

EPIGENETICS

THE SECOND GENOME



Memorial Sloan Kettering
Cancer Center™

MRINAL GOUNDER, M.D.

ATTENDING PHYSICIAN

MEMORIAL SLOAN KETTERING CANCER CENTER

DISCLOSURES



- Scientific Advisory Board: Amgen, Daiichi-Sankyo
- Consultant: Karyopharm Therapeutics
- MSKCC PI: Epizyme, EZH2 inhibitor

GENOTYPE \neq PHENOTYPE

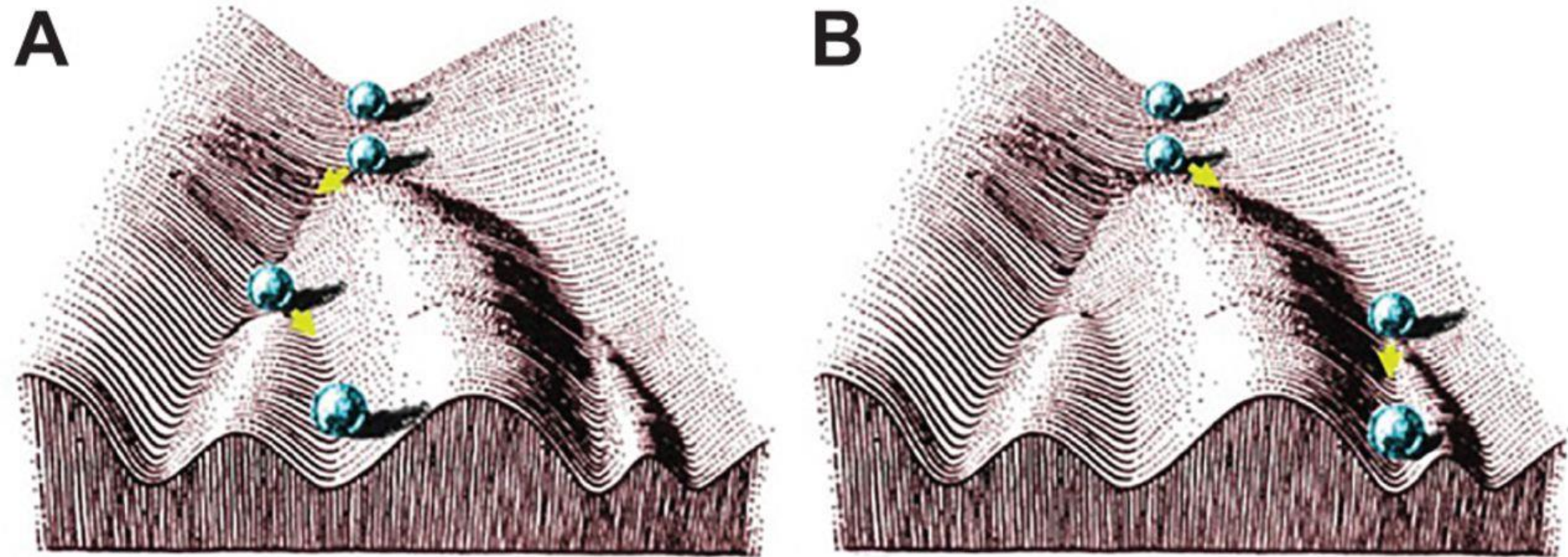


Rainbow



Cc

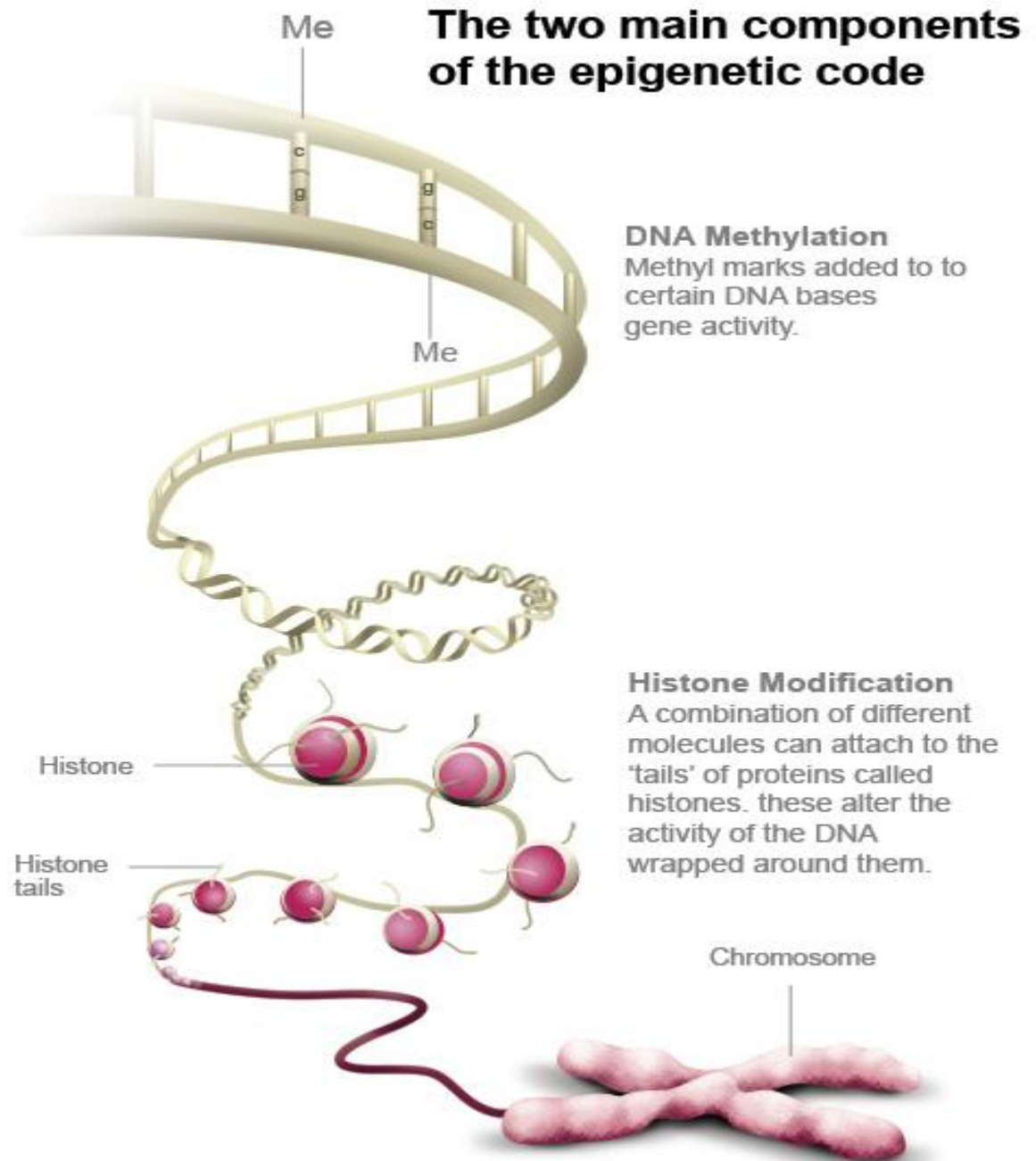
Waddington's Epigenetic landscape



