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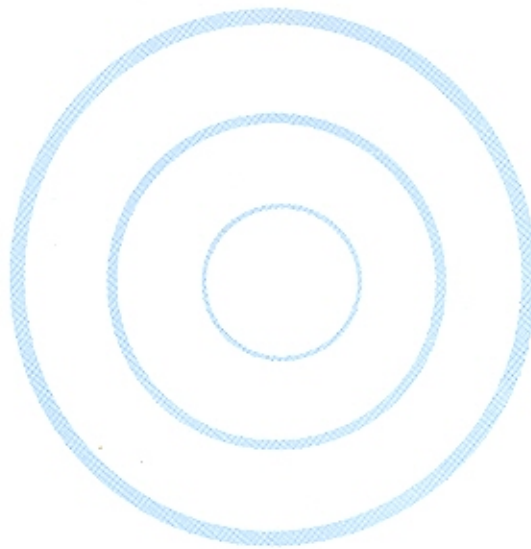

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Environmental and Personal Hygiene Practices: Risk Factors for Diarrhoea Among Children of Nigerian Market Women

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ABSTRACT

A cross-sectional survey was carried out to determine the environmental and personal hygiene practices of mothers of children aged less than five years in two markets in lbadan—one with poor sanitary conditions (Bodija) and the other one with better sanitation facilities (Gbagi). The study sought to identify the risk factors for diarrhoea among these children. Two hundred and sixty-six mothers in Bodija and 260 in Gbagi were interviewed. A questionnaire was used for collecting information on social and demographic characteristics, personal and environmental hygiene practices, including sources of food and water for their children, waste-disposal practices and occurrence of diarrhoea among their children aged less than five years. The educational status of the women in Bodija was lower than that of the women in Gbagi ($p<0.001$). Sixty (23%) women of the Bodija market mentioned that tap water was the source of drinking water for their children, while 91 (34%) brought water from their homes, and 45 (17%) bought it from vendors in the market. The corresponding figures for women of the Gbagi market were 41 (16%), 98 (38%) and 19 (7%). Two hundred and thirty-four (90%) women in Gbagi prepared breakfast at home for their children compared to 216 (81%) women in Bodija. This difference was statistically significant ($p<0.05$). Waste disposal and personal hygiene practices were poorer among the women in Bodija. Yet the occurrence of diarrhoea was not significantly different in both the markets. Risk factors for diarrhoea identified in this study were water and food bought from vendors, child defaecation practices, mothers' cleaning up practices after child's defaecation, and refuse-disposal practices. The inherent risk of sale of unwholesome food and water by vendors is a great concern for public health authorities in Nigeria. Efforts to control diarrhoea must not only be focused on improving mothers' knowledge about food hygiene but also on environmental hygiene practices within the community.

Key words: Diarrhoea, Infantile; Hygiene; Sanitation; Knowledge, attitudes, practice; Environment; Risk factors; Cross-sectional studies

INTRODUCTION

Diarrhoea continues to be a major cause of morbidity and mortality in the developing world (1). In Nigeria, the major single cause of infant and child mortality is death due to dehydration, with an estimated 120,000 deaths due to diarrhoea per year (2). Consequently, development agencies, such as World Health Organization, UNICEF, and USAID introduced oral

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dehydration therapy (ORT) as the main child survival strategy for diarrhoeal diseases, and such preventive measure has been found to be most effective (3). However, ORT cannot prevent subsequent episodes of diarrhoea. There is, therefore, a need to explore factors notably environmental and personal factors, that may predispose children to diarrhoeal diseases.

Most markets in Nigeria are characterized by unsanitary conditions, including poor water supply and poor drainage systems, unsanitary waste disposal, and overcrowding, resulting in poor personal and environmental hygiene. Although infants and young children of the Nigerian market women are constantly exposed to health hazards associated with such an unsanitary environment, the authors are not aware of any study carried out in Nigeria that documents the effects of these adverse environmental conditions on the health of the Nigerian children. The present study attempts to determine the environmental and personal hygiene practices of market women in lbadan and also to establish the risk factors in the occurrence of diarrhoea among their children aged less than five years.

MATERIALS AND METHODS

This cross-sectional study was carried out between September 1996 and March 1997 in the Bodija and Gbagi markets, both situated in different areas of lbadan. The Bodija market is predominantly a food market with 3,596 stalls, while the Gbagi market is mainly a textile market with 2,853 stalls. The Gbagi market is clean and is well laid out with tarred roads, good drainage, and an effective refuse-disposal system. The Bodija market, on the other, is overcrowded with more than half of the traders displaying their food items along the roadside away from the designated stalls. The roads are not tarred and are littered with refuse. Thus, environmental sanitation is poor in the Bodija market.

Permission to carry out the study was sought from the chairpersons of the two local government areas where the markets are located. In addition, the leaders of the Market Women Association in each market were contacted to discuss the objectives of the study and to obtain their consent. Informed consent were also obtained from mothers who participated in the study.

Each market is divided into blocks of stalls which are numbered in a serial order. Only those mothers were included in this cross-sectional survey whose children, aged less than five years, have been staying with them in the market for at least 6 months. A sample-size estimation was calculated for both the markets, using a prevalence of diarrhoea of 8% (4), a precision limit of 1%, and a confidence level of 95%. This gave a minimum sample size of 260 mothers. Stalls were selected using a systematic random-sampling technique in which every 7th stall was chosen. Where no children aged less than five years were found in a particular stall, the next stall was selected. In all, 266 mothers from the Bodija market and 260 from the Gbagi market were included in the survey. A structured questionnaire was administered to these mothers by the trained interviewers. The questionnaire, pretested on 20 mothers in another market before the main study was undertaken, was used for collecting information on the social and demographic characteristics of the respondents, personal and environmental hygiene practices, including the sources of food and water for their children, and the occurrence of diarrhoea among the children. A child was considered to have had diarrhoea if he had passed 3 watery stools within 24 hours. Questionnaires were coded, and statistical analysis was done using the EPI Info software, version 6.02, to calculate frequencies and chi-square analysis to check associations between variables.

RESULTS

The age distribution of 266 women of the Bodija market and 260 mothers of the Gbagi market, who were interviewed, is presented in Table 1, and their educational attainments are presented in Table 2. The age distribution of the women of both the markets was found to be similar. Educational attainment of the women in Gbagi was significantly higher than that of the women of the Bodija market ($p<0.001$).

Table 1. Age distribution of studypopulation

Mother's age (in years)	Bodija (n=266)	Gbagi (n=260)
Under 21	13(5%)	5(2%)
21-25	58(22%)	72(28%)
26-30	89(33%)	94(36%)
31-35	72(27%)	59(23%)
36-40	28(11%)	25(10%)
41-45	4(2%)	3(1%)
46-50	2(1%)	0(0%)
Over 50	0(0%)	2(1%)

Chi-square for trend=0.42; p=0.52

Sources and treatment of water

Table 3 shows the sources of drinking water in the markets for the children, aged less than five years, of

Table 2. Mother's educational status

Educational status	Bodija (n=266)	Gbagi (n=259)
No education	57(21%)	27(10%)
Some primary	32(12%)	23(9%)
Completed primary	81(31%)	42(16%)
Some secondary	29(11%)	47(18%)
Completed secondary	62(23%)	100(38%)
Higher education	5(2%)	20(8%)

Chi-square=46.64; p<0.0001

the market women. Sixty (23%) women of the Bodija market used tap water, 91 (34%) brought water from their homes, and 45 (17%) bought water from market vendors. The corresponding figures for the women of the Gbagi market were 41 (16%), 98 (38%) and 19 (7%) respectively. Wells and tap water are mainly the sources of drinking water at home. Water from wells and tap was treated. One hundred and twenty-six women (47%) of the Bodija market treated drinking water at home by boiling, while 166 (64%) women of the Gbagi market treated drinking water by boiling. One hundred (38%) of the former group and 48 (19%) of the latter group did not treat their water. The proportion of the women who treated their drinking water was significantly higher in Gbagi compared to Bodija ($p<0.001$).

Sources of food

Sources of food given to the children aged less than five years in the market are shown in Table 4. Two hundred and sixteen (81%) of the 266 women of the Bodija market and 234 (90%) of the 260 women of the Gbagi market prepared breakfast at home, while dinner was prepared at home by 235 (88%) women of the Bodija market and 228 (88%) women of the Gbagi market. One hundred and forty-seven (55%) women of the Bodija market and 166 (64%) women of the Gbagi market bought lunch from food vendors. A significantly higher proportion of the women of the Gbagi market prepared breakfast at home compared to Bodija ($p<0.005$). The findings also indicate that a significantly higher proportion of the women of the Bodija market obtained their breakfast from food vendors ($p<0.005$). The proportion of the women of the Bodija market who obtained dinner from various sources was not significantly different from that of Gbagi ($p>0.05$).

Environmental hygiene practices

Refuse disposal. The answers to questions on refuse-disposal practices (Table 5) revealed that refuse was

Table 3. Drinking water sources for children of mothers

At the market

Source of water	Bodija (n=266)	Gbagi (n=260)
Tap water	60(23%)	41(16%)
Brought from home	91(34%)	98(38%)
Well	57(21%)	92(35%)
Bought at the market	45(17%)	19(7%)
Stream/pond	0(0%)	2(1%)
No response	13(5%)	8(3%)

Chi-square (first 4 rows)=22.60; p=0.00005

At home

Source of water	Bodija (n=266)	Gbagi (n=260)
Tap water	117(44%)	96(37%)
Well	136(51%)	148(57%)
Stream/pond	1(0.4%)	7(3%)
No response	12(5%)	9(3%)

Chi-square (first 2 rows) 2.42; p=0.121

Treatment of water	Bodija (n=266)	Gbagi (n=260)
Boiling	126(47%)	166(64%)
Alum	6(2%)	5(2%)
None	100(38%)	48(19%)
No response	34(13%)	41(16%)

Chi-square (first 3 rows)=23.49; p<0.0001

disposed of in personal dust bins by 69 (26%) women of the Bodija market and 84 (32%) in Gbagi; 40 (15%) in Bodija and 44 (17%) in Gbagi discarded refuse into the nearest streams, while 82 (31%) and 21 (8%) in Bodija and Gbagi respectively used the market refuse depots at the end of each day. Fifty-two (20%) Bodija women and 40 (15%) Gbagi women deposited their refuse on a refuse dump.

Child's defaecation practices

One hundred and forty (53%) women of the Bodija market and 63 (24%) in Gbagi indicated that their children were allowed to defaecate on the spot, and the faecal waste was later disposed of. One hundred and forty-six (56%) children in Gbagi and 79 (30%) in Bodija used a 'potty,' while direct defaecation into a pit latrine by the children was reported by 32 (12%) Bodija women and 22 (9%) Gbagi women. The proportion of the mothers reporting on their children's defaecation practices in both the markets was significantly different ($p<0.001$).

Table 4. Sources of food

Source of food	Breakfast		Lunch		Dinner	
	Bodija (n=266)	Gbagi (n=260)	Bodija (n=266)	Gbagi (n=260)	Bodija (n=266)	Gbagi (n=260)
Prepared at home	216 (81%)	234 (90%)	42 (16%)	69 (27%)	235 (88%)	228 (88%)
Food vendors	38 (14%)	17 (6%)	147 (55%)	166 (64%)	8 (3%)	6 (2%)
Food canteens	6 (2%)	5 (2%)	62 (23%)	16 (6%)	2 (1%)	2 (1%)
Other	0	0	2 (1%)	1 (0.4%)	1 (0.4%)	0
No response	6 (2%)	4 (2%)	13 (5%)	8 (3%)	20 (8%)	24 (9%)
Chi-square (p) (first 3 rows)	8.80 (p=0.012)		34.85 (p<0.0001)		1.69 (p=0.79)	

Disposal of child's faeces

One hundred and thirty-six (51%) women of the Bodija market and 83 (32%) of the Gbagi market disposed of their children's excreta by throwing them into a pit latrine, while 80 (30%) Bodija women and 60 (25%) Gbagi women disposed of it in the refuse dump near the market (Table 5).

Personal hygiene practices

Mothers' cleaning up and hand-washing practices: Table 6 shows the cleaning up and hand-washing practices of the mothers. Two hundred and four (77%) women in Bodija and 240 (92%) in Gbagi cleaned their children after defaecation with water using their hands, while others in varied proportions used paper and water, leaf and water, and toilet paper. After cleaning up, 72 (27%) women in Bodija and 138 (53%) women in Gbagi washed their hands with soap and water, while 182 (68%) in Bodija and 112 (43%) in Gbagi washed their hands with water only. The proportion of women in Bodija using the various cleaning and hand-washing methods was significantly different compared to that of Gbagi ($p<0.001$).

Occurrence of diarrhoea

Responses of the women with respect to the occurrence of diarrhoea in the last three months are shown in table 7. A chi-square analysis of the responses categorized into 'diarrhoea' and 'no diarrhoea' groups showed that there was no significant difference between the two markets ($p>0.05$).

Table 8 shows the results of chi-square analysis of various environmental and personal hygiene practices (exposure variables) and diarrhoea (outcome variable). Various categories of hygiene practices shown in table 3-6 were cross-tabulated against the occurrence of diarrhoea in nx2 tables. Chi-square and p values with the degree of freedom for each table are summarized in table 8. The only risk factor that was associated with the occurrence of diarrhoea among children aged less than five years in Gbagi was the refuse-disposal practice. In Bodija, source of water in the market, sources of breakfast and lunch, where child defaecates, mothers' hand-washing practices, and refuse-disposal practices

Table 5. Environmental hygiene practices at the market

Refuse disposal	Bodija (n=266)	Gbagi (n=260)
Personal dust bin	69(26%)	84(32%)
Throw it out of view	10(4%)	25(10%)
Nearest stream	40(15%)	44(17%)
Refuse depot	82(31%)	21(8%)
Refuse dump	52(20%)	40(15%)
Others	10(4%)	19(7%)
No response	3(1%)	27(10%)
Chi-square (first 6 rows)=42.82; p<0.0001		
Child's defaecation	**Bodija (n=266)**	**Gbagi (n=260)**
Potty	79(30%)	146(56%)
Latrine	32(12%)	22(9%)
On the spot	140(53%)	63(24%)
Nearby bush	6(2%)	11(4%)
Other	3(1%)	11(4%)
No response	6(2%)	7(3%)
Chi-sqaure (first 5 rows)=52.08; p<0.0001		
Disposal of child's faeces	**Bodija (n=266)**	**Gbagi (n=260)**
Throw in pit latrine	136(51%)	83(32%)
Throw refuse dump	80(30%)	66(25%)
Throw out of view	15(6%)	39(15%)
Other	25(9%)	47(18%)
No response	10(4%)	25(10%)
Chi-square (first 3 rows)=20.64; p=0.0004		

were found to be significantly associated with the occurrence of diarrhoea ($p<0.05$).

Occurrence of diarrhoea was highest among the children of mothers who bought water from the market and lowest among those who used tap water ($p<0.05$). Diarrhoea was more prevalent among the children of mothers who bought their breakfast and lunch from the market food vendors compared to those who prepared it at home ($p<0.05$). This trend was also observed for sources of dinner, but the difference was not statistically significant ($p>0.05$). Diarrhoea occurred more among the children who defaecated in open spaces, for example on the spot or in the bush, compared to those who defaecated in designated places ($p<0.05$). Diarrhoea was more prevalent among the children of mothers who washed their hands with water only than those who washed their hands with soap and water. Children of mothers with poor refuse-disposal practices recorded the incidence of diarrhoea more ($p<0.05$).

DISCUSSION

The findings of the study indicate that the environmental and personal hygiene practices of the women of the two markets were deficient, and such deficiencies were more pronounced in the women of the Bodija market compared to their counterparts in Gbagi. Surprisingly, no significant difference was found in the occurrence of diarrhoea among the children aged less than five years of the two markets. This finding must, however, be interpreted with caution as responses are subjective, and the frequency of the occurrence of diarrhoea may be under-reported by the less-educated Bodija women, especially when such responses were based on recall.

However, when the association between the various environmental and personal hygiene practices and the incidence of diarrhoea were considered, only refuse-disposal practices were found to be associated with diarrhoea in the Gbagi market with relatively better sanitation facilities. It is possible that the domestic hygiene practices may be the stronger determinants of the occurrence of diarrhoea in this group. Results of several studies have indicated the importance of domestic hygiene and household type as risk factors. Such factors include the type of kitchen (5), dirty feeding bottles (4), disposal of children's faeces (4,6) and of household garbage (6), unhygienic latrines (7), and child eating from floor (7). This study was not aimed at seeking information on the risk factors in the domestic environment.

Over one-third of the respondents of the Bodija market did not treat their drinking water. It is well known that poor water quality is associated with the occurrence of diarrhoea. The use of non-purified water (8) and poor storage of drinking water (9) have been reported to increase the risk of diarrhoea. However, the risk factor found to be associated with diarrhoea in this study was water bought from the market vendors. Similar association has been reported among children aged less than five years in south western Nigeria (10). Water sold by vendors which in the vendors' sales slogan is referred to as 'pure water' is usually packed in clear polythene bags and sold freely for human consumption.

Unfortunately, the source and the quality of such water are not known, and the situation has recently generated a great deal of concerns among the public health authorities. Regular consumption of water from this doubtful source may account for the association

Table 6. Personal hygiene practices

Cleaning child after defaecation	Bodija (n=266)	Gbagi (n=260)
Toilet paper	12(4%)	6(2%)
Scrap paper and water	24(9%)	7(3%)
Scrap paper alone	7(3%)	0(0%)
Leaf and water	2(1%)	2(1%)
Water and hand	204(77%)	240(92%)
Other	7(3%)	1(0.4%)
No response	10(4%)	4(2%)

Chi-square (first 5 rows)=18.93; p=0.0003

Mother's hand cleaning after child defaecation	Bodija (n=266)	Gbagi (n=260)
With water and soap	72(27%)	138(53%)
With water alone	182(68%)	112(43%)
Wipe hands with paper rag or leaf	2(1%)	3(1%)
None	1(0.4%)	5(2%)
No response	9(3%)	2(1%)

Chi-square (first 3 rows, rows 2 and 3 combined)=39.38; p<0.0001

Table 7. Occurrence of diarrhoea in children in 2 markets of Ibadan in the last three months

Response	Bodija (n=266)	Gbagi (n=260)
No diarrhoea	168(63%)	147(57%)
Diarrhoea	82(31%)	87(33%)
No response	16(6%)	26(10%)

Table 8. Risk factors for diarrhoea in children of market women

Risk factors	Bodija			Gbagi		
	Chi-square	P value	Degrees of freedom	Chi-square	P value	Degrees of freedom
Source of water at home	40.08	0.0000	2	0.38	0.94	4
Source of water in the market	20.22	0.00045	4	5.28	0.25	4
Source of breakfast	24.67	0.000004	2	0.48	0.92	3
Source of lunch	13.04	0.023	5	10.09	0.07	5
Source of dinner	7.10	0.068	3	1.37	0.71	3
Source of food on demand	9.42	0.051	4	1.77	0.62	3
Where child defaecates	21.92	0.0002	4	3.41	0.63	5
Disposal of excreta	3.79	0.28	3	3.90	0.28	3
Cleaning up practices	9.84	0.079	5	0.31	0.57	1
Mother's hand cleaning after child defaecation	3.89	0.048	1	0.02	0.98	2
Refuse disposal	23.85	0.00023	5	12.91	0.02	5

observed, and any intervention for the control of childhood diarrhoea must address the problem of water sold to the public by vendors.

We are surprised by our findings that the sources of food for breakfast and lunch were strongly associated with diarrhoea only in Bodija, although a large proportion of the women in each of the two markets obtained their lunch from the food vendors who are mostly women. Unfortunately, many people in the community patronize these women who are known to prepare and handle foods under the most unsanitary conditions. Food bought from food vendors has been shown to be associated with prolonged diarrhoea in the Nigerian children (11). Thus, food vendors play a significant role in transmitting enteric pathogens which cause diarrhoeal illnesses in children. It is pertinent to mention that food vendors often sell the leftover foods from the previous day, and this is more likely to be the case with breakfast and probably lunch. Having exhausted such unwholesome food items, dinner is likely to be prepared fresh, and this is less likely to be associated with diarrhoea. In Gbagi, the source of food from vendors was not associated with diarrhoea. This can be attributed to the fact that the more educated women in Gbagi would be more selective of food items eaten and food vendors used specially with regard to vendor's personal hygiene. In addition, most Gbagi women usually bring their serving bowls from home, whereas food is usually sold to the Bodija women in leaves, paper scraps, or polythene bags which may already be contaminated.

As expected, a strong association was established between the refuse-disposal practices in the Bodija market and the incidence of diarrhoea in children aged less than five years. Such poor refuse-disposal practices include throwing refuse into streams, refuse dumps and anywhere else out of view. Such practices tend to expose children to health hazards as they play around contaminated areas. In addition, indiscriminate refuse disposal can attract flies which serve as vectors of infective agents of diarrhoeal diseases.

