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Epidemiology of Rotavirus Diarrhea in a Prospectively Monitored American Indian Population

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Rotaviral diarrhea is endemic in most areas of the world, yet community-wide epidemics have not been reported in prospectively monitored populations. This prospective study of the etiology of diarrhea included biweekly visits to the homes of 10% of the population of the White Mountain Apache Indians and began in April 1981. During a three-week period beginning 21 October, 1981, 342 new cases of diarrhea were identified on different parts of the reservation. Rotaviral antigen, detected by an enzyme-linked immunosorbent assay, was identified in 169 (73%) of the 233 stool samples that were tested. Rotavirus was not detected in any of the stool samples taken six months before or after the epidemic. During the epidemic, respiratory symptoms were present in 44 (33%) of 135 rotavirus-positive patients compared with 17 (17%) of 98 rotavirus-negative patients ($P < .05$). This rapidly spreading epidemic involving all areas of the reservation, in the absence of a common source of exposure of ill persons, suggests the possibility of respiratory transmission.

Rotavirus is a leading cause of diarrhea in many parts of the world [1–5]. A number of published reports [6–9] have described epidemics of rotaviral diarrhea in hospital-based studies. However, community-wide epidemics of rotaviral diarrhea have not been described in prospectively monitored populations. In this report we describe the occurrence of an epidemic of rotavirus in a prospectively monitored population on an American Indian reservation in the United States.

Patients and Methods

The White Mountain Apache Indian Reservation occupies 1.7 million acres of land in the state of Arizona. Approximately 9,000 individuals reside on the reservation. Most of the medical care for the reservation is provided by the Indian Health Service. The 50-bed hospital on the reservation contains an outpatient clinic where ~200 patients are seen per day. There is also an outpatient clinic 50 miles from the hospital.

Clinical studies. Since 1 April 1981, we have been prospectively monitoring the occurrence of diarrhea in a randomly selected sample of 10% of the households on the reservation (surveillance population). These 201 households (1,103 persons) were visited every two weeks, and each individual living in the household was asked if he or she had had diarrhea, defined as more than three watery stools in a 24-hr period, in the previous two weeks. If diarrhea had occurred during this time, the patients were questioned about the presence of blood in the stool, vomiting, runny nose, cough, and fever. If diarrhea occurred within two days of the visit to the home, a rectal swab was obtained for detection of bacterial pathogens by using modified Stuart's bacterial transport medium (American Scientific Products, Tempe, Ariz). An additional stool swab was collected in 10% PBS for detection of rotaviral antigen. Adult patients were requested to provide a sputum specimen as well for detection of rotaviral antigen. Nasal secretions

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to be assayed for rotavirus were obtained from infants less than three years of age by instilling 0.25 ml of normal saline into the posterior nares by using a small plastic catheter. If surveillance patients were seen for their illness at the hospital or at one of its clinics, all the specimens mentioned above were obtained at that site.

Besides the community-based surveillance population, participants in the study included all patients hospitalized for diarrhea and a 20% sample of all patients with diarrhea attending the outpatient clinic. A complete history was obtained and physical examination performed on these patients. All specimens routinely obtained from surveillance patients with diarrhea were also collected. In hospitalized patients, a whole stool specimen was also obtained, with one aliquot being used for detection of bacterial pathogens and another placed in PBS for detection of rotavirus. Whenever possible, stool specimens were obtained from age- and sex-matched control patients hospitalized for illnesses other than diarrhea. Since most of the hospital beds were occupied by patients with diarrhea during the epidemic, only a few age- and sex-matched control patients were available.

Of the remaining 80% of the outpatients not participating in the study, only rectal swabs were obtained for detection of bacterial pathogens and rotaviral antigen as described above.

Laboratory studies. Specimens were tested in a blinded fashion without knowledge of the presence or absence of clinical illness. All stool specimens, nasal aspirates, and sputa were tested for rotaviral antigen by ELISA [10]. Control assays were performed with nonimmune serum to document the specificity of the assay as described previously [10]. Nasal aspirates and sputum specimens were also tested for the presence of adenovirus, influenza virus, parainfluenza virus, and respiratory syncytial virus by ELISA [11].

