Outside Europe

Value of stool examination in patients with diarrhoea

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Abstract

Findings of stool examinations in 1593 patients with diarrhoea due to a single enteric pathogen—enterotoxigenic *Escherichia coli* rotavirus, *Shigella*, *Campylobacter jejuni*, *Vibrio cholerae* 0:1, *Entamoeba histolytica*, or *Giardia lamblia*—were reviewed to determine how well they predicted the agent associated with the diarrhoea. Specimens were examined visually for blood and mucus, tested for pH, and examined under a microscope for the presence of red and white blood cells, parasites, and stool fat. Although visible blood was more common in specimens from patients infected with *Shigella* (51%) and *Ent histolytica* (39%) than in those from patients infected with other agents (6%; $p < 0.01$), patients infected with *Shigella* were most likely to have numerous faecal leucocytes (> 50 /high power field: 39% v 8% of all patients and 7% of patients infected with *Ent histolytica*, $p < 0.01$ in both cases). Patients infected with enterotoxigenic *E coli*, rotavirus, *V cholerae* 0:1, or *C jejuni* had loose stools with fewer red or white cells. Patients infected with rotavirus and *C jejuni* were more likely to have acid stools with 3 to 4+ fat, but these findings were related to young age and breast feeding.

Stool examination is most useful in establishing a diagnosis of dysentery and in helping to distinguish between patients infected with *Shigella* and *Ent histolytica*; it is of limited usefulness in discriminating between pathogens causing watery diarrhoea.

Introduction

In Bangladesh and other developing countries diarrhoea is a major cause of morbidity and mortality, especially among children.^{1 2} Over the past 10 years two major advances have been made in the treatment of diarrhoeal diseases—namely, the identification of new viral and bacterial agents, which has led to the detection of a pathogen in up to 70% of cases,^{3 4} and the universal acceptance of oral fluid and electrolyte treatment for

watery diarrhoea.⁵ Enterotoxigenic *Escherichia coli*, rotavirus, *Shigella*, *Campylobacter jejuni*, *Vibrio cholerae* 0:1, *Giardia lamblia*, and *Entamoeba histolytica* are the most common pathogens associated with diarrhoea in patients attending treatment centres in Bangladesh.^{4 6} Precise identification of these aetiological agents requires a laboratory that can perform routine cultures⁷ as well as more sophisticated techniques such as enzyme linked immunosorbent assay,⁸ in vivo animal studies,⁹ and tissue culture.^{10 11} The economic and technical limitations in most developing countries make such studies impossible outside reference laboratories. Although an aetiological diagnosis should not influence the decision to give oral treatment to patients with dehydrating diarrhoea, it may influence the decision to prescribe antibiotics or other drugs.

Inspection of stool for the presence of visible blood and mucus, measurement of stool pH, and microscopical examination for fat globules, red and white blood cells, and parasites have been performed for decades to assess colonic inflammatory processes and malabsorption of fat and carbohydrate. Previous studies have shown the value of stool microscopy to look for faecal leucocytes in patients with diarrhoeal diseases, especially shigellosis.^{12–17} Because of recent recognition of the importance of rotavirus, enterotoxigenic *E coli*, and *C jejuni* in Bangladesh we have reviewed the usefulness of direct stool examination in making a diagnosis in patients with these and other agents.

Patients and methods

This study was performed at the Dacca Hospital of the International Centre for Diarrhoeal Disease Research, Bangladesh, between 1 March 1980 and 28 February 1982. A 4% systematic sample of all patients coming to the hospital (Dacca Hospital surveillance system⁴) was interviewed by a trained health worker and examined by a physician. Two rectal swabs were obtained from each patient, one for immediate culture and one for detection of rotavirus. Swabs were processed for *Salmonella*, *Shigella*, *V cholerae* 0:1 and non 0:1, *C jejuni*, rotavirus, and enterotoxigenic *E coli* by procedures described previously.⁴