We could not establish any association between the disposal of excreta and the occurrence of diarrhoea, but an association was established between where child defaecates (on the spot) and the occurrence of diarrhoea. Defaecating on the spot is partly culture-related, and most women, particularly the uneducated ones, have poor perception of the risk associated with indiscriminate waste disposal especially when this is done near the vicinity where the child plays or is fed. Mothers should be educated on safe child-defaecation practices. Since provision of basic water and sanitary facilities is crucial for effective control of diarrhoeal diseases among young children (12), such facilities should be extended to the Nigerian markets.

On personal hygiene practices after cleaning children following their defaecation, we established an association between the hand-washing practices and the occurrence of diarrhoea. The findings of our study agree with the findings of previous studies which showed that hand-washing especially with soap and water has beneficial effect in reducing diarrhoea morbidity (13).

This study has highlighted various risk factors in the occurrence of diarrhoea in the children aged less than five years in both the markets. The apparent lack of an association between the personal hygiene practices and the occurrence of diarrhoea suggests that any intervention should not only address the issue of the personal hygiene practices but must indeed take into consideration the environmental hygiene practices, such as refuse disposal, sale of water and foods by vendors, and child's defaecation practices. To this end, adequate provision must be made in the Nigerian markets for regular supply of piped water and for sanitary disposal of faeces. Emphasis on female education will no doubt equip future mothers to appreciate the health benefits of good personal and environmental hygiene. The present generation of mothers should be taught by health workers that food prepared at home under sanitary conditions is safer than food bought from food vendors. Lastly, the public health authority must urgently address the issue of sale of water and food by vendors.

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Knowledge and Practices of Preventing Diarrhoea in Malnourished Children

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ABSTRACT

Instructions for preventing diarrhoea, based on a knowledge-deficit model, are a common health-promotion approach aimed at the providers of child care attending nutritional rehabilitation centres. However, there is rarely an assessment of baseline knowledge to justify the need for this type of intervention and to guide its form. This study investigated the practice and knowledge of recommended diarrhoea-prevention behaviours of caregivers of 78 malnourished children consecutively admitted to a realimentation programme. Major deficits included: 39% not boiling (or not planning on boiling) drinking water after the child reached two years of age; 35% not always washing children's hands before meals; only 17% reporting that it was rare for their children to go barefoot; and the majority breastfeeding for less than one year. However, almost all measures of knowledge, based on open and closed questions, were not related to the corresponding practice. Several types of barriers to preventive practices were reported on open questions, including, "beliefs," "children as barriers," and "time." This information may be helpful in designing more effective health-promotion programmes.

Key words: Diarrhoea; Health education; Hygiene; Child nutrition disorders; Knowledge, attitudes, practice

INTRODUCTION

Malnutrition continues to be one of the most important health problem of children in developing countries (1-2). Causes of malnutrition in children include inadequate quantity and quality of food and the direct and indirect effects of diseases (1,3-4). Diarrhoea is the most commonly associated illness with malnutrition especially when it is severe, recurrent, and/or chronic (5-7). In addition, diarrhoeal episodes may be more prolonged in malnourished children, thereby increasing the morbidity and mortality rates further (8-11). For this reason in particular, addressing the prevention of diarrhoea in malnourished children is paramount.

One of the key approaches to addressing diarrhoea in malnourished children is to provide instructions relating to prevention of diarrhoea to caregivers attending realimentation centres (1,12). This would seem to be a reasonable intervention given that malnourished children often present to realimentation centres with diarrhoea. However, targeting this population with diarrhoea-prevention instructions makes two assumptions. The first assumption is that the presenting case of diarrhoea is a function of poor prevention practices in the home. The second assumption is that the poor practices in the home are a function of caregiver ignorance versus other factors, such as economic barriers. Although there is support for the former assumption (13-15), there is less so for the later.

Data were collected when the author was at the University of Pittsburgh

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The assumption of ignorance is implicit in health-education messages that rely heavily on providing factual information. Although complex models have been developed for prevention behaviour (e.g. Health Belief Model (16); Theory of Planned Behaviour (17)), health-promotion efforts in some developing countries rely primarily on knowledge-deficit models, as these interventions are facilitated by mass-produced generic information on diarrhoea prevention which allows for a relatively cheap intervention (18,19). However, this is problematic given the lack of evidence linking the knowledge of practice with the engagement in that practice, let alone a link between the change of knowledge and improved practice.

Although a few studies have found a relationship between general diarrhoea knowledge, diarrhoea-prevention practice, and the incidence of diarrhoea (14,20-21), there is a lack of assessment of specific knowledge about specific recommended prevention practices. In addition, there are some negative findings between knowledge and practice. A follow-up study of a comprehensive water supply, sanitation, and hygiene-education programme demonstrated little difference in knowledge about transmission of diarrhoea between the intervention and the control sites, despite the improved health practices and decreased diarrhoea rates in the intervention community (22). Our recent study of a periurban community sample in the Dominican Republic found no relationship between specific diarrhoea-prevention practices and knowledge of these specific practices (23).

Factors that may negate or weaken the relationship between knowledge and practice include various barriers, cultural factors, and beliefs. Gilman and Skillcorn (24) illustrated an economic barrier in their estimates of the cost of boiling drinking water in a fuel scare community. Various qualitative studies have argued the importance of considering cultural factors, such as local beliefs about disease causation and classification that may have important implications for the impact and design of health-promotion interventions (25-27).

Given that diarrhoea continues to be a common disease in malnourished children and that health education has been recommended as an appropriate intervention, we undertook a study of caregivers of malnourished children in a realimentation clinic in the Dominican Republic. We assessed the practice of recommended diarrhoea-prevention behaviours, the understanding of these recommended practices, and the perception of obstacles to performing these practices.

MATERIALS AND METHODS

This study was performed at a nutrition rehabilitation clinic in Los Alcarrizos, a large periurban community on the outskirts of Santo Domingo, the capital of the Dominican Republic. Caregivers attending the clinic with their children were invited to participate and gave verbal consent to be interviewed and to have their children weighed and measured for the study. A committee of Proyecto Niños, the realimentation clinic, approved the study protocol and the questionnaire.

An attempt was made to interview the caregivers of all children aged less than six years who entered the nutrition clinic between 11 August 1995 and 15 December 1995. In total, 100 children entered the nutrition clinic during the study period. Level one and below on the Gomez malnutrition classification system (<80% of the 50th percentile of National Center for Health Statistics [NCHS] weight-for-age) was used as an inclusion criterion for the study (2,28). Nine children who were admitted to the clinic had weights greater than this cut-off and were excluded from the analysis. Seven families had two children each from which one was selected randomly for the interview focus. Caregivers of 78 (93%) of the remaining 84 eligible children were interviewed; the other six had dropped out of the clinic before there was a chance to interview them. Nobody refused to be interviewed.

Caregivers were interviewed using a structured interview schedule designed by the author. Questions were asked about the diarrhoea-prevention practices, knowledge about these recommended practices, and the obstacles impeding their implementation. Questions also included health and socioeconomic issues. Words the mother did not understand were explained to her. Approximately, half of the questions were open ended and the other half closed ended. For open questions, assessing knowledge of a practice (e.g. “What are some of the causes of diarrhoea in children?”), caregivers were encouraged to give more than one answer if they could. Closed questions were composed of yes/no type questions, except for two scaled response questions (e.g. “Do your children go barefoot?: almost always/frequently/sometimes/almost never”). Interviews were conducted in Spanish by three Dominican community health workers who recorded all responses. Each interview took approximately 20 to 30 minutes. The principal investigator monitored the administration of over ten percent of the interviews and reviewed the data from all the interviews with the interviewers.

The children's weights were measured by the paediatrician to the nearest 1/4 lb using either a standing or baby scale (Detecto Scale Company), depending on the patient's ability to stand still. The chi-square statistic with continuity correction and the Fisher's exact test were used for categorical data analysis.

RESULTS

Demography

The age of the 78 children (47% males) ranged from two weeks to 5.86 years with a mean of 1.4 years. The average birth weight, 2.9 kg, is approximately 90% of the 50th percentile of the NCHS standards (28). Eighty-three percent of the caregivers reported that their children had had diarrhoea within the two weeks prior to presentation at the clinic. Forty-seven percent reported vomiting during this same time period.

Biological mothers constituted 93.6% of those interviewed with grandmothers, a father and an aunt constituting the rest. The age of the mothers ranged from 14 years to 40 years, with an average age of 23.6 years. Formal schooling of the mothers averaged just over five years with 15% reporting no formal education at all. The caregivers had, on an average, 2.8 live-births. Most homes were crowded with a mean of 3.8 persons per sleeping room. Forty-two percent reported that they did not have their own water tap, and 26% reported that they did not have their own latrine.

Purification of water

Eighty-three percent of the caregivers reported that they presently purified drinking water for the index child, primarily through boiling (87%). We combined the variables "at what age did you stop boiling water" (for those who had stopped boiling) and "at what age do you plan on stopping boiling water" (for those who were still boiling water at the time of the interview) to create a new variable. Thirty-nine percent either stopped or planned to stop boiling water at or before the time the children reached two years of age.

Twenty-five percent of the caregivers reported "providing good drinking water" to the open question "how can you prevent diarrhoea." Seventy-three percent of the respondents stated that boiling water kills microorganisms or parasites. There was no difference in response on these items by whether the caregiver was presently providing purified drinking water for the child (Table).

Of particular interest is the group that demonstrated a knowledge of the relationship between water and diarrhoea, but still did not provide purified drinking water to the index child. Within this group, the most common reason for not boiling water was that the child was drinking "agua cruda" (non-purified water) in other locations. Some caregivers reported that it is dangerous for the children to switch back from non-purified water to purified water. Others reported that they did not have sufficient time, that someone else cared for the child when the mother was working, that they did not have a stove, or that the child was big enough to drink "agua cruda."

Only 42% of the respondents reported that they used chlorine drops in the drinking water. However, 74% reported that chlorinating water was a good idea. There was a significant relationship between practice and knowledge (Table). Not liking the taste was the most common reason for not using chlorine amongst those who reported that it was a good idea.

Hygiene

Sixty-five percent of the caregivers reported that they "always" wash the children's hands before meals. Thirty-one percent reported that they "sometimes" or "usually" do this, while four percent reported "almost never" engaging in this practice. However, 90% of the total group reported that bacterial or parasitic contamination was the reason for washing hands before eating. There was no relationship between the practice and this measure of knowledge (Table). The mother being away during meals or being too busy were the most commonly reported obstacles to regular hand-washing.

Only 17% of the caregivers reported that it was "rare" for their children to go barefoot. Fourteen percent reported that their children "almost always" go barefoot, and 70% reported that "usually" or "sometimes" their children go barefoot. Ninety-four percent of the caregivers felt the importance of wearing shoes all the time. There was no relationship between agreeing with this recommendation and the practice (Table). There was also no difference between the practice and the frequency of reporting that the wearing of shoes can protect one from getting parasitic or microorganism infection. The main obstacles to wearing shoes by their children all the time were that the family could not always afford to get shoes for their children or that the children would remove the shoes themselves.

Breast-feeding

Almost all the mothers (92.2%) initially breastfed their children; however, only 18% were breast-feeding at the

time of the interview. The mean age of stopping breast feeds for the remaining 82% was 6.1 months. Despite this early age of terminating breast-feeding, 88% of the total group reported knowing that the recommendation is to try to breastfeed for at least one year. Only 27% of those who reported knowing the recommendation breastfed for one year or more. There was no relationship between those reporting knowing the recommendation and the actual practice (Table).

The most common reasons for not breastfeeding for one year for those who knew the recommendation are: the child no longer wanted breast milk; the milk had dried up or was no longer sufficient for the child; the mother was pregnant, or the mother was ill. Of the six mothers who never breastfed, the most common reason was that the mothers did not produce any milk.

All but three caregivers had given their children non-breast milk. The average age of starting milk was approximately three months with a range from 0 to 24 months. The most common reason for starting non-breast milk was that the child was perceived not to be full on breast milk alone.

Table. Diarrhoea-prevention practice variables versus corresponding knowledge variables

	Prevention practice endorsed				
	Yes		No		
Knowledge variable	**%**	**No.**	**%**	**No.**	**p value***
Purifies drinking water					
Good water can prevent diarrhoea†					
Reported	78.9	(15)	21.1	(4)	0.25
Not reported	80.4	(45)	19.6	(11)	
Boiling water can kill germs					
Reported	83.6	(46)	16.4	(9)	0.24
Not reported	80.0	(16)	20.0	(4)	
Chlorinates drinking water					
Chlorinating water is a good idea					
Yes	55.2	(32)	44.8	(26)	<0.001
No	5.0	(1)	95.0	(19)	
Child's hand always washed before meals					
Hands may have microorganism contaminations‡					
Reported	63.6	(28)	36.4	(16)	0.32
Not reported	80.0	(4)	20.0	(1)	
Child always wears shoes					
Children should always were shoes¶					
Agree	17.7	(11)	82.3	(51)	0.47
Disagree	0.0	(0)	100.0	(4)	
Shoes protect from infection¶					
Reported	10.4	(5)	89.6	(43)	<0.10
Not reported	33.3	(6)	66.7	(12)	
Breastfed for one year					
Knew recommendation to breastfeed for minimum one year§					
Yes	27.4	(17)	72.6	(45)	0.26
No	37.5	(3)	62.5	(5)	

*p value based on Fisher's exact test, except for chlorinating water and shoes protect from infection for which the chi-square test was used

†n=only 75, as one did not yet drink water, and two cases had missing data for this question

‡n=only 49, as the practice question was missed on initial interviews due to omission on initial interview forms

¶n=only 66, as 11 children were not yet walking, and one case had missing data for this question

§n=only 70, as six children who were aged less than one year and still being breastfed were excluded, and two cases had missing data for this question

DISCUSSION

This study provides important information about prevention of diarrhoea by caregivers attending a realimentation centre. There was minimal selection bias within the clinic given that we were able to recruit the majority of consecutive admissions to the clinic, hence the results may be generalizable to attendees at this clinic. Although this information may not be generalizable to other realimentation clinics or to the surrounding community, it highlights the need to consider performing a local needs assessment to inform a health-promotion programme.

This study has, however, some limitations. The lack of observational data may have resulted in the inflation of some of the practices, such as hand-washing, given the potential for a socially desirable response bias. However, a large number of the caregivers were still willing to report suboptimal practices. The other major limitation was the high rate of the participants scoring positive on some of the knowledge questions resulting in some small cells for analyses. More complicated questions may have resulted in a greater distribution on knowledge scores. However, the basic questions in our study more closely reflected information in generic health-education programmes than would questions reflecting a more sophisticated understanding of biomedical processes.

The first assumption of generic health-promotion programmes appears to be appropriate, i.e. there are significant practice deficits that increase the likelihood of these children contracting diarrhoea as has been found in many other studies (13-15). (Note: Although going barefoot is not a major risk factor for diarrhoea, it increases the likelihood of contracting hookworm and is a frequent health-promotion target within hygiene lectures). The second assumption that the practice deficits are a function of knowledge deficits does not seem to be supported by this study. The exception to this was for chlorination of water. However, in this one case our "knowledge question" was somewhat different than the others and reflects more opinion than knowledge.

Given that the knowledge levels are high for some of the practices, we cannot say that knowledge is not important. Conceivably, the high rates of knowledge about bacterial and parasitic contamination may have come through previous exposure to health-education messages. Perhaps those engaging in good practices are doing so, at least in part, due to their knowledge of the importance of these practices. However, there are clearly other factors impacting on practice given that many knowledgeable people were not engaging in prevention practices.

Our open questions on barriers provide some possibilities for alternative factors. With regard to providing purified drinking water, one relatively common belief appeared to be that once the child drinks "agua cruda" (non-purified water), they should not go back to purified water, either because there is no subsequent benefit for the child or that this practice is actually dangerous. This may be a reflection of an "all or nothing" belief rather than an appreciation of dose-effect.

Children were also considered a barrier source. Several caregivers reported that children took their shoes off themselves and hence undermined their parents' efforts to keep their feet protected. In addition, "children no longer wanting breast milk" was a frequently reported reason for stopping breast-feeding.

Finally, limited resources were an additional barrier source. Some reported that their children went barefoot as they could not always afford shoes. Although infrequently raised in this study, the cost of boiling water may be a major factor as described by others (24). Another resource limitation is time, with some caregivers indicating that they were too busy to wash the children's hands before meals.

In summary, there is a need to address diarrhoea prevention at this nutrition rehabilitation clinic given several practice deficits. However, there is a need to consider factors beyond knowledge deficits in designing an intervention. A further study of the above barriers and other potential barriers to practice may help inform an effective intervention.

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SHORT REPORT

Detection of *Shigellae* from Stools of Dysentery Patients by Culture and Polymerase Chain Reaction Techniques

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Abstract

In Bangladesh, the isolation rates of *Shigella* spp. range from 11% to 12% by the conventional culture technique. Since the sensitivity of this technique is low, the polymerase chain reaction (PCR) technique was used for detecting small number of *Shigellae* from patients' stools. Sensitivity and specificity of the two techniques were also compared. Stool samples were collected from 41 patients with dysentery who attended the Clinical Research and Service Centre of the ICDDR,B: Centre for Health and Population Research. All stool specimens were directly plated onto MacConkey, Salmonella-Shigella, Xylose lysin deoxycholate and Hectoen enteric agar media, and *Shigellae* were detected following standard procedures. DNA was extracted from the stool samples, and the target sequence of invasive plasmid antigen *(ipa)*H locus was amplified by PCR with 130 ng each of two primers (primer H8 [5'-GTTCCTTGACCGCCTTTCCGATAC-3'] and primer H15 [5'-GCCGGTCAGCCACCCTA-3']) following standard procedures. The amplified product was hybridized using an *ipa*H probe. The isolation rates of *Shigella dysenteriae* type 1, *S. flexneri, S. sonnei,* and *S. boydii* were, respectively, 17.1%, 19.5%, 4.9% and 2.4% by the conventional method. The results of the PCR technique showed that 700 bp fragment was generated in 18 of the 18 culture-positive and in 7 of the 23 culture-negative stools. One hundred twenty-three strains of *Escherichia coli* were also tested by PCR for identifying the enteroinvasive *E. coli,* but none of them yielded any positive result. This study showed that the sensitivity of the culture technique is 72% and specificity is 100%, when the PCR technique was considered as gold standard. Therefore, the PCR may be considered a more sensitive and specific technique than the conventional culture technique and has the potential to be employed in routine diagnosis of dysentery in clinical centres as well as in epidemiologic studies.

Key words: *Shigella*; Dysentery, Bacillary; Culture media; Polymerase chain reaction; Comparative studies

INTRODUCTION

Shigella species are the recognized pathogens of bacillary dysentery and have an isolation rate of approximately 11-12% in diarrhoea patients in Bangladesh, using the conventional culture technique (4). But the performance of the culture technique is limited by its low sensitivity. As a result, shigellosis remains undiagnosed in a significant number of patients (1). Besides, during the convalescent period, *Shigellae* may remain undetected in stools when examined by the conventional culture

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technique due to the presence of fewer organisms as well as non-culturable forms of the organism (5).

The polymerase chain reaction (PCR) technique, on the other, seems ideally suited to detect low number of *Shigellae,* since this method enables the amplification of DNA obtained from as few as a single cell present in the sample (10). This method depends only on the presence of target DNA and allows a specific segment of DNA to be amplified by a factor of 10^6 or more within hours (8). The PCR technique was used in two studies for detecting *Shigellae* from stools (1,2). An invasive-associated locus (*ial*) probe was used in these studies. The *ial* sequence is present in plasmids, as the plasmid is unstable, it can be easily lost (6). To overcome this problem, the invasive plasmid antigen H (*ipa*H) locus probe can be used, since this gene is present in more than one copy on the invasive plasmid and chromosome (3). Although the PCR technique is superior, adequate laboratory trials are required for its optimal application. We, therefore, used both culture and PCR techniques with *ipa*H probe to assess their performance in detecting *Shigellae* from stools of dysentery patients.

MATERIALS AND METHODS

Stool samples, collected from 41 patients with dysentery who attended the Clinical Research and Service Centre of the ICDDR,B: Centre for Health and Population Research, were directly plated onto MacConkey, Salmonella-Shigella, Xylose lysin deoxycholate and Hektoen enteric agar plates (Difco, Detroit, Michigan, USA) to detect *Shigellae* following procedures described by Islam *et al*. (5). In brief, the inoculated plates were incubated at 37 ^{o}C for 18-24 hours. Suspected *Shigella*-like colonies from the plates were subsequently inoculated onto Kligler iron agar, motility indole urea and Simmons citrate agar media. Non-motile organisms showing acid butt and alkaline slant and lack of gas and H_2S production in Kligler iron agar, and negative reaction for urea and citrate utilization were tentatively identified as *Shigellae*. They were then confirmed by slide agglutination with specific *Shigella* antiserum (Wellcome Diagnostics, Dartford, England). Since the *ipa*H genes are also present in enteroinvasive *Escherichia coli,* 123 *E. coli* strains (3 strains each from 41 stool samples) from MacConkey plates were tested by the PCR technique.

