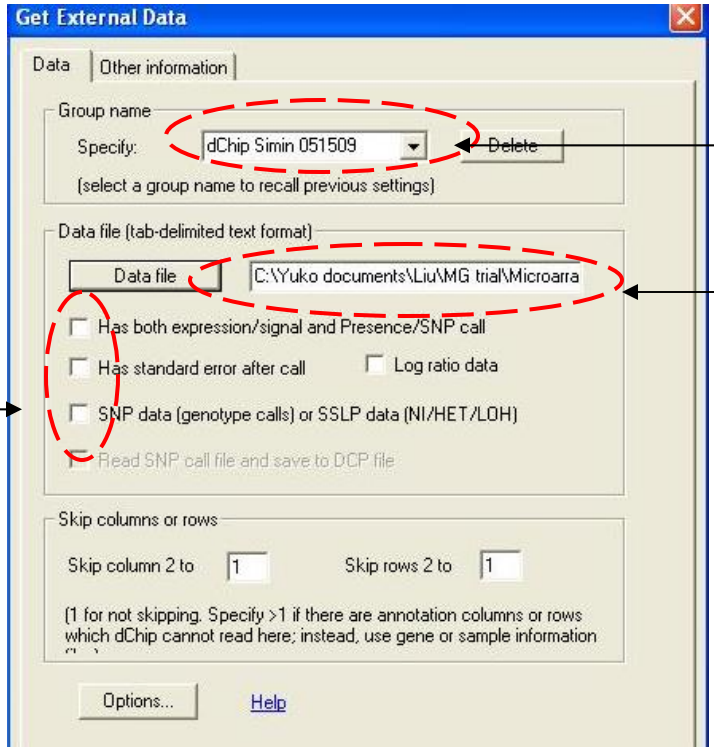
Program of Genomic and Nutrition Laboratory
UCLA Department of Epidemiology
Prepared by Yuko You (yuko@ucla.edu)
05/18/09

Procedures for Microarray Analysis (Mg Trial)

Background information for microarray chip analysis in Mg Trial:
Chip: Affymatrix Human Gene 1.0 ST arrays (exon array)
Instrument: Agilent 2100 DNA analyzer
Specimen: RNA

