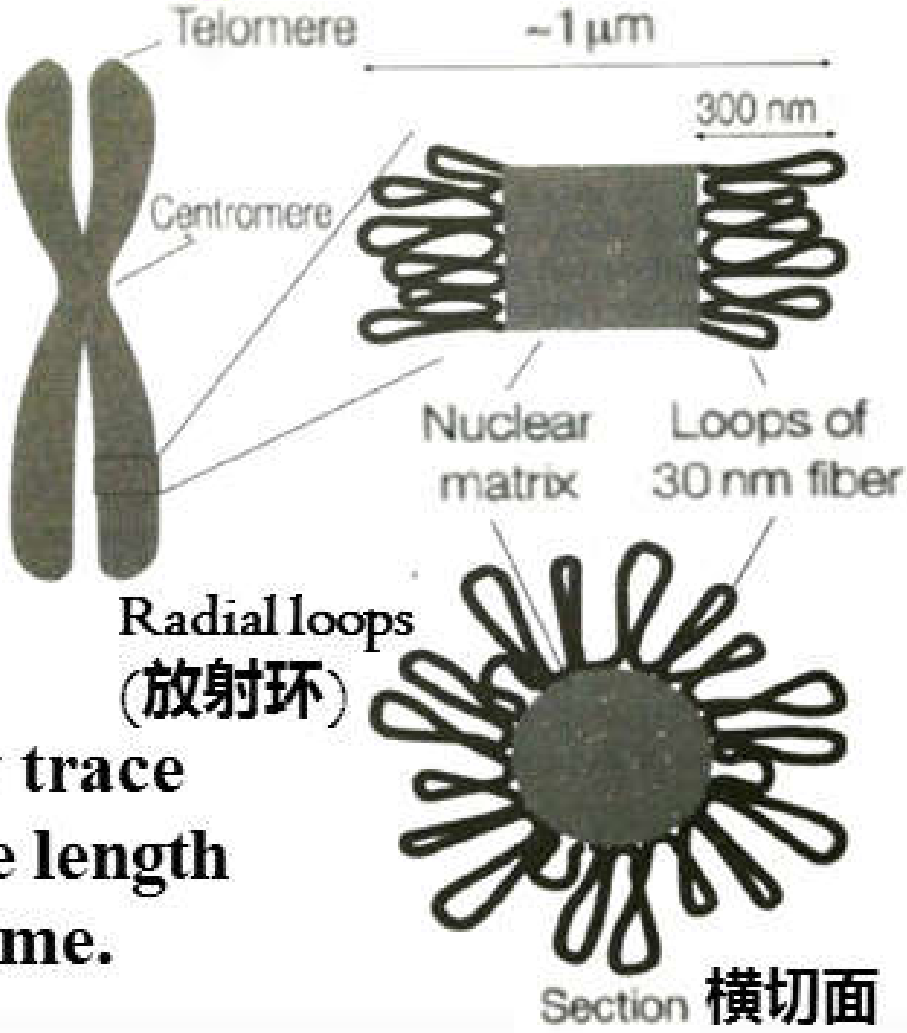
3. Eukaryotic chromosome structure

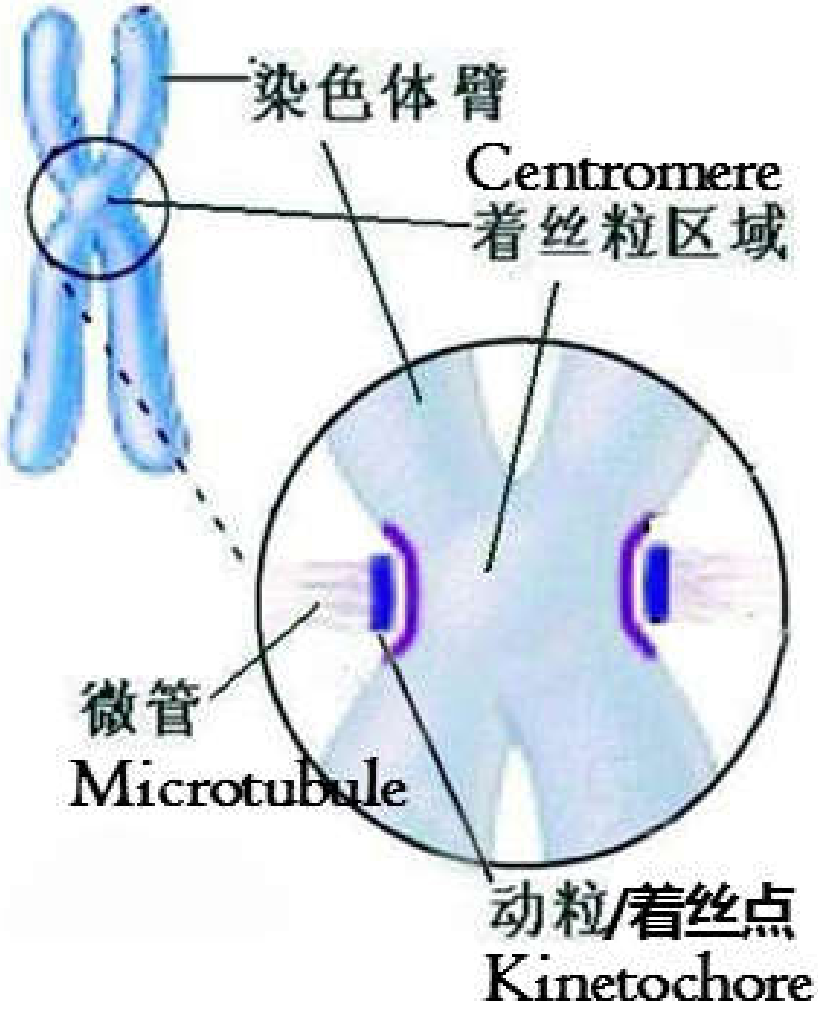
3.1 The mitotic (有丝分裂) chromosome

- Two identical sister **chromatids** (染色单体) joined at their **centromeres** (着丝粒).
- The ends of the chromosomes are the **telomeres** (端粒).
- Consecutive loops may trace a helical path along the length (长轴) of the chromosome.

