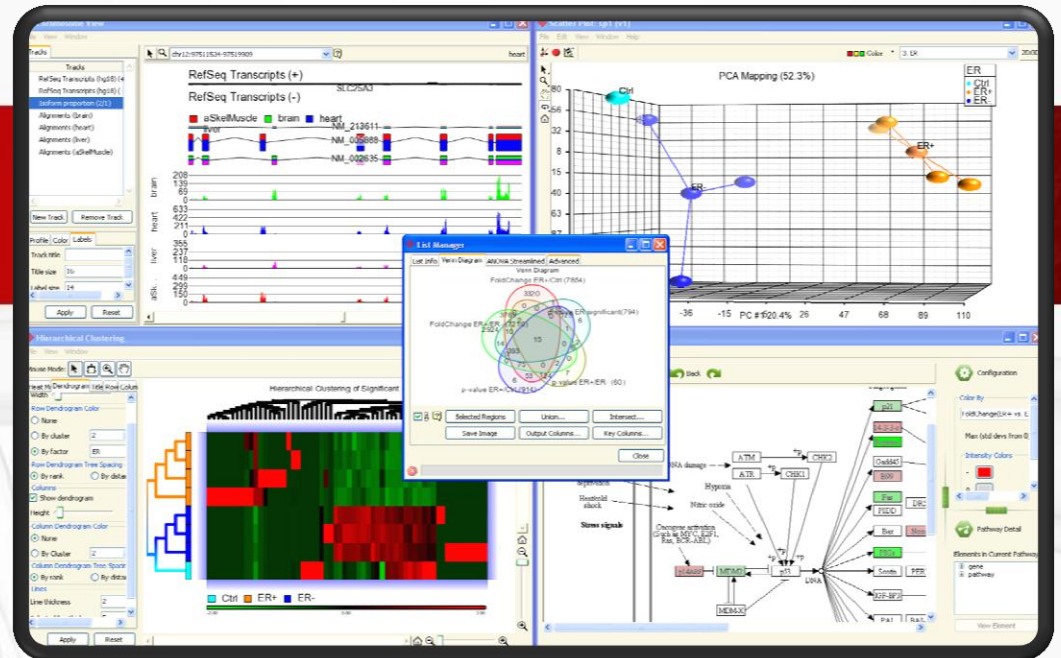


Comparison of Affymetrix PrimeView with U133 Plus 2.0 using PAN Two-cycle Amplification Process and Partek® Genomics Suite Analysis



Objective

- To compare the overall performance between U133 Plus 2.0 array and PrimeView array
- To compare the performance of small input material (1 ng and 10 ng) using the PAN facility's Two-cycle Amplification Process

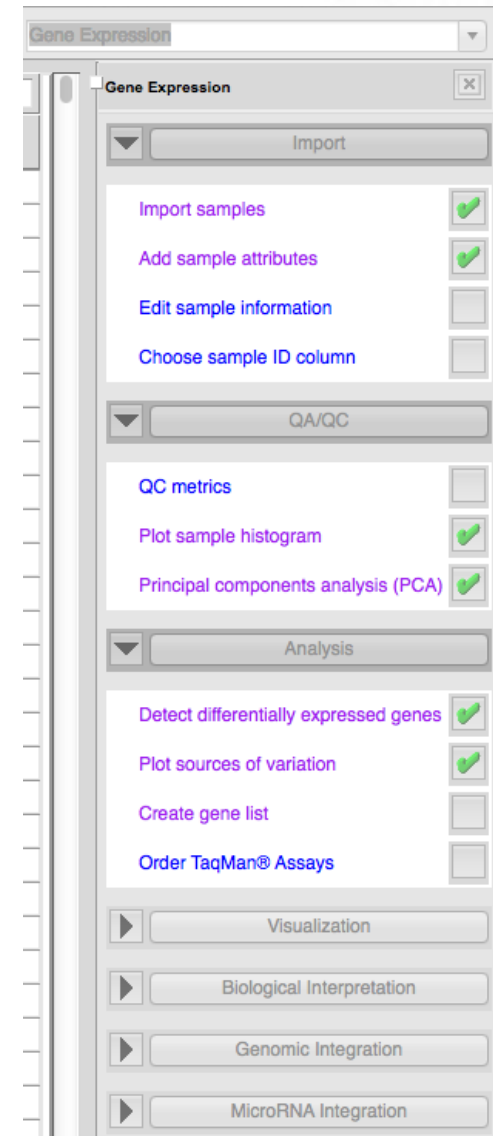
Summary of findings

- U133Plus 2.0 array performance is similar to the PrimeView.
- No gene's were found to be significantly different between the two array types using standard statistical cut offs
- Applying the PAN two cycle amplification process the 1ng and 10ng input performed similarly on both array types
- No genes were found to be significantly different between the two input amounts

