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Clinical diagnosis and surgical approaches of vaginal hyperplasia in bitches

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ABSTRACT

Objective: To describe the breed predisposition, clinical diagnosis, pathological findings and the surgical approach through excision of the hyperplasic mass from the vagina. Methods: Twenty five bitches of different breeds suffering from a protruded mass from the vulva were examined clinically, blood samples were collected to ensure the phase of estrus that were determined by evaluating the exfoliative vaginal epithelium, and a histopathological examination of the hyperplastic mass was done after its surgical excision. Results: The current work revealed that the maximum value of estradiol 17-B was in Alabai breed while the maximum value of progesterone was in Pit bull breed. And Pit bull breed showed cornification and stratification of the vaginal mucosa with abundant eosinophilic cytoplasm and regular round nuclei. Conclusions: As the vaginal hyperplasia is a crucial gynaecological problem that affects different breeds of bitches, the current work provides a comprehensive diagnosis of the case and illustrates the surgical interference for its excision.

1. Introduction

Vaginal hyperplasia is one of the most common clinical disorders affecting bitches during the follicular phase and may be confused with vaginal neoplasia, transmissible venereal tumor and venereal granuloma[1].

Breed and age factors play a crucial role in almost 80% of the occurrence of vaginal hyperplasia in bitches[2]. The protruding mass of the prolapsed part is susceptible to trauma, laceration and ulceration, and can hinder the natural mating[2]. Vaginal hyperplasia is commonly recorded in younger bitches (<2 to 3 years of age) throughout their first three estrous cycles[3].

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This affection is a sequel to the vaginal hyperemia and edema occurred secondary to the sharp rise of estrogen during the proestrous phase[4]. Vaginal hyperplasia mainly occurs during the first estrous period and could be regressed in the luteal phase[1]. Vaginal hyperplasia could diminish spontaneously through the diestrus but usually reoccurs in 66% to 100% of cases during the next estrus[5,6].

Therefore, the aim of the present study was to demonstrate the breed predisposition, clinical diagnosis, pathological findings and illustration of the surgical approach through excision of the hyperplasic mass from the vagina.

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2. Materials and methods

2.1. Animals and diet

Animal care measures followed the ethical protocols approved by the Ethics Committee for Animal Use at Cairo University (approval number: CU II S 4 18).

This study was carried out on 25 bitches of different ages (2-4 years old), body weights and breeds. These bitches were brought to Theriogenology Clinic, Faculty of Veterinary Medicine, Cairo University with history of hemorrhagic discharge and difficult breeding. The initial investigation revealed gross anatomical deformation, protruded mass from the vagina with difficult urination. The full case history including the feeding program was taken.

2.2. Hormonal assay

Estrogen and progesterone were assayed using immunofluorescence assay technique by VIDAS and mini VIDAS system. (Biomèrieux/France)[7].

2.3. Vaginal cytology

Vaginal swabs were introduced to the vagina followed by gentle rolling; swabs were rolled over clean slides and stained using new methylene blue. The epithelial cells were recorded as mentioned by Ajadi $et\ al$ [3].

2.4. Surgical interference

Two surgical approaches were done depending on the type of the hyperplastic mass; type 1 in which hyperplasia occurred only in the ventral portion of the vagina (the most common form), and type 2 in which hyperplasia comprised the whole vaginal wall (lateral, dorsal and ventral).

The bitches were premedicated using atropine sulfate (0.04 mg/kg) followed by xylazine hydrochloride (2 mg/kg) as a muscle relaxant and the anaesthesia was induced by ketamine hydrochloride (10 mg/kg)[8].

In type 1 vaginal hyperplasia, urethra was catheterized using foley catheter to avoid any suturing of the urethral opening, the prolapsed part was crushed using artery forceps for 5 min, mattress sutures were done using vicryl (Ethicon) supported with round needle, and the prolapsed part was carefully amputated using scalpel.

In type 2 vaginal hyperplasia, urethra was catheterized using foley catheter, series of mattress sutures were done in a circumscribed manner all over the whole prolapsed part using vicryl supported with round needle, while making the sutures, the patency of the vaginal lumen was continuously checked, and the prolapsed part was then carefully amputated using scalpel. Post-operative care including antibiotic and non-steroidal anti-inflammatory pain killer was given for five days, and an Elizabethan collar was set.

2.5. Histopathological examination

Tissue specimens were taken from vaginal hyperplastic masses following surgical excision and fixed in 10% neutral buffered formalin (pH 7.0). Tissue processing was routinely performed to obtain paraffin sections, which were stained by haematoxylin and eosin according to Bancroft and Gamble[9].

2.6. Statistical analysis

Simple one way ANOVA was used and data were presented as mean±standard error (mean±SE) supported with the help of computer software SPSS[10].

