Analysis of HapMap Lymphoblastic Cell Line Surface Protein Expression

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Summer Research Program in Genomics

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Project Overview

- Approach: use the HapMap lymphoblastic cell lines as a model system to study gene expression
- Goals: Correlate genetic variation captured by the HapMap with:
  1. cell surface protein expression
  2. poly-marker immunological expression

Data Collection: The Samples

- HapMap Cell Lines (270 Total Samples)
  - 30 Yoruban Trios (YRI)
  - 45 Japanese Individuals (JPT)
  - 45 Han Chinese Individuals (CHB)
  - 30 U.S. Trios (CEU)

Data Collection: The Markers

1. B-cell specific markers
2. Markers expressed on B-cells that showed evidence of heritability in previous RNA expression studies

CD19	CD20
IL6R	IgD
HLA-DQ	HLA-DR
CD227	CD21
CD40	IgG
IgM	CD58
CD95	CD86
CD80	

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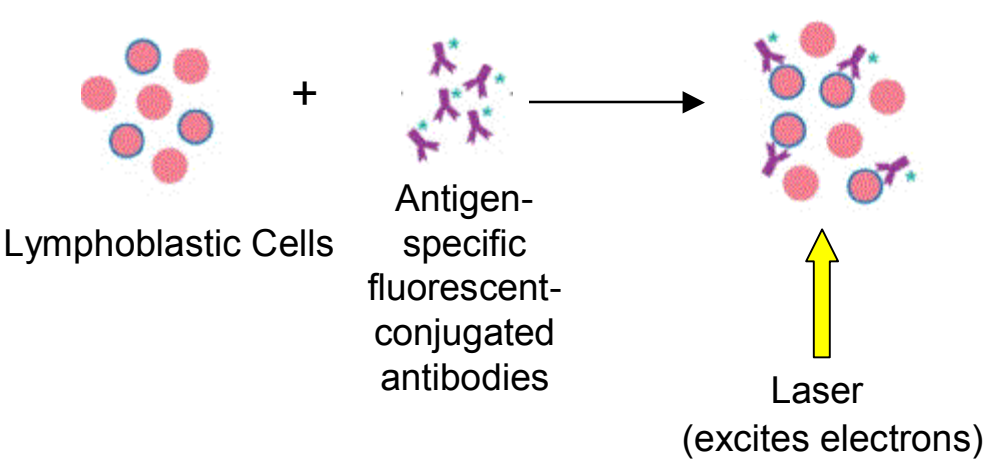
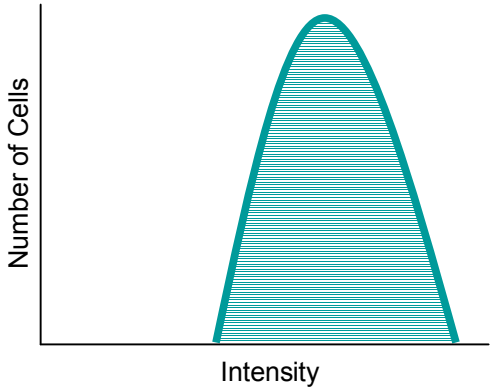
Data Collection: The Method



Cell #	FSC-H	SSC-H	FL1-H	FL2-H	FL3-H	FL2-A	FL4-H
1	484	282	117	195	88	0	192
2	538	308	106	36	49	0	15
3	509	374	227	253	205	0	191
4	595	386	182	188	129	0	237



Fluorescent-activated cell sorting

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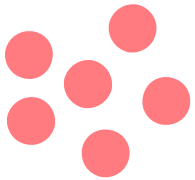
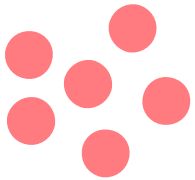
Images courtesy of http://www2.hawaii.edu/~johnb/micro/micro161/antigen-antibody_reactions/Chap5_Ag_Ab_reactions.htm

Data Collection: The Method



Lymphoblastic cells



Laser
(excites electrons)

light
emitted

Detector

Cell #	FSC-H	SSC-H	FL1-H	FL2-H	FL3-H	FL2-A	FL4-H
1	484	282	117	195	88	0	192
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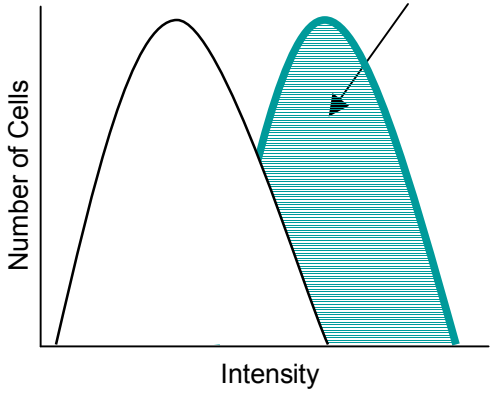
phenotype