- Sample: Control RNA
- Method: PAN Two-cycle Amplification Process
- 4 arrays
 - PrimeView 1ng
 - PrimeView 10ng
 - U133P2.0 1ng
 - U133P2.0 10ng
- Comparisons
 - Primeview vs U133P2.0
 - 1ng vs 10ng
- Analysis was performed using Partek Genomics Suite

Methods in Partek Genomics Suite: Gene expression workflow

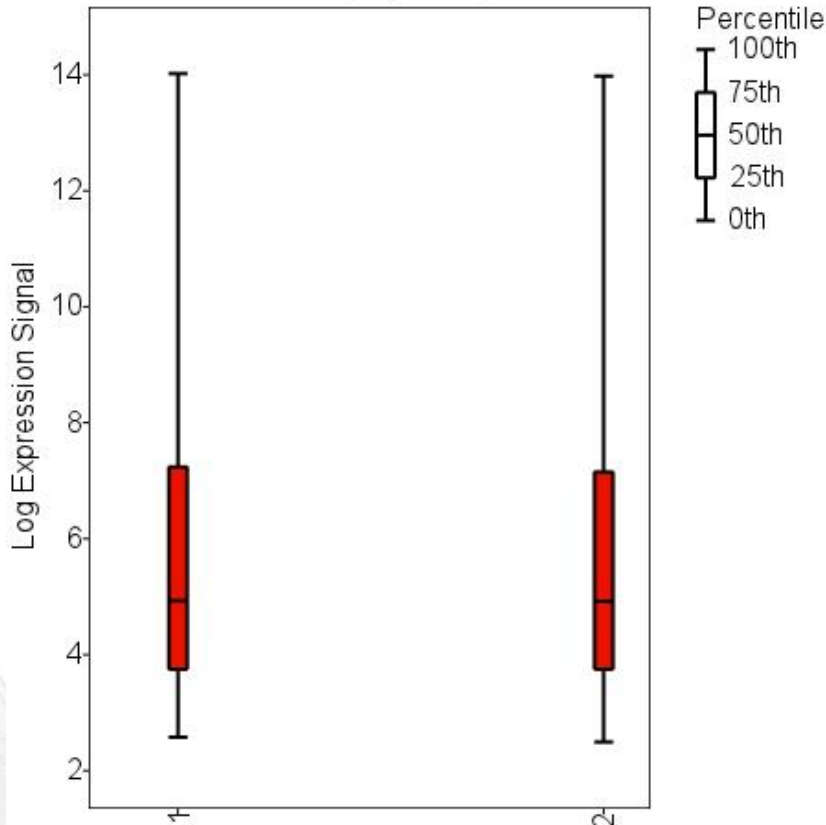
- Four Affymetrix CEL files were imported
 - PrimeView with 1ng and 10ng starting material
 - U133 Plus 2.0 with 1ng and 10ng starting material
- Summarization with RMA
- Compared signal intensity and distribution
- Data was merged and compared at the gene level
- Determine correlations between samples
- Detect differentially expressed genes
 - Genes that appear would indicate a false positive



QA/QC – Intensities (Log based 2)