A single fresh stool sample was examined within one hour of collection for the presence of visible blood and mucus and was tested with litmus paper for an acid or alkaline pH. The stool for microscopical examination was chosen from an area with blood or mucus if present. A wet mount of each specimen was made by mixing a fleck of stool from the tip of a wooden applicator stick with one drop of physiological saline directly on a glass slide so that newsprint could barely be read through the final preparation. An experienced technician examined the preparation under a light microscope for five minutes to determine the numbers of red and white blood cells per high power field, qualitative stool fat (recorded as 0 to 4+), and the presence of parasites. The technician first scanned the slide under low power to detect ova and parasites and to determine areas of increased or decreased cellularity. Two high power fields were examined to obtain a count of the numbers of red and white cells, and other fields were examined to confirm the presence of parasites. If cysts were detected in any sample a wet mount in Lugol's iodine

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was examined to identify the type of cyst. An iodine preparation was examined routinely for all patients aged over 2. If no ova or cysts were found on direct examination, concentration by the flotation method was done and the specimen re-examined.¹⁸ *Ent histolytica* and *G lamblia* were considered to be possible diarrhoeal pathogens only if trophozoites were detected. The examiner had no knowledge of the patient's clinical history.

Results were analysed using the χ^2 , Fisher's exact, and the Kolmogorov-Smirnov goodness of fit tests.

Results

Altogether 6849 of the 171 225 patients who attended Dacca Hospital during the two year study period were enrolled in the surveillance system. A total of 4657 (68%) submitted a stool for examination, and all of the tests were performed on 3558 (76%) of these—that is, stool culture for *Salmonella*, *Shigella*, *Vibrio*, and *C jejuni*; detection of rotavirus by enzyme linked immunosorbent assay; assays for *E coli* toxin; and microscopical examination. A pathogen was identified in 2283 (64%) of the patients screened for all agents, of whom 1622 (71%) were infected with a single agent—namely, rotavirus, 489 patients; enterotoxigenic *E coli*, 353; *Shigella*, 284; *C jejuni*, 173; *V cholerae* 0:1, 112; *Ent histolytica*, 100; *G lamblia*, 82; *V cholerae* non-0:1, 18; and *Salmonella*, 11. Only data on patients screened for all agents in whom a single pathogen was detected were analysed. Altogether 779 of these patients (48%) had an associated helminth or other protozoal agent; these patients were included in the analysis.

Half of the patients were infants and children under 3 years. Rotavirus, enterotoxigenic *E coli*, *Shigella*, and *C jejuni* were the agents most commonly detected (table I). Children under 3 were primarily infected with agents for which antibiotic treatment is not usually indicated (that is, rotavirus, enterotoxigenic *E coli*, *C jejuni*), whereas older children and adults were more likely to be infected with agents

requiring drug treatment (that is, *Shigella*, *V cholerae* 0:1, *Ent histolytica*, *G lamblia*).

Stool with visible blood was commonly found in patients infected with *Shigella* (51%) and *Ent histolytica* (39%) but was uncommon in patients infected with other agents (6%; $p < 0.01$) (table II). Stool with visible mucus occurred in 85% of all patients and did not distinguish between infecting agents. Visible blood correlated closely with the presence of more than 10 red blood cells per high power field (table III). Microscopical examination for faecal red and white

TABLE III—Relation between visible blood in stool and presence of red blood cells in specimens from 3292 patients with diarrhoea, Dacca Hospital surveillance, March 1980 to February 1982

No of red blood cells/high power field	Total No of patients	No (%) with visible blood
0	1985	2 (<1)
1-10	901	57 (6)
11-50	297	246 (83)
51-100	67	64 (96)
101+	42	42 (100)

blood cells was helpful in distinguishing invasive pathogens. Patients infected with *Shigella* or *Ent histolytica* had more faecal erythrocytes (>10 /high power field) than patients with other agents (48% and 39% respectively v 12% of all patients, $p < 0.01$). Although patients infected with either *Ent histolytica* or *Shigella* had more faecal leucocytes (>10 /high power field) than patients infected with other agents (83% and 68% respectively v 48% of all patients, $p < 0.01$), patients infected with *Shigella* more commonly had sheets of leucocytes (>50

TABLE I—Age specific prevalences of infection in patients screened for all pathogens,* Dacca Hospital surveillance, March 1980 to February 1982 (figures are numbers (%) of patients)

