

AGEMAP Data Read Me File

The following header rows are present in each of the data files, for each of the samples. The data from the 2 different membranes have been combined in to one file for each tissue totaling 16,896 cDNA's.

^SAMPLE = 01_AMC185_M_43_A

The Sample Name includes a sample number, 01-67 as well as a mouse ID number, AM... which includes sex, age in days at time of sacrifice, and diet.

!Sample_source_name = Mus musculus Adrenals

!Sample_characteristics = Strain: C57BL/6, Tissue: Adrenals

!Sample_characteristics = Gender: male, Age: 43 days, Diet: Ad lib

!Sample_treatment_protocol = 100% regular feed, ad libitum until age of sacrifice

!Sample_molecule = total RNA

!Sample_extract_protocol = Qiagen RNeasy Mini Kit protocol for animal tissues after disruption using a mini-bead beater-8 and 1.0mm glass beads.

!Sample_label = 33P-dCTP

!Sample_label_protocol = ~5 ug of total RNA was radiolabeled with 33P-dCTP using Superscript II from Invitrogen. Labeled cDNA was separated from unincorporated probe and small nucleotides using a Bio-Rad size exclusion column, Bio-Spin 30 with SSC. 1ul of each sample was counted in a scintillation counter and the remainder was used for hybridization.

!Sample_hyb_protocol = Radiolabeled cDNA was denatured at 95 degrees C for 7 minutes and added to 12 mls of Invitrogen microhyb containing 10% dextran sulfate with Cot-1 DNA (Invitrogen) and Poly-A DNA oligos, (Sigma) added as blocking agents. Arrays were hybridized overnight at 55 degrees C to mouse 17K-A cDNA filters. The hybridized filters were quickly rinsed once and then washed twice at 65 degrees placed onto steel plates covered with plastic wrap and exposed to Kodak storage Phosphorimaging screens for 3-4 days.

!Sample_scan_protocol = Imaging screens were scanned at 50um resolution in a Storm Phosphorimager.

!Sample_data_processing = All data was extracted using ArrayPro Software and processed in Excel spreadsheets using Z normalization of each array. Differently expressed genes were identified between the different sample groups by z-ratio analysis using an in-house microarray analysis program.