

ACUTE GASTROENTERITIS ASSOCIATED WITH ECHOVIRUS TYPE 1

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INTRODUCTION

A wide spectrum of clinical syndromes has followed human infection with different types of echovirus. Symptoms and signs which occur regularly include febrile illness associated with rash, pleurodynia, aseptic meningitis, outbreaks of diarrhoeal diseases and acute respiratory disease, depending upon the target organ of attack (McLean, 1966). For these reasons, clinical symptoms have not been considered satisfactory as a basis of classification. Confirmation, however, must be obtained in the laboratory.

Late in March 1971, a syndrome marked by diarrhoea with mucoid, bloody or watery stools, fever with chills and abdominal pain was experienced by many of the patients in Tri-Service General Hospital, Taipei, Taiwan. Virologic studies were immediately carried out.

MATERIALS AND METHODS

Specimen Collection: Two hundred and ninety-four in-patients of TSG Hospital under 50 years of age with acute diarrhoea were selected for study, and 184 apparently healthy persons closely matched as to time, age and socioeconomic conditions were used as a control. Single rectal swab was collected from all study cases as they reported in with diarrhoea. Similar swab was also collected from the control persons on the same day. The swab was preserved in a vial containing 2 ml. of brain-heart-infusion with 0.5% bovine

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albumin, 200 units of penicillin G, 200 micrograms of streptomycin and 50 micrograms of mycostatin per milliter, and stored in a -70°C freezer until required for testing. A brief clinical history and physical examination were recorded, but patients' serum was not possible to obtain for antibody study.

Virus Isolation and Identification: Virus isolation and identification were carried out in primary Taiwan monkey kidney cells by methods which have been reported previously (Wang *et al.*, 1970, 1973). All cultures were under observation for 28 days through one more blind passage before being considered negative.

Identification of isolated virus was made by cytopathic effect (CPE) of cell culture neutralization procedure (Lim and Benyesh-Melnick, 1960), by using 100 TCID₅₀ of viruses against the type specific hyperimmune rabbit antisera of poliovirus types 1-3, echovirus type 1-32, coxsackievirus A-9 and B-1 to 6. Four-tenth of a ml of mixtures of equal virus and antisera were incubated at 37°C for 2 hours, and 0.2 ml of the material inoculated into 2 monkey kidney culture tubes. Cultures were observed for 7 days and final reading taken 48 hours after the appropriate virus controls showed complete CPE. Virus not neutralized by these antisera remained unclassified.

RESULTS

From late March 1971 to February 1972, rectal swabs were collected from 294 diarrhoeal patients and 184 healthy persons

Table 1

Age and sex distribution of 294 diarrhoeal patients and 184 healthy persons.

Age group (year)	Diarrhoeal patients			Healthy persons		
	Male	Female	Total	Male	Female	Total
< 2	26	10	36	8	5	13
2 - 10	103	72	175	59	37	96
11 - 20	41	13	54	21	10	31
21 - 30	12	2	14	10	2	12
31 - 40	8	1	9	8	3	11
41 - 50	4	2	6	18	3	21
Total	194	100	294	124	60	184

Table 2

Comparison of enteroviruses isolated from 294 diarrhoeal patients and 184 healthy persons studied from March 1971 to February 1972.

Virus types	Virus isolated from			
	Diarrhoeal patients		Healthy persons	
	No.	%	No.	%
Polioviruses	12	4.08	0	0
Type 1	2	0.68	0	0
Type 2	4	1.36	0	0
Type 3	6	2.04	0	0
Echoviruses	25	8.50	2	1.09
Type 1	16	5.44	1	0.54
Type 7	1	0.34	0	0
Type 12	1	0.34	1	0.54
Type 14	1	0.34	0	0
Type 17	1	0.34	0	0
Type 19	2	0.68	0	0
Type 20	1	0.34	0	0
Type 22	2	0.68	0	0
Coxsackieviruses	2	6.68	1	0.54
Type B-4	1	0.34	0	0
Type B-6	1	0.34	1	0.54
Unclassified viruses	4	1.36	2	1.09
Total	43	14.63	5	2.72

(Table 1). From these rectal swabs, 43 strains of enteroviruses including unclassified types were isolated from the diarrhoeal patients and 5 only isolated from the healthy persons. A Chi-square test showed that this difference is highly significant ($P < 0.005$). Among the 43 enteroviruses isolated from diarrhoeal patients 25 were identified as echoviruses, 12 as polioviruses, 2 as coxsackieviruses B, and 4 as unclassified types. The 5 strains of enteroviruses isolated from 184 healthy persons, consisted of 1 strain of coxsackieviruses B and 2 each of echoviruses and unclassified viruses (Table 2). The viral isolation rate of polioviruses ($P < 0.01$) echoviruses ($P < 0.005$) and especial echovirus type 1 ($P < 0.005$) between these 2 groups is also highly significant.

