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Australian Rotavirus Surveillance Program: Annual Report, 2019

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Annual report

Australian Rotavirus Surveillance Program: Annual Report, 2019

Sarah Thomas, Celeste M Donato, Susie Roczo-Farkas, Jenny Hua, Julie E Bines and the Australian Rotavirus Surveillance Group

Abstract

This report, from the Australian Rotavirus Surveillance Program and collaborating laboratories Australia-wide, describes the rotavirus genotypes identified in children and adults with acute gastroenteritis during the period 1 January to 31 December 2019. During this period, 964 faecal specimens had been referred for rotavirus G- and P- genotype analysis, including 894 samples that were confirmed as rotavirus positive. Of these, 724/894 were wild-type rotavirus strains and 169/894 were identified as vaccine-like. A single sample could not be determined as wild-type or vaccine-like due to poor sequencing. Genotype analysis of the 724 wild-type rotavirus samples from both children and adults demonstrated that G3P[8] was the dominant genotype nationally, identified in 46.7% of samples, followed by G2P[4] in 8.8% of samples. The Australian National Immunisation Program (NIP) changed to the exclusive use of Rotarix as of 1 July 2017. The NIP had previously included two live-attenuated oral vaccines: Rotarix (monovalent, human) and RotaTeq (pentavalent, human-bovine reassortant) in a state-based vaccine selection. Continuous surveillance is imperative to determine the effect of this change in rotavirus vaccine schedule on the genotype distribution and diversity in Australia.

Keywords: rotavirus, gastroenteritis, genotype, surveillance, Australia, vaccine, Rotarix, G3P[8]

Introduction

Group A rotaviruses are the most common cause of severe diarrhoea in young children worldwide, estimated to have caused 128,500 deaths and 258 million episodes of diarrhoea among children < 5 years of age in 2016.[1] To reduce this burden, the rotavirus vaccines Rotarix™ [GlaxoSmithKline] and RotaTeq™ [Merck] have been introduced in the national immunisation programs of 102 countries.[2] Both vaccines were included in the Australian National Immunisation Program (NIP) on 1 July 2007, leading to a significant reduction in both rotavirus-coded and non-rotavirus-coded hospitalisations of children ≤ 5 years of age with acute gastroenteritis.[3–5] In the first six years following vaccine introduction, an estimated 77,000 hospitalisations were prevented, 90% of which were in children ≤ 5 years, with indications of herd protection occurring in older age groups.[5] RotaTeq was administered in Queensland, South Australia, and Victoria, whereas Rotarix was administered in the Australian Capital Territory, New South Wales, Northern Territory, and Tasmania. Western Australia initially administered Rotarix and changed to RotaTeq in May 2009. On 1 July 2017, all states and territories in Australia changed to Rotarix.[6,7]

Rotavirus surveillance programs utilise a binary classification system based on the two outer capsid proteins, VP7 (G, glycoprotein) and VP4 (P, protease-sensitive), to describe rotavirus genotypes.[8] Globally, there are five common genotype combinations identified in humans: G1P[8], G2P[4], G3P[8], G4P[8], and G9P[8]; however, G12P[8] has also increased worldwide in recent years.[9,10] Additionally, whole genome classification assigns genotypes to each of the 11 genes: Gx-P[x]-Ix-Rx-Cx-Mx-Ax-Nx-Tx-Ex-Hx,

denoting the VP7-VP4-VP6-VP1-VP2-VP3-NSP1-NSP2-NSP3-NSP4-NSP5/6 genes.[11,12] The majority of human rotavirus genomes fall under two genotype constellations: Wa-like (genogroup 1: G1/3/4/9/12-P[8]-I1-R1-C1-M1-A1-N1-T1-E1-H1), and DS-1-like (genogroup 2: G2-P[4]-I2-R2-C2-M2-A2-N2-T2-E2-H2).[11,12] A third genogroup, AU-1-like, is also detected in humans, though less frequently (genogroup 3: G3-P[9]-I3-R3-C3-M3-A3-N3-T3-E3-H3).[11,12]

Numerous mechanisms contribute to rotavirus diversity including genetic drift, reassortment and zoonotic transmission. The segmented genome allows for reassortment both within and between human and animal strains, leading to the emergence of novel strains and unusual genotype combinations.[13]

The Australian Rotavirus Surveillance Program (ARSP) has characterised rotavirus genotypes causing severe disease in Australian children ≤ 5 years of age since 1999. Genotype surveillance data has revealed changes in diversity, as well as temporal and geographic fluctuations over time.[14] Furthermore, differences in genotype diversity and dominance were observed when comparing vaccine jurisdictions, suggesting that RotaTeq and Rotarix exert different immunological pressures.[15] The continued surveillance and characterisation of rotavirus genotypes circulating in Australia will provide important insights into whether changes in vaccine immunisation programs could impact virus epidemiology and alter strain diversity, which could have ongoing consequences for the success of current and future vaccination programs.

This report describes the G- and P- genotype distribution of rotavirus strains causing severe gastroenteritis in Australia for the period 1 January to 31 December 2019.