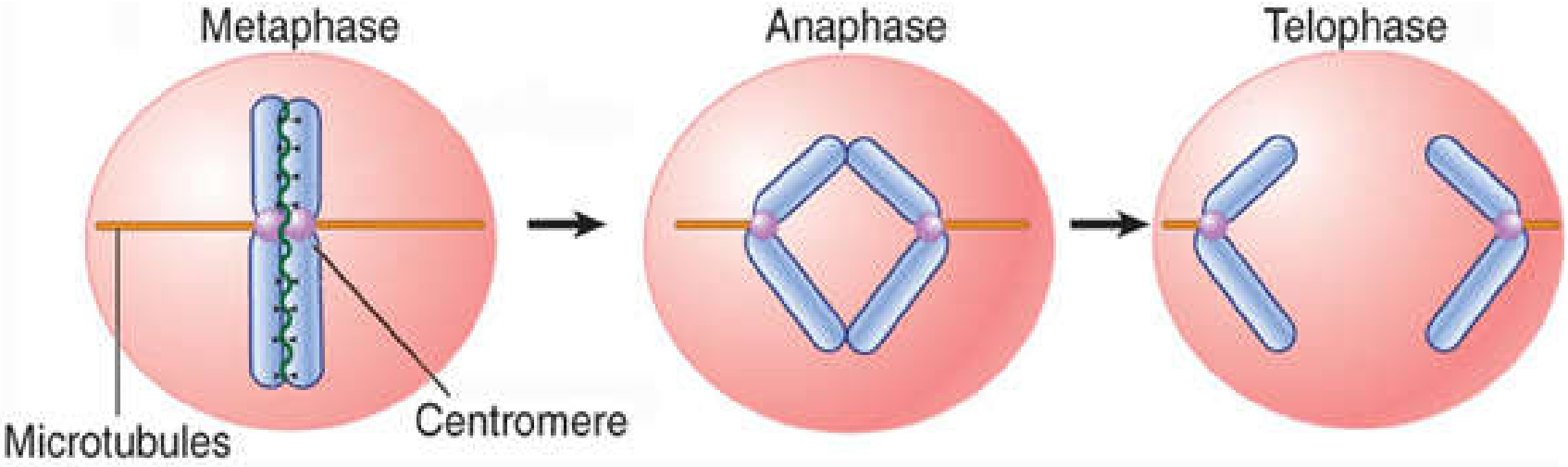


3.2 The centromere

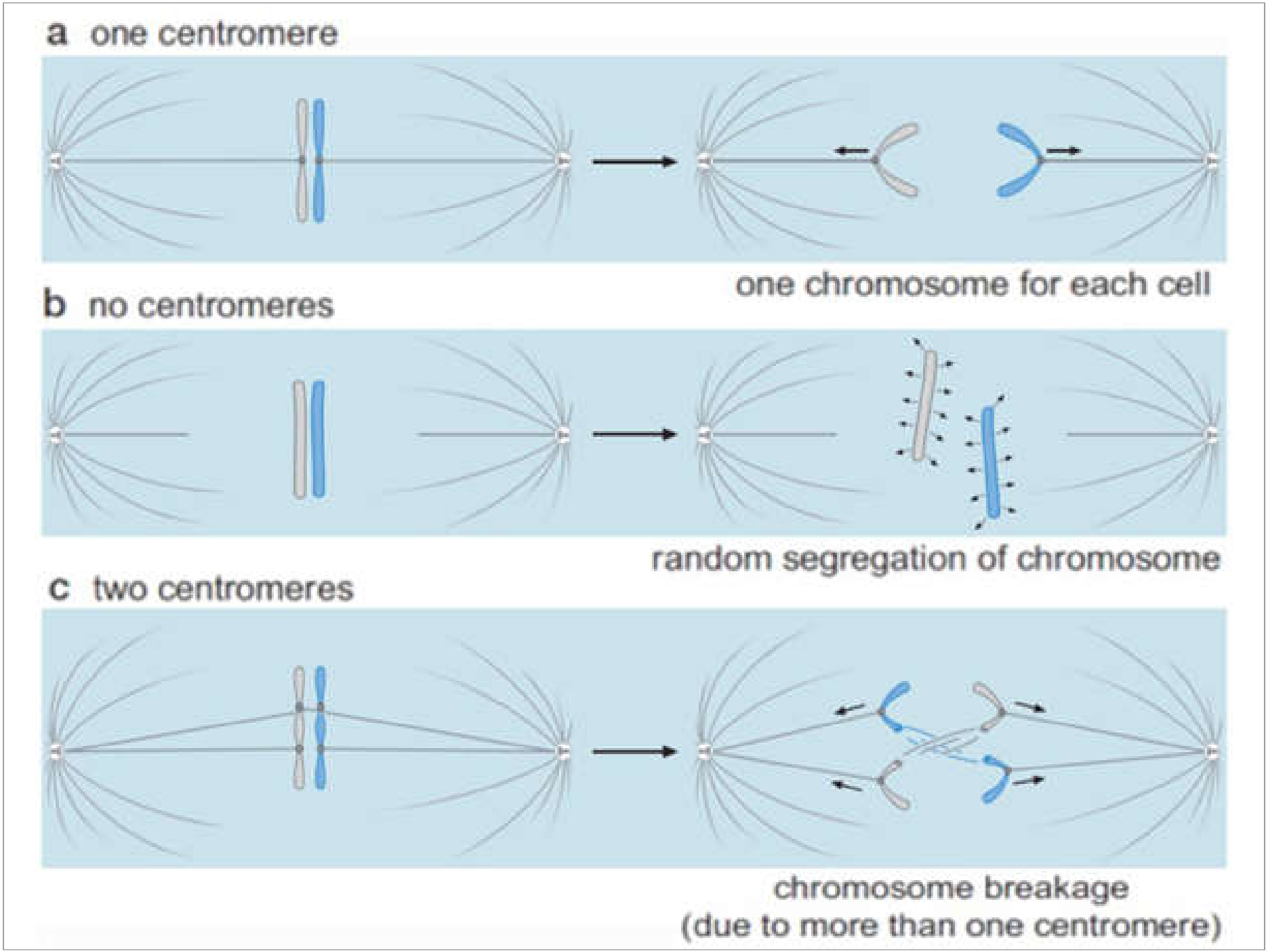
- The centromere is the constricted region where the two sister chromatids are joined in the metaphase chromosome. 着丝粒是分裂中期两条姐妹染色单体相连的紧缩区域。
- It is the site of assembly of the kinetochore (动粒), a protein complex which provides a microtubule (微管) attachment point.



