Introduction

There is wide acceptance of the need for a vaccine to prevent rotavirus disease in children under 5 years of age throughout the world. While there are few deaths in developed countries, there is considerable morbidity, with 10,000 Australian children hospitalised each year.¹ A major outbreak in the Northern Territory during the recent 12 month surveillance period has reinforced the seriousness of this infectious disease in the Australian community, especially in Aboriginal children. The impact on health services in Alice Springs was significant. During May 2001, 246 children with acute gastroenteritis presented to the Emergency Department of the Alice Springs Hospital, resulting in 145 being admitted, of whom 137 were confirmed as having rotavirus infection.²

Surveillance relies upon co-operation of microbiologists from major centres, and they continue to provide very valuable input into the system. Past experience has shown that Brisbane, Sydney, Melbourne, Adelaide and Hobart tend to have similar patterns of disease, while a high number of cases and unusual strains have been identified in Western Australia. The Northern Territory has had very different epidemiology, with outbreaks at unpredictable times of the year and emergence of unusual strains, sometimes in conjunction with Western Australia. Surveillance has continued unchanged in Western Australia and the Northern Territory, but surveillance on the eastern seaboard has been limited to Melbourne.

Methods

Collaborating laboratories undertook rotavirus detection by enzyme immunoassay (EIA) or latex agglutination. Rotavirus positive specimens were collected, stored frozen and forwarded to the Royal Children’s Hospital in Melbourne, together with relevant age and sex details.

Specimens were then tested using an in-house monoclonal antibody (MAb) based serotyping EIA. The EIA employed a panel of MAbs specific for the major glycoprotein VP7 of the outer capsid of the 5 major group A human rotavirus serotypes (G1, G2, G3, G4 and G9). Strains unable to be assigned a serotype were genotyped by reverse transcription/polymerase chain reaction

Abstract

The National Rotavirus Reference Centre together with collaborating laboratories Australia-wide has conducted rotavirus surveillance since June 1999. The serotypes of rotavirus strains that are responsible for the hospitalisation of children with acute gastroenteritis were determined for the period 1 June 2001 to 31 June 2002. We examined 754 rotavirus samples using a combination of monoclonal antibody immunoassay, reverse transcription-polymerase chain reaction, and Northern hybridisation. For the first time, serotype G9 strains were the most prevalent type nationally (40.4%) and found in 8 of the 9 centres. Serotype G1 strains were the second most prevalent type (38.9%), identified in 5 of the centres. These findings have important implications for vaccine development strategies which target serotypes G1-G4.

Keywords: rotavirus; gastroenteritis

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(RT/PCR) using serotype specific oligonucleotide primers.\textsuperscript{3} Northern hybridisation analysis utilising G type specific DNA probes hybridised under stringent conditions was also employed to confirm serotype specificities.\textsuperscript{4} Polyacrylamide gel electrophoresis (PAGE) confirmed the sharing of the same electropherotype between collaborating centres.

**Results**

**Number of isolates**

A total of 847 specimens were received from the collaborating centres. Specimens containing insufficient specimen for testing or specimens that were not confirmed to be positive for rotavirus, were omitted from the serotyping data. A total of 754 positive specimens were analysed over a 13 month period from 1 June 2001 to 31 June 2002.

**Age distribution**

The age distribution of acute gastroenteritis cases were typical of rotavirus infection (Figure). In the reporting period, 39 per cent of cases were from infants 12 months of age or less, 33 per cent were from patients 13–24 months of age, and 15 per cent were from patients 25–36 months of age. Overall, 87 per cent of samples were from children 3 years or less, and 96 per cent were from children 5 years or less.

**Serotype distribution**

Rotavirus serotypes identified in Australia from 1 June 2001 to 31 June 2002 are shown in the Table. Serotype G9 was the most common nationally, representing 40.4 per cent of specimens. It was present in 8 of the 9 centres and was the dominant type in 6 of the centres. G1 was second most common, and represented 38.9 per cent of specimens. G1 was the dominant type in 2 locations (Melbourne and Perth) and was present in another 3 centres (Alice Springs, Darwin-Western Pathology, and WA PathCentre).

