

AperTO - Archivio Istituzionale Open Access dell'Università di Torino

## Canine lymphoma: Going with the flow

### **This is the author's manuscript**

*Original Citation:*

*Availability:*

This version is available <http://hdl.handle.net/2318/93415> since

*Published version:*

DOI:10.1016/j.tvjl.2011.02.017

*Terms of use:*

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)



## UNIVERSITÀ DEGLI STUDI DI TORINO

This Accepted Author Manuscript (AAM) is copyrighted and published by Elsevier. It is posted here by agreement between Elsevier and the University of Turin. Changes resulting from the publishing process - such as editing, corrections, structural formatting, and other quality control mechanisms - may not be reflected in this version of the text. The definitive version of the text was subsequently published in

Canine lymphoma: Going with the flow

The Veterinary Journal, Volume 188, Issue 2, Pages 134–135. May 2011.

<http://dx.doi.org/10.1016/j.tvjl.2011.02.017>

You may download, copy and otherwise use the AAM for non-commercial purposes provided that your license is limited by the following restrictions:

- (1) You may use this AAM for non-commercial purposes only under the terms of the CC-BY-NC-ND license.
- (2) The integrity of the work and identification of the author, copyright owner, and publisher must be preserved in any copy.
- (3) You must attribute this AAM in the following format: Creative Commons BY-NC-ND license  
<http://creativecommons.org/licenses/by-nc-nd/4.0/deed.en>

<http://www.sciencedirect.com/science/article/pii/S1090023311000761>

## **Guest Editorial**

### **Canine lymphoma: Going with the flow**

Similar to the many forms of non-Hodgkin lymphoma (NHL) described in humans, canine lymphoma represents a morphologically, immunologically and clinically heterogeneous group of neoplasms, so diverse in fact that it has been suggested that the ‘one word’ diagnosis ‘lymphoma’ is no longer sustainable (McManus, 2008). The REAL and WHO classifications of human NHL take into consideration multiple features of the disease process (clinical, morphological, immunophenotypic, genetic, epidemiological) that facilitate the identification of the most appropriate therapy and provide the most accurate prognosis. The most recent classifications of canine lymphoproliferative disease, the updated Kiehl and WHO protocols, have taken a step in this direction in more clearly defining tumour sub-types through immunophenotyping (Fournel-Fleury et al., 1997; Valli, 2007).

However, the number of sub-types into which canine lymphoid tumours are classified remains limited (small vs. large cell lymphomas and B vs. T cell lymphomas, respectively) in terms of providing clinically or prognostically useful information. In addition to conventional immunophenotyping, flow cytometric immunophenotyping provides further valuable insights into the character of these neoplasms (Fournel-Fleury et al., 2002; Sozmen et al., 2005; Wilkerson et al., 2005; Comazzi et al., 2006a,b; Riondato et al., 2006a; Gelain et al., 2008). The review by Comazzi and Gelain (2011) in this issue of *The Veterinary Journal*, outlines how, why and when this technique should be deployed in conjunction with cytological evaluation. The authors describe how flow cytometry can provide information in relation to tumour cell lineage, maturation stage and level of antigen expression and highlight the application of this technique in

identifying cell clonality, blood and bone marrow involvement, and in quantifying the numbers of neoplastic cells that survive chemotherapy (minimal residual disease). The review illustrates how such multi-parametric data is invaluable in diagnosing, staging and monitoring canine lymphoma.

Although the advantages of flow cytometry in oncohaematology are acknowledged by many veterinary clinicians, the full potential of the method in clearly differentiating the various sub-types of lymphoma remains to be realised. Recent research suggests we are moving in the right direction with Ponce et al. (2004) describing cytological sub-types that have different prognoses and Marconato et al. (2008) reporting how tumour staging can dictate the chemotherapy protocol. However, to maximise the benefits of such developments, we need to greatly expand the number of tests, such as the flow cytometry, currently available to sub-type canine lymphoma in veterinary diagnostic laboratories.

Studies at The University of Turin are using flow cytometry to investigate if tumour cell DNA content (ploidy and cell cycle), proliferative activity and apoptosis are potentially useful indicators in characterising canine lymphomas and leukaemias. Preliminary findings suggest DNA ploidy and cell cycle analysis with S-phase determination could be useful in discriminating acute from chronic leukaemia and in differentiating acute leukaemia from V-stage lymphosarcoma (Riondato et al., 2010). Flow cytometry is widely used to determine tumour cell DNA content and proliferation in human neoplasms, where these parameters may have use as prognostic markers. DNA aneuploidy has been assessed in both solid tumours and haematological malignancies and a large amount of data exists for this feature in the context of

