

# GENERALIZED SYMMETRIC ALOPECIA AND HYPEROESTROGENISM ASSOCIATED WITH CONCURRENT LYMPHOMA, SERTOLI CELL TUMOUR AND SEMINOMA IN A SAMOYED

Tanja Plavec\*, Nataša Tozon, Tina Kotnik

Clinic for Small Animal Medicine and Surgery, Veterinary Faculty, Gerbičeva 60, 1000, Ljubljana, Slovenia

\*Corresponding author, E-mail: tanja.plavec@vf.uni-lj.si

**Summary:** A 10-year-old Samoyed with unilateral right cryptorchidism was referred to the Clinic for Small Animal Medicine and Surgery, Veterinary Faculty of Ljubljana University, due to a symmetrical, noninflammatory, mildly pruritic alopecia of 6-month duration. It had a history of diarrhoea, which responded to cyclosporine treatment. Lymphoma and testicular neoplasia were suspected based on ultrasonography and fine needle aspiration cytology. The dog was castrated and splenic and gastric lymph node biopsies were obtained. Histopathology revealed three different tumours: Sertoli cell tumour and seminoma were present in the right inguinal testicle, and B cell lymphoma was present in the spleen and lymph node. After two months when the peripheral lymph nodes were considerably enlarged and the owners declined chemotherapy, they were advised to start corticosteroid treatment. Three months after the castration, the hair coat looked normal. Four months after the castration, the dog was euthanized at the owner's request by the referring veterinarian due to a lymphoma related disease.

**Key words:** unilateral cryptorchidism; testicular neoplasms – surgery – chemotherapy; ultrasonography; cytodiagnosis; biopsy, fine-needle; dogs

## Introduction

Noninflammatory alopecias are quite common in dogs and can be congenital, hereditary or acquired. The latter can result from endocrinopathies (e.g. hyperadrenocorticism, hypothyroidism, sexual imbalance), telogen or anagen effluvium, metabolic imbalance (disruptions in protein or fatty acid metabolism) and idiopathic disturbances of the hair growth cycle (1).

Testicular tumours comprise 4-7% of all tumours in male dogs (2). They arise either from Sertoli cells (i.e. Sertoli cell tumours), germinal epithelium (i.e. seminoma) or interstitial cells (i.e. Leydig cell tumours) (3), all of which occur with equal prevalence (2, 4). Other primary tumours may also arise, but are extremely rare. Dogs with non-descended testicles have an increased risk of up to 13.6 fold to develop a testicular tumour compared to dogs with descended testicles (5). Since right-sided cryptorchidism is

more common, the right testis is more often affected (4). Multiple tumours within one testis occur in up to 46% of the cases and are also described in macroscopically normal testes (6). Hyperestrogenism occurs in 25-50% of dogs with Sertoli cell tumour and less frequently with seminomas, and may lead to signs of feminization (7). It can lead to bone marrow hypoplasia with consecutive thrombocytopenia, non-regenerative anaemia and granulocytopenia. Serum testosterone and progesterone concentrations may be increased, but usually testosterone concentration is low to undetectable (5). As these changes may be subtle and are not specific and sensitive changes, castration with a histopathological examination is still the diagnostic and therapeutic approach of choice (8). An alternative and perhaps more sensitive marker for canine Sertoli cell tumour is increased serum inhibin concentration (9).

Malignant lymphoma is the most common canine haemato-lymphatic tumour, most commonly classified based on anatomic location, clinical stage or immunophenotype. The multicentric form with

peripheral lymphadenopathy is the most common form, followed by the alimentary, mediastinal and extranodal (kidneys, skin, and brain) forms (10-12). The aetiology of canine lymphoma is unknown, but hypotheses include retroviral infection, exposure to herbicides and magnetic field, chromosomal abnormalities and immune dysfunction (13).

