Clinical and Molecular Characteristics of Human Rotavirus G8P[8] Outbreak Strain, Japan, 2014

Kenji Kondo, Takeshi Tsugawa, Mayumi Ono, Toshio Ohara, Shinsuke Fujibayashi, Yasuo Tahara, Noriaki Kubo, Shuji Nakata, Yoshihito Higashidate, Yoshiki Fujii, Kazuhiko Katayama, Yuko Yoto, Hiroyuki Tsutsumi

During March–July 2014, rotavirus G8P[8] emerged as the predominant cause of rotavirus gastroenteritis among children in Hokkaido Prefecture, Japan. Clinical characteristics were similar for infections caused by G8 and non-G8 strains. Sequence and phylogenetic analyses suggest the strains were generated by multiple reassortment events between DS-1–like P[8] strains and bovine strains from Asia.

Rotaviruses, the leading cause of acute gastroenteritis in children worldwide, are classified into G and P genotypes on the basis of 2 outer capsid proteins, viral protein (VP) 7 and VP4. A recently established extended rotavirus genotyping system based on the sequence of all 11 genome segments (*1*) grouped most human rotaviruses into 2 genotype constellations: Wa-like (G1/3/4/9-P[8]-I1-R1-C1-M1-A1-N1-T1-E1-H1) and DS-1–like (G2-P[4]-I2-R2-C2-M2-A2-N2-T2-E2-H2) strains.

In industrialized countries, rotavirus genotype G8 infection is common in bovines but rarely occurs in humans; however, the G8 strains are highly prevalent among humans in some countries in Africa (*2*). We investigated the clinical and molecular features of G8P[8] rotavirus, which, we unexpectedly found to be the predominant genotype in southwestern Hokkaido Prefecture, Japan, in 2014.

The Study

During March–July 2014, we obtained rotavirus-positive fecal samples from 165 children in Hokkaido with acute gastroenteritis. The children were receiving care as inpatients or outpatients at 1 of 6 medical facilities (4 hospitals and 2 clinics) in the cities of Sapporo, Tomakomai, Muroran, and Urakawa (Figure 1).

For each fecal sample, we prepared a 10% fecal suspension, from which we extracted viral RNA. We performed reverse transcription PCR on the RNA by using the SuperScript II Reverse Transcriptase (Invitrogen, Carlsbad, CA, USA); PrimeSTAR GXL DNA polymerase (Takara, Shiga, Japan); and previously described primers (*3,4*). We used the BigDye Terminator v.3.1 Cycle Sequencing Reaction Kit (Applied Biosystems, Foster City, CA, USA) to sequence PCR amplicons. For some of the rotavirus samples, next-generation sequencing was performed at the National Institute of Infectious Diseases in Tokyo, Japan, as described previously (*5*). Sequences of the rotaviruses used in this study were submitted to the DDBJ under accession numbers LC102884–LC103134 and LC105000–LC105532.

We successfully determined G and P genotypes for 148 of the 165 rotavirus samples by using the RotaC rotavirus genotyping tool (http://rotac.regatools.be/). The most common genotype was G8P[8], which was identified in 58 samples (39.2%), followed by G1P[8] (25.7%), G9P[8] (20.3%), and G2P[4] (12.8%) (Table 1; online Technical Appendix, https://wwwnc.cdc.gov/EID/article/23/6/16-0038-Techapp1.pdf).

We obtained clinical data for all 84 patients who sought care at the hospital or clinic in Tomakomai. Demographic and clinical characteristics (e.g., age, sex, history of rotavirus vaccination, duration of fever, and duration and frequency of diarrhea and vomiting) were not substantially different between 42 patients with G8P[8] rotavirus infection and 42 patients with non-G8P[8] rotavirus infection. The proportion of patients admitted to hospitals was also similar in the 2 groups (Table 1).

We selected 15 G8P[8] strains for whole-genome analysis. All strains had the same genotype constellation, G8-P[8]-I2-R2-C2-M2-A2-N2-T2-E2-H2, indicating a genomic backbone of the DS-1 genotype constellation. The genomes of these G8P[8] strains shared >99.6% nt identity with each other (Table 2; online Technical Appendix). All 11 genome segments of strain To14-0 (the representative G8P[8] strain in this study) exhibited the highest nucleotide identity to human G8P[8] strains isolated in Southeast Asia in 2014 (represented by strain RVN1149 from Vietnam [>99.4% nt identity] and NP-130 from Thailand [>99.5% nt identity]) (*6,7*) (Table 2). This finding suggests that the strains share a common G8P[8] origin.

Author affiliations: Sapporo Medical University School of Medicine, Sapporo, Japan (K. Kondo, T. Tsugawa, M. Ono, Y. Yoto, H. Tsutsumi); Tomakomai City Hospital, Tomakomai, Japan (T. Ohara); Tomakomai Children's Clinic, Tomakomai (S. Fujibayashi); Steel Memorial Muroran Hospital, Muroran, Japan (Y. Tahara); Japanese Red Cross Urakawa Hospital, Urakawa, Japan (N. Kubo); Nakata Pediatric Clinic, Sapporo (S. Nakata); Sapporo Hokushin Hospital, Sapporo (Y. Higashidate); National Institute of Infectious Diseases, Tokyo, Japan (Y. Fujii, K. Katayama)

