

# Serological survey of selected canine viral pathogens and zoonoses in grizzly bears (*Ursus arctos horribilis*) and black bears (*Ursus americanus*) from Alaska

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## Summary

Between 1988 and 1991, 644 serum samples were collected from 480 grizzly bears (*Ursus arctos horribilis*) and 40 black bears (*Ursus americanus*) from Alaska, United States of America, and were tested for selected canine viral infections and zoonoses. Antibody prevalence in grizzly bears was 0% for parvovirus, 8.3% (40/480) for distemper, 14% (68/480) for infectious hepatitis, 16.5% (79/480) for brucellosis, 19% (93/480) for tularaemia and 47% (225/478) for trichinellosis. In black bears, prevalence ranged from 0% for distemper and parvovirus to 27.5% for trichinellosis and 32% for tularaemia. Antibody prevalence for brucellosis (2.5%) and tularaemia (32%) were identical for grizzly bears and black bears from the geographical area of interior Alaska. Links between differences in prevalence and the origin of the grizzly bears were observed. Antibodies to canine distemper virus and infectious hepatitis virus were mainly detected in grizzly bears from Kodiak Island and the Alaskan Peninsula. Brucellosis antibodies were prevalent in grizzly bears from western and northern Alaska, whereas tularaemia antibodies were detected in grizzly bears from interior Alaska and the Arctic. There was a strong gradient for antibodies to *Trichinella* spp. from southern to northern Alaska. For most diseases, antibody prevalence increased with age. However, for several infections, no antibodies were detected in grizzly bears aged from 0 to 2 years, in contrast to the presence of those infections in black bears. Grizzly bears served as excellent sentinels for surveillance of zoonotic infections in wildlife in Alaska.

## Keywords

Alaska – Black bears – Brucellosis – Canine distemper – Canine infectious hepatitis – Canine parvovirus – Grizzly bears – Trichinellosis – Tularaemia – United States of America – *Ursus americanus* – *Ursus arctos horribilis*.

## Introduction

There are an estimated 30,000 grizzly bears (*Ursus arctos horribilis*) and 140,000 black bears (*Ursus americanus*) in Alaska, United States of America (USA) (18). As predators and scavengers, bears may come into contact with agents of

zoonotic diseases (35). Serological evidence of infection by *Brucella* spp. in grizzly bears in Alaska has been reported previously (29, 43) as well as in black bears from other parts of North America (44). Bears can be infected by *Francisella tularensis*, the agent of tularaemia, which is endemic in Alaska (6): snowshoe hares (*Lepus americanus*) are the major reservoir for this disease (27). However, no extensive

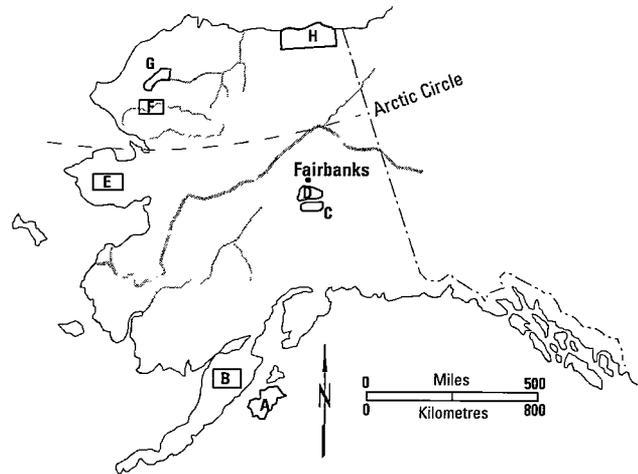
serosurvey of this infection has been performed in bears in Alaska. Trichinellosis is also known to be endemic in Alaska, and antibody prevalence of  $\geq 50\%$  has been reported in brown bears (*Ursus arctos*) (33). Zarnke *et al.* recently reported a seroprevalence of 5% for trichinellosis in grizzly bears from southern Alaska, rising to 83% in grizzly bears from northern Alaska (48). Antibodies to parvovirus have been reported in brown bears from Croatia (22). Antibodies to canine distemper virus and canine parvovirus have been reported from giant pandas (*Ailuropoda melanoleuca*) in China (24), and more recently from brown bears in Italy (25). Similarly, antibodies for distemper were found in polar bears (*Ursus maritimus*) from Alaska and Russia (17). Evidence of canine infectious hepatitis (CIH) antibodies in bears in Alaska has also been reported (46), but no information is available on seroprevalence in grizzly and black bears in Alaska of canine distemper virus and canine parvovirus, infections which have been reported in wolves (*Canis lupus*) from Alaska and Canada (10, 38, 45). The objective of this study was to determine the seroprevalence for various canine viruses and zoonotic agents in bears from Alaska.

