive of death were tachypnoea (respiratory rate >50/min, odds ratio [OR] 4.35), leukopenia (blood leukocyte count <6000 mm^3, OR 6.61), and hypokalemia (serum K^+ <3.0 mmol/L, OR 2.49).

Conclusion: *S. pneumoniae* bacteraemia is a potentially preventable cause of death among patients with diarrhoea in Bangladesh. Preventing such deaths will require earlier identification of these patients.

Azithromycin in the Treatment of Adults with Shigellosis

W.A. Khan, C. Seas, U. Dhar, M.A. Salam, and M.L. Bennish

Objective: Study the efficacy of Azithromycin in the treatment of adults with shigellosis.

Methodology: Multiple-resistant *Shigella* infections are a major public health problem in developing countries. In a randomized, double-blind study at Dhaka hospital of ICDDR,B from February 1995 to March 1996, azithromycin (AZR), 500 mg on day 1 of study, followed by 250 mg daily for 4 days, with ciprofloxacin (CIP), 500 mg every 12 hours for 5 days were compared. Eighty-five patients were enrolled in the study. Patients were eligible for the study if they were men aged 18-60 years, had dysentery for 72 hours, had not taken an effective antimicrobial, trophozoites of *E. histolytica* were not identified on stool examination, and gave informed consent. Patients remained in the hospital for six days. Treatment was considered to have failed clinically if dysentery continued for 72 hours after therapy was initiated, or if on study day 5, patients had >6 stools, or >1 watery stool, or were febrile; and to have failed bacteriologically if *Shigella* could be isolated from a stool culture 72 hours or more after the initiation of therapy.

Results: *Shigella* was isolated from an admission stool culture in 76, of whom 70 were included in the final analysis. Thirty-four received AZR and 36 CIP. Therapy was clinically successful in 28 (82%, 95% CI 66-93%) patients receiving AZR, and 32 (89%, 95% CI 74-97%) receiving CIP (NS); and bacteriologically successful in 32 (94%, 95% CI 80-99%) and 36 (100%, 95% CI 90-100%) patients, respectively (NS). Patients receiving azithromycin had a median of 28 stools during the study period (95% CI 19-35), and those receiving CIP 23 stools (95% CI 18-30) (NS).

Conclusion: AZR is an effective therapy for shigellosis.

Community Participation in Growth Monitoring and Nutrition Education

Moazzem Hossain, M A Hanifi, Abbas Bhuiya, and Peter Eppler

Objective: Measure the extent of community participation in growth monitoring of children aged less than 5 years and nutrition education of the mothers and find out the reasons for both use and non-use of those programmes.

Methodology: The methods included: participatory rural appraisal, people's participatory planning, focus group discussions and individual interviewing in four villages of Chakaria thana under Cox's Bazar district in January-December 1996.

Results: Members of the indigenous village-based organizations have established 4 village health posts. Volunteers were nominated by the organizations and trained by Chakaria Community Health Project of ICDDR,B. These volunteers motivate the parents to bring their less than 5-years old children to the health posts, weigh the babies, record in the growth monitoring chart and counsel the mothers of the malnourished children on nutrition. Although the number of volunteers present during all the sessions were almost same, the attendance of mothers started declining gradually after a few sessions. Main reasons for attendance was their interest in knowing about the progress of their babies and their positive experience in implementing the nutrition advice. On the other hand, the two main reasons for discontinuation were that they did not have someone to carry their babies and their frustration for not finding any medicines at the health posts.

Conclusion: Community members participate actively when it is their own decision and not imposed. For maximum use of the health services, the community perception and expectation about that programme have to be considered and addressed.

Contraceptive Switching Patterns in Rural Areas of Bangladesh

Indrani Haque, Thomas T. Kane, Nikhil Ch. Roy, and Khorshed A. Mozumder

Objective: Investigate contraceptive method switching patterns in rural areas of Bangladesh, including their socio-demographic determinants, and the effect of switching on prevalence, method-mix and continuation of contraceptive use.

Methodology. The longitudinal data from Sample Registration System (SRS) of the MCH-FP Extension Project (Rural) in Abhoynagar and Sirajganj sub-districts, collected bi-monthly over an 11-year period (1983-1993), were used for the analyses of a special cohort of 2,866 married women of reproductive age who were in the 15-39-year age-group in 1983 and were still present in the system in 1993. The frequency of method switching, reasons for switching, and the direction and sequence of switching patterns were analyzed using cross-tabulations. The socio-demographic and programmatic determinants of switching behaviour among contraceptive users are examined using logistic regression procedures.

Results: Method switching is prevalent in rural Bangladesh. More than half of the contracepting women have switched methods at least once. The most frequent method switching occurred between the two hormonal methods – pills and injectables. For the majority of the sterilization users, this was their first method. Traditional method users were more likely to switch to sterilization than were hormonal method users. Education of the women and frequency of contact with government field workers were positively related to the number of switches. Side-effects and the need for more suitable methods were the two main reasons given for switching methods. Husband's objection was an important reason for switching from male methods, like, condoms and withdrawal, and also from 'safe period' to other methods.

Conclusion: Findings show that although method switching is prevalent in rural Bangladesh, switching from hormonal to long-term methods or barrier methods is rare. The family planning programme must underscore the importance of the different types of contraceptive methods available and counsel the eligible couples according to their individual needs.

Clients' and Providers' Perspectives about Low Condom Use

Mehrab Ali Khan, Mizanur Rahman, and Parveen Akhter

Objective: Examine the determinants of low use of condom as a contraceptive method in rural Bangladesh, by documenting reported advantages and disadvantages and accompanying beliefs as reported by users and providers, regarding its use.

Methodology: Over 8,000 married women of reproductive age from four rural thanas of the ICDDR,B extension project were interviewed. Also, focus group discussions (FGD) were conducted among the health and family planning workers to examine possible reasons for low condom use. The MCH-FP Extension Project works with the Government of Bangladesh in rural areas of Abhoynagar, Bagherpara, Keshobpur and Sirajganj thanas, and conducts operations research to improve health and family planning service-delivery. Hence, the study was concluded in these areas. The survey was conducted during April-June 1994. Contraceptive behaviour and attitudes about condom use and its use-effectiveness by rural women were examined.

Results: At the time of the survey, the contraceptive prevalence rate (CPR) was 40%, yet only 3% of the couples used the condom method. The advantages of the condom, as reported by users, were minimal side-effects and protection against STDs. Reported disadvantages included discomfort during use, lack of appropriate disposal facilities and risky as a method due to breakage. FGDs revealed several reasons for low condom use: cost, lack of suitable storage and disposal facilities, and reluctance of women to obtain condoms for the use of their husbands.

Conclusion: Successful promotion of condom use will require innovative approaches for overcoming the barriers, not only for effective contraception among Bangladeshi couples, but for STD and HIV/AIDS prevention as well.

Strategic Issues for a Preventive Intervention against Environmental Hazards in Rural Communities

A. Iqbal, M. Ali, J. Chakraborty, and J. Myaux

Objective: Collect baseline information on drowning deaths to support the strategy for a preventive intervention on targeted population

Methodology: A case study was conducted in Matlab study area on all the accidental drowning cases that occurred in 1995 in children aged between 1 and 4 years. The data were collected from the demographic surveillance system and were plotted on the map. Environmental information was gathered by using Geographical Information System (GIS).

Results: In 1995, 160 deaths were reported from the 1-4-year age group (mortality rate: 7.6 per 1000), among which 51 were caused by drowning (31.9%). Eight cases happened inside the embankment; 62% of the accidents in ponds *vs.* 28% in rivers. In 91% of the cases, the distance to the house was less than 100 m. On an average, drowning occurred in the less crowded areas. Ninety-two percent of the drowning occurred during the rainy season, between April and September. In majority of the cases, the mother was present at the time of the accident, busy with household work. In 3 cases, there was a special event on that day in the household. In 25% of the cases, the mother reports the 'Evil' as cause of the accident.

Conclusion: Drowning remains a very common cause of death in rural setting of Bangladesh. An education programme is expected to reduce the child mortality by 10% to 15%. The strategy should focus on the mothers during their daily home tasks, and on the ponds nearby the house. Key-persons, like traditional healers (*kabiraj* and *jharfuk*) and *Imams* from *madrassas* (Koranic schools) or *Purohits* for the Hindu community ought to be involved in the diffusion of preventive messages.

Quality of Care, Client Satisfaction, and Contraceptive Use in Rural Bangladesh

Thomas T. Kane, Mian Bazle Hossain, Barkat-e-Khuda, and Shameem Ahmed

Objective: Examine the relationship between quality of care and contraceptive use behaviour in rural Bangladesh.

Methodology: A 1994 sample survey of 4,967 currently married women of reproductive age (15-49 years) living in two rural thanas (Mirsarai and Satkania) of Chittagong district is used for both bivariate and multivariate logistic regression analysis of the effects of quality of care provided by field workers (FWAs) and at fixed-site clinics on two outcome variables, namely current use of modern contraceptives, and among non-users, intentions to use modern contraception in the future. Elements of quality of care examined in the study include: adequacy of information provided, method choice, technical competence of provider, provider-client interactions, referrals, and continuity of services, and the constellation of services provided. A composite index of quality of care is constructed for the analysis.

Results: Results of the multivariate analysis indicate that, after controlling for other relevant demographic and socio-economic variables, women's perceptions of quality of care have a significant positive effect on the likelihood of current use of modern contraception, and among current non-users, on the likelihood of women intending to use modern methods in the future. Perception of good quality service at family planning clinics, provision of antenatal care by the field worker, more positive provider-client interactions, and broader choice of methods offered by the provider were positively related to current use or intended use of modern contraception, as were higher parity and education, the desire to delay or limit births, and having permission to visit the health centre.

Conclusion: Improved quality of care, particularly the quality of provider-client interactions, should have a significant positive effect on the use of modern contraceptive methods. Policies designed to strengthen quality standards, and to improve facilities and training of providers in counselling and technical skills are needed in the national programme.

Water Management Practices at Home: A Case Study from Bangladesh on Ingestion of Polluted Water

Shafiul Azam Ahmed, Bilqis Amin Hoque, D. Mahalanabis, and A. Mahmud

Objective: Although Bangladesh has achieved a remarkable success in handpump and piped water coverage, ingestion of unsafe water is still common. This brief study attempted to assess home management practices of water, determine routes of ingesting unsafe water, examine methods of collection and storage, and determine why people use unsafe sources despite having access to sources of safe water.

Methodology: Forty-eight rural and 45 urban slum households were studied. Observations, interviews, and water quality investigations were conducted.

Results: The results showed that the respondents were aware that handpump/tap water was safe. They took efforts to use these safe sources for drinking purposes. However, they continued to use surface water for non-drinking purposes, such as bathing, ablution, and mouthwash. They stated reasons, such as more satisfaction in bathing in surface water, traditional practice to bathe in surface water, staining of food and clothing by groundwater, etc. for not using handpump/tap water. Bacteriological results of water samples indicated that the quality of ingested water, especially in rural areas, was poor. Results also showed that inside bottoms of storage containers were heavily contaminated with bacteria. It was shown that water that was safe at sources became contaminated in the storage containers.

