Global Veterinaria 13 (4): 560-569, 2014
ISSN 1992-6197

DOI: 10.5829/idosi.gv.2014.13.04.8549

Some Prenatal Histological Developmental Features of the Ovary in Dromedary Camel (*Camelus dromedarius*)

[1]A. M. A. Abd-Elrazik, [1]M.F. Nawito, [2]El-Sayed M. Taher,
[1]Karima Gh. M. Mahmoud and [1]Yousef F. Ahmed

[1]Department of Animal Reproduction and A. I,
National Research Center, Dokki, Giza, Egypt
[2]Department of Anatomy, Faculty of Veterinary Medicine, Cairo University, Giza, Egypt

Abstract: A total number of eighty one female embryos and fetuses of the one humped camel was collected from Cairo slaughterhouse in Egypt. The collected specimens were classified into groups representing the progressive stages of development from the 3rd to the 13th month of gestation to cover all the developmental stages. The gonads of camel embryos with Crown Vertebral Rump (C.V.R. length) more than 5.2 cm were found to be closely similar in their structure to those of the indifferent gonad. The gonad of camel fetus 87 days was clearly demarcated into cortical and medullary regions. The ovaries of fetuses at the fourth and fifth months of development were covered by the columnar ovarian epithelial cells. At late fourth month and early fifth one (18.5 to 24 cm C.V.R.L.) more primordial follicles could be observed at the corticomedullary junction of the ovaries of camel fetuses. The ovaries of camel fetuses with more than 24 cm C.V.R.L. up to 49.5 cm (fifth to seventh month), showed more progressive changes in germ cells as well as increased number of primary follicles that were observed with variable sizes during the seventh month. Follicles with more than one layer of folliclar cells surrounding the enlarged oocytes were firstly observed within the medulla during the late seventh month. Elastic -fibers began to appear in ovaries of camel fetuses during 8th and 9th months. During the early eleventh month, growing follicles without antrum, were noticed to increase in number. Also antral follicles with typical stratification of the follicular cells and theca folliculi were noticed for the first time in ovaries of camel fetuses with 90 cm C.V.R. length (during the eleventh month). The ovaries of camel fetuses during the twelfth and thirteen months were noticed to contain many antral follicles with variable sizes, occupying the whole thickness of the cortex. It was concluded that this study form an initial database for camel which may be helpful in the subsequent studies.

Key words: Dromedary · Ovary · Prenatal · Histology

INTRODUCTION

Camel is an important component of the desert ecosystem from time immemorial and is recognized as the "Ship of the desert". Humans depend on this animal not just for meat, milk and hide but also as one of the most important mode of transport in the desert. The genus Camelus has two species, one humped camel found in Africa, Arabia, Iran, Afghanistan and India and two-humped camel found in Central Asia reaching up to Mongolia and Western part of China. Investigations have been carried out to reveal some aspects of the physiological and anatomical mystery of camels in attempt to improve their productive efficiency [1-4].

To exploit the recent advances in techniques such as superovulation and embryo transfer, a sound understanding of the reproductive biology of the camel is essential, including an in-depth appreciation of normal anatomy and histology of the reproductive tract of this species [5-6]. Research work on the morphology, gross and developmental anatomy of ovary of dromedary camel [7-11] has been reported in different countries by many

Corresponding Author: M.F. Nawito, Department of Animal Reproduction & A. I, National Research Center, Dokki, Giza, Egypt. Tel: +2 01001563339, Fax: +2 02 33370931.

researchers. Despite the importance of the ovary for reproduction through the production of oocytes and the secretion of female sex hormones, its development during embryogenesis remains poorly understood. Moreover, information on prenatal morphological development of camel ovary is lacking [12-14].

During the early stages of embryonic development in mammalian species, germ cells migrate from the yolk sac to the mesoderm-derived undifferentiated gonad. In the female fetus, these cells then differentiate into oogonia, proliferate by mitosis and enter the process of meiosis [15]. At this point, the first and largest wave of germ cell death can be identified. The surviving oocytes can then progress to the dictyotene stage of prophase I, when meiosis is arrested until around the time of ovulation later in life [16]. Apoptosis is an organized process of cell death involved in the homeostasis of many tissues and organs, including the ovary. Indeed, apoptosis in camel [12] occurring at similar rates in cattle and buffalo ovary [17], between 4 and 8 months of foetalage. The present study aimed to investigate the histology of fetal ovary of dromedary camel at Egypt at different developmental stages.

MATERIALS AND METHODS

Specimens: A total number of eighty one female embryos and fetuses of the one humped camel was collected from Cairo slaughterhouse. Breeding history was not available. The sex was determined from the external genitalia and the specimens of the early stages of development could not be differentiated grossly. The collected specimens covered most of the developmental stages (5-120 cm Crown Vertebral Rump, C.V.R.).The C.V.R. was measured from the start point of the fore-head up to the base of the tail along the dorsum of the fetus. Fetal age was determined according to the formula given by El-Wishy *et al*. [18]:

$$\text{Fetal age in days} = \frac{\text{C.V.R. (cm)} + 23.99}{0.366}$$

Classification of Developmental Stages: As soon as the uteri were removed from the carcasses, the embryos or fetuses were freed from their fetal membranes. The abdominal cavity of each fetus was evacuated, the two ovaries were examined *in situ*. The ovaries were removed and freed from extraneous tissue. The available specimens were classified macroscopically into groups taking in consideration the position of the gonad and the appearance of the different structures. These groups were classified after the second month (undifferentiated period) into the following:

Group I: 25 specimens ranged from 7.9 cm to 29 cm C.V.R.(early third to late fourth months).

