Life-Threatening Sochi Virus Infections, Russia

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Learning Objectives

Upon completion of this activity, participants will be able to:

- Analyze the demographics of patients infected with the Sochi virus in the current study
- Assess laboratory data available from patients infected with Sochi virus in the current study
- Distinguish the anatomic site of the highest concentration of Sochi virus among infected individuals
- Evaluate the prognosis of infection with Sochi virus.

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Sochi virus was recently identified as a new hantavirus genotype carried by the Black Sea field mouse, *Apodemus ponticus*. We evaluated 62 patients in Russia with Sochi virus infection. Most clinical cases were severe, and the case-fatality rate was as high as 14.5%.

Hantaviruses are zoonotic pathogens transmitted from small animals to humans. Hantavirus disease in the Americas is called hantavirus pulmonary syndrome and in Asia and Europe is called hemorrhagic fever with renal syndrome (HFRS). Both syndromes can lead to cardiopulmonary and renal failure (*1*). Recently we described a new hantavirus, Sochi virus, from the administrative region Krasnodar (including the city of Sochi), southern European Russia, which was isolated in cell culture from a Black Sea field mouse (*Apodemus ponticus*) and a

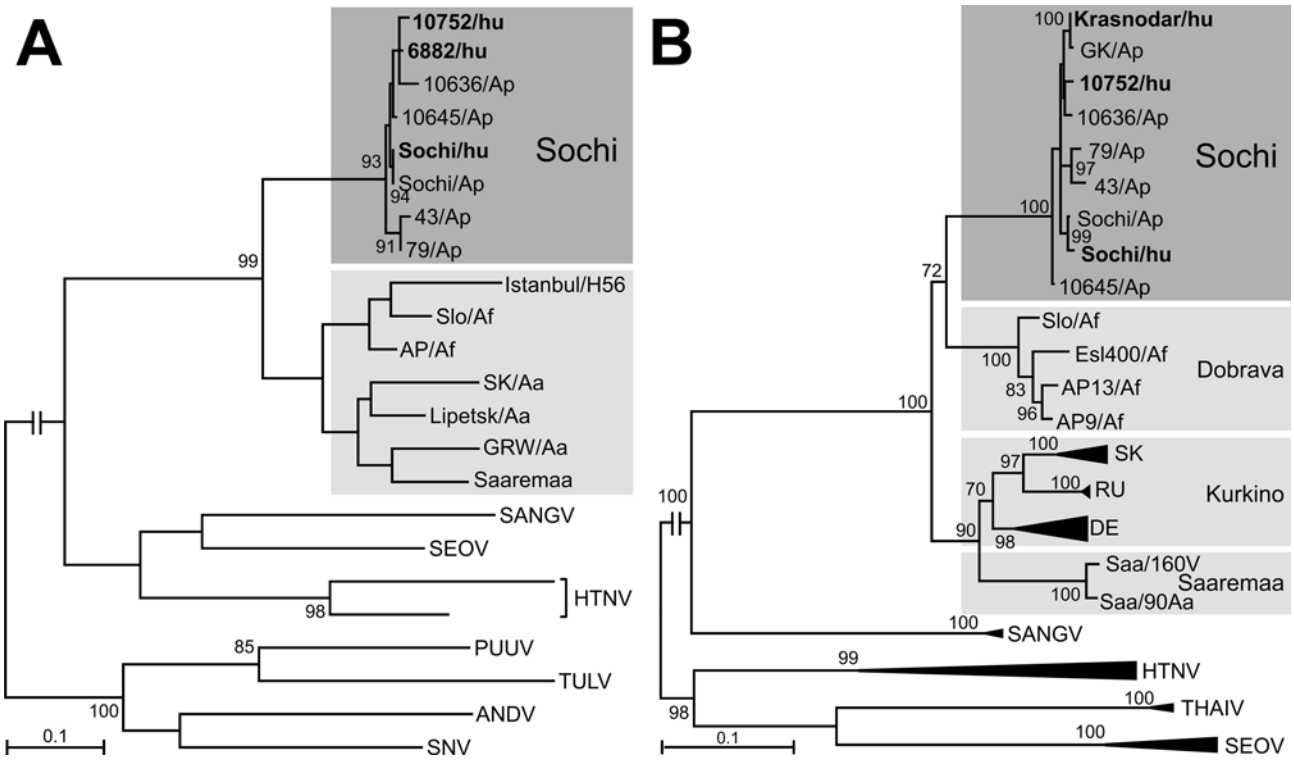

Figure 1. Phylogenetic analysis segment sequences of Sochi virus, Russia: A) 347-bp large (L) segment sequence; B) 1,197-bp small (S) segment sequence. Virus sequences derived from patients (shown in bold type) and *Apodemus ponticus* mice cluster within the Sochi genotype of DOBV. Evolutionary analysis was conducted in MEGA6 (*6*). The evolutionary history was inferred by using the maximum-likelihood method based on the Tamura 3-parameter model with a discrete gamma distribution and 5 rate categories (analysis in panel A) and on the general time reversible model with gamma rates and heterogeneous patterns (analysis in panel B), respectively, which were estimated to be the best-fit substitution model according