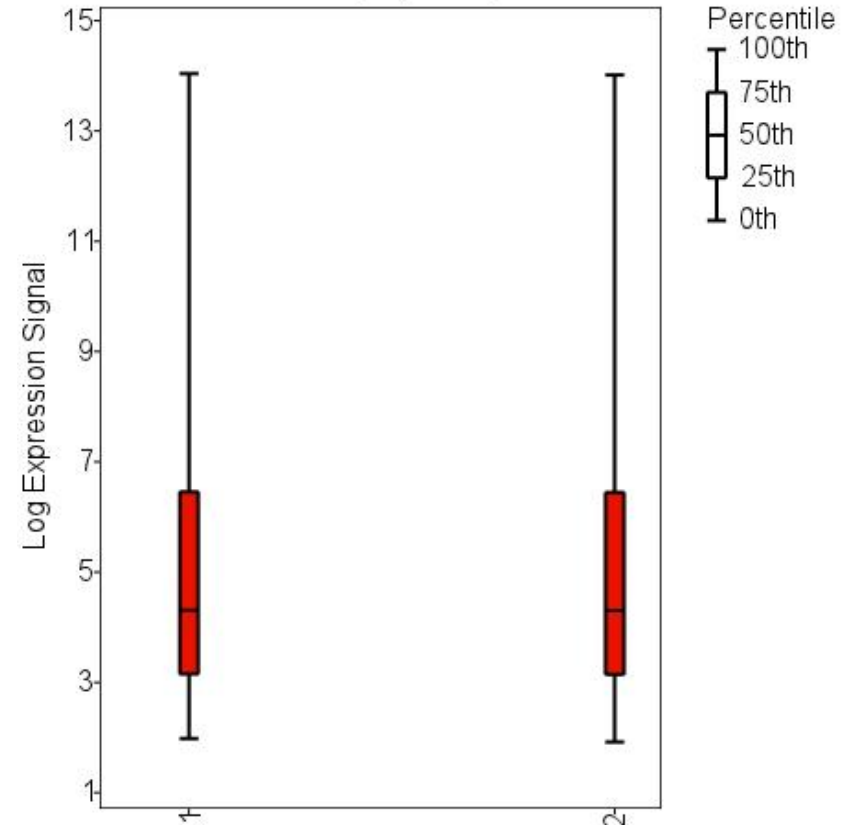
PrimeView

Box Plot on Log Expression Signal



U133 Plus 2.0

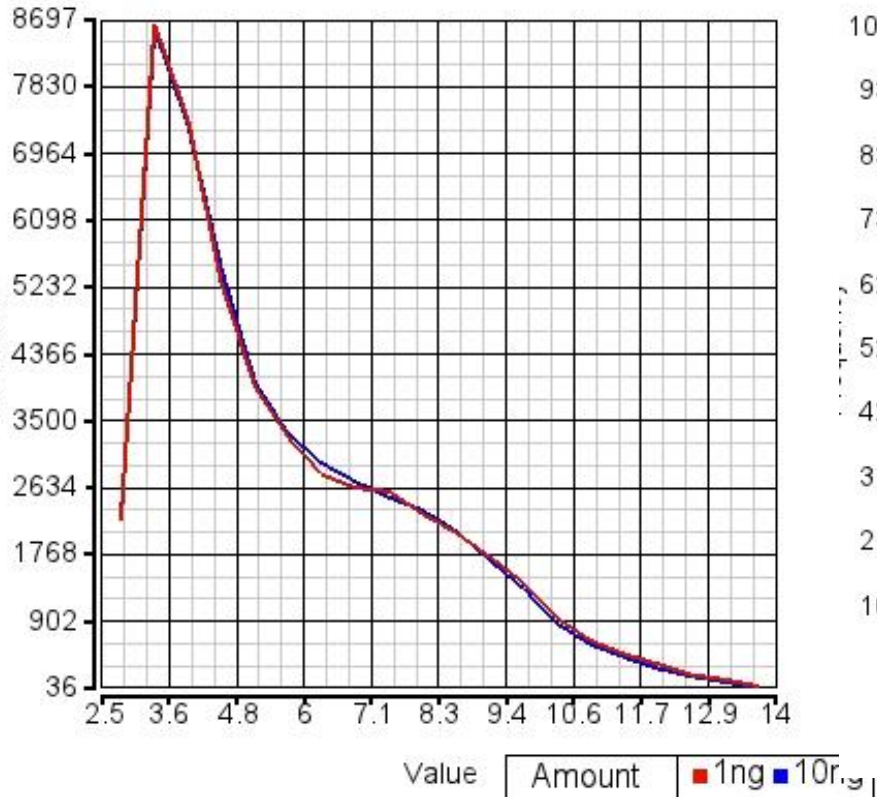
Box Plot on Log Expression Signal



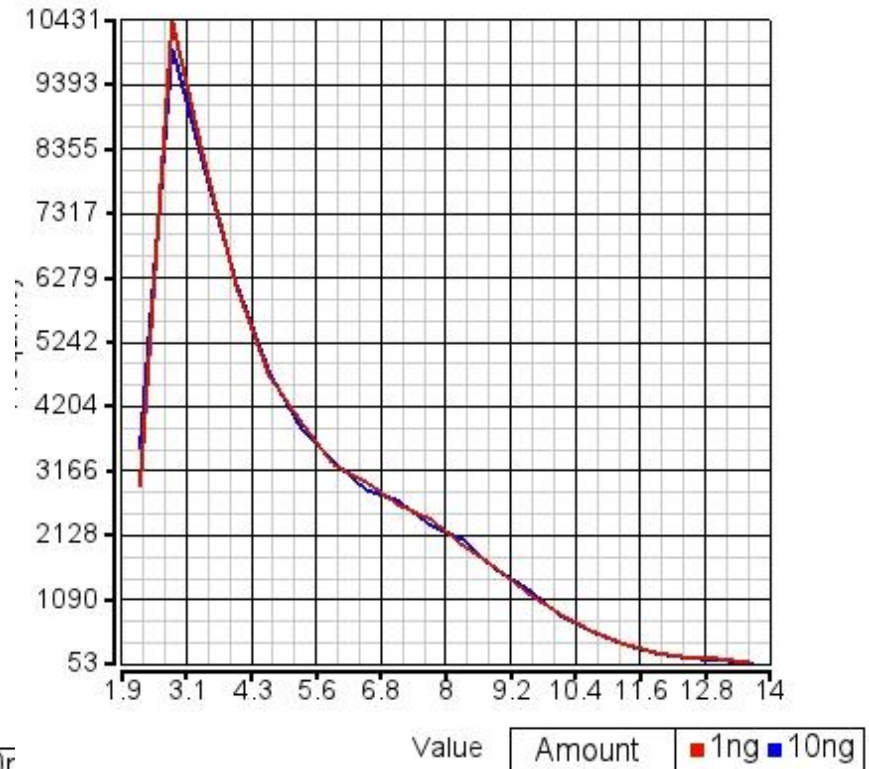
No difference in signal intensity between 1ng and 10ng
