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Seasonal changes in plasma testosterone concentrations in response to administration of hCG in a desert rodent, the sand rat (*Psammomys obesus*)

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Summary. In adult male sand rats inhabiting the Béni-Abbès area (Algeria), testicular endocrine activity increased in early summer (June–July), was highest in autumn–winter and decreased throughout spring. Testosterone secretion by the testis of the sand rat was stimulated (by 10–60-fold) throughout the year by exogenously administered hCG (25 i.u.). However, the response exhibited annual changes mainly characterized by a marked increase in early summer (June–July); the response to hCG was depressed in autumn and became minimal in winter and in early spring. The results strongly suggest that the summer onset of testicular endocrine activity is, at least in part, due to an increase in the testis sensitivity to LH.

Keywords: seasonal changes; testis sensitivity; desert rodent; testosterone; LH; sand rat

Introduction

In previous studies performed over 7 years (Khammar & Brudieux, 1984; Khammar, 1987), we have shown that testicular and seminal vesicle weights and androgen production of the male sand rat (*Psammomys obesus*), a gerbilline rodent inhabiting underground burrows on briny alluvial platforms in the Sahara desert, exhibited in the field (in the Béni-Abbès area, in Algeria) a marked annual cycle, mainly characterized by a rise in early summer (June–July), a maximum in autumn–winter and a decrease during spring. At least two endogenous mechanisms, alone or in combination, may be responsible for this annual endocrine cycle; i.e. seasonal changes (1) in the sensitivity of the testis itself to luteinizing hormone (LH) or (2) in the activity of the hypothalamo–pituitary system. We are not aware of any other reports dealing with annual variations, and their endogenous determinants, in the production of testosterone in desert rodents.

The purpose of the present study was to investigate whether there is annual variation in the sensitivity of the testis of sand rats to hCG.

Materials and Methods

Animals. Adult male sand rats (*Psammomys obesus*) were used. The adult condition was checked according to the criteria previously described (Amirat *et al.*, 1980). They were obtained from the Béni-Abbès area (Wilaya of Béchar, Algeria: 30°7′N; 2°10′W) and were caught from March 1984 to December 1985 (i.e. 21–31 March 1984, N = 5; 5–14 June 1984, N = 10; 3–10 November 1984; N = 6; 21 March–4 April 1985, N = 6; 1–15 June 1985, N = 11; 13–18 July 1985, N = 5; 3–15 November 1985, N = 10; and 19–30 December 1985, N = 12). Immediately after trapping, the animals were kept for 12–24 h in individual cages in the laboratory and fed exclusively fresh plants (*Suaeda mollis*) *ad libitum*. Experiments were always carried out between 09:00 and 13:00 h.

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The sand rats were anaesthetized with an intraperitoneal injection of 4 mg pentobarbitone sodium/100 g body weight (Nembutal, Abbott, St Rémy-sur-Avre, France). The left carotid artery and the right jugular vein were catheterized. Animals were heparinized with an intravenous administration of 50 i.u. (0·2 ml) calcium heparinate/100 g body weight. Then 25 i.u. hCG (Laboratories I.S.H., Paris, France), dissolved in 0·2 ml 0·9% (w/v) NaCl containing 0·2% BSA, were injected through the jugular vein. Immediately (0 min) and 60 and 120 min after the hCG administration, 3 × 1 ml blood samples were withdrawn from the carotid artery catheter. In preliminary experiments it was confirmed that 60 min and 120 min revealed the peak and the duration, respectively, of the testosterone response to this dose of hCG. Plasma samples were stored at −25°C until analysis. Animals were killed by decapitation. Seminal vesicles with secretion and testes were quickly removed and weighed.

Hormone assays. Testosterone concentrations in plasma were measured, after extraction by ethyl acetate–cyclohexane (50:50, v/v) and purification on celite columns, according to the radioimmunoassay technique previously described and validated by Darbeïda & Brudieux (1980). Because of the very low plasma values at 10 min, the purification on celite columns was suppressed. This method has been validated by Khammar & Brudieux (1987): the sensitivity was 7 pg/tube, within-assay variance 3·0% and between-assay variance 10·5%.