to the Bayesian information criterion. Scale bars indicate an evolutionary distance of 0.1 substitutions per position in the sequence. Bootstrap values ≥70%, calculated from 500 replicates, are shown at the tree branches. GenBank accession numbers of all sequences used in the analysis are listed in online Technical Appendix Table 1 (http://wwwnc.cdc.gov/EID/article/21/12/15-0891-Techapp1.pdf). Dark gray shading iindicates cluster of DOBV-Sochi strains; light gray shading indicates different clusters of strains from other DOBV genotypes. ANDV, Andes virus; DOBV, Dobrava-Belgrade virus; HTNV, Hantaan virus; PUUV, Puumala virus; SANGV, Sangassou virus; SEOV, Seoul virus; SNV, Sin Nombre virus; THAIV, Thailand virus; TULV, Tula virus.

patient with fulminant hantavirus disease who died of shock and combined kidney and lung failure (*2–4*). Molecular taxonomical analyses identified Sochi virus as a new genotype within the Dobrava-Belgrade virus (DOBV) species (*5*). Here we show that HFRS caused by Sochi virus infection occurs in the geographic region where *A. ponticus* mice are prevalent. For 62 patients infected by this virus during 2000–2013, we evaluated clinical and epidemiologic data.

The Study

Serum of patients with suspected acute hantavirus disease from the Krasnodar region were screened for hantavirus antibodies by indirect immunofluorescence assays and ELISA. Sixty-two patients showed clear DOBV IgG seropositivity. During the acute phase of illness, all patients tested positive for DOBV IgM (data not shown). For 26 patients, sufficient volumes of follow-up serum were available for additional focus reduction neutralization assays to specify neutralizing antibodies. All serum samples exhibited substantially higher neutralizing titers toward DOBV than toward Puumala virus, Hantaan virus, and Seoul virus. When the neutralizing effect of DOBV-positive patients' serum were compared against the different human pathogenic genotypes of DOBV (Dobrava, Kurkino, and Sochi), all serum predominantly reacted with the Sochi genotype (online Technical Appendix Table 1, http://wwwnc.cdc.gov/EID/article/21/12/15-0891-Techapp1.pdf).

