Cancer Epigenetics

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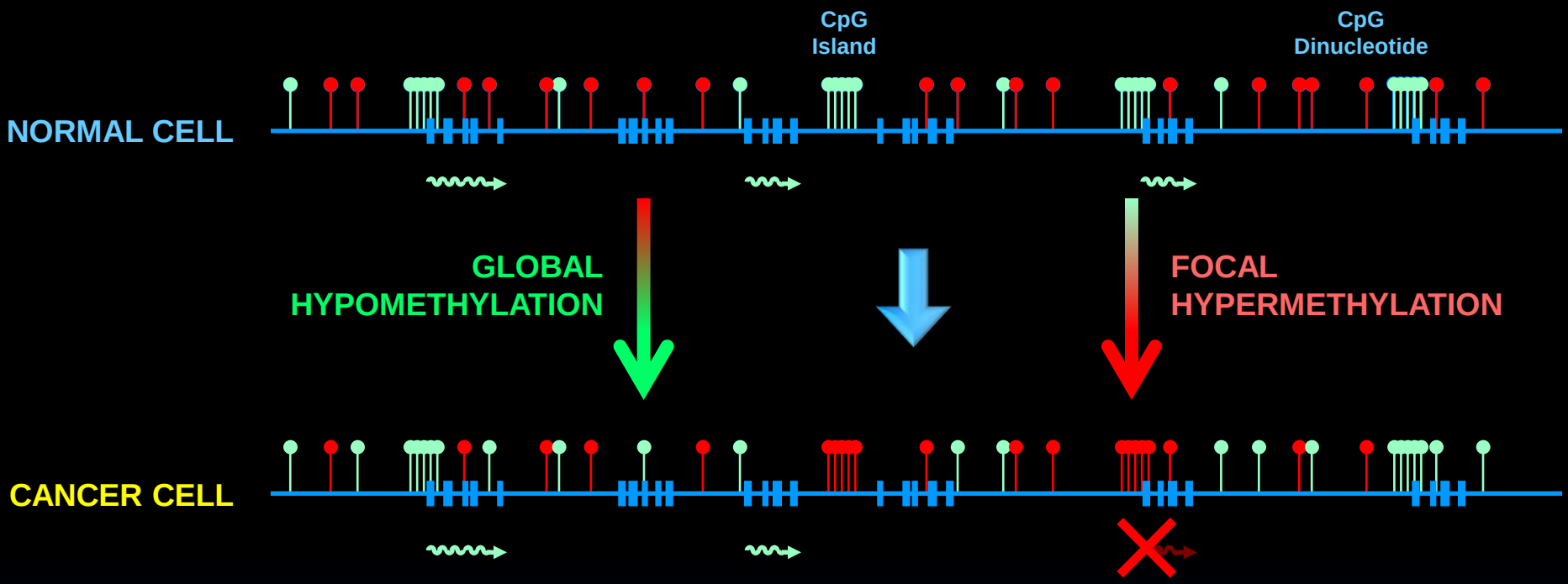
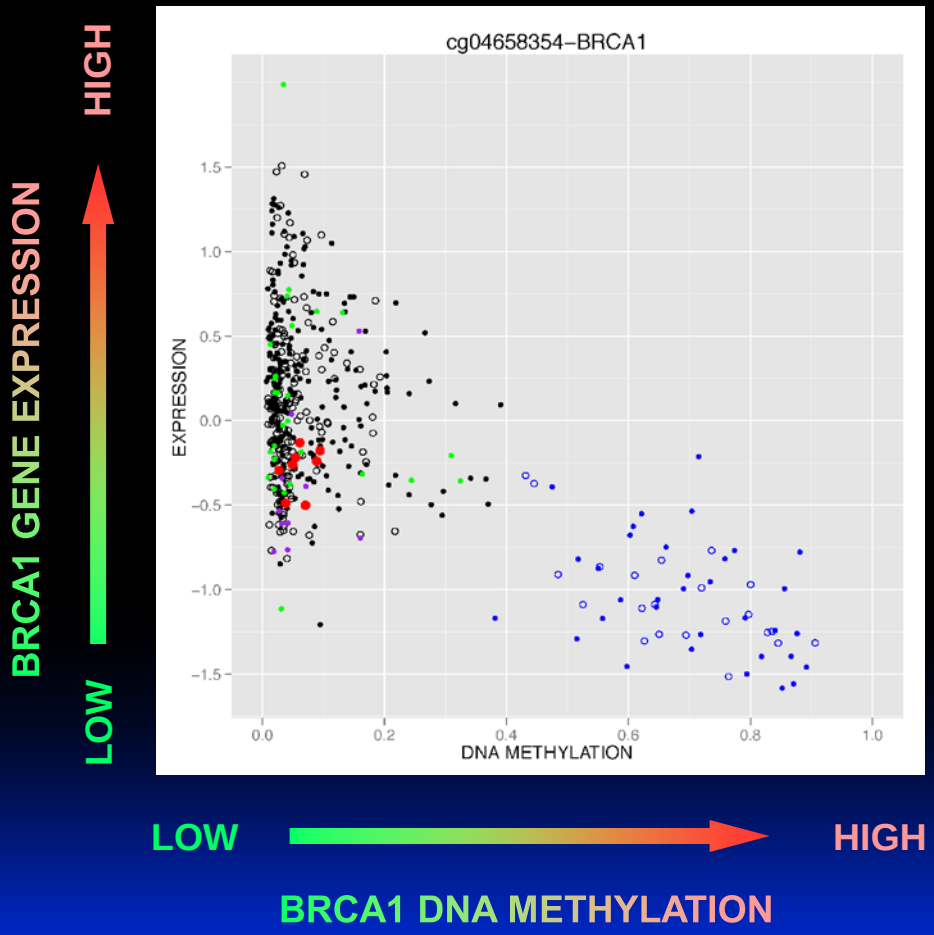
DNA Methylation Alterations in Cancer

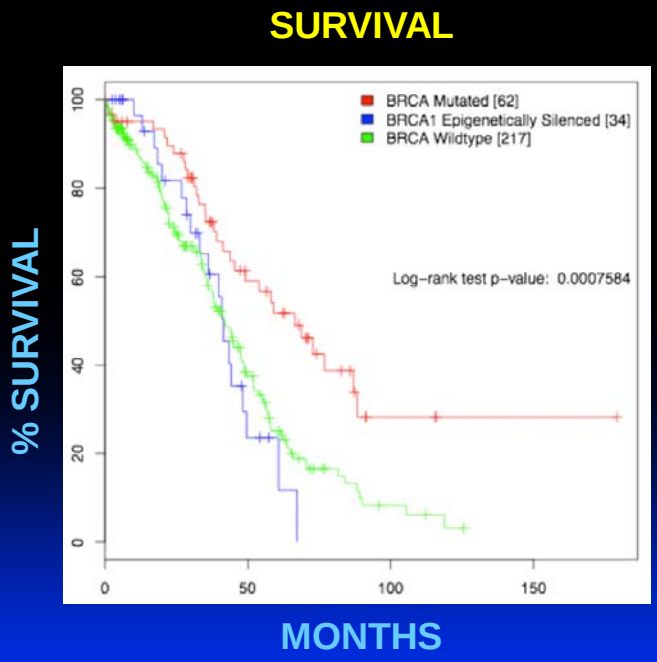


- CpG Islands may acquire abnormal hypermethylation in cancer
- Methylated CpG Island promoters are transcriptionally silenced in cancer
- Areas of low-CpG density may lose DNA methylation in cancer

Epigenetic Silencing of *BRCA1* in Serous Ovarian Cancer



- Red: Fallopian Tubes
- Purple – Somatic Mutation
- Green – Germline Mutation
- Blue – Epigenetic Silencing
- Hollow – Not Sequenced

