RESEARCH ARTICLE Indian Journal of Animal Research, Volume 55 Issue 11: 1352-1355 (November 2021)

Diagnosis of Tvt with Cell Cytology and Efficacy of Treatment with Vincristine Sulfate in Non- Descriptive Indian Canine Breeds

Keshav Kumar, Ajeet Kumar Jha, Kalyani Ray, Avnish Kumar Gautam[1], Dharmendra Singh[1] 10.18805/IJAR.B-4175

ABSTRACT

Background: Canine transmissible venereal tumor (TVT) is a commonly occurring contagious reproductive tumor of dogs affecting both sexes.

Methods: Six female mongrel dogs were brought to Veterinary Clinical Complex, WBUAFS, Belgachia, Kolkata with the history of inappetence, mass swelling on genitalia and blood tinged urine with dysuria for the past two weeks. Following Physical examination, protruded mass was noticed on external genital region. Impression smear was collected from protruded mass for confirmation of vacuole cell. Chemotherapy was applied with Vincristine @ 0.025 mg/kg b.wt; slow IV once a week for 4 weeks along with supportive treatment.

Result: After the end of the 4th week of Chemotherapy animals were showed uneventful recovery.

Key words: Canine, Chemotherapy, Protruded mass, Transmissible venereal tumor, Vincristine.

INTRODUCTION

Canine Transmissible Venereal Tumor (CTVT) is usually a sexually transmitted neoplasm of the external genitalia of dogs which is transmitted between dogs by the allogenic transfer of living cancer cells (Strakova and Murchison). TVTs are usually located on genital, oral, or nasal mucosa (Spugnini *et al.,* 2008). Initially the tumor is small, subsequently progressing to a large, ulcerated and contaminated mass. The lesions are friable, hyperemic, hemorrhagic, multilobular, cauliflower like masses (Johnson, 1994).

A large number of reports have been produced in India (Chauhan *et al.,* 1991; Das *et al.,* 1991; Tiwari *et al.,* 1991; Hoque, 1995; Maiti *et al.,* 1995; Jain *et al.,* 2002; Ray *et al.*, 2019). In India TVT is known to be the most frequently reported tumor in dogs ranging from 23-43% of the total number of tumors in canine population (Gandotra *et al.,* 1993; Chaudhary and Rao, 1982).

Definitive diagnosis is based on physical examination and cytological findings typical of TVT in exfoliated cells obtained by swabs, fine needle aspirations or imprints of the tumors (Richardson, 1981; Moulton, 1978,)

Several treatments including surgery, radiotherapy, immunotherapy, biotherapy and chemotherapy have been applied for TVT. Vincristine sulfate is an alkaloid obtained from Vinca rosea that blocks mitosis by arresting cells in the metaphase (Said *et al.,* 2009).

MATERIALS AND METHODS

This study was conducted at Department of Veterinary Gynaecology and Obstetrics, Faculty of Veterinary and Animal Sciences, West Bengal University of Animal and Fisherry Sciences, 37, K.B. Sarani, Kolkata, West Bengal, India. A total of 6 Non- discriptive Indian canine breeds were selected on the basis of owners history (Uterine bleeding since last 2 months, anorexia, sweling of vulva, protruded mass from vulva). A clean, grease free slide was taken and a smeared at protruded mass of vulva. This slide was then air dried and fixed with 10% ethanol for 10 Minutes then stained with Giemsa stain. After proper staining the slide was observed at 100x objective of Binocular Microscope with oil immersion. Vacuole cells were observed for confirmation of TVT. Furthermore, Haematological test were done at Department of Biochemistry, WBUAFS, Kolkata-700 037 to observe platelets and haemoglobin % of all the 6 dogs.

All the dogs were treated with vincristine sulfate @ 0.025 mg/kg body weight diluted with normal saline at weekly interval. Normal saline according to Company protocol and Thrombofit (A herbal product) @ 10 ml BID was advised to

Department of Veterinary Gynaecology and Obstetrics, West Bengal University of Animal and Fishery Sciences, Belgachia, Kolkata-700 037, West Bengal, India.

[1]Department of Veterinary Anatomy and Histology, West Bengal University of Animal and Fishery Sciences, Belgachia, Kolkata-700 037, West Bengal, India.