0. Getting the dCHIP program (Free) : http://biosun1.harvard.edu/complab/dchip/

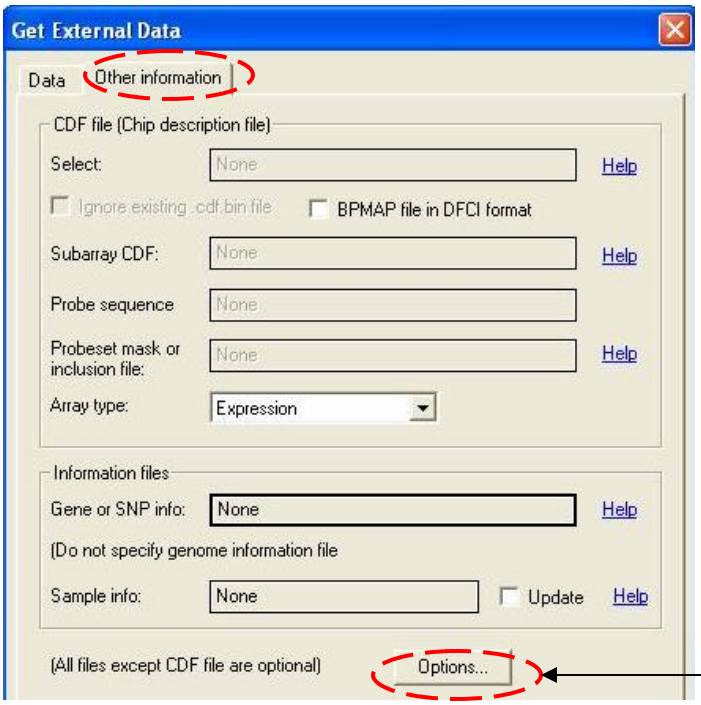
1. Open data file and option setting



Define group name

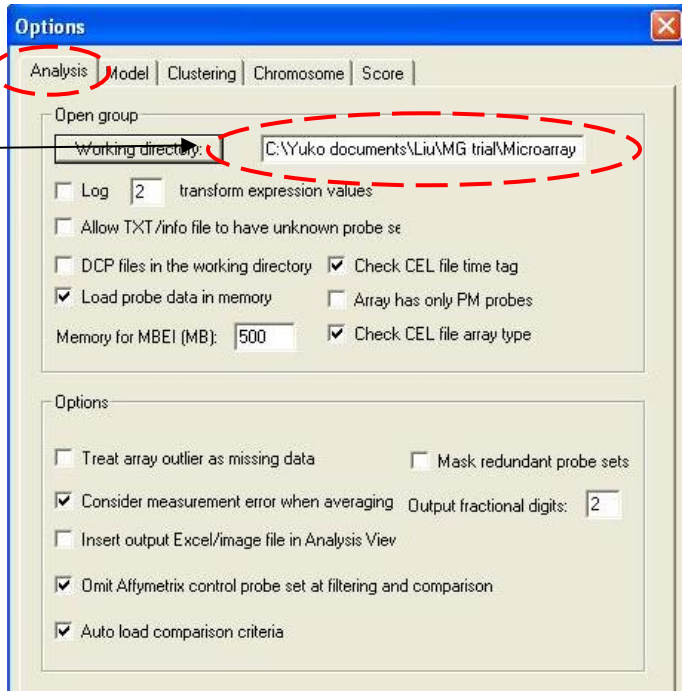
Location of the master data file

Uncheck these 3 options



Press here for the next step

↓

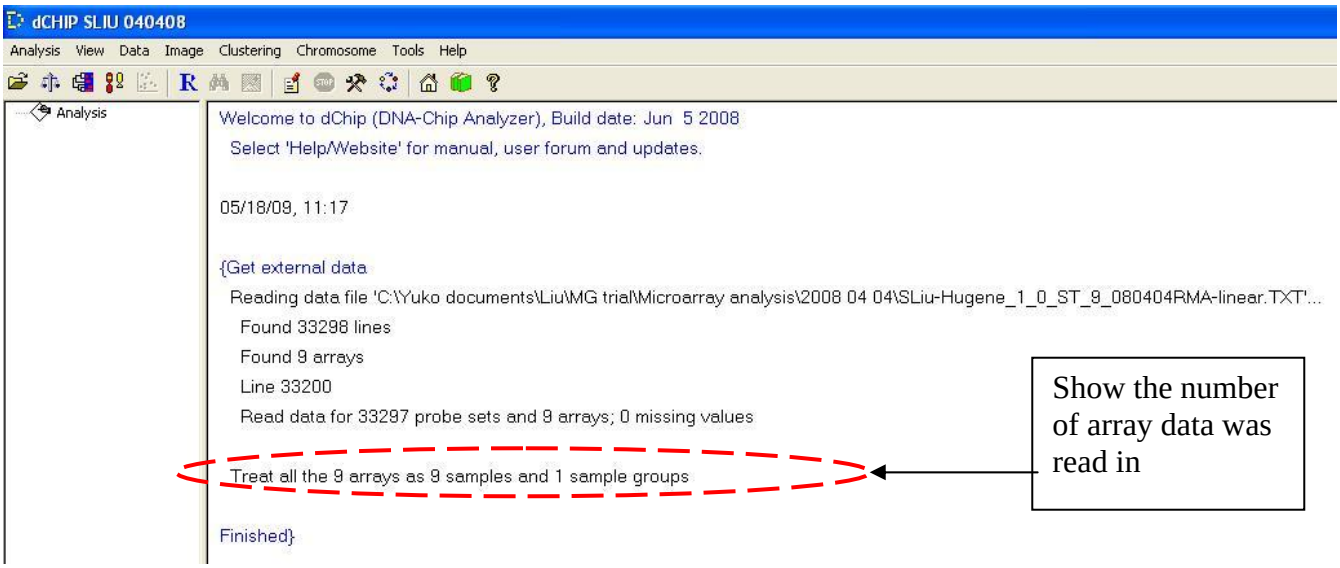


Set up the "working directory" for output files

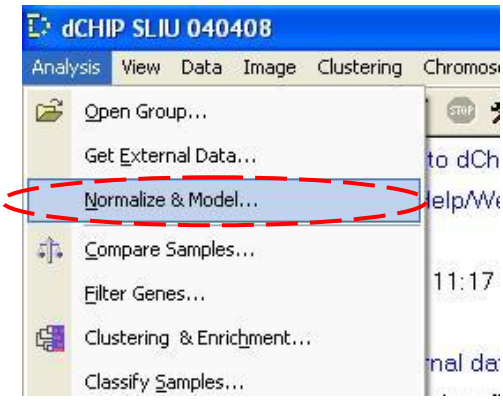
↓

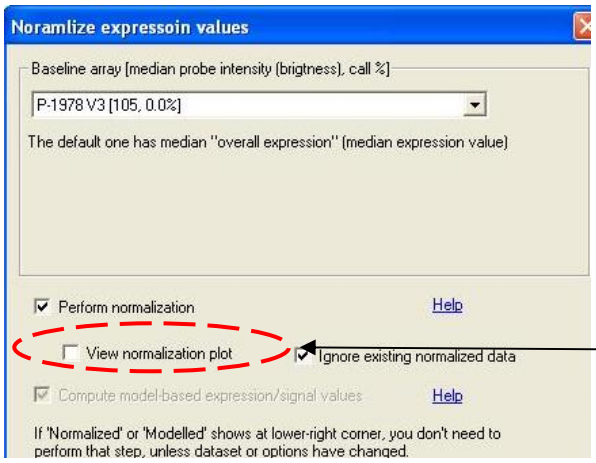
