NOSOCOMIAL ROTAVIRUS GASTROENTERITIS IN A NEONATAL NURSERY

by

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A NEWLY recognised virus has recently been seen in duodenal mucosa from children with acute non-bacterial gastroenteritis (Bishop *et al,* 1973). Subsequently, Flewett, Bryden and Davies (1973) found the virus in negatively stained faecal extracts. Similar viruses have since been detected throughout the world and there is convincing evidence that this virus, provisionally called rotavirus, is an important aetiological agent in acute childhood gastroenteritis. We describe a hospital acquired (nosocomial) outbreak of infective diarrhoea in a newborn maternity unit. This is the first report of a rotavirus outbreak in Ireland.

PATIENTS AND METHODS

Clinical details

The outbreak began on 20th November 1974 in the neonatal unit of the Royal Maternity Hospital, Belfast, and new cases of gastroenteritis occurred until 10th December 1974. During this period 61 babies passed through the unit. Fourteen babies, six of whom were male, were affected (Table 1) and this included two sets of monozygotic twins, one of each sex. All infants had diarrhoea but in only two was there associated vomiting; this occurred at the onset of the illness. Diarrhoea was watery and often green in colour at onset, but it was variable both in frequency and duration. In the group as a whole the mean duration of symptoms was 8.3 days, while in the seven babies of birthweight less than 2.0 kg it was 12.6 days; in heavier infants it was only 4.0 days. Eight infants (five of them less than 2.0 kg birthweight) took more than 2.5 weeks to regain their birthweight. In three there was an electrolyte disturbance, two had hyponatraemia, one had hypokalaemia and two an elevated urea. Eight required intravenous fluid therapy, in some for up to 7 days. In two infants, birthweights 1.8 and 1.2 kg, there was some sugar intolerance, although this was not critically studied. Upper respiratory tract infection was not observed in any of the affected infants. Ultimately all infants recovered and there were no deaths.

TABLE 1: RELATIONSHIP OF DIARRHOEA TO ROTAVIRUS INFECTION

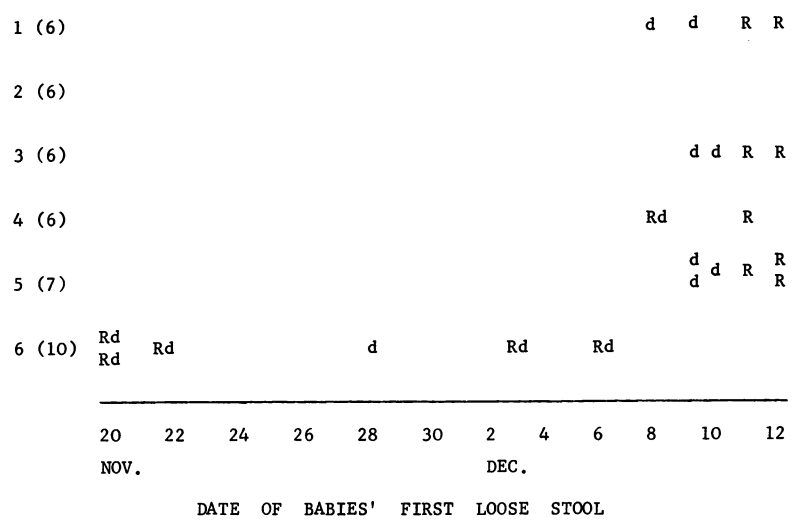
Rotavirus infection	*Diarrhoea mild/severe*	*No diarrhoea*	*Total*
Confirmed	0/6* (3)	8 (4)	14
Not confirmed	6/2* (3)	9 (3)	17

*One infant in each group with vomiting. Number of males shown in brackets.

The neonatal unit has a capacity for 41 babies and is divided up into six cubicles. Diarrhoea commenced in cublcle 6, an area where 10 babies can be nursed, and was confined to this area for 17 days, although six infants had developed symptoms (Table II). Between 8th and 10th December babies in cubicles 1, 3, 4 and 5 began having diarrhoea in spite of great care being taken to confine the infection to cubicle 6. The usual methods of gowning, hand washing and keeping each baby's feeding utensils strictly for the individual concerned were employed. The babies were all bottle fed. The nursing and medical staff involved in the daily care of these infants were also responsible for those in the other five cubicles of the nursery. The only area with no infected babies was cubicle 2. None of the mothers questioned had had recent diarrhoea or vomiting, nor did any of the staff develop symptoms. The outbreak was terminated by closing the unit to further admissions on the 12th December.

