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EFFECT OF PNEUMONIA ON POPULATION SIZE AND LAMB RECRUITMENT IN WHISKEY MOUNTAIN BIGHORN SHEEP

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Abstract: During the winter of 1990-91, a major die-off of Rocky Mountain bighorn sheep (Ovis canadensis canadensis) occurred in the Whiskey Mountain area near Dubois, Wyoming. Onset of the die-off was attributed to increased physiological stress resulting from several weeks of -40 C temperatures and 80 km/hr winds during peak rutting activities and human visitation. Three carcasses and various tissues from 8 additional sheep were collected from affected winter ranges and at necropsy. All lungs were grossly pneumonic microscopically characterized as suppurative to fibrinopurulent bronchopneumonia. Pasteurella haemolytica was isolated from all tonsils cultured, but from only I lung. No bacterial pathogens were isolated from 4 lungs, Moraxella sp. was isolated from 2 lungs and Chlamydia psittaci was isolated from 1 lung. Viral respiratory pathogens, lungworms and other parasites did not appear to be significant in causing mortality. The deaths of 124 bighorn sheep were directly attributed to pneumonia, although computer simulations suggested actual mortality probably exceeded 450 sheep. Animals died in approximately the same sex and age categories as existed prior to the die-off, based on examination of carcasses in the field. Lamb:ewe ratios declined from 44:100 in December 1990 to 16:100 in May 1991, but the percentage of yearling sheep observed in December 1991 indicated lamb mortality during the die-off did not exceed normal levels. Hunting licenses issued in 1991 for 3/4 curl rams were reduced by 22% from previous years. This reduction tended to mask effects of the die-off as success and average age of harvested rams remained similar to previous years. By December 1991, surviving sheep recovered from acute effects of pneumonia. Reduced lamb recruitment during 1991 (6 lambs:100 ewes in December) and predicted sub-normal production in 1992 will pose future management challenges as these age classes carry through the population. However, in spite of losing an estimated 30-40% of the population in 1991, the long-term prognosis is favorable.

<sup>1</sup>Deceased

Pneumonia is a well documented and significant mortality factor in bighorn sheep populations throughout their North American distribution (Spraker 1979, Feuertein et al. 1980, Wishart et al. 1980, Spraker and Hibler 1982, Festa-Bianchet and Samson 1984). Although not completely understood, outbreaks of pneumonia generally occur when animals are stressed above levels which they are accustomed to and/or contact domestic sheep. Further, extant viral and bacterial respiratory pathogens and parasites can affect the severity of a given pneumonia related die-off.

The Whiskey Mountain area in west-central Wyoming supports one of the world's largest wintering concentrations of Rocky Mountain bighorn sheep. Sheep from this area have been utilized to re-establish or supplement populations throughout Wyoming and 5 other western states. They are also becoming more important locally as a source of economic revenue from nonconsumptive wildlife users. One of the reasons this population has been attractive as transplant stock is because it has not undergone a major disease-related die-off in recent history. This paper documents the effects of the first large-scale pneumonia outbreak in Whiskey Mountain sheep.

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## STUDY AREA

The majority of winter habitats utilized by bighorn sheep in the Whiskey Mountain herd lie immediately south of Dubois, Wyoming. Geology, climate, and vegetation of these habitats were summarized by Butler (1977). Within the general wintering area, sheep have historically congregated on 3 "key" sites. These sites include Sheep Ridge, BLM Ridge, and Torrey Rim. The combined number of animals utilizing these sites varies between 600-900 annually.

Livestock grazing has been greatly reduced over the past 20 years as habitats were acquired for management as sheep winter range. Currently, 20-40 domestic horses (Equus caballus) graze portions of the winter range and only 2 forest allotments have active cattle (Bostaurus) preference within the Fitzpatrick Wilderness. There are no domestic sheep (Ovis spp.) grazing permits in the area. Winter and summer ranges of bighorn sheep are also used extensively by backpackers, photographers, fisherman, hunters, and other wildlife enthusiasts.

