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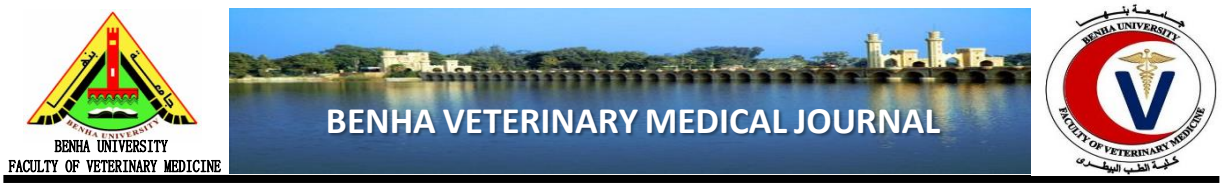

SEASONAL HISTOLOGICAL CHANGES IN THE VESTIBULE OF SHE-CAMELS

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A B S T R A C T

Twenty adult she-camels with ages ranging from 5 to 10 years were used to describe the histological changes in the vestibule during the breeding and non-breeding seasons. The vestibular epithelium and its keratin showed thicker thicknesses during the breeding season. During the breeding season the keratin layer showed strong PAS reaction and the superficial squamous cell layers showed negative PAS reaction, while during non-breeding season the keratin layer showed less PAS reaction and the superficial squamous cell layers showed more PAS reaction. In addition, the vestibular glands and their excretory ducts showed higher activity during the breeding season. These seasonal changes suggest that the vestibule of she-camel may be under the effect of hormonal changes during breeding and non-breeding seasons.

KEYWORDS: Vestibule, Histology, She-camel, Breeding season, Non-breeding season.

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1. INTRODUCTION

Camel is an important livestock species in Egypt. Humans depend on camel not just for meat, milk and hide but also as one of the most important mode of transportation especially in the desert so they are recognized as the "Ship of the desert". Most of the available literature dealt with the histology of the female genital system of any animal including camels were concentrated on the ovary, uterine tube, uterus, cervix and vagina [1, 2, 3, 12, 8, 14, 20, 15, 16 and 23]. Histological studies on the vestibule of she-camels were done in a very narrow scale [4] that motivate us to study it. To explore the effect of seasons on the structure of the she-camel's vestibule, we have preceded to study its histological structure during breeding and non-breeding season.

2. MATERIALS AND METHODS

2.1. Animals

The whole female genital tracts of 20 non pregnant, apparently normal, adult she-camels were collected from El-Warrak abattoir in Giza Governorate, Egypt. The age of the she-camels were ranging from 5 to 10 years. The age of these animals were determined according to [25]. Specimens were collected during the breeding and non-breeding seasons.

2.2.Specimens and processing

At the abattoirs, immediately after slaughter, the female genital tracts of she-camels were opened by median incision from the dorsal commissure of the vulva to the level of the vagina. Small specimens were taken from different parts of the vestibule of each animal.

All the specimens were immediately fixed in formalin, then were dehydrated in ethyl alcohol, cleared in xylene and embedded in paraffin wax.

2.3. Histological stains

Paraffin sections of 5-micrometer thickness from vestibule of the breeding and non-breeding she-camels were cut and stained with Harris's alum haematoxylin and eosin, Masson's trichrome and Periodic acid Schiff (PAS) stains. The fixative and staining methods were used as outlined by [6].

2.4. Morphometry

The thickness of the vestibular epithelium and its keratin during the breeding and non-breeding seasons were measured using Motic Image Plus 0.2 software. Thirty different microscopic fields of H&E stained slides (n=6) for each specimen were used to measure the thickness at magnification X10 and the averages were recorded in microns unit.

2.5. Statistical analysis

Student's t test was used to compare the thickness of the vestibular epithelium and keratin during both the breeding and non-breeding season. $P < 0.05$ was considered statistically significant.