Age (years)	No of patients	Agents for which drug treatment not required			Agents for which drug treatment indicated				Total requiring drug treatment
		Rotavirus	Enterotoxigenic <i>E coli</i>	<i>C jejuni</i>	<i>Shigella</i>	<i>V cholerae</i> 0:1	<i>Ent histolytica</i>	<i>G lamblia</i>	
<1	944	351 (37)	156 (17)	188 (20)	71 (8)	16 (2)	4 (<1)	24 (3)	115 (12)
1-2	840	259 (31)	148 (18)	134 (16)	132 (16)	40 (5)	12 (1)	57 (7)	241 (29)
3-4	291	36 (12)	46 (16)	36 (12)	42 (14)	34 (12)	31 (11)	23 (8)	130 (45)
5-14	410	29 (7)	82 (20)	29 (7)	55 (13)	42 (10)	48 (12)	35 (9)	180 (44)
15-44	872	63 (7)	123 (14)	20 (2)	117 (13)	44 (5)	49 (6)	34 (4)	244 (28)
45+	201	16 (8)	34 (17)	7 (3)	31 (15)	10 (5)	25 (12)	7 (3)	73 (36)
Total	3558	754 (21)	589 (17)	414 (12)	448 (13)	186 (5)	169 (5)	180 (5)	983 (28)

*Includes pathogens listed here plus *Salmonella* and *V cholerae* non 0:1.

TABLE II—Results of stool examination in patients with single enteric pathogen, Dacca Hospital surveillance, March 1980 to February 1982 (figures are numbers (%) of patients)

Characteristic	All patients (n = 3558)	Watery diarrhoea			Dysenteries		Others	
		Rotavirus (n = 489)	Enterotoxigenic <i>E coli</i> (n = 353)	<i>V cholerae</i> 0:1 (n = 112)	<i>Shigella</i> (n = 284)	<i>Ent histolytica</i> (n = 100)	<i>C jejuni</i> (n = 173)	<i>G lamblia</i> (n = 82)
Visual examination:								
Blood	427 (12)	24 (5)**	21 (6)**	9 (8)	145 (51)**	39**	9 (5)**	6 (7)
Mucus	3024 (85)	416 (85)	306 (87)	99 (88)	264 (93)**	90	147 (85)	71 (86)
pH:								
Acid	1921 (54)	357 (73)	165 (47)	23 (21)	77 (27)	22	128 (74)	39 (47)
Alkaline	1637 (46)	132 (27)	188 (53)	89 (79)	207 (73)	78	45 (26)	43 (53)
Microscopical examination:								
No of red blood cells/high power field:								
0	2135 (60)	369 (75)	218 (62)	71 (63)	59 (20)	37	107 (62)	46 (56)
1-10	996 (28)	103 (21)	110 (31)	35 (31)	90 (32)	24	50 (29)	30 (37)
11-50	320 (9)	14 (3)	22 (6)	5 (5)	90 (32)	26	15 (8)	5 (6)
51+	107 (3)	3 (1)	3 (1)	1 (1)	45 (16)	13	1 (1)	1 (1)
No of white blood cells/high power field:								
0	71 (2)	15 (3)	4 (1)	1 (1)	2 (1)	1	9 (5)	0
1-10	1743 (49)	308 (63)	178 (50)	59 (53)	45 (16)	31	84 (49)	34 (41)
11-20	854 (24)	100 (20)	102 (29)	32 (28)	35 (12)	27	41 (24)	26 (32)
21-50	605 (17)	53 (11)	52 (15)	18 (16)	91 (32)	34	32 (18)	21 (26)
51+	285 (8)	13 (3)	17 (5)	2 (2)	111 (39)	7	7 (4)	1 (1)
Fat:								
0 to 2+	3095 (87)	300 (61)	314 (89)	109 (97)	281 (99)	100	140 (81)	82 (100)
3 to 4+	463 (13)	189 (39)	39 (11)	3 (3)	3 (1)	0	33 (19)	0

Significance of difference between all patients and patients infected with a given agent: * $p < 0.05$, ** $p < 0.01$.