Statistical analysis. Values are given as means and standard errors (s.e.m.) and the significance of differences was determined by Student's *t* test.

Results

Testicular and seminal vesicle weights

The results in Table 1 show that the sand rats used in the present experiment exhibited seasonal changes in testicular and seminal vesicle weight similar to those previously reported (Khammar & Brudieux, 1984; Khammar, 1987). Considering all the animals, the heaviest testes and seminal vesicles occurred in autumn and in winter; they declined throughout spring (−52·8%, $P < 0·001$ for the testis; −73·3%, $P < 0·001$ for seminal vesicles). In late March 1984, all the animals trapped had well developed testes and seminal vesicles, but in late March and early April 1985, 2/6 sand rats had regressed genital apparatus. During the first 2 weeks of June, seminal vesicle weights particularly were still low in 4 and 8 animals in 1984 and 1985, respectively, whilst they were increasing in 6 and 3 sand rats; this onset of development was confirmed in the 5 animals caught in July 1985.

Plasma concentrations of testosterone in response to hCG administration (Fig. 1)

Basal plasma concentrations of testosterone were low; they ranged from 0·28 ± 0·04 ng/ml in November 1984 to 0·12 ± 0·01 ng/ml in June 1985 (−57·2%, $P < 0·001$).

Administration of 25 i.u. hCG greatly increased plasma testosterone concentrations, but there were seasonal differences in the response of the testes. At 60 min after hCG administration, plasma concentration of testosterone was 3·2 ± 0·8 ng/ml in March 1984; in June, it increased by 147·0% ($P < 0·02$), regardless of the state of the genital apparatus. The response to hCG was significantly decreased by half in early November 1984 and became minimum for all the animals used in March–April 1985 (2·7 ± 0·2 ng/ml). In June 1985, it remained low in the sand rats in which the genital apparatus was still regressed, but was markedly increased (+182·9%; $P < 0·001$) in the 3 animals which had already exhibited an increase in testicular and seminal vesicle weights. This maximum release was also observed for the animals caught in July 1985 (7·9 ± 2·0 ng/ml). As in 1984, plasma concentrations of testosterone decreased in November 1985 and were minimal in December 1985 (2·0 ± 0·3 ng/ml).

At 120 min after hCG administration, plasma concentrations of testosterone and the pattern of seasonal changes were essentially the same as those observed after 60 min (Fig. 1). In 1984 and in 1985, the highest concentrations were in early summer. However, in June 1984, the 4 animals with the smallest seminal vesicles had the lowest hCG-stimulated testosterone secretion, similar to that in March 1984; moreover, plasma concentrations of testosterone in the sand rats caught in July 1985 were not higher than those of animals in November 1985.

Table 1. Seasonal changes in body weight and testicular and seminal vesicle weights in adult male sand rats (*Psammomys obesus*) live-trapped in the field in the Béni-Abbès area (Algeria)

Time of sample	No. of animals	Body wt (g)	Testicular wt (left testis) (mg)	Wt of seminal vesicle + secretions (mg)
21–31 March 1984	5	110 ± 4	275 ± 13	363 ± 29
5–14 June 1984	6	114 ± 7	210 ± 13	256 ± 16
	4	93 ± 3	167 ± 30	113 ± 13
	(10)	(106 ± 5)	(192 ± 15)	(199 ± 25)
3–10 November 1984	6	141 ± 6	318 ± 20	463 ± 21
21 March–4 April 1985	4	110 ± 9	227 ± 39	190 ± 13
	2	98 ± 1	122 ± 3	48 ± 3
	(6)	(106 ± 6)	(192 ± 33)	(143 ± 31)
1–15 June 1985	3	134 ± 6	221 ± 26	199 ± 15
	8	91 ± 5	123 ± 16	81 ± 14
	(11)	(103 ± 7)	(150 ± 19)	(114 ± 20)
13–18 July 1985	5	88 ± 5	155 ± 12	204 ± 26
3–15 November 1985	10	145 ± 10	274 ± 17	240 ± 28
19–30 December 1985	12	164 ± 7	303 ± 19	327 ± 18

Values are means ± s.e.m.
In June 1984, March–April and June 1985, the criteria for the two separate groups were seminal vesicle weights; numbers in parentheses are relative to all animals caught.

