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ALASKA DEPARTMENT OF FISH AND GAME

JUNEAU, ALASKA

AGE STRUCTURE OF CARIBOU POPULATIONS
IN ARCTIC ALASKA

By: James L. Davis



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FINAL REPORT (RESEARCH)

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Cooperators: James L. Davis, Martha Robus, and Joseph Doerr

Project No.: W-17-9&W-17-10 Project Title: Big Game Investigations

Job No.: 3.22R Job Title: Age Structure of Caribou
Populations in Arctic Alaska

Period Covered: July 1, 1976 to July 30, 1978

SUMMARY

A tooth was sectioned and age was determined for each of 522 caribou taken from the Western Arctic caribou herd between 1959 and 1961 and for 736 caribou taken from the herd in 1975-76. Joseph Doerr, University of Alaska Master's degree candidate, obtained the 1959-61 jaw collection from the University of Alaska Museum. He and Department personnel obtained the 1975-76 collection via village collections. A previously unreported procedure of tooth preparation for aging, using Paragon Multiple Stain for frozen sections, was employed and is described. Age structure of the herd is presented for the two collection periods. A detailed analysis of the data will be presented in a Master's Thesis by Joseph Doerr which will be available from the Cooperative Wildlife Research Unit, University of Alaska, Fairbanks, in late 1978.

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BACKGROUND

Between 1970 and 1976 the Western Arctic caribou (*Rangifer tarandus granti*) herd (WAH) decreased in size from 242,000 (Hemming 1971) to about 65,000 (Davis et al. 1976, Davis et al. in press). The data available implicate human harvest and predation (especially by wolves, *Canis lupus*) as the two greatest mortality factors in this decline. Because annual retrieved human harvest before and after 1970 ranged from 20,000-30,000 animals, many people reasoned that if the herd did not decline earlier at that harvest level then the harvest could not be a factor in the decline. However, with such a heavy harvest, a reduction in recruitment or a recruitment failure during just one year could have precipitated a decline which the unchecked harvest in years following would have sustained. One of the goals of this job was to determine if recruitment had been reduced during the 1970s, using age structure information in lieu of annual herd productivity data. Also, it is essential to have age structure information for computer simulations of possible factors which may have been involved in the herd decline.

OBJECTIVES

To determine the past and present age structure of Alaska's Western Arctic caribou herd by aging hunter-killed animals using the cementum annuli technique.

PROCEDURES

We obtained 522 caribou jaws collected from the WAH between 1959 and 1961 from the University of Alaska Museum collection, and collected an additional 736 jaws from WAH caribou during 1975 and 1976. Most of the jaws in the recent collection were obtained by paying village residents one dollar for each jaw presented. Sex of the caribou was known on about one-half of the jaws in each collection.

We reviewed the tooth sectioning techniques reported by Miller (1974) and Johnson and Lucier (1975) and developed a modified technique that incorporated suggestions and methods from both.

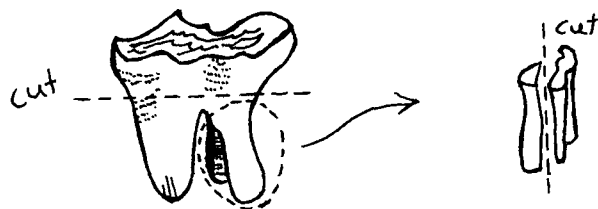
The most discernible tooth to section and read was the first incisor (I_1). When it was not available we used the next longest, I_2 . In some cases we used the I_3 , canine (C), and molars (M_2 and M_3). Some of the

1975-76 collection had been stored dry and some in alcohol. Because traces of alcohol can disrupt the staining procedures, we rinsed the teeth that were in alcohol and allowed them to soak in water for at least a week before going through the decalcification process. The dry teeth from the 1976 collection were taken directly from the envelopes and put into the acid. The older dry teeth (from the 1959-61 collection), however, were so brittle that we finally soaked them in water for several weeks before transferring them into the acid. We hoped that the water would soften them and reduce possible cracking. Nevertheless, the teeth were still badly cracked (especially the molars) and difficult to read.