We successfully obtained virus genomic large (L) segment sequences from 2 patients (no. 51, specimen no. 6882; no. 59, specimen no. 10752). In the neighborhood

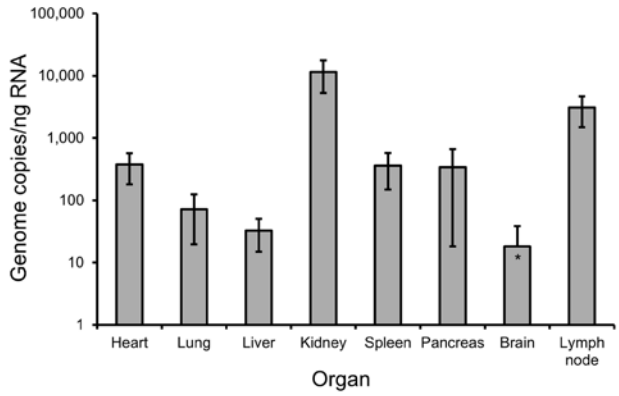

Figure 2. Quantification of hantavirus RNA in tissue biopsies from a 50-year-old Sochi virus–infected man (patient no. 59), Russia. Two independent approaches were performed to extract RNA from each organ. Quantitative reverse transcription PCR previously developed for DOBV (*7*) was used to measure virus load in the analyzed biopsy samples. Three quantitative reverse transcription PCR estimations were conducted for every RNA extraction, followed by calculation of mean values and SDs. Viral RNA levels are shown as genome copies per nanogram of total RNA isolated from the samples. Error bars indicate SD.

of the residence of patient no. 59, mice were trapped, and hantaviral L and small (S) segment regions from 2 *A. ponticus* animals (specimen nos. 10636, 10645) were amplified. The sequences obtained were deposited in GenBank under accession nos. KM192207–09 and KP878308–10 (L segment) and KP878311–13 (S segment) (online Technical Appendix Table 2). Samples from virus-positive mice were phylogenetically characterized by analysis of a 242-bp region of their *cytB* gene; all of them clustered with the previously identified *A. ponticus* animals (*3*) (data not shown). In addition, the *A. ponticus*–derived isolate Sochi/Ap (*4*), the patient-derived isolate Sochi/hu (*5*), an S segment sequence from a mouse (GK/Ap) trapped near the home of the previously described Krasnodar patient (*4*), and sequences originating from 2 *A. ponticus* mice sampled near the Black Sea coast, 43/Ap and 79/Ap, were included in the molecular analyses of the virus.

The patient-derived sequences 6882/hu, 10752/hu, and Sochi/hu clearly cluster with *A. ponticus*–derived sequences 43/Ap, 79/Ap, 10636/Ap, 10645/Ap, and Sochi/Ap (Figure 1, panel A). In the analysis of the S segment, we obtained a very similar result; the patient-derived sequences 10752/hu, Krasnodar/hu, and Sochi/hu cluster with *A. ponticus*–associated sequences 43/Ap, 79/Ap, 10636/Ap, 10645/Ap, GK/Ap, and Sochi/Ap (Figure 1, panel B). In analysis of both L and S segments, the Sochi virus strains form a unique group, clearly distinguishable from all other DOBV genotypes.

Specimens from different organs of deceased patient no. 59 were analyzed for virus load. The highest concentration was detected in kidney (11,446 copies/ng RNA) and lymph node (3,086 copies/ng RNA), whereas the least virus RNA (10–100 copies/ng RNA) was detected in lung, brain, and liver (Figure 2).

The clinical disease severity of the 62 Sochi virus–infected patients investigated (Table 1) was subdivided into mild, moderate, or severe following the standard Russian criteria (i.e., length of febrile phase, minimal blood pressure in the hypotonic phase, extent of hemorrhagic symptoms, minimal urine production, serum creatinine level, and extent of proteinuria) (online Technical Appendix Table 3). The case-fatality rate (CFR) was as high as 14.5% (9/62 patients). Including fatalities, severe disease developed in nearly 60% of patients, whereas the remaining 40% of cases were moderate. The average age of all patients was 33 years. A significantly higher proportion of patients were males ($p = 1.05 \times 10^{-9}$). Moreover, severe disease developed in most affected male patients (66.7%) but in only 35.7% of affected female patients ($p = 0.037$). The fact that only 2 of 9 fatal cases occurred in female patients (Table 1) underscores this finding.

