



NEW CIRCULATING STRAINS OF ROTAVIRUS A

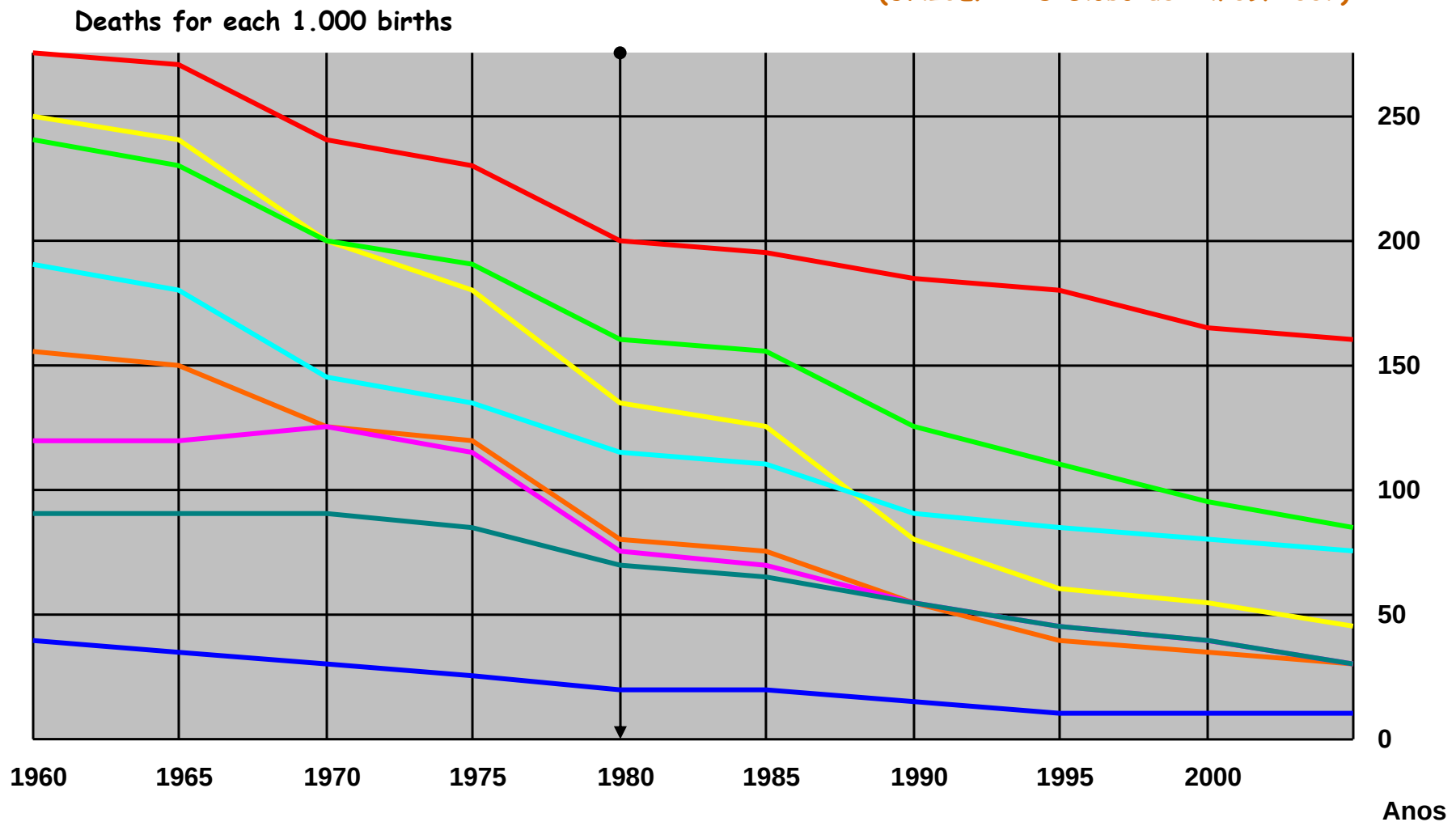
JOSÉ PAULO GAGLIARDI LEITE

LABORATORY OF COMPARATIVE AND ENVIRONMENTAL VIROLOGY

OSWALDO CRUZ INSTITUTE - OSWALDO CRUZ FOUNDATION

Children Mortality by World Region

(UNICEF - "O Globo de 14/09/2007)



— Sub-Saharan Africa

— Middle Orient and North of Africa

— South of Asia

— World

— Latin America and Caribbean

— East of Asia and Pacific Area

— Central and East Europe

— Developed countries



Environmental, Cultural and Health Sanitation Conditions



http://3.bp.blogspot.com/_taeb77q76aI/SQ91IM9FMel/Aia.jpg



<http://g1.globo.com/Noticias/Mundo/foto/0,,17724557-FMM,00.jpg>

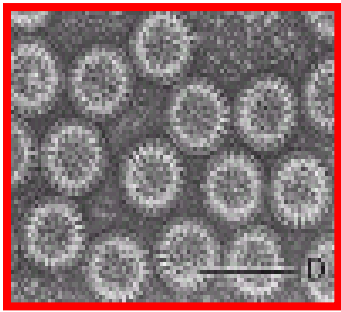


imageeu.homeaway.pt/.../India-Goa_65910.png

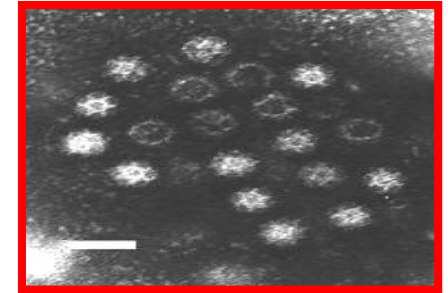


laboratoriodegeografia.blogspot.com





Rotavirus

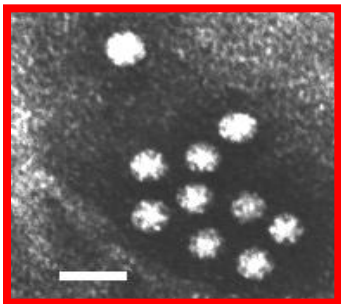


Human
Calicivirus

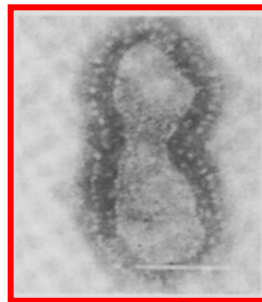
Fastidious!
Experimental model?
Genetic diversity!
Antigenic diversity!
Diversity of hosts!

Interspecies infections!

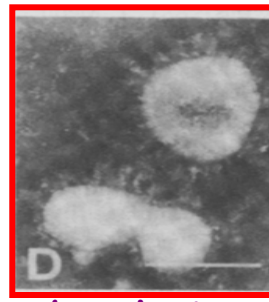
Associated food and water borne
gastroenteritis!



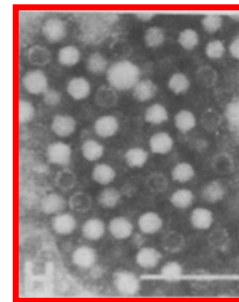
Astrovirus



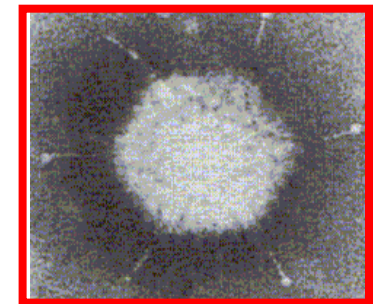
coronavirus



bredavirus



bocavirus



Enteric
adenovirus

Gut metagenomic analysis: new enteric viruses !!



Genetic
Diversity

Antigenic
Diversity

Species
Diversity

Democratic
Viruses



ROTAVIRUS CLASSIFICATION

Family: *Reoviridae*

Genus: *Rotavirus*

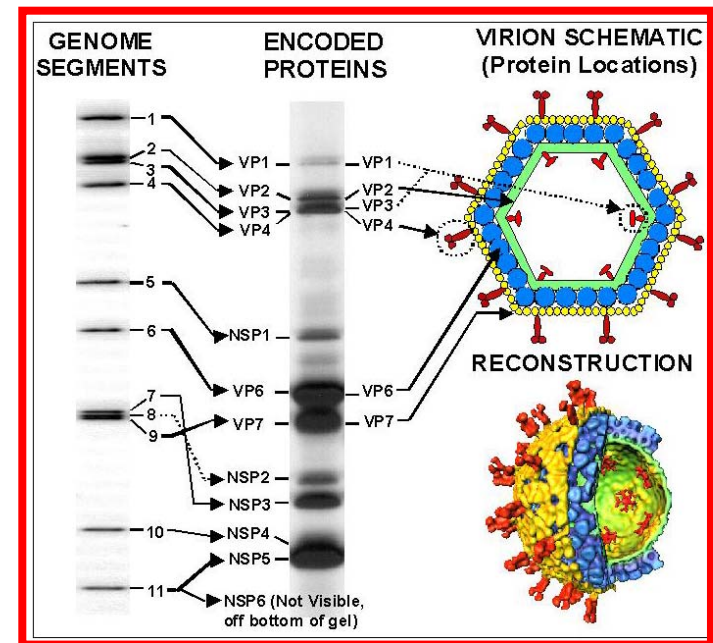
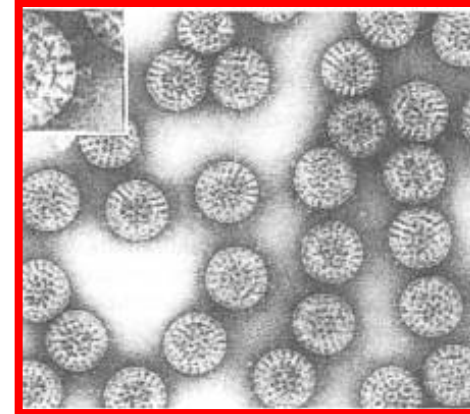
Groups: **A** - G

Virus Particle: 100 nm, non-enveloped,
icosahedral with triple
layer capsid protein.

Genome: dsRNA with 11 segments

Proteins: 6 structural (VP)
6 non-structural (NSP)

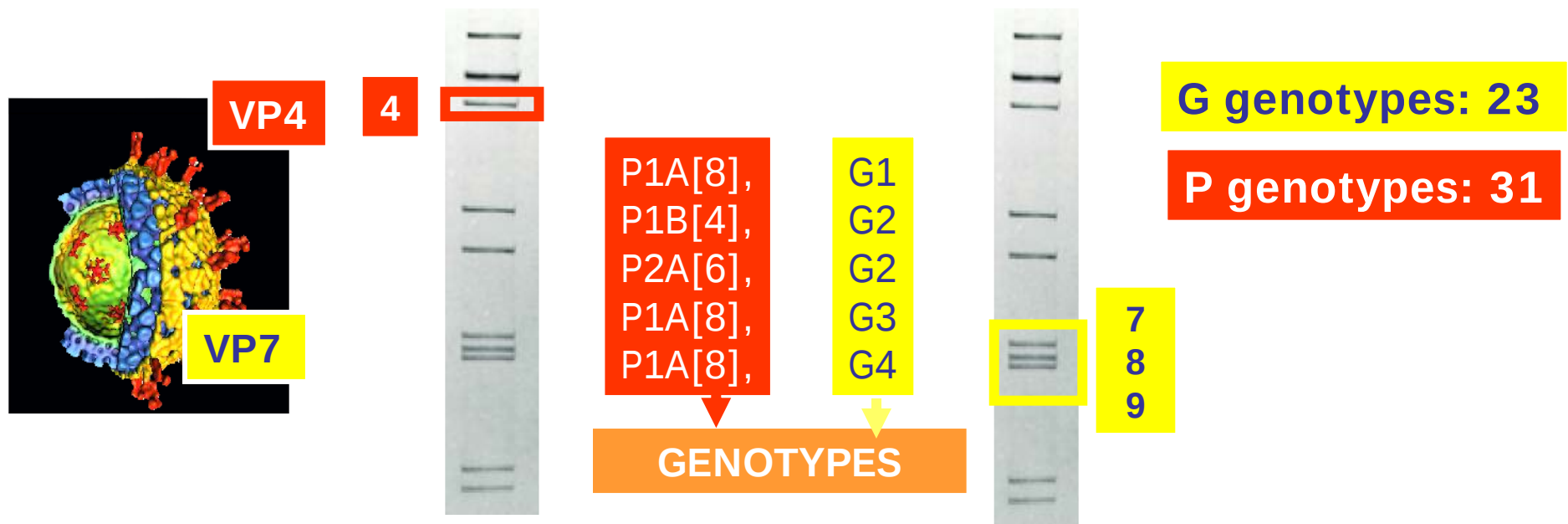
NSP4: enterotoxine
vomiting (Svensson 2009)



