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M.A.M. Alsafy et al., Accessory sex glands of the Barki bucks

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Abstract

The present investigation was prepared to describe the accessory sex glands of the Barki bucks grossly and by light microscopy. There are four sex glands: ampullary, vesicular, prostate, and bulbourethral. The ampullary gland is an enlargement of the terminal part of the ductus deferens, its glandular part has branched tubuloalveolar glands, and its secretory alveoli lined with a pseudo-stratified epithelium composed of cuboidal to columnar cells. The vesicular gland takes the appearance of a cluster of grapes and the left vesicular gland is enlarged and higher than the right one. The vesicular gland is a lobulated tubuloalveolar gland with wide intralobular space and the gland contain a secretory unit which lined by pseudo-stratified columnar epithelium, and the interlobular ductules lined by the stratified epithelium, while the interlobular duct lined by simple cuboidal epithelium moreover, the
lining epithelium of secretory part consists of tall columnar cells. The prostate gland consists only of the disseminated part and is enclosed by a connective tissue capsule that was thin dorsally, thick laterally, and reduced in thickness ventrally. The prostatic acini are lined by simple cuboidal epithelium. The bulbourethral gland was similar in size to the walnut and surrounded by a capsule and there are interlobular connective tissue septa that divided the gland into lobes and lobules of different sizes. The bulbourethral gland contains secretory units lined by the tall simple columnar epithelium of mucous type with basely located nuclei and eosinophilic cytoplasm contain granular secretion. The gross and microscopic examination of the four accessory sex glands gave valuable information in the future pathology diagnosis of the accessory sex glands of the Barki bucks.

Key words: bucks, sex glands, gross morphology, light microscopy, CT scan

INTRODUCTION

The ruminant anatomy was generally concentrated on the bovine especially ox with limited comparative points to the small ruminants, especially goat. Goat is belonging to the Caprini of the Bovidae family in the sub-order Ruminantia of the order Artiodactyla. The goat is considered an important source of meat, milk, and hair in Egypt. The male goat is a very familiar animal in Egyptian farms. There are about seven to ten goat breeds in Egypt; Zaraibi, Barki (Sahrawi), Wahati, Sharkawi, and Black Sinai (1). The three major breeds in Egypt are; Ossimi, Rahmani, and Barki, which are characterized by fat-tailed, their fleece is coarse wool and they are small to medium in size (1).

The male genital systems have to be sound to ensure successful breeding. Ram and buck breeding soundness examination is important to assess the ability to impregnate females during the breeding season (2).

The purpose of the present work was to describe the anatomical position and appearance of the accessory sex glands. In addition to, the microscopic structure of these glands as there is little available published information about the accessory sex gland of the
Barki (Sahrawi) bucks, the study of the accessory sex gland in bucks is very important due to its big role in the fertility rate consideration. Then, comparative the obtained data with the previous published anatomical data about the accessory sex glands.

MATERIALS AND METHODS

Animals and ethics

The present work was carried out on eight normal healthy Barki bucks and weighted about 25–30 kg and the age of these animals between 1-2 years. The Barki bucks were collected from the goat farms in the Matrouh government and transported to the anatomical laboratory in the anatomy and embryology department, Faculty of Veterinary Medicine, Alexandria University.

This study followed the rules for the care and use of animals and intrinsically approved by the Animal Welfare and Ethics Committees, Faculty of Veterinary Medicine, Alexandria University.

Gross morphology

Four Barki bucks were used in the gross morphological descriptions after their sedation with intramuscular injection of the 2% xylazine HCl (0.2 mg/Kg of body weight) and atropine (0.04 mg/kg). Then, these bucks were anesthetized by intravenous injection of Ketamine (5 mg/kg). Then, these bucks were injected with heparin (1.000 IU) to obtain well bleeding and prevent the coagulation. Then, the bucks were sacrificed by the well bleeding through the common carotid artery for injection of 10% formalin, then, the specimens stored for two weeks in formalin 10%. The cadavers injected via a needle in several sites of the body and on the pelvic cavity using the ordinary preservation solution (10% formalin, 2% phenol, and 1% glycerin).

Computed tomography scans
Two bucks were used for carrying the computed tomography. After physical examination of animals, they were anesthetized by using the combination of Ketamine hydrate 0.5-2 mg/Kg of body weight and 2% xylazine HCl 1-3 mg/Kg of body weight. The bucks cadavers were taken freshly to the CT center immediately and fixed in the sternal recumbence position and then, serially sectioned using the Hitachi CT scanner (CT-W450-10A, Hitachi, Japan) (scanning conditions: 120 KV and 200 MA, the width and level of the window (W/L): 2000/250) to examine the accessory male sex reproductive glands. The section thickness was 0.5 cm part interval in the cross-section from the level of the last lumbar to the fourth caudal vertebrae. The CT images photographed helping in the identification of the structures situated from the level of the fifth lumbar and the fourth caudal vertebra. To obtain the good resolute images in the bone windows, the CT machine was adapted with 200 Hounsfield units in the windows width and 1600 Hounsfield units in the level of the windows. While in the soft tissue, we adapted the apparatus with 30 Hounsfield units in the width of the window and 290 Hounsfield in the level of the windows.

