L2.3

Establishment & maintenance of epigenetic programming

Epigenetic inheritance

Textbook

Review

CellPress

Special Issue: Chromatin Dynamics

Epigenetic inheritance: histone bookmarks across generations

Eric I. Campos, James M. Stafford, and Danny Reinberg

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Multiple circuitries ensure that cells respond correctly to the environmental cues within defined cellular programs. There is increasing evidence suggesting that cellular memory for these adaptive processes can be passed on through cell divisions and generations. However, the mechanisms by which this epigenetic information is transferred remain elusive, largely because it requires that such memory survive through gross chromatin remodeling events during DNA replication, mitosis, meiosis, and developmental reprogramming. Elucidating the processes by which epigenetic information survives and is transmitted is a central challenge in biology. In this review, we consider recent advances in understanding mechanisms of epigenetic inheritance with a focus on histone segregation at the replication fork, and how an epigenetic memory may get passed through the paternal lineage.

Please, do not disregard these small yellow boxes.
They are important in order to understand well what the Review tells us.

This Yellow box here contain just «definitions».

Glossary

Epigenetic inheritance: the inheritance of a phenotype in a manner that is independent of the DNA sequence and that remains self-perpetuating in the absence of the initial stimulus that caused the phenotype in the parental cell or organism.

Histone variant: core canonical and linker histones are encoded by a number of different histone genes, resulting in a number of non-synonymous substitutions and divergent domains. This variation adds complexity to the epigenetic landscape.

Histone chaperone: proteins or protein complexes that specifically bind histones, thwarting non-specific interactions, and that promote their deposition or removal from DNA in an ATP-independent manner.

PcG: polycomb Group Proteins. A group of proteins involved in the regulation and transcriptional silencing of key developmental genes, including the *Homeotic* (or *Hox*) gene loci. Human PcG proteins assemble into Polycomb Repressive Complexes (PRCs), of which PRC2 catalyzes the methylation of H3K27 and PRC1 guides the ubiquitin ligation of H2AK119.

Protamine: low molecular weight proteins that tightly package DNA in late spermatids and mature sperm largely due to their arginine-rich DNA anchoring domains. Their precise function is unknown but might include protecting the paternal genome from DNA damage, facilitating formation of a small elongated sperm head for better motility and/or conveying epigenetic information.

Spermatogenesis: the process of generating mature, haploid sperm (spermatozoa) from a diploid spermatogonium. This process initially requires mitosis to create spermatocytes, their subsequent meiotic divisions to create spermatids and finally maturation of spermatids to spermatozoa. During this chain of events, chromatin undergoes dynamic changes whereby canonical histones are largely replaced by protamines through a number of intermediate steps, including histone variant incorporation, nucleosomal destabilization, histone eviction and replacement with transition proteins prior to protamine deposition.

While this one contains an important discussion on trans-generational transmission of epigenetic characters.

This is not relevant to what we discuss today, but it will become very important for Lesson 2.4.

Box 1. Transgenerational inheritance; considering caveats and alternative mechanisms

Non-chromatin based mechanisms likely contribute to transgenerational inheritance. For example, some of these phenotypes might arise from cryptic genetic variation given that inbred strains, nearly identical clones or even neighboring cells in the same organism may possess marked genetic differences [108]. Such genetic variation could be passed on to offspring or arise *de novo* (e.g., transposable elements, mutations) and account for differences. Unfortunately, these alternatives are seldom examined in transgenerational studies. Furthermore, establishing transgenerational inheritance in its purest sense is often confounded by maternal care, social transmission, or other variables that may propagate a phenotype without requirement for epigenetic memory *per se*. Indeed recent studies suggest that maternal care may play a significant role even in the transmission of phenotypes originating from the father [109].

Even if a phenotype is transmitted in a transgenerational epigenetic fashion, chromatin events may not always be responsible for their propagation. Transcriptional loops are one example [110]. As in somatic tissue, noncoding RNAs such as siRNA, piRNAs as well as miRNA contribute to inheritance and might function independently of changes at the level of chromatin (recently reviewed by [63]). In fact, a recent study showed that miRNAs are important for transmitting the experience of trauma to progeny through the paternal lineage [65]. Studying the importance of these varied contributions to transgenerational inheritance is important in understanding whether they are truly epigenetic.

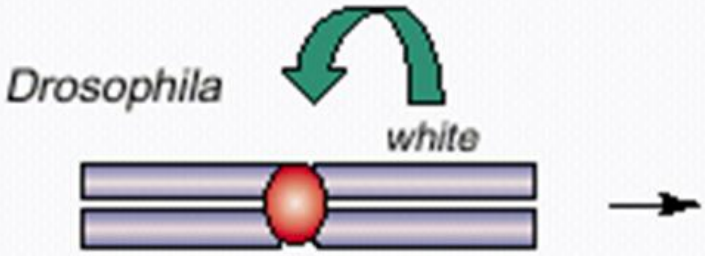
It is useful at this time to go back to very old textbook acquisitions, which can unravel features of chromatin that explain how «heredity» of functional status may work.

There are few **model systems** from which we have learnt several important concepts.

The first data concerns the historical concept in Genetics of the so-called «positional effect variegation» (**PEV)** in *D. melanogaster (…and other model organisms).*

Drosophila ?
Please note that Insects present very low levels of cytosine methylation. It has been mainly attributed to transposon silencing.

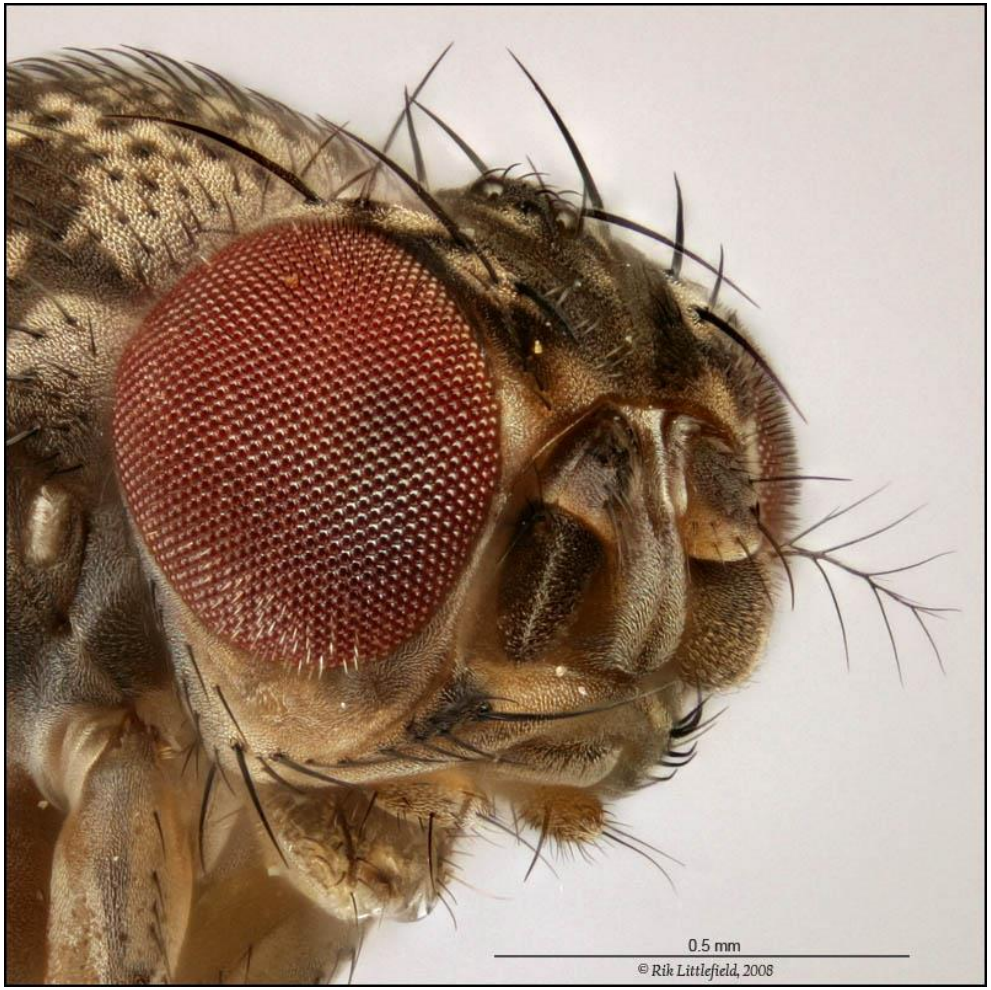
PEV = Positional Effect Variegation Silencing effects of heterochromatin in different organisms.



Translocation of the Drosophila *white* gene close to the centromere results in mosaic (variegated) expression in the eye.

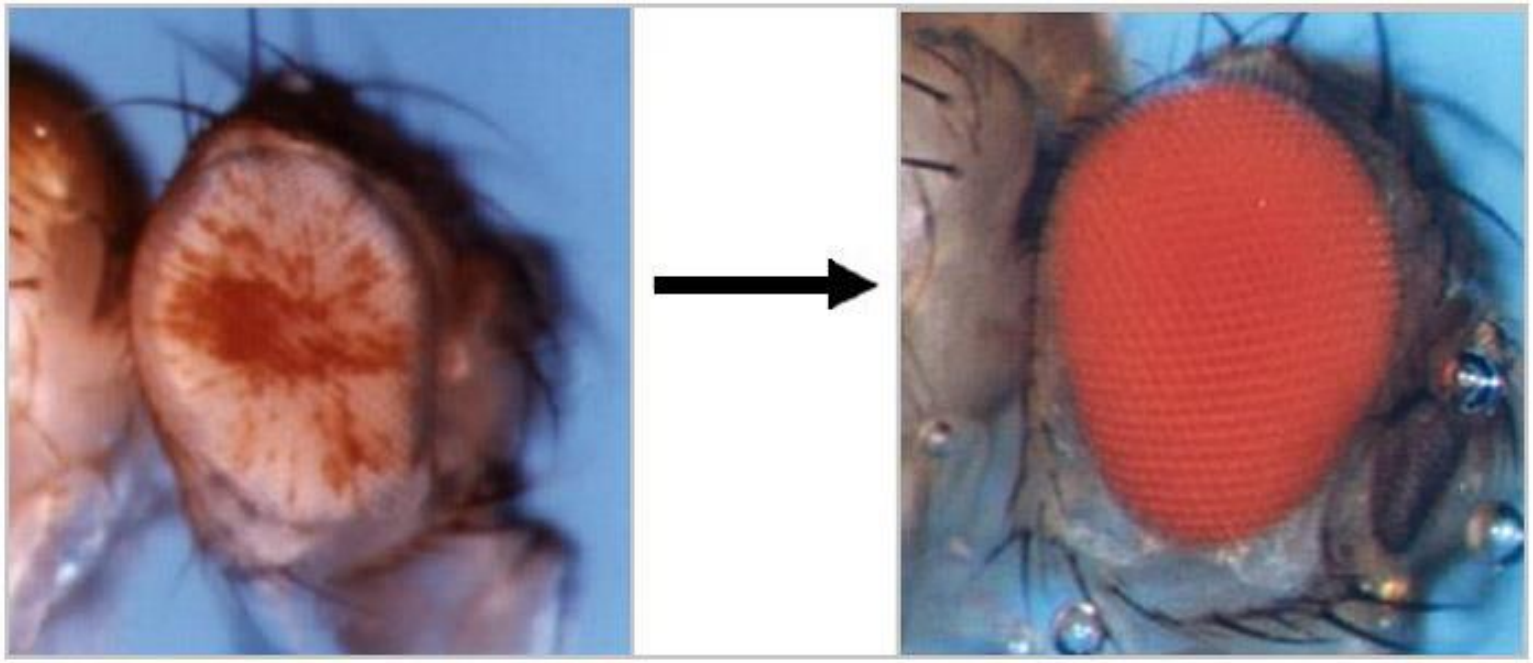
Insertion of transgenes into peri-centromeric regions in mice gives expression in a proportion of cells from expressing tissues. Expression is clonally stable through multiple cell divisions but also undergoes stochastic switching between the active and inactive states.

Insertion of the Ade2 gene into the telomeric region of Saccharomyces cerevisiae causes metastable silencing of the gene resulting in sectored colonies containing white (expressing) and red (nonexpressing) cells.


0.5 mm
© Rik Littlefield, 2008

Variegated

Actually, looking at the eyes of these animals, we observe patches of pigmented cells among a general white color of other cells.



Washington University Department of Biology - Mutations in HP1 lead to a loss of silencing, a suppression of PEV.

Mendez et al, 2011

PEV = Positional Effect Variegation Silencing effects of heterochromatin in different organisms.

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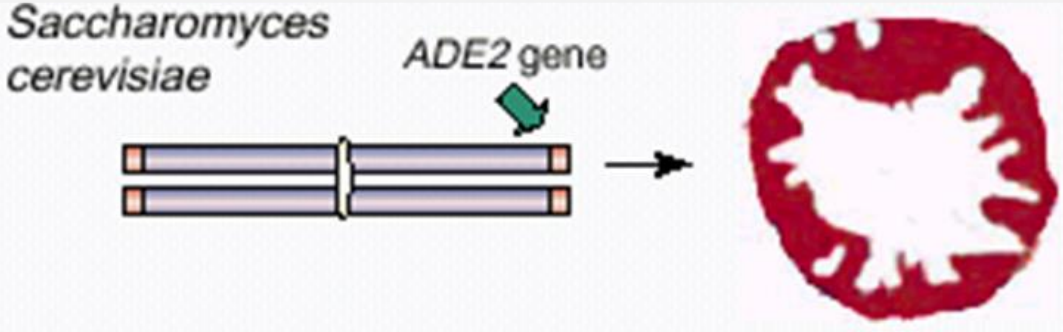

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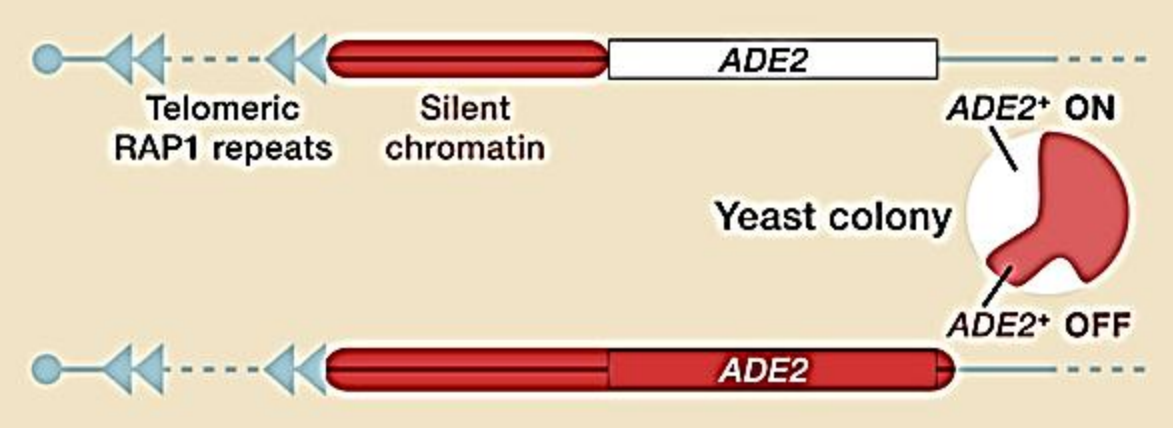
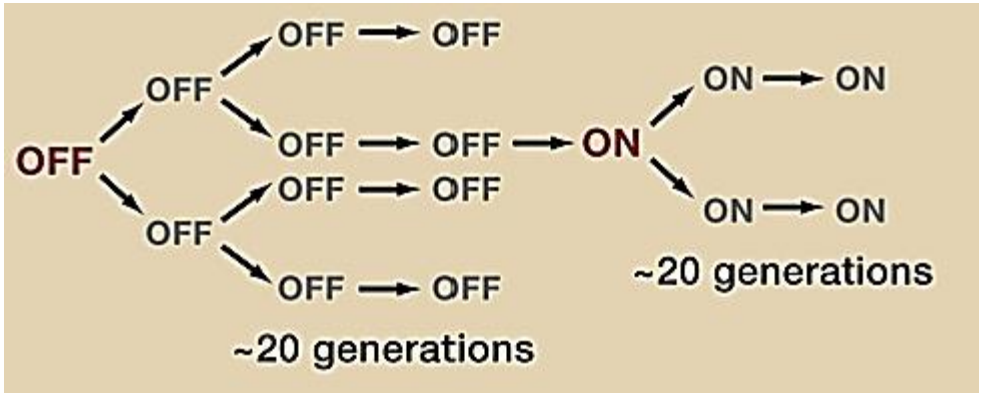
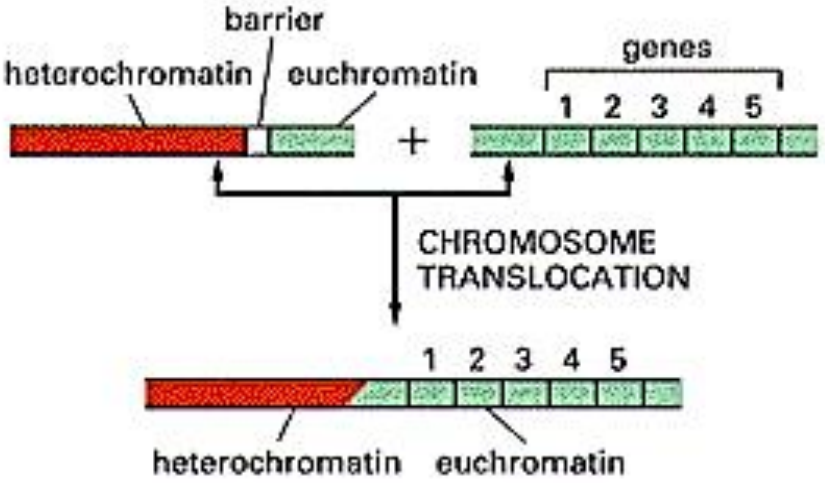


PEV

This variegation results from stochastic loss and re-establishment of silent chromatin and indicates that following a switch in gene expression, the daughters of the switching cell have **a memory of the expression state** of the mother cell.

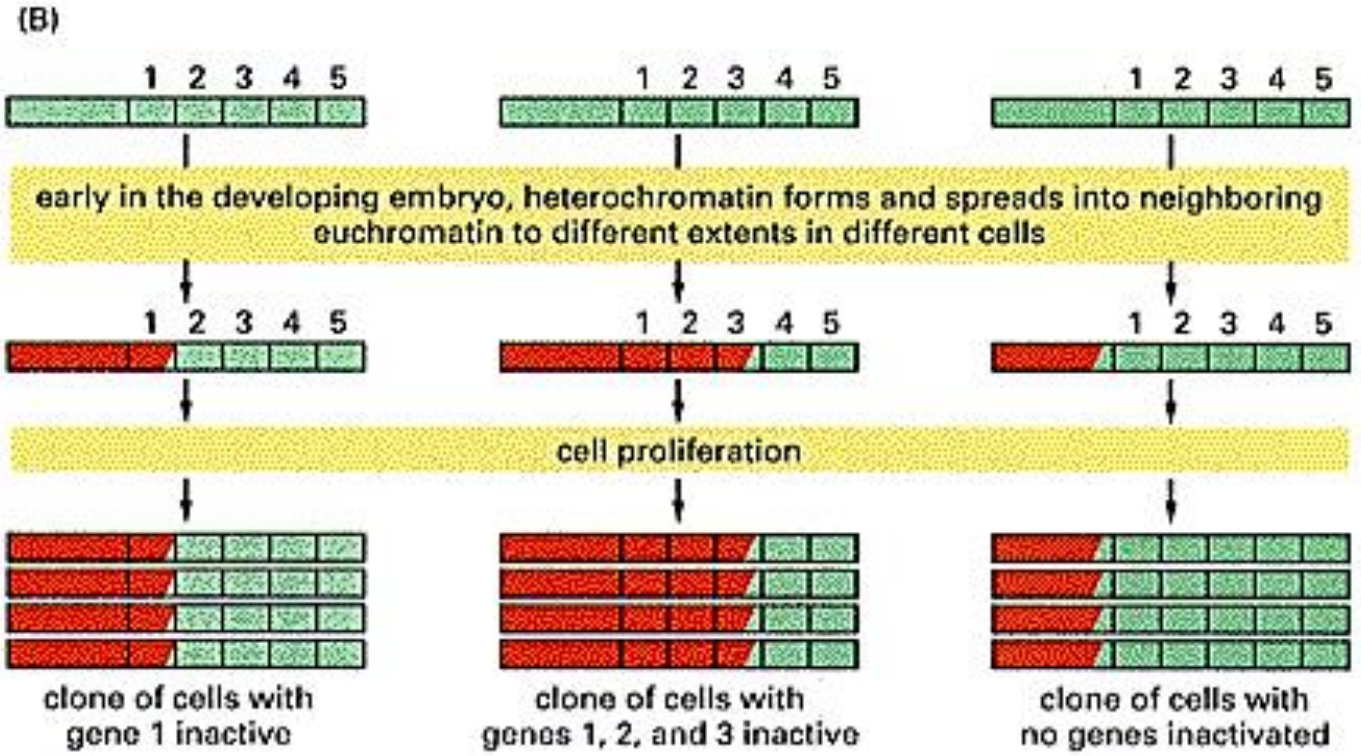


In normal situation, a «barrier» exist between heterochromatic and euchromatic domains
We call it «insulator».



