

Cutaneous nonepitheliotropic B-cell lymphoma in a Golden retriever

Cutaan niet-epitheliotroop B-cellymfoma bij een Golden retriever

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ABSTRACT

The vast majority of cutaneous canine nonepitheliotropic lymphomas are of T cell origin. Nonepithelial B-cell lymphomas are extremely rare. The present case report describes a 10-year-old male Golden retriever that was presented with slowly progressive nodular skin lesions on the trunk and limbs. Histopathology of skin biopsies revealed small periadnexal dermal nodules composed of rather pleomorphic round cells with round or contorted nuclei. The diagnosis of nonepitheliotropic cutaneous B-cell lymphoma was based on histopathological morphology and case follow-up, and was supported immunohistochemically by CD79a positivity.

SAMENVATTING

Het overgrote deel van de cutane caniene niet-epitheliotrope lymfoma's is van T-celorigine. Niet-epitheliotrope B-cellymfoma's zijn zeer zeldzaam. Dit is een casereport van een 10-jaar oude Golden retriever aangeboden met traaggroeiende nodulaire gezwellen in de huid van de romp en ledematen. Histopathologisch werden kleine periadnexale dermale nodulen, samengesteld uit pleomorfe rondcellen met een ronde of niervormige kern, aangetroffen. De diagnose van een niet-epitheliotroop lymfoma werd gesteld aan de hand van de histopathologie en caseopvolging en ondersteund door CD79a-positiviteit via immunohistochemie.

INTRODUCTION

Cutaneous nonepitheliotropic lymphoma has been described in dogs (Moore *et al.*, 1998). The major part of cutaneous canine nonepitheliotropic lymphomas are of T cell origin (Moore *et al.*, 1998). Nonepitheliotropic B-cell lymphomas are extremely rare (Day, 1995; Moore *et al.*, 1998). The etiology of canine nonepitheliotropic lymphoma is unknown. Hypothesized etiologies include retroviral infection, environmental contamination with phenoxyacetic acid herbicides, magnetic field exposure, chromosomal abnormalities, and immune dysfunction (The Merck Veterinary Manual, 2006). Commonly lymphoma affected dog breeds are: Boxer, Scottish terrier, Basset hound, Airedale terrier, Chow Chow, German shepherd dog, Poodle, St. Bernard, Bulldog, Beagle, Rottweiler, Weimaraner, Cocker spaniel and Golden retriever (Beale & Bolon, 1993; Scott *et al.*, 2000). This last breed, as in the present case, is especially susceptible to developing lymphoma, with a lifetime risk of 12.5% (Modiano *et al.*, 2005). Curatively, surgical excision of solitary tumors in dogs has been reported (Brown *et al.*, 1980; McKeever *et al.*, 1982), however the vast majority of cutaneous canine nonepitheliotropic lymphomas exhibit rapid progression and metastasis to draining lymph nodes and subsequent systemic involvement (Moore *et al.*, 1998).

Histologically, nonepitheliotropic lymphomas are classically characterized by predominantly deep dermal and subcutaneous nonencapsulated masses, composed of sheets, clusters or nodular perivascular aggregates of relatively monomorphic cells. However, cells can vary tremendously in morphology. The neoplastic infiltrates have a bottom-heavy or base-wide configuration. With superficial dermal involvement, an area of uninvolved superficial dermis (Grenz zone) is usually seen and adnexal structures are not invaded, although there are exceptions (Gross *et al.*, 2006). Immunohistochemistry is often required to differentiate nonepitheliotropic lymphoma from other round cell tumors.

The present case describes a nonepitheliotropic B-cell lymphoma in a Golden retriever, diagnosed by histopathology, case follow-up, and immunohistochemistry.

CASE REPORT

Case history

A 10-year old intact male Golden retriever, with a previous history of moderate atopic dermatitis, was referred for slowly progressive development of small nodular skin lesions between the shoulders, and on the abdomen, groin, thighs and axillae (Figure 1). Except



Figure 1. Macroscopic appearance of the lesions. Note the presence of small bluish nodular skin lesions on the ventral abdomen and prepuce.

for these skin lesions, the dog was in good health. On clinical examination, small nodular lesions characterized by a bluish color and intact skin surface were found. The peripheral lymph nodes were not enlarged. Cytology revealed a marked presence of mononuclear cells and some binucleated and polynucleated cells. Lymphoglandular bodies were not recognized in the samples. Skin biopsies were taken for routine histopathology examination.

Histopathology

Histopathologically, the lesions were dermal, sub-epidermal nodular masses of 0.5 cm in diameter. They were rather cellular, nonencapsulated, and moderately circumscribed, showing mildly infiltrative growth between the dermal structures. The nodules consisted of a proliferation of large round cells in sheets, with severe anisocytosis and nuclear polymorphism with a variable amount of eosinophilic cytoplasm (Figure 2). Binuclear and polynuclear cells, cells with kidney-like and band-like nuclei, and cells with eccentric nuclei were also observed (Figure 3). An intermittent grade of mitotic activity was noted. Some nodules had localized superficial epidermal ulceration with necrosis and pyogranulomatous infiltrate and secondary epidermal hyperplasia. Giemsa stain did not reveal metachromatic granules in the cytoplasm of the tumor cells. No typical plasma cells were present.