Discussion

The present results show that the testis of wild caught sand rats could be stimulated throughout the year by exogenous hCG; basal values of testosterone were increased by 10–60-fold.

Testicular sensitivity to the gonadotrophin exhibited important seasonal variations. The response increased in early summer, especially in sand rats in which seminal vesicle weights were increasing. The testes of the 4 animals trapped in June 1984, with seminal vesicles just beginning to redevelop, were likewise responsive to hCG since, at the maximal effect, 60 min after administration of hCG, plasma values of testosterone were raised to the same extent as that in the 6 animals with well developed seminal vesicles caught at this period. However, the testosterone response was probably less sustained, because at 120 min after hCG, plasma testosterone concentrations of these 4 sand rats were reduced by half, compared to the 6 other animals. These 4 sand rats were probably therefore at the beginning of their seasonal cycle of sensitivity to LH. On the other hand, in the 8 sand rats trapped in June 1985, which still had low seminal vesicle weights, hCG was no more effective in stimulating testosterone secretion than in animals of March 1985; their seasonal increase in sensitivity to LH had not yet occurred.

We suggest that the early summer onset of sensitivity to LH of the sand rat testis occurred later in 1985 than in 1984; this is supported by the fact that at 120 min after hCG administration there was no significant difference in plasma testosterone concentration between July and November 1985.

The present results suggest that early summer (June–July) is a transition period for seasonal changes in sensitivity of the sand rat testis to LH. However, this summer-enhanced sensitivity was transient, since the testosterone response to LH was depressed in autumn and became minimal in winter and in early spring.