## Materials and methods

Personnel of the Alaska Department of Fish and Game and the United States Fish and Wildlife Service captured 480 grizzly bears and 40 black bears in the course of performing population ecology studies between 1988 and 1991. Sampling was opportunistic and some bears were captured more than once; 644 serum samples were available for testing. 76 blood samples were collected from 40 black bears in interior Alaska on the Tanana Flats, south of Fairbanks. The 568 grizzly bear blood samples were collected from 8 different areas (Fig. 1), as follows:

- southern Alaska: 79 samples were collected on Kodiak Island, and 86 samples from the Alaska Peninsula (Katmai Coast [38 samples] and Black Lake [48 samples])
- interior Alaska: 53 samples were collected in the Tanana Flats, Denali Park and Fairbanks areas
- western Alaska: 40 samples came from the Seward Peninsula and 99 from the Noatak River drainage
- northern Alaska: 133 samples came from the northwest, 6 from the north-central (Prudhoe Bay), and 72 samples from the northeast regions.

Blood samples were collected by femoral, saphenous or cephalic venipuncture. Serum was separated by centrifugation and was stored at  $-20^{\circ}\text{C}$  until tested. Blood was collected from 25 (63%) of the 40 black bears more than once: 4 bears had blood taken four times; 3 bears three times and 18 bears had blood taken twice. Among the grizzly bears, samples were obtained three times from 11 bears and twice from 67 bears. Serological tests were performed for brucellosis, tularaemia and trichinellosis at the Veterinary Public Health Laboratory, Davis, California, USA, and for



- For grizzly bears:
- A: Kodiak Island
  - B: Alaska Peninsula
  - C: Interior Alaska
  - D: Interior Alaska
  - E: Seward Peninsula
  - F: Noatak River drainage
  - G: Western Arctic
  - H: Eastern Arctic
- For black bears:
- D: Interior Alaska

**Fig. 1**

**Location of study sites where samples of grizzly bear (*Ursus arctos horribilis*) and black bear (*Ursus americanus*) sera were collected for serosurvey**

CIH, canine distemper and canine parvovirus at the Virology Laboratory, Merial, Lyons, France, as described below.

### Canine parvovirus

Testing for canine parvovirus was conducted by haemagglutination inhibition, as previously described (8). Briefly, a viral suspension of four haemagglutinin units in a 0.05 ml buffer was added to the various dilutions of the serum samples to be tested for 1 h at room temperature. Then, 0.05 ml of a suspension of  $3 \times 10^7$  sensitised red blood cells were added in the plate wells and left overnight at  $4^{\circ}\text{C}$ . The reaction was read the following day. Any titre  $\geq 1:20$  was considered as a positive result.

### Canine distemper virus

Testing for canine distemper virus used a competitive enzyme-linked immunosorbent assay (ELISA) method. Briefly, 100  $\mu\text{l}$  per well of capture monoclonal anti-H glycoprotein neutralising antibodies at a 1:1,500 dilution in carbonate bicarbonate buffer, pH 9.6, was bound to 96-well flat bottom microtitration plates by overnight incubation at room temperature. On cell culture plates, 50  $\mu\text{l}$  of distemper virus and 50  $\mu\text{l}$  of each serum dilution (0.9, 1.8 and 2.7), and respective controls, were incubated for 1 h at  $37^{\circ}\text{C}$  with continuous shaking. After three washes, 50  $\mu\text{l}$  of the virus-serum mix was transferred onto the monoclonal antibody-sensitised plates and incubated with continuous shaking for 1 h at  $37^{\circ}\text{C}$ . Then, 50  $\mu\text{l}$  of the monoclonal anti-H glycoprotein neutralising antibody labelled with peroxidase

was added to each well and plates were incubated with continuous shaking for 1 h at 37°C. The plates were washed three times, then 100 µl of orthophenylene diamine substrate was added. The reaction was stopped after 25 min with 50 µl of 2.5 M sulphuric acid. Microtitration plates were read at a wavelength of 490 nm. Results were expressed as percent optical density (OD) compared to a control without serum (100%). Serum titres were given as log of the reciprocal dilution with a 50% OD. To validate the ELISA test and define the cut-off point (COP) for seropositivity, 23 serum samples from bears with an ELISA titre  $\geq 0.8$  were also tested by the classical serum-neutralisation (SN) test (4). Titres by SN were usually lower than those obtained by ELISA, which led to defining the COP at  $\geq 1.0$ . For further validation, positive ELISA samples and a random sample of negative ELISA serum samples were also tested using an immunoperoxidase antibody test, which is similar to a fluorescence test but uses immunoperoxidase instead of fluorescein isothiocyanate. Positive ELISA samples were also found to be immunoperoxidase-positive and negative ELISA samples gave negative results to the immunoperoxidase antibody test.

