

Male Pseudohermaphroditism with Clitoromegaly in a Dog: Diagnostic Evaluation and Surgical Correction

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ABSTRACT

Background: Hermaphroditism, or intersexuality, is a pathological condition in which an individual possesses both male and female reproductive organs. Hermaphroditism is classified as true or pseudo-hermaphroditism. True hermaphroditism is referred to an intersex condition in which an ovotestis is present. Pseudohermaphroditism is pathological condition in which an individual has gonads of one sex but the external genitalia of the opposite sex. This case report describes diagnostic procedures, surgical intervention and pathohistological findings in dog with male pseudohermaphroditism and clitoromegaly with penisoid appearance.

Case: A 6-month-old mixed-breed dog was brought to the University Veterinary Clinic (Department of Veterinary Medicine, Faculty of Agriculture, University of Novi Sad) due to frequent urination and licking the vulva. Clinical examination of the reproductive tract revealed an enlarged clitoris. Complete blood count and biochemical analysis of serum did not show any abnormalities in collected blood samples, while the concentrations of estradiol and testosterone were as follows: estradiol 28.5 pg/mL, testosterone 1.8 ng/mL. Castration and clitorectomy were scheduled for the following day. After the midline laparotomy, gonads corresponding macroscopically to testes were observed at the anatomical position of the ovaries. The os clitoris was carefully dissected from the clitoral tissue and completely removed. Pathohistological examination of the left and the right gonads revealed tissue composed of tubules lined solely by Sertoli cells, with no evidence of spermatogenesis. Morphologically, the gonadal tissue (testis) is consistent with the “Sertoli-cell-only” syndrome. No ovarian tissue was identified histologically.

Discussion: Pseudohermaphroditism is generally a rare condition associated with reproductive dysfunction in dogs. Diagnosis of intersex conditions requires a combination of clinical examination, ultrasonography, radiology, endocrinology, and pathohistology, as it has been illustrated in this case. Pathohistological analysis of both gonads confirmed the diagnosis of male pseudohermaphroditism. Microscopically, the presence of Sertoli cells without spermatogenesis and an epididymis without spermatocytes is described as “Sertoli-cell-only” syndrome, indicating that the testes in this case were not functionally developed. Oviductal tissue was also observed histologically. In the serum samples of dog in our case report, the testosterone level was 1.8ng/mL, which probably originates from the secretory role of Leydig cells. On the other hand, since the pathohistological analysis of both gonads did not find ovarian tissue (ovotestis), the question remains is - how the estradiol level was 28.5 pg/mL. Pathophysiologicaly, level of estradiol can be partially explained by pathology of Leydig cells or other physiological sources of the organism. This case confirms that early recognition of intersex conditions prevents complications and improves patient quality of life. Reproductive capability is not expected in such dogs, and surgical intervention ensures a favorable prognosis and preserves quality of life. Male pseudohermaphroditism with pronounced clitoromegaly is unfrequently described in the literature, while recent reports have described this condition in pure-breed dogs but also in one mix-breed, as it is described in this case. In this case report, the symptoms were completely resolved and the animal remained in good general condition after the surgery.

Keywords: reproduction, surgery, sexual disorders, DSD, pseudohermaphroditism.

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INTRODUCTION

Disorders of sexual development (DSD) may occur during the chromosomal sex determination, the gonadal development, or the development of phenotypic sex characteristics, and all are associated with infertility and sterility [4,12]. Hermaphroditism, or intersexuality, is a pathological condition in which an individual possesses both male and female reproductive organs. Hermaphroditism is classified as true or pseudo-hermaphroditism. True hermaphroditism is referred to an intersex condition in which an ovotestis (gonad containing both ovarian and testicular tissue) is present. On the other hand, pseudohermaphroditism is pathological condition in which an individual has gonads of one sex but the external genitalia of the opposite sex. Furthermore, pseudohermaphrodites are categorized as male or female depending on the type of present gonads [3,10]. Male pseudohermaphroditism is characterized by a male chromosomal and gonadal sex (XY karyotype and testes) combined with female external genitalia [7]. In this form of DSD, the presence of clitoromegaly has also been described. Clitoromegaly results from partial masculinization under the influence of androgens during embryonic development. In these individuals clitoris is enlarged, often causing discomfort to the animal and stimulating licking behavior [5]. This case report describes diagnostic procedures, surgical intervention and pathohistological findings in a dog with male pseudohermaphroditism and clitoromegaly with penisoid appearance.

