ENTEROTOXIGENIC ESCHERICHIA COLI (ETEC) DISEASE IN BANGLADESH CLINICAL, THERAPEUTIC, AND LABORATORY ASPECTS

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Many studies have demonstrated the importance of enterotoxigenic E. coli (ETEC) as a cause of diarrhea. In Bangladesh 3 previous studies have shown these organisms to be responsible for many adult cases of cholera-like diarrhea (4, 6, 7). In this study we attempted to define, in hospitalized patients, detailed clinical and laboratory features of ETEC disease, the effect of tetracycline on its clinical course, and efficient means of its diagnosis.

MATERIALS AND METHODS

One hundred seventy-six adult males presenting to the Cholera Research Laboratory (CRL) Hospital in Dacca, Bangladesh, in October through December 1976 with a history of acute watery diarrhea, appearing to have lost 5% or more of their body weight, were studied. All had admission stools examined for Vibrio cholerae by darkfield microscopy and were negative. On admission, a history and physical were performed; stool was obtained by rectal catheter for culture, microscopic analysis, and electrolyte determination; and a venous blood sample was drawn for measurement of electrolytes and specific gravity and for acute serology. Replacement intravenous fluid therapy was administered within four hours, at which time a finger-tip blood specimen was obtained for measurement of specific gravity and a complete blood count. Two hours after admission tetracycline or placebo therapy was initiated with 500 mg capsules for patients weighing 30 or more kgs and 250 mg capsules for those weighing less than 30 kgs. Capsules were given at 6-hour intervals for 72 hours. All subsequent stool losses were replaced with intravenous fluid, and stool volume and fluid replacement were recorded every 2 hours. Follow-up stool cultures were obtained by rectal swabs every 24 hours for 4 days, and 7 and 14 days after admission. Convalescent serum was obtained at 14 days from 102 cases.

All stools were examined for salmonellae, shigellae, and vibrios by standard CRL procedures and for rotavirus by the ELISA assay (11). Patients with those organisms were dropped from further analysis. Up to ten lactose-positive colonies with typical E. coli morphology were individually picked from the MacConkey plate of each stool culture. The first 5 colonies were repicked to form a 5-pool and all 10 colonies were then repicked to form a 10-pool. Isolates and pools from admission cultures were grown in trypticase soy broth with 0.6% yeast extract (TSBY) in a shake (60-70 shakes/min) culture at 37°C and examined for production of heat-labile toxin (LT) by the CHO assay (2) and heat-stable

toxin (ST) by the infant mouse assay (5). Isolates and pools from follow-up cultures of LT-ST and LT E. coli cases were grown in syncase broth in a resting 48 hour culture and examined for LT production in the adrenal cell assay (8). Isolates and pools from follow-up cultures of ST cases were tested for ST production after 18-24 hour growth on a roller drum (22rev/min).

The Sereny test (10) for invasiveness was used to screen lactose-positive and -negative isolates from 100 patients.

One lactose-positive colony from each patient was confirmed as E. coli and serotyped at the WHO Reference Center in Copenhagen. Antibiotic sensitivity tests were carried out by the Kirby-Bauer method (1) on these enterotoxigenic isolates and a sample of non-toxigenic E. coli.

In an attempt to detect LT and ST in the stools of cases 2-3 ml aliquots of admission stools that had been stored at -40°C for approximately 5 months were defrosted and spun at 10,000 rpm for 30 minutes. Supernatants were then tested in the adrenal cell and infant mouse assays. Specimens were run by 2 different procedures. In the first the stools were kept at room temperature for 45-90 minutes after centrifugation and prior to toxin testing (Method A); in the second they were kept constantly at 4°C (Method B).

Anti-toxin neutralization titers were determined for the 102 paired sera by the adrenal cell method (9). Antibody titer measurements to rotavirus were measured by complement fixation (3) and ELISA assays (12).