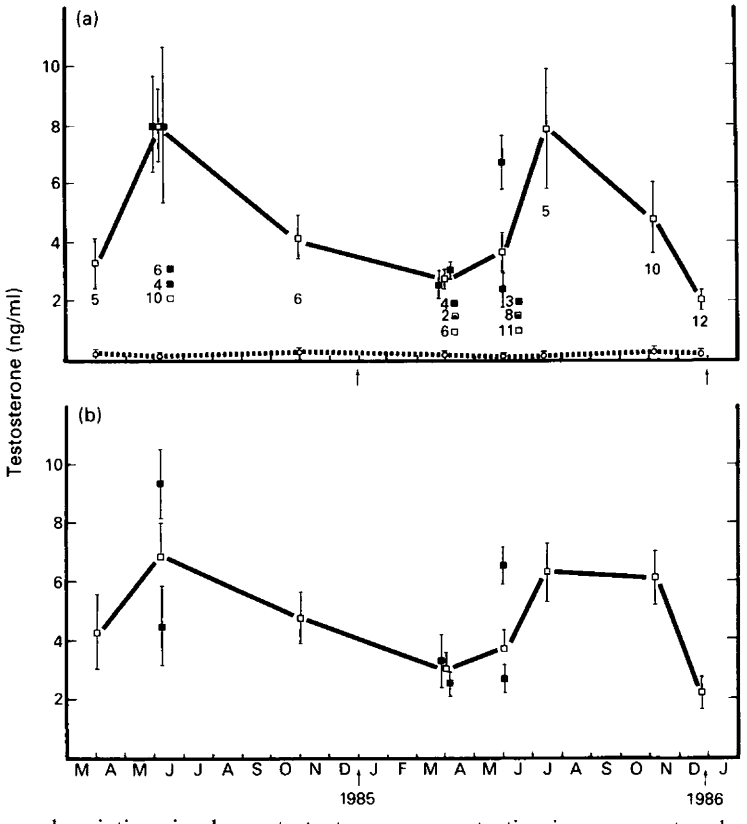

Fig. 1. Seasonal variations in plasma testosterone concentration in response to administration of hCG (a) – – – 0 min and —— 60 min and (b) 120 min after 25 i.u. hCG/100 g body wt, in adult male sand rats. Values are mean ± s.e.m. for the no. of sand rats indicated: ⊡, Animals with low seminal vesicle weights; ■, animals with stimulated seminal vesicles; □, all the animals.

The present report is the first investigation, performed throughout the year, dealing with the in-vivo response to LH of the testis in a wild small mammal live-trapped in the field. Previous studies, in the ferret (Neal & Murphy, 1977), and in Soay (Lincoln, 1981), Finnish-Landrace and cross-bred (Sanford *et al.*, 1984) rams, showed a larger increase in LH-stimulated production of testosterone during the reproductive period than during the non-breeding season. Analysis of the ultradian profiles of plasma LH and plasma testosterone between the active and the inactive seasons in the ram (Katongole *et al.*, 1974; Lincoln, 1976; Schanbacher & Ford, 1976), the red deer (Lincoln & Kay, 1979) and the pigmy goat (Muduuli *et al.*, 1979) revealed that, for a given elevation in plasma LH, the increase in plasma testosterone was largest during the breeding season. Together these data are consistent with the hypothesis of seasonal changes in testis sensitivity to LH in these species.

Annual changes in blood flow through the testis, with maximal values during the breeding season, have been demonstrated in some seasonal breeders such as the dormouse, ferret, fox (Joffre & Joffre, 1973; Joffre, 1977) and ram (Courot & Joffre, 1976); such a feature may be involved in seasonal changes in testosterone responsiveness to LH in the sand rat. However, it will be necessary to investigate in the sand rat the seasonal characteristics (affinity and maximum binding capacity) of LH testis receptors; in other species (hamster: Bex *et al.*, 1978, Bartke *et al.*, 1980; mongoose: Soares & Hoffman, 1982; bank-vole: Tähka *et al.*, 1983; ram: Barenton & Pelletier, 1983) maximum

testosterone responsiveness to LH was associated with an increase in the concentration of testicular LH binding sites. Such a phenomenon is likely at the beginning of summer in the sand rat.

The present work demonstrates clearly that seasonal reactivation in early summer of the endocrine activity of the testis of the sand rat, live-trapped in its desert environment, is, at least partly, due to an increase in the responsiveness of the testis to the stimulatory effect of LH. These changes might be due to seasonal variations in gonadotrophin secretion. Seasonal variations in the production of LH and other hormones such as thyroxine are currently being investigated.

References

Amirat, Z., Khammar, F. & Brudieux, R. (1980) Seasonal changes in plasma and adrenal concentrations of cortisol, corticosterone, aldosterone and electrolytes in the adult male sand rat (*Psammomys obesus*). *Gen. comp. Endocrinol.* **40,** 36–43.

Barenton, B. & Pelletier, J. (1983) Seasonal changes in testicular gonadotrophin receptors and steroid content in the ram. *Endocrinology* **112,** 1441–1446.

Bartke, A., Goldman, B.D., Bex, F.J., Kelch, R.P., Smith, M.S., Dalterio, S. & Doherty, P.C. (1980) Effects of prolactin on testicular regression and recrudescence in the golden hamster. *Endocrinology* **106,** 167–172.

Bex, F.J., Bartke, A., Goldman, B.D. & Dalterio, S. (1978) Prolactin, growth hormone, luteinizing hormone receptors and seasonal changes in testicular activity in the golden hamster. *Endocrinology* **103,** 2069–2080.

