JAX: expression - hu133,

Molecular Methods Description:

• Tissue samples were stored in RNAlater (Ambion) per manufacturer's instructions for later homogenization with TRIzol (Life Technologies) using the GentleMacs dissociator (Miltenyi). Total RNA was isolated using the TRIzol(R) Plus Kit (Life Technologies) according to manufacturer's methods including on the column DNase digestion. * Following reverse transcription with an Oligo(dT)-T7 primer (Affymetrix, Santa Clara, CA), double stranded stranded cDNA was synthesized with the GeneChip Expression 3'-Amplification One-cycle kit (Affymetrix, Santa Clara, CA). In an in vitro (IVT) reaction with T7 RNA polymerase, the cDNA was linearly amplified and labeled with biotinylated nucleotides (Affymetrix). * Ten micrograms of biotin-labeled and fragmented cRNA were hybridized onto U133 Plus 2.0 GeneChip arrays (Affymetrix) for 16 hours at 45 C. Post-hybridization staining and washing were performed according to manufacturer's protocols using the Fluidics Station 450 instrument (Affymetrix). * The arrays were scanned with a GeneChip(TM) Scanner 3000 laser confocal slide scanner.

Analysis Description:

The arrays were processed with the AffyPLM R package, using quantile normalization, no background correction, and fitting to a simple model that treats the log Intensity as a sum of array effect, probe effect, and residual. The array effect is the normalized_expression that is equivalent to the median polished value produced by standard RMA analysis.