VETERINARSKI ARHIV 75 (2), 129-135, 2005

Observations on the morphology of the ovaries of the porcupine (*Hystrix cristata*)

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OZDEMIR, D., A. AYDIN, S. YILMAZ, G. DINC, O. ATALAR: Observations on the morphology of the ovaries of the porcupine (*Hystrix cristata*). Vet. arhiv 75, 129-135, 2005.

ABSTRACT

This study was carried out in order to investigate the structure of ovaries of the porcupine (*Hystrix cristata*). Ovaries were obtained by laparoscopic surgery and were then divided into two sections and fixed in buffered formalin. The sections (5-7 μm) were cut in paraffin blocks, stained with Mayer's haematoxylin stain, Crossmonn's triple stain and McManus's periodic acid Schiff (PAS) technique. In histological examinations it was composed of two main zones, cortex and medulla. The surfaces of the ovaries were wavy and lined by simple cuboidal or columnar epithelium. The tunica albuginea consisted of connective tissue fibres. There were connective tissue cells in the cortex, as well as interstitial cells containing lipid. Primordial, primary, secondary, tertiary follicles, corpus luteum and atretic follicles were seen in the cortex of ovaries. Mean diameters of these follicles varied between 80 and 1600 μm. Medulla consisted of loose connective tissue which contained blood and lymph vessels of varying sizes. The masses and sizes - length, width and thickness - of the right and left ovaries were almost the same.

Key words: *Hystrix cristata*, porcupine, ovary, structure

Introduction

Rodents comprise the largest and most diverse group of mammals, with over 1700 different species (BESSELSEN, 2002). The porcupine belongs to the Hystricidae family, which constitutes a small group of the order Rodentia (DEMIRSOY, 1992; KURU, 1987).

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ISSN 0372-5480
Printed in Croatia

Although numerous papers have been published on the structure of rodent ovaries, rat (O'SHEA, 1970; YECAN and OZAN, 1996), mouse (ODOR and BLANDAU, 1968), guinea pig (ADAMS and HERTIG, 1964; COLLINS and KENT, 1964; DAVIES et al., 1985; DEANESLY, 1972 and 1975; GURAYA, 1977; OZDEMIR and DINC, 2002; OZDEMIR and DINC, 2003; PEDDIE, 1980), hamster (BODEMER et al., 1959; BODEMER and WARNICK, 1961; CHALLONER, 1974; WEAKLEY, 1969) and rabbit (DEANESLY, 1972; DEANESLY, 1975), there are no reports on details of the ovaries of the porcupine. In this study we examined porcupine ovaries macroscopically and by light microscopy to clarify their morphological features.

Materials and methods

Five female adult porcupines (*Hystrix cristata*) of different ages and trapped in Eastern Anatolia (Turkey) were used. The animals were anesthetized with Pentothal (6 mg/kg). The ovarian samples, trimmed of fat and extraneous tissue, were fixed for 24-48 h in 10 % neutral formalin. After fixation the tissues were dehydrated through a series of graded ethanol solutions (50-100%), and embedded in paraffin. The tissue sections (5-7 μm) were stained by Mayer's haematoxylin and eosin, Crossmonn's triple stain or McManus's periodic acid Schiff (PAS) technique for histological examinations. Diameters of follicles were measured using ocular micrometer.

Results

Ovarian anatomy. Ovaries of porcupine were located in the sublumbar area, caudal to the kidneys and under the third or fourth lumbar vertebra. A large bursa ovarica surrounded the ovary completely. Owing to the repeated ovulations that ovaries had gone through, the surface appearance of the ovaries was rough.

Ovaries were bean-shaped and the masses of both ovaries were comparable (0.15 ± 0.2 gr). The length, width and thickness of the ovaries were almost the same for the right (1.5 ± 0.33, 1 ± 0.25, 0.3 ± 0.03 cm) and left ovary (1.5 ± 0.33, 1 ± 0.33, 0.3 ± 0.02 cm).

Ovarian histology. Over the portions of the ovary covered by low columnar or cuboidal epithelium, cells frequently appeared to be detaching from the surface. Tunica albuginea, a poorly vascularized, dense, irregular collagenous connective tissue capsule. Each ovary was subdivided into the highly cellular cortex and medulla, which consisted mostly of a richly vascularized loose connective tissue. The cortex, which was surrounded on the outside by the surface epithelium, contained germ cell clusters, some primordial, primary, secondary, tertiary and atretic follicles, and dilated blood vessels.

Primordial follicles in the ovary of the porcupine were found to be distributed under tunica albuginea and extended deeply for a short distance into the cortical tissue. The primordial follicle was composed of a primary oocyte surrounded by a single layer of

flattened follicular cells cuboidal in shape. Primordial follicles were approximately 80 μm in diameter (Fig. 1).

