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EXPERIMENTAL CONTACT ASSOCIATION BETWEEN BIGHORN SHEEP, ELK, AND DEER WITH KNOWN PASTEURELLA HAEMOLYTICA INFECTIONS.

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Abstract: Rocky Mountain bighorn sheep (Ovis canadensis canadensis), were placed with elk (Cervus elaphus), and deer (Odocoileus spp.) at Washington State University to determine susceptibility of bighorn sheep to pneumonia caused by Pasteurella spp. carried by these animals. Four elk, 2 white-tailed deer (Odocoileus virginianus), and 1 mule deer (Odocoileus hemionus hemionus) were introduced into a 0.72 ha pen which contained 10 resident bighorn sheep. Another group of 4 elk was placed in a 0.4 ha pen with 3 additional bighorn sheep. Pasteurella haemolytica was isolated from the pharyngeal swab samples of all elk and deer and ll of 13 bighorn sheep cultured at the time of elk and deer introduction. An isolate of P. haemolytica which reacted in antisera to serotypes T3, 4 and 10 was the most prevalent serotype detected, and was isolated from 5 of 8 elk, 3 of 3 deer, and 6 of 13 bighorn sheep. Pasteurella multocida was isolated from elk and bighorn sheep, but not from deer. All animals remained clinically healthy during the 6 months of close association. Therefore, no disease was associated with Pasteurella spp. carried by any of the animals included in this study.

The bighorn sheep pneumonia complex is a major mortality factor in bighorn sheep populations, and large scale mortality can result. Viruses, lungworms, stress factors, and bacteria can be important as multifactorial predisposing factors in the disease, and are often isolated or identified during episodes of pneumonia. Bighorn sheep in wild populations (Foreyt and Jessup 1982, Coggins 1988,) and in captive herds (Onderka and Wishart 1988, Foreyt 1989, Foreyt 1990, Callan et al. 1991) have developed bacterial pneumonia and died following exposure to domestic sheep. Domestic and exotic sheep were circumstantially incriminated as the source of bacteria, primarily P. haemolytica, that caused pneumonia in bighorns in the reports cited above. However, the potential role of elk and deer to serve as reservoirs of bacteria which may cause pneumonia in bighorn sheep has not been investigated.

Therefore the objective of this experiment was to determine the effects of close association of elk and deer known to be carriers of P. haemolytica on the health of captive bighorn sheep.

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METHODS

Animals.--Eight elk, 2 white-tailed deer, 1 mule deer, and 13 Rocky Mountain bighorn sheep were used in this experiment. The elk calves, 7 males and 1 female, were captured in the wild as free ranging calves before they were 7 days of age, and raised on unpasteurized goat milk until they were weaned at approximately 10 weeks of age. Calves were originally obtained from southeastern Washington (n = 4), northeastern Oregon (n = 3), or central Idaho (n = 4)The Washington and Idaho calves were captured by searching the elk calving areas on foot and physically catching the calves when found. The Idaho calf was captured after it fell off a cliff and fractured the dorsal spinal process of 3 lumbar vertebrae. recovered with supportive care, but walked with a limp. The other calves were normal and healthy. The deer fawns were orphans from southeastern Washington, and were raised in the same manner as the elk. All elk and deer were raised on a private facility with no contact with other ungulates. The bighorn sheep were 2 captive herds at Washington State University, Pullman, Washington. All bighorn sheep had been born in captivity or had been in captivity for a minimum of 2 years, and were well acclimatized to captivity and their respective pens.

On 16 August 1991 (experimental day 0), all deer and elk were transported to Washington State University and placed in 2 pens with the bighorn sheep. Four elk calves (3 males and 1 female), and the 3 deer were released into pen 1 which was 0.72 ha and contained 10 resident bighorn sheep (7 adult females, 1 adult male, 2 lambs). Four male elk calves were released into pen 2 which was 0.4 ha and contained 3 bighorn sheep (a 2-yr-old castrated male, a 2-yr-old ewe, and a ewe lamb). Both pens had abundant natural grasses, a shelter, feeder, and water. Supplemental feed consisting of alfalfa and barley, and mineralized salt were available at all times. Animals were observed daily for abnormal behavior or signs of disease.

Bacteria sampling procedures.—On experimental day 0, pharyngeal swabs were obtained from each animal. A mouth speculum was used to hold the mouth open while a sterile cotton-tipped swab was used to briskly rub the tonsillar area. Swabs were placed in Amies transport medium (Spectrum Diagnostics, Inc., Houston, Texas 77032, USA) and submitted within 1 hour after collection to the Washington Animal Disease Diagnostic Laboratory (WADDL), Pullman, Washington, for bactera isolations and analysis. Bacterial isolates were confirmed by routine biochemical testing (Carter 1984). Biotyping and rapid plate serotyping methods for P. haemolytica were done according to established formats (Biberstein 1978, Frank and Wessman 1978). All animals were sampled a minimum of 2 additional times between 1 and 6 months after initiation of the experiment (Table 1). Not all animals

were sampled at each sampling period (Table 1).

RESULTS

All animals remained healthy throughout the 6 month experimental period. Signs of respiratory disease were not observed, and all animals survived.