CASE

A 6-month-old mixed-breed dog was brought to the University Veterinary Clinic (Department of Veterinary Medicine, Faculty of Agriculture, University of Novi Sad) due to frequent urination and licking the vulva. Clinical examination of the reproductive tract revealed an enlarged clitoris (Figure 1). After the clinical examination, ultrasonographic (BPU 60 Vet)¹ examination in B-mode in real time, with micro-convex probe 8 MHz, (Figure 2) and radiographic [ZooMax Gold 5G0844]² (Figure 3) examinations were conducted and revealed a shadowing pattern consistent with bone density, which was interpreted as os clitoris. Based on these findings, a presumptive diagnosis of hermaphroditism was made.

Complete blood count and biochemical analysis of serum did not show any abnormalities, while

the concentrations of estradiol and testosterone were as follows: estradiol 28.5 pg/mL and testosterone 1.8 ng/mL (Immunoassay analyzer Access 2)³. Castration and clitorrectomy were scheduled for the following day.

The surgical procedure was performed on the patient under the general anesthesia, following all principles of good veterinary and surgical practice. Premedication included intravenous administration of sedatives. Sedation was achieved using xylazine [Xylased® - 1 mg/kg, IV]⁴. After premedication, the surgical field was prepared according to all principles of antisepsis. Induction was performed with mixture of ketamine and midazolam by intravenous administration [Ketamidor® - 3 mg/kg; Dormicum® - 0.4 mg/kg]^{5,6}. Maintenance of general anesthesia was achieved with sevoflurane [Sevorane®]⁷ with MAC of 2%. Intraoperative analgesia was conducted with bolus intravenous administration of fentanyl - 5 µg/kg [Fentanyl Panpharma]⁸. After the midline laparotomy, gonads corresponding macroscopically to testes were observed at the anatomical position of the ovaries. The gonads were removed along with the organ resembling uterus (Figure 4). Two ligatures were placed proximally to each gonads (at the site which anatomically corresponded to the mesovarium), and 2 ligatures were placed at the caudal end of the organ to which the gonads were anatomically connected, which anatomically corresponded to the body of uterus. Polyglycolic acid was used for all of the ligations (PGA, USP 2/0)⁹ and simple interrupted sutures were made. Following castration, surgical removal of the clitoromegaly was performed. An urinary catheter was placed in the normal direction after cystotomy (Figure 5) to identify the urethral papilla. The os clitoris was carefully dissected from the clitoral tissue and completely removed (Figure 6), and remaining clitoral tissue was closed with simple continuous suture - polydioxanone [Monosorb - USP 2/0]⁹. After castration and removal of the os clitoris, bladder was closed with Lembert continuous suture in one layer manner - polydioxanone [Monosorb - USP 3/0]⁹. Abdomen was closed with simple continuous suture - polydioxanone [Monosorb - USP 2/0]⁹. After the abdomen, subcutaneous tissue was closed with simple continuous suture - polydioxanone [Monosorb - USP 2/0]⁹. Skin was closed with simple continuous suture - polyamide [Nylon, USP 2/0]⁹. Postoperatively, the patient received parenteral administration of benzylpenicillin-procaine in combination with

dihydrostreptomycin-sulfate [Penstrep-ject - 1 mL/10 kg]¹⁰ and carprofen [Rycarfa® - 4 mg/kg]¹¹.

The gonads were fixed for 48 h in 10% formalin after the surgical procedure and sent for pathohistological analysis, while the removed clitoris (Figure 7) was subjected to boiling to remove soft tissue and isolate the bone (Figure 8).

After tissue fixation (gonads), tissue processing and embedding in paraffin were performed. Tissue sections were cut on a microtome at a thickness of 5 µm, stained with hematoxylin and eosin (HE), and then analyzed microscopically. Pathohistological examination of the left gonad (Laboratory of Pathology at the Scientific Veterinary Institute “Novi Sad”, Novi Sad, Serbia) revealed tissue composed of tubules lined solely by Sertoli cells, with no evidence of spermatogenesis (Figure 9). The interstitium around the tubules contained moderate Leydig cell hyperplasia, consistent with the onset of puberty in the animal. Adjacent to the testicular tissue, epididymal tissue with empty tubules was observed (Figure 10), as

well as a portion of tissue histologically corresponding to the oviduct (Figure 11). Morphologically, the gonadal tissue (testis) is consistent with the “Sertoli-cell-only” syndrome. No ovarian tissue was identified histologically.

