Cluster analysis of human and mouse HCC gene expression profiles with survival genes

Our previous study uncovered gene expression signatures of selected "survival genes" that could distinguish between the poor and better prognosis groups[1]. To assess the utility of human survival genes in our mouse models, we analyzed the gene expression patterns of the survival genes in the mouse HCC. Out of the 406 survival genes identified in the human study, 144 orthologous genes were presented in mouse HCC gene expression data. Hierarchical cluster analysis of survival genes from the integrated data also revealed the close relationship of *Myc*, *E2f1*, and *Myc/E2f1* mouse models to human subclass B HCC, and of *Myc/Tgfa* mouse models to human subclass A HCC (**Supplementary Fig. 2**). HCC from *Acox1*$^{-/-}$ mice were well separated from all of the human tumors, indicating again that these mouse tumors had the least similarity to human HCC. These results indicate that a limited number of genes (survival genes) that best represent clinical phenotypes of human HCC are sufficient to determine which mouse models have highest relative similarity to the human HCC.

Quantitative RT-PCR

We assessed the reliability of gene expression measurements in our microarray study as well as our method for standardization of gene expression data by applying quantitative real-time RT-PCR. The relative amount of expressed mRNAs in both subgroups in human and mouse HCC correctly matched with the microarray-based measurement (**Supplementary Fig. 5**). This result not only confirms that the microarray-based measurements of gene expression are reliable, but also supports that the standardization method we applied is adequate to select genes whose relative expression patterns are conserved during tumorigenesis in both species.

1. Lee, J.S. et al. Classification and prediction of survival in hepatocellular carcinoma by gene expression profiling. *Hepatology* **40**, 667-76. (2004).