Polar Biol (2002) 25: 878–883
DOI 10.1007/s00300-002-0430-7

ORIGINAL PAPER

A. Rosing-Asvid · E.W. Born · M.C.S. Kingsley

Age at sexual maturity of males and timing of the mating season of polar bears (*Ursus maritimus*) in Greenland

Received: 17 December 2001 / Accepted: 29 July 2002 / Published online: 28 September 2002

Abstract Age at sexual maturity among male polar bears (*Ursus maritimus*) and timing of the mating season were studied by microscopic and macroscopic analysis of the reproductive organs from 165 male and 92 female Greenland polar bears. Males became sexually mature in the spring following their fifth birthday. Testes sampled during the spring reached 95% of their asymptotic length at 5.8 years. Some spermatozoa were found in 2- to 4-year-old bears during the spring, but only in low concentrations in a mixture of other cell types. The first corpus luteum was found on 1 April and most ovulations were estimated to occur before mid-May which indicate that the peak of the mating season begins in late March and ends in May. These findings are somewhat different from a similar study from Svalbard, where the mating season is estimated to peak later and male polar bears are found to become mature earlier. We hypothesize that these differences occur because adolescent males have their peak testicular activity after the main breeding period and therefore appear to be fully sexually mature during summer and not during spring.

Introduction

Histological examination of testes indicates that male polar bears (*Ursus maritimus*) become sexually mature by 3 years of age in both Svalbard and Alaska (Lønø 1970; Lentfer et al. 1980). However, as a result of competition from older larger males, it is unlikely that these younger males get an opportunity to mate before they reach physical maturity at about 6 years of age (DeMaster and Stirling 1981).

The timing of the polar-bear mating season is somewhat unclear. Lønø (1970) reported that male fighting, courtship and matings have been observed between 8 March and 20 June around Svalbard. However, on the basis of histological examination of testes, he suggested that the mating season commences at the end of April and continues to mid-July, with peak activity during 3 weeks, beginning in the middle of June. In addition, Palmer et al. (1988) found elevated levels of serum steroid concentrations in Canadian male polar bears from April and May compared to March and August/November, and a strong correlation between age and testosterone concentrations in these 2 months. However, their study provided no data from June/July, so it remains unclear whether these results apply in general. There is no study on this subject for bears from Greenland and Alaska, but oestrous females and behaviour related to mating have been reported from early March to mid-May (Pedersen 1931; Lentfer 1982). The combined data suggest that the mating season is spread over a relatively long period and coincides with the time when food is easily accessible and energy resources are accumulated (Stirling and Øritsland 1995). To understand the life-history strategies of male polar bears, it is therefore important to know whether oestrus of polar-bear females is spread evenly over this period or whether there is a short, well-defined mating season, with only a few cases of oestrus outside this period.

In the present study, we investigate the sexual maturity of male polar bears by microscopic and macroscopic analysis of a large set of male reproductive organs from East and Northwest Greenland. Analyses of these organs and a smaller number of reproductive organs from adult females are used to estimate timing and duration of the mating season for polar bears in Greenland.

A. Rosing-Asvid (✉) · E.W. Born · M.C.S. Kingsley
Greenland Institute of Natural Resources,
P.O. Box 570, 3900 Nuuk, Greenland
E-mail: Aqqalu@natur.gl
Fax: +299-325957

A. Rosing-Asvid
University of Copenhagen,
Department of Population Ecology,
Universitetsparken 15, 2100 Copenhagen, Denmark

Materials and methods

Sampling in the field

During the years 1988–1996, reproductive organs, premolar teeth and various soft tissues from polar bears were collected during the subsistence hunt in Northwest and East Greenland (Born 1998). The hunters were asked to retain the testes and epididymides of males and the entire reproductive tract (ovaries and uterine horns down to the entrance to the vagina) of females. Reproductive organs from 165 males and 92 females were collected. However, sample size is smaller in some of the analyses, because many of the female organs were not dissected completely during the sampling. All tissue samples were stored frozen until investigated.

Preparation of organs and tissues in the laboratory

The age of 135 male and 61 female bears was determined by counting annuli in the cementum of the lower 1st premolar tooth (Calvert and Ramsay 1998).