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Data Processing and Analysis

1. Perform quality control analysis
2. Correlate marker expression with genetic variants
3. Define poly-marker immunological expression profiles
4. Correlate immunological expression profiles with genetic variants

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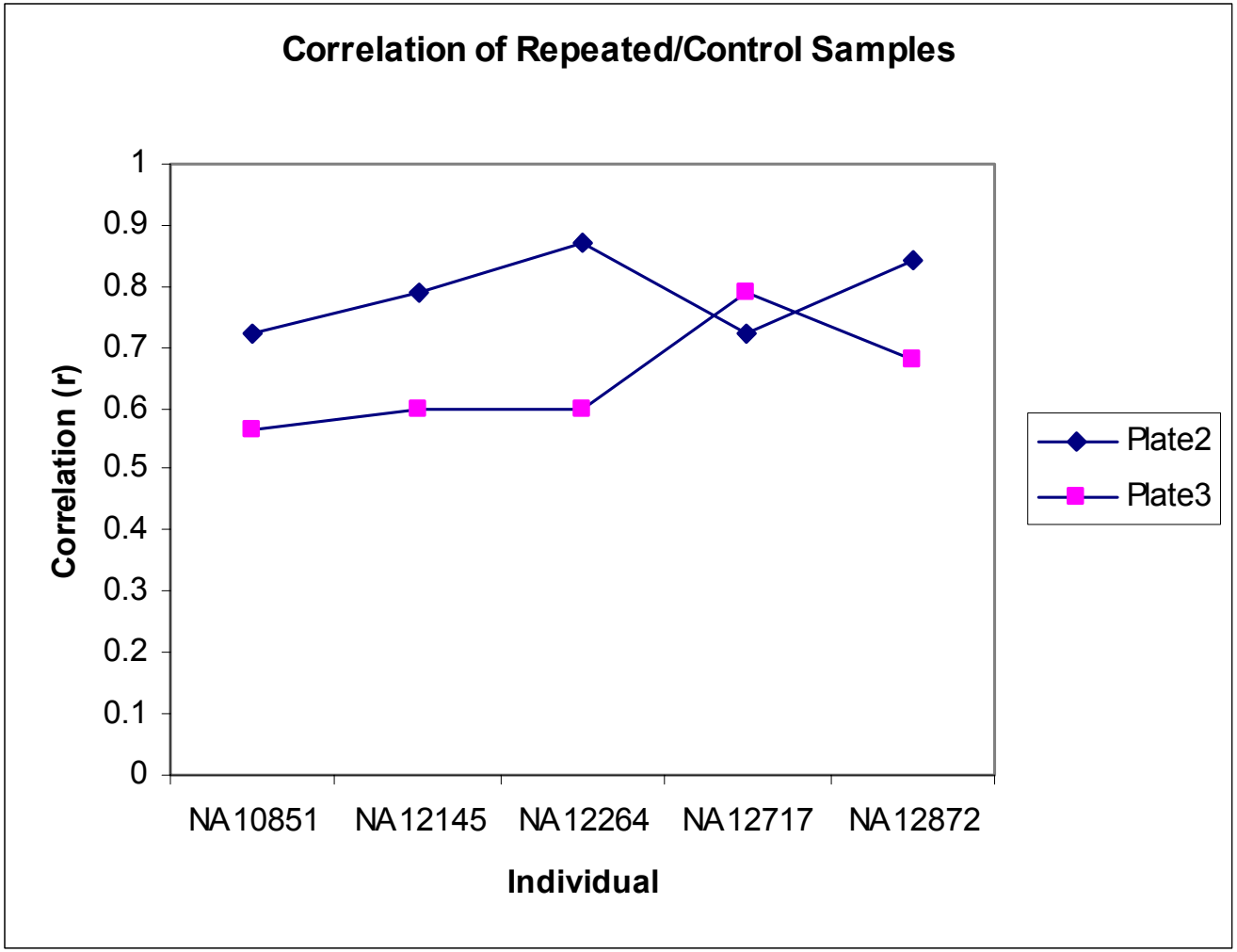
Sources of Variation

