Journal of Clinical Virology 60 (2014) 282–289

Contents lists available at ScienceDirect

Journal of Clinical Virology

journal homepage: www.elsevier.com/locate/jcv

Surveillance of group A Rotavirus in Buenos Aires 2008–2011, long lasting circulation of G2P[4] strains possibly linked to massive monovalent vaccination in the region

Marcelo G. Mandile[a,*], Laura E. Esteban[a], Marcelo H. Argüelles[a], Alicia Mistchenko[b], Graciela Glikmann[a], Alejandro A. Castello[a]

[a] Laboratorio de Inmunología y Virología, Universidad Nacional de Quilmes, Roque Sáenz Peña 352, Bernal 1876, Argentina
[b] Laboratorio de Virología, Hospital de Niños Ricardo Gutiérrez, Sánchez de Bustamante 1330, Buenos Aires 1425, Argentina

ARTICLE INFO

Article history:
Received 10 January 2014
Received in revised form 27 March 2014
Accepted 25 April 2014

Keywords:
Rotavirus
Diarrhea
Vaccines
Phylogeny
Argentina

ABSTRACT

Background: Group A rotaviruses (RVA) are the most frequent single etiological agents of severe diarrhea in infants. Since 2006 RVA vaccines have been introduced in national schedules of middle and high income countries with substantial declines in rotavirus associated disease burden. However, surveillance must be maintained to, eventually, detect emerging types or variants selected by the new pressure imposed by vaccination.
Objectives: To analyze the molecular epidemiology of group A rotavirus after vaccine introduction in the region in the context of data from more than 15 years of continuous surveillance in Buenos Aires.
Study design: RVA positive diarrhea samples collected in Buenos Aires from 2008 to 2011 were genotyped by RT-PCR. Selected samples were sequenced to gain insight on evolution of common and globally emerging human RVA strains.
Results: Lineage III G12P[8] strain emerged in 2008 in Buenos Aires and shared co-dominancy with G3 strains during 2009. An atypical long lasting circulation of G2P[4] strains since 2004 reached rates around 80% in 2011 in Buenos Aires. Sequencing of the VP7 and VP4 genes of representative G2P[4] isolates suggests Brazil as the origin of the 2010–2011 strains.
Conclusions: Globally emergent G12 lineage III strains could be established as dominant strains in a very populated area in two years since emergence. In this work it was also shown that the persistence of G2P[4] strains during 8 years could be related to massive immunization with the monovalent vaccine in the region.

1. Background

Group A rotaviruses (RVA) are the most frequent single etiological agents causing severe diarrhea in infants. It has been estimated that ≈453,000 children under 5 years of age die of rotavirus disease each year worldwide but around 90% of these fatalities occur in poor regions [1]. Since 2006, two RVA vaccines were licensed in many countries: Rotarix (GlaxoSmithKline, Research Triangle Park, NC) and Rotateq (Merck and Co., Whitehouse Station, NJ). Within the first years following vaccine introduction in national schedules of middle and high income countries, substantial declines in all-cause gastroenteritis hospitalization and even larger declines in rotavirus gastroenteritis hospitalizations among children <5 years of age have been observed [2]. Despite this, no RVA vaccine has yet been incorporated in the National Vaccination and Immunization Program in Argentina. Thereby, vaccination is only administered by private prescription covering around 7% of the birth cohort on the basis of sales estimates (E. López, pers. comm.).

Rotaviruses are nonenveloped viruses and their genome consists of 11 double stranded RNA segments. Virus particles possess a triple-layered icosahedral protein capsid. The two outer capsid proteins, VP7 and VP4 elicit neutralizing antibodies and form the basis of a dual classification system in serotypes. The VP7 glycoprotein, defines the G types and VP4, a protease-cleaved protein, the P types [3]. Currently, 27 G genotypes (G1–G27) and 37 P genotypes (P[1]–P[37]) have been described for RVA [4,5]. To obtain a deeper insight in rotavirus evolution, all the genomic RNA segments are similarly analyzed and classified in genotypes [6].

* Corresponding author. Tel.: +54 11 43657100x5634.
E-mail address: mgmandile@gmail.com (M.G. Mandile).

http://dx.doi.org/10.1016/j.jcv.2014.04.022
1386-6532/

Studies on strain spreading and phylodynamics showed that new reassortants including emergent genotypes like G9 and G12 can spread worldwide in approximately one decade [7]. Nevertheless, generalization of the RVA vaccine use could eventually impose a selective pressure on circulating strains, possibly influencing the future selected variants. As an example, there is a current controversy about the possible influence of massive vaccination with the monovalent G1P[8] vaccine in Brazil on the observed high prevalence levels of G2P[4] strains shortly after vaccine introduction [8,9]. However, some investigators have considered that G2P[4] strains naturally appear as dominant strains in a population about every 10 years, a phenomenon observed when long periods of surveillance were sustained in a given region [8,10,11].

Latin America was one of the first regions in the world where vaccination against RVA was implemented and until now, eight South American countries have introduced the monovalent G1P[8] vaccine (Rotarix) in their National Immunization Schedules. Studies from some of these countries have shown significant declines in RVA related hospitalizations and deaths after vaccine introduction [12]. Our neighboring country, Brazil, was one of the first taking this initiative, beginning vaccination in 2006. However, Argentina, Chile and Uruguay have not yet introduced any RVA vaccine in their National Immunization Schedules.

2. Objectives

To analyze the molecular epidemiology of group A rotavirus in Buenos Aires, Argentina and to establish the influence of vaccine introduction in the region in the context of data from more than 15 years of continuous surveillance.

3. Study design

3.1. Sample collection

Six hundred and sixty three fecal specimens were collected between January 2008 and December 2011 from children under 5 years of age with acute diarrhea admitted at the Hospital "Dr Eduardo Oller" in San Francisco Solano (located at 15 km from Buenos Aires city) and Hospital "Dr Ricardo Gutierrez" in Buenos Aires city. This period included 4 winter seasons in which the peak of rotavirus prevalence in Buenos Aires occurred from May to July. Samples were analyzed for the presence of RVA with an in-house ELISA described elsewhere [13].