One hundred µL of stool was mixed in 0.5 mL of phosphate-buffered saline in a 1.5-mL microfuge tube. Suspending particles were removed by centrifugation at 503 g for 3 minutes. The supernatant was again centrifuged at 11,000 rpm for 3 minutes. DNA was extracted from the pellet following procedures described by Echeverria *et al*. (1), and the target sequence of *ipa*H locus was amplified by PCR with 130 ng each of two primers (primer H8 [5'-GTTCCTTGACCGCCTTTCCGATAC-3'] and primer H15 [5'-GCCGGTCAGCCACCCTA-3']) (3) and 1.25 U of Taq polymerase (Perkin-Elmer Cetus, Norwalk, Conn.) per 25 µL of reactions mixture on a Thermal Cycler (Perkin-Elmer Cetus). The PCR reaction was done for 35 cycles of 1 minute each at 94 ^{o}C (for denaturation), 1.5 minute each at 60 ^{o}C (for annealing of primers to single-stranded DNA), and 0.25 to 1 minute each at 72 ^{o}C (for DNA polymerase-mediated extension). The amplified products were then separated by agarose (0.8%) gel electrophoresis, transferred to a nylon membrane by southern blotting and subjected to hybridization with the *ipa*H probe (5). The probe is basically the fragment amplified with primers H8 and H15. Labelling and detection of the hybridized probe were performed with an enhanced chemiluminescence gene detection system following the instructions of the manufacturer (Amersham International, UK). The validity of the primers was previously tested, the results of which indicated that these primers were highly sensitive and specific for detection of *Shigellae* (5,9).

RESULTS

Shigellae were isolated from 18 (43.9%) of the 41 dysentery patients using the culture technique, and the isolation rates of *S. dysenteriae* type 1, *S. flexneri*, *S. sonnei*, and *S. boydii* were 17.1%, 19.5%, 4.9% and 2.4% respectively.

The extracted DNA of the faecal samples was specifically subjected to amplification for *ipa*H sequence using *Shigella*-specific (*ipa*H) primers in Thermal Cycler; it was found that after 35 cycles of amplification, 700 bp fragment was generated in 18 of the 18 culture-positive and in 7 of the 23 culture-negative stool samples (Fig.1A). This amplified fragment was confirmed by hybridization with *ipa*H probe (Fig. 1B). Therefore, *Shigellae* were detected in 25 (61.0%) of the 41 stool samples, using the PCR technique. But none of the 123 *E. coli* strains yielded any positive result with PCR.

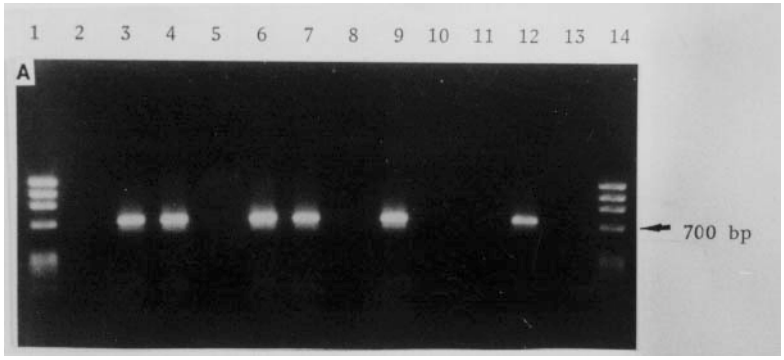

Fig 1A: Specific amplification of invasion plasmid antigen H (*ipa*H) locus DNA by PCR. Amplified products of each reaction were analyzed by electrophoresis through a 0.8% agarose gel. Lanes: 1 and 14, *Hae* III digest of øX 174 replicative form DNA; 2 and 13, negative controls without template DNA; 3, *S. dysenteriae* 1 used as the positive control; 4, 6, 7 and 9, *S. flexneri, S. sonnei, S. boydii,* and *S. dysenteriae* isolated from stools respectively; 5, 8, 10 and 11, PCR negative and no *Shigella* species isolated; 12, PCR positive, but no *Shigella* species isolated.

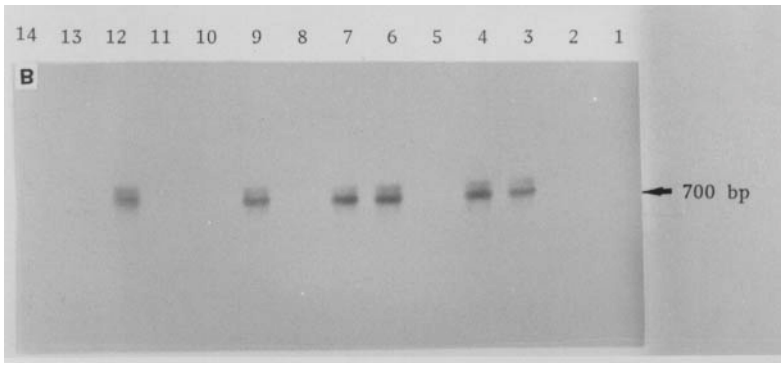

Fig 1B: Hybridization of southern blot shown in panel A with *ipa*H DNA probe as detected by the enhanced chemiluminescence technique. Each signal corresponds to the amplified PCR product.

DISCUSSION

The PCR technique can detect a small number of culturable as well as non-culturable organisms. Such detection is specially important for *Shigellae*, since they can produce disease by as few as 10-100 organisms (7). In our study, the use of PCR technique improved the rate of detecting *Shigellae* in stool samples from 43.9% to 61.0%; which is 17% higher than the conventional culture method. Moreover, the time requirement of this technique is lower compared to that of the culture technique. The detection scheme, including the DNA extraction, DNA amplification and analysis of amplified products, requires only 5-6 hours compared to 18-24 hours of standard stool culture. The specificity of the culture technique is 100%, and the sensitivity is 72%, when the PCR technique is considered as gold standard. Thus, PCR may be judged as superior for its rapidity and sensitivity in the detection of *Shigellae*. Moreover, since this method is applied without employing cultivation of organisms on the synthetic medium, non-culturable population of *Shigellae* can also by detected by this method.

In view of the above, PCR may, therefore, be considered a more sensitive and specific technique than the conventional culture technique, and has the potential in routine diagnosis of dysentery in clinical centres as well as in epidemiologic studies. The PCR technique can be chosen as an alternative to the culture technique, and can further be used for identifying asymptomatic carriers, serving as potential reservoirs of *Shigellae* silently transmitting the disease within communities. Based on the findings of our study, it can be concluded that the PCR technique is more useful than the conventional culture technique in precisely determining the actual prevalence rate of *Shigellae* among dysenteric patients.

ACKNOWLEDGEMENTS

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LETTER TO THE EDITOR

Use of Soursop and Sweetsop Juice in the Management of Diarrhoea in Children

Sir,

Acute diarrhoea is an important cause of morbidity and mortality in children. Although oral rehydration therapy (ORT) is useful for treating diarrhoea, it does not actually reduce the duration of diarrhoea, and people turn to alternative methods of management. This often results in the use of antidiarrhoeal drugs (1). Cereal-based oral rehydration solution (ORS) has been a step forward and is effective in reducing the duration of diarrhoea and in reducing stool output by up to 35% in cases of cholera. However, in diarrhoea due to other causes, the effect is less marked and often is no better than WHO-ORS alone. A well-designed study in Chile looked at 100 children randomly offered rice-based ORS or WHO-ORS and found no difference in outcome (3.5 days in the rice group, 3.9 days in the glucose group). Other approaches to reduction of acute diarrhoea include micronutrient supplementation. Folate supplement in 38 of the 76 South African children resulted in a 43% decrease in time to resolution of diarrhoea. In another study, zinc supplementation in malnourished children resulted in significant reduction in intestinal fluid loss, as well as better height gain at three months.

Soursop *(Annona muricata)* and sweetsop *(A. squamosa)* juice have already been tested for the presence of the elements which are lost from the body during the episode of diarrhoea (in press). The micronutrients that were found to be available in the juice include carbohydrate, glucose, protein, and lipid. (2,3) The elemental composition of the juice includes, among others, sodium, calcium, potassium, phosphorus, chloride, iron, zinc, carbonate, and bicarbonate (3, author's unpublished data). The ORT also contains some of these elements, viz. sodium chloride, sodium bicarbonate, trisodium or citrate dihydrate, potassium chloride, and glucose anhydrous. (4)

The hallmark of diarrhoea, which could be caused by bacteria, fungi, protozoa, and viruses, is the loss of fluids and electrolytes which may eventually lead to dehydration, acidosis, haemoconcentration, and hypovolaemia. (5) This study, therefore, reports on the management of acute watery diarrhoea with fruit juice which is safe for consumption.

The healthy soursop fruit, purchased from the Ekpoma main market, was washed in salt solution. The skin was peeled off and the pulp (mesocarp) squeezed out aseptically to enable the juice to be collected. The juice was sterilized by boiling at a temperature of 100 °C for 30 minutes, and was stored in frozen form in sterilized plastic containers. Alternatively, the juice can be pasteurized at a temperature of 70 °C for 50 minutes.

The procedures followed for the clinical trail were in accordance with the ethical standards of the committee on human experimentation of the institution in which the trial was done.

An ethical certificate was obtained for clinical trial on children and adults, suffering from acute diarrhoea, who visited the Edo State University Health Center, Ekpoma. The services of a physician and a nurse in the health centre were sought for to monitor the progress of the diarrhoea patients.

The juice was administered onto 25 children aged 6-20 months and 5 adults with acute watery diarrhoea. Children with prolonged diarrhoea, malnutrition, or other major systemic illnesses were excluded. Five mL of the juice was administered on to the patients three times daily for up to 72 hours.

The fruit juice was able to resolve the diarrhoeic condition at 48 hours. This conforms with the report that at 48 hours 42% of the diarrhoeic patients treated with smectite were free from diarrhoea. Zinc available in the juice had been reported to bring about significant reduction in intestinal fluid loss. This suggests that soursop juice can be used as a complementary source for ORT in the management of diarrhoea in children and adults. This could reduce morbidity and mortality rate in children, specially in the developing nations. It appears from the previous work of the authors that sweetsop juice has similar elemental composition as the soursop. Therefore, the juice

from the sweetsop can also be used in the management of diarrhoea in human.

There are some advantages in using these fruit juice as a complement for ORT. The moisture content is above 70%, and this can aid rehydration during fluid loss. They contain a greater number of elements than what is stated in ORS (WHO/UNICEF) composition. These elements can replenish the electrolytes lost during the episode of diarrhoea. The taste is better and is well accepted by children. The fruits are common, cheap and readily available in Nigeria. Therefore, the populace can easily accept its use. No additive is needed for its preservation. The juice can readily be squeezed out from the fruit for use without preservation of the juice specially for rural dwellers who might not afford the facilities for preservation. Frozen juice have been reported to maintain the same quality and can be kept for 6 months (personal communication). Boiling and/or pasteurization does not affect the quality of the juice. The micronutrients, e.g. protein, in the juice may help correct malnourishment sometimes associated with diarrhoea.

It is the authors' opinion that people should be educated to subscribe to this complementary method in the management of acute diarrhoea in children and adults.

However, the authors are constrained to make a definite statement on the association between the aetiological agents of diarrhoea and the efficacy of the juice in the resolution of acute diarrhoea. This could be the subject of a further study. Further studies are also required to check the effectiveness of this preparation in children with malnutrition and in persistent diarrhoea.

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LETTER TO THE EDITOR

Cyclospora cayetanensis-associated Diarrhoea in a Patient with Acute Myeloid Leukaemia

Sir,

We report here a case of cyclosporiasis associated with acute myeloblastic leukaemia (AML). A 35-year-old man presented to our hospital on 19 September 1997 with a 15-day history of intermittent fever with chills and rigor. Physical examination revealed pallor and hepatosplenomegaly, while laboratory examination revealed a total peripheral leukocyte count of 20,000/µL with 20% blast cells. Haemoglobin was 8 g/100 mL, and the number of platelets was 100,000/µL. On two separate occasions, the patient's serum was non-reactive for HIV antibodies. Examination of bone marrow, including cytogenetic study, confirmed the diagnosis of AML-M2.

Cultures of blood, throat swab, urine, and stool were inconclusive. Since the patient had fever at presentation, he was started on gentamicin and cefotaxime which resolved the fever. Two courses of anti-cancer chemotherapy (mitoxantrone and cytosine arabinoside) were started on 29 September 1997 and 8 November 1997 respectively.

On the eighteenth day after the last day of chemotherapy, when blood cell counts had returned to normal, and bone marrow was in remission, the patient had 4 loose stools, which were copious and were associated with mucus but no blood. A stool sample was examined for parasites and bacterial pathogens. He was empirically started on metronidazole 400 mg thrice a day for 5 days and ciprofloxacin 500 mg twice a day for 5 days. The gastrointestinal symptoms disappeared within 24 hours. A wet, unstained preparation of the stool revealed non-refractile-round spheres of 8-10 µm in size with granular contents. An aliquot of the stool was stained by the modified acid-fast technique (1). This revealed acid-fast oocysts of 8-10 µm in size. Their staining pattern ranged from unstained to faint pink; the majority being unstained. The acid-fast staining was repeated by the original Ziehl-Neelsen method which involves heating, resulting in uniform dark red staining of the oocysts. Bacterial culture failed to isolate any pathogens. A diagnosis of cyclosporiasis was, thus, established. The patient remained free of gastrointestinal symptoms during his hospital stay, and a repeat stool examination after a week of starting antibiotic was negative.

Cyclospora is a newly recognized cause of diarrhoea (2). The duration of the diarrhoea is known to vary from 2 days to weeks, but resolves without treatment, even in immunocompromised patients (3). Although trimethoprim-sulphamethoxazole is the drug of choice for cyclospora infection, metronidazole has also been used with success (4). Our patient either responded to metronidazole or his diarrhoea was self-limiting. The duration of diarrhoea in this instance lasted for only one day. This may be due to either prompt institution of treatment or improved immune status 18 days after the end of chemotherapy. It has indeed been shown that the severity of cyclosporiasis may be related to the degree of immunosuppression (5).

Cyclospora is widely distributed throughout the world, and has been identified in both residents and travellers from various regions, including North America, Central America, and South America; the Carribean islands, Eastern Europe, India, South Africa, and South-East Asia (3,4). Outbreaks of cyclosporiasis have been reported in Nepal (6). *Cyclospora* has been demonstrated as a cause of diarrhoea in travellers returning from Nepal and Pakistan (7). In addition, it has also been identified from patients with protracted diarrhoea in various places, including Bangladesh (8). Persons of all ages have been infected. Cyclosporiasis may not be uncommon in India, although published reports are rare. Under-diagnosis of cyclospora infection may be due to lack of knowledge on the part of clinicians, failure of clinical laboratories to screen for *Cyclospora*, intermittent shedding of *Cyclospora* oocysts in stools and the acid-fast staining being an insensitive technique for the detection of *Cyclospora*. Recently, microwave heating has been suggested as a modification of the acid-fast staining to overcome the poor staining of the oocysts (9). We obtained good results with simple heating

of the carbol fuchsin as per the original procedure for staining of acid fast-bacilli. Hence, if confirmed, it might become a recommended procedure in laboratories without a microwave oven.

To the best of our knowledge, this is the first case report of cyclospora infection in a cancer patient in our country. The original acid-fast staining technique with heating may easily detect *Cyclospora* even in laboratories with limited facilities. All laboratories should, therefore, routinely screen stool samples of patients to determine the prevalence of cyclospora infection in our country.

ACKNOWLEDGEMENT

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LETTER TO THE EDITOR

Evidence of Involvement of Guanylate Cyclase in the Prolongation of Caeco-colonic Transit Time by Indomethacin in Mice

Sir,

Sodium nitroprusside relaxes the smooth muscle by generating nitric oxide which activates guanylate cyclase, resulting in the formation of cGMP, the second messenger (1). Methylene blue, which inhibits the soluble guanylate cyclase, has been shown to interfere with the action of sodium nitroprusside in some tissues (2, 3). To investigate whether other relaxants of intestinal smooth muscle also act through guanylate cyclase, we measured caeco-colonic transit time after administration of the relaxants using methylene blue. The observations are summarized here.

Male Swiss albino mice (n=50) weighing 25 to 30 g were fitted with intra-caecal cannulae, and the caeco-colonic transit time was measured as described earlier (4). Briefly, the abdomen was opened under nembutal anaesthesia, and a polyethylene tube was inserted into the caecum and secured. The other end of the tube was passed subcutaneously and brought out through the skin at the nape of the neck. After a week of recovery, the mice were used for experimentation. Olive oil 0.2 mL was introduced into the caecum of each fasted mouse through the cannula, after adjusting the dead space. The mouse was immediately placed in a cage lined with a filter paper. The time elapsed between the introduction of oil and the passage of oily stools was taken as the caeco-colonic transit time. The results of a previous work showed that this interval was about 7.5±0.21 minutes (n=52) and that it was prolonged in a dose-dependent manner by the relaxants (4). No attempt was made to look at the tissue damage in the stomach or colon. The animals were not sacrificed at the end of the experiment. They were apparently normal and did not show any ill effects.

Table. Effect of methylene blue on the prolongation of caeco-colonic transit time induced by relaxants

Drug (route)	Control	Test
Saline (i.v.)	7±1	6±1
Sodium nitroprusside (i.v.)	33±1	8±3*
Atropine sulphate (i.p.)	67±4	73±8
Indomethacin (oral)	79±9	18±2*
Loperamide (intra-caecal)	172±27	134±17
Theophylline (i.v.)	28±2	29±3

All values are expressed as mean±SEM of 3-8 experiments in each group. All drugs were given 40 minutes prior to the administration of olive oil. Saline or methylene blue was administered intracaecally 10 minutes after the drug treatment. Drugs were used at the following dosages: atropinesulphate 7 mg/kg, loperamide 2 mg/kg, indomethacin 44 mg/kg, methylene blue 170 mg/kg, theophylline 20 mg/kg, sodium nitroprusside 2.6 mg/kg. Volumes of administrations were 0.3 mL by oral, 0.2 mL by intra-caecal and 0.1 mL by the intravenous routes. Values marked * are significantly different ($p<0.01$) from the controls.

The table shows that administration of agents, known to relax the intestinal smooth muscle, prolonged the caeco-colonic transit of olive oil significantly. Prior administration of methylene blue reduces such prolongation only significantly in case of sodium nitroprusside and of indomethacin.

The ability of methylene blue to inhibit the effect of sodium nitroprusside on caeco-colonic transit is in accordance with the reports on other tissues (2,3). The surprising finding was observed to be the ability of the dye in reducing the effect of indomethacin on the transit time. It is generally believed that indomethacin blocks the synthesis of prostaglandins. Interactions between prostanoids and nitric oxide have been reported. Schirgi-Degen and Beubler report that indomethacin significantly reduces L-NAME-induced fluid secretion in the rat jejunum, probably by generating nitric oxide (5). In canine saphenous vein, endogenous prostanoids inhibit the nitric oxide synthesis. Administration of indomethacin by inhibiting the prostanoid synthesis facilitates expression of the enzyme activity. Nitric oxide thus generated augments the relaxation of sphenous vein (6).

Similar events occurring in the colon may explain the results we obtained with indomethacin. At least, the effect of indomethacin in prolonging the transit time may partly be due to generation of nitric oxide which may lead to formation of cGMP involved in relaxation. Blockade of guanylate cyclase by methylene blue may thereby lead to the inhibition of relaxant action of indomethacin.

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"A total of 500 *Shigella* strains were included in the present study out of which 233 were isolated at National Shigella Centre of Postgraduate Department of Microbiology, K.G. Medical College, Lucknow during last 5 years and 267 strains were received from different parts of the country. Predominent serotypes were *Shigella flexneri* 292 (48.4%) followed by *Shigella dysenteriae* 153 (30.6%), *Shigella boydii* 32 (6.4%) and *Shigella sonnei* 23 (4.6%). All the strains were sensitive to Nalidix acid, Gentamycin, Kanamycin, Neomycin and Furazolidine. Varying degree of resistance was shown to Streptomycin, Chloramphenicol, Tetracycline and Ampicillin. R-plasmid extraction was done and transfer was studied on *E. coli* K-12."

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"Although Bangladesh has achieved remarkable success in extending the availability of hand pumped and piped water, unsafe water is still ingested. This brief study attempted to assess water management practices in rural and urban homes in Bangladesh so as to establish the routes by which unsafe water is ingested, to examine methods of collection and storage, and determine why unsafe water sources are used when safe supplies are available. Forty-eight rural and forty-five urban slum households were studied. Observations, interviews and water quality investigations were conducted. The results show that the respondents were aware that hand pump/tap water is safe and took care to use these safe sources for drinking purposes. However, they continued to use surface water for non-drinking activities such as bathing, washing and rinsing their mouths. Reasons were given that it was a traditional practice to bathe in surface water and was more enjoyable. One of the reasons given for not using hand pumped water to wash clothing and food was that such groundwater caused staining. Bacteriological results from such ingested water showed the quality, especially in rural areas, to be poor. Results also showed the internal surfaces of the base of storage containers to be heavily contaminated with bacteria. This showed that water that was safe when it was first drawn would became contaminated during storage. This study had a limited scope; much further research is needed to find what determines and how water becomes contaminated in containers. These factors include how to reduce contamination of water, in particular the relationship between growth of bacteria in stored water and the material from which the container is made and how to improve the overall quality of water. On the management side, studies could be conducted as to how to improve people's understanding of the use of water for domestic purposes and its safe management."