Estes, 2001



A BINARY CLASSIFICATION SYSTEM HAS BEEN ESTABLISHED FOR GROUP A ROTAVIRUSES : P and G GENOTYPES



Usual human strains: P[8]G1, P[8]G3, P[8]G4, P[8]G9 (recently), and P[4]G2

Unusual human strains: G5, G8, G10, G12, P6, P9, P10

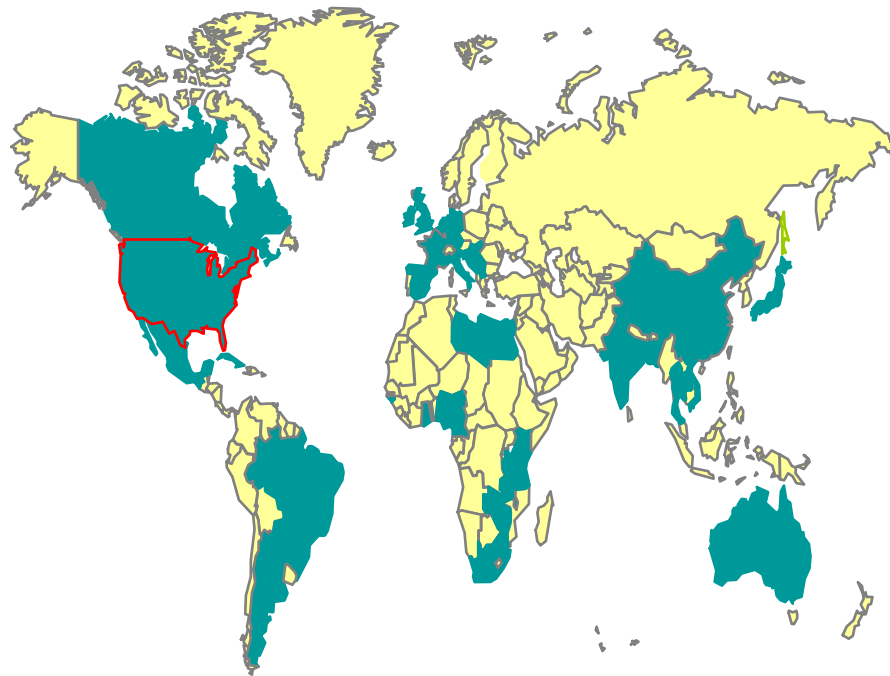


Khamrin et al., 2007; Martella et al., 2007; Matthijnssens et al., 2008; Albe et al., 2009



LESSONS OF INTENSIFIED STRAIN SURVEILLANCE AND MOLECULAR STUDIES

- Large genotype diversity
- Mixed Infections
- Emerging genotypes : globally [G9] or regionally common [G5] in Brazil



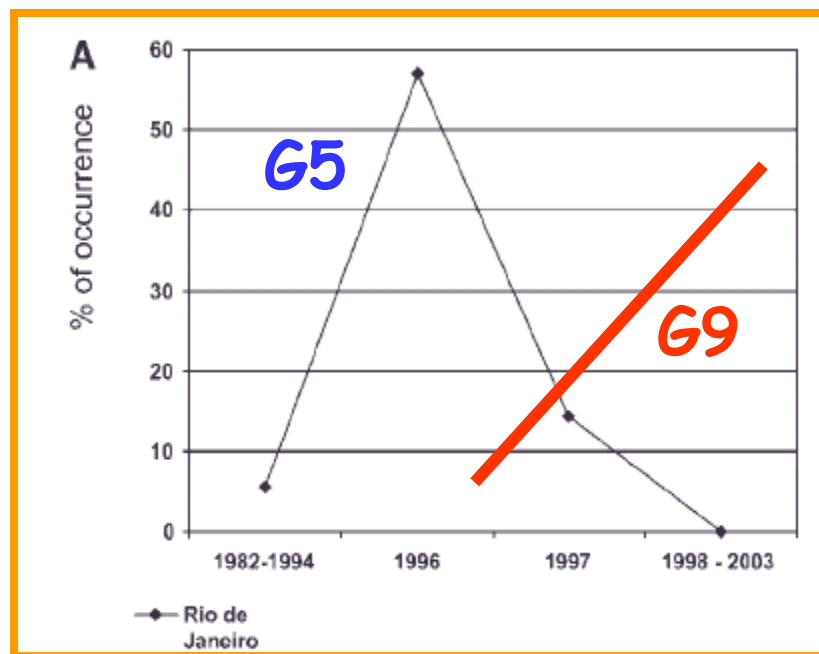
Gentsch et al, 2005





Group A rotavirus genotypes and the ongoing Brazilian experience - A Review

José Paulo Gagliardi Leite^{*,} Filipe Anibal Carvalho-Costa¹, Alexandre C Linhares²



Santos & Hoshino, 2005

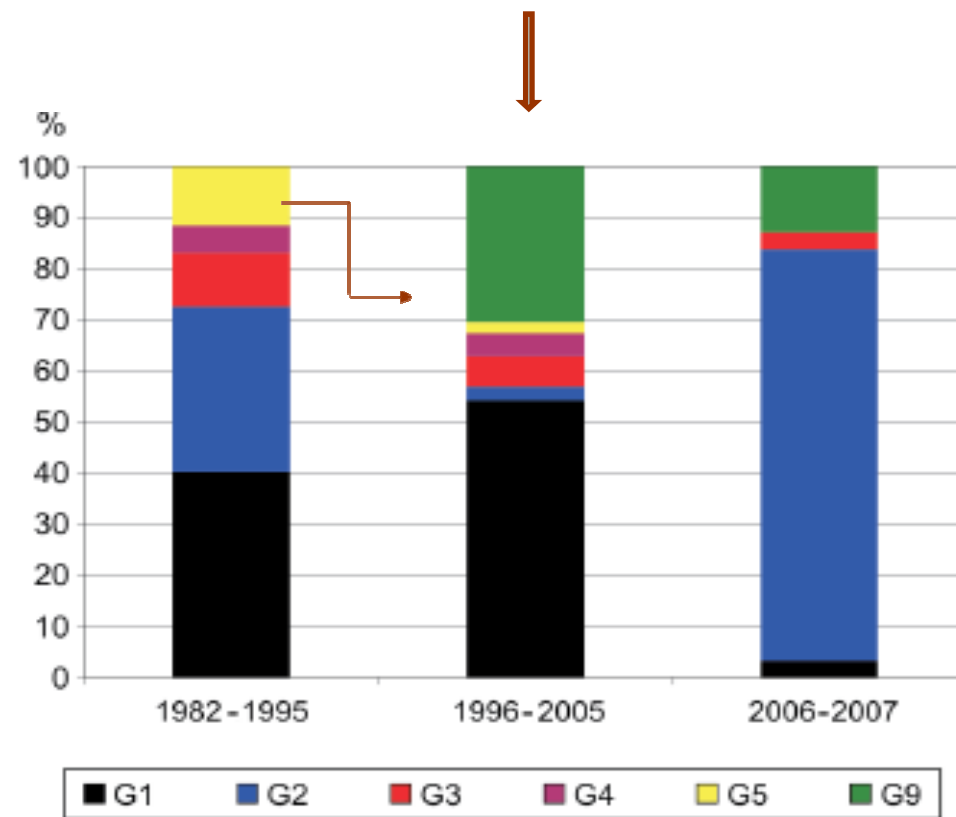


Fig. 3: rates of identification of group A rotaviruses G-genotypes in distinct periods in Brazil.

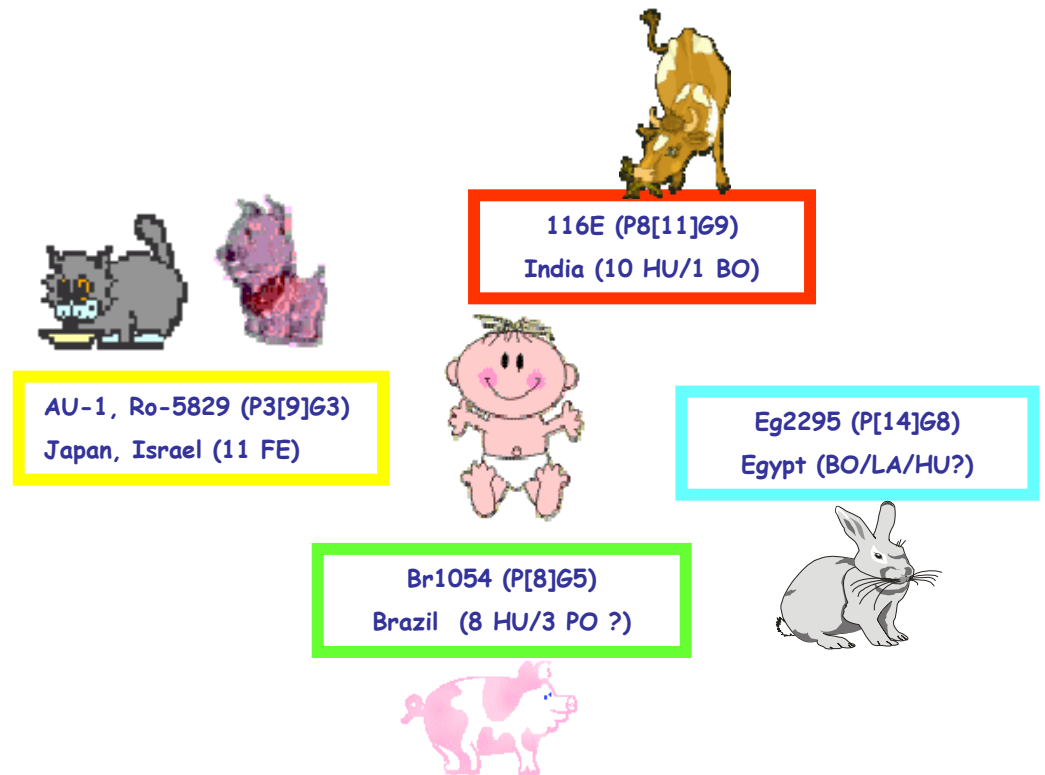
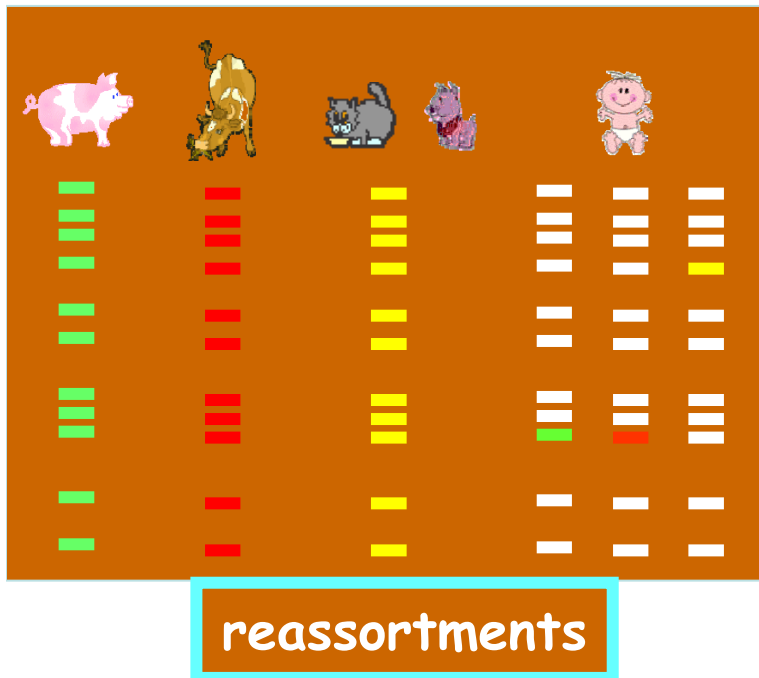
G5

- Camarões (Esona *et al.*, 2004);
- Bangladesh (Hashizume *et al.*, 2007);
- China (Duan *et al.*, 2007);
- Brasil (Carvalho-Costa *et al.*, 2007);
- Vietnã (Ahmed *et al.*, 2007).