Conclusion: This study had a limited scope. Further research is needed to find how water becomes contaminated in containers and its determinants, the effect of container material on bacterial population in stored water, how to reduce contamination of water, and how to improve quality of water. On the management side, studies may be conducted on how to make people understand, use and manage water safely for all domestic purposes.

RTIs and STDs: An Area of Clinical and Human Interest Though not Sychronized

Joachim Victor Gomes and Syeda Nahid Mukith Chowdhury

Objective: RTIs and STDs are a serious concern of the people. The social concern is as great as that of the clinical or research interest. So understanding peoples' perceptions and existing knowledge on STDs/RTIs, along with their explanatory models, is more important than training them on the causes, effects and consequences of STDs/RTIs.

Methodology: The study, therefore, entails representation of illnesses of 500 male patients in two specific urban STD clinics. The study also includes nine in-depth interviews and literature review of existing educational material on sexual diseases that appears in the advertising material of traditional sexual health care providers. The study was launched in July 1996 to be ended by June 1997. The data are based on male patients who came to the STD clinic for services.

Results: The study reveals the need for understanding explanatory models of the general population on sexual diseases. Peoples' concern and language of expression are areas to understand, to build a dialogue between researchers, clinicians, and the general population. This will reveal how and to what extent we can develop education on sexual health in Bangladesh.

Conclusion: The study will help design strategies for clinical management and recording of history, identifying areas where there is need for IEC development and administration on sexual health matters.

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Impact Evaluation and Assessment of the Adolescent Family Life Education Programme of BPHC and Supported-NGOs

Sharmeen Murshid

Objective: Assess the strengths and weaknesses of the Adolescent Family Life Education (AFLE) programme.

Methodology: Participatory research with use of focus group discussions, observation, participatory workshops, case studies, and in-depth interviews. The study was conducted in the three BPHC-funded NGOs who implement AFLE programmes, in Dhaka and Rajshahi districts during 01 March 1996-31 January 1997. All stakeholders of the programme, adolescent girls and boys, trainers, managers, guardians, and community leaders were interviewed. Knowledge, attitudes, and behaviour of adolescents with regard to MCH-FP, social and gender issues; relevance and adequacy of training and IEC material, and gender sensitivity were the outcome variables.

Results: Only adolescent girls were so far enrolled in the AFLE programme; the most noticeable changes in their lives were an improvement in personal hygiene during menstruation and awareness about social issues, such as early marriage, dowry and polygamy. Adolescent boys pointed out to the easy access to drugs and pornographic material in the cities, and the reality of STD among young people. Adolescents, community leaders, and guardians expressed a need for the programme to also reach boys, and their willingness to contribute to its success. The AFLE curriculum, IEC material (especially regarding STDs and HIV/AIDS), and monitoring tools, were designed in a short period and have serious short-falls.

Conclusion: The AFLE, a low-cost programme, has developed rapidly, has proven its impact on the attitude and behaviour of adolescent girls and young women, and is strongly supported by the community. However, the programme needs urgent revision and has to be expanded/modified to target adolescents of both sexes.

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"We followed 1,756 young, rural Bangladeshi children less than five years of age for one month after identification of sentinel *Shigella* patients in their neighborhoods. Two hundred nineteen (12%) children developed *Shigella* diarrhea (shigellosis) and 227 (13%) developed culture-negative dysentery. *Shigella flexneri* (60%) and *S. dysenteriae*, type 1 (15%) were the most common isolates among shigellosis cases. Within individual neighborhoods, there was poor agreement (Kappa = 0.21) between *Shigella* species isolated from sentinel patients and from additional cases detected during surveillance. The risk of shigellosis increased substantially after infancy and peaked in the second year of the life. Severe stunting, as assessed by height-for-age, was associated with an increased risk of shigellosis (adjusted odds ratio [OR_a] = 1.67, 95% confidence interval [Cl] = 1.09-2.57, P<0.05), while breast-feeding was protectively associated (OR_a = 0.40, 95% Cl = 0.24-0.69, P<0.001). Only 43% of the shigellosis cases reported bloody stools; frank dysentery occurred more frequently in *S. dysenteriae* 1 infections than in *S. flexneri* infections (OR_a = 5.04, 95% Cl = 1.76-14.48, P<0.01), and was also associated with severe stunting (OR_a = 2.16, 95% Cl = 1.01-4.58, P<0.05). Our findings show that the high risk of shigellosis in residentially exposed Bangladeshi children results from multiple *Shigella* strains circulating concurrently within the same neighborhood; demonstrate that the risk is notably modified by host age, nutritional status, and dietary patterns; and illustrate that the classic picture of dysentery is relatively infrequent and is correlated with the infecting species and with host nutritional status."

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116. Ananthan S, Dhamodaran S. Toxigenicity & drug sensitivity of *Vibrio mimicus* isolated from patients with diarrhoea. Indian J Med Res 1996 Dec;104:336-41. 23 ref, Eng. Department of Microbiology, Post Graduate Institute of Basic Medical Sciences, University of Madras, Taramani Campus, Madras 600113, India

"In a total of 720 faecal specimens from patients with secretory diarrhoea, vomiting, dehydration, gastroenteritis, cholera and cholera like illnesses, 18 strains of *V. mimicus* were isolated as pure culture. These were characterized for various toxin types and virulence factors using conventional *in vitro* and *in vivo* assays. Labile and stable toxins were elaborated by 15 and 2 strains respectively by ligated rabbit ileal loop (RIL) and suckling mouse assays. While 15 of the whole cell culture elaborated labile toxin, only 7 strains produced the same when culture filtrate was tested in RIL assay. Culture filtrates of 15 strains exhibited vascular permeability factor (PF) on adult rabbit skin, none of the strains were invasive as indicated Sereny's test. Culture supernantants of all strains produced a cytotoxic factor to Vero and Chinese hamster ovary cells. Four of the 18 strains (22%) were resistant to multiple drugs (a combination of 3 or more drugs). The results emphasize the significance of continuous screening and identification of *V. mimicus* and to include in the differential diagnosis of patients with acute diarrhoea."

117. Ansaruzzaman M, Albert MJ, Kühn I, Faruque SM, Siddique AK, Möllby R. Differentiation of *Vibrio cholerae* O1 isolates with biochemical fingerprinting and

comparison with ribotyping. J Diarrhoeal Dis Res 1996 Dec;14(4):248-54. 23 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"The Phene Plate (PhP) system is a commercially available typing system based on the measurements of kinetics of selected biochemical reactions of bacteria grown in liquid medium in 96-well microplates. The system uses numerical analysis to identify biochemical phenotypes among the tested strains. In the present study, a set of 16 discriminatory tests were used to differentiate 117 strains of *Vibrio cholerae* O1 from Mexico and Bangladesh. The stability of PhP types of 16 isolates under different storage temperatures and after repeated subcultures were also evaluated. The PhP system had a reproducibility of 95%. Storage either at +4^{o}C or -70^{o}C, did not affect the reactions of the isolates, whereas 4 strains (25%) stored at room temperature and 5 strains (31%) subjected to 30 consecutive subcultures, exhibited minor changes in their biochemical reactions. Endemic isolates of *V. cholerae* O1 from Bangladesh were more diverse (diversity index = 0.84 to 0.93) than epidemic isolates from Mexico (diversity index = 0.73). Using a collection of 33 heterogeneous isolates of classical biotype of vibrios, PhP typing and ribotyping were compared. PhP typing discriminated more types (n=23) than ribotyping (n=5), whereas a combination of both yielded 27 types. The PhP system appears to be a simple, reliable and highly discriminating method for typing of *V. cholerae*, and may prove especially useful as a first screening method in epidemiological studies of *V. cholerae*."

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"The peripheral blood mononuclear cell (PBMC) phenotype (CD3, CD4, CD8, CD25, CD20 and CD57), spontaneous proliferation of PBMC, proliferation of PBMC in response to Concanavalin A (ConA), pokeweed mitogen (PWM) and phytohaemagglutinin (PHA) and delayed type hypersensitivity (DTH) responses (by skin tests) were measured in 55 children, 12-60 months of age, of whom 17 were children with *S. flexneri* infection and 38 were age-matched uninfected control children. All mitogens were tested in the presence of foetal bovine serum but PHA responses were additionally tested in the presence of autologous plasma. Because the nutritional status (determined as weight-for-age as a percentage of the National Center for Health Statistics median) affected the lymphocyte responses, children were stratified into =65% (poorly nourished) and >65-75% (better nourished) weight-for-age subgroups. Compared with uninfected children, poorly nourished children with *S. flexneri* infection had a lower CD4/CD8 ratio, a higher spontaneous proliferation of PBMC, a lower stimulation index with ConA and a lower stimulation index with PHA in the presence of autologous plasma. In the better nourished subgroup, the only difference was increased numbers of CD25+ PBMC in children with *S. flexneri* infection than in uninfected children. Furthermore, transferrin levels in plasma were found to correlate positively with weight-for-age as well as clinical severity of illness so that poorly nourished children with *S. flexneri* infection were more severely ill. We conclude that malnutrition in children with *S. flexneri* infection is characterized by altered T lymphocyte responses and severe illness."

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"Vitamin A administration in children reduces the incidence of severe diarrhea during the subsequent few months. We therefore examined the effect of treatment with vitamin A during acute diarrhea on the episode duration and severity. In a double-blind controlled field trial, 900 children 1 to 5 y of age with acute diarrhea of =7 d duration were randomly assigned to receive vitamin A (60 mg) or a placebo. Children were followed up at home every alternate day until they recovered from the diarrheal episode. In all study children, those treated with vitamin A had a significantly lower risk of persistent diarrhea [odds ratio (OR) 0.30, 95% confidence interval (CI) 0.07-0.97], but there was no effect on the mean diarrheal duration or the mean stool frequency. In the subgroup of children who were not breast-fed, the mean diarrheal duration [ratio of geometric means (GM) 0.84, 95% Cl 0.72-0.97], mean number of stools passed after the intervention (ratio of GM 0.73, 95% Cl 0.56-0.95), the proportion of episodes lasting =14 d (P = 0.002) and the percentage of children who passed watery stools on any study day (OR 0.40. 95% Cl 0.21-0.77) were significantly lower in those treated with vitamin A. We conclude that administration of vitamin A during acute diarrhea may reduce the severity of the episode and the risk of persistent diarrhea in non-breast-fed children. Similar was not seen in breast-fed children.

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"To characterize the epidemiology of dysentery (defined as bloody diarrhoea) in Burundi, we reviewed national surveillance data and conducted a household cluster survey including two case-control studies: one at the household, the other at the individual level. We estimated that community incidences for dysentery (per 1000 residents) in Kibuye Sector were 15.3 and 27.3 and that dysentery accounted for 6% and 12% of all deaths, in 1991 and 1992, respectively. Factors associated (P=0.05) with contracting dysentery were being female, using a cloth rag after defecation, a history of recent weight loss, and not washing hands before preparing food. The attributable risk, at the household level, of not washing hands before preparing food was 30%. Secondary household transmission accounted for at most 11% of dysentery cases. This study suggests that *Shigella dysenteriae* type 1 may be one of the leading causes of preventable mortality in Burundi and other African countries where effective antimicrobial agents are no longer affordable. Since hands were the most important mode of transmission of *S. dysenteriae* in this study, community-based interventions aimed at increasing hand washing with soap and water, particularly after defecation and before food preparation, may be effective for controlling dysentery epidemics caused by *S. dysenteriae* type 1 in Africa."

