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Peste des Petits Ruminants Virus in Vulnerable Wild Small Ruminants, Iran, 2014–2016

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In 2014–2016, >1,000 wild goats and sheep in 4 northern and central provinces of Iran died from peste des petits ruminants virus (PPRV) infection. Partial nucleoprotein sequencing of PPRV from 3 animals showed a close relationship to lineage 4 strains from China. Control measures are needed to preserve vulnerable ruminant populations.

Peste des petits ruminants virus (PPRV; family *Paramyxoviridae,* genus *Morbillivirus*) causes a highly contagious disease with a high death rate in wild and domestic small ruminants. Four PPRV lineages (L1–L4) exist in Africa and Asia (*1*). The disease was initially recorded in Iran in 1995 (*2*) and subsequently spread throughout the country (*3*). PPRV-L4 infections are endemic in Iran and several neighboring countries (*4*,*5*).

Wild goats (*Capra aegagrus*) and sheep (*Ovis orientalis*), which have become extinct in several West Asia countries, are considered vulnerable species in Iran (*6*,*7*). Although PPRV-associated outbreaks among these ruminants have been suspected since 2000, the virus was not isolated or characterized at that time. In 2001, at least 1,500 wild goats and gazelles (*Gazella subgutturosa*) with clinical signs similar to those caused by PPRV infection died in Kavir National Park (Figure; online Technical Appendix Table, https://wwwnc.cdc.gov/EID/article/23/4/16-1218-Techapp1.pdf). An estimated 25%–40% of the wild goat population in the park was deemed lost as a result of the disease. In 2011, PPRV was the suspected cause of 550–700 deaths among wild sheep in Sarigol National Park (Figure); laboratory investigations using conventional reverse transcription PCR (RT-PCR) confirmed PPRV infection in several dead animals (Iran veterinary organization, pers. comm., 2011 Sep 26).

Beginning in September 2014, park rangers reported and field investigations substantiated mass deaths among wild goats in Bamou National Park (Figure). Clinical signs in affected animals were similar to those reported in wild small ruminants in 2011, and samples we tested from 5 dead goats were positive for PPRV by RT-PCR (online Technical Appendix). In April 2015, a new outbreak started in Haftad Qolleh Arak (Figure) and continued until mid-May, resulting in the death of 428 wild goats and 30 wild sheep. Three more outbreaks occurred in 2015: the first started in August in Kharmaneh-sar Tarom; the second in September in the Alamout Protected Area, 150 km from the previous outbreak in Kharmaneh-sar Tarom; and the third in November in the Taleghan Protected Area, 100 km away from the previous outbreak in Alamout Protected Area (Figure; online Technical Appendix Table).

The last reported outbreak started in April 2016 in Khojir, a national park close to a dam that serves as a water source for wild animals (Figure). In 2015, a total of 110 wild goats and sheep were counted in the park, and by May, 1, 2016, ≈85 were found dead (online Technical Appendix Table).

We detected PPRV genome in 6 oral swab samples and 7 blood and lymph node samples from dead ruminants by using conventional RT-PCR and in 3 oral swab samples by using real-time RT-PCR (quantification cycles 31–34) (online Technical Appendix). In addition, we performed partial nucleocapsid gene sequencing of 3 PPRV isolates from 2015; results showed 100% pairwise nt identity among the isolates (online Technical Appendix). The strains shared highest nt identity (99.4%) with PPRV-L4 strains that were circulating in domestic or wild small ruminants in northwestern and southeastern China during 2013–2015 (*8*) (online Technical Appendix Figure); they were more distantly related to PPRV-L4 strains previously reported from outbreaks in Iran and neighboring countries (*9*,*10*).

Field investigations and laboratory analyses indicated that PPRV was the cause of mass die-offs of wild goats and sheep during 2014–2016 in several national parks in Iran. A risk assessment of PPRV infection in several developing countries in Africa and the Middle East and on the Indian Peninsula indicated that 63% of small ruminant populations

