Genomics & Medicine

http://biochem118.stanford.edu/

Epigenetics

http://biochem118.stanford.edu/16%20Epigenetics.html

Doug Brutlag, Professor Emeritus
Biochemistry and Medicine (by courtesy)
Stanford University School of Medicine

Epigenetics and DNA Methylation on Henry Stewart Talks

- Epigenetics – 22 talks
  http://hstalks.com/main/browse_talks.php?r=18&j=757&c=252

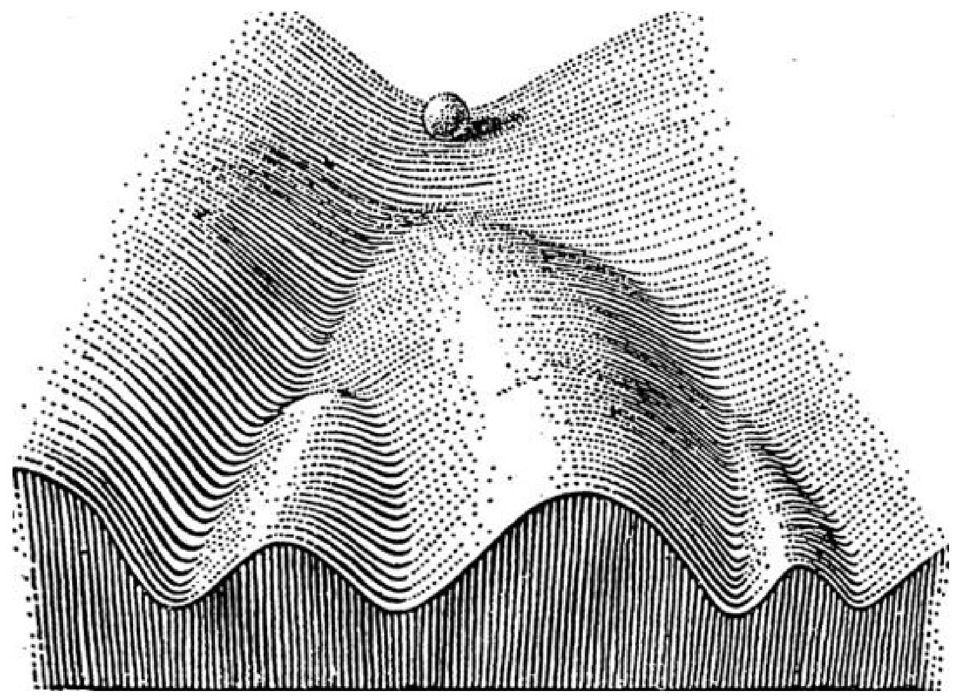
- DNA Methylation – 11 talks
  http://hstalks.com/main/browse_talks.php?r=478&j=756&c=252

- Stem Cells – 10 talks
  http://www.hstalks.com/main/browse_talks.php?r=752&j=761&c=252

What is Epigenetics?

- In 1953, C.H. Waddington coined the term epigenetics to mean above or in addition to genetics to explain differentiation.
- How do different adult stem cells know their fate?
  - Myoblasts can only form muscle cells
  - Keratinocytes only form skin cells
  - Hematopoietic stem cells only become blood cells
  - But all have identical DNA sequences.

C.H. Waddington



Waddington's Epigenetic Landscape

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- Modern definition is non-sequence dependent inheritance.
- How can identical twins have different natural hair colors?

Identical Twins with Different Hair Color

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- How can a single individual have two different eye colors?

An Individual with Two Different Eye Colors

"Diego"

Mosaicism:
An Individual with Two Different Eye Colors

"Josie Too"

Mosaicism:
An Individual Eye with Two Colors

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- How can identical twin liter mates show different coat colors?

Coat Colors of Genetically Identical Agouti Mice Liter Mates

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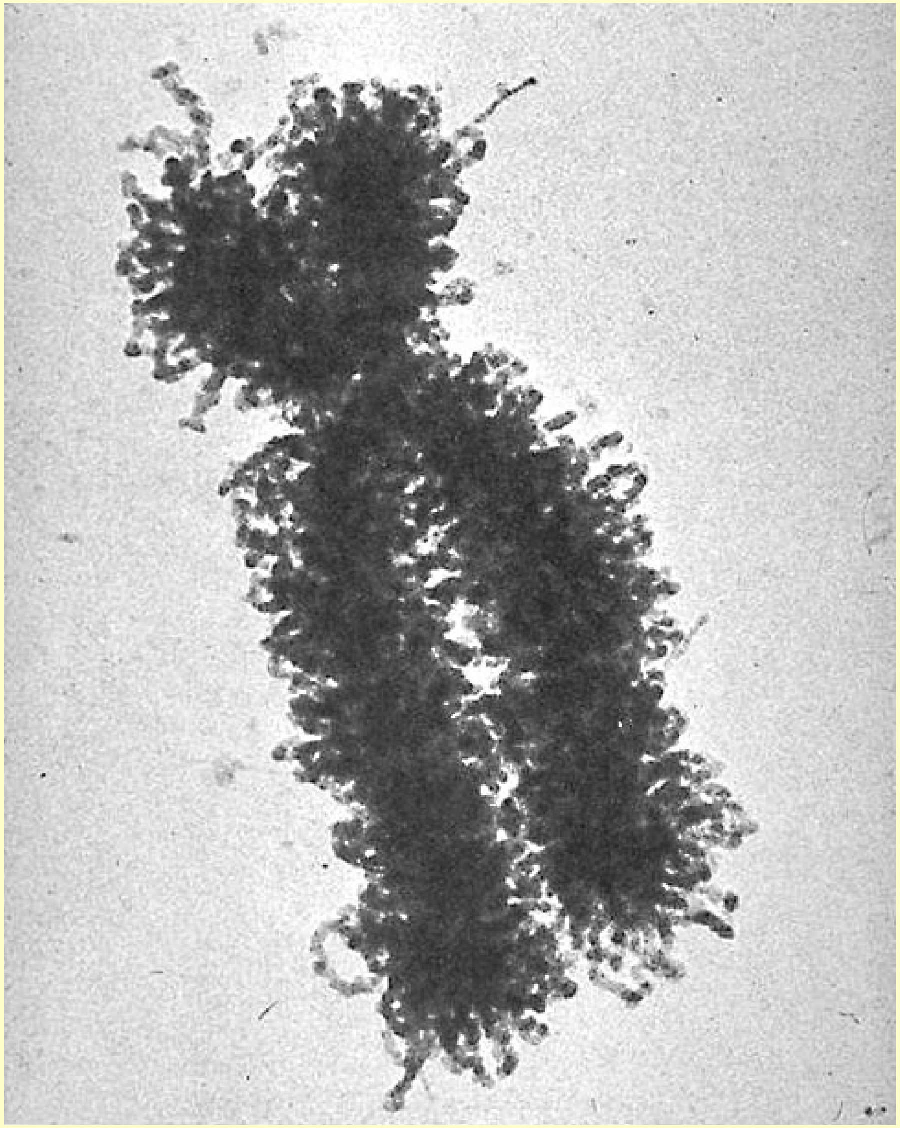
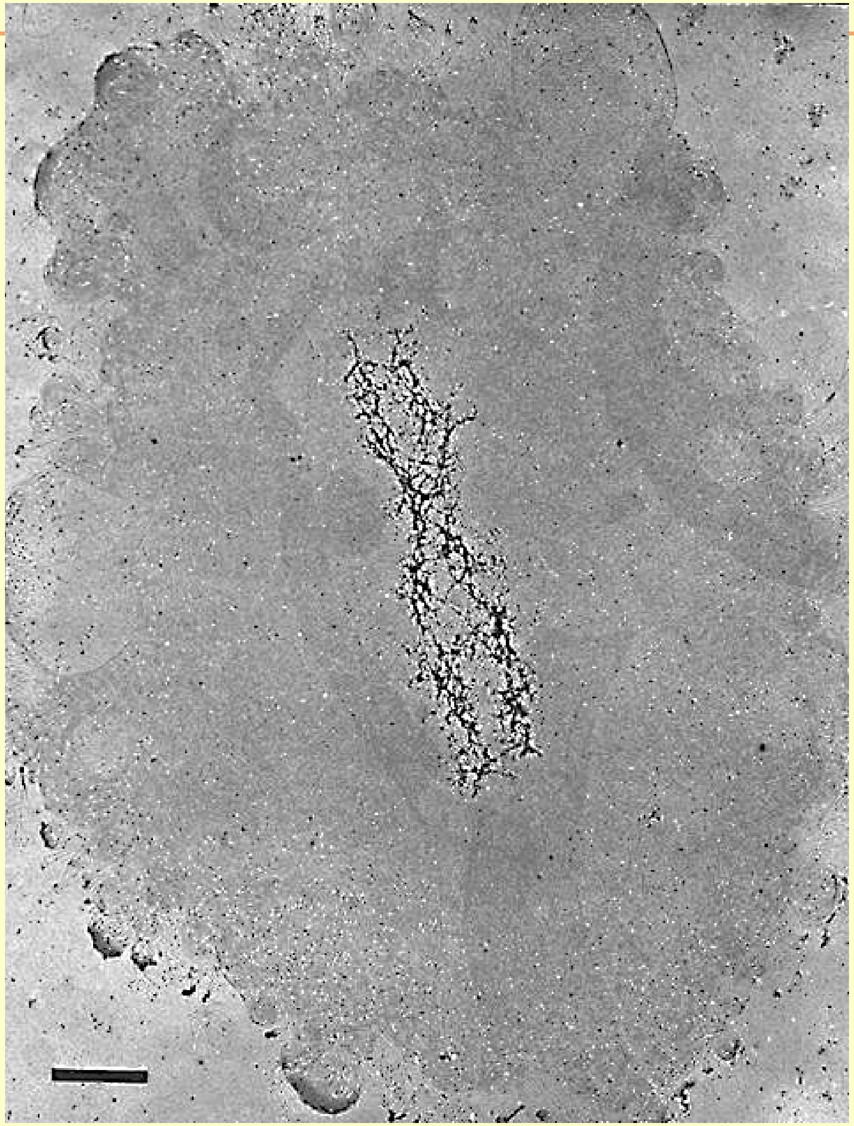
- How can a single individual have two different eye colors?
- How can identical twin liter mates show different coat colors?
- How can just paternal or maternal traits be expressed in offspring? This is called genetic imprinting.
- How can females express only one X chromosome per cell?
- How can acquired traits be passed on to offspring?
- Some changes in gene expression that are, in fact, heritable!

Human Mitotic Chromosome



DNA in a Human Chromosome

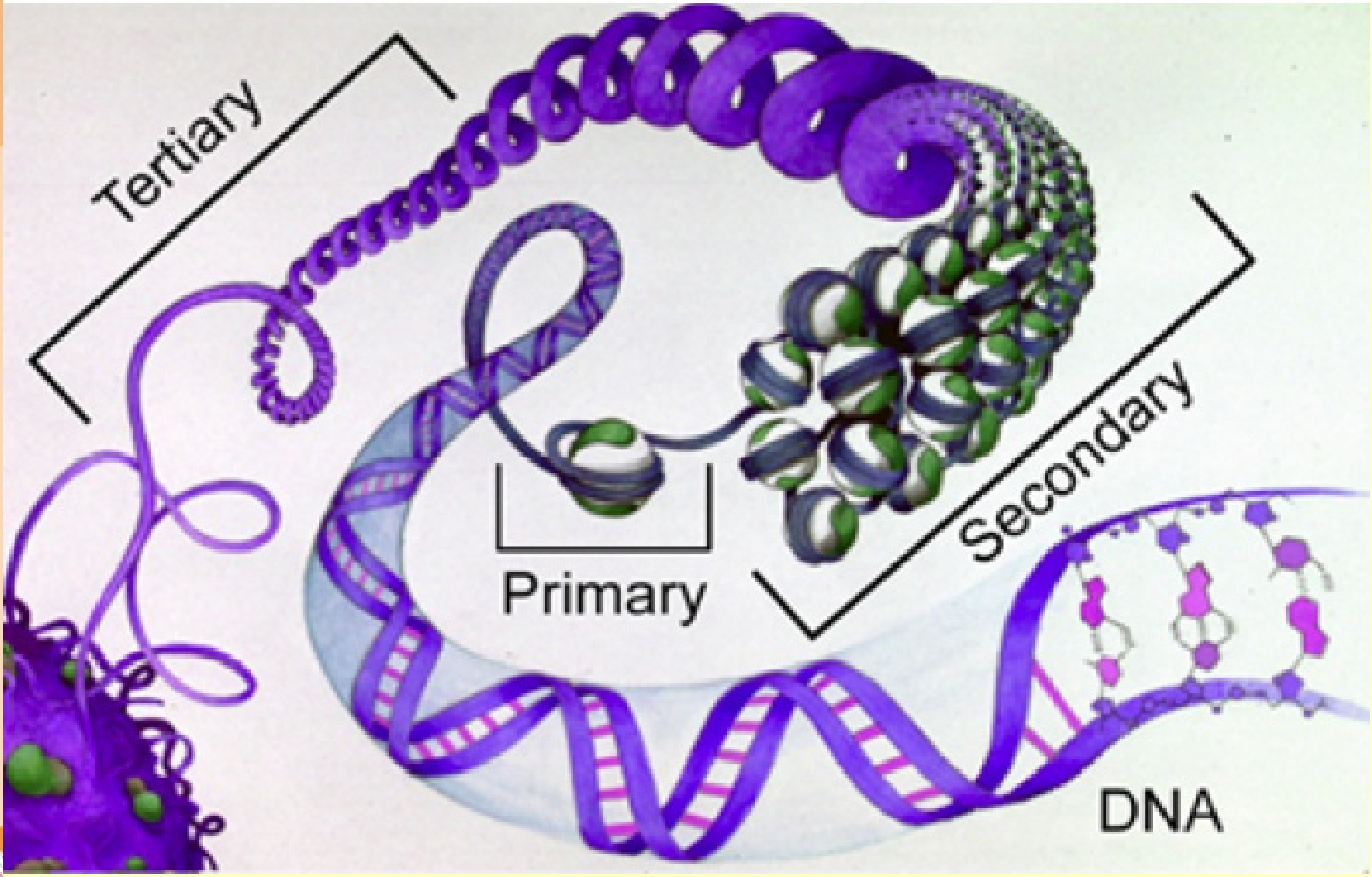


Laemmli: Histone Depleted chromosomes Cell 1977 817-828
https://en.wikipedia.org/wiki/Heparan_sulfate

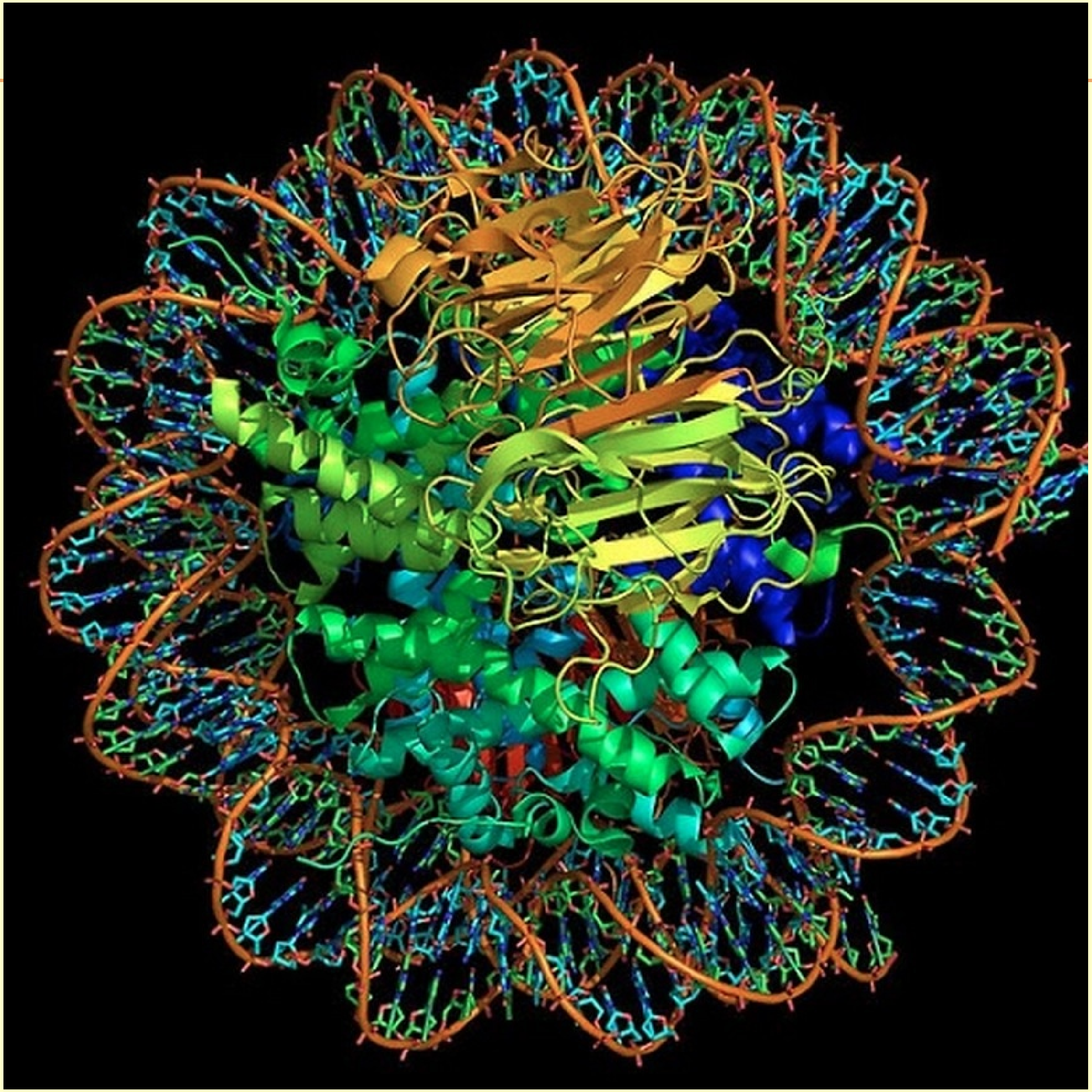
DNA in a Human Chromosome

Laemmli: Histone Depleted chromosomes Cell 1977 817-828

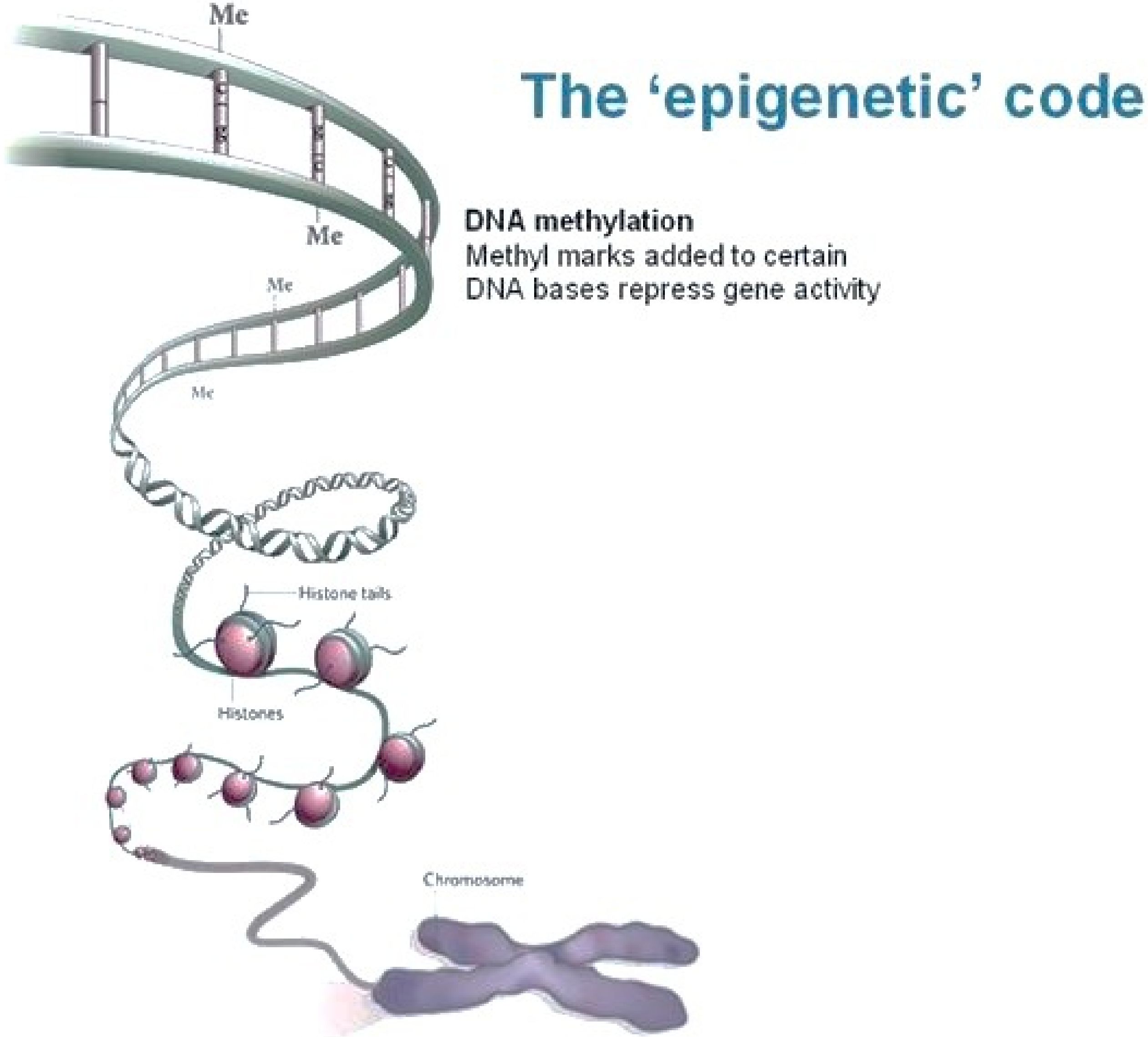
Three Levels of Folding of DNA in Chromatin



