

A devastating outbreak of malignant catarrhal fever in a bison feedlot

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Abstract. In early 2003, an outbreak of malignant catarrhal fever (MCF) occurred in a bison feedlot in southern Idaho. The outbreak resulted in a 51.2% ($n = 825$) mortality rate among bison, which had been exposed to sheep for 19 days. Diagnosis was made by detection of ovine herpesvirus 2 (sheep-associated MCF virus) DNA in tissues or peripheral blood by polymerase chain reaction (PCR), and by histological examination of tissue lesions. Peak losses occurred between 41 and 55 days postmean exposure time (PME), and reached a maximum of 41 head per day. No known cases of MCF were observed among the 177 head of bison that arrived in the lot 3 1/2 weeks after the departure of the sheep. Of the several thousand head of beef cattle in the lot during the outbreak, only a single case of MCF was identified. This outbreak illustrates the devastating impact the MCF virus can have on bison under certain exposure conditions, the high threat posed by adolescent lambs to susceptible species, the significantly greater susceptibility of bison than beef cattle to MCF, and the lack of horizontal transmission from clinically affected bison to herdmates.

Key words: Bison; malignant catarrhal fever; outbreak; ovine herpesvirus-2; sheep.

Malignant catarrhal fever (MCF) is a frequently fatal disease that occurs as a complex of syndromes affecting mainly ruminant species such as domestic cattle, water buffalo, Bali cattle, American bison, and many species of cervids.^{4,14–16} The disease is characterized by inflammation, ulceration, and exudation of the oral and upper respiratory mucous membranes and gastrointestinal tract, ophthalmitis, and nervous system disturbances.¹⁵ MCF occurs worldwide wherever the 2 principal transmitting carrier species exist, domestic sheep (sheep-associated MCF) and wildebeest (the African form, or wildebeest-associated MCF). MCF has long been a major problem in farmed deer in New Zealand,² Bali cattle in Indonesia,²⁰ grazing cattle in Africa,¹⁵ and, in recent years, it has emerged as a severe threat to American bison.^{3,14,19} In this short communication, we report a devastating MCF outbreak in a bison feedlot.

On December 12, 2002, a flock of sheep comprised of 1,375 7-month-old lambs and 375 adult ewes was moved to an area immediately adjacent to a feedlot in southern Idaho that contained 1,610 head of bison and approximately 4,000 head of beef cattle. The sheep were grazed within 600 yards of the lot during the day, and were bedded down adjacent to the feedlot at night, at a distance of approximately 20 and 30 yards from the nearest bison and cattle, respectively; however, there was no nose-to-nose contact between sheep and

bison or cattle. On December 31, 2002, the sheep were moved away after having spent 19 days grazing in the area. An additional 177 head of bison were moved into the feedlot 25 days after the departure of the sheep. During the last week of January, 2003, several sudden bison deaths occurred in the lot. Animals developed depression and diarrhea, corneal opacity, and died within a short period of time (see below). Formalin-fixed and refrigerated tissue samples from 1 of the dead bison were sent to the MCF Research Laboratory, affiliated with the Washington Animal Disease Diagnostic Laboratory, Washington State University, Pullman, Washington. A diagnosis of MCF was made by detection of ovine herpesvirus 2 (OvHV-2, sheep-associated MCF virus) DNA in tissues by polymerase chain reaction (PCR)^{1,8} and by detection of compatible histological lesions.^{14,18}

The first case of MCF was recorded on January 24, 2003. Peak losses occurred between 41 and 55 days postmean exposure (PME \pm 10 days), and reached a maximum of 41 head per day (Fig. 1). Total mortality at 139 days PME was 825 head, or 51.2% of the 1,610 initial, exposed bison. The last case was recorded on May 10, 2003 (139 days PME). Some clinically unaffected bison that were of marketable weight were shipped for slaughter before the last case occurred, to reduce losses. Detailed clinical observations were recorded by the owner of the feedlot. Records made on 328 clinically affected bison revealed that 83.2% of the animals ($n = 273$) were severely depressed and separated themselves from herdmates (Table 1). Corneal opacity was observed in 42.7% of affected bison ($n = 140$). Severe nasal discharge was present in 87.8% ($n = 288$) of affected animals, but only 25.6% ($n = 84$) had ocular discharge (Table 1). The range of

