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CASE STUDY

Two clinical cases of renal syndrome caused by Dobrava/Saaremaa hantaviruses imported to the Netherlands from Poland and Belarus, 2012–2014

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We report the rare event of two imported cases in the Netherlands presenting with renal syndrome caused by Dobrava (DOBV)/Saaremaa (SAAV) hantaviruses. DOBV/SAAV hantaviruses are not circulating in the Netherlands and their clinical manifestation is typically more severe than that of the endemic Puumala virus (PUUV). This report aims to increase awareness among healthcare professionals and diagnostic laboratories to consider different hantaviruses as a cause of renal failure.

Keywords: *hantavirus infections*; *Dobrava*; *Haemorrhagic fever with renal syndrome*; *rodent-borne*; *zoonosis*

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We report two cases of Dobrava (DOBV)/Saarenaa (SAAV) hantaviruses imported to the Netherlands from 2012 to 2014. Both patients presented with renal syndrome following travels to Belarus and Poland, respectively, where DOBV is known to be circulating. This report serves to create awareness among clinicians and diagnostic laboratories to consider different hemorrhagic fever-causing hantaviruses in patients with renal syndrome.

Case 1

In July 2012, a previously healthy 54-year-old Dutch woman was referred to the emergency department because of acute kidney injury. She suffered from confusion, persisting after a period with fever (up to 40°C), vomiting and oliguria 10 days before referral. Temperature (36.8°C) and blood pressure (145/87 mmHg) were normal upon admission but a slight abdominal pain and a slight edema at the ankles were noticed. Laboratory investigation showed a mild normocytic anemia with a Hb of 7.0 mmol/l ($n = 7.5–9.5$); acute renal failure with a serum creatinine of 810 µmol/l ($n = 55–90$); and elevated liver enzymes [ASAT 52 U/l (ULN <31), ALAT 236 U/l (ULN <34), LDH 444 U/l (ULN <247), alkaline phosphatase 165 U/l (ULN <115), γGT 61 mmol/l (ULN <38)]. The C-reactive protein of 13 mg/l and erythrocyte sedimentation rate of 53 mm/hr ($n = 0–20$) were modestly elevated. Ultrasonography showed two edematous kidneys and a normal liver. The patient had travelled to the Republic of Belarus on a bird-watching trip 3 weeks before the onset of symptoms. During her stay in a holiday home located in a woody area, she had cleaned the floor of dust and mouse

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droppings. Infection with a hantavirus was immediately suspected upon anamnesis and thus part of the initial differential diagnosis. Both IgM and IgG antibodies were detected by Puumala virus (PUUV) ELISA screening (Progen) on a serum sample taken 10 days upon onset of illness. Comparative serology using an immunofluorescent antibody mosaic assay (IIFA, Euroimmuun) demonstrated high titers of both IgM and IgG against Dobrava (DOBV) and Saaremaa (SAAV), but no detectable antibodies against PUUV (Table 1). The patient was treated with saline infusion upon which renal function improved; serum creatinine dropped to 174 μmol/l in 4 days and later completely normalized. At a follow-up visit 6 weeks later (52 days after onset of symptoms), convalescent serum was sent to the lab and was again tested positive for both DOBV and SAAV by comparative IFA (Table 1). Virus neutralization as described previously (1, 2) confirmed these results and showed a two-fold higher titer to DOBV than to SAAV. This indicated, but did not confirm, a DOBV infection.

Case 2

In November 2014, a previously healthy 26-year-old Polish seaman visited an outpatient clinic in the Netherlands with symptoms of diarrhea and vomiting since 4 days and fever since 2 days. There was no previous history of disease and no reports of further cases among the patients' crewmates. The ship was reported to navigate exclusively in northern European waters. Besides the travels on the ship, his travel history contained a family trip to the southwest of Poland 3 weeks before presentation, where he had visited a farm with chickens and pigs.