Due to the uniqueness and specificity of the case, the right gonad was sent to another laboratory for analysis (TL VetPath International Consultants, Hiawatha, USA), resulting in 2 independent evaluations. Pathohistological analysis of the right gonad was performed using the same procedure as for the left gonad. The examined tissue consisted of testis, epididymis, with a segment of vascular plexus, and with segment of oviduct. The gonad (testis) was composed of seminiferous tubules lined exclusively by Sertoli cells, with no elements of spermatogenesis (Figure 12). The epididymis consisted of mildly dilated tubules containing small accumulations of eosinophilic material, with no spermatocytes present (Figure 13). The oviductal tissue showed no significant microscopic changes (Figure 14).

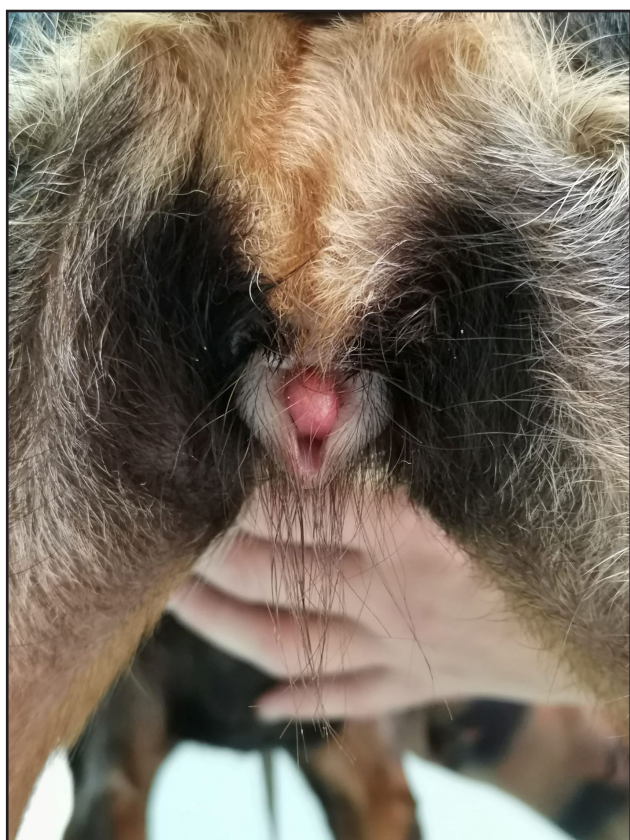


Figure 1. Enlarged clitoris observed during clinical examination.

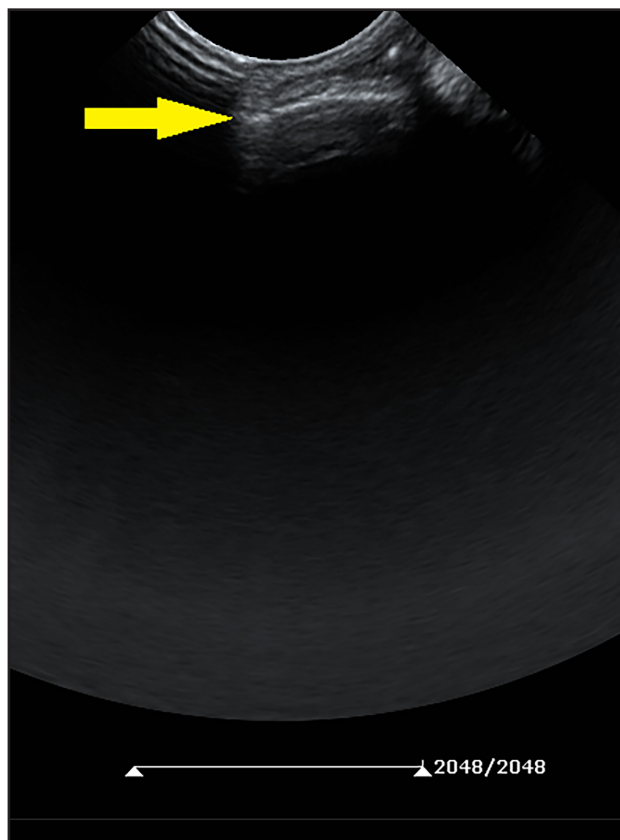


Figure 2. Ultrasonography revealed a clearly identifiable structure with evident bone density (arrow) in the enlarged clitoris.

