

Cutaneous Lymphoma in a Peafowl

Funda YILDIRIM^{1*}, Damla HAKTANIR¹, Aydın GUREL¹, Alper DEMIRUTKU²,
Serhat OZSOY², Gulben ERDEM HUQ³

¹Istanbul University, Faculty of Veterinary Medicine, Department of Pathology

²Istanbul University, Faculty of Veterinary Medicine, Department of Surgery

³Ministry of Health, Istanbul Education and Research Hospital, Pathology Department

*Corresponding Author: Funda YILDIRIM Istanbul University, Faculty of Veterinary Medicine,
Department of Pathology, 34320, Avcılar, Istanbul, Turkey
e-mail: funda@istanbul.edu.tr

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ABSTRACT

Cutaneous lymphoma was clinically, histopathologically and immunohistochemically detected in a 1.5-year-old male peafowl in this case. Histopathologically, intradermally located diffuse cutaneous lymphoma composed of atypical lymphoblastic cells and exhibiting a pattern of follicular development was diagnosed on the right buccal area of the peafowl. Positive reaction with CD3 and CD20 antibodies confirmed the diagnosis immunohistochemically.

Key Words: Peafowl, cutaneous lymphoma, immunohistochemistry, CD3, CD20

ÖZET

BİR TAVUS KUŞUNDA KUTANÖZ LENFOMA OLGUSU

Bu olguda; 1,5 yaşındaki erkek tavus kuşunda klinik, histopatolojik ve immunohistokimyasal yöntemlerle kutanöz lenfoma belirlendi. Tavus kuşunun sağ yanak bölgesinde, histopatolojik olarak atipik lenfoblastik hücrelerden oluşan ve foliküler şekil sergileyen, intradermal yerleşimli kitle diffüz kutanöz lenfoma olarak teşhis edildi. İmmunohistokimyasal olarak CD3 ve CD20 antikorları ile pozitif reaksiyon elde edilmesi sonucu teşhis doğrulandı.

Anahtar Kelimeler: Tavus kuşu, kutanöz lenfoma, immunohistokimya, CD3, CD20

Introduction

Avian lymphosarcoma is frequently reported in poultry and is commonly associated with Marek's disease and Avian Leucosis Virus-Type 1 (Latimer et al., 1998; Miller et al., 1998). Despite the frequency of these tumors in poultry, few reports are available with respect to cutaneous lymphomas in companion and free ranging birds, particularly in peafowl (Jones et

al., 2007; Latimer et al., 1998; Miller et al., 1998). While viral etiology of lymphosarcomas in poultry eventually result in diffuse visceral involvement with commonly affected organs such as the liver and the spleen; solitary lesions are generally located in cutaneous tissues, as periorbital region that has previously described to be the particular site involved in pet birds (Latimer et al., 1998; Miller et al., 1998; Rivera et al., 2009; Souza et al., 2008).

The immunohistochemically labelling of the cell surface receptors for the classification of tumours have been used widely for a long time. The CD3 used in this study is one of the antigen binding receptors located on the T cell surface, which is a complex protein molecule composed of four distinct chains. CD20 and CD21 which are B cell surface receptors are expressed on a wide range normal and neoplastic B cells (Tizard, 2004).

Although the availability of some data the neoplastic lesions in pet birds, in terms with the biological behavior, clinical and histopathological classification have not been elucidated yet. A case of cutaneous lymphoma on the right buccal area of a peafowl was aimed to be presented with its clinical, pathological and immunohistochemical aspects in this report.

Case

A 1.5-year-old male peafowl (*Pavo cristatus*) was referred to the Department of Surgery, Faculty of Veterinary Medicine, Istanbul University with the complaint of a hyperpigmented, and crusted progressive swelling on the right buccal area (Figure 1). Clinical examination revealed a lesion, characterized by swelling, ulceration and encrustation, which were suggestive of a fungal infection. However, despite the symptomatic treatment with antifungal and antibacterial agents the lesion progressed and thus, it was surgically removed. The excised specimen was then submitted to Department of Pathology. Grossly, it was an ulcerative tumoral mass of 2 cm in diameter covered by a dark crust, with its soft, fleshy and grey-white bulging cut surface. Cytologic slides were prepared by making direct imprints of the cut surface, air-dried and stained with May-Grünwald-Giemsa. The biopsy specimen was then fixed in 10% formalin, routinely processed, and stained with hematoxylin-eosin (H&E) to be evaluated by light microscopy. Formalin-fixed paraffin-embedded sections were immunohistochemically stained by streptavidin-biotin immunoperoxidase method, as well, with CD3, CD20, CD21 and pancytokeratin (AE1/AE3)

antibodies (Dako Corporation, Carpinteria, CA, USA) using a commercially available kit Labelled Streptavidin-Biotin2 System, Horseradish Peroxidase (LSAB2 System, HRP) (Dako Corporation, Carpinteria, CA, USA). Immunoreaction was visualized by diaminobenzidine (DAB) and the sections were counterstained with Mayer's hematoxylin.