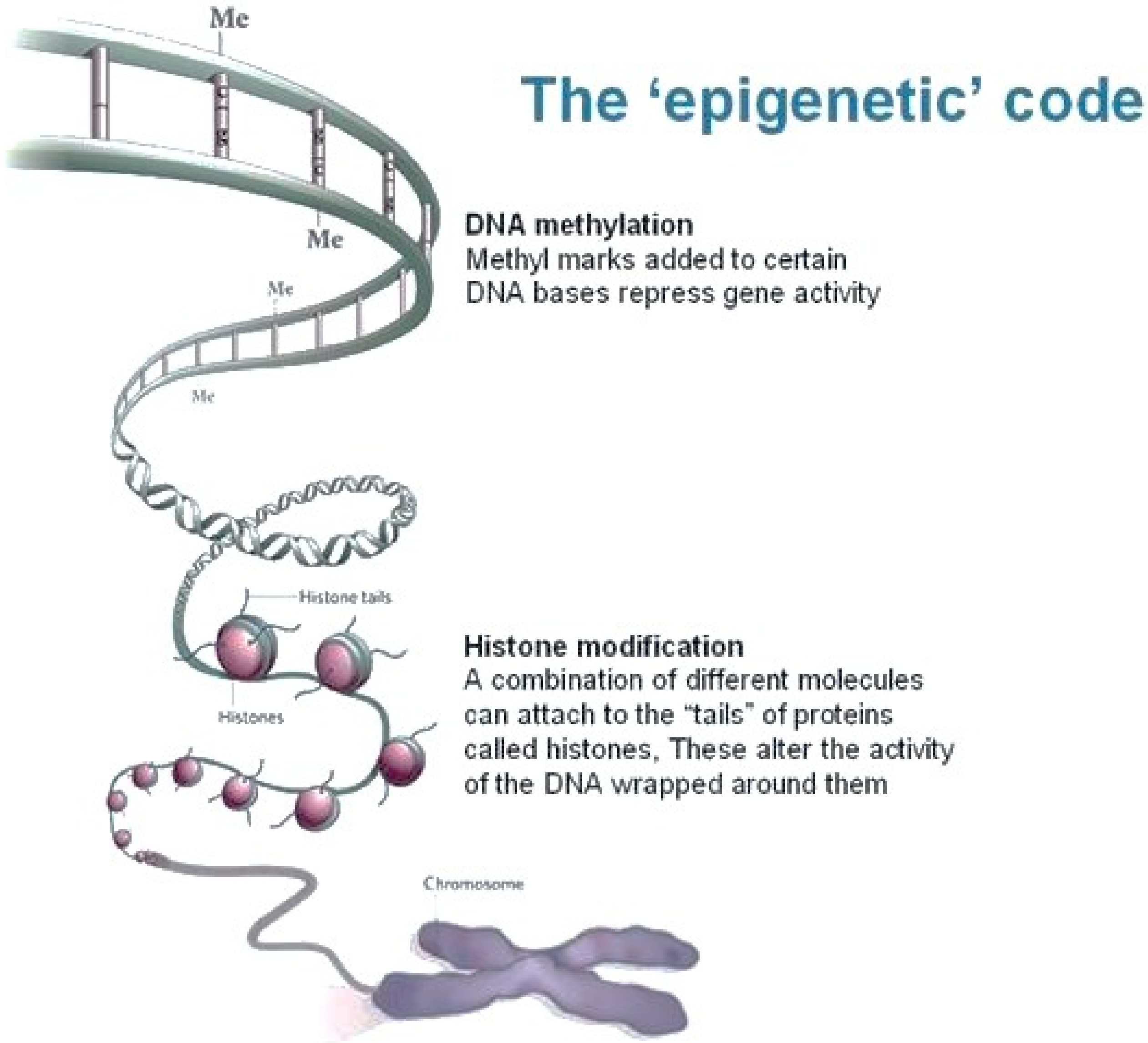
Nucleosome Core Structure



DNA Methylation & the Epigenetic Code

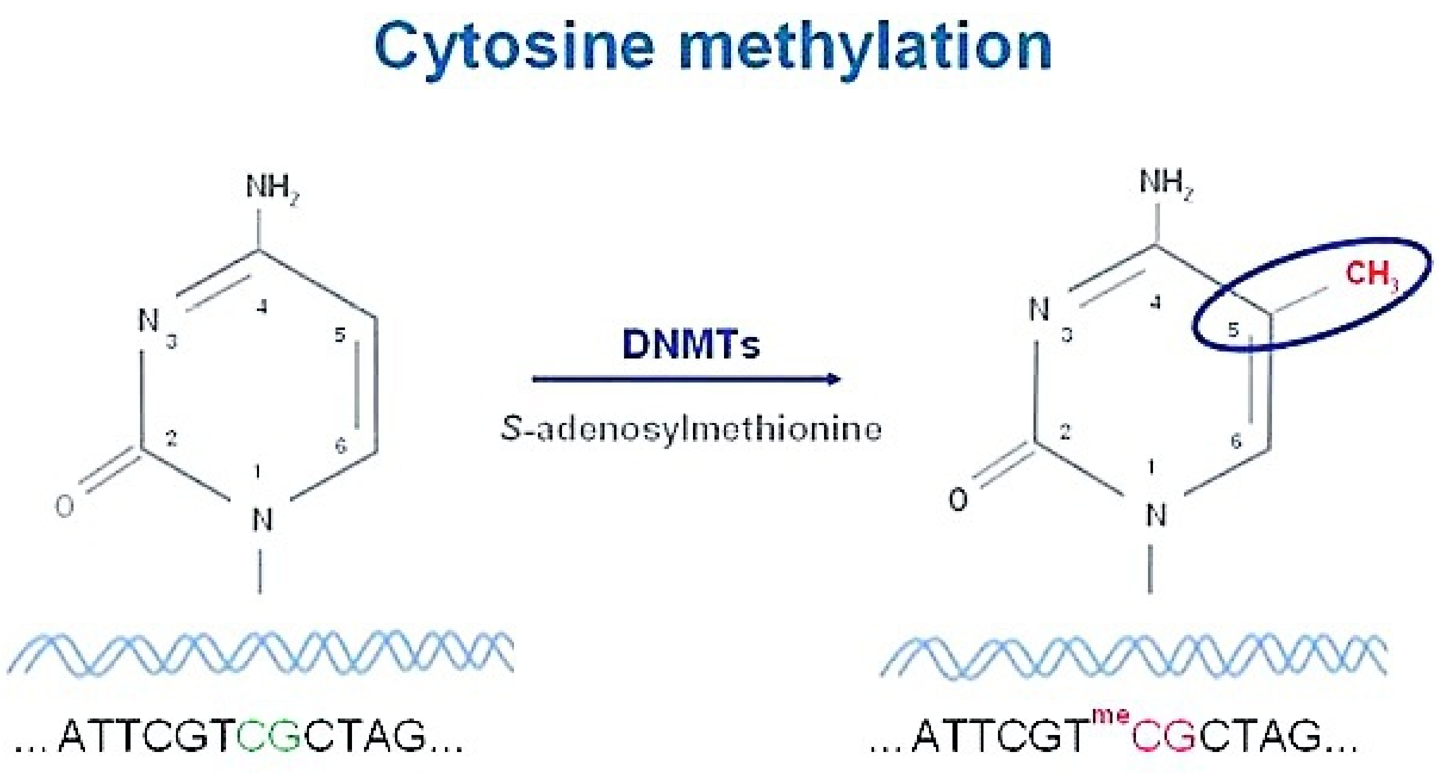


DNA Methylation & Histone Modifications Form the Epigenetic Code



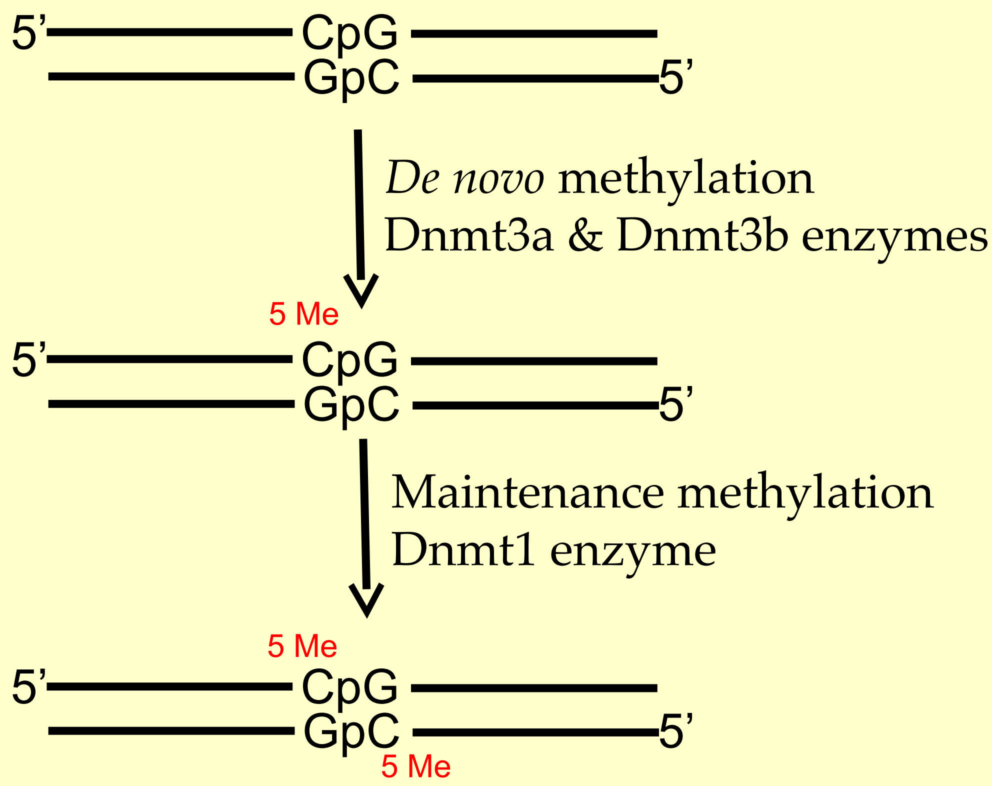
Paula Vertino, Henry Stewart Talks

Methylation of Cytosine in DNA



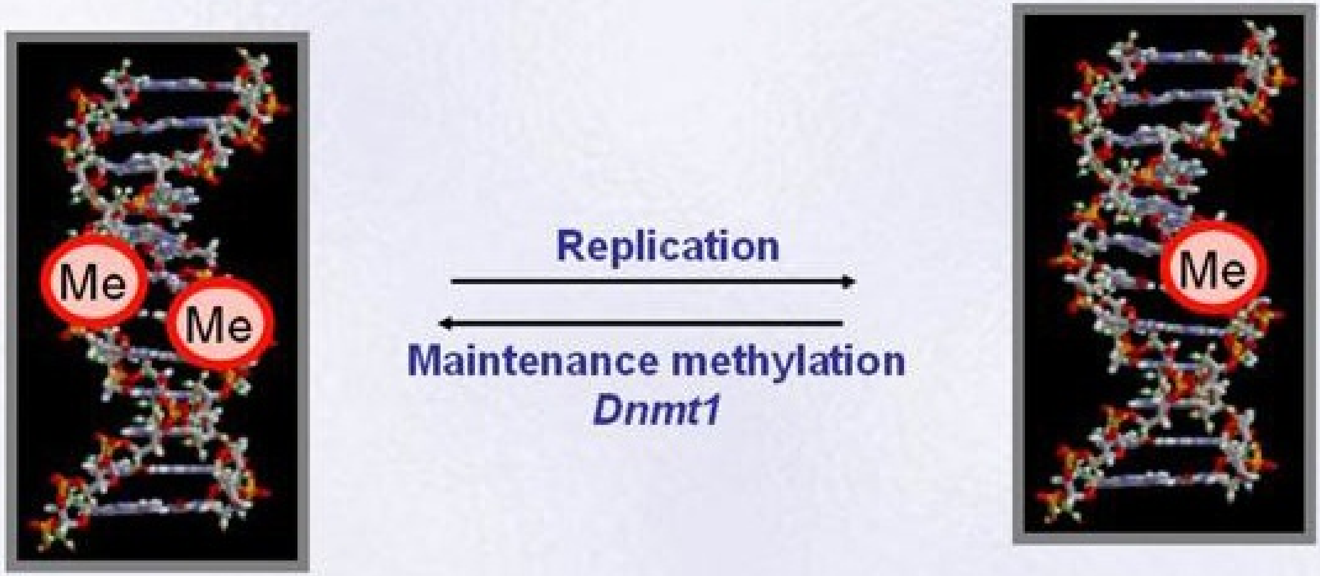
Paula Vertino, Henry Stewart Talks

Only Cs in CG sequences are Methylated

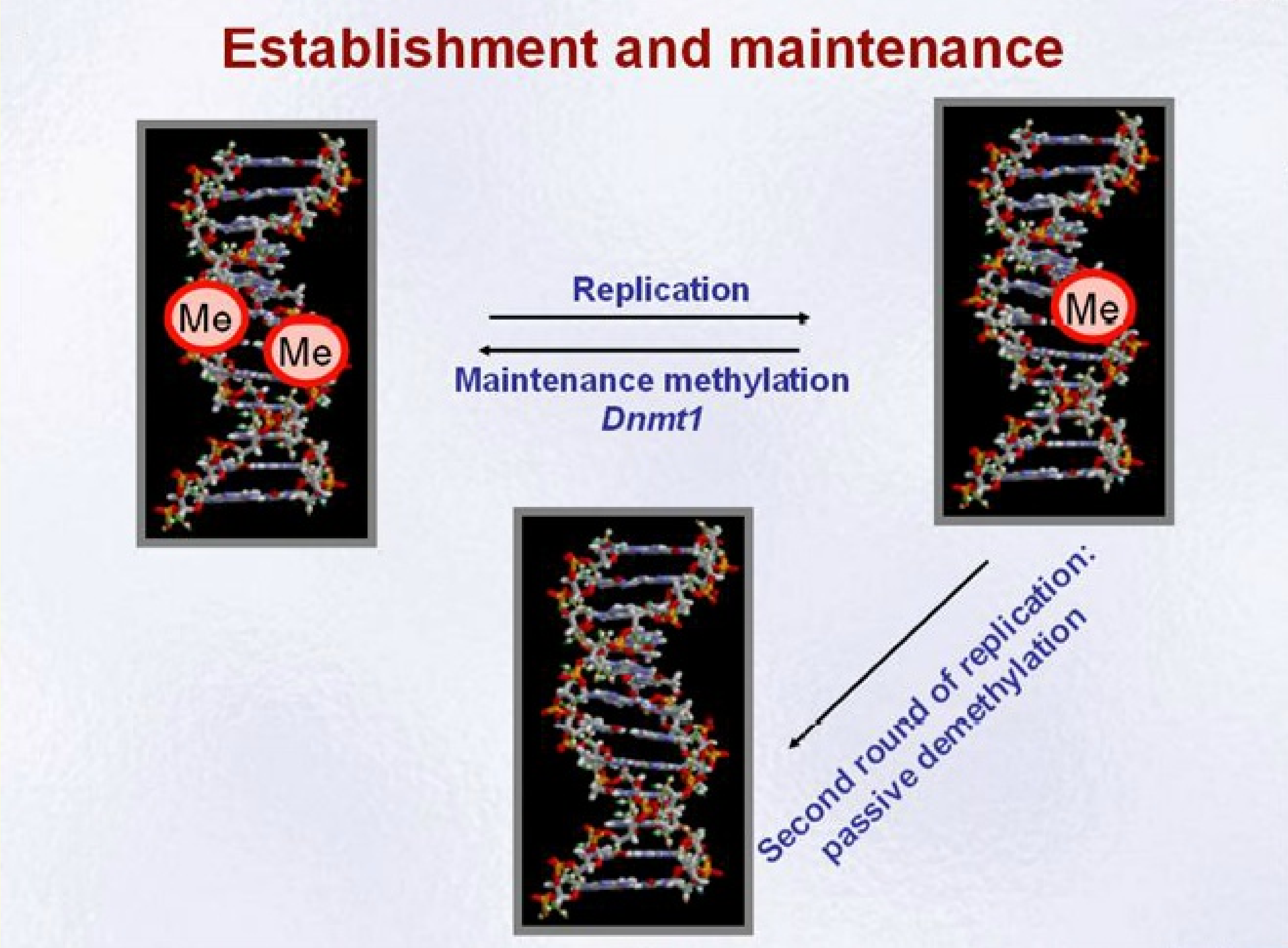


Maintenance of Cytosine Methylation

Establishment and maintenance

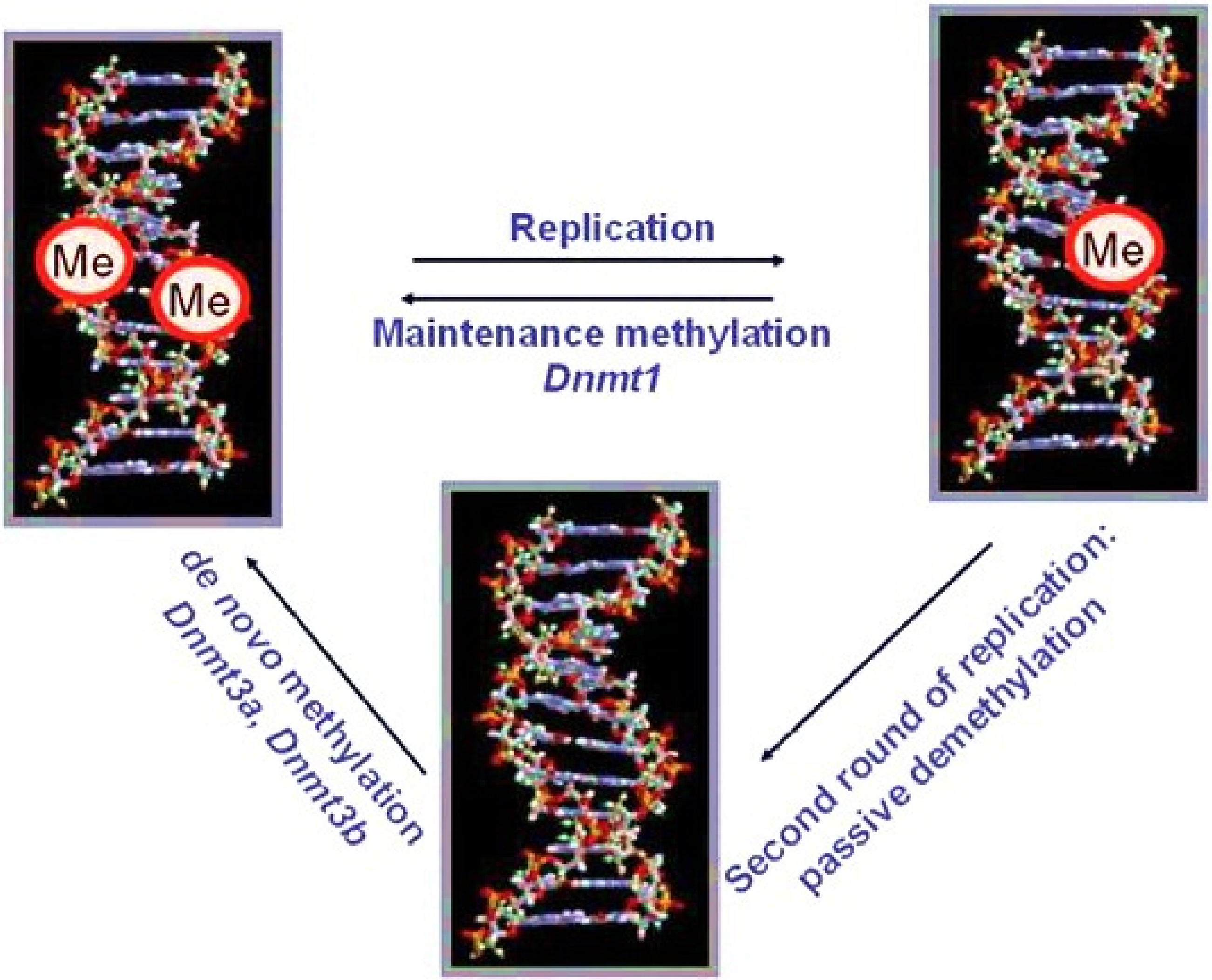


Passive Demethylation of 5-Methyl-Cytosine



Alex Meissner, Henry Stewart Talks

Establishment and Maintenance of Cytosine Methylation



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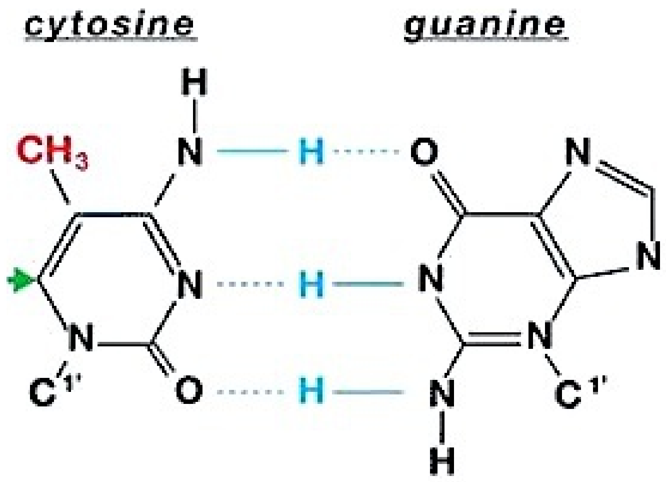
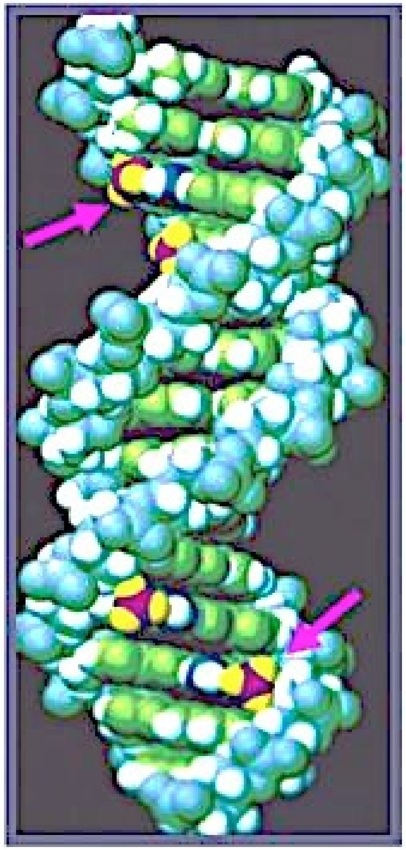
5-Methyl Cytosine in DNA

