Detection of Mycoplasma ovipneumoniae in Pneumonic Mountain Goat (Oreamnos americanus) Kids

Authors: Wolff, Peregrine L., Blanchong, Julie A., Nelson, Danielle D., Plummer, Paul J., McAdoo, Caleb, et al.

Source: Journal of Wildlife Diseases, 55(1) : 206-212

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/2018-02-052

DOI: 10.7589/2018-02-052

Journal of Wildlife Diseases, 55(1), 2019, pp. 206–212

Detection of *Mycoplasma ovipneumoniae* in Pneumonic Mountain Goat (*Oreamnos americanus*) Kids

Peregrine L. Wolff,[1,7] Julie A. Blanchong,[2] Danielle D. Nelson,[3] Paul J. Plummer,[4] Caleb McAdoo,[1] Mike Cox,[1] Thomas E. Besser,[3] Juan Muñoz-Gutiérrez,[3,5] and Christopher A. Anderson[2,6] [1]Nevada Department of Wildlife, 6980 Sierra Center Parkway 120, Reno, Nevada 89511, USA; [2]Department of Natural Resource Ecology and Management, Iowa State University, 339 Science 2, Ames, Iowa 50011, USA; [3]Washington Animal Disease Diagnostic Laboratory, Department of Veterinary Microbiology and Pathology, Washington State University, PO Box 647034, Pullman, Washington 99164, USA; [4]Department of Veterinary and Diagnostic Production Animal Medicine, College of Veterinary Medicine, 1800 Christensen Drive, Ames, Iowa 50011, USA; [5]Current address: Diagnostic Medicine Center, Microbiology, Immunology & Pathology Department, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, 300 W Drake Road, Fort Collins, Colorado 80526, USA; [6]Current Address: Nebraska Game and Parks Commission, 299 Husker Road, Alliance, Nebraska 69301, USA; [7]Corresponding author (email: pwolff@ndow.org)

ABSTRACT: We documented bronchopneumonia in seven mountain goat (*Oreamnos americanus*) kid mortalities between 2011 and 2015 following a pneumonia epizootic in bighorn sheep (*Ovis canadensis*) and sympatric mountain goats in the adjacent East Humboldt Range and Ruby Mountains in Elko County, Nevada, US. Gross and histologic lesions resembled those described in bighorn lambs following all-age epizootics, and *Mycoplasma ovipneumoniae* was detected with real-time PCR in the lower and upper respiratory tracts of all kids. *Mannheimia haemolytica*, with one isolate being leukotoxigenic, was cultured from the upper respiratory tract of five kids, and in one kid, a leukotoxigenic strain of *Mannheimia glucosida* was isolated from both upper and lower respiratory tracts. During this same period, 75 mountain goats within the two populations were marked and sampled for respiratory pathogens, and *M. ovipneumoniae*, leukotoxigenic *Bibersteinia trehalosi*, and *Mannheimia haemolytica* were identified. The *M. ovipneumoniae* recovered from the kid mortalities shared the same DNA sequence–based strain type detected in the adult goats and sympatric bighorn sheep during and after the 2009–10 pneumonia outbreak. Clinical signs in affected kids, as well as decreased annual kid recruitment, also resembled reports in bighorn lambs from some herds following all-age pneumonia-associated die-offs. *Mycoplasma ovipneumoniae*, *Pasteurellaceae* spp., and other respiratory bacterial pathogens should be considered as a cause of pneumonia with potential population-limiting effects in mountain goats.

Key words: Disease, mountain goat, *Mycoplasma ovipneumoniae*, *Oreamnos americanus*, *Pasteurellaceae* spp., pneumonia.

All-age pneumonia epizootics followed in subsequent years by sporadic or recurring summer lamb mortalities are a significant source of mortality and population declines in bighorn sheep (*Ovis canadensis*) throughout western North America (Cassirer et al. 2018). Clinical signs in pneumonic lambs include coughing, nasal discharge, head shaking, ear drooping, and lethargy (Cassirer et al. 2013). Organisms most commonly detected in the lungs of pneumonic lambs include *Mycoplasma ovipneumoniae*, diverse *Pasteurellaceae* spp., and obligate anaerobic bacterial species (Besser et al. 2008).

