B2

Utilizing Fluorescent Transgenic Reporters to Trace the Metastatic Progression of Pancreatic Neuroendocrine Carcinoma

Sean T. Glenn[1]; Calvin Sjaarda[2]; Ping Liang[2]; Karstin Webber[1]; Michael Rusiniak[1]; Caretta J. Reese[1]; Maxwell M. Goodrich[1]; Heinz Baumann[1]; Dominic J. Smiraglia[1]; Kenneth W. Gross[1]

[1]Roswell Park Cancer Institute, Buffalo, NY 14263
[2]Brock University, Ontario, Canada

Background: We have generated a unique mouse model of highly metastatic islet cell carcinoma by selectively abrogating floxed alleles of p53 and Rb using Cre-recombinase driven by the renin promoter. Incorporation of a multi-colored fluorescent reporter, Confetti, confers the ability to trace the lineage of individual renin-expressing cells allowing for observation of hyperplasia within pre-tumorigenic islets, clonality of primary tumor foci, and identification of the primary of origin for observed metastases.

Methods: RenCre activation of the Confetti reporter, randomly labels each cell with one of four fluorescent reporters in concert with concurrent homozygous deletion of Rb and p53. Clonal expansion of the rare cells that progress to primary tumors as a function of additional cooperating mutations results in color-coded primary tumors. Moreover, metastases are also color-coded and matched as clonal derivatives of the independent primary tumors in the same mouse. Next-Generation Sequencing technology is used to study genetic events responsible for progression to metastatic disease.

Results: Dissection of animals of appropriate genetic constitution reveals that most mice harbour multiple primary tumors of different fluorescent color. Remarkably, in the subset of animals that developed metastatic disease, all observed liver metastases are a single color suggesting they derive from only one of multiple primary tumors in each case (Fig 1). Whole Exome Sequencing of DNA from multiple sets of primary and metastatic tumors from individual animals has identified a mutation profile which allows for the tracing of a single primary responsible for seeding distant metastases in the liver, and confirms what is predicted by the reporter. A deeper investigation of both exome and transcriptome data will offer up insight into genes responsible for metastatic progression of disease.

Conclusions: Our utilization of multiple fluorescent reporters for cell lineage tracing of pancreatic renin-expressing cells, is providing unique insights into pancreatic islet carcinogenesis and metastasis.

Figure 1

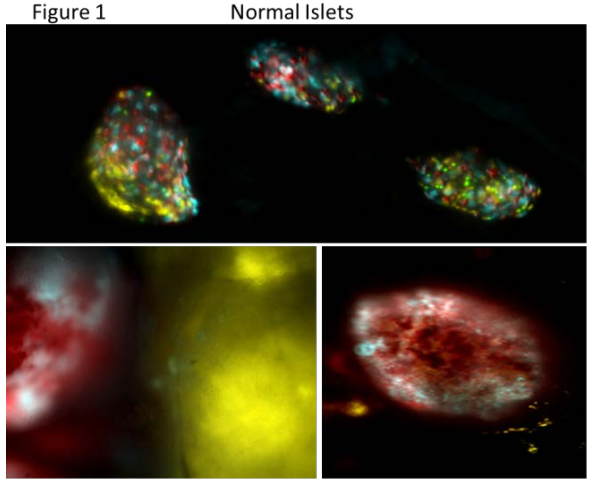

Fig1. Upper panel: confetti labeling of lineal descendants for renin-expressing cells in pancreatic islets. Lower left: Primary pancreatic tumors (2) tagged with confetti. Lower right: Liver metastasis labeled red/blue (all mets in liver same color). Derived from red/blue primary