- Manual handling of samples (staining protocol/loading FACS machine)
- Cell count (sampling)
- Other (cell culture, user manipulation required during detection)

Correlation of Repeated/Control Samples



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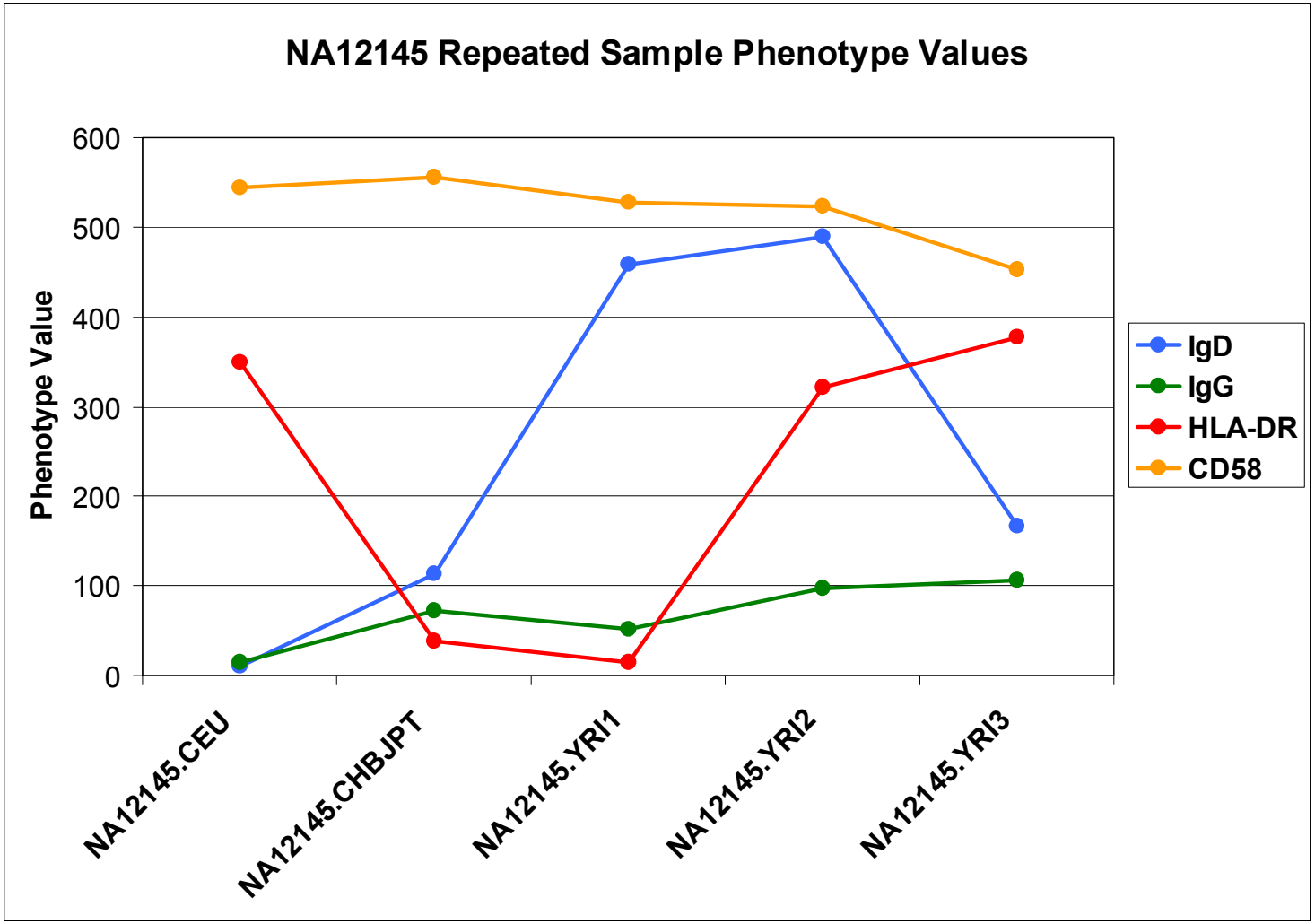
Correlation of Repeated/Control Samples



