Gene Chip® Human Genome U133 Plus 2.0 Array (Affymetrix)

Acute Lymphoblastic Leukemia Phase II and Xenografts (ALL P2 & ALL MDLS) – Gene Expression

*Protocols performed at the University of New Mexico.

cRNA for hybridization to U133_Plus_2.0 arrays was performed according to Affymetrix's recommendations (GeneChip Expression Analysis Technical Manual). First, 300 ng of total RNA was converted to cDNA. Biotinylated cRNA was generated from the cDNA and 15 µg was subjected to fragmentation. Either the Affymetrix One-Cycle Target Labeling Kit or the Affymetrix 3' IVT Express Kit was used. This v02 labeling protcol differs from v01 because the labeling kit changed. Affymetrix changed the IVT kit between 2008 and 2009. While most of the gene expression patterns remain the same, there are some pronounced differences that may result in set effects when trying to merge data generated from the different labeling kits.

Hybridization of 12.5 µg fragmented biotinylated cRNA was performed according to Affymetrix's recommendations (GeneChip Expression Analysis Technical Manual).

Scanning of the microarrays was performed according to Affymetrix's recommended protocol (GeneChip Expression Analysis Technical Manual).

Data were masked according to the method outlined in Harvey et al, Blood 116:4874-4884, (2010) in order to remove uninformative probe pairs. Default MAS 5.0 normalization was performed on the masked data using Expression Console software (Affymetrix).

The non-collapsed GCT file is simply the masked MAS 5.0 data from the CHP files formatted as a GCT file. Level 3 data were generated by using the CollapseDataset algorithm of GenePattern: http://www.broadinstitute.org/cancer/software/genepattern/. (link is external) In applying this software, the "maximum" (as opposed to "median") probeset setting was used, and the gene-to-probeset associations were obtained from the file AFFYMETRIX.chip downloaded from ftp://gseaftp.broadinstitute.org/pub/gsea/annotations/.