LESSONS OF INTENSIFIED STRAIN SURVEILLANCE AND MOLECULAR STUDIES

- **Reassortment of RV-A strains**
 - P and G antigen combinations
 - Intragenogroup reassortment of all genes
- **Role of animal RV-A in diversity**
 - Human-animal RV-A reassortants



Rotavirus Strain Surveillance in Latin America

A Review of the Last Nine Years

Alejandro A. Castello, PhD,† Melissa L. Arvay, MPH,* Roger I. Glass, MD, PhD,* and Jon Gentsch, PhD**

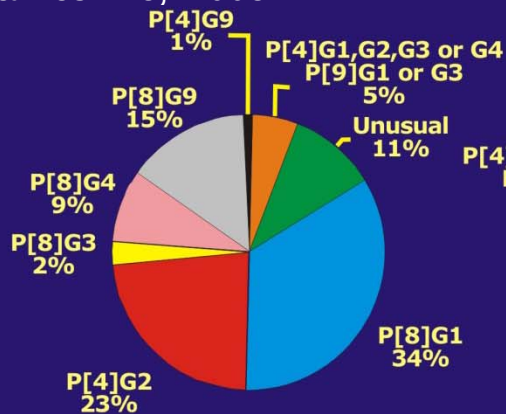
TABLE 2. Findings from G/P Typing Studies in Latin America (Papers Published After 1995)

Finding	Observation
1. Uncommon strains are rare	>70% of strains were common types, ie P[8], P[4] and P[6] with G1, G2, G3 and G4 G5 strains are found throughout Brazil (rates, 4–25%) G9 strains ranged from <1% to 18% of strains in Mexico, Brazil, Argentina and Paraguay G5 (pig, horse) G8 (cow, pig, horse) G10 (cow, pig, horse, lamb) P[1] (cow) P[9] (cat, dog) Human G3 and G4 strains in Mexico had VP7 sequence more similar to animal strains (canine G3, porcine G4) than to human strains G12 appearance in Argentina
2. Some uncommon strains are locally dominant	
3. G9 strains are widely distributed	
4. Many strains are reassortants of animal and human strains	
5. Some common serotypes may have animal origin	
6. Emergence of new, rare human serotypes	

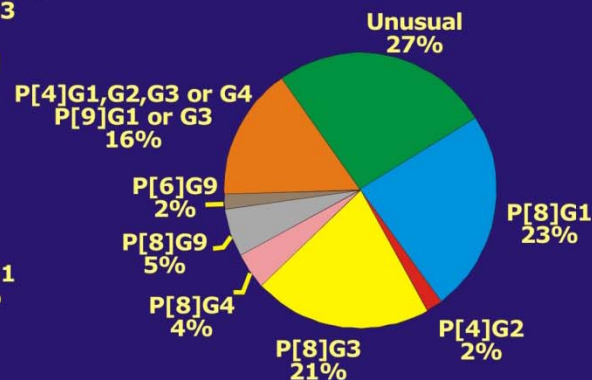


CONTINENTAL / SUBCONTINENTAL DISTRIBUTION OF HUMAN GROUP A ROTAVIRUS P-G COMBINATIONS

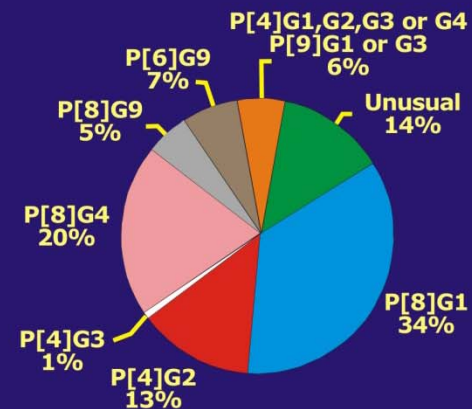
Santos & Hoshino, 2005



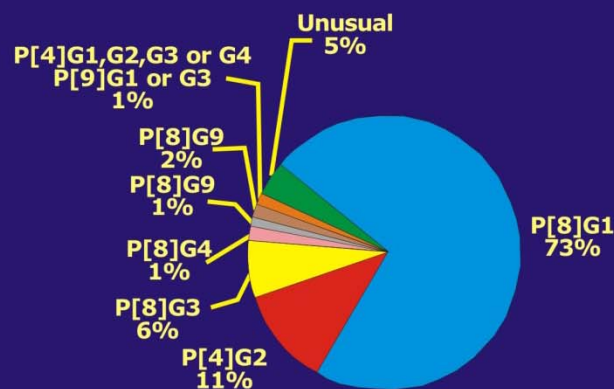
South America
(n=1237)



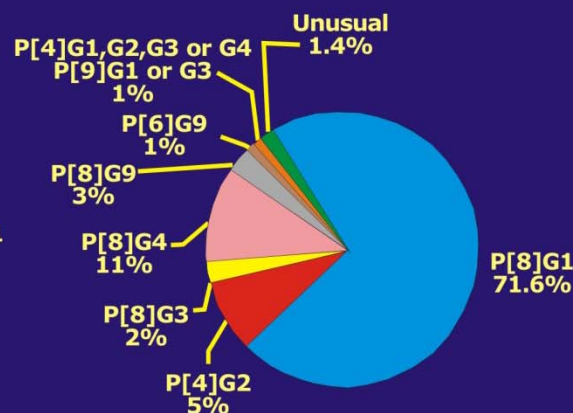
Africa
(n=1077)



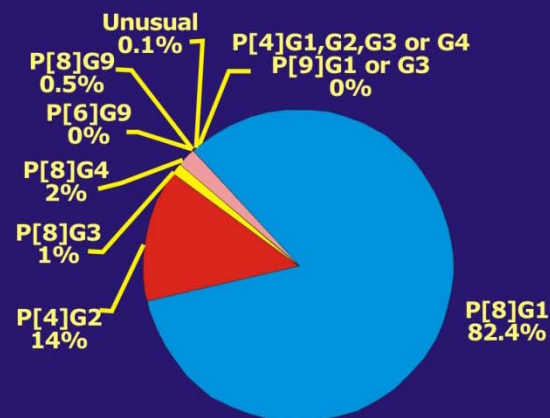
Asia
(n=1567)



North America
(n=1753)



Europe
(n=7024)



Australia/Oceania
(n= 3816)



Group A rotavirus genotypes and the ongoing Brazilian experience - A Review

José Paulo Gagliardi Leite¹*, Filipe Anibal Carvalho-Costa¹, Alexandre C Linhares²

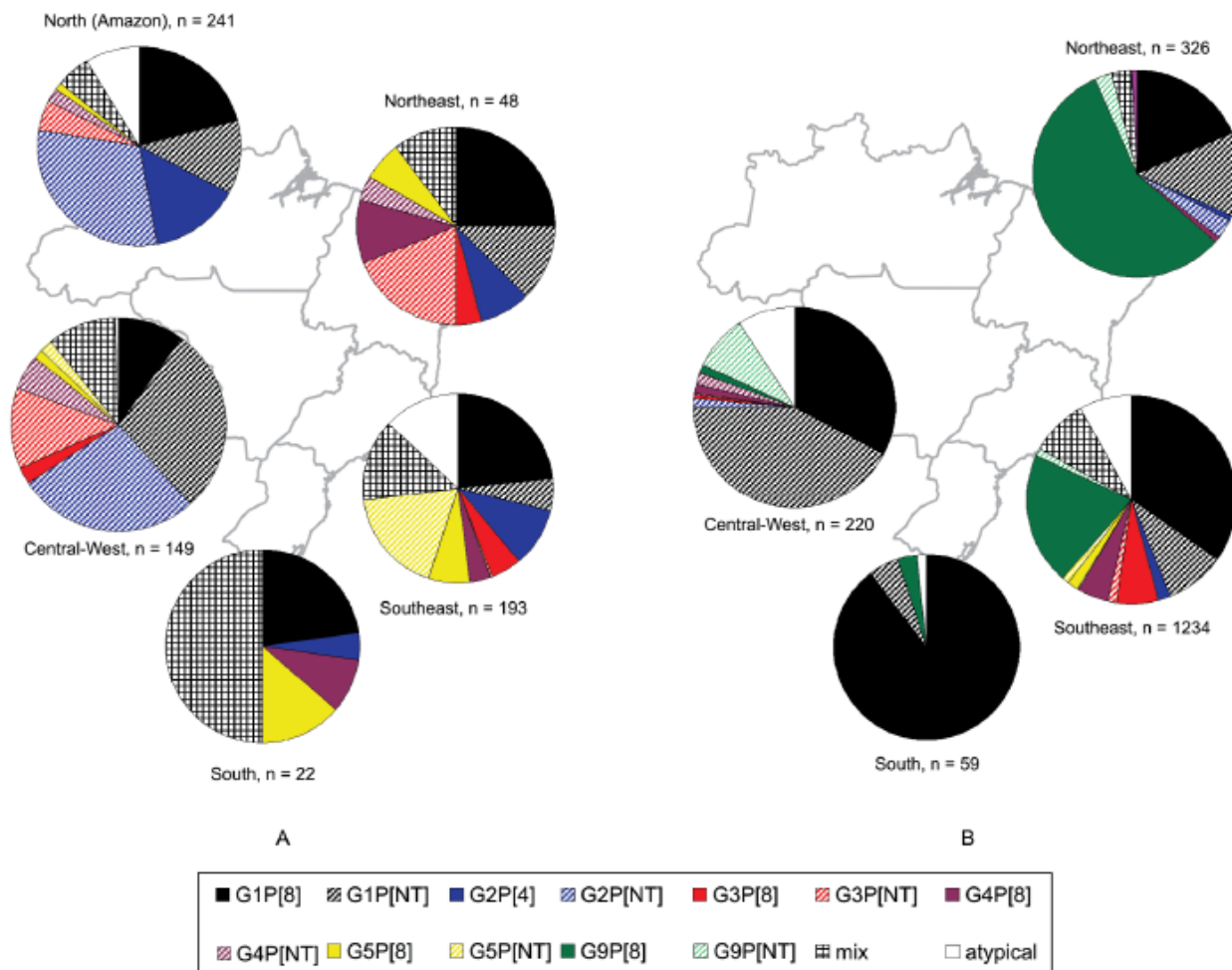


Fig. 2: rotavirus genotypes in Brazil by geographic region. A: 1982-1995 (653 positive stool samples); B: 1996-2005 (1,839 positive stool samples).



The Journal of Infectious Diseases 2009;200:S215–221

Rotavirus Surveillance in Europe, 2005–2008: Web-Enabled Reporting and Real-Time Analysis of Genotyping and Epidemiological Data

M. Iturriza-Gómara,¹ T. Dallman,² K. Bányai,³ B. Böttiger,⁴ J. Buesa,⁵ S. Diedrich,⁷ L. Fiore,⁸ K. Johansen,¹⁰ N. Korsun,¹¹ A. Kroneman,¹² M. Lappalainen,¹³ B. László,³ L. Maunula,¹⁴ J. Matthijnssens,¹⁵ S. Midgley,⁴ Z. Mladenova,¹¹ M. Poljsak-Prijatelj,¹⁶ P. Pothier,⁶ F. M. Ruggeri,¹¹ A. Sanchez-Fauquier,⁶ E. Schreier,⁷ A. Steyer,¹⁵ I. Sitaraviciute,¹⁸ A. N. Tran,¹⁰ V. Usonis,¹⁸ M. Van Ranst,¹⁵ A. de Rougemont,¹⁷ and J. Gray¹