### Canine hepatitis virus

The SN test was used to detect canine hepatitis virus as previously described (7), using canine adenovirus (CAV) type 2 and dog kidney cell line Madin-Darby-canine-kidney (MDCK) ( $10^5$  cells/ml). A cytopathic effect on culture plates was read seven days after infection, and titres were expressed as  $\log_{10}$  protective dose 50% ( $PD_{50}$ ) on MDCK.

### *Brucella* spp.

Brucellosis testing was performed using the buffered acidified card antigen test (2, 3). Any sample which gave positive results was retested to eliminate non-specific reactions.

### *Francisella tularensis* (tularemia)

A commercial slide agglutination test was used to check samples for tularemia (30). Any titre  $\geq 1:20$  after two consecutive tests was considered as a positive result.

### *Trichinella spiralis*

Tests for trichinellosis used the ELISA method. The ELISA was based on the official United States Department of Agriculture method for pseudorabies (5, 37), modified as follows: briefly, 50 µl per well of a *Trichinella spiralis* ES antigen (5 µg/ml) at a 1:1,000 dilution in carbonate bicarbonate buffer, pH 9.6, was bound to 96-well flat bottom microtitration plates by overnight incubation at 4°C. Bear sera were diluted at 1:40 in Tris buffer (pH 7.4) containing 5% skimmed milk, 0.05% Tween 20 and 0.01% bovine serum albumin fraction V. The peroxidase conjugate was a raccoon anti-bear immunoglobulin G antibody used at 1:500. The substrate was 2,2'-azino-bis(3-ethyl benzthiazoline-6-sulfonic acid) (ABTS). The reaction was stopped after 30 min with 100 µl of a 0.1 M solution of hydrofluoric acid (pH 3.3). Each

plate contained known positive and negative control sera. The positive control serum was selected from a bear which gave positive results for both bentonite flocculation and ELISA (OD = 1.0), and the negative controls were selected from three bears that yielded negative results for both ELISA (OD < 0.1) and bentonite flocculation tests. Each serum was tested in duplicate and the mean of the two absorbance values was calculated. Microtitration plates were read at 410 and 450 nm for test and reference respectively on a microELISA autoreader. The COP for a positive test result was OD = 0.3 (mean OD of the negative control population [bears from Kodiak Island] plus 3 standard deviations) (23, 34).

Ages were estimated by examining cementum annuli of premolar teeth for black bears (39) and grizzly bears (12). Black bears were classified into four inclusive age groups: 0 to 2 years, 2.5 to 4 years, 4.5 to 8 years and  $\geq 9$  years. Grizzly bears were classified into five inclusive age groups: 0.5 to 2 years, 2.5 to 4 years, 4.5 to 8 years, 8.5 to 12 years and  $\geq 13$  years (21). All grizzly bears from the Seward Peninsula and Denali Park were reported as being adults (i.e.  $\geq 4.5$  years). In total, 62% (297/480) of the grizzly bears and 55% (22/40) of the black bears were females. Among the grizzly bears, 12% were aged  $\leq 2$  years, compared to 31.5% of the black bears. Demographic data (i.e. species, sex, age) were analysed using Epi Info version 6.02 (14). Frequency distributions were obtained and chi-square statistics for  $2 \times 2$  contingency tables were calculated to obtain measures of association and the statistical significance of such associations.

## Results

### Canine parvovirus

None of the grizzly and black bears tested had canine parvovirus antibody at a significant titre (i.e.  $\geq 20$ ).

### Canine distemper

Prevalence of canine distemper was not significantly different between females and males ( $P = 0.2$ ). It primarily varied according to the geographical origin of the bears (Table I), with grizzly bears from southern Alaska 2.5 times more likely to be seropositive than grizzly bears from other areas (95% confidence interval [CI] = 1.28, 4.96). Among the 41 positive samples (one bear gave positive results twice), 73% (30/41) had titres  $\geq 2.0$ , of which 77% (23/30) were from southern Alaska. Grizzly bears that yielded positive results for distemper were also more likely to give positive results for CIH (odds ratio [OR] = 2.65; 95% CI = 1.18, 5.60). Age prevalences are presented in Table II. The mean age of seropositive grizzly bears was 12.5 years (standard error [SE] = 0.85), whereas the mean age of seronegative bears was  $8.4 \pm 0.27$  years ( $P < 0.05$ ). High titres were mainly observed in grizzly bears of age  $> 8$  years. In southwest and western

**Table I**  
Prevalence of antibodies to *Brucella*, *Francisella tularensis*, *Trichinella spiralis*, canine distemper virus and canine infectious hepatitis virus in black bears and grizzly bears from Alaska by geographical origin, 1988-1991