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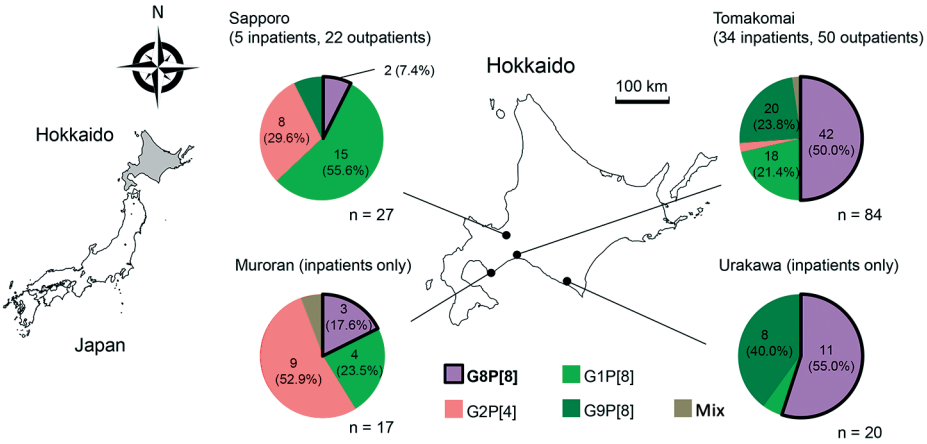

Figure 1. Distribution of rotavirus samples and their G/P genotypes in Hokkaido Prefecture (center map), Japan, 2014. The 4 locations from which the fecal samples were collected are shown on the map. Four hospitals (Tomakomai City Hospital, Japanese Red Cross Urakawa Hospital, Steel Memorial Muroran Hospital, and Sapporo Hokushin Hospital), and 2 clinics (Nakata Pediatric Clinic [Sapporo] and Tomakomai Children's Clinic) participated in the study. Map at left shows location of Hokkaido in Japan (gray shading).

The VP7 gene of rotavirus strain To14-0 shared the highest nucleotide identity with the VP7 genes of human G8P[8] strains from Southeast Asia, including strains RVN1149 and NP-130 (99.4% and 99.7% identity, respectively), and it shared slightly lower identity to the VP7 gene of human strain 04-97s379 (97.8%) from Taiwan, which is speculated to be of bovine origin (*8*) (Figure 2). The VP7 genes of other G8 strains isolated in Japan were more distantly related to the To14-0 VP7 gene (e.g., human AU109 and bovine strains shared 89.5% and 81.9%–85.1%, respectively, with To14-0) (*9*). The VP7 genes of the human G8 strains prevailing in Africa were also distantly related (<90% nt identity) to the VP7 gene of To14-0.

Among the 11 To14-0 genome segments, 8 (VP2–VP4, VP6, nonstructural protein [NSP] 1–3, and NSP5) were highly similar to those of the human DS-1–like P[8] strains that have been isolated in Asia since 2012 (e.g., SKT-109, NT004, and LS-04) (*10–12*), including the strains isolated in this study (e.g., To14-41) (Table 2; online Technical Appendix Figure, panels B–H, J). In addition, the VP6 and NSP5 genes of the strains isolated in this study were also highly similar to those of human G2P[4] strains circulating in South Korea (strain CAU15-11) and Thailand (strain NP-M51) (*12*).

In contrast, the VP1 and NSP4 genes of To14-0 were only distantly related to those of the DS-1–like P[8] strains isolated in Asia (e.g., SKT-109, NT004, and LS-04), including the strains isolated in this study (e.g., To14-41) (Table 2; online Technical Appendix Figure, panels A and I). The To14-0 VP1 gene shared high nucleotide identity with the VP1 genes of human G10P[14] strain PR457 from Italy (98.1%), which are probably the result of independent zoonotic transmissions (*13*). The To14-0 NSP4 gene shared high nucleotide identity with the NSP4 genes of human strains BSGH38 from India (96.7%) and the caprine G6P[1] strain GO34 from Bangladesh (96.0%) (*14*).

Conclusions

The clinical characteristics recorded for patients infected with G8P[8] rotaviruses and those infected with non-G8P[8] rotaviruses did not differ (Table 1). Our findings suggest that the severity of gastroenteritis caused by newly emerging G8P[8] rotaviruses could possibly be attenuated by 1) the existence of VP7/VP4 genotype cross-reactive (heterotypic) protective responses; 2) protective immunity associated with other segments, such as VP6 and NSP4 (*3,15*); or 3) both of these factors combined.

Table 1. Characteristics of patients infected with (G8P[8] and non-G8P[8] rotavirus, Tomakomai, Hokkaido Prefecture, Japan, 2014

Patient characteristic	Patients infected with rotavirus		p value*
	G8P[8], n = 42	Non-G8P[8], n = 42	
Sex, no.			1.0
M	24	24	
F	18	18	
Median age, y	1.8	2.0	0.50
Vaccinated, no (%)	2 (4.8)	1 (2.4)	0.56
Clinical symptoms, mean ± SD			
Duration of fever, d	1.3 ± 1.2	1.7 ± 1.4	0.13
Duration of diarrhea, d	2.4 ± 1.6	2.5 ± 3.2	0.51
Diarrhea episode in 24 h, maximum no.	3.6 ± 3.2	4.3 ± 4.2	0.69
Duration of vomiting, d	1.4 ± 1.2	1.8 ± 2.9	0.79
Vomiting episodes in 24 h, maximum no.	3.6 ± 3.8	3.7 ± 3.5	0.98
Admitted to hospital, no. (%)	18 (42.9%)	16 (38.1%)	0.66

*Categorical data were analyzed by using χ^2 tests; continuous data were analyzed by using Student's *t*-tests or Mann-Whitney U tests.