Standard isolation procedures were performed for the identification of *Shigella*, *Salmonella*, and *Campylobacter* species; pathogenic vibrios; and *Yersinia enterocolitica* [12]. All of the stool specimens were also tested for the presence of enterotoxigenic *Escherichia coli*. The Y1 mouse adrenal cell assay [13] was used for detection of heat-labile enterotoxin, and the infant mouse assay [12] was used for detection of heat-stable enterotoxin.

Determination of rotaviral subgroups was attempted, as previously described [14], with eight rotavirus-positive stool specimens obtained from patients residing on different parts of the reservation. Cultivation of these same eight strain of rotavirus was also attempted by the use of primary cynomolgus monkey kidney cells in roller tube culture; viral growth was detected by enzyme immunoassay [15].

Results

During a three-week period beginning 21 October 1981, there was a sudden increase in the number of cases of diarrhea on the reservation (figure 1). During this epidemic period, 342 new cases of diarrhea occurred; 53 (15%) were diagnosed as having had diarrhea when bimonthly home visits were made, and 289 (85%) were seen at the hospital or at one of its clinics. Rotaviral antigen was detected in the stools of 169 (73%) of the 233 patients that were tested. All but nine of these children were seen in the clinic. Rotaviral antigen, however, was detected in only two of 30 control patients ($P < .001$). One of these two patients experienced rotaviral diarrhea two days after the initial stool specimen was obtained.

The daily occurrence of rotaviral diarrhea during the epidemic is shown in figure 2. The first case occurred on 21 October 1981. The number of cases increased rapidly until it peaked on 26 October when there were 22 cases, and then tapered off gradually until the end of the epidemic. There were no cases of rotaviral diarrhea during the six months before and after the epidemic, although >1,000 diarrheal stool specimens were tested during each of these periods. During the epidemic, 18 patients had a bacterial pathogen isolated in their stool specimens in addition to the rotaviral antigen. The organisms isolated were *Salmonella* (two patients), *Shigella* (14 patients), and *Campylobacter jejuni* (two patients).

The 169 cases of rotaviral diarrhea that occurred within the three-week period were in individuals from all major residential areas of the reservation. These areas are widely separated geographically, and each has its separate source of water. Furthermore, there was no common source of exposure, such as large social gatherings, camp meetings, or rodeos, that could be identified during the epidemic.

The rate of illness with rotaviral diarrhea for the different age groups was calculated by using the surveillance population (figure 3). This rate was highest in patients aged three to five months (67%). There was a progressive decrease in rate of illness in children more than five months of age, with the rate of illness dropping to 1% in children aged 24–59 months. There were no cases of diarrhea in infants

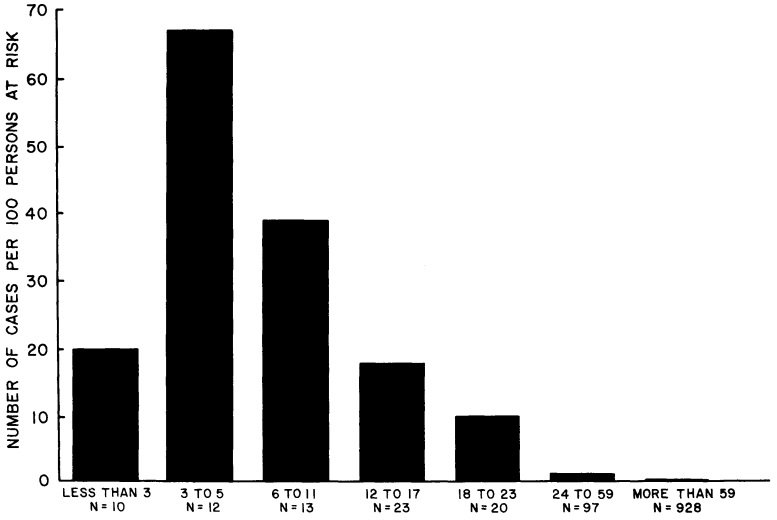

Figure 1. No. of cases of diarrhea by week on the White Mountain Apache Indian Reservation between 1 October 1981 and 2 December 1981. *Solid shading*, stool specimens positive for rotaviral antigen; *dotted hatching*, stool specimens negative for rotaviral antigen; *unshaded areas*, stool specimens not obtained.