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"During 1980-1990 BRAC, a Bangladeshi non-governmental organization, taught over 12 million mothers how to prepare oral rehydration therapy (ORT) at home with lobon (common salt) and gur (unrefined brown sugar). This was followed by a strong promotion and distribution of prepackaged ORS by various agencies including the government. In 1993 we assessed knowledge of ORT preparation, its local availability and its use for the management of diarrhoea. Over 9000 households in 90 villages were revisited; 306 government outreach health workers, 296 drug sellers, and 237 village doctors were interviewed; 152 government facilities and 495 pharmacies/shops were visited. ORT prepared by mothers in a sub-sample of the households was analyzed for chloride content and interviewers collected information on use of ORT for diarrhoeal episodes occurring in the preceding two weeks. The data quality was assessed through a resurvey of sample respondents within two weeks of the first interview. Over 70% of the mothers could prepare a chemically `safe and effective' ORS. A significant proportion of these mothers were very young at the time of the mass campaigns using house to house teaching, implying an intergenerational transfer of the knowledge on ORT. ORT was found to be used in 60% of all diarrhoeal episodes, but the rate varied with the type of diarrhoea, being highest for *daeria* (severe watery diarrhoea) and lowest for *amasha* (dysentery). Drug sellers and village doctors now recommend ORT much more frequently than before. Members of the medical profession (qualified and unqualified) still lag behind in prescribing the use of ORT. The availability of pre-packaged ORS in rural pharmacies has improved enormously. There is convincing evidence that the widescale promotion in the past of ORS for dehydration in diarrhoea has led to this marked improvement today. Nevertheless the use of rice-based ORS, culturally appropriate messages and the promotion of ORS with food offer opportunities to further improve the utilization of ORT."

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"**Background** The aim of this double-blind randomised vaccine-controlled trial was to assess the efficacy of a conjugate vaccine composed of *Shigella sonnei* 0-speciflc polysaccharide bound to *Pseudomonas aeruginosa* recombinant exoprotein A (*S. sonnei*-rEPA) and of an oral, live-attenuated *Escherichia coli/S. flexneri* 2a (EcSf2a-2) hybrid vaccine among military recruits in Isreef at high risk of exposure to *Shigelia* spp. We report here our preliminary findings on the efficacy of *S. sonnei*-rEPA; we have not documented sufficient cases to assess the efficacy of EcSf2a-2. **Methods** Between April, 1993, and August, 1994, male Israeli military recruits aged 18-22 years were asked to take part in our study. We enrolled 1446 soldiers from seven separate field sites (groups A-G). Soldiers were randomly allocated one injection of S sonnei-rEPA and four doses of oral placebo (n=576), four oral doses of EcSf2a-2 and one injection of saline placebo (n=580), or one injection of meningococcal tetravalent control vaccine and four doses of

oral placebo (n=290). Because there were no cases of *S. flexneri* 2a, the EcSF2a-2 and meningococcal vaccines were the control group. We defined *S. sonnei* shigellosis as diarrhoea with a positive faecal culture for *S. sonnei*. Each group of soldiers was followed up for 2.5-7.0 months. The primary endpoint was protective efficacy of *S. sonnei*-rEPA against *S. sonnei* shigellosis. **Findings** Cases of culture-proven *S. sonnei* shigellosis occurred in four groups of soldiers (groups A-D), which comprised 787 volunteers (3l2 received *S. sonnei*-rEPA, 316 received EcSt2a-2, and 159 received meningococcal control veiccine). In groups A-C, cases of shigellosis occurred 70-155 days after vaccination, whereas In group D cases occurred after 1-17 days. In groups A-C, the attack rate of shigellosis was 2.2% in recipients of *S. sonnei*-rEPA compared with 8.6% In controls (protective efficacy 74% [95% Cl 28-1001, p=0-006). *S. sonnei*-rEPA also showed significant protection against shigellosis in group D (43% [4-82), p=0-039). Prevaccination and postvaccination ELISA measurements of antibody to *S. sonnei* lipopolysaccharide among recipients of *S. sonnei*- REPA showed that the vaccinees who developed *S. sonnei* shigellosis had significantly lower serum IgG and IgA responses to the homologous lipopolysaccharide than those who did not (p=<O.05). **Interpretation** One injection of *S sonnei*-rEPA confers type- specific protection against *S. sonnei* shigellosis. The high antibody concentration induced by the conjugate vaccine in volunteers who did not develop shigellosis suggests that there Is an association between serum antibody titre and protection."

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"**Background**: Solar radiation reduces the bacterial content of water, and may therefore offer a method for disinfection of drinking water that requires few resources and no expertise. **Methods**: We distributed plastic water bottles to 206 Maasai children aged 5-16 years whose drinking water was contaminated with faecal coliform bacteria. Children were instructed to fill the bottle with water and leave it in full sunlight on the roof of the hut (solar group), or to keep their filled bottles indoors in the shade (control group). A Maasai-speaking fieldworker who lived in the community interviewed the mother of each child once every 2 weeks for 12 weeks. Occurrence and severity of diarrhoea was recorded at each follow-up visit. **Findings**: Among the 108 children in households allocated solar treatment, diarrhoea was reported in 439 of the 2-week reporting periods during the 12-week trial (average 4.1 [SD 1.2] per child). By comparison, the 98 children in the control households reported diarrhoea during 444 2-week reporting periods (average 4.5 [1.2] per child). Diarrhoea severe enough to prevent performance of duties occurred during 186 reporting periods in the solar group and during 222 periods in the control group (average 1.7 [1.2] vs 2.3 [1.4]). After adjustment for age, solar treatment of drinking water was associated with a reduction in all diarrhoea episodes (odds ratio 0.66 [0.50-0.87]) and in episodes of severe diarrhoea (0.65 [0.50-0.86]). **Interpretation**: Our findings suggest that solar disinfection of water may significantly reduce morbidity in communities with no other means of disinfection of drinking water, because of lack of resources or in the event of a disaster."

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"*Background*. Cholera spread to Latin America in 1991; subsequently, cholera vaccination was considered as an interim intervention until long-term solutions involving improved eater supplies and sanitation could be introduced. Three successive summer cholera outbreaks in northern Argentina and the licensing of the new single-dose oral cholera vaccine, CVD 103-HgR, raised questions of the cost and benefit of using this new vaccine. *Methods*. This study explored the

potential benefits to the Argentine Ministry of Health of treatment costs averted, versus the costs of vaccination with CVD 103-HgR in the relatively confined population of northern Argentina affected by the cholera outbreaks. Water supplies and sanitation in this area are poor but a credible infrastructure for vaccine delivery exists. *Results*. In our cost-benefit model of a 3-year period (1992-1994) with an annual incidence of 2.5 case-patients per 1000 population and assumptions of vaccine efficacy of 75% and coverage of 75%, vaccination of targeted high risk groups would prevent 1265 cases. *Conclusion*. Assuming a cost of US$ 602 per treated case and of US$1.50 per dose of vaccine, the total discounted savings from use of vaccine in the targeted groups would be US$ 132 100. The projected savings would be altered less by vaccine coverage (range 75-90%) or efficacy (60-85%) changes than by disease incidence changes. Our analysis underestimated the true costs of cholera in Argentina because we included only medical expenditures; indirect losses to trade and tourism had the greatest economic impact. However, vaccination with CVD 103-HgR was still cost-beneficial in the base case."

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"Previously we reported that standard oral rehydration salts (ORS) solution is not as effective as a reduced-osmolarity glucose-based ORS for the treatment of children with acute noncholera diarrhoea: with standard ORS the diarrhoea lasts longer, stool output is greater, serum sodium is higher, and there is more need for supplemental intravenous infusion. We studied a reduced-osmolarity maltodextrin (MD)-based ORS to determine whether it had similar benefits, and also the effect of sugar malabsorption on the efficacy of standard and MD-based ORS. A total of 90 boys aged 3-24 months with acute noncholera diarrhoea and moderate dehydration were randomly assigned to either standard ORS (glucose 20 g/l, osmolarity 311 mmol/l) or MD-ORS (MD 50 g/l, osmolarity 227 mmol/l). There were no differences in treatment results. Some 45% of subjects had a high total stool output (>300 g/kg), which was unrelated to the type of ORS given. High stool output was significantly associated with a longer duration of diarrhoea (33 vs. 15 hours; P<0.001), a persistently elevated serum sodium (149 vs. 144 mmol/l at 24 h; P<0.02), the need for intravenous infusion (11/41 vs. 0/48; P<0.002), and an increase in faecal reducing substances (10.8 vs. 3.4 g/l at 24 h; P<0.001). We conclude that some children given standard ORS develop osmotic diarrhoea owing to the combined effect of transient sugar malabsorption and slight hypertonicity of the ORS. Earlier studies show that this adverse outcome can largely be avoided when extra water is given in reduced-osmolarity glucose-based ORS. Reduced osmolarity has no benefit, however, when glucose is replaced by maltodextrin, probably because

the sugars released by hydrolysis of MD, when malabsorbed, raise the intraluminal osmolarity to equal or exceed that of standard ORS. Thus, reduced-osmolarity glucose-based ORS is superior to both standard ORS and reduced-osmolarity solutions based on maltodextrin and probably other complex carbohydrates. Studies are in progress to define the optimal formulation of reduced-osmolarity glucose-based ORS."

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"Two isolates of group A rotaviruses (CR129 and CR156) were isolated from faecal samples of diarrhoeal calves reared in two dairy farnis at Ilisar (Haryana, India) by using MA-104 cell lines. These isolates were compared with three standard reference bovine rotaviruses, UK, NCDV and B223, to reveal differences, if any, in their genome and protein migration profiles. The migration of RNA segment 4 of CR129 was slower than that of NCDV, but faster than that of UK. Segment 10 of CR156 moved faster than that of the reference viruses. The segments 2 and 3 co-migrated in CR129, but resolved separately in CR156. Five protein bands of size 116-120 KD (VP1), 95 KD (VP2), 90 KD (VP3/VP4), 44 KD (VP6) and 34 KD (VP7) were detected by protein analysis. No significant difference was observed in the protein profile of these two bovine rotavirus isolates by immunoblotting. However, VP1 was of approximately 116 KD size in the two isolates, compared to 120 KD in the reference strains. These findings indicate that these rotaviruses isolated from diarrhoeic Indian calves differed from the 3 reference strains."

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"The prevalence of infection by the invasive parasite *Entamoeba histolytica* and the noninvasive parasite *Entamoeba dispar* was determined in 2000 children in Bangladesh. Antigen detection identified more cases of *E. histolytica-E. dispar* infection than did culture or microscopy. Microscopic identification of *E. histolytica-E. dispar* complex infection in stool did not equate with the diagnosis of amebic dysentery because most amebic infections in this population were due to *E. dispar*. Urban children with diarrhea had a 4.2% prevalence of *E. histolytica* infection and a 6.5% prevalence of *E. dispar* infection; rural asymptomatic children had a 1.0% prevalence of *E.*

histolytica infection and a 7.0% prevalence of *E. dispar* infection. *Shigella dysenteriae* and *Shigella flexneri* infections were more frequent in children who also had *Entamoeba* infection, a potentially important consideration for the empiric treatment of dysentery in this population."

