Short Communication

Rotavirus infection of Thai infants
Serotypes and subgroups of cultivated viruses in 1984 and 1985

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In Thailand, rotavirus has been found to be one of the major causative agents of acute diarrhoea during the cool and dry months1,2, and in seroepidemiological studies antibodies to rotavirus were detected in about 80% of Thai infants aged under two years3,4. Although the prevalence of this virus infection has already been reported5-8, no study of serotyping of rotaviruses has not been done in Thailand. In addition to the four distinct serotypes of group A human rotaviruses9-11, some strains 69M12, W1613 and F4514, which did not belong to any of the four serotypes have recently been isolated. In order to develop an appropriate vaccine, it is important to explore the epidemiological characteristics and temporal distribution of the serotypes, because the serotype of rotavirus was originally classified by neutralization. Thus, we studied the serotype of rotaviruses which were isolated in Thailand in 1984 and 1985.

Twenty-four strains of rotavirus isolated in primary Cynomolgus monkey kidney cell cultures from fecal specimens collected in Chiang Mai, Khon Kaen and Bangkok during the period from November, 1984 to December, 198515, were examined. All the specimens were found to contain group A rotavirus antigens by a sandwich enzyme-linked immunosorbent assay (ELISA) according to the method recommended by WHO15.

Serotyping and subgrouping of the virus in the tissue culture fluid were performed by another sandwich ELISA using type- and subgroup-specific monoclonal antibodies developed by Akatani and Ikegami16,17. Briefly, microtiter plates were coated with a monoclonal antibody (Osaka National Hospital, AH6) which captures double-shelled rotavirus particles. Following treatment with 1% bovine serum, the plates were washed with phosphate-buffered saline containing 0.05% Tween 20. Supernatants of the infected tissue culture fluids (50 µl) were then added and incubated for 90 min at room temperature. After incubation, the plates were washed
and each well received one of the biotinylated type- and subgroup-specific monoclonal antibodies. The type-specific monoclonal antibodies were Osaka National Hospital AH49 for serotype 1, AG12 for serotype 2, AC5 for serotype 3 and AE18 for serotype 4. Following incubation for 90 min, the plates were washed and streptavidin-peroxidase conjugates were added. Following incubation for 10 min, the plates were washed and substrate (hydrogen peroxide and o-phenylenediamine) was added, and the $A_{500}$ of each well was measured with a microplate reader. Readings greater than 0.05 were considered positive.

The results are shown in Table 1. We were able to classify 23 out of 24 strains (95.8%) into two of the four previously established serotypes. One specimen, strain CM17-121, could not be serotyped. All of the 19 specimens of serotype 1 were identified, by the used subgroup-specific monoclonal antibodies, as subgroup II and all of the four of serotype 2 as subgroup I. The unserotyped CM17-1 was identified as subgroup I. All of the strains of subgroup I except for CM17-1 showed the S pattern and all of the strains of subgroup II showed the L pattern of the RNA-electropherotype. One strain (S78) isolated from a fecal specimen collected in Bangkok was of serotype 1 and of subgroup II, through the original fecal specimen had shown $L + S$ pattern of the RNA-electropherotype.

Previous studies, one carried out in Italy by solid-phase immune electron microscopy and the others performed in Australia, Central African Republic and Japan by ELISA with serotype-specific monoclonal antibodies have shown that although the majority of stool rotaviruses belong to serotype 1, the frequency of detection of other serotypes varies considerably from one study to another.

In this report, serotypes 1 (79.2%) and 2 (16.7%) strains were detected but neither of the serotypes 3 and 4 were detected in 24 specimens collected in Thailand in 1984 and 1985. Serotype 1 was predominant in three parts (North-Eastern, Central and Northern) of Thailand, and serotype 2 strain was not detected in Chiang Mai in the north. The distances between Khon Kaen, Bangkok and Chiang Mai vary from 440 to 680 km as shown in the map (Fig. 1) and the climate in the cool

### Table 1 Serotypes, subgroups and RNA patterns of rotavirus in Thailand, 1984-1985

<table>
<thead>
<tr>
<th>Place and time of collecting specimens</th>
<th>Numbers of specimens</th>
<th>Serotype Total</th>
<th>Subgroup II</th>
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</table>

*: Unclear (strain CM 17-1)
season is different between each of these areas\textsuperscript{11}. Further work is necessary to clarify the distribution and characteristics of serotypes of rotavirus in Thailand.

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References


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