3.2. RNA extraction and RT-PCR

Viral RNA was extracted from every rotavirus positive stool specimen by the silica powder method [14] and stored. Extracted RNA was denatured at 97 °C for 5 min and a first amplification of VP7 or VP4 segments was performed by RT-PCR with the One-Step RT-PCR kit (QIAGEN GmbH, Germany) with VP7 or VP4 gene specific consensus primers. To determine human G and P genotypes, the first amplicons were submitted to seminested, multiplex PCR with consensus and type specific primers. For G typing, consensus primers VP7F and VP7R [15] were used in the first round RT-PCR and the VP7R consensus primer with a set of G type specific forward primers in the second round PCR. This set contains specific primers for the genotypes G1, G2, G4, G8 [16]; genotypes G3, G9, G10 [17] and genotype G12 [18]. For P typing, consensus primers VP4F and VP4R [19] were used in the first round RT-PCR and the VP4F primer with a set of P type specific primers in the second round PCR. This set contains specific primers for genotypes P[4], P[6], P[9], P[10] [20], and genotype P[8] [21]. Cycle conditions and visualization of PCR products were conducted as reported before [11].

3.3. Nucleotide sequence analysis

The VP7 (from 25 samples) and VP4 (from 19 samples) PCR products genotyped as G2 or G12 were gel purified with the QIAquick gel extraction kit (QIAGEN GmbH, Hilden, Germany) and sequenced with the automated sequencer ABI3730XL under BigDye terminator cycling conditions (Macrogen Inc., Korea). The nucleotide sequences presented in this article have been submitted under GenBank accession nos. KF920553–KF920596.

3.4. Phylogenetic analysis

Phylogenetic trees were reconstructed with the neighbor-joining method and their evolutionary distance was estimated by Kimura-2 parameter at the nucleotide level. The bootstrap probability was calculated for each internal branch of the phylogenetic trees with 1000 times of resampling. These analyses were conducted by using the program MEGA version 5.1 [22].

4. Results

4.1. Genotype frequency fluctuation

The present results along with previous findings [11,13,23] complete 16 years of continuous genotype surveillance in highly populated areas of Greater Buenos Aires, Argentina. Such information allowed us to establish the natural pattern of fluctuations in genotype frequencies in this area for the prevaccination era (1996–2007) in a previous report [11].

In the present study conducted from 2008 to 2011, 21.0% of the fecal specimens screened by ELISA were positive for RVA (Table 1) and the results from genotyping performed on these positive samples are depicted in Table 2. From the total of typified samples, 11.5% were mixed infections (Table 1), while 6.4% were G or P non typeables. Two remarkable features are evident for this period: first, G12 strains emerged in 2008, reaching the highest frequency in 2009 (44.0%) and second, the high rate of G2 strains in 2011 (85.3%) after 8 years of almost continuous circulation. Different to previously characterized G12 strains detected at low frequencies from 1999 to 2007, in the period reported here this genotype emerged at higher rates in 2008, became dominant in 2009, and persisted during 2010 but was not detected in 2011 in our collection. G2 strains were detected at medium to high rates from 2004 to 2008, were not detected in 2009 and reemerged in 2010. In this way, in a lapse of 8 years, two phases of G2 circulating strains can be described according to whether they were isolated before or after 2009.

In 2011 the genotype combination G2P[4] became dominant reaching a frequency of 78.3%, displacing the G3P[8] and G12P[8] genotypes. It is also of note, the detection of G3P[8] as codominant strains in 2008 and 2009 and as the most prevalent strains in 2010 Table 3. This tendency was also detected by the National Rotavirus Surveillance System [24] and is, to the best of our knowledge, the first time that the G3 genotype is detected at high rates in Buenos Aires since middle of the 1990s.

4.2. Nucleotide phylogenetic analysis

G12 strains. A total of 16 G12 strains from our collection were analyzed by VP7 gene sequencing (1 from 2007, 2 from 2008, 11 from 2009 and 2 from 2010). Through phylogenetic analysis of these sequences, as well as of other Argentine, South American and global representative G12 strains, it was observed that the Buenos Aires strains from 2008 to 2010 belonged to the VP7 lineage III. This group appeared clearly separated from the group of strains from our collection isolated in the period 1999–2007 (Fig. 1) previously characterized as G12 VP7 lineage II, most of them combined with

Table 1
Incidence of group A rotavirus from 2008 to 2011 in Buenos Aires.

Year	No. of positive samples/no. of samples tested (%)				
	2008	2009	2010	2011	Total
RVA infections	36/194 (18.6)	19/163 (11.7)	15/79 (19.0)	69/227 (30.4)	139/663 (21.0)
RVA mixed infections	4/36 (11.1)	5/19 (26.3)	2/15 (13.3)	5/69 (7.2)	16/139 (11.5)

RVA – group A rotavirus.

Table 2
Distribution and frequency of G and P genotypes of group A rotavirus from 2008 to 2011 detected in Buenos Aires.

Genotype	2008		2009		2010		2011		Total no.	% Over the 4-year period
	No. of samples	%	No. of samples	%	No. of samples	%	No. of samples	%		
G1	6	15.0	1	4.0	1	5.9	0	0.0	8	5.1
G2	3	7.5	0	0.0	5	29.4	64	85.3	72	45.9
G3	10	25.0	12	48.0	8	47.0	8	10.7	38	24.2
G4	7	17.5	0	0.0	0	0.0	0	0.0	7	4.4
G9	9	22.5	1	4.0	1	5.9	2	2.7	13	8.3
G12	4	10.0	11	44.0	2	11.8	0	0.0	17	10.8
GNT	1	2.5	0	0.0	0	0.0	1	1.3	2	1.3
Total	40	100.0	25	100.0	17	100.0	75	100.0	157	100.0
P[4]	1	2.8	0	0.0	6	40.0	58	84.1	65	46.8
P[6]	1	2.8	0	0.0	0	0.0	0	0.0	1	0.7
P[8]	25	69.4	16	84.2	9	60.0	5	7.2	55	39.6
P[9]	1	2.8	0	0.0	0	0.0	0	0.0	1	0.7
PNT	8	22.2	3	15.8	0	0.0	6	8.7	17	12.2
Total	36	100.0	19	100.0	15	100.0	69	100.0	139	100.0

PNT – P not typeable.
GNT – G not typeable.