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"In May 1996, an outbreak of gastroenteritis occurred among customers who bought dinner from a restaurant that specialised in fried chicken in Abha city, south-west Saudi

Arabia. The median incubation period was 10 hours (range: 3 to 27 hours). Of the 10 food items served, only mayonnaise (RR 2.52; 95% CI 1.71-3.73) and minced garlic (RR 1.20; 95% CI 1.02-1.41) were associated with cases. *Salmonella enterica* was isolated from 124 (84%) of the 159 persons with symptoms of food poisoning, and 91 (73%) were serogroup enteritidis, phage type B 14. Mayonnaise was prepared in the restaurant using a regular blender. Minced garlic was prepared with the same blender immediately after making the mayonnaise. Unsafe storage of the mayonnaise at room temperature for a median of 6 hours could have resulted in overgrowth of bacteria and a high infective dose of bacteria per serving."

243 Ananthan S, Saravanan P. Analysis of human rotavirus serotypes in children with acute diarrhoea in Chennai by monoclonal antibody based ELISA. Indian J Med Res 1998 Aug;108:58-61. 30 ref, Eng. Department of Microbiology, Dr ALM Post Graduate Institute of Basic Medical Sciences, University of Madras, Taramani, Chennai 600113, India

"To determine the individual human rotavirus serotypes prevailing in Chennai, 345 stool specimens obtained from children with acute diarrhoea between March 1996 and November 1997, were screened for the presence of rotavirus by the standardized enzyme linked immunosorbent assay (ELISA). Of the 90 (26%) rotavirus positive specimens, 75 (83.3%) were subgrouped and 65 (72.2%) were serotyped with monoclonal antibody based ELISA. Of the 65 specimens that could be serotyped, 52.3 per cent belonged to serotype 2, 24.6 per cent were serotype 4, 15.4 per cent were serotype 1 and 7.7 per cent were serotype 3. Of the 75 specimens typed for their subgroup specificity, 50.7 per cent were subgroup I, 26.7 per cent were subgroup II, 18.7 per cent equally specific to both subgroups I and II, 4 per cent belonged to nonsubgroups I and II. Our results indicate a predominance of serotype 2 virus. Unusual strain having both subgroups I and II specificity or neither specificity were also encountered."

244 Aysev AD, Guriz H. Drug resistance of *Shigella* strains isolated in Ankara, Turkey, 1993-1996. Scand J Infect Dis 1998;30(4):351-53. 17 ref, Eng. Ankara Universitesi Tip Fakultesi, Cocuk Sagligi ve Hastaliklari Anabilim Dali, Dikimevi, Ankara, Turkey

245 Badawi MH, Zaki M, Ismail EAR, Molla AM. Congenital chloride diarrhoea in Kuwait: a clinical reappraisal. J Trop Pediatr 1998 Oct;44(5):296-9. 7 ref, Eng. AL-Adam Hospital, PO Box 46969, 64020 Hadeia, Kuwait

"Congenital chloride diarrhoea (CCD) is a recessively inherited disorder of chloride transport in the distal ileum and colon. Congenital chloride diarrhoea is a common metabolic disorder in Kuwait with an incidence of 1/3200. Clinical findings in 14 children with CCD are reported over a period of 4 years. Maternal polyhydramnios, abdominal distension, watery diarrhoea, and a high faecal chloride level >90 mmol/l were the cardinal features in the neonatal period. In spite of the classical features of this disease 75 per cent of our cases were diagnosed beyond the neonatal period and all demonstrated chronic diarrhoea and failure to thrive, with hypochloraemia, hypokalaemia, and metabolic alkalosis. The practice of ultrasonic examination for pregnant women with polyhydramnios and, particularly, for those with previously affected siblings led to early identification of new cases among our population recently. The antenatal ultrasonic examination showed dilated intestinal loops which suggest CCD. The diagnosis was confirmed by a high faecal chloride level."

246 Baveja UK, Bhatia VN, Warhurst DC. *Giardia lamblia*: in-vitro sensitivity to some chemotherapeutic agents. J Commun Dis 1998 Jun;30(2):79-84. 16 ref, Eng. Department of Protozoology, London School of Tropical Medicine and Hygiene, Gower Street, London WCIE 7HT, UK

247 Bensoussan A, Talley NJ, Hing M, Menzies R, Guo A, Ngu M. Treatment of irritable bowel syndrome with Chinese herbal medicine: a randomized controlled trial. JAMA 1998 Nov 11;280(18):1585-9. 22 ref, Eng. Research Unit for Complementary Medicine, Faculty of Health, University of Western Sydney Macarthur, PO Box 555, Campbell-town, New South Wales 2560, Australia

248 Bhasin SK, Sharma AK, Agarwal OP. Prevalence of helminthic infections in a resettlement colony of East Delhi. Indian J Preven Soc Med 1998 Jan-Jun;29(1 and 2):29-32. 11 ref, Eng. Department of Preventive and Social Medicine, University College of Medical Sciences & G.T.B. Hospital, Shahdara, Delhi 110095, India

249 Bini EJ, Weinshel EH, Gamagaris Z. Comparison of duodenal with jejunal biopsy and aspirate in chronic human immunodeficiency virus—related diarrhea. Am J Gastroenterol 1998 Oct;93(10):1837-40. 27 ref, Eng. Division of Gastroenterology (IIID), Department of Veterans Affairs Medical Center, 423 East 23rd Street, New York, NY 10010, USA

Objectives: In human immunodeficiency virus (HIV)-infected patients with chronic unexplained diarrhea, upper endoscopy with small bowel biopsy and aspirate is often performed to identify treatable pathogens. The purpose of this study was to compare the diagnostic yield of duodenal with jejunal biopsy and aspirate. ***Methods:*** All HIV-infected patients with chronic unexplained diarrhea who were evaluated by upper endoscopy at Bellevue Hospital Center between January 1992 and January 1997 were identified. Data were collected by reviewing patient charts, endoscopy reports, and pathology records. ***Results:*** During the 5-yr study period, 442 patients underwent upper endoscopy with sampling of the duodenum (N=173) or jejunum (N=269). A pathogen was identified in 123 patients (27.8%). Microsporidia was the most common organism detected (12.2%). The diagnostic yield of jejunal biopsy and aspirate was significantly higher than that obtained from the

duodenum (32.3% *vs* 20.8%, p=0.009). Small bowel aspirates detected a pathogen in only 1.8% of patients evaluated, and there was no difference in the yield of duodenal and jejunal aspirates (1.3% *vs* 2.1%, p=0.7). Patients with a CD4 count of <100 cells/mm^3 were significantly more likely to have a pathogen identified than those with higher CD4 counts (38.8% *vs* 7.1%, p<0.0001). ***Conclusions:*** Upper endoscopy with small bowel biopsy and aspirate identifies a pathogen in 27.8% of individuals with HIV-related chronic unexplained diarrhea. In this patient population, jejunal biopsies acquired by enteroscopy are superior to those obtained from the duodenum. Small bowel aspirates are of little value in the workup of chronic HIV-related diarrhea."

250 Blagburn BL, Drain KL, Land TM, Kinard RG, Moore PH, Lindsay DS, Patrick DA, Boykin DW, Tidwell RR. Comparative efficacy evaluation of dicationic carbazole compounds, nitazoxanide, and paromomycin against *Cryptosporidium parvum* infections in a neonatal mouse model. Antimicrob Agent Chemother 1998 Nov; 42(11):2877-82. 26 ref, Eng. Department of Pathobiology, College of Veterinary Medicine, Auburn University, Auburn, Alabama 36849-5519, USA

251 Boisgerault F, Mounier J, Tieng V, Stolzenberg M-C, Khalil-Daher I, Schmid M, Sansonetti P, Charron D, Toubert A. Alteration of HLA-B27 peptide presentation after infection of transfected murine L cells by *Shigella flexneri*. Infect Immun 1998 Sep;66(9):4484-90. 46 ref, Eng. Institut Nationale Sante & Recherche Medicale U, 396, Hopital Saint-Louis, Centre G. Hayem, 1, Av. C. Vellefaux, 75475 Paris Cedex 10, France

252 Brush CF, Walter MF, Anguish LJ, Ghiorse WC. Influence of pretreatment and experimental conditions on electrophoretic mobility and hydrophobicity of *Cryptosporidium parvum* oocysts. Appl Environ Microbiol 1998 Nov;64(11):4439-45. 51 ref, Eng. Department of Geology and Geophysics, University of Hawaii at Manoa, Honolulu, HI 96822, USA

253 Buchanan RL, Edelson SG, Snipes K, Boyd G. Inactivation of *Escherichia coli* O157:H7 in apple juice by irradiation. Appl Environ Microbiol 1998 Nov;64(11):4533-5. 14 ref, Eng. Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration, Washington, DC 20204, USA

254 Bukhari Z, McCuin RM, Fricker CR, Clancy JL. Immunomagnetic separation of *Cryptosporidium parvum* from source water samples of various turbidities. Appl Environ Microbiol 1998 Nov;64(11):4495-9. 15 ref, Eng. Clancy Environmental Consultants, Inc., 272 North Main St., PO Box 314, St. Albans, VT 05478, USA

255 Buret A, Olson ME, Gall DG, Hardin JA. Effects of orally administered epidermal growth factor on enteropathogenic *Escherichia coli* infection in rabbits. Infect Immun 1998 Oct;66(10):4917-23. 46 ref, Eng. Department of Biological Sciences, University of Calgary, 2500 University Dr. NW, Calgary, AB, Canada T2N 1N4

"The increased intestinal absorption induced by epidermal growth factor (EGF) is associated with diffuse lengthening of brush border microvilli. The aim of this study was to examine the in vivo effects of oral administration of EGF during infection with enteropathogenic *Escherichia coli*. New Zealand White rabbits (4 weeks old) received orogastric EGF daily starting 3 days prior to infection with enteropathogenic *E. coli* RDEC-1 and were compared with sham-treated infected animals and uninfected controls. Weight gain, food intake, fecal *E. coli*, and stool consistency were assessed daily. On day 10, segments of jejunum, ileum, proximal, and distal colon were assessed for gram-negative bacterial colonization, disaccharidase activities, and epithelial ultrastructure. Effects of EGF on *E. coli* RDEC-1 proliferation were studied in vitro. *E. coli* RDEC-1 caused diarrhea and reduced weight gain. Seven days postinfection, the small and large intestines were colonized with numerous bacteria, brush border microvilli were disrupted, and maltase and sucrase activities were significantly reduced in the jejunum. Daily treatment with EGF prevented the occurrence of diarrhea and reduction of weight gain. These effects were associated with significant inhibition of *E. coli* colonization in the small and large intestine, improved jejunal maltase and sucrase activities and reduced microvillous injury. EGF did not affect the proliferation of *E. coli* in vitro. The findings suggest that EGF protects the gastrointestinal tract against colonization by enteropathogenic *E. coli*."

256 Chaicumpa W, Chongsa-nguan M, Kalambaheti T, Wilairatana P, Srimanote P, Makakunkijcharoen Y, Looareesuwan S, Sakolvaree Y. Immunogenicity of liposome-associated and refined antigen oral cholera vaccines in Thai volunteers. Vaccine 1998 Apr;16(7):678-84. 43 ref, Eng. Faculty of Tropical Medicine, Mahidol University, 420/6 Rajvithi Road, Bangkok 10400, Thailand

"A mixture of *Vibrio cholerae* antigens made up of crude fimbrial extract, lipopolysaccharide and procholeragenoid was administered orally to Thai volunteers either as free antigen or associated with liposomes. All vaccinees and controls were administered in three doses given at 14 day intervals. Nine volunteers received liposome-associated vaccine and seven received free vaccine. Liposomes without antigens were given to eight volunteers and seven volunteers received 5% $NaHCO_3$ solution alone. Both vaccines had 100% immunogenicity as determined by serum vibriocidal antibody responses. Liposomes were shown by indirect ELISA to localize the immune response against lipopolysaccharide and fimbriae to the intestinal mucosa. Vaccinees given liposome-associated antigens had a higher rate of antigen-specific antibody response than did individuals who had received free antigens. The vaccines induced intestinal antibodies of IgM and/or IgA isotypes, but not IgG antibody."

257 Checkley W, Epstein LD, Gilman RH*, Black RE, Cabrera L, Sterling CR. Effects of *Cryptosporidium*

***parvum* infection in Peruvian children: growth faltering and subsequent catch-up growth. Am J Epidemiol 1998 Sep 1;148(5):497-506. 34 ref, Eng.** *Department of International Health, Johns Hopkins School of Hygiene and Public Health, Room W3503, 615 N. Wolfe Street, Baltimore, MD 21205, USA

"The authors conducted a 2-year (1989-1991) community-based longitudinal study in a shantytown in Lima, Peru, to examine the effect of *Cryptosporidium parvum* infection on child growth during the year following the onset of infection. A cohort of children, aged 0-3 months at recruitment, was followed monthly for anthropometrics, weekly for stool samples, and daily for diarrheal status. Data from 185 children in the cohort permitted a comparison of growth in *C. parvum*-infected and noninfected children. The analyses fitted smooth, flexible curves with a linear random-effects model to estimate growth differences between *C. parvum*-infected and noninfected children. Children infected with *C. parvum* experienced growth faltering, both in weight and in height, for several months after the onset of infection, followed by a period of catch-up growth. Younger children took longer to catch up in weight than did older children. Catch-up growth, however, did not occur in children infected between ages 0 and 5 months. These children did not catch up in height, and one year after infection they exhibited an average deficit of 0.95 cm (95% confidence interval (CI) 0.38-1.53) relative to noninfected children of similar age. Stunted children who became infected also did not catch up in either weight or height, and one year after infection they exhibited a height deficit of 1.05 cm (95% CI 0.46-1.66) relative to noninfected, stunted children of similar age. These results indicate that *Cryptosporidium parvum* has a lasting adverse effect on linear (height) growth, especially when acquired during infancy and when children are stunted before they become infected."

258 Chen S, Xu R, Yee A, Wu KY, Wang C-N, Read S, Grandis SAD. An automated fluorescent PCR method for detection of Shiga toxin-producing *Escherichia coli* in foods. Appl Environ Microbiol 1998 Nov;64(11):4210-6. 40 ref, Eng. Office of Research, University of Guelph, Reynolds Building, Guelph, Ontario, Canada N1G 2W1

259 Chong C, Friberg M, Clements JD. LT (R192G), a non-toxic mutant of the heat-labile enterotoxin of *Escherichia coli*, elicits enhanced humoral and cellular immune responses associated with protection against lethal oral challenge with *Salmonella* spp. Vaccine 1998 Apr;16(7):732-40. 32 ref, Eng. Department of Microbiology and Immunology, 1430 Tulane Avenue, Tulane University Medical Center, New Orleans, LA 70112, USA

260 Ciarlet M, Crawford SE, Barone C, Bertolotti-Ciarlet A, Ramig RF, Estes MK, Conner ME. Subunit rotavirus vaccine administered parenterally to rabbits induces active protective immunity. J Virol 1998 Nov;72(11):9233-46. 82 ref, Eng. Division of Molecular Virology, Baylor College of Medicine, One Baylor Plaza, Houston, TX 77030, USA

"Virus-like particles (VLPs) are being evaluated as a candidate rotavirus vaccine. The immunogenicity and protective efficacy of different formulations of VLPs administered parenterally to rabbits were tested. Two doses of VLPs (2/6-, G3 2/6/7-, or P[2], G 3 2/4/6/7-VLPs) or SA11 simian rotavirus in Freund's adjuvants, QS-21 (saponin adjuvant), or aluminum phosphate (AIP) were administered. Serological and mucosal immune responses were evaluated in all vaccinated and control rabbits before and after oral challenge with 10^3 50% infective doses of live p[14], G3 ALA lapine rotavirus. All VLP- and SA11-vaccinated rabbits developed high levels of rotavirus-specific serum and intestinal immunoglobulin G (IgG) antibodies but not intestinal IgA antibodies. SA11 and 2/4/6/7-VLPs afforded similar but much higher mean levels of protection than 2/6/7- or 2/6-VLPs in QS-21. The presence of neutralizing antibodies to VP4 correlated ($P<0.001$, $r=0.55$; Pearson's correlation coefficient) with enhanced protection rates, suggesting that these antibodies are important for protection. Although the inclusion of VP4 resulted in higher mean protection levels, high levels of protection (87 to 100%) from infection were observed in individual rabbits immunized with 2/6/7- or 2/6-VLPs in Freund's adjuvants. Therefore, neither VP7 nor VP4 was absolutely required to achieve protection from infection in the rabbit model when Freund's adjuvant was used. Our results show that VLPs are immunogenic when administered parenterally to rabbits and the Freund's adjuvant is a better adjuvant than QS-21. The use of the rabbit model may help further our understanding of the critical rotavirus proteins needed to induce active protection. VLP are a promising candidate for a parenterally administered subunit rotavirus vaccine."

261 Conlan JW, Perry MB. Susceptibility of three strains of conventional adult mice to intestinal colonization by an isolate of *Escherichia coli* O157:H7. Can J Microbiol 1998 Aug;44(8):800-5. 17 ref, Eng. Institute for Biological Sciences, National Research Council Canada, Ottawa, ON K1A OR6, Canada

262 Cooling LLW, Walker KE, Gille T, Koerner TAW. Shiga toxin binds human platelets via globotriaosylceramide (P^k antigen) and a novel platelet glycosphingolipid. Infect Immun 1998 Sep;66(9):4355-66. 100 ref, Eng. Department of Pathology, SUNY Health Science Center at Syracuse, 750 East Adams St., Syracuse, NY 13210, USA

263 Corrao G, Tragnone A, Caprilli R, Trallori G, Papi C, Andreoli A, Paolo MD, Reigler G, Rigo G-P, Ferrau O, Mansi C, Ingrosso M, Valpiani D. Risk of inflammatory bowel disease attributable to smoking, oral contraception and breastfeeding in Italy: a nationwide case-control study. Int J Epidemiol 1998 Jun;27(3):397-404. 45 ref, Eng. Department of Statistics, Università di Milano, Viale Sarca, 202, 20126 Milano, Italy

264 Cortese VS, Grooms DL, Ellis J, Bolin SR, Ridpath JF, Brock KV. Protection of pregnant cattle and their fetuses against infection with bovine viral diarrhea virus type 1 by use of a modified-live virus vaccine. Am J Vet Res 1998 Nov;59(11):1409-13. 16 ref, Eng. Department of Veterinary Microbiology, Western College of Veterinary Medicine, University of Saskatchewan, Saskatoon, Saskatchewan, Canada S7N 5B4

265 Coulson BS. Longitudinal studies of neutralizing antibody responses to rotavirus in stools and sera of children following severe rotavirus gastroenteritis (note). Clin Diagn Lab Immunol 1998 Nov;5(6):897-901. 16 ref, Eng. Department of Microbiology and Immunology, University of Melbourne, Parkville 3052, Victoria, Australia

266 Craven D, Brick D, Morrisey A, O'Riordan MA, Petran V, Schreiber JR*. Low yield of bacterial stool culture in children with nosocomial diarrhea. Pediatr Infect Dis J 1998 Nov;17(11):1040-4. 25 ref, Eng. *Division of Infectious Disease, Rainbow Babies and Children's Hospital, 11100 Euclid Ave., Cleveland, OH 44106, USA

*"**Objective.*** To determine whether bacterial stool cultures (BSC) are useful in initial evaluation of children with symptoms of nosocomial diarrhea. To answer this question we performed a retrospective record review to determine the yield of BSC in children who developed diarrhea after the third hospital day ()HD-3). **Methods.** The hospital computer record keeping system was utilized to compile the result of BSC collected from children and adolescents ages O to 20 years between January 1, 1988, and October 31, 1996. All specimens were analyzed for *Salmonella, Shigella, Yersinia* and *Campylobacter*. We reviewed hospital charts of all children who developed a positive BSC beyond HD-3 to determine the time of onset of diarrhea and clinical circumstances. **Results.** A total of 11 516 BSCs were submitted from 9262 children during the 8½-year period. Five hundred sixty-eight (6.6%) of 9262 children had at least 1 positive BSC. Two thousand five hundred seventy-two children had the first BSC submitted after HD-3 and 13 (0.5%) of these children had a positive result. Chart review of these 13 children demonstrated that 6 had onset of diarrhea during the first 3 hospital days. Therefore only 7 children met our criteria for having nosocomially acquired diarrhea caused by a bacterial pathogen. Children whose first BSC was submitted after HD-3 accounted for 3767 (46%) of the total 8126 inpatient BSCs and in excess of $ 21 000 annually in patient billing charges. *Conclusion.* In the absence of a known exposure the isolation of a bacterial pathogen from the stool of children with onset of diarrhea beyond HD-3 is a rare event. Under most circumstances BSC should not be part of the initial evaluation of children with symptoms of nosocomial diarrhea."