Corresponding Author: Ajeet Kumar Jha, Department of Veterinary Gynaecology and Obstetrics, West Bengal University of Animal and Fishery Sciences, Belgachia, Kolkata-700 037, West Bengal, India. Email: ajitjha2003@gmail.com

How to cite this article: Kumar, K., Jha, A.K., Ray, K., Gautam, A.K. and Singh, D. (2021). Diagnosis of Tvt with Cell Cytology and Efficacy of Treatment with Vincristine Sulfate in Non- Descriptive Indian Canine Breeds. Indian Journal of Animal Research. 55(11): 1352-1355. DOI: 10.18805/IJAR.B-4175.

Submitted: 13-05-2020 **Accepted:** 05-10-2020 **Online:** 21-12-2020

prevent anaemia and thrombocytopenia. Every week haematology and vaginal cytology were done and recorded for any improvement.

RESULTS AND DISCUSSION

TVT is the most prevalent tumor of the genitalia of the either sex of dog, but cutaneous form of TVT is uncommon. The etiology of TVT is not completely known, but it can transmit from affected dogs by coitus and/or by natural contact (Gurel *et al.,* 2002). In the present case the cutaneous mass appears to be due to metastasis and spreading of tumor cells (Fig 1A). TVTs are immunogenic tumors and the immune system of the host plays a major role in inhibiting tumor growth and metastasis (Cohen, 1985). In present study, we observed that the tumour masses on the vagina and vulva which was irregular in shape with cauliflower like appearance and reddish in colour. The consistency of the mass was soft and had a tendency to bleed (Fig 1). The dramatic reduction in gross tumor size was observed by 7 day after the first dose of Vincristine and full-response by 28 day (Fig 1B).

Martins *et al.,* (2008) also did similar treatment and found gradual tumor response although the response was noticeable and significant at the beginning of treatment which regressed gradually later. Valladao *et al.,* (2010) administered similar treatment with Inj. Vincristine @ 0.025 mg/kg i/v at weekly intervals for five weeks with which complete remission was achieved in 75 dogs with a median of 5 Injections. Said *et. al.,* (2009) also treated similarly and found that the treatment was 90% effective. Khan *et al*. (2009) enumerated that Inj. Vincristine gave promising result in respect to granulomatous growth from penis in male, vaginal canal in females which disappeared in 21 days. On cytology, TVT cells showed a predominance of round cells with abundant pale cytoplasm and distinct, punctate cytoplasmic vacuoles. The cytoplasm of the tumour cells was greyish in colour when stained with Giemsa stains (Fig 2). Typical cytological analysis showed slightly polyhedral cells. The nature of cellularity was high with homogenous round individual cells arranged in a sheet-like pattern. A great variation in the cellular (anisocytosis) and nuclear morphology (anisokaryosis) was observed. The nucleus of the tumour cells was round to oval in shape and centrally placed. The most prominent cytological feature of TVT is the presence of distinct, clear, cytoplasmic vacuoles often referred to as punctate vacuoles with a delineated outline. The nuclear to cytoplasmic ratio of tumour cells was large (Fig 2).

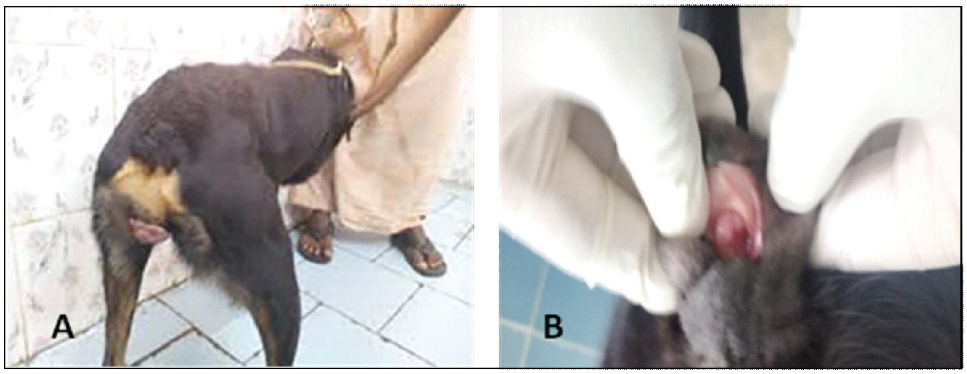

Fig 1: Protrusion of vaginal tumor and its correction using Vincristine sulfate.
A. Protrusion of tumor mass at the time of admission to the Hospital. B. Regression of mass after treatment.