TABLE IV—Comparison of findings on stool examination in patients with *Shigella* infection who presented with dysentery and those who presented with watery diarrhoea, Dacca Hospital surveillance, March 1980 to February 1982 (figures are numbers (%) of patients)

Finding on examination	Presenting complaint		p
	Dysentery (n = 194)	Watery diarrhoea (n = 62)	
Visual blood	124 (64)	14 (23)	<0.01
Visual mucus	188 (97)	52 (84)	<0.01
Alkaline pH	155 (80)	40 (65)	<0.05
>10 white blood cells/high power field	175 (90)	40 (65)	<0.01
>10 red blood cells/high power field	116 (60)	10 (16)	<0.01

TABLE V—Findings on stool examination in children of different age groups infected with single enteropathogen, Dacca Hospital surveillance, March 1980 to February 1982 (figures are numbers (%) of patients)

Age (years)	Characteristic	All patients	Enterotoxigenic <i>E coli</i>	Rotavirus	<i>C jejuni</i>	<i>Shigella</i>	<i>V cholerae</i> 0:1	<i>Ent histolytica</i>	<i>G lamblia</i>
<1	No of patients	944	83	238	85	31	8	1	13
	Visible blood	28 (3)	2 (3)	1 (<1)**	3 (4)	7 (23)**	NA	NA	1 (8)
	Acid pH	727 (77)	69 (83)	193 (81)	66 (78)	20 (65)	NA	NA	11 (85)
	>10 red blood cells/hpf	36 (4)	2 (2)	0**	6 (7)	8 (26)**	NA	NA	1 (8)
	>10 white blood cells/hpf	358 (38)	27 (33)	69 (29)*	31 (36)	24 (77)**	NA	NA	9 (69)*
	3 to 4+ fat	320 (34)	30 (36)	120 (50)**	26 (31)	3 (10)**	NA	NA	0**
1-4	No of patients	1131	106	183	61	93	40	27	28
	Visible blood	119 (11)	2 (2)**	7 (4)**	4 (7)	40 (43)**	2 (5)	9 (33)**	1 (4)
	Acid pH	679 (60)	65 (61)	150 (82)**	47 (77)**	31 (33)**	6 (15)**	3 (11)**	17 (61)
	>10 red blood cells/hpf	118 (10)	1 (1)**	4 (2)**	6 (10)	36 (39)**	1 (3)	11 (41)**	1 (4)
	>10 white blood cells/hpf	548 (48)	36 (34)**	56 (31)**	33 (54)	78 (84)**	16 (40)	19 (70)*	17 (61)
	3 to 4+ fat	145 (13)	10 (9)	68 (37)**	8 (13)	4 (4)**	1 (3)**	0*	0*
5-14	No of patients	410	48	15	9	40	22	23	20
	Visible blood	70 (17)	5 (10)	5 (33)	NA	25 (63)**	1 (5)	8 (35)*	3 (15)
	Acid pH	156 (38)	15 (32)	10 (67)	NA	6 (15)**	2 (9)**	7 (30)	5 (25)
	>10 red blood cells/hpf	63 (15)	6 (13)	2 (13)	NA	22 (55)**	1 (5)	6 (26)	3 (15)
	>10 white blood cells/hpf	215 (52)	30 (63)	10 (67)	NA	33 (83)**	13 (59)	13 (57)	12 (60)
	3 to 4+ fat	2 (<1)	0	1 (7)	NA	0	0	0	0

Significance of differences between all patients and patients infected with each agent: *p<0.05, **p<0.01. NA = Not analysed because there were fewer than 10 patients in the group. hpf = High power field.

