STUDIES ON REPRODUCTION IN THE CAMEL (*CAMELUS DROMEDARIUS*)

V. MORPHOLOGY OF THE TESTIS IN RELATION TO AGE AND SEASON

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Summary. Quantitative and qualitative changes in the morphology of the camel testis were studied in relation to age (6 to 18 years) and season. The diameter of the seminiferous tubules increased up to the age of 9 years. There was little variation in the number of germinal cells (spermatogonia, primary spermatocytes and spermatids) with the advancement of age. The number of spermatozoa increased up to the age of 10 years and decreased thereafter. The number of Sertoli cells was almost constant. Significant monthly and seasonal changes were observed in the criteria studied. The largest seminiferous tubule diameters and the greatest numbers of spermatogonia, spermatids and spermatozoa were found in the material collected during the spring. The numbers of mature Leydig cells, compared to the numbers of pre-Leydig and immature Leydig cells, increased by the end of winter so that, during the spring, the interstitial cells were mainly of the mature type. Degenerative changes with diminished numbers of mature cells were seen in the summer and this trend continued into early and mid-autumn.

INTRODUCTION

The camel is known to be a seasonal breeder (Leese, 1927) and Volcani (1954) and Charnot (1963, 1964, 1965) have studied some aspects of the seasonal changes in the testis. Novoa (1970) reviewed studies on reproduction in the Camelidae and recorded that sections through the camel testis showed monthly and seasonal histological changes.

The aim of the present work was to study the qualitative and quantitative histological changes in the testis of the Arabian camel (*Camelus dromedarius*) in relation to age and season.

MATERIALS AND METHODS

The material included in this study consisted of pairs of testes obtained between

October 1971 and November 1972 from fifty-nine Arabian camels of the native Egyptian breed (Saidi breed) ranging in age from 6 to 18 years.

The testes, obtained shortly after slaughter, were stripped of their tunics and adhering structures, and then divided into two longitudinal halves. Thin slices were taken from the parenchyma parallel to the mediastinum, and after fixing in 10% formalin solution, the tissues were transported to the laboratory. The specimens were then transferred to Bouin's solution for 24 hr, dehydrated and embedded in paraffin wax. Sections about 4 to 6 μm thick were cut and stained with Harris's haematoxylin and eosin.

The diameter of the seminiferous tubules, and the numbers of germinal cells and supporting cells were measured in ten well-defined tubules in each testis. Evaluation of cell morphology was based on the previous descriptions given by Abdel-Raouf (1960, 1961) and Courot, Hochereau-de Reviers & Ortavant (1970). The types and numbers of the cells in the intertubular spaces were analysed according to the description given by Hooker (1944).

RESULTS

Changes in relation to age

The average diameters of the seminiferous tubule at the different ages studied are presented in Table 1. The diameter increased from 6 to 9 years, and then showed slight variations in the different age groups, the average diameter being about 210 μm . There was no significant difference between the average diameters of the tubules measured in the right and the left testes in the different age groups.

The average numbers of germinal cells and Sertoli cells are presented in Table 1. There were no specific differences in the numbers of germinal cells, with the exception of the spermatozoa. The number of Sertoli cells was almost constant. The numbers of germinal cells in the right and left testes appeared unequal but the differences were not statistically significant.

Changes in relation to season

Diameter of seminiferous tubules. The average diameter of the seminiferous tubules was least during the summer and gradually increased throughout the autumn and winter to reach the largest size during the spring (Table 2). Analysis of variance showed that the difference in the diameter of the tubules during the different seasons was highly significant ($P < 0.005$). The diameter was greatest during March, decreased continuously to July, and then started to increase again from September.

Spermatogenesis and number of intratubular cells. In the measured tubules as well as in most of the sections studied in general, the different types of germinal cells were observed in numbers which varied according to season. Spermatozoa were not observed in all the tubules in the different seasons and their presence showed clear seasonal variations. The number of Sertoli cells was almost constant.