### Case report

A 10-year-old, intact male Samoyed with unilateral right cryptorchidism was presented at the Clinic for Small Animal Medicine and Surgery, Veterinary Faculty of Ljubljana University for an investigation of a symmetric alopecia of 6-month duration. It was currently vaccinated and dewormed and has lately gained weight although water and food intake seemed unchanged. The dog had a history of large bowel diarrhoea eight months before the presentation at our clinic, which was treated with metronidazole and spiramycin for 1 month without obvious improvement. This was followed by cyclosporine treatment for 40 days, which led to complete recovery, however, dermatologic abnormalities were observed. Initially, alopecia was present only in the neck region, and later symmetrically spread to the axillae, abdomen as well as the inguinal and tail areas. Mild pruritus was observed during the disease course. An abdominal ultrasound performed by the referring veterinarian did not reveal abnormalities. Serum hormone concentrations were determined and included free T4 (1.1 ng/dl, reference range 0.6 – 3.7), cTSH (0.13 ng/ml reference range <0.40), testosterone (0.0667 ng/ml, reference range 0.3 – 5.8)<sup>1</sup> and 17- $\beta$ -oestradiol (87.9 pg/ml, reference range <15 (9)).

Physical examination revealed, in addition to the alopecia (Figure 1 and 2) and cryptorchidism, a bilateral purulent ocular discharge. Skin scrapings for *Sarcoptes* and *Demodex* were negative. Cytology of the skin was positive for *Malassezia*. Faecal examination was positive for *Giardia*. CBC (Bayer Technicon H\*1, Bayer-Technicon, Germany) and serum biochemistry analysis (biochemical analyzer Technicon RA-XT, Bayer Technicon, Germany) including glucose, urea, creatinine, calcium, phosphorous, alkaline phosphatase, alanine-aminotransferase and cholesterol) were unremarkable, with exclusion of a mild normocytic normochromic anaemia (haematocrit 0.35 l/l, reference range 0.37-0.55 (14)).



Figure 1: Skin at the beginning of the therapy

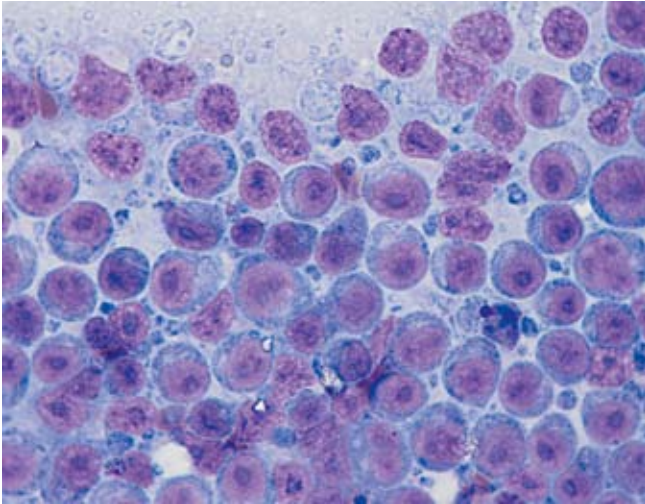


Figure 2: Skin at the beginning of the therapy



Figure 3: Hair re-growth after 3 weeks of the hypoallergenic diet and essential fatty acid supplementation

<sup>1</sup> References values of the Vet Med Lab laboratory in Trieste, Italy.



