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Original Research Article

Histomorphological Developmental Study of Uterus on Local Awassi Sheep Fetuses at Prenatal Period

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Abstract: This study was conducted at Al-Qassim Green University, Collage of Veterinary Medicine, Anatomy and Histology Branch. The experiment was designed to study the uterus of awassi sheep fetuses where take (16) samples sheep fetuses in prenatal stages are distributing into two groups: (second and third trimester of gestation) according to the gestational age, which is determined depending on the crown rump length (CRL) and divided into group I at (90) days of gestation and Group II at (140) days of gestation, containing (8) fetus each group to record the developmental changes in the uterus. In the first group (90) days the uterus appear muscular tubular organ and small in size with thin translucent wall composed of a short body and two short pointed uterine horns that highly attachment with uterine body and finally uterine cervix that has about (5-6) incompletely developed primitive folds that continuous directly with vagina while histological appear uterus wall composed of four undeferential the inner lining epithelium, sub- epithelium layer, primitive musculares layer and outer sub-musculares layer and lumen appear tubular irregular wide comparatively with the next stage. In the second group (140) days, appear the uterus more differentiation and development than pervious stage that appear tubular organ small in size relatively with thick wall comparatively with second trimester of gestation, composed of short uterine body and two short pointed uterine horns and finally uterine cervix that has about (5-6) rings folds connect directly with vagina, While histological, the uterine wall consists of four layer more differentiation: the inner lining epithelium, pluripotential blastemic tissue layer, muscular layer and outer serosa layer, the lumen less wide and deepest portion of the internodular areas expanded parallel with the nodular surface. Keywords: uterus, development. Prenatal, fetuses.

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INTRODUCTION

Sheep (Ovis arise) are the most important and earliest domesticated species of livestock, currently the global sheep population stands at more than one billion head with 19 per cent found in Asia and Africa (Rege and Gibson, 2003) while in Iraq there are three native sheep breeds and awassi sheep is a dominant type of sheep in Iraq and the most famous species; it is mainly raised for the production of meat, milk and wool, which are sheep wool carpet (Al-Dabbagh, S.F. 2009). The female reproductive tract organs of mammals, including the ovary, oviducts, uterus, and vagina, the uterus is bicornuate and body of the uterus is short where the two uterine horns have an inverted comma shape suite in abdomen by the broad ligament (Shehan, N. A., Kareem. *et al.*, 2019). The body of the uterus is relatively short and poorly developed, while the uterine horns are longer and well developed. Uterine cervix of uterus, cylindrical in shape which have thick wall located between uterine body and vagina containing cavity called cervical canal which consist of two opening internal uterine opening connected caudally with uterine body and external uterine opening connected cranially with vaginal (Shehan et al., 2019). Histologically, the uterine wall can divide into: the endometrium and myometrium and outer layer is perimetrium (serosa). The endometrium of domestic animals may be stratified or pseudostratified consists of two epithelial cell types, luminal epithelium, glandular epithelium, stratified stromal compartments that include a densely organized ad luminal zone of fibroblasts extending into a more loosely organized zone in the deeper or basal endometrium blood vessels with

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simple, branched uterine (endometrial) glands extend into the lamina propria. the Myometrium Consist of compact bundles of smooth muscle in different directions separated by thin strands of interstitial connective tissue with numerous blood vessels. The Perimetrium consider the outer serous coat of uterus composed of simple squamous epithelium supported by connective tissue rich in blood vessels and elastic fibers and continuous with the broad ligament (Shehan *et al.*, 2019). The female reproductive system derives from four origins: mesoderm, primordial germ cells, coelomic epithelium, and mesenchyme. Uterine development is initiated in the fetus; however, it is not completed until after birth (Bartoletal.1999, Spencer *et al.*, 2005a).

MATERIAL AND METHODS

This study was designed to described the Histomorphological and development study of uterus in local awassi sheep fetuses (Ovis Aries) at prenatal collected from Najaf abattoir soon after slaughter of the animals. the samples are collection was doing throughout one months extended from September 2024 up to November 2024. The study is performed on (16) sheep fetuses, that are collected from pregnant ewes slaughtered in the abattoirs of Najaf provinces for prenatal study. Which is determined depending on the crown rump length (CRL) using of following formula (Y=2.74X+30.15) where 'Y' is developmental age of fetus in days and 'X' is the crown-rump length in cm (Niyf and Al-Jebori, 2024; Veeresh *et al.*, 2022)

The uterus of sheep's fetuses was fixed in (10%) buffered formalin, dehydrated in a gradual series of alcohol, cleared in xylene the embedded in paraffin wax. The blocks were sectioned at (5-6) um thickness of slice using a rotary microtome. Histological sections were stained with Hematoxylin and Eosin (H&E), periodic acid Schiff, Van Gieson's, allusion blue and trichrome masons stain (Suvarna *et al.*, 2018).

