

## Treatment of canine transmissible venereal tumor using L-asparaginase, prednisone, and surgery in a clinical chemotherapy-resistant case

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**Abstract:** A 4-year-old intact female dog was presented with a vaginal transmissible venereal tumor (TVT) that had clinically developed multidrug resistance to vincristine, vinblastine, and doxorubicin. Complete remission was obtained with combined-modality therapy with surgery, L-asparaginase, and prednisone.

**Key words:** Cancer, Sticker tumor, transmissible venereal tumor

### 1. Introduction

A transmissible venereal tumor (TVT) is a horizontally transmitted venereal round cell tumor diagnosed in dogs (1,2). It is normally transmitted during coitus by viable tumor cells through injured mucosa. TVTs are usually located on genital, oral, or nasal mucosa (2). Initially the tumor is small, subsequently progressing to a large, ulcerated, and contaminated mass. The lesions are friable, hyperemic, hemorrhagic, multilobular, cauliflower-like masses (3). TVTs are locally aggressive and rarely metastatic. Definitive diagnosis is based on cytological and histological findings. Differential diagnosis includes round cell tumors such as lymphoma, plasma cell tumor, and mast cell tumor (4). The most effective treatment for TVT is chemotherapy with vincristine sulfate administered weekly (0.5 to 1 mg/m<sup>2</sup>, IV) for 4 to 6 weeks (5,6). Resistant cases can be treated with doxorubicin (30 mg/m<sup>2</sup>, IV) every 21 days (2 or 3 cycles) (4). When TVT becomes chemotherapy-resistant, treatment becomes a challenge (2,4,7). This report describes a bitch with TVT that had clinically developed resistance to vincristine, vinblastine, and doxorubicin and was successfully treated with surgery resection, L-asparaginase, and prednisone.

### 2. Case history

A 4-year-old, intact female, mixed-breed dog was referred to the Oncology Service of the Veterinary Teaching Hospital of the Federal University of Paraná (HV-UFPR), Curitiba, Brazil, with a history of vaginal TVT previously treated with chemotherapy with partial regression. The

dog had been adopted from the street 6 months before the beginning of treatment. Vincristine sulfate was administered for 8 weeks and partial regression of the tumor was observed. Subsequently, the dog was treated with 2 doses of vinblastine weekly without clinical response. Next, 4 cycles of doxorubicin were administered without tumor regression.

Upon admission at HV-UFPR, multiple hyperemic vaginal nodules were observed. Masses were friable and the tumor size was 5 mm in diameter (Figure 1A). TVT was diagnosed by imprint cytology (Figure 1B). The results of complete blood cell count (CBC) and blood chemistry tests were within reference intervals. Following diagnosis of TVT, surgical excision of tumors and ovariohysterectomy (OH) were performed under general anesthesia. OH was performed before tumor excision via abdominal laparotomy. En bloc excision included the vaginal vestibule, vulvar labia, and tumors and was done after episiotomy.

Histopathology from the vagina and vulva showed confluent sheets of neoplastic round cells growing in cords that were infiltrating into the submucosa and muscle, confirming TVT (Figure 1C). Neoplastic cells were present around the surgical margins. Infiltration of lymphocytes and plasma cells was also observed. Histopathology of the uterus and ovaries was unremarkable. Six days after surgery, chemotherapy was initiated with 1 dose of L-asparaginase (400 UI/kg, SC) and oral prednisone (2 mg/kg, once a day for a week). The prednisone dose was then reduced weekly to 1.5 mg/kg, 1 mg/kg, and 0.5 mg/