RESULTS

A diagnosis was made in 147 (86%) of the 176 cases. Twenty-three (13%) were mixed infections. Data in Table 1 shows the number of cases in which each organism was found as a single pathogen and the total number of cases in which the organism was isolated. ETEC was isolated from 109 (62%) cases and was the only pathogen in 95 (54%). These 95 will be referred to as E. coli cases. Other pathogens isolated included Vibrio cholerae, Vibrio parahaemolyticus, non-cholera Vibrio (NAG), Shigella flexneri, Shigella boydii, Shigella sonnei, Salmonella C and invasive E. coli. In 8 of the 9 rotavirus infections, the virus was detected in stool. Nine (64%) of the 14 NAG infections, 12 (75%) of the 16 Shigella infections and 8 (89%) of the 9 rotavirus infections were mixed infections,

Table 1

FREQUENCY OF ORGANISMS ISOLATED FROM 176 STUDY CASES

Organism	Isolated			
	Single Pathogen		Total Infections	
	No.	%	No.	%
Enterotox. Escherichia coli	95	54	109	62
Vibrio cholerae	12	7	14	8
Vibrio parahaemolyticus	6	3	6	3
NAG (Non-cholera vibrio)	5	3	14	8
Shigella *	4	2	16	9
Invasive E. coli	1	1	1	1
Rotavirus	1	1	9	5
Salmonella C			1	1

* Includes 11 cases with S. flexneri, 3 cases with S. boydii and 2 cases with S. sonnei.

Of the 95 E. coli cases 62 (65%) had LT-ST-producing strains, 30 (32%) had ST-producing strains, and 3 (3%) had LT-producing strains. The clinical features of disease caused by LT-ST E. coli and ST E. coli were similar (Table 2). Almost all the E. coli cases had vomiting with onset on the average 3.8 hours after onset of diarrhea. Two thirds had muscle cramps, 51% had abdominal pain, and 19% had fever lasting an average of 4 hours.

Selected laboratory features of these cases are shown in Tables 3 and 4. For all cases the mean admission serum sodium (134 meq/L), potassium (4.8 meq/L), and chloride (102 meq/L) and the mean hematocrit value (37.9%) measured 4 hours after admission were normal. The mean total white count 4 hours after admission was elevated (14,600); many patients had a shift to the left. Stool microscopic examinations revealed 0 to 5 polymorphonuclear leukocytes per high powered field in 82% of cases, but 8 (9%) cases had 6 to 10 and 7 (8%) cases had greater than 10 per high power field. Over 90% of patients had parasitic infestation. For all these values there was no difference between LT-ST and ST E. coli cases. Data in Table 4 show that the mean stool electrolyte and osmolarity values for the LT-ST and ST E. coli cases were similar to each other and to those of the cholera, other Vibrio, and unknown etiology cases.

Table 2

CLINICAL FEATURES OF ENTEROTOXIGENIC *E. COLI* CASES

	LT - ST * (N=55) No.	%	ST (N=30) No.	%	All ** (N=88) No.	%
Vomiting	53	96	28	94	84	96
D - V (hr) ***	4.1		3.7		3.8	
Muscle cramps	37	67	20	66	58	66
Abdominal pain	25	46	18	60	45	51
Temp. (over 100°F axillary)	9	16	8	27	17	19
Fever dur. (hr)	3.7		4.4		4.0	

* Excludes 7 cases who took tetracycline prior to admission
** Includes 3 LT cases
*** Interval from diarrhea to vomiting

Table 3

ADMISSION LABORATORY FEATURES OF ENTEROTOXIGENIC *E. COLI* CASES

(Mean Values)

	LT - ST * (N=55)	ST (N-30)	All ** (N=88)
Serum Na+ (meq/L)	133	134	134
Serum K+ (meq/L)	4.9	4.6	4.8
Serum Cl- (meq/L)	102	102	102
Hct*** (%)	37.7	38.4	37.9
WBC*** (cells/mm^3)	15,100	13,100	14,600
Fecal Leukocytes (No./hpf) *			
0 - 5	44****	25	72
6 - 10	6	2	8
> 10	5	2	7

* Excludes 7 cases who took tetracycline prior to admission
** Includes 3 LT cases
*** Measured 4 hours after admission
**** No. of cases

Table 4

STOOL ELECTROLYTES* BY ETIOLOGY

Diagnosis	Na (meq/L)	K (meq/L)	Cl (meq/L)	HCO_3 (meq/L)	OSMOL (osm/L)
Enterotox. E. coli					
LT-ST (N=50)	108±4	27±2	78±3	45±4	306±3
ST (N=23)	108±6	24±3	72±4	37±2	327±14
LT (N=3)	129±4	18±3	89±3	42±4	309±3
Cholera (N=11)	115±7	27±4	88±6	38±3	322±16
Other Vibrio (N=10)	124±2	14±2	82±4	36±4	344±15
Unknown (N=26)	114±4	21±2	77±4	39±3	322±8

