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Morphological and Histological Studies of Anogenital Scent Glands of Egyptian Jerboa *Jaculus Orientalis*



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ARTICLE HISTORY	Abstract: Rodents depend on the scent glands and their chemical secretions that stim- ulate the behavior of the same species and other predators in natural environments. The great Egyptian jerboa <i>laculus orientalis</i> were obtained between Aidabiya and Bengha-
Received:	zi, eastern Libva to reveal their morphological and histological constituents. Because
6 October 2022	there are few studies on this subject, the morphological and histological details of the anogenital region glands of adult male and female <i>Jaculus orientalis</i> will be described
Accepted:	in this study. The anesthetized animal with diethyl ether was dissected then the skin
1 June 2023	around the anogenital region and one-third of the anal canals were excised and fixed in
Keywords: Rodents, Scent Glands, Preputial Gland, Perianal Gland, Anal Gland.	10% aldehyde formalin, dehydration, and embedded, then serially sectioned 5µm was cut, stained with eosin, hematoxylin, and trichrome for microscopic examination. A huge circumferential aggregation of modified compound branched sebaceous glands that permeate the submucous and are infused into the internal sphincter muscle layer of the anal canal, with secretory canals that drain to the anal canal orifice's most outer. The preputial glands of male and clitoris glands of female were anatomically appeared in the form of small, separate, scattered, and different sizes of follicular glands and histologically formed compound branched acini sebaceous glands surrounded by connective tissue. However, these results may drop some light on the role of these glands in the scent-marking behaviors of males and females of <i>Jaculus orientalis</i> .

دراسات ظاهرية ونسيجية لغدد الرائحة الشرجية التناسلية للجربوع المصرى Jaculus orientalis

الكلمات المفتاحية : القــوارض، غــدد الرائحة ، غدد القلفة ، الغدد حول الشرج ، الغدد الشرجية

المستخلص : تعتمد القوارض على غدد الرائحة، وإفرازاتها الكيميائية التي تحفز سلوك النوع نفسه، والحيوانات المفترسة الأخرى في البيئات الطبيعية. تم الحصول الجربوع المصري الكبير Jaculus orientalis من المناطق التي بين إجدابيا وبنغازي شرق ليبيا، للكشف عن التراكيب المورفولوجية، والنسيجية لغدد الرائحة، نظرًا لندرة الدراسات حول هذا الموضوع. تم وصف التفاصيل المورفولوجية لغدد المنطقة الشرجية الندد الرائحة، نظرًا لندرة الدراسات حول هذا الموضوع. تم وصف التفاصيل المورفولوجية، والنسيجية لغدد الرائحة، نظرًا لندرة الدراسات حول من الموضوع. تم وصف التفاصيل المورفولوجية لغدد المنطقة الشرجية النداسلية للذكور، والإناث البالغين لله هذا الموضوع. تم وصف التفاصيل المورفولوجية لغدد المنطقة الشرجية التناسلية للذكور، والإناث البالغين للمحول المنطقة الشرجية المندر بايثر ثنائي إيثيل ثم نزع الجلد ول المنطقة الشرجية الترجية التناسلية في ألدهيد فورمالين 10٪، جففت العينة في الكحول الإنتيلي التصاعدي، الترويق بالزايلين، وتم التضمين في البارافين، ثم قطعت بسمك πه معاني اليوسين، والهيماتوكسيلين، والتريكروم الفحص المجهري. يحتوي جلد العانة على تجمع ضخم من الغدد الدهنية الحوسين، والهيماتوكسيلين، والتريكروم الفحص المجهري. يحتوي جلد العانة على تجمع ضخم من الغدد الدهنية الحوسين، والهيماتوكسيلين، والتريكروم الفحص المجهري. يحتوي جلد العانة على تجمع ضخم من الغدد الدهنية الحويصلية والهيماتوكسيلين، والتريكروم الفحص المجهري. يحتوي جلد العانة على تجمع ضخم من الغدد الدهنية الحويصين، والهيماتوكسيلين، والتريكروم الفحص المجهري. يحتوي جلد العانة على تجمع ضخم من الغدد الدهنية الحويصين والهريمة المريكي والمريكري الفعائية، والمغروسة في معارموسة في طبقات العصلة العاصرة الداخلية القناة الشرجية، تصريقان المتفرية في الثرية. أيضا ظهرت غدى المائية العارة من الخور، وغدد البطر في وغدد البغري على المتورعة المريم ألينيكروم الفحص المجهري. وحتوي جلد الوفرانية. أيضا ظهرت غدد القلقة في الذكور، وغدد البظر المتغرعة المتورية المروية في الإفرازية. أيضا ظهرت غد القلقة في الذكور، وغدد البظر في الإرداث تشريحيًا على شكل غدد جرابية صغيرة، منفصلة، متتارة، مختلفة الأحجام، ونسيجيا عبارة عن غدد في الإداث تشريحية متفرة، مخلولة من النسيج ضام، وتصب إفرازاتيا مع الشعرة في منطقة العان عن مرويق قنوات مرو

