SUMMARY

Non-Hodgkin lymphomas cause over 200 thousands deaths every year all over the world. Despite a huge progress in the development and production of many new drugs and therapies, still there is a group of patients who do not respond to standard treatment. Rituximab, a monoclonal antibody to CD20, has significantly improved curability of NHL (non-Hodgkin lymphoma), however new therapeutic targets, giving more treatment opportunities, are still needed. MHC class II molecules have been considered as targets for lymphoma treatment for years. These proteins are responsible mainly for antigen presentation to T cells, but their engagement by antibody may elicit both pro-survival and pro-apoptotic signals. So far, there have been reports on a few anti MHC-II antibodies with some potential for use in human lymphoma therapy (for example Apolizumab or IMMU-14) but none of these are currently in the clinical use due to their limited efficiency or side effects in preclinical studies.

The use of biological drugs in veterinary medicine is completely different – there is only one monoclonal antibody (Lokivetmab, aIL-31) approved for use in atopic dermatitis. Lymphomas in dogs similarly like in humans are common immune system neoplasm. Due to limited possibilities of using high doses of chemotherapy canine lymphomas are considered incurable. On grounds of morphological, epidemiological and clinical similarity and of spontaneous occurrence of lymphomas in dogs, they make great model for testing new medicines and therapies or studying mechanisms in human NHL.

The aim of this work was to obtain antibodies useful in veterinary medicine as therapeutic agent and diagnostic tool. Two monoclonal antibodies B5 and E11 reactive with canine antigen DLA-DR were created and their biological properties and therapeutic potential were analyzed both in vitro and in vivo. It was shown that both of them possess direct and immune system dependent cytotoxic activity demonstrated in vitro. B5 and E11 trigger caspase-dependent apoptosis in about 40% of cells of canine cell lines expressing DLA-DR, but not in DLA-DR deficient control cell lines. B5 and E11 exhibit over 90% cytotoxicity in complement dependent assay and 10% increase in antibody mediated phagocytosis by RAW264 cells. In vivo administration of B5 or methotrexate conjugated B5 mAbs to NOD-SCID mice, xentotransplanted with a canine B-

cell lymphoma cell line (CLBL1-LUC), induced a significant slowdown of the tumor growth and spread to peripheral tissues.

B5 and E11 Mabs were also used to establish an ELISA test enabling evaluation of DLA-DR levels in biopsies and body fluids of dogs. We showed that there was a correlation between the amount of tumor cells and the level of soluble DLA-DR in blood serum of mice engrafted with canine lymphoma CLBL1-LUC. Moreover, we also observed an increased level of soluble DLA-DR in blood serum of lymphoma-bearing dogs in comparison to healthy controls.