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UNIVERSITÀ DEGLI STUDI DI MILANO DIPARTIMENTO DI SCIENZE VETERINARIE PER LA SALUTE, LA PRODUZIONE ANIMALE E LA SICUREZZA ALIMENTARE

Evaluation of CD45 protein expression and transcript in canine small clear cell/T zone lymphoma.

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ABSTRACT

Canine small clear cell lymphoma is a peculiar lymphoma entity with T-zone histopathological pattern andindolent clinical course. From an immunophenotypic point of view the main feature is the lack of CD45 staining by flow-cytometry (FC), which accounts for >95% of cases. Underlying mechanisms have never been investigated. Aim of this work was to evaluate CD45 protein and mRNA expression in small clear cell lymphoma.

Lymph nodes of 18 cases and 11 controls, with either reactive hyperplasia or CD45-positive high grade T-cell lymphoma, were investigated. FC was performed on lymph node fine needle aspiration and CD45 median fluorescence intensity (MFI) was then evaluated on small clear cells and normal residual T-lymphocytes. CD45 surface expression was also evaluated by immunohistochemical reaction on paraffin wax-embedded lymph node sections.

Quantitative real-time RT-PCR was performed on cases and controls. Total RNA was isolated from cell suspension in RNA later. The generated CD45cDNA was amplified and $\Delta\Delta$ Ct method was used for the relative mRNA quantification.

CD45-MFI in neoplastic cells was <1% compared to normal residual T-lymphocytes in the same sample. Cells were also negative for CD45 stain on histopathological preparations. RT-PCR showed a significantly lower amount of CD45 transcript in neoplastic samples compared to controls, likely due to the residual population.

Results showed the lack of CD45 surface antigen and the virtually absence of CD45-mRNA in small clear cell lymphoma. We hypothesize a possible genomic/epigenomic aberration; further studies are in progress to investigate the pathogenesis of this aberrancy and the possible linkage to lymphomagenesis.

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