INTRODUCTION

Scent glands are found in almost all mammals, they secrete chemicals that enable animals communicate with one another. The morphological and histological differences between scent glands vary depending on the animal, gland location, size, and secretion

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technique. Some scent glands are found within the skin's tissue structure, whereas others are found as specialized organs beneath the skin (Vaughan, 1986; Arakawa et. al., 2008; Ralls, 1971; Harriman & Thiessen., 1985; Ferkin & Leonard, 2005; Shimozuru et. al., 2006; Janda et.al., 2019; Yousef et. al., 2022).

The chemical compositions secreted by the scent glands play an important role in the different behaviors of male and female rodents and Ferrets, Rabbits, Muskrat; these secretions may cause different effects in the attraction of females to males and vice versa, but the response varies depending on the quantity and quality of glandular secretions issued by males and females, as well as other sources of smell (Zhang et.al., 2017; Shimozuru et. al., 2006; Quesenberry et. al., 2020; Xie et.al., 2022).

According to previous studies many mammals have scent glands surrounding the anogenital region of rodents in general, rats and mice in particular, which are distinct types of modified sebaceous holocrine glands that create sebum materials such as the preputial gland, which is located in the prepuce of males, the clitoral gland, which is located in the clitoris area of females, the perianal or circumanal glands, and anal glands, that release secretions with urine in addition to the odors of the urine compounds themselves, can all be used as scent marking glands. (Ferkin and Johnston, 1995; Cloe, 2004; Arakawa et. al., 2007; Arakawa, 2008; Mshiriet.al., 2009; Yoshizawa, 2018; Ferkin, 2018; Sialiti, 2021). While both the anal, and the perianal glands in Guinea Pig, and Caviaaperea are situated in the anal sinus area and the two glands are differ in histological structure and size in males and females based on sexual activity and dominance (Arakawa et. al., 2007; Herrera, 1992; Zechman et.al., 1984).

Trevor et. al., (1981) and Budsberg, (1983) also revealed that the perianal gland in some

carnivores consists of one or more lobes, each histologically composed of modified sebaceous glands, and each lobe drains through small openings on both sides of the anus. Also, the anal scent gland in Crocutacrocuta is a swollen anal sac located above the anus and its secretions are used to mark the territory, (Woodmansee et al., 1991), while Mephitidae contains two or three lobes of anal scent glands located on both sides of the anus, secreting an unpleasant odor when attacked by a predator (Heidt and Morgans 1982). As well as (Helder and Fremuller, 1995; Helder Jose et al., 2016) describe the anatomical and histological structure of the paracloacal scent gland in the Brown four-eved opossum / Metachirusnudi caudatus as consisting of two lobes of the gland, one located to the right of the anal canal and the other to the left of it, which affects the mating sexual behavior of the animal, as well as cloacal of Brand's hedgehog consists of the sweat and populated sebaceous glands (Akbariet.al., 2020).