267 Ene-Obong HN, Uwaegbute AC, Iroegbu CU, Amazigo UV. The effect of two child-care practices of market women on diarrhoea prevalence, feeding patterns and nutritional status of children aged 0-24 months. J Diarrhoeal Dis Res 1998 Sep; 16(3):173-9. 21 ref, Eng. Departments of Home Science and Nutrition, University of Nigeria, Nsukka, Enugu State, Nigeria

"A cross-sectional comparative study was conducted in seven markets in Enugu and Nsukka, Nigeria, to evaluate the child-care practices, occurrence of diarrhoea, feeding patterns and nutritional status among 506 and 157 children aged 0-24 months taken to the market (CTTM) by their mothers and those left at home (CLAH) respectively. A lack of a caretaker at home was the most common reason for taking children to the market. The majority (92%) of the caretakers at home were young (<20 years). There was no difference in the occurrence of diarrhoea in the last month between the CTTM (39%) and the CLAH (41%) groups ($p>0.05$). However, there were differences in reported diarrhoea episodes between children aged 0-6 months and higher age classes ($c^2=20.0$; $p=0.003$). Very few children (0.8%) were exclusively breastfed. More CTTM (58%) were still being breastfed than the 42% of CLAH (OR=1.87; 95% C.I.=1.27-2.37; $p<0.001$). Children cared for at home had a slightly better but non-significant ($p>0.05$) anthropometric status as characterised by weight-for-age and height-for-age. There was no relationship between feeding patterns and diarrhoea."

268 Enriquez FJ, Riggs MW. Role of immunoglobulin A monoclonal antibodies against P23 in controlling murine *Cryptosporidium parvum* infection. Infect Immun 1998 Sep;66(9):4469-73. 45 ref, Eng. Department of Veterinary Science and Microbiology, University of Arizona, Veterinary Science and Microbiology Building, Room 202, Tucson, AZ 85721, USA

"*Cryptosporidium parvum* is an important diarrhea-causing protozoan parasite of immunocompetent and immunocompromised hosts. Immunoglobulin A (IgA) has been implicated in resistance to mucosal infections with bacteria, viruses, and parasites, but little is known about the role of IgA in the control of *C. parvum* infection. We assessed the role of IgA during *C. parvum* infection in neonatal mice. IgA-secreting hybridomas were developed by using Peyer's patch lymphocytes from BALB/c mice which had been orally inoculated with viable *C. parvum* oocysts. Six monoclonal antibodies (MAbs) were selected for further study based on indirect immunofluorescence assay reactivity with sporozoite and merozoite pellicles and the antigen (Ag) deposited on glass substrate by gliding sporozoites. Each MAb was secreted in dimeric form and recognized a 23-kDa sporozoite Ag in Western immunoblots. The Ag recognized comigrated in sodium dodecyl sulfate-polyacrylamide gel electrophoresis with P23, a previously defined neutralization-sensitive zoite pellicle Ag. MAbs were evaluated for prophylactic or therapeutic efficacy against *C. parvum*, singly and in combinations, in neonatal BALB/c mice. A combination of two MAbs given prophylactically prior to and 12 h following oocyst challenge reduced the number of intestinal parasites scored histologically by 21.1% compared to the numbers in mice given an isotype-matched control MAb ($P<0.01$). Individual

MAbs given therapeutically in nine doses over a 96-h period following oocyst challenge increased efficacy against *C. parvum* infection. Four MAbs given therapeutically each reduced intestinal infection 34.4 to 42.2% compared to isotype-match control MAb-treated mice ($P<0.05$). One MAb reduced infection 63.3 and 72.7% in replicate experiments compared to isotype-match control MAb-treated mice ($P<0.0001$). We conclude that IgA MAbs directed to neutralization-sensitive P23 epitopes may have utility in passive immunization against murine *C. parvum* infection."

269 Faruque ASG, Salam MA, Faruque SM, Fuchs GJ. Aetiological, clinical and epidemiological characteristics of a seasonal peak of diarrhoea in Dhaka, Bangladesh. Scand J Infect Dis 1998;30(4):393-6. 14 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"During the spring peak of diarrhoea in Bangladesh, 113 consecutive patients who represented a systematic 4% sample of all patients attending an urban diarrhoea treatment facility between 18 and 23 April 1995 were studied. The study was conducted to characterize enteric pathogens associated with the spring peak of the diarrhoea outbreak in Bangladesh and to describe clinical and epidemiological features of the patients. The spring peak is traditionally thought to be mostly due to *V. cholerae* O1. However, the most common cause of diarrhoea among the study patients was enterotoxigenic *Escherichia coli* (36%) followed by *Vibrio cholerae* O1 (23%). The *V. cholerae* O1 patients attended significantly ($p<0.01$) sooner after onset of diarrhoea than enterotoxigenic *E. coli* (ETEC) patients. Studies of behavioural and environmental characteristics are important to determine risk factors for observed higher proportion of ETEC infection during seasonal diarrhoea peaks."

270 Faruque ASG, Teka T, Fuchs GJ. Shigellosis in children: a clinico-epidemiological comparison between *Shigella dysenteriae* type I and *Shigella flexneri*. Ann Trop Paediatr 1998 Sep;18(3):197-201. 16 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh.

"We reviewed the clinical and epidemiological features of 390 children under 5 years of age infected with either *Shigella dysenteriae* type I or *Shigella flexneri* attending a diarrhoea treatment centre from 1993 to 1995 in Dhaka, Bangladesh. Older age (24 months or more), underweight and wasting but not stunting were the host factors significantly more associated with *Shigella dysenteriae* type I infection than in *Shigella flexneri*-infected children. Moreover, use of antibiotics at home, use of water from tubewells or pipe-water for drinking and lack of sanitary facilities for defaecation were the behavioural and environmental factors strongly associated with *S. dysenteriae* type I infection. Children with diarrhoea due to *S. flexneri* presented with more watery/liquid stools and had a shorter duration of illness. Duration of diarrhoea for 4 or more days was typical of *S. dysenteriae* type I infection. Interventions to address malnutrition and to promote environmental hygiene would be predicted to offer greater protection against shigellosis due to *S. dysenteriae* than *S. flexneri*."

271 Fernandez FM, Conner ME, Hodgins DC, Parwani AV, Nielsen PR, Crawford SE, Estes MK, Saif LJ. Passive immunity to bovine rotavirus in newborn calves fed colostrum supplements from cows immunized with recombinant SA11 rotavirus core-like particle (CLP) or virus-like particle (VLP) vaccines. Vaccine 1998 Mar;16(5):507-16. 33 ref, Eng. Food Animal Health Research Program and Veterinary Preventive Medicine Department, Ohio Agricultural Research and Development Center, The Ohio State University, 1680 Madison Avenue, Wooster, OH 44691, USA

272 Fisseha B, Petros B, Woldemichael T. *Cryptosporidium* and other parasites in Ethiopian AIDS patients with chronic diarrhoea. East Afr Med J 1998 Feb;75(2):100-1. 12 ref, Eng. Ethiopian Health and Nutrition Research Institute, PO Box 1242, Addis Ababa, Ethiopia

"Previous studies in other African countries have shown high prevalences of *Isospora belli* and *Cryptosporidium parvum* infections in acquired immunodeficiency syndrome (AIDS) patients with chronic diarrhoea. However, there is lack of information regarding these infectious agents in Ethiopian AIDS patients. Thus, this investigation has been aimed at determining the prevalence of *Cryptosporidium* and other related gastrointestinal parasites in AIDS patients with diarrhoea in seven Addis Ababa hospitals. Stool specimens from 246 clinically diagnosed AIDS patients were parasitologically screened for parasitic infections constituting HIV-negative with diarrhoea and HIV-positive without diarrhoea control groups. A variety of intestinal protozoa and helminths were found in 50% of AIDS patients and 42% of the controls. Cryptosporidiosis was detected in 38 (25.9%), Isosporiasis in two (1.4%), and blastocystosis in one (0.7%) of the AIDS patients with diarrhoea. The occurrence of cryptosporidiosis among the AIDS patients and possible explanation of the increasingly high study subjects with diarrhoea but without identifiable parasites is discussed."

273 Fontaine O. Oral rehydration solution. Bull WHO 1998;76(4):325. Eng. Division of Child Health and Development, World Health Organization, 1211 Geneva 27, Switzerland

274 Forney JR, Vaughan DK, Yang S, Healey MC. Actin-dependent motility in *Cryptosporidium parvum* sporozoites. J Parasitol 1998 Oct;84(5):908-13. 31 ref, Eng. Department of Animal, Dairy, and Veterinary Sciences, College of Agriculture, Utah State University, Logan, Utah 84322, USA

275 Fraser D, Dagan R, Porat N, El-On J, Alkrinawi S, Deckelbaum RJ, Naggan L. Persistent diarrhea in a cohort of Israeli Bedouin infants: role of enteric

pathogens and family and environmental factors. J Infect Dis 1998 Oct;178(4):1081-8. 44 ref, Eng. Epidemiology and Health Services Evaluation Department, Faculty of Health Sciences, Ben-Gurion University of the Negev, PO Box 653, Beer-Sheva 84105, Israel

"This study examined the role of enteric pathogens and infant, family, and household characteristics in persistent diarrhea. Bedouin infants from southern Israel were followed from birth to age 18-23 months. During monthly home visits, stool samples were obtained, and feeding practices and history of diarrhea were determined, and at age 3 months, an environmental assessment was done. Diarrhea surveillance was either via a network covering all community health care facilities or via weekly interviews with the mother. None of the enteric pathogens examined, including *Cryptosporidium parvum* and enteroaggregative *Escherichia coli*, were associated with persistent diarrhea. In multivariate analyses, age at first diarrheal illness and maternal age and maternal education were independently and significantly associated with the risk of persistent diarrhea. These data suggest that persistent diarrhea is a clinical entity that may be related less to a specific enteric pathogen and more to the health experiences of children and their home environment."

276 Fujimoto S, Meno Y, Horikawa K. Heterogeneity in expression of lipopolysaccharide and major outer-membrane proteins by strains of *Escherichia coli* O157 with different H-serotypes. Microbiol Immunol 1998;42(8):527-31. 21 ref, Eng. Department of Bacteriology, Faculty of Medicine, Kyushu University, 3-1-1 Maidashi, Higashi-ku, Fukuoka, Fukuoka 812-8582, Japan

277 Graczyk TK, Farley CA, Fayer R, Lewis EJ, Trout JM. Detection of *Cryptosporidium* oocysts and *Giardia* cysts in the tissues of eastern oysters (*Crassostrea virginica*) carrying principal oyster infectious diseases. J Parasitol 1998 Oct;84(5):1039-42. 19 ref, Eng. Department of Molecular Microbiology and Immunology, School of Hygiene and Public Health, Johns Hopkins University, Baltimore, Maryland 21205, USA

278 Griffiths JK. Treatment for AIDS-associated cryptosporidiosis (editorial). J Infect Dis 1998 Sep;178(3):915-6. 7 ref, Eng. Department of Family Medicine and Community Health, Tufts University School of Medicine, 136 Harrison Ave., Boston, MA 02111, USA

279 Guerin-Danan C, Meslin JC, Lambre F, Charpilienne A, Serezat M, Bouley C, Cohen J, Andrieux C. Development of a heterologous model in germfree suckling rats for studies of rotavirus diarrhea (note). J Virol 1998 Nov;72(11):9298-302. 31 ref, Eng. Unité d'Ecologie et de Physiologie du Système Digestif, Equipe Métabolites Bactériens et Santé, INRA-Domaine de Vilvert, 78352 Jouy-en-Josas Cedex, France

280 Guerin F, Bouguenec CL, Gilquin J, Haddad F, Goldstein FW. Bloody diarrhea caused by *Klebsiella pneumoniae:* a new mechanism of bacterial virulence (brief report)? Clin Infect Dis 1998 Sep;27(3):648-9. 5 ref, Eng. Service de Microbiologie Médicale, Fondation Hôpital Saint Joseph, 185 rue Raymond Losserand, 75014 Paris, France

281 Guerrero ML, Noel JS, Mitchell DK, Calva JJ, Morrow AL, Martinez J, Rosales G, Velazquez FR, Monroe SS, Glass RI, Pickering LK, Ruiz-Palacios GM. A prospective study of astrovirus diarrhea of infancy in Mexico city. Pediatr Infect Dis J 1998 Aug;17(8):723-7. 25 ref, Eng. Instituto Nacional de la Nutricion, Departamento de Infectologia, Vasco de Quiroga 15, Tlalpan 14000, Mexico

"**Aim.** To describe the epidemiologic and clinical characteristics of astrovirus-associated diarrhea in a cohort of young children from a periurban community in Mexico city. **Methods.** From November, 1988, through December, 1991, a total of 214 children were enrolled in a longitudinal study of diarrhea and monitored from birth to 18 months of age. A stool specimen was collected during each episode of diarrhea. Specimens from a total of 510 diarrhea episodes were tested for astrovirus by enzyme immunoassay and examined for other enteric pathogens. The antigenic types of astrovirus were determined by a typing enzyme immunoassay. **Results**. Astrovirus was detected in 26 (5%) of 510 diarrhea episodes, with an incidence rate of 0.1 episode/child year; the highest rate was in children 13 to 18 months of age. Astrovirus-associated diarrhea was characterized by a median of 4 stools (range, 2 to 10) during the first 24 h, a median duration of 3 days (range, 1 to 21), vomiting (20%), and fever (7%). No cases of dehydration or repeat symptomatic infections were observed. Coinfection with another pathogen was detected in 11 of the 26 episodes (42%). Serotype 2 (35%) was most common, followed by serotypes 4 (15%), 3 (11%), and 1 and 5 (4% each); 31% were nontypable. Astrovirus-associated diarrhea was less severe, as measured by the number of stools (4.3 ± 1.9), than diarrhea caused by rotavirus (7.1 ± 2.8) or when coinfections occurred (5.5 ± 1.6; P=0.008). **Conclusions**. Astrovirus was associated with 5% of the episodes of diarrhea in this cohort of young Mexican children and presented as a mild secretory diarrhea. Five predominant antigenic types were detected with type 2 being the most common."

282 Halsey NA, Abramson JS, Chesney PJ, Fisher MC, Gerber MA, Marcy SM, Murray DL, Overturf GD, Prober CG, Saari TN, Weiner LB, Whitley RJ, Baker C, Peter G, Pickering LK, Hirsch A, Jacobs RF, MacDonald NE, Schwartz B, Orenstein WA, Hardegree MC, Rabinovich NR, Breiman RF. Prevention of rotavirus disease: guidelines for use of rotavirus vaccine. Pediatrics 1998 Dec;102(6):1483-91. 73 ref, Eng. Centers for Disease Control and Prevention, 1600 Clifton Road, Mailstop E69, Atlanta, GA 30333, USA

"Virtually all children experience rotavirus (Rv) infection before school entry. In the United States and other temperate

countries, Rv disease peaks in the winter and during this time is responsible for the majority of episodes of diarrhea in infants and young children. Data collected by the Centers for disease Control and Prevention from 1979 through 1992 indicate that approximately 50 000 hospitalizations attributable to Rv occur annually in the United States, a number that approximates about 1 in 78 children being hospitalized with Rv diarrhea by 5 years of age. *RotaShield* (Wyeth-Lederle Vaccines and Pediatrics, Philadelphia, PA) was licensed by the Food and Drug Administration on August 31, 1998, for oral administration to infants at 2, 4, and 6 months of age. The rationale for using Rv immunization for prevention or modification of Rv disease is based on several considerations. First, the rate of illness attributable to Rv among children is comparable in industrialized and developing countries, which indicates that improved public sanitation is unlikely to decrease the incidence of disease. Second, although implementation of oral rehydration programs to prevent dehydration has improved in the United States, widespread use is inadequate to prevent significant morbidity. Third, trials of rhesus rotavirus-tetravelent (Rv) vaccine in the United States, Finland, and Venezuela show efficacy rates of approximately 80% for prevention of severe illness and 48% to 68% against Rv-induced diarrheal episodes. These results are similar to the protection observed after natural Rv infection, which also confers better protection against subsequent episodes of severe disease than against mild illness. This statement provides recommendations regarding the use of Rv vaccine in infants in the United States."

283 Hartman AB, Venkatesan MM. Construction of a stable attenuated *Shigella sonnei* D*virG* vaccine strain, WRSS1, and protective efficacy and immunogenicity in the Guinea pig keratoconjunctivitis model (note). Infect Immun 1998 Sep;66(9):4572-6. 39 ref, Eng. Department of Enteric Infections, Walter Reed Army Institute of Research, Washington, DC 20307-5100, USA

284 Hazeleger WC, Wouters JA, Rombouts FM, Abee T. Physiological activity of *Campylobacter jejuni* far below the minimal growth temperature. Appl Environ Microbiol 1998 Oct;64(10):3917-22. 43 ref, Eng. Wageningen University and Research Centre, Department of Food Technology and Nutritional Sciences, Laboratory of Food Microbiology, Bomenweg 2, NL 6703 HD Wageningen, The Netherlands

"The behavior of *Campylobacter jejuni* at environmental temperatures was examined by determining the physiological activities of this human pathogen. The minimal growth temperatures were found to be 32 and 31°C for strains 104 and ATCC 33560, respectively. Both strains exhibited a sudden decrease in growth rate from the maximum to zero within a few degrees not only near the maximal growth temperature but also near the minimal growth temperature. This could be an indication that a temperature-dependent transition in the structure of a key enzyme(s) or regulatory compound(s) determines the minimal growth temperature. Oxygen consumption, catalase activity, ATP generation, and protein synthesis were observed at temperatures as low as 4°C, indicating that vital cellular processes were still functioning. PCR analysis showed that cold shock protein genes, which play a role in low-temperature adaptation in many bacteria, are not present in *C. jejuni*. The fact that chemotaxis and aerotaxis could be observed at all temperatures shows that the pathogen is able to move to favorable places at environmental temperatures, which may have significant implications for the survival of *C. jejuni* in the environment."

285 Helander A, Wenneras C, Qadri F, Svennerholm A-M. Antibody responses in humans against coli surface antigen 6 of enterotoxigenic *Escherichia coli*. Infect Immun 1998 Sep;66(9):4507-10. 21 ref, Eng. Department of Medical Microbiology and Immunology, Göteborg University, Guldhedsgatan 10A, 413 46 Göteborg, Sweden

"Enterotoxigenic *Escherichia coli* (ETEC) strains expressing only coli surface antigen 6 (CS6) have previously been isolated from patients with diarrhea, but the immunogenicity of CS6 has not been established in humans. We have detected CS6-specific immunoglobulin A responses in the feces and blood of patients convalescing from natural ETEC disease and of volunteers given an oral ETEC vaccine."

286 Högenauer C, Hammer HF, Krejs GJ, Reisinger EC. Mechanisms and management of antibiotic-associated diarrhea. Clin Infect Dis 1998 Oct;27(4):702-10. 90 ref, Eng. Division of Infectious Diseases, Department of Medicine, Karl-Franzens University, Auenbruggerplatz 15, A-8036 Graz, Austria

"Only 10%-20% of all cases of antibiotic-associated diarrhea (AAD) are caused by infection with *Clostridium difficile*. Other infectious organisms causing AAD include *Clostridium perfringens, Staphylococcus aureus, Klebsiella oxytoca, Candida* species, and *Salmonella* species. Most of the clinically mild AAD cases are due to functional disturbances of intestinal carbohydrate or bile acid metabolism, to allergic and toxic effects of antibiotics on intestinal mucosa, or to pharmacological effects on motility. *Saccharomyces boulardii* and *Enterococcus* SF68 can reduce the risk of developing AAD. Patients receiving antibiotic treatment should avoid food containing high amounts of poorly absorbable carbohydrates. Mild cases of AAD that may or may not be caused by *C. difficile* can be resolved by discontinuation of antibiotic therapy and by dietary carbohydrate reduction. Only severe AAD caused by *C. difficile* requires specific antibiotic treatment."