Table 2. Genotype constellations and nucleotide identities of strains closely related to To14-0, the representative G8P[8] rotavirus strain used in a study of the clinical and molecular features of a G8P[8] rotavirus outbreak strain, Hokkaido Prefecture, Japan, 2014

Strain (genotype representative from study/country)	Genotype constellations and nucleotide identities (%), by gene*										
	VP7	VP4	VP6	VP1	VP2	VP3	NSP1	NSP2	NSP3	NSP4	NSP5
Human/To14-0 (G8P[8] in study)	G8 (**100**)	P[8] (**100**)	I2 (**100**)	R2 (**100**)	C2 (**100**)	M2 (**100**)	A2 (**100**)	N2 (**100**)	T2 (**100**)	E2 (**100**)	H2 (**100**)
Human/VNM/RVN1149/2014/G8P[8] (G8P[8] in Vietnam)	G8 (**99.4**)	P[8] (**99.8**)	I2 (**99.8**)	R2 (**99.6**)	C2 (**99.6**)	M2 (**99.8**)	A2 (**100**)	N2 (**99.9**)	T2 (**99.5**)	E2 (**99.9**)	H2 (**99.5**)
Human/THA/NP-130/2014/G8P[8] (G8P[8] in Thailand)	G8 (**99.7**)	P[8] (**99.9**)	I2 (**99.9**)	R2 (**99.9**)	C2 (**99.8**)	M2 (**99.9**)	A2 (**99.9**)	N2 (**99.7**)	T2 (**99.8**)	E2 (**99.7**)	H2 (**99.5**)
Human/THA/KKL-17/2013/G8P[8] (G8P[8] in Thailand)	G8 (**99.5**)	P[8] (**99.8**)	I2 (91.5)	R2 (**99.9**)	C2 (**99.9**)	M2 (**99.9**)	A2 (**99.9**)	N2 (**99.8**)	T2 (**99.9**)	E2 (**99.7**)	H2 (**99.2**)
Human/THA/LS-04/2013/G2P[8] (DS-1–like G2P[8] in Thailand)	G2 (–)	P[8] (**99.8**)	I2 (**99.5**)	R2 (86.0)	C2 (**99.8**)	M2 (97.4)	A2 (**99.9**)	N2 (**99.8**)	T2 (98.6)	E2 (95.5)	H2 (**99.7**)
Human/THA/SKT-109/2013/G1P[8] (DS-1–like G1P[8] in Thailand)	G1 (–)	P[8] (**99.7**)	I2 (**98.7**)	R2 (86.1)	C2 (**99.9**)	M2 (**99.8**)	A2 (**99.8**)	N2 (85.8)	T2 (**99.8**)	E2 (94.5)	H2 (**99.2**)
Human/To14-41 (DS-1–like G1P[8] in study)	G1 (–)	P[8] (98.8)	I2 (98.3)	R2 (86.3)	C2 (**99.4**)	M2 (**99.4**)	A2 (**99.5**)	N2 (85.5)	T2 (**99.6**)	E2 (93.5)	H2 (98.8)
Human/JPN/NT004/2012/G1P[8] (DS-1–like G1P[8] in Japan)	G1 (–)	P[8] (**99.0**)	I2 (98.4)	R2 (86.3)	C2 (**99.7**)	M2 (**99.5**)	A2 (**99.5**)	N2 (85.6)	T2 (**99.6**)	E2 (94.5)	H2 (**99.2**)
Human/THA/NP-M51/2013/G2P[4] (G2P[4] in Thailand)	G2 (–)	P[4] (–)	I2 (**99.5**)	R2 (86.0)	C2 (**99.1**)	M2 (97.5)	A2 (**99.0**)	N2 (85.4)	T2 (98.7)	E2 (95.5)	H2 (**99.7**)
Human/KOR/CAU15–11/2015/G2P[4] (G2P[4] in South Korea)	G2 (–)	P[4] (–)	I2 (**99.5**)	R2 (85.9)	C2 (**99.2**)	M2 (97.4)	A2 (94.0)	N2 (84.8)	T2 (98.1)	E2 (95.3)	H2 (98.6)
Human/MU14114 (G2P[4] in study)	G2 (–)	P[4] (–)	I2 (97.0)	R2 (85.3)	C2 (96.9)	M2 (86.4)	A2 (95.8)	N2 (85.5)	T2 (96.7)	E2 (95.6)	H2 (98.3)
Human/ITA/PR457/2009/G10P[14]	G10 (–)	P[14] (–)	I2 (93.0)	R2 (98.1)	C2 (82.3)	M2 (86.6)	A11 (–)	N2 (86.4)	T6 (–)	E2 (94.3)	H3 (–)
Caprine/BGL/GO34/1999/G6P[1]	G6 (–)	P[1] (–)	I2 (94.9)	R2 (86.3)	C2 (83.4)	M2 (86.8)	A11 (–)	N2 (86.6)	T6 (–)	E2 (96.0)	H3 (–)
Human/COD/DRC88/2003/G8P[8] (G8P[8] in Africa)	G8 (86.0)	P[8] (98.3)	I2 (96.3)	R2 (85.6)	C2 (97.4)	M2 (97.0)	A2 (96.4)	N2 (85.8)	T2 (97.1)	E2 (90.1)	H2 (97.4)
Human/MWI/QEC287/2006/G8P[8] (G8P[8] in Africa)	G8 (85.5)	P[8] (98.3)	I2 (96.3)	R2 (85.6)	C2 (97.8)	M2 (96.0)	A2 (96.5)	N2 (86.9)	T2 (96.6)	E2 (89.2)	H2 (97.6)
Human/MWI/QEC289/2006/G8P[8] (G8P[8] in Africa)	G8 (85.5)	P[8] (97.5)	I2 (96.3)	R2 (85.5)	C2 (97.8)	M2 (95.9)	A2 (96.5)	N2 (86.9)	T2 (96.6)	E2 (89.2)	H2 (97.6)

*Purple indicates G8 genotype; green indicates Wa-like genome segments; red indicates DS-1–like genome segments; bold indicates nucleotide identities >99.0%; – indicates percentages not calculated because the genotype was different that for To14-0.

The VP7 genes of the human G8P[8] strains isolated in this study and in Southeast Asia appear to have a close relationship with bovine strains from Asia but not from Japan, and the VP7 gene of human G8 or bovine G8 strains previously isolated in Japan are distantly related to them. Therefore, the VP7 genes in the G8P[8] strains from this study may have originated from a bovine strain from Asia. As with the VP7 genes, the VP1 and NSP4 genes are also assumed to have been derived from artiodactyl strains.