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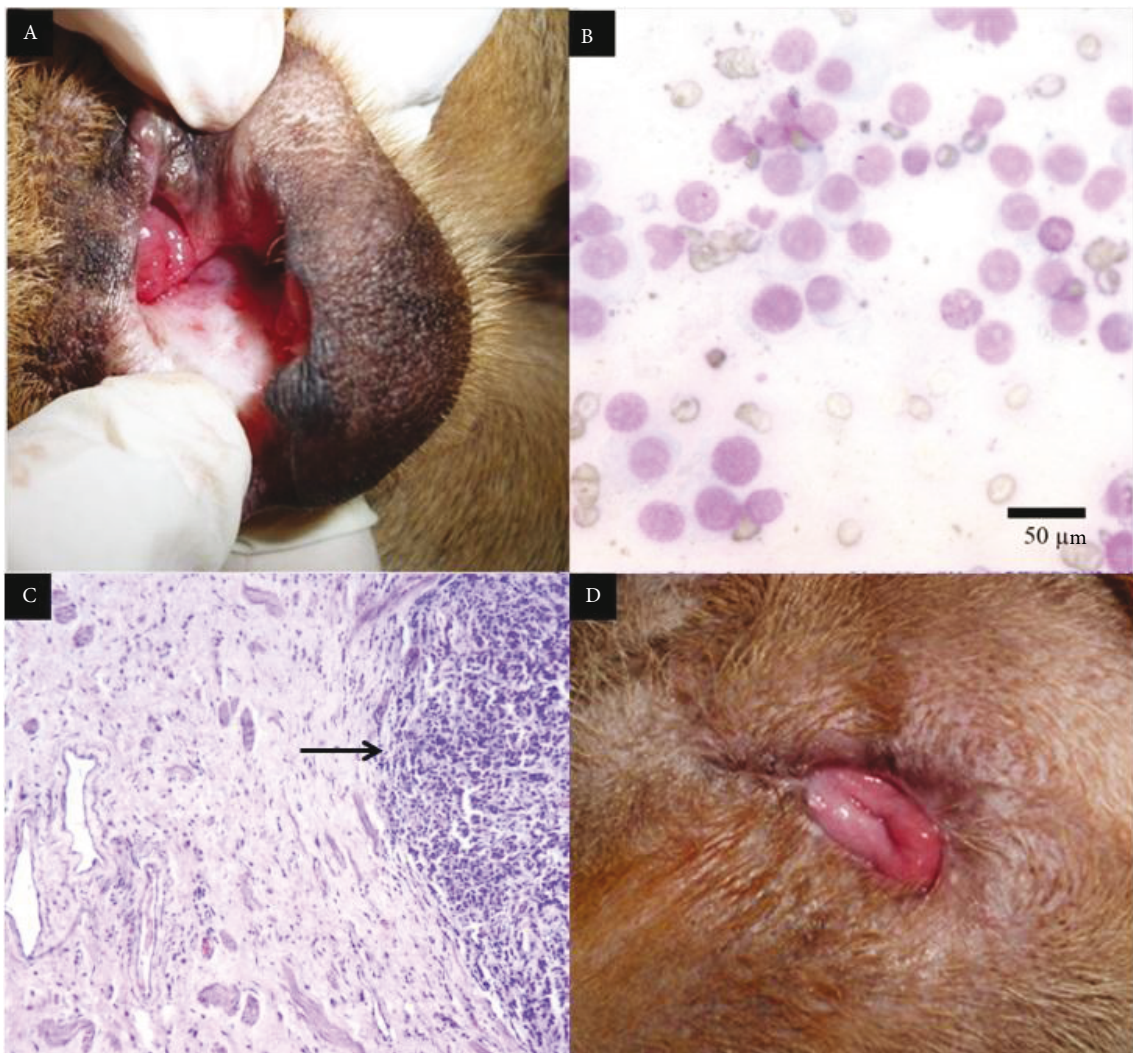
kg. Thirteen days after surgery, the dog was alert and the surgery site had healed (Figure 1D). A CBC disclosed mild neutrophilia [ $15.3 \times 10^3/\mu\text{L}$ , reference range (RR): 3 to  $11.5 \times 10^3/\mu\text{L}$ ] and lymphopenia ( $322/\mu\text{L}$ , RR: 1000 to  $4800/\mu\text{L}$ ). Blood chemistry tests were within reference intervals. Imprint cytology of the vulvar area was performed 1 month after surgery and did not show neoplastic cells.