Establishement →

Maintenance →



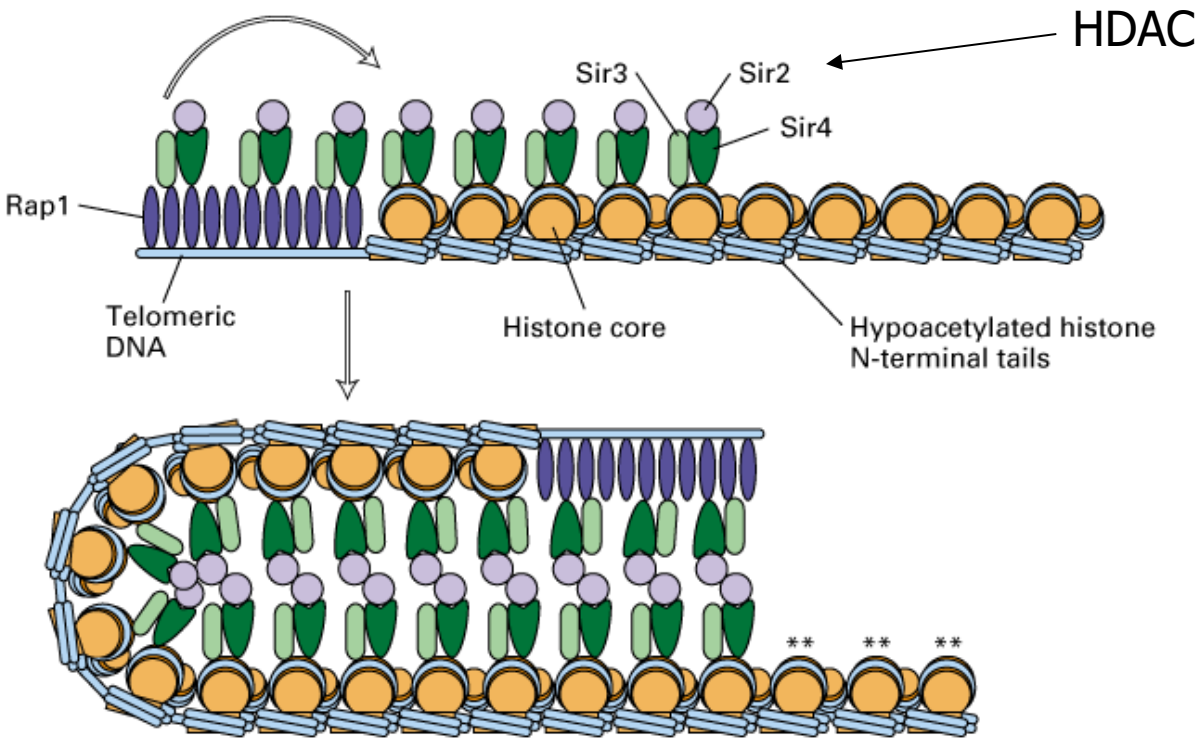
Hetorochromatin starts from one point, then «spreads» until an insulator is reached

Establishment and maintenace (heredity) phases are separated

What are the determinants ?

Old model systems from Yeast:

- Telomeric heterochromatin (S. cerevisiae)
- MAT locus (S. cervisiae)
- Centromeric heterochromatin (S. pombe)



budding yeast

Linear chromosome replication

Telomere structure

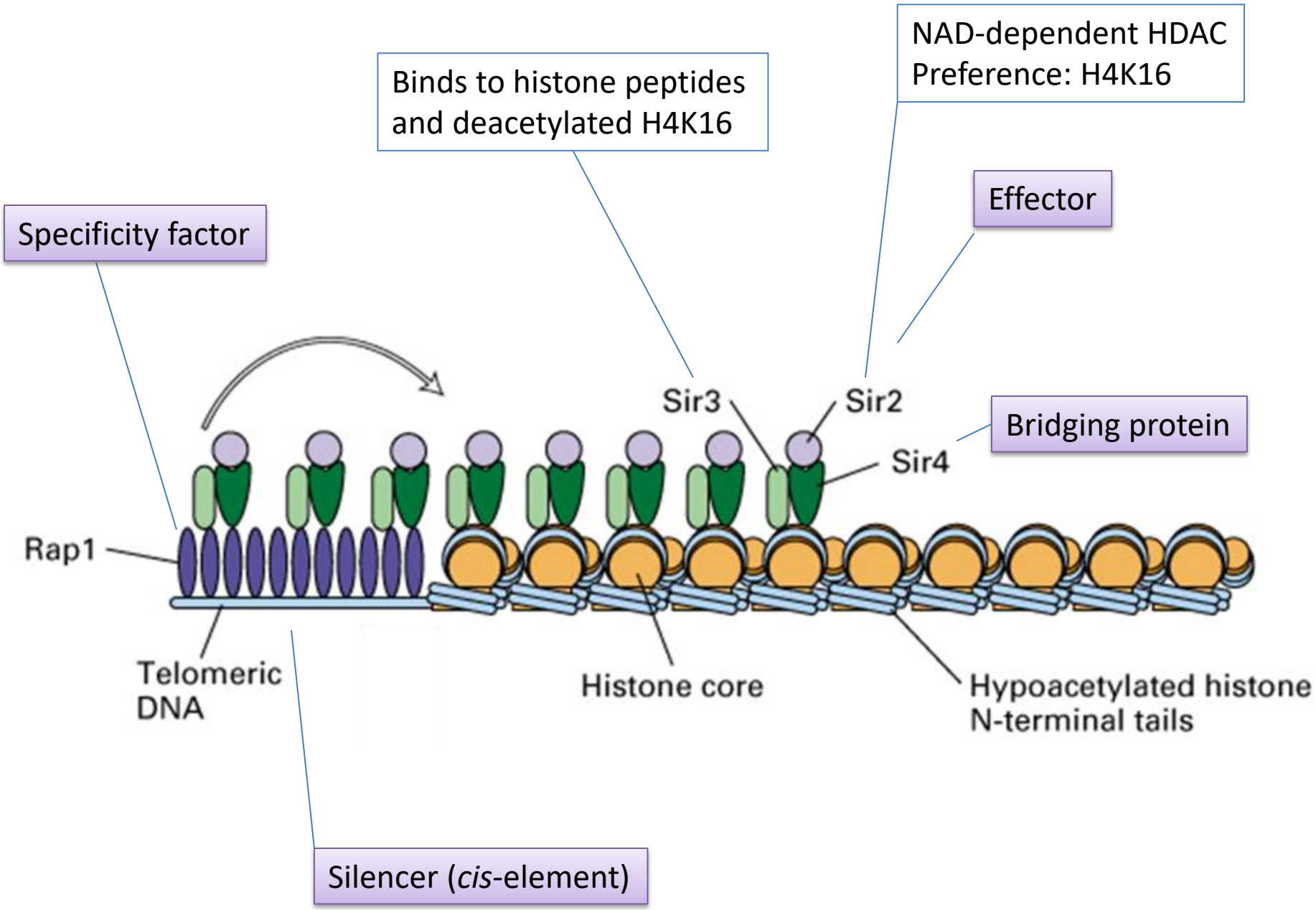
Telomerase

Model for the formation of telomeric heterochromatin. Black lines wrapped around nucleosomes represent DNA.

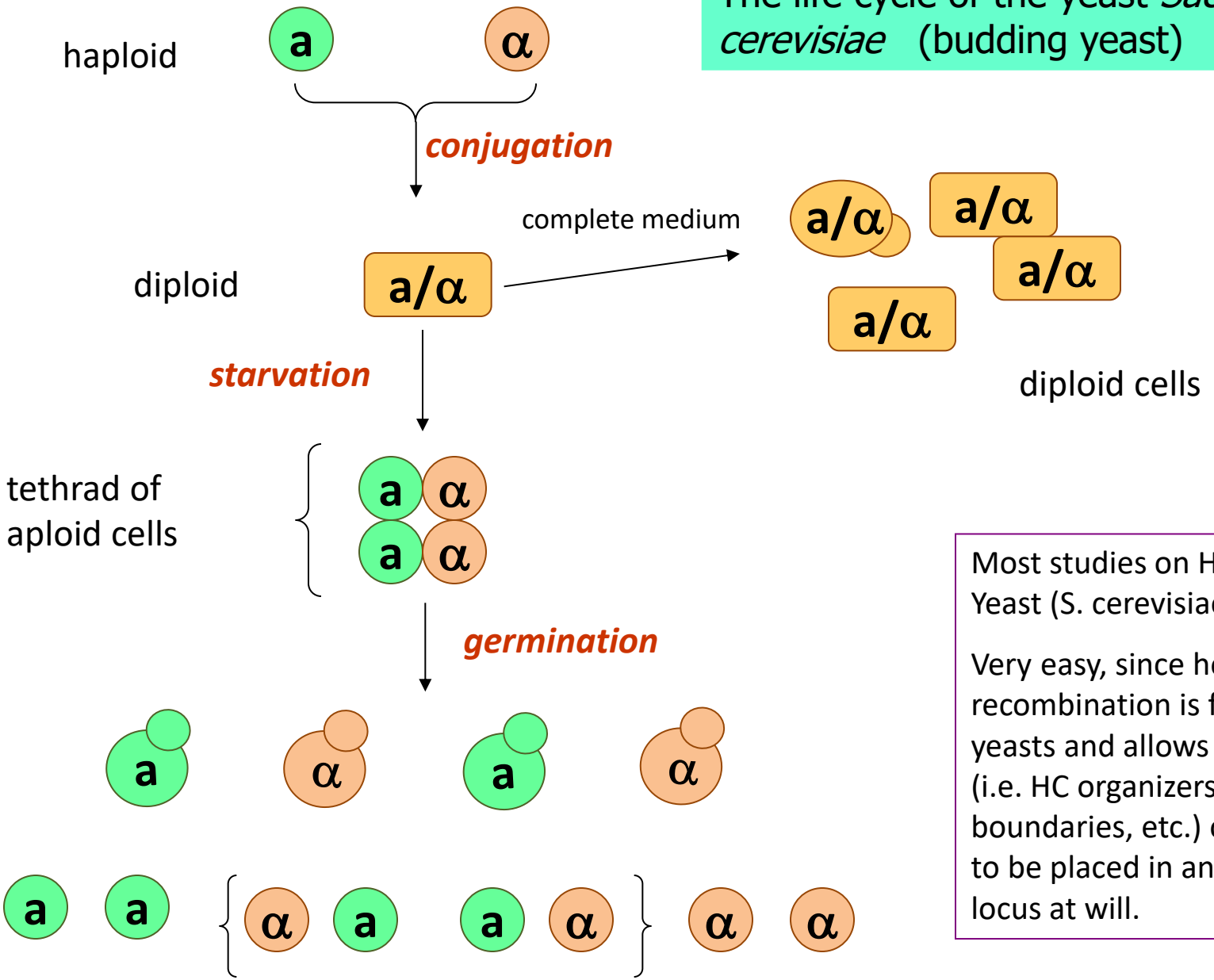
(a) Core telomeric heterochromatin in wild-type cells containing only a single genomic copy of SIR3. It is proposed that the RAP1-containing telosome folds back onto subtelomeric regions. In this manner, RAP1–SIR–histone interactions are all required for stability of the complex.

(b) Upon SIR3 overexpression, telomere position effect and the presence of SIR3 is extended up to some 16–20 kb from the telomere. SIR3 overexpression causes loss of some SIR4 and most SIR2 from the complex. Due to the interdependence of RAP1–SIR3–H4 interactions, and because all three SIR proteins are required for extension of heterochromatin by SIR3, it is proposed that the complex necessary for the initiation of heterochromatin formation requires RAP1, the SIR proteins and H4.

S. Cerevisiae telomere HC



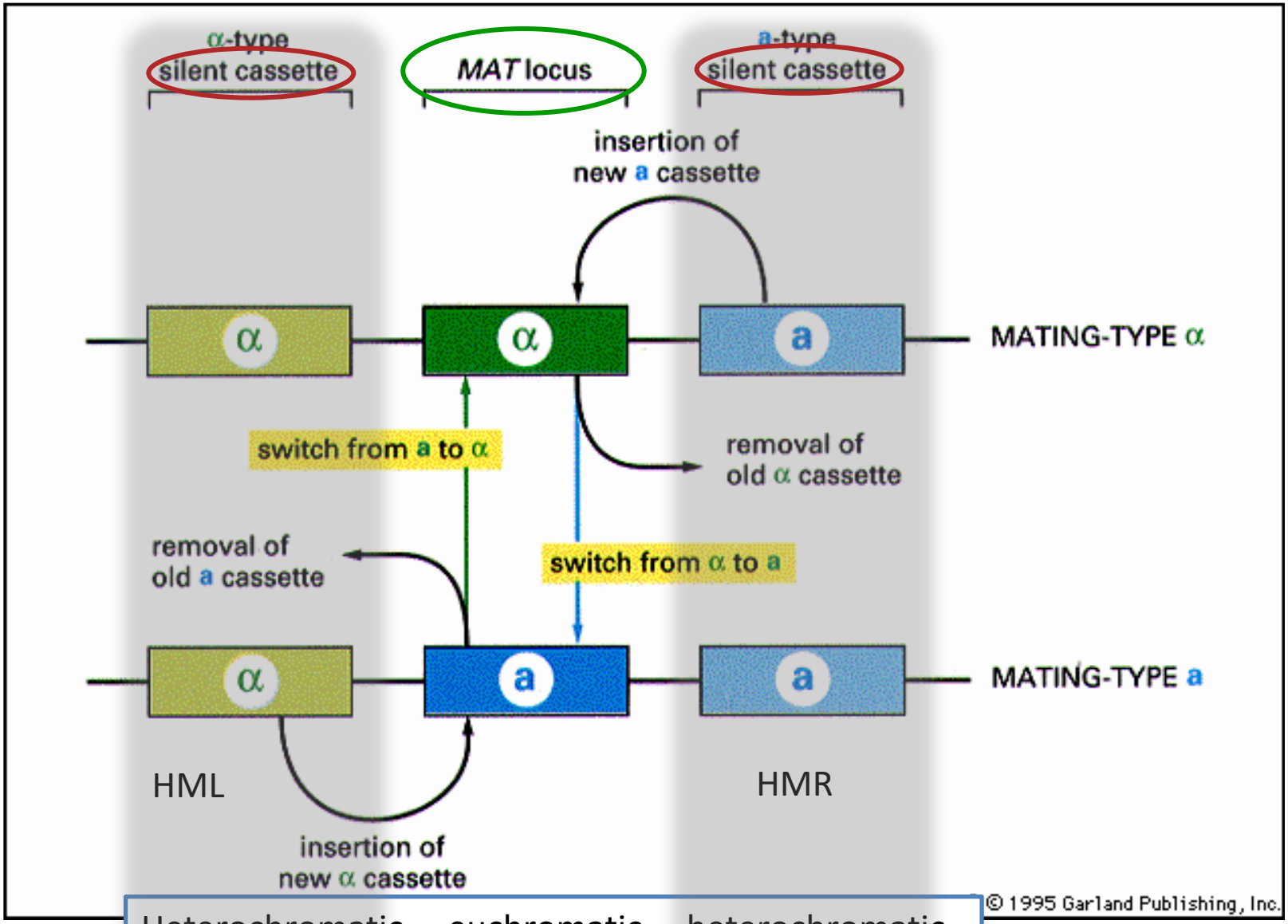
The life cycle of the yeast *Saccaromyces cerevisiae* (budding yeast)



Most studies on HC made in the Yeast (S. cerevisiae / S. pombe)

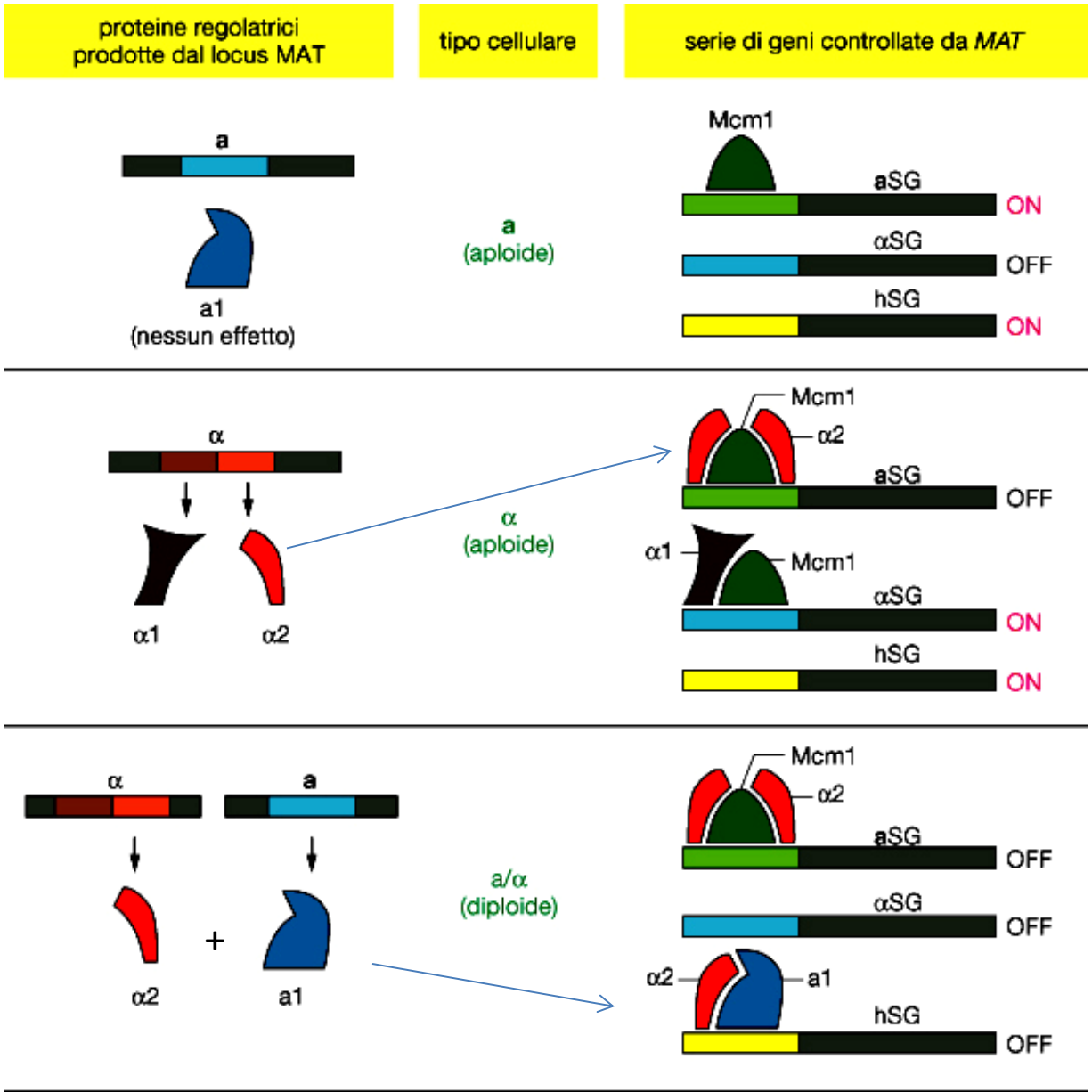
Very easy, since homologous recombination is favored in yeasts and allows HC elements (i.e. HC organizers, silencers, boundaries, etc.) or reporters to be placed in any genomic locus at will.