Press "ok" to close all pop-up windows

↓



2. Data analysis





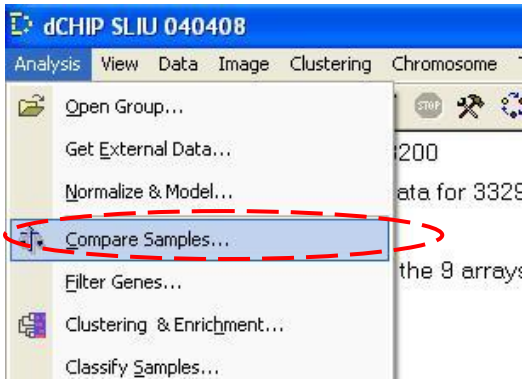
{Normalize arrays using probes of 'Invariant set' and 'Running median' smoothing
Normalize expression values using the Invariant Set method...

Baseline array is P-1978 V3

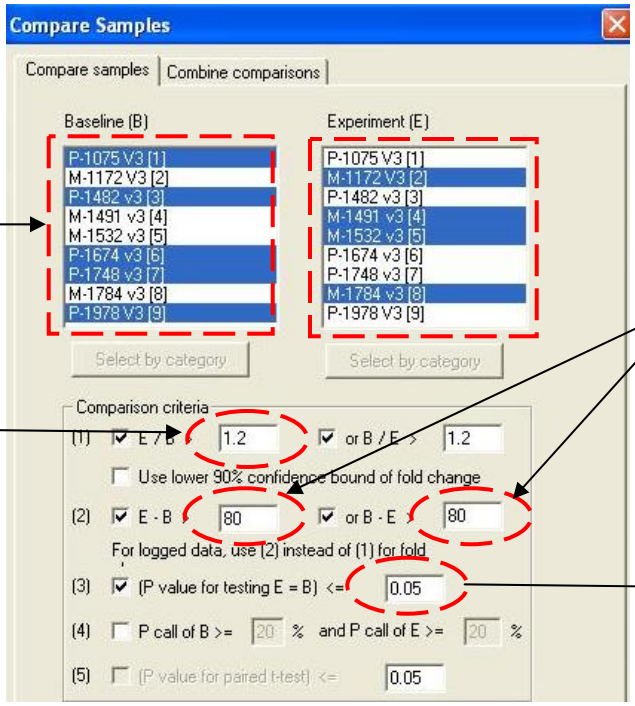
Array P-1075 V3
Searching Invariant-set: 6262
Normalizing arrays...
:
:
Array M-1784 v3
Searching Invariant-set: 6635
Normalizing arrays...
Finished in 00 hours 00 minutes 06 seconds}