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"The antidiarrhoeal activities of leaf extracts of *Ocimum gratissimum* were investigated by disc diffusion and tube dilution methods. The extracts were active against *Aeromonas sobria, Escherichia coli, Plesiomonas shigelloides, Salmonella typhi, and Shigella dysenteriae.* The leaf extracts were most active against *S. dysenteriae* and least active against *S. typhi*. The sensitivity of the organisms measured in terms of zone of inhibition ranged from 8.00 to 19.50 mm. The minimum inhibitory concentrations were from 4.00 to 50.00 mg ml^{-1}, while the minimum bactericidal concentration ranged from 8.00 to 62 mg $m1^{-1}$. The potentials of the leaf extract for the treatment of diarrhoeal diseases is discussed."

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"Described are the findings of a multicentre cohort study to test an algorithm for the treatment of persistent diarrhoea relying on the use of locally available, inexpensive foods, vitamin and mineral supplementation, and the selective use of antibiotics to treat associated infections. The initial diet (A) contained cereals, vegetable oil, and animal milk or yoghurt. The diet (B) offered when the patient did not improve with the initial regimen was lactose free, and the energy from cereals was partially replaced by simple sugars. A total of 460 children with persistent diarrhoea, aged 4-36 months, were enrolled at study centres in Bangladesh, India, Mexico, Pakistan, Peru, and Viet Nam. The study population was young (11.5 ± 5.7 months) and malnourished (mean weight-for-age-Z-score, -3.03 ± 0.86), and severe associated conditions were common (45% required rehydration or treatment of severe infections on admission). The overall success rate of the treatment algorithm was 80% (95% CI, 76-84%). The recovery rate among all children with only diet A was 65% (95% CI, 61-70%), and was 71% (95% CI, 62-81%) for those evaluated after receiving diet B. The children at the greatest risk for treatment failure were those who had acute associated illnesses (including cholera, septicaemia, and urinary tract infections), required intravenous antibiotics, and had the highest initial purging rates. Our results indicate that the short-term treatment of persistent diarrhoea can be accomplished safely and effectively, in the

majority of patients, using an algorithm relying primarily on locally available foods and simple clinical guidelines. This study should help establish rational and effective treatment for persistent diarrhoea."

157. Islam MA, Rahman MM, Mahalanabis D, Rahman AKSM. Death in a diarrhoeal cohort of infants and young children soon after discharge from hospital: risk factors and causes by verbal autopsy. J Trop Pediatr 1996 Dec;42(6):342-7. 24 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"Assessing mortality pattern of children after discharge from hospital is important to guide appropriate management policy. We studied young children aged 1-23 months, who were discharged from an urban Diarrhoea Treatment Hospital. Children were enrolled on discharge from the hospital, and followed at home after 6 and 12 weeks to assess post-discharge mortality. Of 500 children, 427 were available for evaluation at home 6 weeks after discharge. The median age of the children was eight months, 77 per cent of whom were less than 12 months of age. Of the 427 children, 30 (7 per cent) died within 6 weeks and two died within 12 weeks of discharge from hospital. The median survival time of the deceased was 11 days. Children less than 6 months of age had a five times greater risk of death compared with those aged 6 months or older. Malnutrition, non-breastfeeding, and lack of immunization were important risk factors for death. As ascertained by verbal autopsy, the underlying causes of death were respiratory diseases and watery diarrhoea. Malnutrition and low birth weight were the main associated causes. Hospitalized children, especially young infants, should be given special attention and need to be followed preferably within a week of discharge."

158. Islam S, Mahalanabis D, Chowdhury AKA, Wahed MA, Rahman ASMH. Glutamine is superior to glucose in stimulating water and electrolyte absorption across rabbit ileum. Dig Dis Sci 1997 Feb;42(2):420-3. 12 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"L-glutamine is the primary metabolic fuel of the intestinal mucosa. This *in vivo* study compares the effect of L-glutamine 50 mM with that of D-glucose 50 mM on water and electrolyte absorption in jejunal and ileal loops of healthy rabbits. Using polyethylene glycol (PEG) as a nonabsorbable marker and an incubation at 37^{o}C, we found that absorption of water (P=0.000), sodium (P=0.002), potassium (P=0.001), and chloride (P=0.003) from the glutamine electrolyte solution was greater than from the glucose electrolyte solution in the ileum. A similar trend was shown in the jejunum. We conclude that L-glutamine may be a useful component to be tested in oral rehydration solutions for treating diarrheal dehydration."

159. Jahan Y, Hossain A. Multiple drug-resistant *Shigella dysenteriae* type 1 in Rajbari District, Bangladesh. J Diarrhoeal Dis Res 1997 Mar;15(l):17-20. 19 ref, Eng. Microbiology Department, Institute of Epidemiology Disease Control and Research, Mohakhali, Dhaka-1212, Bangladesh

"Twenty-one *Shigellae* isolates were obtained from bloody faecal specimens of diarrhoeal patients at Rajbari District Hospital from January 1994 to June 1995, and serogrouped. Fourteen (67%) isolates belonged to the *Shigella dysenteriae* serogroup and 7 (33%) to *Shigella flexneri* serogroup. *Shigella dysenteriae* strains were further serotyped; all were *Shigella dysenteriae* 1. Each strain was tested for resistance to 6 common antimicrobial agents. The two strains had different antibiotic susceptibility patterns. The 7 *S. flexneri* showed 6 different resistant patterns and the 14 *S. dysenteriae* 1 isolates had 4 resistance patterns. One of the *S. dysenteriae* 1 isolates was resistant to all 6 antimicrobial agents; 10 to 5, and twice to a different combination of 4 antimicrobials. The 14 (100%) *S. dysenteriae* 1 strains were resistant to 3 major antimicrobial agents: ampicillin, tetracycline, and chloramphenicol; 13 (93%) were resistant to 5 agents: ampicillin, tetracycline, chloramphenicol, trimethoprim-sulphamethoxazole, and nalidixic acid. Ciprofloxacin was the only drug active against all 7 *S. flexneri* and 13 of the 14 (93%) *S.*

dysenteriae 1 strains."

160. Jertborn M, Svennerholm A-M, Holmgren J. Intestinal and systemic immune responses in humans after oral immunization with a bivalent B subunit-01/0139 whole cell cholera vaccine. Vaccine 1996 Oct;14(15):1459-65. 35 ref, Eng. Department of Medical Microbiology and Immunology, Göteborg University, S-413 46 Göteborg, Sweden

"There is a need for an effective vaccine that can protect against cholera caused by either *Vibrio cholerae O1* or by the new pandemic serotype O139 Bengal. An oral bivalent B subunit-0110139 whole cell (B-0110139 WC) cholera vaccine has been prepared by adding formalin-killed O139 vibrios to the recently licensed oral recombinant B-O1 WC vaccine. When tested in Swedish volunteers, this B-O110139 WC vaccine was found to be safe and immunogenic. Two vaccine doses given 2 weeks apart induced statistically significant, $P<0.05$, mucosal IgA antibody responses in intestinal lavage fluid against cholera toxin in all of nine vaccinees and against both O1 and O139 vibrios in seven of nine cases. The intestinal responses were associated with similar high frequencies of intestine-derived antibody-secreting cell responses in peripheral blood to the different antigens. A third dose of vaccine given after 5-6 weeks did not result in any further increased response. All of 12 vaccinees responded with significant IgA and IgG antitoxin responses in serum associated with significant vibriocidal antibody titre rises against O1 vibrios in 10 cases (83%) and against O139 vibrios in eight vaccinees (67%). The frequencies and magnitudes of the serological responses to the B subunit and O1 WC components were similar to those induced by the B-O1 WC vaccine. Thus, the O139 component of the vaccine induced intestinal and systemic antibacterial immune responses in the majority of the vaccines, and its addition to the vaccine did not interfere with the immunogenicity of the B subunit or O1 WC components."

161. Kabir I, Khan WA, Haider R, Mitra AK, Alam AN. Erythromycin and trimethoprim-sulphamethoxazole in the treatment of cholera in children. J Diarrhoeal Dis Res 1996 Dec;14(4):243-7. 22 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"To evaluate the efficacy of erythromycin and trimethoprim-sulphamethoxazole (TMP-SMX) in the treatment of cholera in children aged 1-8 years, a randomised clinical trial was conducted at a diarrhoea treatment centre in Bangladesh from December 1991 to June 1992. Fifteen children received erythromycin, 50 mg/kg per day, in four equally divided doses, 18 children received 10 mg/kg per day of trimethoprim and 50 mg/kg per day of sulphamethoxazole in two equally divided doses (12 hourly) for five days, and 15 children received no antibiotic; children in all three groups received intravenous cholera saline for severe dehydration and for mild to moderate dehydration, a rice-based oral rehydration solution. The mean stool volumes in mL/kg body weight in the two treatment groups were less than that of the control group, and there were no significant differences in stool volume among the two treatment groups. However, 67% of the children in the erythromycin group and 82% in the TMP-SMX group recovered within 72 hours compared to 33% in the control group ($p<0.01$). Similarly, the bacteriological cures were 80% in the erythromycin group and 83% in the TMP-SMX group compared to only 27% in the control group ($p<0.001$). These results confirm that both erythromycin and trimethoprim-sulphamethoxazole are effective antimicrobials in the treatment of cholera. These drugs are of value specially in younger children in whom tetracycline is contraindicated or when the infecting *Vibrio cholerae* are resistant to tetracycline.

162. Kariuki S, Gilks CF, Kimari J, Muyodi J, Waiyaki P, Hart CA. Plasmid diversity of multi-drug-resistant *Escherichia coli* isolated from children with diarrhoea in a poultry-farming area in Kenya. Ann Trop Med Parasitol 1997 Jan;91(1):87-94. 29 ref, Eng. Department of Medical Microbiology and Genito-Urinary Medicine, University of Liverpool, PO Box 147, Liverpool L69 3BX, UK

163. Karpman D, Connell H, Svensson M, Scheutz F, Alm P, Svanborg C. The role of lipopolysaccharide and Shiga-like toxin in a mouse model of *Escherichia coli* O157:H7 infection. J Infect Dis 1997 Mar;175(3):611-20. 64 ref, Eng. Department of Pediatrics, University of Lund, S-22185 Lund, Sweden

164. Khan SR, Rehmani NZK, Khan AR, Khan AI. An update on oral hydration therapy for diarrhoea. Int Child Health 1997 Jan;8(1):93-7. 39 ref, Eng. Fatima Memorial Hospital, Lahore, Pakistan

165. Khan WA, Salam MA, Bennish ML. C reactive protein and prealbumin as markers of disease activity in shigellosis. Gut 1995 Sep;37(3):402-5. 23 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"To evaluate serum C reactive protein (CRP) and prealbumin concentration as markers of disease activity in shigellosis this study serially measured serum concentrations of CRP and prealbumin in 39 patients infected with *Shigella* spp, and a comparison group of 10 patients infected with *Vibrio cholerae* serotype 01. On admission, patients with shigellosis had significantly higher median concentrations of CRP (109 v 5 mg/l; $p<0.01$) and significantly lower median concentrations of prealbumin (16 v 23 mg/dl; $p<0.01$) than did patients with cholera. Among *Shigella* spp infected patients, CRP concentrations were significantly lower, and prealbumin concentrations significantly higher, on study days 3 and 5 when compared with admission values. Among *Shigella* spp infected patients, those in whom treatment failed had higher admission CRP concentrations than those in whom treatment was successful ($p=0.142$). An admission CRP concentration =110 mg/1 had a 70% sensitivity and a 61% specificity in predicting failure of treatment among patients infected with *Shigella* spp; the predictive value of a positive and negative test was 14% and 96% respectively. In summary, acute shigellosis elicits an acute phase response, the magnitude of which predicts clinical outcome."