Male reproductive organs were thawed, after which the epididymis was separated from the testis. Both organs were measured to the nearest millimetre and weighed to the nearest milligramme before fixation. A cross section from the centre of 100 sets of testes and epididymides from different age categories and seasons was dissected and fixed with buffered formalin, dehydrated and embedded in paraffin. One- to 2-μm-thick sections were subsequently obtained and stained with haematoxylin and eosin. Some damage to the connective tissue caused by freezing was evident, but the seminiferous and epididymal tubules were intact in most samples. The diameters of these tubules were measured by taking the mean of 5 randomly chosen seminiferous and epididymal tubules at ×250 magnification. The presence or absence of spermatozoa in the epididymis was noted.

The uterine horns of females were dissected longitudinally and their lumens checked for placental scars. The ovaries were preserved in formalin or ethanol, and cut in approximately 1.5-mm-thin slices that were kept attached to the base of the ovary. The number of follicles of various size categories (<1 mm, 1–2 mm, 2–3 mm, 3–6 mm and >6 mm) was counted; corpora lutea were identified and their maximal diameter was measured. No follicles in the interval between 5 and 11 mm were found, indicating that the rapid growth of ripe follicles starts shortly after the follicle has reached 5 mm. The follicles that we found in the 11- to 16-mm interval were therefore considered close to the size of rupture and categorized as ripe follicles. Individuals were considered to be sexually mature if they had either: (1) a ripe follicle, (2) a corpus luteum, (3) a placental scar, (4) were accompanied by cubs, or (5) were older than 5 years of age, at which age most female polar bears from other populations are mature (Lentfer 1982; Ramsay and Stirling 1982, 1988; Furnell and Schweinburg 1984; Derocher et al. 1992; Derocher and Stirling 1995).

Statistical analyses

Sigmoidal growth curves were used to describe the age-related increase in testis length and epididymis weight. Preliminary analyses showed no difference between samples from East and West Greenland, but size of these organs varied seasonally, being larger during the active period in spring and smaller during autumn. Hence growth curves were fitted separately to the two data sets: 105 samples from spring (January/July) and 29 from autumn (August/September). An initial general model fitted a general sigmoidal curve (Richards 1959) with a power-law relation between the deviation from the prediction and the predicted testis length: $y=(\theta,a)+\epsilon$, where ϵ was distributed with variance proportional to $\hat{y}$. This initial model was simplified: the general curve differed only slightly from a Gompertz curve (Gompertz 1825), which was therefore used. Testis size was expected to be the same, on average, at age zero for bears killed in spring as for those killed in autumn, and this expectation was borne out by statistical tests on the difference between separately fitted intercepts. Therefore, the length-age curves for testis size against age for the two seasons were constrained to have the same intercept on the testis-length axis.

Unpaired *t*-tests and one-way ANOVAs were used to test for differences in organ size among various age groups, using GraphPad InStat, whereas regression analyses were performed in Microsoft Excel.

Results

Males

Age-related changes

In the testes of 1-year-old polar bears ($n=4$), no signs of spermatogenesis were found in the seminiferous tubules and the body of the testis was mainly occupied by connective tissue with a few leydig cells.

In all testes of 2-year-old males ($n=9$), spermatogenesis had been initiated as spermatogonia and spermatids were present. Two of the seven 2-year-old males caught from mid-February to end of May had spermatozoa mixed with other unidentified cell types in the epididymis. Five out of ten 3-year-old and four of the nine 4-year-old male polar bears from the same period had spermatozoa in the epididymis, but only in low concentrations and as a mixture with other cell types. Spermatozoa were present in all the 38 examined males 5 years and older caught in the period 13 February to 9 August, except for 1 animal (see next section).

At all ages, the diameter of the seminiferous tubules was positively correlated with testis length. This relationship was closest among sub-adults (1- to 4-year-old bears): $r^2=0.72$, $P<<0.001$, $n=50$, because the width of the tubules and the size of testes in this group are related both to season and maturity (very small testes are immature and therefore have tubules with little diameter), whereas the difference in testes size among adults is smaller and partly related to season and individual differences (5+ years): $r^2=0.14$, $P<0.05$, $n=45$. Similarly, the diameter of the ductus epididymis was correlated with the weight of the epididymis for both groups: sub-adults $r^2=0.44$, $P<<0.001$, $n=42$; adults (5+ years) $r^2=0.13$, $P<0.05$, $n=40$. Samples from West and East Greenland showed no significant difference in mean testis length in any single age group (Table 1). Also, separate fits of the Gompertz growth model to samples from east and west, when compared with the fit to the pooled data (Fig. 1), gave a log likelihood ratio of 3.856, with 5 constraints ($df=5$, $\chi^2=7.712$, $P=0.17$). Data from the two areas were therefore pooled for all analyses.