3. RESULTS

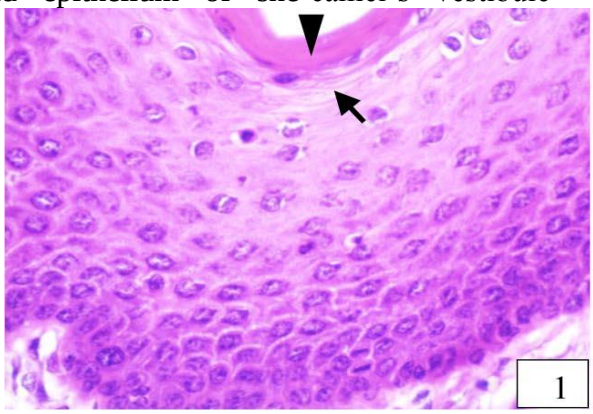
The vestibule of adult she-camels consists of tunica mucosa, muscularis and adventitia. The vestibule is lined with keratinized stratified squamous epithelium (Figs.1,2). During breeding season, the superficial squamous cell layers show pale acidophilic cytoplasm and the keratin layer appears dark acidophilic however, cytoplasm of the superficial squamous cell layers appears dark acidophilic and the keratin layer appears less acidophilic during non-breeding season (Figs.1,2). During breeding season, the keratin layer shows strong PAS reaction and the superficial squamous cell layers show no PAS reaction while during non-breeding season, the keratin layer show less reaction and the superficial squamous cell layers show strong PAS reaction. The basement membrane shows positive PAS reaction during both seasons (Figs.3,4). The average thickness of the vestibular epithelium and keratin show a significant seasonal difference where it is thicker during breeding season than those of non-breeding season (Table 1). Propria-submucosa consists of highly vascularized dense collagenous connective tissues (Fig.5). Lymphocytes are common throughout the propria-submucosa. They are infiltrated superficially just beneath the basement membrane of the vestibular epithelium during both seasons (Fig.5), while lymphocytic nodules are seen deep to propria-submucosa during breeding season (Fig.6). The vestibular glands are present in the deeper part of the propria-submucosa close to muscularis. During breeding season, the vestibular glands are arranged in the form of small isolated groups of few acini and more tubular secretory units which their secretory cells are single layer of columnar cells to large pyramidal cells with foamy cytoplasm, spherical basally located nuclei, and narrow lumen (Fig.7). While during non-breeding season, the vestibular glands are fewer in numbers and appear as isolated acini. They are lined with cuboidal to low columnar cells with acidophilic cytoplasm, oval nuclei and wider lumen than during breeding season (Fig.8) that indicates higher activity of the glands during the breeding season. The excretory ducts of the vestibular glands have variable structures in relation to seasons where they are relatively larger in size, lined with stratified squamous epithelium that rested on PAS positive basement membrane during breeding season, and their lumens are containing secretions of vestibular glands which show strong PAS reaction (Fig.9). During non-breeding season, the excretory ducts of the vestibular glands are relatively

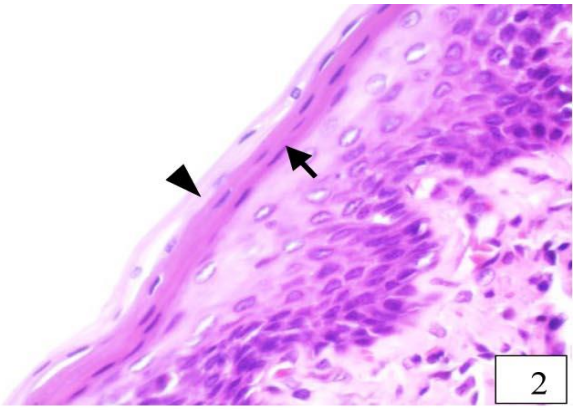
smaller in size and they are lined with stratified cuboidal epithelium with empty lumens (Fig.10). Tunica muscularis is formed of circular and longitudinal smooth muscle bundles embedded in dense collagenous connective tissue (Fig.11). Tunica adventitia is formed of loose connective tissue containing blood vessels and nerve fibers (Fig.12).

