# Annual report of the Rotavirus Surveillance Programme, 1999/2000

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## Abstract

**The National Rotavirus Reference Centre has conducted rotavirus surveillance by means of a collaborative laboratory based initiative started in June 1999. The serotypes of rotaviruses that lead to the hospitalisation of children with acute diarrhoea were determined from June 1999 to May 2000. We examined 1126 rotavirus specimens using a combination of monoclonal antibody immunoassay, reverse transcription-polymerase chain reaction, and hybridisation. The four most common serotypes G1-G4 were represented. More than 50% of isolates tested were serotype G1, with serotype G1 being represented in most centres Australia-wide. Serotype G9 rotaviruses were identified for the first time in Australia, and were second in importance with 10% of samples tested. The significant presence of G9 viruses throughout Australia suggests the emergence of a new serotype and has implications for current rotavirus vaccine strategies that target serotypes G1-G4. *Commun Dis Intell* 2000;24:195-198.**

*Keywords: rotavirus, surveillance, infants, serotypes, vaccine, gastroenteritis*

## Introduction

Rotaviruses are the most important cause of severe gastroenteritis in young children worldwide. The pathogen is believed to be responsible for the annual admission of up to 10,000 children to hospitals nationwide.[1] A national rotavirus surveillance programme was commenced in June 1999 to undertake the surveillance and characterisation of rotavirus strains causing annual epidemics of severe diarrhoea in young children throughout Australia. The programme was designed to monitor the antigenic variation of rotaviruses prior to and after the anticipated rotavirus vaccine release in Australia. The study was reliant on the cooperation and participation of sentinel laboratories from all States and the Northern Territory. It was designed to supplement data from existing notification schemes, by reporting the serotypes circulating in Australian urban centres. The following report covers the period June 1999 to May 2000.

## Methods

A network of laboratories from each State and the Northern Territory undertake rotavirus detection by enzyme immuno-assay (EIA) or latex agglutination. Rotavirus-positive specimens were collected, stored frozen and forwarded to the Royal Children's Hospital (RCH) in Melbourne, together with relevant age and sex details. Representative faecal specimens were then tested using an in-house monoclonal antibody (MAb) based serotyping EIA. The EIA incorporates a panel of MAbs specific for the common group A human rotavirus serotypes (serotypes G1, G2, G3, and G4). Specimens with an absorbance value greater than 0.2 were considered positive for that serotype. Northern hybridisation analysis with G type specific DNA probes using stringent hybridisation conditions was also employed to confirm sero-type specificities. Selected strains unable to be assigned a serotype, were genotyped by reverse transcriptase/ polymerase chain reaction using serotype specific primers.

## Results

### Number of isolates

In all 1545 rotavirus positives were sent to the Royal Children's Hospital. Specimens received from New South Wales made up 30.5% of all specimens received (Newcastle, Narrabri and Sydney). Twenty-five percent of specimens were received from Western Australia (Perth and northern Western Australia), 16% from Victoria (Melbourne and Horsham), 14% from South Australia (Adelaide), 7% from Queensland (Brisbane and Townsville), 7% from the Northern Territory (Darwin, Gove and Alice Springs) and 0.5% from Tasmania (Hobart). Representative specimens were incorporated into the serotyping EIA (1126 specimens). Specimens that were not confirmed to be positive for rotavirus, or had insufficient specimen for testing, were excluded.

### Seasonal occurrence

The overall nationwide rotavirus reports peaked in August (Figure 1). The peak month of activity varied between centres, with temperate regions experiencing a noticeable winter peak between July and October. The rotavirus seasons of combined centres in Victoria, Tasmania and New South Wales peaked in August, followed by Queensland in September (Figure 2). Western Australia shared its rotavirus peak with Alice Springs and Adelaide in October. The tropical Northern Territory rotavirus season peaked in January 2000.

### Age group

The age distribution of rotavirus-positive patients showed the peak incidence occurred in children aged between 1 and 2 years (Figure 3). The male:female ratio was 1.18:1.