166. Khan WA, Seas C, Dhar U, Salam MA, Bennish ML. Treatment of shigellosis: V. Comparison of azithromycin and ciprofloxacin: a double-blind, randomized, controlled trial. Ann Intern Med 1997 May 1;126(9):697-703. 39 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"**Background:** Treatment of shigellosis is currently limited by the high prevalence of multidrug-resistant strains of *Shigella*. **Objective:** To determine the efficacy of azithromycin in the treatment of shigellosis. **Design:** Randomized, double-blind clinical trial. **Setting:** Diarrhea treatment center in Dhaka, Bangladesh. **Patients**: 70 men with shigellosis that had lasted 72 hours or less. **Interventions:** Patients stayed in the hospital for 6 days. Thirty-four patients were randomly assigned to receive 500 mg of azithromycin on study day 1, followed by 250 mg once daily for 4 days; 36 patients were assigned to receive 500 mg of ciprofloxacin every 12 hours for 5 days. **Measurements:** Clinical treatment failure was considered to have occurred if frank dysentery persisted for 72 hours after therapy began or if on study day 5 a patient had more than six stools, had any bloody-mucoid stools, had more than one watery stool, or had an oral body temperature exceeding 37.8^{o}C. Bacteriologic treatment failure was considered to have occurred if *Shigella* strains could be isolated from a stool sample after study day 2. Therapy was considered either clinically or bacteriological successful in patients who completed therapy and did not meet criteria for failure. **Results:** Therapy was clinically successful in 28 (82%) patients who received azithromycin and 32 (89%) patients who received ciprofloxacin (difference, -7% [95% CI, -23% to 10%]). Therapy was bacteriologically successful in 32 (94%) patients receiving azithromycin and 36 (100%) patients receiving ciprofloxacin (difference, -6% [CI, -14% to 2%]). Peak serum concentration of azithromycin were equal to the minimum inhibitory concentration (MIC) of the infecting *Shigella* strains, whereas serum concentrations of ciprofloxacin were 28 times the MIC. Stool concentrations of both drugs were more than 200 times the MIC. **Conclusion:** Azithromycin is effective in the treatment of moderate to severe shigellosis caused by multidrug-resistant *Shigella* strains."

167. Kirk M, Waddell R, Dalton C, Creaser A, Rose N. A prolonged outbreak of *Campylobacter* infection at a training facility. Commun Dis Intell 1997 Mar 6;21(5):57-61. 25 ref, Eng. Communicable Disease Control Branch, South Australian Health Commission, PO Box 6 Rundle Mall, South Australia 5000

168. Kollaritsch H, Que JU, Kunz C, Wiedermann G, Herzog C, Cryz SJ, Jr. Safety and immunogenicity of live oral cholera and typhoid vaccines administered alone or in combination with antimalarial drugs, oral polio vaccine, or yellow fever vaccine. J Infect Dis 1997 Apr;175(4):871-5. 18 ref, Eng. Institute for Specific Prophylaxis and Tropical Medicine, University of Vienna, A-1095 Vienna, Austria

"The effects of concomitant administration of antimalarial drugs, oral polio vaccine, or yellow fever vaccine on the immune response elicited by the *Vibrio choterae* CVD103-HgR and *Salmonella typhi* Ty2la live oral vaccines were investigated. Healthy adults were immunized with CVD103-HgR alone or combined with Ty21a. Subjects were randomized to simultaneously receive mefloquine, chloroquine or proguanil, or oral polio or yellow fever vaccine. The vibriocidal antibody seroconversion rate was significantly reduced (P=.008) only in the group that received chloroqune with the CVID103-HgR alone. The geometric mean vibriocidal antibody titer was significantly decreased in the groups that received chloroquine (P=.001) or mefloquine (P =.02) compared with titers in groups that received CVID103-HgR alone. However, similar immunosuppressive effects were not observed in the groups immunized with Ty2la and CVD103-HgR. Only the concomitant administration of proguanil effected a significant (P=.013) decline in the anti-*S. typhi* lipopolysaccharide antibody response. These results indicate that chloroquine and proguanil should not be simultaneously administered with the CVD103-HgR and Ty21a vaccine strains, respectively.

169. Koo D, Traverso H, Libel M, Drasbek C, Tauxe R, Brandling-Bennett D. [Epidemic cholera in Latin America, 1991-1993: implications of case definitions used for public health surveillance]. Pan Am J Public Health 1997 Feb;1(2):85-92. 17 ref, Eng. Epidemiology Program Office, MS C-08, Centers for Disease Control and Prevention, Atlanta, GA 30333, USA

170. Krishnan T, Naik TN. Electronmicroscopic evidence of torovirus like particles in children with diarrhoea. Indian J Med Res 1997 Mar;105:108-10. 10 ref, Eng. National Institute of Cholera and Enteric Diseases, P-33, CIT Road, Scheme XM, Beliaghata, Calcutta 700010, India

171. Lång H, Korhonen TK. The OmpS maltoporin of *Vibrio cholerae* as carrier of foreign epitopes. Behring Inst Mitt 1997 Feb;(98):400-9. 23 ref, Eng. Division of General Microbiology, Department of Biosciences, University of Helsinki, P.O. Box 56, SF 00014 Helsinki University, Finland

172. Layton MC, Calliste SG, Gomez TM, Patton C, Brooks S. A mixed foodborne outbreak with *Salmonella heidelberg* and *Campylobacter jejuni* in a nursing home. Infect Cont Hosp Epidemiol 1997 Feb;18(2):115-21. 16 ref, Eng. Bureau of Communicable Disease, New York City Department of Health, 125 Worth St., Room 300, Box 22A, New York City, NY 10013, USA

173. Levine MM, Galen J, Barry E, Noriega E, Tacket C, Sztein M, Chatfield S, Dougan G, Losonsky G, Kotloff K. Attenuated *Salmonella typhi* and *Shigella* as live oral vaccines and as live vectors. Behring Inst Mitt 1997 Feb;(98):120-3. 26 ref, Eng. Center for Vaccine Development, University of Maryland School of Medicine, 685 West Baltimore St., Baltimore, Maryland 21201, USA

174. Levine MM. Oral vaccines against cholera: lessons from Vietnam and elsewhere [commentary]. Lancet 1997 Jan 25;349(9047):220-1. 7 ref, Eng. Center for Vaccine Development, University of Maryland School of Medicine, Baltimore, MD 21201-1509, USA

175. Litwin CM, Leonard RB, Carroll KC, Drummond WK, Pavia AT. Characterization of endemic strains of *Shigella sonnei* by use of plasmid DNA analysis and pulsed-field gel electrophoresis to detect patterns of transmission. J Infect Dis 1997 Apr;175(4):864-70. 23 ref, Eng. Department of Pathology, University of Utah, 50 N. Medical Dr., Salt Lake City, UT 84132, USA

"Shigellosis is hyperendemic in Utah. Most isolates are *Shigella sonnei*, making it difficult to identify epidemiologic clustering. To better define transmission, molecular markers and epidemiologic data were examined for 90 cases. Plasmid analysis and pulsed-field gel electrophoresis (PFGE) of the *S. sonnei* isolates identified II and 4 patterns, respectively. Plasmid pattern 1 infections occurred in 8 day care centers over a 6-month period, suggesting spread between centers. Plasmid pattern III was isolated from children at 3 additional centers and pattern IV was associated with another day care center, suggesting different outbreaks. By PFGE, plasmid groups I and XI appeared identical, as were plasmid groups II and V; plasmid group X had a unique pattern. Plasmid groups III, IV, and VII-IX were closely related PFGE subtypes. Both plasmid analysis and PFGE allow better characterization *S. sonnei* transmission patterns of "endemic" strains and could lead to improved control measures."

176. López-Alarcón M, Villalpando S, Fajardo A. Breast-feeding lowers the frequency and duration of acute respiratory infection and diarrhea in infants under six months of age. J Nutr 1997 Mar;127(3):436-43. 31 ref, Eng. Unidad de Epidemiologia, Hospital de Pediatria, Centro Medico Nacional Siglo XXI, Instituto Mexicano del Seguro Social, Mexico, D.F. 06720, Mexico

"It remains unclear whether breast-feeding protects infants against acute respiratory infection (ARI). To determine if breast-feeding protects against ARI as it does against diarrhea, 170 healthy newborns were followed for 6 mo. Feeding mode, incidence and duration of ARI and diarrhea were recorded biweekly. Infants were classified as fully or partially breast-fed, or formula-fed. Incidence and prevalence were computed monthly. The effects of duration of breast-feeding and potential confounders were analyzed by multiple and logistic regression analyses. Incidence and prevalence of ARI were significantly lower in fully breast-fed infants than in formula-fed infants from birth up to 4 mo. as was the mean duration of individual episodes (5.1 ± 3.5 vs 6.4 ± 3.6 d, respectively). Incidence of ARI was negatively associated with duration of breast-feeding and positively associated with the presence of siblings ($P<0.05$). The prevalence of ARI was associated only with the duration of breast-feeding ($P<0.05$). Infants that were never breast-fed and that had one or more siblings were more likely to have an episode of ARI than those fully breast-fed at least 1 mo. Incidence, prevalence, and duration of individual episodes of diarrhea were also lower in breast-fed infants. Incidence ($r=-0.17$, $P<0.02$) and prevalence ($r=0.19$, $P<0.008$) were negatively associated with duration of full breast-feeding. Introduction of solid food was not associated with further episodes of diarrhea. The present results demonstrate protection against ARI as a result of breast-feeding similar to that for diarrhea, i.e., lower incidence and percentage of days ill, and episodes of shorter duration."

177. McClean P, Dodge JA, Nunn S, Carr KE, Sloan JM. Surface features of small-intestinal mucosa in childhood diarrheal disorders. J Pediatr Gastroenterol Nutr 1996 Dec;23(5):538-46. 19 ref, Eng. Department of Paediatrics, St. James's University Hospital, Beckett Street, Leeds LS9 7TF, England, UK

"The pathophysiology of diarrhea, especially in the otherwise healthy child, is still poorly understood. The aim of this study was to use the scanning electron microscope (SEM) to examine the surface of the jejunal mucosa of children with chronic nonspecific diarrhea (CNSD)

(n = 9) and to compare the findings with specimens obtained from children with (n = 21) and without (n = 21) other gastrointestinal diseases. Light microscopy of the specimens from children with CNSD was normal. However, SEM showed the presence of bacterial colonization with predominantly coccoid organisms in 100% of cases. This colonization was associated with loss of glycocalyx and clumping of the microvilli. The children with celiac disease (n = 9) all showed characteristic appearances with light microscopy, but only one had bacterial colonization on SEM. The surface features of specimens from children with other gastrointestinal disorders (food intolerance, postenteritis syndrome, protracted diarrhea of infancy, and immune deficiency states) were very similar to those from the CNSD group. Bacteria were visible on 89% of specimens, and in half of these cases the or- ganisms were bacilli. SEM of specimens from children with no gastrointestinal disease (ages 11-107 months) suggested an increased density of villi/unit area with advancing age. Bacteria were present in only two cases and did not include bacilli. The findings suggest that bacterial colonization of the surface of the small intestine is common in children with several gastrointestinal diseases and may play a part in their pathogenesis. Routine SEM examination of jejunal biopsies provides information not available from standard light microscopy, which may be relevant to the treatment of children with chronic diarrhea."