* Mean values ± 1SE

Twenty three (61%) of the 38 LT-ST cases and 3 of 4 LT cases had 4-fold or greater rise in titer to LT. None of 22 ST patients had a titer rise. Only 1 of 38 other patients with diarrhea due to NAG, Shigella, rotavirus, or of unknown etiology had a titer rise, and this case had a NAG infection and only 1 E. coli isolated from his admission stool specimen which was not enterotoxigenic.

Table 5 presents data that attempt to compare the severity of disease in 31 LT-ST and 16 ST cases. Although none of the differences are significant, the LT-ST cases tended to be older, had greater loss of body weight from their illness, more often appeared severely dehydrated on admission, and had a higher admission serum specific gravity and a lower serum bicarbonate. Although the duration of diarrhea before hospital admission was almost identical, the total duration of diarrhea in the LT-ST cases (32.6 hr) was significantly greater than that in the ST cases (22.5 hr). LT-ST cases also purged a significantly greater volume of stool than ST cases (35ml/kg versus 11ml/kg) and required more intravenous fluid (111ml/kg versus 81ml/kg). The duration of excretion of organisms in stool was almost the same: 2.4 days for LT-ST cases and 2.1 days for ST cases.

The effect of tetracycline on 20 LT-ST cases was compared with 25 cases who received placebo (Table 6). Persons in both groups had tetracycline-sensitive strains and purged for at least 4 hours after admission. In the 2 groups there was no significant differences in clinical or laboratory parameters on admission, although the placebo group had a slight trend toward a more severe illness with a lower mean systolic pressure, greater percent weight loss, more cases appearing severely dehydrated, and a higher admission specific gravity and lower bicarbonate. Compared to the placebo cases the tetracycline cases had a shorter duration of illness in the hospital (25 hr versus 18.8 hr), less stool volume (43ml/kg versus 28ml/kg), and less intravenous fluid requirement (120ml/kg versus 105ml/kg), although these differences were not statistically significant. There was a highly significant difference in mean duration of excretion in the 2 groups - 2.2 days in the placebo group and 0.6 days in the tetracycline group. This effect of

tetracycline on bacterial excretion was apparent after only one day of therapy (Table 7). Twenty-four hours after admission 25 of 29 placebo cases were still culture positive for LT-ST organisms compared to 9 of 22 in the tetracycline group. This difference became more apparent over the next 72 hours. One case in each group was positive on day 7 or 14 follow-up cultures.

Table 5

COMPARATIVE FEATURES OF LT-ST AND ST CASES *

	LT-ST (N=31)	ST (N=16)	
Age (yr)	33	24	p=.07**
Discharge weight (kg)	41.6	38.5	
Admission systolic BP (mmHg)	76	79	
Weight loss (%)	7.5	6.3	p=.4 **
Clinical severity (cases)			
Moderate	10	9	
Moderate-severe	13	6	p=<0.2****
Severe	8	1	
Admission serum spec. gravity	1.0334	1.0327	p=.09**
Admission serum bicarbonate (meq/L)	16.0	17.8	p=<.1***
Duration of diarrhea (hr)			
Pre-admission	11.9	11.7	
Total	32.6	22.5	p=<.05***
Stool volume (cc/kg)	35	11	p=.03**
I.V. volume (cc/kg)	111	81	p=<.05***
Stool excretion (days)	2.4	2.1	

* All values are means except these for clinical severity
** Mann-Whitney U Test
*** t test
**** chi square

Table 6

EFFECT OF TETRACYCLINE ON LT-ST* ENTEROTOXIGENIC *E. COLI* DISEASE

	Placebo (N=25)	Tetracycline (N=20)	
Age (yr)	34	30	
Discharge weight (kg)	41.9	39.4	
Admission systolic (BP (mmHg)	72	88	
Weight loss (%)	7.8	6.4	
Clinical severity (cases)			
Moderate	6	5	
Moderate-severe	11	11	
Severe	8	4	
Admission serum spec. gravity	1.0347	1.0338	
Admission serum bicarbonate (meq/L)	16.1	16.8	
Duration of diarrhea (hr)			
Pre-admission	11.8	11.7	
Hospital	25.0	18.8	p<.2**
Stool volume (ml/kg)	43	28	p=.34***
I.V. volume (ml/kg)	120	105	p<.3**
Stool excretion (days)	2.2	0.6	p<.001**