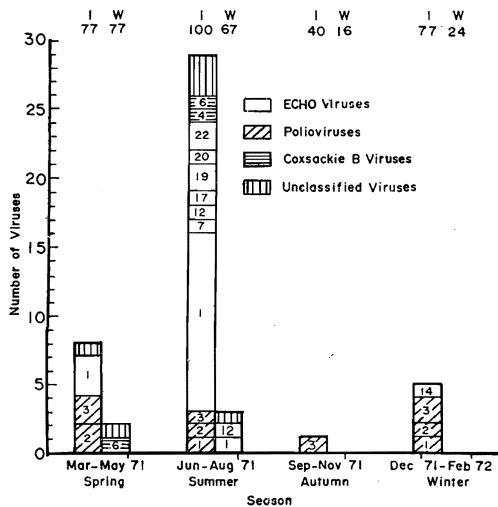


Fig. 1—Seasonal distribution of viruses isolated from 294 patients with acute gastroenteritis and 184 healthy persons. I = Patients with acute gastroenteritis. W = Healthy persons.

The seasonal distribution of types of enterovirus is shown in Fig. 1. Ten strains (8 from 77 diarrhoeal patients and 2 from same number of healthy persons) of enterovirus were isolated in spring, 32 strains (29 from 100 diarrhoeal patients and 3 from 67 healthy persons) in summer, 1 strain from 40 diarrhoeal patients in autumn, and 5

strains from 77 diarrhoeal patients in winter; no virus was isolated from 16 and 24 healthy persons in autumn and winter respectively.

The number and types of enterovirus are shown in Table 2 and Fig. 1. The number of positive isolations in diarrhoeal patients was highest in summer.

The clinical history and physical examination were conducted in 16 diarrhoeal patients from whom echovirus type 1 was isolated. Half of the cases are aged 2-10 (Table 3). The principal symptoms and signs are shown in Table 4.

Table 3

Age and sex of 16 diarrhoeal patients from whom echovirus type 1 was isolated.

Age group (year)	No. of cases		
	Female	Male	Total
< 2		1	1
2 - 10	3	5	8
11 - 20	2	3	5
21 - 30		1	1
31 - 40			0
41 - 50		1	1
Total	5	11	16

Table 4

Symptoms and signs of 16 diarrhoeal patients from whom echovirus type 1 was isolated.

Symptoms & signs	No. cases
Fever (38-40°C) with chills	16
Abdominal pain	12
Stool:	
Mucoïd	10
Bloody	8
Watery	5
Dehydration	3
Vomiting	3

DISCUSSION

There were 27 strains of echoviruses including types 1, 7, 12, 14, 17, 19, 20 and 22 isolated from the rectal swabs. Twenty-five were isolated from diarrhoeal patients and 2 only isolated from healthy persons. The isolation rate of echovirus from diarrhoeal patients was 8.5% in diarrhoeal patients and 1.09% from healthy persons. Among the 27 strains isolated, 17 were type 1, and 16 of them were isolated from diarrhoeal patients and 1 strain only from healthy persons. The isolation rate was ten times higher in patients than that in healthy persons. The higher significant isolation rate ($P < 0.005$) in diarrhoeal cases would suggest that echovirus type 1 played an important role in acute gastroenteritis in Taipei area, during the study period, from June to August 1971.

Echovirus type 19, was first isolated from faeces of an Ohio child with diarrhoea (Ramos-Alvarez and Sabin, 1958), and from cerebrospinal fluid of a man who developed aseptic meningitis in Nova Scotia during 1959 (Faulkner and Ozere, 1960). In this study 2 strains of the echovirus type 19 were isolated from 2 diarrhoeal patients, while echovirus type 20, was recovered from faeces of one case with diarrhoea and fever. The prototype strain of this echovirus was first isolated from a febrile child in the nursery of a Washington D.C. welfare institution (Junior Village), during 1956 (Rosen *et al.*, 1958).