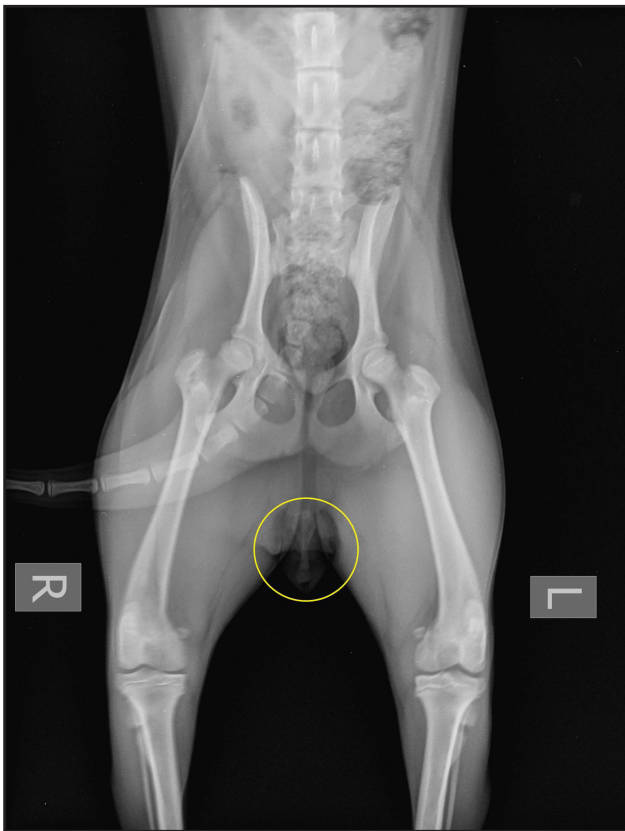


Figure 3. Plain radiography revealed a clearly identifiable structure with evident bone density (circle) in the enlarged clitoris.

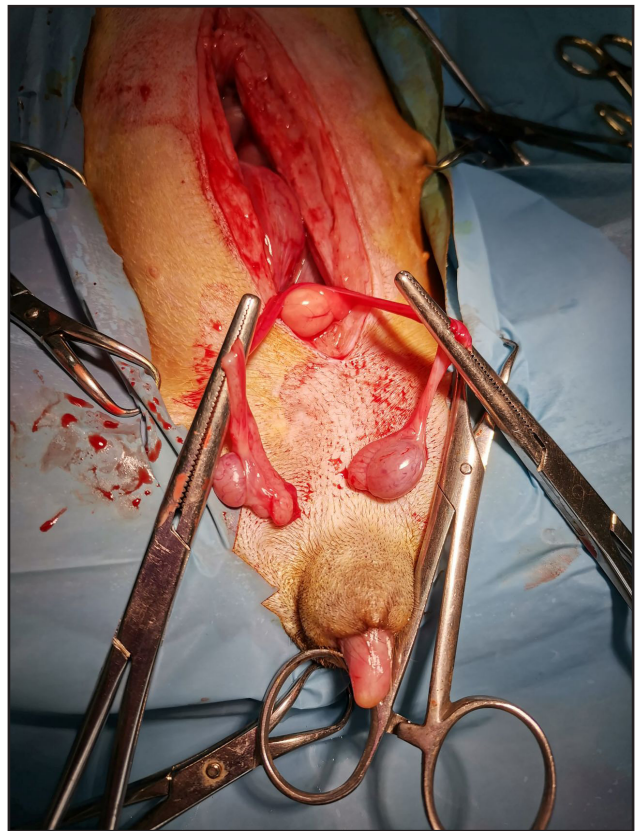


Figure 4. Laparotomy revealed gonads morphologically consistent with testes, located in tissue resembling the uterus.



Figure 5. The normograde passage of the rigid ureteral catheter.