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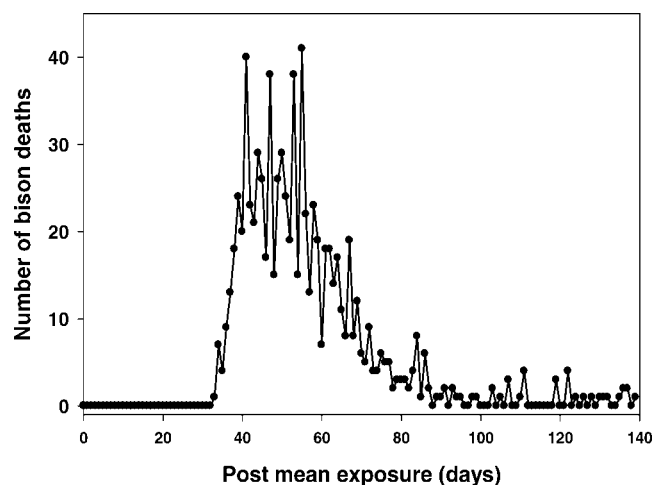


Figure 1. Distribution of numbers of bison that died from MCF in the feedlot during the outbreak. Each dot represents the number of mortalities that occurred on a given day. A total of 825 bison died: the first case was 33 days (January 25, 2003) postmean exposure (PME \pm 10 days) to sheep; the last case recorded was 139 days (May 10, 2003) PME. Peak losses occurred between 41 and 55 days PME, with fatality rate peaking at 41 head per day.

clinical disease duration in these 328 animals was from less than 1 day to 72 days (mean = 5.5, median = 2), with 31% of animals dying in less than 1 day, 50.6% within 2 days, 65.7% within 3 days, and 89.3% within 10 days (Fig. 2), after the onset of clinical signs.

Fifteen MCF suspect bison were randomly selected for histopathological examination and PCR tests. Two sets of tissues, including trachea, lung, liver, lymph node, spleen, kidney, intestine, turbinate, brain, heart, and abomasum, were collected: 1 set was fixed with 10% buffered formalin and the other frozen without fixation. Microscopic examination of the formalin-fixed tissues revealed that all 15 animals had vasculitis compatible with MCF lesions. They were consistent with those previously described in bison,¹⁴ including hemorrhagic cystitis, ulcerative enterotyphlocolitis, and arteritis-phlebitis.^{14,18} OvHV-2 DNA was amplified by nested PCR^{1,8} from all tissues collected from the 15 animals. Selected tissues from 6 of these bison were also examined by real-time PCR^{5,6} for the presence of OvHV-2 DNA. The copy number of OvHV-2 DNA varied among different tissues and even among the

Table 1. Summary of clinical presentations from affected bison during the outbreak.

Clinical signs recorded (<i>n</i> = 328)	Number of bison	% bison
Highly depressed	273	83.2
Nasal discharge	288	87.8
Ocular discharge	84	25.6
Corneal opacity	140	42.7

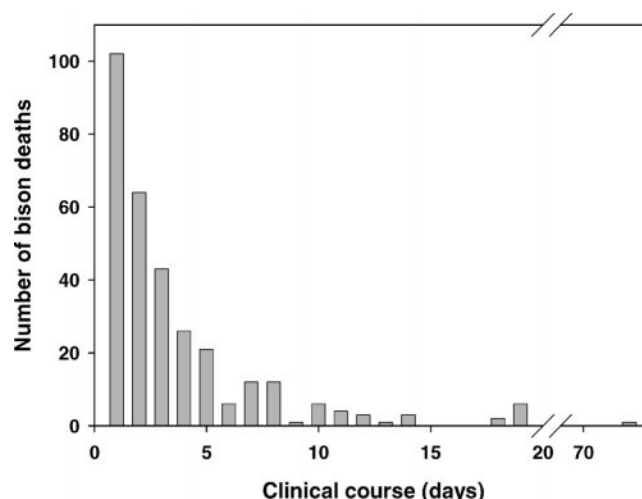


Figure 2. Duration of clinical course for affected bison (*n* = 328) during the outbreak. The bars represent the number of mortalities that occurred on each day following initial onset of clinical signs. The gap covers days 20–69, during which only 16 deaths occurred (not charted).