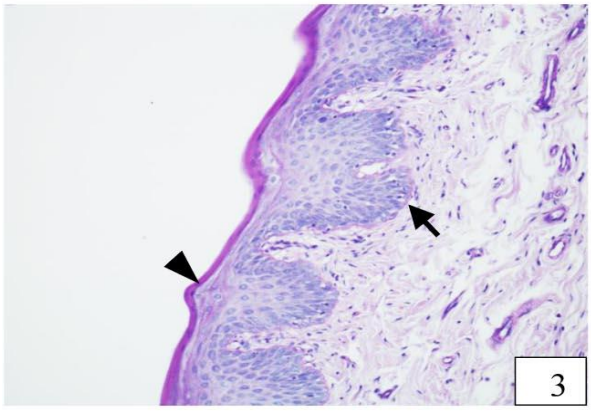
Table (1): Average ± SD thickness of keratin and epithelium of she-camel's vestibule during the breeding and non-breeding season

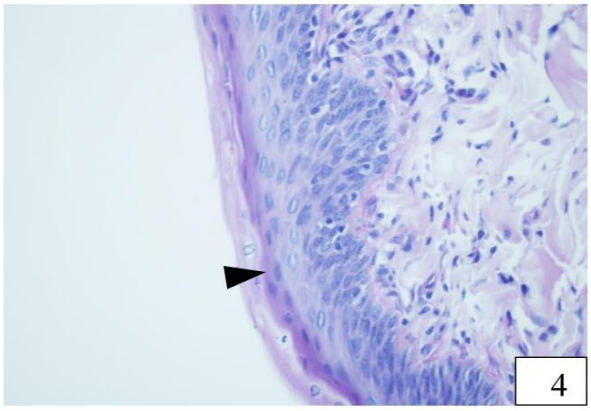
	Epithelium	Keratin
Breeding	705.4±72*	16.6±10*
Non-breeding	325.3±65*	7.7±8*

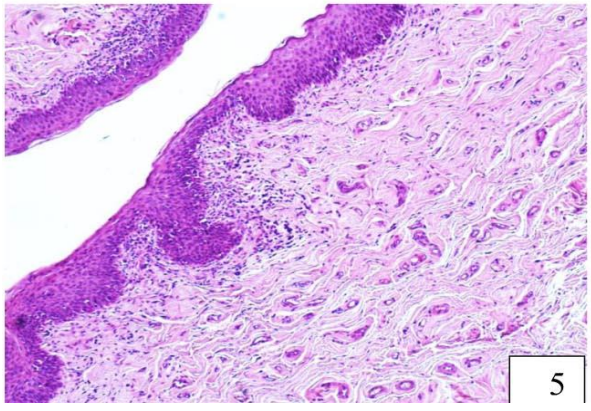
Within a column, means a significant difference ($P < 0.05$).

