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EPIDEMIOLOGICAL SURVEY ON BACTERIAL, VIRAL AND PARASITIC AGENTS IN PATIENTS AFFECTED BY ACUTE ENTERITIS

W. MAGLIANI * [1], P. SOMENZI *, P. VALCAVI *, M. TCHERASSEN *, F. FANTI *, G. MOCCIA ** and C. CHEZZI *

* *Istituto di Microbiologia dell'Università degli Studi di Parma, Ospedale Maggiore, Via Gramsci, 14, 43100 Parma - Italy*
** *2° Laboratorio di Analisi Chimico-Cliniche, Ospedale Maggiore, Via Gramsci, 14, 43100 Parma - Italy.*

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During the period June 1983 - May 1984, faecal specimens from 797 patients with acute enteritis were examined for the presence of bacterial, viral and parasitic agents; 209 (26.2%) enteritic pathogens were identified, of whom 118 (35.4%) in 333 samples from the pediatrics wards. Bacterial agents were detected in 122 (15.3%), viruses in 63 (7.9%) and parasites in 25 (3.1%) of the 797 specimens. LT-producing *E. coli*, *Salmonella* and Rotavirus were the most frequent pathogens. Bacterial agents occurred most frequently in the summer and autumnal months, whereas viruses showed two peaks, the first one in summer due to cultivable agents, the second in winter to Rotavirus mainly.

INTRODUCTION

Acute enteritis is one among the most important causes of illness also in developed countries. However, few epidemiological studies have been carried out in Italy to determine the relevance of the possible different enteric pathogens associated with acute enteritis.

The purpose of this paper is to report the results of a 1-year study on the frequency of bacterial, viral and parasitic agents in patients with acute enteritis admitted to the General Hospital of Parma, Italy, during the period June 1983 - May 1984.

MATERIALS AND METHODS

Clinical specimens. - Faecal specimens from 797 patients with acute enteritis, 333 of whom admitted to the pediatric wards and 464 to the different care units of the Hospital, were examined for the presence of bacterial, viral and parasitic agents.

Bacteriological investigations. - Stool specimens were plated on Hectoen enteric, XLD, Mc Conkey and mannitol-salt agar media. Each sample, previously enriched overnight in sodium selenite broth, was also plated on Hectoen enteric and XLD agar.

Salmonella, Shigella, Staphylococcus spp. were identified by standard methods (4,8).

In order to test the production of heat-labile enterotoxin (LT), ten colonies with the typical appearance of *E. coli* were selected from each Mc Conkey agar plate; the production of LT toxin was determined by the Vero cells assay (9, 10).

[1] Corresponding author.

All enterotoxigenic isolates were finally identified by standard biochemical tests (4).

Each sample was also plated on blood agar with a supplemented selective medium for *Campylobacter* spp. (Butzler supplement; Oxoid Ltd.) (2) and incubated for 48 hours at 42°C in 5% oxygen and 10% carbon dioxide atmosphere. Isolates were Gram stained and tested for motility, oxidase and catalase production, ability to growth in glicine (1%), NaCl (3.5%), 2,3,-5-triphenyltetrazolium chloride medium (0.04%), nalidixic acid sensitivity (8) and morphology by electron microscopy.

For isolation of *Yersinia enterocolitica*, samples were plated on *Yersinia* selective medium (CIN medium; Oxoid Ltd.) and incubated at 32°C for 24 hours. Identification was performed by standard biochemical tests (8); serotyping was kindly provided by the Institut Pasteur, Paris.

Parasitological investigations. - Stools were microscopically examined for intestinal parasites and ova using a wet mounts in Lugol's solution and the migration inhibition factor technique (Marion Scientific Corp.).

Virological investigations. - Faecal specimens were examined for the presence of viral agents by direct electron microscopy, viral isolation in cell cultures and enzyme-linked immunosorbent assay for Rotavirus antigen.

Faecal suspensions (10%) in phosphate buffered saline (pH 7.2) were centrifuged two times for 15 minutes at 2700xg to remove heavy sediments.

The supernatants were tested by commercial Rotazyme EIA (Abbott Laboratories); in addition, formvar-carbon-coated 400 mesh grids for electron microscopy were prepared by the lyphogel method (11) and examined for a minimum of 20 minutes in a Philips EM 300 electron microscope.

For the isolation and identification of viruses by cell cultures, 10% suspensions of stool specimens in Earle's balanced salt solution were centrifuged at 10,000 rpm for 1 hr at 4°C. The supernatant fluid was treated with penicillin (200 units/ml) and streptomycin (200 μg/ml).

Isolation was attempted in VERO, RD, HEp-2 and human foetal diploid cells using the methods previously described by Lennette and Schimdt (7).

RESULTS

From the 797 faecal specimens examined for the presence of bacterial, viral and parasitic agents 209 (26.2%) enteric pathogens were identified, 118 of whom in samples from pediatric wards (Table 1).