HMR/HML loci in yeast have heterochromatic features, e.g. are resistant to endonuclease digestion and **silence constructs placed within**



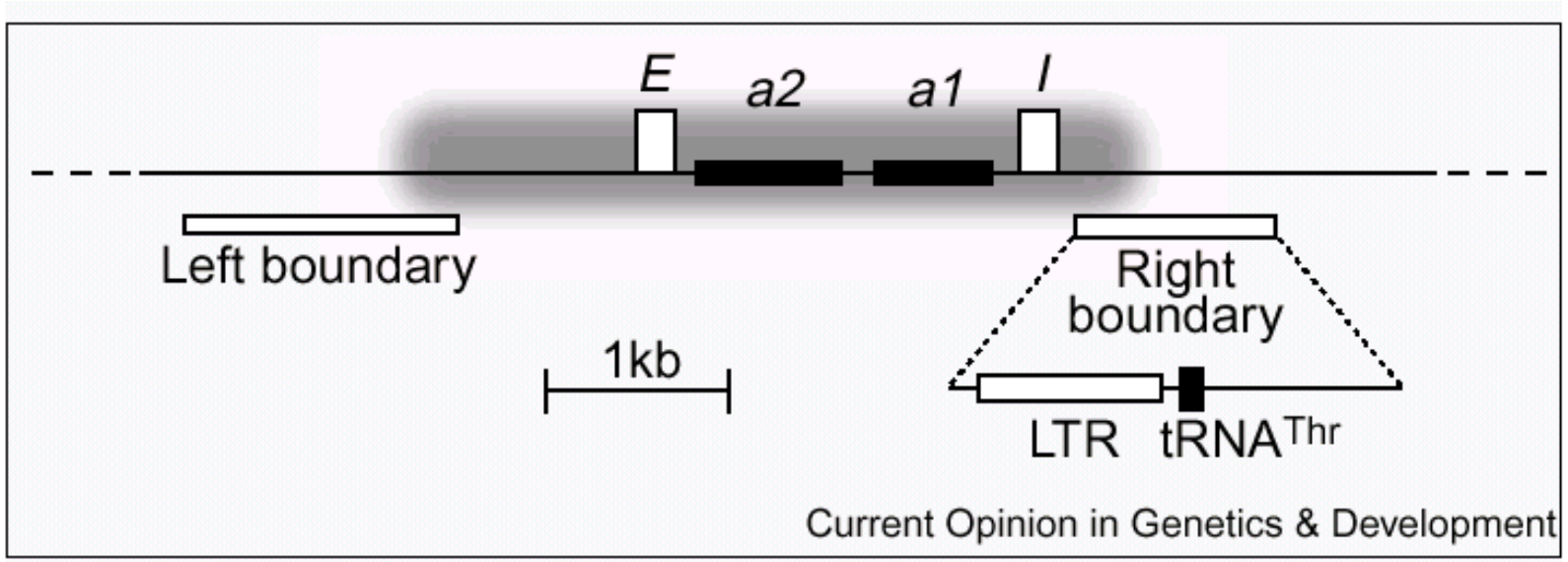
Heterochromatic euchromatic heterochromatic

A and α cassettes encode transcription factors that regulate aploid and mating type-specific genes



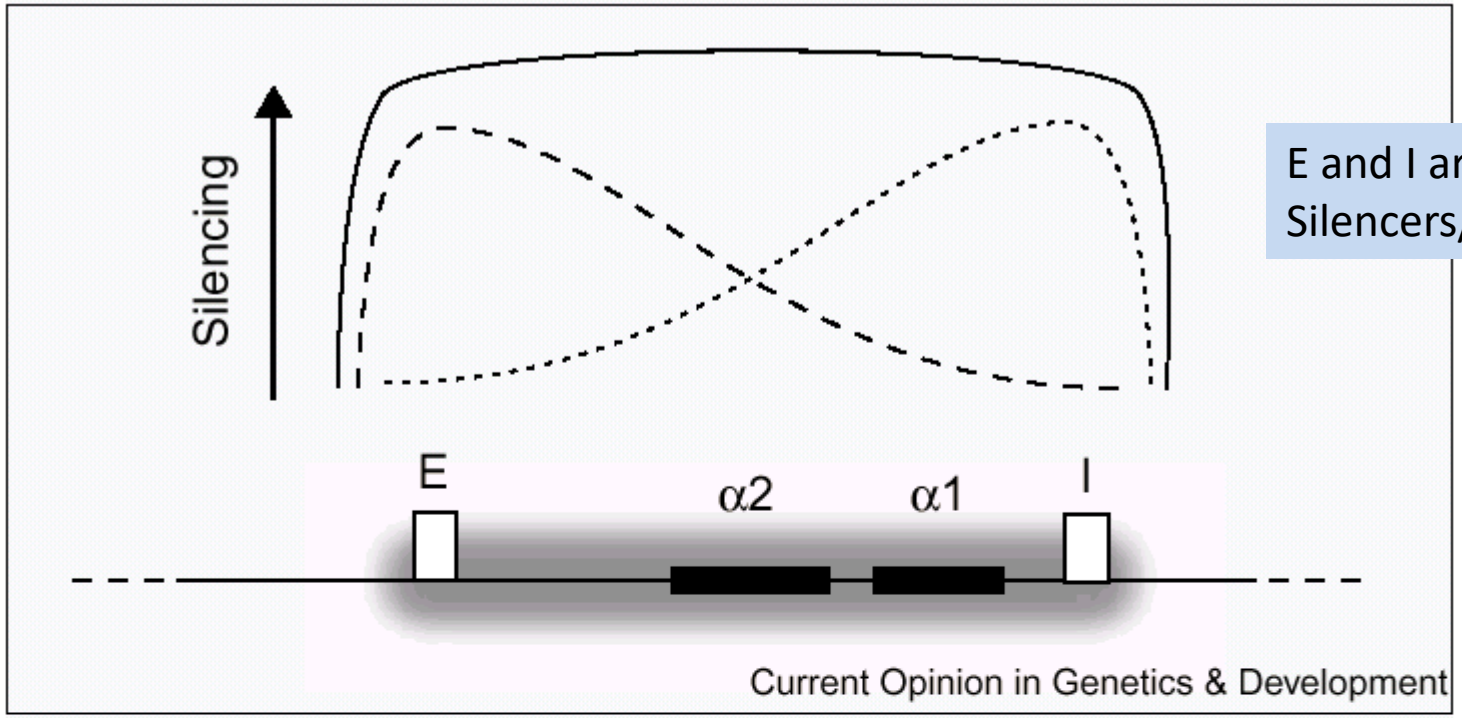
Mcm1 is constitutive

a-specific genes (aSG)
α-specific genes (αSG)
haploid-specific genes (hSG)



HMR

Organization of heterochromatin barriers surrounding *HMR*. The *HMR* locus is diagrammed, showing the location of the mating type genes *a1* and *a2*, the *E* and *I* heterochromatin organizing centers (silencers) and the left and right heterochromatin barriers as defined in [1••]. Background shading indicates the extent of the repressed domain. An expansion of the right barrier shows the location of a Ty1 LTR and the gene for $tRNA^{Thr}$. Earlier evidence suggested that both these elements contribute to barrier activity [1••] but more recent data indicate that the $tRNA^{Thr}$ gene is necessary and sufficient for full barrier activity



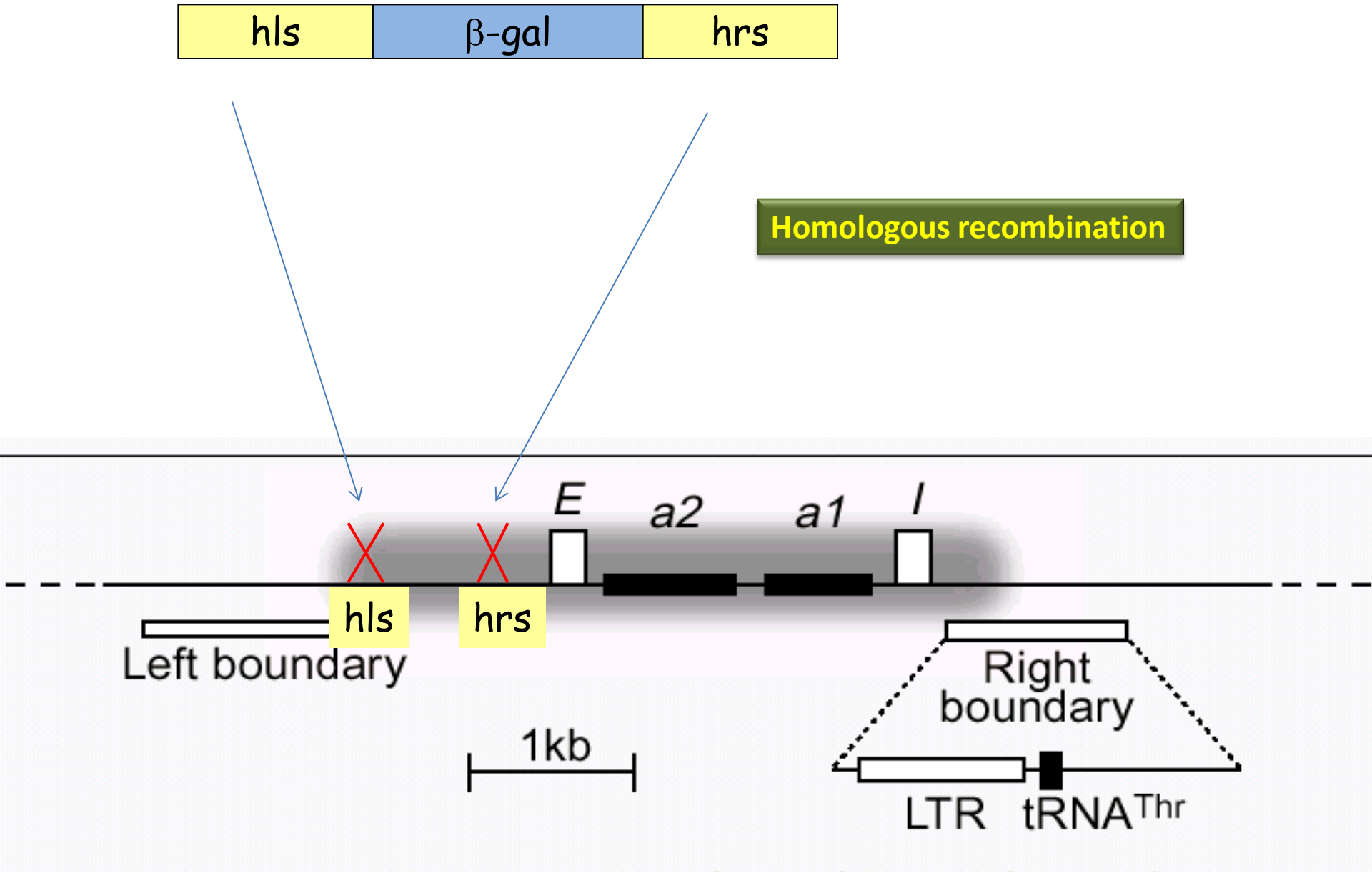
E and I are mixed
Silencers/barriers

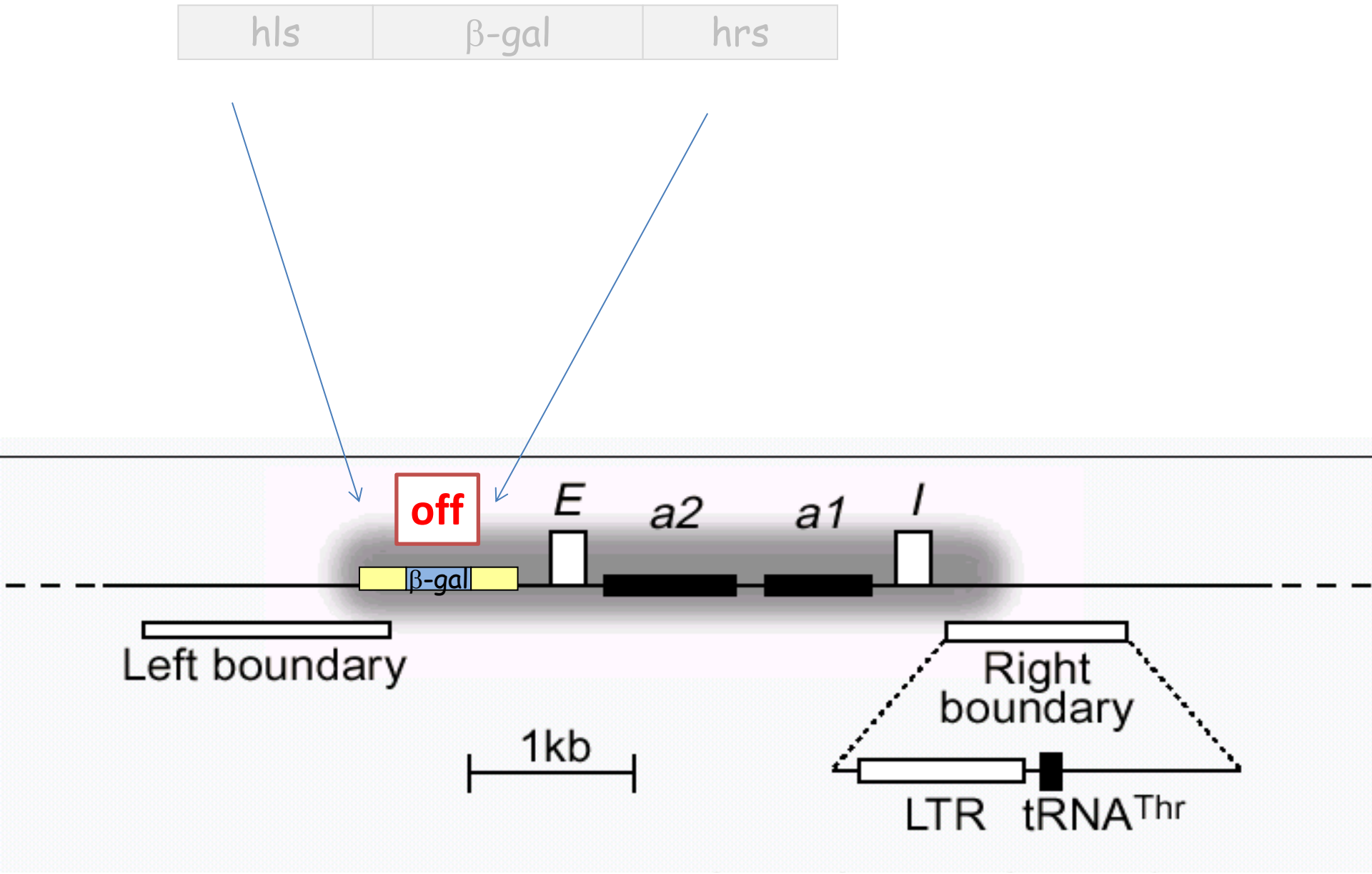
HML

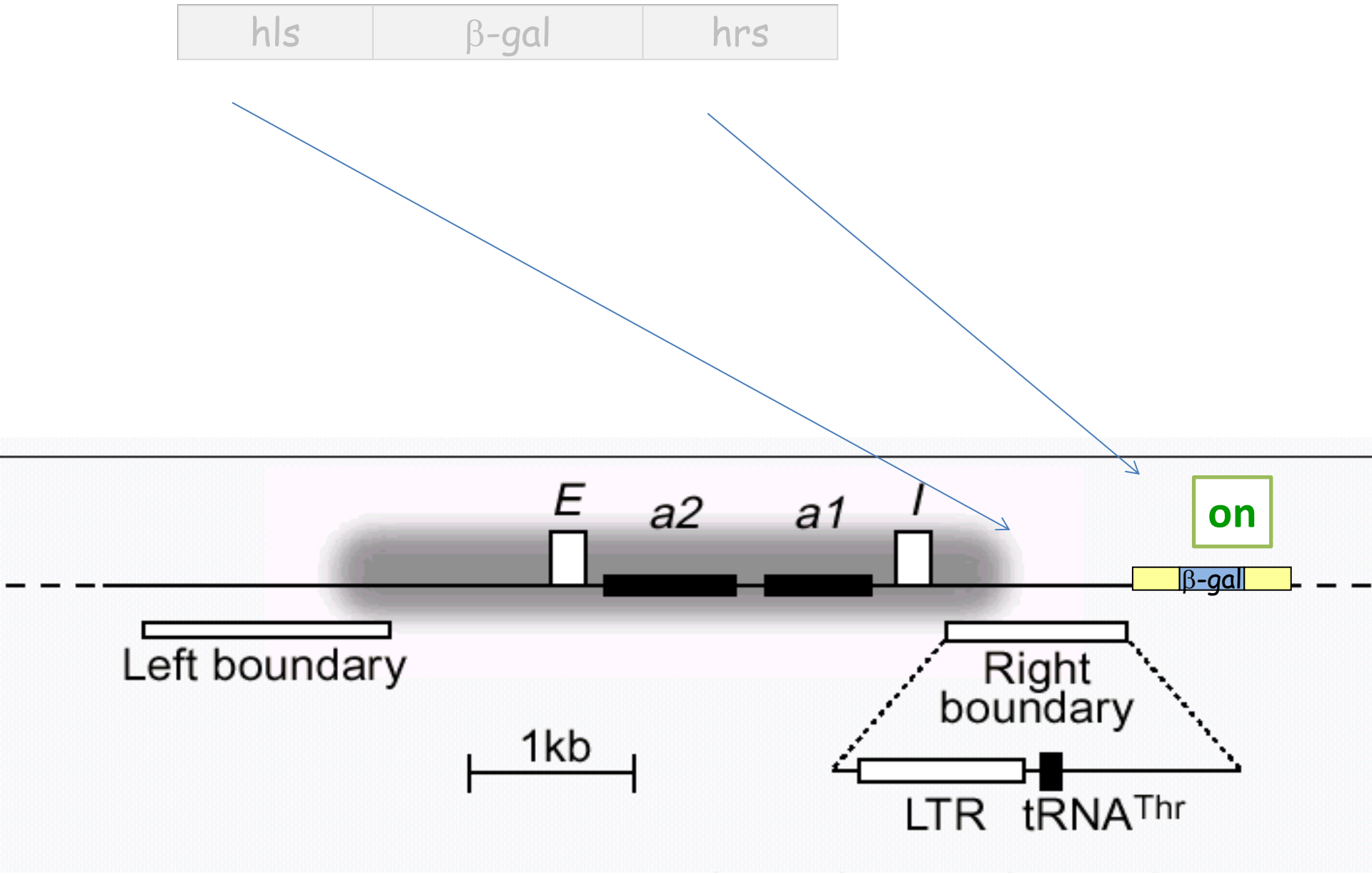
Domain organization by directional initiation of heterochromatin. The *HML* locus is diagrammed, showing the location of the mating type genes, α*1* and α*2*, and the *E* and *I* heterochromatin organizing centers (silencers). Background shading indicates the extent of the repressed domain. Repression emanating from *E* is represented as a dashed line, whereas that emanating from *I* is represented as a dotted line. The sum of the effects of the two organizing centers, shown as a solid line, results in uniformly high repression between the two centers with repression dropping off sharply outside the domain. This model emerges from studies on the domain organization of HML presented in [35•].

High efficiency of Homologous Recombination in Yeast makes easy to study the effects of Heterochromatizatin / euchromatization, using reporter genes such as:

- Beta-galactosidase (blue color)
- Ura4 (S. pombe) Ura3 in S.cerevisiae) is required to grow in absence of uracil and renders cells sensitive to toxicity of 5-FOA
- …other


hls
β-gal
hrs
Homologous recombination
E
a2
a1
I
hls
hrs
Left boundary
1kb
Right boundary
LTR
tRNA Thr


hls
β-gal
hrs
off
E
a2
a1
I
β-gal
Left boundary
Right boundary
1kb
LTR
tRNA Thr


hls
β-gal
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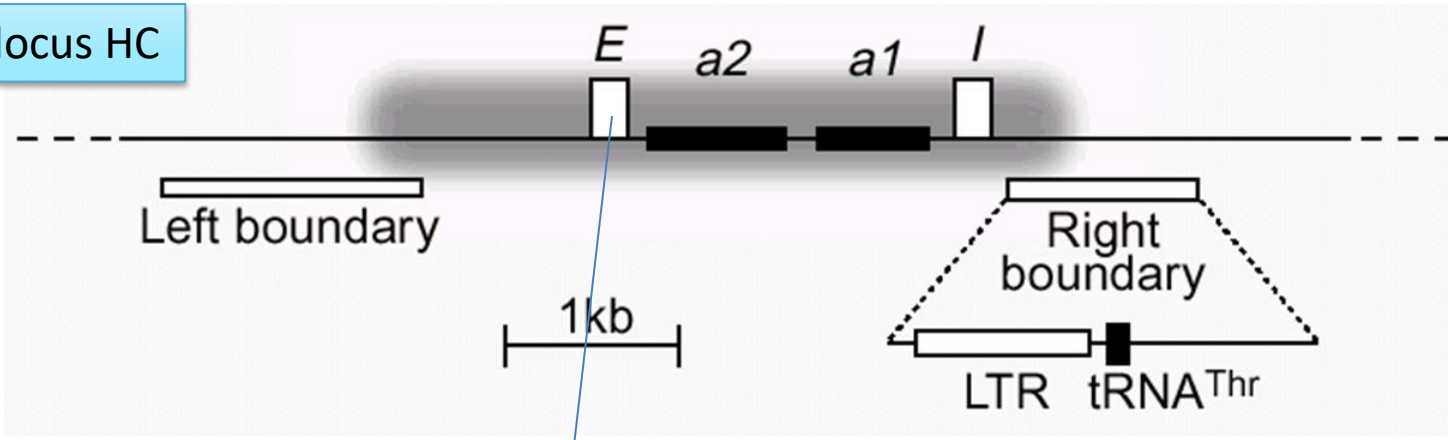
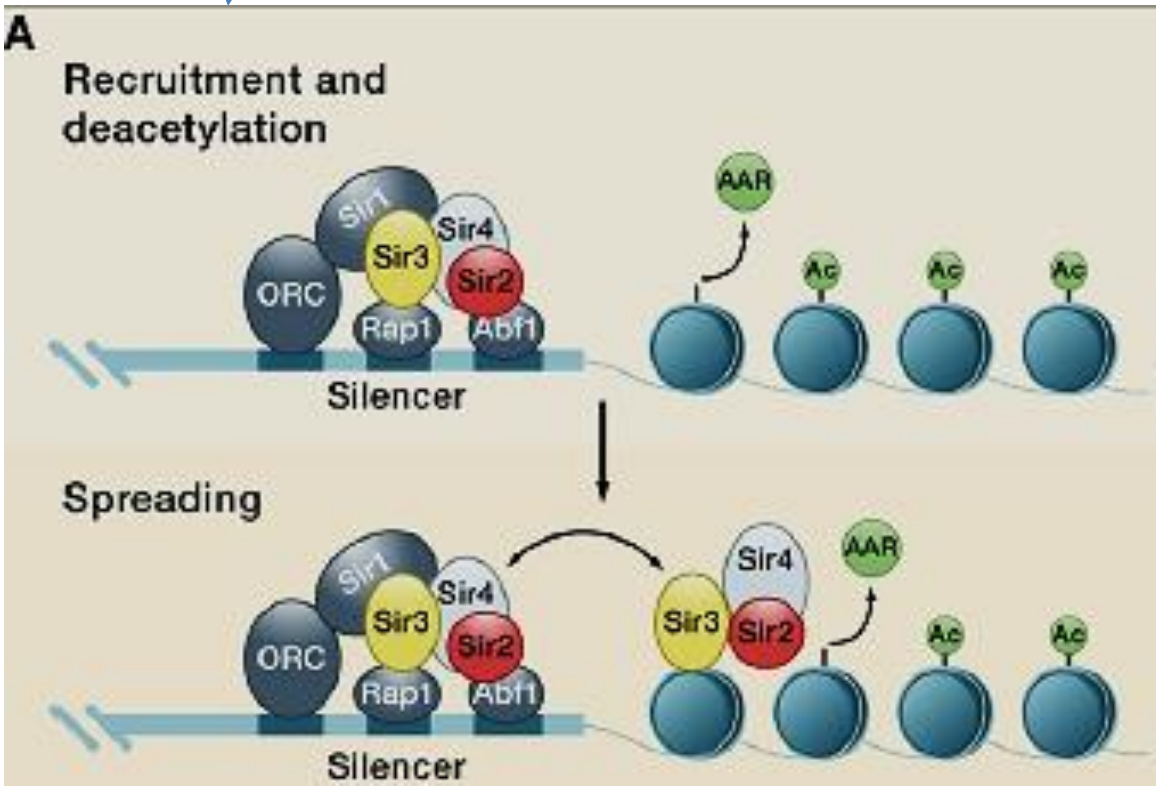
E
a2
a1
I
on
β-gal
Left boundary
Right boundary
1kb
LTR
tRNA Thr