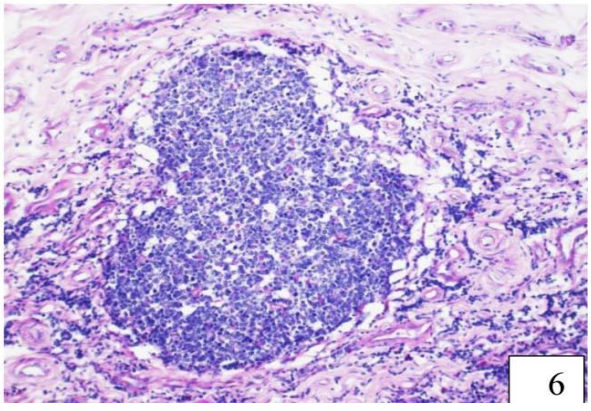












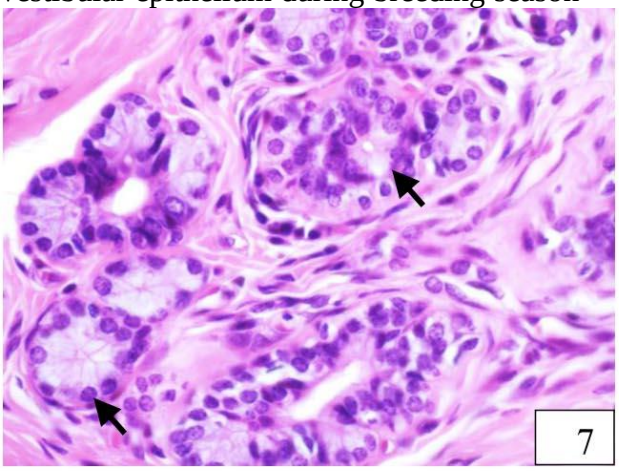
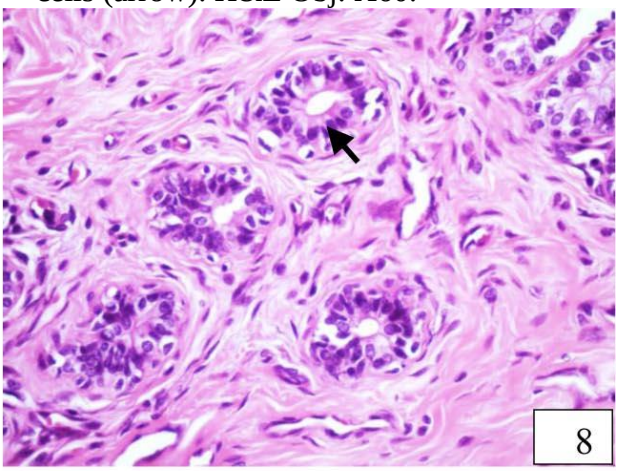
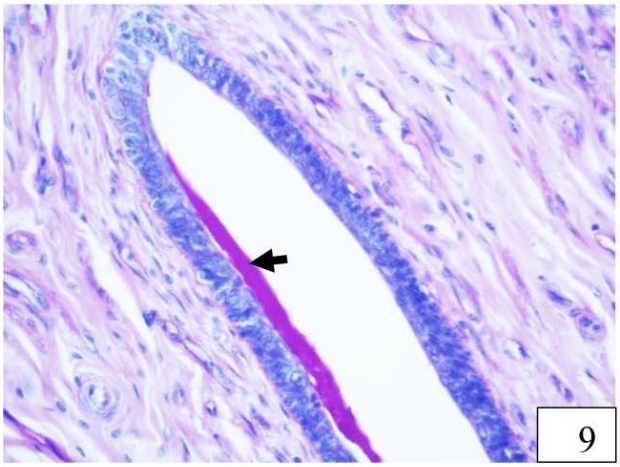
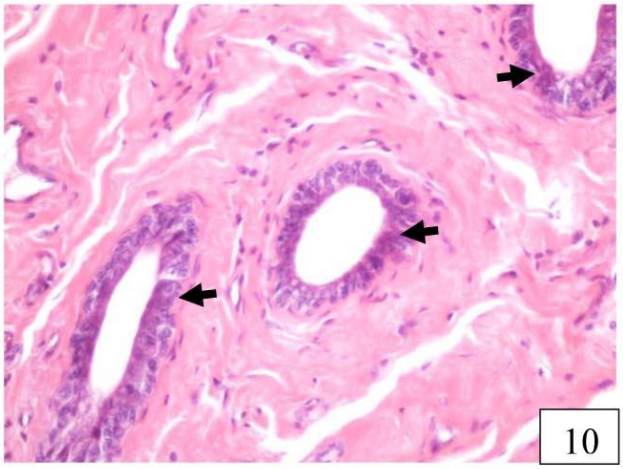
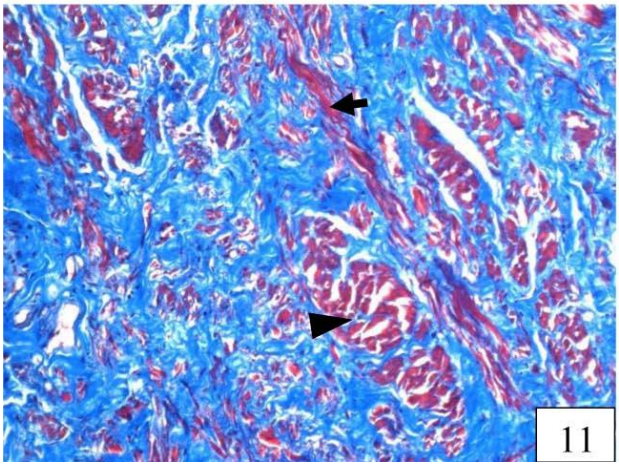