Bacterial agents were detected in 122 (15.3%), viruses in 63 (7.9%) and parasites in 25 (3.1%) of the 797 specimens.

Among bacterial agents, *Salmonella*, *S. aureus* and LT-producing *E. coli* were the most frequent enteropathogens; the incidence of *C. jejuni* and *Y. enterocolitica* was found to be very low (Table 2). Serogroup 09, biotype 2, lysotype X 3 was the unique isolated *Y. enterocolitica* strain.

Rotavirus was the most frequently identified viral agent, mainly occurring in the samples from pediatric wards (Table 3); *Giardia lamblia* and *Enterobius vermicularis* were observed as the prevalent parasitic agents. (Table 4).

Associated agents were detected in 14 patients (Table 5).

TABLE 1.
Bacterial, viral and parasitic agents identified in 797 faecal specimens from patients with acute enteritis.

Agents	Pediatric wards		Other care units		Total	
	No.	%	No.	%	No.	%
Bacteria	62	18.6	60	12.9	122	15.3
Viruses	45	13.5	18	3.9	63	7.9
Parasites	12	3.6	13	2.8	25	3.1
Total	118	35.4	91	19.6	209	26.2

TABLE 2.
Bacterial agents isolated from 797 faecal specimens of patients affected by acute enteritis.

Agents	Pediatric wards		Other care units		Total	
	No.	%	No.	%	No.	%
Salmonella spp.	18	5.4	22	4.7	40	5.0
S. typhimurium	6		9		15	
S. infantis	4		4		8	
S. panama	1		4		5	
S. enteritidis	4				4	
S. derby	1		1		2	
S. berta			1		1	
S. newport			1		1	
S. give			1		1	
S. corvallis			1		1	
S. munchen	1				1	
Salmonella gr. G	1				1	
S. aureus	17	5.1	15	3.2	32	4.1
E. coli LT$^+$	21	6.3	20	4.3	41	5.1
C. jejuni	6	1.8	2	0.4	8	1.0
Y. enterocolitica			1	0.2	1	0.1

TABLE 3. — Viral agents identified by electron microscopy and/or tissue culture and/or enzyme-linked immunosorbent assay for Rotavirus antigen in 797 faecal specimens from patients affected by acute enteritis.

Viruses	Pediatric wards		Other care units		Total		E.M.*	T.C.**	E.I.A.○
	No.	%	No.	%	No.	%	No.	No.	No.
Adenovirus	8	2.4	2	0.4	10	1.3	9	5	—
Rotavirus	26	7.8	4	0.8	30	3.8	30	—	30
Picornavirus	9	2.7	11	2.3	20	2.5	—	20	—
Coronavirus	1	0.5	—	—	1	0.1	1	—	—
Astrovirus	1	0.3	1	0.2	2	0.2	2	—	—
Total	45	13.5	18	3.9	63	7.9	42	25	30

* Electron microscopy.
** Tissue culture.
○ Enzyme-linked immunosorbent assay.

TABLE 4.
Parasitic agents found in 797 faecal specimens from patients affected by acute enteritis.

Parasites	Pediatric wards		Other care units		Total	
	No.	%	No.	%	No.	%
Giardia lamblia	5	1.5	7	1.5	11	1.4
Enterobius vermicularis	7	2.1	1	0.2	8	1.0
Tenia saginata	—	—	2	0.4	2	0.2
Entamoeba coli	—	—	2	0.4	2	0.2
Enteromonas hominis	—	—	1	0.2	1	0.1
Total	12	3.6	13	2.8	25	3.2

TABLE 5.
Associated agents observed in the stool samples from patients affected by acute enteritis.

Patients	Wards	Agents
M.E.	Pediatric	*S. typhimurium, S. aureus*
M.V.	Pediatric	*S. typhimurium, S. aureus*
P.G.	Pediatric	*S. typhimurium, S. aureus*
G.A.	Non-pediatric	*S. berta, S. aureus*
B.M.	Pediatric	*S. infantis, C. jejuni*
B.S.	Pediatric	*S. typhimurium,* Coxsackievirus
S.D.	Non-pediatric	*S. typhimurium,* Echovirus
B.F.	Pediatric	*E. coli LT+,* Rotavirus
T.C.	Pediatric	*S. aureus,* Rotavirus, Adenovirus
M.G.	Non-pediatric	*S. derby, Giardia lamblia*
B.V.	Non-pediatric	*S. typhimurium, S. aureus Enteromonas hominis*
O.S.	Pediatric	*S. aureus, Enterobius*
L.S.	Pediatric	*E. coli LT+, Enterobius*
C.S.	Pediatric	*Giardia lamblia,* Coxsackievirus

The monthly incidence of enteric pathogens is shown in Figures 1, 2 and 3 for all samples, for 333 specimens from pediatric wards and 464 from different care units, respectively. Seasonal variations were noted in the incidence of bacterial and viral agents. Bacteria occurred most frequently in the summer and autumnal months, whereas viruses showed two peaks, the first one in summer, accounting for cultivable agents, the second in winter where mainly Rotaviruses were observed.