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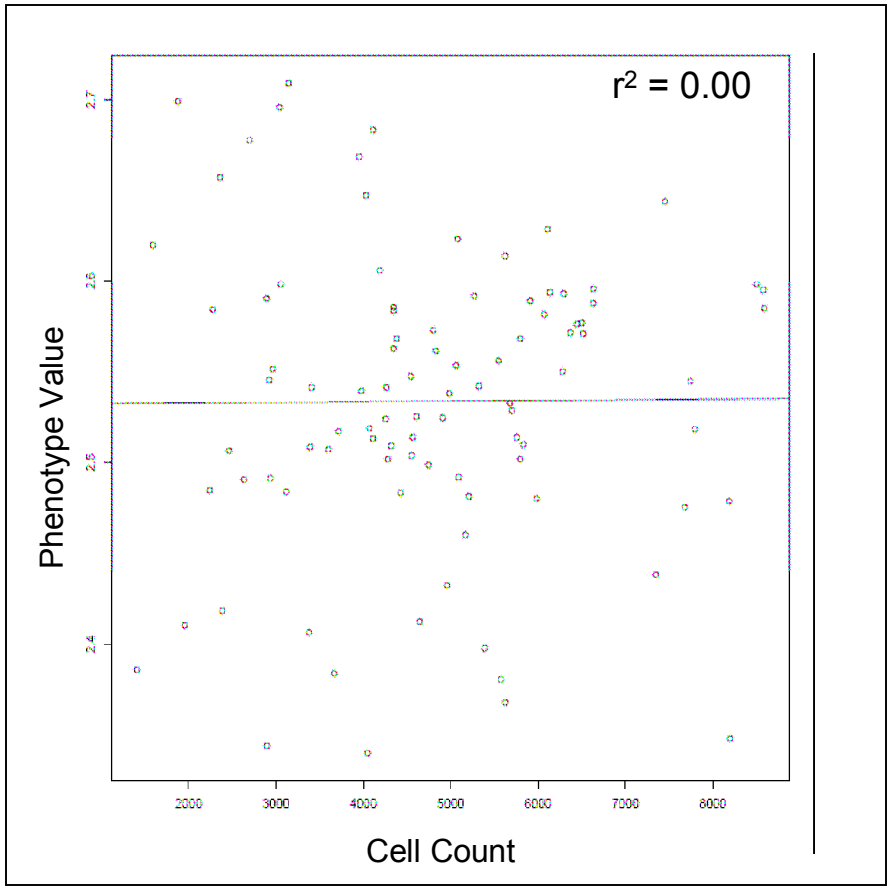
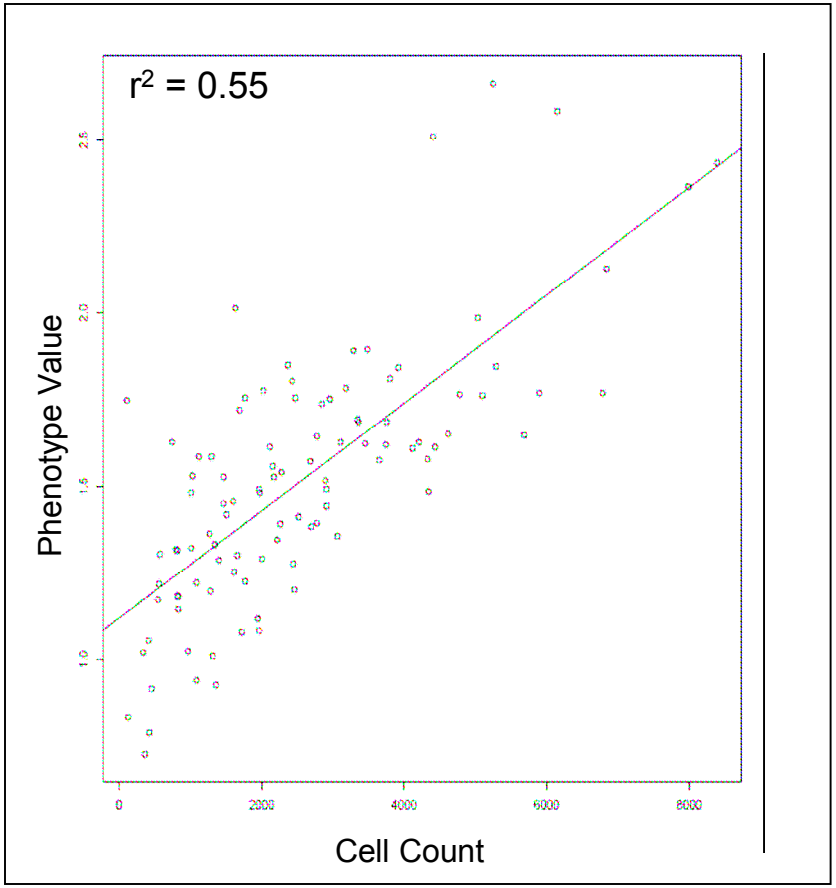
Phenotype vs. Cell Count