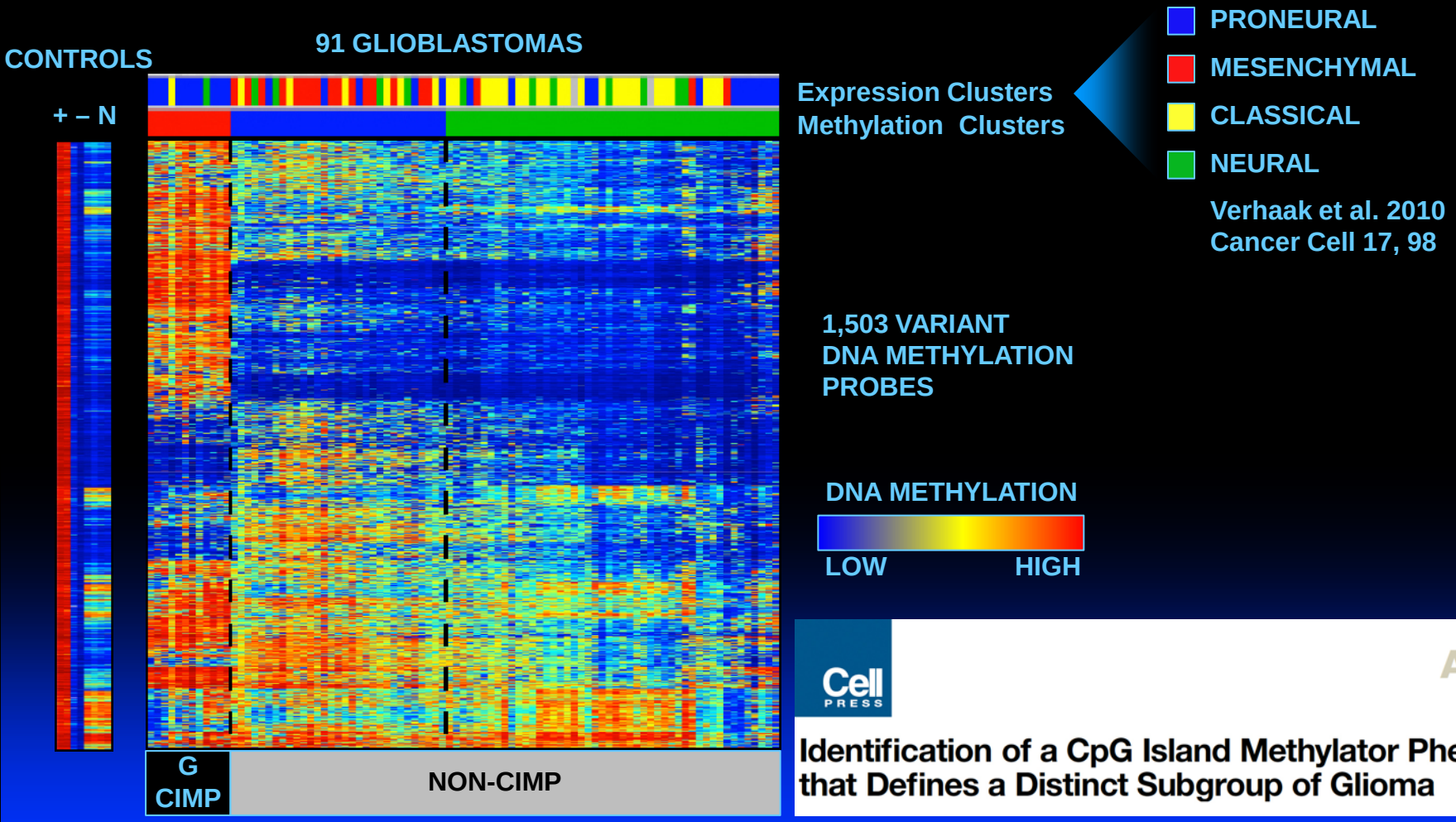


TCGA Research Network (2011) Nature 474, 609

Outline

- CpG Island Methylator Phenotypes - Glioblastoma

Glioma-CpG Island Methylator Phenotype (G-CIMP) (TCGA)

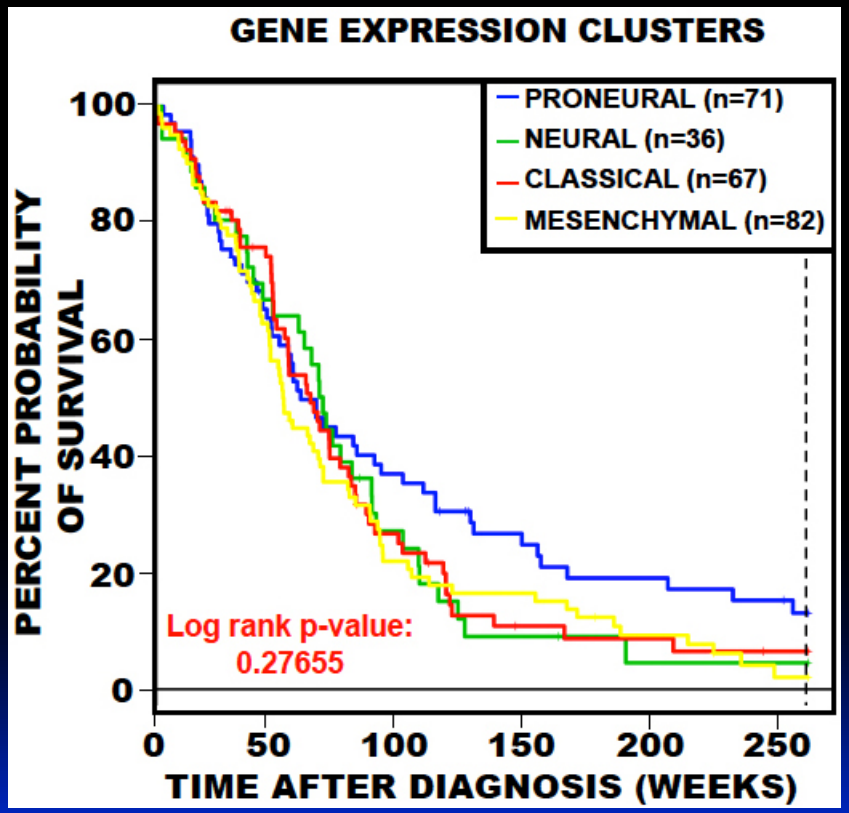


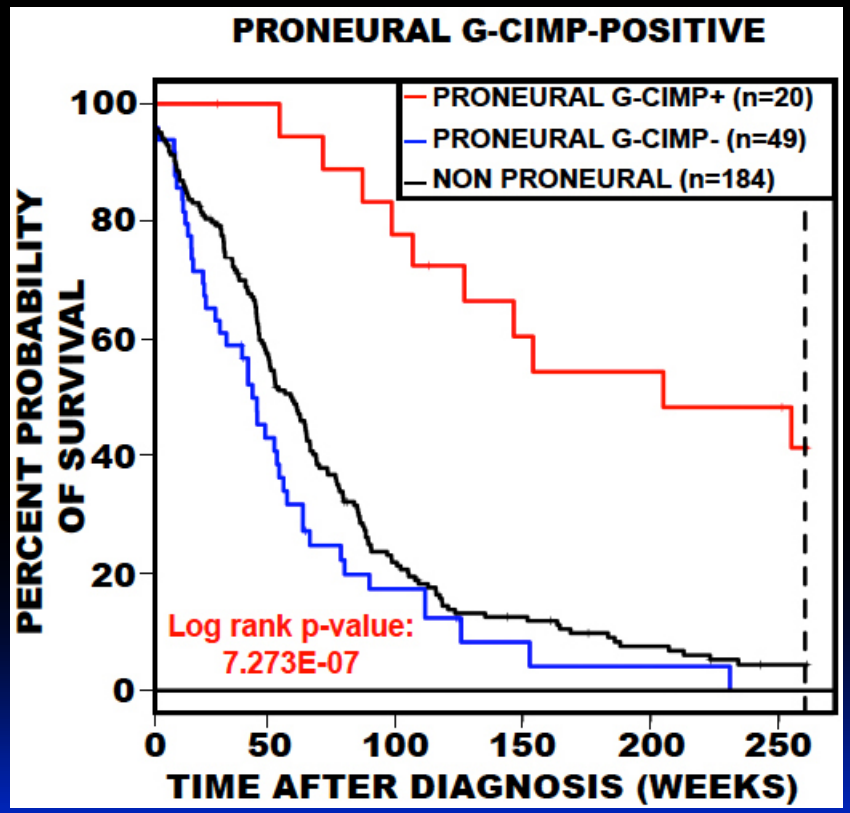
Cancer Cell
Article

Identification of a CpG Island Methylator Phenotype that Defines a Distinct Subgroup of Glioma