All 9 patients with fatal infections died of multiorgan failure and shock (Table 2). Postmortem examination showed multiple hemorrhages and edema in internal organs, including kidneys and lungs. The patients died within 8.2 days (range 3–16 days) after disease onset. An extraordinary fulminant course was observed for patient no. 47, who died 3 days after onset and before he could be hospitalized. This 19-year-old man was the son-in-law of patient no. 48, who also died after Sochi virus infection. Both men lived at the same rural address, and rodent contact during work in haystacks was reported.

Table 1. Comparisons in clinical outcome, age, and sex of 62 patients with Sochi virus infection, Russia*

	Total		Sex, no. (%)		Age, y, n/N (%)	
Characteristic	No. (%)	Median age, y (range)	M, n = 48	F, n = 14	7–15	>15
No. patients	62 (100)	33.3 (7–57)	**48 (77.4)**	**14 (22.6)**	**6/62 (9.7%)**	**56/62 (90.3)**
Outcome						
Died	9 (14.5)	38.6 (19–53)	7 (14.6)	2 (14.3)	0/6	9/56 (16.1)
Survived	53 (85.5)	32.4 (7–57)	41 (85.4)	12 (85.7)	6/6 (100)	47/56 (83.9)
Illness course						
Severe, including fatal	37 (59.7)	33.1 (10–57)	**32 (66.7)**	**5 (35.7)**	3/6 (50)	34/56 (60.7)
Moderate, mild	25 (40.3)	33.6 (7–57)	**16 (33.3)**	**9 (64.3)**	3/6 (50)	22/56 (39.3)

*Bold type indicates statistically significant differences between sex or age groups. Comparison of binomial population proportions analysis as implemented in Statlets (NWP Associates, Inc., http://www.mrs.umn.edu/~sungurea/statlets/statlets.htm) indicates rejection of the null hypothesis (claiming that the 2 proportions are equal) at significance level of $p<0.05$.

Table 2. Characteristics of 9 deceased patients with Sochi virus infection, Russia*

Patient no.	Age, y/sex	Hospitalized, no. d after onset	GI symptoms	Max serum creatinine, μmol/L†	Min platelet count, $\times 10^9$/L‡	Died, no. d after onset	Clinical and postmortem findings
23	33/M	5	No	148	70	8	Pneumonia; renal, cardiovascular, multiorgan failure; multiple internal hemorrhages, edema
29	29/M	Same day	Yes	282	115	6	Renal, cardiovascular, multiorgan failure; multiple internal hemorrhages, edema
30	47/F	5	Yes	391	38	12	Renal, lung failure; shock; coagulation disturbance; hemorrhagic gastroenteritis; multiple internal hemorrhages, edema
34	53/M	3	Yes	250	110	10	Multiorgan failure; coagulation disturbances; multiple internal hemorrhages
42	30/M	14	Yes	186	67	16	Uremic coma; multiorgan failure; multiple internal hemorrhages
47§	41/M	Died before hospitalization	Yes	NR	NR	3	Renal failure; multiple internal hemorrhages, edema
48§	19/M	4	Yes	192	54	6	Renal, cardiovascular failure; RDS, DIC syndrome; bleedings in pituitary, adrenal gland, intestinum, etc.
56	35/F	4	Yes	410	49	6	Cardiovascular, renal, lung, liver failure; renal tubular necrosis; lung, brain edema
59	50/M	5	Yes	310	3	7	Renal,cardiovascular failure; RDS; multiple internal hemorrhages; pleurorrhea; lung, brain edema

*DIC, disseminated intravascular coagulation; GI, gastrointestinal; max, maximum; min, minimum; RDS, respiratory distress syndrome; NR, not reported.
†Reference range <96 μmol/L for female patients, <110 μmol/L for male patients.
‡Reference range 150–400 $\times 10^9$/L
§Patient no. 47 was the father in-law of patient no. 48; both lived in the same rural residence.