Group II: 25 specimens ranged from 29.5 cm to 49 cm C.V.R.(fifth to seventh months).

Group III: 11 specimens ranged from 49.5 to 69 cm C.V.R.(late seventh to early ninth months).

Group IV: 9 specimens ranged from 69.5 cm to 89 cm C.V.R.(late ninth to early eleventh months).

Group V: 8 specimens ranged from 89.5 cm to 109 cm C.V.R.(late eleventh to twelfth months).

Group VI: 3 specimens were obtained with 120 cm C.V.R. (thirteen month).

Histological Examination of Dromedary Ovaries: The fetal ovaries were incised longitudinally and quickly transferred to the fixative used which was either 10% neutral formal saline or Bouin's fluid. The embryos of the early stages up to 6.5 cm C.V.R. were taken, to the fixative, intact. The caudal part of these specimens as well as the ovaries of the other specimens, after being fixed, were processed and embedded in Altmann's paraffin wax with melting point 56-58°C.

Step serial sections with a thickness not more than 5 micrometer were prepared and stained with the following stains:

Haematoxylin and eosin for general studies.

Van Gieson's stain for demonstration of collagenous fibers and muscle cells.

Crossmontrichrome stain for further differentiation.

Weigert's elastic tissue Van Gieson combination stain for demonstration of elastic fibers, collagenous fibers and muscle cells.

Gomorireticulin method for demonstration of reticular fibers.

PAS Alcian blue combination for demonstration of both neutral and acid mucopolysaccharides.

The aforementioned stains were conducted as out-lined by Drury and Wallington [19].

RESULTS AND DISCUSSIONS

Undifferentiated Period: The gonad of camel embryo with C.V.R. length 5-6.5 cm (80-83 days) was found to be covered by elongated wedge shaped coelomic epithelial cells, with narrow proximal end implanted to the underlying gonadal stroma, while its distal end was thick, adapted to the distal end of neighboring cells with spherical or oval lightly stained nucleus (Fig. 1). These findings were in partial agreement with that reported, by other authors, in mammals. George and Fahmy [20], El-Ghannam and El-Naggar [21] and Gondos [22], described the coelomic epithelial cells covering the ovaries of camel fetus (8-12 weeks), buffalo fetus (third month) and human fetus (7-9 weeks) as being cuboidal, rounded and columnar respectively. They reported the presence of primitive germ cells in between this epithelial layer.

The ovarian surface epithelial cells was found to be continuous with the underlying ovarian structures through their cytoplasmic processes and without demarcation by her either basement membrane or tunica albuginea. Such continuity gave an easy way for the primordial germ cells to migrate to the ovarian structure. The primordial germ cells showed large spherical lightly stained nuclei that contain one to three nucleoli and abundant lightly acidophilic granular cytoplasm. The three cell types that constituted the major art of the gonad at that time of prenatal life were: 1- Elongated cells with elongated lightly stained nuclei that showed clear nucleoli and their cytoplasm extended fine threads at the two poles of the nuclei. 2. Cells large oval indented lightly stained nuclei and perinuclear irregular cytoplasm. 3. Cells with angular, large pyramidal, lightly stained nuclei and irregular perinuclear cytoplasm. These cells, most probably, the forerunners of the gonadal stroma cells. Two types of undifferentiated cells in addition to the primitive germ cells were described in the ovary of camel fetus (8-12 weeks stage) by George and Fahmy [20] who observed them to be condensed at the periphery (primary cortex) and loosely arranged in the center (primary medulla).

The gonads of camel embryos with C.V.R. length more than 5.2 cm were found to be closely similar in their structure to those of the indifferent gonad. These specimens might be considered as an extension of the in different gonad as no signs of meiosis of germ cells could be noticed. The cellular elements of the gonad of camel embryo at this stage were supported with a well organized reticular net, the fibers of which were more condensed at the periphery of the gonad as well as around the developing blood cells.

Group I: Early Third to Late Fourth Months: In fetuses with C.V.R.L. 7.9 cm (87 days), the reticular network became more organized and appeared closely meshed. The gonad of camel fetus of 87 days was clearly demarcated into cortical and medullary regions. George and Fahmy [20] stated such demarcation in the ovaries of camel fetus of 56-84 days old. The central part (medulla) showed organized blood vascular bed, delicate collagenous fibers as well as clusters of small darkly stained cells that show dark nuclei presumably rete ovari cells. Byskov [23] stated that the in-growing mesonephric cells form cell cords or cell bodies of varying size, connecting the mesonephros or mesonephric tubules and the gonad.

The ovaries of fetuses at the fourth and fifth months of development were found to be covered by the columnar ovarian epithelial cells, besides the aforementioned cells described in the previous stage. As C.V.R. length increased, this layer was noticed to vary from columnar to cuboidal type almost in mitosis and showed spherical nuclei situated in the distal part of the cells. The inter-posed primordial germ cells increased in number showing mitosis. Invagination of the ovarian surface epithelium was noticed in the ovary of camel fetus with 17 cm C.V.R. length and became prominent and more deeper with 24 cm C.V.R.L (Fig. 2). El-Tayeb [8] mentioned that these infoldings of the germinal epithelium, begin in camel fetus with 16 cm C.V.R.L.