### METHODS

Clinical evidence of pneumonia was first documented in Whiskey Mountain sheep on 13 December 1990. By 6 January 1991, pneumonia was so prevalent that annual trapping operations were cancelled in order to prevent stress and avoid possible introduction by translocation of disease to other bighorn sheep herds. To determine the cause of the pneumonia outbreak, 2 animals shot while exhibiting clinical signs of pneumonia, I complete carcass of an animal found dead, and various tissues from 8 additional bighorn sheep were examined at the Wyoming State Veterinary Laboratory, Department of Veterinary Sciences, University of Wyoming.

Carcasses and tissues were examined grossly and body condition assessed subjectively based on the amount of visceral and bone marrow fat. Representative tissues were fixed in 10% buffered formalin. These were processed routinely for paraffin embedment, stained with hematoxylin and eosin and examined by light microscopy. Special stains included PAS and Gram's.

Tissues collected at necropsy were cultured at 37 C on Columbia agar base with 5% sheep blood and MacConkey agar incubated aerobically in 5% CO2. Isolates were identified using accepted methods (Lennette et al. 1985, Carter and Cole 1990). Lung and/or spleen samples were tested by fluorescent antibody tests for bovine respiratory syncytial virus (BRSV) antigen, bovine virus diarrhea virus (BVDV) antigen, parainfluenza 3 (PI3) virus antigen, infectious bovine rhinotracheitis virus (IBRV) antigen (Carbrey et al. 1971), and Chlamydia psittaci antigen (Riggs 1979). Virus isolation was attempted on ovine embryonic testicle cells using the techniques of Carbrey et al. (1971). Chlamydial isolation was attempted on McCoy cells (Haven et al. 1992). Feces were examined for intestinal parasites by sugar flotation and for lungworm larvae by the Baermann technique (Soulsby 1982).

In addition, to assess what potential respiratory pathogens might have been present in the herds prior to the epizootic and in the subsequent year, sera collected in January 1990 and 1992 were tested by virus neutralization for antibodies against BVDV, IBRV, PI3 virus, and BRSV (Carbrey et al. 1971), by complement fixation for Chlamydia spp. (Texas Veterinary Medical Diagnostic Laboratory, College Station, Texas), and by enzyme-linked immunosorbent assay (ELISA) for Pasteurella haemolytica specific antibody (K. W. Mills, unpubl. data). Sera were not collected during the epizootic in 1991 because trapping operations were cancelled.

From 11 January through 16 May 1991, sheep were surveyed approximately every 12 days. All animals observed were classified as to sex, age (Geist 1968), and whether or not they exhibited signs of pneumonia (i.e., coughing and/or nasal discharge). Percentages of the population infected with pneumonia were estimated using these data. All dead animals found were recorded as to location, sex, age, and, if suitable, submitted for necropsy.

Classification data were compiled and analyzed using Version 6.10 of the computer model POP-II (Barthelow 1990). Simulations were directed at data alignment from 1986-91 using harvest mortality, post-hunting season sex and age ratios, and trapping/transplanting removals. To determine total mortality associated with the pneumonia outbreak, winter Mortality Severity Indices (MSI) in the model were

increased to simulate the observed decline in lamb:ewe ratios from December 1990 (i.e., post-season) to May 1991 (i.e., end-of-biological year). Following these analyses, license quotas were set for 1991. Harvest data for 1991 were collected and analyzed, and sheep were classified again in December 1991. The simulation model was updated in spring 1992 using all 1991 data. Vegetation data presented in this paper were collected using methods outlined in Butler (1977).