Area of origin (species)	No. of animals positive (%)	<i>Brucella</i> positive (%)	Tularaemia positive (%)	<i>Trichinella</i> positive (%)	Distemper positive (%)	Infectious hepatitis positive (%)
Kodiak Island (GB)	77	8 (10)	3 (4)	0 (0)	23 (30)	26 (34)
Alaska Peninsular (GB)	86	11 (13)	12 (14)	17 (20)	7 (8)	8 (9)
Interior Alaska (GB)	40	1 (2.5)	13 (32)	14 (35)	1 (2.5)	5 (12.5)
Interior Alaska (BB)	40	1 (2.5)	13 (32)	11 (27.5)	0 (0)	3 (7.5)
Seward Peninsular (GB)	40	10 (25)	4 (10)	12 (30)	1 (2.5)	3 (7.5)
Noatak River drainage (GB)	87	21 (24)	12 (14)	50 (57)	0 (0)	10 (11.5)
Arctic northwest (GB)	96	15 (16)	34 (35)	86 (91*)	6 (6)	5 (5)
Arctic northeast and central (GB)	54	13 (24)	15 (28)	46 (85)	2 (4)	11 (20)
Overall prevalence (GB)	480	79 (16.5)	93 (19)	225 (47)	40 (8.3)	68 (14)

\* Two young females not tested

BB: black bears

GB: grizzly bears

Alaska, all grizzly bears that gave positive results were >7 years of age, whereas in northern Alaska, 40% of the bears which gave positive results were ≤4 years of age. The annualised prevalence for Kodiak Island was stable (32% in 1988 and 29% in 1989) and ranged from 2.6% to 7.6% in northern Alaska.

### Canine infectious hepatitis

Antibody prevalence for CIH was similar for both sexes. Prevalence varied with the geographical origin and the age of the bears (Tables I, II and III). The mean age of seropositive grizzly bears was 12.2 years (SE = 0.57), whereas the mean age of bears which showed negative results was 8.2 ± 0.28 years ( $P < 0.05$ ). No young grizzly bears (0-2 years) had CIH antibodies (Table II). Adult grizzly bears were more likely to be seropositive (OR = 9.07; 95% CI = 3.3, 34.7) than young bears and subadults. Conversely, none of the adult black bears were seropositive (Table III). Annual prevalence for black bears varied from 0% in 1988 and 1989 to 4.5% (1/22)

in 1990 and 8.7% (2/23) in 1991. Prevalence in grizzly bears decreased from 20% in 1988-1989 to 9.5% in 1990-1991, but varied also from region to region. Among the 78 grizzly bears tested more than once, 7 (9%) gave consistently positive results, 4 (5%) seroconverted between the first and the following collections and 2 bears (2.5%) became seronegative at the next collection. All black bears tested more than once yielded seronegative results.

### Brucellosis

Prevalence for *Brucella* antibody varied widely between geographical areas, but was similarly low in grizzly bears and black bears from interior Alaska (Table I). Grizzly bears from western arid northern Alaska (59/277) were 2.48 times more likely (95% CI = 1.41, 4.50) to be seropositive for brucellosis than grizzly bears from southwestern and central Alaska (20/203). No prevalence differences between the sexes were observed. The mean age of seropositive grizzly bears (8.7 years ± 0.59) was similar to the mean age of seronegative

**Table II**  
Prevalence of antibodies to *Francisella tularensis*, *Brucella* spp., *Trichinella* spp., canine infectious hepatitis and canine distemper by age in grizzly bears\*, Alaska, 1988-1991

Age	Number of serum samples	Tularaemia positive (%)	<i>Brucella</i> positive (%)	<i>Trichinella</i> positive (%)	CIH positive (%)	Distemper positive (%)
0 to 2 years	70	8 (11)	4 (6)	37 (53)	0 (0)	1 (1.5)
2.5 to 4 years	97	33 (34)	13 (13)	48 (49)	4 (4)	3 (3)
4.5 to 8 years	89	17 (19)	19 (21)	40 (45)	12 (13.5)	4 (4.5)
8.5 to 12 years	101	14 (14)	20 (20)	47 (47)	15 (15)	14 (14)
>12 years	167	22 (13)	18 (11)	89 (53)	42 (25)	20 (12)
Adult *	44	7 (16)	10 (23)	16 (36)	4 (9)	2 (4.5)
<b>Total</b>	<b>568</b>	<b>101 (18)</b>	<b>84 (15)</b>	<b>277 (49)</b>	<b>77 (13.5)</b>	<b>44 (8)</b>

\* Forty bears from the Seward Peninsula and 4 bears from Denali Park were only reported as adults

CIH: canine infectious hepatitis

**Table III**  
**Prevalence of antibodies to *Francisella tularensis*, *Brucella* spp., *Trichinella* spp., canine infectious hepatitis and canine distemper by age in black bears, Alaska, 1988-1991**