TABLE VI—Stool pH and fat versus breast feeding in children aged under 3 with diarrhoea and infected with a single enteric pathogen, Dacca Hospital surveillance, March 1980 to February 1982 (figures are numbers (%) of children)

Agent	No of children		Acid pH		≥3+ fat	
	Breast fed	Not breast fed	Breast fed	Not breast fed	Breast fed	Not breast fed
Enterotoxigenic <i>E coli</i>	138	26	112 (81)	9 (35)**	40 (29)	0**
Rotavirus	355	44	298 (84)	26 (59)**	181 (51)	4 (9)**
<i>Shigella</i>	60	40	30 (50)	11 (28)*	2 (3)	1 (3)
<i>C jejuni</i>	108	26	89 (82)	11 (41)*	30 (28)	1 (4)**
All children <3 years†	1385	385	1080 (78)	196 (51)**	443 (32)	12 (3)**

Significance of differences between breast fed and non-breast fed children infected with same agent: *p<0.05, **p<0.01. †Not all infecting agents are listed in table.

white blood cells/high power field) (39% v 7%, p<0.01). Patients infected with *Shigella*, *V cholerae* 0:1, or *Ent histolytica* were more likely to have an alkaline stool pH than those infected with rotavirus and *C jejuni* (p<0.01). Stool fat was absent in most patients, but those infected with rotavirus and *C jejuni* had significantly more fat (3 to 4+) in their stools (rotavirus p<0.01; *C jejuni* p<0.05).

There were no significant differences in findings on stool examination between patients with each pathogen with different severities of illness—that is, between patients in hospital and outpatients. There were also no significant differences in the numbers of faecal red and white cells between patients infected with a given pathogen who had an associated helminth or protozoal agent and those who did not.

Because patients with shigellosis may present with either dysentery or watery diarrhoea we compared findings on stool examination in patients infected with *Shigella* who presented with dysentery with those in patients who presented with watery diarrhoea (table IV). Patients who complained of dysentery were significantly more likely to have stools with visible blood and mucus, an alkaline pH, and more than 10 red and white blood cells per high power field than patients who complained of watery diarrhoea. Patients who presented with watery diarrhoea, however, could still be distinguished from all other patients by having stools with a significantly greater likelihood of having visible blood (23% v 12%, p<0.05), an alkaline pH (65% v

cells/high power field, 65% v 49%, p<0.05; and >50 white blood cells/high power field, 19% v 8%, p<0.01). Of the 284 *Shigella* isolates, 190 (67%) were *Sh flexneri*, 41 (14%) *Sh dysenteriae* type 1, 27 (10%) *Sh boydii*, 14 (5%) *Sh dysenteriae* type 2, and 12 (4%) *Sh sonnei*. Patients infected with *Sh dysenteriae* type 1 were significantly more likely than those infected with *Sh flexneri* to have visible blood (95% v 46%, p<0.01) and numerous red and white cells in their stools (>50 red blood cells/high power field, 29% v 13%, p<0.01; >50 white blood cells/high power field, 63% v 35%, p<0.01).

Results of examinations for stool pH and fat were influenced by age (table V). Infants (<1 year) and young children (1-2 years) more commonly had stools with an acid pH and 3 to 4+ fat than older children and adults (acid pH: 71% v 35%, p<0.01; and 3 to 4+ fat: 25% v <1%, p<0.01). Among infants an acid pH did not help to

46%, p<0.01), and numerous faecal leucocytes (>10 white blood cells distinguish between pathogens. Among children aged 1-4, however, those infected with rotavirus and *C jejuni* were more likely to have stools with an acid pH (82% and 78% respectively v 60%, p<0.01). Patients aged under 4 infected with rotavirus were significantly more likely to have 3 to 4+ fat than those infected with other agents (45% v 25%, p<0.01). Other examination findings were independent of age.

Altogether 1385 (78%) of the 1784 children aged under 3 were at least partially breast fed. To determine whether breast feeding contributed to the frequent finding of acid stools with 3 to 4+ fat in infants and young children we compared stool pH and fat in children under 3 who were breast fed with those in children who were receiving no breast milk (table VI). Breast fed children were more likely to have stools with an acid pH (78% v 51%, p<0.01) and 3 to 4+ fat (32% v 3%, p<0.01). Among breast fed children those infected with rotavirus more commonly had acid stools with 3 to 4+ fat (p<0.01) and those infected with *Shigella* less commonly had acid stools with 3 to 4+ fat (p<0.01) than children without these agents. Among the children who did not receive breast milk, however, those infected with rotavirus were not significantly more likely to have acid stools or 3 to 4+ fat, and those infected with *Shigella* were even less likely to have acid stools than the patients not infected with these agents (p<0.01).