Cytosine methylation





Some DNA Methyl Transferases are Essential

Mammalian Dnmts are essential

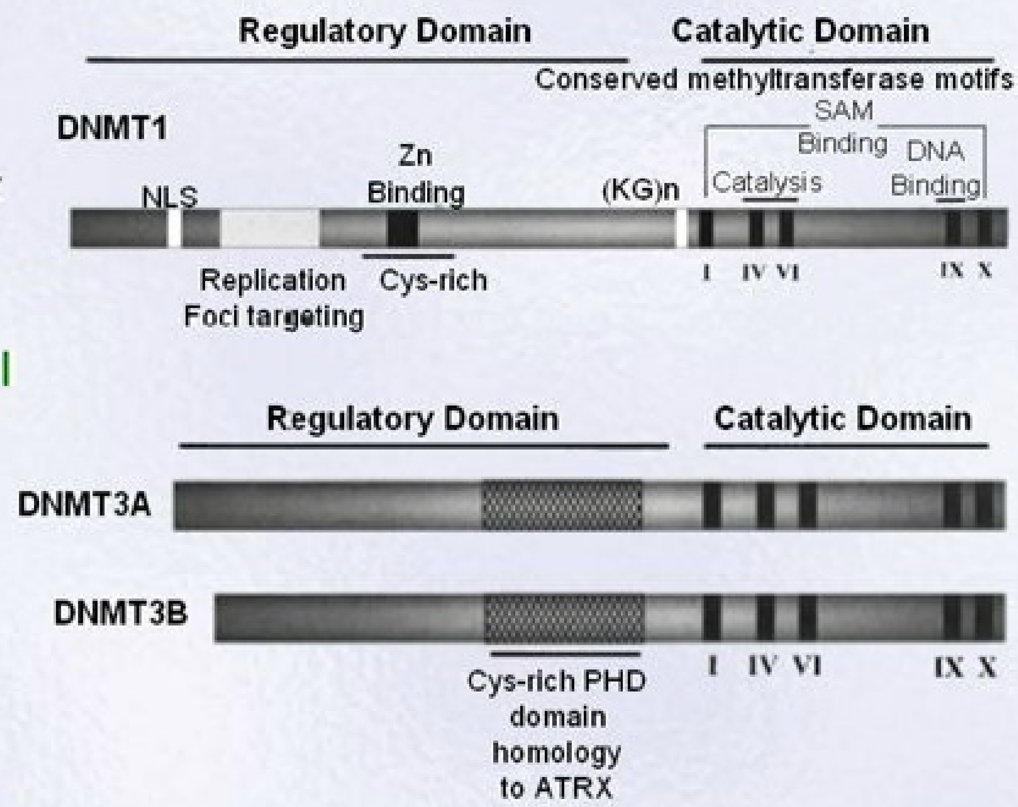
Dnmt1: embryonic lethal

Dnmt2: no obvious effect

Dnmt3a: perinatal death

Dnmt3b: embryonic lethal

Dnmt3l: no imprints

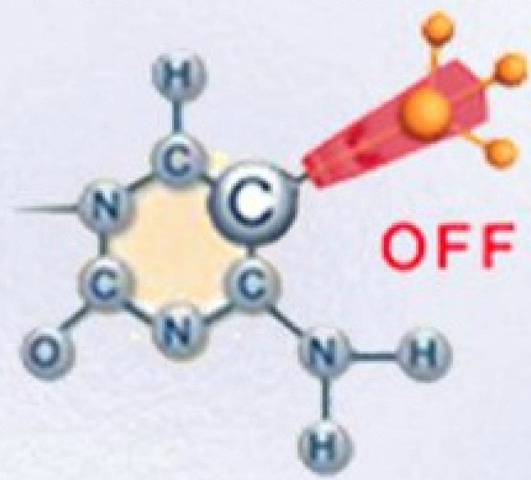


Robertson, KD, *Oncogene* 2002

Some DNA Methyl Transferases are Essential

Cytosine methylation in mammals

- Gene expression
- Chromosomal stability
- Cell differentiation
- Imprinting
- X-Inactivation
- Carcinogenesis
- Aging



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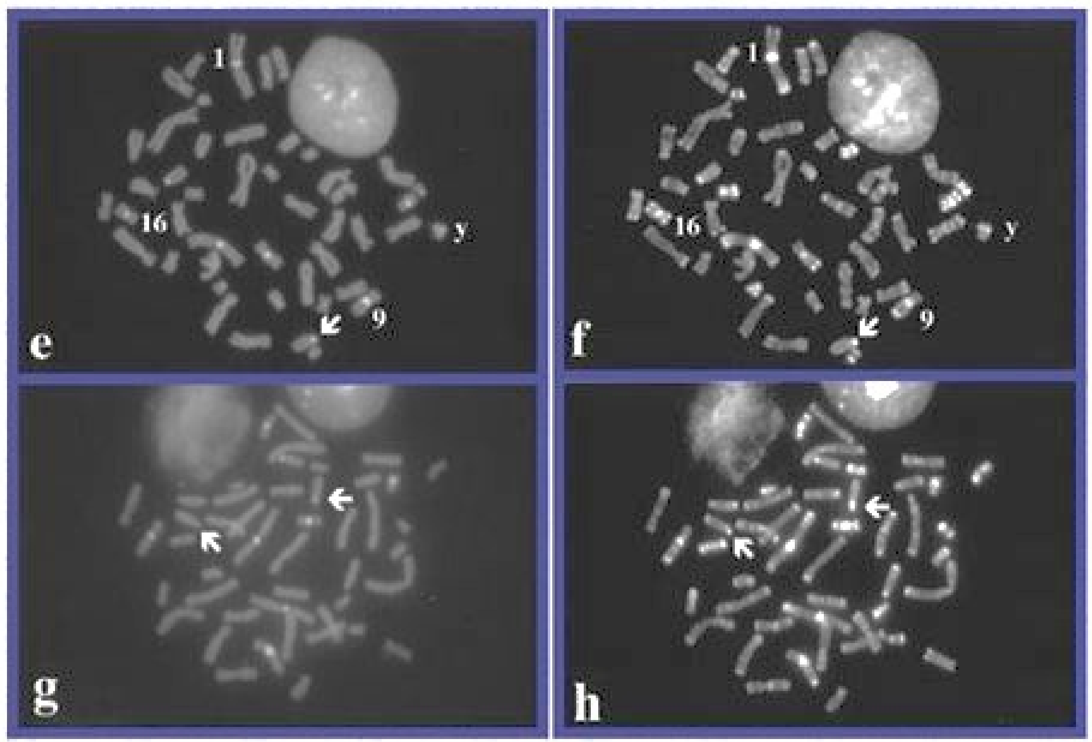
5-Methyl Cytosine is Found in Heterochromatic Regions

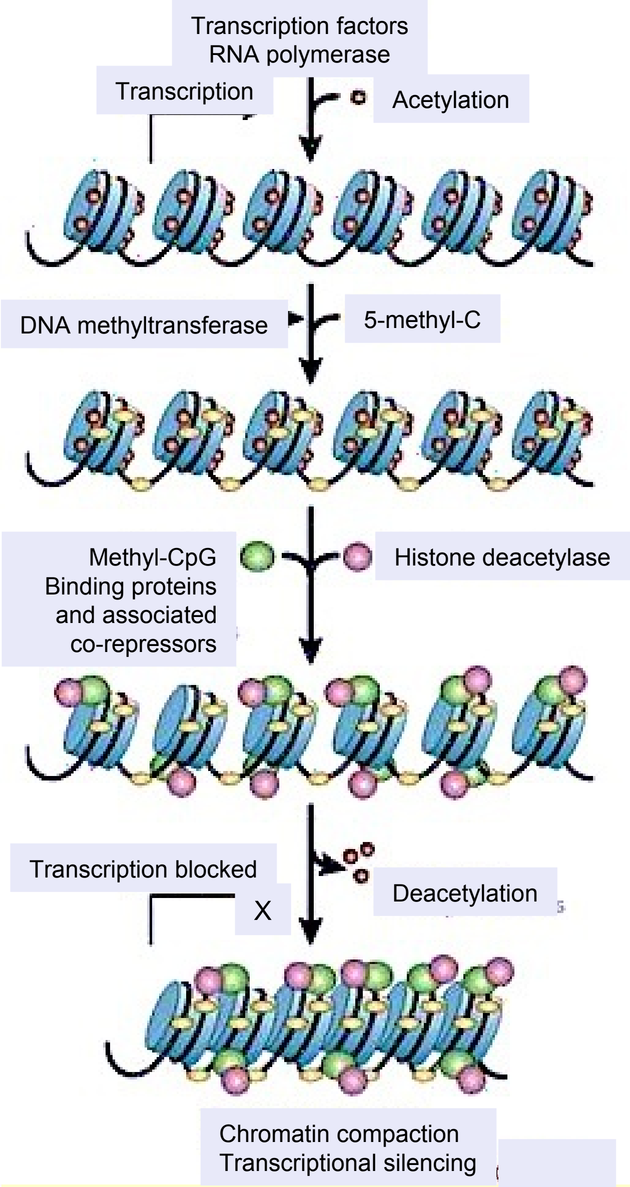
The distribution of cytosine methylation in mammals

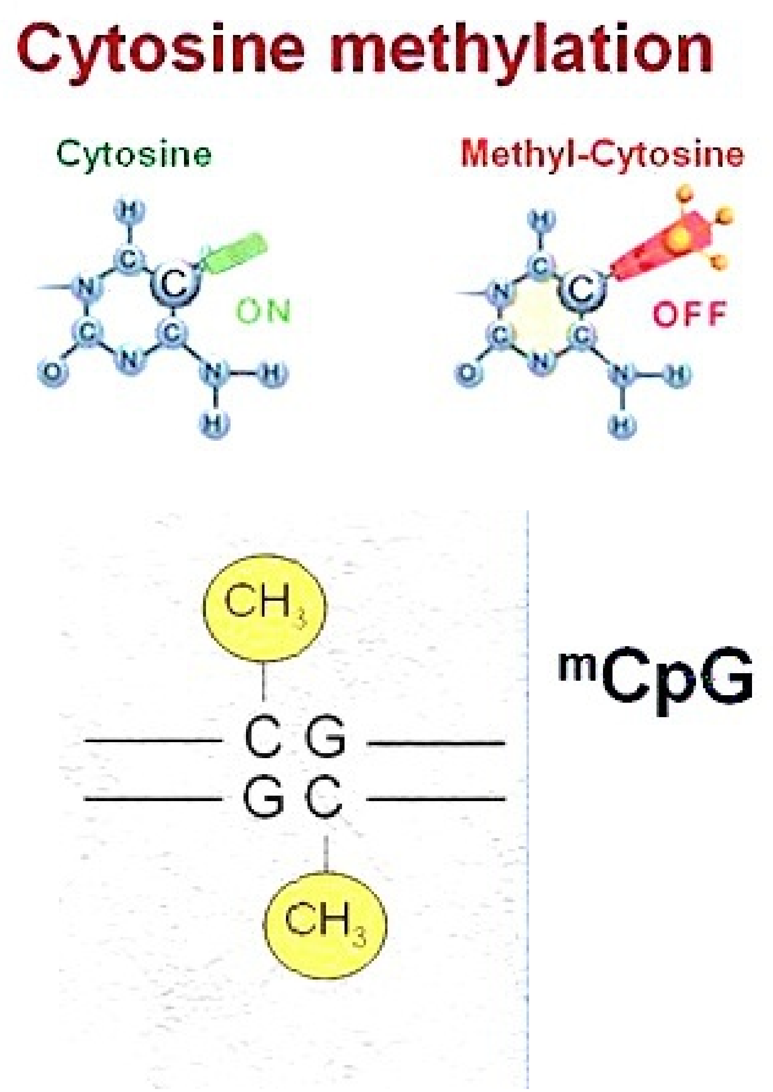
- Heterogeneity visible at cytogenetic scale
- Associated with heterochromatic regions



