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# The Diseases and Parasites of Alaskan Wildlife Populations, Part I.

## Some Observations on Brucellosis in Caribou

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### ABSTRACT

Data on brucellosis in several Alaskan caribou herds during 1962-65 is summarized. During this time agglutination-reactor prevalence rates (1:20 or higher) gradually declined in the Nelchina (6.5 percent to 1.0 percent) and Arctic (30 percent to 12 percent) caribou herds. A simultaneous decline (5 percent to 3.4 percent) in the prevalence of placental retention and/or excessive bleeding at parturition was also observed on the Arctic calving grounds in north-west Alaska in 1963 and 1965. Various additional conditions have been observed, from each of which brucella organisms were isolated on several occasions. These include orchitis-epididymitis, bursitis-synovitis and metritis, singly or in combination. In some cases, the observed lesions no doubt resulted in one or more of the following signs: sterility, lameness, and/or abortion with (probable) subsequent death of the female following putrefaction of retained placental structures. During 1963 about 25 percent of 107 cows showing placental retention and/or "excessive bleeding" were unaccompanied by calves when seen a few days post-partum.

The Russian and American points of view regarding naming the causative organism of rangiferine brucellosis are briefly reviewed. *Brucella suis* biotype *rangiferi* is proposed as a compromise, based on both the principles of bacterial taxonomy and the natural ecology of the organism.

### INTRODUCTION

Although there seems to be reason to believe that brucellosis may have been endemic in caribou and reindeer in North America, at least since the time of the reindeer introductions, it is only recently that the disease has been demonstrated in caribou and finally in reindeer. Tosach<sup>19</sup> concluded from observations on the occurrence of the disease in Eskimos that brucellosis was likely present in Canadian reindeer and/or caribou. Following the diagnosis of the disease in Alaskan Eskimos, Huntley et al.<sup>6</sup> demonstrated the organism in the Arctic caribou herd. Before that publication, the Alaska Department of Fish and Game and the Animal Disease Eradication Division of the U.S. Department of Agriculture independently initiated serological surveys for brucellosis and other diseases in the Nelchina caribou herd in southcentral Alaska and in other game species or caribou herds elsewhere in the State. The purpose of the present paper is to briefly summarize some of the data accumu-

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lated on brucellosis in caribou from 1962-65. A more detailed and extensive consideration of these and related data on caribou, and other species, and also more recent observations, will be reserved for a later date when it may be possible to make meaningful comparisons of the many associated variables (e.g. age, condition, changes in population density, etc.). We are particularly interested in testing the validity of Rosenbusch's<sup>17</sup> "ten-year-cyclicity" hypothesis based on his observations on epidemics of the disease in herds of "semi-wild" range cattle in Argentina. In this respect we expect to continue our field studies in Alaska for some time to come. Cooperative experimental studies of the disease in penned-reindeer with the U.S. Public Health Service, Arctic Health Center, during the past several years will be reported elsewhere by that agency.

#### MATERIAL AND METHODS

Both serology and bacteriological isolation were employed during the course of the study. Serum samples were recovered from whole blood specimens collected by sport hunters, native subsistence hunters, or during scientific collections. Most of these were preserved with phenol at a final concentration of 0.5 percent. Standard rapid-plate and tube agglutination tests using commercial (Lederle or Hyland or U. S. Department of Agriculture) standard *Brucella abortus* antigen were used in the field and laboratory. Standard bacteriological media and guinea-pig inoculations were employed for recovery of organisms from pathological specimens and femur marrow<sup>4</sup>.

Calving ground studies were made with the aid of high performance aircraft (PA-18, Piper, Supercub with large wheels) allowing slow flight for close observation of calving animals. Animals showing signs of disease or other distress that were near to potential landing spots (e.g. gravel bars, ridge tops, frozen lakes, etc.) were collected from the air with a 12 ga. shotgun using Winchester Mark V  $\pm$ 4 buckshot loads. This load is remarkably effective even on large adult males at reasonably close range (20-30 yards) and a marked improvement over older types of ammunition.