same type of tissue from different bison. The copy numbers from the tissues tested ranged from a few hundred copies to over a million copies per 2 μ g tissue DNA (data not shown). These results suggested that most tissues from bison with MCF are useable for diagnosis of MCF by PCR, although brain tissue generally contained relatively lower copy numbers than other tissues. In addition, 122 (97.6%) peripheral blood samples and/or lymph nodes from 125 diseased bison were positive by nested PCR (Table 2). The reason that 3 of the animals with typical clinical disease were OvHV-2 PCR-negative was not apparent. Possible explanations include 1) the clinical disease was induced by another agent, such as bovine virus diarrhea virus (BVDV). In fact, BVDV antigens were demonstrated in some bison with clinical disease in the early stages of this outbreak. The issue of BVDV infection is discussed in the following section. 2) It is

Table 2. OvHV-2 DNA and MCF viral antibody detected in different groups of bison in the feedlot.

Group	Number tested	OvHV-2 DNA		MCF viral antibody	
		Number positive	% positive	Number positive	% positive
Affected*	125	122	97.6	91	72.8
Exposed†	95	20	21.1	43	45.3
Unexposed‡	34	0	0	7	20.6

* Bison with suspect clinical MCF.

† Bison exposed to sheep in feedlot with the samples collected at slaughter.

‡ Bison not exposed to sheep in the feedlot with the samples collected at slaughter.

possible that the OvHV-2 DNA in the samples was below the detectable level by the PCR. Unfortunately, no additional samples were available from these bison that could be examined for further determination of the causes.

During the early phase of the outbreak, BVDV antigens were detected by immunohistochemistry in the tissues of 2 bison with clinical MCF. To determine the role of BVDV, 54 lymph nodes collected from bison with clinical disease were submitted to the Washington Animal Disease Diagnostic Laboratory for detection of BVDV by PCR. Other tissue samples from 21 bison with clinical disease, which included spleen, lymph nodes, liver, and lung, were submitted for BVDV isolation. In addition, 42 serum samples from clinically affected bison and 9 from unaffected bison were tested for BVDV antibody. Of the 54 lymph nodes sampled, 12 were positive for BVDV by PCR. These positive samples were all collected during the first 18 days of the outbreak. Only 1 BVDV isolate was obtained. This was from an animal that died 18 days after the first mortality. Subsequently, the isolate was sent to the National Veterinary Service Center for typing. It was reported that, though not definitively identified, the isolate was compatible with a field strain of the virus. No BVDV DNA was detected by PCR or virus isolation made in any bison after 18 days into the outbreak, even though severe losses from MCF continued for several more weeks. A high percentage of tested animals were seropositive to BVDV (37 out of 42 clinically affected [88%] and 9 out of 9 clinically unaffected [100%] bison were seropositive), but this was of little significance, as all animals had been vaccinated against BVDV, infectious bovine rhinotracheitis virus, and bovine respiratory syncytial virus and parainfluenza-3 virus early in the outbreak. Collectively, the data from the study suggested that the role of BVDV in the outbreak was apparently very minor or negligible. Further studies to determine the role of BVDV in the pathogenesis of MCF would be of value.

The severity of an MCF outbreak is dependent on the identity of the susceptible species involved, the amount of virus that is being shed from the carrier species, and the distance separating the 2. In general, the susceptibility of European breeds of cattle (*Bos taurus*) is low relative to some other ruminants. MCF cases in European breeds usually occurs sporadically.^{4,15} In contrast, Bali cattle of Indonesia,²⁰ American bison,^{14,19} and some cervid species (e.g., Pere David's deer)^{16,17} are highly susceptible. Under the same exposure conditions, in this outbreak, 825 out of 1,610 (51.24%) exposed bison died from MCF, whereas only 1 of 4,000 beef cattle (0.025%) developed clinical MCF (Table 3). As agricultural systems involving bison and deer production have developed, MCF has be-

Table 3. MCF mortality among different groups of animals in the feedlot.