Methods

Faecal samples were tested for the presence of rotavirus by quantitative Reverse Transcription Polymerase Chain Reaction (RT-qPCR), enzyme immunoassay (EIA), or latex agglutination by collaborating laboratories Australia-wide. Positive samples were frozen and sent to the National Rotavirus Reference Centre (NRRC) Melbourne, together with available metadata including: date of collection; date of birth; gender; postcode; and the RT-qPCR cycle threshold (Ct) values generated by the collaborating laboratory. Specimens were received from the following 14 collaborating centres located in the Australian Capital Territory (ACT), New South Wales (NSW), Northern Territory (NT), Queensland (Qld), South Australia (SA), Tasmania (Tas.), Victoria (Vic.), and Western Australia (WA) (n = number of specimens received):

- Microbiology Department, Canberra Hospital, ACT (n = 12).
- Microbiology Department, John Hunter Hospital, Newcastle, NSW (n = 27).
- Microbiology Department, SEALS-Randwick, Prince of Wales Hospital, NSW (n = 43).
- Centre for Infectious Diseases & Microbiology, Westmead, NSW (n = 61).
- Douglass Hanly Moir Pathology, NSW (n = 49).
- Microbiology Department, Central Coast, Gosford, NSW (n = 19).
- Pathology Queensland, Royal Brisbane and Women's Hospital, Herston, Qld (n = 289).
- Microbiology and Infectious Diseases Laboratory, SA Pathology, Adelaide, SA (n = 73).
- Molecular Medicine, Pathology Services, Royal Hobart Hospital, Hobart, Tas. (n = 23).
- Department of Microbiology, Monash Medical Centre, Clayton, Vic. (n = 54).
- Molecular Infectious Department, Australian Clinical Labs, Clayton, Vic. (n = 92).

- Serology Department, Royal Children's Hospital, Parkville, Vic. (n = 59).
- QEII Microbiology Department, PathWest Laboratory Medicine, Nedlands, WA (n = 110).
- Territory Pathology, Royal Darwin Hospital, Tiwi, NT (n = 53).

Samples were allocated a unique laboratory code and entered into the NRRC database (Excel and REDCap). Samples were stored at -80 °C until analysed.

Viral RNA was extracted from 10–20% faecal extracts using the QIAamp Viral RNA mini extraction kit (QIAGEN) according to the manufacturer's instructions. Rotavirus G- and P- genotypes were determined using an in-house hemi-nested multiplex RT-PCR assay. The first-round RT-PCR reactions were performed using the One Step RT-PCR kit (QIAGEN), in conjunction with VP7 (VP7F/VP7R) or VP4 (VP4F/VP4R) conserved primers.[16,17] The second-round genotyping PCR reactions were conducted using specific oligonucleotide primers for G types G1, G2, G3, G4, G8, and G9, or P types P[4], P[6], P[8], P[9], P[10], and P[11].[16,18,19] The G- and P- genotype was determined using agarose gel electrophoresis of second-round PCR products. Samples failing to generate a second-round PCR amplicon or with inconclusive results were further tested by VP6-specific RT-PCR using the Superscript III One-Step RT PCR System with Platinum Taq DNA Polymerase (Invitrogen) and primers Rot3/Rot5 as described previously.[20,21]

Sanger sequencing was used to determine the VP7 and/or VP4 nucleotide sequence for PCR non-typeable or VP6-positive samples. The current set of primers in the second-round G-typing protocol are not able to assign genotypes to equine-like G3, G12, and unusual rotavirus strains. The VP7 gene of each G1P[8] sample was sequenced to determine if wild-type or Rotarix vaccine strain was detected. Samples which had no first-round PCR amplicon were re-amplified using the Superscript III One-Step RT PCR System with Platinum Taq DNA Polymerase (Invitrogen), in conjunction with VP7 (Beg9/End9) or VP4 (Con2/Con3) primers, as described previously.[18,19,22] First-round VP7 or VP4 amplicons were purified using the Wizard SV Gel for PCR Clean-Up System (Promega) or the QIAquick Gel Extraction Kit (QIAGEN), according to the manufacturer's protocol. Purified DNA and oligonucleotide primers (VP7F/VP7R, VP4F/VP4R, Beg9/End9 or Con2/Con3) were sent to the Australian Genome Research Facility (AGRF), Melbourne, and sequenced using an ABI PRISM BigDye Terminator Cycle Sequencing Reaction Kit (Applied Biosystems) in an Applied Biosystems 3730xl DNA Analyzer (Applied Biosystems). Electropherograms were visually analysed and edited using Sequencher v.4.10.1. Genotype assignment was determined using BLAST[i] and RotaC v2.0.[23,ii]

Rotavirus has been a notifiable disease in Australia since 2010, with all states and territories reporting through the National Notifiable Diseases Surveillance System (NNDSS) in 2019.[24]

Results

Number of specimens

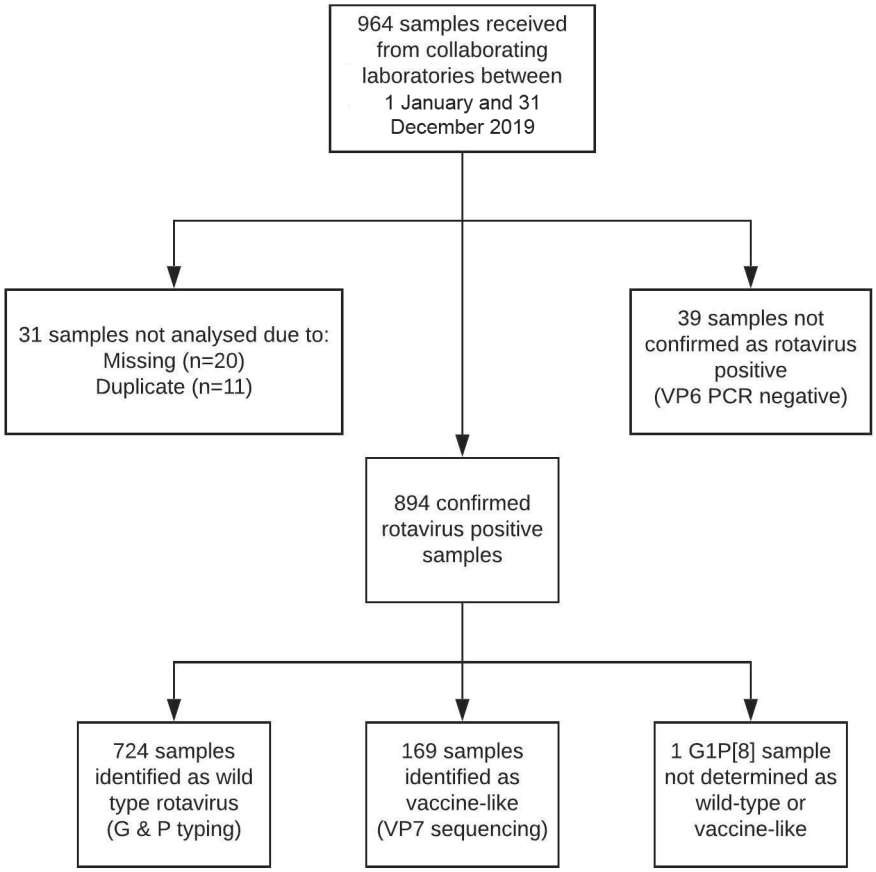
A total of 964 specimens, determined to be rotavirus positive by collaborating laboratories, were sent to NRRC during the period 1 January to 31 December 2019 (Figure 1). A subset of samples were not analysed due to samples missing (not received; n = 20); duplicate (n = 11); or negative by VP6 PCR (n = 39).