#### RESULTS

The distribution of the caribou herds considered herein are shown in Figure 1 which includes all of the Alaskan populations as recognized by Skoog (unpublished data). The data may be conveniently considered under two headings: Serology and Gross Pathology.

##### Serology

The serological data are segregated by area, date, sex, and titre in Table 1. Data on animals collected on the calving grounds are summarized separately in discussing specific lesions in the following section. Since these were for the most part specially selected because of apparent weakness, limping, or other defects, they do not truly represent the prevalence of the lesions and titres encountered.

All animals yielding titres of 1:20 or higher are classified as "positives." Isolations of the organism from animals exhibiting minimal titres of 1:20 support the validity of this practice when surveying the disease in caribou. It is also known from experimental penned reindeer infections that low titres can be associated with serious infections (Rausch et. al., in preparation) and are therefore indicators of exposure, if not significant disease. Golosov and Zabrodin<sup>5</sup> conclude that a titre of 1:25 signifies infection in reindeer. Serological cross reactions with *Pasteurella tularensis* and *Proteus* OX19 as a source of error in caribou-brucella serology have not been demonstrated.<sup>6</sup> However, recent research indicates that employing agglutination tests as the sole measure of prevalence may yield rates which are low. Nicoletti and Muraschi<sup>14</sup> reported that the tube-agglutination test did not detect 39 percent of 135 cattle that yielded *Brucella*, and that complement-fixation was superior to all other tests. LePennec and Goyon<sup>11</sup> reported that of 306 bovine sera positive to Coombs' test



and/or complement-fixation, only 173 were detected by agglutination. Accordingly, while agglutination tests may be satisfactory in situations where periodic retesting can be done (e.g. herds of domestic animals, humans, penned reindeer, etc.) it now appears that it may not be suitable for epidemiological studies of wild populations in which retesting is rarely possible and accurate prevalence rates are vital. However, for the purposes of this communication it is probably satisfactory to assume that any errors arising from the use of the *Br. abortus* agglutination test in our study are both conservative and common to all of the data reported at this time.

Inspection of the data in Table 1 readily reveals differences in prevalence correlated with season and sex. These correlations will be discussed in detail in the next section.

TABLE 1. *The prevalence of brucellosis test reactors in some Alaskan caribou herds, 1962-1965*<sup>①</sup>

Herd	Date	Sex	Titre								Other <sup>②</sup>	
			Negative		Positive							
			Total	Percent	1:20	1:40	1:80	1:160	1:320			
Nelchina	Fall '62	M	155	13	7.7	8	2	2			1:1280(1)	
		F	103	5	4.6	3	1		1			
		Both Sexes	258	18	6.5							
	Fall '63	M	212	8	3.6	1	3	3		1		
		F	107	4	3.6		1	1	2			
		Both Sexes	319	12	3.6							
	Fall '64	Both Sexes	229	1	0.5						1:640(1)	
		Both Sexes	92	1	1.0							
	Arctic	Fall '62	M	22	10	31.3						
			F	17	7	29.2						
Both Sexes			39	17	30.0							
Spring '63		M	15	2	11.7	2						
		F	45	12	21.1	8	2	1	1			
		Both Sexes	60	14	19.0							
Spring '64		M	8	0	0.0							
		F	24	5	17.2	3	1	1				
		Both Sexes	32	5	13.5							
Spring '65		M	58	5	8.0		2	1	1	1		
		F	73	13	15.1	5	6	2				
		Both Sexes	131	18	12.1							
Forty Mile	Fall '63	M	3	2								
		F	5	2								
		Both Sexes	8	5	38	1 <sup>③</sup>						
Alaska Peninsula	1963- 1964	Both Sexes	56	0	0							

<sup>①</sup> Specimens specially selected on the calving grounds not included.<sup>②</sup> Titre with number of reactors in parentheses<sup>③</sup> Sex not recorded

#### Gross Pathology

Three conditions have been found associated with natural infections of brucellosis in caribou: orchitis-epididymitis, bursitis-synovitis, and metritis. The brucella organism has been isolated on one or more occasions from each condition. Each will be briefly discussed below, leaving more detailed descriptions of histo-pathological details for inclusion in a report by the Arctic Health Research Center on penned reindeer and experimental infections.