3. Results

3.1. Mean values of estradiol 17-B (E17-B) and progesterone (P4) in different dog breeds

Mean values of both E17-B (pg/mL) and P4 (ng/mL) were assayed in each breed. The maximum value of E17-B was in Alabai breed while the maximum value of P4 was in Pit bull breed (Table 1).

Table 1Mean values of E17-B and P4 in different dog breeds (Mean±SE).

Breed	Number of cases	E17-B (pg/mL)	P4 (ng/mL)
Rottweiler	2	47.78 ± 17.70	5.29 ± 1.12
German shepherd	4	41.73 ± 2.23	8.88 ± 0.79
Mastiff	5	61.46 ± 19.46	7.77 ± 0.92
Pit bull	4	35.02 ± 12.36	9.10 ± 1.47
Pug	3	56.28 ± 8.28	6.10 ± 0.34
Boxer	3	60.11 ± 15.13	5.22 ± 0.98
Alabai	2	65.16 ± 9.66	4.20 ± 0.87
Saint Bernard	2	64.22 ± 3.92	5.54 ± 0.67

3.2. Examination of the exfoliated vaginal epithelium

The examined vaginal smear revealed presence of cornified epithelial cells and slight intermediate cells. The examined vaginal smears confirmed that the bitches were in the follicular phase of the estrus (Figure 1).

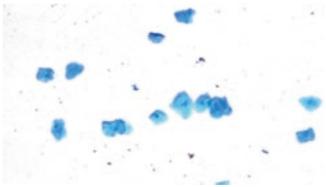


Figure 1. A vaginal smear stained with new methylene blue showing cornified and intermediate cells (×200).

3.3. Surgical approaches

Two surgical approaches were done depending on the site of the protruding mass. Type 1 in which the protruded mass was at the ventral floor of the vaginal lumen (Figure 2 a-d). Type 2 in which the protruded mass comprised the whole vaginal lumen (Figure 3 a-d).



Figure 2. Steps of surgical operation of ventral vaginal hyperplasia.

a) Vaginal hyperplasia protruded ventrally from the vulva; b) Catheter application in the external urethral orifice; c) Suturing the previously crushed hyperplastic mass with a row of mattress sutures using vicryl; d) Excision of the hyperplastic mass.



Figure 3. Steps of surgical operation of complete vaginal hyperplasia.

a) A circumscribed vaginal hyperplastic mass protruded from the vulva;
b) Catheter application in the external urethral orifice; c) A circumscribed suturing the hyperplastic mass with mattress sutures using vicryl; d) Excision of the hyperplastic mass.

3.4. Histopathological alterations

The histomorphological picture of the excised vaginal lesions differed among bitch breeds. The vaginal mucosa of Pit bull breed showed cornification and stratification; the stratum granulosum and germinativum showed hyperplasia and had abundant eosinophilic cytoplasm and regular round nuclei except for some granulosa cells had vacuolated cytoplasm with flattened peripherally located nuclei. The submucosa was composed of hypocellular fibrovascular tissue mainly of collagen containing blood capillaries. The degree of vaginal cornification was marked in German shepherd breed, and the keratin layer retained the nuclei. The histological alteration in vagina of pug breed was the same as previously described cases but the degree of cornification was extremely lower and the stratum granulosum was thinner than previous cases, and the submucosa showed interstitial edema with dilatation of capillaries. The degenerative changes of vaginal stratum granulosum was detected in Mastiff breed, the cellular degeneration was mainly glycogen in nature associated with necrosis. The vaginal mucosa of Rottweiler breed showed neither hyperplasia nor cornification, but ulceration of vaginal epithelium was evident, the inflammatory reaction involving the ulcerated mucosa and extended down ward into the submucosa. The vaginal histomorphological changes in cocker spaniel were extremely different from the previously described cases, the vaginal mucosa showed marked hyperplasia of stratum granulosum with mild keratinization that retained its nuclei, the vaginal epithelium showed not only hyperplasia but also mild dysplastic changes of vaginal epithelium were detected by the increased nuclear/cytoplasmic ratio, hyperchromasia and increased mitosis in lower deeper layers of stratum granulosum and more in stratum germinativum, presence of binucleated cells were observed in vaginal epithelium, and the submucosa was more cellular stromal fibrous tissue that contained numerous spindle shaped cells with inflammatory reaction detected. Marked angiogenesis was recognized in the vaginal stroma with massive interstitial edema, and areas of ulceration and necrosis of vaginal epithelium were detected, which was associated with massive neutrophiles infiltration (Figure 4 a-f) and (Figure 5 a-f).

