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SEROSURVEILLANCE FOR FOOT-AND-MOUTH DISEASE IN MONGOLIAN GAZELLES (*PROCAPRA GUTTUROSA*) AND LIVESTOCK ON THE EASTERN STEPPE OF MONGOLIA

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ABSTRACT: Foot-and-mouth disease (FMD) is a highly contagious, viral disease that affects most ruminant and porcine species, and periodic outbreaks on Mongolia's Eastern Steppe affect Mongolian gazelles (*Procapra gutturosa*) and livestock. During 2005–08, we collected sera from 36 and 57 calf and adult gazelles, respectively, and from adult domestic animals sympatric with the gazelles, including 138 sheep (*Ovis aries*), 140 goats (*Capra aegagrus hircus*), 139 Bactrian camels (*Camelus bactrianus*), and 138 cattle (*Bos taurus*). Our goal was to determine whether the prevalence of the antibody to foot-and-mouth disease virus (FMDV) in gazelles declined relative to previous estimates in the absence of FMD outbreaks. Overall, 2.0% (95% CI 0.7–3.3%, $n=555$) of the four livestock species were antibody-positive for nonstructural proteins of FMDV (FMDV-NS), whereas 30.3% (95% CI 26.5–34.1%, $n=555$) had antibodies for structural proteins (i.e., vaccination-derived antibodies). Seven of 57 free-ranging gazelle calves (7.5%, 95% CI 1.6–12.4%) were FMDV-NS positive. None of 36 adult gazelles sampled in 2008 were antibody-positive for exposure to FMDV, indicating a significant decline ($\chi^2=18.99$; $P<0.001$; df$=1$) in antibody prevalence among gazelles from the same area during a livestock outbreak in 2001. The episodic nature of FMD outbreaks on the Eastern Steppe, Mongolia, with evidence of FMDV exposure in gazelles only during or following concurrent outbreaks in livestock, suggests that FMDV may spill over into the gazelle population during livestock outbreaks and that successful control of FMD on the Eastern Steppe requires a focus on control in livestock populations through vaccination.

Key words: Eastern Steppe, foot-and-mouth disease, Mongolia, Mongolian gazelle, *Procapra gutturosa*.

INTRODUCTION

Foot-and-mouth disease (FMD) is a highly contagious, acute, viral disease that affects most ruminant and porcine species. Given that one-third of the human population of Mongolia depends directly on livestock production for subsistence and a further one-quarter depends on them indirectly (Zahler et al. 2007), past outbreaks of FMD have caused severe disruptions to Mongolia's pastoral economy. Furthermore, FMD directly threatens the long-term persistence of the Mongolian gazelle (*Procapra gutturosa*), a keystone species on the Mongolian Eastern Steppe, directly, through morbidity and mortality, and indirectly, through certain disease-management actions aimed at them (Sokolov and Lushchekina, 1997; Nyamsuren et al., 2006). Mongolian gazelles share the range with domestic livestock, and there is concern that gazelles and other wildlife species may form a reservoir for foot-and-mouth disease virus (FMDV). Consequently, there is a need to understand the potential role of Mongolian gazelles as a reservoir for FMDV on the Eastern Steppe of Mongolia to aid in the design and implementation of disease-management programs. Our goal was to investigate the potential role of Mongolian gazelle in epizootics of FMD in the Eastern Steppe of Mongolia. Specifically, we used these data, and a review of previous outbreaks in Mongolia, to answer the question "given that FMD had not been reported on the Eastern Steppe of Mongolia for 1–4 yr previous to this study, did FMD antibody prevalence decline relative to previously