Age	Number of serum samples	Tularaemia positive (%)		<i>Brucella</i> positive (%)		<i>Trichinella</i> positive (%)		CIH positive (%)		Distemper positive (%)	
0 to 2 years	24	8	(33)	0	(0)	7	(29)	2	(8)	0	(0)
2.5 to 4 years	22	7	(32)	0	(0)	3	(13.5)	1	(4.5)	0	(0)
4.5 to 8 years	17	6	(35)	1	(6)	0	(0)	0	(0)	0	(0)
>8 years	13	0	(0)	0	(0)	1	(8)	0	(0)	0	(0)
<b>Total</b>	<b>76</b>	<b>21</b>	<b>(28)</b>	<b>1</b>	<b>(1.3)</b>	<b>11</b>	<b>(14.5)</b>	<b>3</b>	<b>(4)</b>	<b>0</b>	<b>(0)</b>

CIH: canine infectious hepatitis

grizzly bears (8.8 years  $\pm$  0.29), and prevalence increased with age (Table II). The annual prevalence decreased from 19% (39/202) in 1988 to 6% in 1990 (7/121), and increased again in 1991 (14.5%; 19/131). Among the 78 grizzly bears tested more than once, 4 (5%) gave consistently positive results, 10 (13%) seroconverted between the first and the following collections and 7 bears (9%) became seronegative at the second collection.

### Tularaemia

The overall prevalence of *F. tularensis* antibody was 19% (93/480) in grizzly bears and 32% (13/40) in black bears, with no major difference in the prevalence between the sexes. Seropositive bears were also more likely to yield positive test results for brucellosis (OR = 3.0; 95% CI = 1.75, 5.12). Most of the seropositive bears were from interior Alaska and the northern regions (62/190 bears), compared to grizzly bears from western and southwestern Alaska (31/290) (OR = 4.05; 95% CI = 2.44, 6.77) (Table I). Prevalence was identical (32%) in grizzly and black bears from interior Alaska. In black bears, antibodies were found only in animals <9 years (Table III). The mean age of seropositive grizzly bears (7.3 years  $\pm$  0.6) was slightly lower than the mean age of seronegative grizzly bears (9.1 years  $\pm$  0.3). Young and subadult grizzly bears (41/167) were more likely to be seropositive than adults (60/401) (OR = 1.85; 95% CI = 1.15, 2.95) (Table II). Grizzly bears with high titres ( $\geq$ 1:160) were mainly from the northern regions, the Noatak River drainage and interior Alaska. In black bears, annual prevalence decreased from 39% (12/31) in 1988-1989 to 20% (9/45) in 1990-1991. The same decrease was observed in grizzly bears from interior Alaska (40% [6/15] in 1988-1989 and 24% [9/38] in 1990-1991). However, the yearly overall prevalence in grizzly bears increased from 11% (36/322) in 1988-1989 to 26% (65/252) in 1990-1991. Among the 78 grizzly bears tested more than once, 54% gave negative results at all blood collections, 10% gave positive results at all blood collections, 13% showed positive results at the first blood collection and subsequently showed negative results, and 23% showed negative results at the first blood collection and positive results at following collections. Among the 25 black bears tested more than once, 20% gave positive results at all blood

collections, 12% yielded positive results at the first collection and negative results at the next one, and one bear seroconverted at the second test the following year.

### Trichinellosis

Seroprevalence for trichinellosis showed an increasing gradient from southwest Alaska to the north (Table I), with a similar prevalence in grizzly and black bears from interior Alaska (Table I). 70% of the grizzly bears with high titres (OD  $\geq$ 0.9) were from the northern regions and the Noatak River drainage. In grizzly bears, antibody prevalence was evenly distributed in all age groups (Table II), and the mean age of seropositive bears (8.96  $\pm$  0.38 years) was similar to that of seronegative bears (8.57  $\pm$  0.37 years). In black bears, 10 of the 11 positive bears were <5 years of age (Table III). Yearly prevalence of trichinellosis in grizzly bears was constant at the Noatak River drainage (57% in 1988 and 1989, 59% in 1991), whereas in the northern regions it slightly decreased from 91% in 1988 to 80% in 1990-1991. Of the 78 grizzly bears from which samples were collected more than once, 64% gave positive results, 23% gave negative results at all blood collections and 13% gave negative results at one and positive results at the other. For the 25 black bears tested more than once, 68% regularly gave negative results, 20% gave first negative then positive results and 12% gave first positive results then negative results at the second blood collection.

## Discussion

### Canine parvovirus

None of the bears tested had canine parvovirus antibodies, despite reports of infection of wolves in Alaska with this disease (45). The only reports of possible parvovirus infection in bears were from Croatia, where 7 out of 22 brown bears were found seropositive (22), and from China, where giant pandas were found to be seropositive (24). The data accumulated in the present study seem to indicate that bears in Alaska may not be exposed to canine parvovirus infection.

