MEF-IGF1-Rrep mouse model

REACTION BIOLOGY

IN VIVO TESTING SERVICES

➢ IGF1-R

IGF1-R is a type II tyrosine kinase receptor consisting of two extracellular α-subunits, two transmembrane β-subunits, and an intracellular tyrosine kinase domain. It is ubiquitously expressed and has multiple functions. However, IGF1-R over-expression transforms mouse 3T3 fibroblasts, and high levels of IGF1-R expression have been detected in human tumors, making it a bona fide target for the development of novel anti-cancer treatment.

➢ MEF-IGF1-Rrep

MEF/Toff/IGF1-R cells were generated by stable transfection of MEF/Toff cells (Clontech, Heidelberg, Germany) with the plasmid pTRE-IGF1-R-2myc6his and pSV-hygro. Clones were selected with hygromycin and subclones were selected by picking clones that form colonies when grown in softagar (Graeser et al., unpublished). The cells express the human IGF1-R cDNA under the control of a Tet-inducible promoter. IGF1-R is expressed in the absence of doxycycline and expression is inhibited in the presence of doxycycline.

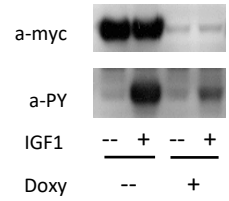
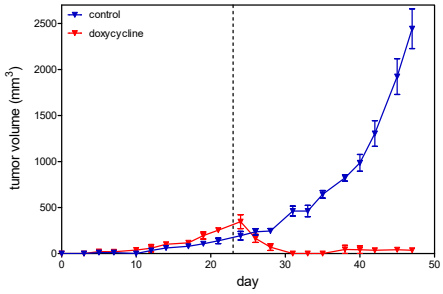


Figure 1:
Characterization of the cell line. The cells were kept either in the presence or absence of the regulator, doxycycline, and IGF1 (100 ng). IGF1-R expression was checked via its myc-tag, and autophosphorylation using an anti-phosphotyrosine antibody.

➢ Tumor growth *in vivo*

MEF-IGF1-Rrep cells harvested from tissue culture flasks were implanted into the subcutaneous space of the left flank of the mice. At an average size of 400 mm3, the mice were randomized into two groups, and one group received doxycycline in the drinking water. As shown in Figure 2, the repression of IGF1-R expression induced by the treatment resulted in tumor regression.



➢ Immunohistochemistry

As shown in figure 3, the addition of doxycycline to the mice in the drinking water ablates exogenous IGF1-R expression in the tumors.

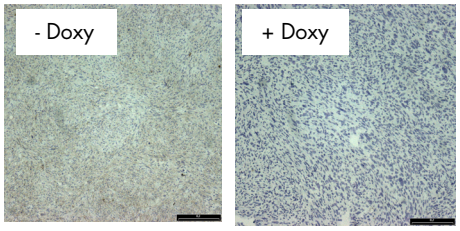


Figure 3: *Immunohistochemistry of MEF-IGF1-Rrep tumors.*
On the left, a tumor of an untreated mouse is shown. The tumor on the right panel was extracted from a mouse treated with doxycycline.
IGF1-R positive cells are brown, the nuclei are counterstained with hematoxylin (blue).