Courot, M. & Joffre, M. (1976) Testicular capillary blood flow in the impuberal lamb and ram during the breeding and non-breeding seasons. *Annls Biol. anim. Biochim. Biophys.* **16,** 171.

Darbeïda, H. & Brudieux, R. (1980) Seasonal variations in plasma testosterone and dihydrotestosterone levels and in metabolic clearance rate of testosterone in rams in Algeria. *J. Reprod. Fert.* **59,** 229–235.

Joffre, M. (1977) Relationship between testicular blood flow, testosterone secretion and spermatic activity in young and adult wild red foxes (*Vulpes vulpes*). *J. Reprod. Fert.* **51,** 35–40.

Joffre, J. & Joffre, M. (1973) Seasonal changes in the testicular blood flow of seasonally breeding mammals: dormouse *Glis glis*, ferret *Mustela furo*, and fox *Vulpes vulpes*. *J. Reprod. Fert.* **34,** 227–233.

Katongole, C.B., Naftolin, F. & Short, R.V. (1974) Seasonal variations in blood luteinizing hormone and testosterone levels in rams. *J. Endocr.* **60,** 101–106.

Khammar, F. (1987) *Variations saisonnières de l'activité endocrine du testicule de deux espèces désertiques, le Rat des sables* (Psammomys obesus) *et la Gerbille* (Gerbillus gerbillus). Thèse Doct. ès sciences, Université d'Alger, 202 pp.

Khammar, F. & Brudieux, R. (1984) Seasonal changes in testicular contents of testosterone and androstenedione and in the metabolic clearance rate of testosterone in the sand rat (*Psammomys obesus*). *J. Reprod. Fert.* **71,** 235–241.

Khammar F. & Brudieux, R. (1987) Seasonal changes in testicular contents and plasma concentrations of androgens in the desert gerbil (*Gerbillus gerbillus*). *J. Reprod. Fert.* **80,** 589–594.

Lincoln, G.A. (1976) Seasonal variation in the episodic secretion of luteinizing hormone and testosterone in the ram. *J. Endocr.* **69,** 213–226.

Lincoln, G.A. (1981) Seasonal aspects of testicular function. In *The Testis*, pp. 255–302. Eds H. Burger & D. M. de Kretser. Raven Press, New York.

Lincoln, G.A. & Kay, R.N.B. (1979) Effects of season on the secretion of LH and testosterone in intact and castrated red deer stags (*Cervus elaphus*). *J. Reprod. Fert.* **55,** 75–80.

Muduuli, D.S., Sandford, L.M., Palmer, W.M. & Howland, B.E. (1979) Secretory patterns and circadian and seasonal changes in LH, FSH, prolactin and testosterone in the male pygmy goat. *J. Anim. Sci.* **49,** 543–553.

Neal, J. & Murphy, B.D. (1977) Response of immature, mature non breeding and mature breeding ferret testis to exogenous LH stimulation. *Biol. Reprod.* **16,** 244–248.

Sanford, L.M., Howland, B.E. & Palmer, W.M. (1984) Seasonal changes in the endocrine responsiveness of the pituitary and testes of male sheep in relation to their patterns of gonadotropic hormone and testosterone secretion. *Can. J. Physiol. Pharmacol.* **62,** 827–833.

Schanbacher, B.D. & Ford, J.J. (1976) Seasonal profile of plasma luteinizing hormone, testosterone and estradiol in the ram. *Endocrinology* **99,** 752–757.

Soares, M.J. & Hoffmann, J.C. (1982) Seasonal reproduction in the Mongoose, *Herpestes auropunctatus*. II. Testicular responsiveness to luteinizing hormone. *Gen. comp. Endocr.* **47,** 226–234.

Tähka, K.M., Ruokonen, A., Wallgren, M. & Teravainen, T. (1983) Temporal changes in testicular histology and steroidogenesis in juvenile bank voles (*Clethrionomys glareolus*, Schreber) subjected to different photoperiods. *Endocrinology* **112,** 1420–1426.

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