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## AGE-RELATED BODY MASS AND REPRODUCTIVE MEASUREMENTS OF GRAY WOLVES IN MINNESOTA

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Based on 65 free-ranging gray wolves (*Canis lupus*) of known age and 25 of estimated age examined during summers of 1970–2004 in northeastern Minnesota, body mass of both males and females peaked at 5 or 6 years of age, with mean masses of 40.8 kg and 31.2 kg, respectively. Testis size varied as a function of age and month through at least 8 years of age, with length plus width ranging from 1.9 to 7.8 cm. Most females aged 4–9 years bred based on assessment of nipple sizes; those that had not bred had average lower body mass than those that had. This is the 1st report of such data from known-aged wolves.

Key words: body mass, *Canis lupus*, development, gray wolf, growth, nipples, reproduction, testes

Information about development and mass changes of free-ranging gray wolves (*Canis lupus*) has been scattered, anecdotal, or piecemeal. Primarily this has been because wolves are long-lived, were scarce and difficult or expensive to live-capture (Mech 1974), and because aging techniques for live wolves had not been developed. Wolf pup growth and development was described for captive (Mech 1970; Pulliainen 1965) and free-ranging (Van Ballenberghe and Mech 1975) animals. Seasonal mass change (Seal and Mech 1983) and nipple measurements (Mech et al. 1993) of wolves with known reproductive histories were documented for captive wolves but were not related to ages. Testis measurements separating yearling wolves from older animals have been published, but no data were presented for various-aged adults (Gese and Mech 1991). Testis size has been said to vary with season, but no data were given (Seal et al. 1987).

However, as a result of a study in which free-ranging wolves have been livetrapped, examined, ear-tagged, and radiotagged from 1968 through 2004 (Mech 1979, 2000), enough known-aged pups were so marked and then recaptured as adults of various known ages that data are now available relating wolf measurements to age. In addition, the development of tooth-wear charts from known-aged wolves (Gipson et al. 2000) allowed accurate age estimates of livetrapped wolves, which yielded additional data (Mech, in press). I thus present here information about the body mass and testis and nipple measurements of free-ranging Minnesota wolves of various ages.

### MATERIALS AND METHODS

The study area encompassed some 2,060 km<sup>2</sup> immediately east of Ely in the east-central Superior National Forest (48°N, 92°W) of northeastern Minnesota. Topography varies from large stretches of swamps to rocky ridges, with elevation ranging from 325 to 700 m above sea level. Winter temperatures below –35°C are not unusual, and snow depths (usually from about mid-November through mid-April) generally ranged from 50 to 75 cm on the level. Temperatures in summer rarely exceeded 35°C.

Conifers predominate in the forest overstory, with the following species present: jack pine (*Pinus banksiana*), white pine (*P. strobus*), red pine (*P. resinosa*), black spruce (*Picea mariana*), white spruce (*P. glauca*), balsam fir (*Abies balsamea*), northern white cedar (*Thuja occidentalis*), and tamarack (*Larix laricina*). However, as a result of extensive cutting and fires, much of the conifer is interspersed with large stands of paper birch (*Betula papyrifera*) and aspen (*Populus tremuloides*). Detailed descriptions of the forest vegetation were presented by Ohmann and Ream (1969).

Wolves in the study area fed primarily on white-tailed deer (*Odocoileus virginianus*), moose (*Alces alces*), and beavers (*Castor canadensis*—Frenzel 1974). The wolves have been legally protected since 1974, although some have occasionally been killed accidentally or illegally by humans (Mech 1977). The wolf population in the study area has remained relatively stable since about 1975, after dropping following a major deer decline (Mech 2000:23; L. D. Mech, in litt.). Because the study population has long been saturated, most of the 940 wolves examined during the tenure of the wolves in the current study possessed little back fat. In the late 1970s, canine parvovirus infected the population, resulting in a strong decrease in pup survival ever since (Mech and Goyal 1995).

The taxonomic identity of the wolves in this study is uncertain. Nowak (1995) considered them *Canis lupus nubilus* based on skull morphology. However, molecular genetic analysis indicated that my study population includes animals identified as being of the same mitochondrial DNA haplotypes as some wolves in Alaska and western

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**TABLE 1.**—Body mass (kg) of free-ranging wolves of known ages ( $n = 65$ ) and estimated ages ( $n = 25$ ) from the Superior National Forest of northeastern Minnesota.  $P$  is probability that difference between males and females is due to chance.