P[9] [11,23,25]. Two additional G12 lineage II strains from 2002 and 2007 sequenced in this work and not previously reported were also included in this analysis.

The VP4 gene was sequenced for a group of samples characterized as single infections by G12P[8] strains to avoid confusing or ambiguous data from mixed infections. There were 6 samples satisfying this condition and all of them were from 2009. Phylogenetic examination indicated that the sequences belonged to the lineage III of the P[8] genotype but clustered in 3 separate groups supported by high bootstrap rates. A first group including strain

Table 3
Distribution and frequency of G and P genotype combinations of group A rotavirus from 2008 to 2011 detected in Buenos Aires.

Strain	2008		2009		2010		2011		Total no.	% Over the 4-year period
	No.	%	No.	%	No.	%	No.	%		
G1P[8]	4	11.1							4	2.9
G1P[6]	1	2.8							1	0.7
G2P[4]	1	2.8			4	26.7	54	78.3	59	42.4
G2P[8]							3	4.3	3	2.2
G2PNT	1	2.8					3	4.3	4	2.9
G3P[4]					1	6.7			1	0.7
G3P[8]	6	16.7	6	31.6	5	33.3	1	1.4	18	12.9
G3PNT	4	11.1	1	5.3			2	2.9	7	5.0
G4P[8]	7	19.4							7	5.0
G9P[8]	5	13.9			1	6.7			6	4.3
G9PNT	2	5.6	1	5.3					3	2.2
G12P[8]			6	31.6	2	13.3			8	5.8
GNTP[4]							1	1.4	1	0.7
GNTP[9]	1	2.8							1	0.7
G1 + 3P[8]					1	6.7			1	0.7
G1 + 12P[8]	1	2.8							1	0.7
G2 + 3P[4]					1	6.7	2	2.9	3	2.2
G2 + 3P[8]							1	1.4	1	0.7
G2 + 12P[8]	1	2.8							1	0.7
G3 + 9PNT							1	1.4	1	0.7
G3 + 12P[8]			4	21.1					4	2.9
G9 + 12P[8]	1	2.8							1	0.7
G9 + 12PNT	1	2.8							1	0.7
G1 + 3 + 12PNT			1	5.3					1	0.7
G2 + 3 + 9P[4]							1	1.4	1	0.7
Total	36	100.0	19	100.0	16	100.0	61	100.0	132	100.0

PNT – P not typeable.
GNT – G not typeable.