During the winter, spermatogenesis was active, although the tubules appeared loosely packed with cells. During the spring, the number of germinal cells increased, the tubules were filled with the different germinal cells and a large

percentage of tubules contained spermatozoa. During the summer, spermatogenesis was reduced and an average of 42% of the tubules contained spermatozoa. The tubules appeared to be loosely packed with the different germinal cells. During the autumn, the number of the tubules containing spermatozoa was reduced compared to the summer, and the number of spermatids and spermatozoa diminished. The intratubular cells appeared loose in the tubules and some of the spermatids and primary spermatocytes showed degenerative changes early in the season. Later in the season, and with the approach of the winter, the condition improved.

Table 1. Age changes in characteristics of the testes of *Camelus dromedarius*

Age (years)	No. of animals	Diameter of seminiferous tubule (μm)	No. of cells per cross-section				
			Spermato- gonia	Primary sperma- toocytes	Sperma- tids	Spermatozoa	Sertoli cells
6	5	193.50 (21.70)	38.36 (12.77)	22.05 (13.74)	60.80 (33.72)	5.69 (6.16)	10.69 (1.23)
7	2	180.70 (46.24)	30.28 (4.12)	16.05 (4.17)	45.50 (25.18)	2.48 (0.99)	9.58 (0.14)
8	7	195.99 (12.69)	39.93 (6.92)	15.76 (4.69)	59.84 (14.97)	8.89 (6.71)	11.02 (1.39)
9	7	215.71 (19.57)	33.40 (8.33)	17.58 (4.17)	60.48 (12.92)	11.98 (9.98)	11.34 (1.12)
10	7	208.13 (22.56)	37.86 (12.00)	25.58 (6.86)	59.86 (27.96)	27.04 (18.78)	11.38 (1.39)
11	2	209.65 (20.86)	34.03 (4.03)	12.40 (2.33)	51.18 (0.14)	10.88 (3.89)	11.68 (0.00)
12	18	213.21 (14.42)	31.58 (7.21)	21.41 (5.39)	67.25 (15.90)	12.97 (8.57)	11.91 (1.59)
13	3	191.03 (10.68)	31.05 (1.45)	17.07 (0.81)	50.32 (4.68)	12.80 (7.66)	10.80 (1.32)
15	5	214.24 (14.04)	31.07 (7.40)	20.52 (3.92)	68.08 (21.49)	6.08 (5.22)	12.37 (2.63)
16	1	207.00	34.65	13.05	51.55	7.55	13.00
18	2	204.50 (19.10)	29.58 (6.36)	17.90 (3.32)	52.90 (0.14)	9.10 (7.35)	11.55 (0.28)

Figures represent the average (and standard deviation) for ten tubules examined in each testis from fifty-nine camels.

Analysis of variance showed that there were highly significant seasonal changes in the number of spermatogonia ($P < 0.005$), the number being greatest during the spring, decreasing during the summer and autumn and then increasing again during the winter (Table 2). The greatest numbers of spermatogonia were found during March, April, May and June.

The average numbers of primary spermatocytes were found to be almost equal in the spring and summer and the numbers counted during the autumn and winter were slightly more than those counted in the other two seasons (Table 2). The seasonal differences were significant ($P < 0.01$). The average numbers during the different months fluctuated, being highest during February.

Significant seasonal differences were recorded for the number of spermatids

Table 2. Monthly and seasonal changes in characteristics of the testes of *Camelus dromedarius*