Age (years)	Males				Females				$P$
	$n$	$\bar{X}$	$SE$	Range	$n$	$\bar{X}$	$SE$	Range	
1	21	30.6	1.2	22–39	11	25.3	1.2	19–20	<0.01
2	5	32.0	2.7	25–41	5	26.4	2.0	19–31	0.07
3	6	36.8	1.5	33–42	3	32.3	1.7	29–34	0.06
4	7	35.1	1.6	30–43	7	28.7	1.9	22–35	0.01
5	4	40.8	1.5	37–44	5	31.2	2.4	24–39	<0.01
6	1	40.0							
7	3	37.3	0.3	37–38	3	27.0	1.0	25–28	<0.001
8	2	38.5	3.5	35–42					
9	2	34.0	5.0	29–39	2	29.0	5.0	24–34	0.28
10				1		26.7			
11				1		28.2			
12				1		25.9			

Canada, as well as animals with coyote (*Canis latrans*)–like haplotypes also found in western Ontario (Lehman et al. 1991). Nevertheless, no morphological differences between wolves of these 2 mitochondrial DNA haplotypes have been recognized, formally or informally, and individuals of both haplotypes inhabit the same packs (Lehman et al. 1992). A newer genetic analysis suggests that Minnesota wolves may be attributable to a newly postulated species, *Canis lycaon* (Wilson et al. 2000), and a less powerful genetic test was consistent with the population being *Canis lupus* or hybrids between *Canis lycaon* and *Canis lupus* (Mech and Federoff 2002).

Wolves were livetrapped in modified steel foot traps (Mech 1974) from about 16 wolf packs throughout the study area from June through November 1968 through 2004. (Movements of wolves between packs and formation of new packs prevent an exact portrayal of the number of packs represented.) The wolves were anesthetized, weighed on a spring scale (Chatillon 160, Largo, Florida, until 1999; and Salter ABS, Santee, California, since then), radiocollared (Telonics Inc., Mesa, Arizona), and examined, and a testis or nipple was measured. For testis measurements, a single testis was forced taut against the scrotum, and a caliper was used to measure its length and then its width while the testis was held taut. The length and width were then added together for analysis. At the suggestion of an anonymous reviewer, I also analyzed differences in testes sizes by using lengths and widths to better represent testes as prolate spheroids, using the formula:

$$\frac{4}{3} \pi ab^2,$$

where  $a$  is testis length and  $b$  is testis width.

On females, the length and width of the largest nipple (usually inguinal) were measured with calipers, and the sum was compared with data from captive wolves of known reproductive history (Mech et al. 1993) to estimate breeding status.

Pups were distinguished by their milk teeth or newly erupted adult canines (Van Ballenberghe and Mech 1975). All animals were ear-tagged (National Band and Tag Co., Newport, Kentucky); most of those >11 kg were radiocollared. Marked pups recaptured later then represented known-aged animals. From 1968 through 2000, individuals older than pups were considered to be of unknown age; however, when these individuals were recaptured, the number of years between captures were added to 1 to estimate their known minimum ages. For analysis, data from only 2 known minimum-aged animals were used,

female wolf 2407 at minimum ages 10 and 12 (2 recaptures), and female 5429 at minimum age 7. To increase the sample for animals 3–9 years old, wolves with estimated ages based on tooth wear were added. From 2000 to 2004, the age of each wolf was estimated in the field by comparing its tooth wear to laminated illustrations of tooth-wear patterns of known-aged wolves (Gipson et al. 2000). For most wolves, a single-year estimate was recorded, but in 6 cases where a range of years was recorded (e.g., 3–5 years), I assumed the midrange to be the actual age. When a range between consecutive years was recorded (e.g., 3–4 years), I allocated the data to the younger age (13 cases).

I used  $t$ -tests to compare male and female mass, simple linear and polynomial regression to assess annual change in mass, and multiple regression to assess differences in testis size. Testis size is thought to change seasonally (Seal et al. 1987), peaking during the breeding season, which in my study area is February (Mech and Knick 1978). Thus, I hypothesized that testis size would decrease to a nadir in July and August and increase thereafter. For the multiple regression, I coded capture months as follows: July and August, 1; June and September, 2; May and October, 3. The 2nd independent variable was age.