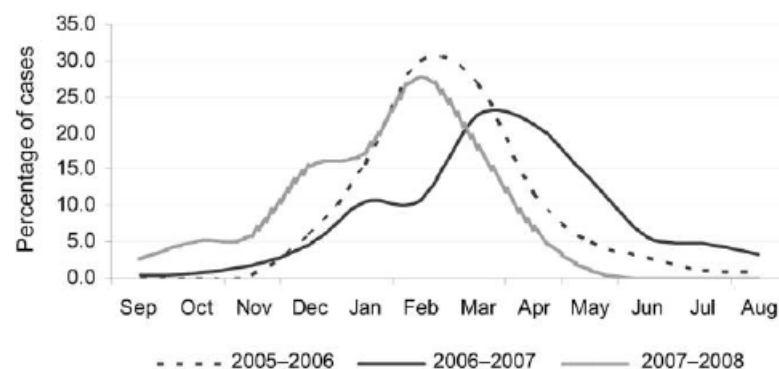


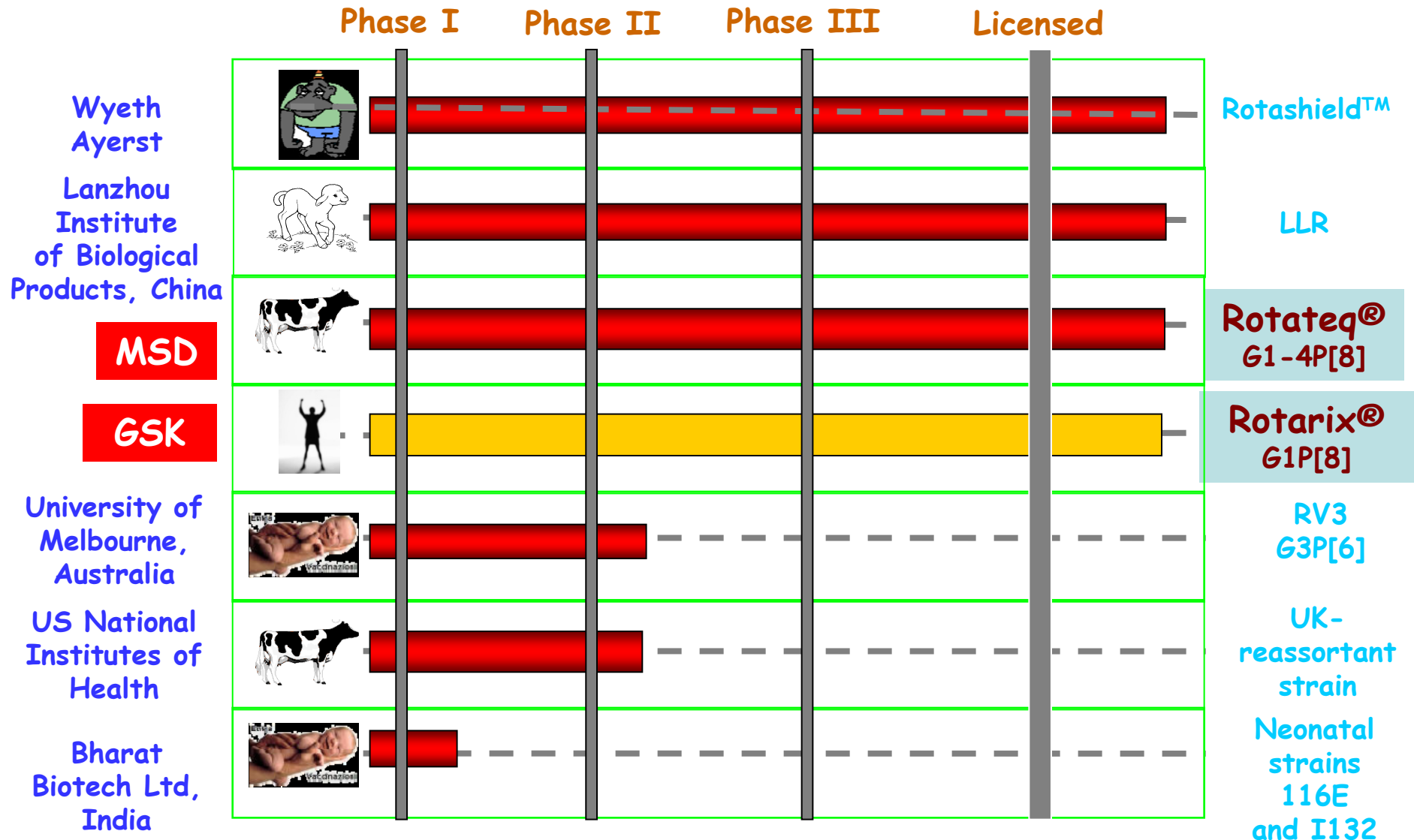
Figure 1. Temporal distribution of rotavirus infections during 3 consecutive rotavirus seasons, 2005–2008. Total numbers of cases analyzed were 2129, 4030, and 2720 for the 2005–2006, 2006–2007, and 2007–2008 seasons, respectively (data for 2007–2008 represent entries uploaded to the database by the end of May 2008).

Table 1. Distribution of Rotavirus Genotypes in the EuroRotaNet Database during 3 Consecutive Rotavirus Seasons from 2005 through 2008 in 19 Countries

Strain group, genotype	No. (%) of strains for season			
	2005–2006 ^a (n = 2129)	2006–2007 ^b (n = 4030)	2007–2008 ^c (n = 2720)	Total (n = 8879)
Common human strains	1939 (91.08)	3552 (88.14)	2514 (92.43)	8005 (90.16)
G1P[8]	922 (43.31)	1686 (41.84)	1690 (62.13)	4298 (48.41)
G2P[4]	51 (2.40)	598 (14.84)	175 (6.43)	824 (9.28)
G3P[8]	81 (3.80)	125 (3.10)	128 (4.71)	334 (3.76)
G4P[8]	90 (4.23)	377 (9.35)	190 (6.99)	657 (7.40)
G9P[8]	795 (37.34)	766 (19.01)	331 (12.17)	1892 (21.31)
Reassortants of common human strains	25 (1.17)	41 (1.02)	16 (0.59)	82 (0.92)
G1P[4]	2 (0.09)	13 (0.32)	8 (0.29)	23 (0.26)
G2P[8]	17 (0.80)	22 (0.55)	6 (0.22)	45 (0.51)
G3P[4]	1 (0.05)	1 (0.02)	0 (0)	2 (0.02)
G4P[4]	3 (0.14)	1 (0.02)	1 (0.04)	5 (0.06)
G9P[4]	2 (0.09)	4 (0.10)	1 (0.04)	7 (0.08)
Potential zoonotic strains	40 (1.88)	56 (1.39)	26 (0.96)	122 (1.37)
G1P[6]	3 (0.14)	0 (0)	0 (0)	3 (0.03)
G2P[6]	1 (0.05)	4 (0.10)	1 (0.04)	6 (0.07)
G3P[6]	1 (0.05)	0 (0)	0 (0)	1 (0.01)
G3P[9]	0 (0)	1 (0.02)	1 (0.04)	2 (0.02)
G4P[6]	6 (0.28)	5 (0.12)	0 (0)	11 (0.12)
G4P[14]	1 (0.05)	0 (0)	0 (0)	1 (0.01)
G6P[8]	0 (0)	0 (0)	2 (0.07)	2 (0.02)
G6P[9]	0 (0)	4 (0.10)	0 (0)	4 (0.04)
G6P[11]	0 (0)	1 (0.02)	0 (0)	1 (0.01)
G8P[4]	2 (0.09)	0 (0)	1 (0.04)	3 (0.03)
G8P[6]	0 (0)	4 (0.10)	1 (0.04)	5 (0.06)
G8P[8]	1 (0.05)	2 (0.05)	1 (0.04)	4 (0.04)
G8P[14]	2 (0.09)	0 (0)	0 (0)	2 (0.02)
G9P[6]	4 (0.19)	5 (0.12)	0 (0)	9 (0.10)
G9P[9]	0 (0)	1 (0.02)	0 (0)	1 (0.01)
G10P[6]	1 (0.05)	1 (0.02)	0 (0)	2 (0.02)
G10P[8]	2 (0.09)	0 (0)	2 (0.07)	4 (0.04)
G10P[14]	0 (0)	5 (0.12)	0 (0)	5 (0.06)
G10P[4]	0 (0)	1 (0.02)	1 (0.04)	2 (0.02)
G12P[6]	0 (0)	4 (0.10)	3 (0.11)	7 (0.08)
G12P[8]	16 (0.75)	18 (0.45)	13 (0.48)	47 (0.53)
Partially typed strains	100 (4.7)	150 (3.72)	104 (3.82)	354 (3.99)
Mixed types	25 (1.17)	231 (5.73)	60 (2.21)	316 (3.56)



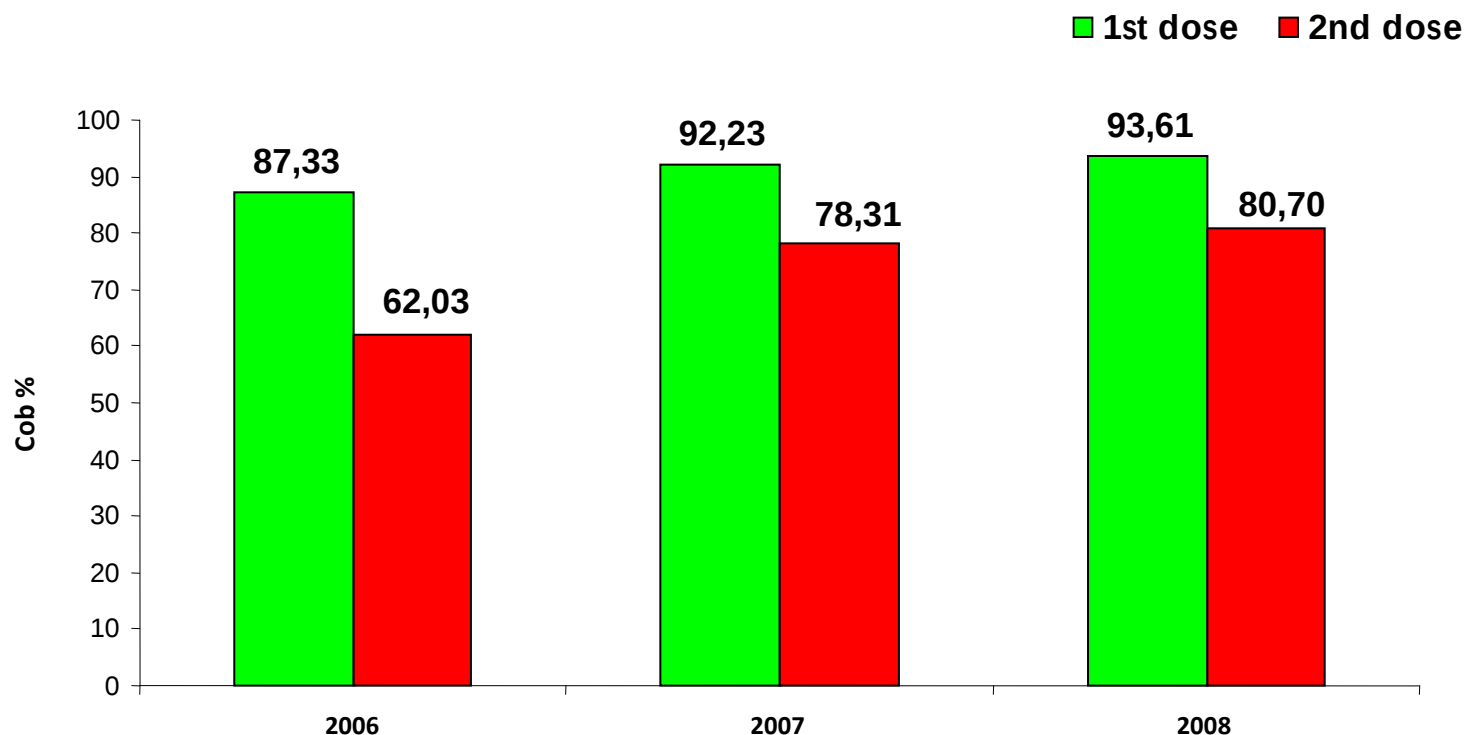
ROTAVIRUS VACCINES



Glass, et al. 2004



Rotarix® vaccination Coverage, March 2006 to December 2008 in Brazil



Número de primeiras e segundas doses registradas da VORH <1 ano, Brasil, 2006 a 2008

Doses/Ano	2006	2007	2008
1ª dose	2.143.078	2.715.991	2.756.724
2ª dose	1.369.812	2.306.181	2.376.559

Fonte: SIAP/CGPNI/SVS/MS



Table. Frequency of rotavirus A infection and distribution of G and P genotypes from February 2005 through December 2007, Rio de Janeiro, Brazil*

Vaccination status and year	No. samples	No. (%) rotavirus positive	No. (%) G1P[8]	No. (%) G2P[4]	No. (%) G3P[8]	No. (%) G9P[8]	No. (%) other genotypes, mixed or not typeable
Ineligible for full vaccination†							
2005	193	73 (38)	10 (14)	1 (1.4)	22 (30)	33 (45)	7 (9.5)
2006	148	34 (23)	1 (2.9)	14 (41)	6 (8)	5 (5)	8 (23)
2007	49	12 (24)	1 (8)	11 (92)	—	—	—
Vaccinated‡							
2006	6	Negative§					
2007	33	4 (12)	—	4 (100)	—	—	—
Not vaccinated							
2006	8	Negative§					
2007	27	10 (37)	—	10 (100)	—	—	—
Total	464	133 (29)	12 (9)	40 (30)	28 (21)	38 (29)	15 (11)

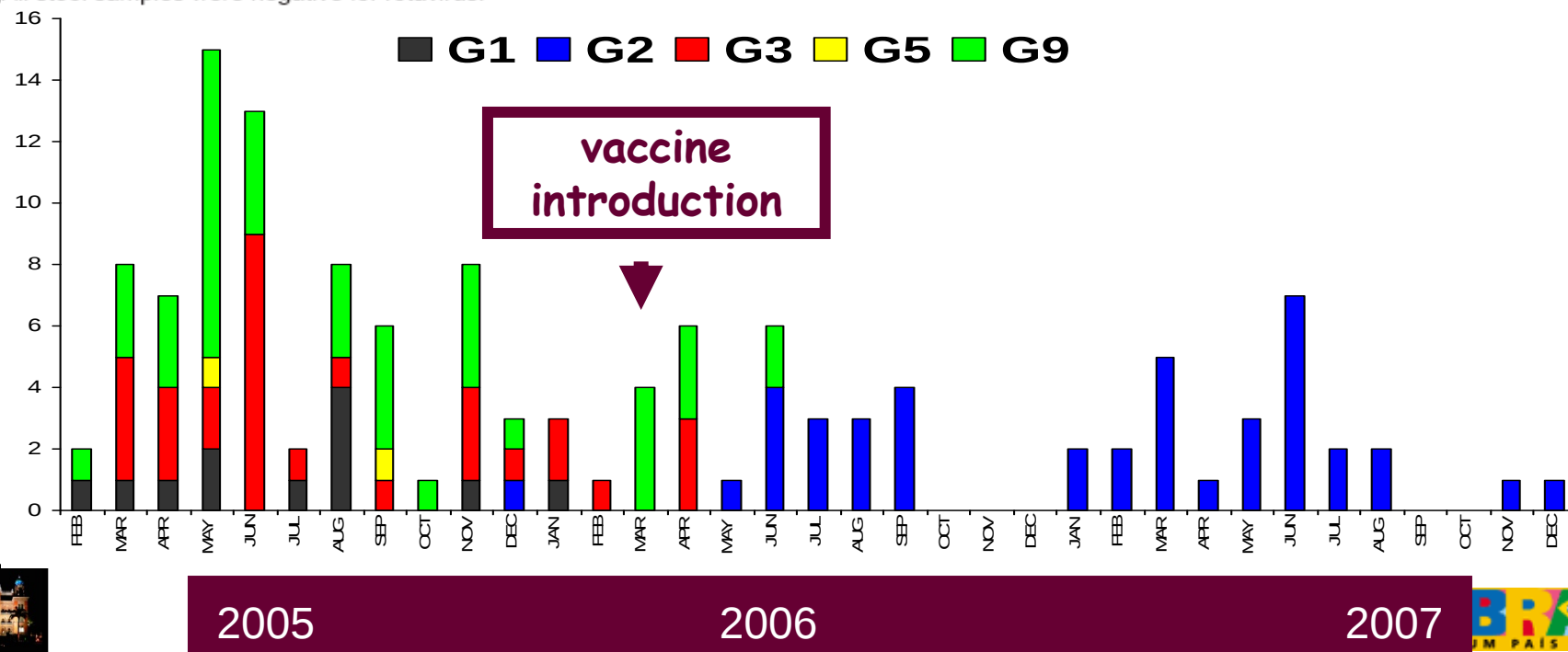
*—, absence of genotypes.