S. Cerevisiae MAT locus HC



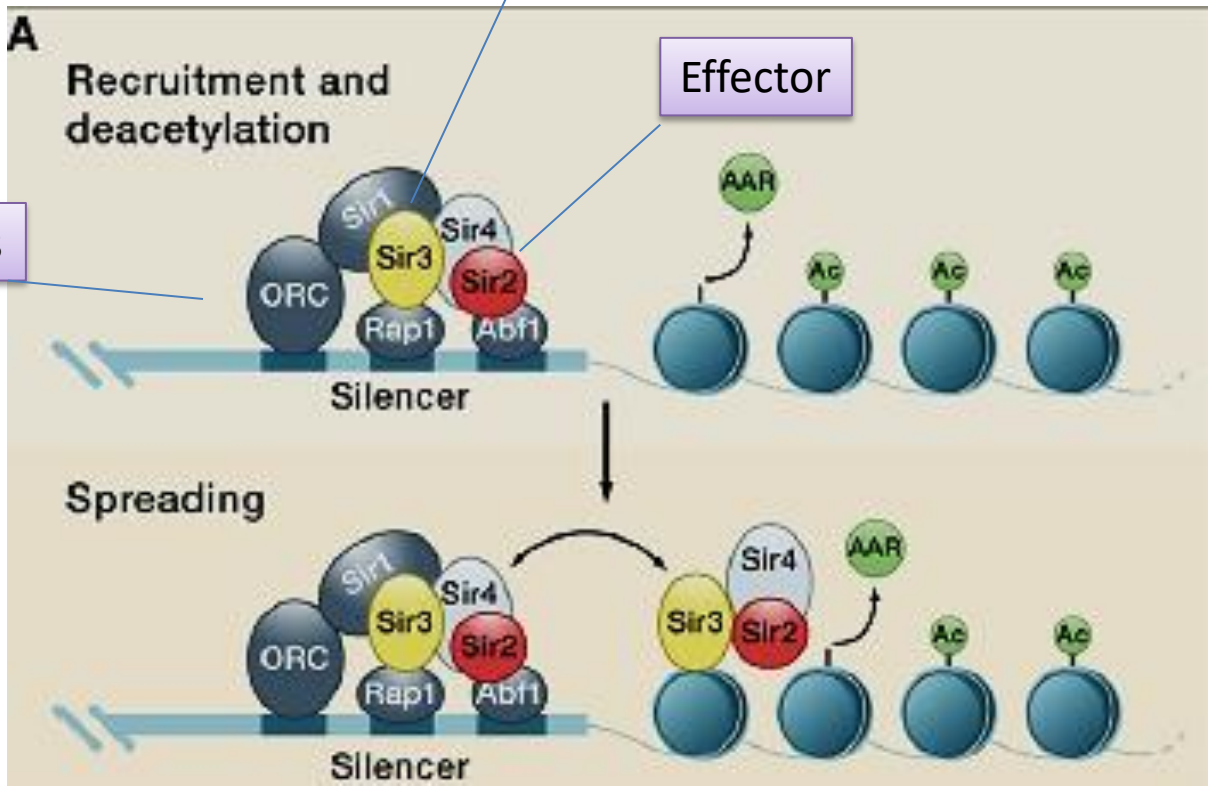
(Top) At the silent mating loci in S. cerevisiae, silencers (DNA regions composed of binding sites) for the origin recognition complex (ORC), Rap1, and Abf1 recruit the Sir1, Sir2, Sir3, and Sir4 proteins through multiple weak interactions. Sir2 uses NAD to deacetylate H4K16, releasing O-acetyl-ADP-ribose (AAR), which binds to one of the Sir proteins and induces a conformational change in the SIR complex that may result in a tighter interaction between Sir3 and Sir4, and Sir3 and the nucleosome.
(Bottom) H4K16 deacetylation promotes binding of Sir3, and sequential cycles of deacetylation and Sir3 binding to deacetylated nucleosomes are proposed to mediate the spreading of the SIR complex away from the silencer. The interaction of Sir3 with Sir4 is also required for spreading.



Bridging protein

Effector

Specificity factors



Silencer (*cis*-element)

Looking into these model systems, we can easily see:

- A DNA element that we can call «silencer» (talking heterochromatin)
- A molecule recognizing this DNA element, called «specificity factor»
- A bridging protein
- A chromatin competent enzyme

Mechanisms that we must understand:

1) How are different chromatin domain established ?
   - pre-existing epigenetic marks
   - DNA sequence
   - Stage- and tissue-specific Transcription Factors

programming

2) How are chromatin domains maintained ?
   - Epigenetic marks inheritance
   - Local spreading

maintenance

1) How can chromatin domains be re-programmed ?
   - Specific combination of Transcription Factors
   - Erasure of histone marks and CpG methylation

reprogramming

Suggested readings, for those of you who are development adepts…

REVIEW FOCUS ON EPIGENETIC DYNAMICS

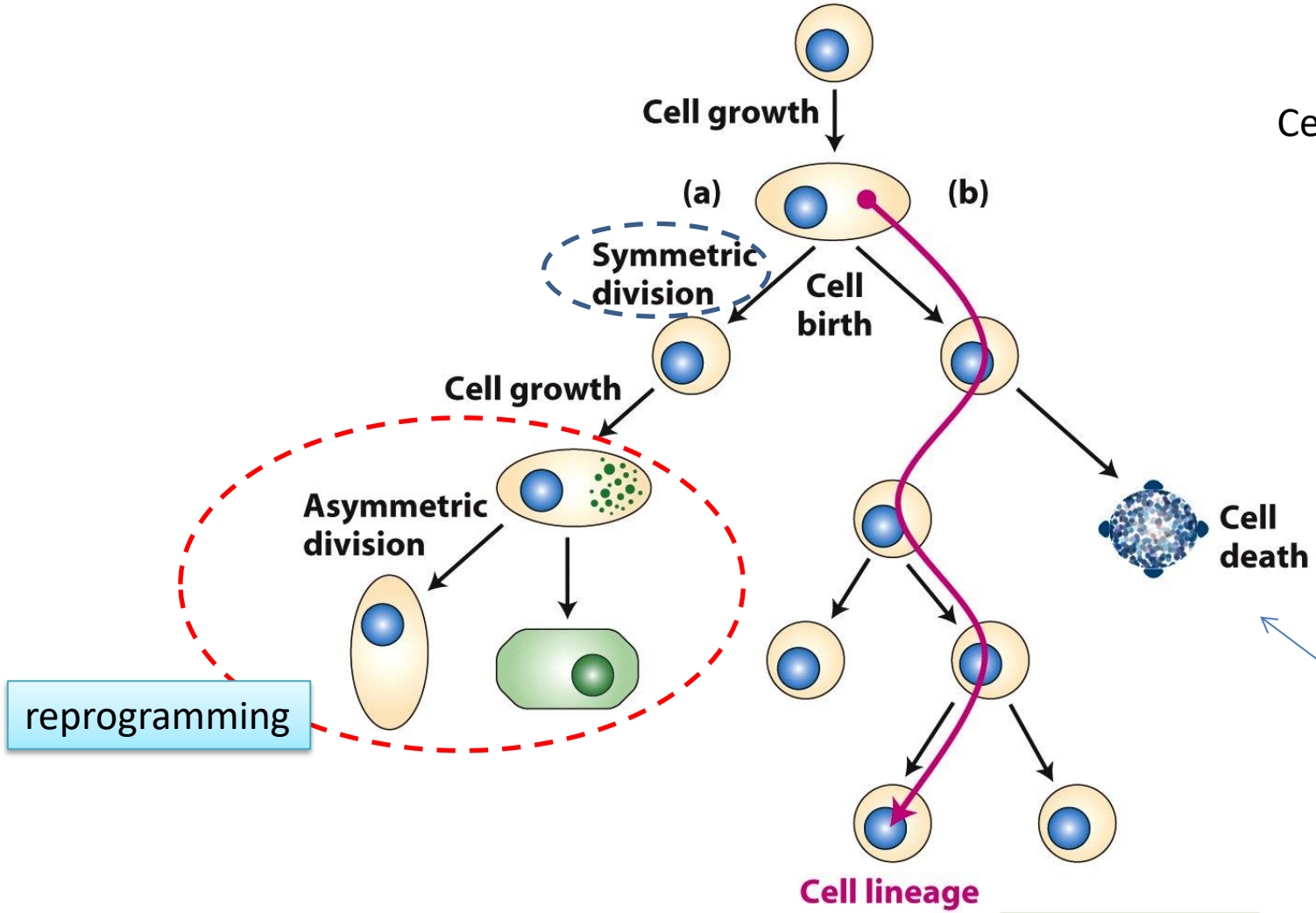
nature structural & molecular biology

Epigenetic programming and reprogramming during development

Irene Cantone & Amanda G Fisher

Cell identity is determined by specific gene expression patterns that are conveyed by interactions between transcription factors and DNA in the context of chromatin. In development, epigenetic modifiers are thought to stabilize gene expression and ensure that patterns of DNA methylation and histone modification are reinstated in cells as they divide. Global erasure of epigenetic marks occurs naturally at two stages in the mammalian life cycle, but it can also be artificially engineered using a variety of reprogramming strategies. Here we review some of the recent advances in understanding how epigenetic remodeling contributes to conversion of cell fate *in vivo* and *in vitro*. We summarize current models of epigenetic erasure and discuss the various enzymes and mechanisms that may operate in cellular reprogramming.

Cellular genealogy

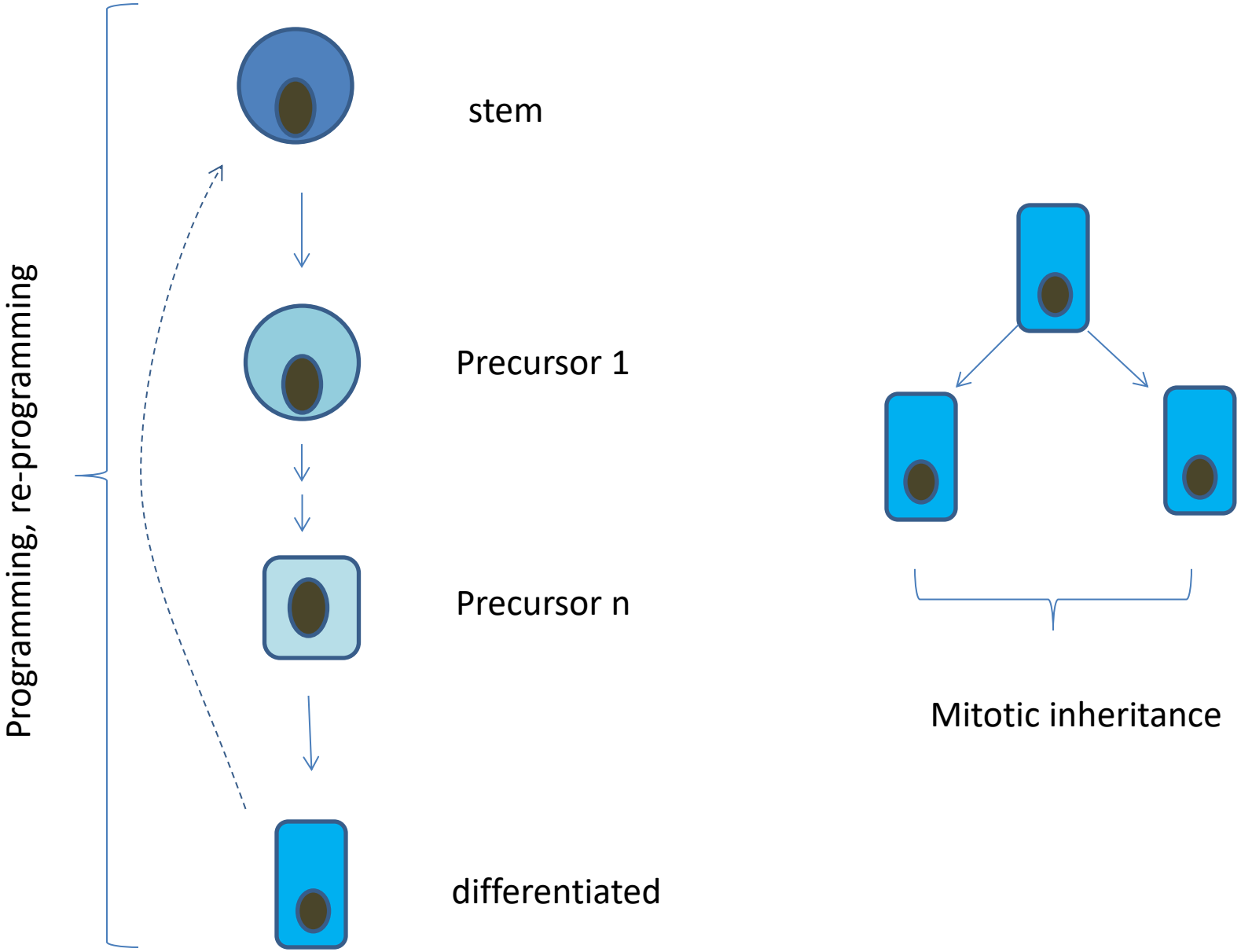


reprogramming

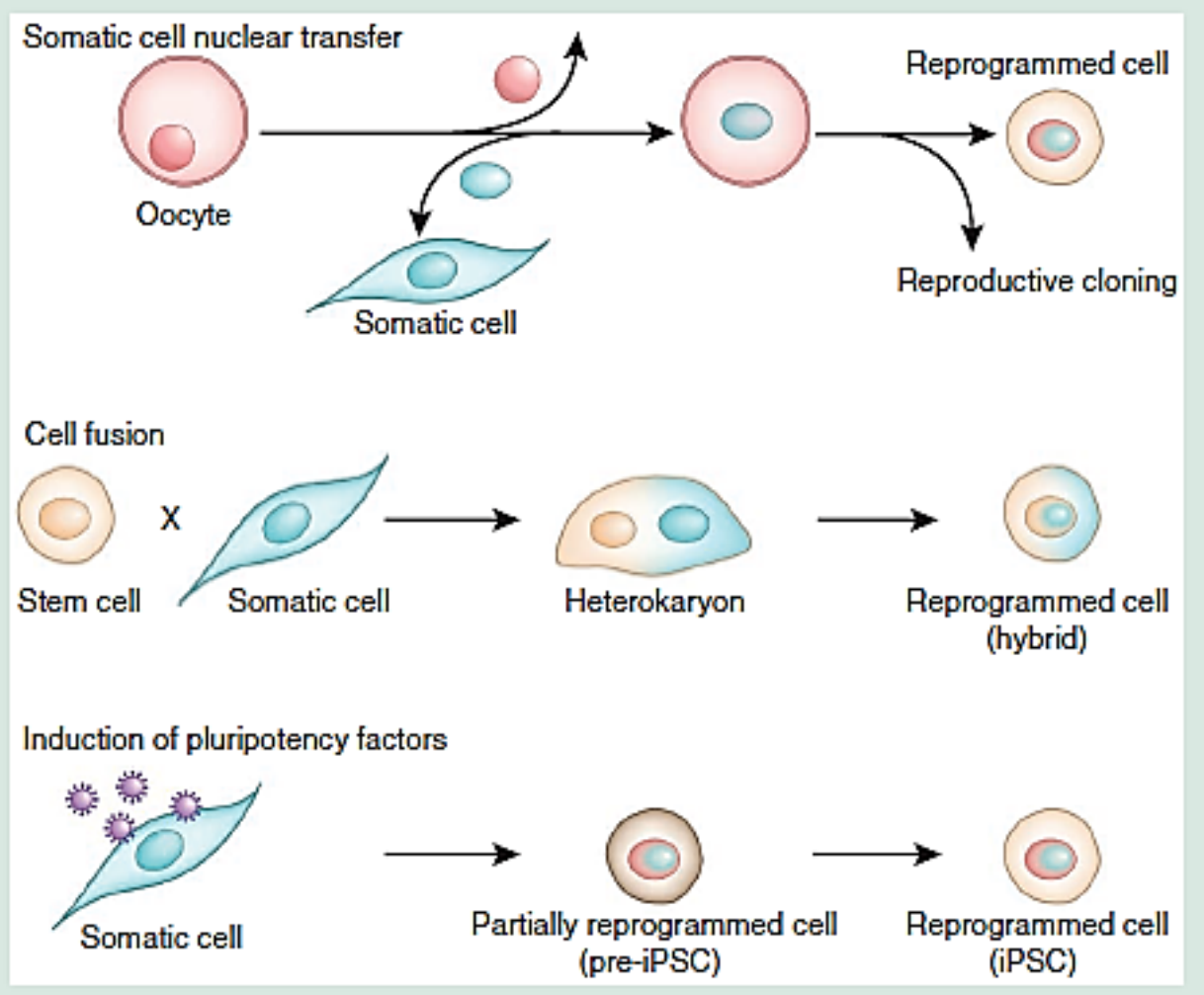
Apoptosis

maintenance

Figure 21-1
Molecular Cell Biology, Sixth Edition


Programming, re-programming
stem
Precursor 1
Precursor n
differentiated
Mitotic inheritance

Re-programming



Cantone & Fisher, 2013

Transcription factors (sequence-specific DNA binding proteins) can induce re-programming of chromatin domains and partial stemness

http://www.genecards.org/cgi-bin/carddisp.pl?gene=NPM1

ES cells

neuronal progenitor cells

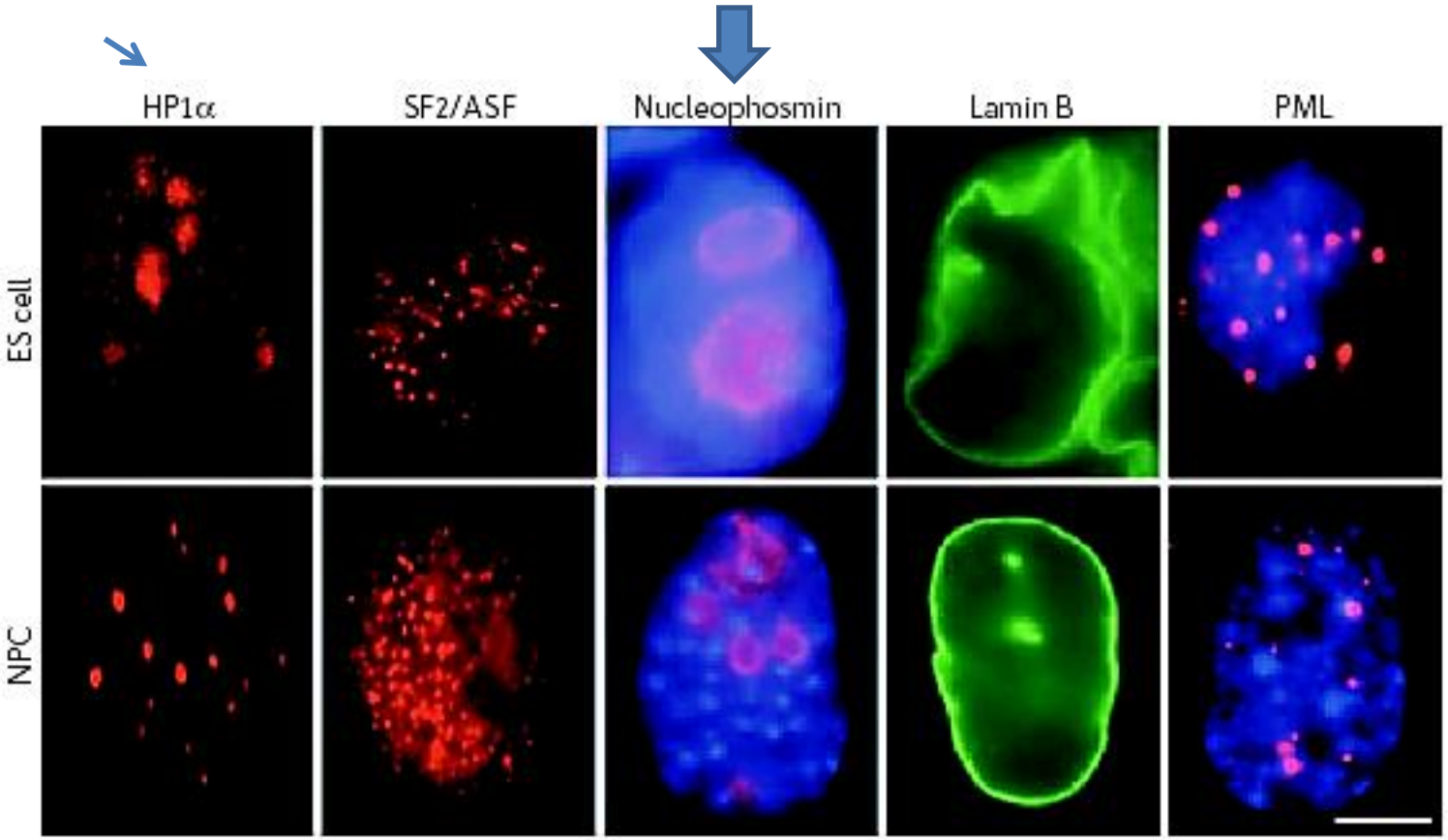


Figure 1 | **Nuclear architecture in ES cells and differentiating ES-derived cells.** Nuclear domains in an undifferentiated embryonic stem (ES) cell (top) and a differentiating ES-derived neuronal progenitor cell (NPC, bottom). From left to right: heterochromatin, as detected with an anti-HP1α antibody, is confined to fewer and larger foci in ES cells compared with NPCs; nuclear speckles, as detected with an anti-SF2/ASF antibody, appear as small, dispersed foci in ES cells and become more conspicuous in NPCs; nucleoli, as identified with an anti-nucleophosmin antibody, appear larger in ES cells compared with NPCs; the ill-defined nuclear lamina in ES cells, stained with an anti-lamin B antibody, becomes round and distinct in NPCs; promyelocytic leukaemia (PML) bodies labelled with an anti-PML antibody show similar patterns in ES cells and NPCs. DAPI, blue. Scale bar, 5 μm.