Discussion

The importance of direct stool examination in evaluating patients with diarrhoea was recognised in the last century by Koch¹² and has been reiterated by recent studies confirming its value in shigellosis and parasitic infections.¹³⁻¹⁷ Its usefulness in diagnosing illnesses caused by the wider range of agents now associated with diarrhoea has not been fully assessed.

Patients with visible stool blood and microscopic red blood cells were most likely to be infected with *Shigella* or *Ent histolytica*, agents for which drug treatment is indicated. As has been shown by previous studies,¹³⁻¹⁶ numerous faecal leucocytes (>50/high power field) were most common in patients infected with *Shigella*, which distinguished them from patients infected with *Ent histolytica*. While patients infected with *Shigella* who presented with watery diarrhoea were less likely than those who presented with dysentery to have numerous faecal red and white blood cells, they were more likely than all patients to have numerous faecal leucocytes, suggesting that an invasive process plays a part.

Patients infected with rotavirus and *C jejuni* were more likely to have acid stools with 3 to 4+ fat, but these findings were related to young age and breast feeding. Infants infected with any pathogen often had fat in the stool and an acid pH, suggesting that fat and carbohydrate malabsorption are common in this age group independent of the pathogen. The high prevalence of breast feeding in our population contributed to the high prevalence of stool fat and acid pH in young children. These findings may explain the observations of Sack *et al.*,¹⁹ who reported a higher concentration of reducing substances after acid hydrolysis in stools of children infected with rotavirus than in those infected with enterotoxigenic *E coli* or *Shigella* and attributed this to rotavirus infection. No age stratification was done in their study, and since patients infected with rotavirus are primarily under 2 years old and therefore often breast fed, the results may possibly be explained more by differences in age and feeding habits than by the infection itself.

The results of this study of patients with diarrhoea in a developing country may not be completely similar to those that would be obtained in a Western population. Although one third of our patients were infected with more than one pathogen, we chose to analyse data on those in whom only a single agent was detected. Some of our patients with non-invasive diseases (*V cholerae* 0:1, enterotoxigenic *E coli*, rotavirus, *G lamblia*) had faecal leucocytes (>20/high power field), which were reported to be absent in specimens from healthy Western volunteers challenged with these same agents¹³ and from Western patients infected with rotavirus.¹⁵ These findings suggest that there might be different host responses in patients with infectious diarrhoea in developing countries, where infection is endemic and repeated and where undernutrition is common, or that other invasive agents not currently being identified might have been present in these patients. The presence of parasites in 48% of the patients did not explain these findings. Finally, we have not discussed dark field techniques, which increase the usefulness of microscopy in diagnosing *V cholerae* 0:1 and *C jejuni*.

In summary, a simple stool examination requiring less than 10 minutes of a technician's time was helpful in establishing a diagnosis of dysentery and in distinguishing between the invasive pathogens *Shigella* and *Ent histolytica*. It was of particular value in older children and adults, who were more likely to have these agents, for which drug treatment may be indicated. Although stool examinations were of limited usefulness in discriminating between pathogens in watery diarrhoea, especially *V cholerae* 0:1 and enterotoxigenic *E coli*, these agents produce self-limited diarrhoeas for which fluid replacement treatment is mandatory but for which antibiotic treatment may serve only to decrease the duration of excretion of organisms and of disease.^{20 21}

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Is the temperature recorded by the clinical thermometer a reliable sign of infection in elderly patients?

The temperature response of the elderly to disease is less constant than it is in younger people. An elderly person may be seriously ill with an infection and yet have no fever. But this does not mean that the temperature is not worth taking since in most cases there is some rise. Occasionally the temperature response in an ill elderly person is paradoxical, an acute illness provoking a hypothermic reaction rather than a fever. In such cases when the hypothermia is treated the temperature often shows an overshoot to a feverish level. There is every justification for continuing to record the temperature when an old person is ill. The pulse rate remains invaluable at any age. Very often in an elderly person an increase in the respiratory rate without a febrile response may be the first sign of pneumonia.—R E IRVINE, consultant physician, Hastings.