**Light microscopy**

Two bucks were used for light microscopic examination. The accessory sex glands were removed from the freshly slaughtered male bucks then, were put on 10% normal buffer formalin solution and transported to the histological lab for the histological preparation of the slides (cutting and staining) to allow the examination of the slides under the light microscope to know the characteristic points of each gland. Then, the samples were put in 70% alcohol solution. Then they were rapidly dehydrated through ascending grades of ethyl alcohol series (30, 50, 70, 90, and 100% for 2 changes) for a half-hour in each. Then cleared by putting in xylene and embedded in paraffin wax. Samples sections of 5 μm were cut by Leica rotatory microtome and mounted on the glass slides. Finally, the paraffin sections were used for ordinary staining by Harris hematoxylin and eosin stain (H&E). The histological techniques are carried according to (3).

The nomenclature used in this study is adapted to (4).
RESULTS

In all examined bucks, there are four accessory sex glands (*Glandulae genitales accessoriae*); the paired ampulla of the ductus deferens (ampullary gland), the paired vesicular gland (*Glandula vesicularis*), the prostate (*glandula prostatica*), and the paired bulbourethral gland (*Glandula bulbourethralis*). All accessory sex glands were located along the pelvic urethra and their ducts opened and empty their secretion into the urethra.

**Ampullary gland (*Glandula ampulla ductus deferentis*)**

The ampullary gland (Fig. 1A\LA and RA; 1B/ R.dd; 6A/4) is a glandular enlargement of the terminal part of the ductus deferens. The length of the ampullary gland reaches 3.5 ± 0.4 cm and 0.5 ± 0.12 cm in width. The ampullary glands were directed ventrocaudally on the dorsal surface of the urinary bladder and attached by the genital fold (Fig. 1A\GF).

The light microscopic examination of the gland (Fig. 2) appeared to consist of the tunica mucosa, tunica muscularis, lamina propria and tunica adventitia, and some blood vessels. The glandular part contains branches of tubuloalveolar glands. The secretory alveoli are lined with a pseudo-stratified epithelium composed of cuboidal to columnar cells with some basal cells.

**Vesicular gland (*Glandula vesicularis*)**

In all investigated bucks, the paired vesicular gland (Fig.1A\LV-RV; 1B/V; 6A/3) is located on the craniodorsal aspect of the neck of the urinary bladder (*Vesica urinaria*) and laterally to the ampullary glands. It can be identified by its appearance that looks like a cluster of grapes and it is important to note that the left vesicular gland is enlarged and higher than the right one in the same buck.

The light microscopic examination clarifies that the vesicular gland (Fig. 3) is a lobulated tubulo-alveolar gland with wide intralobular space or sinuses for storage of a large number of secretions. The gland contains a secretory unit are lined by pseudo-stratified columnar epithelium, and the interlobular ductules are lined by the stratified epithelium while the interlobular duct is lined by simple cuboidal epithelium with intra-
luminal eosinophilic secretion. The lining epithelium of the secretory part consists of tall columnar cells, apical blebs of granular end pieces, and detached blebs in lamina, small and spherical basal cells.

**Prostate gland (glandula prostatica)**

In all examined bucks, the single prostate gland (Figs.1/P; 6B/16) is located close to the junction of the vesicular gland at the pelvic region. It is constituted only from the disseminated part (*pars disseminate*). The pars disseminate surrounds the pelvic urethra which is covered by the urethra masculina.

The light microscopic examination showed that the prostate gland is enclosed by a connective tissue capsule (Fig.4) that is thin dorsally, thick laterally, and reduced in thickness ventrally. The gland capsule is enveloped by a layer of skeletal muscle (muscle urethralis) that surrounded by a layer of loose connective tissue trabeculae extending from the capsule and descending into the parenchyma of the gland forming interlobular connective tissue and divided the gland into lobules. The glandular substance consists of numerous follicles, which open into the elongated canal. The prostatic acini are lined by simple cuboidal epithelium.

**Bulbourethral gland (Glandula bulbourethralis)**

In all investigated bucks, the paired bulbourethral gland is similar in size to the walnut and located dorsal to the urethra in both sides of it, cranial to the ischial arch (Fig. 1A/LB-RB; 1B/ BL; 6C/23). It is closely related to the bulb of the penis and in general, it appears to be covered mostly by the bulbospongiosus muscle. The right and left bulbourethral glands appear to be nearly equal in size and their shape similar to each other.