Table 1: Haematological parameter (Hb, WBC, Neutrophil, Eosinophil, Lymphocyte, Monocyte, Basophil, Platelet, BUN, Creatinine, SGOT and SGPT) before and after treatment (N=6).

Parameter	Mean		P value
	Before treatment	After treatment	
Haemoglobin (gm/dl)	14.10±0.28	13.50±0.29	0.001
WBC (cumm)	10266.66±176.38	11533.33±185.59	0.001
Neutrophil (%)	54.66±2.90	57.33±2.91	0.667
Eosinophil (%)	6.33±0.33	2.67±0.33	0.008
Lymphocyte (%)	33.33±2.67	33.33±1.33	1
Monocyte (%)	2.00±0.57	2.33±0.33	0.423
Basophil (%)	0	0	
Platelet count (cumm)	225.00±14.43	295.00±14.42	
BUN (mg/dL)	7.17±0.20	11.63±0.21	0
Creatinine (mg/dL)	1.1	1.1	
SGOT (IU/L)	43.00±1.73	31.67±1.45	0.001
SGPT (IU/L)	47.00±0.57	37.00±0.58	

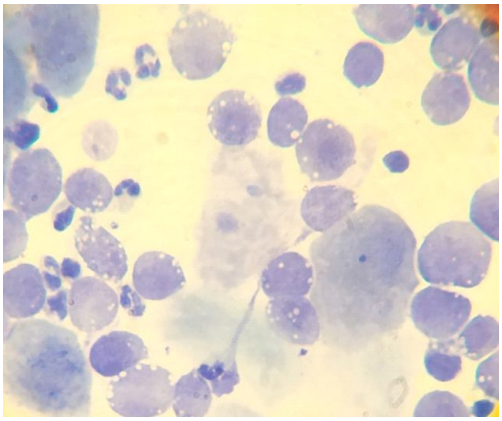

Fig 2: TVT cytology showing cytoplasm with punctate vacuoles, hyperchromatism and multiple nucleoli. Giemsa stain, (100X).

Similar cytological features were reported by many researchers (Alleman and Bain, 2000; Duncan and Prasse, 1979; Fan *et al.,* 2001; Meinkoth and Cowell, 2002). Ulcar Igor *et al.,* (2012) reported cytological evaluation of tumors of genital tract in dogs are essential for diagnosis and the cytological features of the genital tumors have typical criteria of TVT. TVT dogs of the present study showed significant ($P<0.05$) value of Hb, WBC, Eosinophil, BUN and SGOT from day 0 to day 28 during before and after treatment. We observed 14.10±0.28 and 13.50±0.29 gm/dl of Hb before and after treatment respectively. Similarly, WBC count before and after treatment were 10266.66±176.38 and 11533.33±185.59 respectively. The eosinophil value recorded were 6.33±0.33 and 2.67±0.33 respectively before and after treatment. BUN value we found were 7.17±0.20 and 11.63±0.21 whereas, SGOT were 43.00±1.73 and 31.67±1.45 before and after treatment respectively (Table 1).

The hematological findings were in contrast to Das *et al.* (1991). Tella *et al.* (2004), Said *et al.* (2009) and Nak *et al.* (2005) opned that decrease of Hb, PCV and TEC may be attributed to the tumoural bleeding and myelosuppression induced by chemotherapeutic drug. Leucocytosis which was observed in the present study was similarly observed by Girmabirhan *et al.,* (2015) and Behera *et al.,* (2012). Results indicates that all the TVT affected dogs showed significant increase of BUN and no change in serum creatinine, incompatibility. Albanese *et al*. (2006), Gandhimathi *et al*. (2011) and Girmabirhan *et al*. (2015) reported that normal physiological range to mild increase of BUN and serum creatinine was seen in TVT affected dogs. Some researcher also shows that SGPT and SGOT showed significant increase ($P<0.01$) after treatment when compared to day 0 which are at par of our result (Behera *et al*., 2012: Girmabirhan *et al.,* 2015; Albanese *et al.,* 2006 and Gandhimathi *et al.,* 2011). Hematology was indicative of marked microcytosis and thrombocytopenia owing to immunosuppressive nature of the disease, leading to secondary bacterial invasion.