(from Meshorer & Misteli, 2006)

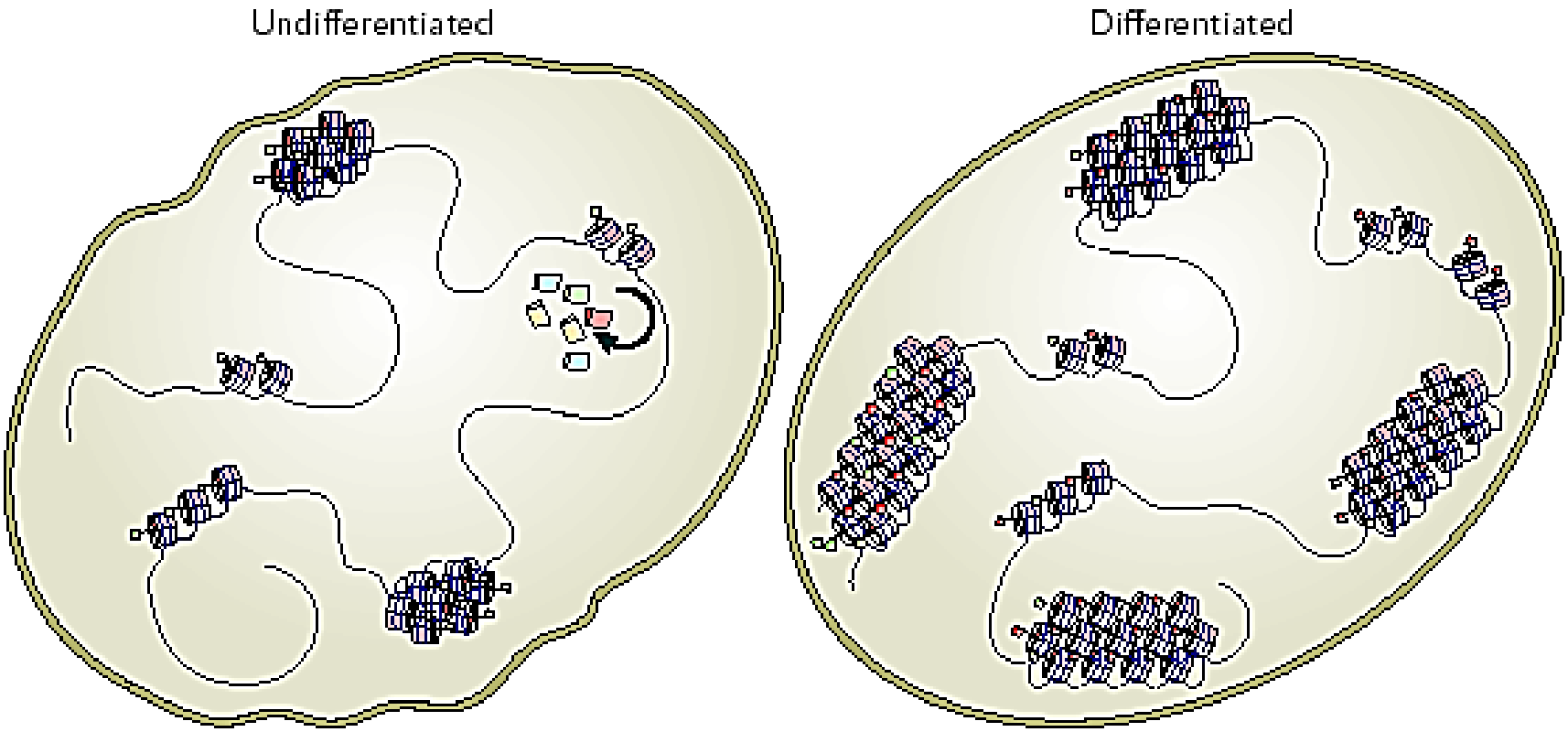
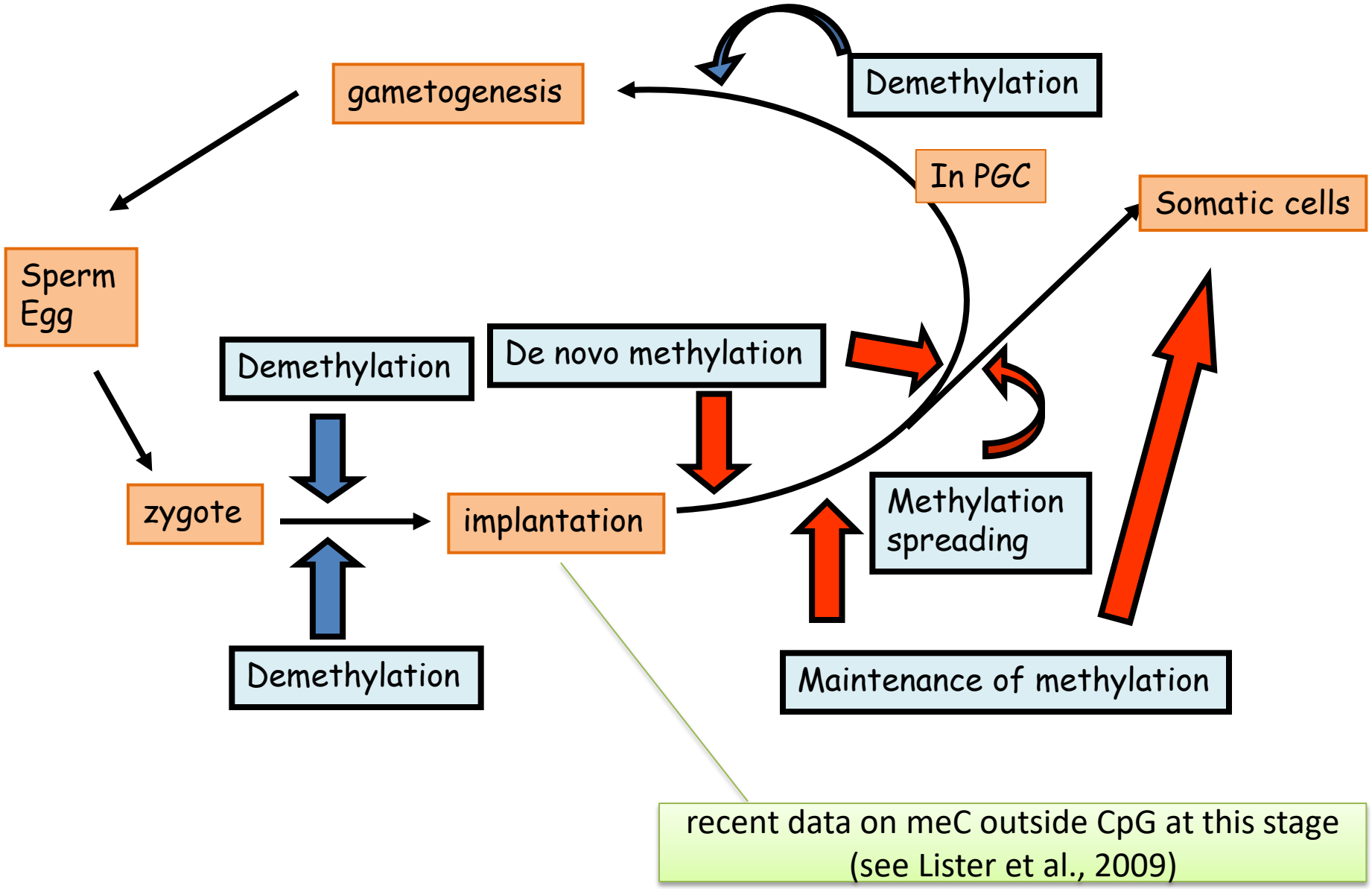
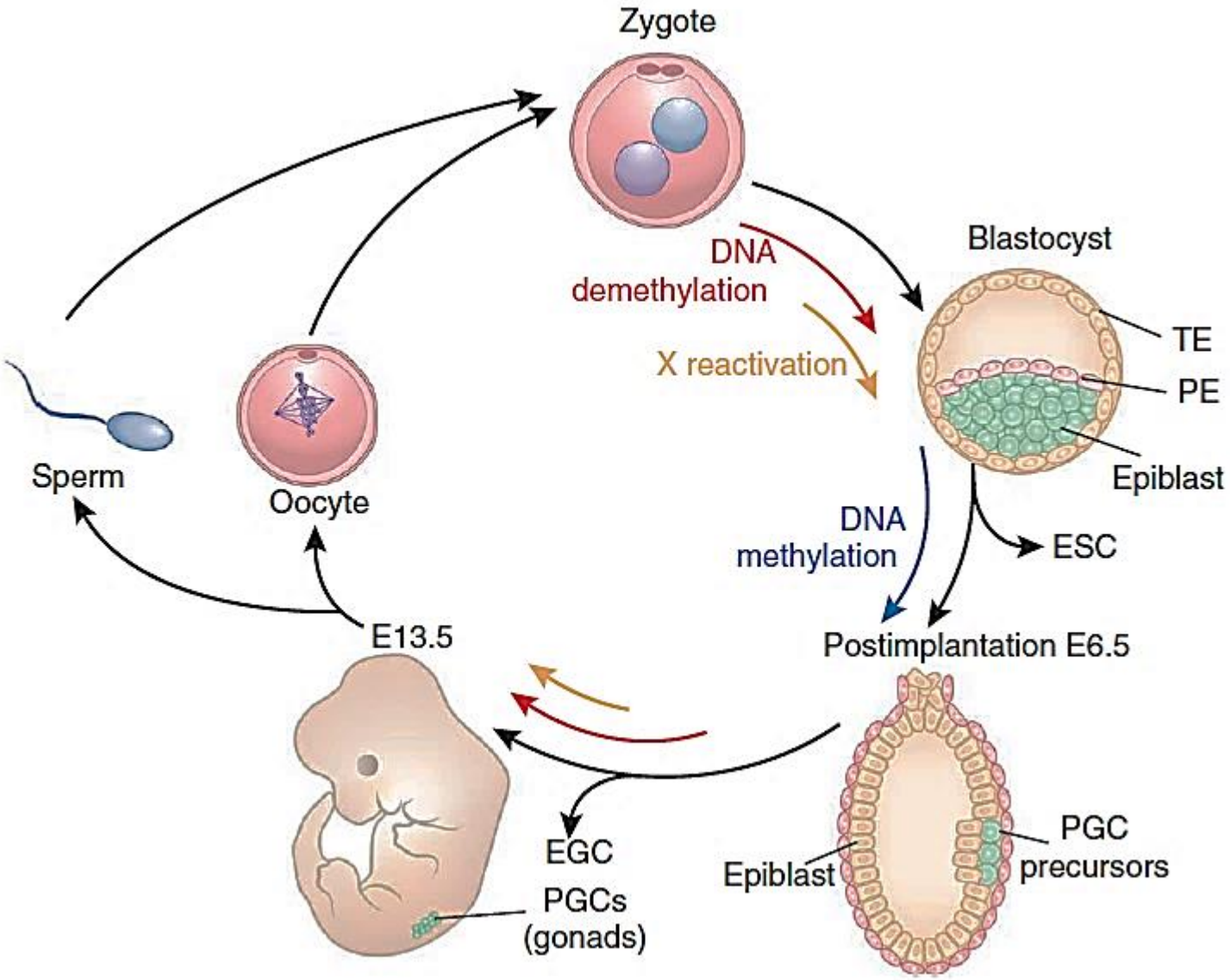


Figure 2 | **Chromatin during ES-cell differentiation.** In pluripotent embryonic stem (ES) cells (left), chromatin is globally decondensed, enriched in active histone marks (green circular tags), and contains a fraction of loosely bound architectural chromatin proteins. As cells differentiate (right), regions of condensed heterochromatin form, silencing histone marks (red circular tags) accumulate, and structural chromatin proteins become more stably associated with chromatin.

(from Meshorer & Misteli, 2006)

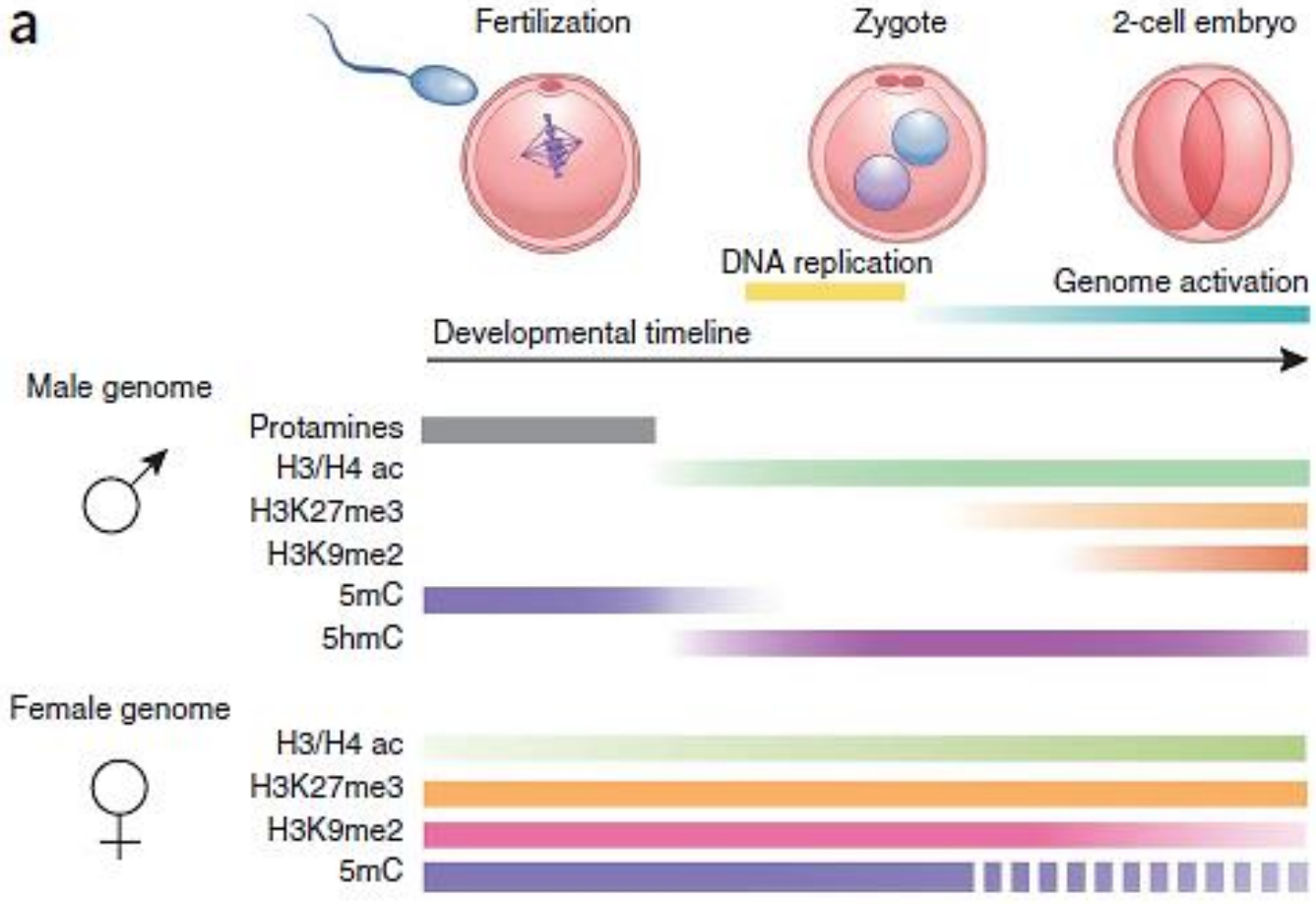
The life cycle of CpG methylation (raw)





Trophoectoderm
Primitive endoderm

Cantone & Fisher, 2013



Cantone & Fisher, 2013

Tet oxidation

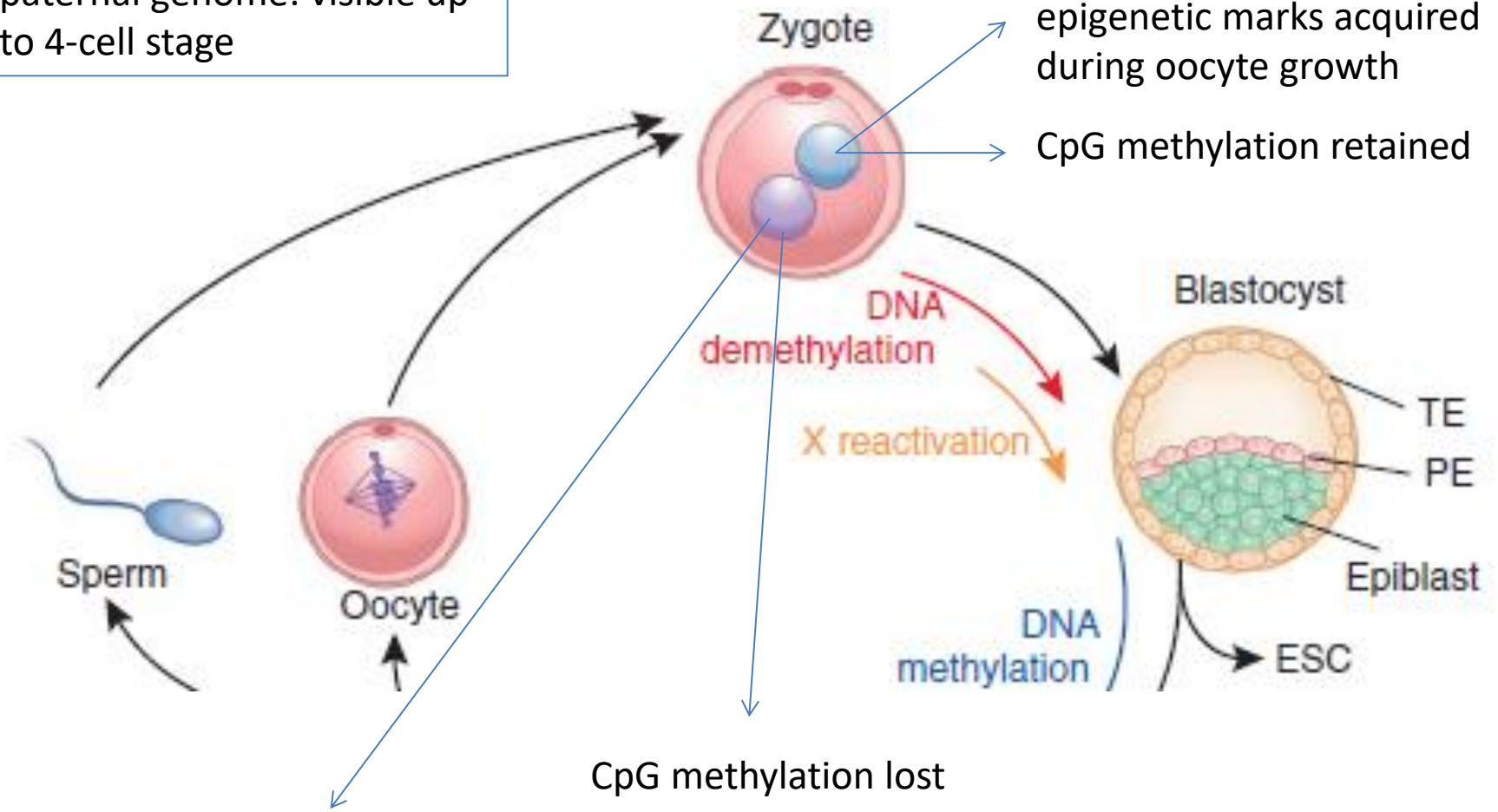
dilution

Figure 2 Epigenetic changes during *in vivo* reprogramming. (**a**) Schematic of global DNA and histone modifications that lead to transcriptional activation of the embryonic genome between the late zygote (paternal genome only) and the 2-cell stage. Gamete genomes undergo different epigenetic programs after fertilization with the paternal genome being mostly subject to epigenetic remodeling at the zygote stage and the maternal genome gradually losing repressive modifications during the subsequent cleavage divisions.

Asymmetry in maternal and paternal genome: visible up to 4-cell stage

Maternal genome: maintains epigenetic marks acquired during oocyte growth

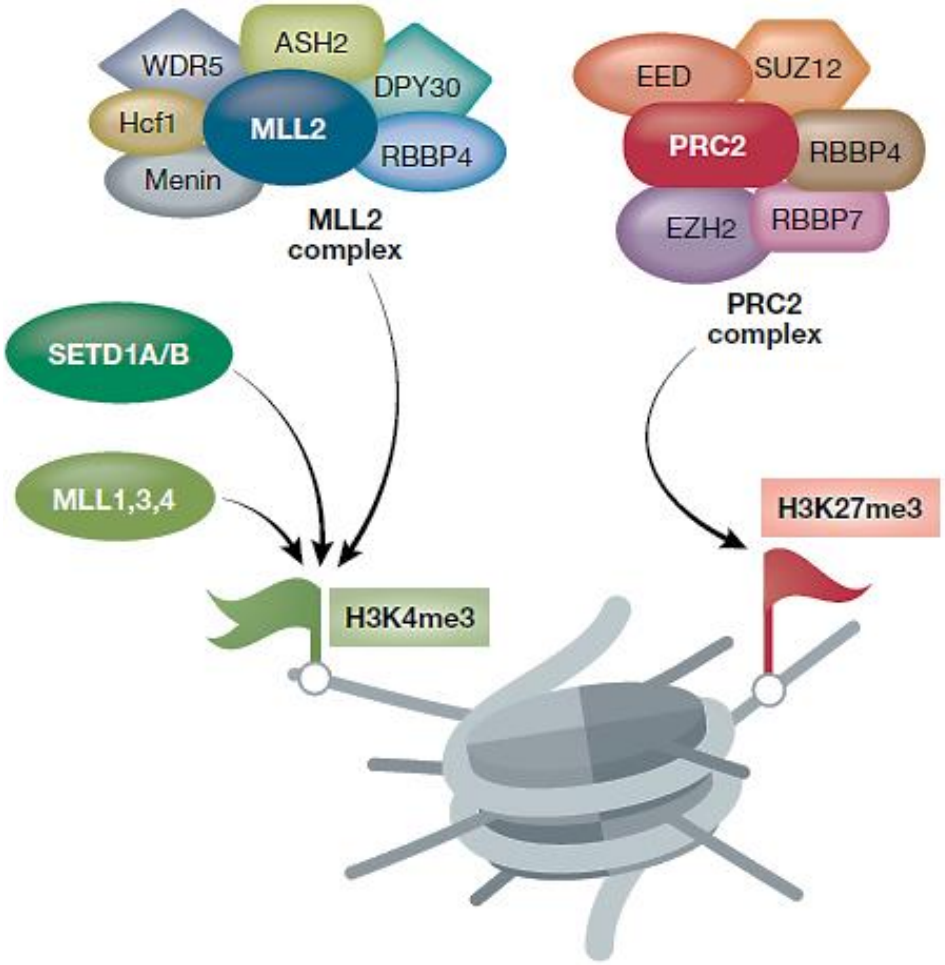
CpG methylation retained



CpG methylation lost

Paternal genome: exchanges protamines with new histones coming from maternal cytoplasm (H4K5ac + H4K12ac).

