Animal Health Prod and Hyg (2021) 10 (1) : 27 - 32

Animal Health, Production and Hygiene

www.aduveterinaryjournal.com

Case Report

Ovarian Metastatic Transmissible Venereal Tumour in a Bitch – A Case Report and Review

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ABSTRACT

Approximately 5% of transmissible venereal tumour can metastatize to various parts of the body. The case presented explains the clinical appearance and histopathology results of the ovarian metastasis of TVT in a female German shepherd. The bitch was brought to the teaching hospital of Ondokuz Mayıs University, Faculty of Veterinary Medicine with a vaginal discharge. A mass with a diameter of 3x1x1 cm was detected on the dorsal vaginal wall. In vaginal cytology, the presence of TVT cells were determined. During ultrasonographic evaluation a mass was detected caudal to the left kidney with irregular margins. The dimension of the mass after ovariohysterectomy was found to be 9x11 cm. Because it was necessary to investigate the possibility of metastasis and to make comparison, the vaginal mass was also removed and sent for histopathological examination and both masses were identified as TVT. According to our knowledge this is the first report presenting a case of TVT with solely ovarian metastasis.

Keywords: bitch; ovarian metastasis; Sticker's sarcoma

Bir Dişi Köpekte Ovaryuma Metastaz Yapan Transmissible Veneral Tümör Olgusu-Olgu Sunumu ve Derleme

ÖZET

Transmissible veneral tümör (TVT) %5 oranında vücudun farklı bölümlerine metastaz yapmaktadır. Sunulan olguda dişi bir Alman çoban köpeğinde TVT'nin ovaryuma metastazı ve klinik görünümü anlatılmaktadır. Uzun süreli kanlı vaginal akıntı şikayetiyle getirilen olgunun vaginal duvarınının dorsalinde 3x1x1 cm çapında kitle belirlendi. Vaginal sitolojide TVT hücreleri görüldü. Ultrasonografik muayenede sol böbreğin kaudalinde, kenarları düzensiz bir kitle tespit edildi. Ovaryohisterektomi sonrası ölçümlerde kitlenin 9x11 cm boyutlarında olduğu belirlendi. Kitlenin metastaz sonucu şekillenme ihtimali nedeniyle, her iki kitlenin karşılaştırılması amacıyla, vaginal kitle de alınarak histopatolojik değerlendirmeye gönderildi. Değerlendirme sonucunda her iki kitlenin de TVT olduğu belirlendi. Yaptığımız araştırma sonucuna göre, sunulan olgu TVT'nin sadece ovaryuma metastaz yaptığı ilk olgu olma özelliğindedir.

Anahtar kelimeler: köpek; ovaryum metastazı; Sticker sarkomu

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Received Date: 11.02.2021 – Accepted Date: 22.03.2021

Introduction

Tumour cells generally originate from long term genetic and epigenetic changes. First phase is the irreversible genetic differentiation, also called promotion, is followed by the end phase, which is the change of the benign tumour into a malign one and gaining metastatic characteristics. Normally, transmission of tumours with contact is not possible. However transmission of tumours that does not fit that explanation were first discovered in dogs as transmissible venereal tumour (TVT) (in 1820), then Syrian hamsters as contagious reticulum cell sarcoma (in 1961) and Tasmanian devils as devil facial tumour disease (in 1996) (Murchison, 2008). Transmissible venereal tumours have special significance because it was the first to be discovered and it was seen globally. The transference of tumour cells were only discovered at 1995 even though the tumour was believed to be around for 2500 years (Murgia et al., 2006; Temitope et al., 2010). Other tumour types usually occur in later stages of life (6-10 years) though TVT happens at an earlier age (2-5 years) because it happens due to dissimilar mechanisms (Gürel et al., 2002). Transmissible venereal tumour has an incidence of 2.8% among all tumours of the dog (Brodey and Roszel, 1967). While TVT is mostly benign, 5% of the cases have the ability to metastasis. The chance of metastasis in males (16%) is greater than females (2%) (Martins et al, 2005), thereby metastatic TVT cases are especially rare in bitches.

