



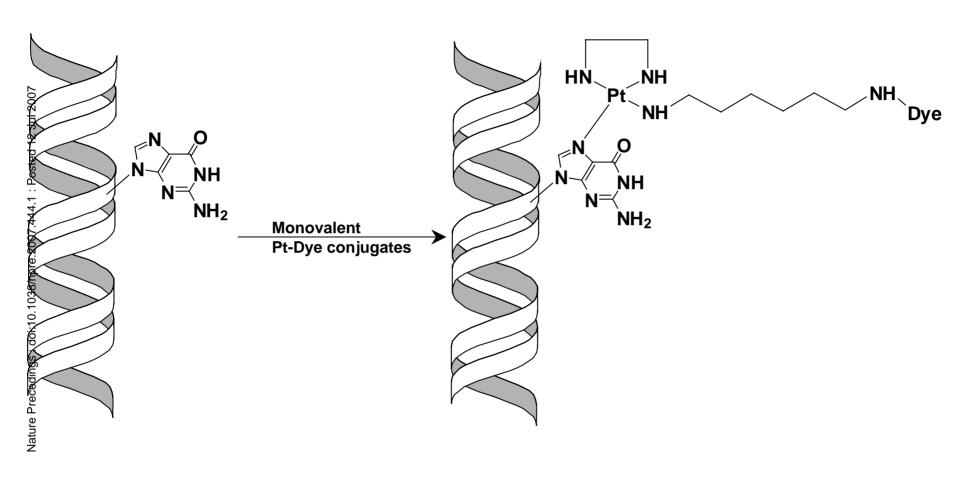
Direct fluorophore conjugation to genomic DNA for microarray-based epigenomic profiling

Vineet Gupta, Ph.D.

Massachusetts General Hospital, Harvard Medical School

http://massgeneral.org/chemicalbiology

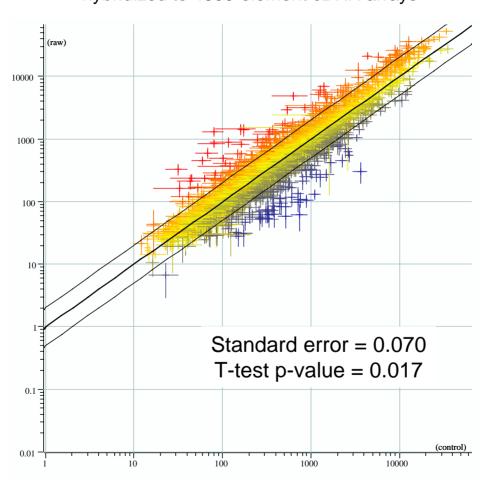
Non-Enzymatic Conjugation of Fluorescent Dyes to Nucleic Acids

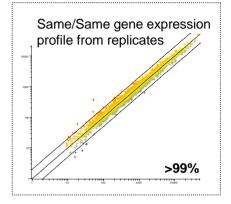


Platinum-based reagents (such as ULS from Kreatech) selectively react with and label the N7 of Guanine residues

Conjugated mRNA Produces Highly Precise Expression Profiles

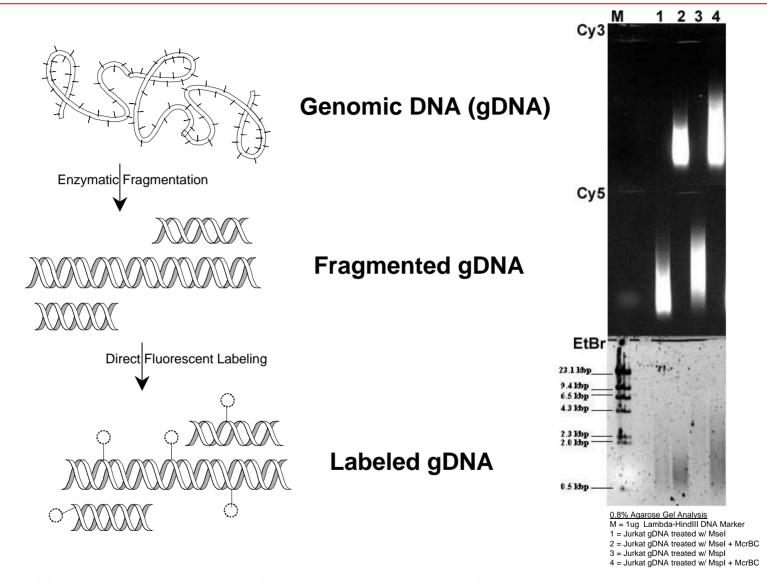
Differential gene expression profile from six replica experiments using HL-60 mRNA (Cy3 labeled) Vs Jurkat mRNA (Cy5 labeled) hybridized to 4800-element cDNA arrays