However, the resent studies about the anatomical and histological structure of the scent glands, which are related to the samegenus rodents in Libva, Jaculus jaculus, described scent glands anatomically and histologically, the perianal glands are composed of several sebaceous glands of different sizes, modified, located in the around the anal region, which are compound acini glands surrounded by a capsule of areolar connective tissue that drains outward on the skin of the anogenital area (Mshiri et. al., 2009). Also, in another study (Mshiri et. al., 2013) on the skin glands of Ctenodactylus gundi, it contains two types of scent glands; The first is large modified sebaceous glands connected to the coarse hair wall around the pubic area (prepuce) and (clitoris) in males and females, respectively, and the second glands are consists of three lobes located around the anus (perianal), both of which are structurally compound branched acini with holocrine secretion, surrounded by compound tubular glands of apocrine that

drain their secretion to the exit through a special duct, generally the skin glands and especially sebaceous glands providing about 90% of its surface lipids (Hoover et.al., 2022).

However, based on what was before mentioned in the previous studies and continuation of the scientific journey, this study aimed to further explore the anatomical and histological structure of the scent glands in the Great Egyptian Jerboa Jaculusorientis.

MATERIALS AND METHODS

For this investigation, twenty male and female Jaculus orientis were captured in the fields between Ajdabiya and Benghazi in eastern Libya Figure 1. The adult male and female animals were anesthetized and slaughtered then placed in a container containing water and ice at a temperature of about five degrees centigrade for 30 min for animal desensitization. The macroscopic aspects of the anogenital region of adult male and female glands were observed fresh under a stereomicroscope (LEICA ES2) and dissected the anogenital area; perianal with one third of anal canal and pubic skin patch, then the removed pieces of skin were positioned in cassettes and then fixed by immersion in a %10 formaldehyde solution and preservation for at least 24 hours. This study was authorized by the System of Authorization and Information on Biodiversity and was certified by Tripoli University/ Zoology department.

longitudinal and transverse cuts across the samples to analyze their morphology under light microscopy. The samples were rinsed in running water for 30 min before being dehydrated in a series of increasing ethanol concentrations (from 70% to 100%), clearing in xylene, embedded in paraffin. Sections were made with thickness of 5 - 6µm using microtome (LAB-MR500). Following that sections were stained with hematoxylin and eosin (H&E) to examine general features and Masson's trichrome stain to examine connective

tissue collagen fibers, according to the standard method in Culling, C.F.A. (1963). The slides were observed using light microscope and photographed using a digital camera. The capture images were being used to examine.



Figure. (1) the geographical distribution of the Egyptian jerboa *Jaculus orientis* in North Africa. http://www. Jaculus orientalis - (7ywn.com).

RESULTS

Gross Anatomy: Regardless of gender, all of the Egyptian great jerboa Jaculus orientis specimens examined have scent glands in the anogenital area Fig. 2 (A); perianal, anal and prepuce glands; which are clitoral glands in females and preputial glands in males. The skin around the female and male genital orifice appears structurally slight grainy in the form of small irregularly sized protrusions of various shapes in prepuce region. They were oval and circular, with dense pale coloration and powerful scents when handled. The perianal glans were tiny protrusions that surrounded the anal orifice and contained lowdensity greasy hair, as shown in Fig. 2 (B and C), but anatomically anal glands are invisible



Figure. (2). A, Great Egyptian jerboa *Jaculus orientis*, B and C the different masse, shape and size of prepuce glandular, B, clitoris glands in female and C, preputial glands in male arrows.