Up until now many canine TVT cases were reported with metastases to many organs and systems like skin and subdermal tissue, mammary glands, brain, eye, palpebral conjunctive tissue, nasal mucosa, soft palate, mediastinum, lungs, liver, spleen, lymph nodes (Higgins, 1966; Adam and Slaughter, 1970; Ayyappan et al., 1994; Baştan et al., 2008; Özyurtlu et al., 2008; Temitope et al., 2010; Behera et al., 2012; Milo and Snead, 2014; Uçmak et al., 2019). To the best of our knowledge the only metastasis of TVT in the reproductive system was the metastasis of TVT to the uterus and ovaries reported by Baştan et al. (2008). This case presented here is the first to report the solely ovarian metastasis of TVT in a bitch. The clinical appearance of the case and the treatment will be explained in this report.

Patient History, Clinical Findings and Treatment

The material of the study was a two years old German shepherd dog weighing 24 kg that was brought to our clinic with a vaginal discharge. The patient's condition got progressively worse, with loss of appetite and lethargy. The bitch had a healthy delivery and litter in the previous cycle, and according to the owner, the vaginal discharge was present for two months as dripping blood. During inspection a trickle of blood could be seen coming from the vulvar lips and palpation revealed a mass in the dorsal wall of the vagina. The dimensions were measured as approximately 3x1x1 cm. The patient had a body temperature of 39.3 oC, a respiratory rate of 32/min, a heart rate of 124 per minute, hyperaemic mucosae and normal sized lymph nodules during clinical examination. Complete blood count (CBC) values were as follows; white blood cell (WBC) 28.31 x103/mcL (Reference Interval, RI, 5-14.1 x103/mcL); red blood cell (RBC) 10.21 x106 /mcL (RI: 4.95-7.87 x106 /mcL); hemoglobin (HgB) 22.1 gr/dL (RI: 11.9-18.9 gr/dL); hematocrit (Ht) 58.59% (RI: 36-60%); lymphocyte (LYM) 8.74% (RI: 8.21%). Serum biochemistry values were; total protein (TP) 8.0 g/dL (RI: 5.4-7.5 g/dL); albumin (ALB) 2.1 g/dL (2.3-3.2 g/dL) ve globulin 5.9 g/dL (RI: 2.7-4.4 g/dL), urea 32.8 mg/dL (RI: 8-28 mg/dL) and creatinine 0.76 mg/dL (RI: 0.5-2,7 mg/dL).

A vaginal smear was taken with a cotton swab and stained with Papanicolau method, then evaluated using a light microscope. There were many lymphocytic cells with intracytoplasmic vacuoles that had a high nucleus vs cytoplasm ratio. This finding confirmed the diagnosis as TVT.

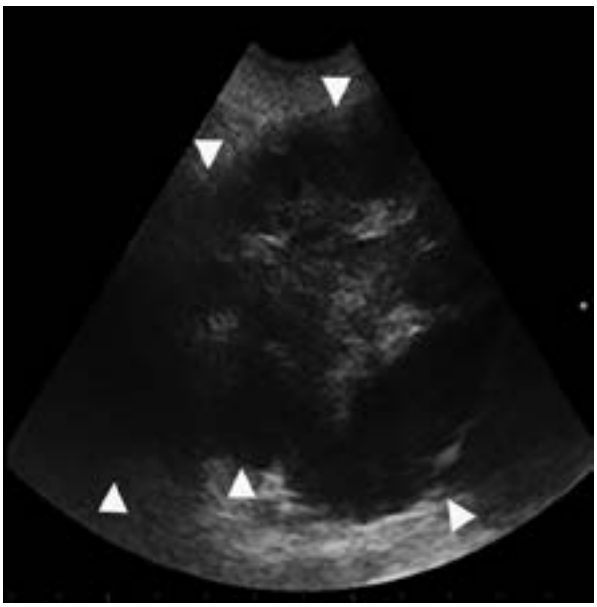

Figure 1. The ultrasonographic appearance of the mass. The mass is marked between the white arrows.