†Born before January 1, 2006, or <4 months of age.

‡Children were considered vaccinated if they had received 2 doses of vaccine.

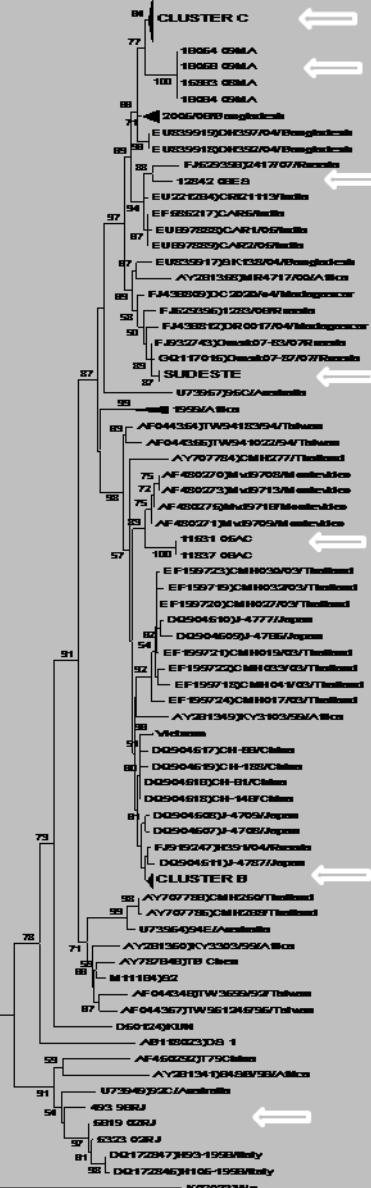
§All stool samples were negative for rotavirus.

Carvalho-Costa et al., 2009



RV-A GENOTYPE G2 CIRCULATING IN BRAZIL: 1996 - 2009

VP7



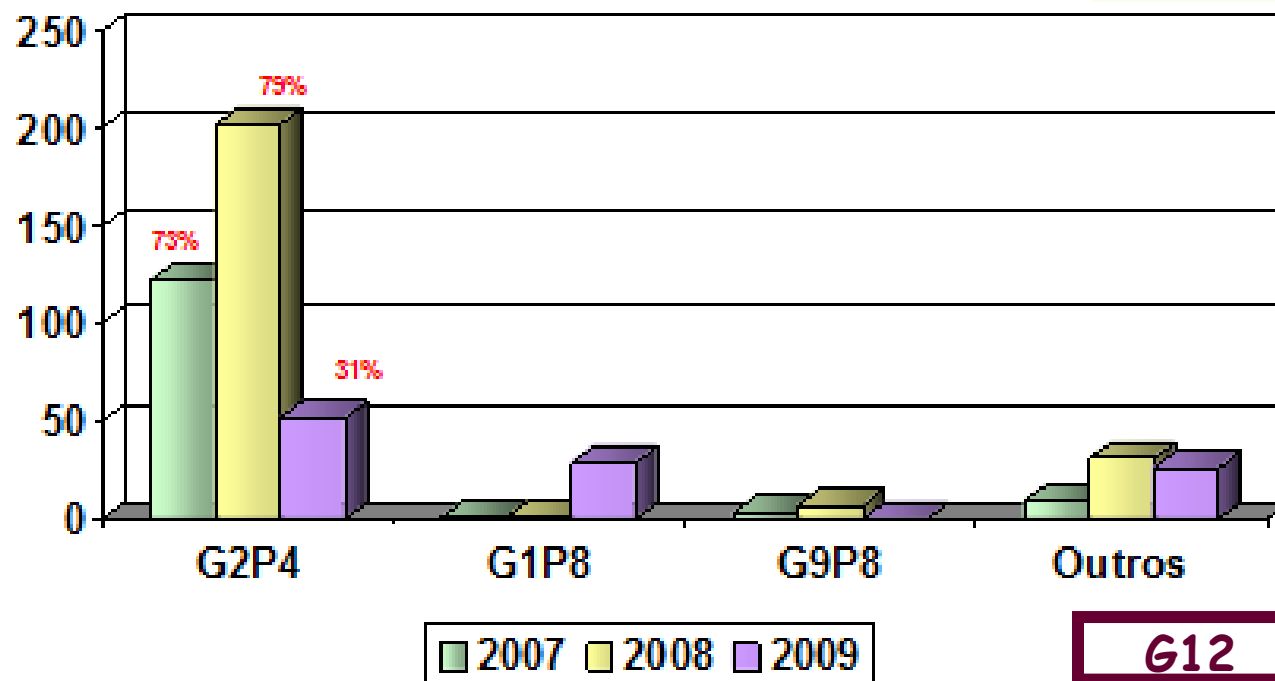
NÚMERO DE AMSOTRA	ANO	ESTADO	VP7	VP4	VP6	NSP4
493_96RJ	1996	RJ			X	
680_96ES	1996	ES	X	X		
685_96ES	1996	ES	X			
957_97RJ	1997	RJ	X		X	
5323_02RJ	2002	RJ			X	X
5619_02RJ	2002	RJ			X	X
11143_05MS	2005	MS			X	
11257_05MS	2005	MS				
11531_05AC	2005	AC		X		
11580_05AC	2005	AC				
11581_05AC	2005	AC	X	X		X
11782_05RJ	2005	RJ	X	X		
11830_06AC	2006	AC	X	X		
11837_06AC	2006	AC		X		
11860_06RJ	2006	RJ				
12220_06RJ	2006	RJ				
12224_06RJ	2006	RJ		X	X	
12287_06BA	2006	BA				
12301_06BA	2006	BA		X		X
12343_06RJ	2006	RJ		X		
12389_06RJ	2006	RJ		X		
12482_06BA	2006	BA				
12522_06BA	2006	BA	X	X		
12540_06RJ	2006	RJ		X	X	
12549_06RJ	2006	RJ				
12585_06ES	2006	ES	X	X		
12589_06RJ	2006	RJ				
12647_06RJ	2006	RJ	X		X	
12684_06RJ	2006	RJ	X		X	X
12774_06RJ	2006	RJ		X		
12775_06RJ	2006	RJ	X	X	X	
12777_06BA	2006	BA	X		X	X
12840_06ES	2006	ES	X			
12842_06ES	2006	ES			X	
12916_06BA	2006	BA	X	X	X	
13039_06RS	2006	RS				X
13079_06RJ	2006	RJ	X		X	X
13081_06RJ	2006	RJ	X		X	X
13151_06RJ	2006	RJ				
13158_06ES	2006	ES				

Gomez et al., 2010





2007/ 2008/ 2009



G12



Seasonal Shifts of Group A Rotavirus Strains as a Possible Mechanism of Persistence in the Human Population

Gabriel I. Parra*

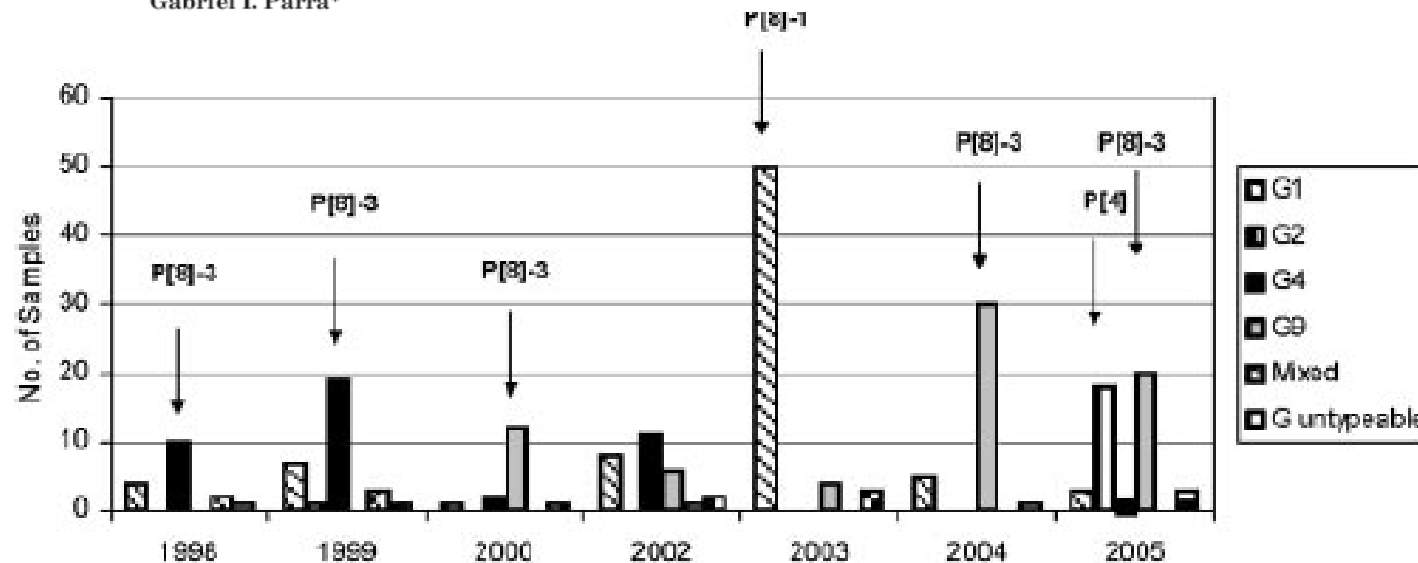


Fig. 1. Yearly variation of rotavirus strains isolated in Asuncion, Paraguay. The number of samples for each G type is shown as bars. The P types associated with the predominant G type are shown above the corresponding bars.

Finally, the immune evasion by rotavirus strains with G and/or P-types different from those present in vaccines is a subject of concern and discussion. Therefore, the continuous monitoring of rotavirus strain diversity during pre- and post-vaccination programs will be helpful for evaluating the efficacy of vaccines available currently.



Molecular Epidemiology of Group A Rotavirus Diarrhea among Children in Buenos Aires, Argentina, from 1999 to 2003 and Emergence of the Infrequent Genotype G12

A. A. Castello,^{1,2*} M. H. Argüelles,² R. P. Rota,² A. Olthoff,³ B. Jiang,¹ R. I. Glass,¹
J. R. Gentsch,¹ and G. Glikmann²

Journal of Medical Virology 81:371–381 (2009)

Characterization of Genotype P[9]G12 Rotavirus Strains From Argentina: High Similarity With Japanese and Korean G12 Strains

Alejandro A. Castello,^{1,2*} Toyoko Nakagomi,³ Osamu Nakagomi,³ Baoming Jiang,² Jung O. Kang,⁴
Roger I. Glass,² Graciela Glikmann,¹ and Jon R. Gentsch²





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



National Rotavirus Surveillance in Argentina: High incidence of G9P[8] strains and detection of G4P[6] strains with porcine characteristics

Juan A. Stupka^a, Paola Carvalho^a, Alberto A. Amarilla^b, Mario Massana^c, Gabriel I. Parra^{b,*}
the Argentinean National Surveillance Network for Diarrheas¹

Table 1

G- and P-type combinations of Argentinean rotavirus strains detected from 2006 to 2007.

G-types	P-types				Total
	P[4]	P[6]	P[8]	P untypeable	
G1	0	0	1 (0.2)	0	1 (0.2)
G2	123 (26.5) ^a	0	0	13 (2.8)	136 (29.3)
G4	0	2 (0.4)	20 (4.3)	3 (0.6)	25 (5.3)
G9	1 (0.2)	0	251 (54.1)	14 (3.0)	266 (57.3)
G3	0	0	2 (0.4)	2 (0.4)	4 (0.8)
G untypeable	2 (0.4)	0	8 (1.7)	22 (4.7)	32 (6.8)
Total	126 (27.1)	2 (0.4)	282 (60.7)	54 (11.6)	464

^a The numbers between parentheses represent percentages.