The cortex appeared to contain numerous germ cells with the least amount of stroma cells, at the peripheral under the ovarian surface epithelium. These germ cells were in direct contact with the surface epithelium. Deeper to these peripherally situated germ cells, oogonia as well as primary oocytes appeared in the form of clusters of cells surrounded by stroma cells with their elongated nuclei arranged in a concentric manner. The aforementioned cell clusters were noticed to constitute most of the cortical region. Towards the medulla the clusters of cells were mostly of the primary oocytes showing the different forms of the transitory stages of the first meiotic prophase. At the corticomedullary junction, the oocytes appeared in late stage of the first meiotic prophase (diplotene) characterized by its large size and, sometimes, by the presence of an incomplete investing layer of flattened

cells (Fig. 3) constituting the primordial follicles. Oogenesis and early follicle formation have been regarded as commencing in the inner part of the cortex in close proximity to the medulla by Erickson [24] in bovine; Black and Erickson [25] in pig; Ghannam and Deeb[26] and El-Ghannam and El-Naggar [21, 27] in buffalo.

The clusters of oocytes at the corticomedullary junction were frequently observed in association with the rete ovarii cells that appeared in the form of cell cords. These oocytes were almost in late stage of the first meiotic prophase. Byskov [28] reported that the first follicles occurring within the ovary of cat, mink and ferret are connected to the contact area with the intraovarian rete system. The same author added that the differentiation of germ cells and granulosa cells are initiated as a result of interaction of medullary factors and the cortex.

The frequent association of the germ cells with the rete ovarii tubules or cords observed in ovaries of camel fetuses during the fourth month would suggest the possibility of the rete cells to be differentiated into granulosa cells and coincides with the results of Byskov and Lintern-Moore [29] who stated that the granulosa cells develop within the rete cords from adjoining rete cells.

Germ cells that migrated through the ovarian surface epithelium were almost observed to be accompanied with cells from that layer. This accompanied cells could differentiated juxta oocytes into layer of granulosa cells as such germ cells came in contact with rete ovarii cords. Byskov [28] in cat, mink and ferret, the granulose cells could originate from both the surface epithelium and the rete ovarii cells.

At 18.5 to 24 cm C.V.R.L., more primordial follicles could be observed at the corticomedullary junction of the ovaries of camel fetuses. These follicles were always noticed in association with rete ovarii. Further growth of these follicles were noticed by the increased size of the oocyte which appeared in the dictyotene stage of meiotic prophase and the change of the surrounding flattened granulosa cells to cuboidal type (Fig. 4). This was observed for the first time in ovaries of camel fetus with 24 cm C.V.R.L. (5th month). Earlier in development (12-16-weeks), George and Fahmy [20] firstly reported primary follicles in the medulla of the ovaries of camel fetuses and added that these folicles which are deeply seated in the medullary tissue show signs of degeneration.

The medulla was noticed to be looser in texture, rich in stroma cells and collagenous fibers, blood vessels as well as rete ovarii and clusters of oocytes were noticed embedded in the medullary stroma. Newly formed blood vessels started, in the form of haemocytopoietic foci. which was supported by reticular net. Some of the germ cells located within the medulla showed signs of degeneration. El-Ghannam and El-Naggar [21] mentioned that high levels of mitosis as well as degeneration are observed in ovaries of buffalo fetuses during the fourth month.

Rete ovarii appeared in the form of cords of cells that showed canalization in ovaries of camel fetuses with 12.5 cm C.V.R. length during the fourth month. At the vicinity of this cords, number of germ cells either in clusters or sporadic form showing the transitory stage of meiotic prophase were observed.

The cortical zone showed reticular fibers only around the clusters of oogonia and oocytes. The sub-epithelial clusters of primordial germ cells were devoid of reticular support. At the corticomedullary junction, the reticular fibers were noticed to support the clusters of oocytes as well as the blood vessels.

Group II: Fifth to Seventh Months: The ovaries of camel fetuses with more than 24 cm C.V.R.L. up to 49.5 cm (fifth to seventh month), showed more progressive change in germ cells as well as increased number of primary follicles that were observed with variable sizes during the seventh month. The surface epithelium was columnar and cuboidal type of cells (late fifth month) changed to columnar cells with the distally situated nuclei (sixth and seventh month). These cells showed mitotic figures that became rarely observed in ovaries of fetuses during the seventh month. The inter-posed germ cells which were numerous during the fifth month, appeared few in number during the seventh month. George and Fahmy[20] mentioned that the germinal epithelium consists of low cubical cells that show considerable mitotic activity, in camel fetuses with 20-24 weeks old. These epithelial cells, begin to assume the columnar shape in some parts although the cuboidal type still predominate, in camel fetuses with 24-48 weeks old. Marai *et al*. [30] reported that the germinal epithelium of flattened and cuboidal cells in the ovary was changed to cuboidal and columnar types, 24-48 weeks of camel fetal age.