During physical examination, an acutely ill patient was observed; he was agitated and his movements were uncoordinated. He was disoriented in place, his consciousness was slow, and he had word finding difficulties in his own language. Physical examination revealed a temperature of 40°C, normal blood pressure (123/70), and tachycardia of 130 beats per minute. There was no evidence of neck stiffness; lymph nodes were palpable, occipital, and cervical; there was conjunctivitis in both eyes and petechiae on the thorax. Laboratory results showed a normal Hb of 10.9 mmol/L ($n = 8.5$–11), thrombocytopenia of 50×10^9 ($n = 150$–370×10^9) with leucocytes of 10.8×10^9 ($n = 3.5$–10×10^9). He had an elevated creatinine of 167 ($n = 65$–115 μmol/L) with an estimated glomerular filtration rate (eGFR) of 43 ($n > 60$ mL/min) and slightly elevated liver enzymes. Cerebrospinal fluid (CSF) examination showed no signs of (meningo-) encephalitis and the CT cerebrum showed no intracerebral pathology.

Serological testing for viral hepatitis, cytomegalovirus, Epstein-Barr virus, Mycoplasma, parvovirus B19, and adenovirus did not show any evidence for a recent infection.

Table 1. Hantavirus diagnostics performed in convalescent sera of two patients in the Netherlands with HFRS after travelling to Belarus and Poland, 2012–2014

			Patient 1		Patient 2	
			Day 10	Day 52	Day 3	Day 9
			After symptom onset		After symptom onset	
ELISA[a]	PUUV	IgM	**17.475**	**17.728**	**1.159**	**1.446**
		IgG	**11.96**	**12.104**	0.295	**2.392**
IFA[b]	DOBV	IgM	**4,000**	**1,000**	**100**	**2,000**
		IgG	**128,000**	**64,000**	**4,000**	**≥16,000**
	SAAV	IgM	**16,000**	**1,000**	**1,000**	**2,000**
		IgG	**4,000**	**64,000**	**4,000**	**≥16,000**
	PUUV	IgM	<100	<100	<100	<100
		IgG	<100	<100	<100	**100**
FRNT[c]	DOBV			**1:320**		**1:160**
	SAAV			**1:160**		**1:160**
	PUUV			<40		<40
PCR[d]	DOBV		Negative		**Positive**	
	PUUV		Negative		Negative	
	SEOV		Negative		Negative	

PUUV, Puumula virus; DOBV, Dobrava virus; SAAV, Saarema virus; SEOV, Seoul virus.

Bold figures indicate positivity. [a]Enzyme-linked immunosorbent assay (Progen), values <1.0 were considered negative (4)]. [b]Indirect immunofluorescence assay (Euroimmuun), titers <100 were considered negative (5). [c]Virus neutralization test, titers <40 were considered negative (1)]. [d]In-house real-time reverse transcriptase-polymerase chain reaction.

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Due to the renal dysfunction and travel history, an acute infection with a pathogenic hantavirus was considered as a differential diagnosis and ascertained in the acute serum sample taken upon admission. DOBV RNA was detected (Ct 32) using an internally controlled in-house real-time reverse transcriptase-polymerase chain reaction (RT-PCR, Table 2), in combination with one-step fast virus mastermix (Applied Biosystems, Bleiswijk, the Netherlands), according to manufacturer's protocol. IgM antibodies were detected in the acute serum sample by the PUUV ELISA (Progen) upon which a mosaic IIFA (Euroimmuun) was performed demonstrating the presence of IgM and IgG antibodies against DOBV and SAAV in both the acute serum and in a serum sample 9 days later (upon discharge). Only a borderline reactivity was observed for anti-PUUV IgG antibodies in the serum at Day 9 (Table 1).

Upon admission, the patients' renal function deteriorated but the patient was hemodynamically stable and improved neurologically. On Day 4, his renal impairment was most severe with an eGFR of 14 ($n > 60$ mL/min) and a creatinine level of 440 ($n = 65$–115 μmol/L). After 1 week of supportive care, his renal function started to improve slightly and the patient requested to be dismissed. A follow-up serum sample for hantavirus serology was taken at discharge, Day 9 after onset of symptoms. Virus neutralization on both sera confirmed the presence of neutralizing antibodies against both DOBV and SAAV but could not distinguish between both viruses based on a four-fold titer difference as the second serum was taken too early after onset of symptoms (1).