We placed each tooth in a 10-dram plastic vial that was one-third to one-half full (depending on the size of the tooth) of a 6 percent nitric acid decalcifying solution. The solution consisted of 85 ml of 70 percent nitric acid, 10 ml of 37 percent formaldehyde and 905 ml of distilled water. We stirred the vials twice a day to release gases produced as a byproduct of decalcification. Stirring speeded the softening process. After 24 hours we checked the teeth for flexibility, softness and yellowness; all signs of decalcification. If these signs were present, we sliced each tooth crown transversely with a new razor blade. If the tooth felt at all gritty, rough to the touch, or quite firm, it was replaced in fresh acid solution.

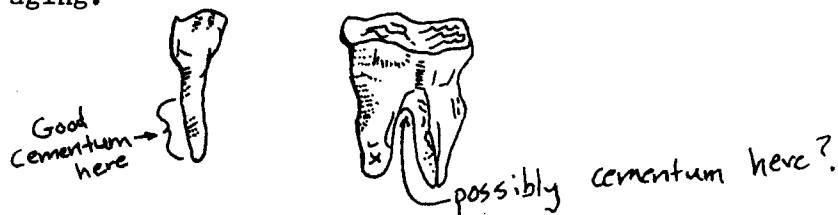
After 24 hours the cementum was usually completely soft because it is located on the tooth surface. The dentine, however, was more apt to contain traces of minerals as it was farther from the acid solution. It was imperative that the tooth be completely soft when sectioned, otherwise slicing it would dull the cryostat blade which resulted in torn and scraped tooth sections. We found that an additional 24 hours of soaking and stirring allowed softening of the tooth interior without cementum damage. Incisors, because of their small size, seldom required decalcification beyond a full 48 hours. Additional decalcification required extreme caution because the cementum was susceptible to disintegration.

Caribou molars are much larger than incisors and we found that they would decalcify more evenly if, when they were soft enough, the crown was cut completely off and the roots were split longitudinally. The crown was discarded and the root pieces were placed back into fresh acid solution. The molars often required up to 96 hours for complete decalcification.



We initially assumed that cementum was deposited on molars the same as on incisors (i.e. thicker, thus more accurately read toward the root apex). We later learned that moose (*Alces alces*) molars have thickest cementum deposition under the crown and along the roots. This might

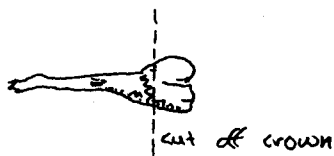
also be true in caribou, as we weren't able to find many molars with sufficient cementum deposition on the lower half of the roots to allow satisfactory aging.



We experimented with a commercial preparation of hydrochloric acid as a decalcifying agent (Scientific Products Decal - 99.2 liter hydrochloric acid) and found no noticeable differences from the nitric acid solution. Since we already had a good supply of nitric acid, we saw no reason to purchase and use the hydrochloric acid preparation. We did not experiment with any other decalcifying agents.

When the teeth were completely decalcified (soft, flexible and yellow) we rinsed them in water for several minutes and then placed them in vials containing water. We again stirred the vials twice a day and substituted fresh water daily. At first we let the teeth soak in water for only 24-48 hours. Later we let the teeth sit for at least a full 72 hours. We found that the stain would not "take" well unless all traces of the acid were removed. As the decalcified tooth soaked in water it gradually became more yellow. After 72 hours a dark yellow color was a good indicator that the tooth was ready to be sectioned.

We employed a freezing microtome (CTD - International Harris Cryostat, at -18 to -20°C) for sectioning the softened teeth. The tooth crown was removed about halfway down as no cementum is deposited here.



Sometimes the peridontal membrane was present. It was desirable to remove this membrane because it got in the way of the processes of mounting and reading the slides. This membrane could be peeled off the teeth in the 1976 collection rather easily. However, we found that forcing it off the older teeth (JD and JGD collections) sometimes resulted in ripping off the cementum also. Therefore we decided that it would be better to leave the peridontal membrane on rather than lose the cementum entirely.