Fig.1: Photomicrograph of she-camel's vestibular epithelium during breeding season showing dark acidophilic keratin (arrowhead) and pale acidophilic superficial squamous cells (arrow). H&E Obj. X60.









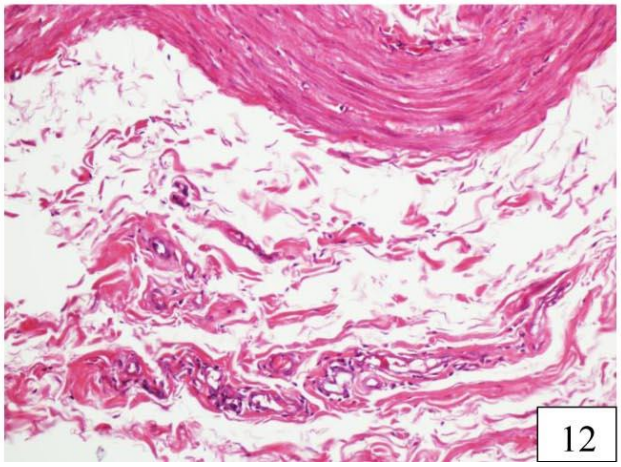


Fig.2: Photomicrograph of she-camel's vestibular epithelium during non-breeding season showing thinner faint acidophilic keratin (arrowhead) and dark acidophilic superficial squamous cells (arrow). H&E Obj. X60.

Fig.3: Photomicrograph of she-camel's vestibular epithelium during breeding season showing PAS positive basement membrane (arrow) and keratin (arrowhead). PAS Obj. X40.

Fig.4: Photomicrograph of she-camel's vestibular epithelium during non-breeding season showing strong PAS reaction in the superficial squamous cells (arrowhead). PAS Obj. X40.

Fig.5: Photomicrograph of she-camel's vestibule showing typical feature of lymphocytic infiltrations under the epithelium. H&E Obj. X10.

Fig.6: Photomicrograph of she-camel's vestibule during breeding season showing nodule of lymphocytic aggregations in deep propria-submucosa. H&E Obj. X20.

Fig.7: Photomicrograph of she-camel's vestibule during breeding season showing many vestibular glands lined with columnar to pyramidal cells with narrow lumen (arrows). H&E Obj. X40.

Fig.8: Photomicrograph of she-camel's vestibule during non-breeding season showing fewer vestibular glands lined with cuboidal to low columnar cells with wider lumen (arrow) compared with breeding season. H&E Obj. X40.

Fig.9: Photomicrograph of she-camel's vestibule during breeding season showing large excretory duct of vestibular gland that contain PAS positive secretion (arrow). PAS Obj. X40.

Fig.10: Photomicrograph of she-camel's vestibule during non-breeding season showing empty, smaller lumen of excretory ducts (arrows) of vestibular gland compared with breeding season. H&E Obj. X40.

Fig.11: Photomicrograph of she-camel's vestibule showing typical feature of circular (arrowhead) and longitudinal (arrow) smooth muscles embedded in dense collagenous tissues. Masson's trichrome Obj. X10.

Fig.12: Photomicrograph of she-camel's vestibule showing typical feature of tunica adventitia of loose connective tissue with blood vessels. H&E Obj. X20.