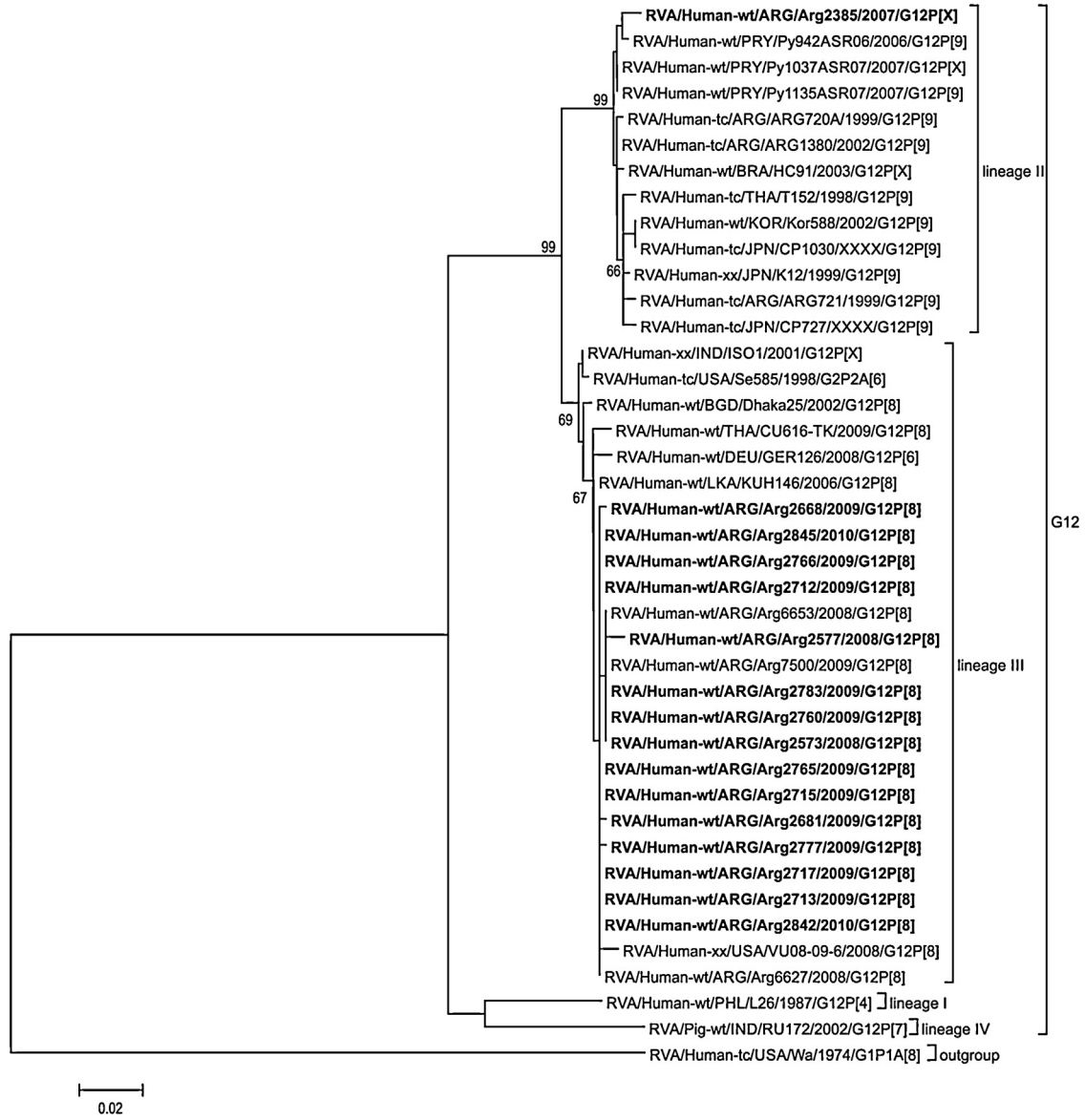

Fig. 1. Phylogenetic analysis of G12 strains. The tree was constructed from nucleotide sequences of VP7 genes of G12 rotavirus strains obtained from the GeneBank database. The lineages are indicated on the right and the bootstrap values are shown at the branch nodes (values <65% are not shown). The Argentine strains analyzed in this study are shown in boldface. Strains are named according to the nomenclature proposed by the Rotavirus Classification Working Group.

Arg2668 (bootstrap value: 99), a second group including strain Arg2765 (bootstrap value: 90) and a third group including strains Arg2712, Arg2713, Arg2717 and Arg2766 (bootstrap value: 99). In a fourth group of the same lineage clustered 2 VP4 sequences from G4P[8] strains and 1 from G1P[8], circulating in our region in 2008 (Fig. 2).

G2 strains. A total of 9 sequences of the VP7 gene were obtained from samples characterized as G2P[4]. Three of them were from 2007 and 6 from 2010 and 2011, representative of both phases of circulating G2 strains mentioned above. Phylogenetic analysis indicated that all the sequences in this collection belonged to the G2 lineage II but segregated in different clusters. Strains from 1997 [11] and 2007 are more related with each other than to the 2010–2011 group containing 6 almost identical VP7 sequences (Fig. 3). Interestingly, strains circulating in Brazil in 2008 and 2009 are closely related to this group, which emerged in Argentina in 2010. In a similar way, some sequences from strains circulating in Brazil in 2005 and 2006 appeared closely related to Argentine strains isolated in 2007 (Fig. 3).

The VP4 gene from 10 samples characterized as G2P[4] were sequenced. From these, 3 were from 2007, 1 from 2008, 3 from 2010 and 3 from 2011. All the sequences belonged to the P[4] lineage V. Phylogenetic analysis confirmed the relationships established on the basis of the VP7 gene of the Argentine strains with each other and with Brazilian strains (Fig. 4). It needs to be highlighted that Figs. 3 and 4 include sequences from the period 2005–2009 from Brazil which are representative of a larger group of strains with almost identical sequences [26]. Additionally, all available sequences in the GenBank were used for phylogenetic analysis but, in the sake of understandability, only representatives of the most closely related sequences are shown in the figures.

5. Discussion

Three genotype constellations have been defined for human RVA: Wa-like, DS-1-like and AU-1-like, being the first two much more common in epidemiologic studies while the last one is rarely found [27]. In general, Wa-like RVA include genotype