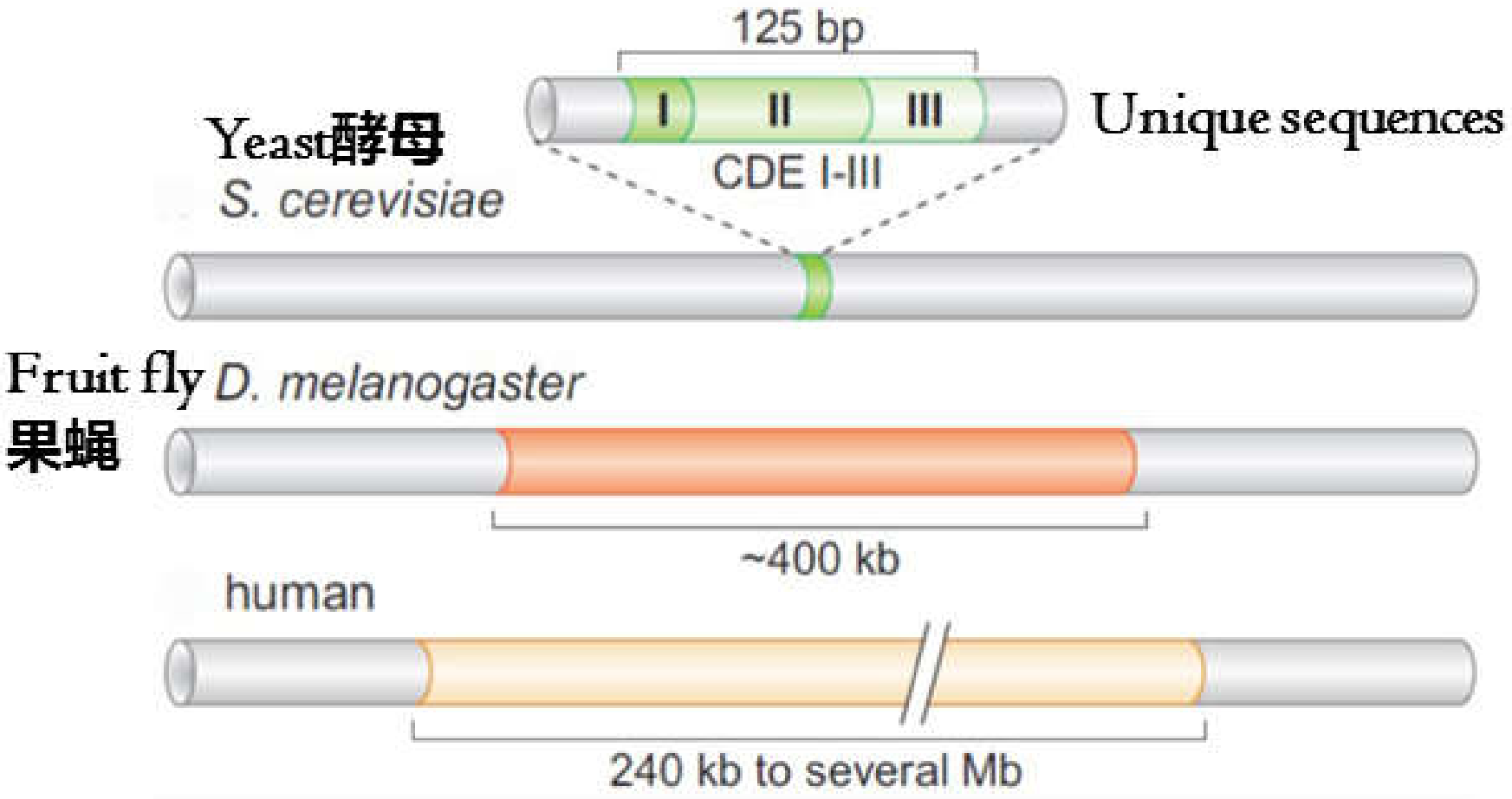
- **The microtubules act as to separate and pull the chromatids (染色单体) to the poles (两极) at anaphase (后期) and telophase (末期).**