Immunohistochemistry

The large cells were positive for CD79a, which indicates either B-cell origin or plasma cell origin.

Case follow-up

As cost was a limiting factor, the dog had only been treated with prednisolone 0.5 mg/kg every other day. Blood analysis three months later was without gross abnormalities. The skin lesions had slowly enlarged,

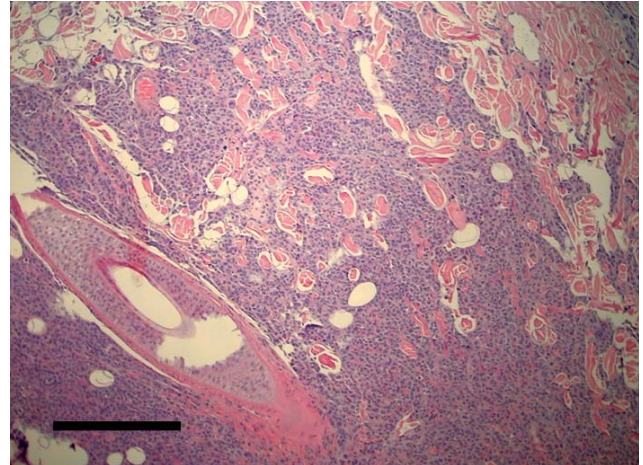


Figure 2. Microscopic presentation of the lesions: the nodules consist of a proliferation of round cells in sheets, with severe anisocytosis and nuclear polymorphism with variable amounts of eosinophilic cytoplasm. HE stain, Bar = 300 μ m.

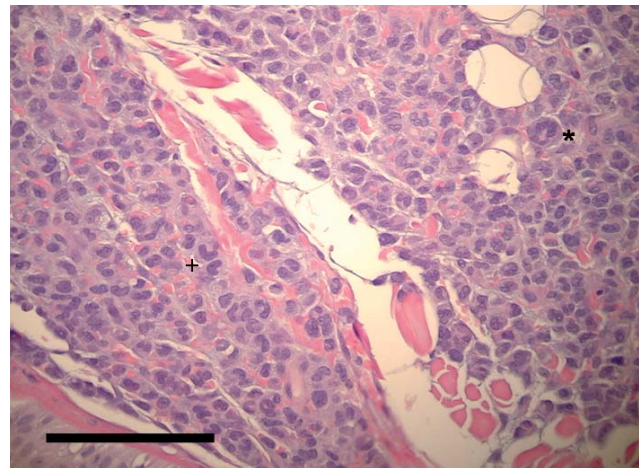


Figure 3. Microscopic presentation of the lesions: note the bi- and polynuclear cells (*), cells with kidney-like and band-like nuclei (+), and cells with eccentric nuclei. HE stain, Bar = 25 μ m.

and at that time the dog still remained in relatively good health. However, another 3 months later (i.e. 6 months after diagnosis), the dog was euthanized as there was a marked enlargement of the skin nodules, severe anemia, mild lymphopenia and lameness. In addition, all the peripheral lymph nodes were greatly enlarged. Unfortunately, section was not allowed.

DISCUSSION

Lymphoma of the skin is an uncommon occurrence. It is one of the 4 variants of the lymphoma classification, next to multicentric, mediastinal and gastrointestinal lymphoma. In humans, lymphomas have been subtyped on the basis of their cellular morphology, which has resulted in several classification systems (Greenlee *et al.*, 1990). Unfortunately, these classification systems of human lymphomas are of limited value for predicting the behavior and prognosis

of nonepitheliotropic lymphomas in dogs (Valli *et al.*, 2002).

Further lymphoma classification is based on the involvement of B-lymphocytes or T-lymphocytes. There are two forms of cutaneous lymphoma: epitheliotropic (closely confined to the epidermis) and nonepitheliotropic. The epitheliotropic form is typically of T-cell origin and has also been called mycosis fungoides. The vast majority of the nonepitheliotropic lymphomas are also of T-cell origin (Moore *et al.*, 1998). The nonepitheliotropic form can present from a single nodule to large areas of bruised, ulcerated, hairless skin (Hoskins, 2006). In the present case, small nodular lesions characterized by a bluish color and intact skin surface were noted.

The cellular morphology of cutaneous nonepitheliotropic lymphoma is highly variable, involving sheets and clusters of neoplastic cells, varying from small to large in size, and even including immunoblastic cell types. Anisokaryosis is a prominent feature. The mitotic activity is highly variable and corresponds to the cell type and/or tumor grade (Gross *et al.*, 2006). In the present case, the cellular pleomorphism was extensive, with bi- and polynuclear cells, and kidney-shaped and band-like nuclei.