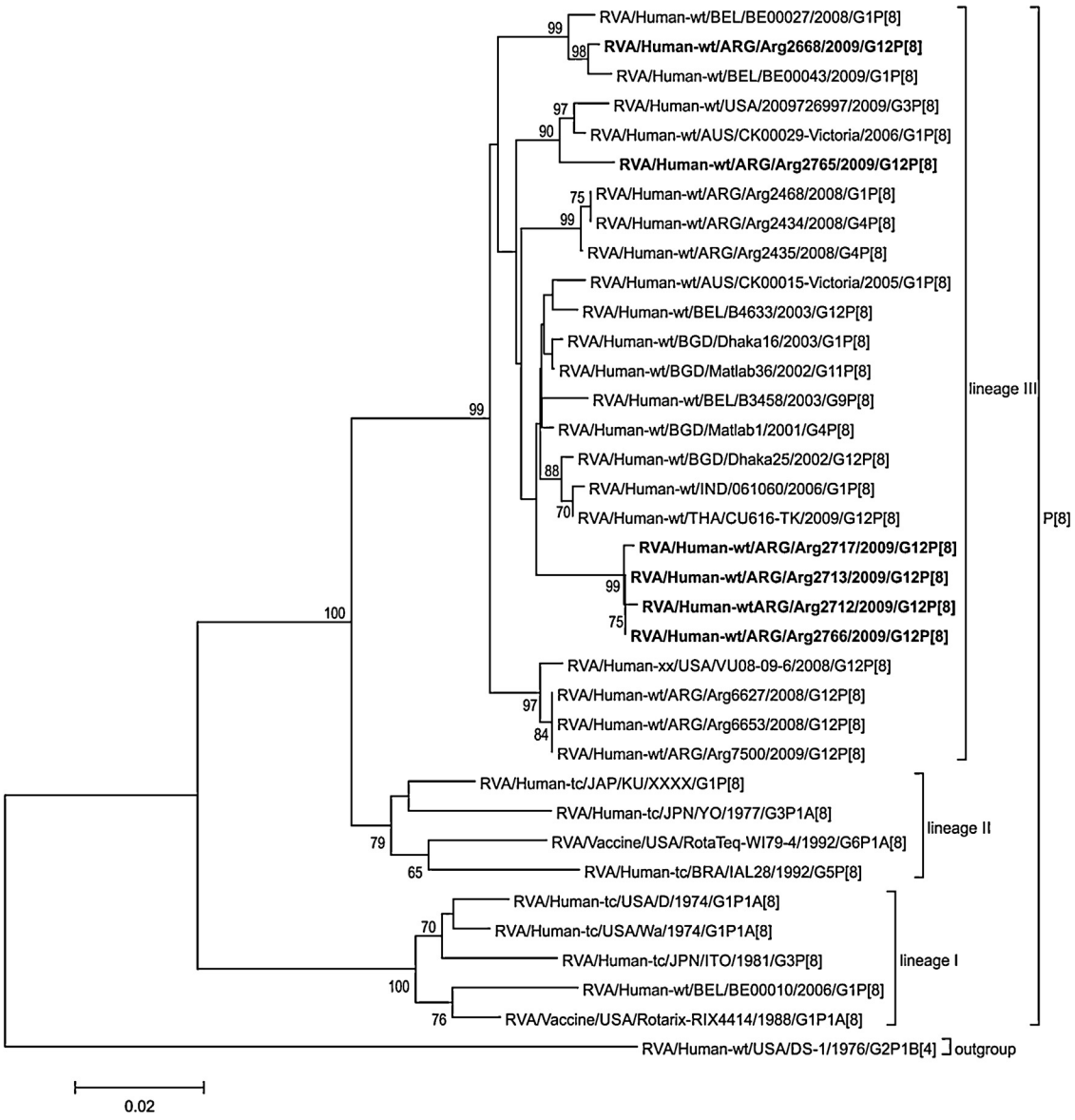

Fig. 2. Phylogenetic analysis of P[8] strains. The tree was constructed from nucleotide sequences of VP4 genes of P[8] rotavirus strains obtained from the GeneBank database. The lineages are indicated on the right and the bootstrap values are shown at the branch nodes (values <65% are not shown). The Argentine strains analyzed in this study are shown in boldface. Strains are named according to the nomenclature proposed by the Rotavirus Classification Working Group.

combinations of G1, G3, G4, or G9 with P[8] (like Rotarix strain) while the DS-1-like RVA typically include the genotype combination of G2 with P[4]. Phylodynamic studies have determined that new strains can spread worldwide in a relatively short time of 10 years [7]. Different G12 strains emerged in several countries during the second half of the 1990s and it was previously demonstrated that the introduction in Argentina of AU-1-like, VP7 lineage II G12P[9] strains, was most probably from Far East countries [23,25]. As shown here, these strains circulated at low rates in Buenos Aires until 2007 and were replaced by lineage III G12P[8] strains in 2008. These last emerging rotavirus strains were also detected at national level by a National Surveillance System study [24] in which the strains were characterized as G12-P[8]-I1-R1-C1-M1-A1-N1-T1-E1-H1, a common human Wa-like genotype constellation. Differently to the AU-1-like G12P[9] strains, these strains were detected at medium to high frequencies in Buenos Aires during three consecutive seasons, suggesting that the G12P[8] strains that emerged in 2008 in Argentina might represent a more cosmopolitan biologically fit G12 strain, well adapted to spread among humans [7,23–25,28]. Thereby, close surveillance should be maintained on this putative globally emergent genotype.

High levels of efficacy and effectiveness of the monovalent vaccine against G2P[4] strains were demonstrated in several studies but some concerns and controversy still remain [9,29,30]. For example, some results suggest that immunity wanes in the second year of life in vaccinated children. Assuming this, generalization of the monovalent vaccine use could hypothetically select G2P[4] (or different DS-1-like strains), increasing the abundance of these strains in the environment as they are excreted during apparent or subclinical infections. In some studies a cycle of recurrence was described for G2P[4] strains about every 10 years [8–10] and the observed increase in South America during the past years has been attributed to this normal fluctuation [8,11]. We showed here a striking and atypically long prevalence period of G2P[4]