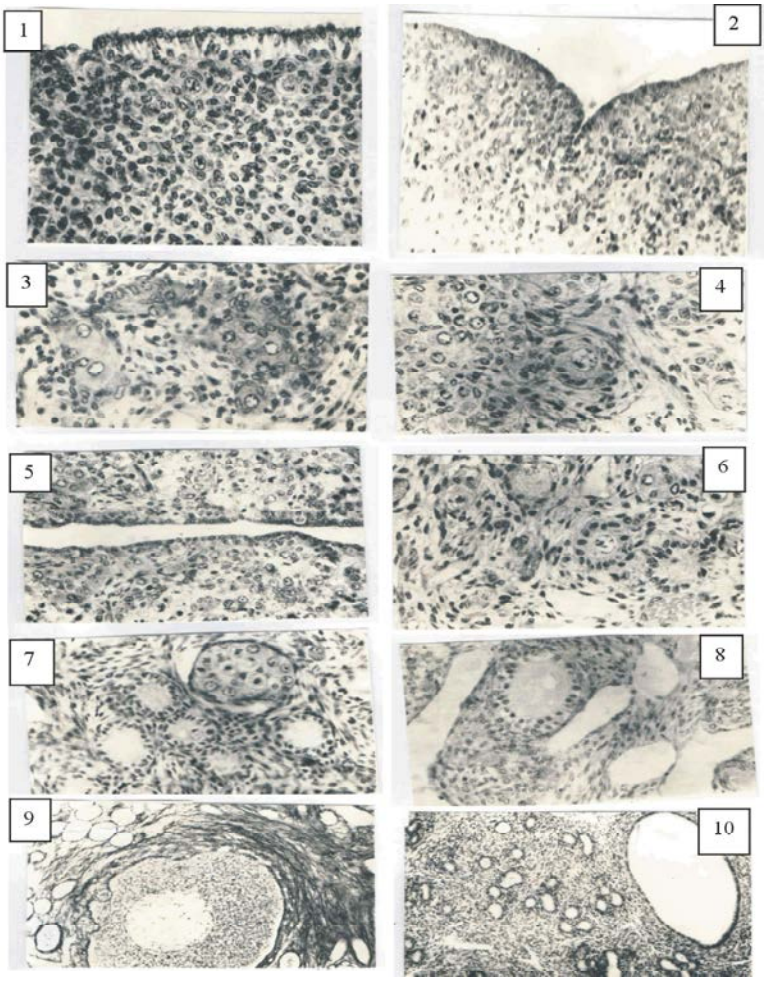

Fig. 1: Photomicrograph of indifferent gonad of camel embryo with 5.0cm C.V.R.l., showing surface epithelial cell and germ cell in outer zone of the gonad (H&E X400).

Fig. 2: Photomicrograph of the ovary of camel fetus with 24.0cm C.V.R.L, showing invagination of the surface epithelium (H&E. X200).

Fig. 3 :Photomicrograph of the ovary of camel fetus during the fourth month, showing primordial follicle, oocyte with peripheral chromatin surrounded by an incomplete layer of flattened cells (Crossmon's trichrome stain X400).

Fig. 4: Photomicrograph of the ovary of camel fetus during the fifth month, showing primary follicle, clusters of oocytes and rete cell clusters (H&E. X 400).

Fig. 5: Photomicrograph of the ovary of camel fetus during the seventh month, showing deep invagination of the surface epithelium lined with columnar cells, germ cells are found in between this cell layer (H&E.X 200).

Fig. 6: Photomicrograph of the ovary of camel fetus during the seventh month, showing primordial and primary follicles (H&E. X40x3.2).

Fig. 7: Photomicrograph of the ovary of camel fetus during eighth month showing primordial and primary follicles and clusters of oogonia and oocytes ensheathed with stroma cells and fibers (Van Gieson's stain X40x2.5).

Fig. 8: Photomicrograph of the ovary of camel fetus during ninth month, showing partial stratification in the follicular cells as well as lymph vessels (Van Gieson's stain X40x2.5).

Fig. 9: Photomicrograph of the ovary of camel fetus during the early eleventh month, showing reticular fibers more condensed in theca interna (Gomori's reticulin method X16x3,2).

Fig. 10: Photomicrograph of the ovary of camel fetus during twelfth months ,showing dilated rete ovarii tubule as well as lymph vessels (H&E X20x2.5).

The invaginations of the surface epithelium increased in number and depth, with the increased fetal age. Some of these invaginations were noticed to go deep in a relative distance through the cortex of the ovaries at the seventh month of fetal life (Fig. 5). The lining epithelium of the deep invagination was of the columnar type that showed, inbetween them, few number of germ cells (Fig. 5). On the contrary, George and Fahmy [20] in camel fetus stated that these infoldings decrease in depth with the increase of fetal age.

The cortex was noticed to consist of germ cells (oogonia and oocytes), occupying most of the cortical region. The oogonia showed mitotic figures while the oocytes were noticed in the transitory stages of the first meiotic prophase. These germ cells were arranged in the form of irregular clusters demarcated by elongated stroma cells that enclosed vascular bed around the deeply situated clusters. The peripherally situated germ cells (oogonia) that appeared in clusters with the least amount of stroma cells, were continued with the underlying clusters of oocytes and incompletely separated from the surface epithelium by the interrupted basement membrane. Towards the end of this stage, oogonia became less in number while oocytes increased in number and showed advanced stage of meiosis. This results coincides with that reported by El-Tayeb [31] who stated that the cortex of ovaries of camel fetuses with 29 to 50 cm C.V.R.L., contains more oocytes than oogonia.

