SOME HISTOLOGICAL, HISTOCHEMICAL, AND ULTRA STRUCTURE STUDIES ON THE VAGINA OF THE EGYPTIAN GEESE

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A B S T R A C T

Ten healthy female geese, aged 10-18 months, were used in this study. The sources of the geese were the local market, and a breeding house in Benha. Immediately after the geese were humanely sacrificed and eviscerated, fresh samples were taken. The samples were obtained from the last segment of the vagina of the left oviduct. Samples were randomly divided into parts: samples from seven geese were fixed in 10% neutral buffered formalin for the light microscope study; and the other three samples were fixed in 2.5 % gluteraldehyde for transmission electron microscopy(TEM) examination. The light microscopic studies revealed that the wall of the vagina in geese is formed of tunica mucosa, tunica sub mucosa, tunica muscularis and tunica serosa. In addition, it was observed that the luminal mucosa of the vagina is folded, and is lined by pseudo stratified columnar ciliated cells. The lamina propria-mucosae is made of loose fibrous connective tissue which is devoid of secretory glands, but contains sperm storage glands. The TEM showed that the lining epithelium is formed of ciliated cells, that exhibit long cylindrical cilia, and the non-ciliated secretory cells. The cells of the lining epithelial possess rounded and oval mitochondria, RER ribosome particles, and small size electron lucent secretory granules in their secretory cells. The nuclei of the epithelial cells are euchromatic, each with a prominent nucleolus.

Key words: Geese, Vagina, L.M, transmission electron microscopy.

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1. INTODUCTION

Compared to other farm animals, poultry population, including geese, can be increased rapidly by economical breeding of poultry on large scale to supply humans with high quality animal proteins (poultry meat and eggs). For this reason, poultry breeding was introduced in Egypt for production of table eggs and meat (El-Habbak, 1990) and it has been expanding since its introduction. It is generally accepted that geese, which belongs to the family *Anatidae and* genus *Anser,* were among the first animals to be domesticated. Geese domestication probably took place in Egypt in about 3000 years ago. Domesticated geese come in a wide range of colors, sizes and shapes. In general, domestic goose breeds are much larger than their wild ancestors, although they have in many cases retained their ability to fly (Buckland, Guy, 2002). Egypt is among the three leading goose meat producers in the world (between1991-2007). However, subsistence goose production is still carried out extensively by small farmers, and is distributed in the middle of Nile delta (Kumar, 2009). The female goose lays an egg every other day. D*omestic breeds of geese in Egypt* can lay 8-10 eggs during the laying period. Eggs are formed in the left oviduct of the

female birds *(*Bradley, 1928*,* Cole, 1938*,* Hodges, 1974*,* Bakst, 1978*,* El-Bargeesy, 1990, El-Habbak, 1990*,* El-Sayed, 1994*)*. In the available literature, only few studies focused on the structure and ultra structure of the vagina (Muwazi et al. 1982; Hicks-Alldredge, 1998, Madekurozwa, 2005, Sharaf, 2005). The present study aims to investigate the histological, histochemical and ultra structure of the vagina of the Egyptian geese. This may be a step for further studies on the anatomy and histology of the oviduct of Egyptian geese.

2. MATERIALS AND METHODS

Ten visibly healthy female geese, aged 10-18 months, were used in this study. The sources of the geese were the local market, and a breeding house in Benha. Immediately after the geese were humanely sacrificed and eviscerated, fresh samples were taken. The samples were obtained from the last segment of the vagina of the left oviduct. Samples were randomly divided into two parts: samples from seven geese were fixed in 10% neutral buffered formalin for the light microscopy study; and the other three samples were fixed with 2.5 % gluteraldehyde for the transmission electron microscopy (TEM) examination.

2.1. Preparation of samples for light microscopy

1 cm² samples of the vagina that were used for the light microscope part of the study, were initially washed in water for removal of blood clots and other debris. Samples were then fixed in 10% neutral buffered formalin, dehydrated, cleaned and embedded in paraffin wax. Serial sections, 5µm thick each, were cut and stained with Harris hematoxylin and eosin, Alcian blue (pH, 2.5) and Periodic acid Schiff technique (PAS), (Bancroft, Cook, 1994).