human breast and colon cancer, as well as for acute lymphocytic leukaemia, lymphoma and multiple myeloma (McCoy and Davis, 2001). Although the results of such studies are often conflicting, the large number of cases where cell DNA content and cell cycle analysis were carried out suggest these parameters are of clinical value in selected tumours and/or localizations (Bagwell et al., 2001; Michels et al., 2003). Aneuploidy is considered a reliable marker of malignancy (Petrozza et al., 2001) and markers of both ploidy and cell proliferation can facilitate both prognosis and therapeutic strategies (Turner and Wass, 1999). In human NHL, the correlation of DNA ploidy patterns with cytological grading is variable and cell cycle analysis has proved more valuable clinically (Ross, 1996; Pinto et al., 2003). In a veterinary context, few studies have focused on cell DNA content or proliferation in canine lymphomas and small numbers of cases have typically been assessed (Hamilton, 1990; Teske et al., 1993; Rutteman et al., 1994; Guglielmino et al., 2001; Riondato et al., 2006b). Most importantly, these cell parameters have not been correlated with current tumour classification schemes.

Alterations to cell apoptotic mechanisms facilitate both carcinogenesis and tumor progression and also contribute to resistance to treatment as many current anti-cancer therapies rely on the activation of programmed cell death in neoplastic cells. Hence, defects in apoptotic molecules may serve as targets in the design of novel therapeutic strategies as well as biomarkers of response to treatment and prognosis. In this context there may be a role for flow cytometry in the evaluation of apoptotic activity in a neoplasm following cell labelling with annexin V. Research into apoptosis as a therapeutic or prognostic marker of canine lymphoma remains a 'green-field site'.

The review by Comazzi and Gelain (2011) in this issue of *The Veterinary Journal* clearly outlines the capacity flow cytometry to characterise canine lymphomas and, together with the judicious use of other ancillary tests, will no doubt pave the way to improved patient care. Collaborative research involving both veterinary and human oncologists is likely to be highly mutually beneficial in this context and the recently established research forum<sup>1</sup> co-sponsored by The Universities of Milan and Vienna, is well placed to promote such efforts.

Fulvio Riondato  
*Department of Animal Pathology  
The University of Turin  
Via L. Da Vinci 44  
10095 Grugliasco  
Italy*  
E-mail address: fulvio.riondato@unito.it

## References

- Bagwell, C.B., Clark, G.M., Spyrtos, F., Chassevent, A., Bendahl, P.O., Stål, O., Killander, D., Jourdan, M.L., Romain, S., Hunsberger, B., Baldetorp, B., 2001. Optimizing flow cytometric DNA ploidy and S-phase fraction as independent prognostic markers for node-negative breast cancer specimens. *Cytometry* 46, 121-135.
- Comazzi, S., Gelain, M.E., Riondato, F., Paltrinieri, S., 2006a. Flow cytometric expression of common antigens CD18/CD45 in blood from dogs with lymphoid malignancies: a semi-quantitative study. *Veterinary Immunology and Immunopathology* 112, 243-252.
- Comazzi, S., Gelain, M.E., Spagnolo, V., Riondato, F., Guglielmino, R., Sartorelli, P., 2006b. Flow cytometric patterns in blood from dogs with non-neoplastic and neoplastic hematologic diseases using double labeling for CD18 and CD45. *Veterinary Clinical Pathology* 35, 47-54.
- Comazzi, S., Gelain, M.E., 2011. Use of flow cytometric immunophenotyping to refine the cytological diagnosis of canine lymphoma. *The Veterinary Journal*, DOI
- Fournel-Fleury, C., Magnol, J.P., Bricaire, P., Marchal, T., Chabanne, L., Delverdier, A., Bryon, P.A., Felman, P., 1997. Cytohistological and immunological classification of canine malignant lymphomas: comparison with human non-Hodgkin's lymphomas. *Journal of Comparative Pathology* 117, 35-59.

---

<sup>1</sup> See: <http://www.eu-can-lymph.net>

Fournel-Fleury, C., Ponce, F., Felman, P., Blavier, A., Bonnefont, C., Cadore, J.L., Goy-Thollot, I., Ledieu, D., Ghernati, I., 2002. Canine T-cell lymphomas: a morphological, immunological, and clinical study of 46 new cases. *Veterinary Pathology* 39, 92-109.

Gelain, M.E., Mazzilli, M., Riondato, F., Marconato, L., Comazzi, S., 2008. Aberrant phenotypes and quantitative antigen expression in different subtypes of canine lymphoma by flow cytometry. *Veterinary Immunology and Immunopathology* 12, 179-188.

Guglielmino, R., Miniscalco, B., Sarotti, D., Buracco, P., Cagnasso, A., Morello, E., 2001. Prognostic value of Ki-67 and DNA flow cytometry in canine lymphoma. *Veterinary Clinical Pathology* 30, 161.