The age-dependent growth in testis length differed significantly between samples taken during spring and autumn, with spring length being greater at all ages. The shapes of both growth curves were similar, with a deceleration in growth between 5 and 10 years of age

Table 1 Length of testes from aged polar bears (in mm) caught in East and West Greenland, 1988–96

		January/July		August/December	
		East	West	East	West
1 year of age	Mean	32.4	36.5	35.2	
	SD	2.6	3.5		
	Min-Max	29–36	34–39	35.2	
	N	5	2	1	
2 years of age	Mean	43.8	47.1	39.2	45.5
	SD	8.0	7.5	5.5	4.9
	Min-Max	32–56	38–57	33–46	42–49
	N	13	6	4	2
3 years of age	Mean	59.0	55.1	52.2	
	SD	9.2	6.0	8.6	
	Min-Max	43–72	45–62	42–62	
	N	18	6	7	
4 years of age	Mean	67.9	59.8	47.0	
	SD	5.1	3.2	6.2	
	Min-Max	57–73	58–62	42–54	
	N	9	2	3	
5+ years of age	Mean	74.5	72.1	66.2	61
	SD	6.1	5.6	4.1	
	Min-Max	65–88	64–80	62–73	61
	N	29	12	9	1

(Fig. 1). This pattern was also seen in the growth in epididymis weight (not shown). In the spring sample, 95% of asymptotic testis length was reached by the age of 5.8 years. In the epididymis, 86% of asymptotic weight (86% because weight goes roughly as the cube of length) was reached by about 5.5 years of age. No correlation was found between age and testis length in bears older than 5 years (spring samples: $df = 37$, $r^2 < 0.007$, $P = 0.62$).

Seasonal changes

January–February. Spermatogenesis had started in the earliest sample from this period (18 January) and, in the next 8 bears from 13–29 February, all stages from germ cells to spermatozoa were found. The epididymis of all individuals contained some spermatozoa together with various other unidentified cell types.

March–May. Twenty-five samples were available from 1 March to 12 May. In March, all but one sample contained highly differentiated seminiferous tubules and spermatozoa in the epididymis. The odd sample was from a 9-year-old male killed on 1 March and reported to be very skinny. In this sample, the seminiferous tubules had formed but the germ cells had not yet started to differentiate. In the other 5 samples from 1–8 March, the epididymides contained a mixture of cells but the 18 samples from 15 March to 12 May all had very dense concentrations of spermatozoa.

June–July. Three samples were available from these months. Spermatozoa were still detected and, in a sample from 24 July, a high concentration was found (similar to what was typically found in April–May).

August–December. Eight samples were available for this period. Few spermatozoa were found in 2 samples from 8 August and 19 September, respectively, but none was found in 4 samples from intermediate dates or in the 2 samples (1 from November and 1 from December). Signs of degeneration of the tubules were apparent in all the samples. The tubules gradually collapsed and loose connective tissue constituted an increasing fraction of the testis.

There was no significant difference between length or weight of testes when comparing the periods January/February, March/May and June/July (One-sided ANOVA; Length: $F_{2,\ 53} = 1.117$, $P = 0.33$, Weight: $F_{2,\ 53} = 0.5130$, $P = 0.60$), but when data from the period August/December were included (Table 2), the test became significant (Length: $F_{3,\ 66} = 7.049$, $P < 0.0004$; Weight: $F_{3,\ 66} = 7.736$, $P < 0.0002$). Mean lengths of testes from the three reproductive stages from January to July were all significantly greater than in the August–December sample ($P < 0.05$).