Eight genome segments (VP2–VP4, VP6, NSP1–NSP3, and NSP5) of the human G8P[8] strains isolated in this study and from Southeast Asia are closely related to those of the DS-1–like P[8] strains that have emerged and spread in Japan and other countries of Asia since 2012 (Table 2). Therefore, these 8 genome segments of the G8P[8] strains from this study may be derived from the DS-1–like P[8] strains in Asia.

For the reasons we have stated, the G8P[8] strains isolated in this study were speculated to be formed outside of Japan by multiple reassortment events between the DS-1–like P[8] strains and bovine strains in Asia. The resulting strain was probably recently introduced into Japan.

The predominance of novel DS-1–like G8P[8] strains noted in this study indicates that these strains are sufficiently adapted to humans to sustain human-to-human transmission in an industrialized country. This finding suggests that these G8P[8] rotavirus strains could spread to other regions in the near future. Continuing surveillance is required to monitor the circulating wild-type strains, and rotavirus genotype constellations and clinical information must be analyzed to understand rotavirus virulence in humans.

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Dr. Kondo is a pediatrician in Department of Pediatrics, Sapporo Medical University School of Medicine. His primary research interests are molecular biology and the epidemiology of rotaviruses.

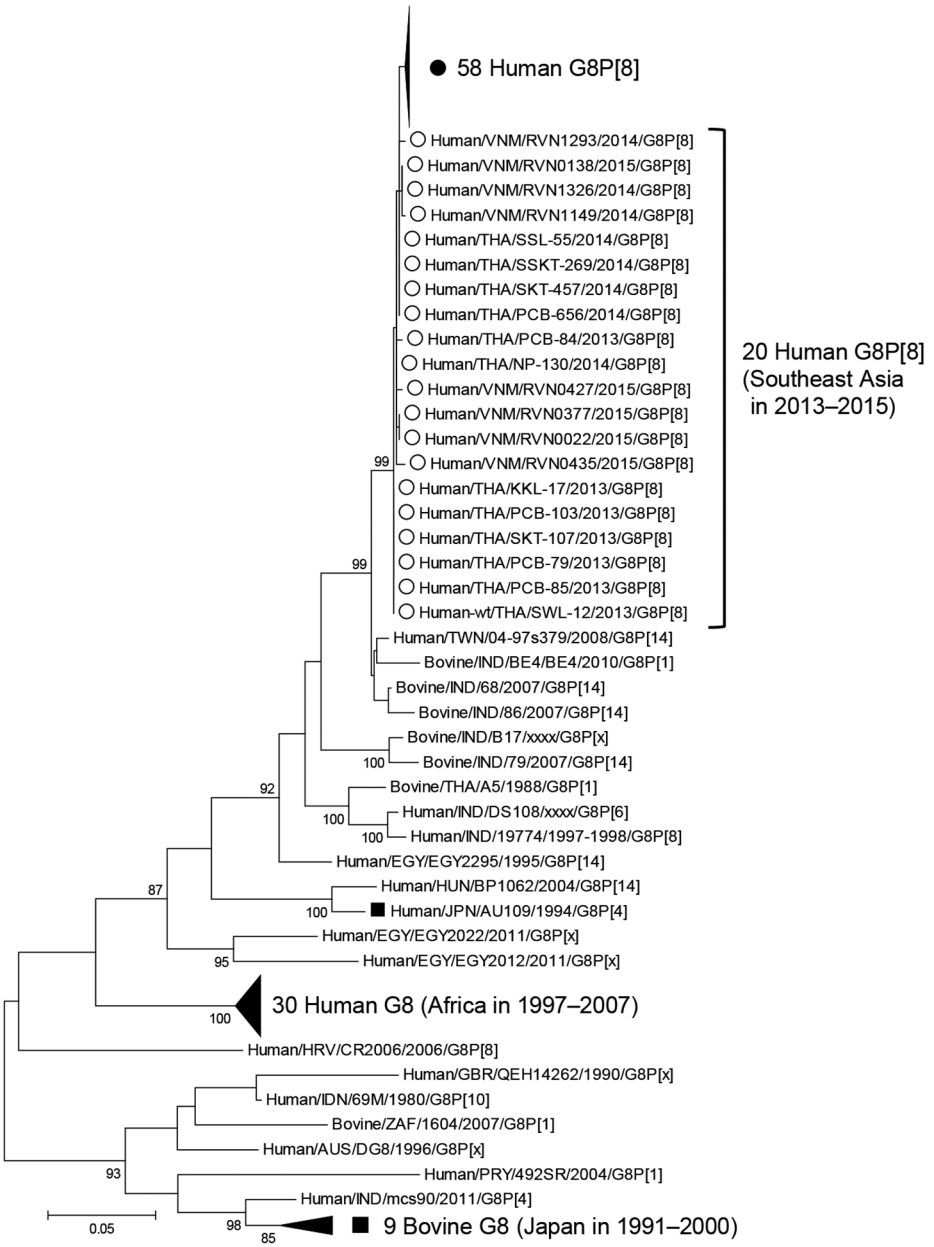

Figure 2. Phylogenetic analysis of the viral protein 7 gene of G8 rotavirus strains used in a study of the clinical and molecular features of a G8P[8] rotavirus outbreak strain, Hokkaido Prefecture, Japan, 2014. Closed circle indicates the G8P[8] rotavirus strain from Hokkaido; open circles indicate human G8P[8] strains from Southeast Asia; and closed boxes indicate other strains from Japan. A Tamura 3-parameter model was used for the maximum-likelihood method. Bootstrap values are shown at the branch nodes (values of <80% are not shown). Scale bar indicates nucleotide substitutions per site.