No difference in signal intensity between PrimeView and U133 Plus 2.0

QA/QC – Histograms

PrimeView



U133 Plus 2.0



Similar intensity distribution

Comparing by Gene Symbol

PrimeView

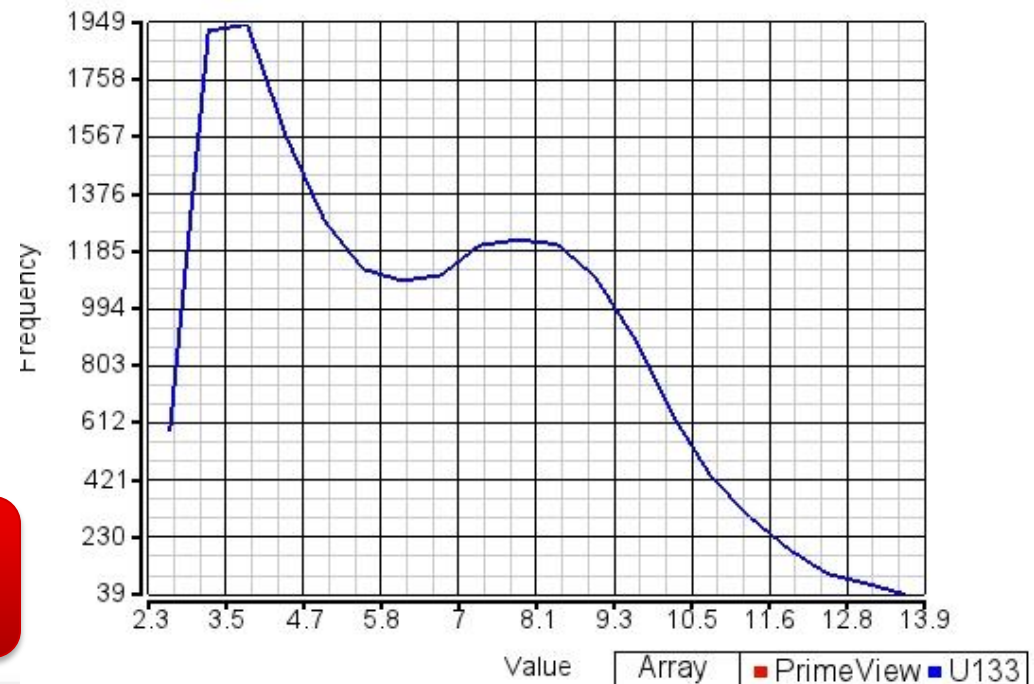
- 49395 Probe sets
- RMA summarized
- 20970 Gene Symbols

