

## Detection of rotavirus in respiratory secretions of children with pneumonia

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SEVERAL ASPECTS OF THE EPIDEMIOLOGY OF ROTAVIRUS, a major cause of pediatric diarrhea, suggest that transmission occurs via the respiratory tract, in addition to the fecal-oral route.<sup>1-4</sup> Recent epidemiologic studies have shown that respiratory symptoms frequently precede or concurrently occur with diarrhea caused by rotavirus.<sup>1,2</sup> Laboratory studies of patients with gastroenteritis have not found rotavirus in respiratory tract secretions<sup>5,6</sup>; the presence of rotavirus in the secretions of patients with respiratory disease has not been studied. Our study was performed to determine whether rotavirus has an etiologic role in pediatric pneumonia.

### MATERIALS AND METHODS

Children, 5 years of age or younger, hospitalized for pneumonia between January 1 and March 31, 1980, at the Social Security Hospital, Panama City, Republic of Panama, were enrolled in the study. Pneumonia was documented radiographically (infiltrates, lobar consolidation, or pleural effusion), and only patients who had been symptomatic for 30 days or less were considered. The degree of respiratory distress was classified as none (no respiratory difficulty except tachypnea), mild (presence of minimally visible intercostal retractions or nasal flaring), moderate (presence of intermediate intercostal retractions), or severe (presence of severe intercostal retractions or cyanosis).

Laboratory specimens obtained within two hours of admission included nasopharyngeal swab, tracheal aspirate (nasotracheal in children older than 6 months, orotracheal in younger patients), and stool swab. Specimens were placed in phosphate-buffered saline and frozen at -70° C

until assayed for rotavirus and adenovirus by the ELISA technique.<sup>6</sup> Specimens were also placed in standard holding medium, held on wet ice, and inoculated within three hours onto HeLa cells and held for 14 days, Vero cells maintained for 21 days and fetal tonsil cells for four weeks.<sup>7</sup> Respiratory tract specimens were also inoculated into the allantoic and amniotic cavities of embryonated chicken eggs to screen for influenza<sup>8</sup> and onto IUDR-treated McCoy cells to assay for chlamydia.<sup>9</sup>

ELISA	Enzyme immunosorbent assay
IUDR	5-Iodo-2-deoxyuridine
PPIO	Pleuropneumonia-like organism

Respiratory specimens for mycoplasma isolation were cultured on PPIO agar as well as PPIO broth.<sup>10</sup> One milliliter blood was obtained at admission for routine bacterial cultures. An additional 3 ml blood was obtained at admission and two weeks after discharge to test for rotavirus antibodies by ELISA.<sup>6</sup>

### RESULTS

We obtained informed consent and successfully followed up on all 45 children eligible for inclusion in this study. An etiologic agent for the pneumonia was identified from the respiratory secretions in 22 of 45 (49%) patients (Table 1). Blood cultures for bacterial pathogens were negative in all patients.

Rotavirus was detected in respiratory tract secretions from four patients: in all three sites (one patient), in tracheal aspirate and stools (one), in nasopharyngeal secretions and stools (one), and in the tracheal aspirate only (one). Mycoplasma was also isolated from the tracheal aspirate of one of the four patients, but the other three had only rotavirus present in their respiratory tract secretions. Clinical presentation on admission was similar for those with and without respiratory tract rotavirus (Table II). The four patients with rotavirus were 3, 22, 24, and 27 months of age, respectively. The 3-month-old infant had mild diarrhea prior to hospitalization, and his stool also contained rotavirus. None of the other three patients had diarrhea before or during hospitalization. Two of the

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**Table I.** Organisms identified among 45 patients admitted with pneumonia

Organism	Tracheal aspirate	Nasopharyngeal aspirate	Rotavirus in tracheal or nasopharyngeal aspirate	Stool
Rotavirus	3	2	4	5
Adenovirus	1	3	4	4
Coxsackie B <sub>1</sub> virus	1	0	1	0
Coxsackie B <sub>2</sub> virus	1	1	1	1
Herpes simplex	1	1	1	0
Rhinovirus	2	2	3	0
Cytomegalovirus	0	1	1	0
Mycoplasma	7	0	7	—
Chlamydiae	0	0	0	—
Influenza	0	0	0	0
Respiratory syncytial virus	0	0	0	—
None	29	35	23	35