Finally, these results highlight the great diversity of rotavirus strains circulating in Argentina, a fact that may have a great impact on vaccination programs; therefore, an in-depth surveillance of human and animal rotaviruses will lead to a better understanding of the complex dynamics of rotavirus epidemiology.



Near Future

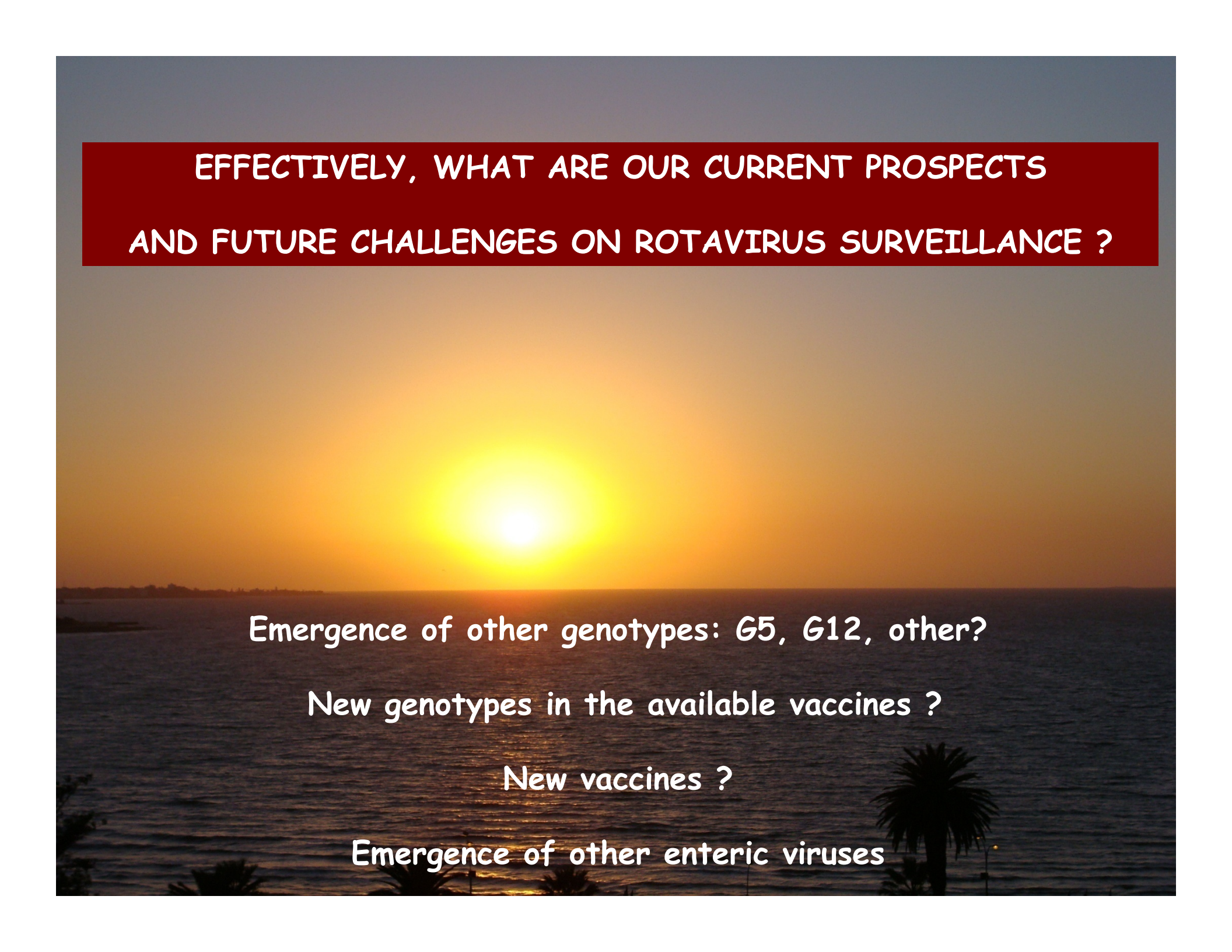


MMMMUUUUUU



Near Future
Business people
X
Gastroenteric Viruses



The background of the slide is a photograph of a sunset over the ocean. The sun is a bright, glowing orb in the center of the horizon, casting a long, shimmering reflection on the water. The sky is a gradient of orange and yellow, fading into a darker blue at the top. In the foreground, the silhouettes of palm trees and other coastal vegetation are visible against the dark water.

EFFECTIVELY, WHAT ARE OUR CURRENT PROSPECTS AND FUTURE CHALLENGES ON ROTAVIRUS SURVEILLANCE ?

Emergence of other genotypes: G5, G12, other?

New genotypes in the available vaccines ?

New vaccines ?

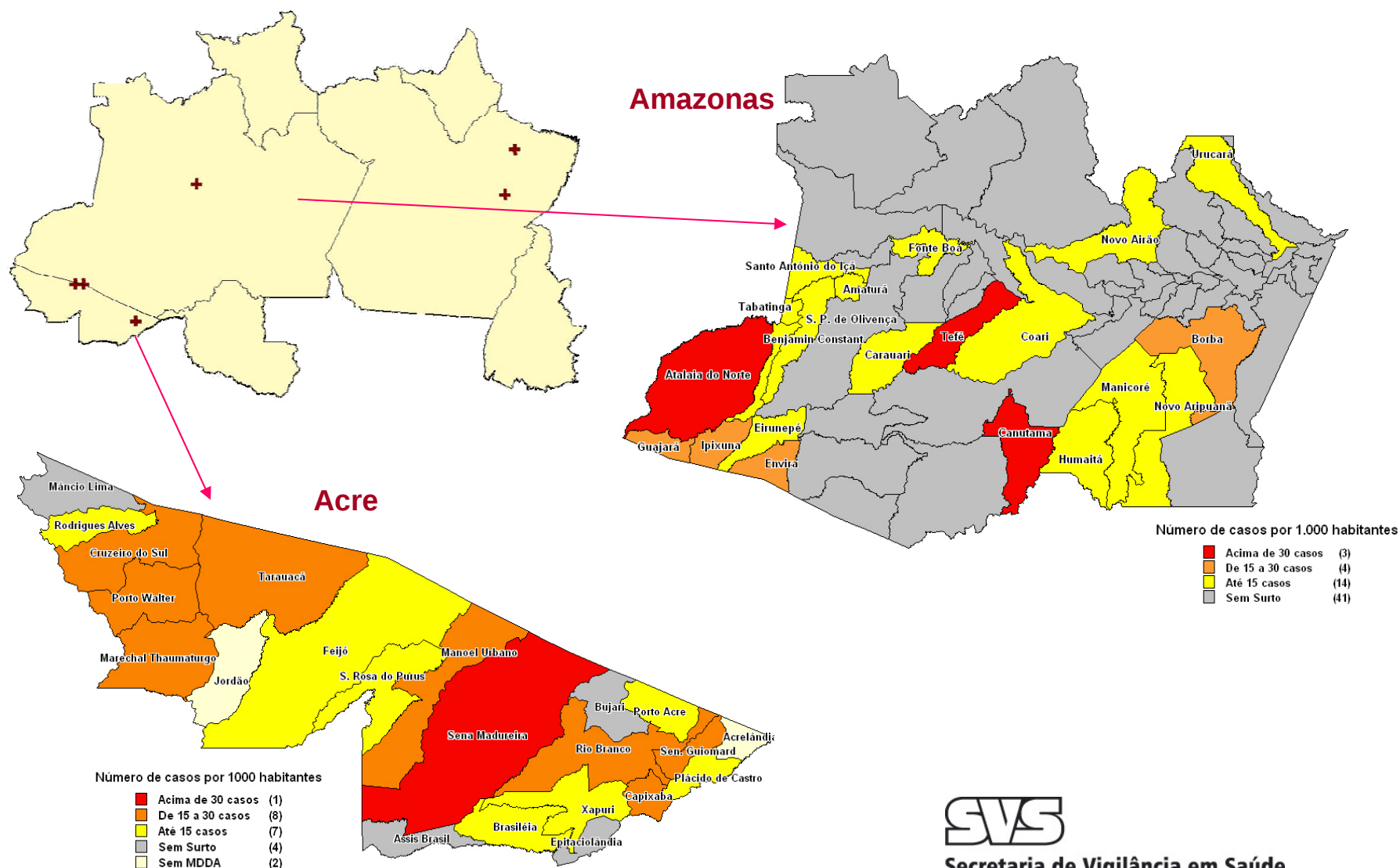
Emergence of other enteric viruses

A background image of a sunset over the ocean. The sun is a bright yellow orb in the center, casting a golden glow across the sky and reflecting on the water. The horizon is visible, and the foreground shows the silhouettes of palm trees and other coastal vegetation.

EFFECTIVELY, WHAT ARE OUR CURRENT PROSPECTS
AND FUTURE CHALLENGES ON ROTAVIRUS SURVEILLANCE ?

Phylogenetic studies: distinct lineages and sub-
lineages for many common genotypes, including
G1, G2, G3, G4, and G9, P4, P6, and P8