A total of 894 samples were genotyped. Samples were then classified as wild-type (no vaccine component identified) or vaccine-like (Rotarix vaccine component identified), based on genotype and the analysis of the top BLAST hits of any G1 VP7 sequence. Of the 724 samples confirmed as wild-type, 338 were collected

i http://blast.ncbi.nlm.nih.gov/Blast.cgi.

ii http://rotac.regatools.be.

Figure 1: Stool sample flowchart



from children < 5 years of age and 384 were from children ≥ 5 years of age and adults. Two further wild-type samples were collected from patients with no recorded age (Table 1). In addition, 169 samples were identified as vaccine-like by VP7 sequencing (Figure 1). A single G1P[8] sample from a 1-month-old failed to yield clean sequencing reads and could not be determined as wild-type or vaccine-like, and was excluded from subsequent analysis (Figure 1).

Rotavirus-positive samples identified by month, compared to national notification rates

Rotavirus-positive samples were analysed by date of collection [month], to determine the peak season (Figure 2). There was a moderate increase in rotavirus detection in June and July, coinciding with the winter months in the southern hemisphere. Most wild-type specimens were collected during September–December, coinciding with the spring-summer period. This trend was also evident in the NNDSS data, where notification rates peaked during September–December (4.0, 3.9, 4.2 and 3.3 per 100,000 population respectively).[24] The September-to-December notification rates were higher than the averages seen for these months in previous years' NNDSS data,[24] indicative of an outbreak.

The NRRC sample numbers for August and October were lower than expected, based on NNDSS data. This may be due to samples from SA and WA not being received for the later months of 2019, as collaborating diagnostic laboratories were focused on SARS-CoV-2 testing when these sample shipments were requested in early 2020. It should be noted that the data between NNDSS and ARSP are not fully reconcilable. Both programs have the potential to underestimate the burden of rotavirus disease: by not all states and territories reporting data to NNDSS; and by ARSP not receiving rotavirus samples for all cases.

Table 1: Age distribution of wild-type rotavirus gastroenteritis cases

Age (months)	Age (years)	Number of cases	Percentage of total	Percentage under 5 years
0–6	≤ 1	47	6.5	13.9
7–12		38	5.2	11.2
13–24	1 – ≤ 2	108	14.9	32.0
25–36	2 – ≤ 3	77	10.6	22.8
37–48	3 – ≤ 4	49	6.8	14.5
49–59	4 – < 5	19	2.6	5.6
Subtotal		**338**	**46.7**	**100**
60–120	5 – ≤ 10	73	10.1	
121–240	10 – ≤ 20	45	6.2	
241–960	20 – ≤ 80	221	30.5	
961+	> 80	45	6.2	
Subtotal		**384**	**53.0**	
Unknown age		2	0.3	
Total		**724**	**100**	

Wild-type rotavirus specimens:

Age distribution for wild-type rotavirus infections

From 1 January to 31 December 2019, 46.7% of rotavirus-positive samples (n = 338/724) were obtained from children < 5 years of age (Table 1). The largest number of positive samples from children < 5 years of age were obtained from the 13–24 month age group, accounting for 32.0% (n = 108/338) of such cases, followed by the 25–36 month age group accounting for 22.8% (n = 77/338) of such cases. In addition, 36.7% of all samples (n = 266/724) were from individuals ≥ 20 years of age.