Noushmehr et al. 2010 Cancer Cell 17, 510

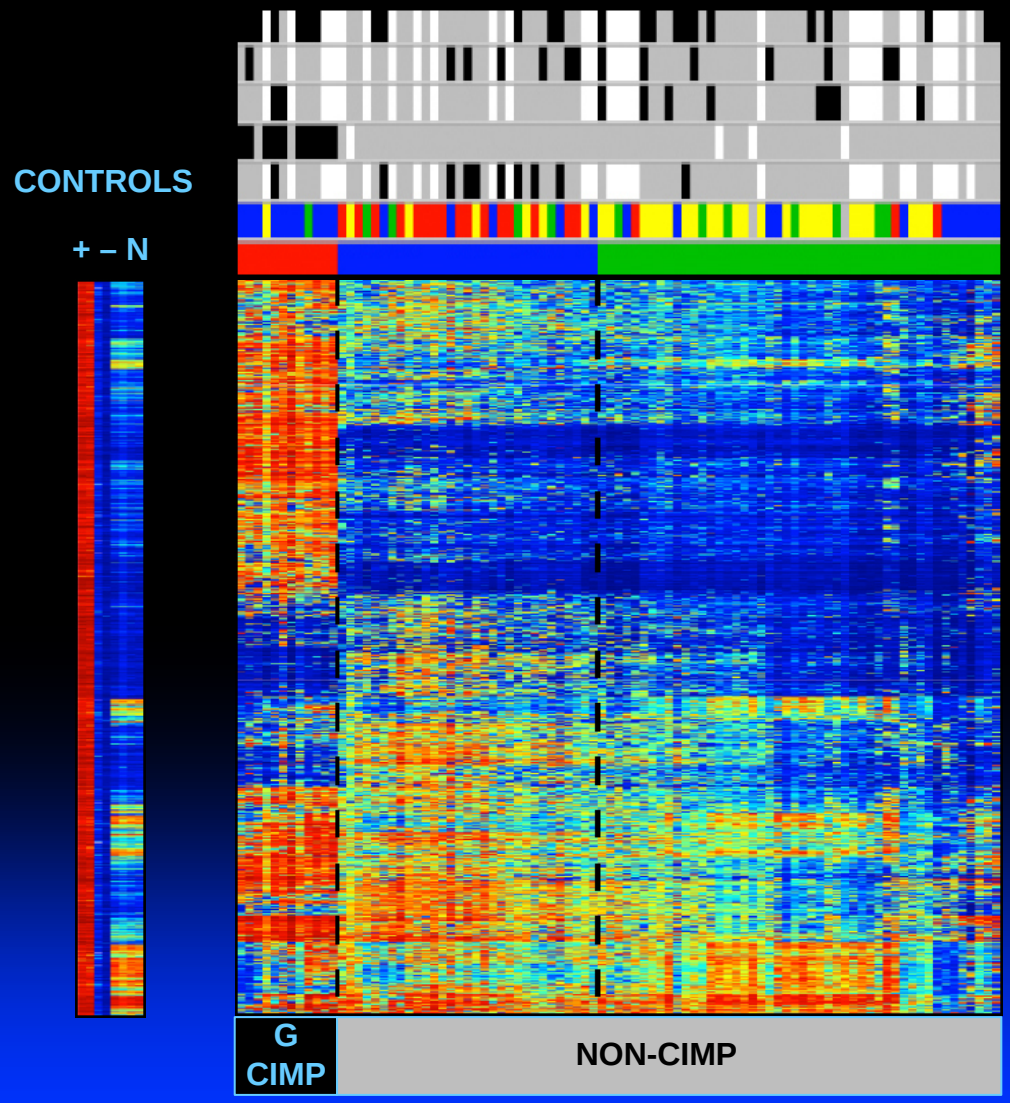
G-CIMP Is a Subset of Proneural Glioblastomas with Better Survival





Noushmehr et al. 2010 Cancer Cell 17, 510

Glioma-CpG Island Methylator Phenotype (G-CIMP) (TCGA)



TP53 Mutation
PTEN Mutation
NF1 Mutation
IDH1 Mutation
EGFR Mutation
Expression Clusters
Methylation Clusters

MUTATION
NO MUTATION
PRONEURAL
MESENCHYMAL
CLASSICAL
NEURAL

G-CIMP is Tightly Linked to *IDH1* Mutation

ALL TUMORS		G-CIMP —	G-CIMP +	TOTAL
IDH1	Wild-type	184	5	189
	Mutant	0	18	18
TOTAL		184	23	207