DISCUSSION

Present microbiological investigations lead us to identify enteric pathogens in 26% of the total faecal specimens; in samples from pediatric wards positive results raised to 35%.

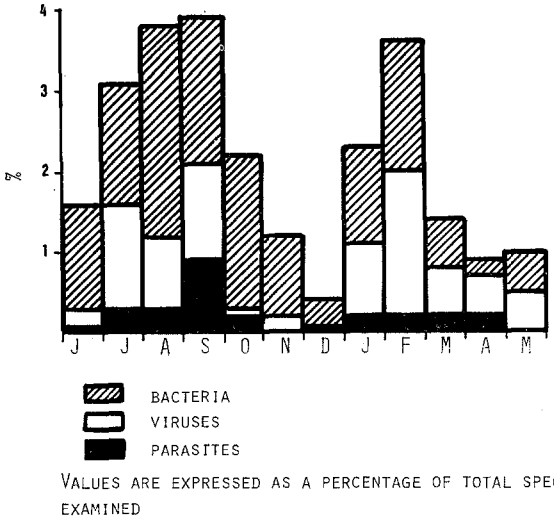
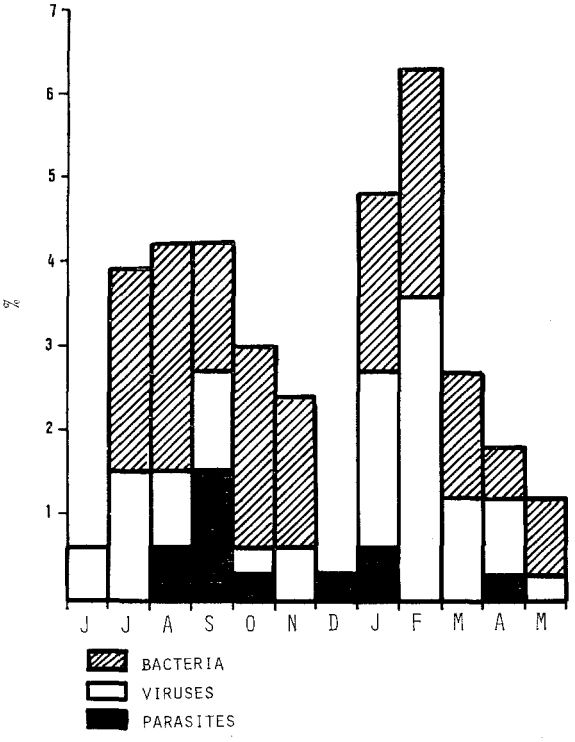


Figure 1. - The monthly distribution of bacterial, viral and parasitic agents among faecal specimens from patients with acute enteritis.

LT-producing *E. coli, Salmonella* and Rotavirus were the most frequently identified pathogens. This result confirms the importance of LT-producing *E. coli* as cause of enteritis in Parma (9).

The highest incidence of Rotavirus was observed during the winter in samples from pediatric wards, in agreement with previous studies in temperate countries (1, 5, 6).

The isolation of Enteroviruses and Adenoviruses by tissue culture methods doesn't seem to be significant, since these viruses appeare to be not important as aetiologic agents in acute enteritis (3).



Fiugre 2. - The monthly distribution of bacterial, viral and parasitic agents in 333 samples from patients admitted to pediatric wards.

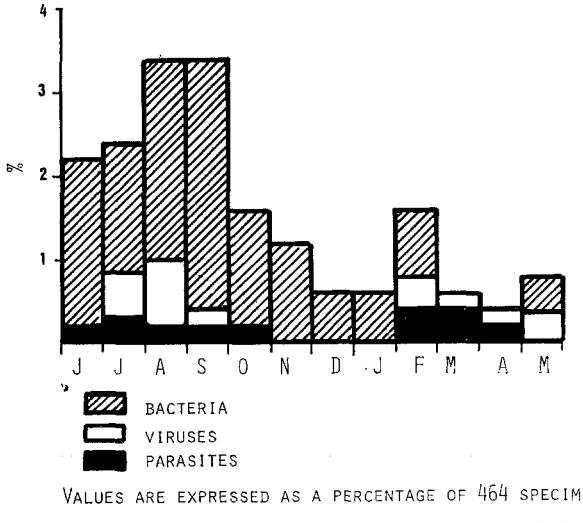


Figure 3. - The monthly distribution of bacterial, viral and parasitic agents in 464 faecal specimens from patients admitted to non-pediatric wards.

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