Directly labeled mRNA produces high quality microarray expression profiles

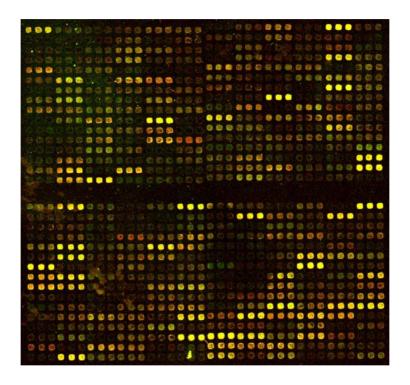
Genomic DNA Can Similarly Be Directly Fluorescently Labeled



Genomic DNA can be differentially digested with methylation-sensitive restriction enzymes and directly fluorescently labeled

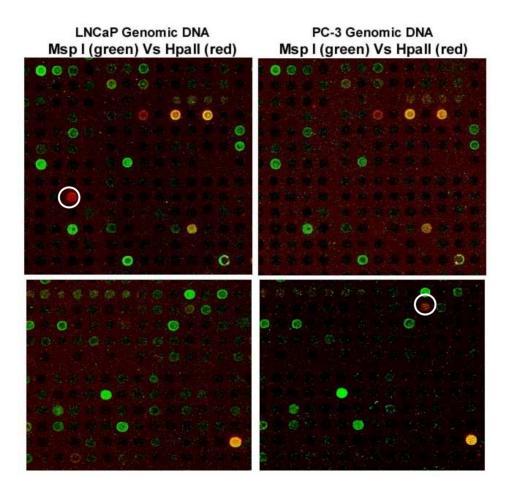
Epigenomic Profiling with Directly Labeled Genomic DNA

Microarray-based epigenomic profiling using Jurkat gDNA and oligo arrays



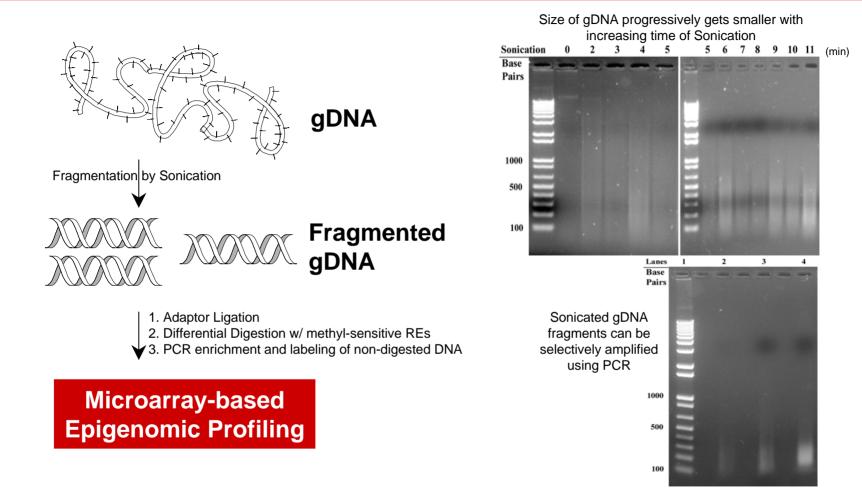
Methylation-dependent gDNA Fragmentation and Direct Labeling (MFDL) can be used for Microarray-based epigenomic profiling

Epigenomic Profiling with Directly Labeled Genomic DNA



Methylation-dependent gDNA Fragmentation and Direct Labeling (MFDL) can be used for Microarray-based epigenomic profiling to identify differentially-methylated loci

Size Fractionation of gDNA by Sonication



Genomic DNA can be size-fractionated by sonication (using published protocols) prior to MFDL for epigenomic profiling