As the oocytes reached the diplotene form of the first meiotic prophase, they were invested with flattened follicular cells. These primordial follicles, that were noticed to be increased in number, with age, proceed in growth to become primary follicles (Fig. 6). These primary follicles appeared in moderate number during the sixth month and some of them were noticed in association with rete ovarii. Byskov [28] stated that several small follicles are visible in the ovary of cat, mink and ferret and some of them are still connected to the rete cords.

Group III: Late Seventh to Early Ninth Months: The ovaries of the camel fetuses during the seventh month, showed increased number of follicles that were noticed to occupy the deeper part of the cortex towards the corticomedullary junction. George and Fahmy [20] stated that the ovarian cortex of camel fetuses with 20 to 24 weeks, is formed mainly from primary follicles and cell cords. El-Ghannam and El-Naggar [27] pointed out that the increase in the primary follicle formation, in buffalo fetuses during the fifth month, is concomitant with the peak level of meiosis.

The tremendous number of follicles that were noticed in ovaries of camel fetuses during the seventh month (49.5 cm C.V.R.L.), appeared with variable sizes; these follicles showed enlarged oocytes with eccentrically situated nuclei, surrounded by cuboidal cells with rounded or somewhat oval nuclei. An increase in the vasculature of the cortical region was noticed through the appearance of blood vessels.

Follicles with more than one layer of folliclar cells surrounding the enlarged oocytes were firstly observed within the medulla during the late seventh month. George and Fahmy [20] stated that the medulla contains great number of primary follicles, in camel fetuses with 16-20 weeks and shows few number of follicles with two layers of follicular cells. These follicles, appear with several follicular cell layers in the medulla of camel fetuses with 24-48 weeks. They added that, at the same age the cortex shows follicles with one or more layers of follicular cells, a condition which was not noticed during the present study in camel fetuses at late seventh month of pregnancy.

The rete ovarii which constituted part of the medulla was noticed as cell cords and tubules almost in association with groups of oocytes at the corticomedullary junction. These were referred to as intraovarian rete cords and tubules according to the nomenclature proposed by Byskov and Lintern-Moore [29] who recorded three parts in the rete system of mouse; the intraovarian rete with connection with oocytes and follicles, extraovarian rete in the periovarian tissue and connecting rete between the intraovarian and extra-ovarian rete. The first part (intraovarian) was firstly noticed in ovaries of camel fetuses during the 4^{th} and 5^{th} month stage. The third part (connecting rete) was clearly observed near the hilus, within the medulla, as a tubular structure that showed argyrophilic. alcianophilic and PAS positive intraepithelial gland cells secretions. Secretory activity in the rete has been reported by Byskov [28] in cat, mink and ferret. Mucin production in the rete tubules of the dog was reported by O'shea [32]. Shehata [33] reported the eosinophilic secretion in the medullary tubules of camel fetus.

The fibrous stroma of the ovaries of camel fetuses, at the fifth to late seventh month, consisted of reticular and collagenous fibers. Atresia of some oocytes in transitory stages of the first meiotic prophase was observed in those located in the deepest part of the cortex. El-Tayeb [31] stated that most of the degenerated and necrotic germ cells are found in the deepest part of the cortex as well as the peripheral part of the medulla of ovaries of camel fetuses with 29-50 cm C.V.R.L.

The ovarian surface epithelium of camel fetuses at the eighth and ninths months of development was noticed to vary from columnar and low columnar cells (eighth month), to columnar, cuboidal and squamous type (ninth month). The basement membrane appeared interrupted. The interposed germ cells were rarely observed. The invaginations showed no difference than that observed in ovaries with the previous state (late seventh month).

George and Fahmy[20] stated that the germinal epithelium consists mainly of columnar cells exceptionally cuboidal type with large primitive oogonia scattered between them in ovaries of camel fetuses with 28-32 weeks and added that the infoldings appear to be much shallower. El-Tayeb [31] mentioned that these infoldings are deep and some of them involve the whole thickness of the cortex of ovaries of camel fetuses with 50.5-75 cm C.V.R. length.

The cortex showed, in its upper most part, few number of oogonia and oocytes. The clusters of germ cells that were noticed to occupy most of the cortical region, were mainly oocytes in the transitory stages of the first meiotic prophase. At the deep cortex and corticomedullary junction great number of primordial and primary follicles of variable sizes were noticed and appeared to form clusters (Fig. 7), during the eighth month. These findings were nearly coincide with El-Tayeb [31]who stated that the primary follicles are clear and abundant in ovaries of camel fetuses with 50.5-75 cm C.V.R.L.

Some of the follicle clusters which were clear during the sixth and seventh months were noticed at the present stage of development to be separated into identical follicles that invaded the medulla. Within the medulla, the follicles appeared, widely separated, embedded in the medullary stroma and some of them showed irregular stratification of the follicular cells (Fig. 8). George and Fahmy [20] pointed that the medulla of the ovaries of camel fetuses with 28-32 weeks, contains a considerable number of primary and secondary follicles, the former are greater in number, near the cortex than the latter. The presence of growing follicles with follicular cavity, reported by George and Fahmy [20], in camel fetuses with 28-32 weeks, were not observed in the present study within the ovaries at this stage (8^{th} & 9^{th} months). El-Tayeb [31] stated that the growing follicles appear in ovaries of camel fetuses with 50.5-75 cm C.V.R.L.