U133 Plus 2.0

- 54675 Probe sets
- RMA summarized
- 19909 Gene Symbols

PrimeView plus U133 Plus 2.0

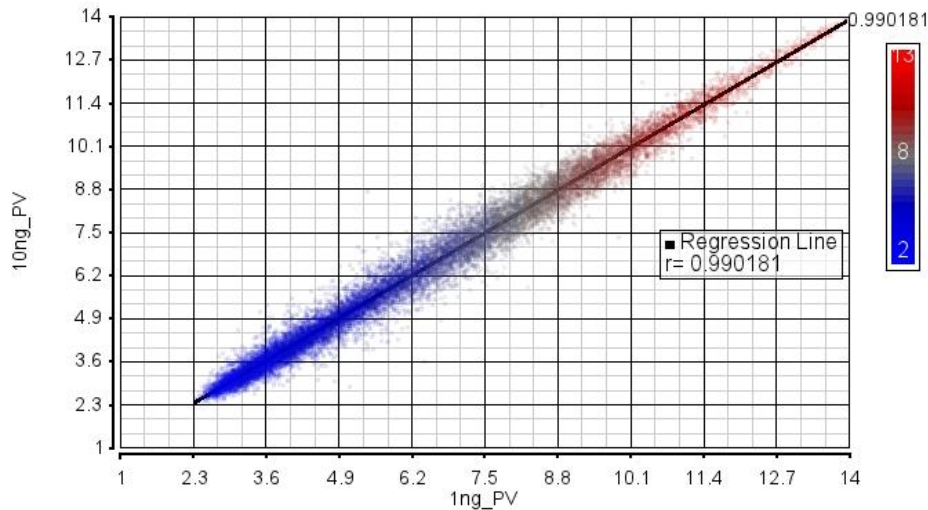
- 18081 common Gene Symbols
- Quantile normalized