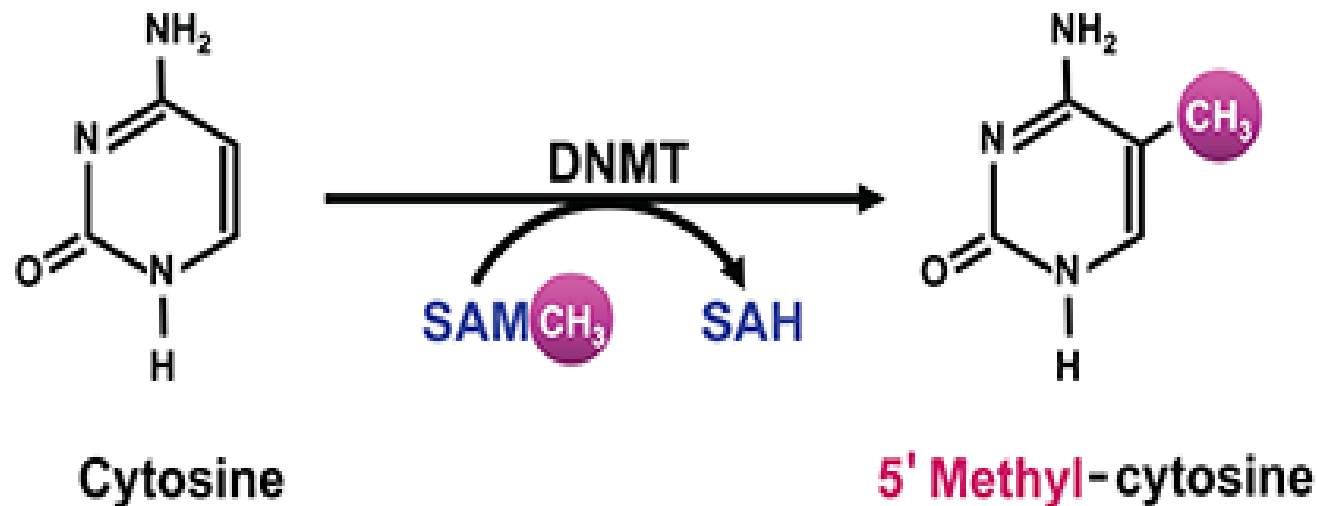
A BRIEF PRIMER ON EPIGENETICS



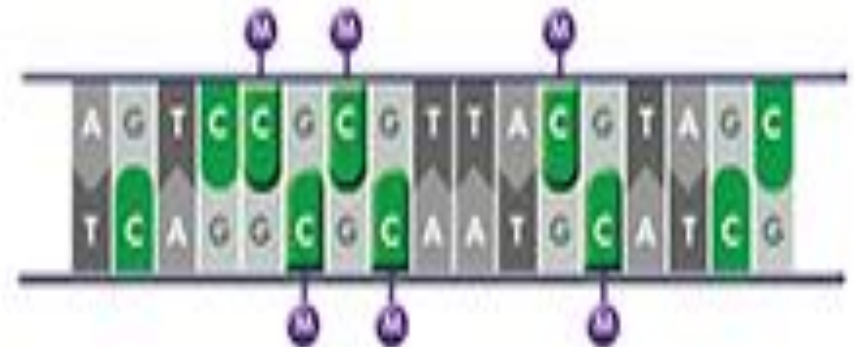
THE EPIGENETIC CODE



DNA METHYLATION :: GENE SILENCING

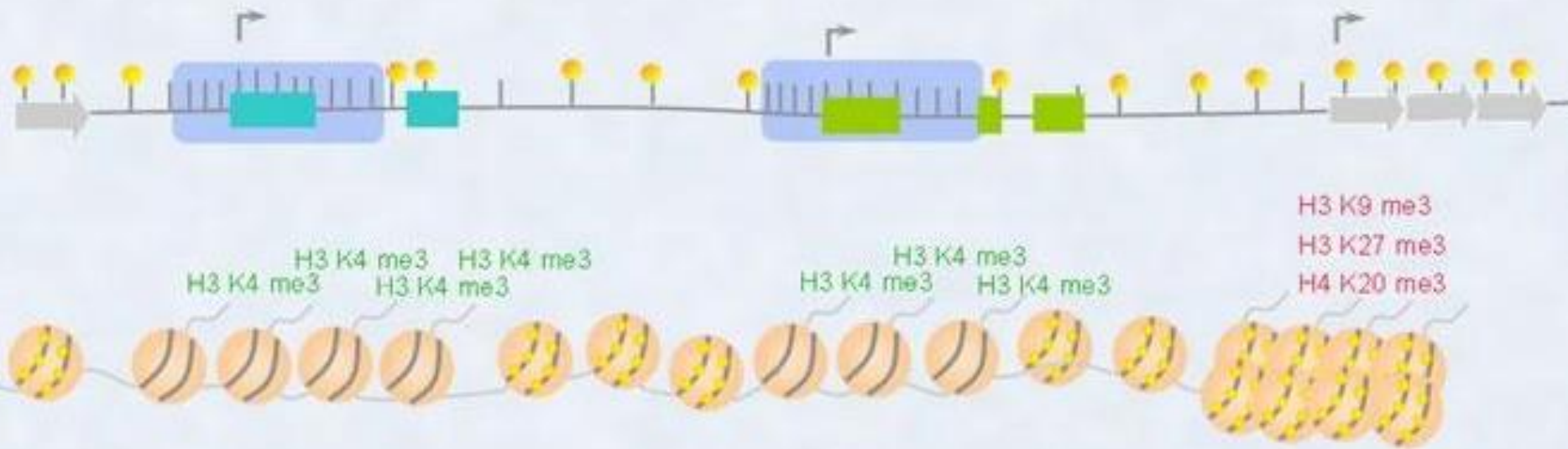


Methylating the cytosine of a CpG motif silences genes



DNA METHYLATION :: GENE SILENCING

Normal Cells



Transcriptional potential

HISTONE MODIFICATION

The **nucleosome** consists of 146bp of DNA wrapped around a protein core of 8 histones