Conclusions

We have demonstrated the occurrence of human infections by Sochi virus and studied the clinical outcome for 62 patients. This virus is carried by the Black Sea field mouse (*A. ponticus*), which occurs naturally in the Transcaucasian region between the Black and Caspian Seas, including a part of southern European Russia. In anecdotal field studies in the coast region near Sochi, *A. ponticus* was the most abundant mouse species (71% of all trapped mice were identified as *A. ponticus*); moreover, 14% of trapped *A. ponticus* mice were serologically proven to be DOBV infected (*8*). This finding indicates that DOBV is the hantavirus indigenous in this geographic area and that *A. ponticus* mice are highly relevant as a hantavirus reservoir. All evidence from the natural virus reservoir, as well as serologic and molecular diagnostics of patients' serum, shows that the virus responsible for the infections is the DOBV genotype Sochi.

Most investigated patients found to be infected by Sochi virus exhibited a severe clinical course. With a calculated CFR of 14.5%, Sochi virus might be the most deadly hantavirus outside the Americas, where 35%–50% of hantavirus infections are fatal (*1*,*9*). Even Asian Hantaan virus is estimated to be less deadly; recent studies show CFRs of 1%–3% in China and South Korea, where Hantaan virus infections play an important role in HFRS morbidity (*10*,*11*). On the other hand, increased awareness in diagnostics, treatment, and prevention by local physicians and public health authorities is expected to improve survival rates for Sochi virus infections.

Among the related viruses of the DOBV species, Sochi virus seems to have the highest level of virulence, similar to Dobrava virus (carried by *A. flavicollis* mice), which has a CFR of up to 10%–12% (*12*,*13*). As shown in larger studies, disease caused by infection with the related Kurkino genotype (carried by the western lineage of *A. agrarius* mice) is associated with a CFR of only 0.3%–0.9% (*3*,*14*). These phylogenetically related viruses exert a quite different pathogenicity in humans.

Acknowledgments

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Dr. Kruger is the head of the Institute of Medical Virology, Charité–University Medicine Berlin. His research focuses on the molecular epidemiology and clinical relevance of emerging virus infections.

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Technical Appendix

Technical Appendix Table 1. Details of serodiagnostic data (reciprocal antibody titers) of the 62 patients*

Patient no.	No. serum samples	Sampling day after disease onset	IFA				ELISA				FRNT					
							PUUV		DOBV			DOBV				
			PUUV	DOBV	HTNV	SEOV	IgM	IgG	IgM	IgG	PUUV	Sochi	Kurkino	Dobrava	HTNV	SEOV
1	1,293	27	<16	1024	1024	512	<128	<128	16384	32768	<40	1280	640	640		
	1312	104	<16	2048	512	512	<128	<128	2048	8192	<40	1280	160	160	160	<80
2	1308	22	<16	2048	1024	1024	256	<128	4096	4096	<40	640	80		160	80
	1310	50	<16	8192	4096	4096					<40	1280	640	640	160	160
	4708†	5 y 9 m	64	2048	2048	2048					<40	2560	1280	2560	80	40
3	1307	15	<16	4096	2048	2048	<128	<128	8196	8196	<40	640	320	320	160	160
4	1335	17	64	8192	4096	4096	512	512	8192	16384	<40	1280	<80	80		
	1336	82	<32	4096	2048	1024	1024	512	4096	32768	<40	640	320	640	<80	160
	4,709†	5 y 9 m	64	4096	4096	2048					<40	5120	1280	2560	160	80
5	1339	11	<32	4096	2048	2048	256	<128	8192	8192	<40	320	80			
6	1642	3	<16	2048	1024	512	<128	<128	8196	2048						
	4716†	5 y 3 m	128	4096	4096	4096					<40	5120	1280	2560	160	160
7	1686	16	<16	2048	1024	512	<128	<128	2048	2048	<40	640	80	80		
8	4712	9	<16	512	256	256	<128	<128	2048	512						
9	3507	15	64	4096	2048	2048	<128	<128	8196	8196	<40	640	160	320		
	4713†	1 y 5 m	1024	8192	4095	4096					<40	20480	5120	10240	320	160
10	3830	13	128	8192	2048	2048	512	256	16384	8192	<40	640	160	640		
	4714†	1y 5m	512	16384	16384	8192					<40	20480	5120	5120	80	40
11	3692	30	64	4096	4096	2048	256	256	4096	16384	<40	1280	160	<160		
12	3693	10	32	2048	1024	1024	<128	<128	16384	8192						
	3694	18	64	2048	2048	2048					<40	640	<80	80		
13	3496	9	<16	4096	4096	2048	<128	<128	16384	16384						
14	3802	11	64	4096	2048	2048	<128	<128	8192	2048	<40	1280	320	640		
	4711†	1 y 6 m	512	8192	8192	4096					<40	20480	20480	20480	160	80
15	3778	10	64	8192	4096	4096	<128	<128	65536	8192						
16	3824	11	64	8192	8192	4096	<128	<128	16384	16384						
17	3813	4	64	8192	2048	2048	<128	<128	16384	16384						
	4710†	1 y 5 m	64	2048	2048	2048					<40	10240	5120	5120		
18	3861	16	256	8192	4096	4096	<128	<128	8192	2048	<40	640	640	640		