Treatment of dog with TVT

Chemotherapy was initiated with Vincristine sulfate @ 0.025mg/kg bwt IV once a week for 4 weeks. Animal showed clinical improvement from 2nd week of chemotherapy and complete uneventful recovery was noticed after 5th weeks of Vincristine sulfate chemotherapy. Vincristine sulfate acts by binding to tubulin dimer which is necessary for mitosis of spindle fibers, contributing to cellular division arrested in metaphase stage (Coppoc, 2009).

CONCLUSION

Vincristine sulfate dose rate of 0.025 mg/kg bwt IV once a week for 4 weeks showed complete recovery from generalized cutaneous and genital form of transmissible venereal tumor (TVT) in a mongrel dog.

REFERENCES

Albanese, F., Salerni, F.L., Giordano, S., Marconato, L. (2006). Extragenital transmissible venereal tumour associated with circulating neoplastic cells in an immunologically compromised dog. Vet. Comp. Oncol. 4: 57-62.

Alleman, A.R., Bain, P.J. (2000). Diagnosing neoplasia: The cytologic criteria for malignancy. Vet. Med. 95: 204-223.

Behera, S.K., Kurade, N.P., Shongsir, W.M., Durga, P.D., Krishna, K.M., Ranjan, K.M. (2012). linico- pathological findings in a case of canine cutaneous metastatic transmissible venereal tumour. Vet. Arhiv. 82: 401-410.

Chaudhary, C., Rao, M.R.K. (1982). Certain canine neoplasms encountered in Andhra Pradesh. Indian Vet. J. 59: 100-102.

Chauhan, H.V.S., Ghosh, R.C., Mascrenhas, A.R., Tiwari, S.K. (1991). Canine venereal sarcoma and its surgical management. Indian Vet. J. 68: 1078-79.

Cohen, D. (1985). The canine transmissible venereal tumor: A unique result of tumor progression. Adv. Cancer Res. 43: 75-112.

Coppoc, G.L. (2009). Chemotherapy of Neoplastic Diseases. In: Veterinary Pharmacology and Therapeutics. 9th edi. [J.E. Riviere and M.G. Papich (eds)] Ames: Willey-Balckwell, 1205-1231.

Das, A.K., Das, U., Das, B.B. (1991). Clinical report on the efficacy of chemotherapy in canine transmissible venereal sarcoma. Indian Vet. J. 68: 249-252.

Duncan, J.R., Prasse, K.W. (1979). Cytology of canine cutaneous round cell tumours. Vet. Pathol. 16: 673-679.

Fan, T.M., Kitchell, B.E., Daliwahl, R.S. (2001). Mast cell neoplasia in dogs. Veterinary Medicine. 96: 919-929.

Gandhimathi, D., Gurupriya, G., Arivucelvan, P., Sridevi, P., Joseph, C. (2011). Effect of doxorubicine therapy on transmissible venereal tumour in dogs. Indian Journal of Animal Reproduction. 32: 2488-2494.

Gandotra, V.K., Chauhan, F.S., Sharma, RD. (1993). Occurrence of canine transmissible venereal tumor and evaluation of two treatments. Indian Vet. J. 70: 854-857.

Girmabirhan, M.C. (2015). A reveiw on canine transmissible venereal tumour from morphologic to biochemical and molecular diagnosis. Academic Journal of Animal Diseases. 4: 185-195.

Gurel, A., Kuscu, B., Gulanber, E.G., Run, S.S. (2002). Transmissible venereal tumors detected in the extra genital organs of dogs. Isr. J. Vet. Med. 57: 23-26.

Hoque, M. (1995). Different modes of therapy for canine transmissible venereal tumor. Veterinarian. 19: 1-2.