Outbreak of G9 in 2005: Acre and Amazonas States, Northern Brazil



Secretaria de Vigilância em Saúde

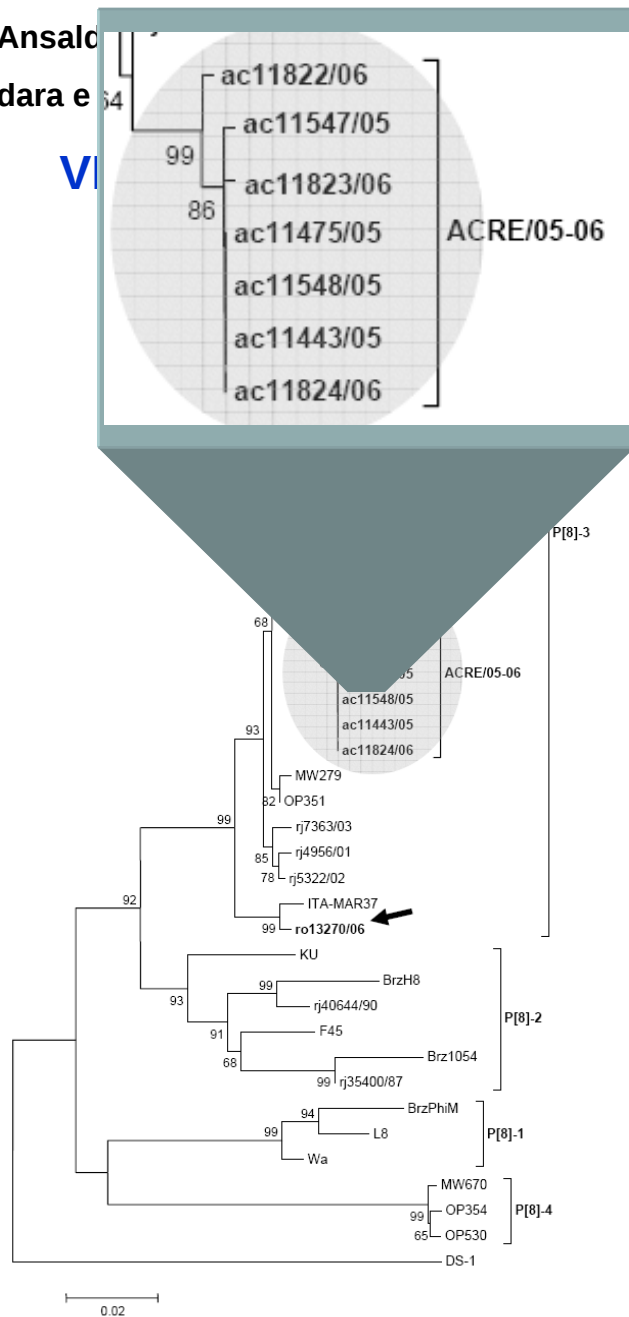


ITA-MAR37: Ansaloni

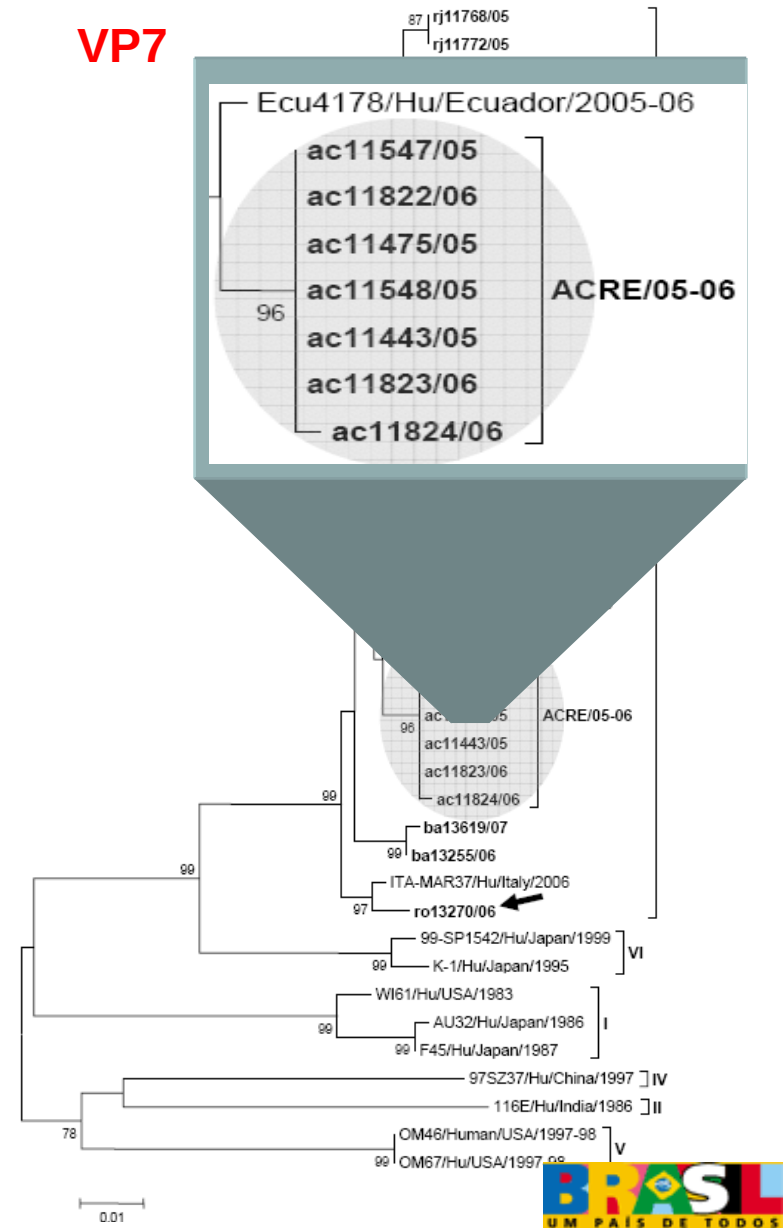
Ecu4178: Endara e

les VP4 e VP7

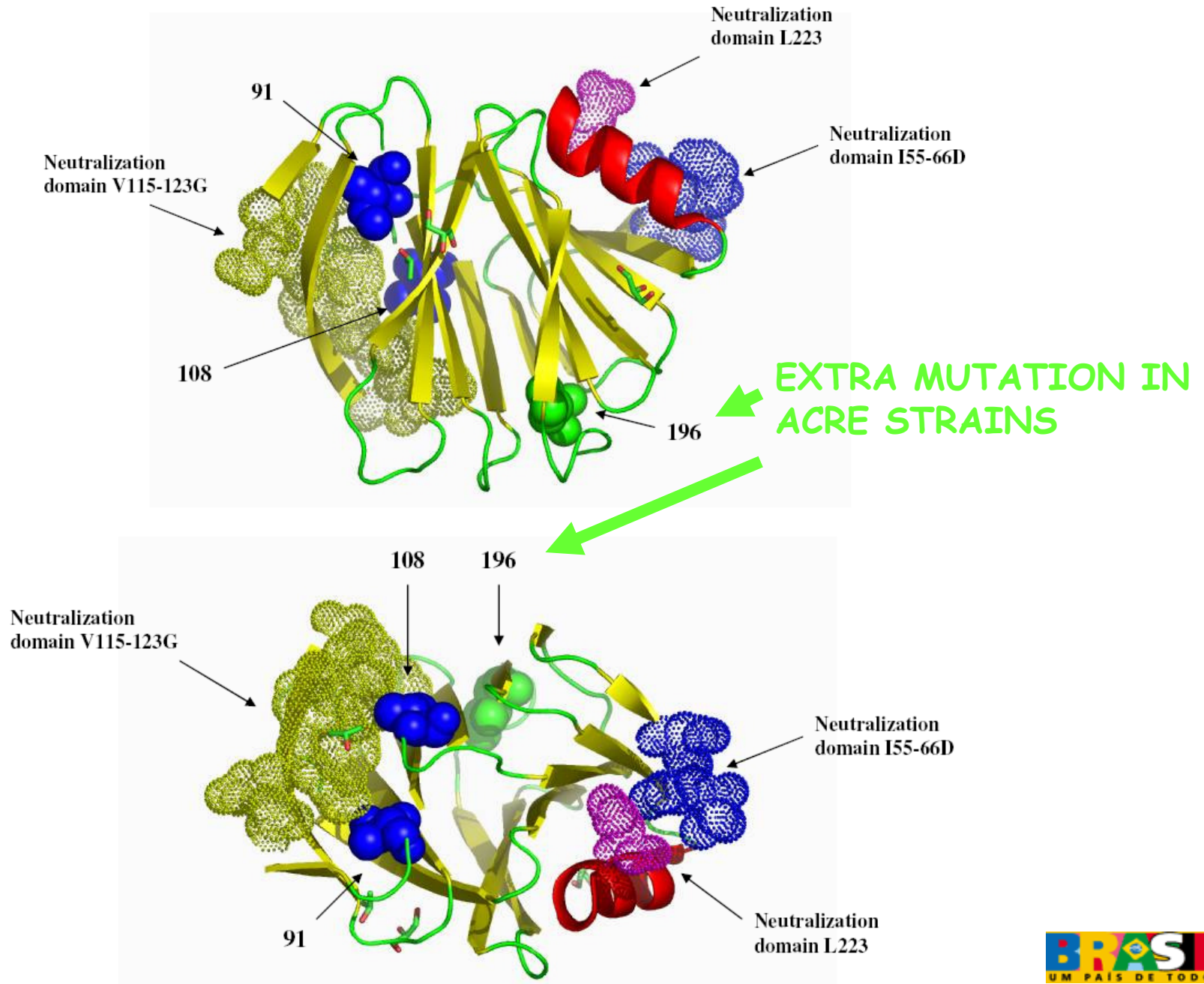
VI



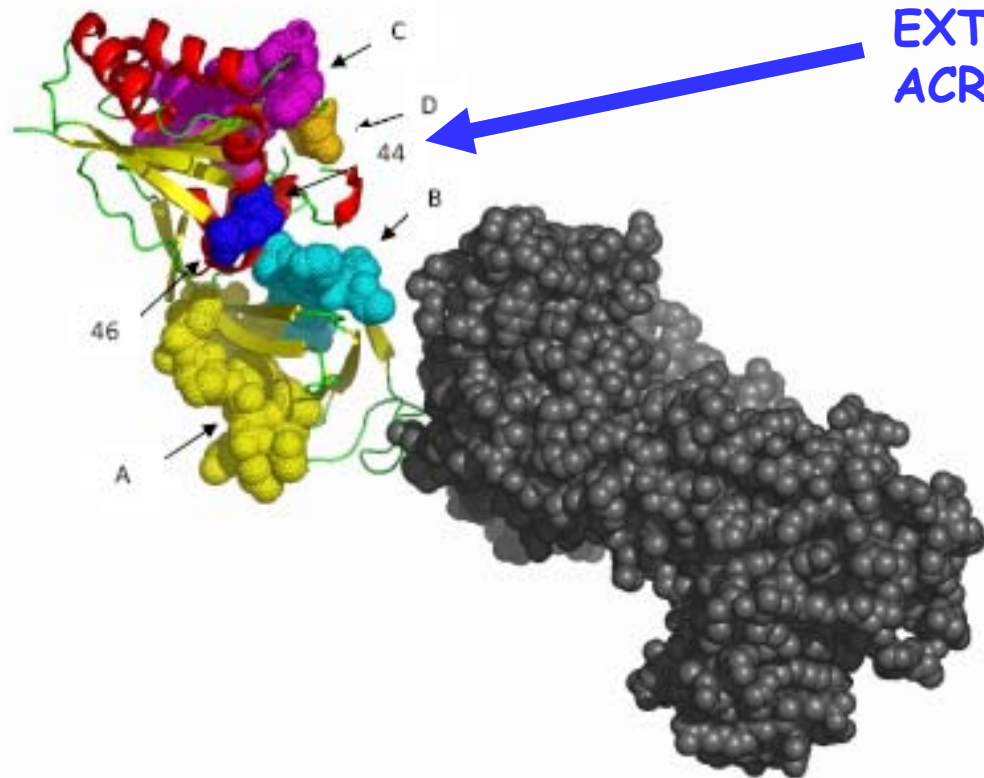
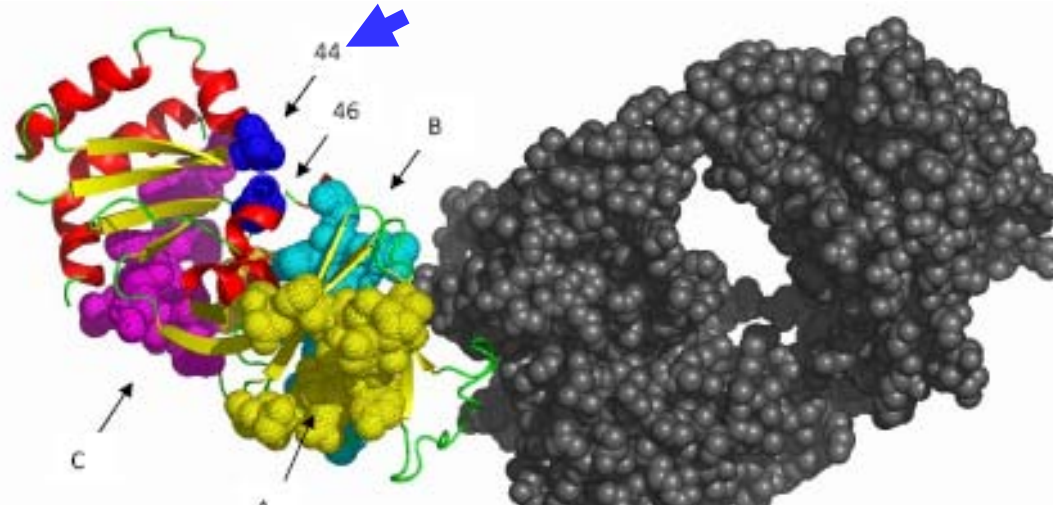
VP7