### Serotype distribution

The serotypes circulating in Australia from June 1999 to May 2000 are shown in Figure 4. Serotype G1 accounted for

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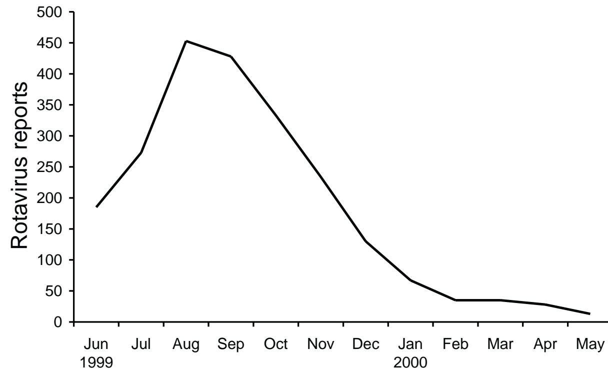


**Figure 1. Rotavirus reports, Australia, June 1999 to May 2000, by month**

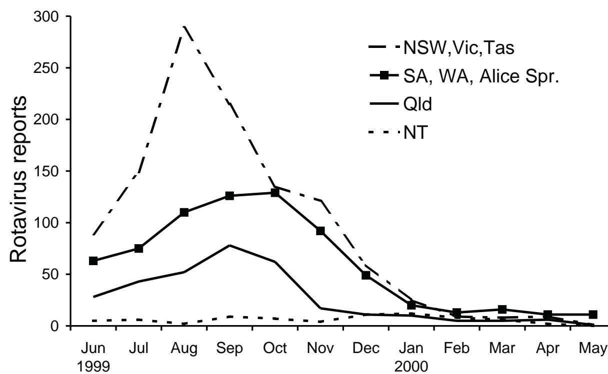


**Figure 2. Seasonal peaks of rotavirus reports, Australia by region, June 1999 to May 2000, by month**

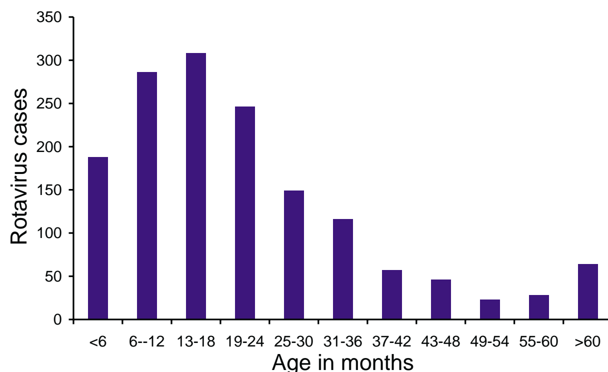


**Figure 3. Rotavirus positive children, Australia, June 1999 to May 2000, by age group in months**

58% of specimens nationally, and was responsible for more than 60% of rotavirus serotypes in some centres; Hobart (100%), Darwin (80%), Northern Western Australia (69%) and Perth (80%) (Table 1). Second in importance to G1 were G9 rotaviruses (10%). The G9 rotaviruses appeared in Sydney and Melbourne in June 1999 and were present in 12 of the 18 centres studied (all centres except Darwin, Townsville, Horsham, Gove, northern Western Australia

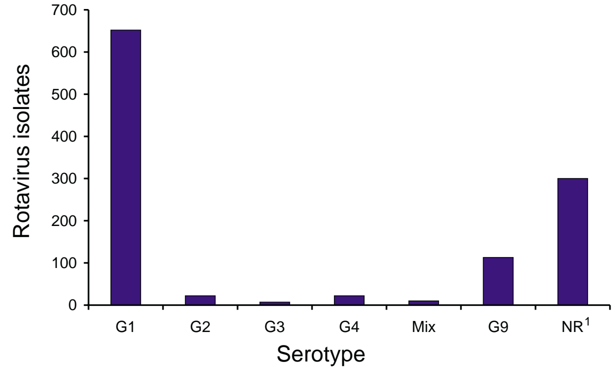


**Figure 4. Rotavirus isolates, Australia, June 1999 to May 2000, by serotype**

1. NR = non-reactive

and Hobart). Serotype G2, G3 and G4 viruses accounted for less than 5% of all serotypes detected. Serotype G2 viruses were detected along the east coast of Australia, Narrabri, Adelaide, Perth and northern Western Australia. Serotype G3 viruses were present in Melbourne and Adelaide only. Serotype G4 viruses were found in all States except Tasmania and the Northern Territory. A serotype could not be assigned to 27% of specimens. Specimens containing mixtures of rotavirus G types were detected in less than 1% of samples tested.