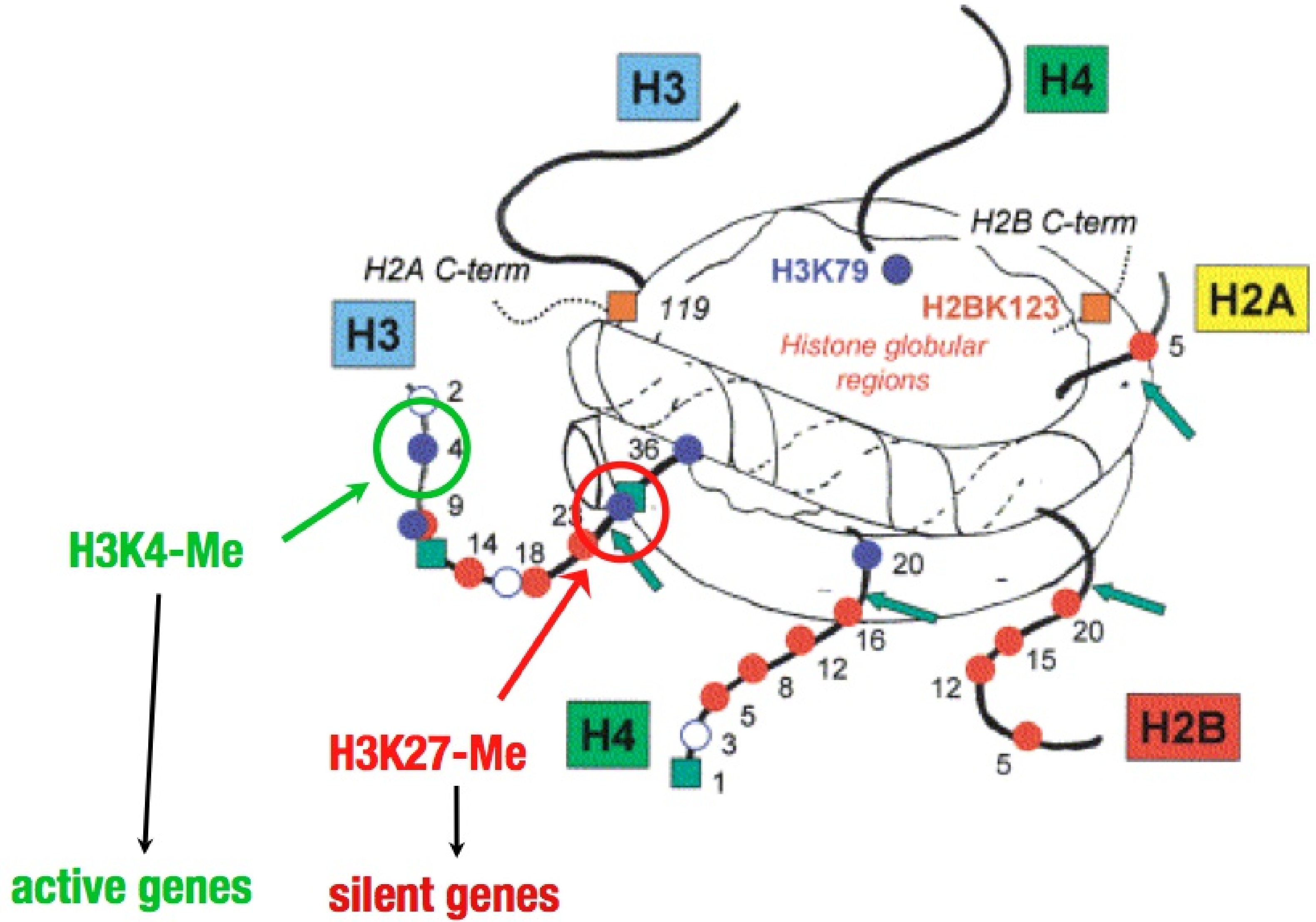
John Greally, Henry Stewart Talks

Cytosine Methylation Maintains Inactive-Condensed Chromatin State

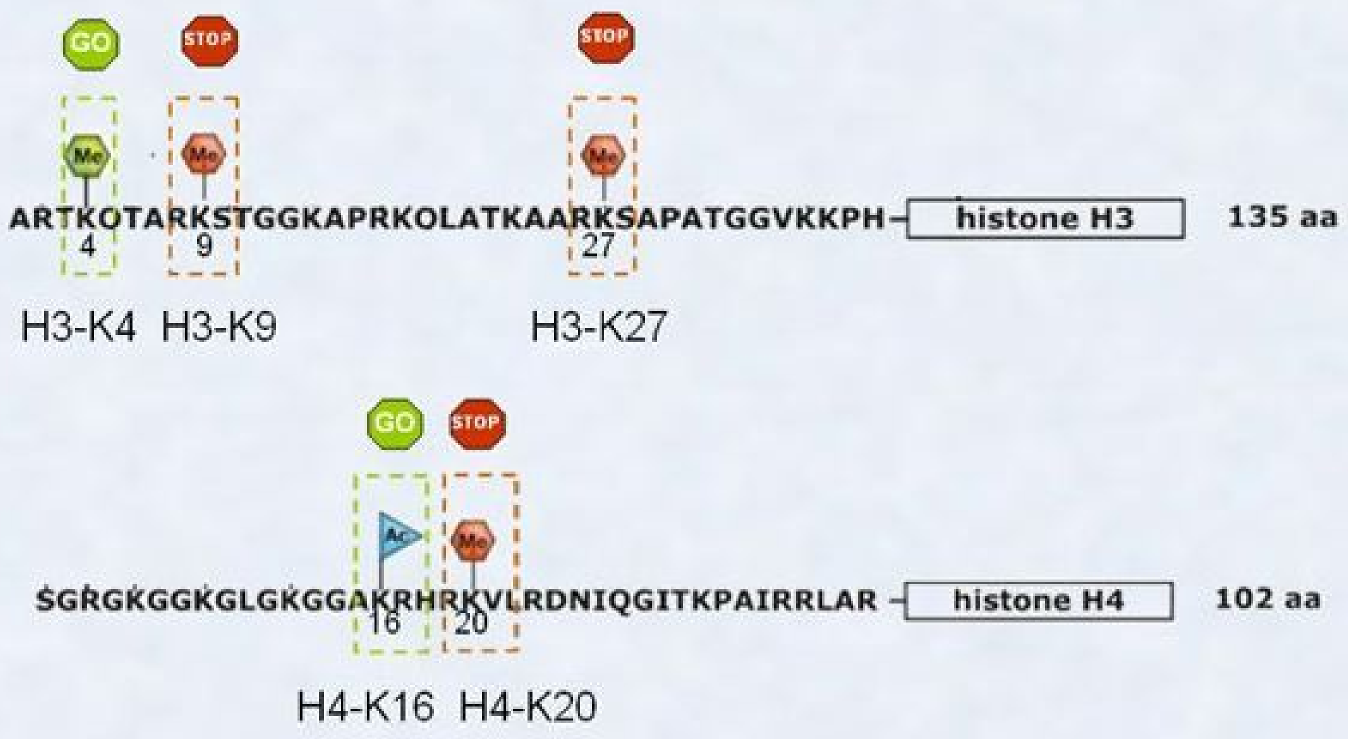




Histone Modifications in Active and Silent Chromatin



Histone Code

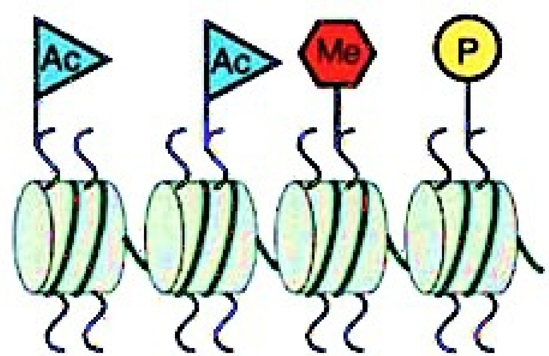


Paula Vertino, Henry Stewart Talks

Structure & Epigenetics of Euchromatin versus Heterochromatin

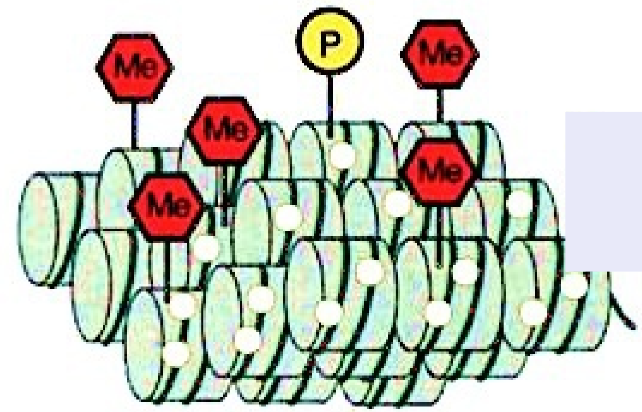
DNA methylation and histone modifications help to compartmentalize the genome into domains of different transcriptional potentials

Euchromatin



- High histone acetylation
- Low DNA methylation
- H3-K4 methylation

Heterochromatin



- Low histone acetylation
- Dense DNA methylation
- H3-K9 methylation

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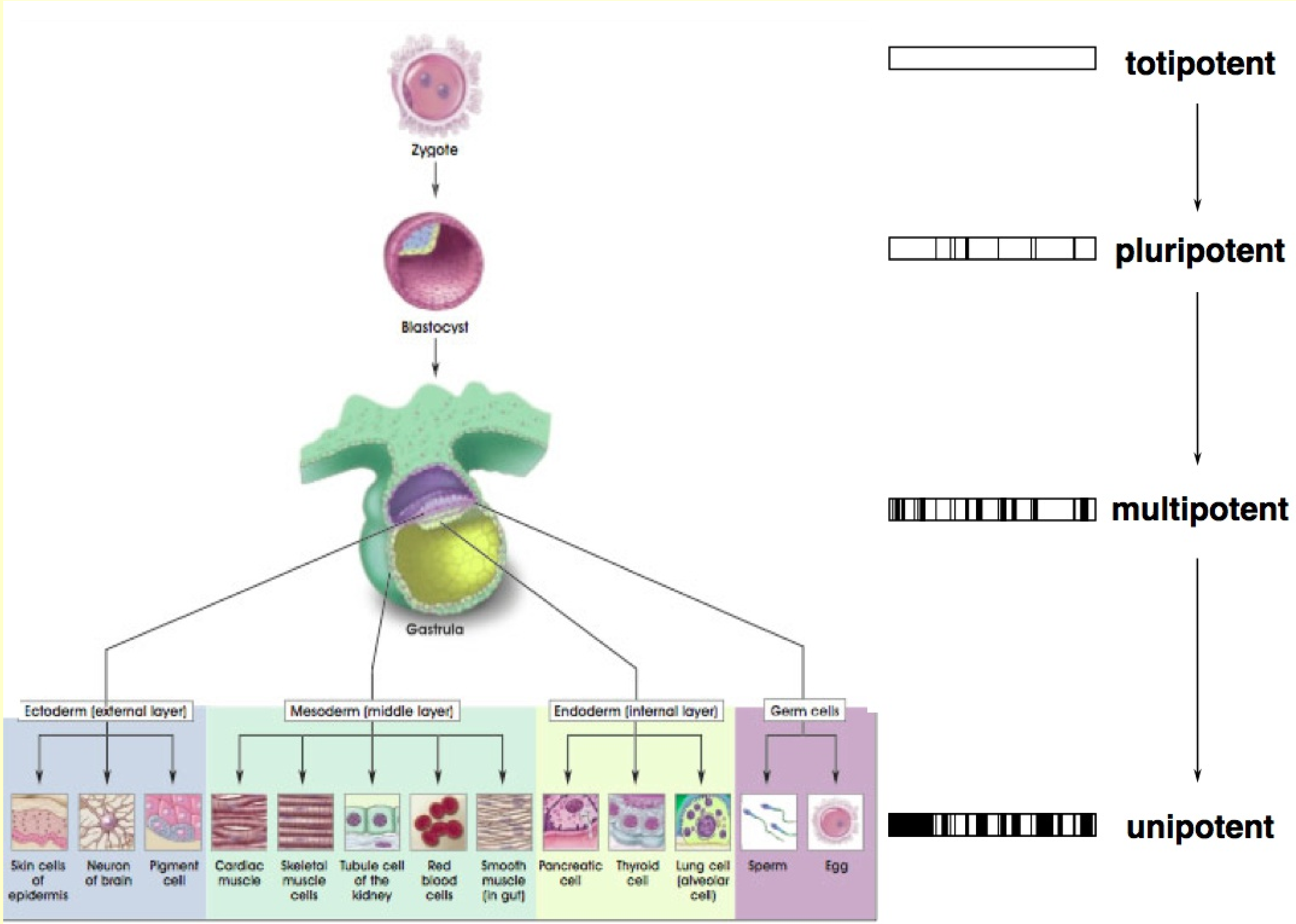
Methylated DNA from Zygote to Adult



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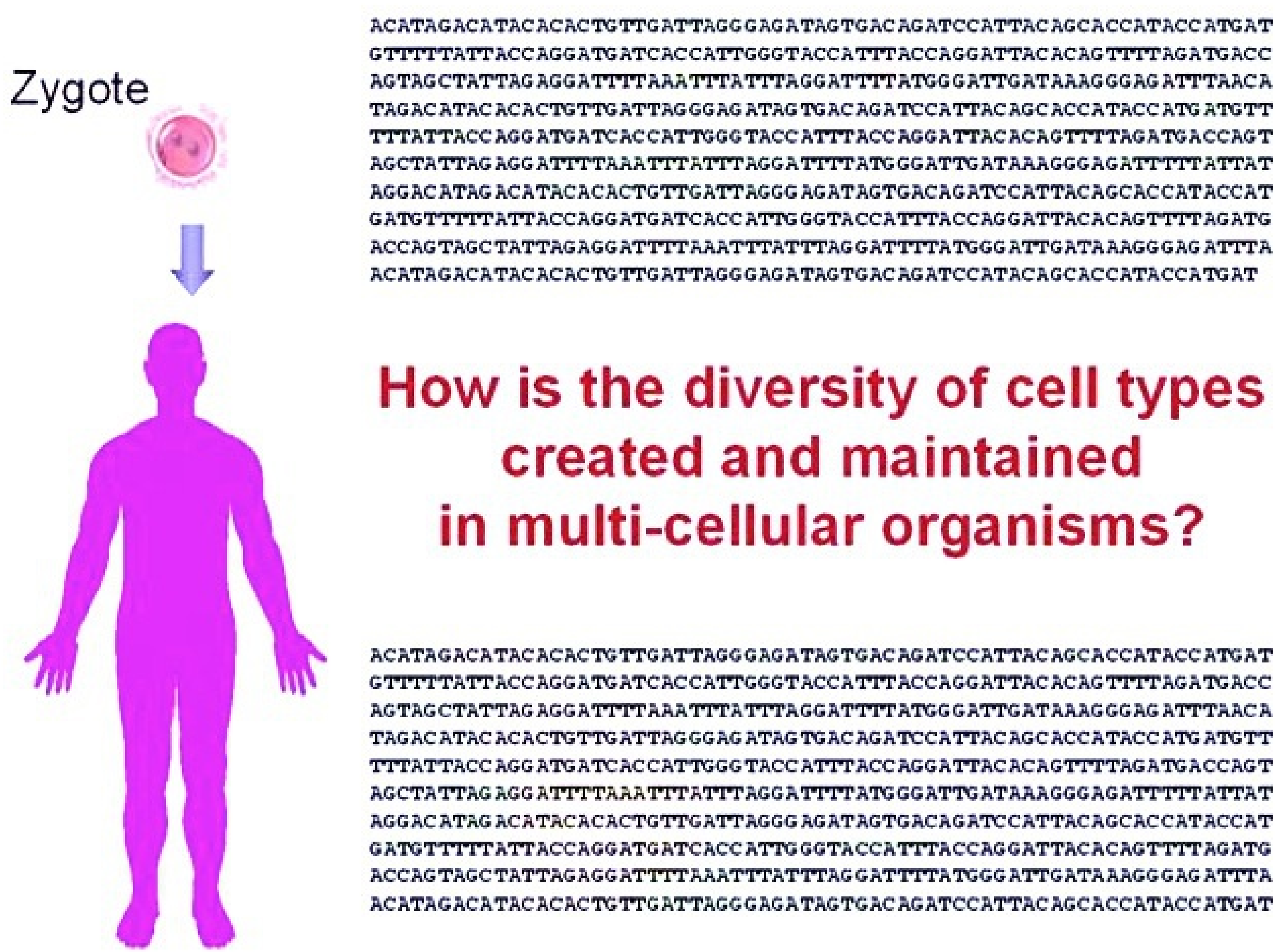

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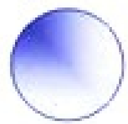
DNA Methylation Differentiates Totipotent Embryonic Stem Cells from Unipotent Adult Stem Cells

Pluripotent cell



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Unipotent cell

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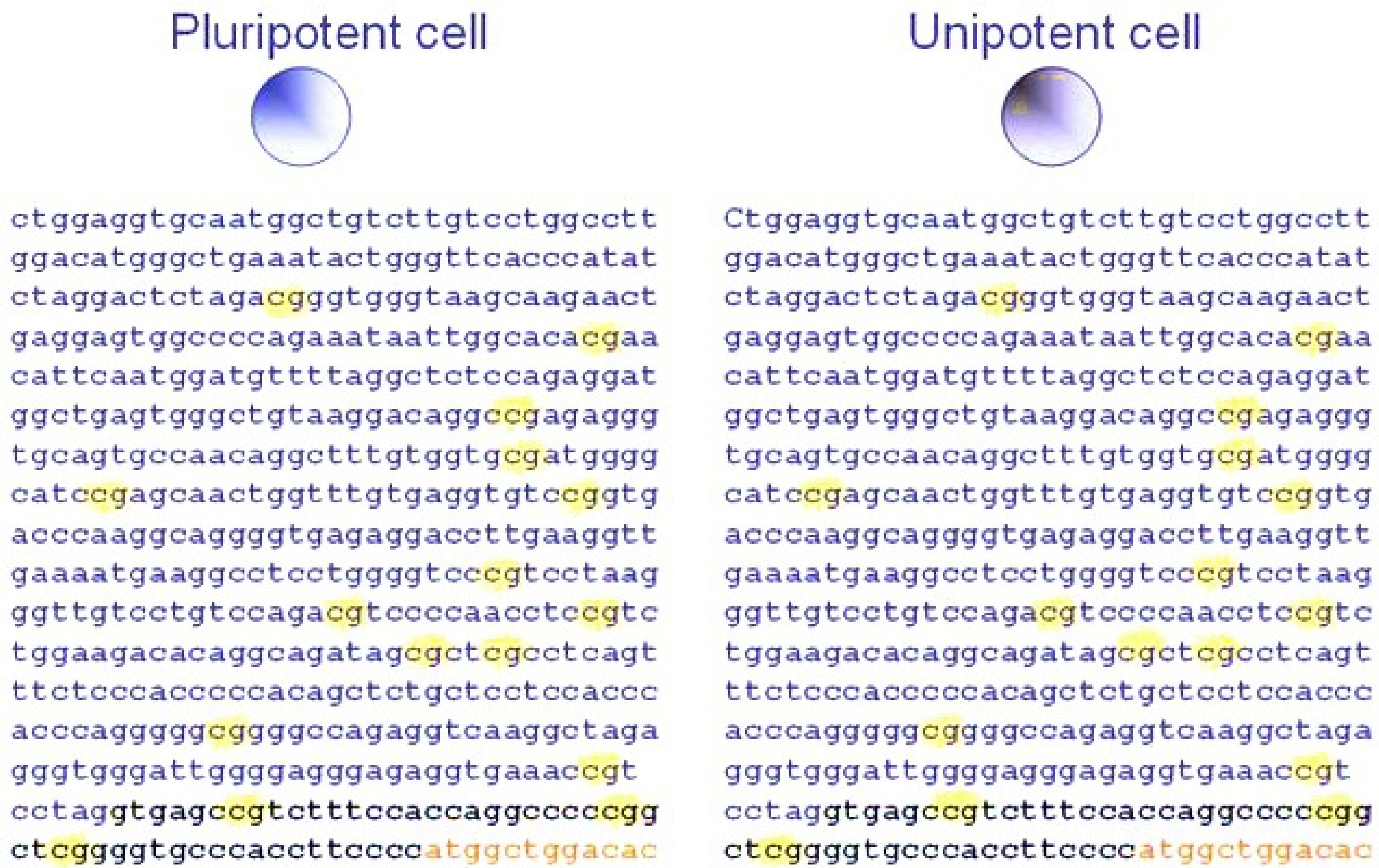
Alex Meissner, Henry Stewart Talks

DNA Methylation Differentiates Totipotent Embryonic Stem Cells from Unipotent Adult Stem Cells



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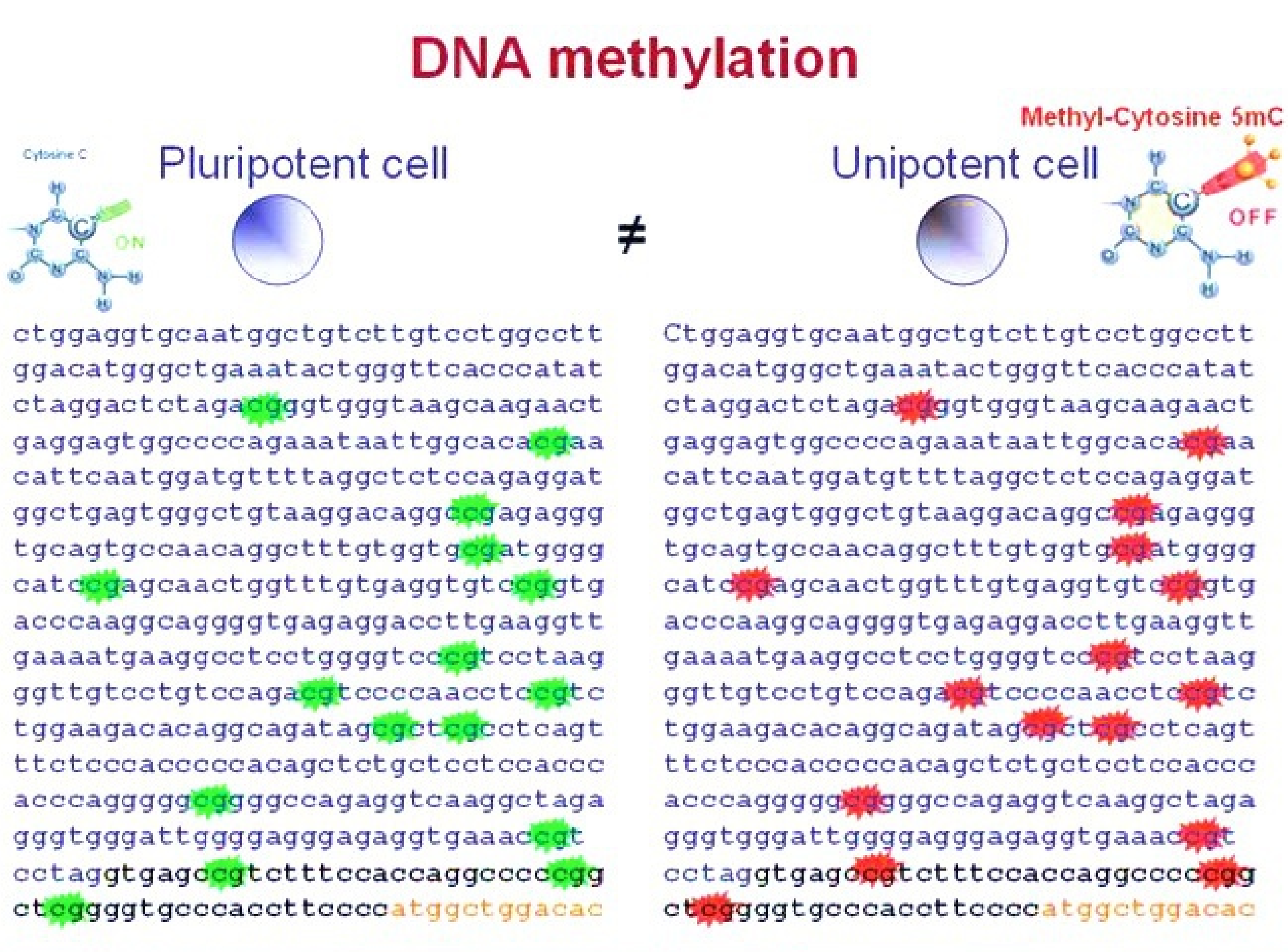