* All values are means except for clinical severity
** t test
*** Mann-Whitney U test

Table 7

NUMBER OF CASES EXCRETING LT-ST *E. COLI*

Culture	Placebo (N=29)	Tetracycline (N=22)	
24 hr	25	9	p≤.01**
48 hr	21	3	p≤.001**
72 hr	16	1	p<.001**
96 hr	5	0	
7 days	1 (24) ***	0 (14)	
14 days	0 (17)	1 (11)	

* Includes 6 cases who did not purge in hospital
** t test
*** Numbers in parenthesis is number cultured

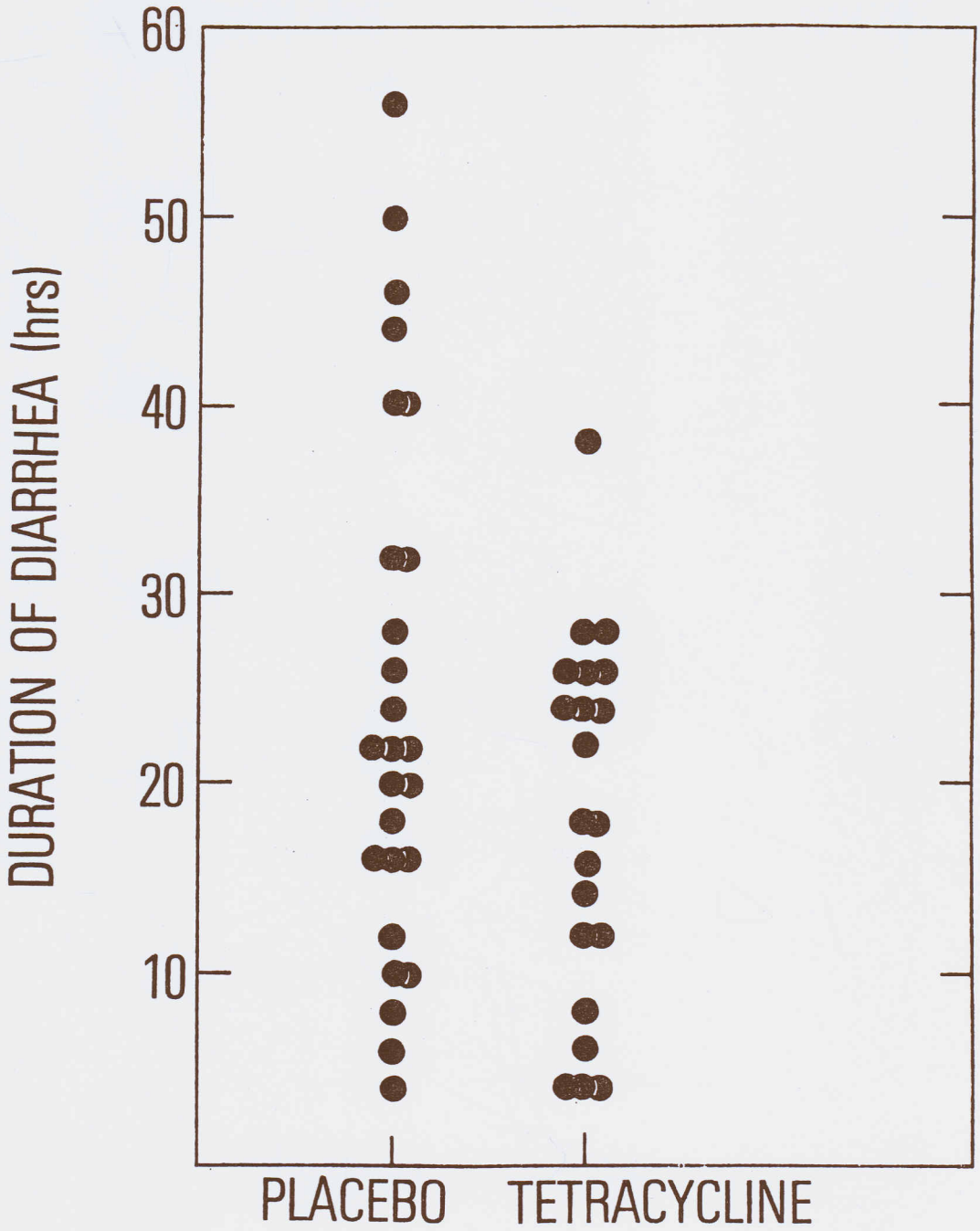

FIGURE 1

DURATION OF DIARRHEA AFTER HOSPITAL ADMISSION
IN ENTEROTOXIGENIC E. COLI LT-ST CASES

In an attempt to further assess the effect of tetracycline on duration of diarrhea, Figure 1 shows the number of cases in the placebo and tetracycline groups by duration of diarrhea in the hospital. Almost all of the cases that purged 28 hours or longer were in the placebo group. As shown in Table 8 there was a significantly greater number of cases in the placebo group than in the tetracycline group whose duration of illness was greater than 28 hours.

Figure 1

Table 8

DURATION OF DIARRHEA IN LT-ST CASES IN PLACEBO AND TETRACYCLINE GROUPS

Duration	Placebo	Tetracycline
$\leq$ 28 hr	17	19
< 28 hr	8	1

p = 0.03, Fischer's exact test

The percentage of cases in the placebo and tetracycline groups who had a 4-fold or greater rise in anti-LT titers and the geometric mean acute phase and convalescent phase titers in the 2 groups are shown in Table 9. Thirteen of 18 (72%) placebo cases and 6 of 12 (50%) of tetracycline cases had a 4-fold or greater titer rise; the difference was not significant. Similarly, the mean convalescent-phase titer in the placebo group (21.2 units) was greater than that in the tetracycline group (16.4 units), but this difference was also not significant.

Table 9

RELATIONSHIP BETWEEN TETRACYCLINE THERAPY AND ANTITOXIN RESPONSE IN LT-ST CASES

	Placebo (N=18)	Tetracycline (N=12)