The light microscopic examination showed that the bulbourethral gland was surrounded by a white fibrous capsule and there are interlobular connective tissue septa that divided the gland into lobes and lobules of different sizes (Fig. 5). The gland contains secretory units lined by the tall simple columnar epithelium of mucous type with basely located nuclei and eosinophilic cytoplasm contain granular secretion.
DISCUSSION

There are four sex accessory glands in most mammalian species; ampullary, seminal vesicle, prostate, and the bulbourethral glands in the Barki bucks as reported in horse (5, 6), bull (6, 7), spotted paca (8), Gracilinanus microtarsus (9), Arabian oryx (10), Elk (Cervus canadensis) (11), red deer (Cervus elaphus) (12). While, the presence of three accessory sex glands was reported by; (6) in; canine (prostate, bulbourethral glands and ampullary gland), canine (prostate, seminal vesicle and ampullary gland), (13) in the pampas deer Ozotoceros bezoarticus (prostate, vesicular and the ampullary glands), (14) in the lesser anteater (prostate, seminal vesicle and bulbourethral glands). However, there are two accessory sex glands only; the prostate and vesicular gland in the capybara (15). Furthermore, (6, 7) reported that the accessory sex glands were fully developed only in the bull.

The present investigation reported that the ampullary gland is a glandular enlargement of the terminal part of the ductus deferens, similar result mentioned by (6, 16-18). Furthermore, (16) added that its size has a species variation among animal species; well-developed in stallion, bull and ram but, it is absent in the boar, however (6, 19) mentioned that it is ill well-developed in the boar but absent in the tomcat and also (9) reported the absence of this gland in Gracilinanus microtarsus. The present histological observation of the ampullary was similar with (20, 21) that, the gland consists of three layers; the tunica mucosa, tunica muscularis and tunica adventitia.

In all investigated bucks, the paired vesicular gland located on the craniodorsal aspect of the urinary bladder and it can easily identify by its appearance which looks like a cluster of grapes and also it's important to note that the left vesicular gland is enlarged and higher than the right one, similar finding obtained by (6, 22, 23). The vesicular gland is the largest accessory sex gland in; the bull (6, 7) and the spotted paca (8). However, the vesicular gland was absent in; the canines (6), Gracilinanus microtarsus (9), some rodents (24, 25). In the current histological study, the vesicular gland is a lobulated alveolar gland containing a secretory unit that lined by pseudo-stratified columnar epithelium, and the interlobular ductules lined by the stratified epithelium while, the interlobular duct lined by simple
cuboidal epithelium, similar finding obtained by (22, 23, 26). While it was lined by simple cuboidal epithelium as reported in the spotted paca was (8) but, it was lined by a simple columnar epithelium as observed in; G. pig (27), and rat (28) and giant rat (29, 30).

Morphologically, the prostate gland may be single as present in our study and by (6, 13, 23, 31-33) or paired as noted by (34). In our work, we found that the single prostate gland has only the pars disseminate that is found in the pelvic urethra and surrounded by the urethral masculina, similar findings obtained by (6, 13, 31-33). While (35) reported that there is an additional part named the corpus prostate present in some gaddi goats, in contrast, this corpus prostate is not reported in small ruminants in all published anatomical textbooks (6, 31, 36).

There is some variation in the portions of the prostate gland among animals species; the present study with (6, 13, 31-33) noted the presence of only one portion called disseminate part while (23) in the Black Bengal buck mentioned that the single prostate gland has two portions; compact (external) and disseminate (internal) portion. But (34) in rat noted that the paired prostate gland has a dorsal and ventral part, also (37) in the G. pig noted that the prostate gland has two portions; the large cranial lobe and the smaller caudal lobe. Moreover (38) in the capybara noted that the prostate gland consists of three parts; intermediate, dorsal and lateral lobes.

According to our histological studies, the prostate gland was enclosed by a connective tissue capsule, which is thin dorsally, thick laterally and reduced in thickness ventrally. The capsule was enveloped by a layer of skeletal muscle surrounded by a layer of skeletal muscle, similar to that reported by (23, 35). In G. pig, the prostate is enveloped by a fibrous muscular layer (39) while in the spotted paca (8) this layer consists of smooth muscle fiber, similar to that observed in the prostate gland of the mouse (40, 41). The present study noted that the prostatic acini lined by simple cuboidal epithelium while in the spotted paca (8) noted that it is simple columnar epithelium but pseudo-stratified in some regions.