There was no discernible pathology in the patient's lateral radiograph. Ultrasonographic examination revealed no pathologies in the uterus but a mass with irregular margins including anechoic areas and having a heterogeneous structure with the dimensions of 5.9x6.5 cm was detected (Figure 1). The mass encountered during the USG examination was suspected to be a tumoral mass an ovariohysterectomy was performed.

Induction of the anaesthesia was performed with propofol (6 mg/kg, i.v. Fresenius Kabi, Sweden) and maintained by isoflurane (2%, Forane, Abbout, England) in oxygen. Meloxicam (0.2 mg/kg, i.v. Maxicam, Sanovel, Turkey) was used for pain management both 45 minutes before surgery and once daily post operatively for 2 days.

The patient was placed in dorsal recumbency and following surgical asepsis, a ventral midline incision was made caudally to provide exposure. During exploratory laparotomy all reproductive organs except the right ovary were seen in their normal anatomic positions and they had normal thickness and structure. The solid mass was at the site of the right ovary, connected to the uterus and had no adhesions to other viscera. The right ovary was not seen at the site; hence the mass was considered as an ovary tumour. After the exploration ovariohysterectomy was initiated. Initially the left suspensory ligament and adjacent broad ligament were ligated and cut. Suspensors of the right ovary could not be reached and thus incision was extended by 3 cm cranially. Same procedures were applied to the suspensors of the right ovarian mass. After releasing both horns, a transfixation ligature was place on the cervix uteri and uterine body was excised, including a portion of cervix uteri. The cervix uteri, muscles and subcutaneous tissue were closed using USP:0, PGA sutures, and the skin was closed with USP: 0, silk sutures. Later on, the vaginal mass was removed with the seemingly healthy mucosa it was attached to, and sent for histopathological evaluation to determine if the two masees were related. The vagina was sutured using USP: 2/0 PGA sutures. During the operation a Ringer Lactate

solution was given as i.v. infusion at a dosage of 10 mg/kg/hour. Following the operation, enrofloxacine (5 mg/kg/day, s.c, Baytril-K, Bayer) was administered for five days.

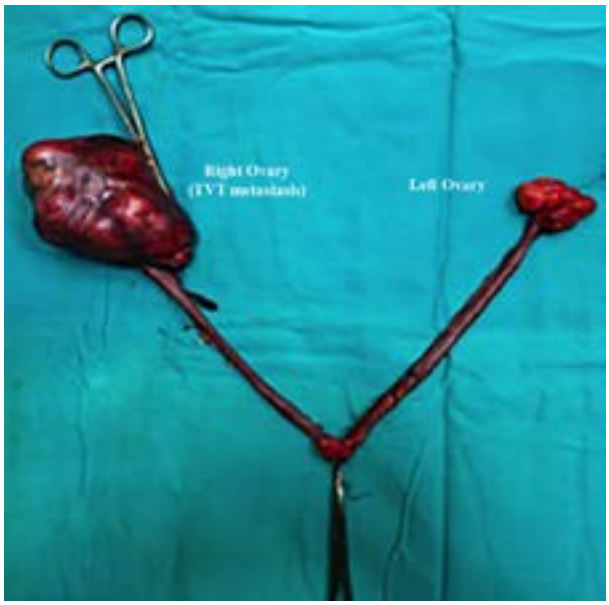

Figure 2. Postoperative image of uterine horns, right ovary and mass.

Macroscopic evaluation of the extirpated tissues were made post operatively, the right uterine horn was measured as 19.7 cm long and 0.8 cm wide, the left as 16.5 cm long and 0.7 cm wide (Figure 2). The ovarian mass was 11x9 cm in size and when cut, the cross section was white gray in colour with a multilobular arrangement showing no apparent cystic formations or fluid accumulation (Figure 3). The vaginal mass was relatively soft and had fresh blood on it.

The ovarian mass, left ovary and vaginal mass was fixed in 10% formaldehyde solution and sent for histopathological examination (Pathology department of Ondokuz Mayıs University, Faculty of Veterinary Medicine). The tissues were further dehydrated inside Histokinet (Leica, TP1020), which included alcohol and xylene series and buried in paraffin. The samples were then cut in sections of 5 microns with a microtome (Leica, RM 2125RT). The sections were dyed using hematoxylene-eozin and were later evaluated with a light microscope (Nikon, Eclipse E600).