Acute gastroenteritis has been often linked with echovirus than with other enterovirus. Type 11, 14, 18, 22 and some of the higher types had been recovered from the faeces of patients during epidemic of gastroenteritis (Lepine *et al.*, 1960; Fenner and White 1971). Eichenwald *et al.*, (1958) identified echovirus type 18 during an outbreak of diarrhoea in a nursery of premature babies, and subsequently in a ward for sick children under 5 months of age. Ramos-Alvarez and

Sabin (1958) presented evidence that several echovirus types 2, 6, 7, 8, 11, 12, 14 may be implicated in summer diarrhoeal diseases of infants and young children. Bergamini and Bonettie (1970) and Klein *et al.*, (1960) also reported acute gastroenteritis caused by echovirus type 11. But these types of echovirus has not been a consistent finding. The present study implicated echovirus type 1 as the causal agent in acute gastroenteritis in children and young adults. The echovirus types 7, 12, 14, 19, 20 and 22 were also isolated from diarrhoeal patients. In association with many echoviruses and coxsackieviruses, no disease entities have been known.

SUMMARY

The incidence of gastroenteritis associated with various enteroviruses was determined in 294 diarrhoeal patients for a year in Taipei, Taiwan, Republic of China, also in a simultaneously control group of healthy persons without diarrhoea matched for age and socioeconomic status.

In the summer of 1971, sixteen strains of echovirus, type 1 were isolated from the rectal swabs of patients with acute gastroenteritis, and one from healthy persons. Most of the patients from whom the virus was isolated suffered from the following principal symptoms: sudden onset, fever with chills, discharges of watery, bloody and mucoid diarrhoea of brief duration, vomiting dehydration and abdominal cramps.

Formerly echovirus, type 18, was commonly associated with acute gastroenteritis. Other workers presented evidence that several, echovirus types: 2, 6, 7, 8, 11, 12, 14, 19 and 22, might be implicated in summer diarrhoeas of infants and young children. The present study implicates echovirus type 1 in acute gastroenteritis in children and young adults. Other types: 7, 12, 17, 19, 20 and 22, of echovirus were also isolated from diarrhoeal patients.

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REFERENCES

- BERGAMINI, F. and BONETTIE, F., (1960). Epidemic episode of acute gastroenteritis from echovirus type 11 in a founding hospital. *Boll. Ist. Sieroter*, Milan., 39 : 510.
- EICHENWALD, H.F., ABABIO, A., ARKY, A.M. and HARTMAN, A.P., (1958). Epidemic diarrhoea in premature and older infants caused by echovirus type 18, *J.A.M.A.*, 166 : 1563.
- FAULKNER, R.S. and OZERE, R.L., (1960). Aseptic meningitis due to echovirus type 19 infection. *New Eng. J. Med.*, 263 : 551.
- FENNER, F. and WHITE, D.O., (1971). *Echovirus in Medical Virology*, pp. 279-282, Academic Press. New York and London.
- KLEIN, A.O., LERNER, A.M. and FINLAND, M., (1960). Acute gastroenteritis associated with echovirus type 11, *Amer. J. Med. Sci.*, 240 : 749.
- LEPINE, P., SAMILE, J., MAURIN, J., DUBOIS, O. and CARRE, M.C., (1960). Isolation of the echovirus type 14 in the course of a nursery epidemic of gastroenteritis. *Ann. Inst. Pasteur*, 99 : 161.
- LIM, K.A. and BENYESH-MELNICK, M., (1960). Typing of viruses by combination of antiserum pools. Application to typing of enteroviruses (coxsackie and echovirus). *J. Immunol.*, 84 : 309.
- MCLEAN, D.M., (1966). Coxsackievirus and echovirus. *Amer. J. Med. Sci.*, 251 : 351.
- RAMOS-ALVAREZ, M. and SABIN, A.B., (1958). Enteropathogenic viruses and bacteria. Role in summer diarrhoeal diseases of infancy and early childhood. *J.A.M.A.*, 167 : 143.
- ROSEN, L., JOHNSON, J.H., HUEBNER, R.J. and BELL, J.A., (1958). Observations on a newly recognized echovirus and a description of an outbreak in a nursery. *Amer. J. Hyg.*, 67 : 300.
- WANG, H.C., TAI, F.H., CHEN, S.T. and WANG, K.Y., (1970). Viruses causing diarrhoeal diseases in infants and children. *Chinese J. Microbiol.*, 3 : 77.
- WANG, H.C., HUNG, S.C. and TAI, F.H., (1973). Studies on viral diarrhoea in Taiwan. 1. Viruses isolated from diarrhoeal cases and control subjects. *Chinese J. Microbiol.*, 6 : 63.