- **Function:** Centromere is responsible for the correct segregation of the chromosome at mitosis and meiosis (减数分裂).


a one centromere
one chromosome for each cell
b no centromeres
random segregation of chromosome
c two centromeres
chromosome breakage
(due to more than one centromere)

- **Centromeres vary greatly in size.**



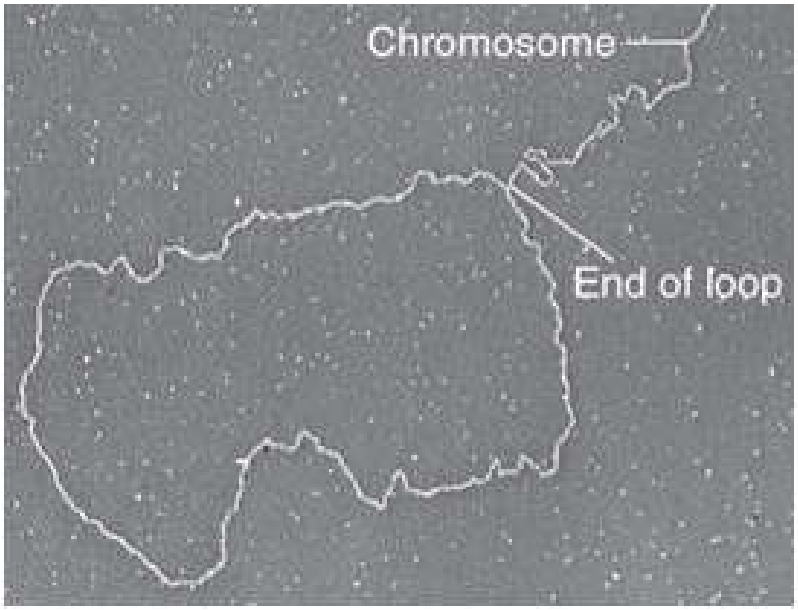
In the majority of eukaryotes, centromeres are >40 kb and are composed of largely repetitive (**重复的**) DNA, known as satellite DNA (**卫星**DNA).