3. Comparison

press 

or 

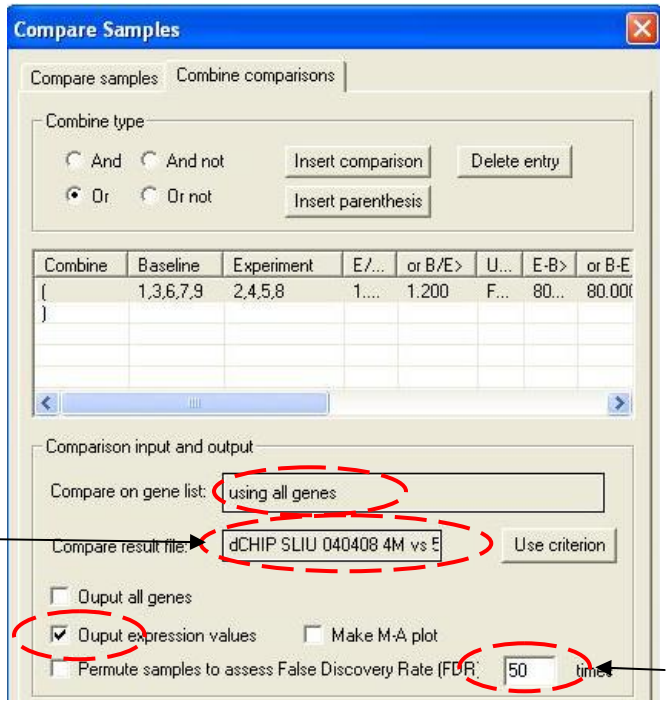
Select compared samples in the "baseline (placebo)" and "experiment (intervention)" by holding CTRL



Effect size cut-offs, ex. 1.2= 1.2 fold change (Dr. Li set 1.2 because expect Mg has low effect on expression

Expression level absolute difference (intensity). Ex. 160/60, effect size is 2.66, differences is 100

Can use p=0.1 for less restrict results or p=1 for all genes fits the first 2 criteria.



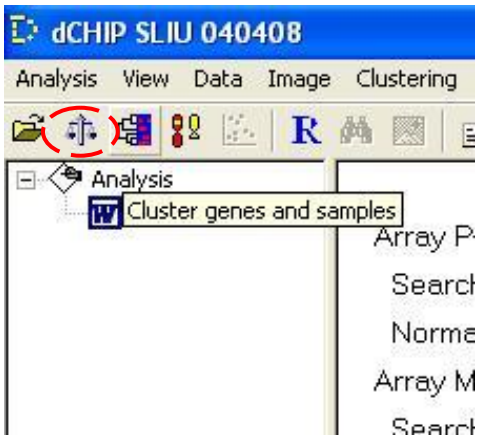
Output file name suggest: "dCHIP SLIU 040408 4M vs 5P at 1.2 fold -80 p005 051809" in this case. Suggest to change the file name according to the selecting criteria