Month and season	No. of animals	Diameter of seminiferous tubules (μm)	No. of cells per cross-section				
			Spermato-gonia	Primary spermatocytes	Spermatids	Spermato-zoa*	Sertoli cells
December	8	215.85 (10.67)	27.58 (4.89)	18.35 (3.32)	59.00 (14.32)	32.54 (77.41)	12.43 (1.29)
January	4	210.98 (14.73)	29.33 (6.63)	22.15 (11.23)	63.00 (23.58)	36.52 (15.56)	11.45 (0.91)
February	4	216.68 (5.16)	32.10 (6.56)	24.30 (5.29)	65.30 (6.25)	26.48 (10.05)	12.40 (0.08)
WINTER	16	214.84 (10.34)	29.14 (5.66)	20.79 (6.56)	61.58 (14.89)	32.29 (9.22)	12.18 (1.09)
March	4	224.05 (20.05)	39.80 (12.20)	21.75 (8.25)	70.53 (11.83)	48.27 (21.28)	12.63 (0.52)
April	5	209.68 (23.26)	37.24 (5.56)	15.20 (4.58)	64.96 (28.76)	36.32 (7.55)	11.88 (0.82)
May	3	225.20 (26.98)	40.27 (7.48)	17.50 (4.24)	65.73 (13.30)	36.58 (9.11)	12.77 (0.50)
SPRING	12	218.35 (22.29)	38.85 (8.80)	17.95 (6.16)	67.01 (19.44)	41.45 (12.88)	12.35 (0.73)
June	5	203.26 (19.69)	38.72 (12.69)	21.16 (8.83)	68.78 (31.06)	35.56 (18.16)	10.88 (0.96)
July	5	189.40 (5.92)	28.74 (6.24)	15.16 (2.45)	52.86 (9.77)	27.85 (8.18)	11.14 (1.11)
August	2	194.05 (12.25)	38.95 (10.25)	20.75 (3.46)	67.85 (31.85)	24.20 (7.28)	10.75 (0.77)
SUMMER	12	195.85 (14.56)	34.60 (10.44)	18.59 (6.40)	61.99 (25.25)	31.93 (12.08)	10.97 (0.93)
September	1	189.00	33.20	18.00	63.30	21.33	9.50
October	12	199.68 (17.63)	35.88 (7.28)	21.91 (3.61)	55.56 (18.63)	29.84 (7.55)	10.54 (1.56)
November	6	198.46 (13.19)	32.43 (7.35)	21.12 (4.24)	57.93 (17.03)	34.45 (12.85)	11.23 (1.94)
AUTUMN	19	198.74 (15.62)	34.65 (7.07)	20.82 (3.74)	56.72 (17.21)	31.47 (9.27)	10.71 (1.54)

Figures represent the average (and standard deviation) for ten tubules examined in each testis from fifty-nine camels.

* The figures shown represent averages based only on those tubules that contained spermatozoa.

($P < 0.05$). The number was greatest during the spring, decreased during the summer and autumn and increased again during the winter (Table 2). The monthly changes showed that the highest numbers of spermatids were found in March, June and August, and the lowest in July.

The average number of spermatozoa was greatest ($P < 0.005$) during the spring and decreased during the summer. There was only a slight increase during the autumn and winter. When the number of spermatozoa was studied on a monthly basis, it was found to increase gradually from December to March and then to decrease gradually to June. The decrease from June to September

was more rapid than in the previous period and was followed by an increase starting in September (Table 2).

The number of Sertoli cells showed no statistically significant seasonal changes, although the numbers in the winter and spring were slightly higher than during the other two seasons (Table 2).

Intertubular cells. At the beginning of the winter, the intertubular spaces were almost filled with closely packed clusters of cells. The interstitial tissue was relatively rich in sinusoids and capillaries. The intertubular cells consisted largely of the pre-Leydig type and some immature Leydig cells. Few mature Leydig cells were seen. Towards the middle of the season, both the mature and immature Leydig cells increased greatly but to a lesser extent than the pre-Leydig cells. At the end of the winter, the number of the mature Leydig cells was almost equal to or slightly more than the number of pre-Leydig cells.

In the spring, the relatively narrow intertubular spaces were packed with interstitial cells, mainly of the mature Leydig cell type. The cells contained large vacuoles evenly interspersed throughout the cell cytoplasm.

At the beginning of the summer, the Leydig cells decreased in number and failed to fill the intertubular spaces. The peritubular spaces were almost empty in most of the regions except for a few connective tissue fibres and fibroblasts. In the spaces containing larger numbers of cells, the interstitial tissue presented a shrunken appearance. Towards the middle of the season, the interstitial cells decreased in amount, became widely separated from each other and were held together by fine connective tissue fibres. At the end of the summer, the mature Leydig cells showed pyknotic changes in their nuclei. A few pre-Leydig and immature Leydig cells were seen between the degenerating mature Leydig cells, and a few fibroblasts were visible between the interstitial cells.

At the beginning of the autumn, the intertubular spaces contained scattered Leydig cells held together by fine connective tissue with relatively few capillaries and sinusoids. During the middle of the season, the interstitial cells increased greatly in amount and appeared in closely adjacent clusters almost filling the intertubular spaces. The blood sinusoids and capillaries were more abundant than at the beginning of the season. The number of cells increased with the approach of winter. Most of the cells observed in the intertubular spaces during this season were of the pre-Leydig type and were replaced by mature Leydig cells as the season progressed.