<45 days of age, in children more than five years of age, or in adults.

The number of patients with rotaviral diarrhea severe enough to require hospitalization is shown in table 1. Eight (47%) of 17 patients less than three months of age required hospitalization as compared with 30 (20%) of 152 patients more than three months of age ($\chi^2 = 5.84$; $P < .02$). None of the patients >11 months of age required hospitalization.

During the epidemic, clinical information was available for 135 patients with rotaviral diarrhea and for 98 patients with diarrhea in whom rotaviral antigen was not detected. The clinical presentation of patients with rotaviral diarrhea was similar to that described previously [1], 44 (33%) of the rotavirus-positive patients and 17 (17%) of the rotavirus-negative patients had associated respiratory symptoms ($P < .05$): 32 (24%) had otitis media, 21 (48%) had upper-respiratory-tract infection, and 1 (1%) had pneumonia. Of the 58 patients with nasal aspirates or 10 patients with sputum specimens, none were positive for rotaviral antigen or other respiratory pathogens.

Subgroup specificity was determined for seven of eight strains of rotavirus isolated from the stool samples; all seven were subgroup 2. Upon attempted cultivation by using cynomolgus monkey kidney cells, all of the eight strains of rotavirus grew after one passage, and seven of the eight grew after two passages. Repeat subgroup determination upon harvests

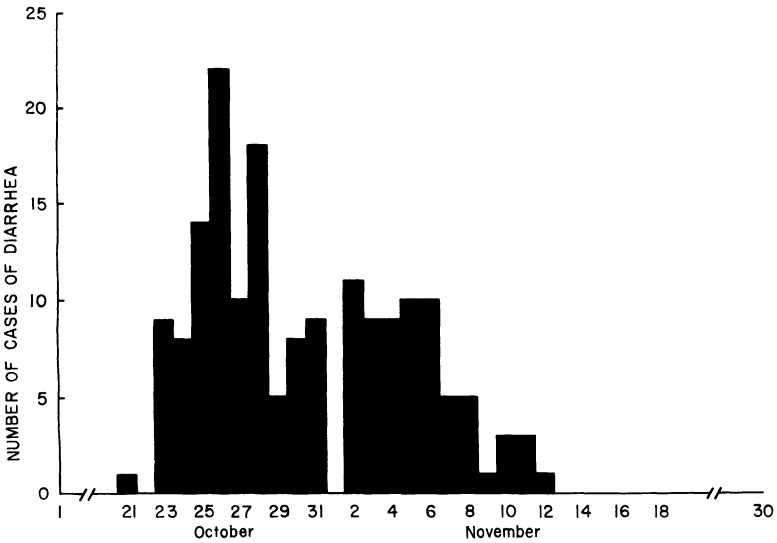

Figure 2. Cases of rotaviral diarrhea per day over a one-month period in 1981 on the White Mountain Apache Indian Reservation.

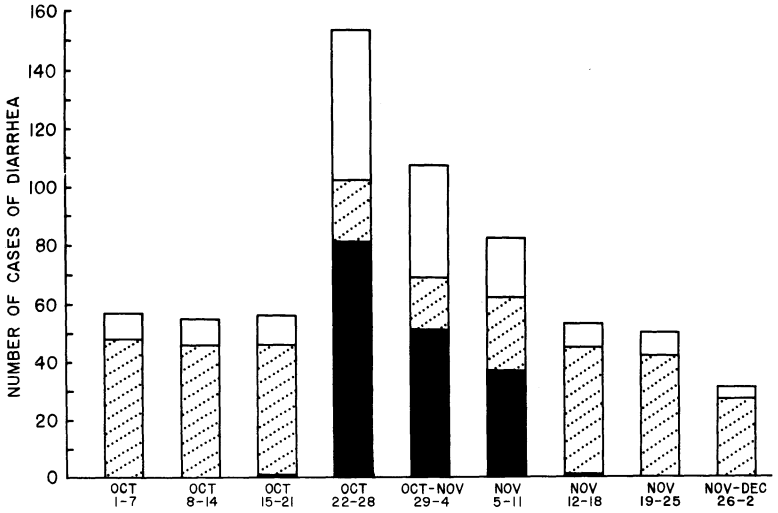

Figure 3. Rate of illness with rotaviral diarrhea by age group on the White Mountain Apache Indian Reservation. The rate of illness was calculated by using the surveillance population.

following the second passage was successful in six of the seven strains, and all six were subgroup 2. The one strain for which the subgroup could not be determined from the stool specimen was determined to be subgroup 2 after cultivation. Thus, all eight strains studied were subgroup 2.