Patient no.	No. serum samples	Sampling day after disease onset	IFA				ELISA				FRNT					
							PUUV		DOBV			DOBV				
			PUUV	DOBV	HTNV	SEOV	IgM	IgG	IgM	IgG	PUUV	Sochi	Kurkino	Dobrava	HTNV	SEOV
	4715†	1 y 4 m	128	8192	8192	4096					<40	5120	2560	5120	80	40
19	3855	9	<16	4096	2048	2048	<128	<128	16384	2048						
20	4387	8	<16	512	256	256	<128	<128	1024	1024						
21	4471	2	<16	512	512	256	<128	<128	4096	1024						
22	4705	?	128	2048	2048	2048	128	<128	4096	4096	<40	640	640	640		
23	5049	7	<16	1024	512	512	<128	<128	4096	1024						
24	5065	8	<16	2048	2048	2048	<128	<128	8192	8192						
25	5066	8	<16	2048	1024	512	<128	<128	4096	8192						
26	5067	8	<16	1024	512	512	<128	<128	4096	1024						
27	5293	12	<16	4096	2048	2048	<128	<128	8192	8192						
28	5312	5	<16	1024	1024	1024	<128	<128	4096	1024						
29	6624	4	<16	4096	2048	2048	<128	<128	16384	4096						
30	6625	6	64	2048	2048	2048	<128	<128	4096	1024						
31	6627	7	64	4096	1024	1024	<128	<128	8192	8192						
	6627-2	73	64	4096	512	512					<40	2560	320	640	160	80
32	6623	13	32	4096	4096	2048	128	128	16384	8192						
33	6626-2	108	256	8192	2048	2048					<40	10240	1280	2560	160	160
34	6930	5	256	4096	2048	2048	<128	<128	8192	8192						
35	6845	4	<16	1024	512	256	<128	<128	8192	4096						
	6845-a	29	<32	1024	512	512					<40	1280	160	320	<40	<40
	6855-2	11 m	512	4096	1024	1024					<40	2560	640	640	80	80
36	6931	7	64	2048	512	512	<128	<128	4096	8192						
37	6918	13	<32	2048	1024	512	<128	<128	8192	16384						
	6918a	27	<32	2048	1024	512					<40	2560	320	1280	40	<40
	6931-2	1y	512	4096	1024	1024					<40	5120	1280	2560	160	160
38	4481	2 m	256	8192	8192	4096	<128	<128	2048	16384		2560	640	640	160	160
40	4482	9	512	8192	4096	2048	<128	<128	16384	16384	<40	2560	2560	2560	<40	<40
41	4483	9	1024	16384	8192	8192	512	256	16384	16384						
42	4484	15	64	8192	2048	2048	<128	<128	16384	32768	<40	320	80	80	<40	<40
43	4485	9	128	8192	4096	4096	256	256	32768	32768						
44		12	512	8192	4096	4096	512	512	16384	16384						
45	6876	6	<16	4096	4096	4096	<128	<128	16384	8192						
46	6878	11	1024	32000	4096	4096	512	256	32768	32768						
	6878-a	33	64	8192	4096	1024					<40	5120	640	2560	160	80
47	6880	3	<16	4096	2048	2048	<128	<128	4096	1024						
48	6929	4	<16	4096	4096	4096	<128	<128	16384	1024						
49	6628	19	<32	2048	1024	1024	<128	<128	4096	8192						
	6875	26	<32	1024	512	512					<40	2560	640	640	160	160
51	6882	17	64	4096	2048	2048	256	<128	8192	8192						