Histopathological examination of the vaginal mass revealed widespread dispersion of uniform, round to oval cells in a fine fibrovascular stroma (Figure 4). Cells nuclei were round, with a single centrally placed nucleolus surrounded by marginated chromatin and mostly located centrally. Small amounts of light amphophilic-to-clear cytoplasm was seen in some areas where the cells are not tightly packed. Moderate rate of mitosis was observed. According to these findings, the mass was diagnosed as TVT. Because the ovarian mass (Figure 5) had the same histopathological characteristics with the vaginal mass it was presumed that the ovarian mass has developed as a result of the vaginal TVT.

The previously planned chemotherapy could not be performed because the patient owner did not bring the bitch to the clinic after the removal of sutures. Patient health status was inquired twice at 3 month intervals via phone surveys. According to this, no complications were observed following suture removal, no further drug administrations were needed and the general condition of the bitch was good.

Discussion

Transmissible venereal tumour also known as sticker sarcoma, venereal granuloma and infectious sarcoma, is still a problem in tropical and subtropical climates that have large populations of uncontrolled stray dogs. Transmission happens during mating or social activity. The transmission rate is greater in sexually active animals (Ganguly et al., 2013).

The lesion usually appears 2-3 weeks as a 1-3 mm mass after transmission and can grow to a size of 15 cm in time. Symptoms change depending on size of the mass, the effected organ or system. In genital TVT the appearance may vary but it usually appears as a cauliflower-like, fragile and bloody (Hoque, 2002). This case's age, discharge and palpation findings were in accordance to other cases in literature.

Ovarian tumours in bitches originate from epithelial cells, germ cells, sex cord stroma or mesenchymal stroma (Kennedy et al., 1998). Ovarian tumours are usually unilateral and on the left. The prevalence of ovarian tumours in bitches is uncertain because they are usually encountered during routine ovariohysterectomy or necropsy (Smith, 2003). In some papers the prevalence of the disease was reported to be between 0.5% and 1.2% (Klein, 1996). These tumours can be encountered during ultrasonographic examination although small masses may not be detected during USG (Smith, 2003).

Transmissible venereal tumour has a biological tendency to stay locally. However, metastases may occur in young, immunosuppressed or malnourished dogs (Maclachlan and Kennedy, 2008). Metastasis requires its originating tumor to be malignant, and can happen with the migration of only a few tumor cells (Cullen et al., 2008). Although it is yet uncertain how the tumour cell chooses its way or complete the process (Veer et al., 2002; Ramaswamy et al., 2003), metastasis is usually on the closer lymph nodes but sometimes far site metastases may happen (Maclachlan and Kennedy, 2008). In TVT cases, metastases to lymph nodes and other organs have been previously reported (Başţan et al., 2008; Milo and Snead, 2014).

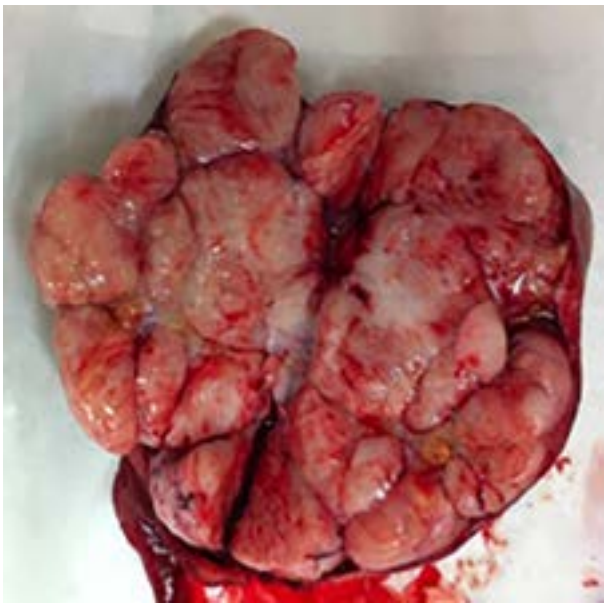

Figure 3. Sagittal section of the mass.