## Canine distemper virus

This is the first report of morbillivirus antibodies in grizzly bears from Alaska. The recent report by Follmann *et al.* (17) of morbillivirus antibodies in polar bears from northern Alaska and Russia clearly indicates that a distemper-like virus is present in different species of bears in North America. This raises questions about the epidemiology of morbillivirus infection in wildlife in Alaska. In contrast to the results reported for polar bears, the seroprevalence of canine distemper virus in grizzly bears was rather low in the northern regions and much higher in southwest Alaska, especially on Kodiak Island. The prevalence was also very low or non-existent in all grizzly and black bears from interior Alaska and grizzly bears from western Alaska. By comparison, antibody prevalence was more evenly distributed among wolf packs in all geographic areas (45). Only adult bears on Kodiak Island and the Alaskan Peninsula had detectable morbillivirus antibodies. This may suggest that a major epidemic occurred several years ago in southern Alaska and the adult bears were the survivors of an infected cohort. On the contrary, in the Arctic region, infection is still active at a low rate in the bear population, as 3% of the juvenile bears and 6% of the subadults and adults  $\geq 8.5$  years had antibodies. The origin of the morbillivirus which affects the grizzly population needs further investigation. It is not possible to determine whether the antibodies were directed against canine distemper virus, phocine distemper virus or a new morbillivirus (17). The distribution of the infection could support a possible relation to phocine distemper, as suggested for polar bears, or the possible spread to bear populations from wild canids, most likely foxes (*Vulpes vulpes*) or possibly wolves (17). For the second hypothesis, a source of infection from wolves seems less likely – despite the fact that distemper antibodies are more widespread and evenly distributed in wolves in Alaska – because of their sparse distribution. However, the much higher geographical density of foxes, which are known to be susceptible to distemper, makes them more likely to be spreading the infection to other wild species. The possibility of infection by a virus of canine origin, as suggested for canine distemper antibodies in giant pandas in China (24), also seems less likely, as domestic dogs are found throughout inhabited areas of Alaska.

## Canine infectious hepatitis

This study confirms previous results obtained for CIH in grizzly bears from Alaska (46). The overall prevalence of 14% (68/480) is similar to the 12% reported by the authors of the previous study, despite a time frame of 4 years in the present study and 15 years in the previous study. Similarly, no differences in prevalence according to sex were found, but age and geographical origin of the bears were significant factors associated with antibody prevalence. Bears from Kodiak Island again had the highest prevalence (31%), and prevalences were very similar to those previously reported in northwest (6%) and northeast Alaska (13%). Prevalence increased with age, but none of the young bears (0-2 years) had any detectable antibodies. Interestingly, none of these

young grizzly bears had *Toxoplasma* antibodies (9). A higher susceptibility of young grizzly bears to CIH was suspected (46), thus leading to the death of infected animals as seen in black bear cubs (11). However, in any epizootic, not all animals will die and a small percentage of the infected animals should have antibodies. Therefore this absence of antibodies may result from a lack of exposure of cubs to the CIH virus or *Toxoplasma* spp. Both the persistent absence of CIH antibodies from young grizzly bears and the high antibody prevalence against morbillivirus and CIH in grizzly bears from Kodiak Island need further investigation to assess the cause.

## Brucellosis

Brucellosis is an endemic disease in the great herds of reindeer and caribou (*Rangifer tarandus*) throughout the world (15). In Alaska, brucellosis is caused by *B. suis* type 4 (26), and major populations of reindeer are found in western (Seward Peninsula) and northern Alaska (40). Prevalences of 30% in the Arctic caribou herd and up to 15% in some of the reindeer herds on the Seward Peninsula have been reported (16). Antibodies against rangiferine brucellosis have been found in wolves, red foxes and bears (28). The prevalence of antibodies to rangiferine brucellosis in grizzly bears is indicative of the prevalence of infection in the reindeer and caribou herds, as the highest prevalence in grizzly bears was found in those from western and northern Alaska. The seroprevalence was somewhat lower than previously reported (28) in grizzly bears from the Alaskan north, but similar to observations by R.L. Zarnke (unpublished data). Prevalence was the lowest in interior Alaska, where brucellosis prevalence is low (1% to 3%) in both reindeer and caribou (R.L. Zarnke, unpublished data) as well as wolves (45). It is not known if brucellosis in bears has any clinical consequences, such as abortion and sterility.