4. DISCUSSION

Histological sections of the vestibule of adult she-camels revealed that its wall consists of tunica mucosa, muscularis and adventitia that agreed with [4] in she-camel, [5] in buffalo and [9] in cow. Vestibule of adult she-camels was lined mainly with keratinized stratified squamous epithelium that agreed with [4] in she-camel however, [7, 5] in buffalo and [13, 9] in cow found that lining epithelium of the vestibule consists of stratified squamous with intraepithelial mucus secreting cells. This work revealed a significant seasonal change ($P<0.05$) in the thickness of the she-camel's vestibular epithelium where both vestibular epithelium. Keratin were thicker during the breeding season (high estrogen concentrations) than those of the non-breeding season (low estrogen concentration) that supports the findings of [4] who identified thicker epithelium and keratin layer of the she-camel's vestibule in the follicular phase (estrogen dominant phase) than in the luteal phase (progesterone dominant season). In addition, our results revealed pale acidophilic cytoplasm of the superficial squamous cell layers and dark acidophilic keratin layer during the breeding season, while during the non-breeding season our results showed dark acidophilic cytoplasm of the superficial squamous cell layers and less acidophilic keratin layer. This work showed seasonal variations in the reaction of the vestibular epithelium for PAS where during the breeding season, the keratin layer showed strong PAS reaction, but the superficial squamous cells layers shows no PAS reaction. However, during the non-breeding season, the keratin layer showed less reaction and the superficial squamous cell layers showed strong PAS reaction. Propria-submucosa of she-camel's vestibule was consisted of vascularized collagenous

connective tissues that was similar to [4] in she-camel, [7, 5] in buffalo, [21, 13] and [9] in cow, [26] in women. Lymphocytes were common and present as lymphocytic infiltration just beneath the basement membrane of vestibular epithelium during both seasons that agreed with [10] in cow. Also, nodules of lymphocytic aggregations were seen deep to propria-submucosa during the breeding season that was similar to previous observation [13 and 10] in cow vestibule. The seasonal change in the lymphocytic amount and distribution indicated increased immune defense in the vestibule during the breeding season where the mating and pregnancy occurred. Our results demonstrated the vestibular glands mainly within the lateral wall of middle part of the vestibule, however [7] identified the vestibular glands in both dorsal and ventral walls of the vestibule of buffalo. The vestibular glands of she-camels were present in the deeper part of the propria-submucosa close to muscularis, arranged in form of small isolated groups of acini that agreed with [4]. However, in buffalo [7, 5] detected two types of vestibular glands; large subepithelial glandular masses and small glandular masses. The activity of she-camel's vestibular glands appeared to be higher during the breeding season (high estrogen concentration) than the non-breeding season (low estrogen concentration) that was supported by findings of [5] where they observed higher gland activity in the follicular phase (estrogen dominant phase) than in the luteal phase (progesterone dominant phase). During the breeding season, the secretory cells of vestibular glands were single layer of columnar cells to large pyramidal cells with foamy cytoplasm, spherical basally located nuclei, and narrow lumen, while during the non-breeding season the vestibular glands were fewer in numbers and lined with cuboidal to low columnar cells with acidophilic cytoplasm, oval nuclei and wider lumen. The muco-serous nature of the glandular masses simulated that of the glandular structures described by [22] in cat and [7] and [5] and buffalo. Our results revealed no minor vestibular glands in the she-camel that was similar to [4], but differed from [24, 11, 17 and 9] who identified minor in addition to major vestibular glands in cow, however [18, 11 and 17] in bitch identified only minor vestibular glands. The cells of excretory duct of the she-camel's vestibule were not secretory, while [19] reported that tubular cells of female calves produced neutral mucins. The excretory ducts of the vestibular glands showed seasonal variation in their structures where they were relatively larger in size, they were lined with stratified squamous epithelium that rested on PAS positive basement membrane during the breeding season and, their lumens were containing secretions of vestibular gland which showed strong PAS and alcian blue reactions. During the non-breeding season, the excretory ducts of the vestibular glands were relatively smaller and they were lined with stratified cuboidal epithelium with empty and narrow lumens. Tunica muscularis of she-camel's vestibule was formed of longitudinal and circular smooth muscle bundles embedded in dense collagenous connective tissue that simulated to findings of [4] in she-camel, [7, 5] in buffalo. Sometimes, nerve bundles were sandwiched between the muscular layers of she-camel's vestibule. Tunica adventitia covered the muscularis of the vestibule externally and was formed of loose connective tissue containing blood vessels and nerve fibers that agreed with [4] in she-camel and [5] in buffalo.