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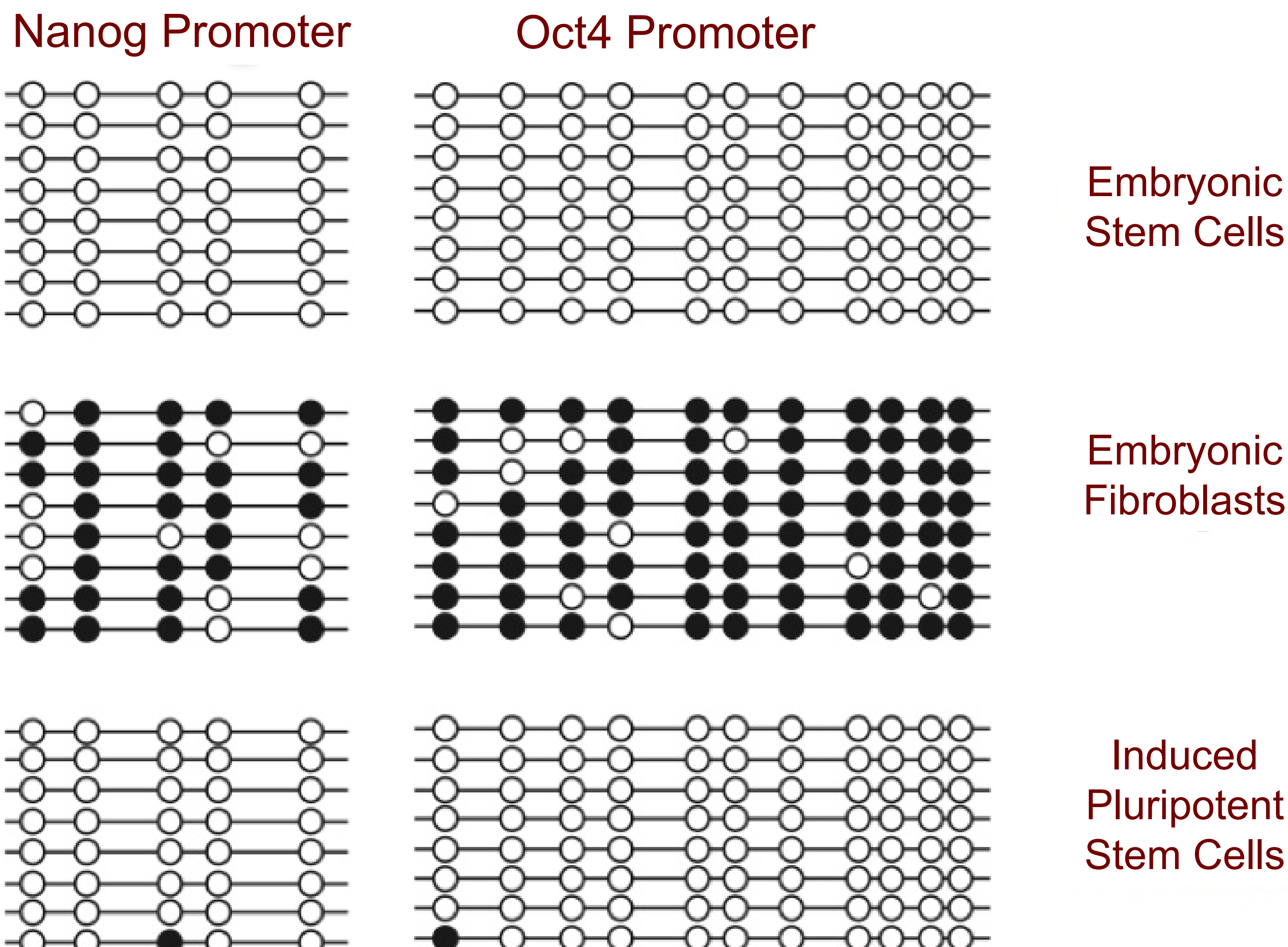
Nanog and Oct4 Promoter Methylation in Embryonic and Induced Stem Cells



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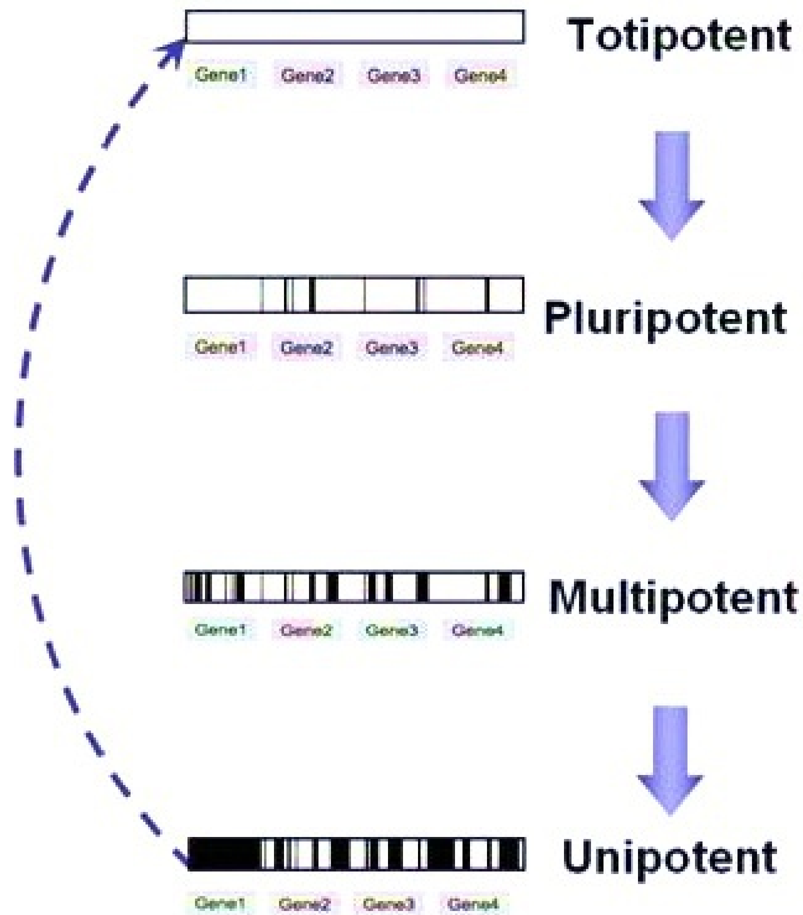
Differentiated Cells can Become Totipotent Again

Nuclear transplantation demonstrates nuclear equivalence

Briggs and King, 1952
Gurdon, 1960s

"Dolly"

Differentiated cells maintain the potential to generate an entire organism



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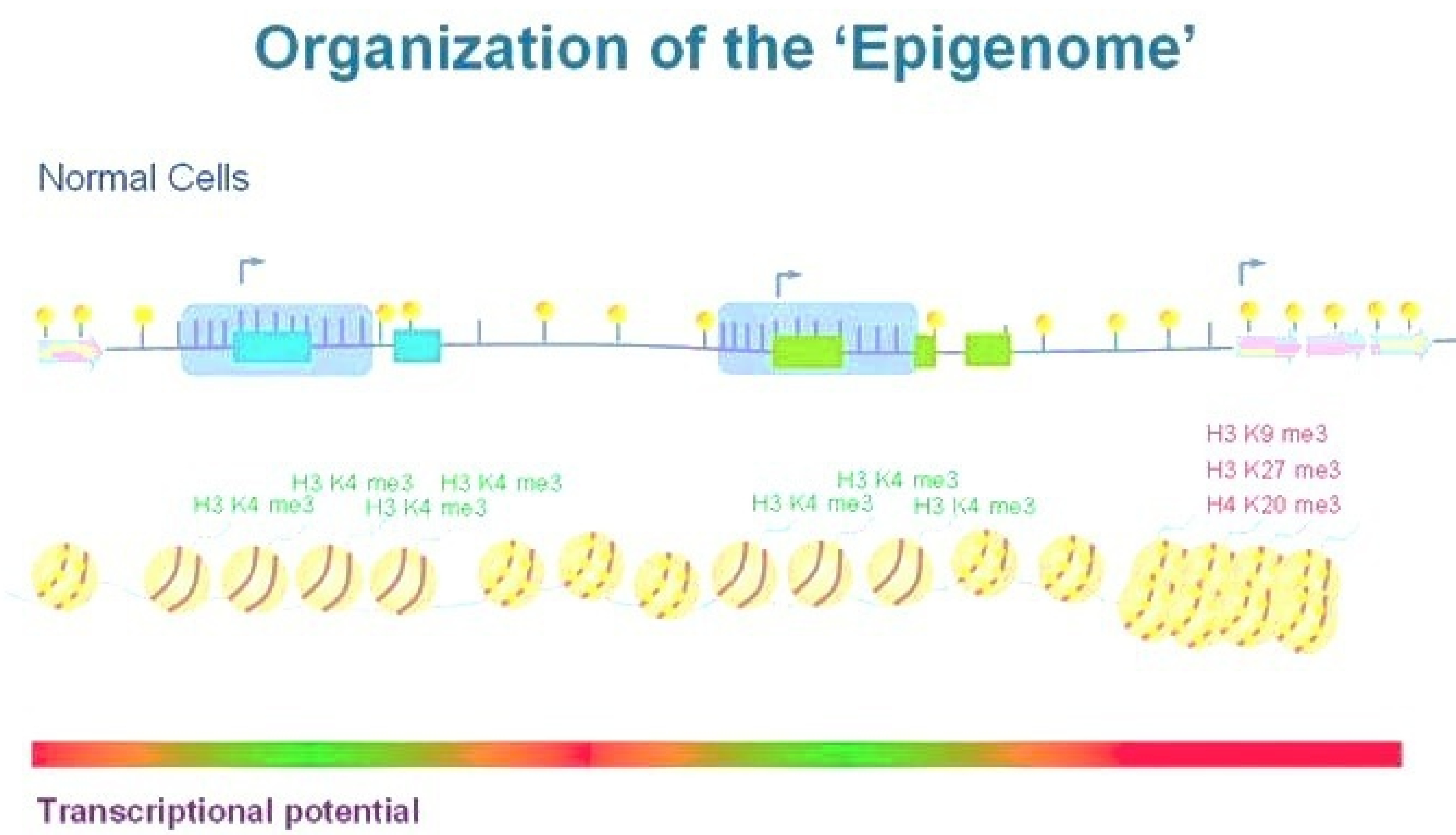
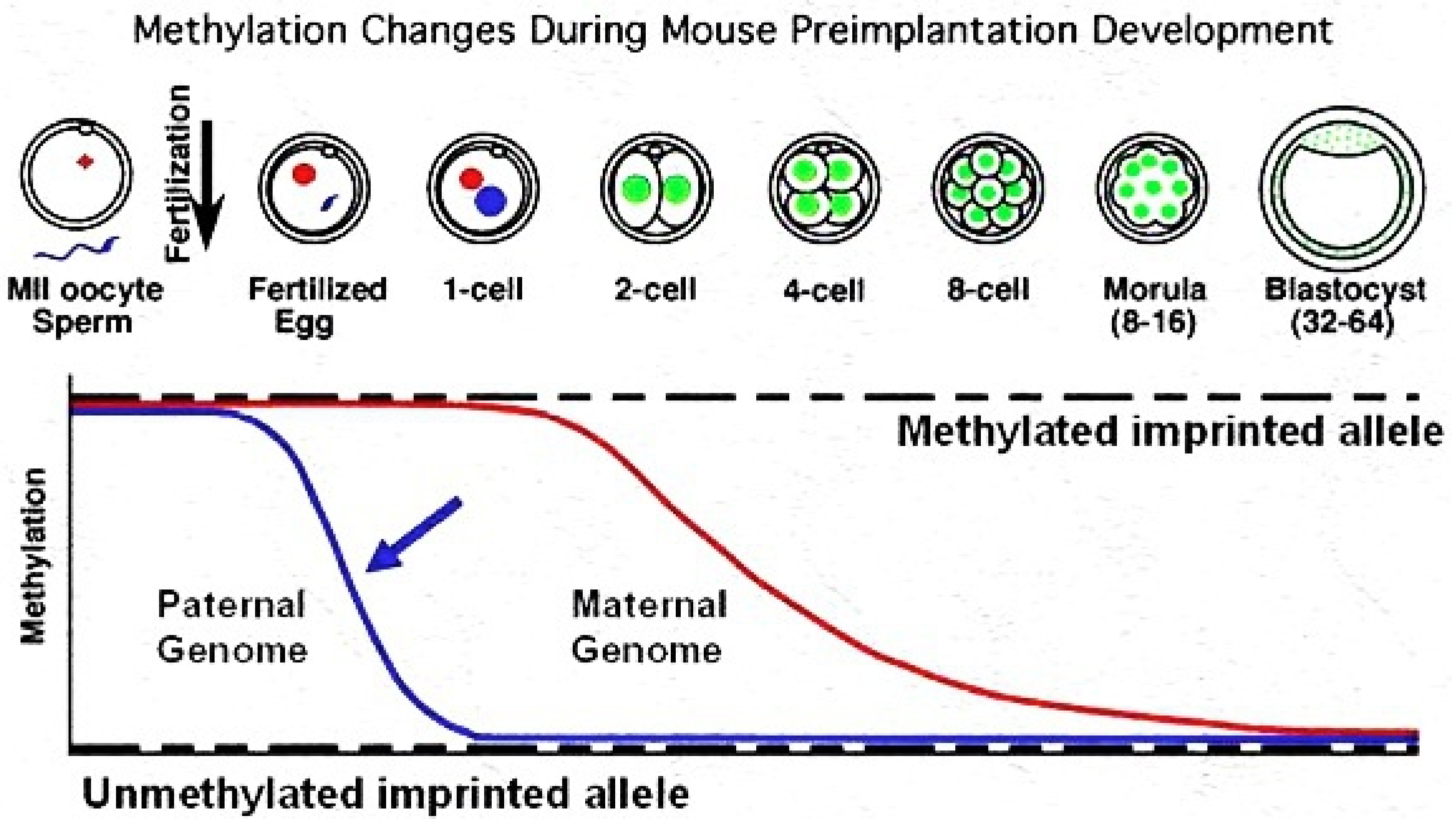
Organization of the Epigenome