## Tularaemia

*Francisella tularensis* was first recorded in Alaska in 1937, when it was isolated from a hare tick (*Haemaphysalis leporispalustris*) (31). Tularaemia is endemic in Alaska (27), but data on the prevalence of infection in wildlife are scarce. Large predators, such as wolves or bears, constitute very good sentinels of the infection, as they prey on infected hares and rodents. A high prevalence of infection (30% or more) was found in bears in interior Alaska and in the northern region. Conversely, prevalence was much lower on Kodiak Island (4%) and was moderate in the southwestern and western parts of the State. In interior Alaska, antibody prevalence in grizzly and black bears was identical. Central Alaska is an active focus of tularaemia, as illustrated by the recent report of human cases resulting from exposure to domestic cats in the Fairbanks area (20). The prevalence of infection in bears (32%) was also close to that reported for wolves (25%) from interior Alaska (45). In contrast to brucellosis, which seems to be prevalent mainly in western and northern Alaska, tularaemia appears to be more prevalent in interior and northern Alaska. Bears which gave seropositive results for tularaemia were more likely also to give seropositive results

for brucellosis. Antibody cross-reactivity between *Brucella* spp. and *F. tularensis* may be of concern (20). It could explain the reason for the reactions of the three positive bears on Kodiak Island, as two of these had *Brucella* antibodies and low titres for *F. tularensis*. However, the fact that prevalence was very high for tularaemia and very low for brucellosis in bears tested from interior Alaska is supportive of a rather specific test for both infections. The high prevalence of both infections, especially in northern Alaska, could be related to the scavenging habits of the bears. The yearly variations observed in seroprevalence may reflect the cyclical pattern of tularaemia in hare and rodent populations (32).

### Trichinellosis

*Trichinella spiralis* is a common parasite of free-ranging carnivores and omnivores (49). Bears are particularly at risk of acquiring trichinellosis (13), and many reports have been made on the prevalence of infection in black bears in North America (36, 50), as well as in grizzly bears in Alaska (33) and the northern Rocky Mountain area (42). In polar bears, Weyermann *et al.* reported a 61% (56/92) infection rate by *T. spiralis* var. *nativa*, by artificial digestion of masseter muscle samples (41). Serological tests are not able to distinguish between *T. spiralis* and *T. nativa* infection. In Arctic regions, the latter is more commonly found (1). *Trichinella spiralis* var. *nativa* is characterised by low infectivity in laboratory mice, resistance to freezing and confinement to the Arctic and subarctic regions of the world (19). A strong latitudinal trend in the frequency of infection has been reported in Alaskan red foxes, where 46.3% (25/54) of foxes collected in the Brooks Range harboured *T. spiralis*, while only 15.4% (2/13) of those captured from the Copper River drainage showed positive results (33). The present study confirms this observation in grizzly bears. An increasing gradient of infestation from south to north could be observed. The data collected in the present study also support the study recently published by Zarnke *et*

*al.* on seroprevalence in grizzly bears in Alaska captured between 1973 and 1987, which found an antibody prevalence that increased from 5% in the southern region of Alaska to 83% in the northern region (48). However, the present study did not find a major difference in prevalence by age group. In Alaskan lynx (*Felis lynx*), the prevalence of *Trichinella nativa* larvae was the highest (27%) in animals from northern Alaska (47), where a very high seroprevalence in grizzly bears was also found. None of the young grizzly bears (0-2 years) from interior Alaska or the Alaskan Peninsula had *Trichinella* antibodies, whereas most of the black bears which gave positive results were young bears. This observation, which correlates with data on CIH antibody prevalence in grizzly bears, could indicate that young grizzly bears are exposed to infectious diseases much later than black bears and start foraging at a later age. Usually, black bears leave their mother after their second year in the den, whereas young grizzly bears stay with their mother for an additional year (18). However, 13.5% of the grizzly bears from the northern regions were seropositive, which suggests a much earlier exposure to the parasite in these areas. The other possibility could be a different age susceptibility to *T. spiralis* and to *T. nativa*, which is mainly found in the Arctic regions. Rausch *et al.* reported that most infections in man have occurred in the northern regions of the State. The seroprevalence of *T. spiralis* in bears strongly supports that observation, as most of the wildlife species used as a source of meat by humans in these areas are likely to be infected (33).

