



Predicting rapamycin response using pathway profile in a population of genetically engineered HER2 driven breast tumor model

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Abstract (#3615)

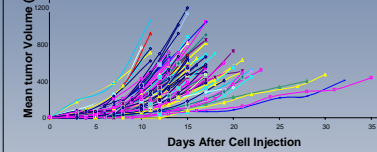
The limited utility of traditional preclinical models of human cancer in guiding clinical development of anti-cancer agents and the significant variation in response seen even among subpopulations of human tumors, warrants development of pathway based models that better represent the genetic diversity and as a result the response variation in human patient populations. In an effort to more accurately model human HER2 amplified breast cancer populations, we have established a platform of *in vivo* genetically engineered tumors in which defined genetic alterations (inducible mutant *hher2* transgene (V84E) on an *lnk4a/ARF-* background) are combined with spontaneous and naturally arising variation to create populations of *in vivo* tumors suitable for preclinical drug response testing.

Utilizing a chimeric approach, we have generated over 100 murine breast adenocarcinomas engineered to combine HER2 overexpression and *lnk4a/ARF-* knockout. Each of these primary tumors was then expanded and propagated *in vivo* to generate an archive of frozen tumor material suitable for *in vivo* drug testing. Tumors from these archives were found to show significant variation in growth rate, stromal involvement, angiogenesis, response to taxotere, and microarray profile. Using a novel computational approach we assessed the coordinated variation in gene expression among genes involved in various biological functions and generated expression based indices that revealed variation in several signal transduction pathways, including mTOR pathway signaling. The mTOR signaling index enabled us to make a prospective prediction that a subset of HER2 driven breast tumors should be sensitive to *in vivo* rapamycin treatment. Upon testing the select tumors with Rapamycin for tumor growth inhibition, we were able to confirm that the PI3K index we created could indeed be used to predict responsiveness in our murine model of human breast cancer. Variation in the same mTOR signaling index is observed in human breast cancers providing a hypothesis for predicting rapamycin response in subsets of human breast cancer. Thus the current platform that we have generated in this work, could be used effectively as a better predictive model for testing known or unknown cancer drugs in order to prospectively predict drug responses.

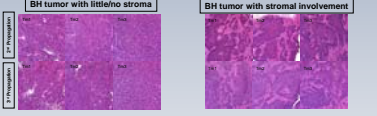
Natural variation in Inducible Breast Her2 Tumors

Natural variation in the growth profile of ~107 murine inducible Breast Her2 tumors

N = 5 individual tumors for each tumor line implanted subcutaneously from archived material



Primary tumor dependent variation in the histopathology of BH tumor cohorts



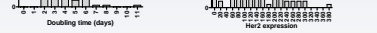
Histological variations are stably inherited through 3rd propagation

Diversity in the tumor doubling time, Her2 expression, pAKT and stromal involvement among archived ~107 murine inducible Breast Her2 tumors

Doubling times across tumor set



hher2 expression by TaqMan



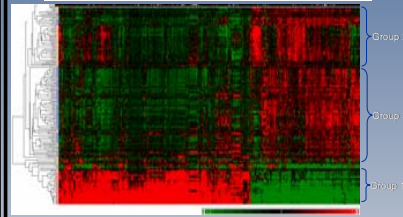
pAKT staining signal intensity



Variation in stromal involvement



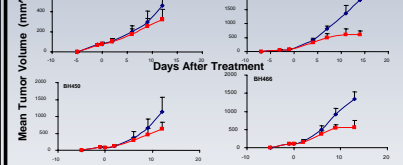
Expression profile analysis using microarray of 107 murine inducible breast Her2 tumors reveal diversity among these tumors



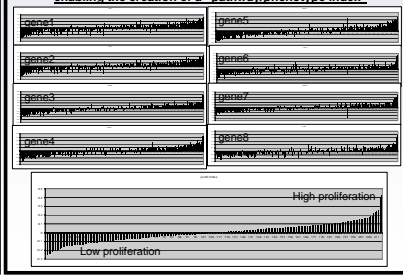
*Biological reproducibility observed by duplicates clustering together (55/107)