#### RESULTS

Winter, 1990-91

From 1985-89, western Wyoming experienced above normal daily temperatures and below normal precipitation patterns (U.S. Department of Commerce 1992). Drought conditions peaked during 1988 and contributed to the massive Yellowstone wildfires. In the Wind River Mountains, drought appeared to moderate during the summer of 1990. As a result, lamb survival through the 1990 post-hunt period was slightly higher than the previous 3-yr average (44 lambs:100 ewes versus 39 lambs:100 ewes). Late-winter snows and heavy rains just prior to the birth of lambs apparently resulted in good post-partum foraging conditions. Better than average forage production on summer range was reflected in data collected on key winter use sites (Table 1). Herbaceous forage production improved dramatically above long-term average levels for sampling sites on BLM and Sheep Ridge. However, production was well below the long-term average for sampling sites on Torrey Rim.

Table 1. Comparison of herbaceous forage production (kg/ha) among Whiskey Mountain bighorn sheep winter use sites.

Site	1990	1991	5 yr. avg
Torrey Rim West	302	537	417
Torrey Rim East	373	837	540
Sheep Ridge East	563	567	451
Sheep Ridge West	484	359	312
BLM Ridge East	998	639	573
BLM Ridge West	499	476	404
Averages	537	569	450

The winter of 1990-91 was generally mild except for a 2-week interval in December 1990. Temperatures during this time dropped as low as -40 C. Under these conditions, winds are usually negligible. However, during the December cold period, winds in excess of 80 km/hr were common. Thus, wind chill factors of -85 C occurred during the rut when animal energy expenditures were high and human visitation was at a peak. The combination of high animal energy expenditures, increased human disturbance, extremely cold temperatures, and below normal forage production on Torrey Rim corresponded to the appearance of clinical pneumonia within that segment of the herd. By mid-February, the disease appeared in animals on BLM Ridge and Sheep Ridge.

Since pneumonia was not diagnosed in sheep on BLM or Sheep Ridges until mid-February, those areas were not surveyed as intensively until time (Table 2). Several trends are evident when comparing classification data collected from 13 December 1990 to 16 May 1991. First, rams left wintering areas in large numbers following cessation of the rut. Total ram:ewe ratios declined significantly late-January, February, and April and those rams observed were primarily yearlings and Class I animals. Second, on Torrey Rim where the disease was first diagnosed, the 1990 lamb crop was essentially eliminated by May. Lamb mortality was not as significant on BLM and Sheep Ridges. Finally, numbers of sheep observed in the area declined steadily until May. Declines were probably caused by a combination of increasing sheep mortality and movement of some animals off primary wintering sites. Increased numbers of sheep observed in May were thought to be the result of early plant green-up at lower elevations. Lush, succulent vegetation probably attracted animals which had previously left these areas.

A total of 124 dead bighorn sheep were found, mostly by horn hunters during and after the die-off. Thirty-six percent of all rams observed during 1990 post-season classifications were yearlings. Only 11% of documented ram mortalities were yearlings. Thus, yearlings died at a lower rate than they existed in the population prior to onset of pneumonia, while older, breeding-age rams succumbed at a higher rate.

Class I rams comprised 35% of all rams aged 2+ and older. Yet, only 19% of adult ram mortalities occurred in this age category. Conversely, Class II rams made up 30% of the classification sample, but 51% of the documented adult ram mortalities. Class III and Class IV rams died at rates similar to percentages of the population observed in December (24% observed versus 22% of mortalities and 11% observed versus 8% of mortalities, respectively). Total ewe mortality was not determined through data collection efforts, but is thought to have occurred in proportion to this sex class's occurrence in the herd.

Late, heavy snow accumulations in April 1991, coupled with heavy rains and more high elevation snow in May, forced sheep to remain on winter habitats much later than in most years. By 31 May, snow depths exceeded 1 m at elevations above approximately 2,900 m and many animals remained at low elevations. As a result, early lambs were being born on traditional wintering sites.

Using a POP-II computer model, post-season lamb:ewe ratios from 1986-90 were aligned to within I lamb:100 ewes of observed values. Simulated post-season ram:ewe ratios were left to track approximately II rams:100 ewes above observed values because some ram bands winter at high elevations and were missed during ground surveys.