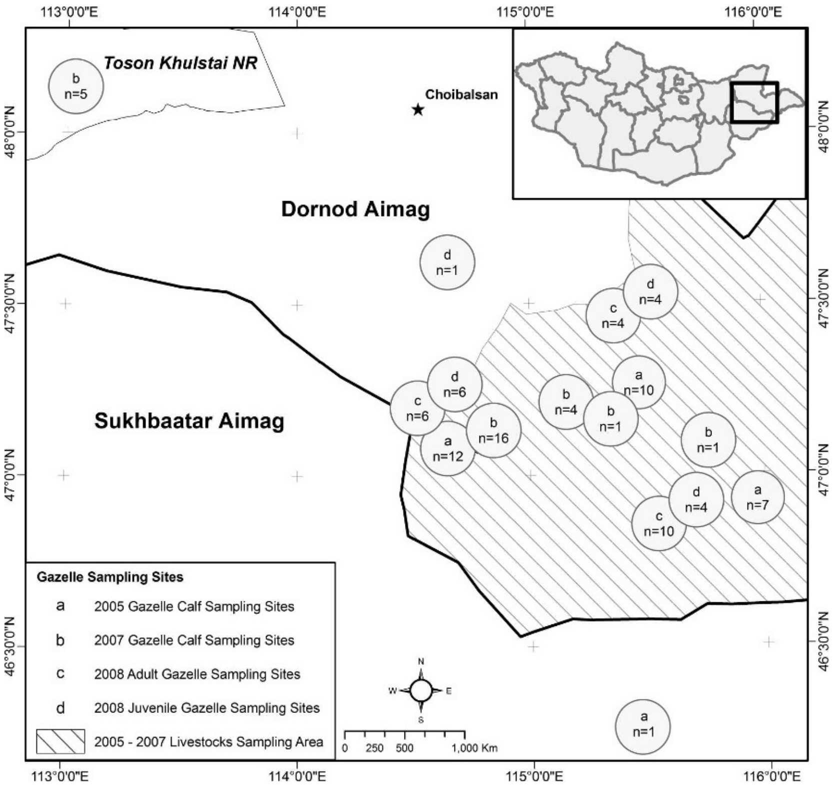

FIGURE 1. Map of livestock and Mongolian gazelle (*Procapra gutturosa*) sample collection sites from Dornod province of Mongolia. Livestock sampling site is marked as a polygon because herders move around at different seasons. Sampling years are provided for livestock and Mongolian gazelle sampling sites.

reported estimates from 2001, a year in which gazelles were sampled during a livestock outbreak?" We focused on FMDV type O (FMDV-O) as the prominent type identified in Mongolia since FMD reemergence in Mongolia in 2000, but other serotypes, such as Asia-1, C, and A, were screened for as well.

MATERIALS AND METHODS

Field sampling was conducted in Dornod Province, Mongolia, during 2005–08 (48°N, 114°E; Fig. 1). Gazelle and livestock serum samples were collected from 57 wild Mongolian gazelle calves, 36 adult Mongolian gazelles, and 555 adult domestic animals syntopic with the Mongolian gazelle, including 138 sheep (*Ovis aries*), 140 goats (*Capra aegagrus hircus*), 139 Bactrian camels (*Camelus bactrianus*), and 138 cattle (*Bos taurus*).

Domestic livestock samples were collected in Ehen Hudag of Matad soum from Dornod province, Mongolia (Fig. 1), and Mongolian gazelle samples were collected within an approximately 100-km radius from Matad soum, Mongolia. Domestic livestock samples were collected monthly from March through December 2005 (excluding October), January–February 2006, and May–July 2007. Mongolian gazelle calf samples were collected in June 2005 and June 2007, and adult gazelle samples were collected in September 2008. In 2005 and 2007, we captured gazelle calves ranging from 30 min to 2 days old while they were hiding in tall grass for protection and not yet able to run. Age of the calves was determined based on umbilical cord healing, dryness of hair, and agility. Most calves had nursed, and it was assumed that they had acquired maternal colostral immunity at the time of capture. Adult gazelles were captured by a team of 17 people using drive nets for live capture and were released in <1 hr.

Blood samples were collected using 9-ml vacuum tubes. The amount of whole blood collected (5–15 ml) varied by age and physical condition of the animal. Collected blood samples were kept at ambient temperature for 10 min, followed by serum harvesting by centrifuge. In the field, harvested sera were kept cool in summer and prevented from freezing in winter and were later transferred to a −20 freezer at the veterinary laboratory.

All harvested sera from gazelle calves (n=57), adult gazelles (n=16 juveniles and n=20 adults), and domestic animals (n=555) were first tested at the local immunology laboratory of the Mongolian Institute of Veterinary Medicine (Ulaanbaatar, Mongolia) to determine the presence of antibodies to FMDV-O using an enzyme-linked immunosorbent assay (ELISA; Yondondorj et al., 2006). Additional screening to differentiate exposure to nonstructural proteins of FMDV (FMDV-NS; i.e., natural exposure to wild-type virus) from exposure to structural proteins (vaccinated animals) was performed using Cedi Diagnostics FMDV-NS test kit (Prionics Lelystad BV, Lelystad, The Netherlands). All samples from gazelle calves and adult gazelles and all FMDV-NS–positive livestock samples (excluding one sheep and one goat sample that were mistakenly left out of the shipment) were sent for confirmation testing to the US Department of Agriculture (USDA), Foreign Animal Disease Diagnostic Laboratory (USDA-FADDL, Plum Island, New York, USA). The following tests were conducted at Plum Island: 3ABC ELISA (to detect the polypeptide 3ABC), virus infection–associated antigen (VIAA), and serotyping against O, Asia-1, A, and C serotypes using tissue culture virus neutralization (TC-VN) tests. We considered samples found positive at either laboratory by FMDV-NS, 3ABC ELISA, with a cutoff value ≥50% and VIAA with titer ≥32 as being positive. Unfortunately, FMDV serotyping at Plum Island of the gazelle calf samples collected in 2007 was not possible because of insufficient serum quantity.