#### Orchitis-Epididymitis

This condition has been observed on several occasions in animals from the Nelchina and Arctic herds. In one instance the infection was obviously of long standing. The testis-epididymis of a Nelchina bull taken in 1962 was greatly enlarged, purulent, evidently partly calcified and about 15 cm. long by 3.5 cm. in diameter. Bacteriological examination yielded a pure culture of *Brucella*. Unfortunately, other data on this hunter-killed animal are not available.

Meaningful estimates of the frequency of testicular lesions of brucellosis (or any other etiology) are not available. Confirmed cases have been encountered only twice, once each in the Arctic and Nelchina ranges. The Nelchina case has already been cited. The Arctic case involved a specially selected animal, one of seven males collected on the calving grounds during June 1963 and 1964. A titre of 1:160 was associated with an abscess of the epididymis from which the organism was isolated. However, careful examination of 52 bulls (yearlings or older) collected more or less at random at Anaktuvuk Pass during 1962-64 failed to reveal any testicular lesions. Cherchenko<sup>3</sup> reports that this is one of the less frequent conditions in Siberian reindeer, as it appears to be in Alaskan caribou.

#### Bursitis-Synovitis

Examination of 164 Arctic caribou (yearlings or older) taken at Anaktuvuk Pass during 1962-64 revealed only two instances of swollen carpal or tarsal joints. One of these was associated with a titre of 1:80 and was bacteriologically confirmed. In addition, one of 41 specially selected animals taken in June 1963 and 1964 on the Arctic calving grounds near the Upper Colville River also exhibited a swollen hock and an inflamed uterus, from both of which the *brucella* organism was later isolated. This animal, an adult female in poor condition, had a titre of 1:1280 and apparently had recently aborted. A mammary abscess was also present but culture was not attempted.

A particularly interesting example of carpal bursitis (hygroma) was brought in by a sport hunter. An adult female taken from the Nelchina herd in September 1961 bore large, identical sized, hygromata protruding from the anterior surfaces of the leg adjacent to each carpal joint. One was opened and found to contain 1576 gms. of clear fluid and 326 gms. of loosely aggregated solids. The greatly enlarged bursa appeared mildly inflamed. Unfortunately, neither serology nor bacteriological examination were available at that time as diagnostic aids. However, according to Jubb and Kennedy<sup>7</sup> in cattle, "large hygromata should be suspected of being secondarily infected with *Brucella abortus*." Cherchenko<sup>3</sup> reports that sero-fibrinous bursitis is one of the typical conditions associated with brucellosis in Siberian reindeer and Serova and Serova<sup>18</sup> claim that the most frequently affected bursae are those associated with the carpal joints. Accordingly, it seems reasonable to conclude that this spectacular pair of "caribou knees" represents a case of sero-fibrinous, carpal bursitis which perhaps had a traumatic origin, but which grew to relatively enormous size following invasion by the *brucella* organism. Another interesting case of bursitis was found in a hunter-killed animal also taken from the Nelchina herd in September 1961. This adult bull in otherwise excellent condition was found to have a large encapsulated, suppurative lesion containing about a quart of creamy, mayonnaise-like pus in the region of the *ligamentum nuchae*. No fistulae were present and no skeletal or other bursal involvement was evident either in the neck or elsewhere. A blood sample for serology could not be obtained from the bled-out animal and attempts to isolate an organism were unsuccessful. According to Jubb and Kennedy<sup>7</sup>, "progression of the reaction as a suppurative and granulomatous process is apparently due to

*Brucella abortus* or *Actinomyces bovis*, since these two organisms can be isolated rather consistently from the closed lesions." Since both brucellosis in caribou and actinomycosis (lump jaw) in wild sheep and caribou are known to be present in Alaska, and without other evidence, one can only conclude that either are possibilities.