**Figure 4:** Lymphoblasts in the spleen ( $\times 1000$ )



**Figure 5:** Dog three months after surgery



**Figure 6:** Dog three months after surgery

Treatment was initiated using anthelmintics praziquantel 5 mg/kg and fenbendazole 50 mg/kg (Zantel, CAM) PO q24h for 5 days, ketoconazole 14 mg/kg PO q24h (Oronazol, Krka) and topical ophthalmic gentamicin (Garamycin, Krka) ointment bilaterally q12h. After one month, there was no improvement and total serum T4 and TSH analyses were repeated (30.6 nmol/l and 0.47 ng/ml, respectively). A hypoallergenic diet (Eucanuba FP dermatosis and home-made diet (horse meat, rabbit meat, fish) and an oral omega 3 and omega-6 fatty acids preparation (Dermanorm, Vetoquinol, 2 capsules/dog PO q24h) were prescribed for three weeks, when an improvement in general health and growth of hair was seen (Figure 3). The same therapy was continued for 2 additional months. At that time, the dog was rechecked due to deterioration in the condition of the skin. It had pruritus, alopecia, erythema, ventral hyper- and hypopigmentation and dorsal hypotrichia, seborrhoea and crusts. An abdominal ultrasound revealed nonhomogeneous splenomegally and nonhomogenous masses: two cranio-dorsal to right and two cranio-dorsal to the left kidney. These masses were interpreted as lymph nodes by the radiologist. The testicle present within the scrotum had a hypoechoic discrete nodule of 8 mm diameter. A nonhomogeneous 3 $\times$ 4 cm mass in the right inguinal area was observed. Other abdominal organs were ultrasonographically normal. A fine needle aspiration of the inguinal mass (probably testicle) and the spleen was performed. The first was non-diagnostic while the latter revealed a population of large lymphoid cells with large nuclei, scant deeply blue cytoplasm, some of which had a pronounced perinuclear reactive zone and 1-3 prominent nucleoli of different size and shape (Figure 4).

According to these findings we planned to castrate the dog and take samples of the spleen, enlarged lymph nodes and skin.

A preoperative CBC showed a mild normochromic normocytic nonregenerative anaemia (RBC  $5.02 \times 10^{12}/l$ , reference range  $5.5-8.5 \times 10^{12}/l$ ; haemoglobin 115 g/l, reference range 120-180 g/l; haematocrit 0.32 l/l, reference range 0.37-0.55 l/l) and mild thrombocytopenia (platelets  $141 \times 10^9/l$ , reference range  $200-500 \times 10^9/l$ )(14). Serum biochemistry was unremarkable. Under general anaesthesia, the dog was castrated and the right inguinal mass was excised. This mass was 5 cm in diameter and had an appearance of a testicle. Then, a midline exploratory laparotomy was performed. The spleen as well as the gastric lymph node were enlarged and were removed. The mesenteric lymph nodes were also



mildly enlarged (5 cm diameter). No other abnormalities were observed. The biopsy of the skin was also performed.

Histopathology of the right inguinal mass revealed the presence of seminoma and Sertoli cell tumour in the right testis. The left testis was not examined. Lymphoma was present in the spleen and gastric lymph node. Subchronic dermatitis with a mixed cell (predominantly eosinophils) population was present in the skin biopsies. Immunohistochemistry examination of splenic and gastric lymph node biopsies with CD3 and CD79 antibodies (DAKO REALTM, EnVision Detection System) confirmed a B cell lymphoma.

Postoperatively a bloody-purulent urine was noticed. Dipstick and urine sediment analyses confirmed a diagnosis of purulent cystitis. Culture and sensitivity were not performed. Amoxicillin with clavulanic acid (Synulox, Pfizer) 20 mg/kg PO q12h for 14 days was prescribed.

The dog was rechecked one month later, seemed healthy, gained two kilograms of body weight and its hair-coat showed signs of improvement. The palpable peripheral lymph nodes were unchanged and CBC was unremarkable (mild anaemia, haematocrit 0.36 l/l). A month later, the dog had mild mandibular and praescapular lymphadenopathy. CBC was normal, with the exception of a mild anaemia (haematocrit 0.34 l/l). The owners were offered a multidrug chemotherapy, but declined and were thus advised to start corticosteroid treatment - methylprednisolone (Medrol, Pfizer; 2 mg/kg/day PO until relaps). An additional month later, three months after the operation, Lord's hair looked normal again (Figure 5, 6). A month later, after two months of methylprednisolone treatment, the dog deteriorated, was depressed and anorexic and was euthanized at the owner's request by the referring veterinarian.

## Discussion

This is an unusual case with 3 different neoplasias occurring concurrently in a single dog. Multiple tumours occurring in the brains (15, 16) or in the mammary tissue (17) were reported, but authors are not aware of any report about concurrent multiple tumour occurrence in different tissues.