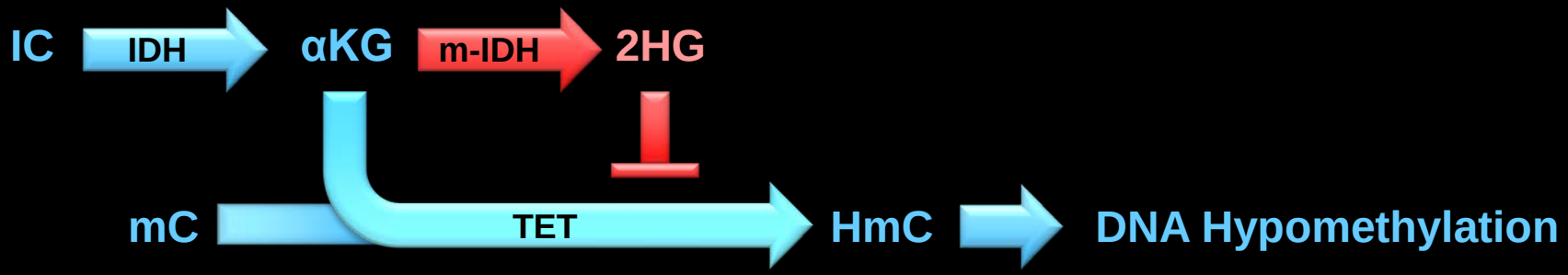
DNA METHYLATION

LOW HIGH

Noushmehr et al. 2010 Cancer Cell 17, 510

Model for G-CIMP

IDH1 Mutation Causes Aberrant CpG Island Methylation

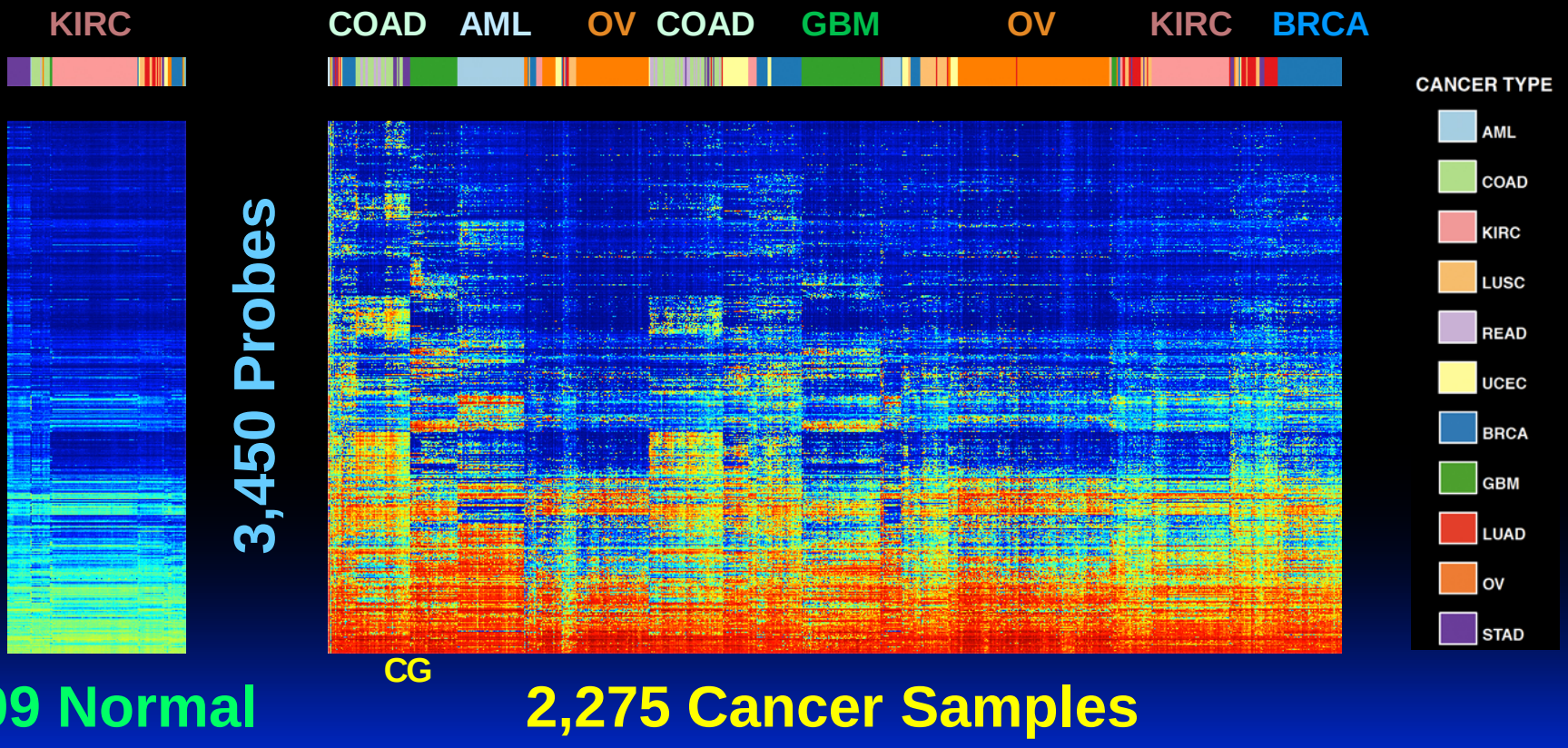


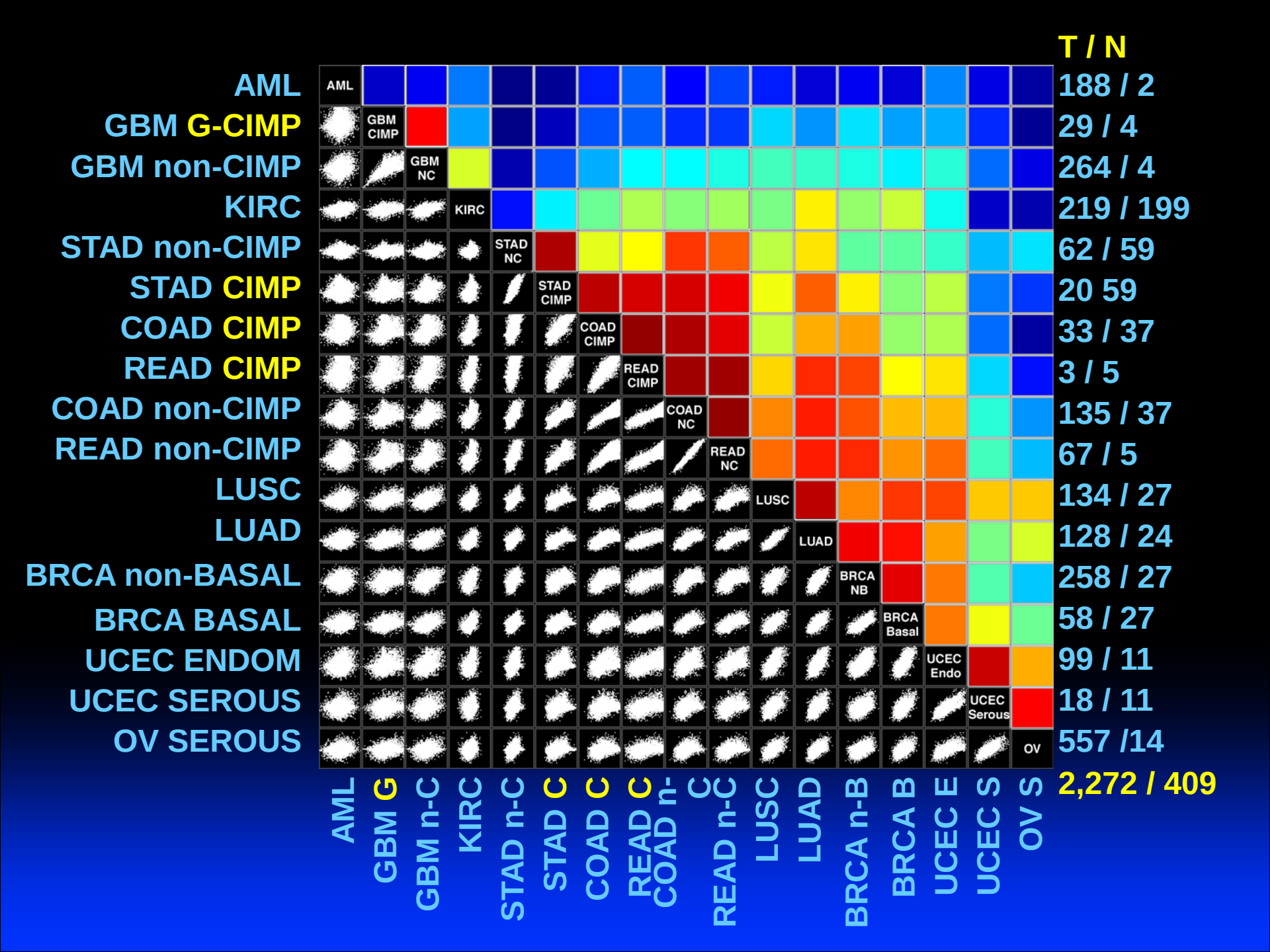
…..Does not explain G-CIMP IDH1^{wt} cases

Outline

- CpG Island Methylator Phenotypes - Glioblastoma
- Cross-tumor Comparisons

Comparison of 2,275 TCGA Cancer Samples and 409 Normal Tissues




T / N
AML
GBM G-CIMP
GBM non-CIMP
KIRC
STAD non-CIMP
STAD CIMP
COAD CIMP
READ CIMP
COAD non-CIMP
READ non-CIMP
LUSC
LUAD
BRCA non-BASAL
BRCA BASAL
UCEC ENDOM
UCEC SEROUS
OV SEROUS
188 / 2
29 / 4
264 / 4
219 / 199
62 / 59
20 59
33 / 37
3 / 5
135 / 37
67 / 5
134 / 27
128 / 24
258 / 27
58 / 27
99 / 11
18 / 11
557 /14
2,272 / 409
AML
GBM CIMP
GBM NC
KIRC
STAD NC
STAD CIMP
COAD CIMP
READ CIMP
COAD NC
READ NC
LUSC
LUAD
BRCA NB
BRCA Basal
UCEC Endo
UCEC Serous
OV
AML
GBM G
GBM n-C
KIRC
STAD n-C
STAD C
COAD C
READ C
COAD n-C
READ n-C
LUSC
LUAD
BRCA n-B
BRCA B
UCEC E
UCEC S
OV S

Outline

- CpG Island Methylator Phenotypes - Glioblastoma
- Cross-tumor Comparisons
- Bisulfite Sequencing - Epigenetic Origins of Cancer

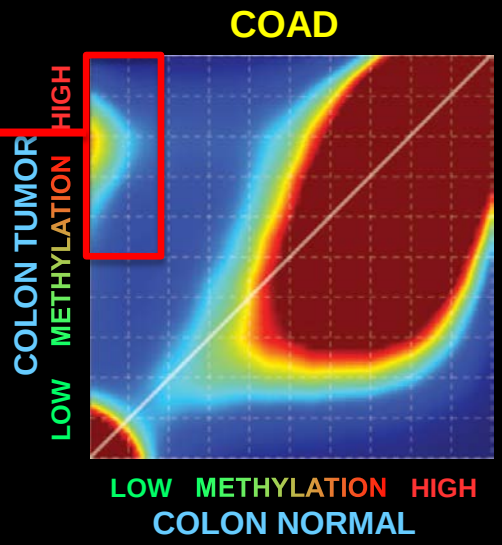
TCGA Whole Genome Bisulfite Sequencing (WGBS)

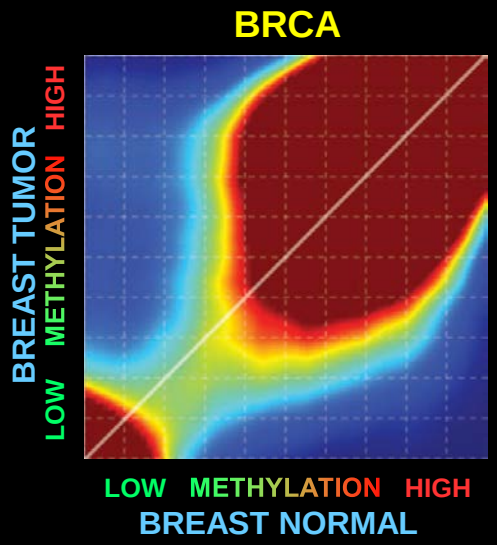
TCGA Sample	Type	Description	Bisulfite non-conversion	Mean cvg	# CpGs	1x cvg (% CpGs)	5x cvg (% CpGs)
AA-3518-01A	**COAD**	MSI-H	0.92%	23x	51.8M	92%	90%
AA-3518-11A	**COAD - N**		0.86%	22x	51.5M	91%	90%
A7-A0CE-01A	**BRCA**	Basal-like subtype	0.31%	19x	50.7M	90%	86%
A7-A0CE-11A	**BRCA - N**		0.36%	19x	50.3M	89%	85%
AA-3518-01A	**UCEC**	Grade 1 endometrioid	0.31%	19x	52.1M	92%	90%
AA-3518-11A	**UCEC - N**		0.31%	18x	51.8M	92%	89%
60-2722-01A	**LUSC**	Classical subtype	0.30%	21x	51.8M	92%	89%
60-2722-11A	**LUSC - N**		0.61%	5x	39.3M	69%	33%

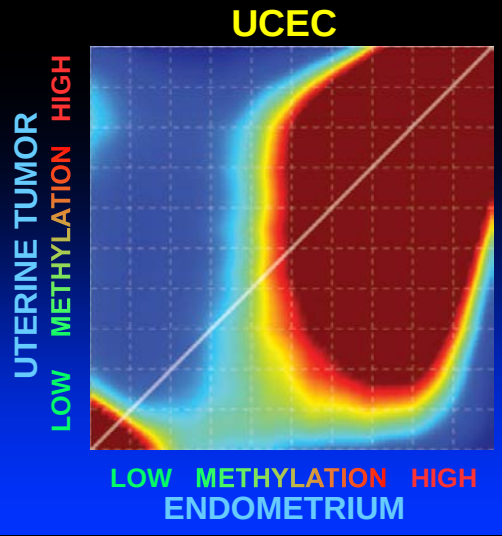
- **In Production: 3 Lung squamous Tumors, 3 Breast Tumors**
- **In Sample Selection: 2 GBM Tumors, 3 Renal Cell Kidney Pairs**