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Address for correspondence: Takeshi Tsugawa, Department of Pediatrics, Sapporo Medical University School of Medicine, Minami 1-jo, Nishi 16-chome, Chuo-ku, Sapporo, Hokkaido 060-8543, Japan; email: tsugawat@sapmed.ac.jp

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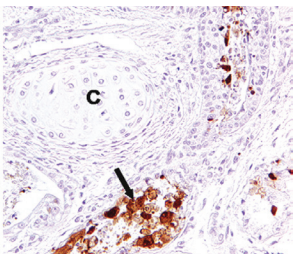
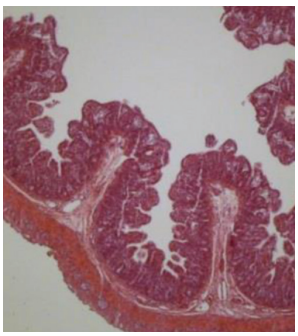
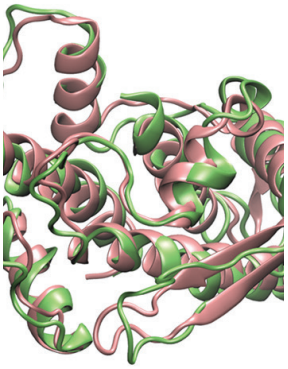
- Hantavirus Pulmonary Syndrome, Southern Chile, 1995–2012



- Animal-Associated Exposure to Rabies Virus among Travelers, 1997–2012
- Evolution of Ebola Virus Disease from Exotic Infection to Global Health Priority, Liberia, Mid-2014
- Influenza A(H7N9) Virus Transmission between Finches and Poultry
- Population Structure and Antimicrobial Resistance of Invasive Serotype IV Group B *Streptococcus*, Toronto, Ontario, Canada
- Norovirus Genotype Profiles Associated with Foodborne Transmission, 1999–2012
- Sequence Variability and Geographic Distribution of Lassa Virus, Sierra Leone
- Highly Pathogenic Avian Influenza A(H5N1) Virus Infection among Workers at Live Bird Markets, Bangladesh, 2009–2010
- Deaths Associated with Respiratory Syncytial and Influenza Viruses among Persons >5 Years of Age in HIV-Prevalent Area, South Africa
- Nairobi Sheep Disease Virus RNA in Ixodid Ticks, China
- Increased Risk for Group B Streptococcus Sepsis in Young Infants Exposed to HIV, Soweto, South Africa, 2004–2008
- Bat Coronavirus in Brazil Related to Appalachian Ridge and Porcine Epidemic Diarrhea Viruses
- Tandem Repeat Insertion in African Swine Fever Virus, Russia, 2012
- Norovirus GII.21 in Children with Diarrhea, Bhutan





- Enterovirus D68 Infection, Chile, Spring 2014
- Zika Virus Infection, Philippines, 2012
- Chikungunya Outbreak, French Polynesia, 2014
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- Hepatitis E Epidemic, Biratnagar, Nepal, 2014

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Clinical and Molecular Characteristics of Human Rotavirus G8P[8] Outbreak Strain, Japan, 2014

Technical Appendix

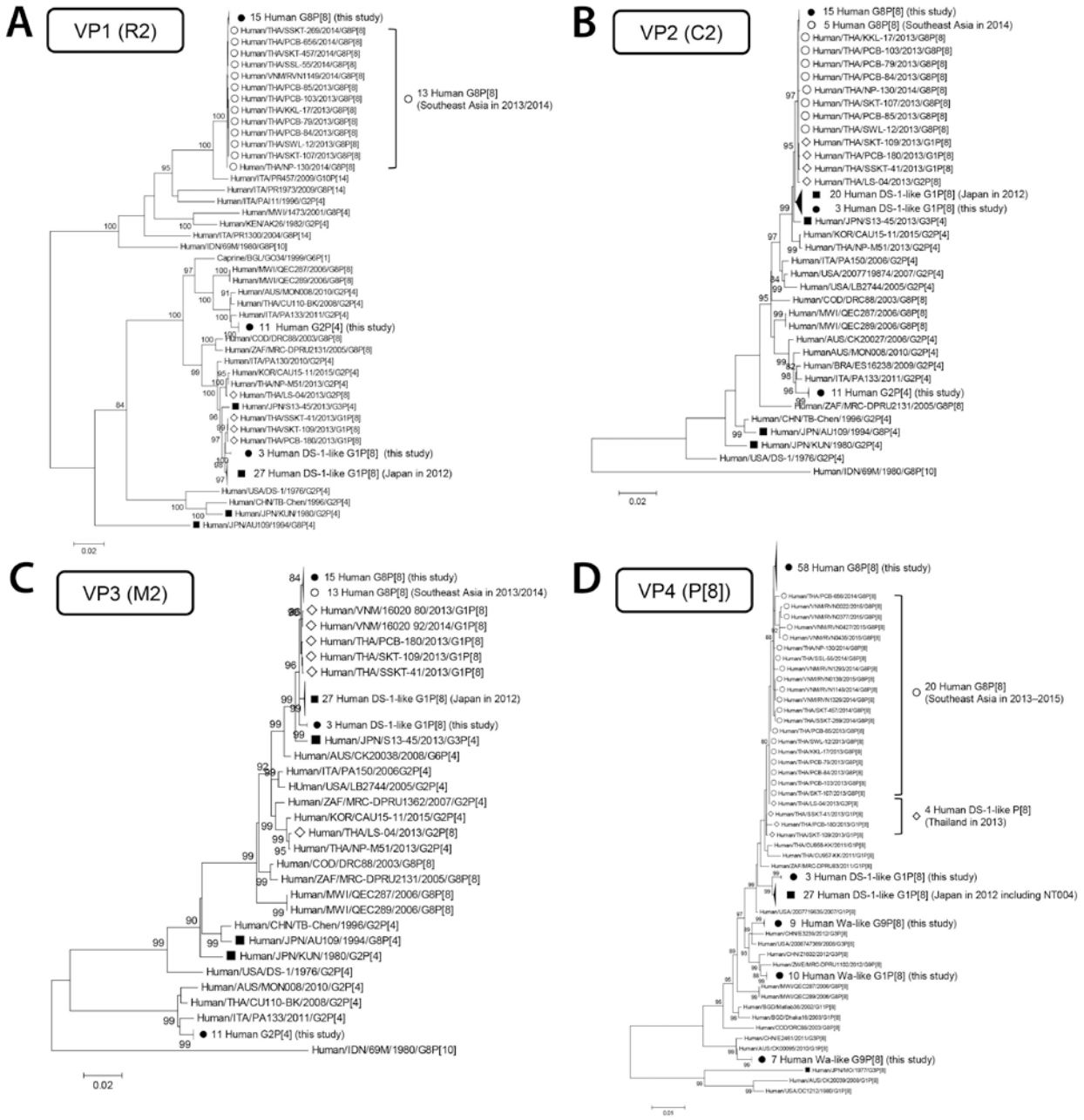
Technical Appendix Table 1. Distribution of G/P genotype combinations detected in Hokkaido, Japan in 2014