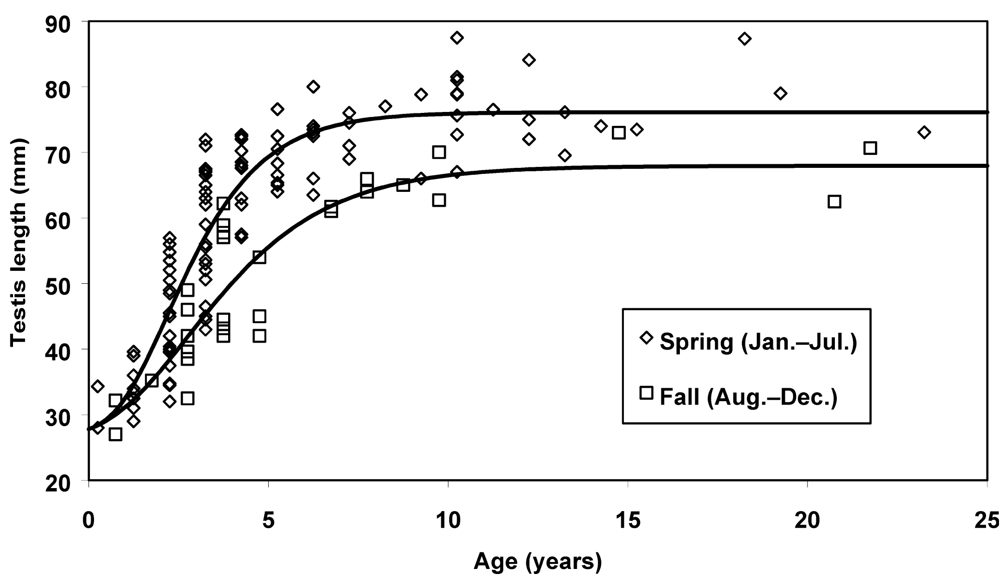

Fig. 1 Gompertz growth model fitted to seasonal data on testis length of 134 polar bears killed in Greenland 1988–1996. Growth curves, same starting values (each *symbol* represents the mean of the testes of an individual)

Table 2 Size and weight of the reproductive organs of adult polar-bear males (5 years+) in relation to season. Length and weight are the mean of the two organs

		Jan–Feb	Mar–May	Jun–Jul	Aug–Dec
Testis length (mm)	Mean	76.0	73.1	75.6	65.9
	SD	7.5	6.4	6.1	4.5
	Min-Max	67–98	63–90	70–84	61–74
	N	15	37	4	14
Testis weight (g)	Mean	70.7	65.7	65.9	45.4
	SD	14.8	16.7	18.0	11.2
	Min-Max	53–109	35–112	47–90	32–68
	N	15	37	4	14
Epididymis weight (g)	Mean	9.0	9.0	9.2	7.0
	SD	1.73	2.18	3.05	1.74
	Min-Max	6.1–11.5	5.0–13.9	5.3–12.8	4.5–9.7
	N	15	37	4	14

Timing of ovulation in females

During January, most follicles were less than 1 mm in diameter. The three available January samples from adult females that might come into oestrus the next spring (one without cubs and two with 2-year-old cubs) showed a largest follicle size of 1.7, 1.8 and 2.6 mm. Single, mature females are likely to come into oestrus in spring and so are females with 2-year-old cubs, because most females wean their cubs as 2-year-olds (Ramsay and Stirling 1988). However, none of the four single, adult females and none of the three females with 2-year-old cubs caught between 28 February and 30 March had ovulated or were about to do so (the largest measured follicle being between 1 and 4.9 mm). Corpora lutea were found in females ranging from four to ten years of age. The earliest was found in a female caught together with a male on 1 April 1995, in Nuuk (central West Greenland). Five out of seven adult females caught between 1 April and 19 May had either corpora lutea or ripe follicles. Two ripe follicles were found in a female killed on 9 April (diameters: 11 and 14 mm) and one in a female killed on 5 May (diameter: 16 mm) (both females were from Northeast Greenland, Fig. 2). The female caught on 5 May also had a fresh placental scar, indicating that she had lost an infant cub or aborted at a late stage and come into oestrus shortly after. Eight bears had corpora lutea with maximum diameters between 8 and 14 mm. In the seven samples where both ovaries were sampled, six had two corpora and the seventh had only one (average ovulation rate is thus 1.86). In two instances, there were two corpora lutea in one ovary and none in the other. Of the three single, adult females caught in June/July, one had ovulated and two had follicle sizes that indicated that they were not in oestrus.