Patient no.	No. serum samples	Sampling day after disease onset	IFA				ELISA				FRNT					
							PUUV		DOBV			DOBV				
			PUUV	DOBV	HTNV	SEOV	IgM	IgG	IgM	IgG	PUUV	Sochi	Kurkino	Dobrava	HTNV	SEOV
	6882-a	34	64	4096	2048	1024					<40	1280	320	640	40	<40
52	6883	13	256	2048	1024	1024	<128	<128	8192	8192						
	6883-2	10 m	256	4096	2048	512					<40	2560	80	160	40	40
53	7105	10	256	4096	4096	2048	256	128	32768	32768						
54	7110	9	64	1024	1024	1024	<128	<128	2048	4096						
55	7111	5	64	1024	1024	512	<128	<128	8192	4096						
56	8385	4	<16	1024	512	512	<128	<128	8192	2048						
57	8386	8	64	1024	1024	512	256	128	4096	8192						
58	8380	6	32	2048	1024	1024	<128	<128	8192	8192						
59	8381	5	<16	128	64	64	<128	<128	1024	128						
60	8382	9	32	4096	4096	1024	<128	<128	32768	16384						
61	8383	16	<16	4096	4096	2048	<128	<128	16384	16384						
62	8384	24	64	4096	4096	2048	256	256	8192	32768						
63	8388	5	<16	4096	2048	2048	<128	<128	8192	8192						

*Patient serum samples were screened for hantavirus antibodies by indirect IFAs by using spot-slides containing a mixture of Vero-E6 cells infected by PUUV, DOBV, HTNV, and SEOV, as previously described (Dzagurova et al., Zh Mikrobiol Epidemiol Immunobiol, 2008; No.1: 12-16). Serum found to be positive was confirmed and further typed on IFA with “monovalent” spot-slides containing cells infected by only 1 of the mentioned viruses. Thereafter, IgG and IgM were determined by ELISAs on the basis of PUUV and DOBV antigens by established methods (Meisel et al., Clin Vaccine Immunol, 2006;13: 1349-57). For further serotyping, the neutralizing activity of serum was investigated by FRNT (Dzagurova et al., Zh Mikrobiol Epidemiol Immunobiol, 2008; No.1: 12-16), using Vero-E6 cells to propagate the following virus stocks; PUUV strain K-27/Ufa-85, DOBV (genotype Sochi) strain Ap1584/Sochi-01 (named Sochi/Ap), DOBV (genotype Kurkino) strain Aa1854/Lipetsk-02, DOBV (genotype Dobrava) strain Bel-1, HTNV strain P-88/ Khabarovsk-89, SEOV strain SR-11. DOBV, Dobrava-Belgrade virus; FRNT, focus-reduction neutralization assay; HTNV, Hantaan virus; IFA, immunofluorescence assay; PUUV, Puumala virus; SEOV, Seoul virus. Blank cells indicate testing not done.

†FRNT was performed with addition of 10% guinea pig blood serum (as complement).