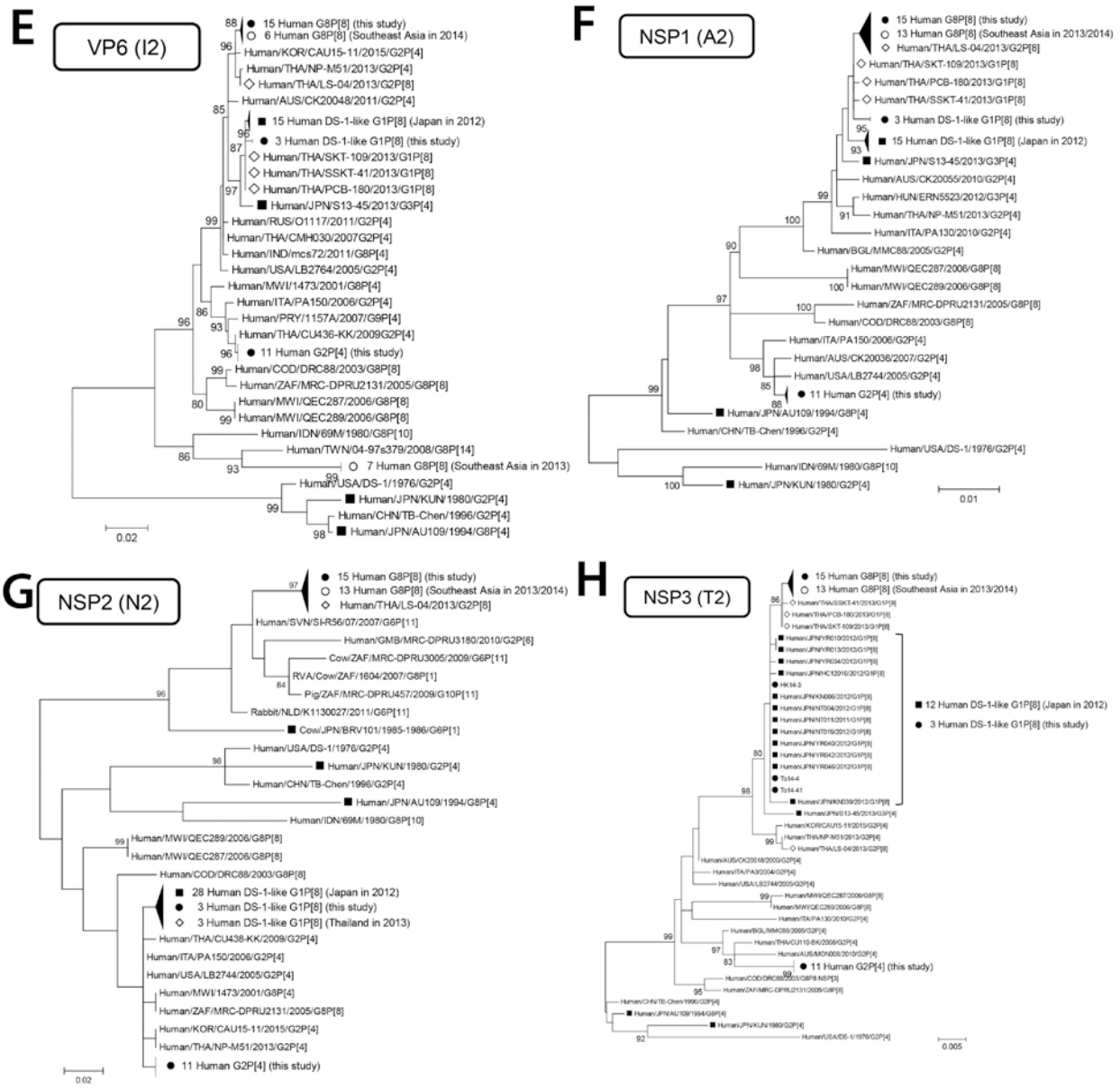
City	No. (%) typeable sample	G8P[8]	G1P[8]	G9P[8]	G2P[4]	Mixed*
Tomakomai	84	42 (50.0)	18 (21.4)	20 (23.8)	2 (2.4)	2 (2.4)
Urakawa	20	11 (55.0)	1 (5.0)	8 (40.0)	0	0
Muroran	17	3 (17.6)	4 (23.5)	0	9 (52.9)	1 (5.9)
Sapporo	27	2 (7.4)	15 (55.6)	2 (7.4)	8 (29.6)	0
Total	148	58 (39.2)	38 (25.7)	30 (20.3)	19 (12.8)	3 (2.0)