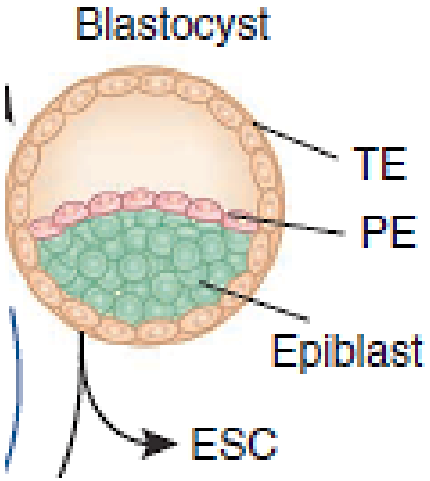
In **ESC** (but not only there) there are many «**bivalent**» chromatin domains. This kind of domain are also found in precursors and in general in cells that are not completely differentiated.



Drosophila nomenclature is Tritorax and Polycomb group proteins. In Human, they are called MLL2 complex and PCR2.

Tritorax (TrxG) and MLL2 complex are responsible of H3K4me3.
Polycomb group proteins (PcG) and PCR2 write H3K27me3.

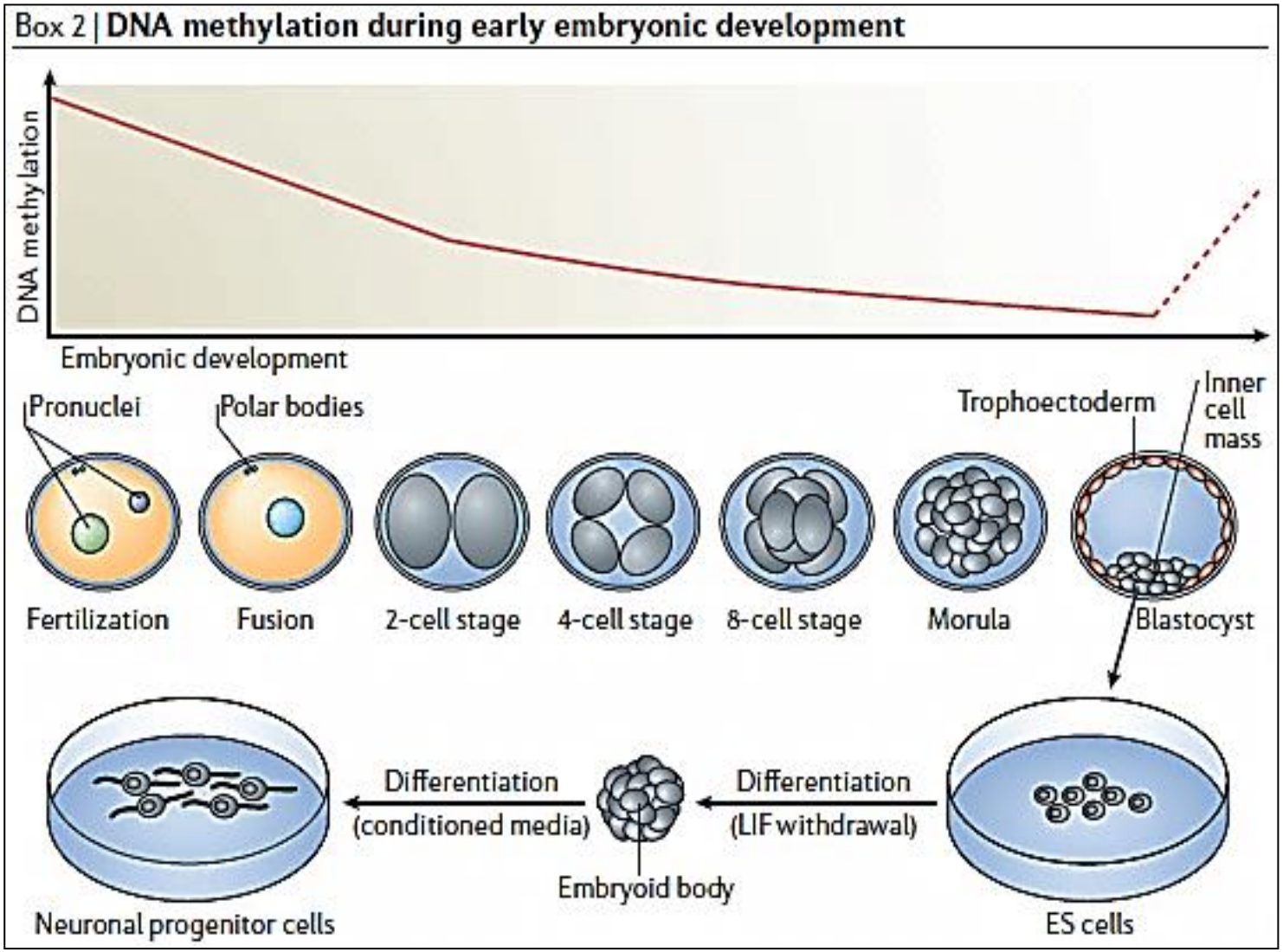
From Harikumar & Meshorer, 2015



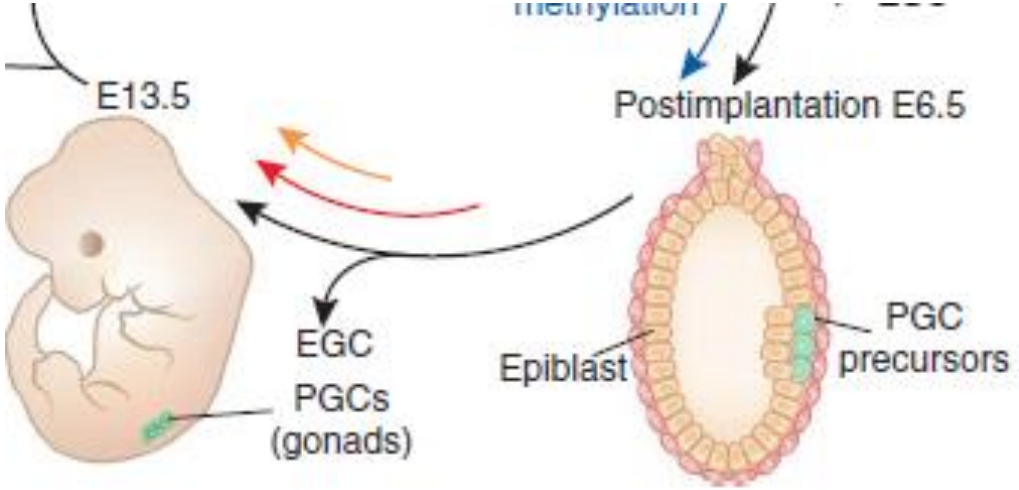
Embryonic stem cells

Since we can culture ES cells and differentiate in vitro, there are many studies on chromatin dynamics in this model.
Be aware that ESC represent an «in vitro» model system

TE= trophoectoderm
PE=primitive endoderm

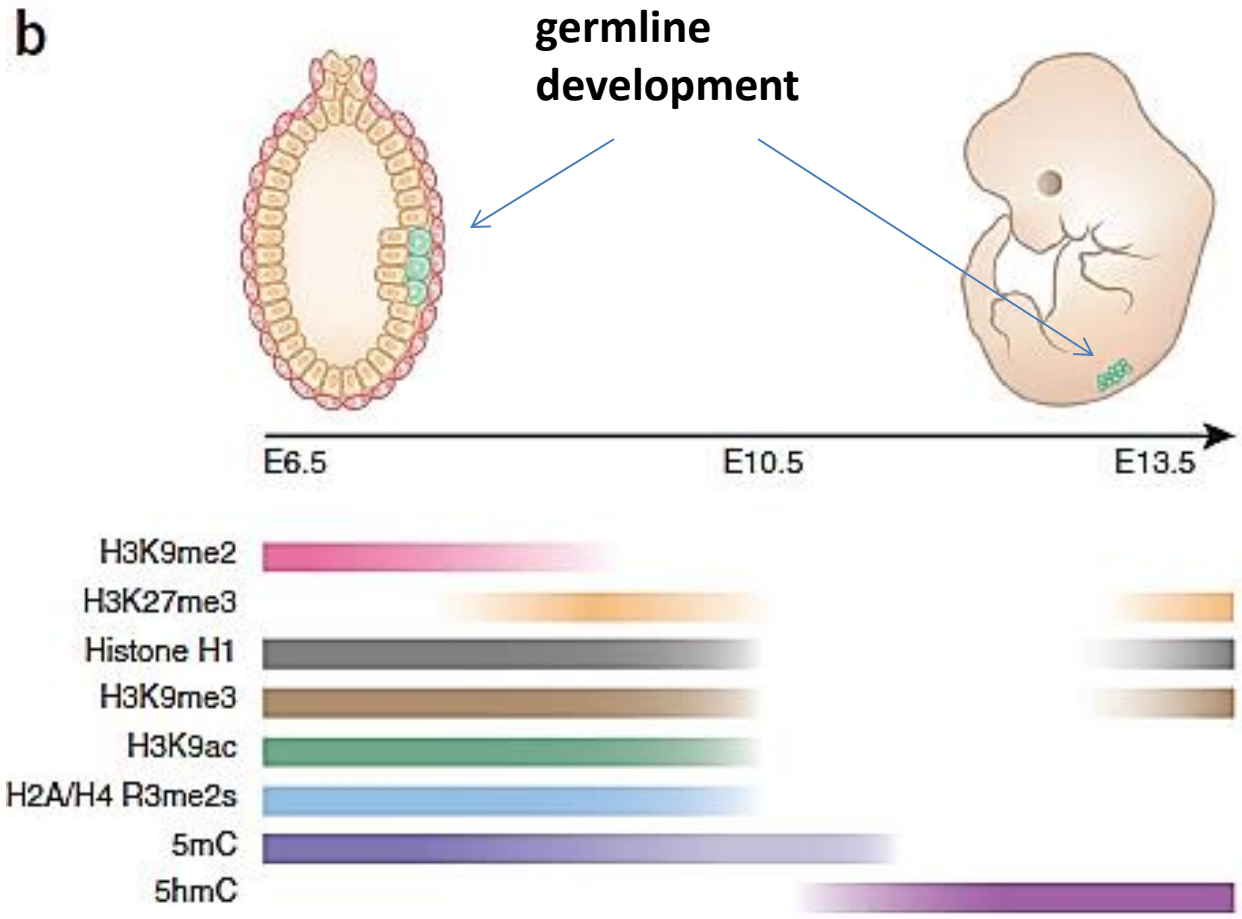

Box 2 | DNA methylation during early embryonic development
DNA methylation
Embryonic development
Pronuclei
Polar bodies
Trophoectoderm
Inner cell mass
Fertilization
Fusion
2-cell stage
4-cell stage
8-cell stage
Morula
Blastocyst
Differentiation (conditioned media)
Differentiation (LIF withdrawal)
Embryoid body
Neuronal progenitor cells
ES cells

Re-programming



Primordial Germ Cell (PGC) precursors will start to be re-programmed around E6.5 AT this stage they **are already marked as somatic**, but as soon as they migrate to destination they **start loosing H3K9me2 and acquiring H3K27me3**.

Note that when PGC enter the gonads (E11.5-E12.5) there is rapid and extensive **CpG demethylation** reaching complete at day E13.5.



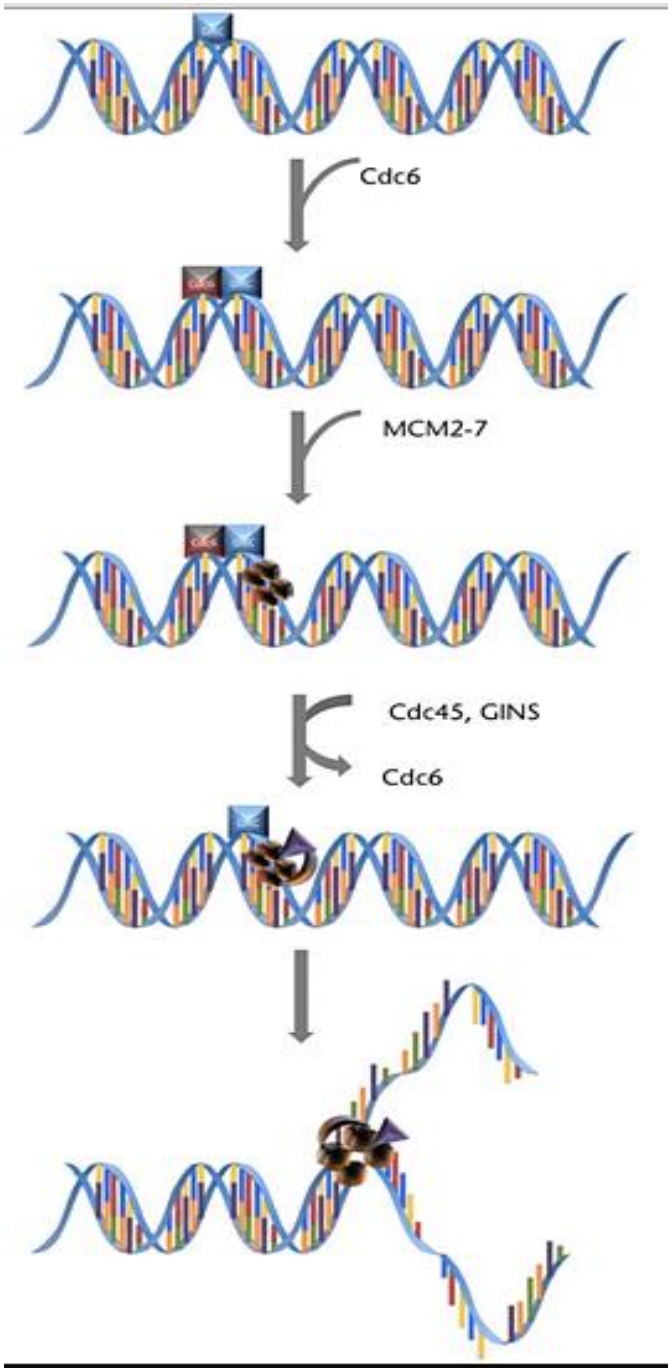
Cantone & Fisher, 2013

Tet enzymes

Figure 2 Epigenetic changes during *in vivo* reprogramming

(**b**) Global epigenetic changes during **germline development** from PGC specification (E6.5) to the mitotic/meiotic arrest at E13.5. Two major reprogramming phases can be distinguished during PGC migration toward the genital ridges (E7.5–E10.5) and upon their arrival into the gonads (E10.5–E12.5).

Maintenance
or
Mitotic epigenetic inheritance



DNA Replication

Mammalian origin recognition

From Smith et al., 2014

DNA Replication

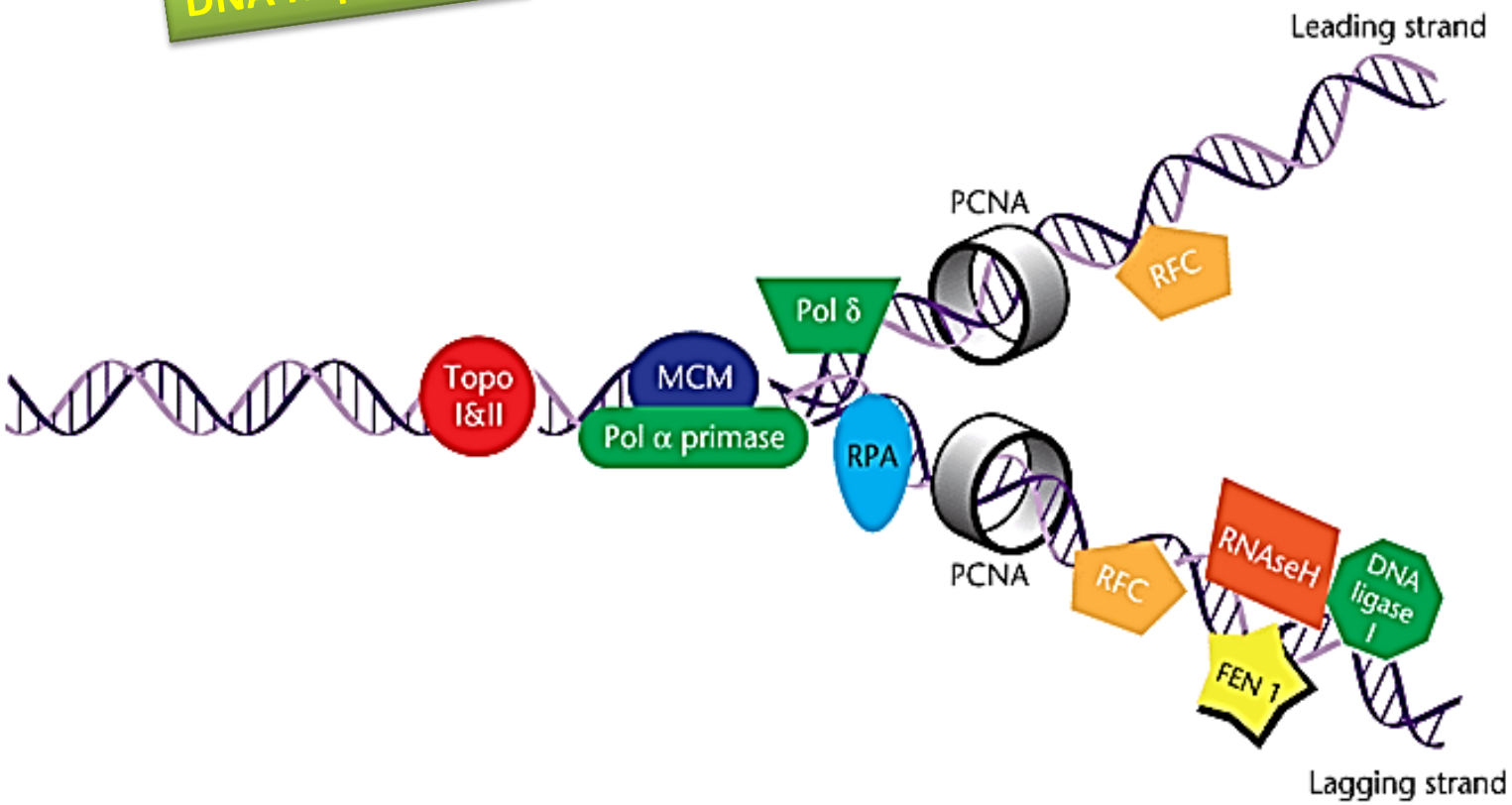
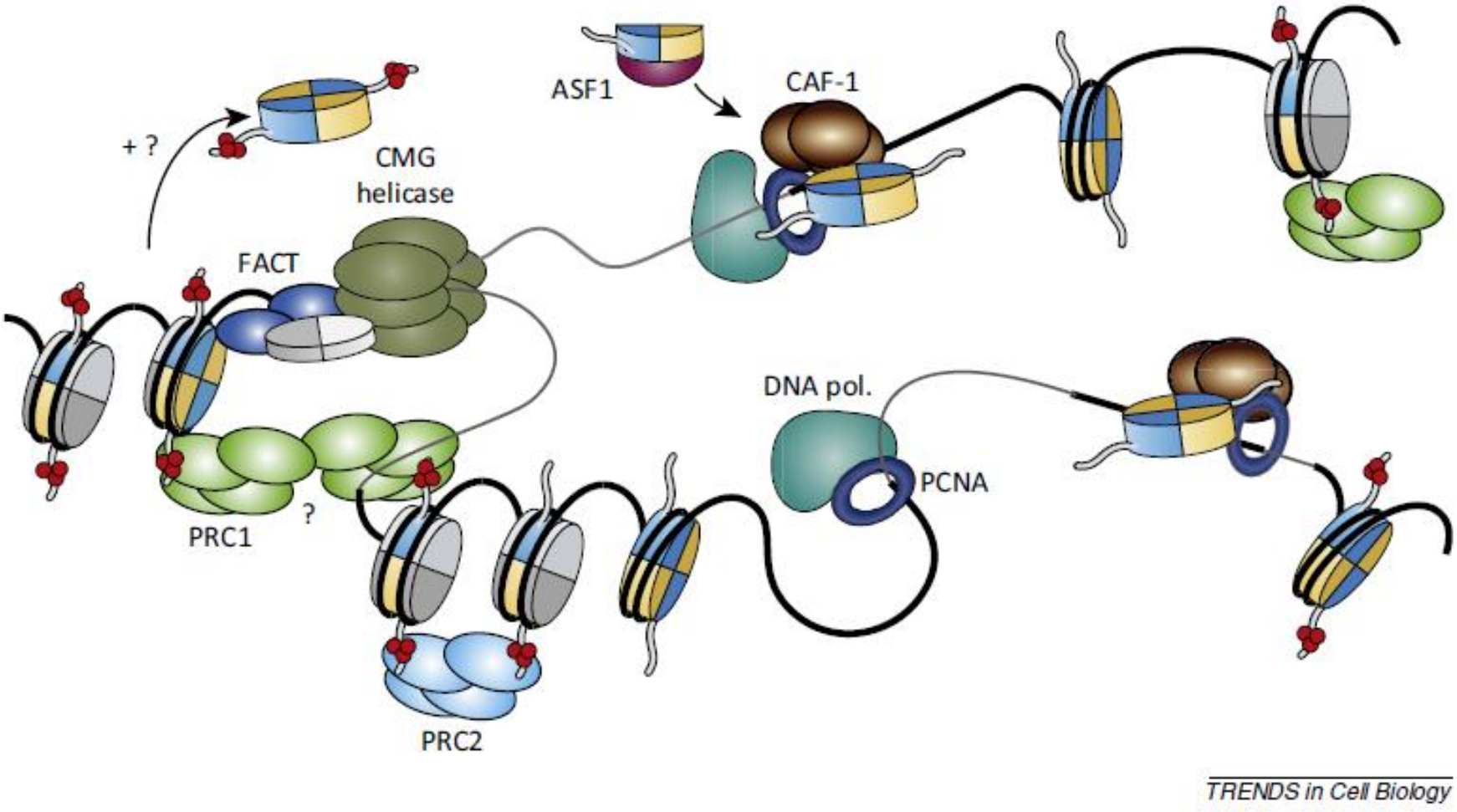


Figure 2.
Leading and lagging strand synthesis of mammalian DNA at the replication fork. After unwinding of the parental DNA, replication protein A (RPA) binds, stabilizing the DNA pol α-primase complex. Following primer synthesis, replicating factor C (RFC) loads proliferating cell nuclear antigen (PCNA) onto the leading strand. PCNA then acts as a scaffold to load polymerase δ (pol δ), continuing synthesis in the 5′→3′ direction. On the lagging strand, pol α-primase creates Okazaki fragments, which are extended by pol δ. When these fragments converge, a single-stranded flap is formed. This flap is then cleaved by flap endonuclease 1 (FEN1) and ribonucleic acidase (RNAse H). The resulting nick is sealed by DNA ligase 1.