In parts of their range, mountain goats (*Oreamnos americanus*) are sympatric with bighorn sheep. They may also contact domestic livestock on public grazing allotments or private land. Mountain goats are potential hosts for respiratory pathogens that are of concern for bighorn sheep (Lowrey et al. 2018). A single adult goat mortality related to *Pasteurella* pneumonia has been reported (Brandborg 1955). We report the diagnosis of bronchopneumonia and the detection of *M. ovipneumoniae* by real-time (RT)-PCR from upper and lower respiratory tract samples of seven dead kids from the East Humboldt Range (EHR), coordinates 40°55′16″N, 115°7′7″W, and the adjacent Ruby Mountains (RM), coordinates 40°37′19″N, 115°28′30″W in Elko County, Nevada US.

In the winter of 2009–10, an all-age pneumonia epizootic with consistent detection of *M. ovipneumoniae* in the upper and lower respiratory tracts was documented in bighorn sheep (McAdoo et al. 2010). The Nevada Department of Wildlife (NDOW) estimated that 10% (population estimated at

TABLE 1. Diagnostic summaries for *Pasteurellaceae* spp. (*Bibersteinia trehalosi* and *Mannheimia haemolytica*), *lktA* (the gene encoding leukotoxin A), and *Mycoplasma ovipneumoniae* (Mo) detected in mountain goats (*Oreamnos americanus*) sampled between 2010 and 2015 in the East Humboldt Range (EHR) and Ruby Mountains (RM) by the Nevada Department of Wildlife during an investigation of an outbreak of respiratory disease. Samples were tested by the Washington Animal Disease Diagnostic Laboratory (Pullman, Washington, USA).[a]

			Pharyngeal cultures for *Pasteurellaceae* spp.			*Pasteurellaceae lktA* detected		Mo detected	
Location	Year	Number tested[a]	*B. trehalosi*	*M. haemolytica*	*Mannheimia* spp.	*B. trehalosi*	*M. haemolytica*	Real-time PCR	Mo enzyme-linked immunosorbent assay
EHR	2010	3	3	1	—	NT	NT	1	3
	2012	2	1	—	—	NT	NT	0	2
	2013	15	13	9	—	NT	NT	1	14
	2014	16	11	15	1	NT	NT	2	14
	2015	11	11	7	1	4	8	2	9
RM	2012	12	8	6	—	NT	NT	5	11
	2013	2	NT	NT	—	NT	NT	0	2
	2014	11	8	7	3	NT	NT	1	10
	2015	3	3	1	—	0	1	0	3

[a] A total of 75 animals were tested for *Pasteurellaceae* and Mo. — = not sampled; NT = not tested.

220) and 13% (population estimated at 130) of sympatric mountain goat populations also died during this epizootic in the RM and EHR, respectively (Cox et al. 2017). During the disease event, one adult mountain goat carcass was recovered, and gross necropsy results documented bronchopneumonia. Due to advanced autolysis, no samples were collected for analysis. A total of 75 goats were captured by helicopter net gunning during (n=3) and after (n=72) the outbreak for marking and sample collection for pathogen surveillance. Nasal and pharyngeal swabs and blood samples were collected from goats in both mountain ranges and submitted to the Washington Animal Disease Diagnostic Laboratory for detection of *M. ovipneumoniae* by RT-PCR, identification of *Pasteurellaceae* spp. by bacterial culture as described by Roug et al. (2017), and *M. ovipneumoniae*–specific antibodies by enzyme-linked immunosorbent assay (Ziegler et al. 2014), respectively. The laboratory detected *M. ovipneumoniae* from nasal swabs of 12 goats and *M. ovipneumoniae*–specific serum antibodies in 68 goats. Pharyngeal bacterial cultures yielded *Bibersteinia trehalosi* in 77% (58/75) and *Mannheimia haemolytica* in 61% (46/75) of the animals sampled during this period. In 2015, RT-PCR targeting *lktA*, the gene encoding leukotoxin A (Walsh et al. 2016), was performed on the *B. trehalosi* (n=14) and *Mannheimia haemolytica* (n=8) pharyngeal isolates, detecting this gene in four and eight isolates, respectively (Table 1). Aerial surveys and ground observations conducted from 2011 to 2015 indicated decreased annual kid recruitment in both the RM and EHR populations (Cox et al. 2017; Blanchong et al. 2018), as has been documented for bighorn lambs following pneumonia epizootics (Cassirer et al. 2018).