287 Hong M, Gleason Y, Wyckoff EE, Payne SM. Identification of two *Shigella flexneri* chromosomal loci involved in intercellular spreading. Infect Immun 1998 Oct;66(10):4700-10. 68 ref, Eng. Department of Microbiology, University of Texas at Austin, Austin, TX 78712-1095, USA

"The ability of *Shigella flexneri* to multiply within colonic epithelial cells and spread to adjacent cells is essential for

production of dysentery. Two *S. flexneri* chromosomal loci that are required for these processes were identified by screening a pool of Tn*phoA* insertion mutants. These mutants were able to invade cultured epithelial cells but could not form wild-type plaques. Analysis of the nucleotide sequence indicated that the sites of Tn*phoA* insertion were within two different regions that are almost identical to *Escherichia coli* K-12 chromosomal sequences of unknown functions. One region is located at 70 min on the *E. coli* chromosome, upstream of *murZ*, while the other is at 28 min, downstream of *tonB*. The mutant with the insertion at 70 min was named *vpsC* because it showed an altered pattern of virulence protein secretion. The *vpsC* mutant formed pinpoint-sized plaques, was defective in recovery from infected tissue culture cells, and was sensitive to lysis by the detergent sodium dodecyl sulfate. Recombinant plasmids carrying the *S. flexneri vpsA, -B,* and -C genes complemented all of the phenotypes of the *vpsC* mutant. A mutation in *vpsA* resulted in the same phenotype as the *vpsC* mutation, suggesting that these two genes are part of a virulence operon in *S. flexneri*. The mutant with the insertion at 28 min was interrupted in the same open reading frame as *S. flexneri ispA*. This *ispA* mutant could not form plaques and was defective in bacterial septation inside tissue culture cells."

288 Huttly SRA, Lanata CF, Yeager BAC, Fukumoto M, del Aguila R, Kendall C. Feces, flies, and fetor: findings from a Peruvian shantytown. Pan Am J Public Health 1998 Aug;4(2):75-9. 7 ref, Eng. Maternal and Child Epidemiology Unit, London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT, UK

"Sanitary disposal of feces is vital to combat childhood diarrhea, and its promotion is key to improving health in developing countries. Knowledge of prevailing feces disposal practices is a prerequisite to formulation of effective intervention strategies. Two studies were conducted in a shantytown area of Lima, Peru. First, information was gathered through in-depth interviews with mothers and structured observations (4 hours) of young children and their caretakers. Data on beliefs and practices related to feces disposal behaviors were obtained. Excreta were deposited by animals or humans in or near the house in 82% of households observed. Beliefs about feces depended on their source and were reflected in how likely the feces were to be cleared. While 22% of children aged ³18 months were observed to use a potty for defecation, 48% defecated on the ground where the stools often remained. Although almost all children were cleaned after defecation, 30% retained some fecal matter on their body or clothes. Hand-washing after the child's defecation was extremely rare for both children (5%) and caretakers (20%). The hygienic disposal of feces poses problems in this type of community. Nevertheless existing practices were found that show promise for promotion on a wider scale, including greater use of potties."

289 Iijimia H, Takahashi I, Hiroi T, Shimaoka M, Kawano S, Nagano K, Hori M, Kiyono H. Orally administered cholera toxin prevents murine intestinal T cells from staphylococcal enterotoxin B-induced anergy. Gastroenterology 1998 Nov;115(5): 1197-1204. 25 ref, Eng. Department of Mucosal Immunology, Research Institute for Microbial Diseases, Osaka University, Osaka 565-0871, Japan

290 Jalgaonkar SV, Ingole KV, Ambhore NA, Fule RP. Re-emergence of *Vibrio cholerae* serogroup O139 during June-August, 1997 in Yavatmal (Maharashtra). Indian J Med Res 1998 Jul;108:1-2. 5 ref, Eng. Department of Microbiology, Shri V.N. Government Medical College, Yavatmal 445001, India

291 Jamal WY, Pal T, Rotimi VO, Chugh TD. Serogroups and antimicrobial susceptibility of clinical isolates of *Salmonella* species from a teaching hospital in Kuwait. J Diarrhoeal Dis Res 1998 Sep;16(3):180-6. 31 ref, Eng. Department of Microbiology, Faculty of Medicine, Kuwait University, PO Box 24923, Safat 13110, Kuwait

"*Salmonella* strains isolated in a teaching-cum-general hospital in Kuwait during 1990-1993 and 1996 were analysed to determine the trend in the prevalence of the serogroups and their changing pattern of susceptibility. The records were reviewed for all the 661 isolates encountered during these periods. The most prevalent serogroup in both children and adults was serogroup B, followed by serogroup C and D. A sizeable proportion of the strains were resistant to first-line drugs. About 39% of the isolates were resistant to ampicillin, 17% to co-trimoxazole, 13% to chloramphenicol, and 15% to cephalothin. The majority were, however, susceptible to the other drugs with low to very low resistance rates: 7% to amoxicillin/clavulanic acid, and 0.3% to cefotaxime. All the strains were susceptible to ciprofloxacin. In all, resistant strains were more prevalent among children than adults."

292 James VLA, Lambden PR, Caul EO, Clarke IN. Enzyme-linked immunosorbent assay based on recombinant human group C rotavirus inner capsid protein (VP6) to detect human group C rotaviruses in fecal samples. J Clin Microbiol 1998 Nov;36(11):3178-81. 22 ref, Eng. Public Health Laboratory, Southampton General Hosptial, Southampton SO16 6YD, UK

"A recent study showed that 43% of a population in the United Kingdom were seropositive for group C rotavirus. The higher than expected incidence may be due to limited diagnosis of acute human group C rotavirus infections because no routine test is available. Human group C rotavirus infections are routinely diagnosed by electron microscopy (EM) and a negative group A rotavirus enzyme-linked immunosorbent assay (ELISA) result. An antigen-detection ELISA was developed with hyperimmune antibodies raised to human group C rotavirus recombinant VP6 (Bristol strain) expressed in inset cells. The assay was used to screen fecal samples to determine the prevalence of gorup C rotavirus infection. Samples positive by ELISA were confirmed by EM, polyacrylamide gel electrophoresis of double-stranded RNA, or detection of the VP6 gene by reverse transcription-

PCR. Retrospective analysis indicated a 1 to 2% detection rate of positivity among samples from patients with acute diarrhea."

293 Jenkin GA, Tee W. *Campylobacter upsaliensis*—associated diarrhea in human immunodeficiency virus—infected patients. Clin Infect Dis 1998 Oct;27(4):816-21. 33 ref, Eng. Victorian Infectious Diseases Reference Laboratory, Locked Bag 815, Jane Bell House, 10 Wreckyn Street, North Melbourne, Victoria 3051, Australia

"*Campylobacter upsaliensis* was isolated from the feces of 20 human immunodeficiency virus (HIV)-infected patients with diarrhea over a 67-month period, representing 18.5% of fecal *Campylobacter* isolates from our HIV—seropositive patients. All isolates were recovered from feces by a 0.45-mm-membrane filtration method. Culture on Skirrow's selective medium incubated at 42°C failed to support any growth. The mean CD4 cell count of the patient group was 185/mm^3 (range, 0-840/mm^3), and all patients had diarrhea for 1-60 days (mean, 19 days). Thirteen had no other enteric pathogens isolated, and three patients (CD4 counts, £70/mm^3) had persistent carriage for between 5 weeks and 3 months. All tested isolates were susceptible to erythromycin and doxycycline, but three isolates from two patients were resistant to ciprofloxacin. *C. upsaliensis* is associated with prolonged diarrhea of mild to moderate severity in HIV-infected patients."

294 Jernigan JA, Siegman-Igra Y, Guerrant RC, Farr BM*. A randomized crossover study of disposable thermometers for prevention of *Clostridium difficile* and other nosocomial infections. Infect Cont Hosp Epidemiol 1998 Jul;19(7):494-9. 14 ref, Eng. *University of Virginia Health Sciences Center, Box 473, Charlottesville, VA 22908, USA

295 Johnson JL, Brooke CL, Fritschel SJ*. Comparison of the BAX for screening/*E. coli* O157:H7 method with conventional methods for detection of extremely low levels of *Escherichia coli* O157:H7 in ground beef. Appl Environ Microbiol 1998 Nov;64(11):4390-5. 17 ref, Eng. *Qualicon, Inc., Route 141 & Henry Clay Rd., Wilmington, Delaware 19880-0357, USA

296 Jongpiputvanich S, Veeravongs S, Wonsekiarttirat W. Difficulties in conducting participatory action research to prevent diarrhoea in a slum area of Bangkok. J Diarrhoeal Dis Res 1998 Sep;16(3):187-93. 12 ref, Eng. Department of Pediatrics, Faculty of Medicine, Chulalongkorn University, Bangkok 10330, Thailand

"Childhood diarrhoea is a major cause of childhood morbidity and mortality in Thailand. During April 1988-April 1990, a study was conducted to evaluate participatory action research as a method of intervening on unhygienic practices in two communities in the Klong Toey slum of Bangkok (intervention group) and compared these practices in two other communities of the same slum where there was no intervention (control group). The following variables were compared in two groups: hygiene behaviour and factors associated with participation of community leaders and mothers with children aged less than five years. Two hundred and twenty-one mothers of the original 606 (36%) were still available at the end of the study. It was found that the mothers in the intervention groups were more likely to wash their hands before feeding and use cupboards and covered containers for keeping food. Several factors negatively affected community participation: matters concerning the slum culture and organization, community leadership, family problems, occupational problems, and physical environment of the community. The conclusions of the study are limited because of the considerable proportion of the subjects that was lost to follow-up. The study points at some of the methodological issues when conducting research in challenging and difficult to reach communities that have important health problems."

297 Kabir I, Rahman MM, Haider R, Mazumder RN, Khaled MA, Mahalanabis D. Increased height gain of children fed a high-protein diet during convalescence from shigellosis: a six-month follow-up study. J Nutr 1998 Oct;128(10):1688-91. 21 ref, Eng. International Centre for Diarrhoeal Disease Research, Banglaesh, GPO Box 128, Dhaka 1000, Bangladesh

"The impact of dietary supplementation on catch-up growth was evaluated in 69 malnourished children ages 24-60 mo after recovery from shigellosis. They were fed either a high-protein (HP) diet with 15% of energy as protein, or a standard-protein (SP) diet with 7.5% energy as protein, for 3 wk in a metabolic study ward. Children were followed up bi-weekly for 6 mo by trained health assistants when anthropometric measurements and information of any illness were collected. Thirty-one children in the HP group and 28 children in the SP group completed 6-mo follow-up. The increase in height (mean ± SD) was 5.3 ± 1.0 cm vs. 4.1 ± 1.1 cm for HP and SP groups, respectively (P<0.001), whereas increase in body weight was 1.39 ± 0.58 and 1.29 ± 0.72 kg for children fed HP and SP, respectively (P=0.59). The proportion of children who were severely stunted (<-2 SD height-for-age) decreased from 45 to 29% in the HP group compared to 50 to 46% in the SP group (P<0.05) at 6-mo follow-up. The number of diarrheal episodes per child tended to be lower in the HP vs. SP than in the SP group (1.9 vs 2.3, P=0.41). These results demonstrate that feeding an HP diet to the malnourished children during recovery from shigellosis enhanced linear growth with a modest reduction in diarrheal morbidity during the 6-mo follow-up period."

298 Katz JP, Lichtenstein GR. Rheumatologic manifestations of gastrointestinal diseases. Gastroenterol Clin North Am 1998 Sep;27(3):533-62. 188 ref, Eng. Division of Gastroenterology, Department of Medicine, Hospital of the University of Pennsylvania, University of Pennsylvania School of Medicine, 3-Ravdin Building, 3400 Spruce Street, Philadelphia, PA 19104-4283, USA

299 Kaur H, Lal M. Typing and antibiotic susceptibility patterns of *Vibrio cholerae* during six consecutive cholera seasons in North India. Trop Gastroenterol 1998 Apr-Jun;19(2):59-61. 11 ref, Eng. Department of Microbiology, Christian Medical College, Ludhiana 140008, Punjab, India

"A total of 10,427 diarrhoeal stool specimens were cultured for *Vibrio cholerae* between 1992 and 1997. The isolation rates were 2%, 2.6%, 6.7%, 7.08%, 0.9% and 2.6% in the years from 1992 to 1997 respectivelly. Till 1992, *Vibrio cholerae* O1 ogawa was the predominant strain. In 1993, 81.3% of the isolates were of 0139 Bengal strain and the rest were *V. cholerae* 01. From 1994 to 1997, *V. cholerae* 01 ogawa was the predominant strain and there were no isolation of 0139 strain. The predominant phage type in 1992 and 1993 were T2 and T27 thereafter. Most *Vibrio cholerae* strains were sensitive to tetracycline, gentamycin, netromycin, norfloxacin and furazolidine. Strains were resistant to cotrimaxozole till 1996, but were 100% sensitive in 1997. Strains were sensitive to chloramphenicol till 1993 but acquired resistance thereafter.

300 Khanduja V, Kang G, Rajan DP, Balasubramanian KA. Oxidative stress response in *Shigella* & nonpathogenic gut bacteria. Indian J Med Res 1998 Jul;108:3-7. 21 ref, Eng. Department of Gastrointestinal Sciences, The Wellcome Trust Research Laboratory, Christian Medical College & Hospital, Vellore 632004, India

"The effect of oxidative stress in the form of exogenous H_2O_2 on the survival of four species of *Shigella* and two nonpathogenic gram negative gut bacteria and the role of catalase as an antioxidant enzyme, neutralizing the effect of H_2O_2 were examined. A significant decrease in the number of colony forming units (CFUs) after exposure to exogenous H_2O_2 (122 ± 37, compared to control bacteria (218 ± 63, P<0.001) was observed. There was an induction of catalase activity after exposure to exogenous H_2O_2 and the specific activity of catalase in H_2O_2 exposed bacteria was significantly increased (2.88 ± 1.25), compared to control bacteria (1.5 ± 0.44; P<0.05). A direct correlation was observed between the decrease in bacterial counts and increase in catalase activity after exposure to H_2O_2 (regression coefficient (0.56). Gut bacteria appear to be susceptible to oxidative stress and inducible catalase activity may form an important part of the antioxidant defence mechanism against oxidative stress."

301 Kim LS, Hadley WK, Stansell J, Cello JP, Koch J. Declining prevalence of cryptosporidiosis in San Francisco (brief report). Clin Infect Dis 1998 Sep;27(3):655-6. 7 ref, Eng. San Francisco General Hospital, NH-3D7, 1001 Potrero Avenue, San Francisco, CA 94110, USA

302 Kimsey HH, Waldor MK. *Vibrio cholerae* hemagglutinin/protease inactivates CTXF. Infect Immun 1998 Sep;66(9):4025-9. 29 ref, Eng. New England Medical Center 041, 750 Washington St., Boston, MA 02111, USA

"Pathogenic strains of *Vibrio cholerae* are lysogens of the filamentous phage CTXf, which carries the genes for cholera toxin (*ctxAB*). We found that the titers of infective CTXf in culture supernatants of El Tor CTXf lysogens increased rapidly during exponential growth but dropped to undetectable levels late in stationary-phase growth. When CTXf transducing particles were mixed with stationary-phase culture supernatants of El Tor strains, CTXf infectivity was destroyed. Our data indicate that this growth phase-regulated factor, designated CDF (CTXf-destroying factor), is the secreted hemagglutinin/protease (HA/P) of *V. cholerae*. A strain containing a disrupted *hap* gene, which encodes HA/P of *V. cholerae*, did not produce CDF activity in culture supernatants. Introduction of the HA/P-expressing plasmid pCH2 restored CDF activity. Also, CDF activity in culture supernatants of a variety of pathogenic *V. cholerae* isolates varied widely but correlated with the levels of secreted HA/P, as measured by immunoblotting with anti-HA/P antibody. CDF was purified from *V. cholerae* culture supernatants and shown to contain a 45-kDa polypeptide which bound anti-HA/P antibodies and which comigrated with HA/P in sodium dodecyl sulfate-polyacrylamide gel electrophoresis. The production of high levels of secreted HA/P by certain *V. cholerae* strains may be a factor in preventing CTXf reinfection in natural environments and in the human host."

303 Kirkwood CD, Bishop RF, Coulson BS. Attachment and growth of human rotaviruses RV-3 and S12/85 in Caco-2 cells depend on VP4. J Virol 1998 Nov;72(11):9348-52. 40 ref, Eng. Department of Microbiology and Immunology, The University of Melbourne, Royal Parade, Parkville 3052, Victoria, Australia

"Studies with human neonatal rotaviruses RV-3 and S12/85 and their reassortants showed that VP4 is a determinant of rotavirus attachment to and growth in Caco-2 cells. The binding of these viruses to MA104 and Caco-2 cells correlated with their growth ability. Virus sensitivity to trypsin and the VP4 fusion region may be implicated in these processes."

304 Klose KE, Novik V, Mekalanos JJ. Identification of multiple σ^{-54}-dependent transcriptional activators in *Vibrio cholerae* (note). J Bacteriol 1998 Oct;180(19): 5256-9. 17 ref, Eng. Department of Microbiology, University of Texas Health Science Center, 7703 Floyd Curl Dr., San Antonio, TX 78284-7758, USA

305 Ko W-C, Wu H-M, Chang T-C, Yan J-J, Wu J-J. Inducible ß-lactam resistance in *Aeromonas hydrophila*: therapeutic challenge for antimicrobial therapy. J Clin Microbiol 1998 Nov;36(11):3188-92. 35 ref, Eng. Department of Medical Technology, National Cheng Kung University Medical College, No. 1 University Rd., Tainan, Taiwan

"Despite the abundant amount of knowledge about inducible chromosomally mediated ß-lactamases among *Aeromonas*

species, extended-spectrum ß-lactam-resistant *A. hydrophila* strains selected in clinical practice were rarely reported. In the present study, two strains of *A. hydrophila*, A136 and A139, with markedly different susceptibilities to extended-spectrum cephalosporins were isolated from blood and the tip segment of an arterial catheter of a burn patient. Another strain (A136m) was selected in vitro by culturing A136 in a subinhibitory concentration of cefotaxime, the ß-lactam agent administered for the treatment of *Aeromonas* bacteremia in this patient. Typing studies by arbitrarily primed PCR and pulsed-field gel electrophoresis indicated a clonal relationship among strains A136, A136m, and A139. These strains were identified to be of DNA hybridization group 1. Wild-type strain A136 was resistant only to ampicillin and cephamycins, but A136m and A139 were highly resistant to the expanded- and broad-spectrum cephalosporins. The presence of increased ß-lactamase activity in A139 suggests that A139 is a derepressed mutant which overexpresses ß-lactamases. These results call attention to the use of ß-lactam agents for the treatment of invasive *Aeromonas* infections."

306 Ko W-C, Chuang Y-C, Huang G-C, Hsu S-Y. Infections due to non-O1 *Vibrio cholerae* in southern Taiwan: predominance in cirrhotic patients. Clin Infect Dis 1998 Oct;27(4):774-80. 26 ref, Eng. Department of Internal Medicine, National Cheng Kung University Hospital, 138 Sheng Li Road, Tainan, Taiwan

"Although Taiwan is not an area where cholera is endemic, from October 1988 to October 1997 30 episodes of non-O1, non-O139 *Vibrio cholerae* infection were noted at the National Cheng Kung University Hospital in Taiwan. Infections generally occurred in hot seasons, and two episodes were concomitant with *Vibrio vulnificus* infection. Three major clinical presentations were found: bacteremia with concurrent spontaneous bacterial peritonitis or invasive soft-tissue infections that occurred solely in cirrhotic patients; self-limited acute febrile gastroenteritis that occurred in patients with no underlying medical disease; and necrotizing fasciitis or cellulitis that often resulted from a would on extremities. Other manifestations included fatal pneumonitis in a drowned man and acute pyosalpinx. The differential diagnosis of invasive infections in cirrhotic patients should include infections due to non-O1 *V. cholerae* or *V. vulnificus*, and a third-generation cephalosporin and a tetracycline analogue or a fluoroquinolone alone is recommended for treatment of severe vibrio infections."

307 Kresse AU, Schulze K, Deibel C, Ebel F, Rohde M, Chakraborty T, Guzman CA. Pas, a novel protein required for protein secretion and attaching and effacing activities of enterohemorrhagic *Escherichia coli*. J Bacteriol 1998 Sep;180(17): 4370-9. 64 ref, Eng. Division of Microbiology, GBF-National Research Centre for Biotechnology, Mascheroder Weg 1, D-38124 Braunschweig, Germany

**308 Leder K, Ryan N, Spelman D, Crowe SM*. Microsporidial disease in HIV-infected patients: a report of 42 patients and review of the literature. Scand J Infect Dis 1998;30(4):331-8. 52 ref, Eng. ***AIDS Pathogenesis Research Unit, MacFarlane Burnet Centre for Medical Research, Yarra Bend Road (PO Box 254), Fairfield Vic 3078, Australia

309 Legros D, Ochola D, Lwanga N, Guma G. Antibiotic sensitivity of endemic *Shigella* in Mbarara, Uganda. East Afr Med J 1998 Mar;75(3):160-1. 5 ref, Eng. Epicentre, PO Box 2362, Kampala, Uganda

"We analysed the chimio-sensitivity to antibiotics of endemic strains of *Shigella* isolated in Mbarara district, southwest Uganda. Twenty four strains were isolated, of which none was sensitive to cotrimoxazole and eight (33.4%, 95% CI [15.6-55.3]) to ampicillin, the two antibiotics recommended to treat dysentery during non epidemic periods in Uganda. Two isolates were resistant to nalidixic acid and none was resistant to the fluoroquinolones (Ciprofloxacin®, Norfloxacin®). It is concluded that the results of this survey could be used to facilitate the elaboration of a new treatment protocol to treat endemic dysentery cases in Uganda."