2.2. Preparation of samples for electron transmission microscopy (TEM)

1 mm³ samples were used for transmission electron microscope study. Vagina samples were fixed with 2.5% glutaraldehyde in 0.1 M sodium cacodylate buffer, and post-fixated for 2 h in 1% osmium tetroxide solution buffered to pH 7.4 at 4°C. The samples were then dehydrated in a graded series of ethyl alcohol and embedded in epoxy resin embedding medium to produce medium hardness blocks prior to sectioning. Semi thin sections 0.5 µm in thickness were used for histological observation after toluidine blue staining. The ultrathin sections were prepared with Ultracut microtome, stained with uranyl acetate and lead citrate (Chiu et al. 1993) and examined by a Jeol JEM-100S transmission electron microscope (25KV) in Military veterinary hospital.

3. RESULTS

The vaginal wall is the thickest part of geese oviduct. The wall formed of tunica mucosa, tunica sub mucosa, and tunica muscularis. Externally, it is formed of tunica serosa in the anterior part and adventitia in the posterior part where it joined with adjacent structures. The tunica mucosa projects into the lumen giving rise to primary, secondary, and sometimes tertiary folds. The vaginal folds are long, narrow and run longitudinally. On the other hand, the tunica muscularis is composed of the thick inner circular layer, and outer longitudinal layer, which are made of smooth muscle fibers. There is a band of smooth muscle extending from the inner circular muscle layer to the center of the fold. The tunica serosa is made up of fibrous connective tissue and is covered externally by simple squamous epithelium (Fig.1). The epithelial lining of the vaginal fold is formed of ciliated pseudo stratified columnar epithelium. The lamina propria-mucosaeis made up of fibrous connective tissue with blood vessels, lymphatic