Four-fold or greater titer rise	13/18 (72%)[1]	6/12 (50%)[1]
Geometric mean titer (unit/ml)		
Acute-phase	3.0	6.1
Convalescent-phase	21.2[2]	16.4[2]

1.2
Difference in placebo and tetracycline groups not significant

Only 18 cases were included in the assessment of tetracycline on ST disease - 10 received placebo and 8 received tetracycline. Except for age and clinical severity these groups were well matched (Table 10). Tetracycline had no effect on disease duration, stool volume, or intravenous fluid requirement. However, tetracycline did significantly shorten the duration of bacterial excretion from 1.8 to 0.4 days. As was the case of LT-ST disease this effect was seen within 24 hours after admission (Table 11). 12 of 14 placebo cases were positive for ST organisms at 24 hours compared with 4 of 10 tetracycline treated cases. By 72 hours only placebo patients were still culture positive. No cases were culture positive on day 7 or day 14.

Table 10

EFFECT OF TETRACYCLINE ON ST-DISEASE*

	Placebo (N=10)	Tetracycline (N=8)	
Age (yr)	25	40	p=.03**
Discharge weight (kg)	41.0	43.3	
Admission systolic BP (mmHg)	88	93	
Weight loss (%)	5.2	4.8	
Clinical severity (cases)			
Moderate	8	4	
Moderate-severe	1	4	
Severe	1	0	
Admission plasma spec. gravity	1.0326	1.0320	
Admission serum bicarbonate (meq/L)	18.3	19.2	
Duration of diarrhea (hr)			
Pre-admission	10.5	8.0	
Hospital	17.2	15.5	
Stool volume (ml/kg)	17	24	
I.V. volume (ml/kg)	85	86	
Stool excretion (days)	1.8	0.4	p=.01**

* All values are means except for clinical severity
** Mann-Whitney U test

Table 11

NUMBER OF CASES EXCRETING ST E. COLI

Culture	Placebo (N=14)	Tetracycline (N=10)	
24 hr	12	4	p<.05*
48 hr	8	1	p<.05*
72 hr	6	0	p<.05*
96 hr	2	0	
7 days	0 (12) **	0 (9)	
14 days	0 (11)	0 (7)	

Includes 6 cases who did not purge in hospital
* t test
** Number in parenthesis is number cultured