5. REFERENCES

1. Abdalla, O. 1965. Anatomical study of the female genital system of the one-humped camel (Camelus dromedarius). I. The ovaries. Sud. J. Vet. Sci. Anim. Husb. **6**: 41-52.

2. Abdalla, O. 1967. Anatomical study of the female crenital system of the one-humped camel (Camelus dromedaries). II. The oviducts. Sud. J. Vet. Sci. Anim. Husb. **8**: 67-77.
3. Abdalla, O. 1968. Anatomical study of the female genital system of the one-humped camel (Camelus dromedaries). III. The uterus and placenta. Sud. J. Vet. Sci. Anim. Husb. **9:** 47-57.
4. Awad H. H., El-Hariri M. N. and Bareedy M. H. 1982. Macro-and Micromorphological study of the female copulatory organs of the one-humped camel (camelus dromedarius). Egyptian J. Histo. **5**(1): 85-94.
5. Badawy, Y. H., Ewais, M. S., Mahmoud, S. A. and Bareedy M. H. 1978. A Micromorphological study of the female copulatory organs of buffaloes in Egypt (Bos bubalus L.). Gegenbaurs Morph. Jahrb., Leipzig **124**(5): 663-679.
6. Bancroft, J. D., Cook, H. C., Striling, R. W. and Turner, D. R. 1994. Manual of hidtologivcal techniques and their diagnostic application. Second Ed., Churchill Livingston, Edinburgh, London, Madrid, Melbourne, New York and Tokyo.
7. Bareedy, M. H. 1977. Some micro-morphological studies of the female copulatory organs of buffaloes in Egypt (Bos bubalis L.) with reference to the vestibular glands. M.V.SC. Fac. Vet. Med. Zagazig Univ.
8. Bezrukov, N. I. and Smidt, G. A. 1970. Some functional changes in the ovaries and uterus of Bactrian camels. Arkh. Anat. Histol. Embryol. **58**(l): 64-73.
9. Blazquez, N. B., Batten, E. H., Long, S. E. and Perry, G. C. 1987a. Histology and histochemistry of the bovine reproductive tract caudal to the cervix. Part I. The vestibule and associated glands. Br. Vet. J. **143**:328-337.
10. Blazquez, N. B., Batten, E. H., Long, S. E. and Perry, G. C. 1987b. Amount and distribution of vestibular associated lymphoid tissue in calves and adult cows. Res. Vet. Sci. **43**(2): 239-242.
11. Dellman, H. D. 1971. Veterinary histology, an outline text and atlas, Lea and Febiger, Philadelphia, USA.
12. El-Hag Mousa, B. (1969): A study of some aspects of the reproduction in the female camel (Camelus dromedarius). M. V. SC. Fac. Vet. Med. Khartum, Sudan.
13. El-Hagri, M. A. A. 1967. Splanchnology of domestic animals, 1st Ed., The public organization for book and scientific applicances, University Press, Cairo.
14. El-Wishy, A. B. and El-Sawaf, S. A. 1971. Functional activity of the ovaries and uterine horns in fat-tailed sheep and camels (Camelus dromedarius). Fortpflanzung. Besamung und Aufzucht der Haustiere **7**(3): 181-187.
15. Ewais, M. S. S., El-Hariri, M. N., Yousef, H. I., Affiefy, M. M. and Omar, M. A. 1979. Cyclic variation in the structure of the cervix of the she-camel (Camelus dromedarius). Egypt. Vet. Med. J. Vet. Sci. **27**:183-201.
16. Ewais, M. S. S., Yousef, H. I., Affiefy, M. M. and Abu-El-Fadle, W. S. 1979. Some macro and micromorphlogical studies on the uterine tube of the camel (Camelus dromedarius) in different phases of the estrous cycle and pregnancy. Egypt. Vet. Med. J. Vet. Sci. **27**:202-223.
17. Getty, R. 1975). The anatomy of the domestic animals, volume I and II ,5th Ed. W. B. Saunders Comp. Philadelphia, London, Toronto, Mexico city, Rio de Janeiro, Sydney and Tokyo.
18. Miller, M. E., Christensen, G. and Evan, H. 1964. Anatomy of the dog, W. B. Saunders Comp. Philadelphia and London.
19. More, J., Van Haverbeke, G., Cabanie, P., Benazet, F. and Sautet J. 1986. Secretory glycoproteins of major vestibular glands (Bartholin's glands) in female calves. Studies with usual histochemical methods and with lectin-horseradish peroxidase