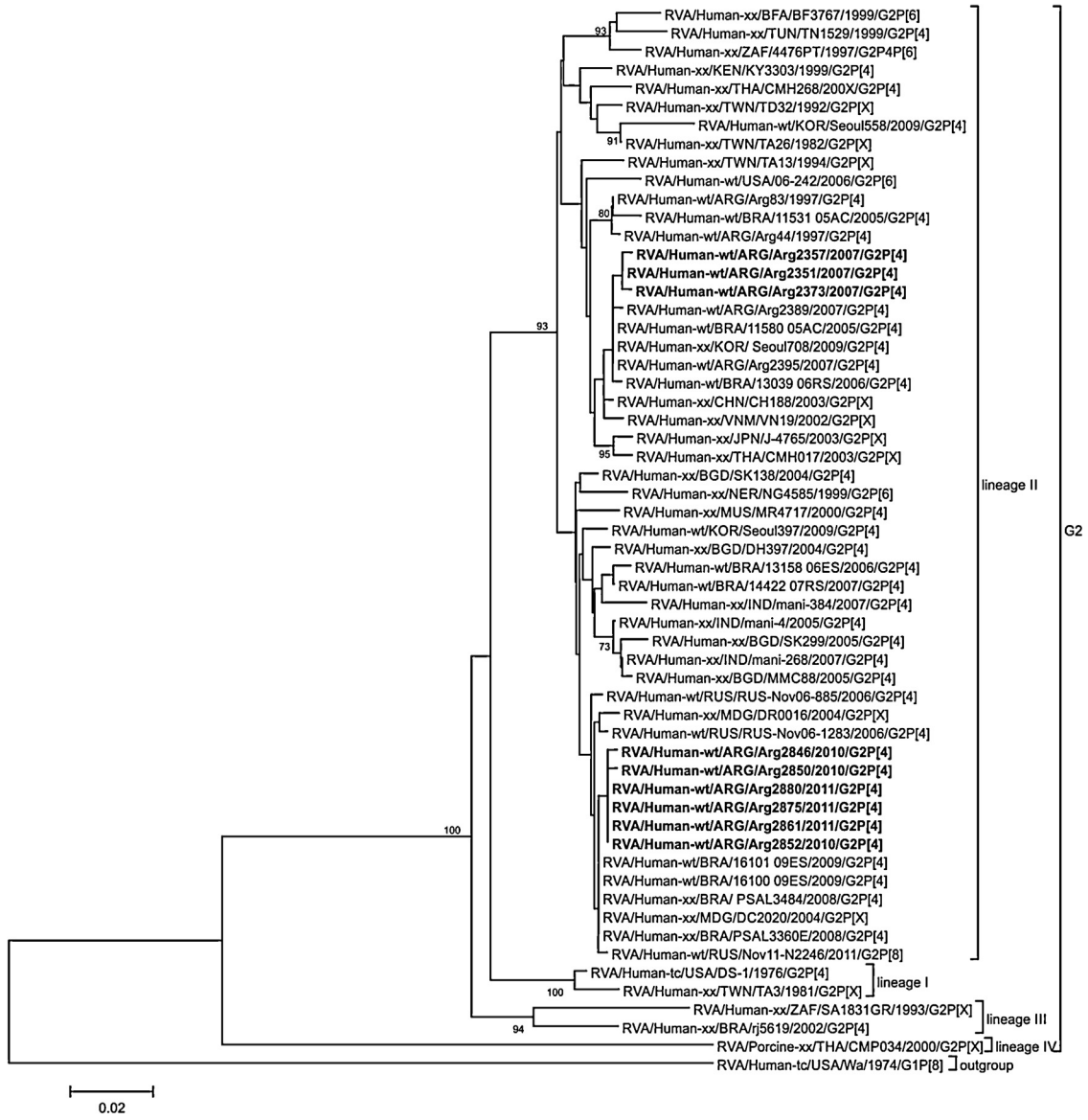

Fig. 3. Phylogenetic analysis of G2 strains. The tree was constructed from nucleotide sequences of VP7 genes of G2 rotavirus strains obtained from the GeneBank database. The lineages are indicated on the right and the bootstrap values are shown at the branch nodes (values <65% are not shown). The Argentine strains analyzed in this study are shown in boldface. Strains are named according to the nomenclature proposed by the Rotavirus Classification Working Group.

strains in Buenos Aires in 2 phases since 2004, with reemerging strains becoming highly prevalent in 2010–2011. After just 1 year of absence in 2009, these strains reached almost 30% of the G types detected in 2010 and around 80% in 2011. The VP4 and VP7 sequences from these strains circulating in 2010 and 2011 were highly similar to a subgroup of strains circulating in Brazil in 2008 and 2009. These results are consistent with the hypothesis that these G2P[4] strains were introduced from Brazil.

The analysis of deduced amino acid sequences of the B, C and F antigenic regions on VP7 showed only one consistent substitution (242 S → N) between strains circulating in both phases in Argentina. This change is also shared by strains circulating in Brazil after 2006 (data not shown). It is difficult to link this kind of changes to strain selection by vaccine induced immunity. Significant changes in neutralization assay titers of sera from vaccinated individuals against both variants of G2 strains or against two reassortants bearing both VP7 variants in a common genome background could help clarify this point. However, since T or B epitopes on several rotavirus proteins not related to classical VP7 neutralization regions, could be implicated in cross-protection elicited by a Wa-like vaccine strain against DS-1 like infecting strains [31,32], it would be very interesting to extend the analysis to a complete genome comparison.

Neighboring countries in the Southern Cone of Latin America are epidemiologically connected and the circulation of strains is shared to some extent as clearly illustrated by the introduction of unusual G12P[9] strains in Argentina and their ulterior passage to subsequent spread to Brazil and Paraguay [7,23,33,34]. Hence, it is possible that the relative abundance of DS-1-like or decline of Wa-like strains in the largest and most populated country of our region could introduce a bias on the usual serotype substitution. Argentina, as a country without universal vaccination, may be acting as an amplifier of this effect visualized through the striking reduction in prevalence of G1P[8] strains and the persistence of G2P[4] in the past few years. To determine if this is a bias of the normal fluctuation of genotype frequencies caused by the