On day 0, \underline{P} . $\underline{haemolytica}$ was isolated from 11 of 13 bighorns, and all deer and elk. An isolate of \underline{P} . $\underline{haemolytica}$ that reacted in antisera to serotypes T3, 4 and 10 was the most common, and was detected in 6 of 13 bighorns, 5 of 8 elk, and 3 of 3 deer. Other serotypes in bighorns included A7 ($\underline{n}=1$), and serotypes that were untypeable because they did not agglutinate with known antisera ($\underline{n}=8$); in elk: untypeable serotypes ($\underline{n}=5$); in deer: T3 ($\underline{n}=1$ white-tailed deer), T3, 4 and 10 ($\underline{n}=1$ mule deer), and an untypeable serotype ($\underline{n}=1$ white-tailed deer). \underline{P} . $\underline{multocida}$ was isolated from 1 of 13 bighorn sheep and 2 of 8 elk, but not from the deer (Table 1).

For the remainder of the 6 month experiment, P. haemolytica was isolated at least once from every animal. An isolate of P. haemolytica that reacted in antisera to serotypes T3, 4 and 10 was the most common in all 3 animal species. Other isolates of P. haemolytica reacted in antisera to serotypes T3 and 4; T3, 4 and 10; and untypeable isolates in bighorn sheep; T3; T3 and 4; T3, 4 and 10; and untypeable in elk; and T3, T3, 4 and 10; and untypeable in elk; and T3, T3, 4 and 10; and untypeable in deer. P. multocida was isolated from bighorns and elk, but not from deer (Table 1). Other bacteria of potential importance that were isolated included Moraxella sp. from 2 elk, and Hemophilus sp. from 2 elk.

DISCUSSION

Close association between bighorn sheep, elk, and deer in this experiment did not result in pneumonia or death of any animal. Throughout the experiment, P. haemolytica and P. multocida were isolated commonly, but attempts to track transmission of the isolates between species was not attempted. However, future studies using DNA analysis techniques may indicate which bacteria were cross-transmitted between species, but without evidence of disease.

Previous research involving close association between bighorn sheep and domestic sheep has clearly indicated that bighorn sheep often die after association, most likely due to transmission of specific types of \underline{P} . haemolytica from domestic sheep to bighorn sheep. When I strains of \underline{P} . haemolytica from domestic sheep were given intratracheally to \underline{P} bighorns at a concentration of \underline{P} organisms, both bighorns developed respiratory disease and died within 42 h postinoculation (Onderka et al. 1988). Recent results from my laboratory indicated that a strain of \underline{P} . haemolytica A2 from healthy domestic sheep is usually lethal to bighorn sheep within 48 h when administered intratracheally at a concentration of less tha \underline{P} organisms. The same strain did not affect domestic sheep at the same inoculum level (Foreyt, Wash. State Univ., unpubl. data). Based on

Table 1. Isolations of <u>Pasteurella haemolytica</u> and <u>P. multocida</u> from captive bighorn sheep, elk, and deer.

	Bighorn (n =13	n Sheep	E1k (<u>n</u> = 8)	3)(8	White-ta	White-tailed Deer $(\underline{n} = 2)$		Mule Deer $(\underline{n} = 1)$
inoculation	P. haem	P. mult	P. haem	haem P. mult	Р. нает	P. mult	Р. наеш	haem P. mult
Day 0	11/13	1/13	8/8	2/8	2/2	2/0	1/1	0/1
Day 27	9/13	3/13	NStr	NS	NS	NS	NS	NS
Day 55	NS	MS	2/2	1/2	2/2	0/2	1/1	0/1
Day 87	NS	NS	3/8	3/8	NS	NS	NS	NS
Day 123	NS	MS	NS	NS	NS	NS	NS	NS
Day 146	3/3	6/0	8/9	3/8	NS	NS	NS	NS
Day 172	10/13	2/13	8/8	3/8	2/2	0/2	1/1	0/1

On Number of animals positive for <u>Pasteurella</u> sp. / Number of animals sampled b NS = No sample

experimental data and data collected from dieoffs of bighorn sheep in the field after exposure to domestic sheep, it has become a widely accepted tenet to prevent contact between domestic sheep and bighorn sheep in order to minimize domestic sheep induced mortality in bighorn populations. Similar recommendations could not be made regarding elk or deer and bighorn sheep based on the results of this experiment because all animals remained clinically healthy.

Information regarding <u>Pasteurella</u> spp. in wild ruminants is less extensive than for domestic animals (Rosen 1970, Thorne 1982, Biberstein 1990), but based on unpublished data from our ongoing investigations, it is likely that most wild ruminants carry a variety of strains of <u>Pasteurella</u> spp., and if samples are collected and processed according to established recommendations (Wild and Miller 1991), the probability of isolating <u>P. haemolytica</u> is high. It has been my experience that the most reliable method of isolation of <u>P. haemolytica</u> from bighorn sheep and other wild ungulates is to collect pharyngeal swabs and streak them directly on blood agar for bacterial isolation. This has resulted in significantly more isolations from live animals and a greater percentage of positive animals.

Based on the limited results from the current experiment, the strains of \underline{P} . haemolytica and \underline{P} . multocida that were identified in the elk and deer did not result in clinical disease in the bighorn sheep. Because the elk and deer in these studies were reared in captivity, it is not known if the bacterial isolates identified in the experiment are representative of the isolates found in wild populations of elk and deer. Future studies that compare isolates from wild populations and the isolates from animals in these studies will clarify that question. Further research is necessary to evaluate the association between bighorn sheep, elk, deer and other ungulates.

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