Our study revealed that the paired bulbourethral gland is small in size similar to hazelnut, and lies dorsal to the pelvic urethra and more obvious as the gland surrounded by a white fibrous capsule, similar observations noted by (7, 16, 22, 23). In the spotted paca,
the bulbourethral gland presented at the terminal part of the rectum and dorsal to the urethra and caudal to the prostate gland (8). The bulbourethral gland is absent in the canine and present in the feline as noted by (6, 19). The bulbourethral gland was surrounded by striated skeletal muscle as noted by (8, 29, 30). Our work mentioned that the bulbourethral gland contains secretory units lined by the tall simple columnar epithelium of mucous type. The lining simple columnar epithelium was also noted by (30). The bulbourethral gland appears to be covered mostly by the bulbospongious muscle and the gland itself is covered by a white fibrous capsule, similar to that noted by (7).

Availability of data and materials
The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request. Research data are not shared.

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**Figure 1.** Morphological appearance of the position of the accessory sex reproductive glands of the buck:

**View (A)** of the isolated genital organs with the accessory sex reproductive glands of the buck: U.B- Urinary bladder, LD-RD- Left and Right ductus deference, GF- Genital fold, LU-RU- Left and Right ureter, LA-RA- Ampulla of ductus deference, LV- RV- Left and Right vesicular gland, P- Prostate gland, U- Urethra, LB-RB- Left and Right bulbo urethral gland. **View (B):** represented the left side of the pelvic cavity showing the topography of the accessory sex reproductive glands of the buck: 1- Pararectal fossa, 2- Vesicogenital pouch, 3- Rectogenital pouch, 4- Pubovesical pouch, 5- Ampulla recti, 6- Muscle sphincter ani internus, 7- Muscle sphincter ani externus, R- Rectum, R.dd- Right Ampulla of ductus deference, R.U- Right ureter, U.B-Urinary bladder, U- Urethra, IM- Mesorectum, V- Vesicular gland, P- Prostate gland, BL- Bulbourethral gland, L - Lumbar vertebrae, S- Sacral vertebrae, C- Caudal vertebrae.

**Figure 2.** Micrograph of the ampullary gland of the adult buck showing:
Alveoli (As); Tunica muscularis (Ms); Tunica adventitia (Ad); Blood vessels (Bv); Pseudo-stratified epithelium (St); Basal cell (Bc); Lamina propria (L). H&E, X 160.

**Figure 3.** Cross-section micrograph of the vesicular gland of the mature buck showing:
**View (A) clarify;** SU- Secretory units lined by pseudo stratified columnar epithelium, intralobular ductules lined by stratified epithelium (**black arrow**) and intralobular duct lined by simple cuboidal epithelium (**red arrow**) with intraluminal eosinophilic secretion (As). H&E. X160. **View (B) clarify;** The lining epithelium of secretory part consists of tall
columnar cells (arrow), Apical blebs of granular end pieces (red headarrows) and detached blebs in lamina (blue headarrows), Small and spherical basal cells (black headarrows). H&E. X400

Figure 4. Histological micrograph of the prostate gland (pars dessiminata) from the mature buck:
View (A): Lamina muscularis (LM), Prostatic acini (PA) Surrounded by fibrous capsule (C) and propria submucosa (PS) with smooth muscle bundles (black headarrows), Excretory ducts of the prostate gland (Ed). H&E, X160. View (B): Prostatic acini lined by simple cuboidal epithelium with glandular epithelium (Ge). H&E, X400.

Figure 5. Histological cross section micrograph of the bulbourethral gland of the mature buck:
View (A): Adventitia (Ar), Capsule (C) and Secretory unit (SU). H&E, X160. View (B): The lining epithelium of secretory unit is a tall simple columnar epithelium of mucous

Figure 6. Computed tomographical images of the male buck's pelvis;
View (A) represent the Transversal CT scan at the level of the 3rd sacral vertebra.
View (B) represents the Transversal CT scan at the level of the 5th sacral vertebra.
View (C) represents the Transversal CT scan at the level of the 3rd caudal vertebra.
1- 3rd sacral vertebra; 2- Rectum; 3-Vesicular gland; 4-Ampulla of ductus deference; 5- Neck of urinary bladder; 6- Pubic symphysis; 7- Ilio psoas muscle; 8- Right ilium shaft; 9- Superficial gluteal muscle; 10- Testis; 11- Right adductor muscle; 12- External obturator muscle; 13- 3rd caudal vertebra; 14- Muscle sphincter ani internus and externus; 15- Pelvic urethra; 16- Prostata; 17-Penis; 18- Right sacrocaudalis dorsalis lateralis muscle; 19- Right sacrocaudalis ventralis medialis and lateralis muscle; 20- Anal canal; 21- Right ischium; 22- Ischiatic arch; 23- Bulbo urethral gland; 24- 5th sacral vertebra; 25- Head of femur; 26- Greater trochanter of femur.