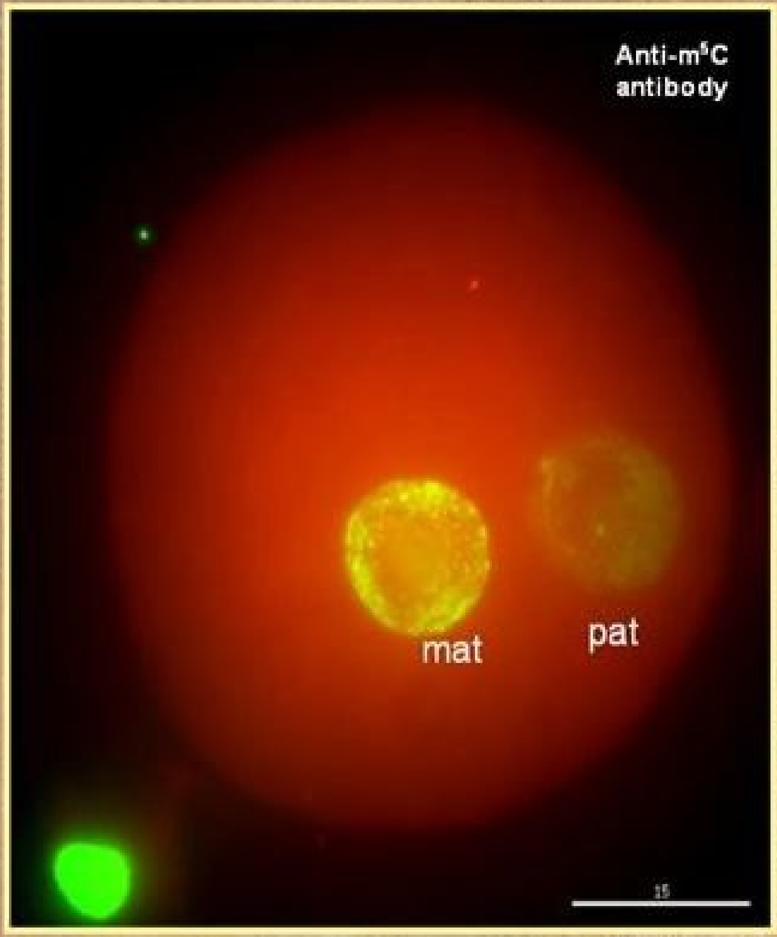
Methylation Changes During Development

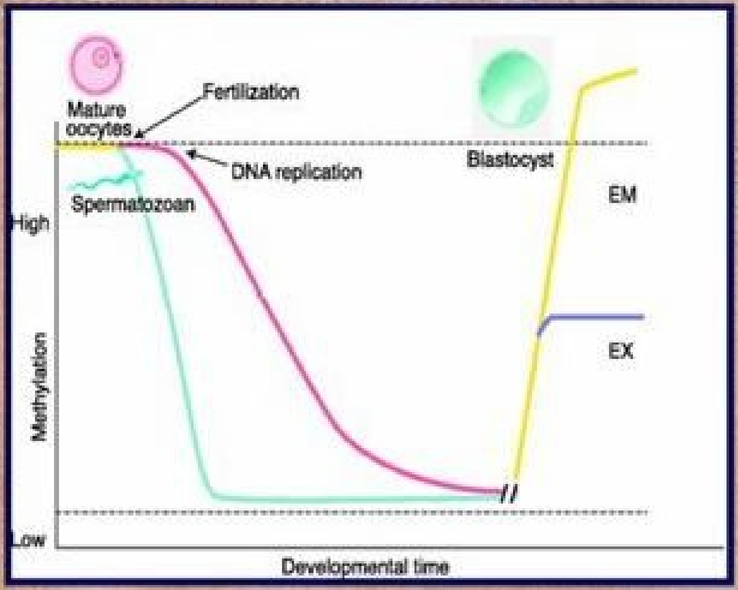


Marisa Bartolomei, Henry Stewart Talks

Demethylation of the Paternal Genome

De-methylation of the paternal pronucleus in the one-cell embryo of mouse





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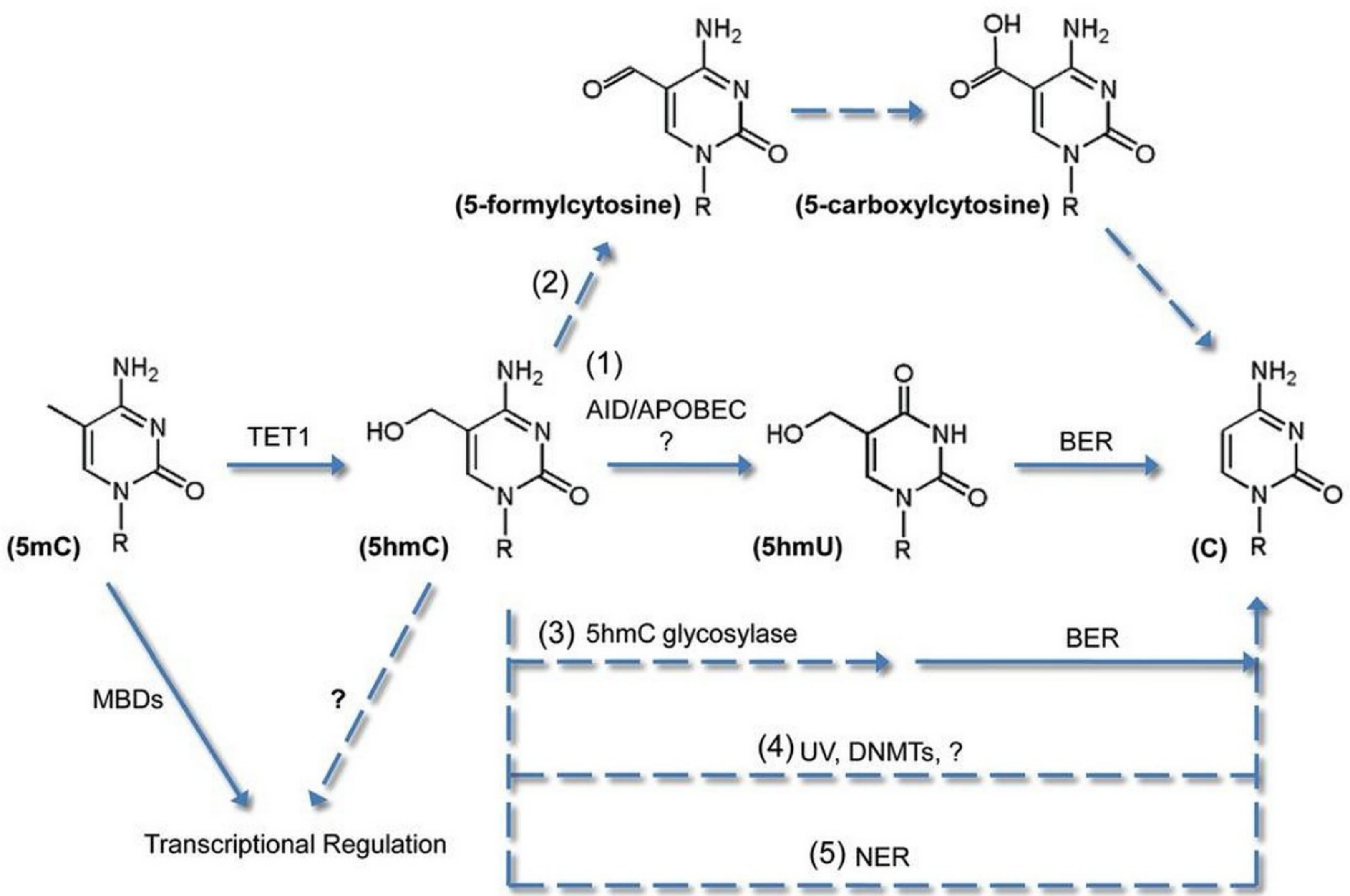
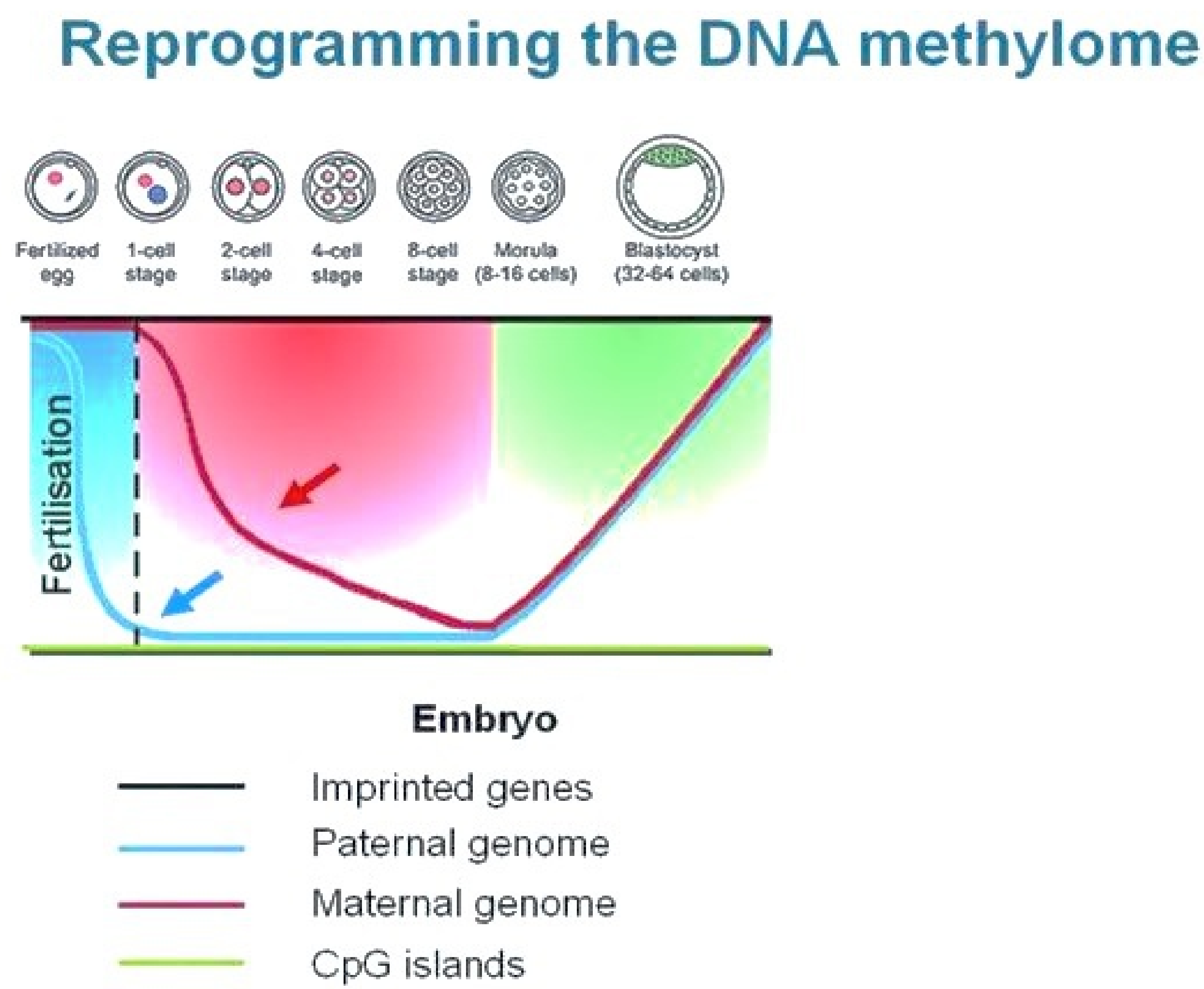
Tet Proteins Modify 5-Methyl-Cytosine Leading to Removal by DNA Repair



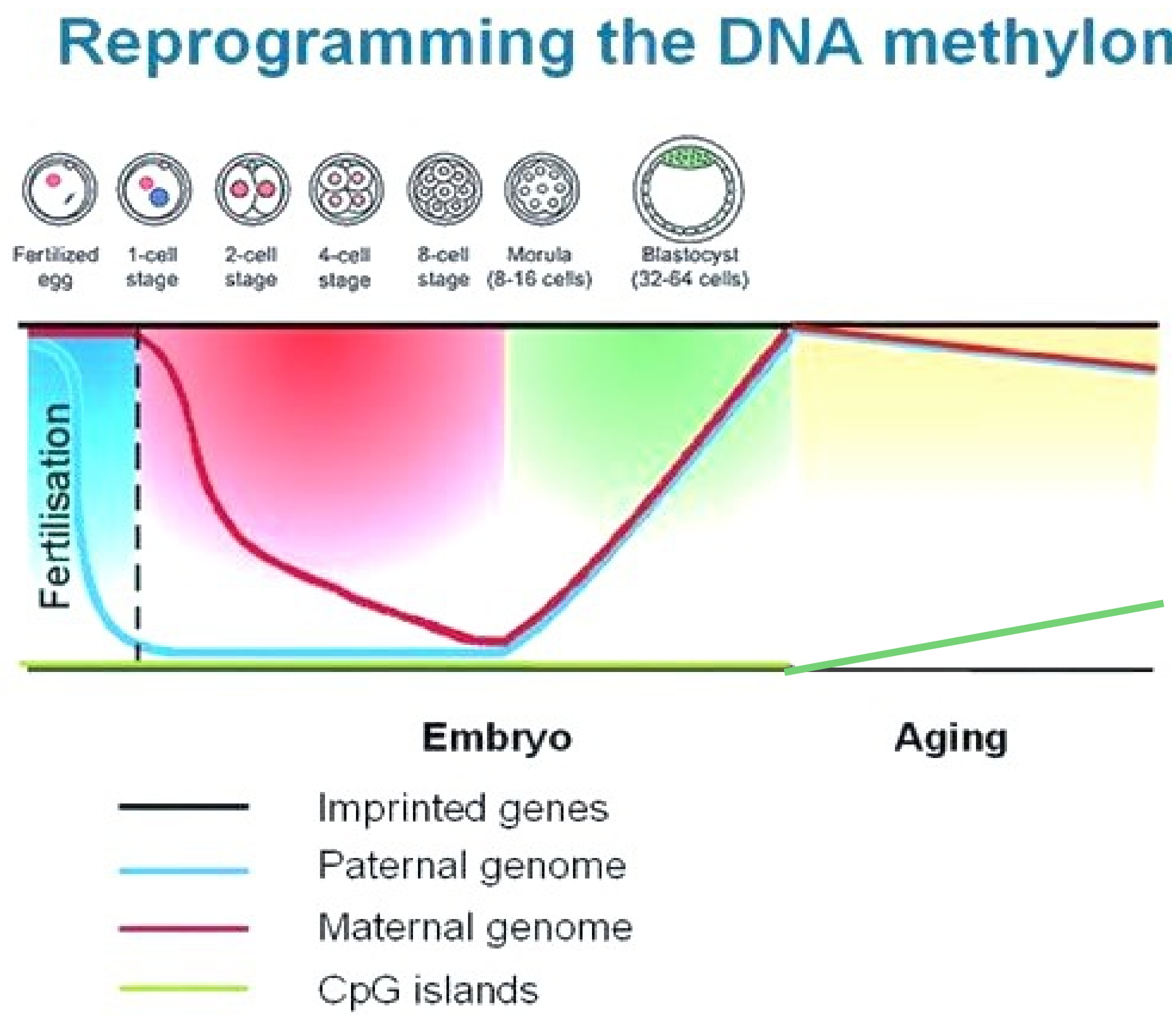
Guo et al, Cell Cycle. 2011 August 15; 10(16): 2662–2668.

Methylation Changes During Development



Paula Vertino, Henry Stewart Talks

Methylation Changes During Development



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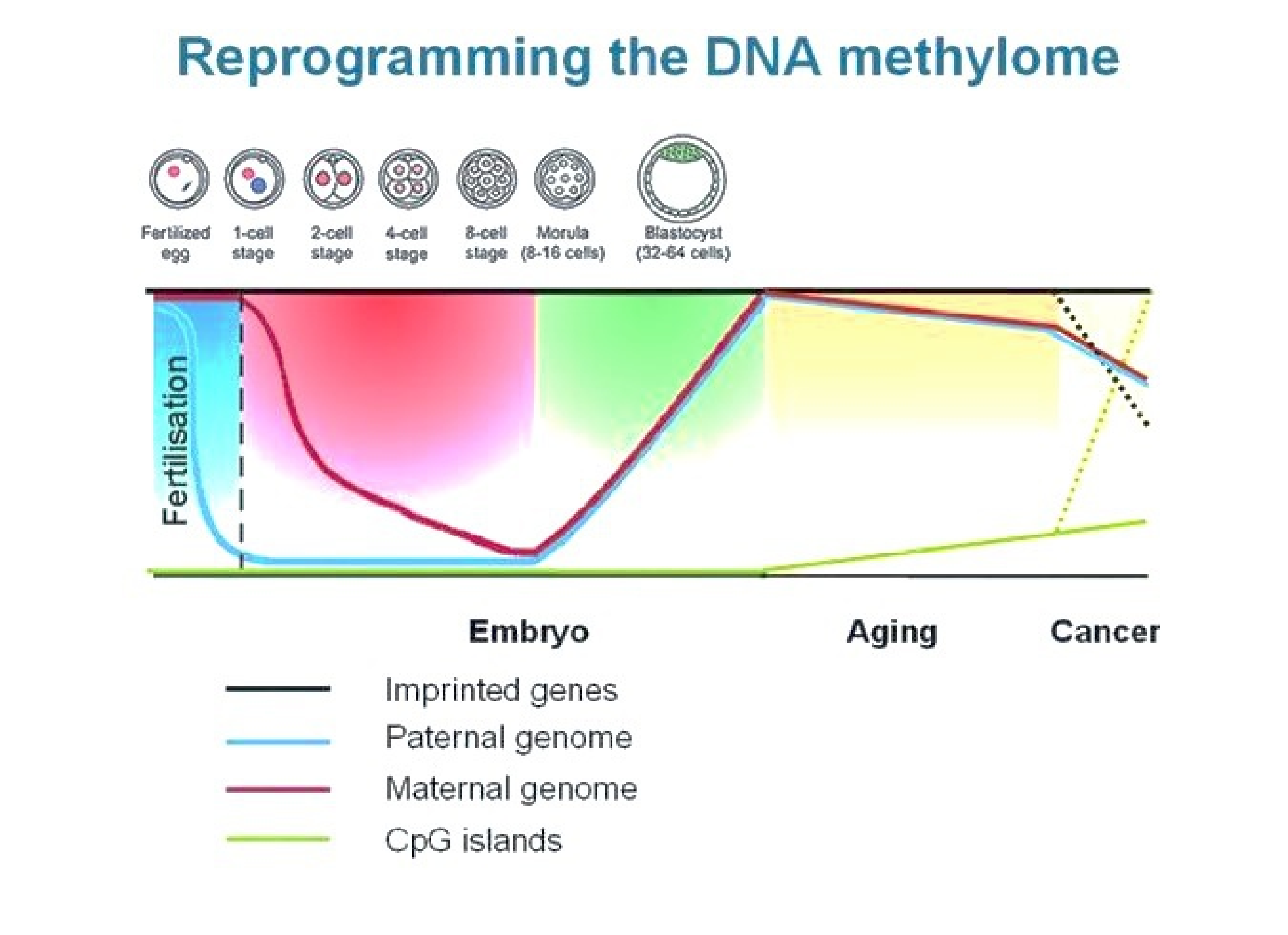
Methylation Changes During Development



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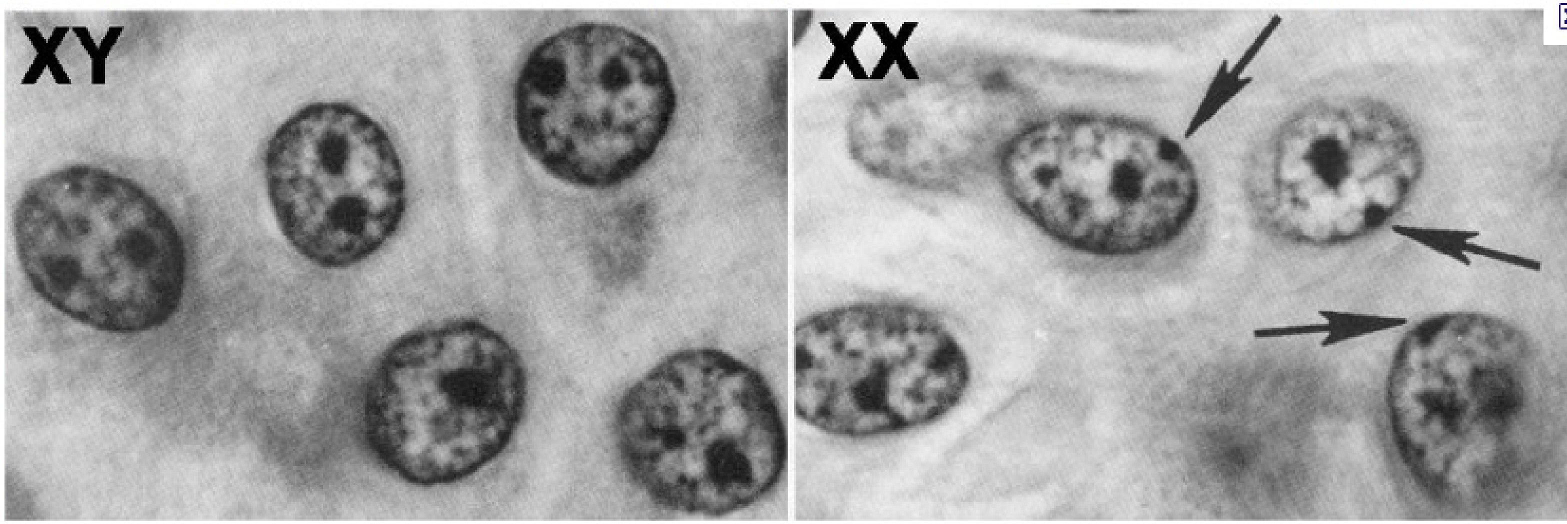
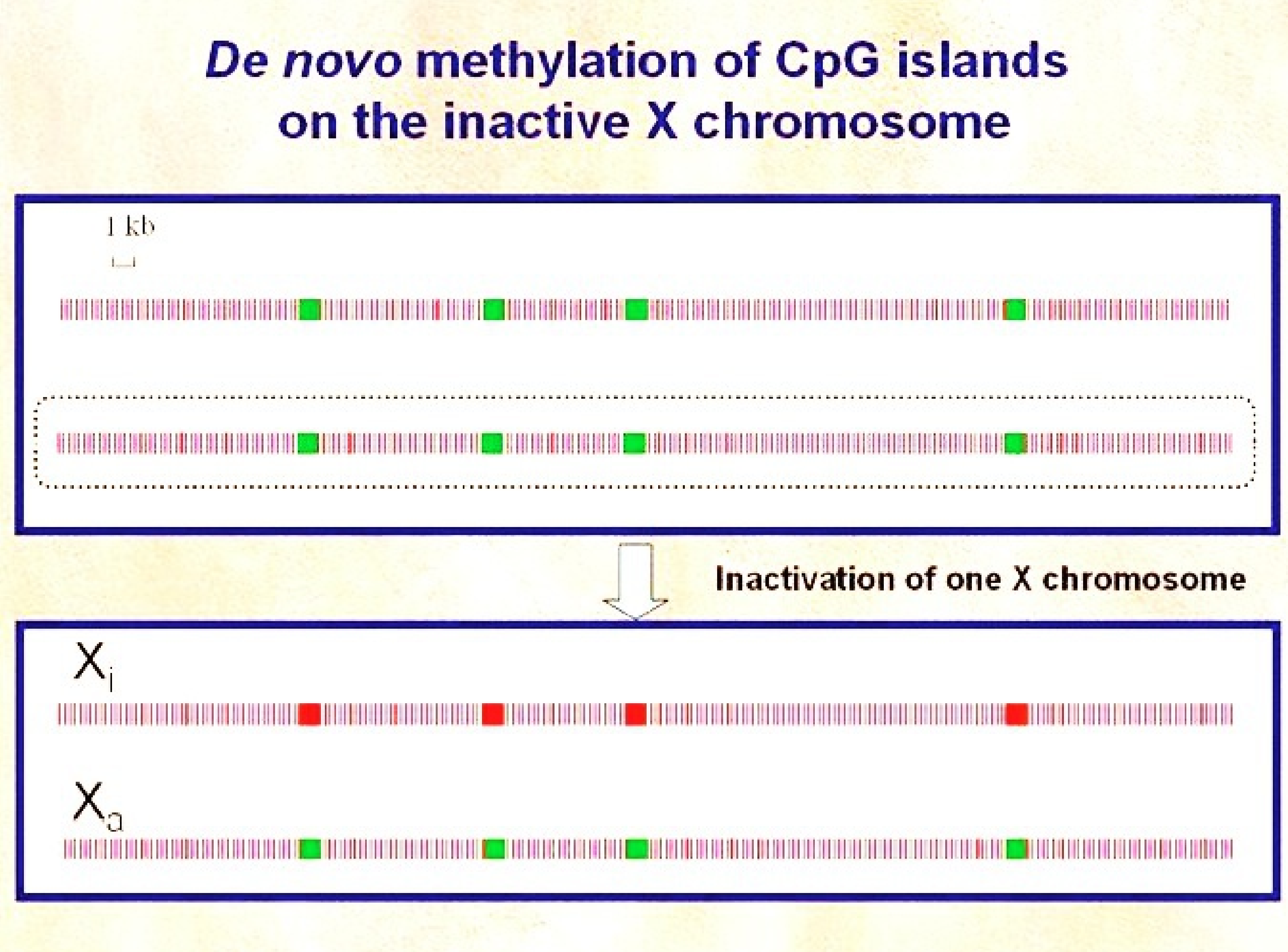
X Chromosome Inactivation: Barr Bodies



Barr, M. L., Bertram, E. G., (1949), A Morphological Distinction between Neurones of the Male and Female, and the Behaviour of the Nucleolar Satellite. *Nature.* ***163*** *(4148): 676-7.*