Table 12 shows the antibiotic resistance patterns of the ETEC and a group of 76 nontoxigenic E. coli isolated from admission stool specimens of 11 ETEC and 63 non-E. coli cases. The most common resistance pattern was sulfonamide/streptomycin in the LT-ST E. coli, tetracycline alone and sulfonamide/streptomycin in the ST E. coli and sulfonamide/streptomycin/tetracycline in the control strains. Eleven (16%) of 69 LT-ST E. coli were resistant to one or more antibiotics compared to 11 (32%) of 34 ST E. coli and 28 (37%) of 76 control strains. The difference in resistance between LT-ST and ST strains was of borderline statistical significance (p=06); the difference between the LT-ST and controls was highly significant (p<.01). Significantly more of the LT-ST E. coli (96%) were sensitive to tetracycline than the ST E. coli (80%) (p<.05 by chi-square analysis) or control strains (67%) (p<.001 by chi-square analysis).

Table 12

ANTIBIOTIC SENSITIVITY OF ENTEROTOXIGENIC

E. COLI AND CONTROL STRAINS

Antibiotic resistants‡	LT/ST(N=69)	ST(N=34)	LT(N=6)	Control(N=76)
S/ST	7	4		3
S/ST/TET	2	1		14
S/ST/TET/CH		1		
S/ST/TET/KA	1			
S/ST/TET/AMP			1	2
S/ST/AMP/KA	1			
S/ST/SM/CH/TET				4
TET		5		3
TET/S				1
TET/CH				1
Total:	11*#	11*	1	28#
Percent:	16	32	17	37

* chi square = 3.34, p=.06
\# chi square = 7.00, p<.01
‡ S = sulfonamide
ST = streptomycin
TET = tetracycline
KA = kanamycin
AMP = ampicillin
CH = chloromycetin

The O serogroups of the ETEC strains are shown in Table 13. Although the 69 LT-ST strains included 11 serogroups, 59 (85%) of the strains were in one of 4 serogroups - 06, 08, 078, 0115. In contrast the 34 ST strains were distributed among 15 O serogroups, and the two most common serogroups were 078, one of the 4 groups that included the majority of the LT-ST strains, and 0128, a classical enteropathogenic serotype. The 6 LT strains were distributed among 6 O serogroups.

Table 13

O GROUP AND ENTEROTOXIN TYPE OF E. COLI STRAINS

ISOLATED FROM 109 ENTEROTOXIGENIC E. COLI

O Group	Enterotoxin Type LT-ST	ST only	LT only
04		1	
06	9		
07		1	
08	19(1)	1	1
015	1		
020	1	3(2)	
025	2		
029		2	
034		2	
048			1
063	1	2	1
078	20(3)	5	
085	2	1	
096			1
0114		1	1
0115	11	2	
0123		1	
0126	1	1	
0128		5	
0148		1	
0159			1
OX2	1		
Neg 01-0163	1	3	
Rough		2	

(1) Includes one 08:060
(2) Includes three 020;0153
(3) Includes three 078:044 and one 078:0137

To determine the most expedient way to diagnose ETEC disease in these patients we compared results from toxin testing 10 individual isolates, a pool of 5 isolates and a pool of 10 isolates from each stool culture. Of the 109 E. coli cases detected on admission by testing 10 individual picks, the 10-pool was positive in 105(96%) and the 5-pool in 104 (95%). If only the first pick from each patient had been toxin-tested, 100 (92%) of the cases would have been identified; if the first 2 picks had been tested 104 (95%) would have been diagnosed.

Table 14

SENSITIVITY OF POOLING ISOLATES FOR LT TESTING*

Positive Picks (of 10)	Cases (no.)	Pool Positive No.	Pool Positive %
1	16	9	56
2	9	6	66
3	4	3	75
4	6	5	83
5	2	1	50
6	7	5	71
7	6	6	100
8	7	7	100
9	7	7	100
10	24	24	100

* Resting 48 hr culture in syncase broth; adrenal cell assay; 36 LT-ST and 2 LT only strains

Table 15

SENSITIVITY OF POOLING ISOLATES FOR ST TESTING *

Positive Picks (of 10)	Cases (no.)	Pool Positive No.	Pool Positive %
1	5	2	40
2	9	8	89
3	2	2	100
4	1	1	100
5	1	1	100
6	2	2	100
7	2	1	50
8	8	8	100
9	2	2	100
10	6	6	100

