

P002 Analysis Of Knock-in Mice For A Constitutively Active Lipid Kinase Form Of PI3K γ

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PI3K γ activity is triggered by the activation of G protein-coupled receptors. To study the in-vivo effects of a constitutively activated PI3K γ , we decided to generate a knock-in mouse strain expressing a membrane bound PI3K γ . For this purpose we used a mutant PI3K γ containing the K-Ras CAAX box and showing a constitutive lipid kinase activity *in vitro*. Two recombinant ES cell clones gave germ line transmitting chimeras. Homozygous animals, where the Neomycin resistance cassette was removed by Cre-mediated recombination, appeared viable, fertile and did not show increased incidence of tumorigenesis. Peritoneal macrophages derived from PI3K γ -CAAX homozygous mice normally expressed the mutant form of PI3K γ . Moreover, macrophages derived from PI3K γ -CAAX homozygous mice displayed basal levels of Akt/PKB phosphorylation higher than wild-type controls, but were still able to increase Akt/PKB phosphorylation after chemokine stimulation. Despite these findings, PI3K γ -CAAX homozygous showed several phenotypes typical of mice completely lacking PI3K γ . For example, the thymus weight in PI3K γ -CAAX homozygous mutants was 45% smaller than in wild-type mice. In addition, knock-in mice showed reduced levels of peritoneal recruitment after peritonitis induction. Further studies will be focused to understand the mechanism underlying these apparently contradictory results. As cells with increased PI3K γ activity could lose directed movement, a first attempt would be to study the chemotactic response of knock-in leukocytes.