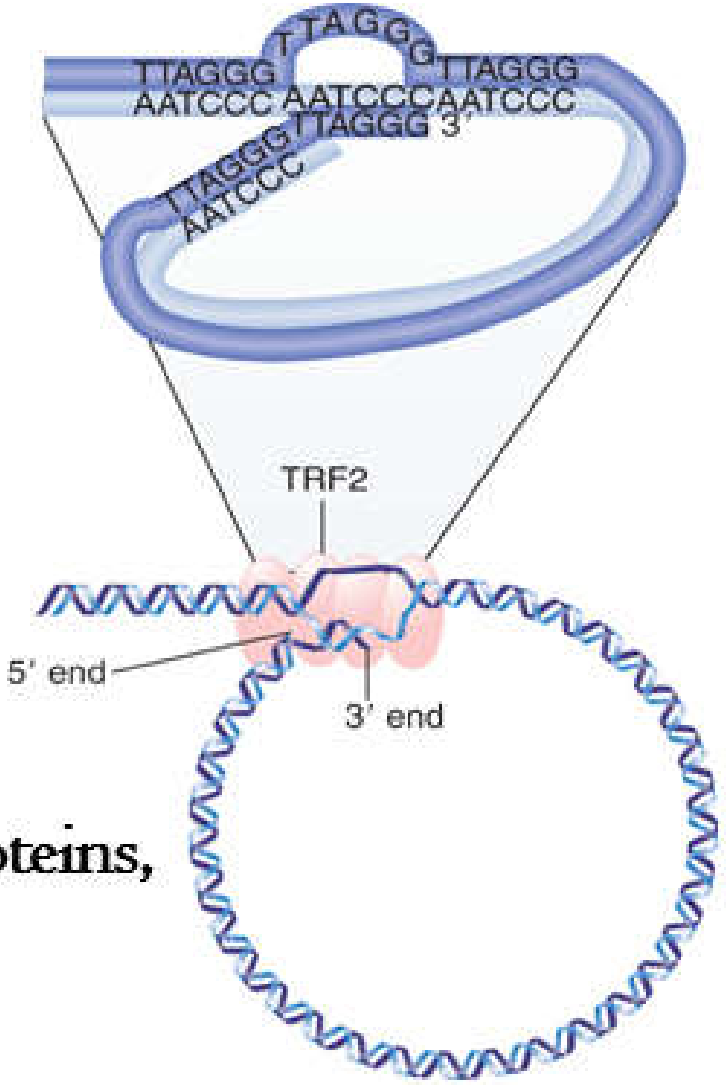
3.3 Telomeres

- **Telomeres** are specialized DNA sequences that form **the ends** of the linear DNA molecules of the eukaryotic chromosomes. **端粒**是形成真核生物染色体线性DNA分子**末端**的特化了的DNA序列。
- Telomeres consist of hundreds of copies of **a simple, non-informational repetitive sequence** (TG-rich repeat) with the 3'-end overhanging to (突出于) the 5'-end.

Structure of a typical telomere

- **TG-rich strand forms a loop by displacing its homolog (同源) in an upstream region of the telomere.**





Telomere binding proteins, e.g. TRF2

- **Functions of telomeres**

(1) Seal and stabilize the chromosome ends

(The ends of linear chromosomes cannot be fully replicated.)

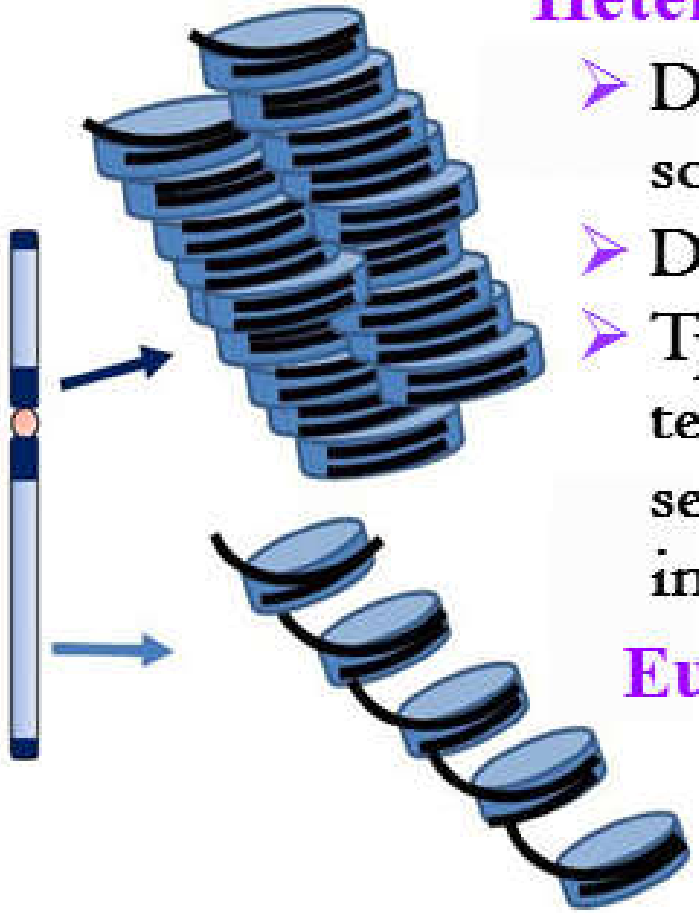
(2) Related to aging and cancer

- **Telomeres are synthesized by telomerase (端粒酶).**
- **Telomerase is a ribonucleoprotein enzyme.**

RNA+Protein

3.4 Heterochromatin and euchromatin

- Chromosomes consist of heterochromatin (异染色质) and euchromatin (常染色质).