Six months after surgery, the patient returned for an oncological evaluation and a vaginal nodule of 3 mm in diameter was observed (Figure 2A). Cytological examination of the lesion was unremarkable. An excisional biopsy revealed lymphocytes and plasma cell infiltration, indicating a subacute inflammatory process, without neoplastic cells (Figure 2B). Therefore, no further

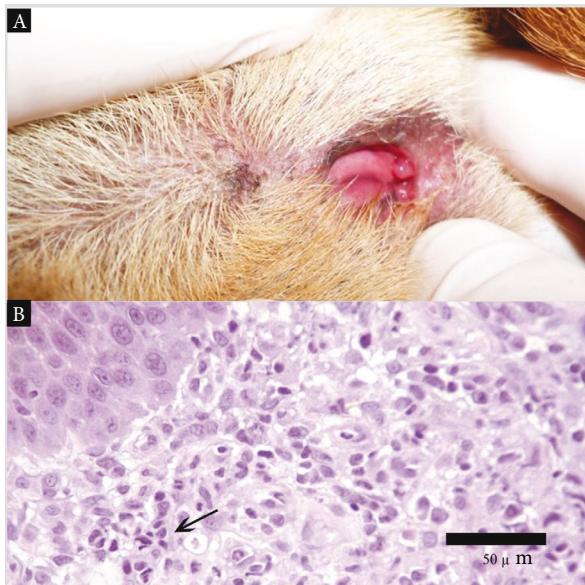
treatment was introduced. One year after treatment the tumor had not recurred and there was a remarkable improvement in the patient's quality of life.

### 3. Discussion

In general, vincristine sulfate as a single agent administered weekly for up to 6 cycles yields a cure for TVT. However, resistance to vincristine has been frequently observed in TVT patients. Resistance to chemotherapy should be considered when a residual tumor is present after 6 treatment cycles (7,8). In the present case, previous treatment details such as chemotherapy doses, application interval, clinical remission, and tumor reappearance were unknown. These factors might have predisposed TVT



**Figure 1.** A) Multiple hyperemic vaginal nodules. B) Cytology of canine transmissible venereal tumor from a vaginal mass aspiration. Moderate amount of cells with coarse chromatin and smoky vacuolated cytoplasm. Wright's stain, 40 $\times$ . C) Histology of canine transmissible venereal tumor. Round to ovoid neoplastic cells (arrow) arranged in short cords. H&E stain, 10 $\times$ . D) Surgical site 13 days after surgery.



**Figure 2.** A) Nodule on vaginal mucosa 6 months after TVT treatment. B) Histopathology from nodule located on vaginal mucosa showing presence of mild lymphoplasmacytic infiltration (arrow) just below the epithelial lining; neoplastic cells were not observed. H&E stain, 40 $\times$ .

chemotherapy resistance to vincristine and doxorubicin. There are several mechanisms by which tumor cells acquire drug resistance. In dogs with TVT, overexpression of permeability glycoprotein (P-gp) is one of the major

factors leading to multidrug resistance (MDR) (9,10). MDR may occur with vincristine, vinblastine, doxorubicin, prednisolone, and others (11). The dog in this case report possibly had P-gp-mediated MDR, but this hypothesis was not tested.

In contrast to antimicrotubule and anthracycline agents, L-asparaginase is not affected by MDR. L-asparaginase is an asparagine-specific enzyme whose mechanism of action is based on depletion of plasma asparagine. Some cancer cells are unable to synthesize asparagine and are dependent on an exogenous source of asparagine for survival, like lymphoma and mast cell tumors (12). A protocol combining L-asparaginase and vincristine was described as a successful treatment for vincristine-resistant TVT cases (7). Since the dog in this case had been previously treated with antimicrotubule and anthracycline agents, the protocol established was based on surgical cytology reduction obtained with surgery in association with L-asparaginase and prednisone. No data are available on the use of this protocol for treating dogs with TVT, and so it is possible that the use of either treatment alone could cause the response seen in this case. The benefit of including prednisone in this protocol is uncertain, because many vincristine-resistant TVT cases may also be resistant to prednisone mediated by P-gp (13).

Surgical resection in combination with L-asparaginase and prednisone may be an option for dogs with TVT that does not respond to standard chemotherapy treatment with vincristine and/or doxorubicin.

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