Figure 1. Gross appearance of tumoral mass on the right buccal area.

Şekil 1. Sağ yanak bölgesinde tümöral kitlenin makroskopik görünümü.

Large, round, pleomorphic lymphoblastic cells, some of which had large and cleaved nuclei with prominent or multiple nucleoli and small lymphocytes suggestive of normal T cells and clusters of erythrocytes were the cytological findings. Histopathology revealed a tumoral mass, arranged in a distinctive follicular pattern (Figures 2 and 3) composed mostly of large, pleomorphic lymphoid cells with multinucleolated cleaved vesicular nuclei and numerous mitotic figures (Figure 4). Intradermally located neoplastic cells were observed to have destructed the epidermal layer in some areas, yielding hemorrhage and ulceration on the surface. Wide necrosis was evident beneath these areas. Numerous fungal hyphae were found scattered on the epidermal surface. Grocott's stain was used to display the fungal elements. The sections showed positive immunoreaction for CD3 and CD20 (Figure 5A, 5B), while no reaction was obtained with CD21 and pancytokeratin antibodies. On the basis of histomorphology and immunohistochemistry, the neoplastic entity was

diagnosed as non-epitheliotrophic cutaneous lymphoma of B type. The patient's outcome was followed up. However; the patient died 4 weeks after the surgery, rendering a detailed postmortem examination possible, as well. An extensive wounded area was replaced by the excised tumoral mass. Necrotizing changes accompanied by edema and hemorrhage were detected to have been expanded deep into the subdermal and muscular tissues. Perilaryngeal

region was affected due to remarkable swelling of these tissues. Histopathologically, edema, hyperemia, necrosis, and fibrin fibers along with few clusters of atypical lymphoid cells were visible at the wounded site. No gross lesions in association with the primary entity were observed at necropsy and also histopathologically, ruling out a putative visceral involvement.

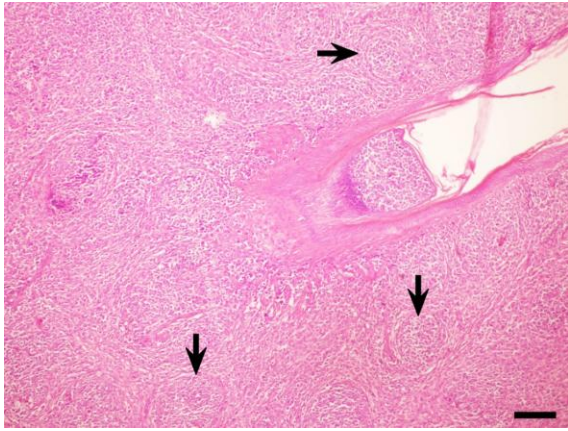


Figure 2. Subepidermal pleomorphic lymphoid cells arranged in follicular pattern (arrows) H&E, Bar = 100 µm.

Şekil 2. Foliküler şekilde düzenlenmiş subepidermal pleomorfik lenfoid hücreler (oklar) H&E, Bar = 100 µm.

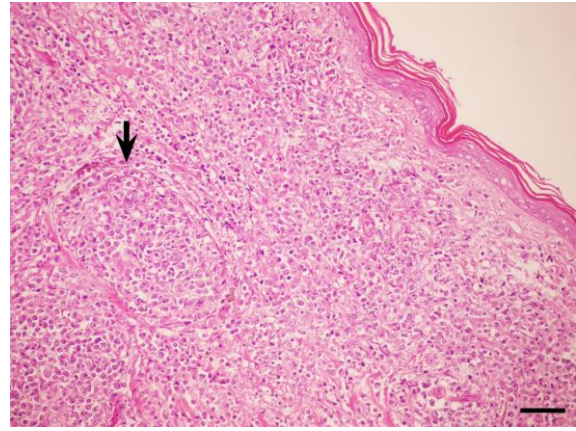


Figure 3. Subepidermal pleomorphic lymphoid cells arranged in follicular pattern (arrow) H&E, Bar = 50 µm.

Şekil 3. Foliküler şekilde düzenlenmiş subepidermal pleomorfik lenfoid hücreler (ok) H&E, Bar = 50 µm.