Preoperatively seen mild nonregenerative anaemia with the haematocrit of 0.32 l/l and thrombocytopaenia could have developed secondarily to lymphoma (18,19), but haematocrit of 0.35 l/l was also seen 3 months earlier, when lymphoma was not

diagnosed yet. Such a long course of the disease is unusual for lymphoma (18).

In addition to symmetrical truncal alopecia other male patients, but not the present case, may also present other signs of feminization (8). The subsequent improvement of the skin after surgery in contrast to the partial response to previous therapy supports the assumption that these changes were secondary to hyperoestrogenism due to Sertoli cell tumour. Plasma oestradiol concentrations in male dogs should be less than 15 pg/ml. In dogs with Sertoli cell tumours, oestradiol concentrations are usually 10 to 150 pg/ml, as was in the present case (87,9 pg/ml) (9).

Sertoli cell tumours metastasize in 2-14% of cases; seminomas do so even less frequently. Most commonly they metastasize to the sublumbar (3), inguinal and iliac lymph nodes, lungs, liver, spleen, kidneys, pancreas (9), subcutis, brain and eyes (20). The above mentioned lymph nodes were not enlarged in our case, as observed upon the exploratory laparotomy. However, gastric and mesenteric lymphadenopathy was observed. While the gastric lymph node presented lymphoma and had no evidence of a testicular cancer metastasis, the mesenteric lymph nodes were not examined histologically. Although we cannot rule out that they were enlarged due to metastasis of the testicular tumours, it is more likely that lymphoma was the cause. A postoperative serum oestrogen could have aided in determination of Sertoli cell tumour metastasis. Retrospectively, considering the chronic diarrhoea, unresponsive to antibiotics, but responsive to cyclosporine, the mesenteric and gastric lymphadenopathy, and diagnosis of lymphoma in the latter as well as in the spleen, we believe this was a case of alimentary lymphoma. It is presumed that they are most likely B-cell in origin (18), but newer studies are conflicting (21). However, it could be also a stage IVa of the multicentric form (18), which is perhaps more likely regarding subsequent development of generalised lymphadenopathy.

The chronic diarrhoea, mentioned in the history, could develop due to other causes such as lymphocytic-plasmatic enteritis (LPE) or giardiasis, as the dog was positive for the latter. Giardiasis seems to be a less likely cause for the diarrhoea as the dog failed to respond to a prolonged metronidazole and spiramycin treatment, and did respond to cyclosporine (22, 23). It has been suggested, that LPE may be a prelymphomatous change in the gastrointestinal tract (18). Cyclosporine treatment has been reported to induce lymphoma or other tumours oc-

asionally (18, 24, 25). The drug hinders lymphocyte T activity and recognition of neoplastically changed cells (26), which, presumably, can consequently induce additional concurrent neoplastic disease.

Dogs with lymphoma are known to be less immunocompetent because of the incompetence of neoplastic lymphocytes/lymphoblasts (11, 18) or fewer total lymphocytes, especially T cells (27). This could have led to the observed postoperative cystitis.

Lymphoma is usually treated with combination chemotherapy protocol (28-30), rather than corticosteroids a single agent, however, the latter was attempted due to the owner's decline of the former. Corticosteroid effects are usually short-lived in canine lymphoma, and relapses are extremely common (18), and thus it is not surprising that the present dog was represented shortly later with signs of systemic disease, most probably a relapse of the lymphoma.

## Conclusion

We presented a case of multiple neoplasia in a dog including B cell lymphoma, Sertoli cell tumour and seminoma. The dog had a chronic dermatopathy, probably due to hyperoestrogenism secondary to the Sertoli cell tumour. The latter is supported by recovery of the skin following castration. The lymphoma was probably alimentary, although a multicentric lymphoma cannot be ruled out. The cause of lymphoma is unknown, but it may have been associated with a previous cyclosporine therapy.

## Acknowledgement

We would like to thank Prof. Dr. Polona Junes for her contribution to the pathohistologic diagnosis and Assist. Prof. Dr. Aleksandra Domanjko Petrič for the ultrasonographic examination.