178. Mahalanabis D, Faruque ASG, Islam A, Hoque SS. Maternal education and family income as determinants of severe disease following acute diarrhoea in children: a case control study. J Biosoc Sci 1996 Apr;28(2):129-39. 13 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"In a case-control study among the urban poor of Dhaka, Bangladesh, the association of maternal education and family income with severity of disease due to diarrhoea in children was examined. After adjusting for family income, 7 or more years of school education was associated with 54% reduced risk of severe disease as indicated by the presence of dehydration. Income in the uppermost quartile of this population, independently of maternal education, was associated with 41% reduced risk of severe disease compared to the lowest quartile. In the logistic regression model the effect of maternal education remained high after adjustment for several confounders. Based on the concept that socioeconomic variables operate through a set of proximate variables it is contended that maternal education, independently of economic power, through its impact on disease from acute diarrhoea, favourably influences child survival."

179. Mahendru M, Prasad KN, Dhole TN, Ayyagari A. Rapid identification of *Campylobacter jejuni* strains by polymerase chain reaction & their restriction fragment length polymorphism analysis. Indian J Med Res 1997 Jan;105:9-14. 22 ref, Eng. Department of Microbiology, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow 226014, India

180. Mazumder RN, Hoque SS, Ashraf H, Kabir I, Wahed MA. Early feeding of an energy dense diet during acute shigellosis enhances growth in malnourished children. J Nutr 1997 Jan;127(1):51-4. 33 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"In a controlled clinical trial, we examined the effect of the short-term feeding of an energy-dense milk cereal formula in malnourished children with clinically severe dysentery due to acute shigellosis. Seventy-five malnourished children, aged 12-48 mo, passing blood or blood with mucous in the stool for =96 h, were offered a hospital diet. In addition, study children (n=36) were offered a milk-cereal formula with an energy of 5 kJ/g (an 11% protein diet); similarly, control children (n=39) were offered a milk-cereal formula with an energy content of 2.5 kJ/g (an 11% protein diet). Patients were admitted to the metabolic ward of the Clinical Research and Service Centre, Dhaka, at the International Centre for Diarrhoeal Disease Research, Bangladesh. Patients were studied for 10 hospital days and were then followed up at home after 30 d. After 10 d of dietary intervention, children in the study group had a significantly greater increase vs. controls in weight-for-age (6 vs. 3%, P<0.001) and in weight-for-height (7 vs. 3%, P<0.001). Serum prealbumin concentrations were significantly higher (study vs. control) after 5 d (0.214 vs.

0.170 g/L, P=0.01) and after 10 d (0.244 vs. 0.193 g/L, P=0.006) of the study. Greater weight-for-age was sustained at home 1 mo after discharge (8 vs. 5%, P=0.005) from the hospital. Similarly, higher weight-for-height was sustained 1 mo after discharge (8 vs. 5%, P=0.01). During their stay at home, there was no dietary intervention. The results of this study suggest that short-term feeding of an energy-dense diet enhances growth in malnourished children with acute dysentery due to shigellosis."

181. Mazumder RN, Salam MA, Ali M, Bhattactharya M. Reactive arthritis associated with *Shigella dysenteriae* type 1 infection. J Diarrhoeal Dis Res 1997 Mar;15(1):21-4. 10 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"*Shigella dysenteriae* type 1 causes the most severe form of bacillary dysentery. The spectrum of illness ranges from mild watery diarrhoea to severe bloody diarrhoea. Shigellosis is often associated with intestinal complications, including intestinal perforation, intestinal obstruction, toxic dilatation of the colon, and prolapse of the rectum; systemic complications include septicaemia, hyponatraemia, hypoglycaemin, seizure, encephalopathy, haemolytic-uraemic syndrome, and malnutrition. Arthritis and conjunctivitis are rare extra-intestinal complications of shigellosis. Annually, about 110,000 patients receive treatment in the Dhaka Hospital of the International Centre for Diarrhoea] Disease Research, Bangladesh for diarrhoea and diarrhoea-associated illnesses, of which 11% are due to shigellosis. However, arthritis associated with shigellosis has not been reported from this population. Arthritis has been reported in association with infection due to *S. flexneri* and *S. sonnei* from other places. We are unaware of any reported case of arthritis in association with *S. dysenteriae* type 1 infectious. In this report, we describe the clinical and laboratory features of a young woman who developed arthritis following *S. dysenteriae* type 1 infection."

182. Mellander A, Mattsson A, Svennerholm A-M, Sjövall H. Relationship between interdigestive motility and secretion of immunoglobulin A in human proximal small intestine. Dig Dis Sci 1997 Mar;42(3):554-67. 39 ref, Eng. Department of Internal Medicine, Sahlgrenska University Hospital, S-413 45 Göteborg, Sweden

183. Moser I, Schröder W. Hydrophobic characterization of thermophilic *Campylobacter* species and adhesion to INT 407 cell membranes and fibronectin. Microb Pathog 1997 Mar;22(3):155-64. 20 ref, Eng. Institut für Mikrobiologie und Tierseuchen, Freie Universität Berlin, Fabeckstr, 36A, 14195 Berlin, Germany

184. Mourad FH, O'Donnell JD, Dias JA, Ogutu E, Andre EA, Turvill JL, Farthing MJG. Role of 5-hydroxytryptamine type 3 receptors in rat intestinal fluid and electrolyte secretion induced by cholera and *Escherichia coli* enterotoxins. Gut 1995 Sep;37(3):340-5. 38 ref, Eng. Digestive Diseases Research Centre, Medical College of St. Bartholomew's Hospital, Charterhouse Square, London EC1M 6BQ, UK

"Cholera toxin and *Escherichia coli* heat labile toxin (LT) induced intestinal secretion has in the past been attributed exclusively to an increase in intracellular cAMI' whereas *E. coli* heat stable toxin (ST) induced secretion is mediated through cGMP. Evidence is accumulating on the importance of 5-hydroxytryptamine (5-HT) in cholera toxin induced secretion, but its role in LT and ST is not well established. This study therefore investigated in vivo the effect of 5-HT_3 receptor antagonist, granisetron, on intestinal fluid and electrolyte secretion induced by cholera toxin, LT, and ST. Granisetron (30, 75, 150, or 300 µg/kg) was given subcutaneously to adult male Wistar rats 90 minutes before instillation of 75 µg cholera toxin or 50 µg LT in isolated whole small intestine. In situ small intestinal perfusion was performed with iso-osmotic plasina electrolyte solution (PES) to assess fluid movement. In second group of animals, granisetron (300 µg/kg) was given subcutaneously and two hours later small intestinal perfusion with PES containing. Cholera toxin induced net fluid secretion (median -50.1 µl/min/g (inter- quartile range -

59.5 to -29.8)) was found to be dose dependently decreased or abolished by granisetron (plateau effect at 75 µg/kg: 18 (-7.8 to 28), p<O-01). Granisetron in high dose (300 µg/kg), however, failed to prevent LT or ST induced secretion (-52 (-121 to -71) v -31 (-44 to -18), and (-39 (-49 to 17) v (-22 (-39 to -3)), respectively. Sodium and chloride movement parallel that of fluid. In conclusion, these data show that 5-HT and 5-HT_3 receptors play an important part in cholera toxin induced secretion but are not involved in *E. coli* heat stable or heat labile toxin induced secretion."

185. Nagy B, Whipp SC, Imberechts H, Bertschinger HU, Dean-Nystrom EA, Casey TA, Salajka E. Biological relationship between F18ab and F18ac fimbriae of enterotoxigenic and vertoxigenic *Escherichia coli* from weaned pigs with oedema disease or diarrhoea. Microb Pathog 1997 Jan;22(1):1-11. 29 ref, Eng. Veterinary Medical Research Institute, Hungarian Academy of Sciences, Budapest, Hungary

186. Naravane A, Lindo JF, Williams LAD, Gardener MT, Fletcher CK. *Ascaris lumbricoides* in the paranasal sinuses of a Jamaican adult. Trans R Soc Trop Med Hyg 1997 Jan-Feb;91(1):37. 3 ref, Eng. Department of Anatomy, The University of the West Indies, Mona, Kingstom 7, Jamaica

187. Nath G, Panda S, Sharma BM. Epithelial adherence of *Candida albicans* is enhanced by passage through rat small intestine. J Diarrhoeal Dis Res 1996 Dec;14(4):286-8. 14 ref, Eng. Department of Microbiology, Institute of Medical Sciences, Banaras Hindu University, Varanasi 221 005, India

"Seven *Candida albicans* isolates (four from patients with diarrhoea and three from healthy persons) underwent two passages through rat ileal loop (RIL) to see the effect of consecutive passages on the adherence to rat intestinal epithelium. The isolates from patients with diarrhoea showed a significant enhancement in adherence after the first passage ($1.95x10^4$ cfu/cm^2 versus $3.67x10^4$ cfu/cm^2). There was no further increase between the first passage ($3.67x10^4$ cfu/cm^2) and the second one ($3.61x10^4$ cfu/cm^2). A similar pattern was observed with the three non-diarrhoeal isolates. Animal passage of this fungus probably leads to better interactions between the cell surfaces causing the enhanced adherence."

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189. Pasquali P, Fayer R, Almeria S, Trout J, Polidori A, Gasbarre LC. Lymphocyte dynamic patterns in cattle during a primary infection with *Cryptosporidium parvum*. J Parasitol 1997 Apr;83(2):247-50. 24 ref, Eng. Istituto di Parassitologia, Facolta' di Medicina Veterinaria, Universita delgi Studi di Perugia, Perugia, Italy

190. Peina Y, Jianhui L, Siguo D, Lijun J, Bingrui W, Lijun J, Rong Q, Yi R, Jinrong C. Detection of serological antibodies after immunization with a killed whole-cell vaccine against *Vibrio cholerae* O139 in human. Prog Microbiol Immunol 1997 Mar;25(1):6-12. 10 ref, Eng. National Institute for the Control of Pharmaceutical and Biological Products, Beijing 100050, People's Republic of China

"50 students (middle school) were immunized by intramuscular injection with a killed whole-cell vaccine against vibrio cholerae 0139, each received a close of 4.5 x lO^9 cells in 0.5 ml. In another 50 students, each received a dose of 9.O x 10^9 cells in l ml. After a month, respectively 80% vaccinees had serum vibriocidal antibody, the titres ranged from 1:20 to 1:160. It had a little declined by 3 months, however this antibody still gave positive reaction in 68-72% students. Serum agglutination antibody and anti-CT antibody were detected and increased at same times.

These results indicated that killed whole-cell vaccine against 0139 cholera, should be used as short-time effective vaccine and emergency vaccine."