Between January 2011 and August 2015, seven mountain goat kids (two from the RM and five from EHR), approximately 56–217 d of age, were presented to NDOW for necropsy. In 2011, one kid from the RM was found dead in January, and another alive in August that died 36 h after surrender to NDOW. The five kid mortalities from the EHR were collected in the summers of 2014 and 2015, coincident with weekly ground field observations to document kid behavior, signs of respiratory disease, and timing of kid mortalities (Cox et al. 2017; Blanchong et al. 2018). Clinical signs observed included coughing, head shaking, extension of the neck,

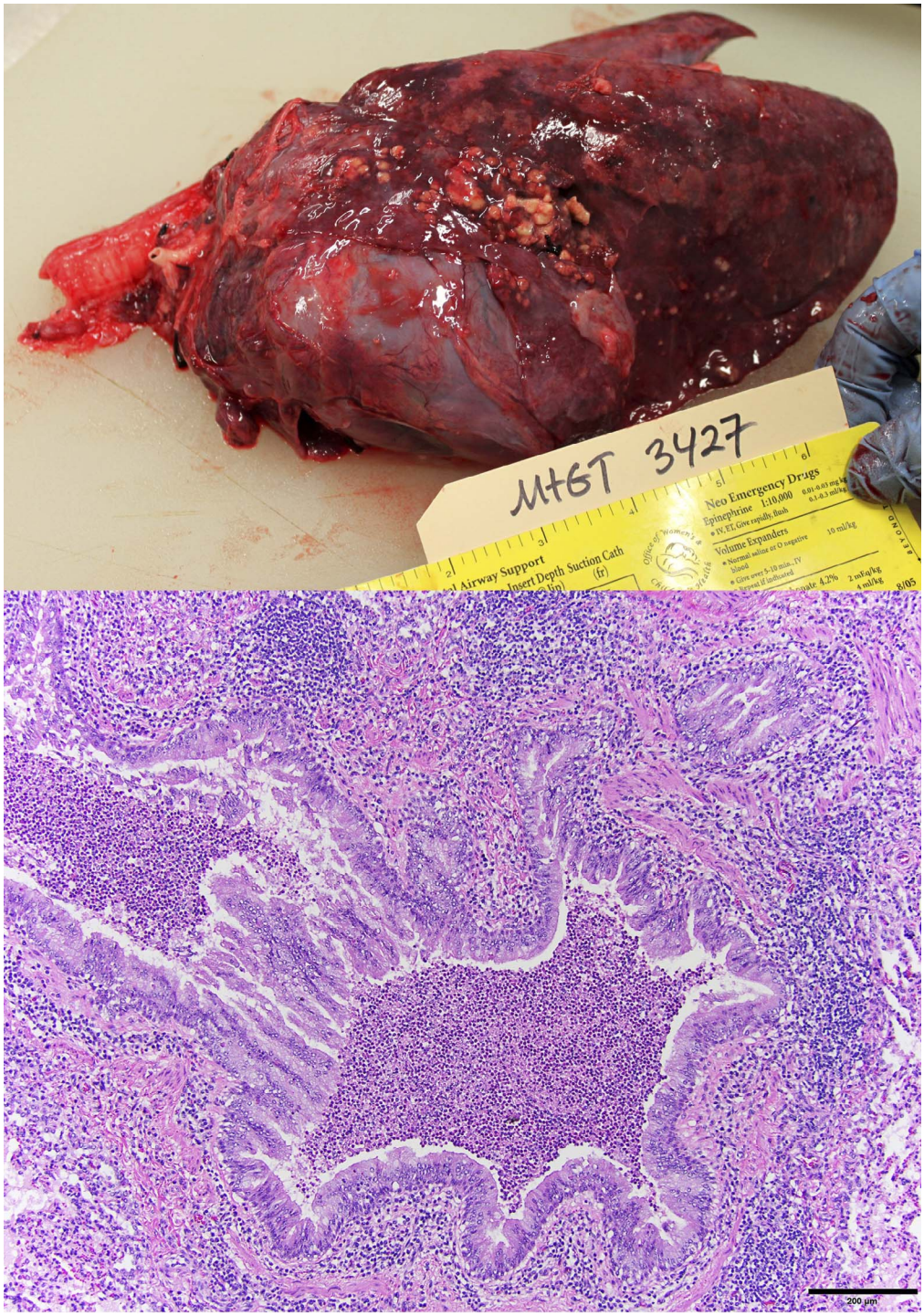

FIGURE 1. Gross necropsy findings of 56-d-old male mountain goat *(Oreamnos americanum)* kid identified as 3427 collected in 2014 following a bighorn sheep (*Ovis canadensis*) pneumonia epizootic in the Ruby Mountains and East Humboldt Range, Nevada, USA. (A) Abscessing bronchopneumonia of the heart and lung. (B) Histologic findings from the lungs of 77-d-old, male mountain goat kid identified as 4248 include bronchiolar lymphatic tissue hyperplasia, perivascular lymphoid cuffing, bronchiolar mucosal hyperplasia, and suppurative inflammation involving bronchioles and adjacent alveoli. Bar=200 µm.