Smith, SJ, Li, CM, Hickey, RJ, and Malkas, H(Nov 2014) DNA Replication: Mammalian. In: eLS. John Wiley & Sons Ltd, Chichester. http://www.els.net [doi: 10.1002/9780470015902.a0001041.pub3]

Your textbook, Figure 1

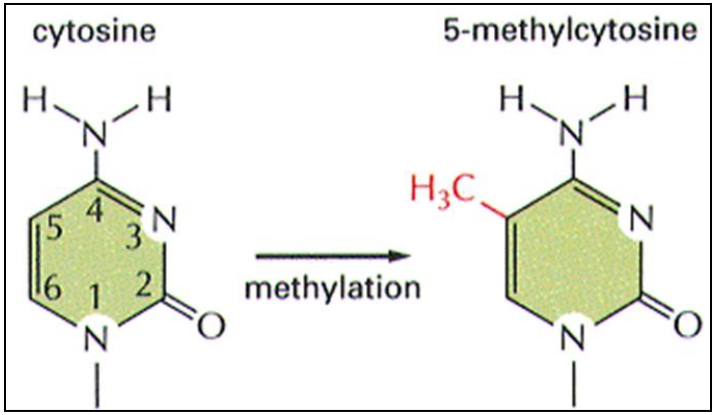


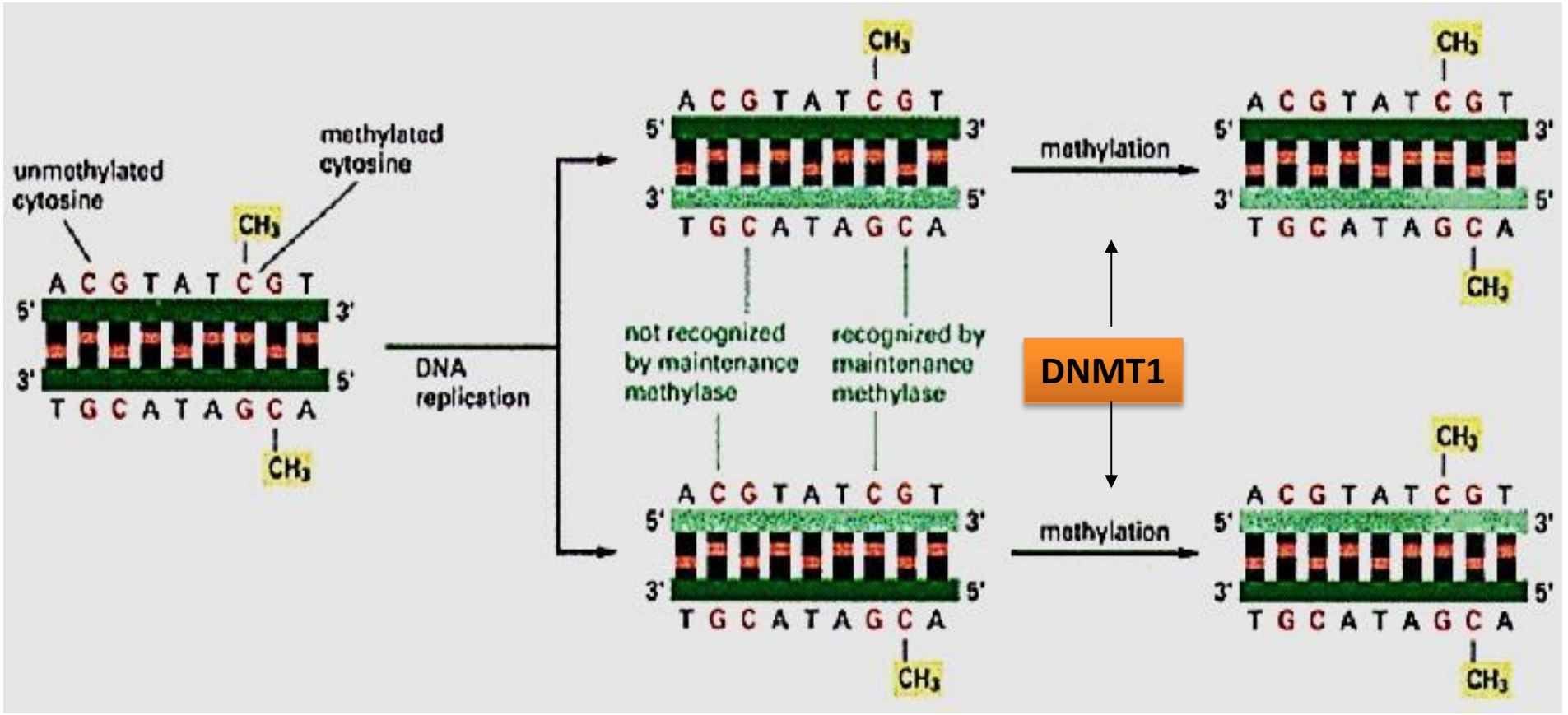
Your textbook, 2nd paragraph

Dismantling and restoring chromatin throughout DNA replication

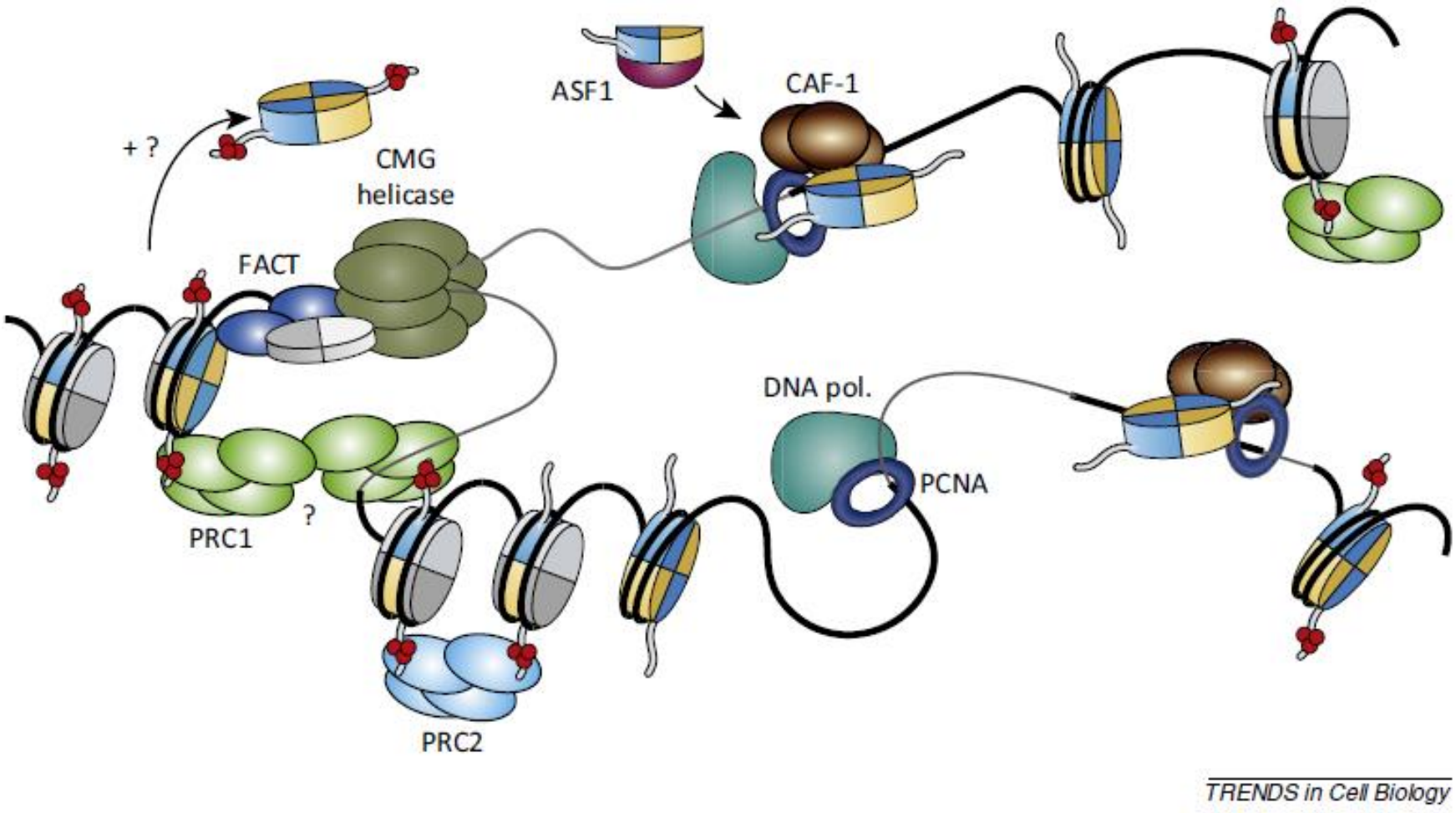
The post-replicative restoration of DNA methylation on the newly synthesized DNA via the maintenance DNA methyltransferase, DNMT1, is perhaps one of the better-understood examples of epigenetic inheritance (recently reviewed elsewhere [3]). By contrast, other epigenetic

DNA CpG methylation is propagated at cell division using a very simple mechanism:
DNMT1 is a methylation-dependent cytosine methyl transferase.

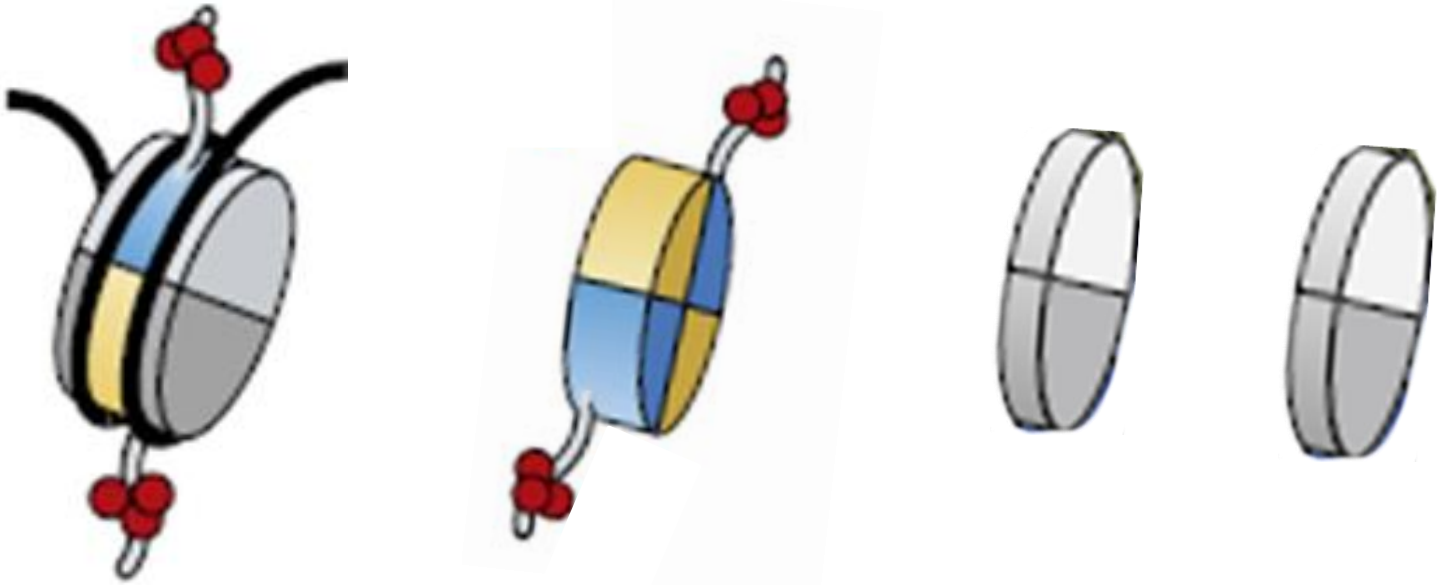




considering nucleosomes ….



Nucleosome octamer dissociation at the replication fork



Octamer

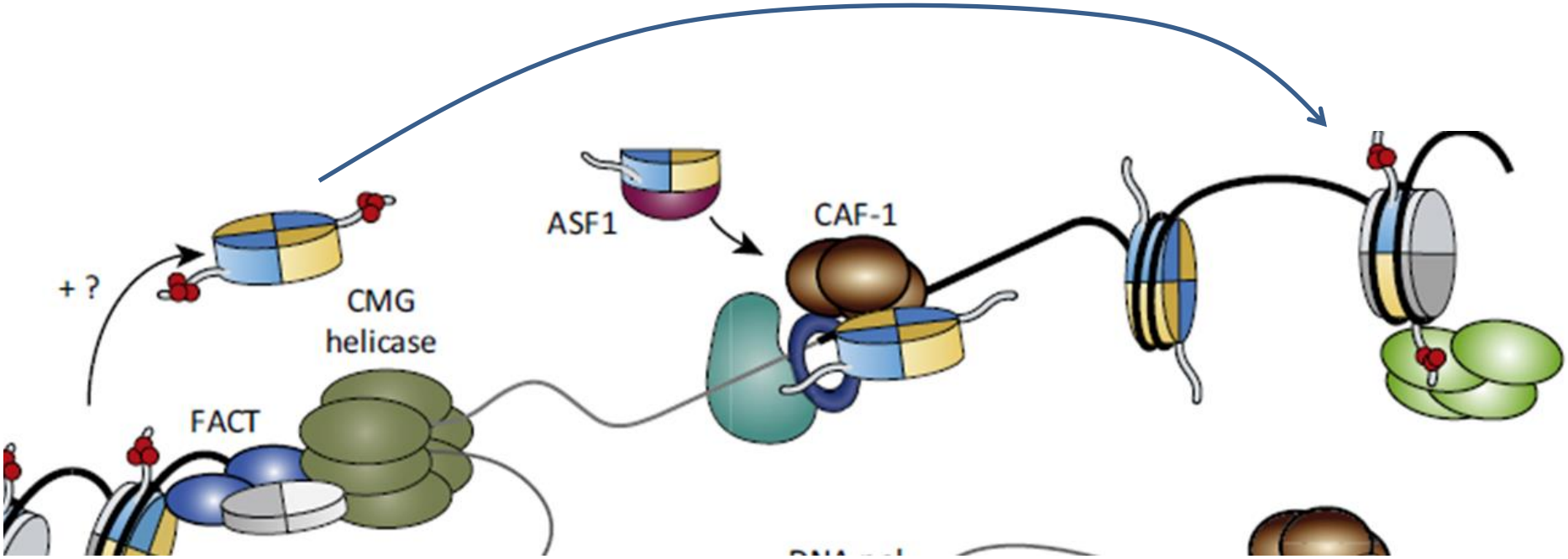
H3-H4 tetramer

H2A-H2B dimers

Histome PTNs are maintained in dissociated histones

H3-H4 tetramer immediately reassembles after the replication fork has passed, followed by 2x H2A-H2B dimers addition .

Histones **redistribute equally** to the daughter strands, so that new histones should be synthesized and incorporated. H3-H4 dimers arrive carried by ASF, then CAF-1 chaperones tetramer formation.



H3.1 and H3.2 are the replication-dependent H3 isoforms, whereas H3.3 is incorporated post-replicationally.

ASF1 (anti-silencing factor 1) is the carrier for all isoforms, though.

ASF-1 interacts also with

- RFC replicative clamp loader (clamp is PCNA, the ring in figures)
- MCM subunits of replicative Helicase (CMG)

Two competing models for H3-H4 redistribution:

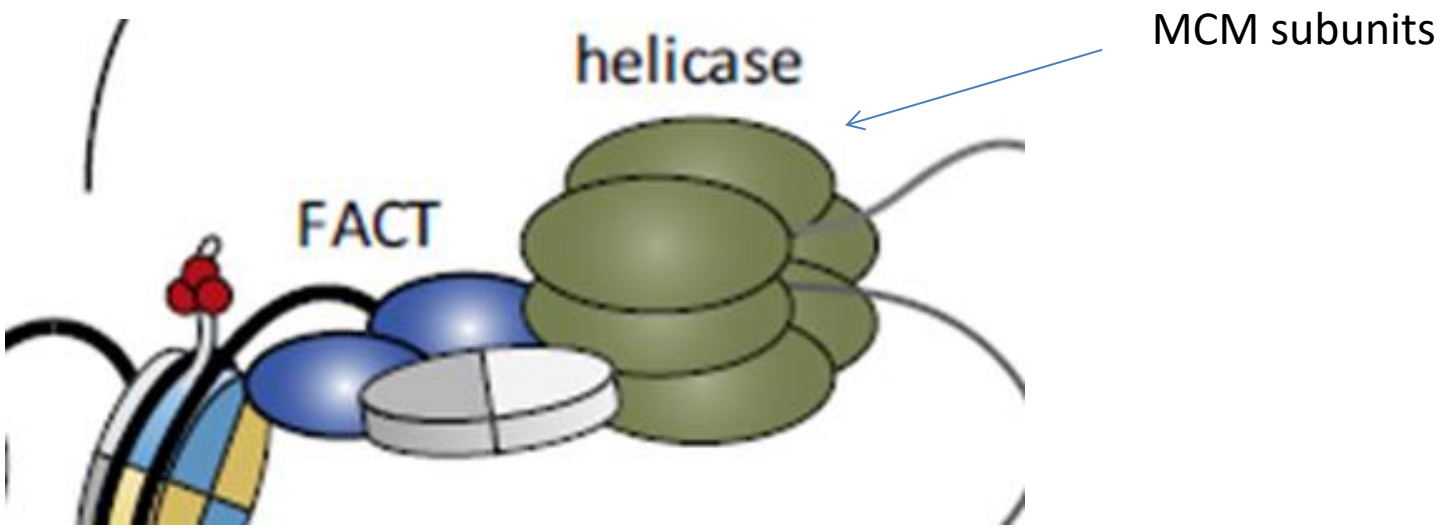
Splitting model:

ASF1 may split H3-H4 tetramer in two and distribute equally

Random tetramer model:

ASF1 distribute randomly tetramers

The **latter** is favoured today, since PTMs are non symmetrical in the two half-tetramers, whereas the PTMs are indeed conserved equally after replication in the two daughter chromatin molecules.



FACT is histone chaperone: it interacts mainly with H2A-H2B dimers and dissociates them from the core. MCM also interacts with H2A-H2B.

MCM mutants unable to interact with H2A-H2B show defects in telomeric heterochromatin.

Thus, this interaction possibly plays a role in correctly passing on PTMs-containing histones equally to daughter cromatin molecules.

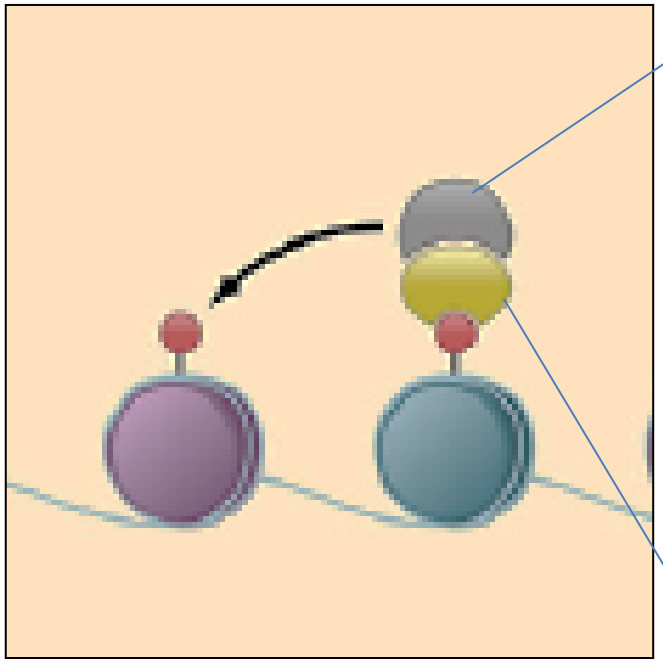
Of course, this kind of mechanism is fully compatible with the observation that chromatin domains are quite conservatively inherited by the two daughter cells after mitosis.