Variation in efficacy for taxotere treatment among ~30 tumors tested from murine inducible breast Her2 tumor model

N = 10 individual tumors for each study arm implanted subcutaneously from archived material

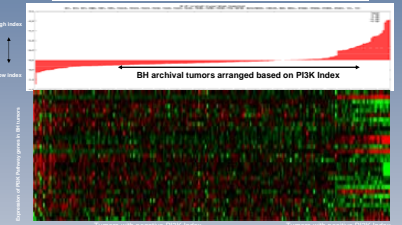


Expression level of co-correlated genes varies across tumors enabling the creation of a "pathway/phenotype index"

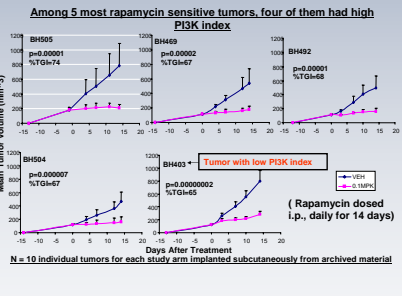


Validation of Rapamycin Response Prediction

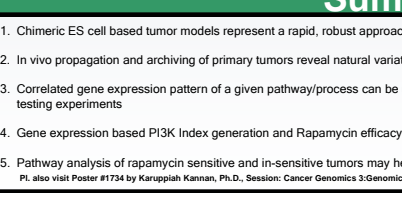
Archived 107 BH tumors can be ranked based on PI3K pathway activation assessed through gene expression



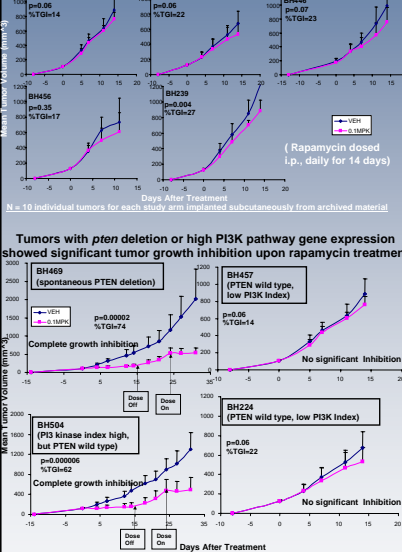
All of the five rapamycin resistant tumors showed low/negative PI3K index



Among 5 most rapamycin sensitive tumors, four of them had high PI3K index

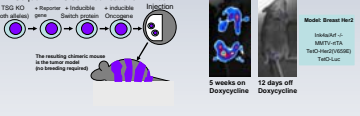


Tumors with pten deletion or high PI3K pathway gene expression showed significant tumor growth inhibition upon rapamycin treatment



Chimeric Inducible Models

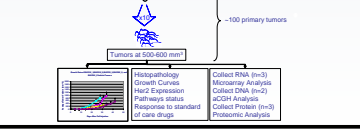
Complex Models Can Be Generated Rapidly - Inducible Breast Tumors (HER2)



In vivo Propagation of primary tumors creates stable "archive"



Characterization of Breast Her2 Model Tumors to determine diversity



Summary

1. Chimeric ES cell based tumor models represent a rapid, robust approach for generating complex inducible tumors with precise genetic alterations
2. In vivo propagation and archiving of primary tumors reveal natural variation and also enable the quantitative comparison of drug effects *in vivo*
3. Correlated gene expression pattern of a given pathway/process can be utilised to identify and select tumor sub-populations for prospective drug testing experiments
4. Gene expression based PI3K Index generation and Rapamycin efficacy study may validate the use of this model for response prediction studies
5. Pathway analysis of rapamycin sensitive and in-sensitive tumors may help us in identification of more robust predictive biomarkers

Pl. also visit Poster #1734 by Karupiah Kannan, Ph.D., Session: Cancer Genomics 3:Genomic Profiling III, Monday, April 14th 8AM-12Noon, Exhibit Hall B.F, Section 4, Board#22