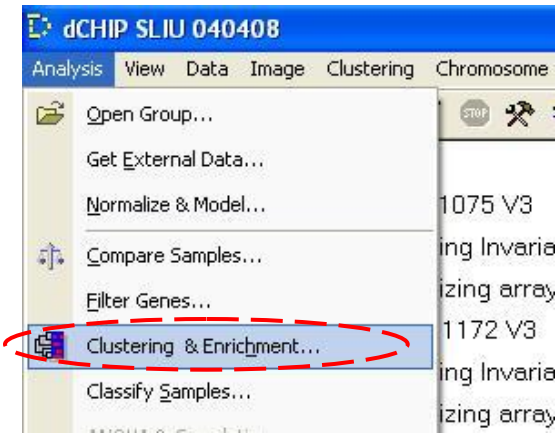
Use it for manuscript

{Compare samples
Writing comparison result in 'C:\Yuko documents\Liu\MG trial\Microarray analysis\2008 04 04\Output\dCHIP SLIU 040408 4M vs 5P at 1.2 fold -80 p005 051809' (file pathname is in clipboard)...
23 genes satisfied the comparison filtering criteria

Finished}

4. Cluster and enrichment

press 

or 

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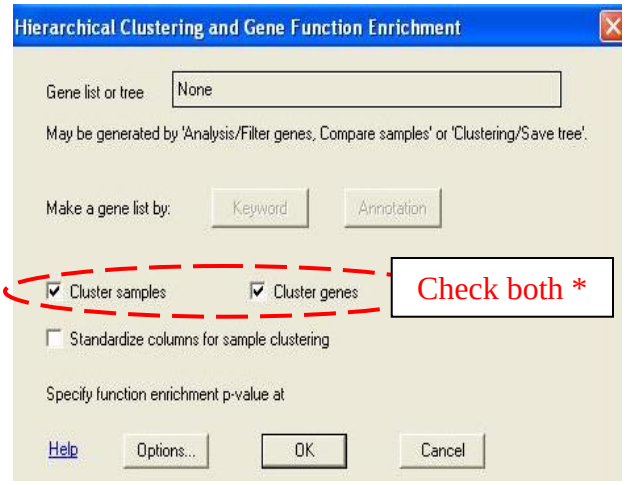
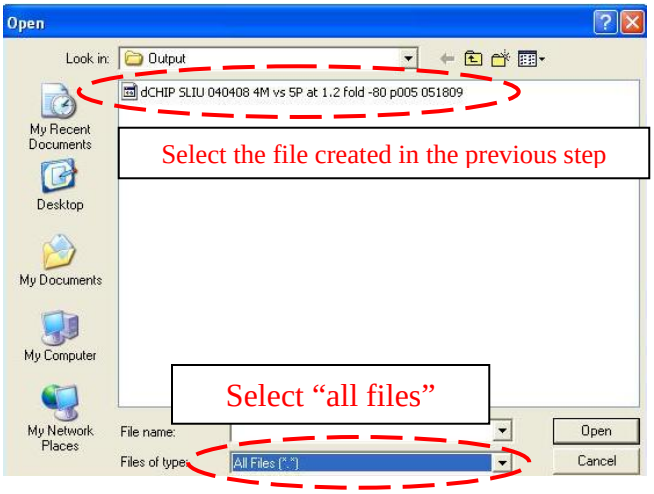
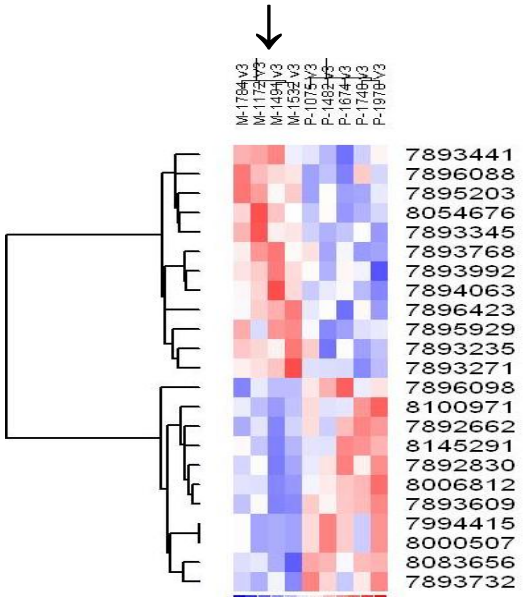
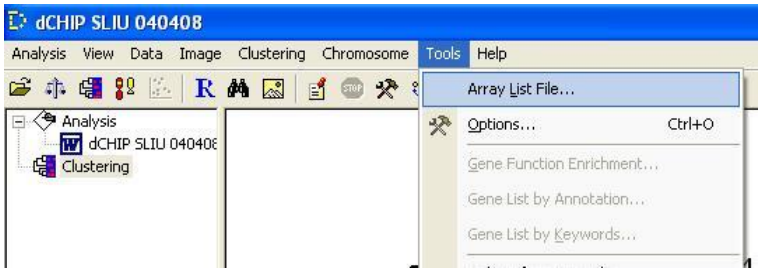