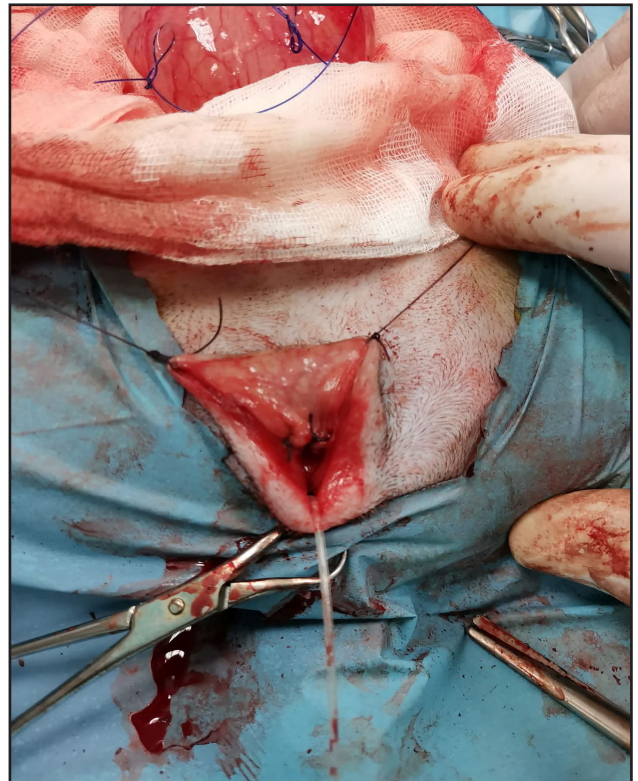


Figure 6. Vagina after surgical removal of the clitoris.



Figure 7. Clitoris after surgical removal.



Figure 8. Os clitoris after removal of surrounding soft tissue by boiling.

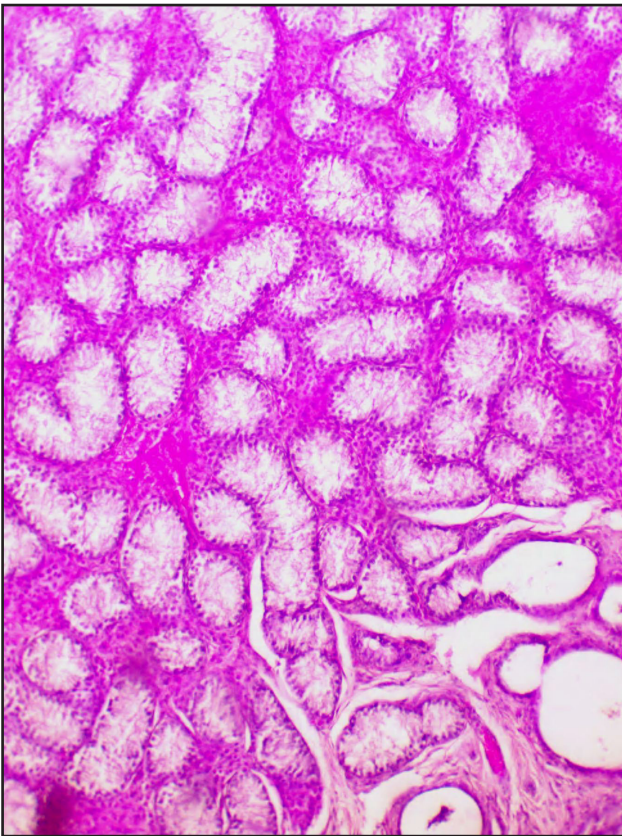


Figure 9. Testicular tissue composed of tubules lined only with Sertoli cells, without evidence of spermatogenesis [HE; 40x].