#### Metritis-Abortion

Only two cases of brucellar metritis have been bacteriologically confirmed. Both of these animals were among 21 specially selected females taken on the Arctic calving ground in 1964. One case, already cited in the previous section, which had a titre of 1:1280, displayed confirmed uterine, tarsal, and mammary (not confirmed) infections and apparently may have recently aborted. Another which had lost her calf had a titre of 1:40 and an isolation was made from an infected cotyledon.

Fifty-nine cows (yearlings or older) taken at Anaktuvuk Pass in April 1963 were examined for abnormal uteri. Seven of these yielded inflamed uteri, two of which had serologic titres of 1:20 and 1:80.

Lent,<sup>10</sup> during his studies on the Arctic herd in 1961-1963, noticed abnormal retention of placental membranes, but apparently did not appreciate the possible epidemiological significance of his observation. During calving in the Arctic in 1963 and 1965, aerial inspection of 700 and 2,000 animals that had recently calved revealed 5 percent and 3.4 percent respectively with retained placental material and/or signs of excessive bleeding (Table 2). In some instances it was observed that placental retention continued to the point of extreme putrefaction and probable mortality. Occasionally cows without calves have been seen which, judging from the dried-shriveled appearance of the retained afterbirth, evidently produced a non-viable (dead pre or post partum) calf a day or so before. Other calves produced by such animals, though born alive, were stunted and appeared weak and unlikely to survive. It was noted during 1963 that about 25 percent of 107 cows with placental retention or excessive bleeding had lost their calves. Since only four of ten cows with retained afterbirth and/or bleeding collected during our studies were brucellosis reactors, it seems that other factors may also be involved in this condition. However, not all cases of brucellosis are detected by agglutination tests, so we can only conclude that at least some of the present instances of abortion and/or placental retention and bleeding are the result of this disease. Table 2 shows the relationship between spring, female reactor rates and the occurrence of animals with retained placentas and/or bleeding on the Arctic calving grounds. The simultaneous decline in both reactors and retained placentas seems to further implicate the disease as a probable cause of the placental problems (this trend continued in 1966 and 1967, unpublished data). Perhaps "excessive bleeding" and placental retention sometimes also involves poor nutrition. Indeed, animals in very poor condition have been commonly encountered during our studies and the Nunamiut residents of Anaktuvuk Pass claim that poor caribou are much more common now than a decade ago. We have a limited amount of data (unpublished) suggesting that there are disproportionately fewer agglutination reactors among animals in the very poorest (or best) condition. Accordingly, it seems safe to conclude that the present epidemics in the Arctic, particularly calving problems, are not simply a matter of pathogen biology, but also include secondarily related aspects of host biology (e.g. nutrition, etc.).

Cherchenko<sup>3</sup> reports that in Siberian reindeer herds only 1-5 percent of the animals will show any of the obvious brucellar conditions (e.g. bursitis, abortion, metritis, orchitis, etc.) at any one time. Also, that although abortions are frequently observed, their number cannot always be precisely established. Zabrodin<sup>22</sup> has observed morbidity rates of about 15 percent among reindeer older than 6 months.

TABLE 2. Comparison of female reactor<sup>①</sup> prevalence rates and placental retention and/or bleeding at parturition in the Arctic Caribou Herd, 1963 and 1965.

Year	Reactor Prevalence Sample		Retention and/or Bleeding Sample	
	Size	Percent	Size <sup>②</sup>	Percent
1963	45	21.7	2,130	5.0
1965	73	15.1	780	3.4

① Does not include animals specially selected on the calving grounds.

② Only cows with calves tallied.