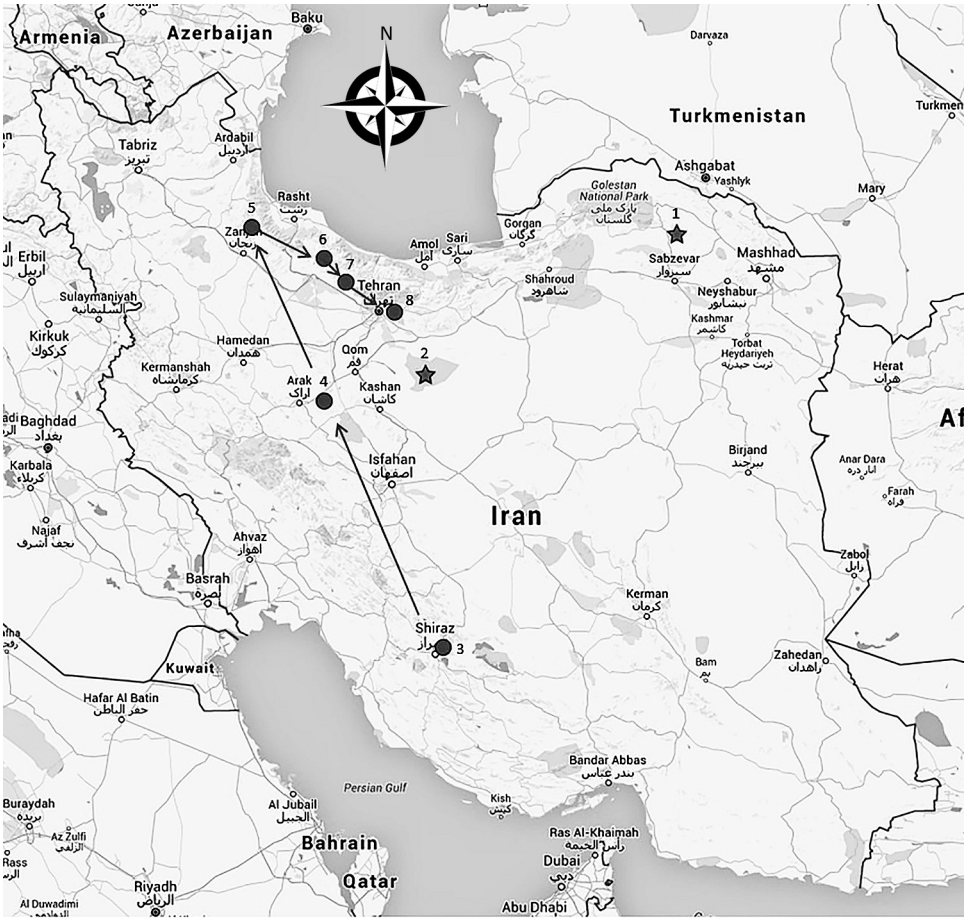

Figure. Geographic distribution of peste des petits ruminants virus outbreaks in Iran since 2000. Stars indicate outbreaks that occurred in 2000 and 2011; circles indicate outbreaks investigated during this study in 2014–2016. Arrows indicate chronologic order of the outbreaks: 1, Sarigol National Park; 2, Kavir National Park; 3, Bamou National Park; 4, Haftad-qolleh; 5, Kharmaneh Sar Tarom; 6, Alamout Protected Area; 7, Taleghan Protected Area; 8, Khojir National Park. Map generated using Google Maps (interactive map available at https://www.google.com/maps/d/viewer?mid=1GsluO7SZ2z_SBUawdPHsDF6s7ww). Details on the number of animals and dates of outbreaks are available in the online Technical Appendix Table (https://wwwnc.cdc.gov/EID/article/23/4/16-1218-Techapp1.pdf).

are at risk for infection (*4*). Legal and illegal movement of domestic small ruminants into wildlife territories over short and long distances, within and across borders, increases the possibility of transmission of various pathogens, including PPRV, to wild small ruminants, which may threaten vulnerable species. Transboundary circulation between China and Kazakhstan was recently shown for PPRV strains closely related to the PPRV Iran/2015 strains, suggesting that these closely related strains have been circulating in central and western Asia for a few years (*5*).

Clinical signs similar to those caused by PPRV infection were observed in domestic small ruminants in villages around the Kharmaneh-sar Tarom region before deaths were noted among wild goats in the area, and the samples collected from domestic animals tested positive for PPRV. It is unknown whether PPRV-infected wild small ruminants may contribute to PPRV spread by spillback to domestic small ruminants.

Comprehensive field studies of PPRV infection in domestic and wild small ruminants are necessary to evaluate the occurrence and origin of PPRV infections and of different PPRV strains in domestic and wild small ruminants in Iran. Emerging PPRVs can potentially spread to all susceptible small ruminant populations in the region and cause extinction of local subpopulations. Furthermore, control measures, such as vaccination against PPRV and movement control of domestic small ruminants around protected areas, would facilitate the preservation of vulnerable wild small ruminant populations and reduce the economic effect of PPRV infection on small ruminant production in affected regions.

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Discussion of Average versus Extreme Case Severity in Pandemic Risk Communications

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To investigate determinants of the public's perceptions of disease threat, in 2015 we conducted a randomized survey experiment in the Netherlands. Adults who read a mock news article describing average or extreme outcomes from a hypothetical influenza pandemic were more influenced by average than by extreme case information. Presenting both types of information simultaneously appeared counterproductive.