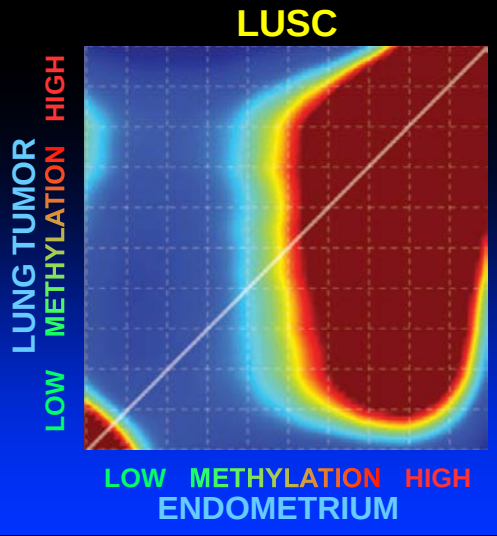
Whole Genome Bisulfite Sequencing of TCGA Tumors and Normal Tissues

MP: Methylation Prone Regions

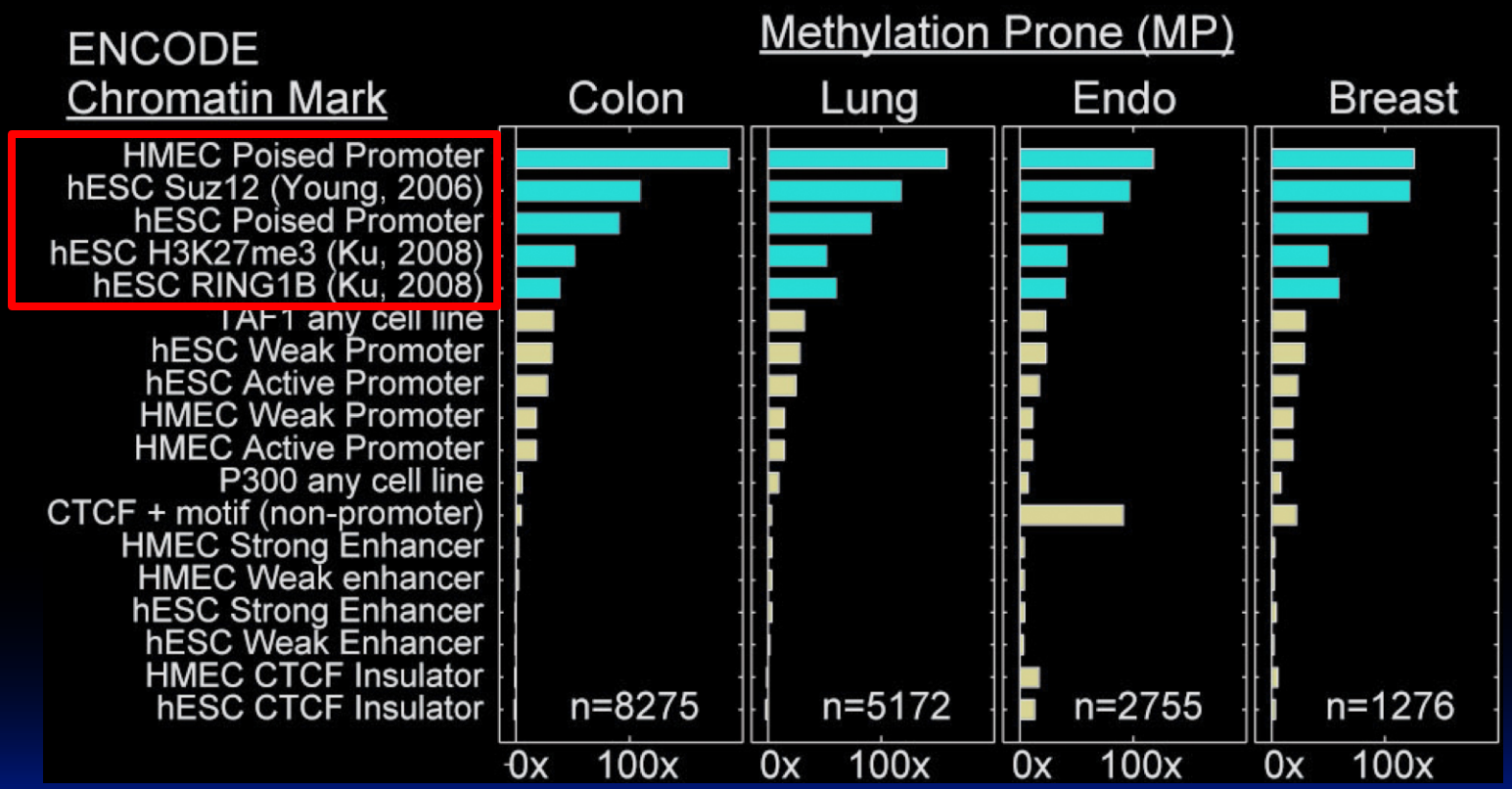








Methylation-Prone Elements are Enriched for Stem-Cell Polycomb Marks



ENCODE chromatin types from J. Ernst *et al*. *Nature* 2011

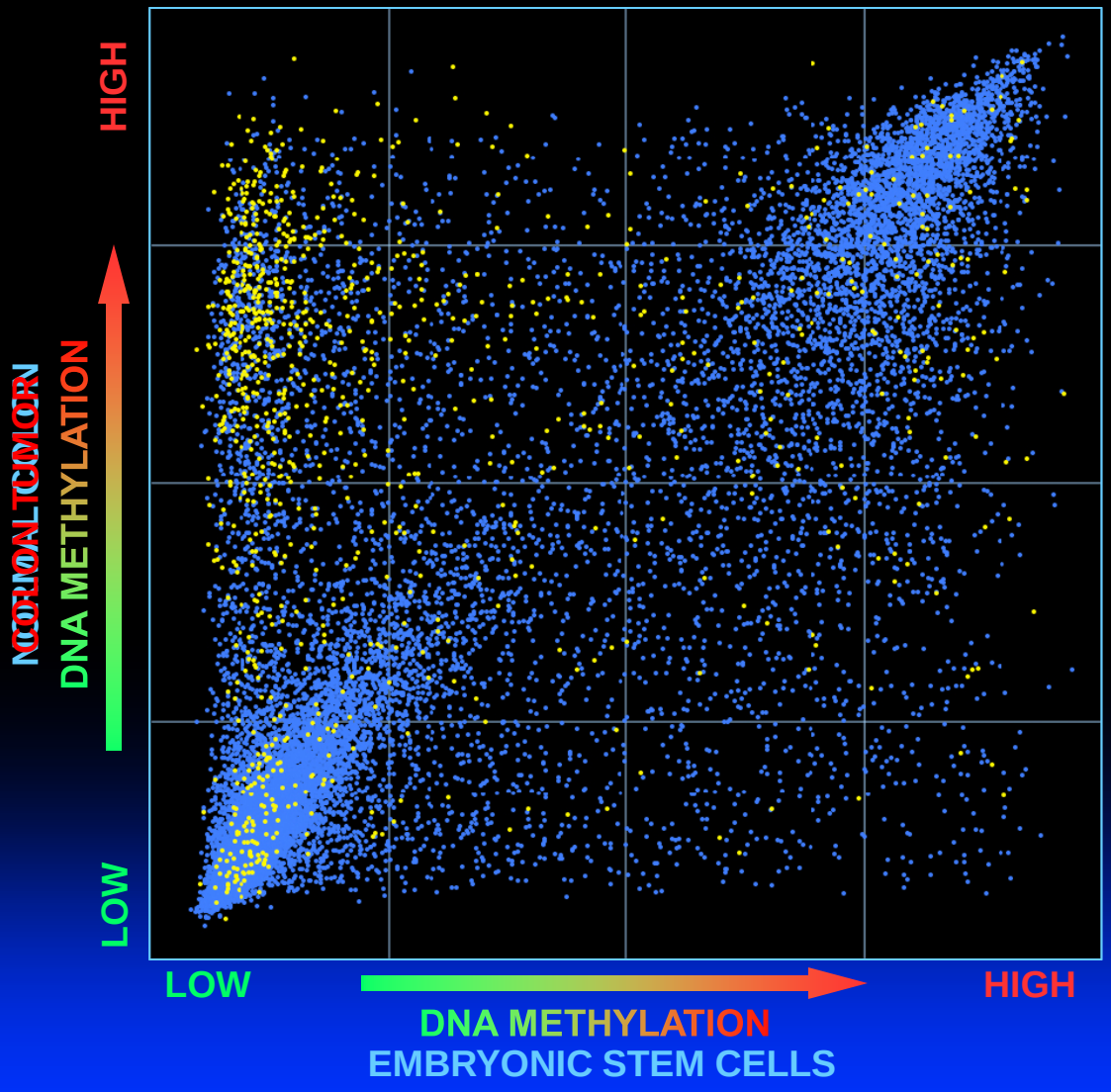
- Active promoter: K4me3, K9ac, K27ac
- Weak promoter: K4me3, K9ac
- Poised promoter: K4me1/2, K27me3
- Strong enhancer: K4me1/2, K9ac, K27ac
- Weak enhancer: K4me1/2
- CTCF Insulator: CTCF