A total of 39 known-aged males 1–9 years old and 26 known-aged females 1–12 years old were captured from 1970 to 1995 and were weighed and measured. There was a dearth of known-aged animals after 1997 in an ongoing study, which may reflect a reduced number of pups captured after canine parvovirus affected the study population (Mech and Goyal 1995). Body mass of 12 males and 13 females with estimated ages (Gipson et al. 2000; Mech, in press) or whose minimum ages were known also were available (Table 1). Testis measurements were available for 43 animals, and nipple sizes for 35. This research was conducted under both state and federal endangered species permits and complied where applicable with guidelines of the American Society of Mammalogists (Animal Care and Use Committee 1998).

## RESULTS

Body mass of males was significantly higher than mass of females for most ages, although both were highly variable (Table 1). Mass of males and females increased annually from 1 year of age to 5 or 6 years and then appeared to decline (Figs. 1 and 2). Testis measurements (length plus width) increased significantly through at least 8 years of age ( $r^2 = 0.19$ ,  $P < 0.01$ ) from 1.9 cm for a yearling in September to 7.8 cm for a 3-year-old in September (Fig. 3). Adding code for month of capture to the analysis increased the relationship to  $R^2 = 0.34$  ( $P = 0.0001$ ). Representing the testes as prolate spheroids, the comparable figures were  $r^2 = 0.07$  ( $P = 0.09$ ) for relationship between testes and age, and  $R^2 = 0.27$  ( $P = 0.0007$ ) when month of capture was added.

Nipples on all but 1 yearling and on 6 females 3–9 years old were inconspicuous and unmeasurable (Table 2). Those that were measurable varied (length plus width) from 0.5 cm on a 3-year-old caught in June to 3.3 cm on the same individual as a 4-year-old caught the following June (Table 2). Based on nipple size (Mech et al. 1993), females estimated to have produced pups were all 4–9 years old, except for two 2-year-olds. Both of these apparently produced pups, but, if they did, they had lost them by early summer (Tables 2 and 3). The 4- to 9-year-old females that had produced pups were an average of 4% above the average mass for their age, whereas three 5- to 9-year-old females estimated by nipple size to not have produced pups averaged 21% below the mean mass for their ages ( $P = 0.06$ ).

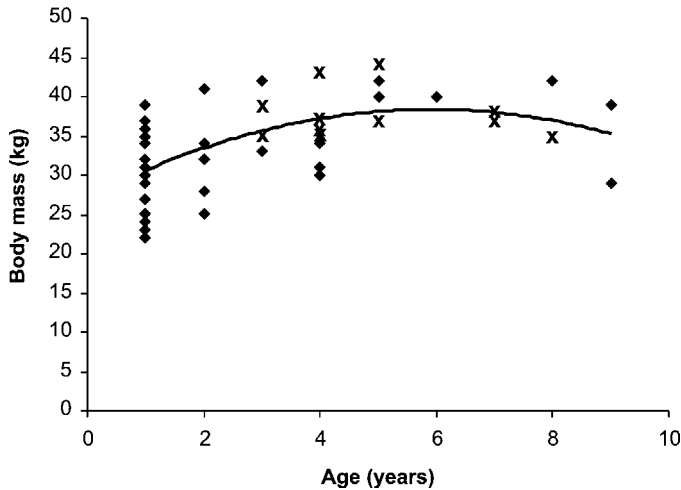


FIG. 1.—Relationship between body mass and age in male wolves from the Superior National Forest of northeastern Minnesota. Diamonds = known-aged wolves, and X = wolves of estimated age ( $r^2 = 0.31$ ,  $P = 0.0002$ ,  $y = 27.14 + 3.8x - 0.32x^2$ ).

### DISCUSSION

These findings indicate that wolves probably are not fully mature developmentally until about 5 years old. Body mass of both males and females peaked at 5 years of age. Because only 1 wolf known to be 6 years old was examined, conceivably mass could have peaked at 6 years. In any case, mass appeared to decline after 5 years of age in both males and females, although samples of older animals were small. As wolves grow and develop, their milk canines are replaced by adult teeth at about 6 months of age (Mech 1970; Van Ballenberghe and Mech 1975). The epiphyseal cartilage of their long bones ossifies by 12–14 months (Rausch 1967), so their stature is fixed by then. However, examination of my data shows that, at least from summer to summer, wolves continue to increase in mass until 5 or 6 years old, presumably by increasing muscle, bone, and fat mass.

Both males and females can breed at 10 months of age (Medjo and Mech 1976), although in the wild they do not usually breed until at least 2 years of age (Rausch 1967), and females, sometimes not even when 3 years old (Mech and Seal 1987). In the present study, all known-aged females at least 4 years old had nipple measurements indicative of having bred (Mech et al. 1993), but females with estimated ages of 4–9 years had nipples indicating that they had not bred (see below).