Lyon, M. F., (2003), The Lyon and the LINE hypothesis. j.semcdb 14, 313-318. (Abstract)

X Chromosome Inactivation: CG Island Methylation



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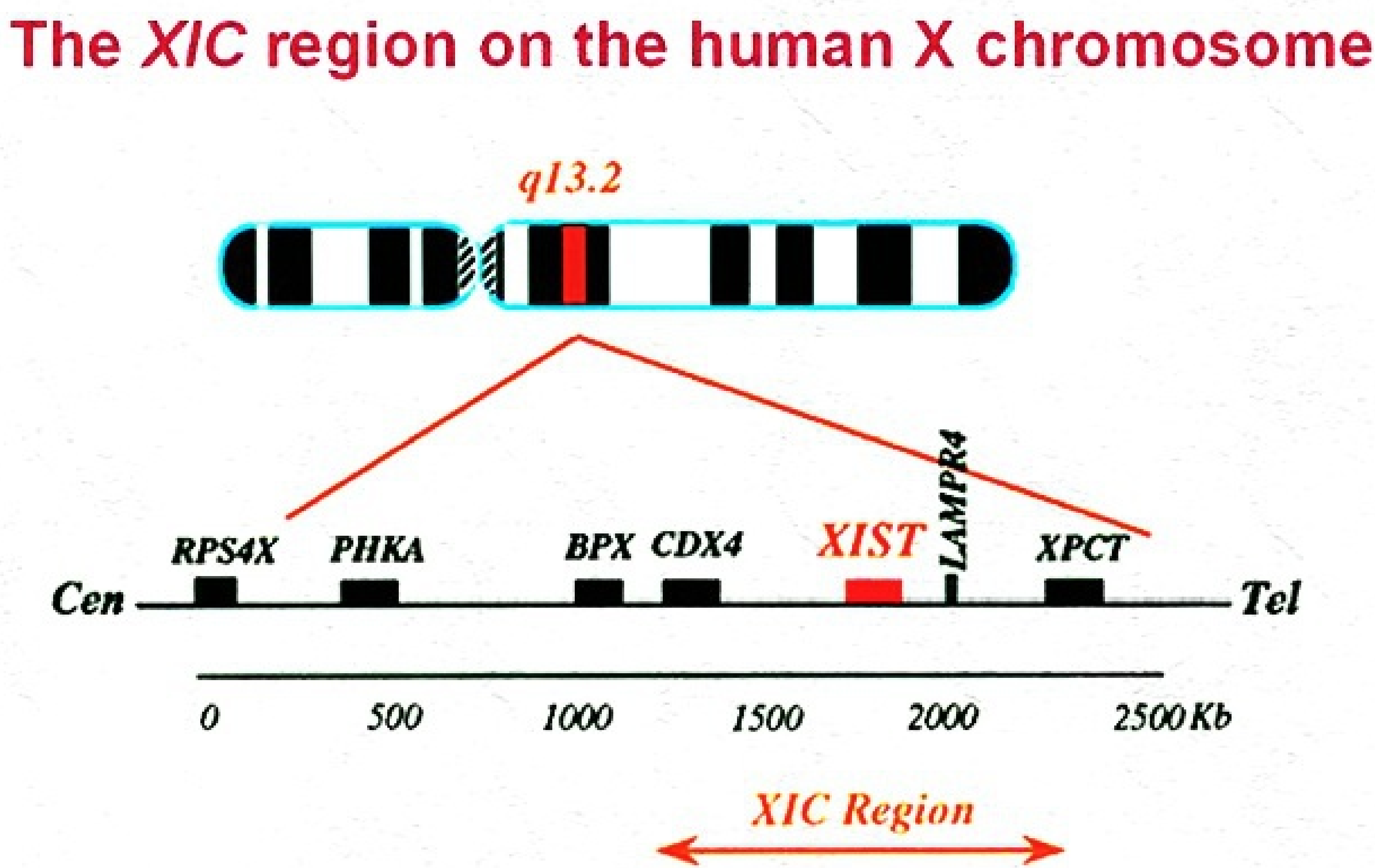
XIC Region



Barbara Migeon, Henry Stewart Talks

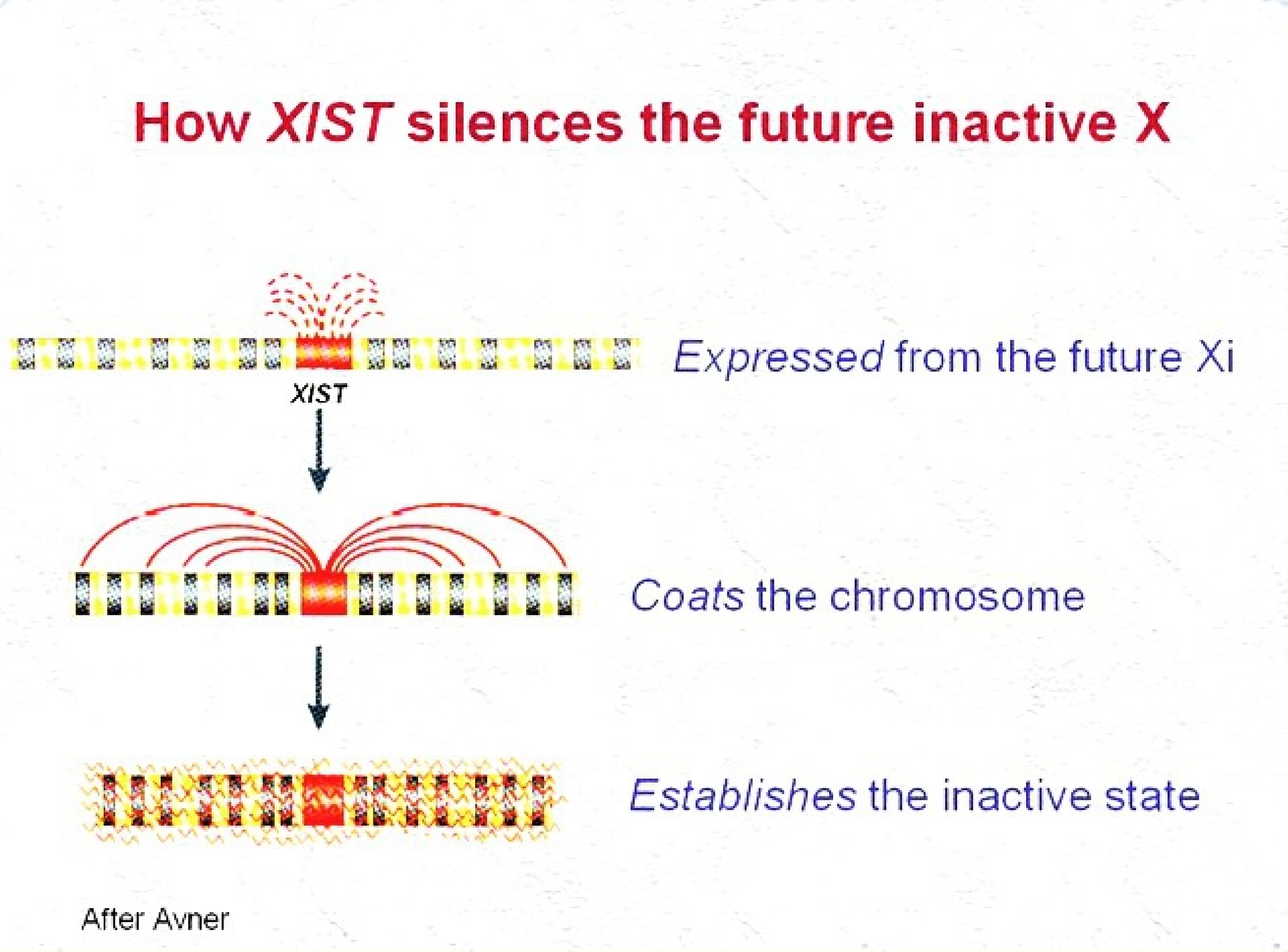
Characteristics of XIST Gene

Characteristics of *XIST*

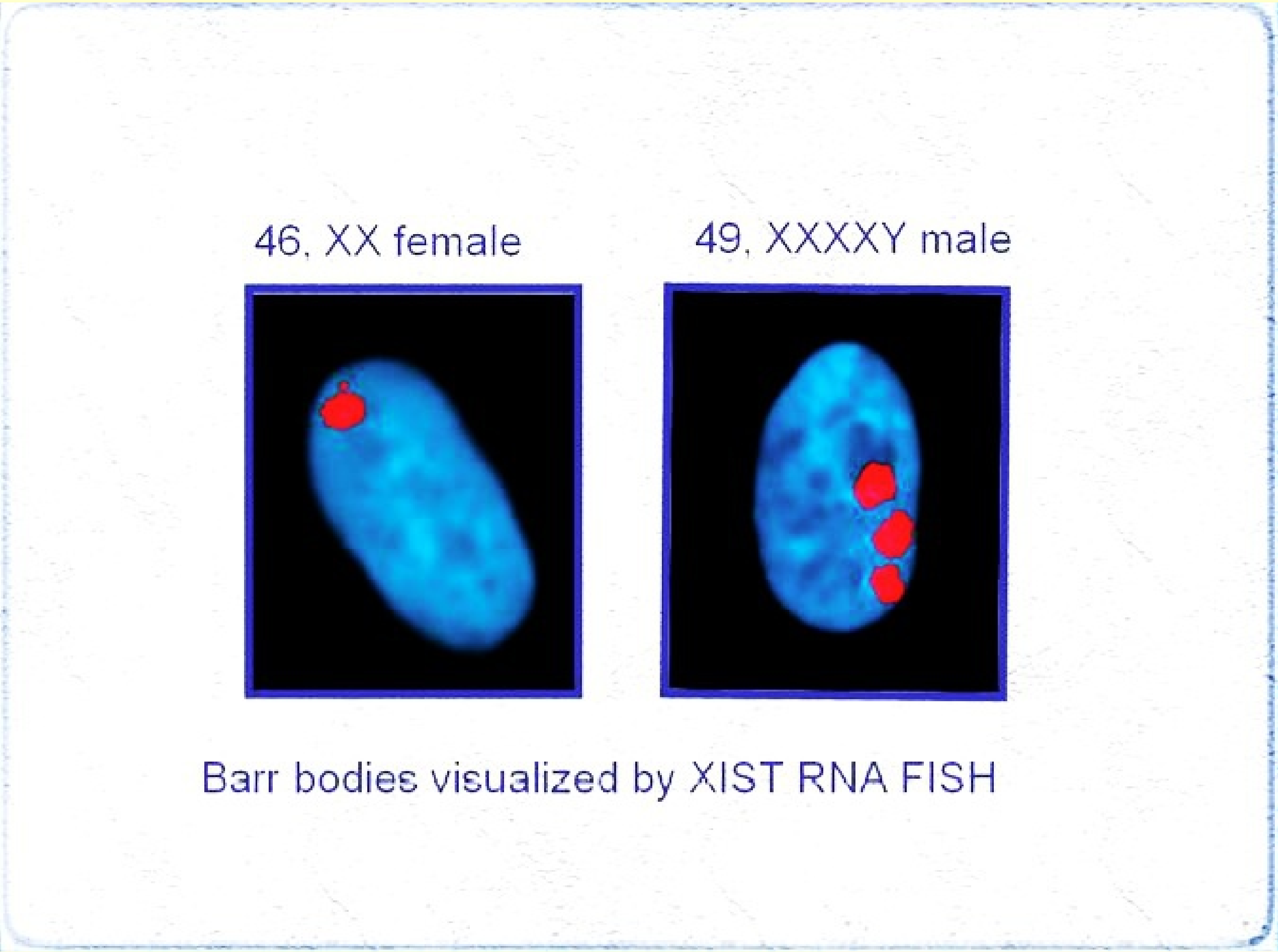
- Located in the XIC
- Transcribed *only* from the *inactive* X
- 20kb cDNA with no ORF, remains intranuclear, surrounding the *Barr body*
- *The* XIC gene responsible for Cis inactivation
- If transcribed (at critical time) it invariably inactivates its X by modifying chromatin