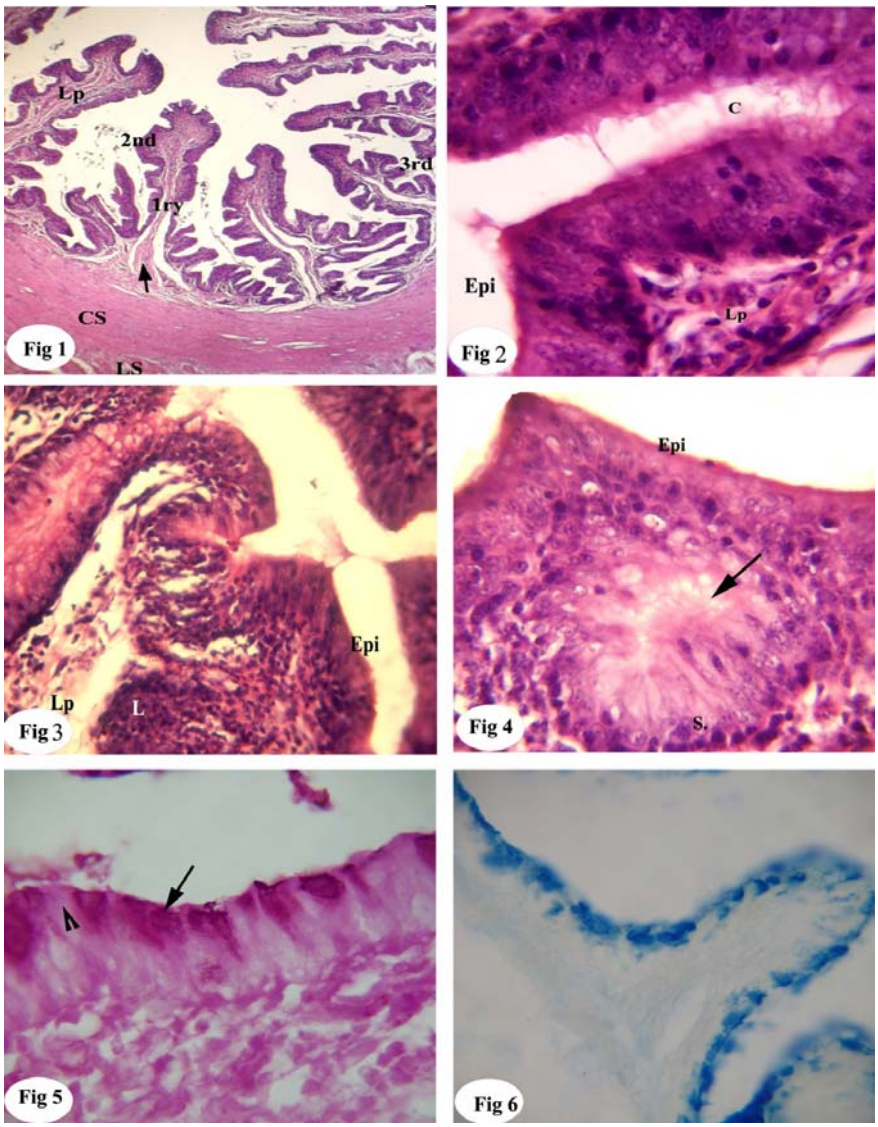

Fig. 1. Photo micrograph showed 4 layers of vaginal wall, primary (1ry), secondary (2nd) and tertiary folds (3rd), thick tunica muscularis (C.s, L.s) ,central band (arrow) which extend from the inner circular smooth muscles to the center of the fold H&E X 10. Fig. 2. Photo micrograph showed the lining epithelium is pseudo stratified columnar ciliated(C) epithelium and fibrous lamina propria (L.p) X 100. Fig. 3. Photo micrograph showed the lamina propria (L.P) is fibrous connective tissue containing lymphatic follicles (L) and devoid of secretory glands H&E X 40. Fig. 4. Photo micrograph showed the sperm storage gland in the lamina propria (arrow) which is lined by simple columnar epithelium H&E X 100. Fig. 5. Photo micrograph showed PAS positive substances in the secretory epithelial cells (arrow) and negative reaction in ciliated cells (arrow head) PAS stain X 100. Fig. 6. Photo micrograph showed alcianophilic substances in the lining epithelium of the vaginal wall Alcian blue stain X 40.

