

Toxicoepigenomics

Transcriptional and epigenetic profiles of primary liver cells and *in vitro* models

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HepaRG cells, unlike the widely used HepG2 cell line, might be a promising candidate for toxicological studies, considering its ability to maintain the activity of many liver-specific factors in a two-dimensional culture system. In this study we compared genome-wide DNA methylation and RNA expression patterns of HepG2 and HepaRG cells to that of freshly isolated as well as seven days cultivated primary human hepatocytes obtained from four donors (PHH.7d.cult) (Fig. 1). In a second step, selected differentially methylated loci identified by Illumina Bead Chip arrays will be subjected to high-resolution bisulfite profiling using the 454 GS-FLX Titanium platform.

Fig. 1: Experimental Pipeline

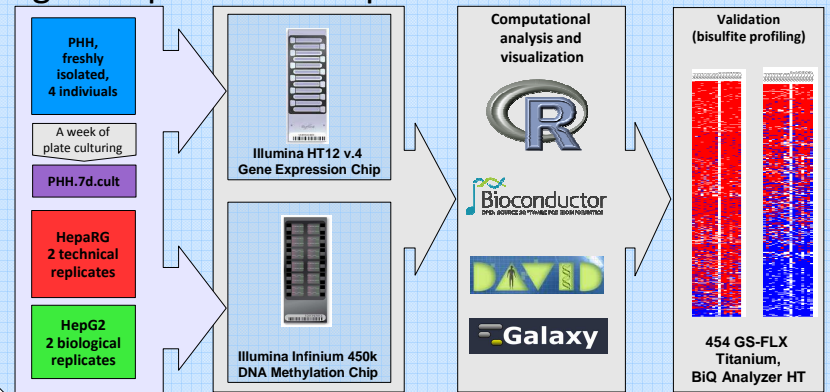
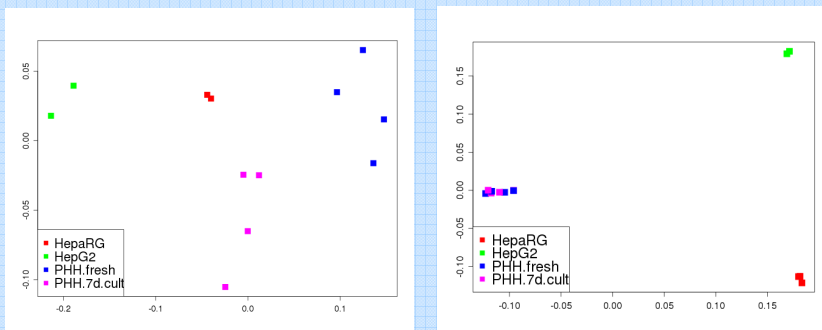


Fig. 2: Exploratory Analysis

A. Gene Expression

B. DNA Methylation



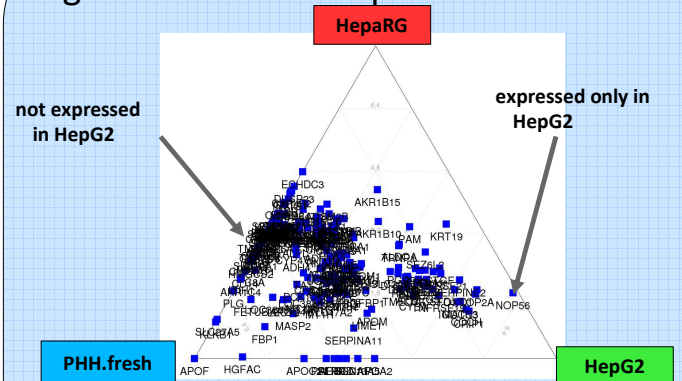
The plots were generated via Multidimensional Scaling of a Pearson correlation similarity matrix

Multidimensional scaling shows that the transcription profile of HepaRG is more similar to that of freshly isolated and cultivated PHH samples than the HepG2 cells (Fig. 2A). The cultivated PHH differed significantly from freshly isolated PHH. PHH individual variation was rather high. In contrast, the DNA methylation profiles formed compact clusters (Fig. 2B). Differential methylation analysis yielded thousands of CpGs significantly different between the cell types, while only very small differences were seen between cultivated and fresh PHH, suggesting that short term culturing has moderate effects upon DNA methylation.

We refined the set of differential transcripts to more than two hundred of high-confidence targets with an expression change of an order of magnitude or higher (Fig. 3). The dominating bulk of the differential transcripts can be classified as being specifically expressed or silenced in HepG2.

Finally, we applied correlation analysis to establish correspondence between the observed transcriptional and DNA methylation changes. An obvious tendency towards anti-correlation was observed for the top-scoring differential transcripts suggesting that DNA methylation changes may be associated with the observed gene expression changes.

Fig. 3: Differential Expression



219 differential transcripts with changes over an order of magnitude