Technical Appendix Table 2. Virus sequences and corresponding GenBank accession numbers used in the phylogenetic analyses

Virus species	Abbreviation	Strain name	GenBank accession no.	
			S segment	L segment
Dobrava-Belgrade virus	DOBV	10636/Ap	**KP878311**	**KP878308**
		10645/Ap	**KP878312**	**KP878309**
		10752/hu	**KP878313**	**KP878310**
		6882/hu	**–**	**KM192207**
		43/Ap	JF920151	**KM192209**
		79/Ap	JF920152	**KM192208**
		Sochi/hu	JF920150	JF920148
		Sochi/Ap	EU188449	EU188451
		GK/Ap	AF442622	–
		Krasnodar/hu	AF442623	–
		SK/Aa	AY961615	GU904039
		Esl29/Aa	AY533118	–
		Esl81/Aa	AY533120	–
		Kurk44/Aa	AJ131672	–
		Kurk/53/Aa	AJ131673	–
		Esl856/Aa	AJ269549	–
		Esl862/Aa	AJ269550	–
		Lipetsk/Aa	EU188452	EU188454
		GER/08/118/Aa	GQ205407	–
		GER/08/131/Af	GQ205408	–
		GER/07/293/Aa	GQ205401	–
		GER/07/607/Af	GQ205402	–
		GER/07/1064/Aa	GQ205404	–
		GER/05/239/Aa	GQ205405	–
		GER/05/477/Af	GQ205406	–
		Esl400/Af	AY168576	–
		AP9/Af	AJ410615	AJ410617
		AP13/Af	AJ410619	–
		Slo/Af	L41916	AJ009779
		Istanbul/H56	–	KF039740
		Saa/160V	AJ009773	AJ410618
		Saa/90Aa	AJ009775	–
Sangassou virus	SANGV	SA14	JQ082300	JQ082302
		SA22	JQ082303	–
Hantaan virus	HTNV	76–118	M14626	NC_005222
		SC-2	–	AY675354
		Z10	AF184987	–
		LR1	AF288294	–
		AH09	AF285264	–
		84FLi	AY017064	–
Thailand virus	THAIV	741	AB186420	–
		NR/Bi0017	AM397664	–
Seoul virus	SEOV	80–39	NC 005236	NC_005238
		Gou3	AF184988	–
		Z37	AF187082	–
		L99	AF488708	–
Puumala virus	PUUV	CG1820	–	M63194
Tula virus	TULV	Mo5302	–	NC_005226
Andes virus	ANDV	Chile-9717869	–	NC_003468
Sin Nombre virus	SNV	NM H10	–	NC_005217

*The nucleotide sequences generated in this study were based on PCRs amplifying a 390-nt conserved region within the L segment (Klempa et al. Emerg Infect Dis. 2006;12:838–40) or a 599-nt fragment of the S segment (Klempa et al., J Clin Microbiol, 2005;43:2756–63). Bold type indicates new sequences. Dashes indicate that no sequence information was available.

Technical Appendix Table 3. Classification criteria of clinical severity of hemorrhagic fever with renal syndrome*

Symptoms	Clinical course		
	Mild	Moderate	Severe
Duration of fever, d	3–4	5–6	>6
Systolic blood pressure, mm Hg	100	<100	<80
Hemorrhagic syndrome	Scleral, subcutaneous hemorrhages	Bleedings, not life threatening	Bleedings, life threatening
Duration of oliguria <500 mL/24 h	24–48 h	49-96 h	>96 h
Anuria >50 mL/24 h	–	–	Positive
Serum creatinine, μmol/L	Normal (<96/F, <110/M)	120–250	>250
Proteinuria, g/L	<1	<5	>5
Pulmonary edema	–	–	Positive
Cerebral edema	–	–	Positive
Kidney rupture	–	–	Positive

*Classification according to Russian standard criteria (Leshchinskaia et al., Vopr Virusol. 1990;35:42–5; Klempa et al., Emerg Infect Dis.2008;14:617–25). Dashes indicate that these symptoms were not observed.