Barbara Migeon, Henry Stewart Talks

Xist Works in Cis

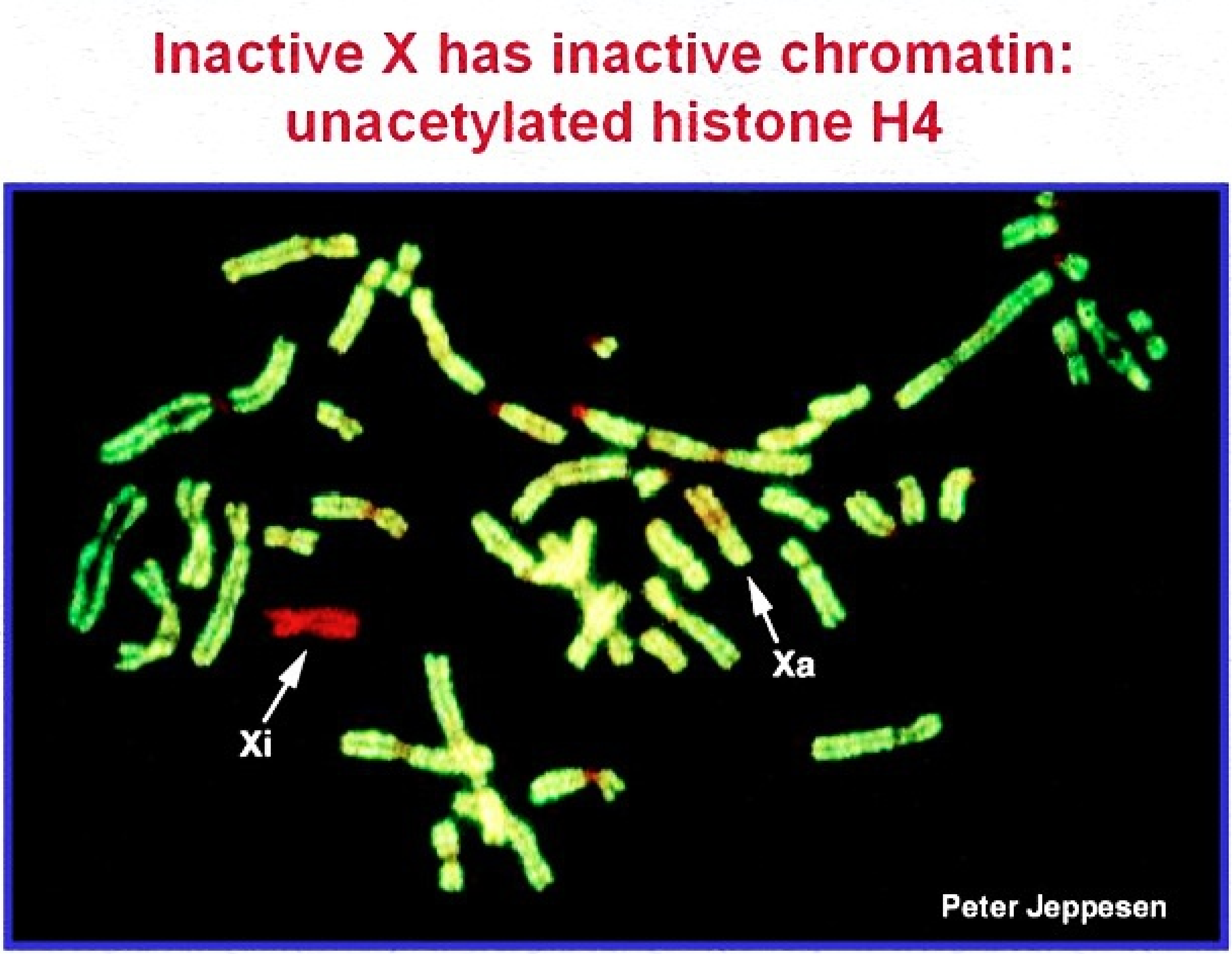


Only one X is active



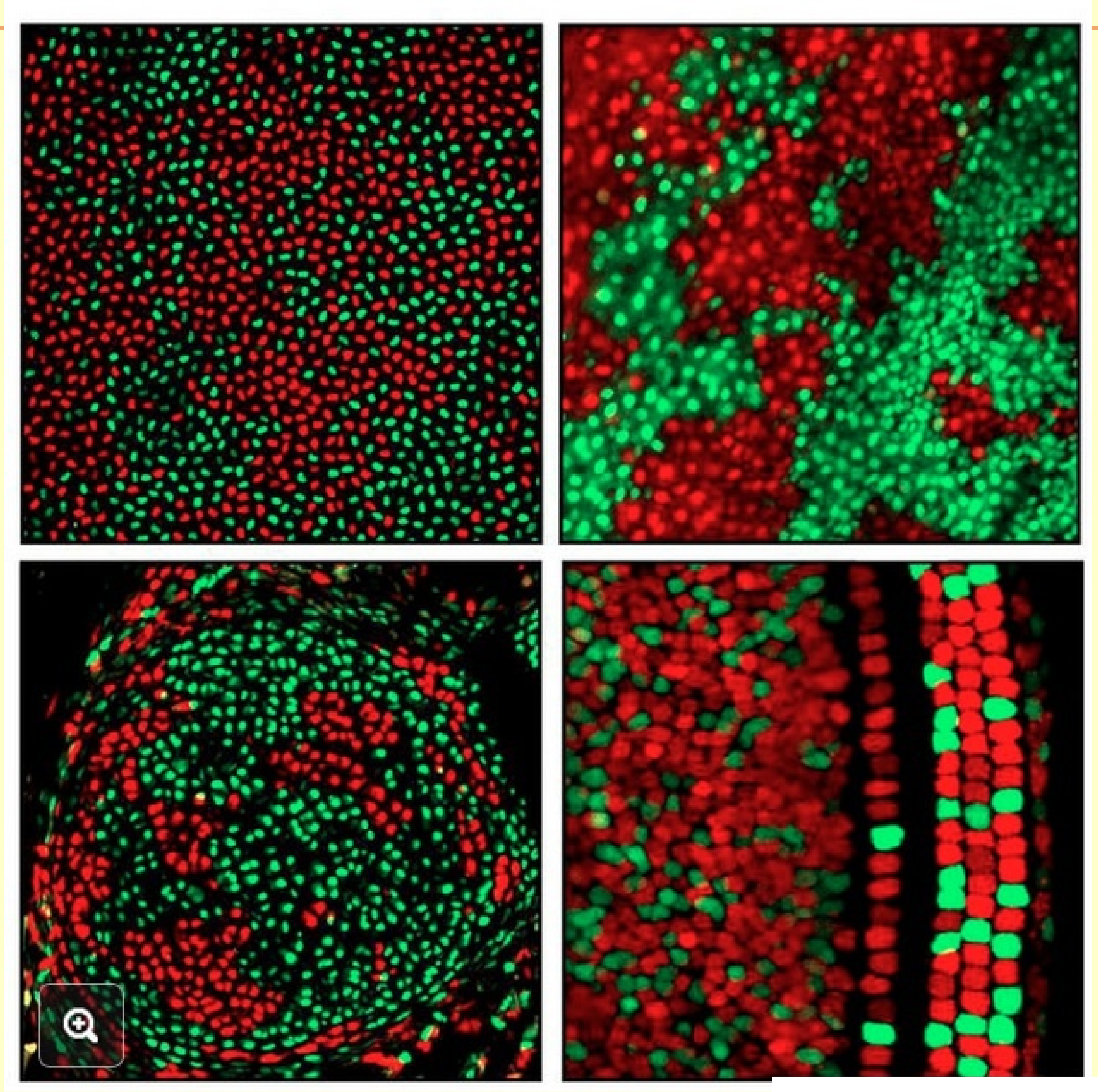
Barbara Migeon, Henry Stewart Talks

Inactive X has unacetylated histone H4



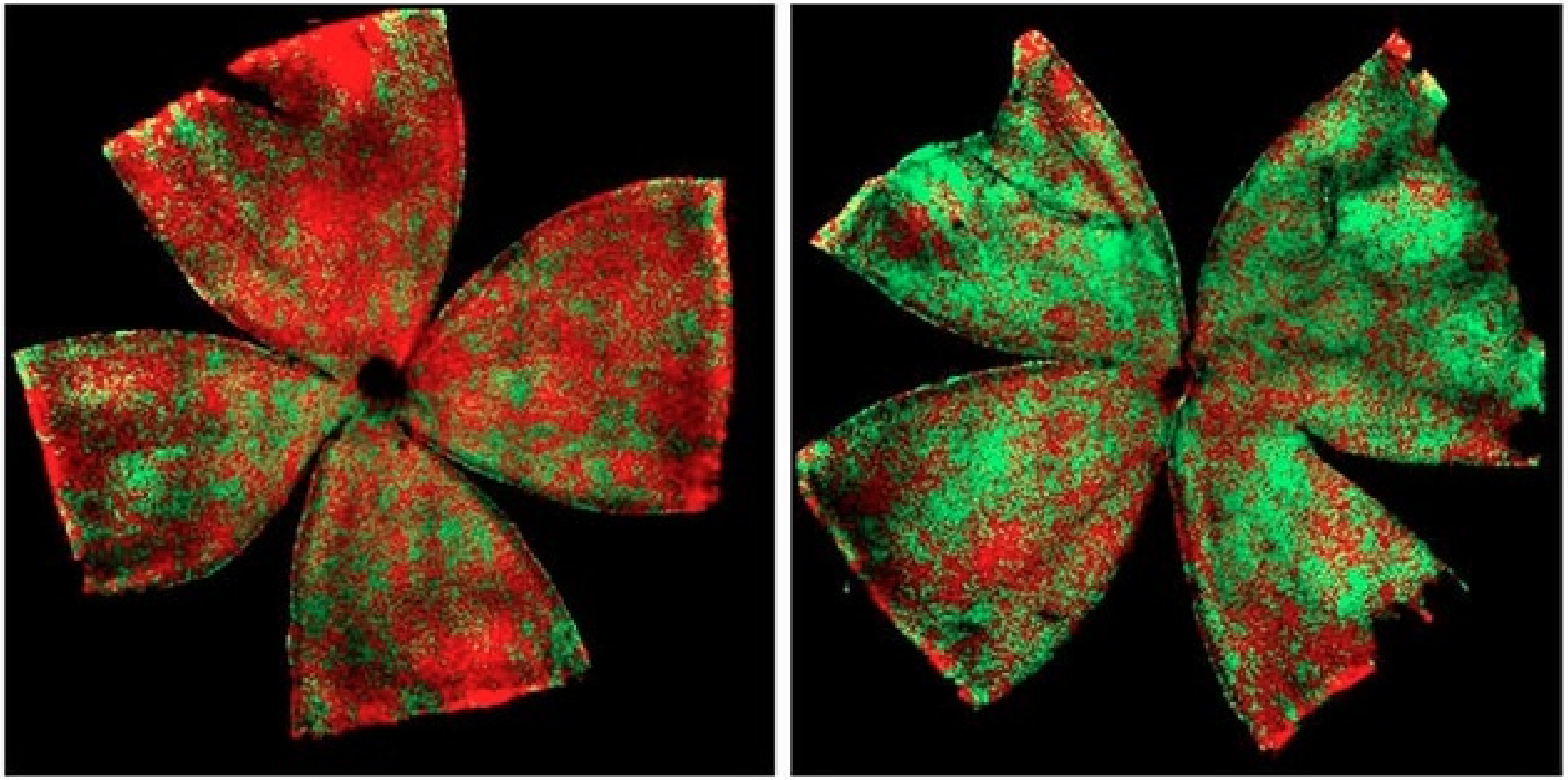
Barbara Migeon, Henry Stewart Talks

Female X chromosome Mosaicism (cornea, skin, cartilage & inner ear)



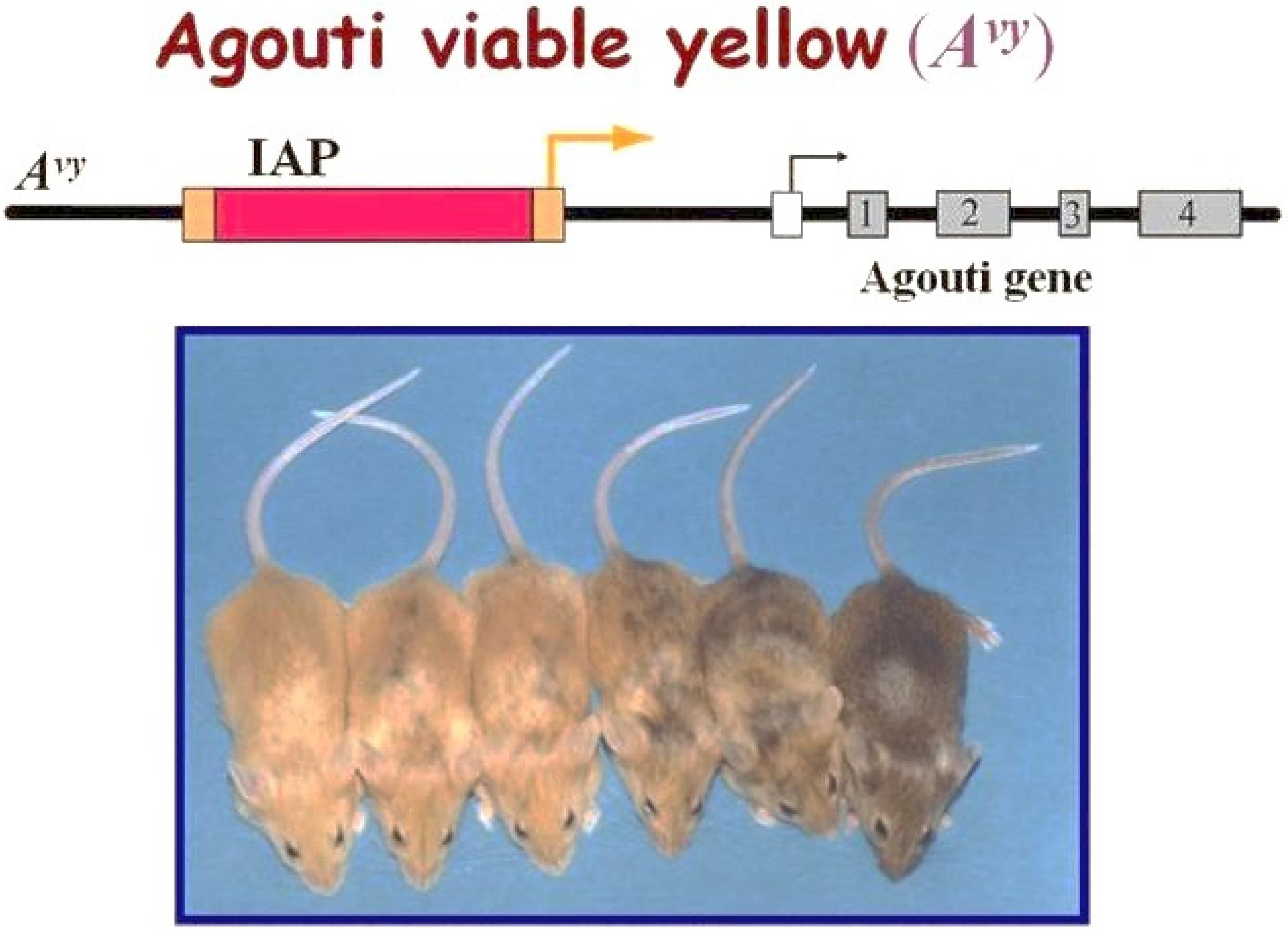
Wu et al., Neuron (2014) 81: 103-119.

Female X chromosome Mosaicism Left and Right Retina



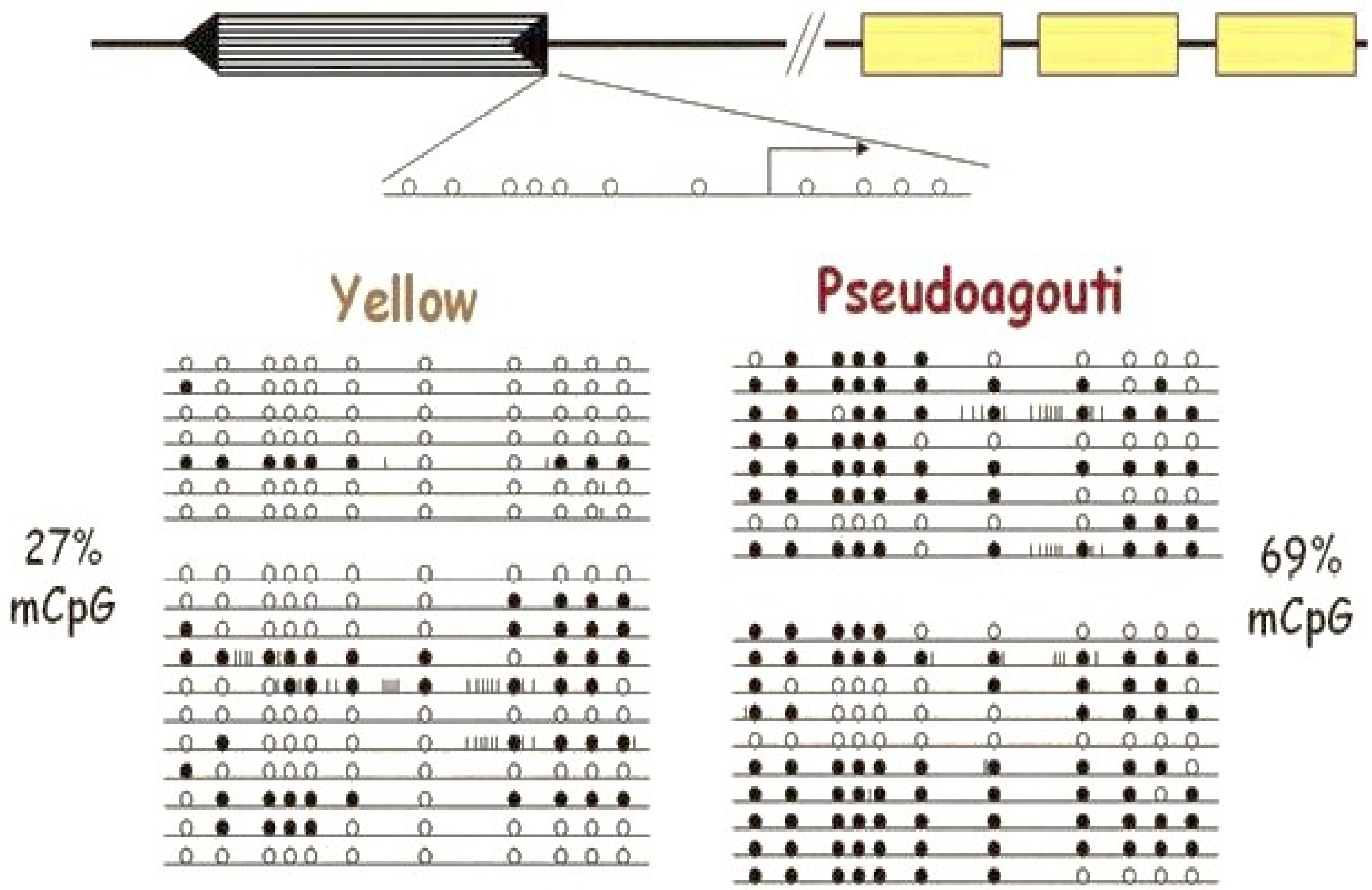
Wu et al., Neuron (2014) 81: 103-119.

Agouti Genes in Mice



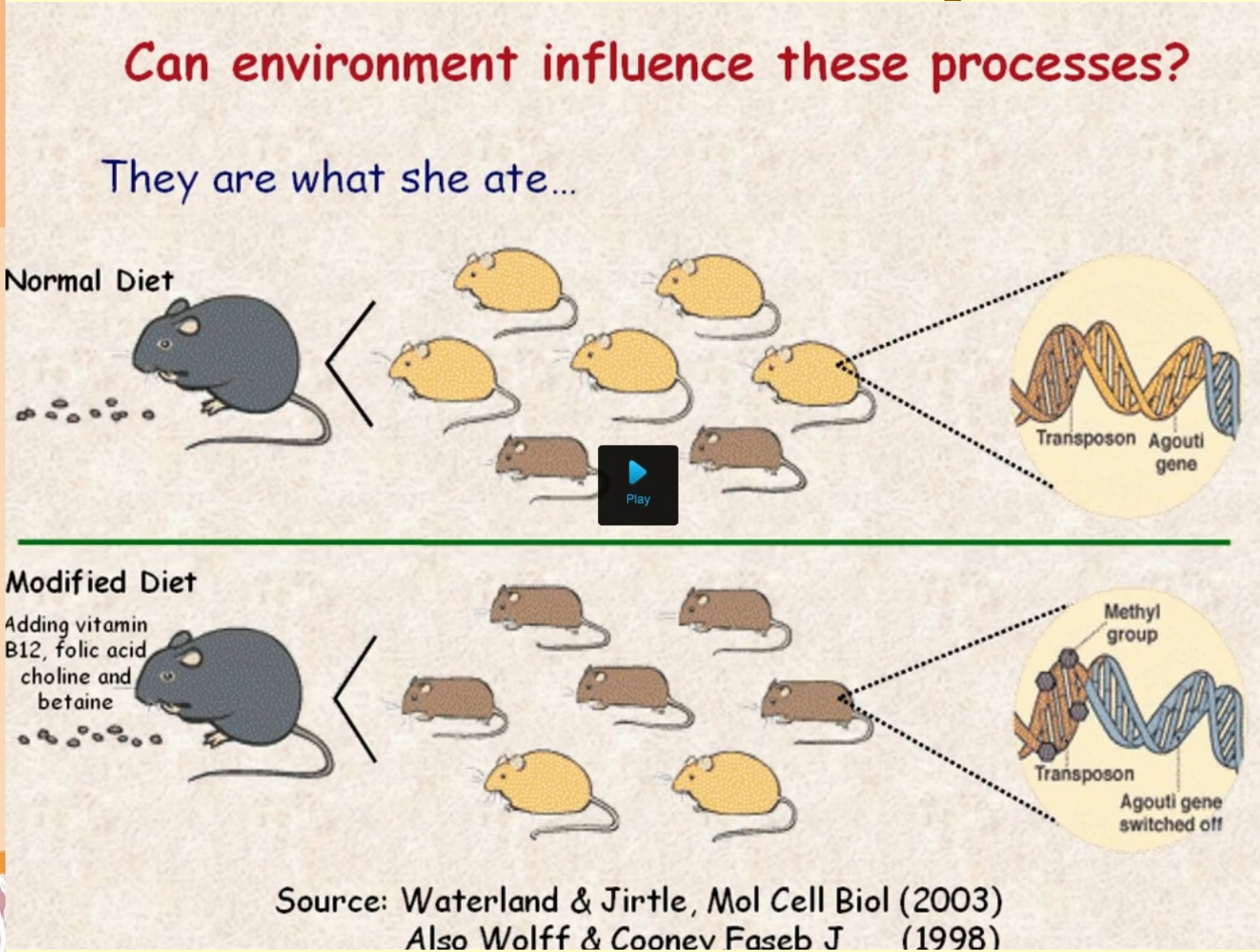
Emma Whitelaw, Epigenetic regulation of phenotype. Henry Stewart Lectures

Methylation of A^{vy} Agouti Genes in Mice



Emma Whitelaw, Epigenetic regulation of phenotype. Henry Stewart Lectures

Environment influences this process



Environment can Affect Epigenetics

- Feed pregnant mice folic acid, vitamin B12, choline, betaine the offspring have more methylation of agouti yellow promoter
- Mothers who lick offspring decreased stress in offspring and decreased methylation of promoters
- Stress also increase methyl state of a number of promoters including the promoter of the glucocorticoid receptor.
- Pattern of epigenetics can be passed on from mother to offspring.

Hongerwinter 1944

- German's blocked food to Belgium and Holland in the winter of 1944.
- Calorie consumption dropped from 2,000 to 500 per day for 4.5 million.
- Children born or raised in this time were small, short in stature and had many diseases including, edema, anemia, diabetes and depression.
- The Dutch Famine Birth Cohort study showed that women living during this time had children 20-30 years later with the same problems despite being conceived and born during a normal dietary state.

Summary of Epigenetic Gene Regulation

- Patterns of DNA methylation in adult cells parallels cell fate, chromatin structure and gene activation.
- Most DNA methylation is removed at fertilization and re-established during embryogenesis.
- Imprinted genes keep their parental pattern of methylation giving rise to parental patterns of expression.
- Patterns of histone modifications parallel DNA methylation.
- Methylated gene regions are genetically inactive, highly condensed and special histone modifications.
- Active gene regions have little DNA methylation and distinctive histone modifications (acetyl groups and H3K4 and H3K27 methyl).
- X chromosome inactivation in females is correlated with extensive CG island methylation on one chromosome, condensation, inactivation and Barr body formation.
- Alterations in gene and CG island methylation patterns are seen in aging and in cancer.
- Most CG islands are not methylated except for X chromosome inactivation and tumor suppressors in cancer.

Henry Stewart Talks: DNA Methylation

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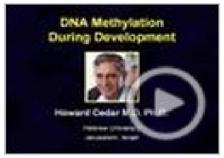
Introduction



1. DNA methylation during development (38 mins)
Prof. Howard Cedar – Hebrew University, Hadassah Medical School, Israel

The Nuts and Bolts of DNA Methylation

2. DNA methylation patterns in mammals (54 mins)
Prof. John Greally – Albert Einstein College of Medicine, USA

3. Proteins that Bind Methylated DNA (31 mins)
Dr. Pierre-Antoine Defossez – CNRS, Paris, France

DNA Methylation and Normal Physiology

4. Genomic Imprinting (25 mins)
Prof. Marisa Bartolomei – University of Pennsylvania, USA

5. DNA methylation and stem cells (32 mins)
Dr. Alexander Meissner – Harvard University, USA

Henry Stewart Talks: Epigenetics

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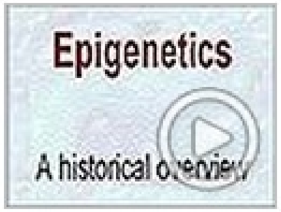
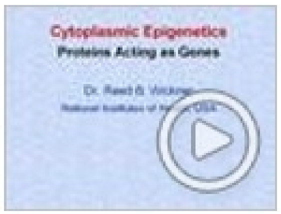
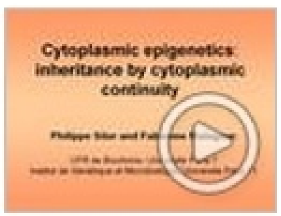
The Notion of Epigenetics



1. Epigenetics: A Historical Overview (24 mins)
Dr. Robin Holliday – National Institute for Medical Research, Mill Hill, London, UK



2. Cytoplasmic Epigenetics: Proteins Acting as Genes (36 mins)
Dr. Reed Wickner – National Institutes of Health, USA



3. Cytoplasmic Epigenetics: Inheritance by Cytoplasmic Continuity (43 mins)
Prof. Philippe Silar – University of Paris, France
Dr. Fabienne Malagnac – University of Paris, France

Epigenetics: Paradigms

4. A Historical Perspective on Ideas on X-Chromosome Inactivation (34 mins)
Dr. Mary Lyon – Mammalian Genetics Unit, Medical Research Council, UK

Henry Stewart Talks: Eukaryotic Gene Regulation

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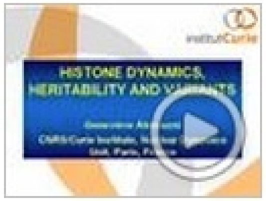
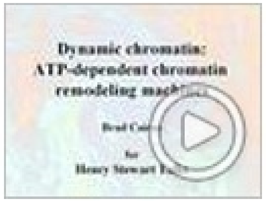
Chromatin and Epigenetics

8. Introduction to Chromatin Structure (41 mins)
Prof. Karolin Luger – Colorado State University, USA



9. Histone Dynamics, Heritability and Variants (36 mins)
Dr. Genevieve Almouzni – Curie Institute/CNRS, France



10. Dynamic chromatin: ATP-dependent chromatin remodeling machines (56 mins)
Prof. Bradley Cairns – University of Utah School of Medicine, USA

11. Epigenetic Information in Gene Expression and Cancer (34 mins)
Prof. Siavash Kurdistani – University of California, Los Angeles, USA

12. Gene Silencing by Polycomb Complexes (27 mins)
Prof. Yi Zhang – University of North Carolina at Chapel Hill, USA