Transcriptional Potential Associated with Histone H3 Methylation

Polycomb Target Genes in Embryonic Stem Cells:

- Master regulators of differentiation and development
- Poised to be turned on during differentiation
- Bivalent epigenetic state: Active (H3K4me3) and Repressive Marks (H3K27Me3)

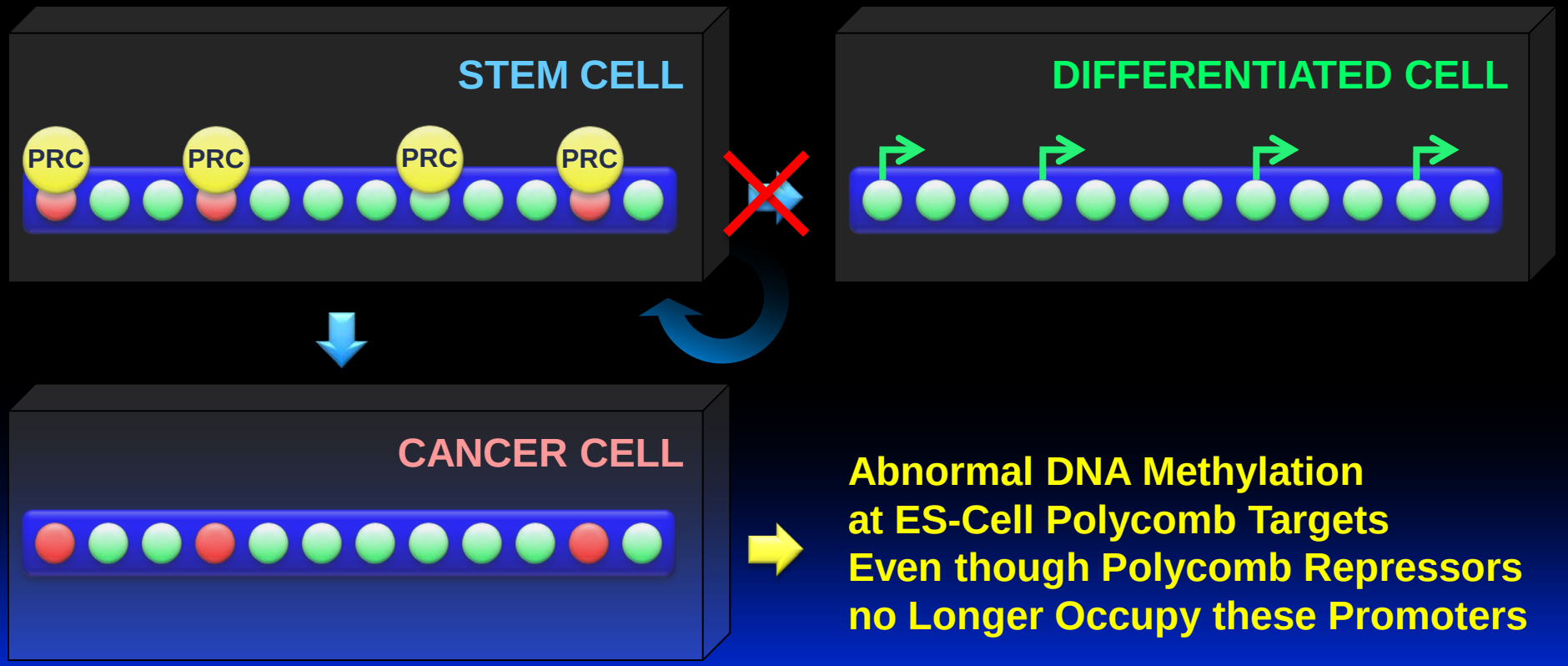
Polycomb Target DNA Methylation Starts in Normal Tissues



16,846 CpG Probes

1,000 ES-Cell Polycomb Targets

Model: Polycomb Crosstalk Leads to Cumulative Stochastic Methylation



Abnormal DNA Methylation at ES-Cell Polycomb Targets Even though Polycomb Repressors no Longer Occupy these Promoters

Widschwendter at al. (2007) Nature Genetics 39, 157

This Model….

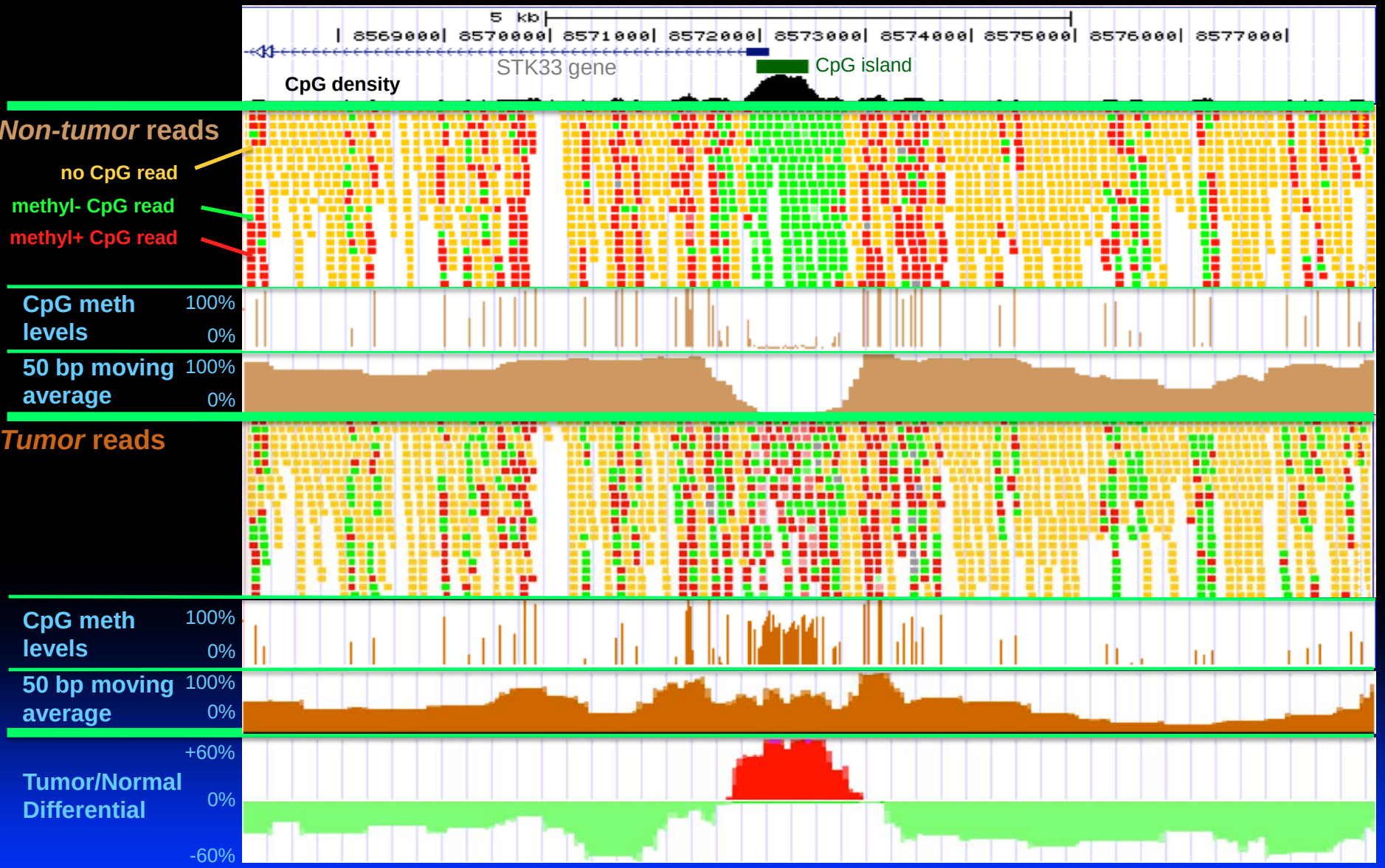
- Would explain the DNA methylation behavior for about half of cancer-specifically methylated genes
- Is consistent with the observation of epigenetic field effects adjacent to tumors
- Is consistent with the stem-cell like behavior of cancer cells and with the evidence for tumor-initiating cells
- Suggests that therapeutic cloning strategies using human ES cells or IPS cells should incorporate screening for PRC2 DNA methylation abnormalities
- Suggests that the first steps of oncogenesis may be epigenetic

Widschwendter at al. (2007) Nature Genetics 39, 157
Hinoue et al. (2011) Genome Research, In Press