* Roller drum culture (22 rpm) in trypticase soy broth with 0.6% yeast extract

Results of the follow-up cultures in which most patients had less number of toxin-positive organisms provided an assessment of the sensitivity of pooling 10 isolates for LT and ST testing. As shown in Table 14 when 7 or more of the 10 isolates were positive, the 10-pool was always positive. The diagnostic yield gradually decreased as less picks became positive and was only 56% when 1 of 10 picks was positive. Similar data for ST testing were more limited because of the few number of cases (Table 15). Except for 1 pool of 7 positive picks, all pools with 3 or more of 10 ST-positive isolates were positive. Eight of nine pools with 2 positive isolates and 2 of 5 pools with 1 positive isolate were toxin-positive.

Results of testing directly for LT and ST in admission stools are shown in Tables 16 and 17. Significantly more stools known to contain LT-ST organisms were positive for LT by Method B (85%) than by Method A (51%) ($p<.01$). One of 6 cases with LT *E. coli* and 11 of 13 cholera cases were also positive. None of 33 ST cases, 22 cases with other diagnoses, or 29 cases with no known diagnosis were positive. Twelve (36%) of 33 LT-ST cases were positive for ST by Method A and 14 (44%) of 32 by Method B. Six (21%) of 29 ST cases were positive. No LT or cholera cases were positive, but 2 cases with mixed *Shigella* and rotavirus infections and 4 (17%) of 24 cases with no known diagnosis were positive.

Table 16

DETECTION OF LT IN STOOL SUPERNATANTS

Laboratory Diagnosis	Specimens (No.)	Positive No.	Positive %
Enterotoxigenic *E. coli*			
LT-ST (Method A)	35	18	51*
LT-ST (Method B)	33	28	85*
ST	33	0	0
LT	6	1	17**
Cholera	13	11	77**
Others	22	0	0
No diagnosis	29	0	0

* Difference in Method A and Method B significant, chi square = $p<.01$
** All positive stools run by Method B

Table 17

DETECTION OF ST IN STOOL SUPERNATANTS

Laboratory Diagnosis	Specimens (No.)	Positive No.	Positive %
Enterotoxigenic *E. coli*	33	12	36
LT-ST (Method A)	33	12	36
LT-ST (Method B)	32	14	44
ST	29	6*	21
LT	5	0	0
Cholera	13	0	0
Others	10	2**	20
No diagnosis	26	4***	15

* 1 of 7 positive by Method A and 5 of 21 positive by Method B
** Includes two with mixed shigellae and rotavirus infections positive by Method B
*** 0 of 6 positive by Method A and 4 of 20 by Method B

CONCLUSIONS

This study has demonstrated that in the time period under study a diagnosis could be made in 86% of adult cases of cholera-like diarrhea admitted to the Cholera Hospital with moderate to severe dehydration. In up to 62% of cases ETEC was responsible. In 13% of the cases more than 1 pathogen was isolated. More than 60% of the cases in which rotavirus, shigellae, or NAG organisms were isolated were mixed infections.

Of the ETEC cases LT-ST strains were isolated from about 66%, ST strains from about 33% and LT strains were found in less than 5%. Frequent clinical features of the E. coli cases included vomiting, muscle cramps, and abdominal pain; fever was documented in 20% of cases. Serum electrolytes were normal and stool electrolytes were similar to those seen in cholera. Antibody rise to LT was found in 61% of LT-ST cases but in none of the ST cases. LT-ST E. coli disease was clinically more severe than ST E. coli disease.

Tetracycline treatment of LT-ST disease resulted in a slightly earlier termination of diarrhea. Tetracycline had no effect on ST disease. Tetracycline did however shorten the duration of excretion of organisms in both diseases. Tetracycline had no significant effect on antibody response to LT. LT-ST strains had significantly less antibiotic resistance than ST or non-toxigenic strains; these data were not controlled for serotype. LT-ST strains but not ST strains appeared to be clustered in a few O serogroups.