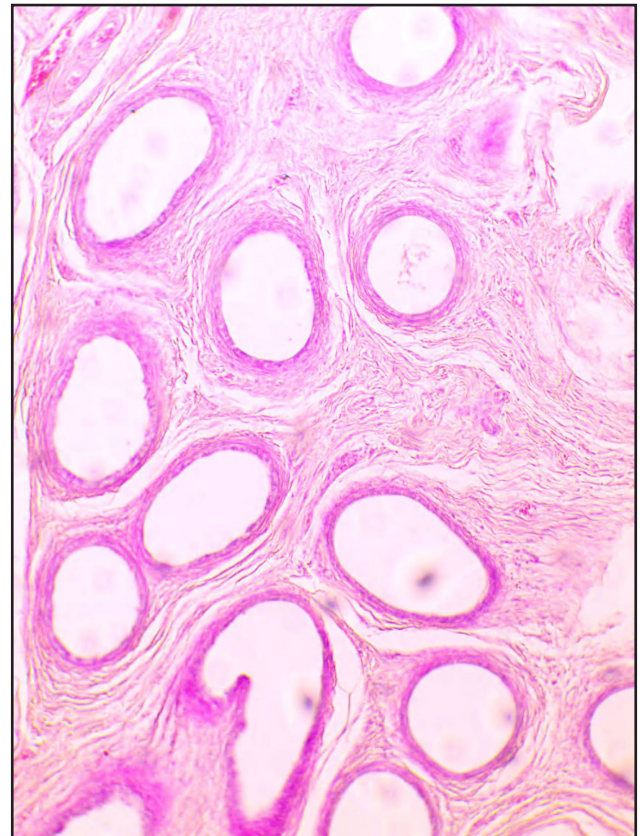


Figure 10. Epididymal tissue with empty tubules [HE; 100x].

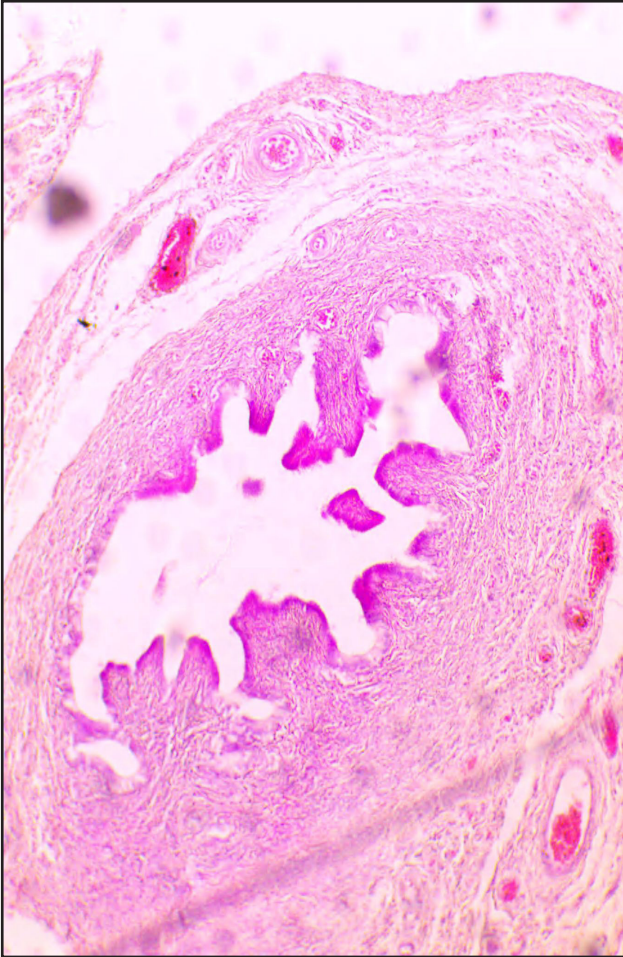


Figure 11. A portion of tissue that histologically corresponds to the oviduct [HE; 100x].

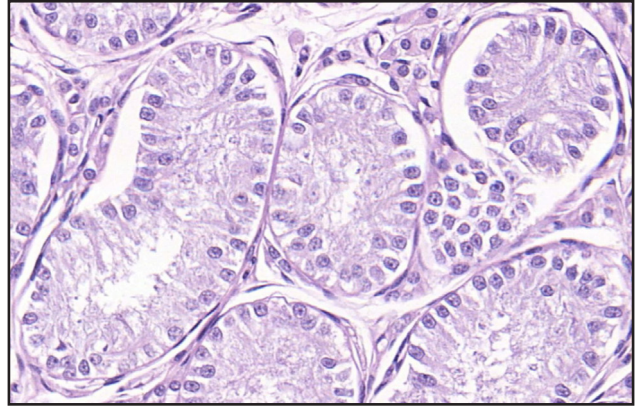


Figure 12. Testicular tissue with presence of Sertoli cells without spermatogenesis [HE; 400x].