#### DISCUSSION

During the past several years brucellosis has been epidemic in both the Nelchina and Arctic caribou herds. It appears that the disease has now declined to a low endemic level in the Nelchina herd, but still remains in essentially epidemic proportions in the Arctic herd. Because of the vast areas (tens of thousands of square miles) and large numbers of animals (about 100,000 and 250,000 respectively) involved, it is very difficult, if not impossible on practical grounds, to confidently estimate the true extent of the disease or its biological consequences. Indeed a sample of 66 animals taken from the Arctic herd in October 1965, yielded no serological reactors, even though prevalence rates obtained before and after in the springs in 1965 and 1966 were about 10-15 percent. We assume that this unexpected result happened by chance and does not represent a lasting, disease-free sub-population. The data do suggest, however (see Table 1), that there are distinct seasonal variations in reactor rates for males and females. It appears that in the fall male reactors are more common than female reactors and that the reverse is true in the spring. The additional stresses of rutting in the fall and pregnancy during late spring probably have some bearing on the health of each sex. Also, it is likely that the disease in males may be essentially chronic in nature (e.g. orchitis, bursitis, etc.) while the disease in females (e.g. metritis, abortion, placental retention) takes on primarily a short-term, acute form. In this event, the spring rise in numbers of female reactors would follow either infection (or reinfection) through sexual contact during the rut; or relapse of old infections under the stress of pregnancy. Experimental infections of pregnant reindeer resulted in abortions of the first fetuses, but not (without reinfection) those of the following year (to be published elsewhere in detail as noted above). It may be that the populational effect of this disease is determined by the number of infected, though sexually active bulls in the herd, perhaps at least sometimes in conjunction with poor nutrition and severe weather.

Whenever one is suddenly confronted with an unexpected epidemic of a disease like brucellosis, even in the wild, there is a temptation to wonder whether it was introduced or whether it is normally endemic in the region in question in an inapparent form in the primary host or a reservoir. Because most animal disease research in the past has dealt with domestic animals, wild animals have often been thought of as reservoirs but not primary hosts. Accordingly, when natural epizootics of brucellosis were first recognized in Siberia in 1949<sup>3</sup> it was hypothesized that the disease was introduced by infected cattle. It is now known that the organism from Siberian, Alaskan, and Canadian sources is a bacteriologically distinct form common to these areas.<sup>13</sup> Further, the disease in Siberian reindeer is reported not to involve reservoir species in that area,<sup>4</sup> and this may be the case in Alaska. It appears that brucellosis is a distinct, circumpolar disease of *Rangifer tarandus* subspp. Since symptoms in reindeer suggestive of brucellosis have been known in both Siberia<sup>3</sup> and Alaska (see



Reports on the Reindeer Industry, Sheldon Jackson, 1902-1906; Hadwen and Palmer, 1922<sup>[1]</sup>) since earlier times, it appears that rangiferine brucellosis may also be a disease of long standing, and not just a recently acquired biochemical and immunological variant of one of the well known forms causing disease in domestic animals. When one considers the history of our knowledge of brucellosis in domestic animals, and of Arctic medicine, it is little wonder that this seemingly distinct zoonotic disease of the Arctic has been recognized only quite recently. Indeed, only a few years ago in an article dealing in part with Arctic caribou in Alaska<sup>16</sup> it was concluded that, "infectious diseases can hardly survive in such a moving herd. . . ." In any event whether the disease has been endemic in caribou since their prehistoric migrations to North America or was introduced with reindeer at the turn of the century is unknown. However, it does not appear that the disease was recently introduced.