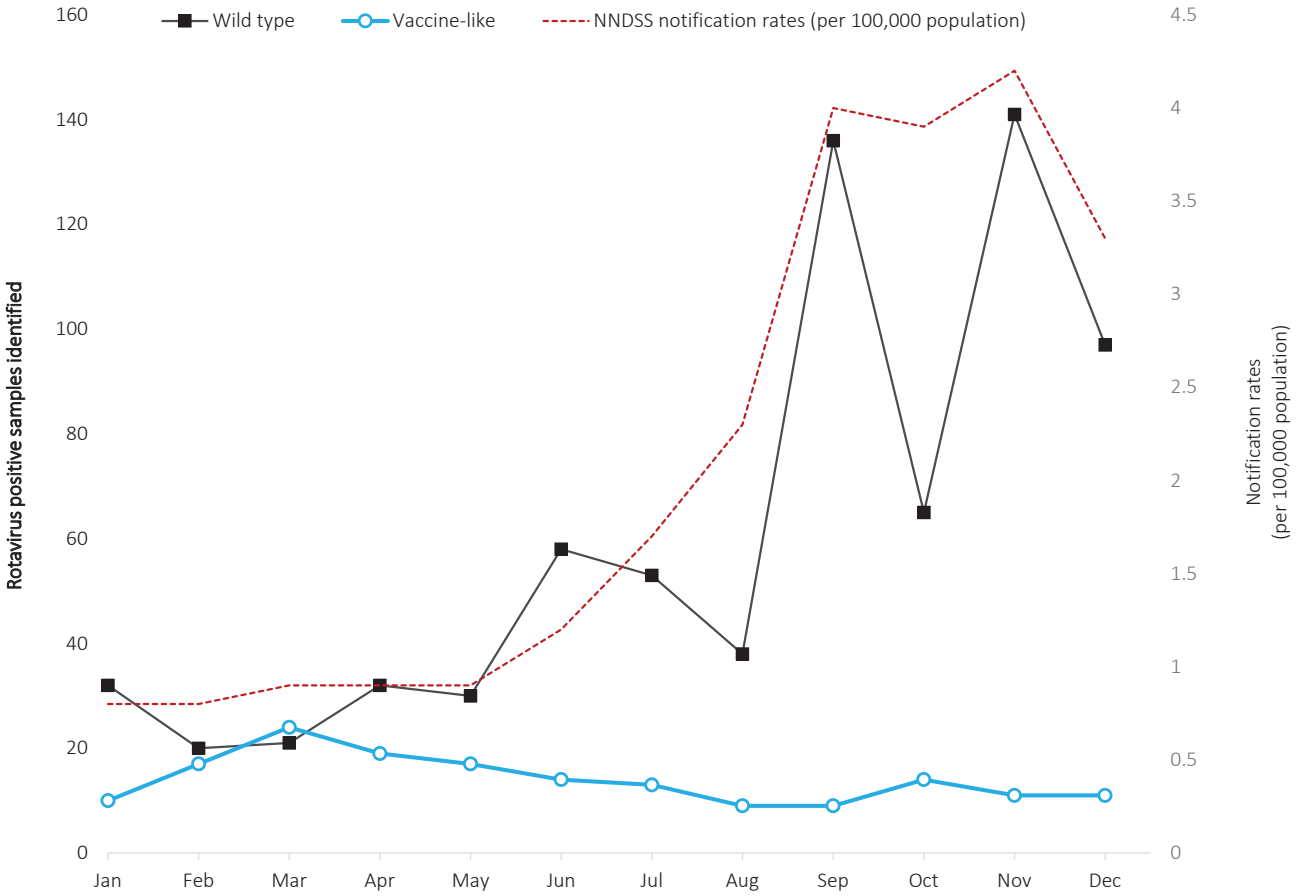
Wild-type rotavirus genotype distribution

Genotype analysis was performed on 724 confirmed rotavirus-positive samples from children and adults (Table 2). G3P[8] was the most common genotype identified nationally, representing 46.7% of all wild-type specimens analysed. G3P[8] was the dominant genotype in New South Wales, Queensland, South Australia, Tasmania, Victoria, and Western Australia, representing 26.3%, 74.0%, 32.5%, 31.3%, 38.1% and 39.1% of samples respectively.

G2P[4] was the second most common genotype, representing 8.8% of all samples nationally. This was the dominant genotype in the Northern Territory, representing 48.1% of samples from that jurisdiction; it was also prevalent in Tasmania (25.0%) and Victoria (16.5%). Other common genotypes nationally included G9P[8] (8.1%), G8P[8] (5.8%), equine-like G3P[8] (5.7%), and the previously uncommon genotypes G2P[8] (8.1%), G9P[4] (3.6%) and G3P[4] (4.3%).

Of the 17 specimens identified as mixed or 'other' (2.3% of wild-type samples), six were uncommon or unusual genotype combinations, including G1P[4] (n = 1), G2P[6] (n = 1), G3P[9]

Figure 2: Number of analysed wild-type and vaccine-like specimens compared to NNDSS rotavirus notification rates per 100,000 population,[a,b] Australia, 1 January to 31 December 2019



a NNDSS: National Notifiable Diseases Surveillance System notification rates for rotavirus.[24]

b Note: 1 wild-type sample and 1 vaccine-like sample had no date of collection recorded.

(n = 1), G8P[4] (n = 2) and G12P[8] (n = 1). The remaining ten samples exhibited an animal VP7 and/or VP4 gene: canine/feline-like G3P[3] (n = 4), equine-like G3P[4] (n = 1), canine/feline-like G3P[8] (n = 2), feline-like G3P[8] (n = 1) and bovine-like G10P[14] (n = 2) (Table 3). One sample with a mixed genotype (G3/G4P[8]) was identified (Table 3).

Genotypes identified in samples from children < 5 years of age

A total of 338 wild-type samples were collected from children < 5 years of age (Table 4). Within this subset, G3P[8] was the most common genotype, found in 51.2% of all samples, followed by G2P[4] (7.7%), equine-like G3P[8] (6.8%), G9P[8] (6.5%), G2P[8] (6.5%) and G8P[8] (6.2%). G1P[8], G3P[4] and G9P[4] represented minor genotypes (0.3–3.6%).

Genotypes identified in samples from individuals ≥ 5 years of age

A total of 384 rotavirus-positive samples were collected from children ≥ 5 years of age and adults (Table 5). Similar to the < 5 years cohort, G3P[8] was the dominant genotype (42.7%), followed by G2P[4] (9.9%), G2P[8] (9.6%) and G9P[8] (9.4%). G1P[8], G3P[4], equine-like G3P[8], G8P[8] and G9P[4] represented minor genotypes (1.6–5.5%).