conjugates in normal animals and in animals implanted with anabolic drugs. Acta Anat, (Basel) **126**(3): 199-204.
20. Nayak, R. K. 1977. Scanning electron microscopy of the camel uterine tube (oviduct). Am. J. Vet. Res. **38**(7): 1049-1054.
21. Raghavan, D. and Kachroo, P. 1964. Anatomy of the ox. First Ed., Indian council of agricultural research, New Delhi.
22. Shehata, E. (1973): Glandular acini in the wall of the lower part of the domestic cat vagina. Acta Anat. **84**: 52-76.
23. Skidomore, J. A., Billah, M. and Allen, W. R. 1995. The ovarian follicular wave pattern in the mated and non-mated dromedary camel (Camelus dromedarius). J. Reprod. Fertil. Suppl. **49**:454-458.
24. Trautmann, A. and Fiebiger, J. 1957. Fundamentals of the Histology of domestic animals, 4th Ed., Ithaca, New York, Comstock.
25. Williamson, G. and Payne, W. J. A., 1978. An Introduction to Animal Husbandry in the Tropics. Third Ed. England and New York.
26. Yang, C. C., Cold, C. J., Yilmaz, U. and Maravilla, K. R. 2005. Sexually responsive vascular tissue of the vulva. B.J.U. International **97**: 766-772.

مجلة بنها للعلوم الطبية البيطرية عدد 24 (1)، يونيو 2013: 150-158

BENHA UNIVERSITY
FACULTY OF VETERINARY MEDICINE

مجلة بنها للعلوم الطبية البيطرية

تغيرات هستولوجية موسمية فى دهليز النوق

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الملخص العربي

تم استخدام عشرين ناقة بالغة، تراوحت أعمارها بين 5 إلى 10 سنوات لوصف التغيرات النسيجية في الدهليز خلال مواسم التوليد وعدم التوليد. ولقد كانت ظهارة وكيراتين الدهليز أكثر سمكا خلال موسم التوليد. وكان خلال موسم التوليد تفاعل قوى لطبقة الكيراتين مع صبغة شيف وحامض البيرأيوديك بينما طبقات الخلايا الحرشفية السطحية أظهرت تفاعل سلبي مع الصبغة، أما في غير موسم التوليد فقد أظهرت طبقة الكيراتين تفاعل أقل مع صبغة شيف وحامض البيرأيوديك بينما زاد تفاعل الصبغة مع طبقات الخلايا الحرشفية السطحية. بالإضافة إلى ذلك، فإن غدد الدهليزي وقنواتها الإخراجية أظهرت نشاطا عاليا خلال موسم التوليد. ومن المقترح أن هذه التغيرات الموسمية تشير إلى أن دهليز النوق قد يكون تحت تأثير التغيرات الهرمونية أثناء مواسم التوليد وعدم التوليد.

(مجلة بنها للعلوم الطبية البيطرية: عدد 24 (1)، يونيو 2013: 152-160)