The cortical stroma cells and fibers were noticed to be increased gradually from the surface toward the deep part of the cortex and at the corticomedullary junction. The increase of the vasculature of the cortex, were noticed through the appearance of plexus of blood vessels in the outer part of the cortex. George and Fahmy [20] and El-Tayeb [31] reported the increase of the stroma cells and fibers as well as the vasculature of the cortical region of ovaries of camel fetuses with 28-32 weeks and 50.5-75 cm C.V.R.L. respectively.

At the periphery of the cortex, more concentration of stroma, fibers and cells, appeared underneath and parallel to the surface epithelium indicating the differentiation of the tunica albuginea in ovaries of camel fetuses with 67.5 cm C.V.R. length (ninth month). The tunica albuginea, at that age, was noticed to be interrupted by clusters of oogonia and oocytes that were observed in direct contact with the surface epithelium. Similar results were reported by George and Fahmy [20]and El-Tayeb [31].

The medulla of the ovaries of camel fetuses within this stage, showed increase in the spindle shaped stroma cells and fibers (collagenous and reticular) and blood vessels. Great number of lymph vessels were also noticed, follicles with different sizes were noticed widely separated, embedded in the medullary stroma and stratification of the follicular cells was observed in some follicles in the outer medulla. At the hilus region, rete ovarii in the form of cell cords and tubules, were noticed. The rete cells were columnar in shape, with ovoid lightly stained, basally situated nuclei with condensed chromatin. George and Fahmy [20] stated that the rete ovarii, in camel fetuses with 28-32 weeks, appears, at the hilus region, as epithelial strands. Byskov [28] stated that the coiled cords of cells (hilar gland), at the hilus region of the ovaries of mink and ferrent, are in continuation with the extraovarian rete. The same author added that this tissue may have a different appearance in different mammalian species. Elastic -fibers began to appear in ovaries of camel fetuses during this stage of development (8^{th} and $9t^{h}$ months) and increased in amount, number and thickness in advanced stages.

Group IV: Late Ninth to Early Eleventh Months: The ovaries of camel fetuses during late ninth, tenth and early eleventh months, were found to be covered with low columnar, cuboidal and flattened epithelial cells, that became cuboidal and flattened in ovaries of camel fetuses during late eleventh and twelfth months of pregnancy. The interposed germ cells that were noticed with very few number during tenth and eleventh months, were not observed during the twelfth month. The germinal epithelium was reported by George and Fahmy [20] to consist mainly of columnar cells exceptionally cuboidal in ovaries of camel fetuses with 32-36 weeks old that was considered by the authors to be the last stage of prenatal

development. They pointed out that the cuboidal cells are observed only where large secondary follicles lie close to the surface. Ghannam and Deeb [26] and El-Ghannam and El-Naggar [27] reported that the covering epithelium in ovaries of buffalo fetuses with more than 500 mm C.R. length and during the last trimester, is of low cuboidal and flattened epithelial cells, respectively.

The invaginations that appeared wide and deep during late ninth month (73 cm C.V.R.L.), became more deeper and frequent in ovaries of camel fetuses during the tenth month (85 cm C.V.R.L.). These invaginations were noticed to be lined with low columnar and cuboidal cells. Oogonia and oocytes appeared to lie close to the surface epithelium in some areas that showed the flattened type of cells. These findings were in accordance with that reported by El-Tayeb [8] and Abdel- Elrazik [12]and contradict the results of George and Fahmy [20] who pointed out that these invaginations become much shallower and some of them disappear, in ovaries of camel fetuses with 32-36 weeks old that was considered by the same authors to be the last stage of prenatal development.

The tunica albuginea during late ninth month, appeared more organized but still interrupted, in very few areas, by oogonia and oocytes that were noticed in direct contact with the surface epithelium. During early eleventh month (90 cm C.V.R.L.), the tunica albuginea became a continuous fibrous connective tissue layer, mostly of collagenous fibers, underneath the surface epithelium. Nearly similar results were reported by El-Tayeb [88] and Abdel- Elrazik [12].

The primary follicles which were newly enclosed by rete ovarii clusters were surrounded by PAS positive basement membrane. Such demarcation between the follicular epithelial cell layer and the rete cell mass was interrupted in parts and gradually disappeared. During the early eleventh month, growing follicles without antrum, were noticed to increase in number, occupying the deeper part of the cortex and the corticomedullary junction, besides the increased number of rete ovarii cords and clusters. Also antral follicles with typical stratification of the follicular cells and theca folliculi were noticed for the first time in ovaries of camel fetuses with 90 cm C.V.R. length (during the eleventh month) beside which rete ovarii cords of cells could be observed. The aforementioned findings supported the suggestion that, at least, the outer layer of follicular epithelial cells in growing follicles and the follicular epithelial membrane in the antral follicles were derived from the rete cells and differ in origin from the cells of the corona radiata and those of the cumulus oophorus.