In human medicine, there is a relationship between the immunodysregulation observed in atopic dermatitis and the development of epitheliotropic lymphoma (Santoro *et al.*, 2007). Santoro *et al.* (2007) detected an association between atopic dermatitis and epitheliotropic lymphoma in dogs. Although, the dog in the present case had a nonepitheliotropic lymphoma, it also had a history of moderate atopic dermatitis.

The diagnosis of lymphoma is classically made by cytologic or histopathologic evaluation of the affected organ system. Immunohistopathology can provide a definite diagnosis of lymphoma, and it can distinguish B-cell from T-cell lymphomas. Cytology is unable to differentiate or categorize the wide spectrum of lymphomas. Due to these constraints, histopathologic tissue evaluation remains the gold standard for the diagnosis of lymphoma, providing the additional morphologic information required for definitive classification. Nonepitheliotropic lymphomas should be differentiated from other round cell tumors, such as canine histiocytoma, plasmacytoma, mast cell tumor and amelanotic melanoma.

Canine histiocytomas have a “top-heavy” configuration and sometimes intraepidermal tumor cells, nevertheless the cytologic features may be indistinguishable between histiocytoma and lymphoma.

Plasmacytomas often display marked anisocytosis and anisokaryosis, and bi- and multinucleated cells are also common, as in the present case. Proliferating plasma cells are arranged in tight packets and well-differentiated plasma cells can usually be identified along the tumor margins. Well-differentiated plasma cells were not observed in the present case. The mitotic index is usually low, in contrast to an intermittent grade of mitotic activity in the present case.

Additional stainings such as toluidine blue and

giemsa will usually identify the metachromatic granules of poorly granulated mast cell tumors.

Round cell amelanotic melanomas may be characterized by cells with a larger amount of cytoplasm when compared to lymphoma, and there may be few admixed spindle tumor cells. Fontana-Masson stain may assist in detecting small amounts of melanin within the tumor cells (Gross *et al.*, 2006).

However, in the present case, B-cell lymphoma and plasmacytoma cannot be distinguished on the basis of immunohistochemistry. Although the cells were CD79a positive and the incidence of CD79a expression varies in plasma cells, plasmacytoma could not be ruled out. The diagnosis was mostly based on the histopathological morphology and case follow-up. Immunohistochemistry showed B-cell origin (B-cell or plasma cell) and clonality supported the clonal expansion (unpublished data) (can be B-cell or plasma cell). It was the morphologic appearance that pointed towards a B-cell lymphoma (Verena K. Affolter, personal communication). The majority of cutaneous plasma cell tumors in the dog are benign. Metastases of plasmacytomas have been reported, though rarely, while cutaneous lymphoma tends to be a progressive disease, with the development of multicentric skin tumors and ultimately involving the regional lymph nodes and viscera, as in the present case (Goldschmidt and Hendrick, 2002).

The treatment of lymphoma tends to be palliative, as complete cure is rare, but long remission times are possible with chemotherapy. With effective protocols, the average first remission times are 6 to 8 months. Second remissions are shorter and harder to accomplish. With combination chemotherapy, the expected survival time for dogs with B-cell lymphoma is approximately 9 to 12 months. For dogs with T-cell lymphoma, the expected survival times are shorter (6 months). The most common treatment is a combination of cyclophosphamide, vincristine, prednisolone, L-asparaginase, and doxorubicin (Morrison, 1998). Other chemotherapy drugs such as chlorambucil, lomustine (CCNU), cytosine arabinoside, and mitoxantrone are sometimes used in the treatment of lymphoma by themselves or in substitution for other drugs. In most cases, appropriate treatment protocols cause few side effects, but white blood cell counts must be monitored (Wikipedia, the free encyclopedia, 2007).

If therapy is restricted (finances), prednisolone used alone can improve the symptoms dramatically, but it does not significantly affect the survival rate. The average survival times of dogs treated with prednisolone and untreated dogs are both one to two months (Morrison, 1998). Nevertheless, the present patient was still in relatively good health after three months. Using prednisolone alone can cause the cancer to become resistant to other chemotherapy agents, so it should only be used if more aggressive treatment is not an option (Wikipedia, the free encyclopedia, 2007). Untreated dogs have an average survival time of sixty days (siedlecki *et al.*, 2006). Dogs with B-lymphocyte

tumors have a longer survival time than those with T-lymphocyte tumors (Morrison, 1998).

By way of summary, this dog was still in relatively good health three months after diagnosis, but had to be euthanized six months after diagnosis due to widespread lymphonodular metastasis. Nevertheless, the survival time in the present case was longer than the one or two months of expected survival time reported in the literature. This is probably due to the fact that the greater part of nonepitheliotropic lymphomas are of T-cell origin (Moore *et al.*, 1998) and that nonepitheliotropic T-cell lymphomas have a shorter expected survival time compared to nonepitheliotropic B-cell lymphomas. This is an argument for further differentiating lymphomas after the initial histopathological diagnosis.

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