Background

Hantaviruses are enveloped viruses with a segmented negative-strand RNA genome belonging to the family *Bunyaviridae*. Within Europe, at least four hantavirus species cause infections in humans. Puumula (PUUV) and the closely related Dobrava-Belgrade virus (DOBV) and Saaremaa (SAAV) (6) are mostly reported, while recently the first cases of Seoul virus (SEOV) infection have been described (7, 8). The close relationship between DOBV and SAAV is reflected by a high level of serological crossreactivity between these viruses. A further hantavirus, Tula virus (TULV), has been detected in vole species and its rodent reservoir in several European countries (9). The pathogenicity of TULV in humans is considered to be low, but has been described in an immunocompromised patient (10).

The total recorded number of hantavirus infections in Europe has been steadily increasing during the past years; an average of 3,138 cases were recorded from 2000 to 2009 versus 1,671 in the period 1990–1999 (11). The highest number of reported cases is found in Finland, Sweden, Germany, France, and Belgium, but severe underdiagnosis is suspected in several other European countries (12). The majority of clinically apparent human hantavirus infections in Western Europe are caused by PUUV (6, 11). In the Netherlands, from 2008 to 2013, the number of yearly reported PUUV cases varied between 4 and 24 cases per year, but seroprevalence studies suggest that this number is an underestimation as well with a peak of 36 notified cases in 2014 (13–15). DOBV cases have thus far mostly been reported in the Balkan countries and the Alpe-Adrian region (16).

Each hantavirus is associated with a specific natural reservoir (17, 18). For the hantaviruses causing hemorrhagic fever with renal syndrome (HFRS), these include the *Apodemus*, *Rattus, Myodes*, and *Microtus* species. Hantaviruses infect humans primarily from aerosolized rodent excreta. Particular risk has been associated with opening, occupying, and cleaning structures, such as summer cottages, barns, and cellars, which have been infested by rodents. There is no specific vaccination or antiviral therapy in use and so infection should be avoided by not inhaling unventilated air in such structures, using personal protection such as surgical masks and by rodent control measures (12). As humans are generally considered dead-end hosts for hantaviruses, there is no risk of ongoing transmission of DOBV upon import of infected patients.

Diagnosis of HFRS/NE currently relies on serology. The viremic stage in hantavirus infections is short and diagnostic requests for HFRS/NE are often too late in the course of disease to enable a diagnosis by RT-PCR, outside of endemic areas. The most commonly used methods for verifying a hantavirus infection are indirect IgG- and IgM-enzyme-linked immunosorbent assays (ELISA), IgM capture ELISAs, or IIFAs. With these routine serology

Table 2. Primer and probes used for internally controlled DOBV real-time RT-PCR

	Name	Sequence 5′-3′	Conc (pmol/μl)	References
Dobrova	Dobrava -rev-TM	AGACATTCAGGAAGCAAATYAATGA	30	In-house
	Dobrava-fwd-short	GGTGGTTTAGGAYGTCACCTTAAGTG	45	
	Dobrava-probe-new	FAM-ACAACAACTAYCTACC[a]CAAAACAACAACACTACCTCA	10	
PDV	PDV fwd	CGGGTGCCTTTTACAAGAAC	30	(3)
	PDV rev	TTCTTTCCTCAACCTCGTCC	7.5	
	PDV probe	CY5-ATGCAAGGGCCAATT-MGB	10	

[a]BHQ1, internally coupled quencher. PDV, phocine distemper virus, used as an internal process control.

techniques, it is impossible to distinguish between hantavirus species with known crossreactivity such as seen between DOBV and SAAV (19, 20). Comparative virus neutralization tests are the gold standard to confirm an infection with a specific hantavirus species (21).