Testes increased throughout the 8-year span for which I had both known-aged animals and testis measurements. The true relationship between age and testis size was no doubt tighter than examination of my data showed (Fig. 3); the relationship was obscured by the seasonal changes that wolf testes undergo (Seal et al. 1987) because specimens were measured from May through October. These seasonal changes were documented by the increased strength of the relationship between age and size when capture month was considered.

Of further interest were apparent discrepancies between nipple measurements of my animals and similar measurements of captive wolves of known breeding histories. All the apparent discrepancies involved 3 animals aged 4–9 years according to

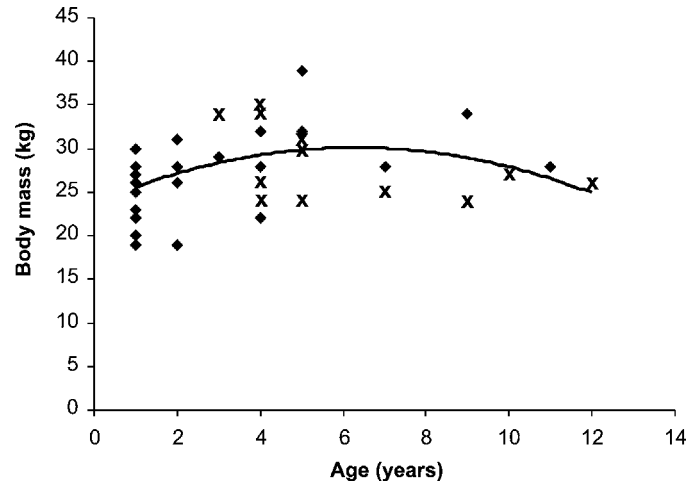


FIG. 2.—Relationship between body mass and age in female wolves from the Superior National Forest of northeastern Minnesota. Diamonds = known-aged wolves, and X = wolves of estimated age ( $r^2 = 0.15$ ,  $P = 0.05$ ,  $y = 23.74 + 2.02x - 0.16x^2$ ).

tooth wear (Gipson et al. 2000). These animals were all deemed by field technicians to have nipples too small to measure (Table 2), which indicated that they had never bred (Mech et al. 1993). Such a conclusion conflicts with the fact that all known-aged females of these ages had nipples of sizes indicating that they had bred (Table 2). The females of estimated age were examined in 3 different summers, during which 3 different groups of field technicians estimated their ages. Thus, biased observers could probably be ruled out. In addition, the estimated ages included those showing so much tooth wear that it seems highly unlikely that the technicians would have mistakenly judged nonbreeding 1- to 3-year-old wolves, which generally show little tooth wear (Gipson et al. 2000), to be several years older. Evidence that the age estimates of the 4- to

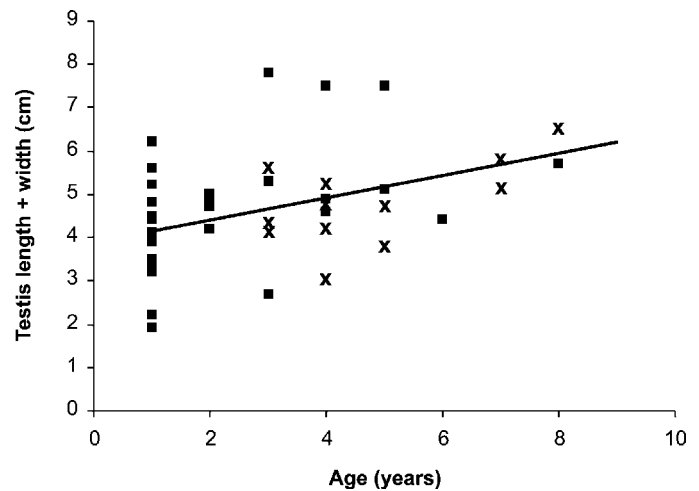


FIG. 3.—Relationship between testis size and age of wolves of known and estimated age in the Superior National Forest of northeastern Minnesota. Squares = known-aged wolves, and X = wolves of estimated ages ( $r^2 = 0.19$ ,  $P < 0.01$ ). When code for month of capture is used in a multiple regression,  $R^2 = 0.34$ ,  $P = 0.0001$ .