310 Louie M, Read S, Simor AE, Holland J, Louie L, Ziebell K, Brunton J, Hii J. Application of multiplex PCR for detection of non-O157 verocytotoxin-producing *Escherichia coli* in bloody stools: identification of serogroups O26 and O111 (note). J Clin Microbiol 1998 Nov;36(11):3375-7. 27 ref, Eng. Department of Microbiology, Sunnybrook Health Science Centre, 2075 Bayview Ave., Room B130, Toronto, Ontario, Canada M4N 3M5

311 Ludwig K, Petric M, Blanchette V, Karmali M. Isolated thrombocytopenic purpura associated with infection due to verocytotoxin (Shiga toxin)—producing *Escherichia coli* serotype O26:H11 (brief report). Clin Infect Dis 1998 Sep;27(3):660-1. 10 ref, Eng. Division of Microbiology, The Hospital for Sick Children, 555 University Avenue, Toronto, Canada M5G 1X8

312 McCormick BA, Siber AM, Maurelli AT. Requirement of the *Shigella flexneri* virulence plasmid in the ability to induce trafficking of neutrophils across polarized monolayers of the intestinal epithelium. Infect Immun 1998 Sep;66(9):4237-43. 58 ref, Eng. Department of Pediatric Gastroenterology and Nutrition, Massachusetts General Hospital—East, Charlestown Navy Yard Bldg. 149 (1493404), Charlestown, MA 02129, USA

"Attachment of an array of enteric pathogens to epithelial surfaces is accompanied by recruitment of polymorphonuclear leukocytes (PMN) across the intestinal epithelium. In this report, we examine how *Shigella*-intestinal epithelium interactions evoke the mucosal inflammatory response. We modeled these interactions in vitro by using polarized monolayers of the human intestinal epithelial cell line, T84, isolated human PMNs, and *Shigella flexneri*. We show that *Shigella* attachment to T84-cell basolateral membranes was a necessary component in the

signaling cascade for induction of basolateral-to-apical directed transepithelial PMN migration, the direction of PMN transepithelial migration in vivo. In contrast, attachment of *Shigella* to the T84-cell apical membrane failed to stimulate a directed PMN transepithelial migration response. Importantly, the ability of *Shigella* to induce PMN migration across epithelial monolayers was dependent on the presence of the 220-kb virulence plasmid. Moreover, examination of *Shigella* genes necessary to signal subepithelial neutrophils established the requirement of a functional type III secretion system. Our results indicate that the ability of *Shigella* to elicit transepithelial signaling to neutrophils from the basolateral membrane of epithelial cells represents a mechanism involved in *Shigella*-elicited enteritis in humans."

313 McKillip JL, Jaykus L-A, Drake M. rRNA stability in heat-killed and UV-irradiated enterotoxigenic *Staphylococcus aureus* and *Escherichia coli* O157:H7. Appl Environ Microbiol 1998 Nov;64(11):4264-8. 21 ref, Eng. Department of Food Science and Technology, Southeast Dairy Foods Research Center, Box 9805, Mississippi State University, Mississippi State, MS 39762-9805, USA

314 Manabe YC, Clark DP, Moore RD, Lumadue JA, Dahlman HR, Belitsos PC, Chaisson RE, Sears CL. Cryptosporidiosis in patients with AIDS: correlates of disease and survival. Clin Infect Dis 1998 Sep;27(3):536-42. 34 ref, Eng. Division of Infectious Diseases, Department of Medicine, Ross Research Building, Room 933, 720 Rutland Avenue, Baltimore, Maryland 21205, USA

315 Mascolo R, Saltzman JR. Lactose intolerance and irritable bowel syndrome. Nut Rev 1998 Oct;56(10):306-8. 15 ref, Eng. Division of Digestive Disease and Nutrition, University of Massachusetts Memorial Health Care, 55 Lake Avenue North, Worcester, MA 01655, USA

316 Mead JR, You X. Susceptibility differences to *Cryptosporidium parvum* infection in two strains of gamma interferon knockout mice. J Parasitol 1998 Oct;84(5)1045-8. 23 ref, Eng. Department of Pediatrics, Emory University School of Medicine, Atlanta, Georgia 30322, USA

317 Medema GJ, Schets FM, Teunis PFM, Havelaar AH. Sedimentation of free and attached *Cryptosporidium* oocysts and *Giardia* cysts in water. Appl Environ Microbiol 1998 Nov;64(11):4460-6. 13 ref, Eng. Department of Chemistry and Biology, Kiwa Research and Consultancy, PO Box 1072, 3430 BB Nieuwegein, The Netherlands

318 Moss DM, Chappell CL, Okhuysen PC, DuPont HL, Arrowood MJ, Hightower AW, Lammie PJ*. The antibody response to 27-, 17-, and 15-kDa *Cryptosporidium* antigens following experimental infection in humans. J Infect Dis 1998 Sep;178(3):827-33. 19 ref, Eng. *Division of Parasitic Diseases, MS F-13, Centers for Disease Control and Prevention, 4770 Buford Highway, Atlanta, GA 30341, USA

319 Moxley RA, Berberov EM, Francis DH, Xing J, Moayeri M, Welch RA, Baker DR, Barletta RG. Pathogenicity of an enterotoxigenic *Escherichia coli* hemolysin (*hlyA*) mutant in gnotobiotic piglets (note). Infect Immun 1998 Oct;66(10):5031-5. 45 ref, Eng. Department of Internal Medicine, Division of Rheumatology, University of Michigan Medical Center, Ann Arbor, MI 48109, USA

320 Murphy MS. Guidelines for managing acute gastroenteritis based on a systematic review of published research. Arch Dis Child 1998 Sep;79(3):279-84. 70 ref, Eng. Institute of Child Health, Clinical Research Building, Whittall Street, Birmingham B4 6NH, UK

"This paper is intended to provide evidence-based recommendations about the assessment and clinical management of infants and children with acute gastroenteritis. These guidelines were derived from a systematic review of published research. The diagnosis of gastroenteritis is not addressed; this is often presumptive and is based on a history of acute diarrhoea in the absence of other likely explanations. Microbiological investigation is not necessary in every case, but may be important in patients who require admission to hospital, in those who have bloody or mucoid diarrhoea suggesting colitis, in high risk patients such as those with an immune deficiency, and in cases where there is diagnostic uncertainty. Clinicians should apply general medical knowledge and clinical judgment in using these guidelines."

321 Murray J, McFarland DA, Waldman RJ. Cost-effectiveness of oral cholera vaccine in a stable refugee population at risk for epidemic cholera and in a population with endemic cholera. Bull WHO 1998;76(4):343-52. 52 ref, Eng. BASICS, 1600 Wilson Boulevard, Arlington, VA 22209, USA

"Recent large epidemics of cholera with high incidence and associated mortality among refugees have raised the question of whether oral cholera vaccines should be considered as an additional preventive measure in high-risk populations. The potential impact of oral cholera vaccines on populations prone to seasonal endemic cholera has also been questioned. This article reviews the potential cost-effectiveness of B-subunit, killed whole-cell (BS-WC) oral cholera vaccine in a stable refugee population and in a population with endemic cholera. In the population at risk for endemic cholera, mass vaccination with BS-WC vaccine is the least cost-effective intervention compared with the provision of safe drinking-water and sanitation or with treatment of the disease. In a refugee population at risk for epidemic disease, the cost-effectiveness of vaccination is similar to that of providing safe drinking-water and sanitation alone, though less cost-effective than treatment alone or treatment combined with the provision of water and sanitation. The implications of these data for public health decision-makers and programme

managers are discussed. There is a need for better information on the feasibility and costs of administering oral cholera vaccine in refugee populations and populations with endemic cholera."

322 Nakasone N, Iwanaga M. Characterization of outer membrane protein OmpU of *Vibrio cholerae* O1. Infect Immun 1998 Oct;66(10):4726-8. 16 ref, Eng. Department of Bacteriology, Faculty of Medicine, University of the Ryukyus, Nishihara, Okinawa 903-0215, Japan

"The outer membrane protein OmpU of *Vibrio cholerae* O1 strain 86B3 was characterized with reference to colonization of the intestine by the organism. The purified OmpU exhibited a pl of 3.6. Upon sodium dodecyl sulfate-polyacrylamide gel electrophoresis, it migrated to 38, 32, and 110 kDa when the sample was heated at 100^0C for 2 min, 50^0C for 15 min, and room temperature for 30 min, respectively. The purified OmpU was not hemagglutinative. Anti-OmpU serum did not agglutinate strain 86B3 or other *V. cholerae* organisms. OmpU adhered to the brush border of the rabbit small intestine; adhesion of the organisms to the intestine treated in advance with OmpU was not inhibited. Treating the organisms in advance with anti-OmpU Fab did not inhibit adhesion to the intestine. These results obtained in vitro suggest that OmpU is not involved in the adhesion of *V. cholerae* to the intestinal epithelium."

323 Nchito M, Kelly P, Sianongo S, Luo NP, Feldman R, Farthing M, Baboo KS. Cryptosporidiosis in urban Zambian children: an analysis of risk factors. Am J Trop Med Hyg 1998 Sep;59(3):435-7. 15 ref, Eng. Department of Pathology, University Teaching Hospital, PO Box 50110, Lusaka, Zambia

"In four crowded townships of Lusaka, Zambia, the prevalence of cryptosporidiosis in 222 children with diarrhea was 18%, with marked temporal and geographic variation over the course of one rainy season. Using data on the finding of oocysts of *Cryptosporidium parvum* in urban water supplies, the areas under study were categorized as high or low risk. Prevalence of cryptosporidiosis in children with diarrhea was higher in high risk areas after stratification by early/late stage of the rains (Mantel-Haenszel odds ratio [OR] = 2.9, 95% confidence interval [CI] = 1.3, 6.7; *P*=0.008). Cryptosporidiosis was not associated with keeping animals, nutritional status, or parental education, but was apparently more common in breast fed children (OR = 2.7, 95% CI = 1.1, 6.9; *P*=0.01), although the proportion of exclusively breast fed children was not measured. Since most of these infections were of short duration, we conclude that transmission of *C. parvum* can vary dynamically within one city and over short periods of time, and that water-borne contamination may be a substantial influence."

324 Ogutu EO, Kanja C, Kang'ethe SK, Nyong'o A. Prevalence of intestinal parasites in Kenyans with dyspepsia. East Afr Med J 1998 Jan;75(1):16-8. 9 ref, Eng. Department of Medicine, College of Health Sciences, University of Nairobi, PO Box 19676, Nairobi, Kenya

325 Omokhodion FO, Oyemade A, Sridhar MKC, Olaseha IO, Olawuyi JF. Diarrhoea in children of Nigerian market women: prevalence, knowledge of causes, and management. J Diarrhoeal Dis Res 1998 Sep;16(3):194-200. 9 ref, Eng. Department of Preventive and Social Medicine, University College Hospital, Ibadan, Nigeria

"A cross-sectional survey was carried out among mothers of children aged less than five years in two markets in Ibadan, one with poor environmental sanitation and the other clean and well maintained. The study took place between September 1996 and March 1997. The questionnaire used for this survey sought information about the occurrence of diarrhoea among children aged less than five years, their mothers' knowledge about the management of diarrhoea and their practices, including care-seeking practices, and the use of oral rehydration solutions. Two hundred and sixty-six mothers were interviewed in the first market (Bodija) and 260 in the other (Gbagi). Thirty-seven percent of the children in the cleaner market (Gbagi) were said to have had diarrhoea within the last 3 months compared to 33% of the children in the unhygienic Bodija market ($p>0.05$). These results suggest that environmental sanitation may not be a major determinant of diarrhoea among children of the two groups of market women. When their children had diarrhoea, 44% (Bodija) and 40% (Gbagi) of the mothers attended health centres, 33% (Bodija) and 32% (Gbagi) gave ORT at home, and 12% (Bodija) and 19% (Gbagi) purchased drugs at a chemist. The study further showed that, while only one-third of all respondents resorted to home-treatment of diarrhoea with ORS, more than 80% of them knew the components and composition of ORS solution. There is a need to continue to encourage mothers to use ORS and, thus, bridge the knowledge-practice gap in mothers' management of diarrhoea at home."

326 O'Ryan ML, Vial PA, Mamani N, Jiang X, Estes MK, Ferrecio C, Lakkis H, Matson DO. Seroprevalence of Norwalk virus and Mexico virus in Chilean individuals: assessment of independent risk factors for antibody acquisition. Clin Infect Dis 1998 Oct;27(4):789-95. 26 ref, Eng. Department of Microbiology, University of Chile, Condell 303, Santiago, Chile

"Norwalk virus (NV) and Mexico (MX) virus represent distinct genetic clusters within the same genus of human caliciviruses (CVs), a major cause of diarrhea in adults. The magnitude and potential risk factors of human CV infection in populations from Santiago and Punta Arenas, Chile, were assessed. Individuals (*n*=1,864) gave a blood sample and answered a questionnaire during a household survey. Sera were tested for antibody to NV and MX virus with use of recombinant capsid antigens. Overall, NV and MX virus seroprevalence rates were 83% and 91% in Santiago vs. 67% and 90% in Punta Arenas, respectively (*P*<.001 for NV virus). Lower socioeconomic status and increasing age were risk factors for infection with both viruses (*P*<.001). Consumption of seafood, consumption of vegetables, and

child care center attendance were population risk factors for infection, but the association of a factor with a virus depended on the city. Prevention of human CV infections will require individual assessment in different communities."

327 Peterson EA, Roberts L, Toole MJ, Peterson DE. The effect of soap distribution on diarrhoea: Nyamithuthu refugee camp. Int J Epidemiol 1998 Jun;27(3):520-4. 13 ref, Eng. Public Health Consultant, 11211 Sorrel Ridge Lane, Oakton, VA 22124, USA

"**Background**: In January 1993, Nyamithuthu Camp in Malawi housed 64 000 Mozambican refugees. Communicable diseases, primarily diarrhoea, pneumonia, malaria and measles, contribute to substantially higher mortality rates in refugee populations compared to similar non-displaced populations. **Method**: A systematic sample of 402 households in one portion of the camp were surveyed for diarrhoeal risk factors, and then interviewed twice weekly for 4 months regarding new diarrhoea episodes and the presence of soap in the household. Two-hundred grams of soap per person was distributed monthly. **Results**: Households had soap on average only 38% of the interview days. Soap was used primarily for bathing and washing clothes (86%). Although 81% of mothers reported washing their children's hands, only 28% of those mothers used soap for that purpose. The presence of soap in a household showed a significant protective effect: there were 27% less episodes of diarrhoea in households when soap was present compared to when no soap was present (RR=0.73, 95% CI: 0.54< RR<0.98). Potential confounding factors were assessed and did not appear to be responsible for the association between the presence of soap and reductions in diarrhoea incidence. **Conclusion**: In summary, our findings suggest that the provision of regular and adequate soap rations, even in the absence of a behaviour modificiation or education programme, can play an important role in reducing diarrhoea in refugee populations. If subsequent study confirms the soap as a cheap and effective measure to reduce diarrhoea, its provision in adequate amounts should be a high priority in refugee settings."

328 Pierard D, Muyldermans G, Moriau L, Stevens D, Lauwers S. Identification of new verocytotoxin type 2 variant B-subunit genes in human and animal *Escherichia coli* isolates. J Clin Microbiol 1998 Nov;36(11):3317-22. 39 ref, Eng. Department of Microbiology, AZ-VUB, Laarbeeklaan 101, B-1090 Brussels, Belgium

"The sequence of a verocytotoxin 2 (VT2) variant gene that was untypeable by the B subunit PCR and restriction fragment length polymorphism analysis (PCR-RFLP) method described by Tyler *et al*. (S.D. Tyler, W.M. Johnson, H. Lior, G. Wang, and K.R. Rozee, J. Clin. Microbiol. 29:1339-1343, 1991) was determined and compared with published sequences. It was highly homologous to two recently reported VT2 variant sequences. The PCR-RFLP method described by Tyler *et al*. was extended to include these new sequences. New VT2 variants were identified in 65 of 359 VT-producing *Escherichia coli* (VTEC) with newly designed primers (VT2-cm and VT2-f) and were characterized as well by restriction analysis of the amplification products obtained with another VT2-specific primer pair (VT2-e and VT2-f). The VT genes harbored by 64 of these isolates proved to be untypeable by Tyler's PCR-RFLP method because no amplification was obtained with the primers used with this method (VT2-c and VT2-d). The last isolate harbored the new variant gene in addition to VT2vh-a. None of the isolates harboring these new toxin genes belonged to serogroups O157, O26, O103, O111, and O145. All 65 isolates were negative for the *eaeA* gene and were significantly less frequently enterohemolytic or positive for the enterohemorrhagic *E. coli* (EHEC) virulence plasmid than non-O157 VTEC isolates harboring other VT2 genes. They were also less frequently isolated from patients with EHEC-associated symptoms. The extended PCR-RFLP typing method is a useful tool to identify less-virulent VTEC isolates and for VT genotyping in epidemiological studies with non-O157 strains."

329 Rahman MM, Mahalanabis D, Sarker SA, Bardhan PK, Alvarez JO, Hildebrand P, Beglinger C, Gyr K. *Helicobacter pylori* colonization in infants and young children is not necessarily associated with diarrhoea. J Trop Pediatr 1998 Oct;44(5):283-7. 20 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"A cohort of 151 infants and young children aged 1-23 months from a poor peri-urban community of Bangladesh was studied to determine the relationship between *Helicobacter pylori* colonization and morbidity due to diarrhoea. A ^{13}C urea breath test was performed to detect the presence of *H. pylori*. Children were followed up at home every alternate day for 6 months and diarrhoeal morbidity data were collected. Diarrhoeal morbidity was compared between *H. pylori*-positive and *H. pylori*-negative children. Sixty-eight (45 per cent) children were *H. pylori* positive and 83 (55 per cent) were *H. pylori* negative. During the first 1-month period following the breath test, three (4.4 per cent) *H. pylori*-positive and four (4.8 per cent) *H. pylori*-negative children had diarrhoea. Thirty-two (47 per cent) of the children in the positive group and 43 (52 per cent) in the negative group had one or more episodes of diarrhoea during the 6-month follow-up period. Median number of diarrhoeal episodes was 1.0 (range 1.0-4.0) in the *H. pylori*-positive children and 2.0 (range 1.0-5.0) in the *H. pylori*-negative children (p=0.19). No significant difference was observed in the cumulative days with diarrhoea. The results of this study suggest that *H. pylori* colonization is not associated with diarrhoeal morbidity in infants and young children."

330 Rautanen T, Halme S, Vesikari T. Community-based survey of paediatric diarrhoeal morbidity and home treatment practices in Finland. Acta Paediatr 1998 Sep; 87(9):986-90. 26 ref, Eng. Department of Paediatrics, Jorvi Hospital, Turuntie 150, FIN-02740 Espoo, Finland

"To determine total diarrhoeal morbidity and current home case management practices, a recall study was conducted among 345 mothers whose 406 children under 5 y of age had had diarrhoea in the past 4 months. The seasonally adjusted annual diarrhoea incidence rate was 0.48 episodes/child/y. For home treatment, increased amounts of fluid were given in 93% and oral rehydration solution (ORS) in 37% of cases. However, when given, ORS was diluted with other fluids in 41% of cases. More than half (55%) of the children received increased or normal amounts of food during the diarrhoeal episode, but 7% of the children were kept fasting for at least 1 d. Use of antidiarrhoeal drugs was minimal (0.7%), but products containing lactic acid bacteria were given in 44% of cases. Case management practice in cases of diarrhoea at home have much improved during the last 20 y, but are still not optimal."

331 Reilly A. Prevention and control of enterohaemorrhagic *Escherichia coli* (EHEC) infections: memorandum from a WHO meeting. Bull WHO 1998;76(3):245-55. 17 ref, Eng. World Health Organization, 1211 Geneva 27, Switzerland

"*Escherichia coli* is a commonly occurring inhabitant of the intestine of humans and other animals, but there are several pathogenic types of *E. coli* which cause a variety of human diseases. One of these pathogenic types *E. coli* O157:H7, belongs to the group of enterohaemorrhagic *E. coli* (EHEC) which produce potent toxins and cause a particularly severe form of disease, haemorrhagic colitis (HC). About 10% of patients with HC can go on to develop haemolytic uraemic syndrome (HUS), a life-threatening complication of *E. coli* O157:H7 infection that is characterized by acute renal failure, haemolytic anaemia, and thrombocytopenia. These sequelae are particularly serious in young children and older people. On average, 2-7% of patients with HUS die, but in some outbreaks among the elderly the mortality rate has been as high as 50%. This memorandum reviews the growing importance of *E. coli* O157:H7 as a foodborne pathogen and reports on the issues of surveillance, outbreak investigation, and control strategies with respect to EHEC infections that were discussed at the WHO Consultation on Prevention and Control of EHEC infections, held in Geneva on 28 April to 1 May 1997. Recommended measures for prevention and control include the following: use of potable water in food production; presentation of clean animals at slaughter; improved hygiene throughout the slaughter process; appropriate use of food processing measures; thorough cooking of foods; and the education of food handlers, abattoir workers, and farm workers on the principles and application of food hygiene."