DISCUSSION

The diameter of the seminiferous tubules increased slightly from 6 to 9 years of age, though this picture might have been different if the material had included animals younger than 6 years old. Similar changes in the diameter of the seminiferous tubules with age have been reported for bulls (Michatsch, 1933; Abdel-Raouf, 1960) and rams (Carmon & Green, 1952). The value given by Abdel-Raouf (1965) for the average diameter of the seminiferous tubules of a 7-year-old camel was confirmed for animals of the same age in the present study.

No data are available in the literature concerning differences in the tubular diameter or number of cells in the right and left testes.

The quantitative study of different germinal cells showed that there was little

variation with the advancement of age in the numbers of spermatogonia, primary spermatocytes and spermatids. This may indicate that by the age of 6 years spermatogenesis has reached its upper limits and that all the animals included in this study had reached puberty. Leese (1927) mentioned that the male camel may breed when 3 years old but that full reproductive maturity was not reached until 6 years of age. The present study therefore concerns postpubertal animals. The number of spermatozoa did, however, increase up to the age of 10 years, i.e. even after reaching full reproductive status. This agrees with the previous findings of Abdel-Raouf (1960) in bulls. The number of Sertoli cells seemed to be constant without any change related either to age or season and the averages obtained were approximately similar to those previously counted in normal tubules in the camel testis (Abdel-Raouf, 1965).

The highly significant seasonal changes in the diameter of the seminiferous tubules are similar to those reported for camels by Bodenheimer (1954) and Volcani (1954). They found that the diameter of the tubules measured 183 μm during December to March and were reduced to 131 μm during May. Variations similar to those reported here in the camel have also been reported in the buffalo, in which the diameter of the tubules reached a maximum during the winter and spring, and a minimum during the autumn and summer (Bhatnagar, Mukherjee & Bhattacharya, 1955).

The numbers of spermatogonia, spermatids and spermatozoa were significantly higher in the spring than in any other season, denoting that spermatogenesis is active during this season. All the germinal cells were found during the different seasons with the exception of the spermatozoa which showed considerable seasonal variations in number, and the tubules containing these cells diminished greatly in number during the late summer, autumn and early winter. This means that the non-breeding season in the camel is characterized only by reduced spermatogenesis, never by complete aspermatogenesis. These changes are in accord with the description given by Bodenheimer (1954), Volcani (1954) and Charnot (1964).

The findings observed here for the Arabian camel resemble to a great extent the condition in rams as described by Maqsood (1951). In some non-domesticated ungulates, complete aspermatogenesis occurs during the non-breeding season. Thus, in the roe-buck (Short & Mann, 1966), in the fallow deer (Chaplin & White, 1972) and in the reindeer (Meschaks, 1966), the degenerative changes include the spermatozoa and the spermatids and only spermatocytes and spermatogonia are found in the tubules during the non-breeding season. In many seasonally breeding animals, e.g. the ferret (Allanson, 1934) and fox squirrel (Kirkpatrick, 1955), the retrogressive changes result in a testis containing only spermatogonia at the basement membrane after sloughing of the remainder of the germinal cells.

Seasonal changes in the contents of the intertubular spaces confirmed the report of Charnot (1964, 1965) for the camel and have also been found in the Virginia deer (Wislocki, 1943) and in the reindeer (Meschaks, 1966). Other wild mammals, such as the bat, common European mole, European hedgehog, marmot, ferret and flying fox (see Asdell, 1946), show clear seasonal changes in the interstitial tissue.

Wislocki (1949) studied the histochemical reactions in the interstitial tissue of deer and found changes indicating the formation of steroid hormones during the breeding season. Since the Leydig cells are mainly responsible for androgen production, the camel could only breed during the season when mature Leydig cells are abundant in the intertubular spaces, i.e. during late winter and spring. This is in agreement with previous studies which have shown that the male camel ruts in the spring in Egypt (Abdel-Raouf & El-Naggar, 1964) and in Somaliland (Mares, 1954), during November to February in India (Hira, 1947; Singh & Brakash, 1964; Khan, 1971), from December to March in Pakistan (Yasin & Wahid, 1957), and in the winter and spring in Morocco (Charnot, 1963).

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