PUUV typically causes a mild form of HFRS called *Nephropathica epidemica* (NE). The course of NE/HFRS has been divided into febrile, hypotensive, oliguric, diuretic, and convalescent phases, but these phases are not always clinically evident (6). HFRS is characterized by fever and renal failure associated with hemorrhagic manifestations. In NE, renal failure and fever (above 38.5°C) dominate, and hemorrhagic signs are rare. The described case fatality rate of NE is low and varies from 0.1 to 0.4% (6). More recently, PUUV has been demonstrated as a cause of hantavirus pulmonary syndrome as well, which was previously thought to be caused solely by American hantaviruses (22). DOBV, in contrast to PUUV, is usually characterized by more severe HFRS, with a case fatality rate up to 12% (23). Although genetically resembling DOBV, clinical data indicate that SAAV infections are much less severe than DOBV (24–26).

During the acute phase of HFRS, mild neurological symptoms such as headache, vertigo, and nausea are common and direct PUUV infection of CNS has been demonstrated (27). Data on the occurrence of severe, potentially life-threatening neurological manifestations is rather scarce; since only a few cases of severe neurological manifestation in DOBV have been described (28–30).

Discussion and conclusion

Routine diagnostics of NE/HFRS in the Netherlands and the majority of northwestern European countries are based on serology and relies on the assumption that PUUV is the most common causative agent. Even during the first days of a hantavirus disease, IgM and usually even IgG antibodies are present. Extensive crossreactivity of antibodies to the various hantaviruses species circulating in Europe complicates interpretation of routine serology tests. However, the cases presented here illustrate that the crossreactivity of antibodies to PUUV and DOBV/SAAV is sometimes weak or completely absent (1, 21). This reflects their antigenic distance and also emphasizes the need for use of multiplex antigens (at least PUUV and DOBV/SAAV) to cover HFRS diagnostics within Europe. Possible SEOV infections will usually be detected by this panel as well, as SEOV is antigenically similar to DOBV (5) which is of importance seeing the increasing evidence for SEOV circulation in wild and pet rats in Europe (31). Definite serological diagnosis of a specific hantavirus infection can be established only by virus neutralization assay. The neutralizing antibody titer to the causative hantavirus should be four-fold higher in comparison to all other relevant hantaviruses. Alternatively, RT-PCR and subsequent sequencing can be performed on acute phase samples, although viral genome sequences can be detected in less than two-thirds of the acute serum samples of suspect cases (30, 32).

The two presented cases emphasize the challenge of definitely diagnosing a specific hantavirus infection based on serology, as both patients were reactive in common routine PUUV diagnostic tests and we could not unambiguously conclude whether infection was caused by DOBV or SAAV (although they are sometimes considered to be lineages of one hantavirus species). In the acute serum of the second patient, hantavirus genome was detected by our DOBV-specific in-house RT-PCR. This, in combination with the clinical signs (severe renal failure and neurological symptoms), made the diagnosis of a DOBV infection instead of a SAAV infection highly probable, although virus neutralizing antibodies were detected at the same end-point dilution for DOBV and SAAV (1:160).

The presented cases furthermore underscore that health care workers in regions where typically PUUV related, and possibly SEOV, infections may be detected should be aware of other circulating hantaviruses that may be imported to non-endemic regions. DOBV is known to cause severe and fatal HFRS and requires more intensified supportive care than the less severe PUUV- and SAAV-infected patients. For an adequate diagnosis, multiplex testing targeting different strains of hantavirus is required in clinical suspect cases. Recently published quality assessments have demonstrated that within Europe, the overall specificity and sensitivity of detecting hantavirus infection is acceptable but that the exact serotyping can be problematic with currently available serodiagnostic methods. Especially, the misclassification of DOBV cases by routine diagnostics based on ELISA warrants vigilance and more studies into the burden of DOBV infections in Europe (33, 34).