Table 2: Rotavirus G and P genotype distribution in infants, children and adults, 1 January to 31 December 2019

Jurisdiction	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Non-type[b]		Other[c]	
	n	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Australian Capital Territory	9	0	–	0	–	1	11.1	0	–	3	33.3	0	–	5	55.6	0	–	0	–	0	–	0	–
New South Wales	171	2	1.2	10	5.8	23	13.5	13	7.6	45	26.3	33	19.3	12	7.0	7	4.1	7	4.1	14	8.2	5	2.9
Northern Territory	27	0	–	13	48.1	1	3.7	0	–	4	14.8	0	–	0	–	1	3.7	0	–	2	7.4	6	22.2
Queensland	254	0	–	13	5.1	15	5.9	8	3.1	188	74.0	2	0.8	14	5.5	1	0.4	7	2.8	4	1.6	2	0.8
South Australia	40	0	–	1	2.5	5	12.5	1	2.5	13	32.5	1	2.5	3	7.5	0	–	9	22.5	7	17.5	0	–
Tasmania	16	0	–	4	25.0	1	6.3	0	–	5	31.3	0	–	0	–	2	12.5	3	18.8	1	6.3	0	–
Victoria	97	3	3.1	16	16.5	5	5.2	4	4.1	37	38.1	3	3.1	5	5.2	2	2.1	14	14.4	6	6.2	2	2.1
Western Australia	110	2	1.8	7	6.4	8	7.3	5	4.5	43	39.1	2	1.8	3	2.7	13	11.8	19	17.3	6	5.5	2	1.8
Total	**724**	**7**	**1.0**	**64**	**8.8**	**59**	**8.1**	**31**	**4.3**	**338**	**46.7**	**41**	**5.7**	**42**	**5.8**	**26**	**3.6**	**59**	**8.1**	**40**	**5.5**	**17**	**2.3**

a Equine-like G3P[8].
b A specimen where G- and/or P- genotype was not determined.
c See Table 3.

Table 3: Mixed and unusual G and P genotypes identified in infants, children and adults, 1 January to 31 December 2019

Genotype	NSW	NT	Qld	Vic.	WA	Total
G1P[4]	1	–	–	–	–	1
G2P[6]	–	–	–	1	–	1
G3P[3] canine/feline-like	1	3	–	–	–	4
G3P[4] equine-like	–	–	1	–	–	1
G3P[8] canine/feline-like	–	2	–	–	–	2
G3P[8] feline-like	–	1	–	–	–	1
G3P[9]	–	–	–	–	1	1
G8P[4]	2	–	–	–	–	2
G10P[14] bovine-like	1	–	–	–	1	2
G12P[8]	–	–	1	–	–	1
Mixed[a]	–	–	–	1	–	1
Total	**5**	**6**	**2**	**2**	**2**	**17**

a G3/G4P[8].

Vaccine-like rotavirus specimens:

Age distribution for rotavirus vaccine cases

All G1P[8] samples (n = 177) were analysed by VP7 sequencing to identify vaccine-like strains. A single G1P[8] sample from a 1-month-old failed to yield clean sequencing reads and could not be determined as wild-type or vaccine-like.

A total of 176 samples were successfully sequenced: 169 were confirmed to be Rotarix vaccine-like strains and seven were wild-type. Of the vaccine-like samples, 94.7% (n = 160/169) were from the 0–6 month age group. Vaccine-like rotavirus was also detected in patients aged 2, 11, 26, 56, and 57 years old (Table 6). One G1P[8] vaccine-like sample was detected in a patient whose age was unknown.

Discussion

The 2019 ARSP report describes the distribution of rotavirus genotypes causing gastroenteritis in Australia for the period 1 January to 31 December 2019, the second full year of exclusive use of Rotarix within the NIP.[7,14] Rotavirus vaccines have been reported to alter rotavirus epidemiological patterns from annual to biennial peaks, a trend previously observed by ARSP.[6,25] In 2019, an increase in notifications for rotavirus disease was reported in most states/territories, with a peak in notifications and samples submitted to ARSP in September, November and December. Compared to previous years (2010–2018), the notification rates for July to December 2019 were higher than average, with rates for November and December 2019 the highest reported for these months to date (4.2/100,000 and 3.3/100,000 population respectively).[24] This is highly suggestive of a rotavirus outbreak. The predominant rotavirus genotype reported in Queensland between September and December was G3P[8] (82.6–89.3% of samples). In New South Wales, G3P[8] was dominant between September and November (28.9–45.8% of samples), with equine-like G3P[8] increasing in November (21.1% of samples) and becoming dominant in December (60.0% of samples) (Appendix 1). The link between the high rate of notifications with the dominance of human and equine G3P[8] suggests that these genotypes were likely to be responsible for the outbreaks observed in New South Wales and Queensland.

Table 4: Rotavirus G and P genotype distribution in infants and children under 5 years of age, 1 January to 31 December 2019

Jurisdiction	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Non-type[b]		Other[c]	
	n	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Australian Capital Territory	5	0	–	0	–	0	–	0	–	1	20.0	0	–	4	80.0	0	–	0	–	0	–	0	–
New South Wales	90	0	–	3	3.3	11	12.2	7	7.8	26	28.9	21	23.3	7	7.8	1	1.1	3	3.3	8	8.9	3	3.3
Northern Territory	25	0	–	13	52.0	1	4.0	0	–	4	16.0	0	–	0	–	1	4.0	0	–	1	4.0	5	20.0
Queensland	119	0	–	6	5.0	4	3.4	3	2.5	92	77.3	0	–	7	5.9	1	0.8	4	3.4	1	0.8	1	0.8
South Australia	14	0	–	1	7.1	1	7.1	0	–	5	35.7	0	–	2	14.3	0	–	3	21.4	2	14.3	0	–
Tasmania	2	0	–	0	–	0	–	0	–	1	50.0	0	–	0	–	0	–	0	–	1	50.0	0	–
Victoria	35	1	2.9	2	5.7	3	8.6	1	2.9	18	51.4	2	5.7	0	–	1	2.9	5	14.3	2	5.7	0	–
Western Australia	48	0	–	1	2.1	2	4.2	1	–	26	54.2	0	–	1	2.1	7	14.6	7	14.6	3	6.3	0	–
Total	**338**	**1**	**0.3**	**26**	**7.7**	**22**	**6.5**	**12**	**3.6**	**173**	**51.2**	**23**	**6.8**	**21**	**6.2**	**11**	**3.3**	**22**	**6.5**	**18**	**5.3**	**9**	**2.7**