Hamilton, T.A., Morrison, W.B., Robinson, J.P., Teclaw, R.F., DeNicola, D.B., Carlton, W.W., Hahn, K.A., 1990. Prognostic value of flow cytometric DNA analysis of canine lymphoma. In: *Proceedings of the 10th Annual Conference of Veterinary Cancer Society, Alabama, USA*, p.63.

Marconato, L., Bonfanti, U., Stefanello, D., Lorenzo, M.R., Romanelli, G., Comazzi, S., Zini, E., 2008. Cytosine arabinoside in addition to VCAA-based protocols for the treatment of canine lymphoma with bone marrow involvement: does it make the difference? *Veterinary and Comparative Oncology* 6, 80-89.

McCoy, J.P., Davis, B.H., 2001. Report of the Clinical Practice Task Force Survey of the Clinical Cytometry Society. *Cytometry (Communications in Clinical Cytometry)* 46, 177-183.

McManus P., 2008. Lymphoma in veterinary medicine: no longer a one-word diagnosis. *Veterinary Clinical Pathology* 37, 360-362.

Michels, J.J., Duigou, F., Marnay, J., Denoux, Y., Delozier, T., Chasle, J., 2003. Flow cytometry in primary carcinomas. Prognostic impact of multiploidy and hypoploidy. *Cytometry* 55, 37-45.

Petrozza, V., Verna, F., Lenti, L., Carpino, F., 2001. Prognostic value of flow-cytometric DNA analysis in breast cancer. *Rivista di Medicina di Laboratorio - Journal of Laboratory Medicine* 2, 17-21.

Pinto, A.E., Cabecadas, J., Nobrega, S.D., Mendonça, E., 2003. Flow cytometric S-phase fraction as a complementary biological parameter for the cytological grading of non-Hodgkin's lymphoma. *Diagnostic Cytopathology* 29, 194-199.

Ponce, F., Magnol, J.P., Ledieu, D., Marchal, T., Turinelli, V., Chalvet-Monfray, K., Fournel-Fleury, C., 2004. Prognostic significance of morphological subtypes in canine malignant lymphomas during chemotherapy. *The Veterinary Journal* 167, 158-166.

Riondato F., Gelain M.E., Miniscalco B., Guglielmino, R., Comazzi, S., 2006a. Utility of flow cytometry in the diagnosis of canine small cell lymphoma. *Veterinary Clinical Pathology* 35, 475-476.

Riondato, F., Gianella, P., Miniscalco, B., Maggi, E., Martano, M., Morello, E., 2006b. Flow cytometric DNA ploidy in canine tumours compared to human: preliminary data. *Cytometry* 69, 444-445.

Riondato, F., Poggi, A., Maggi, E., Morello, E., Miniscalco, B., 2010. Flow cytometric DNA ploidy and S-phase fraction in canine leukaemias. In: *Proceedings of the 20th ECVIM-CA Congress, 9-11th September 2010, Toulouse, France*, pp. 305-306.

Ross, J.S., 1996. DNA ploidy and cell cycle analysis in cancer diagnosis and prognosis. *Oncology* 10, 867-887.

Rutteman, G.R., Teske, E., Verschueren, C.P., Cornelisse, C.J., 1994. DNA flow cytometry in canine neoplasms. In: *Proceedings of the 14th Annual Conference of Veterinary Cancer Society, Tennessee, USA*, pp. 88-89.

Sözmen, M., Tasca, S., Carli, E., De Lorenzi, D., Furlanello, T., Caldin, M., 2005. Use of fine needle aspirates and flow cytometry for the diagnosis, classification, and immunophenotyping of canine lymphomas. *Journal of Veterinary Diagnostic Investigation* 17, 323-330.

Teske, E., Rutteman, G.R., Kuipers-Dijkshoorn, N.J., van Dierendonck J.H., van Heerde P., Cornelisse C.J., 1993. DNA ploidy and cell kinetic characteristics in canine non-Hodgkin's lymphoma. *Experimental Hematology* 21, 579-584.

Turner, H.E., Wass, J.A., 1999. Are markers of proliferation valuable in the histological assessment of the pituitary tumours? *Pituitary* 1, 147-151.

Valli, V.E., 2007. *Veterinary Comparative Hematopathology*. First ed. Blackwell Publishing, Ames, Iowa, USA.

Wilkerson, M.J., Dolce, K., Koopman, T., Shuman, W., Chun, R., Garrett, L., Barber, L., Avery, A., 2005. Lineage differentiation of canine lymphoma/leukemias and aberrant expression of CD molecules. *Veterinary Immunology and Immunopathology* 106, 179-196.