TABLE 2. Histopathology findings by the Washington Animal Disease Diagnostics Laboratory (Pullman Washington, USA) for seven mountain goat (*Oreamnos americanus*) kids collected in 2011–15 following a bighorn sheep (*Ovis canadensis*) pneumonia epizootic in the Ruby Mountains and East Humboldt Range, Nevada, USA. Except for kid 917, all were recovered and submitted for necropsy to the Nevada Department of Wildlife within 24 h of death.

Year of death	Kid identification	Age (d)	Sex[a]	Histopathology
2011	612	66	M	Chronic-active suppurative bronchopneumonia with bronchiolar lymphatic tissue (BALT) hyperplasia; enteritis, moderate, multifocal, acute; lymphoid necrosis, mild to moderate, mesenteric lymph node
2011	917	217	F	Acute suppurative bronchopneumonia; acute tracheitis; necrotizing lymphadenitis
2014	3427	56	M	Abscessing bronchopneumonia; chronic, multifocal, tracheitis; otitis media, suppurative; portal hepatitis, suppurative, acute, multifocal, mild
2014	3434	63	F	Severe, chronic-active, bronchopneumonia with BALT hyperplasia; chronic, diffuse tracheitis; chronic, suppurative, bilateral, otitis media; acute, moderate, suppurative, multifocal hepatitis
2014	3435	66	M	Severe chronic-active bronchopneumonia with BALT hyperplasia and pleuritic; epicarditis, lymphoplasmacytic, multifocal, chronic, moderate; chronic, suppurative otitis media
2015	4241	66	M	Severe, chronic, suppurative bronchopneumonia with BALT hyperplasia; chronic, diffuse tracheitis
2015	4248	77	M	Severe, chronic, suppurative bronchopneumonia with BALT hyperplasia; chronic, diffuse tracheitis

[a] M = male; F = female.

nasal discharge, and lethargy (Blanchong et al. 2018). In July–August 2014 and August 2015, kids exhibiting severe clinical signs of respiratory disease were opportunistically collected; two immediately after natural death and three following euthanasia by gunshot. Whole carcasses were packed in ice and transported to NDOW for necropsy within 24 h.

Gross necropsy lesions (Fig. 1), histopathology (Fig. 1 and Table 2), and culture results (Table 3) for all kids were consistent with acute to chronic suppurative bronchopneumonia, including varying degrees of bronchiolar mucosal hyperplasia and bronchiolar lymphatic tissue hyperplasia. Five of the seven kids had histologic lesions consistent with acute secondary septicemia. There was no histologic evidence of predisposing immunosuppression, and all kids were negative for bovine virus diarrhea virus by immunohistochemistry or PCR. *Mycoplasma ovipneumoniae* was detected in all kids tested by RT-PCR in both the upper (nasal, sinus, tonsil, and tympanic bullae) and lower (lung) respiratory tract. The *M. ovipneumoniae* strain detected in the mountain goats (adults and kids) shared the 16S-23S intergenic spacer region DNA sequence previously detected in sympatric bighorn sheep during and after the 2009–10 pneumonia outbreak (Besser et al. 2012). As with other reports of pathogens recovered from the respiratory tract of pneumonic bighorn lambs (Besser et al. 2008; Grigg et al. 2017; Wood et al. 2017), *M. ovipneumoniae* was a consistently detected respiratory pathogen in the lungs of these pneumonic kids. However, the *Pasteurellaceae* spp. isolated from the pneumonic lung tissue of the kids were largely discordant with those detected in the pharyngeal cultures from the goats sampled during the same period (Table 1) and also with those detected in some studies of pneumonic bighorn lambs (Grigg et al. 2017; Wood et al. 2017). However, it is likely that additional *Pasteurellaceae* spp. were present but not detected by