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References

1. Lundkvist A, Hukic M, Horling J, Gilljam M, Nichol S, Niklasson B. Puumala and Dobrava viruses cause hemorrhagic fever with renal syndrome in Bosnia-Herzegovina: evidence of highly cross-neutralizing antibody responses in early patient sera. J Med Virol 1997; 53: 51–9.
2. Lundkvist A, Scholander C, Niklasson B. Anti-idiotypic antibodies against Puumala virus glycoprotein-specific monoclonal antibodies inhibit virus infection in cell cultures. Arch Virol 1993; 132: 255–65.
3. Hoek RA, Paats MS, Pas SD, Bakker M, Hoogsteden HC, Boucher CA, et al. Incidence of viral respiratory pathogens causing exacerbations in adult cystic fibrosis patients. Scand J Infect Dis 2013; 45: 65–9.
4. Muyldermans A, Lagrou K, Patteet S, Van Esbroeck M, Van Ranst M, Saegeman V. Comparison of two automated immunoassays for the determination of Puumalavirus IgM and IgG. J Clin Virol 2014; 60: 165–7.
5. Lederer S, Lattwein E, Hanke M, Sonnenberg K, Stoecker W, Lundkvist A, et al. Indirect immunofluorescence assay for the simultaneous detection of antibodies against clinically important old and new world hantaviruses. PLoS Negl Trop Dis 2013; 7: e2157.
6. Vapalahti O, Mustonen J, Lundkvist A, Henttonen H, Plyusnin A, Vaheri A. Hantavirus infections in Europe. Lancet Infect Dis 2003; 3: 653–61.
7. Mace G, Feyeux C, Mollard N, Chantegret C, Audia S, Rebibou JM, et al. Severe Seoul hantavirus infection in a pregnant woman, France, October 2012. Euro Surveill 2013; 18: 20464.
8. Jameson LJ, Logue CH, Atkinson B, Baker N, Galbraith SE, Carroll MW, et al. The continued emergence of hantaviruses: isolation of a Seoul virus implicated in human disease, United Kingdom, October 2012. Euro Surveill 2013; 18: 4–7.
9. Schlegel M, Kindler E, Essbauer SS, Wolf R, Thiel J, Groschup MH, et al. Tula virus infections in the Eurasian water vole in Central Europe. Vector Borne Zoonotic Dis 2012; 12: 503–13.
10. Zelena H, Mrazek J, Kuhn T. Tula hantavirus infection in immunocompromised host, Czech Republic. Emerg Infect Dis 2013; 19: 1873–5.
11. Heyman P, Ceianu CS, Christova I, Tordo N, Beersma M, Joao Alves M, et al. A five-year perspective on the situation of haemorrhagic fever with renal syndrome and status of the hantavirus reservoirs in Europe, 2005–2010. Euro Surveill 2011; 16(36): pii: 19961.
12. Vaheri A, Henttonen H, Voutilainen L, Mustonen J, Sironen T, Vapalahti O. Hantavirus infections in Europe and their impact on public health. Rev Med Virol 2013; 23: 35–49.
13. Sane J, Reimerink J, Harms M, Bakker J, Mughini-Gras L, Schimmer B, et al. Human hantavirus infections in the Netherlands. Emerg Infect Dis 2014; 20: 2107–10.
14. Goeijenbier M, Hartskeerl RA, Reimerink J, Verner-Carlsson J, Wagenaar JF, Goris MG, et al. The hanta hunting study: underdiagnosis of Puumala hantavirus infections in symptomatic non-travelling leptospirosis-suspected patients in the Netherlands, in 2010 and April to November 2011. Euro Surveill 2014; 19(32): pii: 20878.
15. Rijksinstituut voor Volksgezondheid en Milieu. In 2014 meer mensen ziek door wilde knaagdieren. 2015. Available from: http://www.rivm.nl/Documenten_en_publicaties/Algemeen_Actueel/Nieuwsberichten/2015/In_2014_meer_mensen_ziek_door_wilde_knaagdieren [cited 17 December 2015].