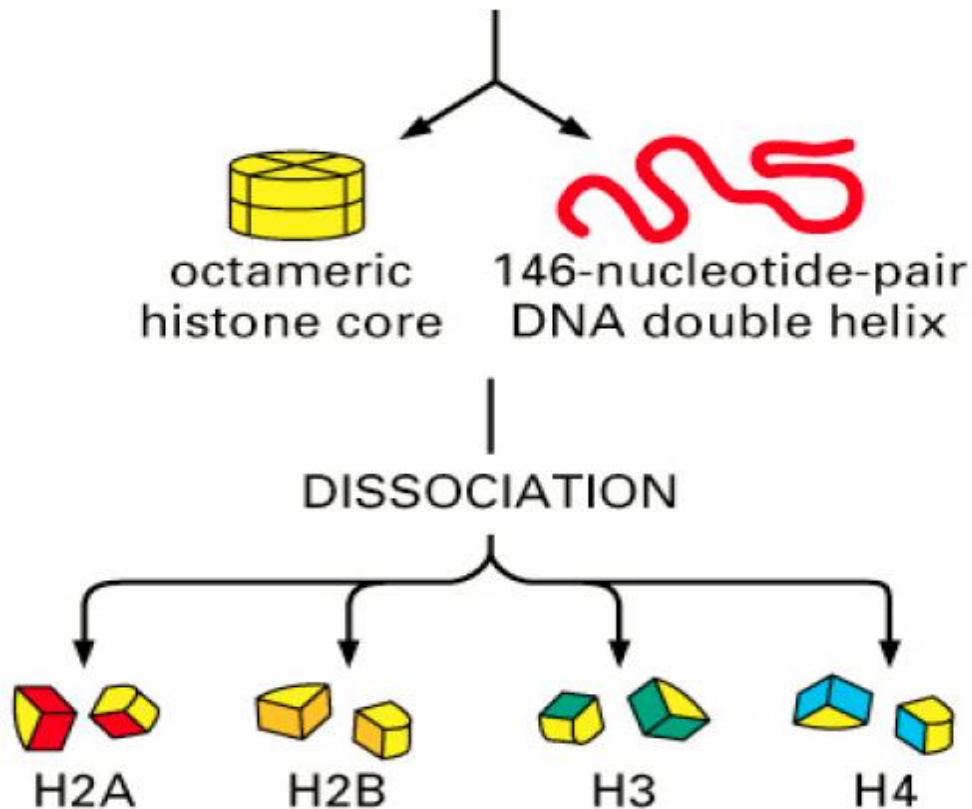


Figure 4-24 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

HISTONE TAILS:: MODIFICATIONS

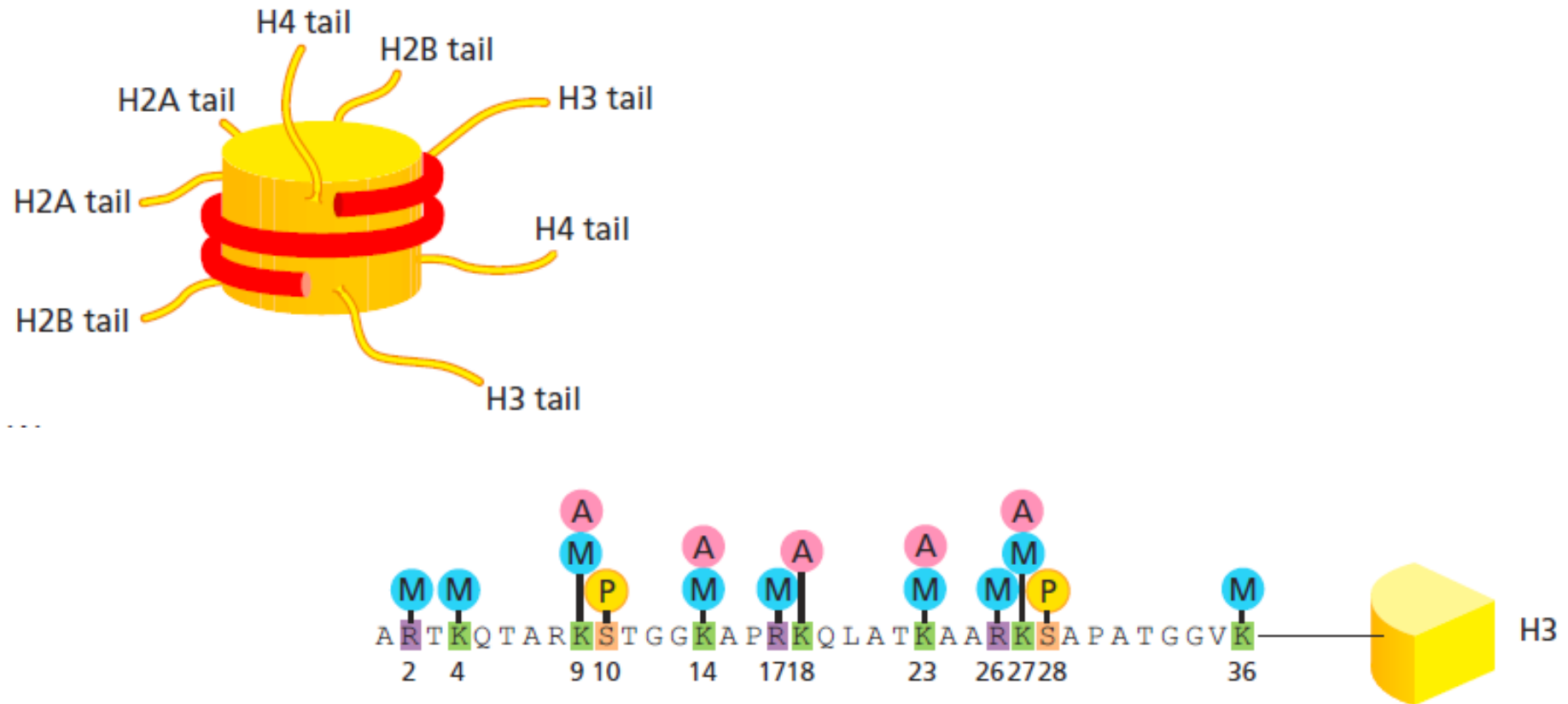


Figure 4-29 and 4-34: Molecular Biology of the Cell, 6th Edition

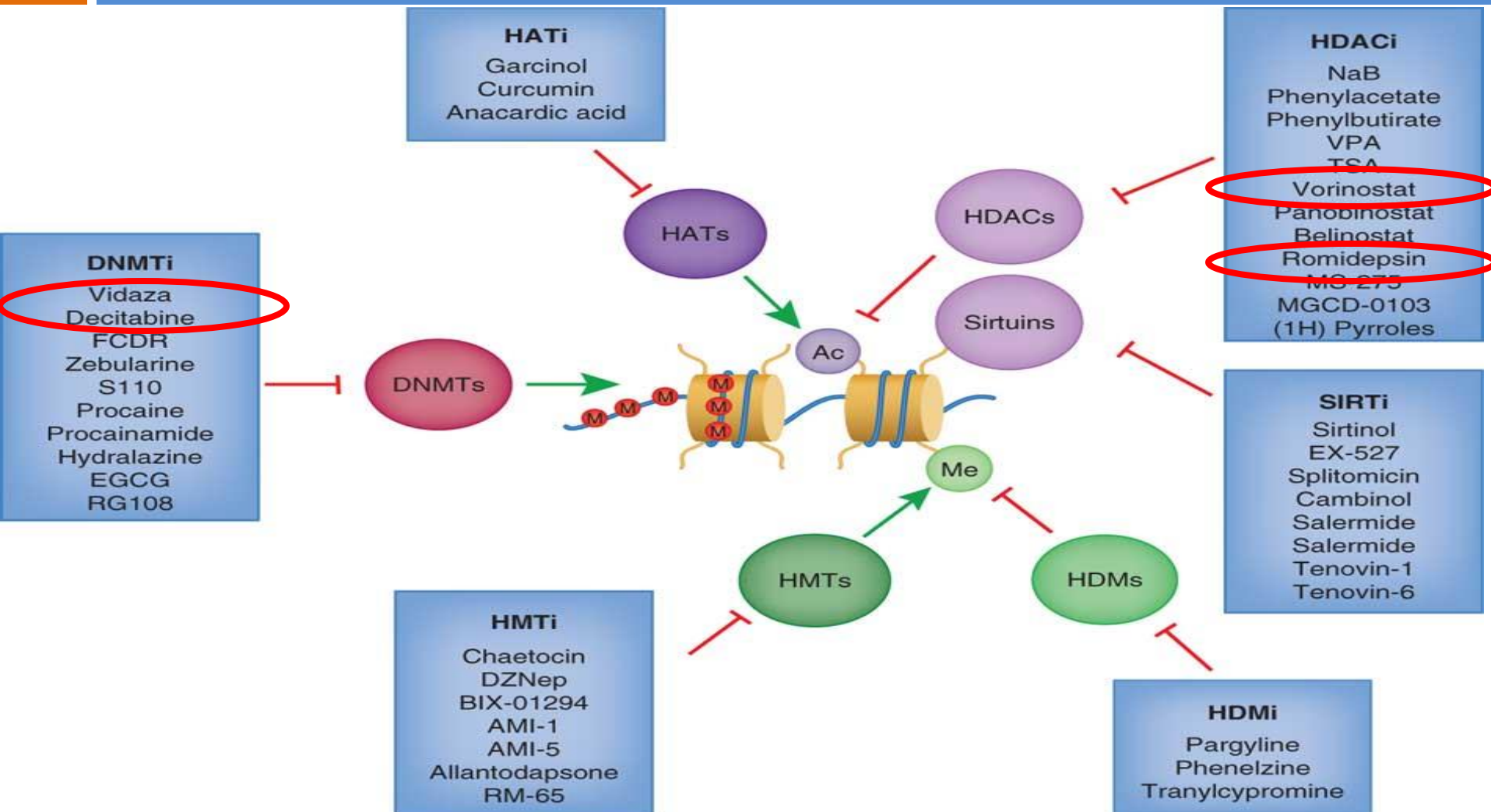
HISTONE “CODE”

<u>Mark</u>	<u>Transcriptionally relevant sites</u>	<u>Biological Role</u>
Methylated cytosine (meC)	CpG islands	Transcriptional Repression
Acetylated lysine (Kac)	H3 (9,14,18,56), H4 (5,8,13,16), H2A, H2B	Transcriptional Activation
Phosphorylated serine/threonine (S/Tph)	H3 (3,10,28), H2A, H2B	Transcriptional Activation
Methylated arginine (Rme)	H3 (17,23), H4 (3)	Transcriptional Activation
Methylated lysine (Kme)	H3 (4,36,79) H3 (9,27), H4 (20)	Transcriptional Activation Transcriptional Repression
Ubiquitylated lysine (Kub)	H2B (123/120) H2A (119)	Transcriptional Activation Transcriptional Repression
Sumoylated lysine (Ksu)	H2B (6/7), H2A (126)	Transcriptional Repression