TABLE 3. Bacterial culture and real-time PCR findings of the Washington Animal Disease Diagnostics Laboratory (Pullman, Washington, USA) for seven mountain goat (*Oreamnos americanus*) kids collected in 2011–15 following a bighorn sheep (*Ovis canadensis*) pneumonia epizootic in the Ruby Mountains and East Humboldt Range, Nevada, USA. *Mycoplasma ovipneumoniae* (Mo) was detected by real-time PCR in the lungs and upper respiratory tract (nose, tonsil, sinus, or middle ear [bulla]) of all seven kids tested. The PCR detections of *lktA*, the gene encoding leukotoxin A, conducted on upper respiratory tract or lung *Pasteurellaceae* isolates are indicated. Bacterial culture results conducted on nonrespiratory tissues (other tissues) are identified.

				Pathogens and leukotoxin A gene (*lktA*+) detected[a]						
Year of death	Kid identification	Age (d)	Sex[b]	Lung	Liver	Middle ear (bulla)	Tonsil	Nasal swab	Sinus swab	Other tissues in which pathogens were cultured
2011	612	66	M	Mo, ND	—	—	Mh	Mo	—	—
2011	917	217	F	Mo, ND	—	—	Mo	Mo, Mh	—	—
2014	3427[c]	56	M	Fn	—	Mo, Mh	—	—	Mo	Bronchial lymph nodes (Mh)
2014	3434	63	F	Mo, ND	Ps	Mo	Mh	—	Mo, Mh	Heart (Mh); kidney (Ms)
2014	3435	66	M	Mo, Ms	—	—	—	—	Mo, Pm	—
2015	4241	66	M	Mo, ND	ND	Mo,	*lktA*+, Mh	—	Mo, *lktA*+, Mh	Heart and kidney (Mg)
2015	4248	77	M	Mo, *lktA*+, Mg	Ahs, Ec	*lktA*+, Mg	*lktA*+, Mg	*lktA*+, Mg	Mo, *lktA*+, Mg	Heart (Ahs); kidney and spleen (Ec)

[a] ND = no pathogen detected; — = not sampled; Mh = *Mannheimia haemolytica*; Fn = *Fusobacterium necrophorum*; Ps = *Pasteurellaceae* sp.; Ms = *Mannheimia* sp.; Pm = *Pasteurella multocida*; *lktA*+ = gene encoding leukotoxin A detected; Mg = *Mannheimia glucosida*; Ec = *Escherichia coli*; Ahs = *Alpha-hemolytic streptococci*.

[b] M = male; F = female.

[c] This was the only kid to have anaerobic culture performed on the lung. The testing was conducted at Iowa State University, College of Veterinary Medicine (Ames, Iowa, USA).

culture (Butler et al. 2017). Four kids had additional organs submitted for aerobic culture, and in three, pathogenic bacteria were recovered, supporting the histologic finding of septicemia.

To our knowledge, respiratory disease as a cause of kid mortality has not been previously documented in mountain goats. Histopathology, bacterial culture, and agent-specific, as well as leukotoxin PCR assays, coupled with direct animal observations and decreasing recruitment, strongly suggest that mountain goat kids can have a similar clinical presentation, etiology, and population response as bighorn lambs following an all-age pneumonia epizootic (Besser et al. 2008; Wood et al. 2017). However, further research is needed to define the population impacts and epidemiology of respiratory disease in this species. Where mountain goats are sympatric with domestic or wild members of the Caprinae subfamily, wildlife managers should consider mountain goats to be susceptible to and potentially carriers of, important respiratory pathogens that may lead to population-limiting disease.

Funding for this project was provided by the Elko Bighorns Unlimited, Nevada Bighorns Unlimited, the Nevada Department of Wildlife Heritage Trust Fund, the Rocky Mountain Goat Alliance, and Iowa State University. Without the hard work of our field technicians, Joe Hoffmann, Wade McCammond, and Cole Bleke, this work would not have been possible. We are grateful to Washington Animal Disease Diagnostic Laboratory anatomic pathologist Kyle Taylor and Nevada Department of Wildlife personnel, Shane Talley, Tyler Nall, and especially, Chris Morris, for their dedication and support.

LITERATURE CITED

Besser TE, Cassirer EF, Potter KA, VanderSchalie J, Fischer A, Knowles DP, Herndon DR, Rurangirwa FR, Weiser GC, Srikumaran S. 2008. Association of *Mycoplasma ovipneumoniae* infection with population-limiting respiratory disease in free-ranging Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*). *J Clin Microbiol* 46:423–430.