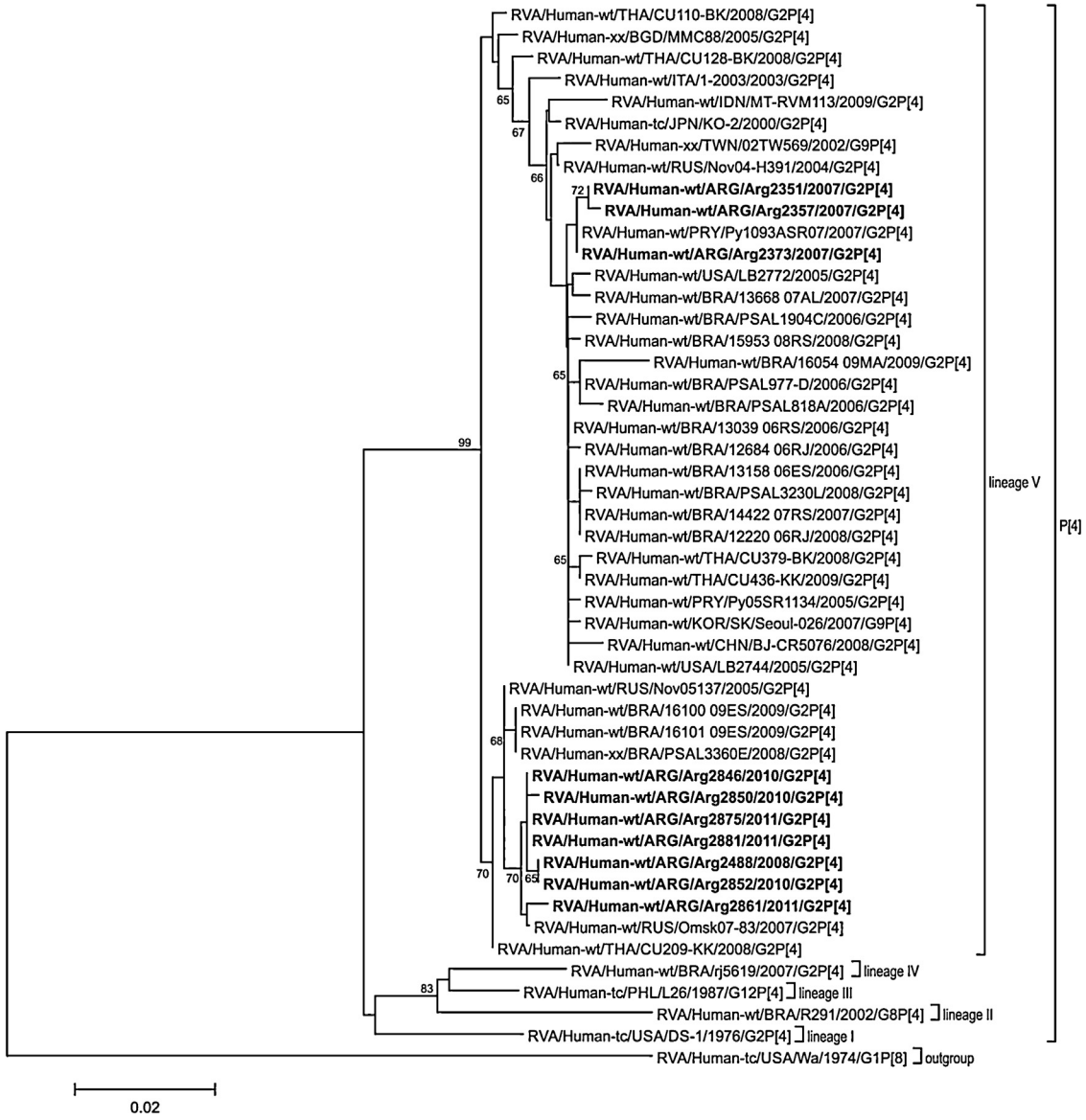

Fig. 4. Phylogenetic analysis of P[4] strains. The tree was constructed from nucleotide sequences of VP4 genes of P[4] rotavirus strains obtained from the GeneBank database. The lineages are indicated on the right and the bootstrap values are shown at the branch nodes (values <65% are not shown). The Argentine strains analyzed in this study are shown in boldface. Strains are named according to the nomenclature proposed by the Rotavirus Classification Working Group.

monovalent vaccine selection, further follow-up of genotypes in Brazil and other regional countries will be required. Furthermore, strain characterization by full genome sequencing of Argentine and Brazilian strains will allow us to confirm the identity and origin of rotaviruses spreading in the region.

Funding

This work was supported by research grants from the Universidad Nacional de Quilmes.

Competing interests

None declared.

Ethical approval

Not required.

Contributors

M.G.M. performed the experiments, analyzed data and drafted the manuscript. L.E.E. and M.H.A. drafted and revised the manuscript. A.M. collected samples and analyzed clinical data. G.G. and A.A.C. designed the study and revised the manuscript. All the authors approved the final manuscript as submitted.

Acknowledgements

Marcelo G. Mandile and Laura E. Esteban are recipients of doctoral and postdoctoral fellowships, respectively, granted by CONICET (Comisión Nacional de Investigaciones Científicas y Técnicas, Argentina). Alejandro A. Castello has received honoraria from MSD Argentina for lectures on rotavirus epidemiology.

References

[1] Tate JE, Burton AH, Boschi-Pinto C, Steele AD, Duque J, Parashar UD. 2008 estimate of worldwide rotavirus-associated mortality in children younger