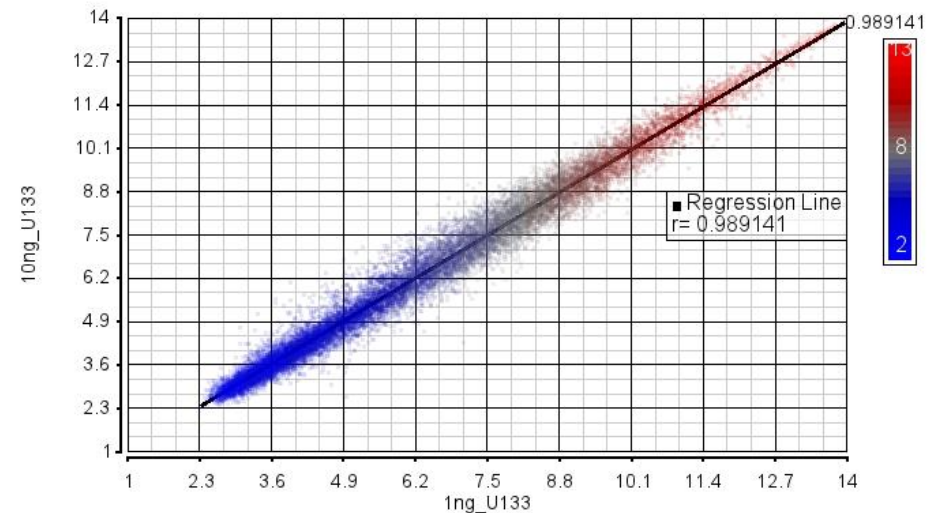
All four samples overlap

Correlations between different input amounts

Correlation PrimeView 10ng_vs_1ng



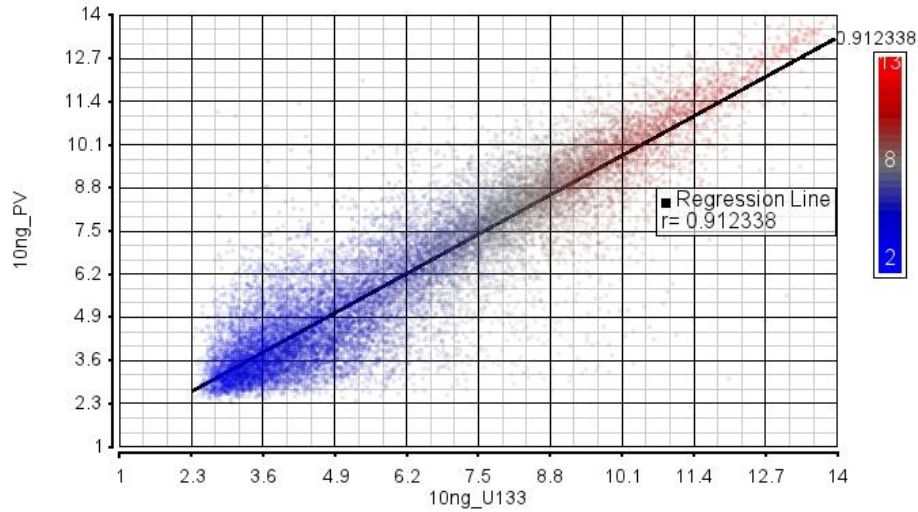
Correlation U133 10ng_vs 1ng



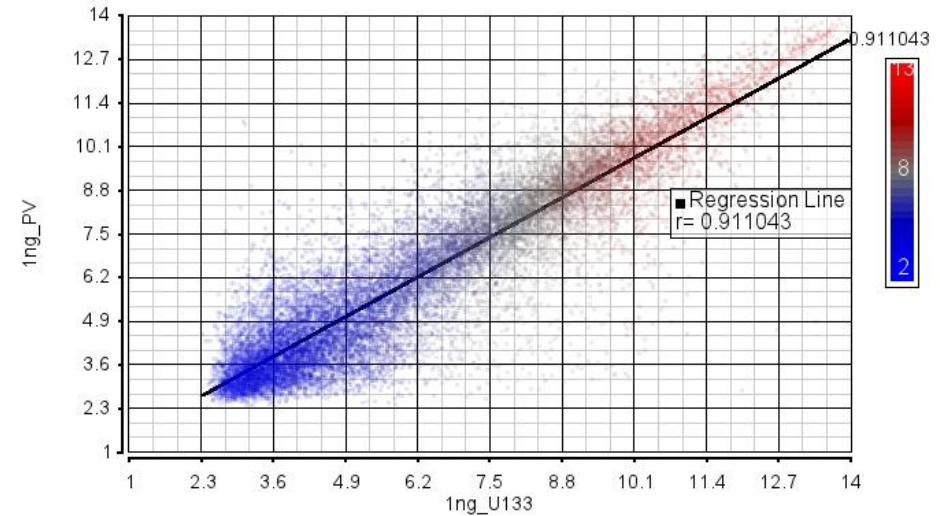
Very good correlation between the different starting amounts (10ng and 1ng)
Very similar performance with PrimeView and U133 Plus 2.0