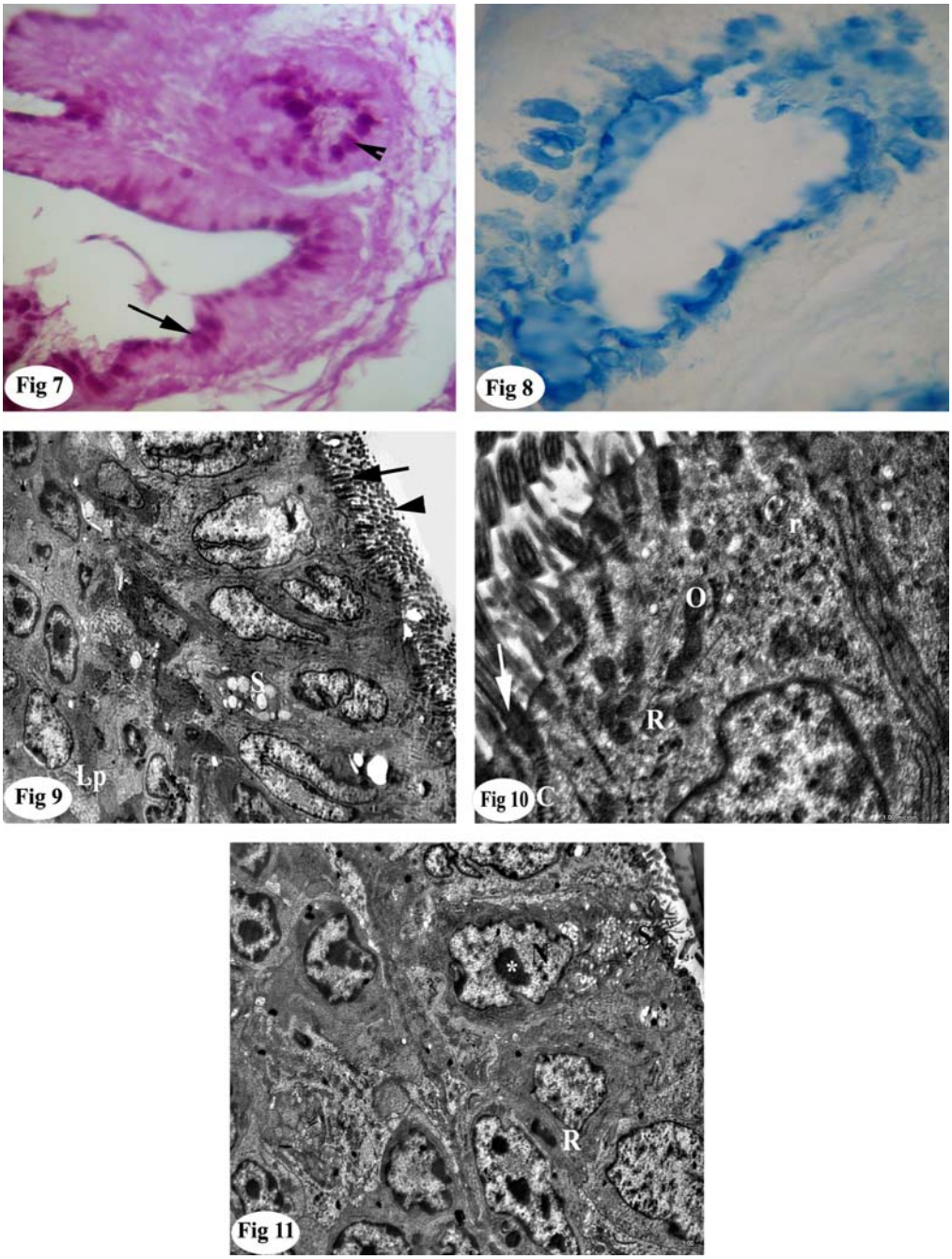

Fig. 7. Photomicrograph showed PAS positive substances in the lining epithelium of the vaginal wall (arrow) and in sperm storage gland (arrow head) PAS stain X 100. Fig. 8. Photo micrograph showed alcianophilic substances in the lining epithelium of the sperm storage gland Alcian blue stain X 100. Fig. 9. Electron micrograph revealed that the lining epithelium formed from ciliated cells which exhibit long cylindrical cilia (L.s arrow, C.s arrow head) and the non ciliated secretory cells and infra nuclear secretory granules (S) in the secretory cells x 2000. Fig. 10. Electron micrograph revealed that the ciliated cells (C) exhibit long cilia (arrow), rounded (R) and oval mitochondria (O), RER, ribosome particles and the nucleus are euchromatic with prominent nucleolus x 15000. Fig. 11. Electron micrograph showed supra nuclear small size electron lucent secretory granules (S) in the secretory cells and basal cell with rounded nucleus (R) x 3000.

vessels, nerves, and lymphatic nodules (Fig.2 and Fig.3).The lamina propria is devoid of secretory glands but contains sperm storage glands, which are lined by simple columnar epithelium (Fig.4). The lining epithelium shows strong PAS positive materials containing neutral mucopolysaccharides (Fig.5). The lining epithelium also shows alcianophilic substances containing acidic mucopolysaccharides (Fig.6). In addition, the sperm storage gland reveals the presence of both neutral and acidic mucopolysaccharides (Fig.7 and Fig.8). The transmission EM revealed that the lining epithelium is formed of ciliated cells, which exhibit long cylindrical cilia; and the non-ciliated secretory cells which contain infra-nuclear small size electron-lucent secretory granules (Fig.9). The cells exhibit rounded and oval mitochondria, RER, ribosome particles and the nucleus are euchromatic with prominent nucleolus (Fig.10). The secretory cells exhibit supra-nuclear small size electron-lucent secretory granules in secretory cells (Fig.11).