RESULT AND DISCUSSION

1. Second trimester of gestation (90) days.

The body weight of sheep embryo at (90) days of gestation is about (560 ± 30.99) gram and crown rump length are about (250±3.53) mm (figure, 1). Morphologically, the morphometric measurement of uterus appears in group 1 as following, the total length of the uterus in sheep fetus (12.48±0.462) mm. The length of horn (5.54±0.248) and dimeter (0.900±0.070). The length of body (3.80±0.094), width of the body (4.40±0.221) and thickness (2.30±0.114). The length of cervix (3.14±0.128) and width (4.28±0.159) and thickness (1.11±0.098) while in camel at same trimester as following, at (4-7) months the left and right horn measured (4.44 \pm 0.17) cm and (4.15 \pm 0.15) cm, respectively, the uterine body, cervix measured (2.85 \pm 0.11) cm, (3.15 ± 0.10) cm, respectively (Jaji, et al., 2022). The reason for this difference may be the breed, size of the animal, gestation period and type of animal. At this age of embryo, the uterus of sheep fetuses, muscular tubular organ and small in size with thin translucent wall composed of a short body and two short pointed uterine horns that highly attachment with uterine body and finally uterine cervix that has about (5-6) incompletely developed primitive folds that continuous directly with vagina, the uterus located in the abdominal cavity completely, small in size with narrow tubular lumen in gross sections in different regions and become more narrowing and twisted toward uterine horns and complete closing toward cervix (figure, 2). The bicornuate uterus of the African buffalo consisted of parallel and curved uterine horns connected via the uterine body. The cranial portion of the uterine body was divided by a distinct septum according to (Schmidt, S., 2006). While internal lining of uterus horn and body contain on primitive or anlage elevation areas called caruncles. In bovine, fourth month of gestation from (CRL of 31.0 cm) onwards, prominent caruncle's can be observed in bovine uterus (Kenngott, R. A. M., et al., 2016).



Fig. 1: Photograph showing the method of measurement of crown rump length by measure tape



The histological observations in current appear approved that uterine horns was tubular irregular wide lumen comparatively with the next stage and itis wall composed of four undeferential layer from inner to outer configuration as following: the inner lining epithelium, sub- epithelium layer, primitive musculares layer and outer sub-musculares layer (figure,3) and this agreement with In goats, (Cavalcanto, 2007) who mntioned that uterine mucosa was more folded and serosa well restricted, the uterus composed of three differentiation layers, perimetrium, myometrium endometrium and were clearly distinguishable.

Uterine body wall composed of undifferentiation four layer; the inner lining epithelium,

sub-epithelium layer, primitive musculares layer and outer sub-musculares layer (figure,3). The lumen of uterine body appears irregular wide, almost star in shape with caruncles that appear as nodular or flattened elevation areas and internodular or inter-elevation areas more prominent making clearly distinguishable that in uterine horn (figure, 4). In camel, a occur increase in the number and size of the endometrial undulations of the uterine body to a result of progressive invagination of the epithelial lining into the superficial layer of the underlying propria-submucosa. The uterine body showed moderate alkaline phosphatase and moderate ATPase reaction in its epithelial lining, gradual increase in the thickness of the uterine wall in both uterine body at 465 - 630 mm CVRL stage (Farouk *et al.*, 2012).



(E) the inner lining epithelium, (P) sub-epithelium layer, (M) musculares layer, (F)outer sub-musculares layer (H&E. 4X)

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uterine cervix, musculomembrane tubular opening organ more development comparatively with uterine body and horn composed of four undifferentiation layers; inter inner epithelium layer, sub-epithelium layer, musculares layer and outer submusculares layer. The luminal surface of the inner epithelium layer has numerous primordial folds with irregular length and borders partially close of uterine cervix lumen(figure,5,6). in goats, the cervix, muscle layer was evident at 90 days which increased in thickness and number with advancement of age. Serosa also developed simultaneously without true glands were not found in uterine wall (Abiaezute & Nwaogu, 2021).



Fig (5) Histological cross section at 90 days of gestation explain uterine cervix layer; (D) folds, (E) inner epithelium layer, (P)sub-epithelium layer, (M) muscular layer (F) mesothelium (H&E. 4X).



Fig (6) Histological cross section at 90 days of gestation represent uterine cervix layer; (M) circular packages of smooth muscle fiber contain blood vessel in middle muscular layer (F) collagen in submuscular (B) blood vessel, (N) nerve (allusion blue. 10X).