The histological ultrastructure of the scent glands :First: the anal and perianal are group of separated glands located in the mucosa and submucosal layer of the anal canal and anus (which are compound branched acini)sebaceous glands) with holocrine secretion Fig.3 (A, B and C), each gland is consisting of mature acini with pale and shattered cells (immature acini which consisting of dark stained cells. Also, the acini are surrounded by a capsule of connective tissue that rich in

'secretions were drained through a central canal lined with stratified nonkeratinized epithelial cells that opened within the anal canal to expel their secretions with excreta. The glandular acini cells' secretions are drained by displacement with the aid of myoepithelium cells surrounding the acini, as shown in Fig.3 (D), as well as the skeletal muscles of the anal canal as shown in Fig.3 (B and C).

collagen fibers Fig.3 (C and D).



Figure. (3). A, B and C, shows the various structures of large modefied sebaceous glands in the mucosal and submucosal layer of the anal canal (H, E and Trichrome, 10X, 20X and 40X respectively). D, shows mature glandular acini (M ac) consisting of pale and shattered cells, immature glandular acini (IM ac) consisting of dark stained cells. C, capsule consisting of areolar connective tissue with myoepithelelial cells (arrows, H & E 40X).

It also contains a group of modified sebaceous glands of the holocrine type that are distributed between the longitudinal and circular skeletal muscle layers of the anal sphincter structure. These glands are encapsulated in a thick outer capsule of skeletal muscles that is considered part of the anus Fig.4 (A and B). In addition, each of these glands is encased by an inner capsule composed of areolar connective tissue, from which the trabeculae subdivide the glands lobes into lobules as in Fig.4 (B and C). Each gland is made up of a cluster of glandular lobes; each glandular lobe is made up of a cluster of glandular lobules, which are includes a glandular acinus that made up of two types of cells: basal flattened cells with clear basophilic stain central nuclei, mature cells with unclear nucleus and rich in fatty substances and other ruptured cells Fig.4 (C). The secretions of the acini drain into the lateral secretory ducts, which are lined with unkeratinized stratified squamous epithelium and then into the central canal, which is lined with unkeratinized stratified squamous epithelium. The central canal ends

at the anus orifice, which is lined with a keratinized stratified squamous epit helium Fig.4 (D).



Figure. (4). Perianal glands large modified sebaceous glands (MSG), **A** and **B**, circumferential arranged around the anus (AN), between the anal orifices sphincter muscles (SM) and in hypodermis (H). **C**, the gland is surrounded by an external capsule (Ec) and the internal capsule (Ic). the mature acinus (M ac) is consisted of pale cells and shattered cells, and immature acini (IM ac) consisting of dark stained cells. **D**, central canal is lined by the keratinized stratified squamous epithelial tissue arrow. **A**, (H&E and Masson's Trichrome) and B, C and D (H&E) (20X, 20X, 20X and 40X).

Second: Prepuce glands (preputial and clitoral glands: Prepuce glands are a protrusion consisting of various shapes lobes of a large separate modified sebaceous gland Fig.5 (A, B and C) which are surrounded by areolar connective tissue, forms the outer capsule of each glands Fig.5 (A and C). Each glandular lobe is made up of glandular lobules consisted of acini that are made up of immature, mature, and exploding cells Fig.5 (E and F). All glandular acini are linked to the lateral secretory ducts 'these ducts are lined by unkeratinized stratified squamous epithelium. Their secretions drain into the main ducts, which is lined by unkeratinized stratified squamous epithelial cells Fig.5 (C, E and F) and the keratinized stratified squamous epithelium towards the peripheral orifices of the main ducts Fig.5 (D), the secret ions of these glands are drained through the main ducts of each gland on the surface of the skin of the prepuce. However, each gland connected with hair which

secretions flow to the outer surface of the prepuce the secretion is displaced by the help of myoepithelial cells) surrounding the glandular acini Fig.5 (B). Secretion is displaced by the help of myoepithelial cells that are surrounding the glandular acini Fig.5 (B).