Outline

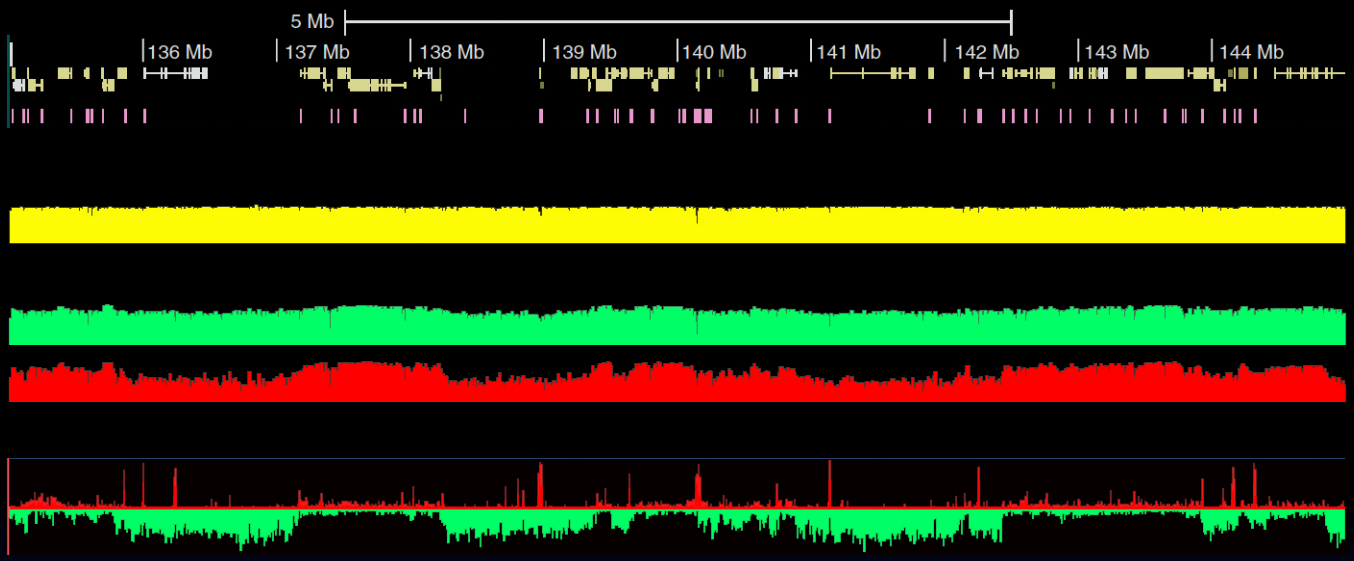
- CpG Island Methylator Phenotypes - Glioblastoma
- Cross-tumor Comparisons
- Bisulfite Sequencing - Epigenetic Origins of Cancer
- Bisulfite Sequencing – Long Range Instability

Methylation-Prone CpG Islands



Berman et al. 2011 Nature Genetics 43, In Press

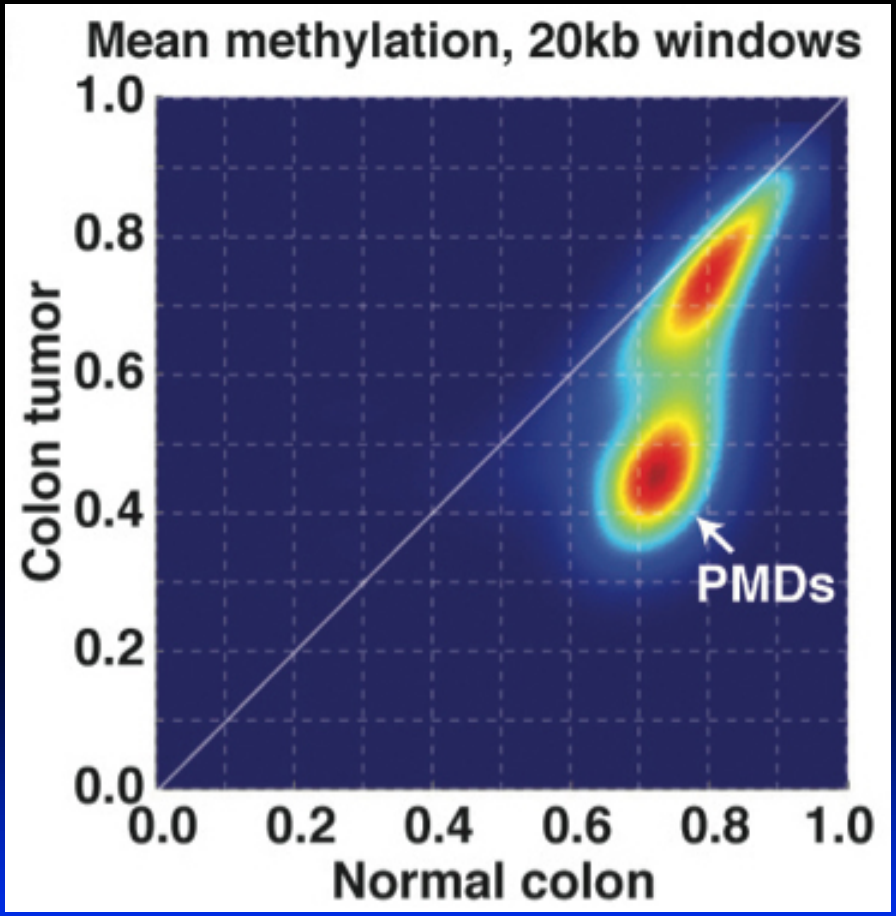
Regions of Focal Hypermethylation and Long-Range Hypomethylation Coincide



Berman et al. 2011 Nature Genetics 43, In Press

A Subset of the Cancer Epigenome Has Partially Lost Methylation

20-kb Windows



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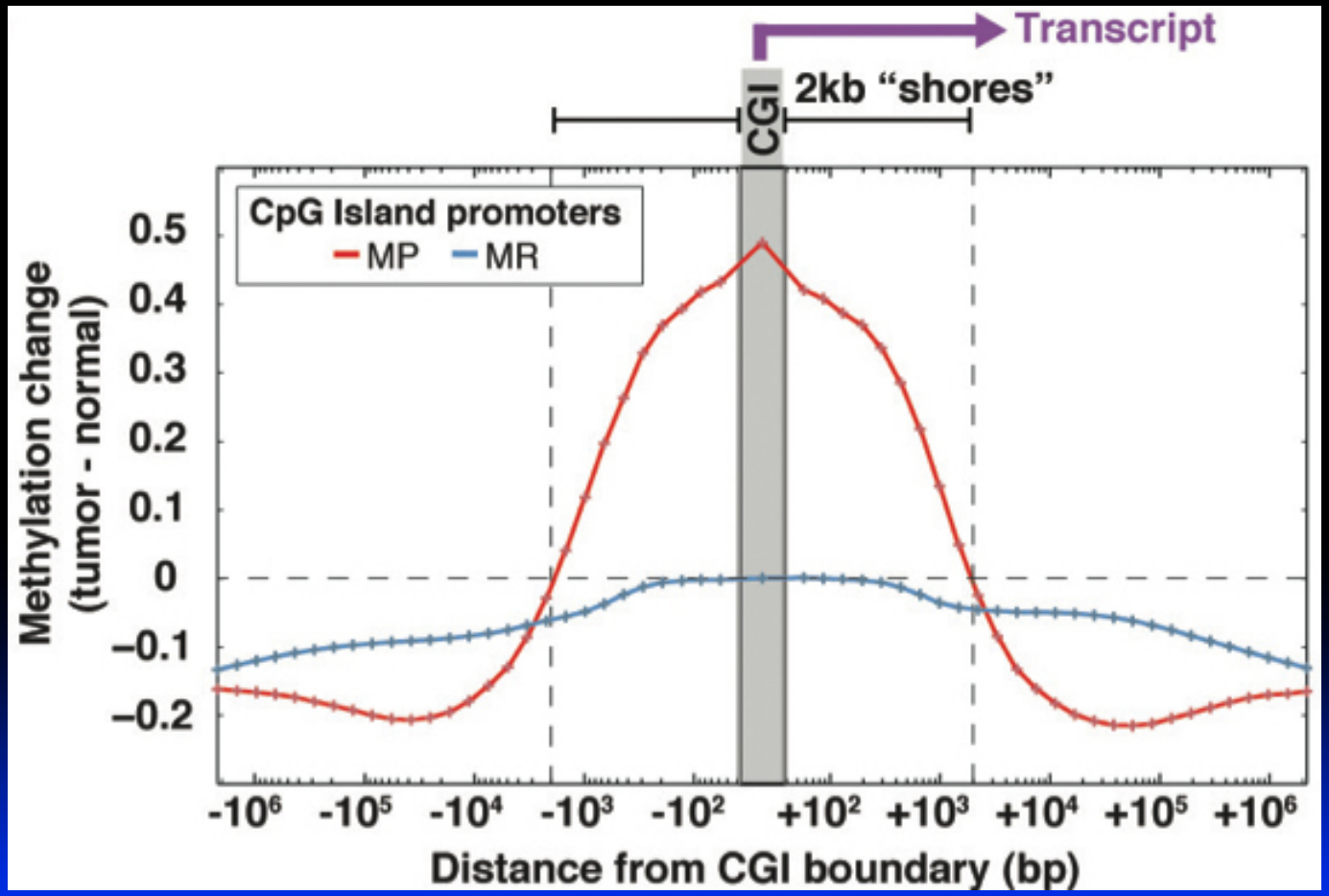