Discussion

Age at sexual maturity

Males

Some spermatozoa were found in bears younger than 5 years of age, but only in a mixture with other cell types, and not in the high concentrations found in older bears. It cannot be excluded that some 3- to 4-year-old males might be able to mate, but the average male polar bear is not fully sexually mature before 1 or 2 years later and 95% of the asymptotic testis length was not reached before 5.8 years of age.

Females

The female samples were few and therefore not used to calculate a mean age of sexual maturity. However, our analyses show that at least some of the females from Greenland are able to give birth around their 5th birthday, which is in agreement with findings from other parts of the Arctic (Lentfer 1982; Ramsay and Stirling 1982, 1988; Furnell and Schweinburg 1984; Derocher and Stirling 1995).

Timing of the mating season

Adult males and females are frequently caught together in March (Rosing-Asvid and Born 1990) and the fact that all males had a high concentration of spermatozoa in the epididymis from mid-March onwards in both East

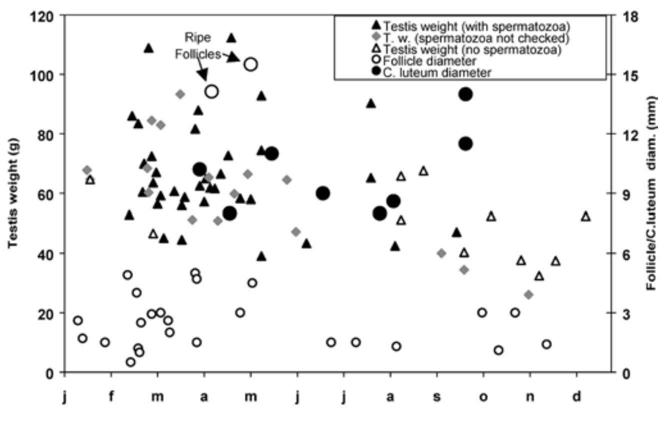

Fig. 2 Testis weight and diameter of the largest follicle or corpus luteum in relation to season in mature polar bears killed in Greenland, 1988–1996

and West Greenland indicates that the mating season has started by then. Our material, however, indicates that ovulations cannot be very common in March. The first corpus luteum was registered on 1 April and the fact that five out of seven single, adult females caught between 1 April and 19 May had either ovulated or were about to do so (two had ripe follicles and three had corpora lutea) indicates that April and the beginning of May is likely to be the peak season for ovulations.

Malyov (1988) found that female polar bears in Kazan Zoo start to become excited 5 to 22 days before the start of coupling. They also renounce food in this period and their external genitals swell noticeably. He further recorded 4 female copulation periods of 7, 14, 21 and 22 days, respectively. This indicates that the mating period if polar bears has induced ovulation, as suggested by Lønø (1970) might start up to a month earlier than ovulation.

In June and July, high concentrations of spermatozoa may still be found in the epididymis in some males, but our April/May samples indicate that most females have ovulated by then, and the two summer samples from females that had not ovulated (21 June and 13 July) had follicle sizes that indicated that they were not in oestrus. So, although males may still be fertile in June/July, their chance of finding a receptive mate at this time in the year is likely to be small.

Differences from the study in Svalbard

Lønø (1970) found that the two 3.5-year-old male polar bears he had from June were both fully mature. We examined eight samples from 3-year-old males caught in April and May (the period found to be the peak of the mating season in Greenland) and only four had spermatozoa in their epididymides. In all these cases, spermatozoa were found in low densities in a mixture with other cell types, quite unlike the high densities found in 5+-year-old animals. We did not have samples of 3-year-olds from June and July, but if the breeding pattern of polar bears resembles that of black bears (*U. americanus*), then it is likely that the testis activity of juveniles and adolescents peaks later than it does in mature animals, which means that males might become fully sexually mature at a mean age somewhat less than the 5.8 years found in this study.

Garshelis and Hellgren (1994) found age- and status-related variation in the serum testosterone in black bears, which indicated that secretion might be modulated through social interactions. The serum testosterone level in 3-year-old black bears was found to decrease at the time of maximal levels in older bears, possibly as a consequence of male-male contests. Some of these young bears, however, were accompanied by and probably mated with single, adult females late in the season, when older bears may be exhausted, as they reduce their food intake during the peak of the breeding season.