The theca folliculi consisted of collagenous and reticular fibers, the latter type was found to be more condensed in the theca interna (Fig. 9). El-Tayeb [8] reported that growing follicles with acidophilic liquour folliculi are observed in ovaries of camel fetuses, with 94 cm C.V.R. length corresponding to the eleventh month, while George and Fahmy [20]; Abdin[7] and Marai *et al.* [30]mentioned that growing follicles with follicular cavity are observed in ovaries of camel fetuses with 32-36 weeks.

Group V: Late Eleventh to Twelfth Months: In camel fetuses, during late eleventh month, the ovaries showed two or even four oocytes invested in one layer of follicular cells. Black and Erickson [25] recorded that polyoocytic follicles with as many as "20" oocytes in a single follicle, are found in ovaries of 10 days post partum pig. The indented appearance of the reticular net that surrounded the polyoocytic follicle would indicate the expected separation of these follicles into identical ones. The ovaries of camel fetuses during the twelfth month were found to contain many antral follicles with variable sizes, occupying the whole thickness of the cortex. The small antral follicles showed small antrum filled with alcianophilic liquour folliculi and the follicular cells were noticed to contain in its intercellular tiny spaces alcianophilic substance. These findings added further support to the aforementioned suggestion that the follicular cells are rete cells derivative. These intercellular secretory substance, with further follicular growth, accumulated in the more developed larger antrum. The oocytes appeared lightly PAS positive in reaction. In the more larger follicules, the liquor folliculi was alcianophilic and PAS positive. The addition of neutral mucopolysaccharides to the follicular fluid was possibly a product of the cumulus oophorous and corona radiata. In the most larger follicles, liquor folliculi was PAS positive in reaction. The graduation in reaction of the liquor folliculi from alcianophilic, mixed (alcianophilic and PAS) and finally PAS positive that correlated with the gradually increased follicular. size, might be attributed to the gradual differentiation of the alcianophilic secretory rete cells that constituted the follicular wall, side by side with the increase of the already differentiated cell layer that was observed in direct contact with the PAS positive oocyte.

The follicular wall, of the antral follicle, was found to consist of several layers of follicular cells, next to the antrum, showing mitotic figures and resting on a basement membrane (external limiting membrane) and surrounded theca folliculi that appeared more organized

and differentiated into theca interna and outer theca externa. Theca interna was mostly cellular less fibrous, theca externa was mainly fibrous with fusiform cells. This findings were in agreement with that reported in ovaries of camel fetuses with 32-36 weeks, by George and Fahmy [20] and Abdel- Elrazik [12]. El-Tayeb [8] mentioned that the theca folliculi is very clear around the vesicular follicle, in camel fetuses with more than 94 cm C.V.R. length.

The cortical stroma, cells and fibers were found to be increased and appeared in the form of bundles running in all directions, joined peripherally the tunica albuginea and surrounded the ovarian follicles, blood vessels and rete ovarii cords and tubules. The medulla showed medullary stroma with its associated blood vessels, lymphatic vessels and rete ovarii, the fibrous stroma, appeared in the form of loosely arranged irregularly interwoven collagenous fibers that joined the adventitia of the enclosed blood vessels and pass towards the cortex where they joined the cortical stroma fibers. The vasculature of the camel fetal ovaries during this stage was noticed to increase in the medullary and cortical regions. These findings were consistent with that reported by George and Fahmy [20] who added that, primitive oogonial cords are still found in ovaries of camel fetuses with 32-36 weeks. In the medulla, the rete ovarii was found as convoluted tubules lined with columnar cells, besides, dilated tubules were noticed (Fig. 10). Byskov and Lintern-Moore [29] stated that the connecting rete tube in the mouse ovary, during the second week of life, is frequently dilated.

Group VI: Thirteen Month: In the examined sections of ovaries of camel fetuses with 120 cm C.V.R. length (thirteen month), no histological changes could be noticed than those of the previous stage.

CONCLUSION

The findings of this study form an initial database for camel which may be helpful in the subsequent studies. So ovary differentiation by molecular markers to identify early ovarian genes besides histological and immunohistochemical study must be performed specially in camel.