For laboratory diagnosis of ETEC disease pooling was a less sensitive method than testing individual strains for LT when less than 6 of 10 isolates were enterotoxigenic and for ST when less than 3 were toxigenic. However, for work-up of admission specimens in our hospitalized patients there was no advantage to pooling as diagnosis could be made with nearly equal frequency by testing only 1 or 2 isolated colonies. As an alternative diagnostic approach we found LT could be detected in stool supernatants in 80-90% of our hospitalized cases. Testing for ST in supernatants appeared to be less sensitive and perhaps less specific.

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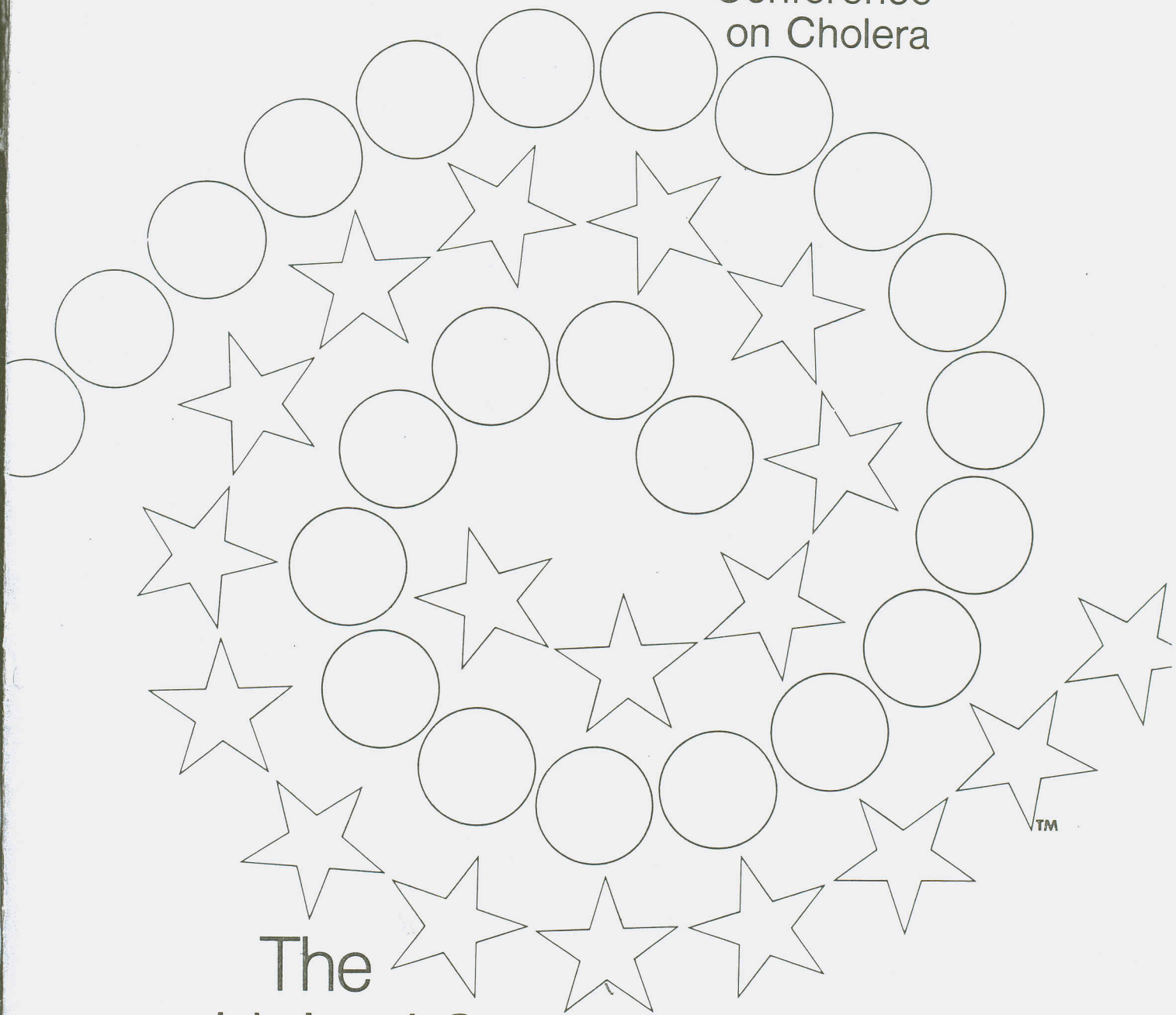

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