Group	Total number	Total mortality	% mortality
Exposed bison	1,610	825	51.24
Unexposed bison	177	0	0
Exposed cattle	4,000	1	0.025

come more troublesome for these species. It is the leading cause of infectious disease losses on New Zealand deer farms.² With the increase of the American bison population in recent years, MCF has emerged as an extremely serious threat for bison producers. Virtually all sheep living under natural flock conditions that are older than 3–4 months of age are infected with the virus and can serve as sources for virus transmission.¹⁰ However, adolescent sheep between 6 and 9 months of age shed virus significantly more often and in greater amounts than adults.¹¹ Hence, sheep in this age range are the highest risk group for causing MCF outbreaks. The 1,375 lambs involved in this outbreak averaged about 7 months of age. This and the relatively short distance between sheep and bison in the lot were critical factors leading to the devastating losses. Several other significant MCF outbreaks in bison have been observed, all of which have been associated with congregations of young sheep (Crawford and Li, unpublished data). The 6–9-month range encompasses the age at which most lambs are confined for fattening, which makes sheep feedlots particularly rich sources of MCF virus.

The data from this study further suggested that bison suffering from clinical MCF do not transmit the disease to herdmates. Previous studies have shown that cattle were dead-end hosts for the wildebeest strains of MCFV, and no cell-free virus was demonstrable in the naso-ocular secretions of affected animals.^{13,15} However, whether animals with sheep-associated MCF also are dead-end hosts has remained uncertain. The potential for bison with clinical MCF to transmit the disease to their normal herdmates is of great concern for bison producers. A recent report³ on a natural MCF outbreak in bison after a 1-day exposure to sheep at an auction sale showed unequivocally that bison with sheep-associated MCF do not transmit MCF to other bison. This is further confirmed by data herein, in that 825 (51.24%) out of 1,610 head that were exposed to sheep developed clinical MCF, whereas among 177 bison that were moved to the feedlot 25 days after the departure of the sheep, not a single animal developed MCF, even though they were closely exposed to diseased pen mates.

Current diagnosis of sheep-associated MCF relies on detection of OvHV-2 DNA in affected animals by

PCR.^{12,14} cELISA serology, using a monoclonal antibody against an epitope conserved among the MCF viruses,⁷ detects antibody, which is a reliable marker for current infection because herpesviruses are almost universally persistent infections. Used alone, however, it does not establish the cause of the disease. Without PCR data, serology must be interpreted in light of clinical and histological evidence of MCF. As shown in Table 2, 122 (97.6%) out of 125 clinically affected bison were PCR positive but only 91 (72.8%) of these animals were seropositive. This percentage is similar to previous studies on diseased bison.¹⁴ The seropositivity among affected bison in this study was lower than usually seen in affected cattle.⁷ Although firm data are lacking, the explanation may lie in the shorter disease course of MCF in bison than in cattle not allowing sufficient time for antibody production. This area needs further study, as conflicting data exist. For example, in the present study, although only limited numbers were available, there was no apparent difference in seropositivity between bison clinically affected less than 3 days and those affected more than 7 days (data not shown).

A recent and as yet unpublished study revealed that the appearance of detectable antibody in bison experimentally infected with OvHV-2 ranged from 7 to 30 days before the onset of clinical signs and appeared to be related to challenge dose (O'Toole, unpublished data). A significant percentage of clinically normal bison on range or in feedlots are MCF seropositive.¹⁴ Only about 10% of those clinically normal bison had detectable OvHV-2 DNA in peripheral blood. A similar percentage (20.6%) of seropositivity as previously described¹⁴ was also observed in this study among the unaffected bison that were not exposed to sheep, but none of these seropositive animals were PCR positive. This may reflect the limited number of animals available for observation ($n = 34$). In contrast, among the 95 unaffected bison examined that had been exposed to the sheep, 45.3% ($n = 43$) were seropositive. Of a subset of these animals, 21.1% ($n = 20$) had detectable OvHV-2 DNA in peripheral blood at the time of slaughter (Table 2). The difference was statistically significant ($P < 0.05$) (Epi Infor, version 6.04, CDC, U.S.A & WHO, Switzerland) between the exposed and unexposed groups. Because all the clinically unaffected bison from the feedlot were of marketable weight and were shipped for slaughter to try to minimize losses, all the samples from these 2 groups were collected at slaughter. The difference between these 2 groups may have been due to the fact that some of the sheep-exposed bison may have been in a preclinical stage at slaughter and could have developed clinical MCF within a few days or weeks. A recent study on experimental induction of clinical MCF in bison revealed