a Equine-like G3P[8].
b A specimen where G- and/or P- genotype was not determined.
c See Table 3.

Table 5: Rotavirus G and P genotype distribution in children ≥ 5 years of age and adults, 1 January to 31 December 2019

Jurisdiction	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Non-type[b]		Other[c]	
	n	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
Australian Capital Territory	4	0	–	0	–	1	25.0	0	–	2	50.0	0	–	1	25.0	0	–	0	–	0	–	0	–
New South Wales	81	2	2.5	7	8.6	12	14.8	6	7.4	19	23.5	12	14.8	5	6.2	6	7.4	4	4.9	6	7.4	2	2.5
Northern Territory	2	0	–	0	–	0	–	0	–	0	–	0	–	0	–	0	–	0	–	1	50.0	1	50.0
Queensland	135	0	–	7	5.2	11	8.1	5	3.7	96	71.1	2	1.5	7	5.2	0	–	3	2.2	3	2.2	1	0.7
South Australia	27	0	–	0	–	4	14.8	1	3.7	8	29.6	1	3.7	1	3.7	0	–	6	22.2	6	22.2	0	–
Tasmania	15	0	–	4	26.7	1	6.7	0	–	4	26.7	0	–	0	–	2	13.3	3	20.0	1	6.7	0	–
Victoria	58	2	3.4	14	24.1	2	3.4	3	5.2	18	31.0	1	1.7	5	8.6	1	1.7	8	13.8	2	3.4	2	3.4
Western Australia	62	2	3.2	6	9.7	6	9.7	4	6.5	17	27.4	2	3.2	2	3.2	6	9.7	12	19.4	3	4.8	2	3.2
Total	**384**	**6**	**1.6**	**38**	**9.9**	**37**	**9.6**	**19**	**4.9**	**164**	**42.7**	**18**	**4.7**	**21**	**5.5**	**15**	**3.9**	**36**	**9.4**	**22**	**5.7**	**8**	**2.1**

a Equine-like G3P[8].
b A specimen where G- and/or P- genotype was not determined.
c See Table 3.

Table 6: Age distribution of vaccine-like rotavirus gastroenteritis cases

Age (months)	Age (years)	Number of cases	Percentage of total	Percentage under 5 years
0–6	≤ 1	160	94.7	97.6
7–12		3	1.8	1.8
13–24	1 – ≤ 2	0	–	–
25–36	2 – ≤ 3	1	0.6	0.6
37–48	3 – ≤ 4	0	–	–
49–59	4 – < 5	0	–	–
Subtotal		**164**	**97.0**	**100**
60–120	5 – ≤ 10	0	–	
121–240	10 – ≤ 20	1	0.6	
241–960	20 – ≤ 80	3	1.8	
961+	> 80	0	–	
Subtotal		**4**	**2.4**	
Unknown age		1	0.6	
Total		**169**	**100**	

In New South Wales, an increase in G2P[8], G3P[8], equine-like G3P[8] and a decrease of G2P[4] was observed in 2019 compared to 2018 (Table 2). A reduced number of samples were submitted from both South Australia and Western Australia, as a result of logistical issues due to the COVID-19 pandemic (Table 2).

During this reporting period, human G3P[8] was the predominant genotype circulating nationally, comprising 46.7% of all samples, and was the dominant genotype in six of the eight states and territories (Table 2). G2P[4] was the second most common genotype identified nationally (8.8%), detected in all states and territories except the Australian Capital Territory (Table 2). An increase in G2P[8] and G3P[4] genotypes was observed in comparison to previous years (Table 2).

Differences in genotypes observed in 2019 versus 2018 were most apparent in the < 5-year-old age group, where increases in G3P[8], equine-like G3P[8], G2P[8] and G3P[4] were observed. This is of particular interest as the patients had most likely received a recent rotavirus vaccine. Within this age group, the proportion of G3P[8] decreased from 65% in 2018 to 51.2% in 2019 (Table 4). However, in the older age group (≥ 5 years), G3P[8] was seen in a similar proportion to 2018, although the distribution between states varied substantially. In New South Wales and Queensland, the increase in G3P[8] was likely associated with an outbreak during the later months of the year. In the overall annual period in Queensland and Tasmania, G3P[8] was seen in a similar proportion to 2018 in both the < 5 and ≥ 5 years of age groups (Tables 4 & 5). G2P[4] genotypes decreased in New South Wales in both the < 5 years and ≥ 5 years of age groups, compared to 2018 (Tables 4 & 5). In Queensland, an increase in G3P[8] and a decrease in G2P[4], equine-like G3P[8] and G9P[8] in both the < 5 year and ≥ 5 years of age groups was observed (Tables 4 & 5). In children < 5 years of age, an increase in G8P[8] and G9P[8] was observed when compared to 2018. In 2019, children aged 13–24 months, and adults aged 20–80 years were key age groups reported with rotavirus disease, similar to that observed in 2018 (Table 1). This shift in age towards an older population has previously been observed in Australia and worldwide.[6,26–29] It is possible that waning immunity (both vaccine and naturally

acquired) and child-to-adult transmission may contribute to an increase of rotavirus disease in the older population.