Besser TE, Highland MA, Baker K, Cassirer EF, Anderson NJ, Ramsey JM, Mansfield K, Bruning DL, Wolff P, Smith JB, et al. 2012. Causes of pneumonia epizootics among bighorn sheep, western United States, 2008–2010. *Emerg Infect Dis* 18:406–414.

Blanchong JA, Clark CA, Klaver RW, Plummer PJ, Cox M, McAdoo C, Wolff PL. 2018. Respiratory disease, behavior, and survival of mountain goat kids. *J Wildl Manag* 82:1243–1251.

Brandborg SM. 1955. Life history and management of the mountain goat in Idaho. *Project Completion Report, PR Project 98-R. Bulletin No. 2*. State of Idaho Department of Fish and Game, Boise, Idaho, 142 pp.

Butler CJ, Edwards WH, Jennings-Gaines JE, Killion HJ, Wood ME, McWhirter DE, Paterson JT, Proffitt KM, Almberg ES, White PJ, et al. 2017. Assessing respiratory pathogen communities in bighorn sheep populations: Sampling realities, challenges, and improvements. *PLoS One* 12:e0180689.

Cassirer EF, Manlove KR, Almberg ES, Kamath PL, Cox M, Wolff P, Roug A, Shannon J, Robinson R, Harris RB, et al. 2018. Pneumonia in bighorn sheep: Risk and resilience. *J Wildl Manag* 82:32–45.

Cassirer EF, Plowright RK, Manlove KR, Cross PC, Dobson AP, Potter KA, Hudson PJ. 2013. Spatiotemporal dynamics of pneumonia in bighorn sheep. *J Anim Ecol* 82:518–528.

Cox M, McKee C, Schroeder C, Jackson P, Wakeling B, Scott M, Donham T, Kimble S. 2017. *2016–2017: Big game status*. Nevada Department of Wildlife. http://www.ndow.org/uploadedFiles/ndoworg/Content/Wildlife_Education/Publications/2017%20Big%20Game%20Status%20Book.pdf. Accessed February 2018.

Grigg JL, Wolfe LL, Fox KA, Killion HJ, Jennings-Gaines J, Miller MW, Dreher DP. 2017. Assessing timing and causes of neonatal lamb losses in a bighorn sheep (*Ovis canadensis canadensis*) herd via use of vaginal implant transmitters. *J Wildl Dis* 53:596–601.

Lowrey B, Butler CJ, Edwards WH, Wood ME, Dewey SR, Fralick GL, Jennings-Gaines J, Killion H, McWhirter DE, Miyasaki HM, et al. 2018. A survey of bacterial respiratory pathogens in native and introduced mountain goats (*Oreamnos americanus*). *J Wildl Dis* 54:852–858.

McAdoo C, Wolff P, Cox M. 2010. Investigation of Nevada's 2009–2010 East Humboldt Range and Ruby Mountain bighorn sheep die-off. In: *Proceeding of the 17th biennial symposium of the northern wild sheep and goat council*, Oregon Department of Fish and Wildlife, Hood River, Oregon, 7–11 June, pp. 51–52.

Roug A, Diaz-Campos D, Teitzel C, Besser TE. 2017. Comparison of two bacterial transport media for culture of tonsilar swabs from bighorn sheep (*Ovis canadensis*) and mountain goats (*Oreamnos americanus*). *J Wildl Dis* 53:188–192.

Walsh DP, Cassirer EF, Bonds MD, Brown DR, Edwards WH, Weiser GC, Drew ML, Briggs RE, Fox KA,

Miller MW, et al. 2016. Concordance in diagnostic testing for respiratory pathogens of bighorn sheep. *Wildl Soc Bull* 40:634–642.

Wood ME, Fox KA, Jennings-Gaines J, Killion HJ, Amundson S, Miller MW, Edwards WH. 2017. How respiratory pathogens contribute to lamb mortality in a poorly performing bighorn sheep (*Ovis canadensis*) herd. *J Wildl Dis* 53:126–130.

Ziegler J, Lahmers K, Barrington G, Parish S, Kilzer K, Baker K, Besser T. 2014. Safety and immunogenicity of a *Mycoplasma ovipneumoniae* bacterin for domestic sheep (*Ovis aries*). *PLoS One* 9:e95698.

Submitted for publication 21 February 2018.
Accepted 26 July 2018.