Correlations between different Arrays

Correlation 10ng PrimeView_vs_U133



Correlation 1ng PrimeView vs U133



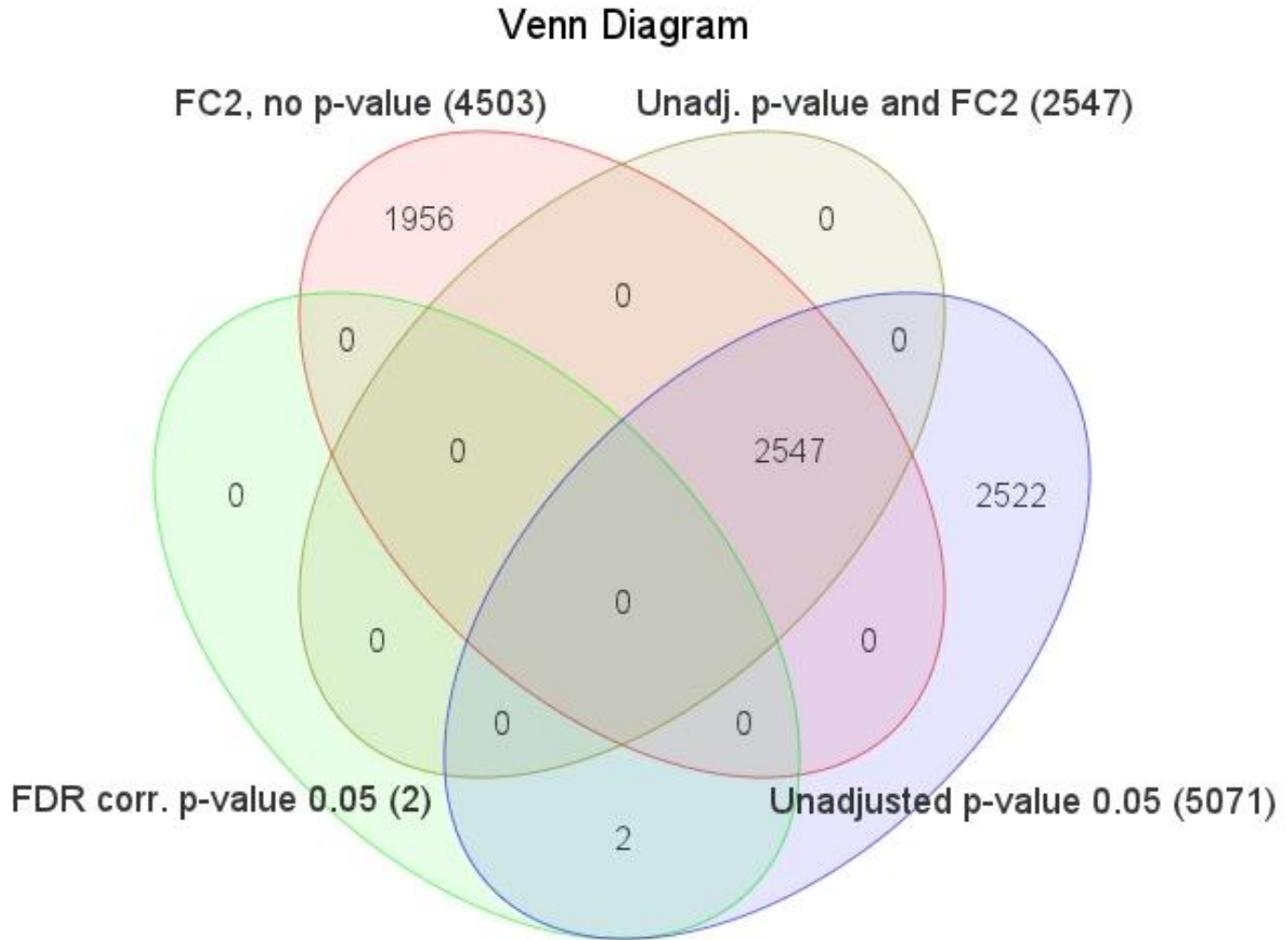
High correlation between PrimeView and U133 Plus 2.0 for both 10ng and 1ng input

No significant genes found between array type or input amount

Differential Expression Calculations

- ANOVA was run based two factors: Array type and starting amount
- Comparisons made:
 - 10ng vs 1 ng
 - PrimeView vs U133 Plus 2.0
- Significant genes found would be considered false positives
 - **Based on p-value (FDR corrected 0.05):**
 - **10ng vs 1ng: 0 genes**
 - **PrimeView vs U133 Plus 2.0: 0 genes**
 - Based on unadjusted p-value (0.05) and fold change higher abs.2
 - 10ng vs 1ng: 68 genes
 - PrimeView vs U133 Plus 2.0: 2547
 - Based on p-value (0.05, no FDR correction):
 - 10ng vs 1ng: 1897 genes
 - PrimeView vs U133 Plus 2.0: 5071 genes
 - Fold Change higher abs. 2, no p-value:
 - 10ng vs 1ng: 169 genes
 - PrimeView vs U133 Plus 2.0: 4503 genes

Venn Diagramm for PrimeView vs U133 Plus 2.0



- Correlation between 10ng and 1ng is very good on both array types
- Correlation between PrimeView and U133 Plus 2.0 is also very good
 - No false positive differential expressed genes could be found with standard thresholds (FDR corrected p-Value of 0.05)
 - But with lowered thresholds some could be found
- Array comparison can be carried out with Partek Genomics Suite
 - Removing probe set redundancy by the highest expressed probe set for each Gene Symbol
 - Merging array results based on commonly represented Gene Symbols
 - Quantile Normalization for direct array comparison
 - Comparison done based on 18081 common Gene Symbols

Contact Information

Sample Processing

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Data Analysis

Partek: Providing software for genomic data analysis

More Info

- www.partek.com
- inquiry@partek.com

Free Trial

- www.partek.com/freetrial

Global Technical Support

- Email: support@partek.com
- North America: 314-878-232