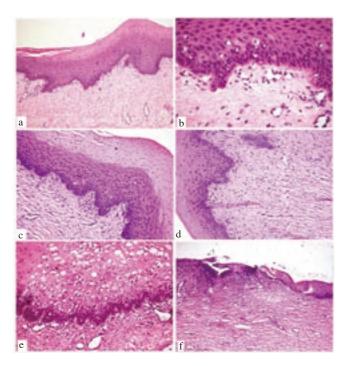


Figure 4. Histological section from vagina (haematoxylin and eosin). a)Partial stratification and hyperplasia of stratum granulosm associated with moderate cornification with partial nucleation of stratum corneum (×200); b) Mild vacuolization of granulose cells note the hypocellular submucosal fibrous collagen deposition (×400); c) A cornification of vaginal mucosa with associated nuclei (×200); d) A thin stratum coreum note the necrosis of granulose cells (×200); e) A diffuse vacuolization and glycogen infiltration of granuolsa cell with necrosis (×400); f) A vaginal mucosal ulceration with inflammatory cell (×100).

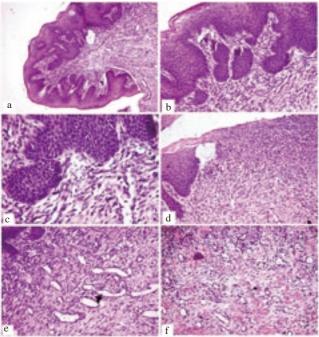


Figure 5. Histopathological alterations in case of cocker spaniel.
a) Vaginal mucosa displays hyperplasia of stratum granulosum and germinativum (×100); b) Downward hyperplastic growth of vaginal epithelium into underlying submucosa with focal dysplasia (×200); c) The hyperplastic cells showing individual dysplasia with mitotic figures, increased N/C ratio and hyperchromasia (×400); d) Ulceration of vaginal mucosa with massive neutrophiles infiltration (×200); e) Vaginal sub mucosa composed of numerous spindles shaped cells with numerous blood capillaries (×200); f) Deeper muscle showing extensive necrosis, edema, angiogenesis and inflammatory cells (×200).

4. Discussion

Canine vaginal hyperplasia is commonly detected in bitches at proestrous and estrous phases, normally the vaginal size and vascularization increase with the increased serum estrogen level during proestrus[11], thus detection of vaginal hyperplasia was common at follicular phase of estrous cycle and reduced at diestrous phase[12].

The current work revealed appearance of a protruded mass varying in size from small bulge to a pear-shaped structure that protrudes from the vulva, also in some cases the whole boundary of the vaginal mucosa was involved. These findings were parallel to the results found by Antonov *et al*[8] who described the vaginal hyperplasia as a small bulge to a tongue-shaped mass.

As breed and age resemble, there is nearly 80% of occurrence of vaginal hyperplasia in bitches. Our results showed that most of bitches with vaginal hyperplasia their ages ranging from one to four years old, and our findings were nearly similar to those found by Nak *et al*[13].

Serum estradiol increases throughout from 5 to 15 pg/mL to reach peaks of 40–120 (mean 70) pg/mL. Serum progesterone rapidly increases above 1–3 ng/mL during the preovulatory luteinizing hormone surge, and immediately (or after a 1–3 days pause) rapidly increases further, reaching 10–25 ng/mL by day 10, at or shortly after the end of estrus[14]. This could illustrate that vaginal hyperplasia is not only affected by the hormonal profile but also the breed predisposition plays a crucial role.

Vaginal cytology as a tool for confirming the follicular phase of estrous cycle was done based on the presence of cornified cells and red blood cells. Similar to our results, Turmalay *et al*[15] described the vaginal cytology as a confirmatory method for determination the phases of the estrous cycle.

The present study showed that there was a variation in histomorphological picture of vagina according to serum level of estrogen. In Pit bull, German, Rottweiler, pug and boxer breed with relatively lower serum level of estrogen compared with mastiff and cocker spaniel breeds, the vaginal hyperplasia is characterized by stratification of vaginal mucosa with mild edema and hypocellular submucosal connective tissue, while in mastiff and cocker spaniel breeds ulceration of vaginal mucosa with subsequent inflammation and degenerative histopathological alterations were evident in these breeds and this is related to higher serum level of estrogen in addition to excessive mitosis and mild dysplastic changes that was observed in Cocker spaniel with marked rise in serum estrogen level. Previous research clarified that persistent rise of serum estrogen level induced marked folding of vaginal floor cranial to urethral papilla and protruded out the vulvar labia with subsequent trauma and development of inflammation[1] and added that canine vaginal hyperplasia was regressed in luteal phase and recurrent with successive periods of estrus and developed after parturition.

In conclusion, the current work provides a comprehensive diagnosis of vaginal hyperplasia in bitches and illustrates the surgical interference for its excision.

Conflict of interest statement

The authors declare that there is no conflict of interest.

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