After aligning the model with observed data by adjusting winter Mortality Severity Indices, it calculated a total mortality of 467 animals. Estimated ewe and lamb losses appeared feasible when compared with changes in observed age ratios from December 1990 to May 1991 and documented mortalities. However, the model could not accurately simulate documented ram mortality with its existing initial age

Table 2. Bighorn sheep classifications conducted on Whiskey Mountain winter use sites during the pneumonia outbreak of 1990-91.

Location and date	Rams	Ewes	Lambs	Rams:100 ewes: lambs	Total counted (% sick)
Torrey Rim:					
12/13/90 1/11/91 1/18/91 1/24/91 1/30/91 2/08/91 2/15/91 2/22/91 4/15/91 5/16/91	71 37 50 52 31 55 21 11 29 29	195 125 151 162 124 188 164 82 139 115	86 64 47 42 40 52 42 23 12 8	36:100:44 30:100:51 33:100:31 32:100:26 25:100:32 29:100:27 13:100:26 13:100:28 21:100:9 25:100:7	352 (0%) 226 (9%) 248 (8%) 256 (5%) 195 (4%) 295 (4%) 227 (4%) 116 (3%) 180 (2%) 152 (0%)
BLM Ridge/S	heep Ridge	e:			
12/15/90 1/11/91 <sup>a</sup> 1/18/91 <sup>a</sup> 1/24/91 <sup>a</sup> 1/30/91 <sup>b</sup> 2/08/91 <sup>b</sup> 2/15/91 <sup>c</sup> 2/22/91 <sup>d</sup> 4/15/91 <sup>d</sup> 5/16/91 <sup>d</sup>	82 42 46 28 27 26 21 60	221 59 97 91 233 207 138 210	94 12 33 35 95 77 32 52	37:100:43 105:100:20 47:100:34 31:100:38 12:100:41 13:100:37 15:100:23 29:100:24	397 (0%) 113 (0%) 176 (0%) 154 (0%) 130 (0%) 156 (10%) 355 (1%) 310 (2%) 191 (3%) 322 (2%)
Area-wide S			-771		
12/15/90 2/15/91 2/22/91 4/15/91 5/16/91	153 48 37 50 89	419 397 289 277 320	180 137 100 44 60	37:100:43 12:100:35 12:100:35 16:100:16 27:100:18	749 (0%) 582 (2%) 426 (3%) 371 (2%) 474 (1%)

Ridge.

dBLM & Sheep Ridge surveyed, sick sheep observed on both.

<sup>&</sup>lt;sup>8</sup>BLM Ridge surveyed only. <sup>b</sup>BLM Ridge surveyed only, none classified because no sick sheep

observed.

CBLM & Sheep Ridge surveyed, no sick sheep observed on Sheep

structure. Thus, total estimated mortality was considered conservative.

All carcasses and tissues examined at necropsy were collected on the Torrey Rim portion of the winter range from 4 January to 5 February 1991. Three carcasses and scavenged remains from 8 bighorn sheep were examined. Eight samples were from ewes, 1 was from a ram, and the sex of 2 were unidentified. Four of these samples were from lambs, 1 was from a yearling, 5 were from adults, and age of 1 specimen was not determined. Body condition was determined to be excellent in 2 cases, good in 4 cases, poor in 1 case, and 1 animal was judged to be emaciated. Two adult ewes were pregnant. Other documented carcasses were discovered, but were not suitable and/or not submitted for examination.

No bacteria were isolated from 4 of 7 lungs cultured. Moraxella-like bacteria were isolated from 2 lungs, and P. haemolytica was isolated from 1. Tonsils from 3 bighorn were cultured and all were positive for P. haemolytica, including the animal that had this bacteria in the lung. Actinomyces pyogenes was also isolated from the lung of this affected animal. All P. haemolytica isolates were nonhemolytic and typical of T types. The Moraxella-like organism was identified based on the following biochemical and other criteria: Gram negative, oxidase positive, catalase negative, vancomycin resistent, TSI K/K, urea negative, motility negative, nitrate negative, and no growth on MacConkey agar (R. Ellis, Colorado State Univ., pers. commun.).