Discussion

Community-wide epidemics of rotaviral diarrhea such as the one described in the present study have been described previously in another American Indian population [11] and in other geographically isolated populations [17, 18]. However, none of the published reports of community-wide epidemics have occurred in a prospectively monitored population.

The mode of spread of rotavirus in the present epidemic is unknown. Transmission by food or by the fecal-oral route seems unlikely since cases of diarrhea occurred simultaneously in all areas of the reservation even though individual residential areas were widely separated in distance by five to 50 miles, and there was no common meeting site. Transmission by contaminated water also seems unlikely since the different geographic areas on the reservation had their own separate sources of water. The rapid spread of rotavirus throughout the community in this epidemic is similar to that of other viruses such as influenza and measles, which are both spread by the respiratory route. There were no unusual climactic conditions that could be identified during or immediately preceding the epidemic. The high incidence of symptoms involving the respiratory tract seen in the present epidemic has also been documented by other investigators [19, 20]. However, the presence of rotavirus in the respiratory secretions could not be documented in patients with diarrhea either in the present epidemic or in other studies. In contrast to the findings in patients with diarrhea, we have recently demonstrated the presence of rotaviral antigen in the respiratory secretions of children diagnosed as having pneumonia without symptoms of diarrhea [21]. This finding suggests that rotavirus is capable of colonizing the respiratory tract and could, therefore, be transmitted by this route.

Table 1. Patients requiring hospitalization for rotaviral diarrhea.

Age of patient (months)	No. with rotaviral diarrhea	No. hospitalized (%)
<3	17	8 (47)
3–5	42	12 (29)
6–11	75	18 (24)
Total	134	38 (28)

Previous studies have identified at least two antigenic variants of rotavirus [22] that are now considered to represent two distinct subgroups characterized by the antigenic specificity of the sixth gene product [23]. In addition, four serotypes of human rotavirus have been characterized on the basis of the neutralization phenotype, which is mainly characterized by the eight or ninth gene product [23]. We did not process the sera for subgroup- or serotype-specific antibody responses. However, eight strains of rotavirus from different geographic areas on the reservation belonged to subgroup 2. An attempt was

made to grow a sufficient quantity of the rotaviral isolates to study the RNA pattern and to evaluate the serotype in order to determine whether the viruses were of the same or different strains or serotypes. Unfortunately, there were not sufficient quantities of RNA in the preparations to make that evaluation.

As documented by other studies [1–5], most of the cases of rotaviral diarrhea occurred in children less than two years of age. However, the highest incidence of rotaviral diarrhea in the present study occurred in children aged three to five months. This finding is in contrast with the incidence in other populations in the United States in which the highest rate of illness is in children aged 13–15 months [24]. This may be a result of the low prevalence (<10%) of breast feeding in this Apache population. The rate of hospitalization for this illness in children less than one year of age with rotaviral diarrhea was 28%, whereas none of the patients older than this required hospitalization. This observation suggests that the disease is relatively severe in young Apache children. In addition, the rapid spread, high rates of illness, and short duration of the epidemic suggest that rotavirus rapidly infected all susceptible infants on this reservation. Once the pool of the susceptible population was exhausted, the epidemic apparently stopped. Since none of the children more than five years of age or adults were found to have rotaviral diarrhea, we assume that they had probably acquired immunity to rotavirus during previous exposure to the disease.

In summary, we have described an epidemic of rotaviral diarrhea in a prospectively monitored American Indian population. The outbreak was brief in duration; the highest rate of illness was in children aged three to five months; the illness was most severe in children <12 months of age; and there was a high frequency of symptoms involving the respiratory tract associated with this illness. The mode of spread of the epidemic remains unknown, although transmission by the respiratory route is suggested.

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