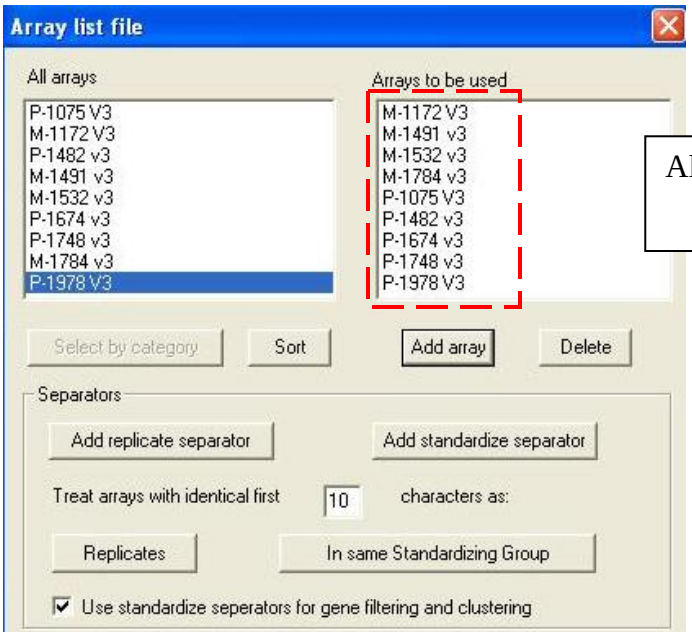




-Use ←↑→↓ keys to change image size.
-if replace the "probe set" to "gene symbol" in the master data file, in this graph will show the gene names on the righthand side instead of probe ID

If not satisfy with the sample align, then do the following steps

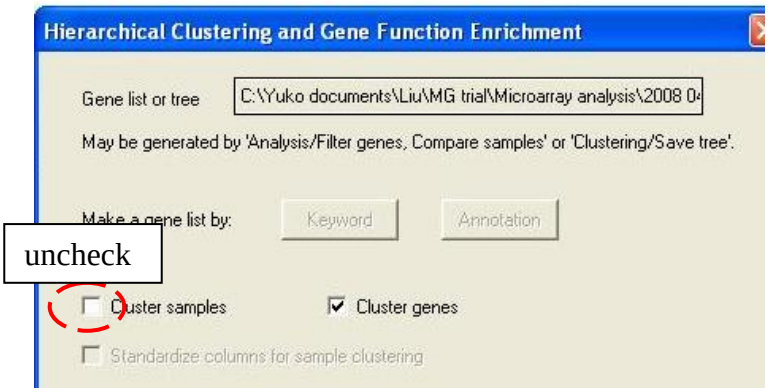




Align sample ID as needed

↓

Go back to "Cluster & Enrichment"



uncheck

And the samples will be arrayed as needed.

6. Check the result file

Go to the output folder (as assigned in the "working directory" in step 2) to find the file and open using Excel with the selected gene list.