All fluorescent antibody tests on lung tissues for respiratory viruses and Chlamydia were negative as were attempts at virus isolation. However, <u>C. psittaci</u> was isolated from the lung of l animal. Parasite burdens were considered low to moderate; 10, 80, and 150 larvae of <u>Protostrongylus</u> sp. per gram of feces were found in 3 animals. Eggs and oocysts of <u>Nematodirus</u> sp., <u>Trichuris</u> sp., and <u>Eimeria</u> sp. were detected in fecal samples, but numbers were low. Sarcocysts were identified in skeletal and/or cardiac muscle of 4 animals by microscopic examination.

Bronchopneumonia was grossly apparent in all lungs examined. Some were autolytic and many had been frozen and thawed prior to examination. The anteroventral regions were involved in all animals for which distribution could be determined. In those animals, grossly recognizable lungworm nodules were in the dorsal diaphragmatic lobes. Microscopically, the lesions in lung were characterized by vascular congestion and exudation of edema, neutrophils and macrophages into bronchi and alveoli. Fibrin within alveoli and on the pleura was observed in some animals. Areas of necrosis occurred in 2 animals, including the animal from which P. haemolytica was isolated from the lung. Mild bronchial epithelial hyperplasia was recognized in 1 animal. Bacteria were observed within pulmonary parenchyma, but large colonies were not common. Mild multifocal granulomatous to pyogranulomatous inflammation was associated with eggs, larvae, and adult lungworms in alveoli.

Results of virus neutralization tests indicated that BVDV was not present in the Whiskey Mountain herd (0 of 12 seropositive in 1990 and 0 of 27 seropositive in 1992). However, serologic evidence indicates other potential respiratory pathogens are very common. Eleven (92%) of 12 bighorns were seropositive for antibodies against PI3 in 1990 (reciprocal geometric mean titer [GMT] = 106, range 1:16 - 1:256) and 27 (100%) of 27 were seropositive in 1992 (GMT = 131, range 1:16 - 1:1024). Similarly, 11 (92%) of 12 bighorns were seropositive (GMT = 60, range 1:16 - 1:512) for respiratory syncytial virus antibodies in 1991 and 27 (100%) of 27 were seropositive (GMT = 69, range 1:16 - 1:2048) in 1992. Twelve (100%) of 12 bighorn were seropositive (GMT = 30, range 1:16 - 1:64) in 1990 and 25 (93%) of 27 were seropositive (GMT = 59, 1:16 - 1:64) for antibodies against Chlamydia spp. in 1992. In 1990, 9 (69%) of 13 bighorns were positive for P. haemolytica antibodies by ELISA and in 1992, 25 (93%) of 27 animals were seropositive. Seropositive animals for respiratory pathogens in 1992 included 1 lamb, indicating that transmission occurred between the time of it's birth in June 1991 to January 1992.

# Winter, 1991-92

Because preliminary data analyses from 1990 suggested a substantial die-off had occurred, the number of hunting licenses for fall 1991 was reduced from 82 to 64. Reducing total licenses by 22% was successful in lowering the harvest of 3/4 curl or larger rams by 17% from the previous 3-yr average (39 versus 47, respectively). However, hunter success increased from 57% to 61%, the average age of harvested rams remained at 6.5 yrs, and the percent of Class III and IV rams in the harvest increased from 33% to 46%. Thus, reduced license numbers effectively masked any detectable influence of the die-off on harvest statistics.

On 17 December 1991, bighorns were again classified on lowelevation winter ranges. A total of 608 sheep was observed during these classifications. An additional 354 animals were observed during aerial surveys. Only 1 animal exhibited signs of pneumonia and it was shot; necropsy revealed chronic resolving pneumonia. Lamb production declined to 6 lambs:100 ewes, the lowest level ever documented for this sheep population. Ram:ewe ratios were similar to those observed in 1990 (i.e., 39 rams:100 ewes each year).