Two of the three samples from June that Lønø (1970) used to estimate the peak of the mating season were the 3.5-year-old males mentioned earlier, so if testicular activity in adolescent males peaks later than in older mature bears, than this could explain why Lønø found the mating season to peak approximately 2 months later than we did.

Acknowledgements This study was financed by the Åge V. Jensen foundation and the Greenland Institute of Natural Resources. Among the many people who in some way or another have helped during this study, we wish to gratefully acknowledge the many hunters who collected the samples, Hanne Røddik who did the preparations for histology, and Ian Stirling, Garry Stenson, Arild Landa, Mads Forchhammer, Koos Boomsma and Sylvia Mathiasen who all provided valuable suggestions or comments for this paper.

References

Born EW (1998) Research on polar bears in Greenland, primo 1993 to primo 1997, 97–100. In: Derocher A, Garner GW, Lunn NJ, Wiig Ø (eds) 1998. Polar bears. Proceedings of the 12th Working Meeting of the IUCN Polar Bear Specialist Group. Occasional Paper of IUCN/SSC no. 19. IUCN, Gland

Calvert W, Ramsay MA (1998) Evaluation of age determination of polar bears by counts of cementum growth layer groups. Ursus 10:449–453

DeMaster D, Stirling I (1981) Mammalian species – *Ursus maritimus*. American Society of Mammalogists, no. 145

Derocher AE, Stirling I (1995) Temporal variation in reproduction and body mass of polar bears in Western Hudson Bay. Can J Zool 73:1657–1665

Derocher AE, Stirling I, Andriashek D (1992) Pregnancy rates and serum progesterone levels of polar bears in western Hudson Bay. Can J Zool 70:561–566

Furnell DJ, Schweinburg RE (1984) Population dynamics of central Arctic polar bears. J Wildl Manage 48:722–728

Garshelis DL, Hellgren EC (1994) Variation in reproductive biology of male black bears. J Mammal 75:175–188

Gompertz B (1825) On the nature of the function expressive of the law of human mortality, and on a new mode of determining the value of life contingencies. Philos Trans R Soc Lond 115:513–585

Lentfer JW (1982) Polar bear (*Ursus maritimus*). In: Chapman JA, Feldhammer GA (eds) Wild mammals of North America: biology, management and economics. The John Hopkins University Press, Baltimore, pp 557–566

Lentfer JW, Hensel RJ, Gilbert JR, Sorensen FE (1980) Population characteristics of Alaskan polar bears. Int Conf Bear Res Manage 4:109–115

Lønø O (1970) The polar bear (*Ursus maritimus* Phipps) in the Svalbard area. Nor Polarinst Skr 149:103

Malyov AV (1988) Reproduction and sexual behavior of polar bears *Ursus maritimus* Phipps in the Kazan Zoo botanical garden. Paper presented to the 10th Meeting of IUCN Polar Bear Specialist Group, Sochi, USSR, 24–30 October 1988

Palmer SS, Nelson RA, Ramsay MA, Stirling I, Bahr JM (1988) Annual changes in serum sex steroid in male and female black (*Ursus americanus*) and polar (*Ursus maritimus*) bears. Biol Reprod 38:1044–1050

Pedersen A (1931) Fortgesetztte Beiträge zur Kenntniss der Säugetier und Vogelfauna der Östküste Grönlands. Medd Grønl 77:344–506 (In German)

Ramsay MA, Stirling I (1982) Reproductive biology and ecology of female polar bears in Western Hudson Bay. Rev Ecol Syst 109:941–946

Ramsay MA, Stirling I (1988) Reproductive biology and ecology of female polar bears (*Ursus maritimus*). J Zool Lond 214:601–634

Richards FJ (1959) A flexible growth function for empirical use. J Exp Bot 10:290–300

Rosing-Asvid A, Born EW (1990) Catch of polar bears in the municipalities of Avanersuaq and Upernavik: an interview survey (in Danish with English summary). Report no. 23. Grønlands Hjemmestyre. Miljø og Naturforvaltning

Stirling I, Øritsland NA (1995) Relationships between estimates of ringed seal (*Phoca hispida*) and polar bear (*Ursus maritimus*) populations in the Canadian Arctic. Can J Fish Aquat Sci 52:2594–2612