Jain, A., Tiwari, R.P., Tiwari, S.K., Gupta, N., Awasthi, M.K. (2002). Histopathological studies in canine transmissible venereal tumor. Indian Journal of Animal Reproduction. 23: 60-62.

Johnson, C.A. (1994). Infecções Genitais e Tumor Venéreo Transmissível. In: Fundamentos de Medicina Interna de Pequenos Animais. [Nelson, R.W., Couto, C.G., editors]. Rio de Janeiro, Brazil: Guanabara Koogan. 522-525.

Khan, L.A., Khante, G.S., Raut, B.M., Bodkhe, A.M., Chavan, M.S., Pagrut, N.S. (2009). Incidence of Venereal Granuloma and its Medicinal treatment in stray Dogs of Nagpur City. Vet. World. 2: 13-14.

Maiti, S.K., Roy, S., Ali, S.L., Ghosh, R.C. (1995). Therapeutic management of transmissible venereal tumor with vincristine in dog. A case report. Indian Vet. J.72: 614-615.

Martins AMCRPF, Vasques-Peyser, A., Torres, L.N., Matera, J.M., Dagli, M.L.Z., Guerra, J.L. (2008). Retrospective systematic study and quantitative analysis of cellular proliferation and apoptosis in normal hyperplastic and neoplastic perianal glands in dogs. Vet. Comp. Oncol. 6: 71-79.

Meinkoth, J.H., Cowell, R.L. (2002). Sample collection and preparation in cytology: Increasing diagnostic yield and recognition of basic cell types and criteria of malignancy. Veterinary Clinics of North America: Small Animal Practice. 32: 1187-1235.

Moulton, J.E. (1978). Tumor of Genital Systems. In: Tumors in Domestic Animals. [Moulton, J.E., ed]. 2. ed. California: University of California. 326-330.

Nak, D., Nak, Y., Cangul, L., Tuna, B. (2005). A clinicopathological study on the effect of vincristine on transmissible venereal tumour in dogs. J. Vet. Med. 52: 366-370.

Ray, K., Jha, A.K., Biswas, P., Basu, S. (2019). Study on physico-morphological seminal characters of German Shepherd canine breed. Indian Journal of Animal Heaith. 58: 95-100.

Richardson, R.C. (1981). Canine Transmissible Venereal Tumor. Compendium on Continuing Education for the Practising Veterinarian -North American Edition. 3: 951-956.

Said, R.A., Silva, L.F., Albuquerque, AROL, Sousa-Neta, E.M., Lavinsky, M.O. (2009). Efficacy and Side Effects of Vincristine Sulphate Treatment on Canine Transmissible Venereal Tumour- 438. In: Proceedings of the 34th World Small Animal Veterinary Congress. São Paulo, Brazil. WSAVA.

Spugnini, E.P., Dotsinsky, L., Mudrov, N., Citro, G., D´Avino, A., Baldi, A. (2008). Biphasic pulses enhance bleomycin efficacy in a spontaneous canine genital tumor model of chemoresistance: Sticker sarcoma. Exp. Clin. Cancer Res. 27: 58.

Strakova, A., Murchison, E. (2014). The changing global distribution and prevalence of canine transmissible venereal tumour. BMC Vet. Res. 10: 1-10.

Tella, M.A., Ajala, O.O., Taiwo, V.O. (2004). Complete response of transmissible venereal tumour (TVT) in Nigerian mongrel dogs with vincristine sulfate chemotherapy. African Journal of Biomedical Research. 7: 133-138.

Tiwari, S.K., Ghosh, R.C., Mascasenhas, A.R., Chauhan, H.V.S. (1991). Canine venereal sarcoma and its surgical management. Indian Vet. J. 68: 1078-1079.

Ulcar, L, Celeska, L., Ilievska, K., Atanaskova, E., Nikolovski, G., Trojacanec, P. (2012). Cytological diagnostic of canine transmissible Venereal tumor - Case report. Macedonian Veterinary Review. 35: 91-96.

Valladao, M.L., Scarpelli, K.C., Metze, K. (2010). Clinical utility of a life quality score in dogs with canine transmissible venereal tumour treated by Vincristine chemotherapy. Arq. Bras. Med. Vet. Zoo. 62: 1086-1093.