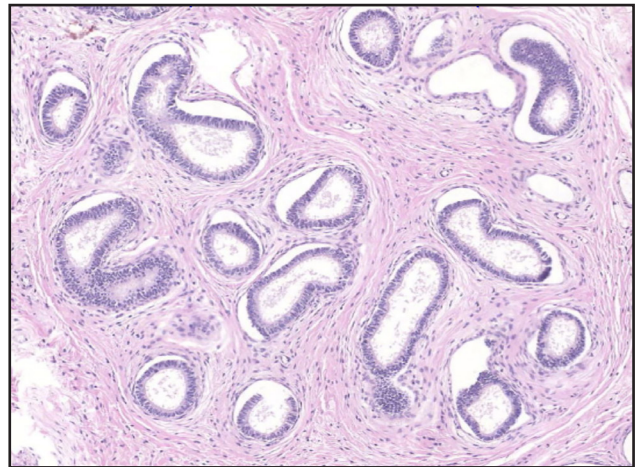


Figure 13. The epididymis consists of mildly dilated ducts containing small accumulations of eosinophilic material, with no spermatozoa present [HE; 100x].



Figure 14. Oviductal tissue showing no significant microscopic alterations [HE; 100x].

DISCUSSION

Pseudohermaphroditism is generally a rare condition associated with reproductive dysfunction in dogs. With the increasing number of dogs as social companions, recent literature reports such disorders in purebred dogs, whereas they are less common in mixed breed dogs [4,5,10], as demonstrated in this case report. A multimodal diagnostic approach is crucial in diagnosis of disorders of sexual development. Diagnosis of intersex conditions requires a combination of clinical examination, ultrasonography, radiology, endocrinology, and pathohistology, as it has been illustrated in this case. Only complete diagnostics can lead to an accurate diagnosis. Surgical intervention for clitoral amputation is justified in cases such as clitoral hypertrophy, which can result in constant licking, chronic inflammation, pain, and trauma [4,5]. Gonadectomy is also recommended as essential, since without it various reproductive and endocrine disorders may occur, and it also improves the animal's quality of life and prevents reproduction [2,4,10]. The surgical outcome in this case was entirely favorable, with no postoperative complications. Pathohistological analysis of both gonads confirmed the diagnosis of male pseudohermaphroditism. Microscopically, the presence of Sertoli cells without spermatogenesis and an epididymis without spermatocytes is described as "Sertoli-cell-only" syndrome [13], indicating that the testes in this case were not functionally developed. Oviductal tissue was also observed histologically. The presence of oviductal tissue in this case may be pathophysiologically attributed to an intrauterine failure of Müllerian duct regression, due to a lack of Müllerian-inhibiting factor (secreted by Sertoli cells of the testes) caused by functionally inactive testes, or the inability of androgens to masculinize target organs, as previously described in some cases [4,6,9]. However, in our case, this remains only a hypothesis. In the serum samples of a dog in our case report, the testosterone level was 1.8 ng/mL, which probably originates from the secretory role of Leydig cells. On the other hand, since the pathohistological analysis of both gonads did not find ovarian tissue (ovotestis), the question remains is - how the estradiol level was 28.5 pg/mL. Pathophysiologically, level of

estradiol can be partially explained by pathology of Leydig cells [8] or other physiological sources in the organism [11]. In our case report, we could not find the correlation between pathohistological findings of male pseudohermaphroditism and levels of estradiol. In addition, the owners of the dog in this current report declined further diagnostic tests, and correlation of hormonal and pathohistological findings could not be evaluated in more detail, and also will not be discussed furthermore, to avoid assumptive conclusions. This case confirms that early recognition of intersex conditions prevents complications and improves patient quality of life. Reproductive capability is not expected in such dogs, and surgical intervention ensures a favorable prognosis and preserves quality of life. Male pseudohermaphroditism with pronounced clitoromegaly is unfrequently described in the literature, while recent reports have described this condition in pure-breed dogs [1,4,5] but also in 1 mix-breed [10], as described in this case. In this case report of a mixed-breed dog, the symptoms were completely resolved and the animal remained in good general condition after the surgery. Additional case reports of this disorder could contribute to the standardization of diagnostic and surgical protocols.

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