TABLE II

TEMPOROSPATIAL SPREAD OF ROTAVIRUS INFECTION IN NURSERY



DATE OF BABIES' FIRST LOOSE STOOL

The origin of infection in our infants was not discovered, although the source may have been a baby initially thought to have subacute bowel obstruction admitted to the nursery from a postnatal ward on 17th November. This infant

had mild diarrhoea, vomiting and abdominal distension. Bacterial cultures were negative. Symptoms abated within 48 hours and the baby was discharged. Three days later, however, infants in the same nursery cubicle of the unit developed diarrhoea. Moreover, two weeks after discharge an infant who had been in an adjacent cot to this baby in the postnatal ward was admitted to the local fever hospital with gastroenteritis. Neither was examined for rotavirus infection, but no other cause for the symptoms was found.

LABORATORY STUDIES

On 11th and 12th December suitable faecal samples were obtained from 29 infants and blood samples were taken from 11 infants. Both faeces and sera were stored at —20°C.

Bacteriological culture of faeces was carried out on blood agar and MacConkey's agar. Electron microscopy of five faecal samples was performed after differential centrifugation and negative staining, using the method of Bishop *et al,* 1974. For rotavirus culture faecal extracts were centrifuged on to coverslip monolayers of LLC-MK2 cells, and after incubation the cells were stained with a fluorescein isothiocyanate conjugated (FITC) rabbit anti-rotavirus serum to detect rotavirus antigen (Bryden *et al,* 1977). Faeces were tested after 18 months storage at —20°C. Rotavirus antiserum was prepared as follows. Purified Nebraska calf diarrhoea rotavirus in Freund's complete adjuvent was injected intramuscularly into each limb of a rabbit. After one month a further dose was given intravenously and the antiserum was removed a week later. Rotavirus specific immunoglobulins IgM and IgG: the infants' sera and FITC anti-human IgM or anti-human IgG were absorbed with LLC-MK2 cells. In addition, the infants' sera were inactivated at 56°C for 30 minutes. For detection of rotavirus specific IgM, the sera were further absorbed with *Staphylococcus aureus* protein A and heat aggravated human immunoglobulin, using the method of Thompson *et al,* 1975. Rotavirus specific IgM or IgG was measured by the indirect fluorescent antibody technique with suitable controls. Coverslip cultures of acetone fixed LLC-MK2 cells infected with Nebraska calf diarrhoea virus were covered with FITC anti-human IgM or IgG for one hour at 37°C. After further washing, the coverslips were mounted and viewed with a fluorescence microscope.

RESULTS

All bacterial cultures were negative for faecal pathogens, whether from infants with or without diarrhoea. Rotaviruses were detected by electron microscopy in three out of the five faeces examined. Of faecal samples from 29 infants, rotaviruses were cultured from 11 infants; 18 were negative. Rotavirus specific IgM was detected in sera from five of 11 children (Table 3). Of these, nine had faeces tested for rotavirus which was detected in two infants who had rotavirus specific IgM and in one infant who had no detectable specific IgM. Three babies had rotavirus specific IgM in their sera but rotavirus was not found in the faeces. Rotavirus specific IgM was negative in a further five infants, in three of whom

rotavirus was not detected in faeces. Two babies had no faeces samples tested. Rotavirus specific IgG was found in the acute phase sera of six out of six children tested. In two of these rotavirus was not cultured nor was rotavirus specific IgM found in serum.

TABLE 3: RELATIONSHIP OF ROTAVIRUS STOOL CULTURE TO ROTAVIRUS SPECIFIC IgM

Rotavirus stool culture (*Twenty-nine babies*)	*Rotavirus Specific IgM* (*Eleven babies*)	
	Positive	*Negative*
11 Positive	2	1
18 Negative	3	5

Fourteen of 31 children (45 per cent) had laboratory confirmation of rotavirus infection in infants who had diarrhoea, and those who remained well is shown in Table 1. Rotavirus infection was confirmed in six of eight infants with severe diarrhoea, but in none of the six babies who had mild diarrhoea. In those with diarrhoea there was a confirmation rate for rotavirus infection of 43 per cent. Of particular interest were the eight of 17 (47 per cent) asymptomatic infants who were found to be infected with rotavirus on the 11th and 12th December. Of the two pairs of twins with diarrhoea, rotavirus infection was confirmed in the male twins but not in the female pair.

DISCUSSION

In this outbreak rotaviruses were strongly associated with gastroenteritis since faecal bacterial pathogens were excluded. The fluorescent antibody method for detecting rotavirus antigen in infected cells was more convenient than direct electron microscopy of faeces and is known to give similar isolation rates (Bryden *et al,* 1977). This should not be influenced by storage of faeces at —20°C because the virus is thermostable. Moreover, the detection of rotavirus specific IgM in five out of 11 infants tested indicates recent infection with rotaviruses since IgM, unlike IgG, does not pass the placenta (Davidson *et al,* 1975). Some of the sera, however, may have been taken too early for detection of rotavirus specific IgM.