### DISCUSSION

Acute to subacute, suppurative to fibrinopurulent bronchopneumonia was responsible for the mortality observed in the Whiskey Mountain herd in 1991. No known respiratory pathogen was consistently isolated from lungs and in 4 cases no bacteria were isolated. Microscopic lesions were most consistent with P. multocida or other bacterial induced bronchopneumonia and were not typical of lesions usually observed with P. haemolytica lung infection (Dungworth 1985). There was no evidence to suggest that viral respiratory pathogens were involved in this epizootic, even though the serologic data indicate that these viruses are ubiquitous in this herd. The role of Chlamydia spp. is not clear. It may act synergistically with bacteria to produce pneumonia and may

have been important in at least I animal.

P. haemolytica was isolated from the tonsils of 3 bighorns, but it was only present in the lung of 1 animal. Apparently, even though this bacteria is carried in the oropharynx, it may not be important in initiating bronchopneumonia even in the face of considerable environmental stress. The bighorn that had P. haemolytica in the lung was in poor body condition, also had A. pyogenes and Chlamydia in the lung, and the course of clinical disease was more prolonged than observed in the other animals. Perhaps, multiple factors allowed P. haemolytica to colonize the lung.

Moraxella and Pasteurella are taxonomically similar bacteria, though Moraxella is not considered a primary respiratory pathogen in domestic ruminants (Timoney et al. 1988). Moraxella liquifaciens was isolated from healthy bighorns from Whiskey Basin in 1976 and 1977 (Thorne et al. 1979) suggesting that it is probably not a primary pathogen. Similarly, PI3 virus appears to have been present in the Whiskey Mountain herd for a long period of time without causing appreciable clinical disease. Thorne et al. (1982) reported 23 (77%) of 30 bighorns seropositive by virus neutralization in 1976 and 1977. Long-term data are not available on the other potential respiratory pathogens, but there was little difference in seroprevalence to BRSV, Chlamydia, and P. haemolytica between 1990 and 1992. This, along with no evidence these were active in animals necropsied, suggests they were unimportant in the epizootiology of this pneumonia outbreak.

The epizootic occurred following a period of severe environmental stress. Animals varied in body condition. Two animals shot because of pneumonia were judged to be in excellent body condition. Thus, body condition alone was not a predisposing factor. However, a comparison of 1990 post-season ram classifications with the age structure of ram mortalities suggest that males may have been the first animals to become sick during the December cold period. Since Class II, III, and IV rams perform most of the breeding, these age classes would be expected to be in poorer physical condition than yearling and Class I rams. Thus, they should be more susceptible to a stress-related disease outbreak and would theoretically be more likely to succumb at higher rates than other ram age classes.

Originally, lambs were thought to have died at a much higher rate than they occurred in the population. However, the percentage of yearling sheep observed in December 1991 surveys was not significantly lower than average. Thus, lamb mortality during the die-off may have been compensatory to normal natural mortality in this segment of the herd. Although relatively few lamb mortalities were documented, avian scavengers and other predators rapidly removed all evidence of a carcass soon after death. During several sheep surveys, lambs which were discovered within hours of death were almost completely consumed.

During the pneumonia outbreak, wintering animals foraged in large, compact herds. This herding behavior probably facilitated spread of the disease among animals. Because several dead rams were found in Dry Whiskey Creek between Torrey Rim and BLM Ridge, males traveling between

ewe-lamb groups in these areas during the rut may have carried the disease to western portions of the area.

Despite death of an estimated 30-40% of the population, the long-term prognosis for this herd is favorable. Since approximately 1,000 animals were observed 1 yr after the die-off, adequate numbers of sheep remain to retain learned migratory behavior. We feel this behavior is essential to reduce forage use on winter ranges and maintain high physiological and genetic fitness. In addition, winter habitats at Whiskey Mountain continue to be intensively managed to increase forage production and expand sheep distribution into suitable, but currently unused areas. Reduced lamb recruitment during 1991 and predicted sub-normal production in 1992, however, will pose future management challenges as these age classes carry through the population.

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