4. DISCUSSION

Vagina is the last portion of the oviduct. It is short S-shaped tube which connects the uterus to the cloacae, and it is responsible for the selection, storage, and transport of sperms. The wall of the vagina shows branched and convoluted folds which project in to the luminal surface and this in agreement with the results obtained by El-Habbak, (1990), Sharaf, (2005) and this is consistent with the findings of the present study. This study showed that the lamina epithelialis of the vagina is formed of pseudo stratified ciliated columnar cells. The propria-submucosa devoid of secretory glands, and contains many lymphocytes and lymphoid follicles. These findings are in agreement with Paria, (2011), however, the results of this investigation disagrees with Hodges, (1974) who stated that there are predominant goblet cells in the epithelial folds which were crowded at the bottom of the grooves between the mucosal folds. The lamina propria-submucosae has sperm storage glands which are lined by simple columnar epithelium, and these findings are in agreement with Fujii, (1963) who found that the mucosal folds of vagina have simple tubular glands lined with tall columnar cells, however, Verma, Cherms, (1964), Das, Biswal, (1968), Hodges, (1974), El-Habbak, (1990) stated that the vagina was devoid of any glands. The tunica muscularis in vagina is the thickest layer than any other part of the geese oviduct, it is made of thick inner circular and outer longitudinal smooth muscle fibers with stratum vascular in between the two layers. It was observed that the vagina is bounded by serosa cranially and adventitia caudally. The findings of this study agree with what was published by Paria, (2011).

It is interesting to note that, our study documented that the lining epithelium showed PAS positive and alcianophilic substances in the apical part of the non-ciliated cells as an evidence of the secretory activity of these epithelial cells, which contained both neutral and acidic mucopolysaccharides. In addition, the epithelial cells of the sperm storage glands have both neutral and acidic mucopolysaccharides . These observations disagrees with Asuman et al. (2009) who argued that there is no neutral or acidic mucopolysaccharides in the vagina, While El-Habbak, (1990) contended that the vaginal epithelial cells gave a strong staining reaction with PAS and alcian blue stains. The present study revealed that the ultra structure of the surface epithelium showed two types of cells, ciliated and non-ciliated granular cells or secretory cells. The ciliated cells are narrow with an expanded apex carrying the cilia. Similar results were recorded by Fertuck , Newstead (1970), Wyburn et al. (1970), Muwazi, (1982). It noteworthy

that, the secretory cells have supra-and infra-nuclear electron-lucent secretory granules of small size, and have rounded shape and oval mitochondria, RER, and free ribosome's.

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دراسات هستولوجية وهستوكيميائية وتركيبية دقيقة على المهبل فى الأوز المصري

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الملخص العربى

استخدمت الباحثة 10 من أنثى الأوز فى الفترة العمرية من عشرة إلى ثمانية عشر شهرا. لقد جمعت الباحثة العينات من الأسواق المحلية وقامت بتربيتها فى المنزل ومباشرة بعد الذبح وإزالة الأحشاء أخذت الباحثة أخر جزأ من قناة البيض اليسرى(المهبل) وبعد ذلك حفظت العينة فى 10% فورمالين متعادل لأجراء دراسات باستخدام المجهر الضوئى وحفظت فى 2.5% جلوترالدهيد لأجراء دراسات تركيبية باستخدام المجهر الألكترونى. باستخدام المجهر الضوئى وجدت الباحثة أن جدار المهبل يتكون من طبقة مخاطية، طبقة تحت المخاطية، طبقة عضلية وتغطى من الخارج بطبقة مصلية فى الجزأالأمامى وغلالة برانية فى الجزأ الأخير من المهبل. كما لاحظت الباحثة وجود ثنايات فى جدار المهبل من الداخل وهذه الثنايات مبطنة بنسيج طلائى مصفف كاذب لديه أهداب. تختلط الطبقة المخاطية والتحت مخاطية وتتكون من نسيج ضام ليفى خالى من الغدد الإفرازية ولكن يحتوي على غدد تخزينية للحيوانات المنوية. كما أوضحت الدراسات المجهرية الألكترونية أن النسيج الطلائى يتكون من خلايا هدبية لها أهداب طويلة أسطوانية وخلايا إفرازية غير هدبية. كما لوحظ أن هذه الخلايا تحتوي على ميتوكوندريا دائرية وبيضاوية، شبكة إندوبلازمية محببة وريبوسومات وحبيبات إفرازية فاتحة صغيرة الحجم داخل الخلايا الأفرازية كما وجد أن النواة فاتحة مع وجود نوية واضحة.

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