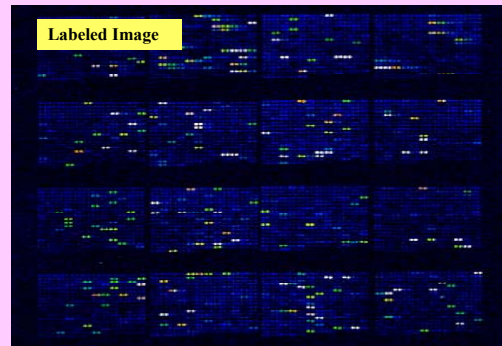


Quality Monitoring on Human 14K Oligo Chips at the University of Calgary

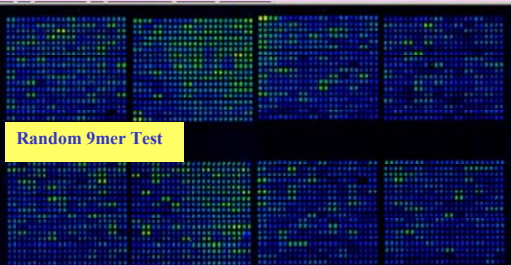
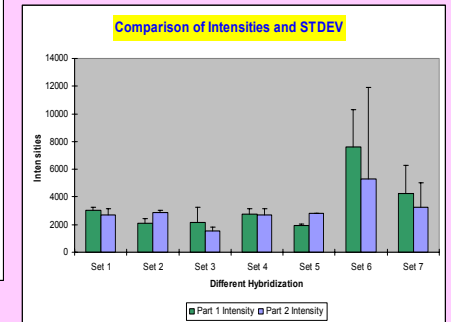
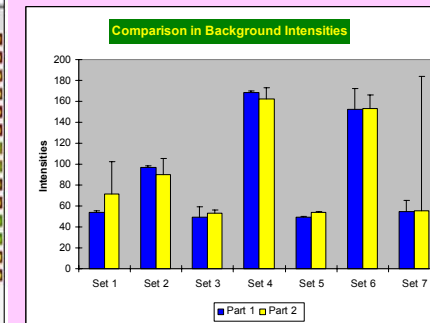
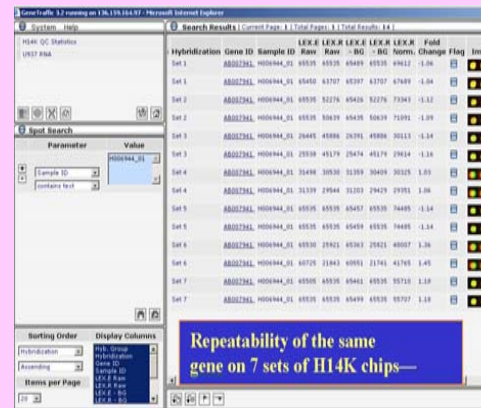
Xiuling Wang, Stephen M. Robbins, Mayi Arcellana-Panlilio, Southern Alberta Microarray Facility,

The University of Calgary, 3330 Hospital Drive, N.W., Calgary, Alberta T2N 4N1 Email: xiuwang@ucalgary.ca Tel: +1(403) 210-3858

The production of microarrays needs to be carefully monitored and quality control tests done to track systematic effects and performance variations over time to maximize the possibility of getting high quality data.

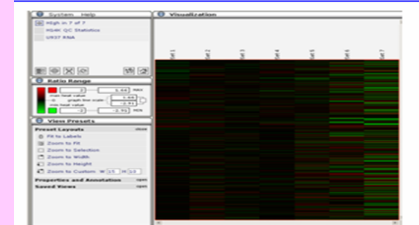


Labeling Efficiency: We use U937 cell-line RNA to test array quality after each print run. Seven such sets of H14K chips have been analyzed to compare the consistency and variation from different production dates and different operators. Scanned images were quantified using *QuantArray* and further analysis was done with *Gene Traffic*. The variations from different printing dates and operators are compared by background and foreground intensities and presence of biological functional genes in each

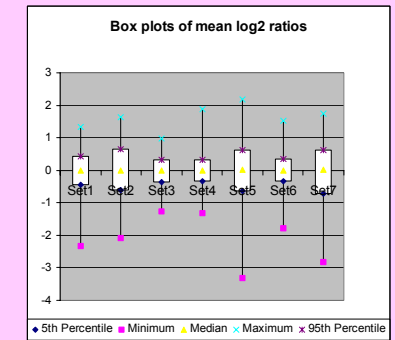
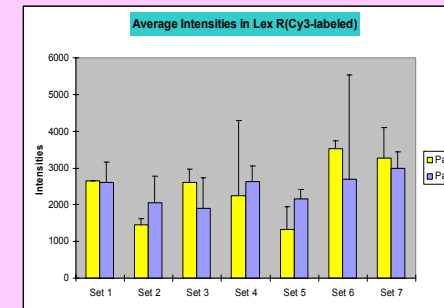


Morphology: We test the quality of each printing run with a random Cy3-labeled 9mer hybridization to evaluate spot morphology (size, circularity, uniformity).

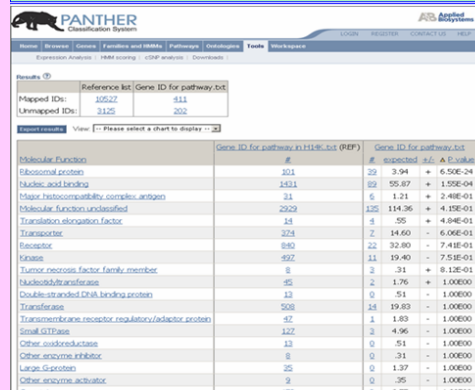
Variations by Operators



Since we have been using same cell line RNA samples labeled with Cy3/Cy5 to test the label efficiency on each print, it is reasonable that we don't have higher or lower ratio (fold changes). But there are intensities differences between the two different operators, set 6 and 7 show brighter and higher intensities, especially in Cy3-label channel.



Consistency in H14K Chips



Biological Function	Gene ID for pathway in H14K-chip (PEP)	#	Gene ID for pathway in H14K-chip (PEP)	#	expected z-score	A-E value
Ribosomal protein	201	39	3.94	6.50E-24		
Nucleic acid binding	1431	89	55.87	1.95E-04		
Transmembrane protein	11	6	1.21	2.48E-01		
Other enzyme	508	14	19.49	7.53E-01		
Transmembrane receptor/regulator/adaptor protein	42	1	1.83	1.00E00		
Small GTPase	127	3	4.96	1.00E00		
Other oxidoreductase	13	0	.51	1.00E00		
Other enzyme inhibitor	6	0	.31	1.00E00		
Large G-protein	25	0	1.37	1.00E00		
Other enzyme activator	9	0	.35	1.00E00		

There are over 500 genes showed higher intensities in all sets analyzed which related to the biological functions of U937 cell line.

Expression Information:

Box plots of the mean log₂ ratios show that 90% of the mean ratios fall within a tight range for each of the sets. The min and max whisker lines encompass the outliers. A total of 36 spots showed fold changes higher than 3 in the 7 hybridizations. After excluding dust and single spots, only 4 pairs of genes remained. On the other hand, 186 spots showed fold change less than -3. Over half (95/186) of these spots were from set 6 and set 7, which may reflect operator variability.

Conclusions:

- Morphology parameters remain consistent from print to print.
- The same genes (>500) are labeled with high intensity. These genes are related to the biological functions of the U937 cell line.
- Ninety percent of the mean log₂ ratios stayed within a tight range for each of the hybridizations. However outliers were present.