191. Pérez-Cuevas R, Guiscafré H, Romero G, Rodríguez L, Gutiérrez G. Mothers' health-seeking behaviour in acute diarrhoea in Tlaxcala, Mexico. J Diarrhoeal Dis Res 1996 Dec;14(4):260-8. 37 ref, Eng. Inter-institutional Health Systems Research Group, The Ministry of Health, Mexican Social Security Institute, Mexico DF, Mexico

"This study, a cross-sectional survey, was conducted to assess how mothers take care of their children with diarrhoea and to develop a model of health-care seeking behaviour. Multistage sampling was used. Mothers whose children aged less than five years had suffered from diarrhoea in the last fortnight were included. Nurses interviewed the mothers to collect data. Variables included in the interview were: mothers' characteristics, children's characteristics, clinical data, treatment given by the mother, maternal health-seeking behaviour and mothers' information about diarrhoea and dehydration. Variables corresponding to the clinical data were grouped to identify dehydration signs and the need for medical care. Dehydration was defined as the presence of two or more of the following reported signs: thirst, sunken eyes, sunken fontanelle, or scanty urine. The need for medical care was defined as the presence of one or more of the following characteristics: illness lasting more than three days, vomiting, fever, bloody diarrhoea or dehydration. A sample of 747 mothers was obtained. Household treatments consisted of herbal teas to stop diarrhoea (52.3%), liquids to prevent dehydration (92.2%), symptomatic drugs (35.2%) and changes in feeding patterns (36.3%), which consisted in suppressing milk and dairy products and interrupting breast feeding (12.2%). Mothers sought medical assistance when they perceived a worsening of clinical conditions. Clinical signs statistically associated with their decision were: bloody diarrhoea, vomiting, illness longer than three days, weight loss, and fever. The signs of dehydration were not associated with health care-seeking because the mother did not recognise them. It is concluded that maternal educational programmes should emphasise, besides the proper use of oral rehydration therapy, teaching mothers to identify signs of dehydration as an indication to seek timely medical care."

192. Prasad KN, Dhole TN, Ayyagari A. Adherence, invasion and cytotoxin assay of *Campylobacter jejuni* in HeLa and HEp-2 cells. J Diarrhoeal Dis Res 1996 Dec;14(4):255-9. 21 ref, Eng. Department of Microbiology, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow 226014, India

"*Campylobacter jejuni* is an important human enteropathogen worldwide. Chickens are the major reservoir and source of campylobacter infection. Ten clinical isolates from human and five chicken strains were tested for the adherence, invasion and cytotoxin assay in HeLa and HEp-2 cells. All human strains adhered to both the HeLa (10^3 to $3x10^4$ bacteria/mL of cell lysate) and HEp-2 cells ($2x10^3$ to $4x10^4$ bacteria/mL of lysate). All chicken strains also adhered to the HEp-2 cells (10^2 to 10^3 bacteria/mL), but only two strains adhered to the HeLa cells. Six clinical and none of the chicken strains invaded the mammalian cells. Both the adherence and invasion were better observed in HEp-2 than in HeLa cell lines. All three isolates from patients having invasive diarrhoea and only one strain from a patient having watery diarrhoea produced cytotoxin. All three invasive strains also adhered to polystyrene surface after the localised destruction of the HEp-2 cells, a phenomenon not reported earlier. Adherence was markedly inhibited by the whole cell lysate and the acid glycine extracts, and the results were comparable. This study indicates that the clinical isolates of *C. jejuni* are more virulent than the chicken strains, HEp-2 is better for the adherence/invasion assay and HeLa is better for cytotoxin assay. The acid glycine extracts probably contain the key adhesins for *C. jejuni*."

193. Rahman MM, Mitra AK, Mahalanabis D, Wahed MA, Khatun M, Majid N. Absorption of nutrients from an energy-dense diet liquefied with amylase from germinated wheat in infants with acute diarrhea. J Pediatr Gastroenterol Nutr 1997 Feb;24(2):119-23. 15 ref,

Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"**Background**: Addition of a small amount of amylase rich flour (ARF) to a thick porridge instantly liquefy the porridge and increase the energy intake even by sick children. The present study examined the absorption of macronutrients and calorie from an energy dense diet liquefied with ARF in children aged 6-11 months with acute watery diarrhea. **Methods**: After adequately hydrated with oral rehydration fluid over a period of 24 hours children were randomly assigned to receive either an ARF treated liquefied porridge (test diet) or a porridge diluted with water (control diet). A 72-hour metabolic balance was performed to determine the absorption of carbohydrate, fat, protein, and calorie. **Results**: Thirteen infants received the test diet, and 15 infants received the control diet. The intake of protein (g/kg/d), carbohydrate (g/kg.d), fat (g/kg.d) and calorie (kj/kg.d) were 1.97, 20.6, 4.3 and 548 respectively in the test group and those in the control group were 1.12, 13.3, 2.8 and 356. The stool loss of protein, carbohydrate and fat were comparable in the two groups. The absorption coefficient (%) of carbohydrate, fat and energy were 69.6, 61.3 and 65.4 in the test group and were 73.2, 58.6 and 66.7 in the control group. The coefficient of absorption of protein was significantly higher in the test group (37.7% vs. 21.7%). The mean (95% CI) nitrogen balance (g/kg.d) in the test and control groups were 0.064 (0.026, 0.102) and -0.029 (-0.055, 0.003) respectively. **Conclusions**: The results suggest that energy dense diet liquefied with ARF was well absorbed in children with acute diarrhea and there was a positive nitrogen balance that may have a positive impact in preventing weight loss during acute illness."

194. Rensheng J, Yiwen N, Keliang H, Jinye Y, Wenwu L, Guangzhan S, Shaobei L, Jianqiang H, Peina Y, Siguo D, Jianping W, Jianhui L, Bingrui W, Lijun J, Yan W, Jinrong C. Reaction observation of human immunized with cholera vaccine type O139. Progr Microbiol Immunol 1997 Mar;25(1):1-5. 7 ref, Eng. Epidemic Prevention Station of Guangxi Zhuang Autonomous Region, Nanning 530021, People's Republic of China

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196. Robertson LJ. Severe giardiasis and cryptosporidiosis in Scotland, UK. Epidemiol Infect 1996 Dec;117(3):551-61. 20 ref, Eng. Scottish Parasite Diagnostic Laboratory, Department of Bacteriology, Stobhill NHS Trust, Glasgow G21 3UW, Scotland, UK

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"The IgG subclass response to *Shigella sonnei* and *Shigella flexneri* 2a lipopolysaccharide (LPS) was examined in subjects naturally exposed to these organisms. Affinity-purified LPS antibodies obtained using a column of *Shigella* LPS bound to epoxy-activated sepharose 6B were used as standards to calibrate the serum antibody response to natural *Shigella* infection. The geometric mean concentrations of specific IgG in sera from those not exposed to *Shigella* organisms were 7.9 µg/mL against *S. sonnei* LPS and 18.6 µg/mL against *S. flexneri* 2a LPS. After natural exposure to *S. sonnei* or *S. flexneri* 2a, the concentrations rose to 30.3 and 127.9 µg/mL, respectively. IgG2 was the major component in the anti-*S. flexneri* subclass response, while the anti-*S. sonnei* response was dominated by IgGI. High levels of IgGI antibodies before exposure to organisms from either *Shigella* serogroup correlated with a lower risk of developing symptomatic infection."

198. Ronan A, Azad AK, Rahman O, Phillips RE, Bennish ML. Hyperglycemia during childhood diarrhea. J Pediatr 1997 Jan;130(1):45-51. 19 ref, Eng. Department of Emergency Medicine, John Hunter Hospital, Locked Bag 1, Hunter Region Mail Centre, Newcastle, New South Wales 2310, Australia

"*Objective*: To determine the cause of hyperglycemia in childhood diarrhea. *Methods*: During an 8-month period, patients admitted to a diarrhea treatment center in Bangladesh had their blood glucose concentrations determined. Sixteen patients aged 2 to 10 years with hyperglycemia (blood glucose concentration >10.0 mmol/L) and 20 patients in the same age group with a normal blood glucose concentration (3.3 to 9.0 mmol/L) had blood samples obtained on admission and 4 and 24 hours later for determination of glucoregulatory hormones and gluconeogenic substrates. *Results*: Prevalence of hyperglycemia among patients aged 2 to 10 years was 9.4%. Compared with the normoglycemic patients, hyperglycemic patients more often had severe dehydration (100% versus 10%, p<0.001), infection with *Vibrio cholerae* 01 or toxigenic *Escherichia coli* (94% vs 25%, p<0.001), and had similar duration of fasting (16 vs 14 hours, p=0.677). Concentrations of epinephrine (7.15 vs 2.00 µmol/L), glucagon (36 vs 14 pmol/L), and C-peptide (1.22 vs 0.35 nmol/L) were all significantly (p=0.014) higher in patients with hyperglycemia than in normoglycemic patients. *Conclusions*: The development of hyperglycemia in diarrhea is caused by a stress response to hypovolemia."

199. Rotman HL, Schnyder-Candrian S, Scott P, Nolan TJ, Schad GA, Abraham D. IL-12 eliminates the Th-2 dependent protective immune response of mice to larval *Strongyloides stercoralis*. Parasite Immunol 1997 Jan;19(1):29-39. 51 ref, Eng. Department of Microbiology and Immunology, Thomas Jefferson University, Philadelphia, PA 19107, USA

200. Sack RB, Rahman M, Yunus M, Khan EH. Antimicrobial resistance in organisms causing diarrheal disease. Clin Infect Dis 1997 Jan;24(1 Suppl):S102-5. 11 ref, Eng. Johns Hopkins University, School of Hygiene and Public Health, 615 North Wolfe Street, Baltimore, Maryland 21205, USA

"Antimicrobial resistance is becoming increasingly important in the treatment of enteric infections, particularly those due to *Shigella, Vibrio cholerae*, enterotoxigenic *Escherichia coli* (associated with traveler's diarrhea), and *Salmonella typhi*. The rate of antimicrobial resistance is highest in the developing world, where the use of antimicrobial drugs is relatively unrestricted. Of greatest immediate concern is the need for an effective, inexpensive antimicrobial that can be used safely as treatment for small children with dysentery due to *Shigella*, primarily *Shigella dysenteriae* type 1."

201. Sharma C, Nair GB, Mukhopadhyay AK, Bhattacharya SK, Ghosh RK, Ghosh A. Molecular characterization of *Vibrio cholerae* O1 biotype El Tor strains isolated between 1992 and 1995 in Calcutta, India: evidence for the emergence of a new clone of the El Tor biotype. J Infect Dis 1997 May;175(5):1134-41. 28 ref, Eng. Institute of Microbial Technology, Sector 39A, Chandigarh 160036, India

202. Singh DV, Sanyal SC. Production of haemolysin and enterotoxin by *Aeromonas jandaei* and *Aeromonas trota* strains after animal passage. J Diarrhoeal Dis Res 1996 Dec;14(4):274-9. 40 ref, Eng. Department of Microbiology, Institute of Medical Sciences, Banaras Hindu University Varanasi 221 005, India

"Five *Aeromonas jandaei* and 12 *Aeromonas trota* isolates were tested for the production of haemolysin and enterotoxin, and the correlation between these two properties. The majority (10 isolates) of the strains produced ß-haemolysis. The titres of haemolytic activity for both species were 8-64 HU/mL. In the initial ileal loop test, only two (*A. trota*) of the 17 isolates produced enterotoxin. One each of these 2 *A. trota* strains was ß-haemolytic and non-haemolytic. The remaining isolates of *A. trota* and *A. jandaei* included a-, ß-and non-haemolytic strains, and failed

to cause any fluid accumulation in the initial tests, but did so after one-to-five sequential passages through the rabbit ileal loops. Three a- and 4 non-haemolytic strains switched over to the production of ß-haemolysis when they showed the positive ileal loop reaction. However, on repeated subcultures or on storage in the laboratory, all of them reverted back to their original a- or non-haemolytic character and no longer produced enterotoxin."