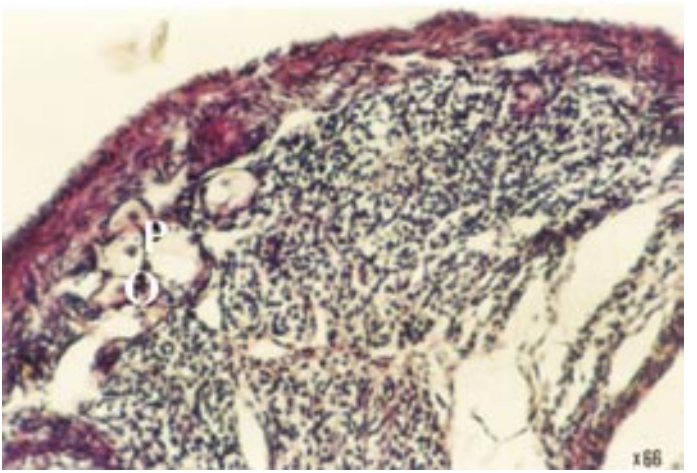

Fig. 1. Portion of porcupine ovary, showing surface epithelium. A few primordial follicles (P) and their oocyte (O) were seen. H&E.

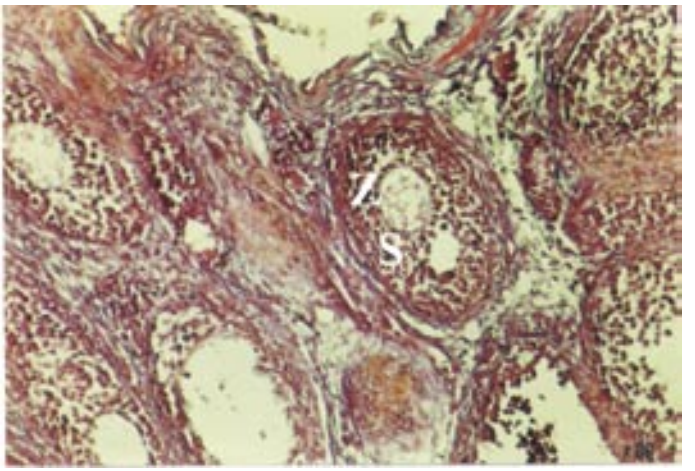

Fig. 2. Secondary follicles (S), porcupine. The primary oocyte was surrounded by a zona pellucida (Z) and a stratified epithelium of polyhedral cells. Triple stain.

Primary follicles were composed of a primary oocyte surrounded by a simple cuboidal and a simple columnar epithelium of follicular cells. Primary follicles were approximately 150 μm.

Secondary follicles were composed of a primary oocyte surrounded by a stratified epithelium of granulosa cells (Fig. 2) and were approximately 210 μm in diameter.

Tertiary follicles were composed of a primary oocyte surrounded by a stratified epithelium of follicular cumulus cells. The stratum granulosum was surrounded by the theca, which in tertiary follicles differentiates into two layers: an inner vascular theca interna and an outer supportive theca externa (Fig. 3). The theca interna cells were spindle-shaped and located in a delicate reticular fibre. The theca externa consisted of a thin layer of loose connective tissue with fibroblasts arranged concentrically around the theca interna. In these follicles, PAS positive reaction was seen in the zona pellucida. Tertiary follicles were approximately 1600 μm in diameter.

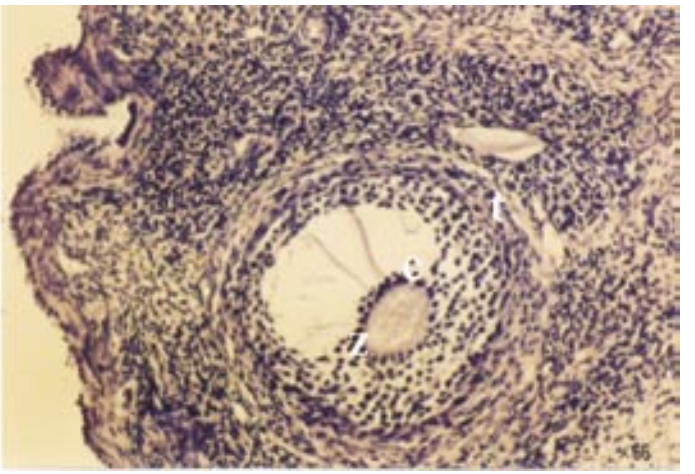

Fig. 3. Vesicular follicle from a porcupine ovary. The follicular antrum has formed. c-cumulus oophorus; z-zona pellucida; t-thecal cells. P.A.S.

Numerous atretic follicles were observed in different periods of development. Atretic follicles were small in the early period of follicle development, but large in the latter stages of development of the atresia of the follicle. Large luteal cells contained eosinophilic granules in the cytoplasm, whereas there were no granules in the cytoplasm of small luteal cells, fibroblasts or vascular endothelial cells.