16. Heyman P, Vaheri A, Lundkvist A, Avsic-Zupanc T. Hantavirus infections in Europe: from virus carriers to a major public-health problem. Expert Rev Anti Infect Ther 2009; 7: 205–17.
17. Schmaljohn C, Hjelle B. Hantaviruses: a global disease problem. Emerg Infect Dis 1997; 3(2): 95–104.
18. Plyusnin A, Sironen T. Evolution of hantaviruses: co-speciation with reservoir hosts for more than 100 MYR. Virus Res 2014; 187: 22–6.
19. Klempa B, Stanko M, Labuda M, Ulrich R, Meisel H, Kruger DH. Central European Dobrava Hantavirus isolate from a striped field mouse (Apodemus agrarius). J Clin Microbiol 2005; 43: 2756–63.
20. Klempa B, Tkachenko EA, Dzagurova TK, Yunicheva YV, Morozov VG, Okulova NM, et al. Hemorrhagic fever with renal syndrome caused by 2 lineages of Dobrava hantavirus, Russia. Emerg Infect Dis 2008; 14: 617–25.
21. Sjolander KB, Golovljova I, Vasilenko V, Plyusnin A, Lundkvist A. Serological divergence of Dobrava and Saaremaa hantaviruses: evidence for two distinct serotypes. Epidemiol Infect 2002; 128: 99–103.
22. Rasmuson J, Andersson C, Norrman E, Haney M, Evander J, Ahlm C. Time to revise the paradigm of hantavirus syndromes? Hantavirus pulmonary syndrome caused by European hantavirus. Eur J Clin Microbiol Infect Dis 2011; 30: 685–90.
23. Papa A. Dobrava-Belgrade virus: phylogeny, epidemiology, disease. Antiviral Res 2012; 95: 104–17.
24. Plyusnin A, Vaheri A, Lundkvist A. Saaremaa hantavirus should not be confused with its dangerous relative, Dobrava virus. J Clin Microbiol 2006; 44: 1608–9; author reply 1609–11.
25. Nemirov K, Andersen HK, Leirs H, Henttonen H, Vaheri A, Lundkvist A, et al. Saaremaa hantavirus in Denmark. J Clin Virol 2004; 30: 254–7.
26. Golovljova I, Sjolander KB, Lindegren G, Vene S, Vasilenko V, Plyusnin A, et al. Hantaviruses in Estonia. J Med Virol 2002; 68: 589–98.
27. Mahonen SM, Sironen T, Vapalahti O, Paakko E, Hautala N, Ilonen J, et al. Puumala virus RNA in cerebrospinal fluid in a patient with uncomplicated nephropathia epidemica. J Clin Virol 2007; 40: 248–51.
28. Cerar D, Avsic-Zupanc T, Jereb M, Strle F. Case report: severe neurological manifestation of Dobrava hantavirus infection. J Med Virol 2007; 79: 1841–3.
29. Avsic Zupanc T, Korva M, Markotic A. HFRS and hantaviruses in the Balkans/South-East Europe. Virus Res 2014; 187: 27–33.
30. Markotic A, Nichol ST, Kuzman I, Sanchez AJ, Ksiazek TG, Gagro A, et al. Characteristics of Puumala and Dobrava infections in Croatia. J Med Virol 2002; 66: 542–51.
31. Verner-Carlsson J, Lohmus M, Sundstrom K, Strand TM, Verkerk M, Reusken C, et al. First evidence of Seoul hantavirus in the wild rat population in the Netherlands. Infect Ecol Epidemiol 2015; 5: 27215, doi: http://dx.doi.org/10.3402/iee.v5.27215.
32. Plyusnin A, Mustonen J, Asikainen K, Plyusnina A, Niemimaa J, Henttonen H, et al. Analysis of puumala hantavirus genome in patients with nephropathia epidemica and rodent carriers from the sites of infection. J Med Virol 1999; 59: 397–405.
33. Hofmann J, Grunert HP, Donoso-Mantke O, Zeichhardt H, Kruger DH. Does proficiency testing improve the quality of hantavirus serodiagnostics? Experiences with INSTAND EQA schemes. Int J Med Microbiol 2015; 305: 607–11.
34. Escadafal C, Avsic-Zupanc T, Vapalahti O, Niklasson B, Teichmann A, Niedrig M, et al. Second external quality assurance study for the serological diagnosis of hantaviruses in Europe. PLoS Negl Trop Dis 2012; 6: e1607.