Mouse Models

**Trangenic / Knockout Mouse Models (for Inflammation and Cancer)**

Recently, we generated transgenic mice that carry a targeted deletion of either the Klk5 or the Klk6 protease, in collaboration with Dr. Iacovos Michael and Prof Andras Nagy (University of Toronto, Canada). The scheme below shows the targeting cassette used for the generation of Klk5/ mice, which allows for a global or a conditional knockout.

In addition, we have generated a novel transgenic/knockout mouse (NGL Klk5/-) to monitor Nφ-κB activation *in vivo* in the whole animal (shown in the figure below). In transgenic Ngl mice a reporter luciferase gene is integrated. The Nf-kb reporter is under the control of a minimal promoter carrying eight Nf-kb consensus sequences upstream of the firefly luciferase gene. Skin inflammation can be induced by TPA following established protocols and NF-kb activity is monitored with an IVIS bioluminescence imaging under anesthesia.

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**Cancer models**

1. **Tumor xenografts in immunocompromised mice**

Our group uses SCID mice to study cancer cell growth following implantation into SCID mice. Cancer cells (either parental or genetically modified by specific gene transfer) are orthotopically injected into SCID mice and primary tumor formation is monitored for several weeks depending on tumor growth rates. At the end of the experiment, mice are euthanized and tumor progression to metastatic sites in vital organs (lung, liver, brain, bone) is recorded. Tumors and their normal adjacent tissues are resected for histological observation by E/H staining and immunohistochemistry. We employed such models to elucidate the role(s) of KLK5 and KLK6 in human breast cancer. A typical experiment is shown in the following figure.

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**Relevant References**


2. Tumor Metastasis Assay - Tail Vein Assay

Metastatic breast cancer cell lines (e.g. MDA-MB-231, MDA-MB-468) are injected via the mice tail vein, as shown in the figure below. Eight weeks post-injection visible tumors are present in the lungs (and other vital organs) of the injected mice, which are excised, quantified, and biopsied to assess metastatic disease.

3. Chemical carcinogenesis in transgenic mice

We use knockout models to delineate the role of proteases in cancer development. To investigate the possible role(s) of KLK5 and KLK6 proteases in skin cancer we employ chemical carcinogenesis in Klk5<sup>-/-</sup> and in Klk6<sup>-/-</sup> mice. Skin tumors are induced by two different schemes: DMBA/TPA and DMBA/DMBA. Our goal is to find new KLK-mediated pathways that are associated with the different stages of tumorigenesis, i.e. initiation, promotion, and metastatic progression. Since chemical carcinogenesis in mice is an inflammation-driven process it is expected to reveal the inflammatory mechanisms implicated in skin cancer. In this direction we also apply two well-establish systems of chemical-induced inflammation, i.e. the irritant and the allergic contact dermatitis, in order to further elucidate the role of KLKs in skin inflammation. Finally, we also employ the model of methylcholanthrene-induced fibrosarcomas to delineate the role of KLKs in this type of cancer.