that detectable OvHV-2 DNA appeared in peripheral blood before the onset of clinical signs (O'Toole, unpublished data). This suggests that, had the outbreak been allowed to run its complete course without premature removal of any bison from the lot, the actual mortality of this MCF bison outbreak may have been higher than observed.

Most MCF cases in bison can be linked to degree of association with domestic sheep. Although a full description of all potential transmission routes from sheep to bison awaits further studies, our understanding has advanced materially in recent years. A recent study on experimental induction of MCF in bison by aerosolizing nasal secretions from sheep that were experiencing a viral shedding episode supported the concept that aerosol transmission during close contact is the major route for transmission (O'Toole, unpublished data). Major factors affecting virus transmission to bison include the number and age of sheep involved and the distance separating the 2 species. Other factors, such as the presence of common water sources, mechanical carriers such as caretakers and vehicles, and prevailing climatic conditions also can affect the efficiency of virus transmission.

No vaccine is currently available for MCF. The most practical control measure for bison at present is to separate the animals from sheep. For small-scale application, sheep can be produced that are free of virus by early weaning and isolation,⁹ for use in mixed species operations, such as petting zoos and game farms. The establishment of effective yet practical separation distance guidelines represents an important issue facing sheep and bison producers. In the meantime, more widespread awareness and dissemination among both sheep and bison producers and involved veterinarians of currently available information about the disease and the factors influencing virus transmission would further the interests of both the sheep and bison industries.

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Survey of equine cutaneous neoplasia in the Pacific Northwest

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Abstract. A retrospective study examined data regarding equine cutaneous and mucocutaneous neoplasms submitted to the Veterinary Diagnostic Laboratory at Oregon State University in a 3.5-year period. A total of 536 neoplasms were identified, accounting for 30% of the total equine pathology submissions. Sarcoïd, squamous cell carcinoma, melanocytic tumors, papillomas, and mast cell tumors were the most common neoplasms, constituting 87.5% of all cutaneous neoplasms. Sarcoïds represented 51.4% of all neoplasms and 15.18% of total equine accessions. Sarcoïd was most common in paints, quarter horses, and Arabians, and was the only common tumor in donkeys and mules. Mean age at diagnosis of equine sarcoïd was 9 years. Squamous cell carcinoma constituted 18.3% of all neoplasms and 5.41% of total equine accessions. Ocular squamous cell carcinoma was most common in paints and quarter horses, and penile/preputial squamous cell carcinoma was most common in appaloosas and quarter horses. The mean age of horses with ocular squamous cell carcinoma (13 years) and squamous cell carcinoma of the skin (15 years) was significantly less ($P < 0.5$) than that of horses with squamous cell carcinoma of the penis and prepuce (21 years) or vulva, anal, and perianal skin (19 years). Findings suggest that equine sarcoïd and squamous cell carcinoma occur more frequently in the Pacific Northwest than in the northeastern United States.

Key words: Cutaneous neoplasms; equine.

Several prior studies have provided information regarding incidence of cutaneous neoplasia in horses.^{1–}

¹⁰ Previously reported incidence of all types of neoplasia in horses is up to 3% of horses in veterinary practice populations⁵ and up to 18% of all equine cases examined by veterinary pathologists.^{6,10} Cutaneous neoplasia has been reported to account for 50% of all equine neoplastic disease.¹ Prior survey studies are

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