2. Third trimester of gestation (130-140) days

The body weight of sheep embryo in the current study at (130-140) days of gestation about (4360 ± 136.3) gram and crown rump length about (404±5.09) cm. Morphologically, the morphometric measurement of uterus appears in group 2 as following, the total length of the uterus in sheep fetus (32.2±0.481) mm. The length of horn (15.1±0.331) and dimeter (2.960±0.243), The length of body (7.56±0.150), width of the body (7.52±0.177) and thickness (3.34±0.143). The length of cervix (9.54±0.273) and width (6.68±0.188) and thickness (3.30±0.164) while In camel at (10-13) months the left and right horn measured (7.50 ± 1.86) cm and (5.80 ± 0.79) cm, respectively, the uterine body, cervix measured (4.28 ± 0.17) cm, (4.69 ± 0.09 cm), respectively (Jaji *et al.*, 2022).

At this age of embryo, the uterus of sheep fetuses more differentiation and development than pervious stage that appear tubular organ small in size relatively with thick wall comparatively with second trimester of gestation, composed of short uterine body and two short pointed uterine horns that highly attachment with uterine body and finally uterine cervix that has about (5-6) rings folds connect directly with vagina, and these folds facilitate the function of the cervix and prevent access of foreign bodies such as bacteria and viruses into the uterus, suspended in the abdominal cavity by broad ligament (figure,7). At four to seven months of gestation conform that the uterine horn is the longest of all the parts of the genital tract, and the left uterine horn is longer than the right, the cervix had definite transverse folds (muscular annular rings) (Jaji, *et al.*, 2022).

Internal lining of horn and body appear numerous prominent rounded or oval small in size called caruncles. These caruncles were distributed in more than one rows in the middle and caudal thirds of the uterine horns and less in number and disappear toward tip or end of uterine horn. In bovine Large and well-differentiated caruncles are seen in the fetal corpora uteri as well as in the caruncles at eight of gestation (CRL of 90.0 and 94.0) cm, in uteri of fetuses with CRLs of 57.6 (58.7, 66.0, and 72.5 cm, the caruncles increase in number and size according to age. (Kenngott & Sinowatz, 2016).





The histological observations appear that uterine horn, more growth and development with advancement of gestation, the lumen less wide and deepest portion of the internodular areas expanded parallel with the nodular surface. The wall consists of four layer more differentiation from inner to outer configuration as following: the inner lining epithelium, pluripotential blastemic tissue layer, muscular layer and outer serosa layer (figure,8). (Wiley et al., 2011) show that in sheep at 140 day of gestation the deepest portion of the internodular areas expanded parallel with the nodular surface. Slight epithelial undulations, confined to the deep internodular mucosal surface, were seen by d 135. These undulations were never more than one or two cells deep, nor did they involve more than two or three mucosa epithelial cells cellular relationships within the mesenchymal, myometrial and outer cell layers remained unchanged from those seen at (100-115) days fetal uteri while;Uterine body, appear the histological results of uterine body composed of four layers; the inner lining epithelium, pluripotential blastemic tissue layer (submucosa), the middle myometrium and outer perimetrium. The lumen of uterine body irregular wide, almost star in shape contain the nodular and internodular areas more differentiation and clearly distinguishable

than the previous stag (figure,9). In buffalo, at (240) day of gestation, the uterine lumen approximal irregular, the endometrium of development to superficial and deep zone, lined by simple tall columnar to pseudostratified. The myometrium be thickness and perimetrium became fibrous with appear few smooth muscle fibers, endometrium, myometrium and perimetrium resembled to that of adult uterus at (274 -300) day and the uterine glands show at (7) months of gestation (Gbaishyn *et al.*, 1997).

The uterine cervix, the wall of uterine cervix consists of four layer more differentiation than the second trimester from inner to outer configuration as following: the inner lining epithelium, sub epithelium layer, muscular layer and outer layer (serosa). the uterine cervix has narrow lumen with numerous polymorphic folds with different sizes and length that closed partially uterine cervix lumen (figure,10,11). in camel the uterine cervix composed of the lining epithelial (mucosa), submucosa, muscular and sub-serosal layer, contain numerous of primary, secondary and tertiary fold that gradually increased in size and close partially (Sameh M *et al.*, 2013)



Fig (8) Histological cross section at 140 days of gestation explain uterine horn layer; (E) lining epithelium, (P) subepithelium layer, (M) muscular layer (F) sub musculares layer, (L) lumen, (Q) caruncle (H&E. 4X).



Fig (9) Histological cross section at 140 days of gestation explain uterine body layer; (E) lining epithelium, (P) subepithelium layer, (M) muscular layer (F) submusculares layer, (L) lumen, (B) blood vessel (H&E. 4X).





CONCLUSION

In conclusion, the study unveils key aspects of uterus development in sheep embryos across the second and third trimesters of gestation. It delineates morphological changes, including the emergence of distinct folds and caruncles in uterus part. Histological insights underscore the differentiation of mesenchymal tissue into muscle fibers and the simple columnar epithelial tissue lining role. These findings contribute to our understanding of uterus development in sheep, offering parallels and distinctions with related species. Such comprehension is pivotal for veterinary and medical research, shaping insights into uterus anatomy and paving the way for further investigations into embryonic development.

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