Figure. (5). A, the various large modified sebaceous glands in the prepuce. **B**, each gland consists of many lobes (**lo**) connected with hair (H), and lobe subdivided to lobules that are surrounded by a areolar connective tissue containing myoepithelial cells (arrows). **C**, the gland surrounded by dense connective tissue capsule (**c**) rich in collage fibers. **C**, **D**, **E** and **F**, each lobule is consisting of mature glandular acini (M ac) consisting of pale cells and disruption cells (dc) and immature acini (IM ac) consisting of dark stained cells, the lateral ducts (Lc) and the main duct (Md) lined with unkeratinized and keratinized stratified squamous epithelium. (A, C, D. E and F stained with (H&E), 20X, 20X and 40X) (B stained with (Masson's Trichrome) 10X).

DISCUSSION

The anatomically study of the great Egyptian jerboa *Jaculus orientis* indicated the presence of glandular masses of numerous shapes arranged around the genital area, randomly arranged in the prepuce of the male and the clitoris of the female. This result agreed with the

results that mentioned by (Mshiri et. al., 2009; Nelson and Thiboul, 2012) in terms of the locations of the preputial glands in Libyan *Jaculusjaculus* as well as with the skin glands of the foreskin *Ctenodactylus gundi* (Mshiri et. al., 2013), while it differed anatomically with what Previously published on the location of the foreskin gland in the Norwegian rat *Rattus norvegicus* and the white laboratory rats *Rattus rattus*, where the preputial gland consists of two glands resembling a ball racquet, surrounded by a thick capsule and drained its secretions through the urethra of the male and female organ (Mshiri et. al.,2005; Yoshizawa, 2018).

The histological study of the anal and the skin of the genital region (uroano-genetal) organ of the great Egyptian Jaculus orientis have two types of scent glands were found; the anal glands which are located in the mucous and submucosal layer of the anal canal surrounded by areolar connective tissue and the perianal glands which are a group of separate glands located between the muscular layer around the anus and each gland is surrounded by a layer of skeletal muscle serve as capsules, both of which are modified sebaceous glands with holocrine secretion however this result agree with the findings regarding the anal scent glands in carnivores as they are consists of compound modified sebaceous glands (Budsberg, 1983 'Trevor et .al., 1981), while contradicted with the histological structure of the anal glands of Ctenodactylus gundi which are consist of compound sebaceous glands with holocrine secretion and compound tubular glands with apocrine secret ion (Mshiri et. al., 2013).

Also this result differs with the anal scent glands of skunks, which are composed of two or three lobes and the glandular tissue consists of compound tubular alveolar glands with apocrine secretion (Heidt and Morgan, 1982), and differ with the anal pouch (acini) glands of aardwolf *Proteles cristatus* that are histologically consists of compound tubular glands with apocrine secretion (Stoeckelhuber et.al., 2000). Also, it's defers from the Brand's hedgehog cloacal populated sebaceous gland (Akbari et. al., 2020). In addition, the current study indicates that each gland of the prepuce area is composed of lobes and each lobe composed of lobules, and each lobule consists of acini which are consist of immature, mature cells. The acini of the gland are connected to the lateral and central canal that lined with unkeratinized stratified squamous epithelial cells. The orifice of the central duct is lined with keratinized stratified squamous epithelium. The secretions displacement with the help of myoepithelial cells surrounded the acini, this finding is agreed with the results of the histological structure of the prepuce skin in Libyan gerbil Jaculus jaculus (Mshiri et. al., 2009), while disagree with the foreskin of the gundi Ctenodactylus gundi (Mshiri et. al., 2013) and also with foreskin of the Norway rat Rattus norvegicus and white rat Rattus rattus (Yoshizawa, 2018; Mshiri et.al., 2005).

CONCLUSION

The anatomical and scatological characteristics of the scent glands are differed slightly between males and females of the same species and also differ between species (but no specific model has been described, necessitating additional research in the same area as well as more through studies to comprehend the transmission of animal behaviors and to increase the local database in Libya.

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ETHICS

The permission for animal ethics and Behavioral Science were taken from the Zoology

department at the University of Tripoli.

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