**Table II.** Clinical characteristics of patients

	With rotavirus-associated pneumonia (n = 4)		Without rotavirus-associated pneumonia	
		(n = 41)	(%)	
Age (mo)				
Mean	19	19		
Range	3 to 27	1 to 59		
History of fever	3	40	98	
Duration of fever prior to admission (days)				
Mean	7.7	3.5		
Range	1 to 19	1 to 15		
History of diarrhea	1	4	10	
History of vomiting	0	2	5	
History of cough	4	39	71	
Duration of cough prior to admission (days)				
Mean	4.5	6.2		
Range	2 to 10	1 to 30		
History of rhinorrhea	3	32	71	
Duration of rhinorrhea prior to admission (days)				
Mean	2.7	5.7		
Range	2 to 3	1 to 30		
Respiratory rate on admission				
Mean	50	42		
Range	44 to 58	20 to 60		
Respiratory distress				
None	0	12	29	
Mild	4	27	66	
Moderate	0	2	5	
Severe	0	0		

Differences between groups not significant by two-tailed *t* test or  $\chi^2$  test.

patients with rotavirus had received antibiotics prior to admission. Three of the four had a history of fever for one, three, and 19 days, respectively, prior to admission, and all four had been coughing for from two to 10 days. Three had a history of rhinorrhea. All had mild respiratory distress on

admission. On roentgenographic examination of the chest, two had lobar consolidation of the lung (left lower lobe in one, right upper lobe in one), one had bilateral diffuse parenchymal infiltrate of both lung fields, and one had a diffuse parenchymal infiltrate of the right middle lobe.

In two additional patients, aged 26 months and 17 months, respectively, rotavirus was detected only in the stools. In one of these patients, coxsackie B<sub>1</sub> virus also was detected from the tracheal aspirate. Neither patient had symptoms of diarrhea.

Blood for convalescent titers was available from 25 (56%) of the children. Two of the four patients with rotavirus in their respiratory tract secretions had fourfold rotavirus antibody titer rises. Convalescent sera were not available in the other two. One of the two patients with fecal rotavirus also had a fourfold rise in antibody titers to rotavirus; convalescent serum was not available in the other. None of the other patients had significant rise in antibody titers to rotavirus during convalescence.

## DISCUSSION

Our data demonstrate that rotavirus may be found in the respiratory tract secretions of children. The occurrence of both rotavirus isolates and specific titer rises in two patients suggests that rotavirus may occasionally be associated with pneumonia. Previous studies<sup>2,3</sup> have failed to find rotavirus in respiratory tract secretions of patients with gastroenteritis, although many such patients also have had respiratory symptoms. At least three factors might explain this discrepancy. (1) To our knowledge, no detailed etiologic studies have been published concerning pediatric pneumonia in Latin America, and geographically different determinants of rotavirus colonization of the respiratory tract may exist. (2) The previous studies involved patients with diarrhea, not patients with pneumonia; rotavirus may have a different anatomic distribution in the two diseases. (3) Other host differences, such as age, could determine

whether rotavirus infects the gastrointestinal or respiratory tract.

It is possible that rotavirus was only colonizing the respiratory tract in our patients and that the pneumonia was caused by a bacterial pathogen: two of the patients with rotavirus had received antibiotics prior to admission, and we did not perform bacterial cultures on the respiratory tract specimens. However, the occurrence of a fourfold rise in antibody titers to rotavirus in two of the four children with rotavirus in the respiratory tract suggests that rotavirus was the probable cause of their pneumonia. It is unfortunate that convalescent blood was not available for the other two. Mycoplasma was detected in the tracheal aspirate of one of these patients, and may have been the cause of that child's pneumonia.

In addition, we did not look for rotavirus antigen in the respiratory tract secretions of children hospitalized for diseases other than pneumonia. To be certain that rotavirus can cause pneumonia in children, a larger number of children with the disease and their age-matched controls need to be studied.

Our data demonstrate that pneumonia of childhood may occasionally be associated with detection of rotavirus antigen in the respiratory tract. It also supports previous evidence<sup>1,2,4</sup> that rotavirus may be transmitted by the respiratory route. Further studies are needed to outline the incidence of rotavirus in other respiratory tract infections and to study the clinical course of respiratory illness associated with rotavirus.

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## Chronic active Epstein-Barr virus infections in two immunodeficient patients

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ALTHOUGH THE LYMPHOPROLIFERATION in Epstein-Barr virus infections is almost always self-limiting, there are instances of uncontrolled proliferation resulting in death.<sup>1,2</sup> In addition, there are a few reports of patients with chronic active EBV infection in which there is clinical

EBNA	Epstein-Barr nuclear antigen
EBV	Epstein-Barr virus
PBMC	Peripheral blood mononuclear cells
VCA	Viral capsid antigen