Mutations in TP53 gene in canine mammary tumor and their clinical significance.

Human Breast Cancer is still one of the leading diseases in mortality. Although, with early diagnosis and treatment, full recovery is obtainable. Nevertheless in search for new therapy, a well fit animal model is needed. While mice and rats as models are still in use, they are provided with limitations such as differences in the tumor pathology. Dogs (*canis lupus familiaris*) were proposed as a more fitting model. This particular model is extensively studied for differences and similarities to human breast cancer and it seems to be a promising model candidate.

Breast cancer may occur due to the non-genetic factors such as exposure to carcinogens, and genetic which include ethnicity and family history of tumor occurrence. Dogs share with humans the same environment and also may develop spontaneous mammary cancer which is similar in many aspects to the human breast cancer. In every case of neoplasia, at least one mutation must occur in order to transform the normal cell into cancer cell. The most frequently mutated gene in all types of cancer is TP53. Its product, protein p53 normally functions as the guardian of the genome, protecting the cell from the transformation. If mutation in this gene occurs, tumor can develop. When breast cancer is already developed, the outcome of the disease could be predicted using the PAM50 testing. It provides the information about the molecular subtype of the cancer, which helps to determine the right therapy. The panel contains expression profiles for 50 genes related to proliferation and hormone receptors. The performance of this assay has been extensively studied and its relevance has already been proven, but only few studies tried to verify its validity in canine mammary cancer and still no clear answer has been established.

Scientific goal of our study is to describe the spectrum of mutations in TP53 in canine mammary tumors and investigate whether it is similar to that found in human breast cancers. Subsequently we will test whether mutations in p53 correlate with clinical course of the disease. Biological material, for research purposes will be acquired by a veterinarian during therapeutic resection of canine mammary tumors. Cells isolated from the sample will be tested for the TP53 mutation. Afterwards, the cells will be propagated, until there is enough material to measure such parameters as invasiveness, motility and chemoresistance for commonly used therapeutics in 2D and 3D culture (mammospheres). The results will be correlated with the clinical condition of the patient. Data on the presence of mutations in TP53 will be correlated with changes in the expression of 10 genes belonging to the PAM50 panel. Selected genes: MYBL2, CCNE1, PHGDH, CDC20, CDH3, ESR1, MAPT, FOXA1, MLPH and SLC39A6 show the greatest change in expression in the presence of the TP53 mutation in humans. Additionally, the expression level of MDM2 will be measured. MDM2 is the primary ubiquitin ligase responsible for p53 degradation and its transcription is positively regulated by p53 creating a negative feedback loop.

Scientific research, both in vitro and utilizing animal models, resulted in development of treatments that significantly improved clinical outcomes in breast cancer. In addition, recent developments in creation and analysis of large-scale clinical-molecular databases allowed for identification of molecular changes with clinical relevance. Clarification of molecular and physiological mechanisms underlying these associations is a necessary step preceding any diagnostic testing or drug development efforts. Therefore a high-quality research model is required. The data generated by this project may also contribute to the field of veterinary medicine. If the results of ex vivo treatments with chemotherapeutics used in the clinic indicate that patient’s cells are particularly resistant (or sensitive) to a certain drug, then this information may be a basis for therapeutic decisions made by the veterinarian.