An Outbreak of Norwalk-like Virus Infection in Tokyo and Saitama in Late 1995

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From the end of October to December, 1995, outbreaks of gastroenteritis were prevalent in Japan. The appearance of this gastroenteritis seemed to be one month earlier than the annual average. The outbreaks were unusual because rotavirus and enteric adenovirus, which are usually detected around this time of the year, were not found. Both school children and adults had also gastroenteritis although infants and young children were predominantly infected.

In this study, we examined Norwalk-like virus (NLV) and astrovirus (AV) from stool samples collected in Tokyo and Saitama by reverse transcription and polymerase chain reaction (RT-PCR).

Materials and Methods

Sixteen stool samples were collected from 6-month to 2-year-old infants at Akabane Pediatric Clinic, in the northern part of Tokyo. Twenty stool samples were collected from 9-month to 2-year-old infants at Saitama Kyodo Hospital, in the southern part of Saitama. The samples were obtained from the end of October to the beginning of December. The stool samples were previously determined to be nonbacterial by culture, nonrotavirus by latex agglutination and nonadenovirus by latex agglutination.

Viral RNA was extracted from stool specimens with guanidin thiocyanate and glass powder, and RT-PCR was then conducted as previously described1,2). The RNA was reverse transcribed with three sets of the desired first PCR primer mixtures for AV (Gr15-Gr12)3) and NLVs (P35-P364) and P81-P824), separately. The second PCR was conducted with P69-P39 following the first PCR with P35-P36. Both the first and second PCR products were electrophoresed in agarose gel containing ethidium bromide and photographed under UV light.

Results and Discussion

From Akabane Pediatric Clinic, NLVs were detected in six samples among sixteen with P81-P82. However, NLVs were not detected with P35-P36 (the first PCR) and P69-P39 (second PCR). AV was not found with Gr15-Gr12. From Saitama Kyodo Hospital, NLVs were detected in six samples among
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twenty with P81-P82. However, NLVs and AV were not found with the other primers, as in the Akabane cases.

We have been studying the molecular epidemiology of NLVs and AV by RT-PCR. We have detected NLVs and AVs in about 10% of diarrheal samples obtained during the winter season, sporadically, in outpatient clinics1,2). This was our first experience with an outbreak of NLVs in outpatient clinics, with detection in approximately 40 to 30% of samples. Moreover, it was the first time that NLVs were not detected with P35-P36 and P39-P69. We are now sequencing the PCR to assertain differences in DNA sequences in comparison with those of previous reports3,4) and to determine whether we can make new primers for the second PCR following the first PCR with P81-P82. We will examine all samples by immune electron microscopy and enzyme immunoassay in the near future.

References