than 5 years before the introduction of universal rotavirus vaccination programmes: a systematic review and meta-analysis. Lancet Infect Dis 2012;12:136–41.
[2] Parashar U, Steele D, Neuzil K, Quadros C, Tharmaphornpilas P, Serhan F, et al. Progress with rotavirus vaccines: summary of the tenth international rotavirus symposium. Expert Rev Vaccines 2013;12:113–7.
[3] Estes MK, Kapikian AZ, Rotaviruses. In: Fields BN, Knipe DM, Howley PM, editors. Fields virology, vol. 2, 5th ed. Philadelphia, PA: Wolters Kluwer Health/Lippincott Williams & Wilkins; 2007.
[4] Matthijnssens J, Ciarlet M, McDonald SM, Attoui H, Banyai K, Brister JR, et al. Uniformity of rotavirus strain nomenclature proposed by the Rotavirus Classification Working Group (RCWG). Arch Virol 2011;156:1397–413.
[5] Trojnar E, Sachsenroder J, Twardziok S, Reetz J, Otto PH, Johne R. Identification of an avian group A rotavirus containing a novel VP4 gene with a close relationship to those of mammalian rotaviruses. J Gen Virol 2013;94:136–42.
[6] Matthijnssens J, Ciarlet M, Rahman M, Attoui H, Banyai K, Estes MK, et al. Recommendations for the classification of group A rotaviruses using all 11 genomic RNA segments. Arch Virol 2008;153:1621–9.
[7] Matthijnssens J, Heylen E, Zeller M, Rahman M, Lemey P, Van Ranst M. Phylodynamic analyses of rotavirus genotypes G9 and G12 underscore their potential for swift global spread. Mol Biol Evol 2010;27:2431–6.
[8] Leite JP, Carvalho-Costa FA, Linhares AC, Group. A rotavirus genotypes and the ongoing Brazilian experience: a review. Mem Inst Oswaldo Cruz 2008;103:745–53.
[9] Justino MC, Linhares AC, Lanzieri TM, Miranda Y, Mascarenhas JD, Abreu E, et al. Effectiveness of the monovalent G1P[8] human rotavirus vaccine against hospitalization for severe G2P[4] rotavirus gastroenteritis in Belem, Brazil. Pediatr Infect Dis J 2011;30:396–401.
[10] Bishop RF, Unicomb LE, Barnes GL. Epidemiology of rotavirus serotypes in Melbourne, Australia, from 1973 to 1989. J Clin Microbiol 1991;29:862–8.
[11] Esteban LE, Rota RP, Gentsch JR, Jiang B, Esona M, Glass RI, et al. Molecular epidemiology of group A rotavirus in Buenos Aires, Argentina 2004-2007: reemergence of G2P[4] and emergence of G9P[8] strains. J Med Virol 2010;82:1083–93.
[12] CDC. Progress in the introduction of rotavirus vaccine – Latin America and the Caribbean, 2006–2010. MMWR Morb Mortal Wkly Rep 2011;60:1611–4.
[13] Argüelles MH, Villegas GA, Castello A, Abrami A, Ghiringhelli PD, Semorile L, et al. VP7 and VP4 genotyping of human group A rotavirus in Buenos Aires, Argentina. J Clin Microbiol 2000;38:252–9.
[14] Boom R, Sol CJ, Salimans MM, Jansen CL, Wertheim-van Dillen PM, van der Noordaa J. Rapid and simple method for purification of nucleic acids. J Clin Microbiol 1990;28:495–503.
[15] Gomara MI, Cubitt D, Desselberger U, Gray J. Amino acid substitution within the VP7 protein of G2 rotavirus strains associated with failure to serotype. J Clin Microbiol 2001;39:3796–8.
[16] Gouvea V, Glass RI, Woods P, Taniguchi K, Clark HF, Forrester B, et al. Polymerase chain reaction amplification and typing of rotavirus nucleic acid from stool specimens. J Clin Microbiol 1990;28:276–82.
[17] Iturriza-Gomara M, Kang G, Gray J. Rotavirus genotyping: keeping up with an evolving population of human rotaviruses. J Clin Virol 2004;31:259–65.
[18] Banerjee I, Ramani S, Primrose B, Iturriza-Gomara M, Gray JJ, Brown DW, et al. Modification of rotavirus multiplex RT-PCR for the detection of G12 strains based on characterization of emerging G12 rotavirus strains from South India. J Med Virol 2007;79:1413–21.
[19] Simmonds MK, Armah G, Asmah R, Banerjee I, Damanka S, Esona M, et al. New oligonucleotide primers for P-typing of rotavirus strains: strategies for typing previously untypeable strains. J Clin Virol 2008;42:368–73.
[20] Gentsch JR, Glass RI, Woods P, Gouvea V, Gorziglia M, Flores J, et al. Identification of group A rotavirus gene 4 types by polymerase chain reaction. J Clin Microbiol 1992;30:1365–73.
[21] Iturriza-Gomara M, Green J, Brown DW, Desselberger U, Gray JJ. Diversity within the VP4 gene of rotavirus P[8] strains: implications for reverse transcription-PCR genotyping. J Clin Microbiol 2000;38:898–901.
[22] Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S. MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol Biol Evol 2011;28:2731–9.
[23] Castello AA, Arguelles MH, Rota RP, Olthoff A, Jiang B, Glass RI, et al. Molecular epidemiology of group A rotavirus diarrhea among children in Buenos Aires, Argentina, from 1999 to 2003 and emergence of the infrequent genotype G12. J Clin Microbiol 2006;44:2046–50.
[24] Stupka JA, Degiuseppe JI, Parra GI. Increased frequency of rotavirus G3P[8] and G12P[8] in Argentina during 2008–2009: whole-genome characterization of emerging G12P[8] strains. J Clin Virol 2012;54:162–7.
[25] Castello AA, Nakagomi T, Nakagomi O, Jiang B, Kang JO, Glass RI, et al. Characterization of genotype P[9]G12 rotavirus strains from Argentina: high similarity with Japanese and Korean G12 strains. J Med Virol 2009;81:371–81.
[26] Gómez MM, de Mendonca MC, Volotao Ede M, Tort LF, da Silva MF, Cristina J, et al. Rotavirus A genotype P[4]G2: genetic diversity and reassortment events among strains circulating in Brazil between 2005 and 2009. J Med Virol 2011;83:1093–106.
[27] McDonald SM, Matthijnssens J, McAllen JK, Hine E, Overton L, Wang S, et al. Evolutionary dynamics of human rotaviruses: balancing reassortment with preferred genome constellations. PLoS Pathog 2009;5:e1000634.
[28] Rahman M, Matthijnssens J, Yang X, Delbeke T, Arijs I, Taniguchi K, et al. Evolutionary history and global spread of the emerging g12 human rotaviruses. J Virol 2007;81:2382–90.
[29] Correia JB, Patel MM, Nakagomi O, Montenegro FM, Germano EM, Correia NB, et al. Effectiveness of monovalent rotavirus vaccine (Rotarix) against severe diarrhea caused by serotypically unrelated G2P[4] strains in Brazil. J Infect Dis 2010;201:363–9.
[30] Snelling TL, Andrews RM, Kirkwood CD, Culvenor S, Carapetis JR. Case–control evaluation of the effectiveness of the G1P[8] human rotavirus vaccine during an outbreak of rotavirus G2P[4] infection in central Australia. Clin Infect Dis 2011;52:191–9.
[31] Vizzi E, Calvino E, Gonzalez R, Perez-Schael I, Ciarlet M, Kang G, et al. Evaluation of serum antibody responses against the rotavirus nonstructural protein NSP4 in children after rhesus rotavirus tetravalent vaccination or natural infection. Clin Diagn Lab Immunol 2005;12:1157–63.
[32] Patel M, Glass RI, Jiang B, Santosham M, Lopman B, Parashar U. A systematic review of anti-rotavirus serum IgA antibody titer as a potential correlate of rotavirus vaccine efficacy. J Infect Dis 2013.
[33] Pietruchinski E, Benati F, Lauretti F, Kisielius J, Ueda M, Volotao EM, et al. Rotavirus diarrhea in children and adults in a southern city of Brazil in 2003: distribution of G/P types and finding of a rare G12 strain. J Med Virol 2006;78:1241–9.
[34] Martinez M, Amarilla AA, Galeano ME, Aquino VH, Farina N, Russomando G, et al. Predominance of rotavirus G2P[4] and emergence of G12P[9] strains in Asuncion, Paraguay, 2006–2007. Arch Virol 2010;155:525–33.