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## Enquête sérologique portant sur certains virus et agents de zoonoses chez les ours grizzly (*Ursus arctos horribilis*) et les ours noirs (*Ursus americanus*) en Alaska

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Cet article reprend une communication présentée lors de la 8<sup>e</sup> rencontre de la Société internationale d'épidémiologie vétérinaire et d'économie à l'Institut Pasteur, Paris, France, 8-11 juillet 1997

### Résumé

Six cent quarante-quatre échantillons de sérum récoltés en Alaska (États-Unis d'Amérique) sur 480 ours grizzly (*Ursus arctos horribilis*) et 40 ours noirs (*Ursus americanus*) entre 1988 et 1991 furent testés pour la présence d'anticorps contre diverses zoonoses et infections virales canines. La prévalence chez les ours grizzly était de 0 % pour la parvovirose, de 8,3 % (43/480) pour la maladie de Carré,

de 14 % (68/480) pour l'hépatite infectieuse canine, de 16,5 % (78/480) pour la brucellose, de 19 % (93/480) pour la tularémie et de 47 % (225/478) pour la trichinellose. La prévalence en anticorps chez les ours noirs variait de 0 % pour la parvovirose et la maladie de Carré à 27,5 % pour la trichinellose et à 32 % pour la tularémie. Les prévalences en anticorps vis-à-vis de *Brucella* (2,5 %) et de l'agent de la tularémie (32 %) étaient identiques chez les ours grizzly et les ours noirs de la région centrale de l'Alaska. Des différences notables de prévalence furent observées selon l'origine des ours. Les anticorps vis-à-vis des virus de la maladie de Carré et de l'hépatite infectieuse canine étaient surtout observés chez les ours grizzly de l'île Kodiak et de la péninsule d'Alaska. Les anticorps vis-à-vis de *Brucella* étaient principalement observés chez les ours grizzly de l'ouest et du nord de l'Alaska, et les anticorps vis-à-vis de l'agent de la tularémie chez les ours grizzly des régions centrales et arctiques. Un gradient croissant Sud-Nord a été observé pour la présence d'anticorps vis-à-vis de *Trichinella* spp. La prévalence en anticorps augmentait avec l'âge pour la plupart des infections testées. Toutefois, pour quelques-unes de ces maladies aucun anticorps n'était détectable chez les ours grizzly de moins de deux ans alors que ces mêmes maladies affectaient les ours noirs. Les ours grizzly apparaissent être d'excellentes sentinelles pour la surveillance épidémiologique des zoonoses dans la faune sauvage d'Alaska.

#### Mots-clés

Alaska – Brucellose – États-Unis d'Amérique – Hépatite infectieuse canine – Maladie de Carré – Ours grizzly – Ours noir – Parvovirus canin – Trichinellose – Tularémie – *Ursus americanus* – *Ursus arctos horribilis*.

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## Encuesta serológica sobre determinadas infecciones víricas caninas y zoonosis en osos grizzly (*Ursus arctos horribilis*) y osos negros (*Ursus americanus*) de Alaska

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#### Resumen

Entre 1988 y 1991 se tomaron 644 muestras de suero de 480 osos grizzly (*Ursus arctos horribilis*) y 40 osos negros (*Ursus americanus*) de Alaska, Estados Unidos de América. Dichas muestras se sometieron a pruebas de detección de ciertas infecciones víricas caninas y zoonosis. La prevalencia de anticuerpos en los osos grizzly resultó de 0% para los parvovirus, de 8,3% (40/480) para el moquillo, 14% (68/480) para la hepatitis infecciosa, 16,5% (79/480) para la brucelosis, 19% (93/480) para la tularemia y 47% (225/478) para la triquinelosis. En el oso negro, por su parte, la prevalencia oscilaba desde un 0% para el moquillo y los parvovirus hasta el 32% para la tularemia, pasando por un 27,5% para la triquinelosis. Los osos grizzly y negros procedentes del área geográfica del interior de Alaska mostraron idéntica tasa de prevalencia de anticuerpos para la brucelosis (2,5%) y la tularemia (32%). Se observó correlación entre las diferencias en prevalencia y el origen de los osos grizzly. Los anticuerpos contra los virus del moquillo y de la hepatitis infecciosa se detectaron principalmente en osos grizzly de la isla de

Kodiak y la península de Alaska. Los anticuerpos contra la brucelosis, por su parte, eran prevalentes en osos grizzly de las regiones occidental y septentrional de Alaska, mientras que los anticuerpos contra la tularemia se observaron en osos grizzly del interior de Alaska y la zona del Ártico. Se observó asimismo un fuerte gradiente de Sur a Norte de Alaska en el caso de los anticuerpos contra *Trichinella* spp. Para la mayoría de las enfermedades, la prevalencia de anticuerpos aumentaba con la edad. Sin embargo, la ausencia de anticuerpos contra diversas infecciones en osos grizzly de entre 0 y 2 años de edad contrastaba con la presencia de esas infecciones en osos negros. Los osos grizzly resultaron ser excelentes centinelas para vigilar la aparición de infecciones zoonóticas entre la fauna salvaje de Alaska.

#### Palabras clave

Alaska – Brucelosis – Estados Unidos de América – Hepatitis infecciosa canina – Moquillo – Oso grizzly – Oso negro – Parvovirus caninos – Triquinelosis – Tularemia – *Ursus americanus* – *Ursus arctos horribilis*.

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