## References

1. Heripret D. Alopecia. In: Ettinger SJ, Feldman EC, eds. Textbook of veterinary internal medicine. 6<sup>th</sup> ed. St. Louis: Elsevier Saunders, 2005: 34-7.
2. Spugnini EP, Bartolazzi A, Ruslander D. Seminoma with cutaneous metastases in a dog. *J Am Anim Hosp Assoc* 2000; 36 (3): 253-6.
3. Cooley DM, Waters DJ. Tumors of the male reproductive system. In: Withrow SJ, MacEwen EG, eds. Small animal clinical oncology. 3<sup>rd</sup> ed. Philadelphia: Saunders, 2001: 478-89.
4. Kim O, Kim KS. Seminoma with hyperestrogenemia in a Yorkshire terrier. *J Vet Med Sci* 2005; 67 (1): 121-3.
5. Morrison WB. Cancer of the reproductive tract. In: Morrison WB, ed. Cancer in dogs and cats: medical and surgical management. 2<sup>nd</sup> ed. Jackson: Teton NewMedia, 2002: 555-64.
6. Peters MAJ, de Rooij DG, Teerds KJ, van der Gaag I, van Sluijs FJ. Spermatogenesis and testicular tumours in ageing dogs. *J Reprod Fertil* 2000; 120 (2): 443-52.
7. Chun R, Garrett. Urogenital and mammary gland tumors. In: Ettinger SJ, Feldman EC, eds. Textbook of veterinary internal medicine. 6<sup>th</sup> ed. St. Louis: Elsevier Saunders, 2005: 784-9.
8. Scott DW, Miller WH, Griffin CE. Small animal dermatology. 6<sup>th</sup> ed. Philadelphia: W.B. Saunders, 2001: 780-885.
9. Feldman EC, Nelson RW. Canine and feline endocrinology and reproduction. 3<sup>rd</sup> ed. St. Louis: Saunders, 2003: 961-76.
10. Tozon N, Samardžija P, Prijič S, Fazarinc G. Canine lymphoma: cytologic study and response to therapy. *Slov Vet Res* 2006; 43 (3): 127-33.
11. Vonderhaar MA, Morrison WB. Lymphosarcoma. In: Morrison WB, ed. Cancer in dogs and cats: medical and surgical management. 2<sup>nd</sup> ed. Jackson: Teton NewMedia, 2002: 641-70.
12. Lowe AD. Alimentary lymphosarcoma in a 4-year-old Labrador retriever. *Can Vet J* 2004; 45 (7): 610-2.
13. Fan TM. Lymphoma updates. *Vet Clin North Am Small Anim Pract* 2003; 33 (3): 455-71.
14. Bush BM. Interpretation of laboratory results for small animal clinicians. Oxford: Blackwell Science, 1991: 35-131.
15. Stacy BA, Stevenson TL, Lipsitz D, Higgins RJ. Simultaneously occurring oligodendroglioma and meningioma in a dog. *J Vet Int Med* 2003; 17 (3): 357-9.
16. Alves A, Prada J, Almeida JM, et al. Primary and secondary tumours occurring simultaneously in the brain of a dog. *J Small Anim Pract* 2006; 47 (10): 607-10.
17. Ferguson HR. Canine mammary gland tumors. *Vet Clin North Am Small Anim Pract*. 1985; 15 (3): 501-11.
18. Vail DM, MacEwen EG, Young KM. Canine lymphoma and lymphoid leukemias. In: Withrow SJ, MacEwen EG, eds. Small animal clinical oncology. 3<sup>rd</sup> ed. Philadelphia: Saunders, 2001: 558-90.
19. Gaschen FP, Teske E. Paraneoplastic syndrome. In: Ettinger SJ, Feldman EC, eds. Textbook of veterinary internal medicine. 6<sup>th</sup> ed. St. Louis: Elsevier Saunders, 2005: 789-95.
20. Dhaliwal RS, Kitchell BE, Knight BL, Schmidt BR. Treatment of aggressive testicular tumors in four dogs. *J Am Anim Hosp Assoc* 1999; 35 (4): 311-8.
21. Coyle KA, Steinberg H. Characterization of lymphocytes in canine gastrointestinal lymphoma. *Vet Pathol* 2004; 41 (2): 141-6.
22. Leib MS, Dalton MN, King SE, Zajac AM. Endoscop-

ic aspiration of intestinal contents in dogs and cats: 394 cases. *J Vet Int Med* 1999; 13 (3): 191-3.