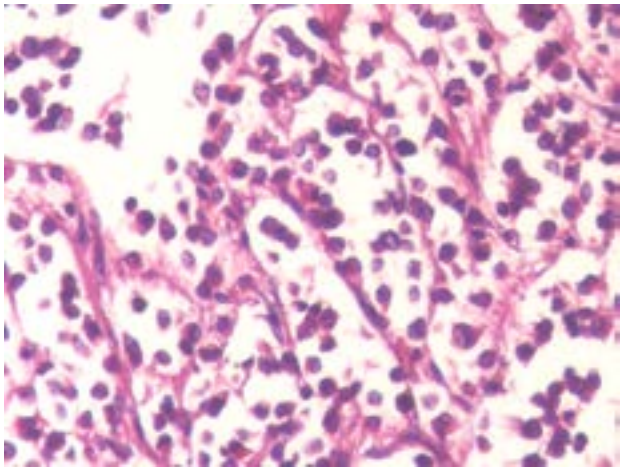

Figure 4. Uniform, round to oval cells placed on a fine fibrous stroma. Vaginal mass. TVT, Bitch, HxE, x40.

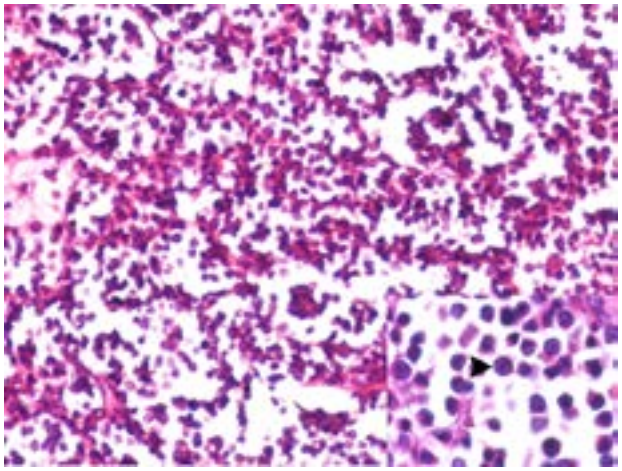

Figure 5. Histological characteristics are similar to vaginal mass. Same round to oval cells with scant amphophilic to clear cytoplasm are seen on a fine fibrous stroma. Inset: The nucleoli are big in relative to cell size and a single nucleolus placed centrally or peripherally (arrow head).Ovarian mass.TVT, Bitch. HxE, x20, Inset: x100.

The clinical findings of ovarian tumours differ depending on the type of the tumour. Symptoms like anoestrus, nymphomania, masculinisation, vaginal discharge, abdominal masses/distension (due to tumours or effusions), anorexia, vomiting, weight loss, hyperestrogenism in uterus and related pathologies such as pyometra, cystic endometrial hyperplasia (CEH) and metaplasies (Robbins, 2003) were reported. In the present clinical case clinical evaluation have revealed that the metastasis was on the right ovary and no clinical dysfunctions, discomfort or abdominal distension were seen. The patient history was incomplete due to the owner's lack of attention to the animal, so the previous cyclic activity of the bitch was unknown. Also no laboratory findigs concerning oestrogen levels were available so only the clinical findings were used to estimate the presence of hyperoestrogenism. Absence of vulvar swelling and alopecia led us to conclude that hyperoestrogenism was not present. In addition, macroscopic examination of the uterus after OHE showed there was no CEH, endometritis or metaplasia present.