203. Singh J, Bora D, Sachdeva V, Sharma RS, Verghese T. *Vibrio cholerae* 01 and 0139 in less than five years old Children Hospitalised for Watery Diarrhoea in Delhi, 1993. J Diarrhoeal Dis Res 1997 Mar;15(l):3-6. 14 ref, Eng. National Institute of Comniunicable Diseases, 22 Shamnath Marg, Delhi 110054, India

"In Delhi, patients with cholera-like illness are admitted to the Infectious Diseases Hospital. In 1993, rectal swabs from 836 such patients aged less than five years were examined for the presence of *Vibrio cholerae* 01 and 0139. Of them, 232 (28%), 180 (22%), and 424 (51%) were found suffering from 01 cholera, 0139 cholera, and non-cholera watery diarrhoea respectively. Twelve children (1.4%) excreted both *V. cholerae* 01 and 0139. Both types of cholera were similarly distributed by age, with 19% of the cases occurring in infants. The findings indicate that cholera should be suspected in children aged less than two years and in infants with acute watery diarrhoea. For both serotypes, males were more represented than females; the differences were, however, not significant. Clinical features of patients with *V. cholerae* 0139 and 01 were indistinguishable, except that a significantly higher percentage of the former had fever. Potential risk factors for cholera were almost equally prevalent in the families of children aged less than 5 years having either 01 or 0139 cholera. The results suggest a similar mode of transmission of the two serotypes in children. By inference, the preventive and control measures are also likely to be similar."

204. Slutsker L, Ries AA, Greene KD, Wells JG, Hutwagner L, Griffin PM. *Escherichia coli* O157:H7 diarrhea in the United States: clinical and epidemiologic features. Ann Intern Med 1997 Apr 1;126(7):505-13. 43 ref, Eng. Foodborne and Diarrheal Diseases Branch, National Center for Infectious Diseases, Centers for Disease Control and Prevention, Atlanta, GA 30333, USA

205. Sodeinde O, Adeyemo AA, Gbadegesin RA, Olaleye BO, Ajayi-Obe KE, Ademowo OG. Interaction between acute diarrhoea and falciparum malaria in Nigerian children. J Diarrhoeal Dis Res 1996 Dec;14(4):269-73. 20 ref, Eng. Department of Paediatrics, University College Hospital, P.M.B. 5116, Ibadan

"Although both malaria and diarrhoea are major public health problems in developing countries, and separately each has been the subject of intense research, few studies have investigated the interaction between these two conditions. The interaction between diarrhoea and malaria among children aged 4 months to 12 years in two tertiary health-care facilities, University College Hospital, Ibadan, and Lagos University Teaching Hospital, Lagos, Nigeria was studied. In Ibadan, the prevalence of diarrhoea among the cerebral malaria patients on admission was 11.7% (7/60) compared to 9.3% (215/2312) among other admissions in 1990 (chi square=0.16; p=0.6913). Similarly, no significant difference in the prevalence of diarrhoea was found between the cerebral malaria patients (14.3%) and other patients (16.1%) seen in Lagos in 1992 (chi square=0.06, p=0.81). Thus, cerebral malaria does not seem to be associated with an increased or decreased prevalence of diarrhoea when compared with other conditions. The prevalence of malarial parasitaemia among the 554 diarrhoea patients studied in Ibadan during 1993-1994 was 13.6% compared with 17.9% among the 347 controls (chi square=3.75, p=0.053). However, of the children with diarrhoea, malarial parasitaemia was more common among the dehydrated patients (25.4%) than among the well-hydrated patients (11.6%) (chi square=8.11, p=0.004). These data suggest that diarrhoea is merely coincidental in severe malaria and conversely, malarial parasitaemia is similarly coincidental in children with acute diarrhoea, although it may be more frequent among dehydrated diarrhoea patients than well-hydrated ones."

206. Starr JM, Rogers TR, Impallomeni M. Hospital-acquired *Clostridium difficile* diarrhoea and herd immunity. Lancet 1997 Feb 8;349(9049):426-8. 16 ref, Eng. Department of Geriatric Medicine, University of Edinburgh, Royal Victoria Hospital, Edinburgh EH4 2DN, UK

207. Sun WM, Read NW, Verlinden M. Effects of loperamide oxide on gastrointestinal transit time and anorectal function in patients with chronic diarrhoea and faecal incontinence. Scand J Gastroenterol 1997 Jan;32(1):34-8. 11 ref, Eng. Department of Medicine, Royal Adelaide Hospital, University of Adelaide, Adelaide, South Australia

Background: Loperamide improves anorectal function in patients with chronic diarrhoea. We wished to investigate whether the prodrug loperamide oxide has similar effects. *Methods:* Eleven patients with chronic diarrhoea and faecal incontinence participated in a randomized, placebo-controlled, double-blind, crossover study of the effects of loperamide oxide (4 mg twice daily for 1 week). *Results:* Loperamide oxide reduced wet stool weight and improved the patients' ratings of symptoms. Mouth-to-caecum transit time was not altered, but whole-gut transit time was prolonged. There were limited effects on anorectal function, but the mean minimum basal pressure mainly contributed by the internal anal sphincter (IAS) was increased, as was the mean volume infused before leakage occurred in the same continence test. *Conclusion:* Loperamide oxide is effective in the treatment of diarrhoea with faecal incontinence; normalization of colon transit time and an increase in the tone of the IAS seem to be the main determinants of efficacy."

208. Tamura S-i, Hatori E, Tsuruhara T, Aizawa C, Kurata T. Suppression of delayed-type hypersensitivity and IgE antibody responses to ovalbumin by intranasal administration of *Escherichia coli* heat-labile enterotoxin B subunit-conjugated ovalbumin. Vaccine 1997 Feb;15(2):225-9. 25 ref, Eng. Department of Pathology, National Institute of Health, 1-23-1 Toyama, Shinjuku-ku, Tokyo 162, Japan

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210. Thomson ABR, Wild G. Adaptation of intestinal nutrient transport in health and disease. Pt. I. Dig Dis Sci 1997 Mar;42(3):453-69. 254 ref, Eng. 519 Robert Newton Research Building, University of Alberta, Edmonton, Alberta T6G 2C2, Canada

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212. Tsuji T, Yokochi T, Kamiya H, Kawamoto Y, Miyama A, Asano Y. Relationship between a low toxicity of the mutant A subunit of enterotoxigenic *Escherichia coli* enterotoxin and its strong adjuvant action. Immunology 1997 Feb;90(2):176-82. 30 ref, Eng. Department of Microbiology, Fujita Health University, School of Medicine, Toyoake, Aichi 470-11, Japan

213. Vanderhoof JA, Murray ND, Paule CL, Ostrom KM. Use of soy fiber in acute diarrhea in infants and toddlers. Clin Pediatr 1997 Mar;36(3):135-9. 32 ref, Eng. Department of Pediatrics, University of Nebraska College of Medicine, 8300 Dodge Street, Suite 330, Omaha, NE 68114, USA

214. Vartanian MD, Girardeau J-P, Martin C, Rousset E, Chavarot M, Laude H, Contrepois M. An *Escherichia coli* CS31A fibrillum chimera capable of inducing memory antibodies in outbred mice following booster immunization with the entero-pathogenic coronavirus transmissible gastroenteritis virus. Vaccine 1997 Feb;15(2):111-20. 28 ref, Eng. Laboratoire

de Microbiologie, Institut National de la Recherche Agronomique, Centre de Recherches de Clermont-Ferrand-Theix, 63122, Saint-Genes-Champanelle, France

215. Victora CG, Fuchs SC, Kirkwood BR, Lombardi C, Barros FC. Low body weight: a simple indicator of the risk of dehydration among children with diarrhoea. J Diarrhoeal Dis Res 1997 Mar;15(l):7-11. 8 ref, Eng. Departamento de Medicina Social, Universidade Federal de Pelotas, CP 464, 96001-970, Pelotas, RS, Brazil

"The early identification of children at high risk of dehydration as a consequence of diarrhoea would be of great value for health care workers in developing countries. By comparing children aged less than two years with diarrhoea and moderate to severe dehydration with matched controls who had uncomplicated diarrhoea, a number of prognostic factors were assessed. Low body weight, regardless of age, was strongly associated with the risk of dehydration; using 7.0 kg as a cut-off, it had a sensitivity of 75% and a specificity of 68%. Low body weight was superior to more complex anthropometric indices, including weight for age, weight for length or length for age, and also to early signs and symptoms during the episode. By reflecting the effects of both young age and those or malnutrition, low body weight may prove to be a simple indicator for predicting dehydration among children with diarrhoea presenting at a health service."

216. Wenzl HH, Fine KD, Ana CA, Porter JL, Fordtran JS. Effect of fludrocortisone and spironolactone on sodium and potassium losses in secretory diarrhea. Dig Dis Sci 1997 Jan;42(1):119-28. 45 ref, Eng. Baylor University Medical Center, Department of International Medicine, 3500 Gaston Ave., Dallas, Texas 75246, USA

"The response of the colon to aldosterone is believed to be an important adaptive mechanism to excessive sodium losses in diarrhea. However, the degree to which mineralocorticoid activity actually influences fecal output of sodium in people with diarrhea is unknown. To gain insight into this question, 10 normal people were treated with placebo, fludrocortisone (an aldosterone agonist), and spironolactone (an aldosterone antagonist) during three experimental periods lasting nine days. On days 5-8, diarrhea was induced by ingestion of phenolphthalein. Diet was controlled. Fecal sodium was 40 meq/day on placebo and 29 meq/day on fludrocortisone, consistent with mineralocorticoid stimulation of intestinal sodium absorption. However, contrary to our expectations, spironolactone therapy was also associated with a fall in fecal sodium output, to 28 meq/day. To explain this paradoxical effect of spironolactone, we suggest that sodium depletion caused by spironolactone's natriuretic action on the kidney caused the release of an unknown stimulant of intestinal sodium absorption, whose action more than overcame the reduced colonic absorption resulting from inhibition of aldosterone activity by spironolactone. This interpretation implies that the intestinal adaptation to sodium depletion in diarrhea involves both aldosterone and an aldosterone independent factor, working in concert to reduce fecal sodium output."

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219. Ye X-P, Donnelly CA, Fu Y-L, Wu Z-X. The non-randomness of the distribution of *Trichuris trichiura* and *Ascaris lumbricoides* eggs in faeces and the effect of stirring faecal

specimens. Trop Med Int Health 1997 Mar;2(3):261-4. 13 ref, Eng. The Wellcome Trust Centre for the Epidemiology of Infectious Disease, Department of Zoology, University of Oxford, South Parks Road, Oxford OX1 3 PS, UK

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