We fixed the prepared teeth to freezing disks with OCT embedding medium and cut longitudinal sections of a thickness of 20 microns. To have a good selection we cut as many sections as possible from the pulp area. Any remaining sections (ones not mounted on slides) were saved in glass vials when there were not extra teeth for a certain jaw.

The cutting process had to be done carefully because if the cryostat blade was dull or dirty it would rip the tissue and leave scratches

across the face of the sections. If the bevel of the blade was improper the sections might be of uneven thickness. This also occurred when the freezing disk was not secured tightly and shifted around as the tooth touched the blade. Sometimes sections might be cut alternately too thick and too thin which could be caused by any one or all of these problems.

We placed the cut sections of each tooth in separate petri dishes of clean water on a black background (which made it easier to see the light-colored sections), and then floated them onto clean glass microscope slides with a dissecting needle. We then blotted the slides dry with a paper towel using firm, gentle pressure, and they were allowed to air-dry for one-half to one hour. It was imperative that the slides dry completely. If they were at all damp the sections would float off the slides when stained. Sometimes when drying, the sections would curl at the edges and might even lift off the slide. This indicated an unclean slide, too much or too little pressure when blotting the sections, or sections with an uneven thickness.

When the sections were completely dry we stained them with Paragon Multiple Stain for Frozen Sections, by flooding each slide with several drops of the stain for 5-10 seconds. The staining time seemed to vary from tooth to tooth. For some it might even take over 10 seconds to darken satisfactorily (later we found these to be teeth that probably did not rinse long enough in water). Excess stain was drained back into the bottle and the slide was dipped in water to halt the staining process. The slide was then placed in another beaker of water for 5-10 minutes to wash off the remaining stain. Leaving the slide in the water for a longer period also tended to bleach it.

We removed the slides from the beaker, blotted them, and allowed them to air-dry overnight. At one point, we tried to speed up this process by coverslipping while the slides were still damp. We discovered that any moisture clouded the sections, so for a better slide, it was worth waiting until they were completely dry.

When the slides were thoroughly dry, we applied coverslips with Permunt permanent mounting medium and allowed them to dry for 24 hours. Later, we stored them in slide boxes.

On an average day, we could section, mount and stain the equivalent of 40 to 50 teeth. A calculation of the cost per slide appears in Appendix I.

We read the slides with a Carl Zeiss microscope at 80x and when necessary at 200x. The best slides resulted from the 1976 collection probably because they had been freshly-pulled teeth. Since these were the first teeth that we processed, some variation in quality occurred because we experimented with decalcification time, cutting, staining time, etc. The older teeth (1959-61 collection) produced much poorer slides probably due to drying, cracking, etc. The most difficult teeth to read were the molars from this collection. They were very brittle and the cementum tended to split, shatter and slough off.

At many times it appeared that some teeth were just plain "ornery": no matter what special precautions were taken the tooth would still be difficult to read. Teeth preserved under similar conditions and treated similarly (decalcified, cut and stained in the same batch) had individual problems. For example, one slide might stain too dark and another too light with the same staining time. One tooth might have distinct, readable cementum annuli where another would have no visible annuli although cementum was present.

RESULTS

Summaries of the age structure in 1959-61 and 1975-76 are presented in Tables 1 and 2. Data from the aged teeth were analyzed by Joseph Doerr, as part of a Master's Thesis to be completed in summer 1978. His thesis contains a detailed discussion of the past and present age structure of the WAH, and it will be available in late 1978 through the Alaska Cooperative Wildlife Research Unit, University of Alaska, Fairbanks.

These data will also be used in the future in computer modeling programs which will be used to simulate the past, present, and future population dynamics of the WAH.