Antibody prevalence in serum of adult gazelles (n=36, sampled in 2008) were compared with prevalence estimates (67%, n=33) from adult gazelles sampled in 2001 during an outbreak in livestock (Nyamsuren et al., 2006) using a χ^2 test of goodness of fit and independence (Preacher, 2001) to see whether there was a temporal change in FMDV antibody prevalence in the gazelle population in the absence of FMD outbreaks in livestock.

RESULTS

Overall, 1.9% (95% CI 1.1–3.5%, n=555) of the four livestock species had detectable antibody to FMDV-NS, whereas 23.2% (95% CI 20.3–26.5%, n=555) had antibodies for structural proteins (likely from vaccination). Seven of 57 free-ranging gazelle calves (10.9%, 95% CI 5.4–20.9%) had FMDV-NS antibody (Table 1). The presence of FMDV-NS antibodies in the serum of newborn gazelles (<2 days old) likely indicates the serologic status of the mother (Stone et al. 1960; Graves 1963) from a previous outbreak. The FMDV neutralization test results showed that antibodies to serotype O were present in three of seven gazelle calves. Results of FMDV neutralization test in livestock showed exposure to serotype O in one camel and four cattle (that were negative to the FMDV-NS tests and likely showed reaction because of the vaccine). Additionally two cattle had antibodies reacting to serotypes O, Asia-1, A, and C; four more cattle to serotypes O and Asia-1; and one cow to serotypes O, Asia-1, and A. All adult gazelles (16 juveniles and 20 adults) sampled in 2008 were negative for FMDV exposure. The χ^2 test results (χ^2=18.99; P<0.001; df=1) show the antibody prevalence in adult gazelles was significantly lower in 2008 compared with the 2001 study.

DISCUSSION

For logistic reasons, we relied on serum collected from newborn calves in 2005 and 2007. The presence of FMDV-NS antibodies in the serum of newborn gazelles likely indicates maternal antibodies to FMDV, given the young age of the calves (<2 days). We make this assumption based on studies demonstrating that antibody status of the calves of domestic cattle reflects that of the mother (Graves, 1963; Stone and DeLay, 1960). This maternal antibody may be acquired via the consumption of colostrums, within 30 min of birth. In addition, buffalo (*Syncerus caffer*) calves have maternal antibodies for the first 3–8 mo of life (Bastos et al., 2000).

Our goal was to investigate the potential

TABLE 1. Prevalence of antibody to foot-and-mouth disease virus type O (FMDV-O) in Mongolian gazelles (*Procapra gutturosa*), sheep (*Ovis aries*), goats (*Capra aegagrus hircus*), Bactrian camels (*Camelus bactrianus*), and cattle (*Bos taurus*) on the Eastern Steppe of Mongolia, 2005–07. Antibody prevalence due to vaccination (FMDV-O vaccinated) and natural infection with wild virus (FMDV-NS) are provided separately. Percentages in parentheses are 95% adjusted Wald confidence intervals (Agresti and Coull, 1998).

Species	Year	Number tested	FMDV-O vaccinated	% antibody-positive (CI)	FMDV-NS	% antibody-positive (CI)
Camel	2005	89	9	10 (3–17)	1	1.1 (0–5)
	2006	20	0	0 (0–12)	0	0 (0–12)
	2007	30	11	37 (19–54)	0	0 (0–9)
Camel total		139	20	14.4 (8.4–20.4)	1	0.7 (0–3.1)
Cattle	2005	89	34	38 (28–48)	4	4.5 (0–9.6)
	2006	19	4	21 (1–41)	0	0 (0–13)
	2007	30	23	77 (61–93)	3	10 (0–23)
Cattle total		138	61	44.2 (35.9–52.5)	7	5.1 (1–9.2)
Goat	2005	90	28	31 (22–41)	0	0 (0–3)
	2006	20	4	20 (1–39)	1	5 (0–19.5)
	2007	30	11	37 (19–54)	0	0 (0–8)
Goat total		140	43	30.7 (23–38.4)	1	0.7 (0–3.1)
Sheep	2005	88	29	33 (23–43)	0	0 (0–3)
	2006	20	1	5 (0–20)	1	5 (0–20)
	2007	30	14	47 (29–65)	1	3 (0–13)
Sheep total		138	44	31.9 (24.1–39.7)	2	1.4 (0–4.2)
Livestock Total		555	168	30.3 (26.5–34.1)	11	2 (0.7–3.3)
Mongolian gazelle, calves	2005	30	0	0 (0–8)	4	13 (0–26.9)
	2006	0	—	—	—	—
	2007	27	0	0 (0–9)	3	11 (0–25)
Adults	2008	36	0	0 (0–7)	0	0 (0–7)
Gazelle total		93	0	0 (0–2)	7	8 (2–12)