Vaccine-like G1P[8] was consistently detected at a low level throughout the year (Figure 2). Vaccine-like G1P[8] strains were not only detected in the expected cohort of recently vaccinated children (0–8 months of age), but also in five individuals that ranged in age from two to 57 years. The horizontal transmission of vaccine strains from vaccinated infants to close contacts has been reported elsewhere.[30–32]

Since vaccine introduction in the Australian NIP, G1P[8] has drastically decreased in prevalence, from 53.4% in the pre-vaccine era to 26.2% in the vaccine era.[14] This trend was also observed during this reporting period, where G1P[8] was only detected in 1.0% of samples, similar to 2018. Despite being a dominant genotype during 2013–2015, G12P[8] prevalence has continued to decline in recent years, accounting for 0.1–1% of samples between 2017 and 2019.[6,14] This highlights the ongoing fluctuations in genotype diversity in Australia over time, with the seemingly periodic replacement of genotypes in the population.

Both Rotarix and RotaTeq provide broad homotypic and heterotypic protection against common genotypes (i.e. G1P[8], G2P[4], G3P[8], G4P[8], and G9P[8]); however, the increase in inter-genogroup reassortant strains, unusual genotypes, and zoonotic strains, including equine-like G3P[8] and G12P[8], create uncertainty as to whether these vaccines will perform against these emerging strains.[33,34] Of the 724 rotavirus-positive samples presented in this report, 17 were mixed or unusual G and P genotypes (Table 3). These unusual genotype combinations could be inter-genogroup reassortants, such as Wa-1-like undergoing reassortment with DS-1-like or AU-1-like strains resulting in genotypes such as G2P[8], G3P[4], G3P[8] or G9P[4]; or zoonotic in nature, including canine/feline-like G3P[8], canine/feline-like G3P[3], equine-like G3P[4] and bovine-like G10P[14]. Of interest, G2P[8], G3P[4] and G9P[4], which were previously considered unusual or rare,[6] comprised 16% of samples (116/724). As seen in previous ARSP reports, these rare/unusual genotypes appear to be increasing in frequency. It is yet to be determined if this is a natural phenomenon or is influenced by the Australian NIP.

In this 2019 annual report, we document the variation in circulating rotavirus genotypes during the second full year of surveillance following the change of all states and territories to Rotarix within the NIP. An increase in rotavirus disease was reported by NNDSS in 2019, coinciding with an increase in rotavirus-positive specimens submitted to ARSP, with an outbreak of rotavirus disease observed in September, November and December. The pattern observed in 2019 was not dissimilar to 2017, when a higher rate of rotavirus-positive samples and outbreaks was reported.[6] Genotypes associated with these 2017 outbreaks included G2P[4], G3P[8] equine-like and G8P[8].[6] However, in 2019, G3P[8] was the dominant genotype across six out of eight states and territories and likely responsible for the outbreaks observed in New South Wales and Queensland. Equine-like G3P[8] was also observed in association with the New South Wales outbreak. G2P[4] was the second most prominent genotype identified across the year throughout Australia. ARSP monitors the shift in genotypes causing disease in Australia with the aim to inform disease surveillance activities and maintain an effective vaccination program.

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Appendix A: Rotavirus G and P genotype distribution in infants, children and adults, by jurisdiction and month of collection, 2019

Australian Capital Territory	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	0																						
February	1													1	100.0								
March	1													1	100.0								
April	3									1	33.3			2	66.7								
May	0																						
June	1													1	100.0								
July	0																						
August	0																						
September	0																						
October	1									1	100.0												
November	2					1	50.0			1	50.0												
December	0																						
Total	**9**	**0**	**0**	**0**	**0**	**1**	**11.1**	**0**	**0**	**3**	**33.3**	**0**	**0**	**5**	**55.6**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**

a G3P[8] equine-like.

New South Wales	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	2					1	50.0							1	50.0								
February	4							1	25.0	1	25.0			1	25.0							1	25.0
March	2							1	50.0							1	50.0						
April	4					1	25.0							1	25.0					1	25.0	1	25.0
May	4					1	25.0			1	25.0					1	25.0					1	25.0
June	8					2	25.0	1	12.5	4	50.0											1	12.5
July	10			1	10.0					2	20.0			1	10.0	2	20.0	2	20.0			2	20.0
August	21			2	9.5	8	38.1			4	19.0			2	9.5	1	4.8	2	9.5	1	4.8	1	4.8
September	24	1	4.2			5	20.8	2	8.3	11	45.8	1	4.2			1	4.2			1	4.2	2	8.3
October	14			4	28.6			1	7.1	6	42.9			3	21.4								
November	38	1	2.6	1	2.6	4	10.5	5	13.2	11	28.9	8	21.1	3	7.9	1	2.6	1	2.6	1	2.6	2	5.3
December	40			2	5.0	1	2.5	2	5.0	5	12.5	24	60.0					2	5.0	1	2.5	3	7.5
Total	**171**	**2**	**1.2**	**10**	**5.8**	**23**	**13.5**	**13**	**7.6**	**45**	**26.3**	**33**	**19.3**	**12**	**7.0**	**7**	**4.1**	**7**	**4.1**	**5**	**2.9**	**14**	**8.2**

a G3P[8] equine-like.