Berman et al. 2011 Nature Genetics 43, In Press

Outline

- CpG Island Methylator Phenotypes - Glioblastoma
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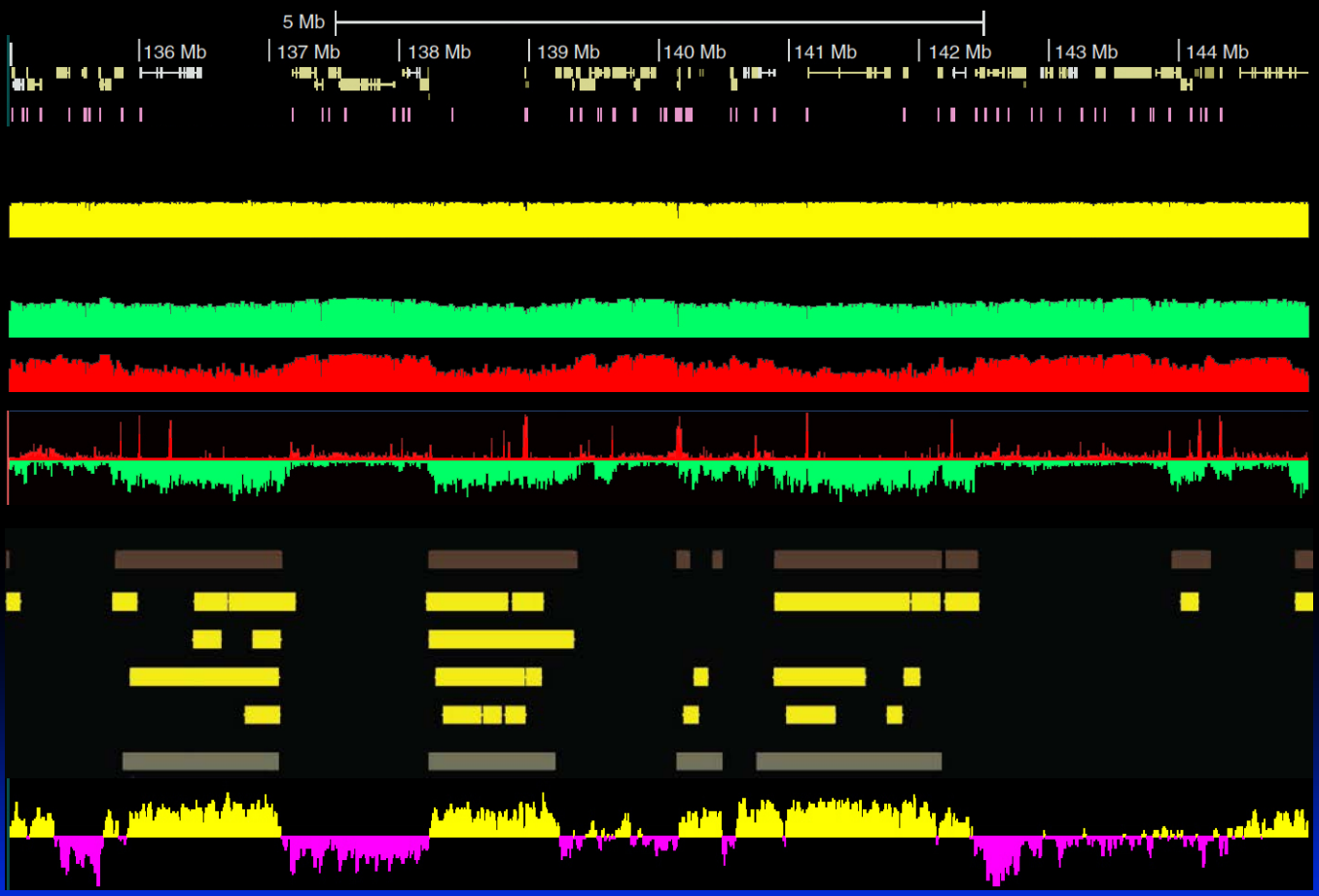
- Bisulfite Sequencing - Epigenetic Origins of Cancer
- Bisulfite Sequencing – Long Range Instability
- Bisulfite Sequencing – Nuclear Architecture

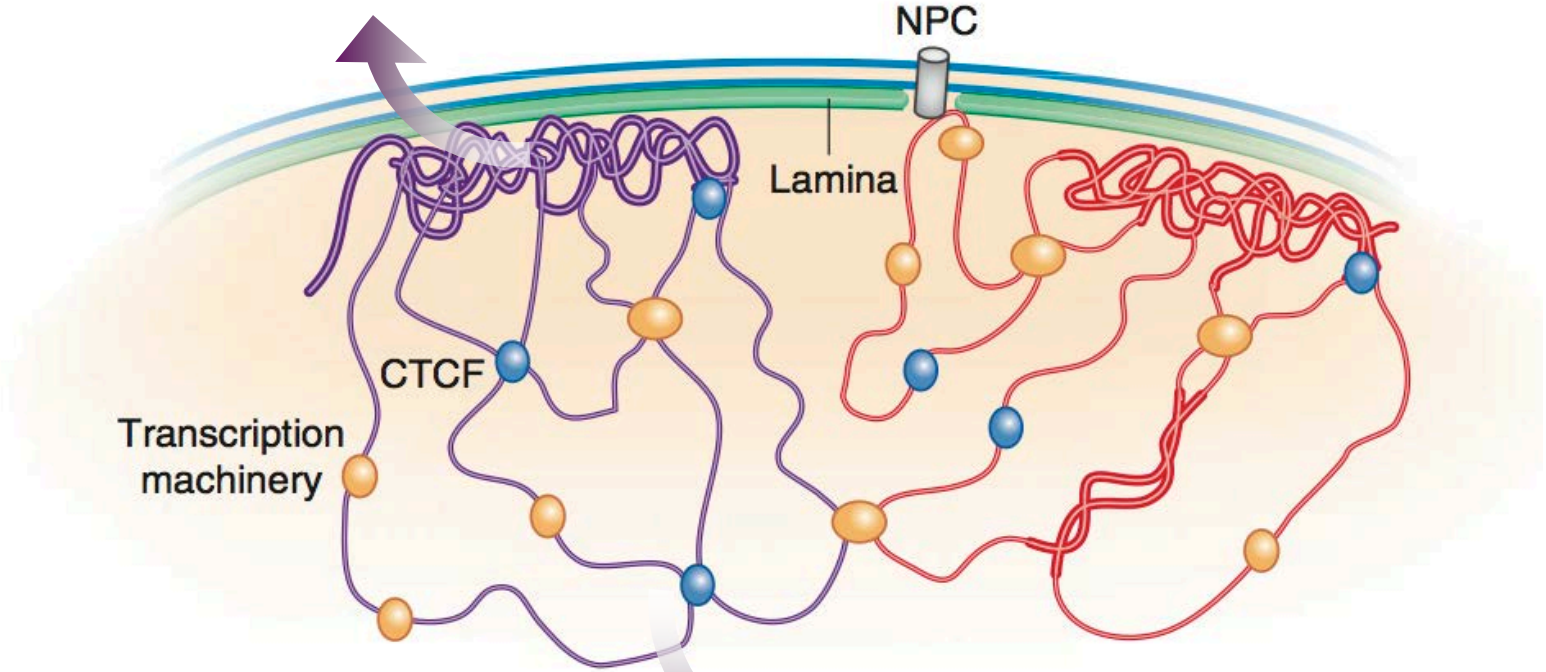
Hypomethylated “Oceans” Correspond to Lamin Attachment Domains



Berman et al. 2011 Nature Genetics 43, In Press

Spatial Organization of the Epigenome

- **Lamin Attachment**
- **Late Replication**
- **Epigenetic Instability in Cancer**



Bas Van Steensel,
Curr Opin Cell Biol 2010

- **Active Transcription**
- **Epigenetically Stable in Cancer**

SUMMARY

Epigenetic Subtypes

- CpG Island Methylator Phenotype in Glioblastoma – *IDH1* Mutation

Epigenetic Origins of Cancer

- Polycomb Repressor Binding in ES-Cells Predisposes to Aberrant DNA Methylation in Cancer
- Polycomb Repressor Predisposition Seen Across Cancer Types

The Role of Nuclear Architecture in Epigenetic Instability

- Focal Hypermethylation and Long-Range Hypomethylation Coincide in Partially Methylated Domains (PMDs)
- Epigenetically Unstable PMDs are Associated with Nuclear Lamina Attachment and Late-Replicating Regions

Acknowledgements

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