Copying mechanisms rely on complex Writers / Readers / / Eraser systems

as examplified in the case of PCR2 and HP1-Suv39H1/2 complexes

maintenance

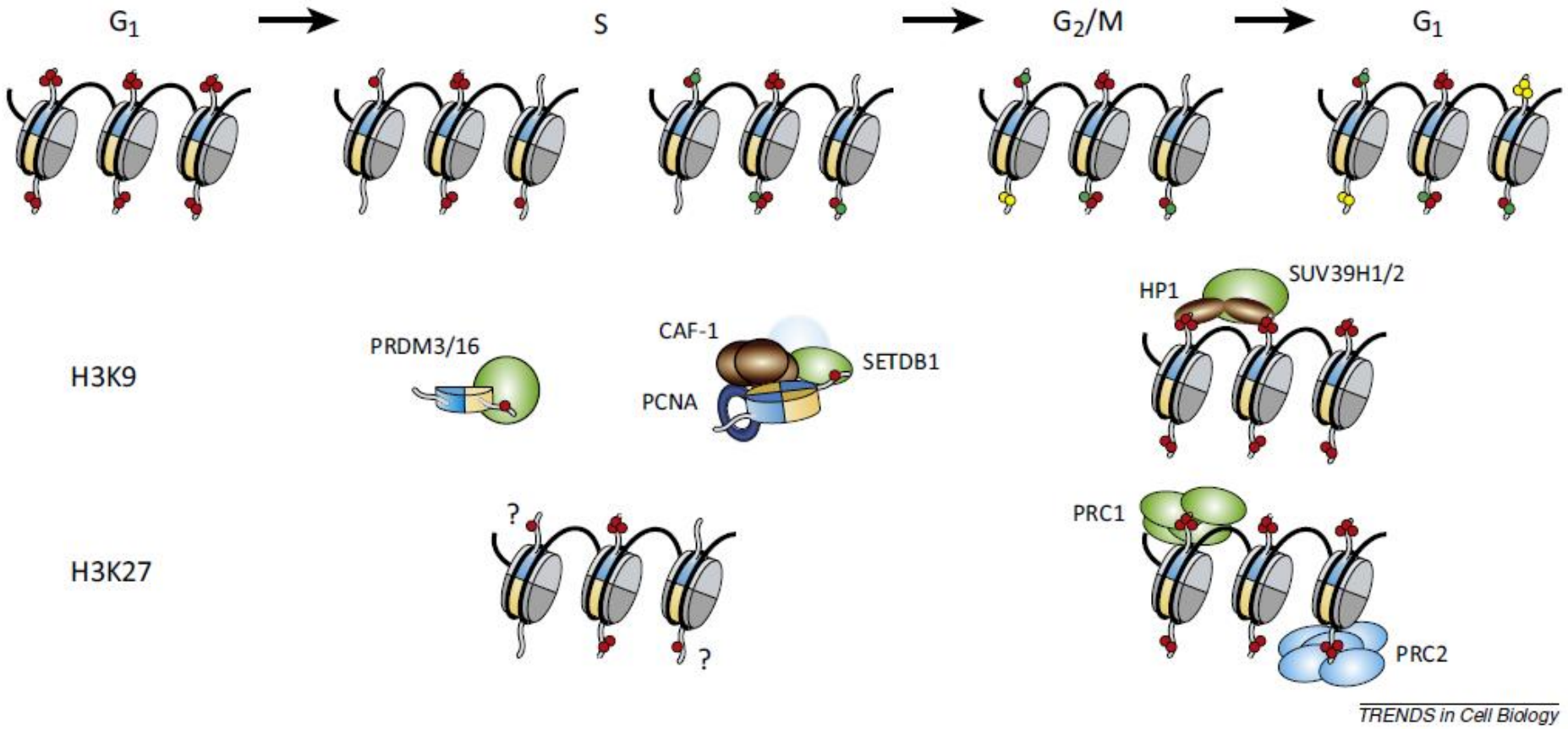
The R/W/E complex model



PTM enzyme (W)

PTM-binding protein (R)

Moazed 2011, Cell, 146:510-8


G1
S
G2/M
G1
H3K9
PRDM3/16
CAF-1
SETDB1
PCNA
HP1
SUV39H1/2
H3K27
?
?
PRC1
PRC2
TRENDS in Cell Biology

first

The **HP1 – Suv39H1 – HDAC1** complex in **fission yeast**

Swi6 (HP1) possesses a chromodomain that is a «reader» of H3K9me2/3

Swi6 (HP1) interacts with Suv39H1

Suv39H1 and Suv39H2 are enzymes that methylate H3K9

Swi6 (HP1) also interacts with HDAC1 or SIRT1

The same in Mammals
with different names and components

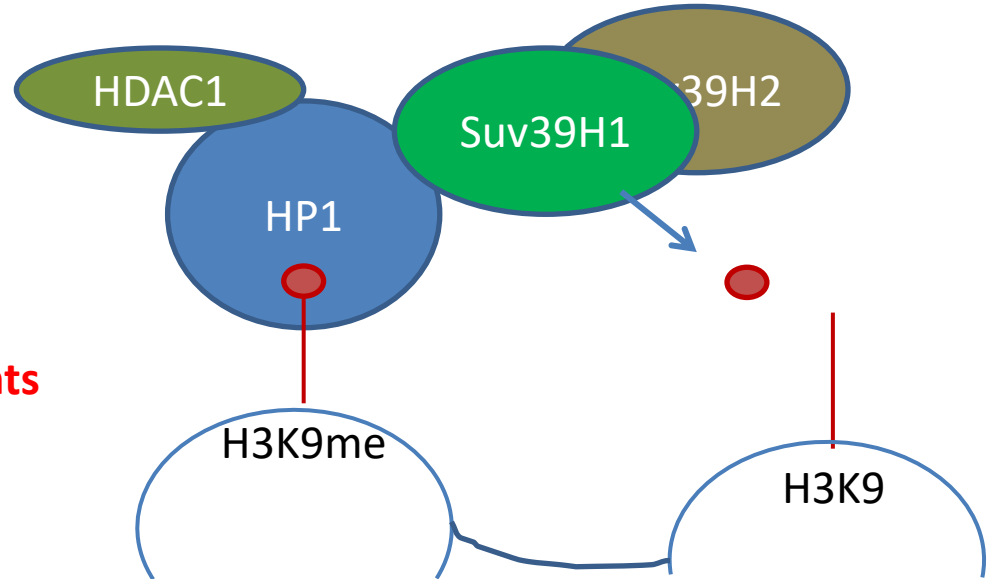
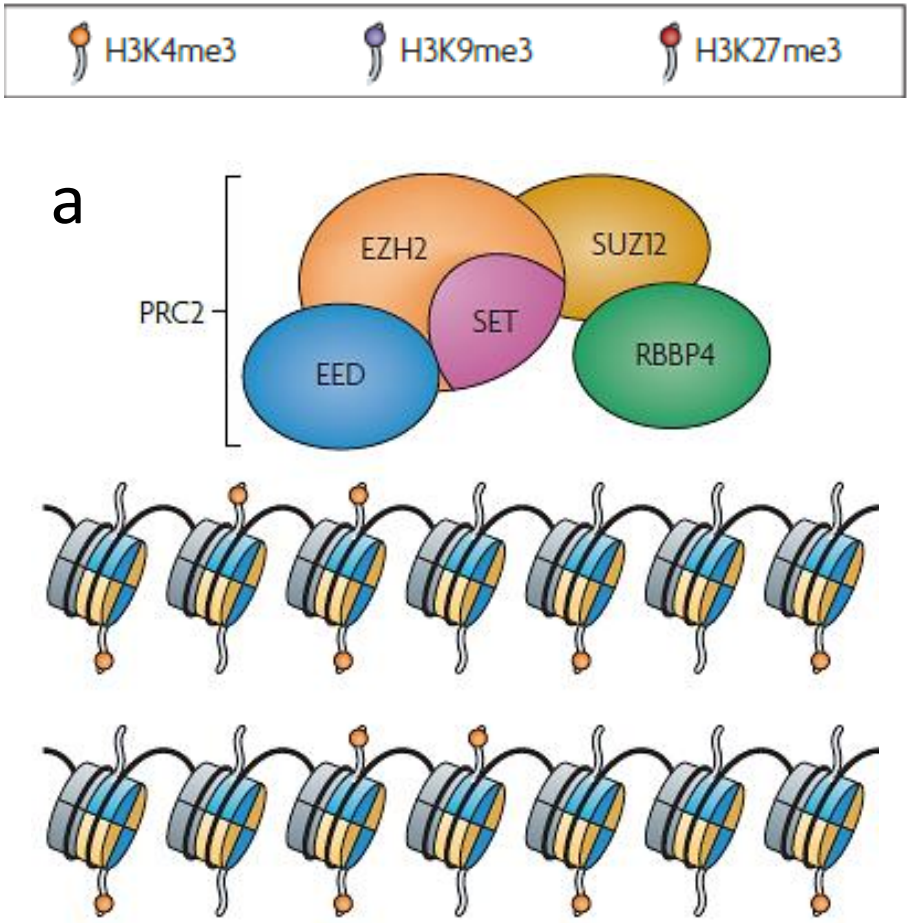


Figure 4 | **Propagation of histone 3 lysine 27 trimethylation by polycomb repressive complex 2.**
This scheme shows how pre-existing histone methylation marks regulate the polycomb repressive complex 2 (PRC2)-mediated spread of histone 3 lysine 27 methylation (H3K27me). For simplicity, only one type of histone methylation is presented for each domain, although *in vivo* there might be combination of these marks. Importantly, this scheme does not consider the recruitment of PRC2.
The components of PRC2 are indicated. Three examples are envisioned.

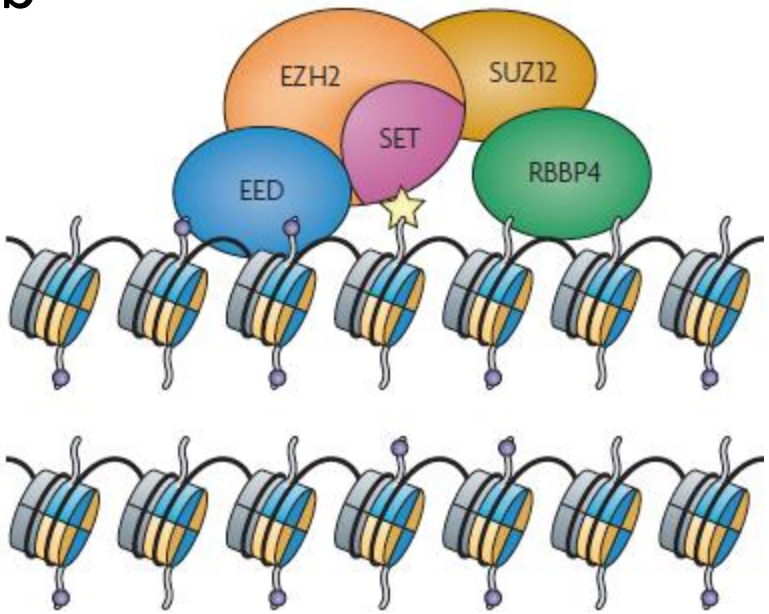
a A chromatin domain is enriched for an 'active mark' — such as H3K4 trimethylation (H3K4me3) — that is not recognized by PRC2 and therefore H3K27 is **not** methylated.



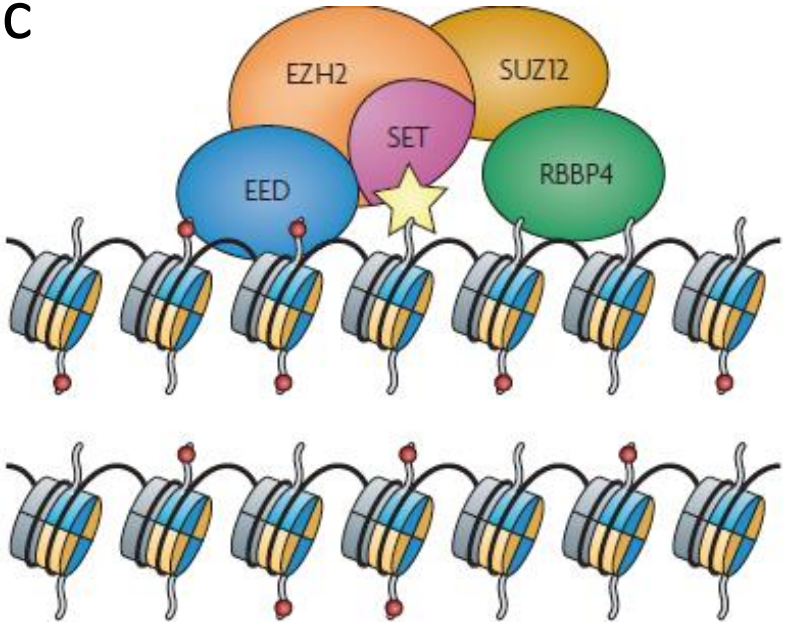
EED, embryonic ectoderm development;
EZH2, enhancer of zeste homologue 2;
RBBP4, retinoblastoma binding protein 4;
SUZ12, suppressor of zeste 12.

Margueron & Reinberg, Nature Rev Genet. 2010, 11:285-98

b



c



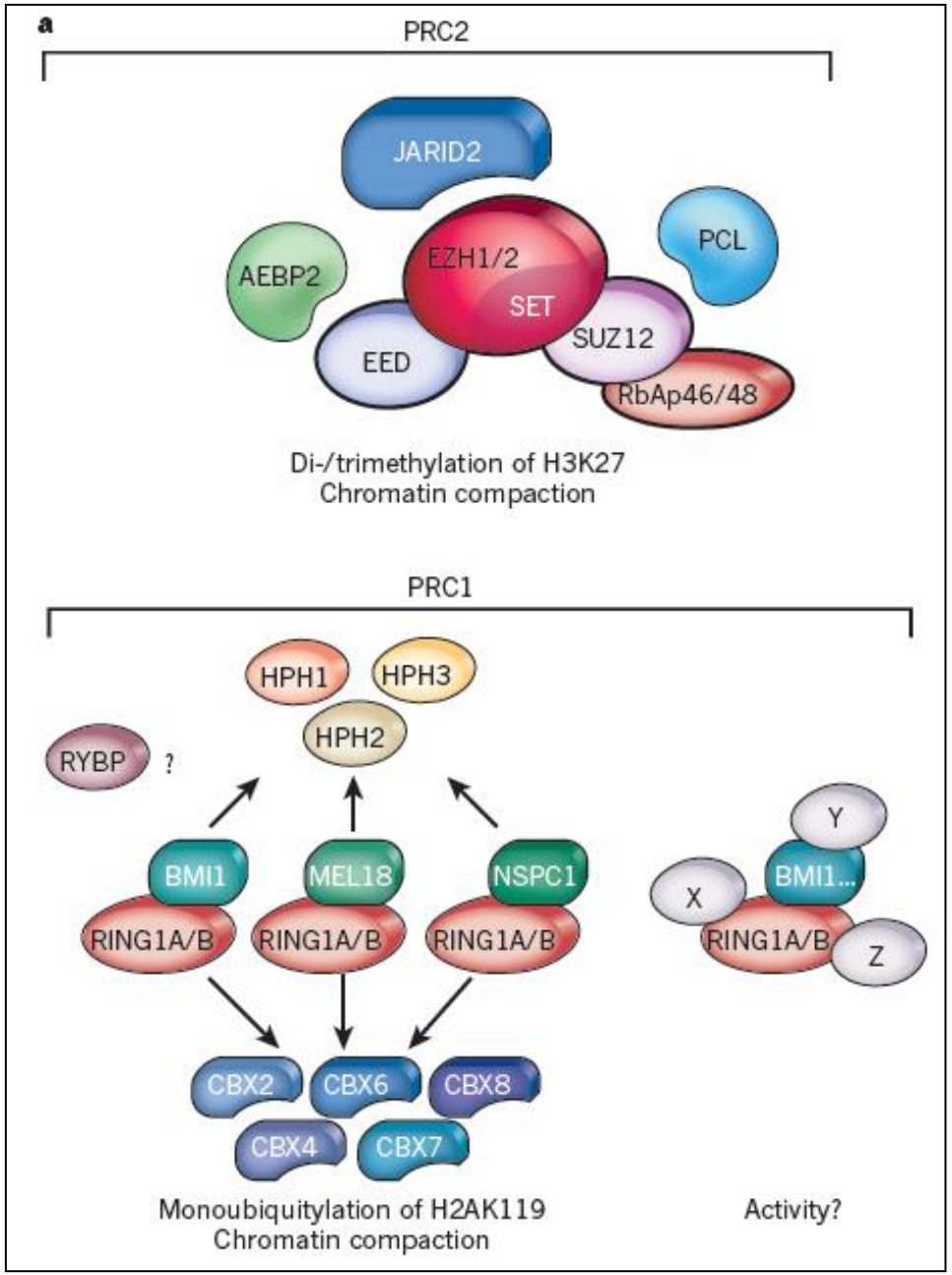
b | A chromatin domain is enriched for repressive marks — such as H3K9me3 (shown), H1K26me3 or H4K20me3 (not shown) — that are recognized by PRC2, but the enzymatic activity of PRC2 is only modestly increased (small yellow star).

c | A chromatin domain is enriched for **H3K27me3**, which is recognized by PRC2 and stimulates a robust increase in its enzymatic activity (large yellow star).

EED, embryonic ectoderm development; EZH2, enhancer of zeste homologue 2; RBBP4, retinoblastoma binding protein 4; SUZ12, suppressor of zeste 12.

Margueron & Reinberg, Nature Rev Genet. 2010, 11:285-98

Figure 1 | The Polycomb complexes PRC1 and PRC2. a, Diagrams representing the composition of PRC2 and PRC1 are shown. In PRC1, the diagrams shown on the left correspond to the classical PRC1 complexes, whereas those on the right correspond to the so-called PRC1-like complexes.
Owing to their homology with the *Drosophila* PSC protein, we assumed that the BMI1-, MEL18- and NSPC1-containing PRC1 complexes could compact chromatin. The 'pocket' shape of the CBX proteins represents the chromodomain that specifically recognized H3K9/27me3. HPH1, 2 and 3 denote human polyhomeotic homologue 1, 2 and 3. X, Y and Z denote various proteins such as SCMH1/2, FBXL10, E2F6 and JARID1D that could contribute to the formation of PRC1-like complexes, whose exact composition is still enigmatic.



From Margueron & Reinberg (2011) Nature 469: 343-49

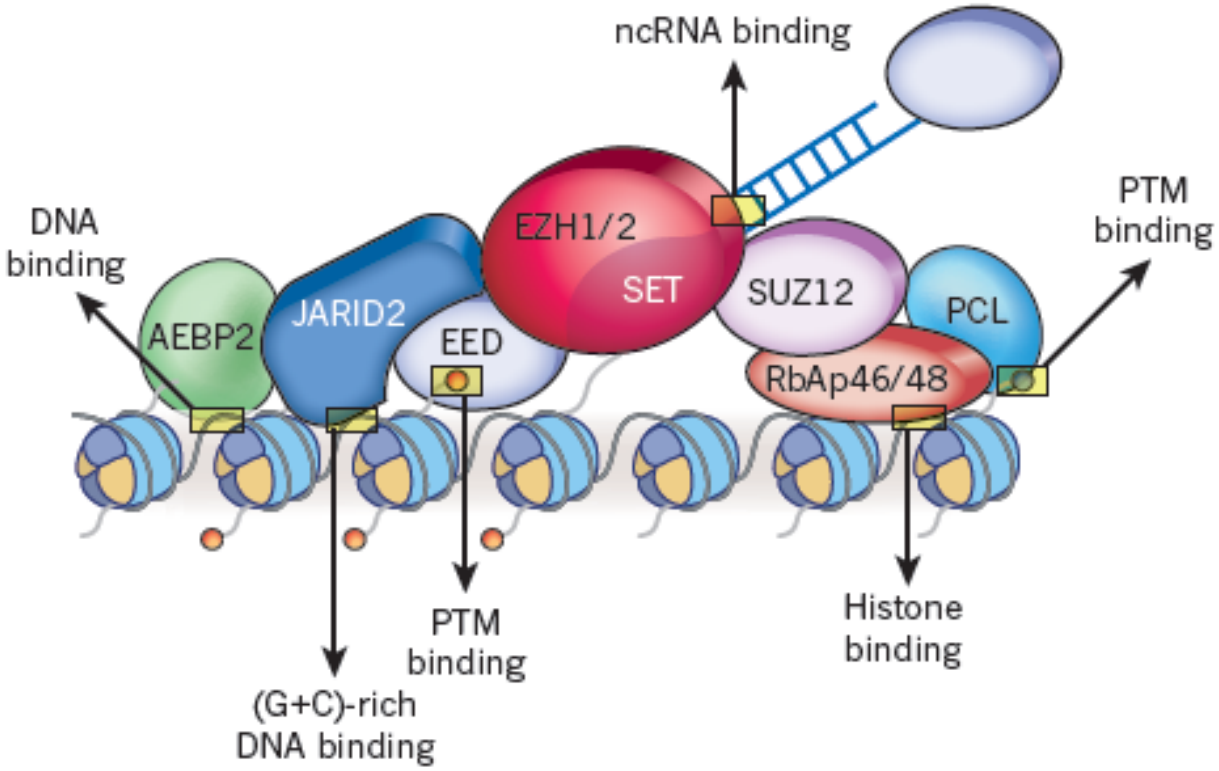


Figure 3 | The many interactions of PRC2 with chromatin. Schematic representation of the PRC2 holoenzyme at chromatin. Putative interactions with either DNA or histones that could explain PRC2 recruitment are highlighted.

From Margueron & Reinberg (2011) Nature 469: 343-49