EPIGENETICS AND CANCER

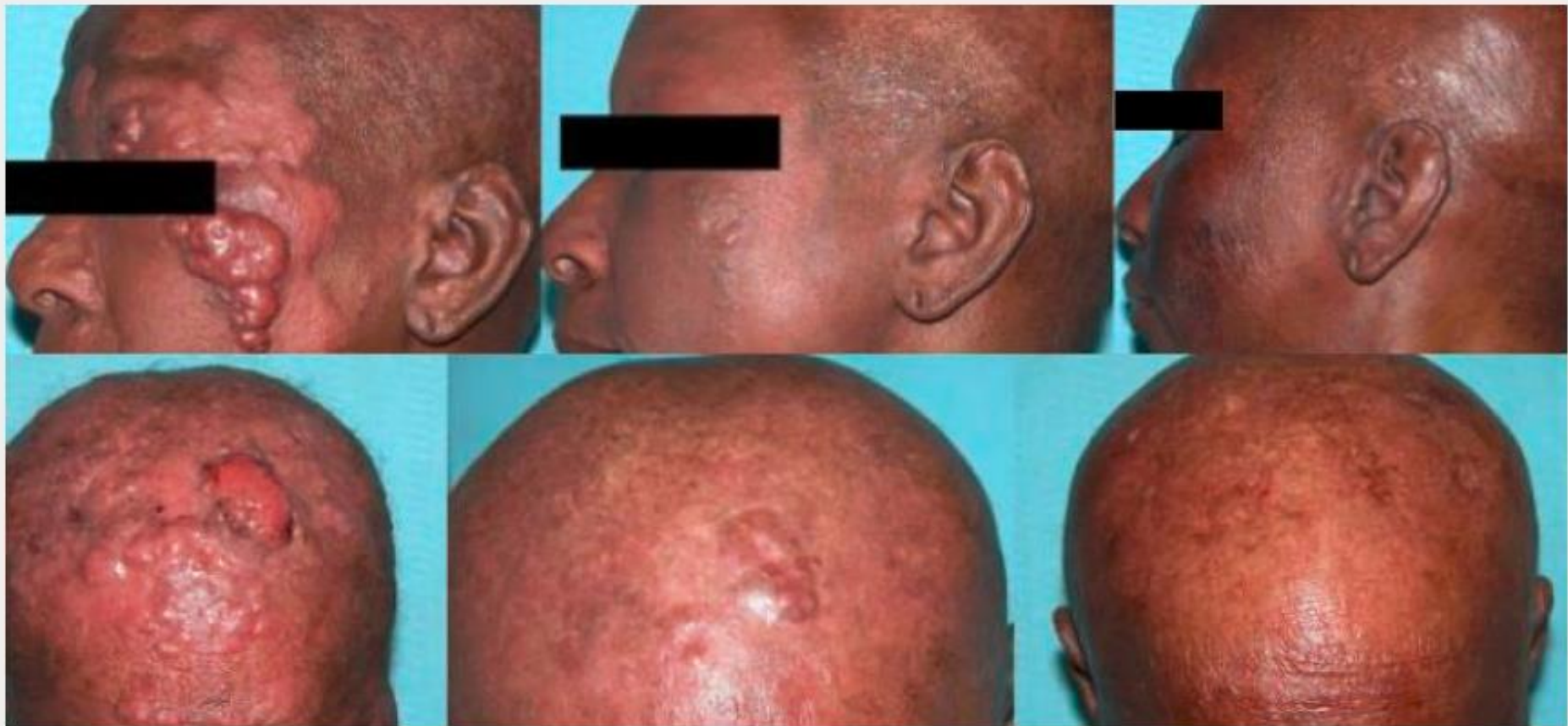


EPIGENETIC DRUGS IN CANCER



HDAC inhibitor: SAHA

Partial response to vorinostat (400 mg qd) in a patient with stage IVB CTCL



Baseline

Week 8

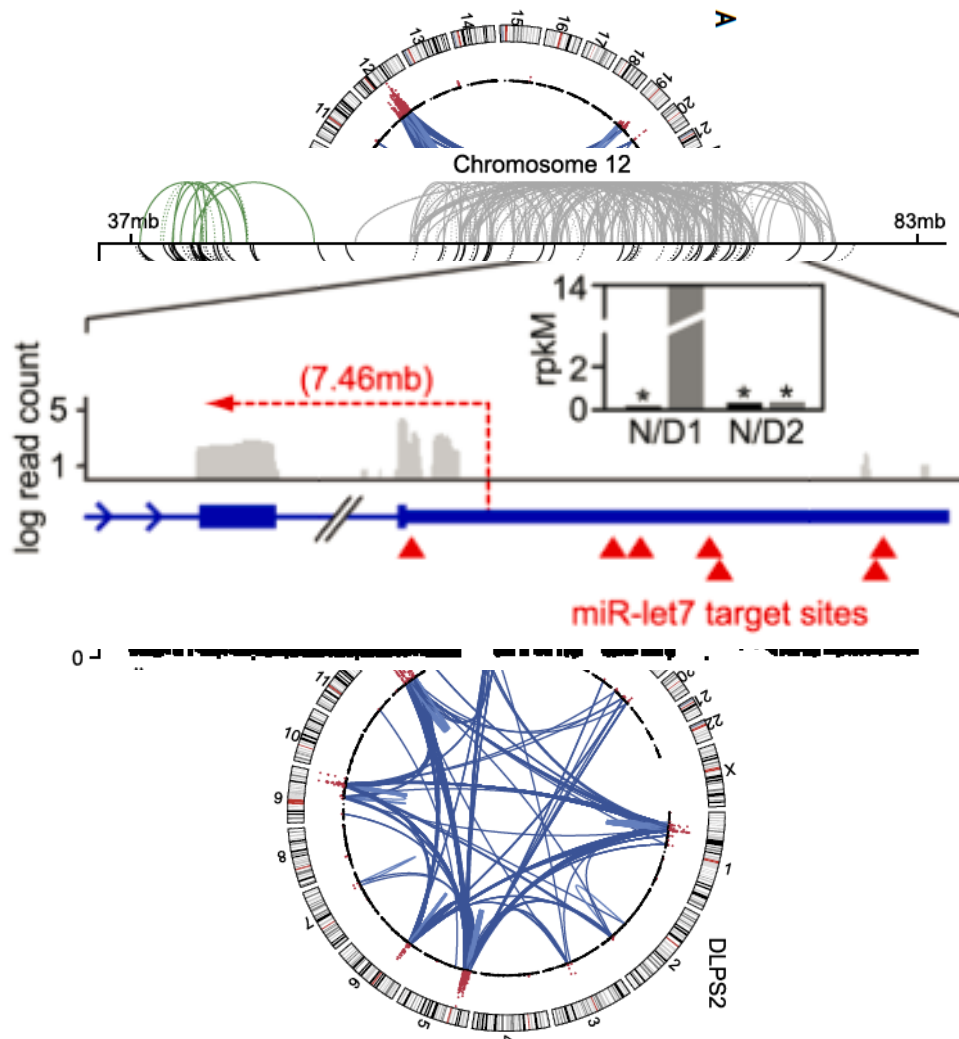
Week 24

Duvic M, et al. *Blood* 2007; Kelly WK et al *JCO* 2005

EPIGENETICS and LIPOSARCOMA

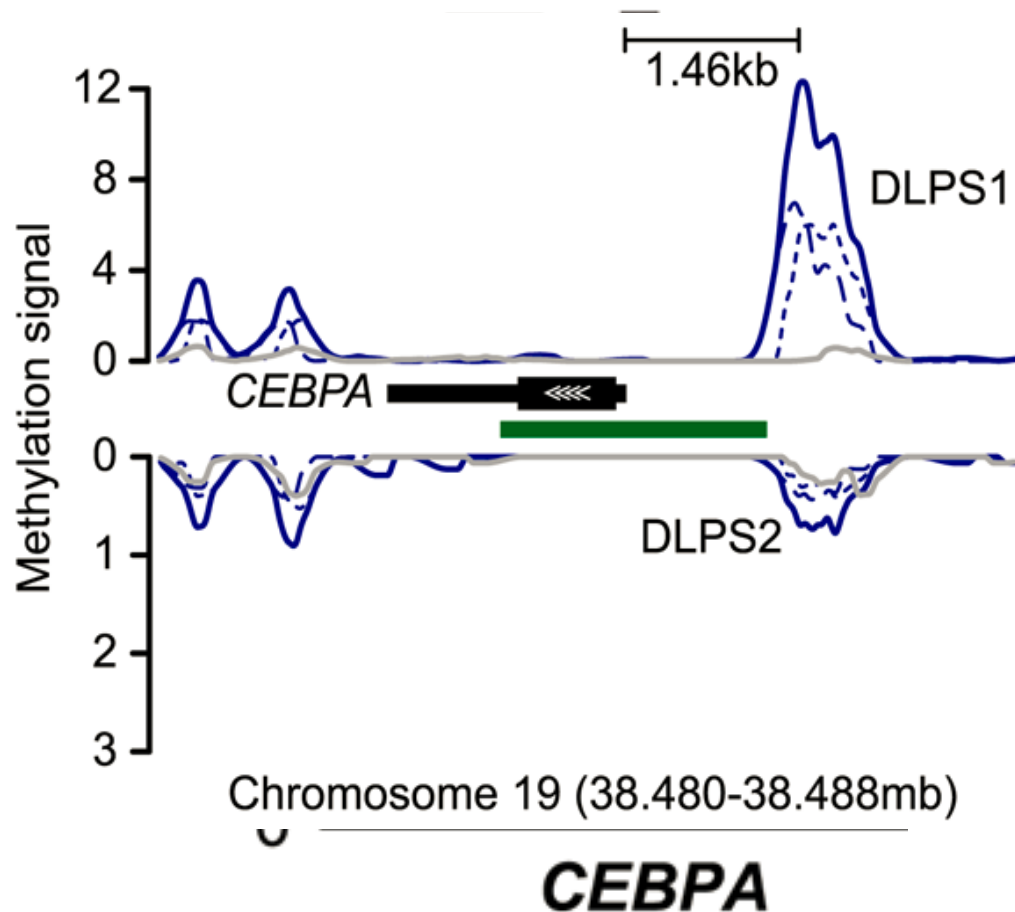


HMGA2 :: CHROMATIN :: miRNA INHIBITION



- 350 – 550 chr rearrangements.
- Majority non-functional
- Hallmark: genomic amplification
- 12q :: CDK4 and MDM2 genes
- HMGA2: High Mobility Group A2
- Mutations in NON-CODING region of DNA
- High mRNA expression: amplification and loss of negative regulatory region (microRNA mediated)

CEBPA METHYLATION:: INHIBITS ADIPOCYTE DIFFERENTIATION



CEBPα is master regulator of adipocyte differentiation.