ACKNOWLEDGMENTS

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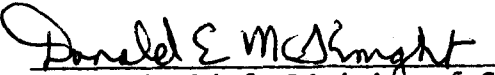

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Table 1. Estimated ages of hunter-killed caribou from the Western Arctic herd (1959-61).¹

Age	Male		Female		Unknown Sex		Total	
	No.	%	No.	%	No.	%	No.	%
2-3	14	(10.8)	38	(21.6)	37	(17.1)	89	(17.0)
3-4	12	(9.2)	29	(16.5)	43	(19.9)	84	(16.1)
4-5	24	(18.5)	33	(18.8)	41	(19.0)	98	(18.8)
5-6	30	(23.1)	19	(10.8)	30	(13.9)	79	(15.1)
6-7	19	(14.6)	25	(14.2)	19	(8.8)	63	(12.1)
7-8	9	(6.9)	13	(7.4)	14	(6.5)	36	(6.9)
8-9	9	(6.9)	11	(6.2)	9	(4.2)	29	(5.6)
9-10	4	(3.1)	5	(2.8)	10	(4.6)	19	(3.6)
10-11	5	(3.8)	1	(0.6)	5	(2.3)	11	(2.1)
11-12	4	(3.1)	1	(0.6)	4	(1.9)	9	(1.7)
12-13	0	(0.0)	1	(0.6)	2	(0.9)	3	(0.5)
13-14	0	(0.0)	0	(0.0)	1	(0.4)	1	(0.2)
14-15	0	(0.0)	0	(0.0)	1	(0.4)	1	(0.2)
Total	130		176		216		522	

¹ Collected in the villages of Anaktuvuk Pass, Kivalina, Noatak, and Point Hope.

Table 2. Estimated ages of hunter-killed caribou from the Western Arctic herd (1975-76).¹

Age	Male		Female		Unknown Sex		Total	
	No.	%	No.	%	No.	%	No.	%
2-3	17	(8.6)	14	(11.7)	64	(15.3)	95	(12.9)
3-4	28	(14.2)	11	(9.2)	64	(15.3)	103	(14.0)
4-5	34	(17.3)	18	(15.0)	80	(19.1)	132	(17.9)
5-6	45	(22.8)	13	(10.8)	65	(15.5)	123	(16.7)
6-7	35	(17.8)	20	(16.7)	41	(9.8)	96	(13.0)
7-8	19	(9.6)	11	(9.2)	39	(9.3)	69	(9.4)
8-9	11	(5.6)	11	(9.2)	35	(8.4)	57	(7.7)
9-10	4	(2.0)	10	(8.3)	15	(3.5)	29	(3.9)
10-11	2	(1.0)	6	(5.0)	5	(1.2)	13	(1.8)
11-12	0	(0.0)	4	(3.3)	6	(1.4)	10	(1.4)
12-13	1	(0.5)	2	(1.7)	3	(0.7)	6	(0.8)
13-14	0	(0.0)	0	(0.0)	2	(0.5)	2	(0.3)
14-15	1	(0.5)	0	(0.0)	0	(0.0)	1	(0.1)
Total	197		120		419		736	

¹ Collected in the villages of Ambler, Barrow, Evansville, Kiana, Kivalina, Kotzebue, Noorvik, Point Hope, Selawik, Shungnak, and in the Kiana Hills.

Appendix I.

COST/SLIDE

Assuming : 1 slide/tooth

Slides:	1 gr. @ \$6.10	6.10/144 slides	= \$.04
Coverslips:	1 oz. @ \$2.25	2.25/48 slides	= .05
Vials:	1 gr. @ \$19.40	19.40/144 slides	= .13
Oct Comp'd:	12 bottles @ \$26.00	2.17/100 slides	= .02
Paragon stain:	50 ml bottle @ \$2.50	2.50/300 slides	= .01
70% HNO ₃ :	7 lb. @ \$17.64	17.64/550 slides	= .03
Slide boxes:	1 box @ \$5.75	5.75/100 slides	= .06
Permout:	1 pt. @ \$3.90	3.90/1661 slides	= .01
Temp. Help:	\$58.65/day	58.65/25 slides*	= <u>2.35</u>
			\$ 2.70

* Includes both sectioning and reading and miscellaneous lab work.