**TABLE 2.**—Nipple measurements (length plus width in centimeters of largest nipple) and inferred reproductive status (Mech et al. 1993) of wolves of known and estimated ages from the Superior National Forest of northeastern Minnesota, 1976–2004.

Wolf no.	Capture date	Age (years)	Nipple (cm)	Breeding status	Body mass (kg)	% Difference from mean mass for age
401	28 April 1992	1 <sup>a</sup>	0.6	Nonbreeder	28	+11
413	26 June 1987	3	0.5	Nonbreeder	34	+5
413	22 June 1988	4	3.3	Breeder	32	+11
413	15 November 1989	5	2.5	Breeder or former	39	+25
743	5 August 2002	4	1.5	Breeder or former	28	-2
827	10 August 2002	5 <sup>b</sup>	1.1	Unknown	30	-4
845	7 June 2002	3 <sup>b</sup>	<sup>c</sup>	Nonbreeder	34	+5
879	13 June 2002	5 <sup>b</sup>	1.7	Former breeder	31	-1
887	15 August 2002	9 <sup>b</sup>	<sup>c</sup>	Nonbreeder	24	-34
893	23 August 2004	4 <sup>b</sup>	1.9	Breeder	35	+22
903	21 June 2003	4 <sup>b</sup>	<sup>c</sup>	Nonbreeder	34	+18
905	25 June 2004	4 <sup>b</sup>	2.0	Breeder	24	-16
909	5 July 2003	5 <sup>b</sup>	<sup>c</sup>	Nonbreeder	24	-23
913	22 July 2003	7 <sup>b</sup>	<sup>c</sup>	Nonbreeder	25	-7
923	17 July 2004	4 <sup>b</sup>	<sup>c</sup>	Nonbreeder	26	-9
2407	14 August 1980	10 <sup>d</sup>	1.0	Former breeder	27	
2407	28 August 1982	12 <sup>d</sup>	1.1	Former breeder	26	
5139	29 July 1976	2	1.6	Former breeder <sup>e</sup>	19	-28
5176	26 June 1976	2	1.5	Former breeder <sup>e</sup>	31	+17
5176	11 September 1979	5	1.2	Breeder	32	+3
5176	26 July 1981	7	1.3	Breeder	28	+4
5176	12 October 1983	9	1.2	Former breeder	34	+17
5176	22 August 1985	11	1.2	Former breeder	28	
5415	16 September 1978	3	1.2	Nonbreeder	29	-10
5429	6 September 1981	7 <sup>d</sup>	0.9	Unknown	28	+4
6439	14 August 1986	4	1.2	Former breeder	22	-23

<sup>a</sup> Ten other known-aged yearlings all had nipples too small to measure.

<sup>b</sup> Estimated age based on tooth wear (Gipson et al. 2000).

<sup>c</sup> Too small to measure.

<sup>d</sup> Known minimum age.

<sup>e</sup> Pups lost before summer.

9-year-old animals based on tooth wear were probably accurate and that they actually had not bred could be found in the fact that the mass of those animals had averaged 21% below the means for their ages. The known breeders of known age averaged 4% above. Thus, the apparent lack of breeding in the 3 animals of estimated age may have resulted from their poor body condition (Boertje and Stephenson 1992).

The known-aged females were all examined before 1993 (Table 2), whereas the animals of estimated ages were examined in 2002–2004. Conceivably some recent unexplained population phenomenon resulted in the 3 animals ostensibly born from 1993 to 2000 never having bred. However, if so, this phenomenon did not result in a measurable population change (Mech 2000; L. D. Mech in litt.). Thus, this finding remains an enigma.

Because no other literature is available relating wolf age to body mass and reproductive measurements (Kreeger 2003), this study provides new insight into the life history and reproductive physiology of wolves.

#### ACKNOWLEDGMENTS

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**TABLE 3.**—Summary of female wolf breeding status as inferred from nipple measurements (Table 2).

Age (years)	<i>n</i>	Nulliparous	Parous
1	11	11	0
2	2	0	2 <sup>a</sup>
3	3	3	0
4	7	2	5
5	4	1	3
7–12	7	2	5

<sup>a</sup> Nipples indicated they had produced pups, but if so, pups did not survive into summer.

Agriculture North Central Research Station. M. E. Nelson and numerous volunteers livetrapped the study animals and performed the age estimates. G. D. DelGiudice, D. R. MacNulty, and M. E. Nelson reviewed the manuscript and offered helpful suggestions for improving it.

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