Low expression = distant relapse
18 – 25 fold reduction in expression
(compared to fat or WWDL).

No mutations or deletions

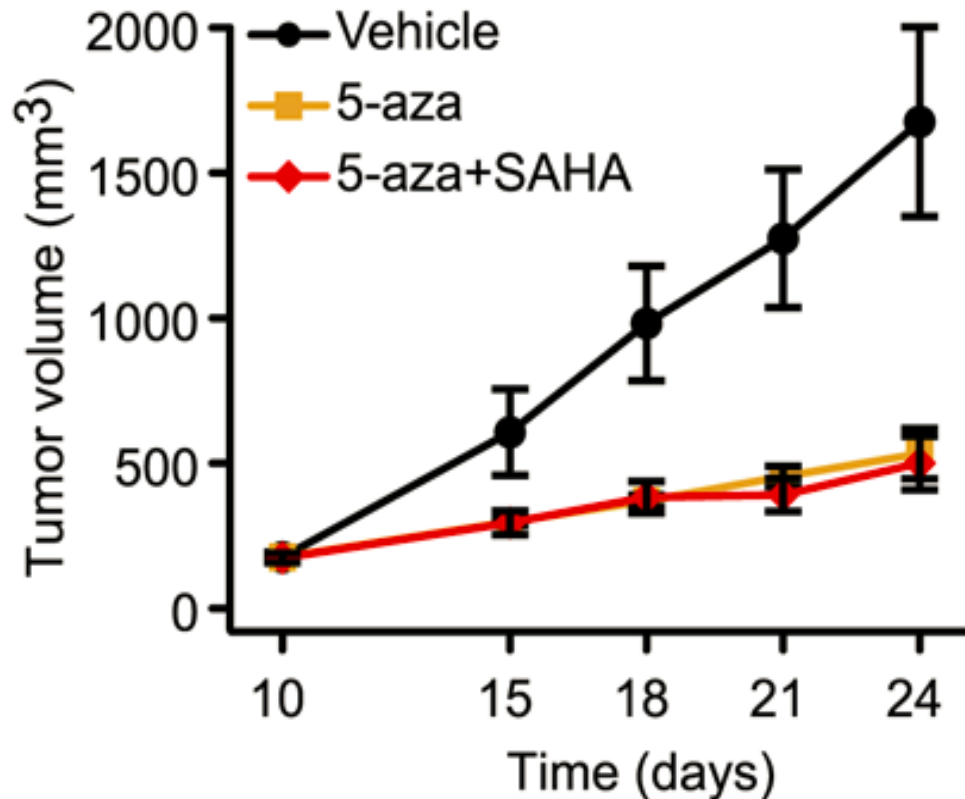
Tumors expressed 2 – 12 fold
higher methylation.

24% of DDLP was methylated

Can we reverse methylation?

Yes, 5-AZA, Decitabine

SOMATIC MUTATIONS IN HDAC1



8 – 13 somatic mutations detected and some in adipogenic genes.

96 tumors and cell lines

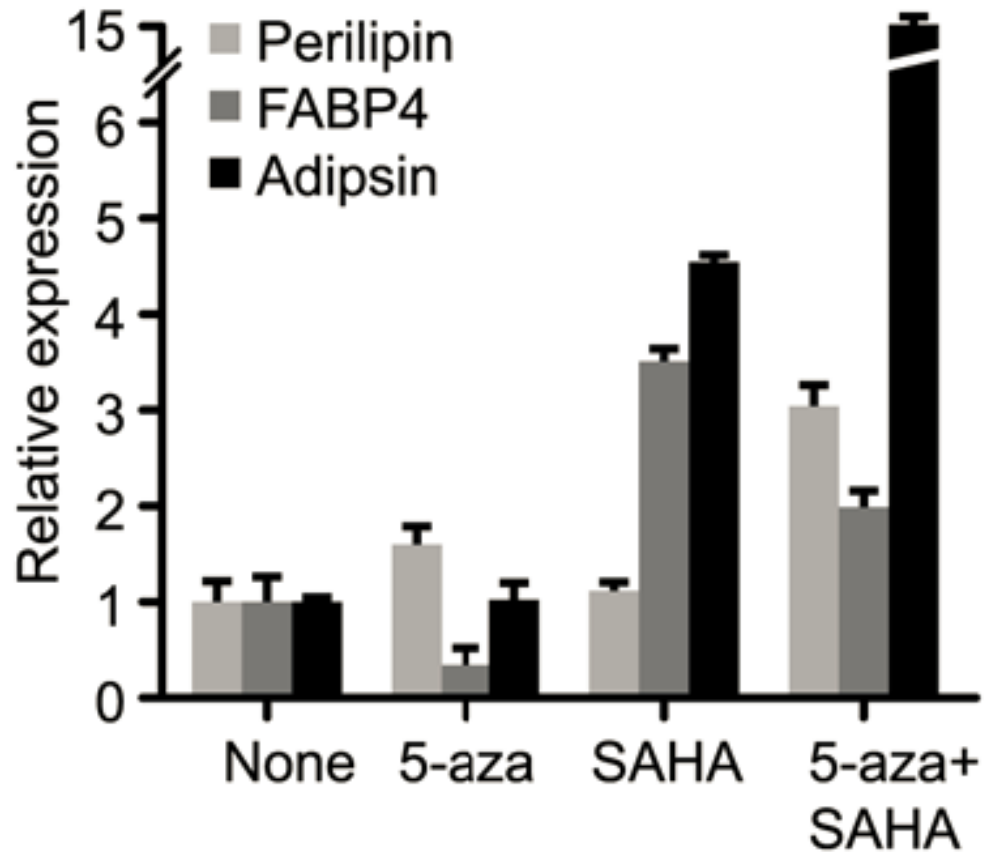
mutHDAC1: 8.1%

Can de-acetylation be reversed?