The was showing advanced levels of anorexia and lethargy, however these may be attributed to the presence of chronic blood loss. Some previous TVT studies show no difference in CBC values (Das et al., 1991) while others report leucocytosis (Behera et al., 2012). The leucocytosis seen in this case was interpreted as stress related due to lymphocytosis. The increased values of RBC, HGB and HCT values combined with steady values of urea creatinine suggested primary absolute polycythemia. Large tumours are known to induce polycythaemia, increasing erythropoietin and erythropoietin extraction (Maclachlan nad Kennedy, 2008). The change to CBC in this case may be due to the metastatic mass. Low albumin count in CBC was thought to be due to an inflammatory reaction (negative acute phase proteins), the increased globulin levels were interpreted to be the result of a chronic antigenic effect. Immunologic studies concerning TVT also state that it has an antigenic effect and that effect increases as the tumour size grows (Ganguly et al., 2013). This finding may explain the increased levels of globulin in our case. However Behera et al. (2012) did not see any increase in globulin levels while there was a decrease in albumin. The retrospective study done by Kabuusu et al. (2010) show lymphopenia and mild anaemia in the CBC results.

Presence of the metastatic mass was revealed with the routine gynaecologic examination. Transmissible venereal tumour cases were also seen with uterus pathologies, ovarian remnant syndrome, abdominal/reproductive metastases and rarely urinary obstruction in previous literature which also suggests that abdominal/reproductive USG should be performed in such cases (Feldman and Nelson, 2004; Başţan et al., 2008; Sontaş et al., 2010; Chikweto et al., 2013). Healthy ovaries, especially in anoestrus, are quite difficult to detect with USG. However ovarian tumour can include solid or cystic masses. Those with cystic characteristics may be confused with non-neoplastic structures such as follicular or luteal cysts. Tumour structures are usually bigger in size and include liquid filled compartments (Smith, 2003). This data shows the accuracy of the diagnosis made with this case as an ovarian mass. In this case report, the ultrasonographic findings, the size of the tumours and sighting of the anechoic areas are similar to the aforementioned literature.

Occasionally TVT cases may recover spontaneously and most are in benign nature but all of them should receive treatment because of their progressive characteristics (Das and Das, 2000). There are many treatment methods like radiotherapy, chemotherapy, surgery, immunotherapy, cryotherapy or their combination for this purpose (Martins et al., 2005). Recently the most commonly preferred treatment choice is chemotherapy which requires weekly use of vincristine sulphate (i.v. infusion, 0.025 mg/kg) for both primary and secondary masses (Bhatia et al., 2010). However, surgery has been reported to be the first choice for ovarian tumours (Smith, 2003). In the present case, initial treatment was to continue chemotherapy using vincristine following surgical removal of the tumour mass. The resection of the vaginal mass was performed to compare the ovarian mass histopathologically to determine if metastasis was present. Resection was performed rather than a biopsy because of the small size of the vaginal mass. Unfortunately, vincristine sulphate treatment could not be carried out due to the decision of the patient owner. Surgical intervention is not recommended in TVT cases because recurrence is seen in 18-60% of cases (Ganguly et al., 2013). However, it is applicable to smaller sized masses such as in our case (Martins et al., 2005). In the bitch presented in the present study surgery seemed to be succesful for the treatment of TVT. In this case, the failure of the vaginal mass to recur later can be evaluated as the success of the surgical intervention. However, considering the high recurrence rate of surgical intervention, it can be predicted

that spontaneous recovery may play a role in remission. In TVT cases, remission starts after a two months long progressive phase14 and 16% of cases spontaneously recovery (Feldman and Nelson, 2004). Our opinion is, owing to the antigenic nature of the mass, the response of T and B-lymphocytes, TGF-B1 and NK cells might have a supportive effect during this period (Murchison, 2008).

As a result, our opinion was that sole ovarian metastases TVT are possible and could metastasize only to the ovary and a thorough clinical and gynaecologic examination; a thorough utrasonographic evaluation of the reproductive system should be performed even though the patient shows no outward clinical symptoms of any disorder.

Acknowledgement

This case report has been presented at "IV Veterinary Gynaecology Congress, Fethiye, Turkey, 15-18 October 2015" and "8th International Symposium on Canine and Feline Reproduction with XIX EVSSAR Congress, Vienna, Austria, 29 June-1 July 2017". The authors would like to thank Dr. Taylan ÖNYAY for the language edition.

Ethical approval

This work involved the use of non-experimental animals only (including owned). Established internationally recognized high standards ('best practice') of individual veterinary clinical patient care were followed. Ethical approval from a committee was therefore not necessarily required.

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