**Table 1. Reports of rotavirus G serotypes in Australia, 1 June 2001 to 31 June 2002**

<table>
<thead>
<tr>
<th>Centre</th>
<th>Total number</th>
<th>G1</th>
<th>G2</th>
<th>G3</th>
<th>G4</th>
<th>G9</th>
<th>Mixed serotypes</th>
<th>No result*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Melbourne</td>
<td>166</td>
<td>47.6</td>
<td>4.2</td>
<td>0</td>
<td>7.2</td>
<td>13.3</td>
<td>5.4</td>
<td>22.3</td>
</tr>
<tr>
<td>Hobart</td>
<td>6</td>
<td>0.0</td>
<td>33.3</td>
<td>0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>66.7</td>
</tr>
<tr>
<td>Perth</td>
<td>224</td>
<td>72.3</td>
<td>2.2</td>
<td>0</td>
<td>0.0</td>
<td>11.6</td>
<td>1.8</td>
<td>7.1</td>
</tr>
<tr>
<td>WA Pathcentre</td>
<td>94</td>
<td>37.2</td>
<td>0.0</td>
<td>0</td>
<td>1.1</td>
<td>40.4</td>
<td>4.3</td>
<td>17.0</td>
</tr>
<tr>
<td>Darwin</td>
<td>22</td>
<td>0.0</td>
<td>0.0</td>
<td>0</td>
<td>0.0</td>
<td>95.5</td>
<td>0.0</td>
<td>4.5</td>
</tr>
<tr>
<td>Darwin-Western Pathology</td>
<td>44</td>
<td>6.8</td>
<td>0.0</td>
<td>0</td>
<td>0.0</td>
<td>70.5</td>
<td>4.5</td>
<td>18.2</td>
</tr>
<tr>
<td>Alice Springs</td>
<td>118</td>
<td>2.5</td>
<td>0.0</td>
<td>0</td>
<td>0.0</td>
<td>80.5</td>
<td>5.9</td>
<td>11.1</td>
</tr>
<tr>
<td>Gove</td>
<td>30</td>
<td>0.0</td>
<td>0.0</td>
<td>0</td>
<td>0.0</td>
<td>86.7</td>
<td>0.0</td>
<td>13.3</td>
</tr>
<tr>
<td>Mt Isa</td>
<td>50</td>
<td>0.0</td>
<td>0.0</td>
<td>0</td>
<td>0.0</td>
<td>92.0</td>
<td>0.0</td>
<td>8.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>754</strong></td>
<td><strong>38.9</strong></td>
<td><strong>1.9</strong></td>
<td><strong>0</strong></td>
<td><strong>1.7</strong></td>
<td><strong>40.4</strong></td>
<td><strong>3.4</strong></td>
<td><strong>13.7</strong></td>
</tr>
</tbody>
</table>

* No result – unable to be serotyped with monoclonal antibodies or genotyped by RT/PCR.
Serotypes G2 and G4 each represented less than 2 per cent of the specimens. G2 was identified in 3 centres (Melbourne, Hobart and Perth), while G4 was identified in Melbourne and Western Australia. Serotype G3 was not identified during this surveillance period in any centre.

These serotyping results illustrate important differences between the distribution of serotypes in different parts of Australia. Serotype G9 was the dominant type in Central and northern Australia, while G1 was dominant in west and eastern Australia. Melbourne was the only centre where the four circulating serotypes were identified.

In the reporting period, 3.4 per cent of the rotavirus samples analysed contained multiple serotypes. The presence of mixed infections provides the opportunity for rotavirus to undergo reassortment, potentially resulting in new strains. In 13.7 per cent of the samples a serotype was unable to be assigned. These could represent unusual serotypes not identified using standard methods, or samples with low virus numbers which are below the detectable limits of our assays.

**Discussion**

National rotavirus surveillance from 1 June 2001 to 31 June, 2002 highlighted the emergence of serotype G9 as the nationally dominant serotype. This corresponded with the large outbreak of acute gastroenteritis (caused by rotavirus G9) that occurred in Alice Springs, where 145 children (137 were rotavirus positive) were hospitalised and several hundred more were affected during May 2001. This outbreak continued to spread northward to Tenant Creek, Katherine, Darwin and Gove during June and July 2001. In addition, cases of acute gastroenteritis in several remote Western Australian, South Australian and Queensland locations were also attributed to this serotype. PAGE analysis of samples from each of these locations indicated that the same strain was responsible for the initial outbreak in Alice Springs and the subsequent spread.

Serotype G9 was first identified in 3 children with severe gastroenteritis during Australia-wide surveillance in 1997. Serotype G9 strains were not identified during 1998. During surveys conducted in 1999/2000 and 2000/2001, serotype G9 was the second most prevalent serotype nationally, representing 10 per cent and 18.1 per cent respectively of specimens collected in those years. Serotype G9 strains have persisted since 1996 to 1997 in many countries including the United States of America, Bangladesh, and the United Kingdom, and their occurrence has now been documented in all continents.

For the first time since national rotavirus surveillance began in 1993, serotype G1 was not the dominant national type, being detected in 38.9 per cent of samples. Previously serotype G1 was the most prevalent serotype in Australia, representing 58 per cent and 49.5 per cent of specimens during 1999/2000 and 2000/2001. Serotype G1 was also dominant in a study conducted from 1993 to 1996. The decline in the prevalence of serotype G1 strains around Australia can be attributed to the relative increase in the prevalence of serotype G9 strains.

The prevalence of serotype G4 increased in Melbourne from 5.2 per cent in 2000/2001 to 7.2 per cent. During the previous year (2000/2001), serotype G4 was the second most common type identified in Darwin and Sydney. Whether the Melbourne serotype G4 strains identified in 2001/2002 are related to those earlier strains from Darwin and Sydney requires further analysis. The decrease of serotype G2 in Melbourne (from 15.3% to 4.2%) continues the sporadic occurrence of this serotype from both a local and national perspective.

Ongoing surveillance of the seasonal variation in rotavirus is warranted. The identification of emerging serotypes highlights the continued evolution of rotavirus. Epidemiological knowledge of serotype prevalence will assist in the design of future vaccine strategies.

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K Dempsey, Mt Isa Centre for Rural and Remote Health

Victoria
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References