Throughout the preceding text we purposely have not used a specific name for the organism under discussion. Two schools of thought on this point seem to be current, each with considerable apparent justification. The American or "bacteriological school" having demonstrated that but a single pathogen is involved throughout the Arctic regions<sup>13</sup> proposes to name the organism strictly in accordance with bacteriological principles of taxonomy, i.e. *Brucella suis* type 4. The Russian or "ecological school," while employing bacteriological means to demonstrate the relative distinctness of the organism, gives greater weight to the ecology of the organism and thus names it *Brucella rangiferitarandi* or *Br. rangiferi*.<sup>4,21,15</sup> Indeed, anyone who is primarily concerned with the study of the disease in the field, and particularly in wild or semi-wild species, cannot help but be more impressed with the ecology of the organism and related natural lesions than with the techniques which prove useful in identifying the organism in laboratory cultures. Perhaps the most biologically meaningful name for the agent of rangiferine brucellosis would be *Brucella suis* biotype *rangiferi*. The biological, if not bacteriological, utility of this taxonomic compromise is obvious. Meyer<sup>12</sup> has commented in an abstract to her paper, "The Epizootiology of Brucellosis and its Relationship to the identification of *Brucella* Organisms," that ". . . each species of *Brucella* had a decided host preference" and that ". . . when a species does induce disease outside the preferential host, the organisms usually localize in the mammary gland and reticuloendothelial system rather than in the uterus and fetal membranes." In this respect *Brucella suis* biotype *rangiferi* appears to have as clear-cut preferences as the bovine, porcine, and caprine species of *Brucella*.

In discussing the epidemiology of human brucellosis in Alaska, Brody, et. al.<sup>2</sup> concluded, ". . . that intimate contact with caribou is responsible for a great proportion of brucella infections in northern regions." However, they are of the opinion that this may not be the case for the residents of Fort Yukon who consume more moose than caribou but also have the highest (21 percent of 174) brucella reactor rate of any Eskimo or Indian group studied. While they recognize that this nevertheless may be through contact with caribou, they also suggest that "other sources" may be involved and cite unpublished serological data implicating moose and Dall sheep. Rodents were also considered as possible reservoirs. While we don't have any data on rodents, we do have considerable serological data on moose and bison, and some Dall sheep. A Dall sheep ewe and lamb were taken on the John River in April 1963 while collecting Arctic caribou. The ewe was not a reactor, but the lamb unexpectedly tested 2<sup>+</sup>1:40 with *Br. abortus* antigen (Lederle). The only

[1] Unpublished reports on file in the Bureau of Animal Industry, Nome, Alaska.

other evidence at hand involving non-rangiferine reservoirs is a positive titre given by a moose killed on the Seward Highway on the outskirts of Anchorage. We have tested many other moose (to be published elsewhere), many of which were taken in caribou country, but all have been negative. Perhaps it is only a curious coincidence that this apparently rare reactor-moose was found within a few miles of the site of the reindeer pens in which experimental brucellosis studies were being carried out. In any case, these two natural infections (moose and sheep), both of which are unusual in some respect, are scant evidence at best that other species are natural reservoirs of brucellosis at this time. The demonstration that moose and sheep can be infected experimentally is not surprising nor is it evidence that such infections do occur except perhaps rarely in nature. Indeed, a moose kept in contact with infected reindeer for a year failed to develop a titre, while at the same time exposed reindeer became infected and in one case apparently died from the disease and perhaps other complications.

A possible reservoir of human disease seems to have been overlooked in Alaska, if not elsewhere. Since 1906 evidence has accumulated that dogs, though more or less resistant, can be naturally or experimentally infected, but usually without clinical symptoms of disease.<sup>10</sup> Most recently Kimberling, et al.<sup>8</sup> reported natural canine infections of *Br. abortus* and Kolesnik et al.<sup>9</sup> reported on experimental *Br. melitensis* in canines. The latter organism also has been reported in natural canine infections recently in Turkey.<sup>1</sup> Accordingly, there appears to be ample evidence that canines can be infected without always showing clinical symptoms and that they may also excrete organisms in urine or feces. When one also considers the fact that each year Eskimos feed thousands of pounds of fresh or frozen caribou meat and offal to their dogs and that 15-30 percent of the caribou are infected with *Br. suis* biotype *rangiferi*, it seems highly probable that at least occasionally dogs become infected. Since hydatid infections (*Echinococcus granulosus* and *E. multilocularis*) are more or less common in Eskimos and are unquestionably derived from unsanitary contact with egg-bearing dog feces, this same contact should result at least occasionally in exposure to brucellosis. Only if dogs are completely resistant to rangiferine brucellosis would this be entirely unlikely. Up to now no one appears to have recognized this obvious and apparently easily tested possibility.

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