23. Lappin MR. Giardia infections. In: World congress 2006 WSAVA/FECAVA/CSAVA. Proceedings of the 31<sup>st</sup> World Small Animal Veterinary Association. Prague: WSAVA, 2006: 50-1.

24. Blackwood L, German AJ, Stell AJ, O'Neill T. Multicentric lymphoma in a dog after cyclosporine therapy. *J Small Anim Pract* 2004; 45 (5): 259-62.

25. Callan MB, Preziosi D, Mauldin E. Multiple papillomavirus-associated epidermal hamartomas and squamous cell carcinomas in situ in a dog following chronic treatment with prednisone and cyclosporine. *Vet Dermatol* 2005; 16 (5): 338-45.

26. Knapp DW: Immunotherapy and biologic response modifiers. In: Morrison WB, ed. *Cancer in dogs and cats: medical and surgical management*. 2<sup>nd</sup> ed. Jackson: Teton NewMedia, 2002: 425-39.

27. Walter CU, Biller BJ, Lana SE, Bachand AM, Dow SW. Effects of chemotherapy on immune responses in dogs with cancer. *J Vet Int Med* 2006; 20 (2): 342-7.

28. Chun R, Garrett LD, Vail DM. Evaluation of a high-dose chemotherapy protocol with no maintenance therapy for dogs with lymphoma. *J Vet Intern Med* 2000; 14 (2): 120-4.

29. Garrett LD, Thamm DH, Chun R, Dudley R, Vail DM. Evaluation of a 6-month chemotherapy protocol with no maintenance therapy for dogs with lymphoma. *J Vet Intern Med* 2002; 16 (6): 704-9.

30. Simon D, Nolte I, Eberle N, Abbrederis N, Killich M, Hirschberger J. Treatment of dogs with lymphoma using a 12-week maintenance-free combination chemotherapy protocol. *J Vet Intern Med* 2006; 20 (4): 948-54.

## GENERALIZIRANA SIMETRIČNA ALOPECIJA IN HIPERESTROGENIZEM PRI SAMOJEDU Z LIMFOMOM, SEMINOMOM IN TUMORJEM SERTOLIJEVIH CELIC

T. Plavec, N. Tozon, T. Kotnik

**Povzetek:** 10 let star samojed, desnostranski kriptorhid, je bil napoten na Kliniko za kirurgijo in male živali Veterinarske fakultete zaradi šest mesecev trajajoče nevnetne alopecije. V anamnezi živali so lastniki navajali drisko, ki je bila uspešno zdravljena s ciklosporinom. Na podlagi ultrazvočne preiskave in citološke preiskave tankoigelnega punktata smo postavili sum na limfom in neoplazijo mod. Psa smo kastrirali ter odvzeli biopsat vranice in bezgavke. Patohistološka preiskava je potrdila tumor sertolijevih celic in seminom desnega moda ter B celični limfom vranice in bezgavk. Dva meseca po operativnem posegu smo opazili povečane periferne bezgavke, vendar se lastniki niso odločili za citostatično zdravljenje. Svetovali smo jim začetek zdravljenja s kortikosteroidi. Tri mesece po kirurškem posegu je bila odlakanost psa spet normalna, evtanaziran je bil štiri mesece po kastraciji zaradi obolenja, povezanega z limfomom.

**Ključne besede:** enostranski kriptorhidizem; testis, novotvorbe – kirurgija – kemoterapija; ultrazvok; citodiagnostika; biopsija, tankoigelna; psi