3D Model of VP8*Sub-Unit of VP4 Protein



3D Model of VP7 Protein

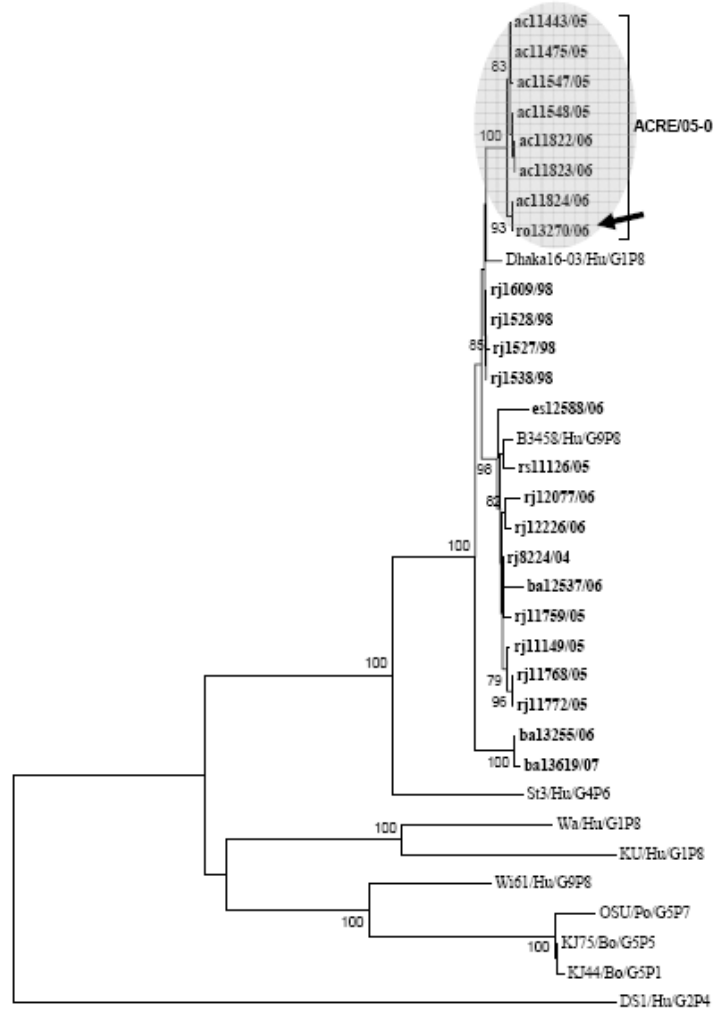


EXTRA MUTATION IN
ACRE STRAINS

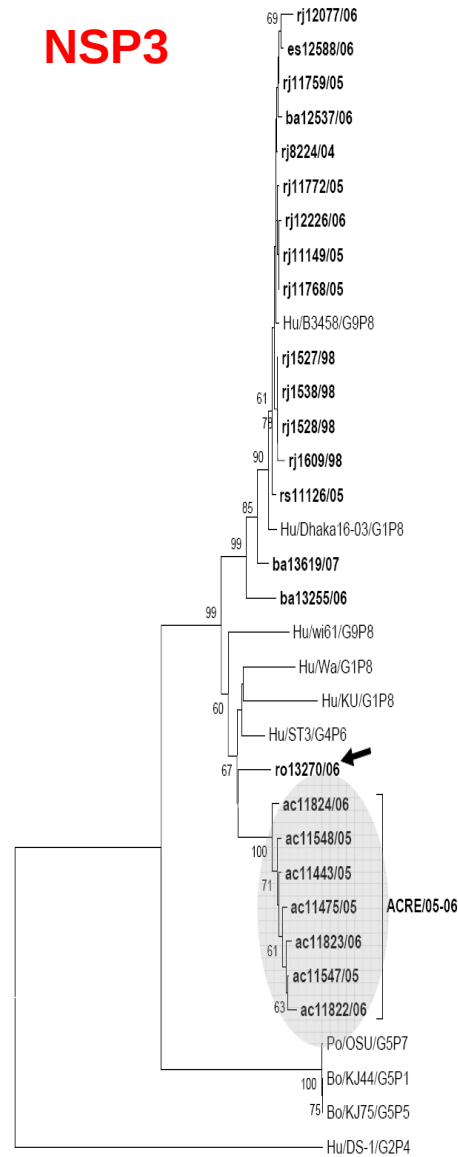


Genes NSP1, NSP3 e NSP5

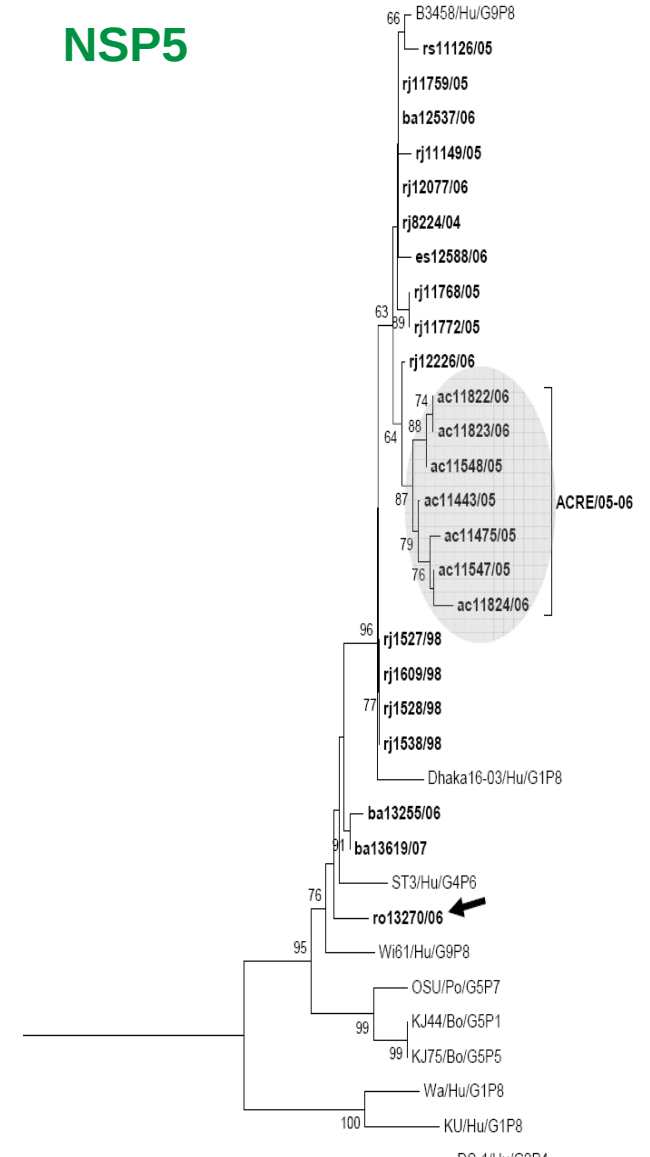
NSP1



NSP3



NSP5



Phylogenetic analysis of human P[8],G9 rotavirus A strains circulating in Brazil reveals the presence of a novel genetic variant

Article Type: Full Length Article (2000 words)

Keywords: Rotavirus A; genotypes; phylogenetic analysis; structural genes; non-structural genes; genetic variant

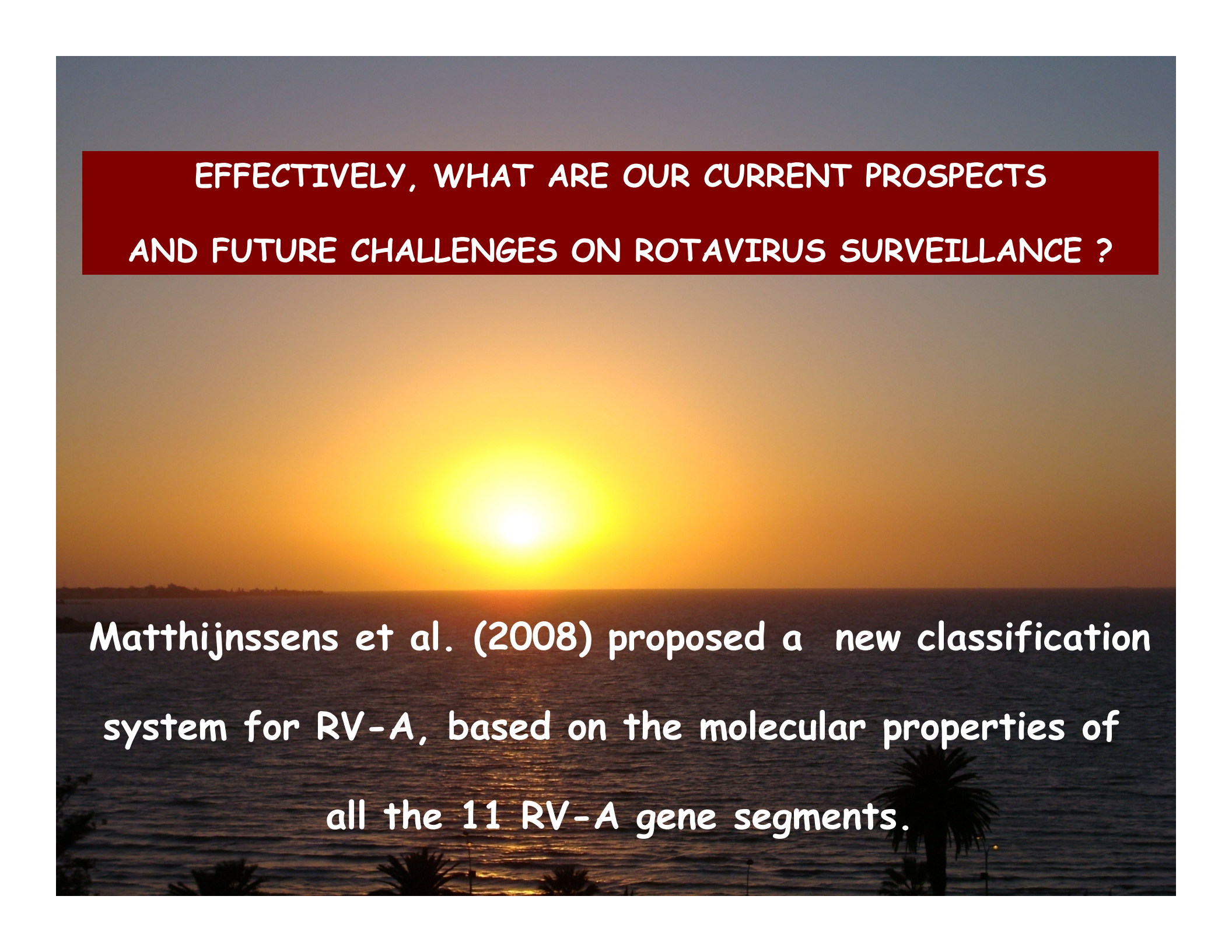
Corresponding Author: PhD José Paulo Gagliardi Leite,

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Order of Authors: Luis Fernando L Tort, Ms.C; Eduardo M Volotão, Ph.D; Marcos César L Mendonça, Ph.D; Marcelle F Silva, Ms.C; Rosane Maria S Assis, Bs.C; Alessandra A Siqueira, Ms.C; Gonzalo Moratorio, Ms.C; Juan Cristina, Ph.D; José Paulo Gagliardi Leite



A sunset over the ocean with palm trees in the foreground. The sun is a bright yellow orb in the center of the frame, casting a long, shimmering reflection on the dark blue water. The sky transitions from a deep orange near the horizon to a pale blue at the top. In the foreground, the silhouettes of palm trees and other coastal vegetation are visible against the dark water.

EFFECTIVELY, WHAT ARE OUR CURRENT PROSPECTS AND FUTURE CHALLENGES ON ROTAVIRUS SURVEILLANCE ?

Matthijnssens et al. (2008) proposed a new classification system for RV-A, based on the molecular properties of all the 11 RV-A gene segments.

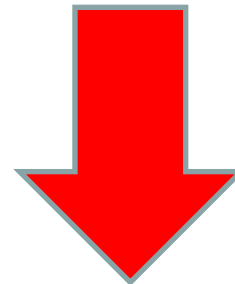
Recommendations for the classification of group A rotaviruses using all 11 genomic RNA segments

Jelle Matthijnssens · Max Ciarlet · Mustafizur Rahman · Houssam Attoui · Krisztián Bányai · Mary K. Estes · Jon R. Gentsch · Miren Iturriza-Gómara · Carl D. Kirkwood · Vito Martella · Peter P. C. Mertens · Osamu Nakagomi · John T. Patton · Franco M. Ruggeri · Linda J. Saif · Norma Santos · Andrej Steyer · Koki Taniguchi · Ulrich Desselberger · Marc Van Ranst

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Gene	Valores cut-off de identidade nucleotídica (%)	Genótipos	Designação dos nomes dos génotipos
VP7	80	19G	Glicoproteína
VP4	80	27P	Sensível a Protease
VP6	85	11I	Capsídeo Interno
VP1	83	4R	RNA polimerasa – RNA dependente
VP2	84	5C	Proteína do Core
VP3	81	6M	Metiltransferase
NSP1	79	14A	Antagonista do Interferon
NSP2	85	5N	NTPase
NSP3	85	7T	Intensificador da Tradução
NSP4	85	11E	Enterotoxina
NSP5	91	6H	Fosfoproteína (<i>pHosphoprotein</i>)

Gx-P[x]-Ix-Rx-Cx-Mx-Ax-Nx-Tx-Ex-Hx



VP7-VP4-VP6-VP1-VP2-VP3-NSP1-NSP2-NSP3-NSP4-NSP5/6

EFFECTIVELY, WHAT ARE OUR CURRENT PROSPECTS
AND FUTURE CHALLENGES ON ROTAVIRUS SURVEILLANCE ?

Environmental RV-A investigation should be
also considered

Molecular Detection and Characterization of Gastroenteritis Viruses Occurring Naturally in the Stream Waters of Manaus, Central Amazônia, Brazil[¶]

Marize P. Miagostovich,¹ Fabiana F. M. Ferreira,¹ Flávia R. Guimarães,¹ Túlio M. Fumian,¹
Leonardo Diniz-Mendes,¹ Sérgio Luiz B. Luz,² Luciete A. Silva,²
and José Paulo G. Leite^{1*}

TABLE 2. Frequencies of the viruses investigated and fecal coliforms detected in the areas studied

Pathogen	No. (%) found in:		Chi square	<i>P</i> value
	Urban area (<i>n</i> = 24)	Rural and forest areas (<i>n</i> = 28)		
Any enteric virus	21 (87.5)	10 (35.7)	14.4	<0.001
RV	15 (62.5)	8 (28.6)	6.0	0.014
NoV	3 (12.5)		3.7	0.09
HAsTV	6 (25.0)	2 (7.1)	3.2	0.08
HAdV	15 (62.5)	1 (3.6)	21.1	<0.001
Fecal coliforms	24 (100.0)	14 (50.0)	16.4	<0.001

RV = HAdV > HAsTV > NoV



IN CONCLUSION

VACCINES WILL PREVENT MORTALITY ASSOCIATED WITH ROTAVIRUS A INFECTION IN DEVELOPING COUNTRIES



Children should be saved
independently of their race,
nationality, religion, social
and economic conditions !



LABORATORY OF COMPARATIVE AND ENVIRONMENTAL VIROLOGY





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**Ministério
da Saúde**



Thank You and Welcome
Gracias y Bienvenido