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Courtesy of Roman Yelensky

Markers that Passed QC in CEU

CD19	**CD20**
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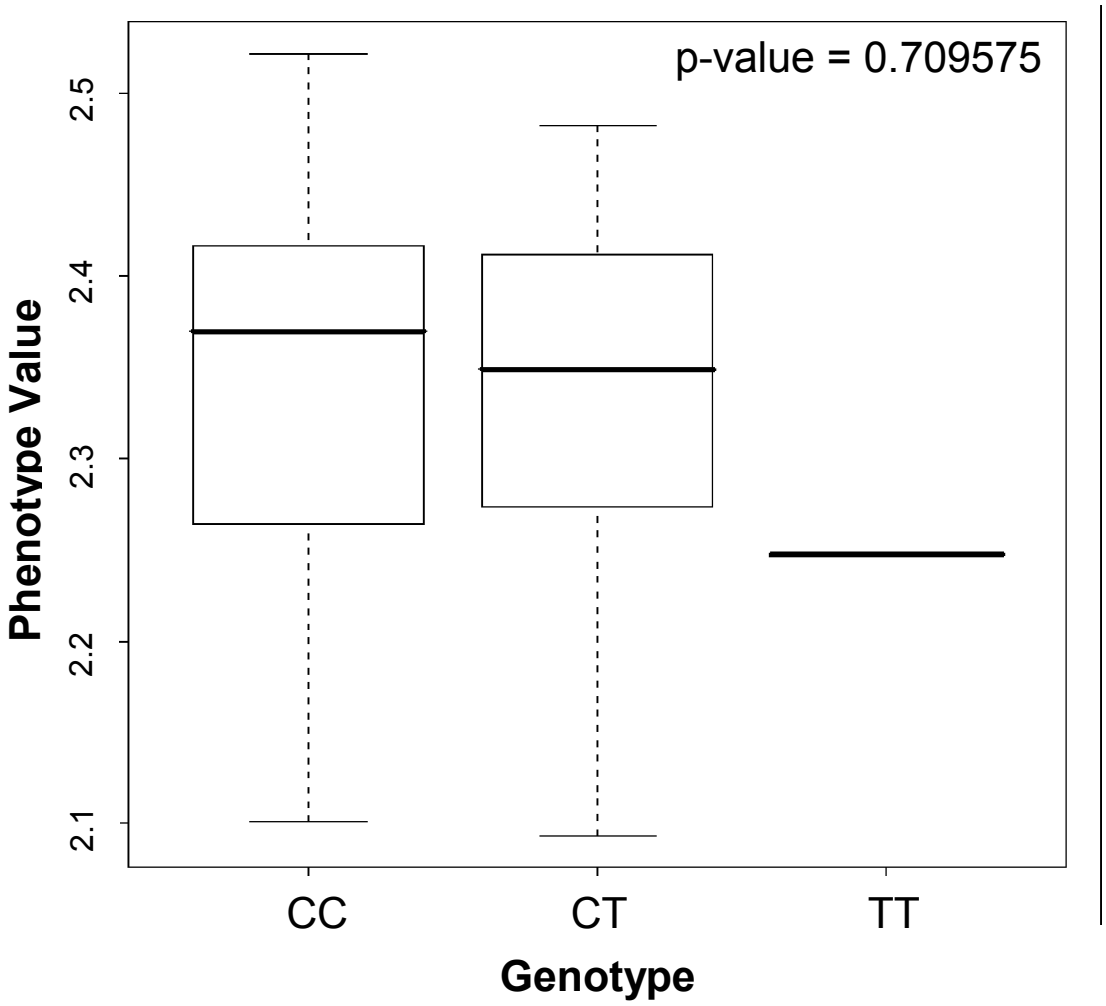
Correlate Marker Expression with Genetic Variation



Attempt to validate a known correlation between CD40 protein expression and dose of the rs1883832^{C} allele (in Kozak sequence)