When pandemics strike, clear and timely communication is essential to raising public awareness of disease threat and motivating preventive behaviors (*1*). Yet, in most pandemics, the experience of affected persons is heterogeneous: a subset of persons have severe symptoms or sequelae, whereas most affected persons have much milder symptoms or sequelae. This heterogeneity creates a dilemma: Should communications about new infectious disease threats emphasize the character and severity of modal cases, which represents what most persons will experience, or should they focus on the severity of extreme cases to make clear the potential threat, even if that threat is highly unlikely? Both types of information are clearly important. Yet, risk messages are inherently difficult to understand, and providing multiple types of information simultaneously might undermine the public's understanding of a threat. Simplicity of message enables communications to stick with target audiences, and limiting communications to fewer, clearly contextualized, issues can increase efficacy (*2*,*3*).

To begin to address this communications dilemma, during 2015 we conducted a randomized survey experiment with adult residents of the Netherlands who participate in an online panel administered by Survey Sampling International (https://www.surveysampling.com/). We established quotas for age and sex that approximated the distributions of these characteristics in the population of the Netherlands (online Technical Appendix, https://wwwnc.cdc.gov/EID/article/23/4/16-1600-Techapp1.pdf). Upon completing the survey, participants received modest prizes.

Participants read a mock news article about a new pandemic (referred to as H7N3 influenza) spreading within the Netherlands. We randomly varied how the article discussed the average case severity, which was 1) not discussed, 2) described as mild (moderate fever and cough; generally goes away by itself), or 3) described as moderately severe (high fever, cough, vomiting; generally requires intravenous medication and hospitalization). We also independently varied the description of extreme cases, which were 1) not discussed, 2) described as (relatively) mild (requiring 1–2 days of hospitalization because of difficulty breathing, dizziness, and persistent coughing), or 3) described as moderately severe (requiring hospitalization [and causing 1 death] because of difficulty breathing, dizziness, severe coughing, and fluid in the lungs). This randomization resulted in a 3 × 3 between-subjects factorial design. Following guidelines for effective health messages (*4*), all articles

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Technical Appendix

Materials and Methods

Within the framework of outbreak investigation, a standard operating protocol of field epidemiologic data collection, post-mortem examination of carcasses, sample collection and submission, and laboratory analyses were carried out in this study.

RNA was extracted from oral swabs, tissue (lymph node and spleen) samples and whole blood samples obtained from more than 22 dead wild goats/sheep (found during last six outbreaks between September 2014 and April 2016, Table 1) using RNeasy® Mini Kit (Qiagen) and QIAmp® Viral RNA Mini Kit (Qiagen).

The samples were analyzed for PPRV-RNA using conventional RT-PCR (*1*) and real-time RT-PCR (SensiFAST Probe No-ROX Kit, Bioline) based on a published PCR-assay (*2*) that detects partial nucleocapsid protein (N) of PPRV. PPRV genome was detected in 6 oral swabs and 7 blood and lymph node samples using conventional RT-PCR and later in 3 oral swabs using real-time RT-PCR (quantification cycle (C_q) values: 31 to 34).

Partial PPRV nucleocapsid (N) sequences of 3 real-time RT-PCR-positive oral swabs of a length between 948 and 1097 bp were obtained with published (*3*–*5*) and unpublished (available on request) forward and reverse primers and used for sequencing. A pairwise identity of 100% was found between the three PPRV-RNA positive samples. Since only part of the PPRV genomes was sequenced, we cannot exclude that the three samples may contained three different, but very closely related PPRV strains. Three representative sequences was compared to a selection of PPRV sequences available in the GenBank (http://www.ncbi.nlm.nih.gov/genbank)

and used for phylogenetic analysis (Figure 1). The representative sequence is available with GenBank accession number KY550670.