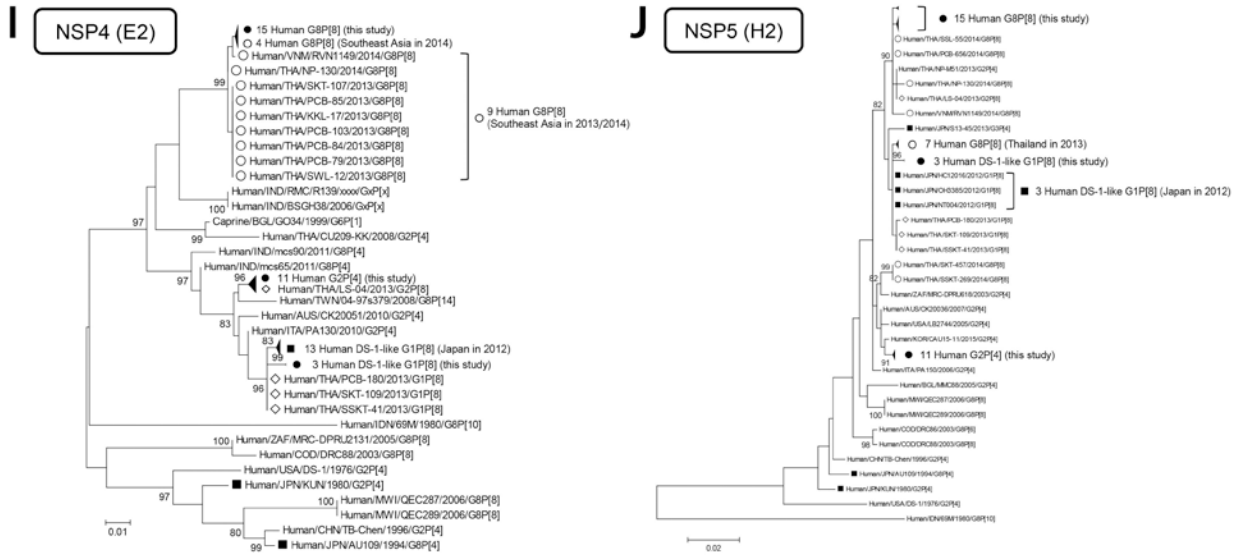
*Mixed infections: Tomakomai, G1P[8]+G9P[8]; Muroran, G2P[4]+G9P[8].

Technical Appendix Table 2. Nucleotide identities (%) at each genome segment of G8P[8] strains from this study compared with To14-0 (the representative G8P[8] strain in this study)

Location, strain name	VP7	VP4	VP6	VP1	VP2	VP3	NSP1	NSP2	NSP3	NSP4	NSP5
Tomakomai											
To14-0	100	100	100	100	100	100	100	100	100	100	100
To14-2	99.9	100	100	99.9	100	99.9	100	100	99.9	100	100
To14-14	99.9	99.9	99.9	100	100	100	100	100	99.9	100	100
To14-36	99.8	100	100	99.9	100	99.9	99.9	100	99.9	99.8	99.7
TKC14-6	99.9	99.6	100	100	99.9	100	100	99.8	99.9	100	100
TKC14-16	99.9	99.6	99.9	99.8	99.9	100	99.8	100	99.8	99.9	99.7
TKC14-37	99.9	99.6	100	99.9	100	99.9	100	100	99.8	99.9	100
Muroran											
MU14-0	99.9	99.6	99.9	100	100	99.9	100	99.9	99.9	99.9	100
MU14-2	99.9	99.9	100	99.9	100	100	99.9	99.9	99.8	99.7	100
MU14-3	99.9	99.6	99.9	99.7	100	99.9	100	100	99.9	100	99.6
Urakawa											
UR14-10	99.9	100	100	100	100	100	100	100	99.9	100	100
UR14-14	99.9	100	99.8	100	100	100	99.9	100	99.9	100	99.9
UR14-29	99.9	99.9	99.8	99.9	100	99.8	99.9	99.9	99.9	100	100
Sapporo											
N14-22	99.9	100	100	100	100	100	99.9	99.9	100	100	100
HK14-2	99.9	99.7	99.7	99.9	100	100	99.9	99.8	99.9	100	100


A
VP1 (R2)
15 Human G8P[8] (this study)
13 Human G8P[8] (Southeast Asia in 2013/2014)
11 Human G2P[4] (this study)
3 Human DS-1-like G1P[8] (this study)
27 Human DS-1-like G1P[8] (Japan in 2012)
0.02
B
VP2 (C2)
15 Human G8P[8] (this study)
5 Human G8P[8] (Southeast Asia in 2014)
20 Human DS-1-like G1P[8] (Japan in 2012)
3 Human DS-1-like G1P[8] (this study)
11 Human G2P[4] (this study)
0.02
C
VP3 (M2)
15 Human G8P[8] (this study)
13 Human G8P[8] (Southeast Asia in 2013/2014)
27 Human DS-1-like G1P[8] (Japan in 2012)
3 Human DS-1-like G1P[8] (this study)
11 Human G2P[4] (this study)
0.02
D
VP4 (P[8])
58 Human G8P[8] (this study)
20 Human G8P[8] (Southeast Asia in 2013–2015)
4 Human DS-1-like P[8] (Thailand in 2013)
3 Human DS-1-like G1P[8] (this study)
27 Human DS-1-like G1P[8] (Japan in 2012 including NT004)
9 Human Wa-like G9P[8] (this study)
10 Human Wa-like G1P[8] (this study)
7 Human Wa-like G9P[8] (this study)


