Summary report by Montana Fish, Wildlife and Parks to the Environmental Quality Council addressing statutory reporting requirements relative to bighorn sheep health sampling (87-2-702, MCA).

## September 2020

Before September 1 of each even-numbered year, the department shall report to the environmental quality council information on:

## (A) mountain sheep harvested pursuant to this subsection (4) from the Tendoy Mountain herd;

The Tendoy bighorn sheep herd was depopulated with a combination of an unlimited hunt and MFWP management removals in 2015 and 2016. A total of 34 bighorn sheep were removed through these processes. Public hunters harvested the bulk of the bighorn sheep herd, accounting for 23 bighorns in 2015 and 5 in 2016. About 360 hunters participated in the depopulation hunts. MFWP management removals accounted for another 6 bighorns sheep over the two-year period. The last management removals were in December of 2016 when the removal portion of that project was concluded.

The department is prepared to begin restoration efforts through translocation of bighorn sheep into this historical habitat. During winter 2020–2021, the department intends to capture 20–30 bighorn sheep from Wildhorse Island, of which four to five will be males that are 2 years old or less. The remaining bighorn sheep will be predominately ewes with some lambs. Another capture of 20–30 bighorn sheep from the same area is likely in 2021–2022, although another similar source may be used as well.

The bighorn sheep on Wildhorse Island have a long history of biological sampling without detection of important pathogens. Most recently, in winter 2020-2021, twenty-six bighorn sheep were captured for health evaluation. All captured animals appeared healthy upon examination, and all tested negative for exposure to *Mycoplasma ovipneumoniae* via PCR and serology. Only 1 animal tested positive for Leukotoxin A on PCR. *Bibersteinia trehalosi* and *Mannheimia haemolytica* were detected and are commonly detected in bighorn sheep herds in the absence of respiratory disease.

## (B) efforts to collect tissue samples and other biological information from mountain sheep harvested from the Tendoy Mountain herd to determine the immunity of surviving herd members to pneumonia outbreaks; and

MFWP sent letters to hunters requesting samples and field staff were supplied with kits to collect samples from harvested sheep. Harvested bighorns were sampled by collecting lungs, nasal and tonsil swabs, and a tissue sample for DNA analysis. Hunter participation in sampling was exceptional and a very high percentage of bighorns were sampled during the hunt. MFWP

conducted the standard suite of tests to look for respiratory pathogens. Bighorn sheep respiratory disease is a complex issue which is still very much in the research/learning stage with no definitive conclusion as to whether individual animals have immunity or if other factors resulted in survival.

Many harvested animals in the Tendoy herd had visible evidence of pneumonia upon examination of the lungs as well as moderate to heavy lungworm burdens. Of 33 animals sampled, 33% tested positive for *Mycoplasma ovipneumoniae* by PCR on either lung tissue or nasal swabs, and 44% tested positive on nasal swabs alone (Table 1). The Leukotoxin A gene, *Mannheimia haemolytica, Pasteurella multocida* and *Bibersteinia trehalosi* were also detected at low levels (Table 1), although true prevalence was almost certainly higher given the low detection probabilities associated with culture.

Table 1. Results from the sampling of hunter harvested and agency-removed bighorn sheep from the Tendoy Mountain herd, 2015-2016. A total of 33 animals were tested by PCR, and 32 animals were tested by culture. Nasal and tonsil swabs as well as lung tissue were collected for testing.

Pathogen/Disease	Test Type	N	Positive	Prevalence	95% CI	Comment
Mycoplasma ovipneumoniae	PCR	33	11	33%	20-50%	This represents PCR testing on either nasal swabs, lung tissue, or lung swabs. Prevalence on nasal swabs alone was 44% (11/25). We did not obtain blood, so we were unable to run serological assays for <i>M. Ovipneumoniae</i> .
Pasteurella						This test was only run on tonsil swabs
Leukotoxin A						that tested positive for Pasteurella
Gene	PCR	13	7	54%	29-77%	species.
Mannheimia haemolytica	Culture	32	1	3%	0-16%	
Mannheimia sp.	Culture	32	5	16%	7-32%	
Mannheimia glucosida	Culture	32	3	9%	3-24%	
Mannheimia ruminalis	Culture	32	5	16%	7-32%	
Pasteurella multocida	Culture	32	6	19%	9-35%	
Bibersteinia trehalosi	Culture	32	7	22%	11-39%	

(C) attempts by the department to share tissue samples and other biological information collected from the Tendoy Mountain herd with Washington State University, other public entities, and private entities that research the interaction between mountain sheep and domestic sheep.

Samples (lung and swabs) collected from harvested bighorn sheep were submitted to WADDL (Washington Animal Disease Diagnostic Laboratory) for testing, and genetic sequencing was attempted on any Mycoplasma ovipneumoniae PCR-positive samples. This data was subsequently used in an analysis of the genetic structure of Mycoplasma ovipneumoniae across western bighorn and domestic sheep (Kamath, P.L., Manlove, K., Cassirer, E.F., Cross, P.C. and Besser, T.E., 2019. Genetic structure of *Mycoplasma ovipneumoniae* informs pathogen spillover dynamics between domestic and wild Caprinae in the western United States. *Scientific reports*, *9*(1), pp.1-14.)

MFWP entered into a data sharing agreement with U.S. Department of Agriculture-Agricultural Research Station (ARS) in January 2017. The title of the ARS project was: *Identification of Host Factors and Immunopathogenesis of Pneumonia in Domestic and Bighorn Sheep*. This project was funded for 5 years and requested that FWP collect and share bighorn sheep samples from herds handled across Montana during this time. FWP collected and shared samples over three capture seasons (Feb-March 2017, Jan-March 2018, and Dec 2018) from 6 herds. No additional sheep from the Tendoy Mountain herd were removed during that time. Samples were processed according to ARS instructions, and shipped to the ARS lab. In addition, lab results for nasal swabs submitted by MFWP to WADDL for *Mycoplasma ovipneumoniae* PCR were shared with the ARS lab. The original data sharing agreement has expired, and ARS staff are in the process of finalizing their analyses.