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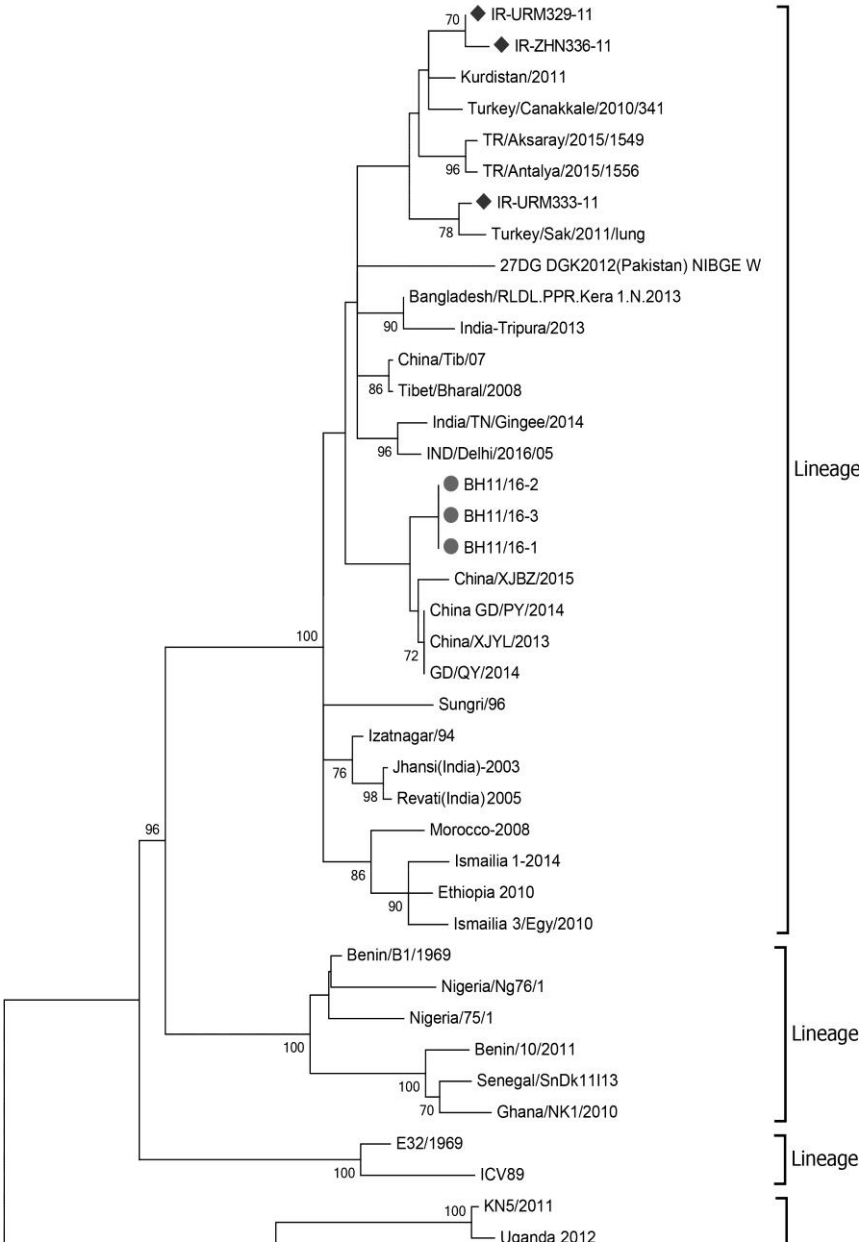
Technical Appendix Table. Location and number of dead wild goats and wild sheep associated with PPRV outbreaks reported in domestic small ruminants in Iran since 2000*

Year	Month	Outbreak region	Total no. dead	No. wild goats	No. wild sheep	No. gazelle
2001	May–Jun	Kavir National Park[1]	1,500–2,500	×		×
2011	Aug/Sep	Sarigol National Park[2]	550–700	–	550–700	–
2014	Sep–Nov	Bamou National Park[3]	400	×	×	–
2015	Apr/May	Haftad-qolleh[4]	458	428	30	–
2015	Aug/Sep	Kharmaneh sar tarom[5]	126	126	–	–
2015	Sep–Nov	Alamout Protected Area[6]	30	30	–	–
2015	Nov–Dec	Taleghan Protected Area[7]	204	204	–	–
2016	Mar–Apr	Khojir National Park[8]	85	85	–	–

*x, no. of dead species not exactly defined; –, dead species not reported; [1–8], numbers correspond to outbreak areas on the map in main text.



Technical Appendix Figure. Phylogenetic tree of Peste des Petits Ruminants viruses (PPRV) lineages 1 to 4 based on the N gene, including the "PPRV Iran/2015" strains (red circle marked) obtained from three wild goats in this study (laboratory submission numbers BH11/16–1 to –3) and isolates previously reported from PPRV outbreaks in Iran (blue marked). The closest relationship of PPRV Iran/2015 was found with PPRV strains from the provinces Xinjiang (China/XJBZ/2015 and China/XJYL/2013) and Guangdong (China GD/PY/2014 and GD/QY/2014) in northwestern and southeastern China, respectively. The tree was constructed using maximum-likelihood approach (MEGA 6.0 software) based on the GTR+I+G model. Numbers indicate the bootstrap values of 1000 replicates (only values >75% are shown) and the scale bar nucleotide substitutions per site.