C allele = susceptibility to Grave's disease, high protein expression (*in vitro* & B cells *ex vivo* – 23 individuals)

T allele = low expression

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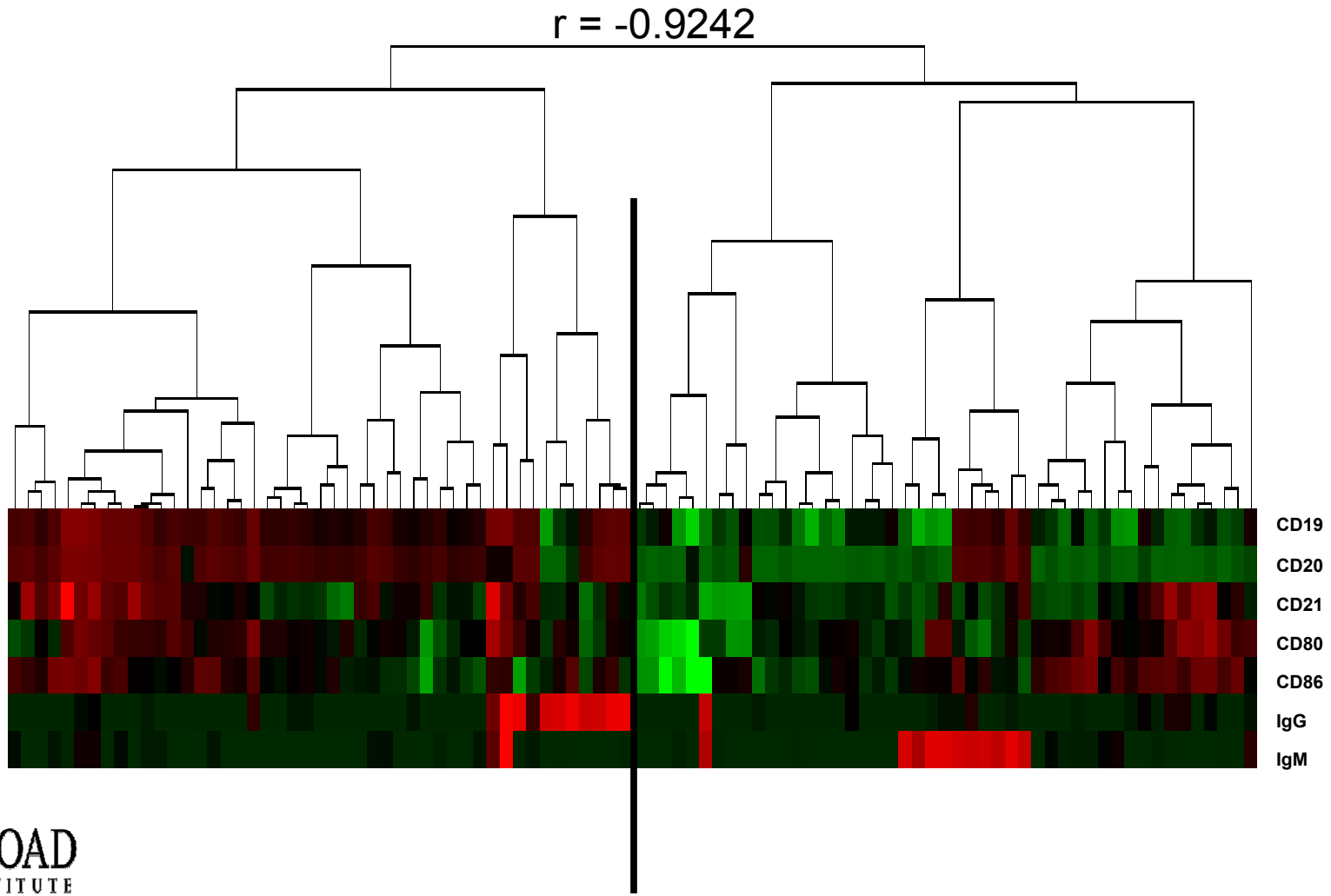
Courtesy of Roman Yelensky
Jacobson *et al., Endocrinology* 2005, 146:2684

Define poly-marker immunological expression profiles



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Conclusion

- Completed quality control analysis
  - Defined a curated marker set
- Sub-populations of cell lines that share particular immunological profiles may exist

Future Work

- Further define poly-marker immunological expression profiles
- Further correlate marker expression phenotype with genetic variation
- Correlate marker expression phenotype with other relevant phenotypes

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