role of Mongolian gazelle in epizootic FMD in the Eastern Steppe of Mongolia, specifically to determine whether FMDV antibody prevalence in gazelles declined in the absence of ongoing outbreaks of FMD in livestock and whether FMDV antibody prevalence in gazelle reflects the dynamics in livestock. The prevalence of antibody to FMDV in gazelles declined between 2001 (during an active outbreak of FMD) and 2008, when no outbreaks of FMD were detected on the Eastern Steppe of Mongolia subsequent to February, 2004 (Table 2). This study, along with previous work in Mongolia, also demonstrated that patterns of antibody prevalence in gazelle reflect dynamics of FMD in livestock across the Eastern Steppe of Mongolia: 0% prevalence during outbreak-free years in livestock, 1998–99 (Deem et al., 2001, this study); 67% prevalence during a concurrent FMD outbreak in livestock in 2001 (Nyamsuren et al. 2006); and declining prevalence in the gazelle population following periods without livestock outbreaks, during which livestock vaccination occurred. Based on these observations, we hypothesize that the Mongolian gazelle population is not a reservoir for FMDV on the Eastern Steppe of Mongolia, but rather, the virus enters the gazelle population after spillover from livestock outbreaks.

The episodic history of FMD in our study region supports this hypothesis of spillover to gazelles from domestic livestock. Outbreaks of FMDV serotypes O and A occurred periodically in both domestic livestock and gazelles from 1931–73, but subsequently, there was a gap in outbreak occurrence in both livestock and gazelles for almost 30 yr. The FMDV

TABLE 2. Timing of foot and mouth disease outbreaks in Mongolia and in neighboring countries from 1999–2010, reported to the World Organization for Animal Health (OIE, 2011).

Year	Outbreaks in Mongolia	Serotype	Previous outbreaks in neighboring countries
1999	None reported	—	China PR
2000	April, May	O	Russia, Kazakhstan, China-Taipei, Korea, Japan
2001	February-March, May	O	Kazakhstan
2002	July	O	Korea
2003	None reported	—	None
2004	February	O	Russia
2005	August	Asia 1	Russia, China PR
2006	None reported[a]	—	Russia, China PR
2009	None reported	—	China PR
2010	May	O	China PR, Korea, Japan

[a] Foot-and-mouth disease virus type O was reported among the livestock owned by the School of Veterinary Medicine and Biotechnology within Ulaanbaatar, the capital city (ProMED-Mail Archive Number 20060424.1201). The outbreak was not reported to the OIE and was contained to these captive livestock.

serotype O reemerged in Mongolia in 2000, after outbreaks occurred elsewhere in Central Asia (Leforban and Gerbier, 2002; Sakamoto and Yoshida, 2002), and was present until 2004, infecting both livestock and gazelles (Sodnomdarjaa, 2005; Shiilegdamba et al., 2008). Furthermore, FMDV serotype Asia-1 emerged in August 2005 on the Eastern Steppe of Mongolia as part of an Asia-wide panzootic, and the genetic lineages showed close connections with that of China and Russia (Valarcher et al., 2009). The latest FMD outbreak occurred in Mongolia in May 2010 (FMDV-O) on the Eastern Steppe of Mongolia and also followed outbreaks caused by FMDV-O in China, Russia, Korea, and Japan. Based on reports by the World Organization for Animal Health (OIE), it appears that FMD outbreaks in Mongolia tend to follow outbreaks in neighboring countries (Table 2), suggesting that livestock and wildlife in Mongolia may be exposed to FMDV during pan-Asia epizootics. A longitudinal study of FMD infection in livestock and gazelle, alongside genetic comparison of viral isolates to isolates from elsewhere in Asia during a panzootic, is necessary to fully understand FMD presence and circulation on the Eastern Steppe of Mongolia.

If our hypothesis that Mongolian gazelles become exposed to FMD after spillover from domestic livestock is correct, then management actions targeting gazelles, such as culling or fencing to control movements (Taylor and Martin, 1987) are unnecessary and are likely to be ineffective in controlling FMD on Mongolia's Eastern Steppe. Mongolian gazelles are one of the few remaining species that maintains a long-distance migration (Berger, 2004) and have declined greatly in numbers. Management measures that decrease their numbers or limit their access to current habitat could have disastrous consequences for the species. The FMD prevention and eradication activities should rely on standard livestock disease-management actions that have been successful in controlling FMD elsewhere: serologic surveillance, vaccination, and, when necessary, judicious culling of livestock.

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