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Northern Territory	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	0																						
February	0																						
March	0																						
April	0																						
May	1					1	100.0																
June	0																						
July	1																			1	100.0		
August	2																					2	100.0
September	1																			1	100.0		
October	2																			2	100.0		
November	4			2	50.0											1	25.0			1	25.0		
December	16			11	68.8					4	25.0						0.0			1	6.3		
Total	**27**	**0**	**0**	**13**	**48.1**	**1**	**3.7**	**0**	**0**	**4**	**14.8**	**0**	**0**	**0**	**0**	**1**	**3.7**	**0**	**0**	**6**	**22.2**	**2**	**7.4**

a G3P[8] equine-like.

Queensland	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	n	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%	n	%
January	23			12	52.2	6	26.1	2	8.7	2	8.7											1	4.3
February	2					1	50.0			1	50.0												
March	5											1	20.0					3	60.0	1	20.0		
April	5					2	40.0											2	40.0	1	20.0		
May	7					1	14.3	1	14.3	4	57.1							1	14.3				
June	0																						
July	0																						
August	1																					1	100.0
September	86					3	3.5	3	3.5	71	82.6			6	7.0			1	1.2			2	2.3
October	31			1	3.2			1	3.2	27	87.1			1	3.2	1	3.2						
November	75							1	1.3	67	89.3			7	9.3								
December	19					2	10.5			16	84.2	1	5.3										
Total	**254**	**0**	**0**	**13**	**5.1**	**15**	**5.9**	**8**	**3.1**	**188**	**74.0**	**2**	**0.8**	**14**	**5.5**	**1**	**0.4**	**7**	**2.8**	**2**	**0.8**	**4**	**1.6**

a G3P[8] equine-like.

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South Australia	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	6					2	33.3	1	16.7	2	33.3											1	16.7
February	3									2	66.7											1	33.3
March	5					2	40.0					1	20.0					1	20.0			1	20.0
April	7													2	28.6			3	42.9			2	28.6
May	1																					1	100.0
June	2									1	50.0							1	50.0				
July	5									4	80.0							1	20.0				
August	1			1	100.0																		
September	7					1	14.3			2	28.6			1	14.3			2	28.6			1	14.3
October	3									2	66.7							1	33.3				
November	0																						
December	0																						
Total	**40**	**0**	**0**	**1**	**2.5**	**5**	**12.5**	**1**	**2.5**	**13**	**32.5**	**1**	**2.5**	**3**	**7.5**	**0**	**0**	**9**	**22.5**	**0**	**0**	**7**	**17.5**

a G3P[8] equine-like.

Tasmania	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	0																						
February	0																						
March	0																						
April	0																						
May	0																						
June	0																						
July	1																	1	100.0				
August	4					1	25.0									1	25.0	1	25.0			1	25.0
September	2			1	50.0											1	50.0						
October	3			2	66.7													1	33.3				
November	2			1	50.0					1	50.0												
December	4									4	100.0												
Total	**16**	**0**	**0**	**4**	**25.0**	**1**	**6.3**	**0**	**0**	**5**	**31.3**	**0**	**0**	**0**	**0**	**2**	**12.5**	**3**	**18.8**	**0**	**0**	**1**	**6.3**

a G3P[8] equine-like.

Commun Dis Intell (2018) 2021;45 (https://doi.org/10.33321/cdi.2021.45.4) Epub 29/1/2021

Victoria	Total[a]	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][b]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	1									1	100.0												
February	1																					1	100.0
March	1																	1	100.0				
April	1											1	100.0										
May	3	1	33.3	1	33.3											1	33.3						
June	12	1	8.3	3	25.0	4	33.3			1	8.3			1	8.3			2	16.7				
July	14	1	7.1	1	7.1	1	7.1			6	42.9			2	14.3			3	21.4				
August	5			2	40.0					2	40.0			1	20.0								
September	10			3	30.0					3	30.0							1	10.0	1	10.0	2	20.0
October	11			2	18.2			2	18.2	5	45.5							2	18.2				
November	19			2	10.5					11	57.9			1	5.3			3	15.8	1	5.3	1	5.3
December	18			2	11.1			2	11.1	8	44.4	2	11.1			1	5.6	1	5.6			2	11.1
Total	**96**	**3**	**3.1**	**16**	**16.7**	**5**	**5.2**	**4**	**4.2**	**37**	**38.5**	**3**	**3.1**	**5**	**5.2**	**2**	**2.1**	**13**	**13.5**	**2**	**2.1**	**6**	**6.3**

a No date of collection given for one sample.
b G3P[8] equine-like.

Western Australia	Total	G1P[8]		G2P[4]		G2P[8]		G3P[4]		G3P[8]		G3P[8][a]		G8P[8]		G9P[4]		G9P[8]		Other		Non-type	
Month	**n**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**	**n**	**%**
January	0																						
February	9					1	11.1	1	11.1	2	22.2					2	22.2	2	22.2			1	11.1
March	7	1	14.3	1	14.3	1	14.3			2	28.6	1	14.3									1	14.3
April	12					1	8.3	1	8.3	8	66.7			1	8.3			1	8.3				
May	14					3	21.4	1	7.1	4	28.6			1	7.1	2	14.3			1	7.1	2	14.3
June	35	1	2.9	3	8.6	1	2.9	2	5.7	11	31.4	1	2.9	1	2.9	3	8.6	10	28.6	1	2.9	1	2.9
July	22			1	4.5	1	4.5			10	45.5					5	22.7	4	18.2			1	4.5
August	4									4	100.0												
September	6			1	16.7					2	33.3					1	16.7	2	33.3				
October	0																						
November	1			1	100.0																		
December	0																						
Total	**110**	**2**	**1.8**	**7**	**6.4**	**8**	**7.3**	**5**	**4.5**	**43**	**39.1**	**2**	**1.8**	**3**	**2.7**	**13**	**11.8**	**19**	**17.3**	**2**	**1.8**	**6**	**5.5**

a G3P[8] equine-like.