Diarrhoea was the prime clinical manifestation. This was marked in eight babies (57 per cent), which included five babies less than 2 kg birthweight, indicating that smaller, less mature infants are more at risk. Vomiting occurred in only two of the 14 babies with diarrhoea but may be a more common symptom in older children (Shepherd *et al,* 1975). Upper respiratory signs were not noted in this outbreak but have been documented in 42 per cent of older babies with rotavirus gastroenteritis (Carr, McKendrick and Spyridakis, 1976).

Outbreaks of gastroenteritis in newborn nurseries have been reported from various cities: London, Glasgow, Sydney, Melbourne and Paris (Chrystie *et al,* 1975; Madeley and Cosgrove, 1975; Murphy, Albrey and Hay, 1975; Cameron *et al,* 1975; and Weekly Epidemiological Record, 1977a). The incidence of rota-

virus infection in the community is highest during winter months, and hospital outbreaks may reflect this, but nosocomial rotavirus infections are found at other times of year (Chrystie *et al,* 1975). During this outbreak in the winter of 1974 there was a marked increase in rotavirus infections reported in England and Wales (Weekly Epidemiological Record, 1977b).

Of the 31 infants tested, whether symptomatic or not, rotavirus infection was proven in 45 per cent. This is the same rate of confirmation as reported by Chrystie *et al,* 1975. Of greater interest were the eight of 17 asymptomatic infants who were infected with the virus, although there was circumstantial evidence that an infant with gastroenteritis may have started this outbreak. Other possibilities include an asymptomatic mother or member of staff who may have been excreting rotavirus. This is particularly relevant since it has been reported that only 11 per cent of adult family contacts of infected children were symptomatic, although 41 per cent had serological evidence of infection. These adults also had rotavirus antibody in acute phase sera, suggesting that previous childhood infection with the virus may modify a serious infection to a mild or inapparent one (Kim *et al,* 1977).

The mode of spread of rotaviruses through the cubicles is also unknown, although similar rapid spread has been reported in other neonatal outbreaks. The temporal and spatial spread is characteristic of a nosocomial infection. Infectious droplets are created during changes of napkins, hence airborne infection is possible, but it is more likely that the virus was transmitted on the hands of attending staff despite meticulous hand-washing. This is probable because rotavirus is thermostable, with up to ten thousand million viruses present per gram of faeces, so that transmission on the hands is virtually inevitable (Lancet, 1976). Furthermore, asymptomatic attending staff may have been excreting rotaviruses after being infected by the patients.

Since the newborn gut is microbiologically sterile, it seems likely that rotavirus caused the first infection in these infants. The newborn is therefore analagous to the immune adult in that most babies will have circulating transplacental rotavirus specific IgG. Indeed, rotavirus specific IgG was found in the acute phase sera of six infants who were tested, but it clearly did not provide absolute protection against infection. Adults who have had rotavirus infection during childhood, when reinfected, also mount a rapid secondary rotavirus specific IgA response in the gut which may modify symptoms. The newborn, however, is at a disadvantage because virus induces a primary IgA response in the gut which is slower and may be further delayed by immunological immaturity of the host. Breast-fed infants, on the other hand, ingest antibody which is predominantly IgA and presumably contains a rotavirus specific portion. Our babies were all fed modified cow's milk formula, but in a similar outbreak in a London maternity hospital, rotavirus was isolated less frequently from breast as compared to bottle-fed babies (Chrystie *et al,* 1975). There is as yet no explanation for the fact that eight babies infected with rotavirus were asymptomatic, although such patients are common in many other virus infections.

We would recommend, therefore, that in future those infants with infective diarrhoea and adjacent asymptomatic babies be immediately isolated from other infants. All of these should be cared for by medical and nursing personnel who have no responsibility for other infants in the nursery. Disposable gowns and gloves should be worn during handling. Finally, potentially infected infants should be tested for rotavirus infection before being admitted to the general nursery area.

SUMMARY

A nosocomial outbreak of rotavirus gastroenteritis is described in a neonatal nursery. Fourteen infants had diarrhoea but only two infants had associated vomiting. Six of 14 infants (43 per cent) with diarrhoea and eight of 17 (47 per cent) infants without symptoms had evidence of rotavirus infection. The infection was confined to one cubicle for 17 days, then spread rapidly into four other cubicles. Suggestions are made for containing future outbreaks.

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ADDENDUM

Since this paper was prepared, rotavirus antibody has been found in human colostrum (Thouless, Bryden and Flewett, 1977; Simhon and Mata, 1978).

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