332 Rocha MFG, Soares AM, Flores CA, Steiner TS, Lyerly DM, Guerrant RL, Ribeiro RA, Lima AAM. Intestinal secretory factor released by macrophages stimulated with *Clostridium difficile* toxin A: role of interleukin 1b**. Infect Immun 1998 Oct;66(10):4910-6. 52 ref, Eng.** Clinical Research Unit, Federal University of Ceara, PO Box 3229, CEP 60 436-160, Fortaleza, CE, Brazil

333 Root GPM. Evidence of malnutrition having a threshold effect on the risk of childhood diarrhoea in Zimbabwe. Cent Afr J Med 1997 Jul;43(7):185-8. 13 ref, Eng. Population Studies Unit, Department of Geography, University of Liverpool, Roxby Building, L69 3BX, UK

"**Objective:** Diarrhoeal disease is a major cause of childhood morbidity and mortality in Zimbabwe. This paper examines the relationship between diarrhoea and malnutrition to determine whether there is a threshold effect in operation. **Design:** Multivariate analysis of a retrospective survey. **Setting/Subjects:** Using a nationally representative sample, the Zimbabwe Demographic and Health Survey, 1994, collected diarrhoeal, anthropometric and socio-economic data for 2,073 children aged 0 to 35 months. **Results:** Age of the child, residence, and severe stunting and wasting were found to be significant predictors of childhood diarrhoea. However, moderate stunting and wasting failed to show any relationship. **Conclusions:** Moderate malnutrition is the main nutritional complaint in Zimbabwe. Thus malnutrition may have a relatively small role in determining the prevalence of childhood diarrhoea. Instead, factors related to exposure - namely sanitation, water supply, population density and hygiene - may be more important. Consequently supplementary feeding programmes should work in tandem with initiatives to reduce exposure if the morbidity burden of children is to be reduced."

334 Roze C, Varannes SBD, Shi G, Geneve J, Galmiche JP*. Inhibition of prostaglandin-induced intestinal secretion by igmesine in healthy volunteers. Gastroenterology 1998 Sep;115(3):591-6. 34 ref, Eng. *Centre Hospitalier Universitaire, Hotel-Dieu 44035, Nantes, Cedex, France

335 Samadi AR, Islam R, Huq MI. Replacement of intravenous therapy by oral rehydration solution in a large treatment centre for diarrhoea with dehydration. Bull WHO 1998;76(4):319-24 (reprinted—the article first appeared in: Bull WHO 1983;61:471-6). 21 ref, Eng. International Centre for Diarrhoeal Disease Research, Bangladesh, GPO Box 128, Dhaka 1000, Bangladesh

"Intravenous fluid therapy is particularly effective in reversing severe dehydration due to diarrhoea, but it can be replaced by oral rehydration, with the advantages of lower risks and costs, in the treatment of mild, moderate and some severe cases of dehydration. In this study, the efficacy of oral rehydration solution was compared with that of intravenous fluid in the treatment of moderate and some severe cases of dehydration due to diarrhoea in a treatment centre in Bangladesh during a period of 5 months in 1980 and the same months in 1981. The results of this study show that rehydration by oral rehydration solution (ORS) is as effective as intravenous fluid therapy. ORS can be used for the treatment of diarrhoeas in all age groups including young infants. The costs were reduced by 33% despite an increase in the duration of stay of some patients."

[Please see O Fontaine's commentary on "Oral rehydration solution" published in the same issue at page 325]

336 Sarkar MAK, Shakur MS, Doherty C, Price V, Alam HSK. The use of anti-microbials: in the treatment of diarrhoea and the clinical distinction between invasive and non-invasive disease—an audit of current practice at Dhaka Shishu Hospital. Dhaka Shishu (Child) Hosp J 1997 Dec;13(1-2):18-25. 21 ref, Eng. Bangladesh Institute of Child Health, Dhaka Shishu Hospital, Dhaka, Bangladesh

337 Silva A, Fando R, Benitez JA*. Overexpression of a mutant B subunit in toxigenic *Vibrio cholerae* diminishes production of active cholera toxin in vivo. Curr Microbiol 1998 Oct;37(4):231-5. 22 ref, Eng. *Centro Nacional de Investigaciones Cientificas, PO Box 6990, La Habana, Cuba

338 Siu YK, Ng PC*, Fung SCK, Lee CH, Wong MY, Fok TF, So KW, Cheung KL, Wong W, Cheng AFB. Double blind, randomised, placebo controlled study of oral vancomycin in prevention of necrotising enterocolitis in preterm, very low birthweight infants. Arch Dis Child Fetal Neonatal Ed 1998 Sep;79(2):F105-9. 23 ref, Eng. *Department of Paediatrics, Level 6, Clinical Science Building, Prince of Wales Hospital, Shatin, New Territories, Hong Kong, People's Republic of China

339 Smith NH, Cron S, Valdez LM, Chappell CL, White AC*, Jr. Combination drug therapy for cryptosporidiosis in AIDS. J Infect Dis 1998 Sep;178(3):900-3. 15 ref, Eng. *Department of Medicine, Baylor College of Medicine, One Baylor Plaza, 561E, Houston, TX 77030, USA

340 Sobel J, Mahon B, Mendoza CE, Passaro D, Cano F, Baier K, Racioppi F, Hutwagner L, Mintz E. Reduction of fecal contamination of street-vended beverages in Guatemala by a simple system for water purification and storage, handwashing, and beverage storage. Am J Trop Med Hyg 1998 Sep;59(3):380-7. 20 ref, Eng. Foodborne and Diarrheal Diseases Branch, Centers for Disease Control and Prevention, 1600 Clifton Road, Mailstop A-38, Atlanta, GA 30333, USA

"Street-vended foods and beverages, an integral part of urban economics in the developing world, have been implicated in cholera transmission in Latin America. To improve the microbiologic quality of market-vended beverages in Guatemala, we tested a simple system consisting of dilute bleach (4.95% free available chlorine) for water purification, narrow-mouth plastic vessels with spigots for disinfecting and storing water and for preparing and storing beverages, handwashing soap, and education in using the system. We conducted a randomized controlled intervention trial among 41 vendors who received the intervention and 42 control vendors, comparing total and fecal coliform bacteria and *Escherichia coli* contamination of market-vended beverages, stored water, and vendors' hands. Samples were obtained at baseline and at each of six weekly follow-up visits. At baseline, fecal coliform bacteria were found in 40 (48%) market-vended beverages and *E. coli* in 14 (17%). When compared with samples from control vendors, a significant decrease in total coliform (P<0.001) and fecal coliform (P<0.001) bacteria in samples of stored water and beverages sold by intervention vendors was observed over the course of the study. The vessel system was well accepted by vendors. This simple inexpensive system consisting of hypochlorite disinfectant, plastic vessels, soap, and education can significantly reduce fecal contamination of market-vended beverages."

341 Sood A, Midha V, Sidhu SS. A case of enteric fever with unusual manifestations. Trop Gastroenterol 1998 Sep;19(3):114. 9 ref, Eng. Department of Medicine, Dayanand Medical College and Hospital, Ludhiana 141001, India

342 Sood S, Kapil A, Das BK, Seth P. The survival strategy of *Vibrio cholerae*. Trop Gastroenterol 1998 Jul-Sep;19(3):96-7. 17 ref, Eng. Department of Microbiology, All India Institute of Medical Sciences, Ansari Nagar, New Delhi 110029, India

343 Spano F, Putignani L, Crisanti A, Sallicandro P, Morgan UM, Blancq SML, Tchack L, Tzipori S, Widmer G*. Multilocus genotypic analysis of *Cryptosporidium parvum* isolates from different hosts and geographical origins. J Clin Microbiol 1998 Nov;36(11): 3255-9. 24 ref, Eng. *Division of Infectious Diseases, Tufts University School of Veterinary Medicine, 200 Westboro, Rd., North Grafton, MA 01536, USA

344 Svenungsson B, Capraru T, Evengardi B, Larsson R, Lebbad M. Intestinal microsporidiosis in a HIV-seronegative patient. Scand J Infect Dis 1998;30(3):314-6. 16 ref, Eng. Department of Infectious Diseases, Huddinge University Hospital, S-141 86 Huddinge, Sweden

345 Turunen UM, Farkkila MA, Hakala K, Seppala K, Sivonen A, Ogren M, Vuoristo M, Valtonen VV, Miettinen TA. Long-term treatment of ulcerative colitis with ciprofloxacin: a prospective, double-blind, placebo-controlled study. Gastroenterology 1998 Nov;115(5): 1072-8. 34 ref, Eng. Maria Hospital, Sisatautien Poliklinikka, Lapinlahdenkatu 16, 00180 Helsinki, Finland

346 Turvill JL, Mourad FH, Farthing MJG. Crucial role for 5-HT in cholera toxin but not *Escherichia coli* heat-labile enterotoxin-intestinal secretion in rats. Gastroenterology 1998 Oct;115(4):883-90. 49 ref, Eng. Digestive Diseases Research Centre, St. Bartholomew's and The Royal London School of Medicine and Dentistry, Turner Street, London E1 2AD, England, UK

"**Background & Aims***:* Many consider cholera toxin (CT) and *Escherichia coli* heat-labile enterotoxin (LT) to be functionally identical. Both increase intracellular adenosine 3', 5'-cyclic monophosphate concentration; however, differences between the two and the severity of the diseases they cause have been reported. The secretagogue 5-hydroxytryptamine (5-HT) is implicated in CT-induced

secretion, but its role in LT-induced secretion is unclear. We tested the hypothesis that LT fails to recruit 5-HT in its secretory processes. **Methods:** In vivo small intestinal perfusions were undertaken in adult male Wistar rats after incubation with equipotent doses of CT or LT, or saline. Small intestinal 5-HT release and the effect on net small intestinal water and electrolyte transport of (1) pharmacological depletion of 5-HT; (2) blockade of 5-HT type 2, 3, and 4 receptors; and (3) pretreatment with lidocaine, hexamethonium, and atropine were determined. **Results:** CT-but not LT-induced secretion was accompanied by 5-HT release, reduced by 5-HT depletion, and inhibited by each 5-HT antagonist. By contrast, lidocaine and hexamethonium inhibited secretion induced by both toxins. **Conclusions:** LT induces secretion without recruiting a 5-HT-dependent cascade. This may account for differences in clinical severity of the diseases CT and LT cause and has implications for the development of antisecretory therapies."

347 Verweij WR, de Haan L, Holtrop M, Agsteribbe E, Brands R, van Scharrenburg GJM, Wilschut J. Mucosal immunoadjuvant activity of recombinant *Escherichia coli* heat-labile enterotoxin and its B subunit: induction of systemic IgG and secretory IgA responses in mice by intranasal immunization with influenza virus surface antigen. Vaccine 1998 Dec;16(20):2069-76. 50 ref, Eng. Department of Physiological Chemistry, Groningen-Utrecht Institute for Drug Exploration (GUIDE), University of Groningen, Ant. Deusinglaan 1, 9713 AV Groningen, The Netherlands

"The *Escherichia coli* heat-labile enterotoxin (LT) is a very potent mucosal immunogen. LT also has strong adjuvant activity towards coadministered unrelated antigens and is therefore of potential interest for development of mucosal vaccines. However, despite the great demand for such mucosal vaccines, the use of LT holotoxin as an adjuvant is essentially precluded by its toxicity. LT is composed of an A subunit, carrying the toxic ADP-ribosylation activity, and a pentamer of identical B subunits, which mediates binding to ganglioside G_{M1}, the cellular receptor for the toxin. In this paper, we demonstrate that recombinant enzymatically inactive variants of LT, including the LTB pentamer by itself, retain the immunoadjuvant activity of LT holotoxin in a murine influenza model. Mice were immunized intranasally (i.n.) with influenza virus subunit antigen, consisting mostly of the isolated surface glycoprotein hemagglutinin (HA), supplemented with either recombinant LTB (rLTB), a nontoxic LT mutant (E112K, with a Glu112®Lys substitution in the A subunit), or LT holotoxin, and the induction of systemic IgG and local S-IgA responses was evaluated by direct enzyme-linked immunosorbent assay (ELISA). Immunization with subunit antigen alone resulted in a poor systemic IgG response and no detectable S-IgA. However, supplementation of the antigen with E112K or rLTB resulted in a substantial stimulation of the serum IgG level and in induction of a strong S-IgA response in the nasal cavity. The adjuvant activity of E112K or rLTB under these conditions was essentially the same as that of the LT holotoxin. The present results demonstrate that nontoxic variants of LT, rLTB in particular, represent promising immunoadjuvants for potential application in an i.n. influenza virus subunit vaccine. Nontoxic LT variants may also be used in i.n. vaccine formulations directed against other mucosal pathogens. In this respect, it is of interest that LT<B)-stimulated antibody responses after i.n. immunization were also observed at distant mucosal sites, including the urogenital system. This, in principle, opens the possibility to develop i.n. vaccines against sexually transmitted infectious diseases."

348 Wai SN, Mizunoe Y, Takade A, Kawabata S-I, Yoshida S-I. *Vibrio cholerae* O1 strain TSI-4 produces the exopolysaccharide materials that determine colony morphology, stress resistance, and biofilm formation. Appl Environ Microbiol 1998 Oct;64(10):3648-55. 42 ref, Eng. Department of Bacteriology, Faculty of Medicine, Kyushu University, Fukuoka 812-8582, Japan

"*Vibrio cholerae* O1 strain TSI-4 (El Tor, Ogawa) can shift to a rugose colony morphology from its normal translucent colony morphology in response to nutrient starvation. We have investigated differences between the rugose and translucent forms of *V. cholerae* O1 strain TSI-4. Electron microscopic examination of the rugose form of TSI-4 (TSI-4/R) revealed thick, electron-dense exopolysaccharide materials surrounding polycationic ferritin-stained cells, while the ferritin-stained material was absent around the translucent form of TSI-4 (TSI-4/T). The exopolysaccharide produced by *V. cholerae* TSI-4/R was found to have a composition of *N*-acetyl-D-glucosamine, D-mannose, 6-deoxy-D-galactose, and D-galactose (7.4:10.2:2.4:3.0). The expression of an amorphous exopolysaccharide promotes biofilm development under static culture conditions. Biofilm formation by the rugose strain was determined by scanning electron microscopy, and most of the surface of the film was colonized by actively dividing rod cells. The corresponding rugose and translucent strains were compared for stress resistance. By having exopolysaccharide materials, the rugose strains acquired resistance to osmotic and oxidative stress. Our data indicated that an exopolysaccharide material on the surface of the rugose strain promoted biofilm formation and resistance to the effects of two stressing agents."

349 Wanke CA, Gerrior J, Blais V, Mayer H, Acheson D. Successful treatment of diarrheal disease associated with enteroaggregative *Escherichia coli* in adults infected with human immunodeficiency virus. J Infect Dis 1998 Nov;178(5):1369-72. 16 ref, Eng. Tufts University School of Medicine, 136 Harrison Ave., Boston, MA 02111, USA

"The presence of enteroaggregative *Escherichia coli* (EAggEC) in stool has been strongly associated with persistent diarrhea. No treatment trials have been done to demonstrate that clearance of EAggEc results in an improvement of diarrheal symptoms. Twenty-four adults infected with the human immunodeficiency virus (HIV) with diarrhea and EAggEc were randomized to a double-blind

placebo-control cross-over treatment trial (cirprofloxacin 500 mg orally twice daily for 7 days vs. placebo). After treatment with ciprofloxacin, the subjects had significantly fewer (50%) stools per day (from 5.0 ± 2.9 to 2.4 ± 1.9). Intestinal symptoms decreased by 42% after active treatment. EAggEc were eradicated from stool of all participants after active treatment. These data strengthen the link between the presence of the EAggEc in stool and their role in the pathogenesis of diarrheal disease. It is likely that EAggEc are a treatable cause of diarrheal disease in some persons with HIV and no other apparent enteric pathogen."

350 Watarai M, Sato T, Kobayashi M, Shimizu T, Yamasaki S, Tobe T, Sasakawa C, Takeda Y. Identification and characterization of a newly isolated Shiga toxin 2-converting phage from Shiga toxin-producing *Escherichia coli*. Infect Immun 1998 Sep;66(9):4100-7. 59 ref, Eng. Research Institute, International Medical Center of Japan, 1-21-1, Toyama, Shinjuku-ku, Tokyo 162, Japan

351 Widmer G, Tzipori S, Fichtenbaum CJ, Griffiths JK. Genotypic and phenotypic characterization of *Cryptosporidium parvum* isolates from people with AIDS. J Infect Dis 1998 Sep;178(3):834-40. 23 ref, Eng. Division of Infectious Diseases, Tufts University School of Veterinary Medicine, 200 Westboro Rd., North Grafton, MA 01536, USA

352 Willingham FF, Chavez ET, Taylor DN, Bowen AB, Crane AR, Gottlieb AL, Gayles MK, Grahn KF, Perez VMC, Apolinario IS, Gilman RH. Diarrhea and *Clostridium difficile* infection in Latin American patients with AIDS. Clin Infect Dis 1998 Sep;27(3):487-93. 41 ref, Eng. Department of International Health, Johns Hopkins University School of Hygiene and Public Health, 615 North Wolfe Street, Baltimore, MD 21205, USA

"Diarrhea and wasting are among the most debilitating and deadly manifestations of AIDS, yet only limited information is available regarding the etiology, clinical consequences, and immunologic effects of infection with diarrheal agents. Peruvian AIDS patients presenting with and without diarrhea were followed prospectively to examine the relations among diarrheal pathogens, clinical presentations, CD4 lymphocyte count, weight loss, and survival. Patients with chronic diarrhea had lower CD4 lymphocyte counts (P=.001) and lost more weight (P<.001). Weight loss and a decreased CD4 lymphocyte count were associated with increased mortality (P=.001 and P=.003, respectively). Mean CD4 lymphocyte count varied significantly by diarrheal agent. *Clostridium difficile* was the most prevalent pathogen and was associated with significantly increased mortality before and after adjustment for coinfection, length of follow-up, CD4 lymphocyte count, and weight loss (P=.006). *C. difficile* may be a more important and more prevalent etiologic agent in AIDS than previously recognized and may represent a preventable cause of death in patients with immuno-suppression."

353 Yang DP, Goldberg KM, Ma X-D, Magargle W, Rappaport R. Development of a fluorescent focus identification assay using serotype-specific monoclonal antibodies for detection and quantitation of rotaviruses in a tetravalent rotavirus vaccine. Clin Diagn Lab Immunol 1998 Nov;5(6):780-3. 13 ref, Eng. Wyeth Ayerst Research, PO Box 8299, Philadelphia, PA 19101, USA

"A fluorescent focus identification assay (FFIDA) was developed for use in experimental studies and for quantitation of the components in a tetravalent live oral rotavirus vaccine. The assay utilizes four serotype-specific neutralizing monoclonal antibodies (MAb) to detect and quantify individual rotaviruses by immuno-fluorescence staining of fixed virus-infected monkey kidney cells. In mixed virus infections, all four MAb, W1 (serotype 1), 1C10 (serotype 2), R1 (serotype 3), and S4 (serotype 4), specifically stain the relevant homologous serotype without exhibiting any cross-reactivity against the other serotypes. Furthermore, the test is sensitive enough to differentiate at least twofold (0.3 log) differences in virus titer. The results of testing four individual experimental vaccine lots three or more consecutive times showed that all four lots contained similar proportions of the four vaccine strains as detected by the classical plaque neutralization identification test. The rapidity and efficiency of the FFIDA are desirable attributes that make it suitable for use in studies requiring identification and quantitation of one or more of the four major rotavirus serotypes."

354 Zhong W, Gutshall LL, Vecchio AMD. Identification and characterization of an RNA-dependent RNA polymerase activity within the nonstructural protein 5B region of bovine viral diarrhea virus (note). J Virol 1998 Nov;72(11):9365-9. 22 ref, Eng. Department of Molecular Virology and Host Defense, SmithKline Beecham Pharmaceuticals, 1250 S. Collegeville Rd., PO Box 5089, Collegeville, PA 19426-0989, USA

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World Health Organization. Scientific Working Group. Rotavirus and other viral diarrhoeas. *Bull WHO* 1980;58:183-98.

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Walker-Smith J. Diseases of the small intestine in childhood. 2d ed. Kent: Pitman Medical, 1979:171-249.

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(8) *Chapter in a book*

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Hamill PW. NCHS growth curves for children birth - 18 years - United States. Hyattsville, MD: National Center for Health Statistics, 1977. iv, 74 p. (DHEW publication no. (PHS) 78-1650) (Vital and health statistics, series 11, no. 165).

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Other articles

(13) *Newspaper article*

Azad AS. Water pollution and health hazards. *Bangladesh Observer* 1982 Dec 11:5(col 3-5).

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Roueche B. Annals of medicine; the Santa Claus culture. *The New Yorker* 1971 Sep 4:66-81.

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