It was found that the medulla was structured from loose connective tissue, blood and lymph vessels. The interstitial endocrine cells were able to be distinguished from ovarian stromal cells.

Discussion

The morphological structure of ovaries in the porcupine has been studied by light microscopy. The cortex of ovaries had many follicles which were in various stages of development, and many corpora lutea, interstitial cells, and stromal elements. A number of investigators and authors (DELMAN and BROWN, 1981; HAFEZ, 1970; OZDEMIR and DINC, 2002 and 2003; WILLIAM, 1986) have reported that the ovaries of domestic mammalian and laboratory animals, which have exocrine and endocrine functions, consist of an outer cortex and an inner medulla; the surface epithelium was simple cuboidal in young animals and simple squamous in older animals; the cortex contained follicles, which were in various stages of development, while the medulla consisted of loose connective tissue, as well as blood and lymph vessels. In the present study we observed similar histological findings in the ovaries of porcupine.

Polyovular follicles in the ovaries of some rodents have been described previously, for instance, in hamster (BODEMER et al., 1959; BODEMER and WARNICK, 1961; WEAKLEY, 1969), guinea pig (COLLINS and KENT, 1964). However, there are several reports that contradict the results on polyovular follicles. OZDEMIR and DINC (2002), and ODOR and BLANDAU (1968) have not observed polyovular follicles in the guinea pig and mouse, respectively. In the presented study, polyovular follicles were not observed in the ovaries of porcupine, which is consistent with the latter reports.

Many studies measuring follicle diameter have been carried out in various animals and was found to be: 18-300 μm in guinea pig (OZDEMIR and DINC, 2002), 100-399 μm in mouse (NUMAZAWA and KAWASHIMA, 1982) and 0.1-0.8 mm in rat (O'SHEA, 1970). We measured follicle diameter as 80-1600 μm.

At all stages studied the epithelial cells vary in shape over the surface of the same ovary, from squamous to low columnar. The differences in shape appear to result from simple mechanical deformation of otherwise identical cells. It was ascertained that ovarian volume and number of mitoses in the surface epithelium varies in the adult with the oestrus cycle. Thus, the change in cell shape from squamous to low columnar could be a response to local volume changes on the one hand, or to crowding resulting from mitotic activity on the other. Similar observations have been described for the ovaries of guinea pig (OZDEMIR and DINC, 2002 and 2003) and hamster (WEAKLEY, 1969).

The cortex contained oocytes surrounded by one layer of follicular cells, which were either flattened, cuboidal or columnar in shape. The zona pellucida was forming in most follicles with cuboidal or columnar cells. Zona pellucida was found to be PAS-positive. In the developing follicles, the follicular epithelium rests on a thin basal lamina and the follicles were separated from one another only by thin processes of connective tissue cells. Similar findings have also been observed in mouse (ODOR and BLANDAU, 1968) and guinea pig (OZDEMIR and DINC, 2002).

In conclusion, it was determined that the morphology and constituent cell types of the ovaries were similar to those described for other rodents.

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Received: 8 January 2004
Accepted: 1 March 2005

OZDEMIR, D., A. AYDIN, S. YILMAZ, G. DINC, O. ATALAR: Morfologija jajnika dikobraza (*Hystrix cristata*). Vet. arhiv 75, 129-135, 2005.

SAŽETAK

Istraživanje je provedeno u svrhu proučavanja građe jajnika dikobraza (*Hystrix cristata*). Jajnici su izvađeni laparoskopskim zahvatom, prerezani u dva dijela i fiksirani u puferiranom formalinu. Rezovi od 5-7 µm izrezani su od parafinskih blokova te bojani hematoksilinom po Mayeru, Crossmonn-ovim trostrukim bojanjem i McManus-ovom PAS tehnikom. Histološkom pretragom ustanovljeno je da se sastoje od kore i srži. Površina jajnika bila je valovita i obrubljena jednoslojnim kubičnim ili cilindričnim epitelom. Tunica albuginea bila je građena od vezivnotkivnih vlakana. Kora se sastojala od stanica vezivnog tkiva i intersticijskih stanica koje su sadržavale lipide. Primordijalni, primarni, sekundarni i tercijarni folikuli, zatim žuto tijelo i atretični folikuli bili su nađeni u kori jajnika. Prosječan promjer ovih folikula iznosio je između 80 i 1600 µm. Srž je bila sastavljena od rahlog vezivnog tkiva s krvnim i limfnim žilama različite veličine. Masa i veličina (duljina, širina i debljina) desnih i lijevih jajnika bile su uglavnom jednake.

Ključne riječi: *Hystrix cristata*, dikobraz, jajnik, građa