## *Discussion*

The surveillance study was marked by the appearance of G9 rotaviruses in Australia for the first time.[2] The virus was responsible for at least ten percent of acute diarrhoea rotaviral hospital admissions. Retrospective analysis showed two G9 viruses were present in Perth and Melbourne in 1997. The subsequent spread of G9 viruses appears to be almost nationwide with the increase in numbers making G9 viruses second in importance to G1 viruses. G9 viruses were found in all States except Tasmania. G9 viruses have been reported in India (1993-1994),[3] Bangladesh (1987-1997),[4] Malawi (1997-1998),[5] the USA (1996-1997)[6] and the UK (1996).[7] The rapid emergence of G9 as a major infecting serotype has important implications for rotavirus vaccine strategies. Current candidate vaccines target only serotype G1-G4 infections. Ongoing surveillance is warranted to obtain a clearer picture of the importance of the spread of G9 viruses.

Serotype G1 viruses were the most prevalent serotype in Australia during the sampling period. This G1 dominance was consistent with recent studies undertaken in Australia (1993-1996) (Personal communication, Professor Ruth Bishop, Department of Gastroenterology, Royal Children's Hospital. Parkville Victoria), the UK (1996)[7] and the USA (1996-1997)[6]. The virus was present in all centres tested except Townsville and Narrabri.

Centres that showed evidence of more than one serotype generally had larger populations. There appeared to be more serotypes circulating and presenting as hospital admissions in the bigger centres. Smaller population centres such as Hobart, Townsville and Darwin had only one serotype detected and appear to have insufficient population size to sustain multiple G types. Serotype G2, G3

**Table 1. Rotavirus positive specimens, Australia, June 1999 to May 2000 by typing centre and serotype**

| | Serotype | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Centre | G1 | G2 | G3 | G4 | G9 | Mix | NR[1] | Total |
| Brisbane | 31 | 2 | | 2 | 5 | 1 | 34 | 75 |
| Townsville | | 3 | | | | | 16 | 19 |
| South-eastern Sydney | 123 | | | 1 | 31 | | 52 | 207 |
| Western Sydney | 46 | 1 | | 2 | 20 | 1 | 23 | 93 |
| Newcastle | 2 | | | 2 | 7 | | 5 | 16 |
| Narrabri | | 2 | | | 2 | | 19 | 23 |
| Melbourne[2] | 117 | 1 | 5 | 8 | 26 | 5 | 72 | 234 |
| Melbourne[3] | 3 | | | | | | 1 | 4 |
| Horsham | 6 | | | 2 | | | | 8 |
| Hobart | 5 | | | | | | | 5 |
| Adelaide | 52 | 5 | 2 | 2 | 3 | 2 | 11 | 77 |
| Perth | 199 | 4 | | 3 | 7 | 1 | 32 | 246 |
| WA PathCentre | 18 | 4 | | | | | 4 | 26 |
| Darwin[4] | 22 | | | | | | 2 | 24 |
| Darwin[5] | 6 | | | | | | 5 | 11 |
| Gove | 2 | | | | | | 1 | 3 |
| Alice Springs | 20 | | | | 12 | | 23 | 55 |
| Total | 652 | 22 | 7 | 22 | 113 | 10 | 300 | 1,126 |

1. NR = Non reacting to G1, G2, G3, G4 and G9 monoclonal antibodies.
2. Royal Children's Hospital
3. Southern Cross Pathology
4. Royal Darwin Hospital
5. Western Diagnostic Pathology

and G4 viruses were present in small numbers and appeared sporadically.

Antigenic similarities between G4 and G9 viruses were noted. There were a number of specimens that reacted to both G4 and G9 serotyping MAbs. The use of Northern hybridisation and RT/PCR analysis clarified any serological cross reactivities.

Several specimens were unable to be assigned a serotype by EIA. These non-reactive (NR) specimens, generally contained minimal amounts of viral capsid protein and were unable to be detected by the serotyping MAbs. Electrophoretic analysis of the RNA from some of them showed that the viruses circulating in Townsville, Brisbane and Narrabri all shared the same electrophoretic profile. RT/PCR typing of limited numbers showed the samples were serotype G2. Type G2 viruses were under represented in the serotyping EIA suggesting genetic changes in the epitopes targeted by G2-specific MAbs. Specimens received from remote regions of northern Western Australia shared serotype similarities with specimens collected from Perth children. This coexistence of virus serotypes in geographically diverse locations suggests rapid spread of virus strains across the country. The incorporation of remote locations into the study helped to gain a better insight into the pattern of spread.

Ongoing surveillance of seasonal rotavirus serotype patterns is warranted, in particular to monitor the spread of new or emerging serotypes. Such information will influence the strategy for development of second and third generation rotavirus vaccines, and will show whether Australia's requirements differ from those of other parts of the world. This is particularly important now that the first G1-G4 targeted vaccine has been withdrawn from sale in the USA due to its apparent association with intussusception - a form of bowel obstruction.

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