REFERENCES

1. Shalash, M.R., 1965. Some Reproductive aspects in female camel. World Rev. J. Anim Prod., 4: 103-108.
2. Nawito, M.F., M.R. Shalash, R.H. Hoppe and A.M. Rakha, 1967. Reproduction in female camel. Anim. Sci. Res. Inst., Bull., pp: 2.
3. Abdealla, O., 2000. Anatomical study of the female genital system of the one – humped camel. Sudan J. Vet. Sci. Anim. Husb., 6: 41-52.
4. Khanvilkar, A.V., S.R. Samant and B.N. Ambore, 2009. Reproduction in camel. Veterinary World, 2(2): 72-73.
5. Srikandakumar, A., E.H. Johnson, O. Mahgoub, I.T. Kadim and D.S. Al-Ajmi, 2001. Anatomy and histology of the female reproductive tract of the Arabian camel. Emir. J. Agric.Sci., 13: 23-26.
6. Abdel-Elrazik A.M.A., M.F. Nawito, S.E.l. Taher and K.Gh. M. Mahmoud, 2013. Prenatal morphological development of the ovary in dromedary camel (Camelus dromedaries). Global Veterinaria, 11(4): 372-376.
7. Abdin, A. El.M. M., 1978. Developmental changes of the genitalia and adenohypophysis in dromedary camel. M.Sc. Thesis, Faculty of Agriculture, Zagazig University.
8. El- Tayeb, M.M., 1981.The evaluation of the genitalia and endocrine glands of camel. Ph. D. thesis, Faculty of Veterinary Medicine, Zagazig Univ.
9. EL-Wishy, A.B., 1988. A study of genital organs of female dromedary (camelus dromedaries) J. Reprod Fert., pp: 587-593.
10. El-Wishy, A.B., 1992. Functional morphology of the ovaries of the dromedary camel. Proc. J. Anat. Anz., 131: 140.
11. Umaru M.A. and A. Bello, 2012.A study of the biometry of the reproductive tract of the one-humped camel (Camelus dromedarius) in northern Nigeriamore. Scientific Journal of Microbiology, 1(5): 82-86.
12. Abd-Elrazik, A.M.A., 1984. Some studies on the prenatal development of the ovary of one humped camel "camelus dromedaries". Ph.D thesis (anatomy), Faculty of Veterinary Medicine, Cairo University.
13. El Harairy, M.A., A.E.A. El Khalek, A.Z. Mehrez and H.E. El Metwaly, 1998. Prenatal development of the ovary of the camel Camelus dromedarius. First international conference on animal production and health in semi arid areas, El-Arish, Egypt, pp: 203-211.
14. Jaji, A.Z., H.D. Kwari, A.Y. Ribadu and M.N. Sivachelvan, 2010. Studies on prenatal development of dromedary ovaries in Maiduguri, Borno state. Sahel J. Vet. Sci., 10: 45-49.
15. Baker, T.G., 1963. A quantitative and cytological study of germ cells in human ovaries. Proc R Soc. London B, 158: 417-433.

16. Hirshifield, A.N., 1991. Development of follicles in the mammalian ovary. Int Rev. Cytol., 124: 43-101.
17. Santos, S.S.D., M.A.P. Ferreira, R.V. Sampaio, N.N. Costa, D.C.N. Santos, P.P.B. Santana, A.L.A. Sá, B.B. Silva, M.S. Cordeiro, T.V.G. Silva, M.S. Miranda and O.M. Ohashi, 2013. Evaluation of apoptosis as a mechanism of follicular cell atresia in the ovaries of cattle (bo sindicus) and buffalo (bubalus bubalis) fetuses. Anim. Reprod., 10: 55-61.
18. El-Wishy, A.B., N.A. Hemeida, M.A. Omar, A.M. Mobarak and M.A.I. El-Sayed, 1981. Functional changes in the pregnant camel with special reference to fetal growth. Brit. Vet. J., 137: 527-537.
19. Drury, R.A.B and E. A. Wallington, 1980. General histology procedures in Carleton's histological techniques 5 thEd. Oxford Univ Press, 7: 139.
20. George, A.N. and M.F.A. Fahmy, 1966. Histological study of the developing ovary of the dromedary (Camelus dromedaries). J. Vet. Sci., u.A.R., 3: 93-100.
21. El-Ghannam, F. and M.A. El-Naggar, 1974. The prenatal development of the buffalo ovary. J. Reprod. Fert. 41: 479-483.
22. Gondos, B., 1975. Surface epithelium of the developing ovary. Amer. J. Pathol., 81: 303-312.
23. Byskov, A.G., 1982. Primordial germ cells and regulation of meiosis, In Reproduction in mammals, Book I: Germ cells and fertilization, Chapter I, pp: 1-16, Ed. By Austin, C. R. and Short, R. V. Cambridge University Press.
24. Ereckson, B.M., 1966. Development and radio-response of the prenatal bovine ovary. J. Reprod. Fertility, 10: 97-105.
25. Black, J.L. and B.M. Ereckson, 1968. Oogenesis and ovarian development in prenatal pig. Anat. Rec., 161: 45-56.
26. Ghannam, S.A.M. and S. Deeb, 1969. Prenatal development and activity of the ovaries in buffaloes. J. Vet. Sci.(Cairo), 6: 105-119.
27. El-Ghannam, F. and M.A. El-Naggar, 1975. Studies of oocytogenesis of buffalo ovaries. ZbL. Vet. Med. Series A., 22: 248-255.
28. Byskov, A.G., 1975. The role of rete ovarii in meiosis and follicle formation in cat, mink and ferret. J. Reprod. Fertil., 45: 201-209.
29. Byskov, A.G. and S. Lintern-Moore, 1973. Follicle formation in the immature mouse ovary: the role of the rete ovarii. J. Anat., 116: 207-217.
30. Marai, I.F., T.M. elEnany and A.M. Abdine, 1990. Prenatal development of adenohypophyseal cell types, ovary and uterus of dromedary camel. Arch ExpVeterinar. med., 44: 581-589.
31. El-Tayeb, M.M., 1981. The evaluation of the genitalia and endocrine glands of camel. Ph. D. thesis, Faculty of Vet. Med., Zagazig University.
32. O'Shea, J.D., 1966. Histological observation on mucin secretion by subsurface epithelial structures in the canine ovary. J. Morph., 120: 347-358.
33. Shehata, R., 1964. Medullary tubes in the ovary of the camel and other animals. Vet. Rec., 76: 750-753.