Yes, HDAC inhibitor: SAHA

Combine: de-Methylation and Acetylation drugs

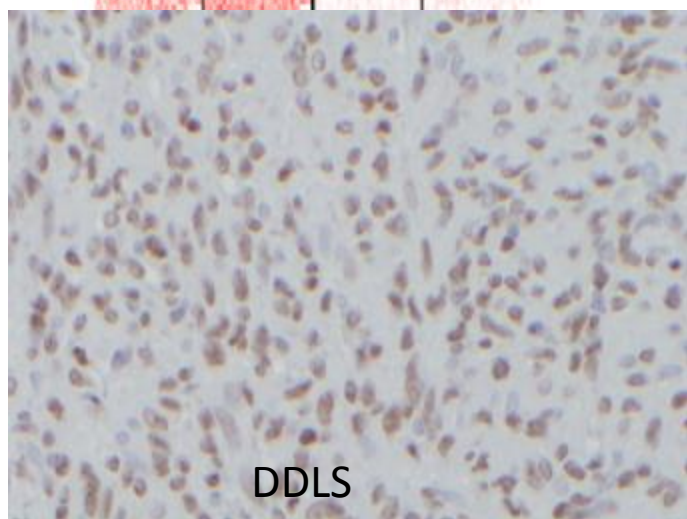
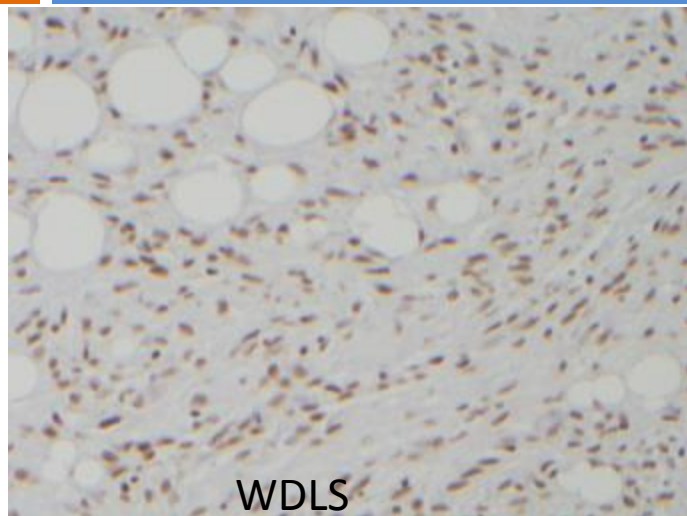
EPIGENETIC REACTIVATION OF ADIPOGENIC PATHWAYS



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DDLs HAS DISTINCT HISTONE MODIFICATIONS



- 151 pt: WWDS, DDLS
- Higher expression of H3K9me3 in DDLS
- H3K9me3 affects different sets of genes between DDLS and WDLS (ChIP-on-chip)
- DDLS and WDLS have different gene expression profiles.

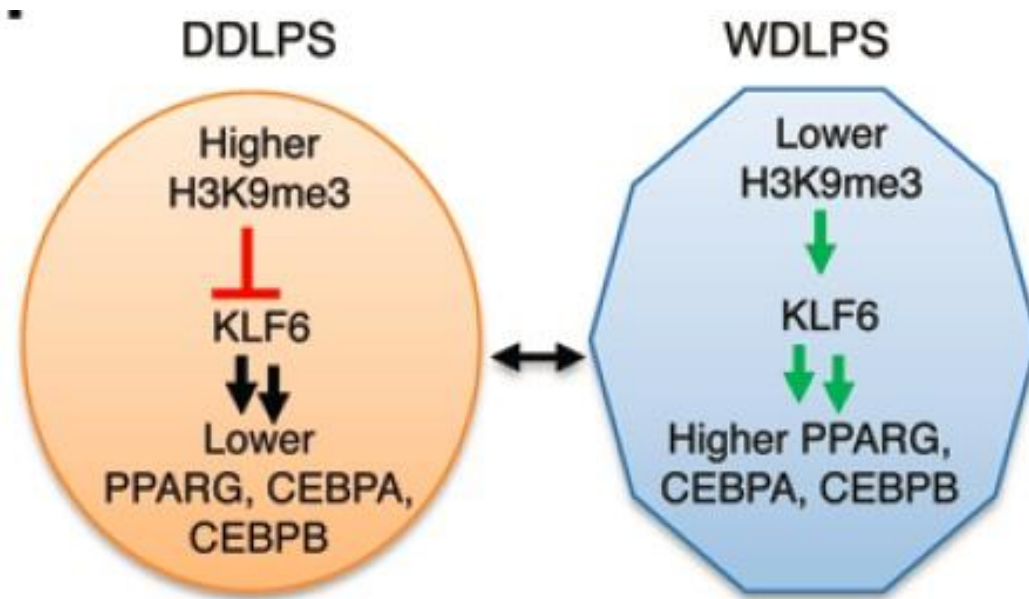
Increased H3K9me3 drives dedifferentiated phenotype via KLF6 repression in liposarcoma

Emily Z. Keung,^{1,2,3} Kadir C. Akdemir,¹ Ghadah A. Al Sannaa,⁴ Jeannine Garnett,² Dina Lev,⁵ Keila E. Torres,² Alexander J. Lazar,⁴ Kunal Rai,¹ and Lynda Chin¹

J Clin Invest. 2015;125(8):2965–2978. doi:10.1172/JCI77976.