Discussion and Conclusion

Since no accurate classification system is available for avian species in terms of the neoplastic entities, the mass was evaluated on the basis of the principles of Histopathological Classification of Hematopoietic Tumors in Domestic Animals, established by World Health Organization (Valli et al., 2002). Cutaneous lymphomas in cat and dog are allocated into two categories as epitheliotrophic and non-epitheliotrophic tumors (Valli et al., 2002). The epitheliotrophic type, also known as mycosis fungoides is a common entity in dog, mostly of T-cell origin, where as B-cell cutaneous lymphomas are non-epitheliotrophic tumors, affecting the deep dermal layers (Ettinger, 2003). This case is different from

Mycosis fungoides which located within the dermis and epidermis remained intact. So, the present case, as its canine counterpart, was diagnosed to be non-epitheliotrophic cutaneous lymphoma which has been reported as extremely rare canine lymphomas (Day, 1995). Histomorphological features of large neoplastic cells were compatible with those of lymphoblastic B cells, therefore the tumor was considered to be of B-cell origin. Besides, the most common lymphoid tumors in primary skin tumors of human are reported to be B-cell follicle center lymphoma (Horwitz, 2008). Histopathological appearance of this case was found to be compatible with human Primary cutaneous follicle center lymphoma.

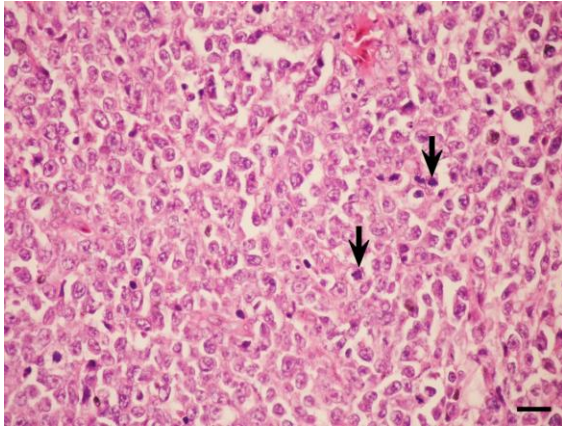


Figure 4. Large, pleomorphic lymphoid cells with cleaved vesicular nuclei and mitotic figures (arrows), H&E, Bar = 20 μ m.

Şekil 4. Çentikli veziküler çekirdekli, iri pleomorfik lenfoid hücreler ve mitotik figürler (oklar), H&E, Bar = 20 μ m.

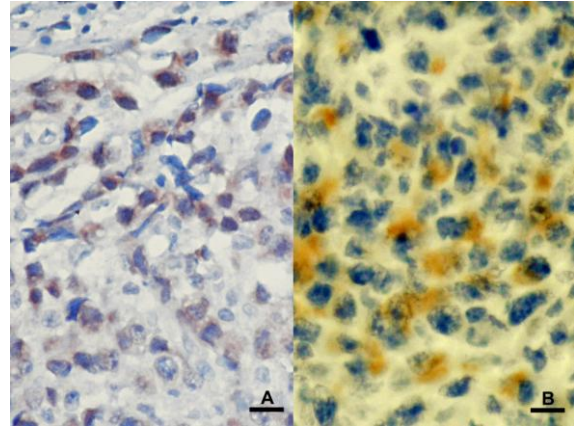


Figure 5A. CD3 positive staining, IHC, Bar = 20 μ m.

Şekil 5A. CD3 pozitif boyanma, IHC, Bar = 20 μ m.

Figure 5B. CD20 positive staining, IHC, Bar=20 μ m.

Şekil 5B. CD20 pozitif boyanma, IHC, Bar = 20 μ m.

Inflammatory lesions and xanthoma have been reported to likely accompany the primary tumoral entities in avian species (Jones et al., 2007; Rivera et al., 2009). In the present case, fungal hyphae were detected within the foci of ulceration, which was demonstrated by Grocott's stain. After the diagnosis was confirmed both histologically and immunohistochemically as B-cell type lymphoma, the presence of fungal elements were considered to be a secondary incident.