E
VP6 (I2)
15 Human G8P[8] (this study)
6 Human G8P[8] (Southeast Asia in 2014)
Human/KOR/CAU15-11/2015/G2P[4]
Human/THA/NP-M51/2013/G2P[4]
Human/THA/LS-04/2013/G2P[8]
Human/AUS/CK20048/2011/G2P[4]
15 Human DS-1-like G1P[8] (Japan in 2012)
3 Human DS-1-like G1P[8] (this study)
Human/THA/SKT-109/2013/G1P[8]
Human/THA/SSKT-41/2013/G1P[8]
Human/THA/PCB-180/2013/G1P[8]
Human/JPN/S13-45/2013/G3P[4]
Human/RUS/O1117/2011/G2P[4]
Human/THA/CMH030/2007G2P[4]
Human/IND/mcs72/2011/G8P[4]
Human/USA/LB2764/2005/G2P[4]
Human/MWI/1473/2001/G8P[4]
Human/ITA/PA150/2006/G2P[4]
Human/PRY/1157A/2007/G9P[4]
Human/THA/CU436-KK/2009G2P[4]
11 Human G2P[4] (this study)
Human/COD/DRC88/2003/G8P[8]
Human/ZAF/MRC-DPRU2131/2005/G8P[8]
Human/MWI/QEC287/2006/G8P[8]
Human/MWI/QEC289/2006/G8P[8]
Human/IDN/69M/1980/G8P[10]
Human/TWN/04-97s379/2008/G8P[14]
7 Human G8P[8] (Southeast Asia in 2013)
Human/USA/DS-1/1976/G2P[4]
Human/JPN/KUN/1980/G2P[4]
Human/CHN/TB-Chen/1996/G2P[4]
Human/JPN/AU109/1994/G8P[4]
0.02
F
NSP1 (A2)
15 Human G8P[8] (this study)
13 Human G8P[8] (Southeast Asia in 2013/2014)
Human/THA/LS-04/2013/G2P[8]
Human/THA/SKT-109/2013/G1P[8]
Human/THA/PCB-180/2013/G1P[8]
Human/THA/SSKT-41/2013/G1P[8]
3 Human DS-1-like G1P[8] (this study)
15 Human DS-1-like G1P[8] (Japan in 2012)
Human/JPN/S13-45/2013/G3P[4]
Human/AUS/CK20055/2010/G2P[4]
Human/HUN/ERN5523/2012/G3P[4]
Human/THA/NP-M51/2013/G2P[4]
Human/ITA/PA130/2010/G2P[4]
Human/BGL/MMC88/2005/G2P[4]
Human/MWI/QEC287/2006/G8P[8]
Human/MWI/QEC289/2006/G8P[8]
Human/ZAF/MRC-DPRU2131/2005/G8P[8]
Human/COD/DRC88/2003/G8P[8]
Human/ITA/PA150/2006/G2P[4]
Human/AUS/CK20036/2007/G2P[4]
Human/USA/LB2744/2005/G2P[4]
11 Human G2P[4] (this study)
Human/JPN/AU109/1994/G8P[4]
Human/CHN/TB-Chen/1996/G2P[4]
Human/USA/DS-1/1976/G2P[4]
Human/IDN/69M/1980/G8P[10]
Human/JPN/KUN/1980/G2P[4]
0.01
G
NSP2 (N2)
15 Human G8P[8] (this study)
13 Human G8P[8] (Southeast Asia in 2013/2014)
Human/THA/LS-04/2013/G2P[8]
Human/SVN/SI-R56/07/2007/G6P[11]
Human/GMB/MRC-DPRU3180/2010/G2P[6]
Cow/ZAF/MRC-DPRU3005/2009/G6P[11]
RVA/Cow/ZAF/1604/2007/G8P[1]
Pig/ZAF/MRC-DPRU457/2009/G10P[11]
Rabbit/NLD/K1130027/2011/G6P[11]
Cow/JPN/BRV101/1985-1986/G6P[1]
Human/USA/DS-1/1976/G2P[4]
Human/JPN/KUN/1980/G2P[4]
Human/CHN/TB-Chen/1996/G2P[4]
Human/JPN/AU109/1994/G8P[4]
Human/IDN/69M/1980/G8P[10]
Human/MWI/QEC289/2006/G8P[8]
Human/MWI/QEC287/2006/G8P[8]
Human/COD/DRC88/2003/G8P[8]
28 Human DS-1-like G1P[8] (Japan in 2012)
3 Human DS-1-like G1P[8] (this study)
3 Human DS-1-like G1P[8] (Thailand in 2013)
Human/THA/CU438-KK/2009/G2P[4]
Human/ITA/PA150/2006/G2P[4]
Human/USA/LB2744/2005/G2P[4]
Human/MWI/1473/2001/G8P[4]
Human/ZAF/MRC-DPRU2131/2005/G8P[8]
Human/KOR/CAU15-11/2015/G2P[4]
Human/THA/NP-M51/2013/G2P[4]
11 Human G2P[4] (this study)
0.02
H
NSP3 (T2)
15 Human G8P[8] (this study)
13 Human G8P[8] (Southeast Asia in 2013/2014)
12 Human DS-1-like G1P[8] (Japan in 2012)
3 Human DS-1-like G1P[8] (this study)
11 Human G2P[4] (this study)
0.005



Technical Appendix Figure. Phylogenetic analyses of rotavirus genome segments. Phylogenetic trees of the VP1–3 (A–C), VP4 (D), VP6 (E), NSP1–5 (F–J) genes of the human strains sequenced in this study (closed circles) and other reference strains. The human Southeast Asian G8P[8] strain, the human Southeast Asian DS-1-like P[8] strains, and the Japanese strains are indicated by an open circle, open diamonds, and closed boxes, respectively. A Tamura 3-parameter model was used for the maximum-likelihood method. Bootstrap values are shown at the branch nodes (values of <80% are not shown). The scale bars indicate genetic distance.