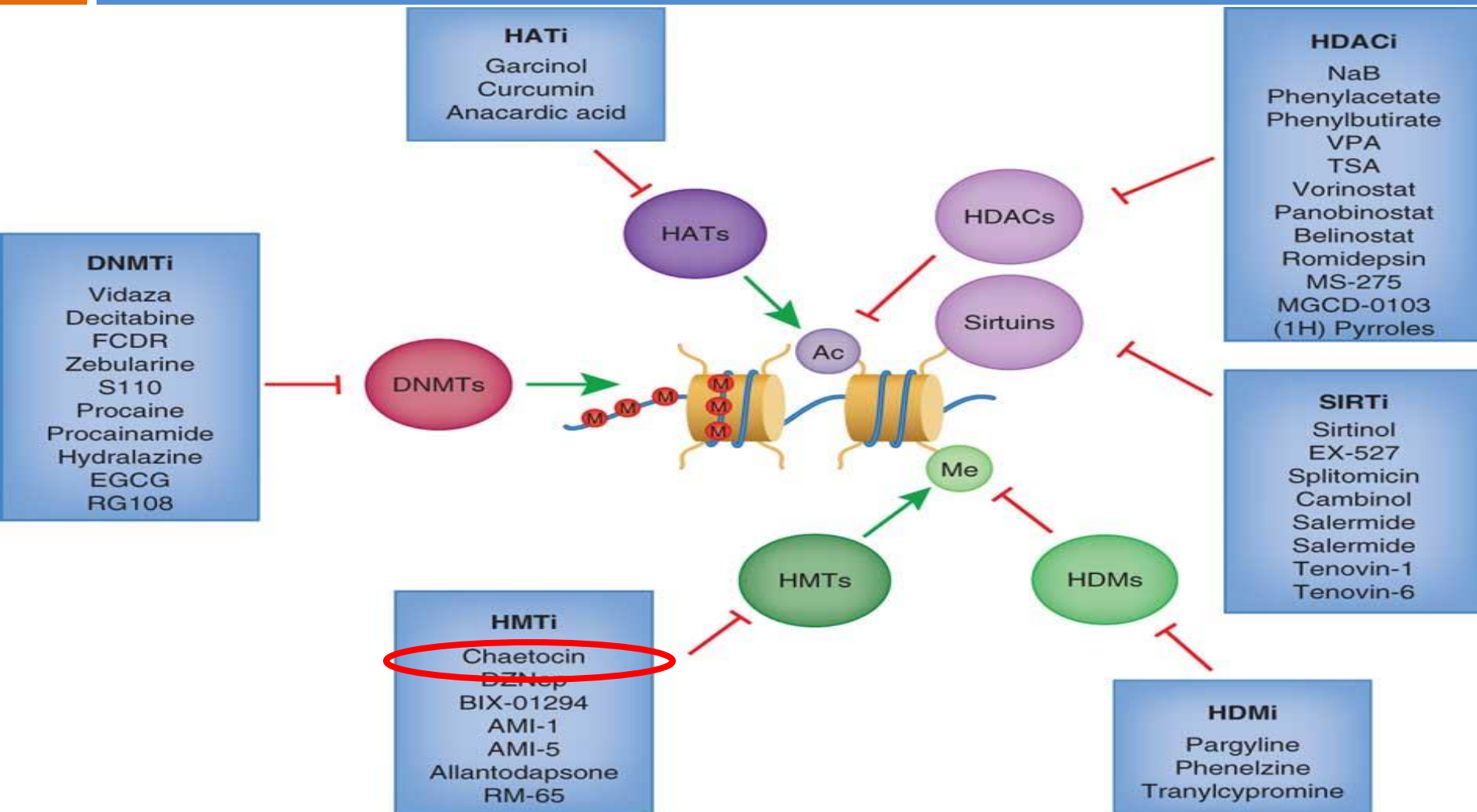
HIGH H3K9me3 AND LOW GENE TRANSCRIPTION

- KLF6: Kruppel-like factor 6
- Tumor suppressor, adipogenesis, migration, metastases, cell cycle, apoptosis.

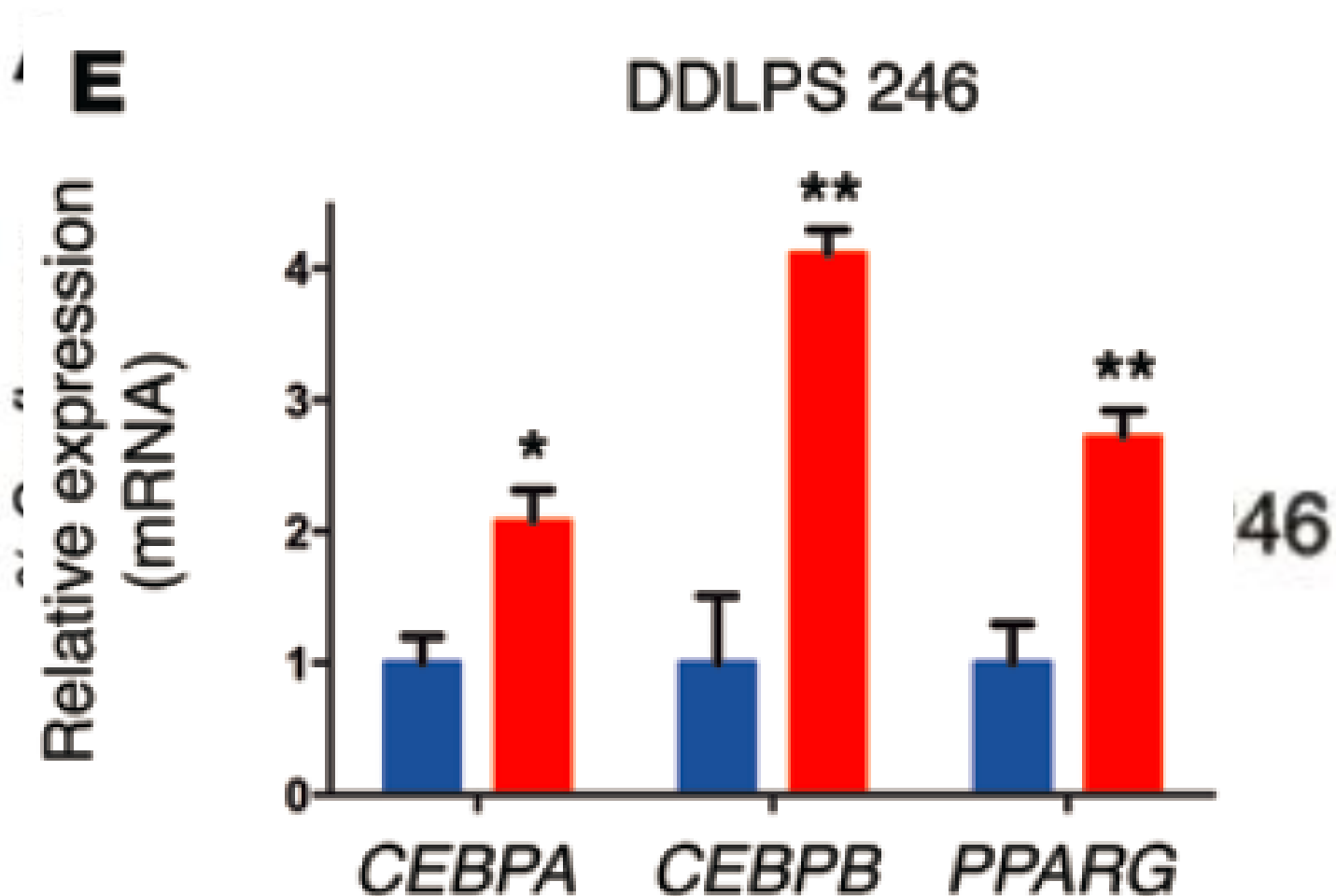


Increased H3K9me3 drives dedifferentiated phenotype via KLF6 repression in liposarcoma

EPIGENETIC DRUGS IN CANCER



Chaetocin: inhibits methylation



SUMMARY: EPIGENETICS AND LIPOSARCOMA

- Complex chromosomal rearrangements
- microRNA controls expression on non-coding DNA
- There are differences in methylation in many genes involved in adipogenesis (CBEPa, KLF4, KLF6).
- Mutations: HDAC1 (8%)
- Combination of de-methylating, HDAC, de-methylating agents :: growth arrest, tumor regression and adipogenic differentiation.

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THANK YOU