While CD20 is a well-known antigen expressed on the surface of B-type lymphocytes including follicular and dendritic cells, thus rendering CD20 antibody a useful marker for B cell lymphomas (Kojima et al., 2005), CD21 antibody is used in the differential diagnosis of follicular dendritic cell sarcomas (Kojima et al., 2005) and CD3 antibody reacts with T-type lymphoid cells (Souza et al., 2008). In the present case, immunohistochemistry demonstrated substantial positive immunoreaction for CD20 confirming the cell of origin as B-type, while no immunoreaction was obtained with CD21 Ab. CD3 positive cells were rather observed at the periphery of the tumoral mass, where a demarcation line occurred, revealing that those were normal T cells, dispersed

throughout the inflammatory zone. And finally the lack of immunoreaction with pancytokeratin antibody, except for the epidermal cells, excluded the possibility of a round cell tumor of epithelial origin. Although morphological and immunohistochemical findings of the present case were similar to those of its canine and feline counterparts to some extent, the prominent follicular growth pattern of the neoplastic cells, yet the lack of immunoreaction for CD21 pointed out the distinctive feature of follicular type B-cell lymphomas in this avian species. The presence of T-cell lymphocytes demonstrated by CD3 positive immunostaining was also a significant finding, distinguishing B-type non-epitheliotropic cutaneous lymphoma in the peafowl from that of other domestic animals. CD3 positivity had been previously reported also in cutaneous B-cell lymphoma of an umbrella cockatoo which was associated with the presence of reactive T cells among the neoplastic B lymphocytes (Rivera et al., 2009). Although there are some reports available with respect to histomorphological and immunohistochemical aspects of malignant lymphomas in exotic companion birds, no classification scheme peculiar to avian species has been established in terms of neither histo-

morphological features nor immuno-histochemical staining patterns of lymphoid tumors. Therefore, the present case was considered to be worthy of presentation due to its contributory value to literature.

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REFERENCES

- Day, M.J., 1995.** Immunophenotypic characterization of cutaneous lymphocytic neoplasia in the dog and cat. *Journal of Comparative Pathology* 122, 79-96.
- Ettinger, S.N., 2003.** Principles of treatment for canine lymphoma. *Clinical Techniques in Small Animal Practice* 18(2), 92-97.
- Horwitz, S.M., 2008.** Fast Facts: Primary Cutaneous B-Cell Lymphoma. *Cutaneous Lymphoma Foundation*: http://www.clfoundation.org/about/cutaneous_lymphoma/CLFCBCL_Fast_Facts.pdf. (Erişim tarihi: 01.06.2011).
- Jones, A., Kirchgessner, M., Mitchell, M.A., McLaughlin, L.D., Guzman, D., 2007.** Diagnostic challenge. *Journal of Exotic Pet Medicine* 16(2), 122-125.
- Kojima, M., Nakamura, S., Murase, T., Motoori, T., Murayama, K., Lijima, M., Itoh, H., Sakota, N., Masawa, N., 2005.** Follicular colonization of nodal marginal-zone B-cell lymphoma resembling follicular lymphoma: Report of 6 cases. *International Journal of Surgical Pathology* 13(1), 73-78.
- Latimer, K.S., Ritchie, B.W., Campagnoli, R.P., Harris, D.J., 1998.** Cutaneous T-cell-rich B-cell lymphoma and leukemic blood profile in an umbrella cockatoo (*Cacatua alba*). In: *Annual International Virtual Conference in Veterinary Medicine (IVCVM)*, College of Veterinary Medicine, University of Georgia: <http://www.vet.uga.edu/vpp/archives/ivcvm/1998/latimer01/index.php>. (Erişim tarihi: 01.06.2011).
- Miller, P.E., Paul-Murphy, J., Sullivan, R., Cooley, J., Dubielzig, R.R., Murphy, C.J., Fadly, A.M., 1998.** Orbital lymphosarcoma associated with reticuloendotheliosis virus in a peafowl. *Journal of American Veterinary Medicine Association* 213(3), 377-380.
- Rivera, S., McClearen, J.R., Reavill, D.R., 2009.** Treatment of nonepitheliotrophic cutaneous B-cell lymphoma in an umbrella cockatoo. *Journal of Avian Medicine and Surgery* 23(4), 294-302.
- Souza, M.J., Newman, S.J., Greenacre, C.B., Avenell, J.S., Wall, J.S., Phillips, J.C., Fry, M.M., Donnell, R.L., Daniel G.B., 2008.** Diffuse intestinal T-cell lymphosarcoma in a yellow-naped Amazon parrot (*Amazona ochrocephala auropalliata*). *Journal of Veterinary Diagnostic Investigation* 20, 656-660.
- Tizard, I.R., 2004.** *Veterinary Immunology: An Introduction*, Seventh Edition. Saunders, Philadelphia.
- Valli, V.E., Jacobs, R.M., Parodi, A.L., Vernau, W., Moore, P.F. 2002.** *Histological Classification of Hematopoietic Tumors of Domestic Animals*. In: Schulman, F.Y. (Ed), *WHO International Histological Classification of Tumors of Domestic Animals. VIII. Armed Forces Institute of Pathology*. Washington, D.C., Series 2, pp. 21-63.