Heterochromatin

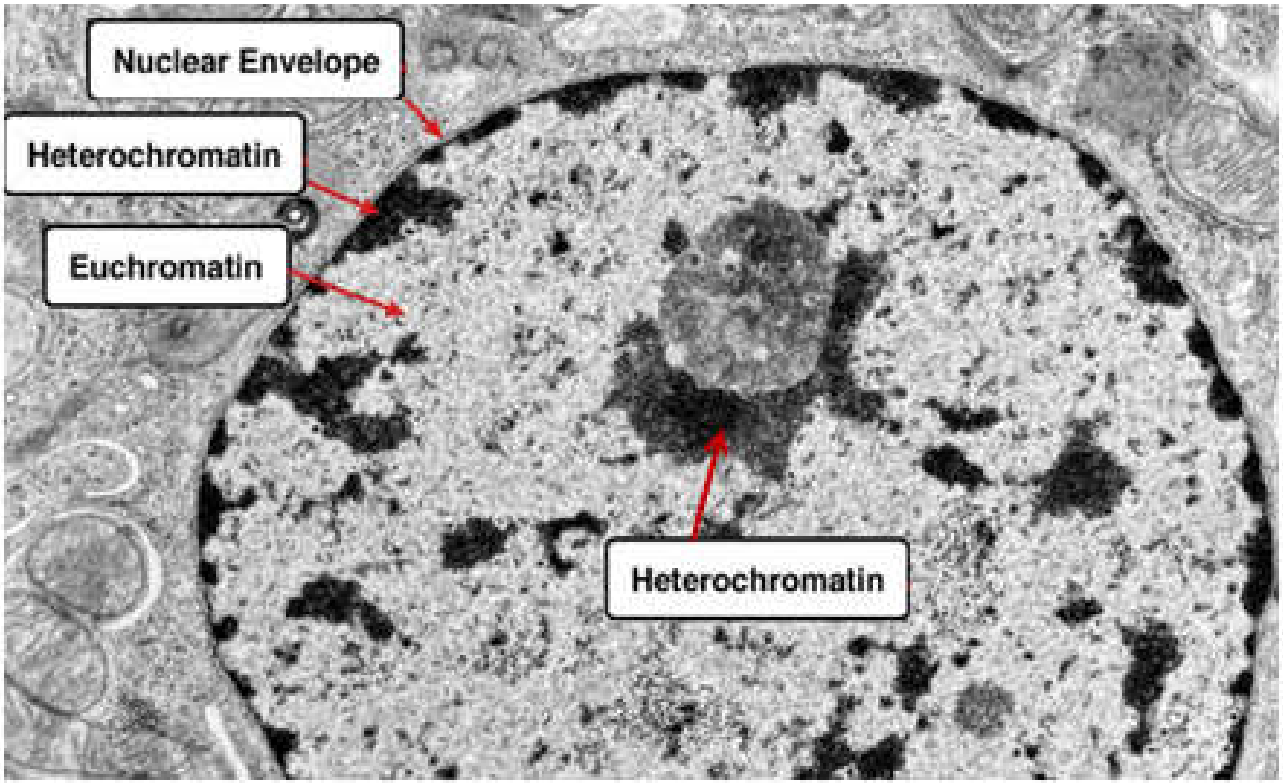
间期

- Densely packaged in interphase (not so compacted as at metaphase)
- Dense staining with dyes
- Typically found at centromeres, telomeres and other highly repetitive sequences. (one of X chromosomes in female mammals)

Euchromatin

- Less densely packaged
- Staining poorly with dyes

- **Heterochromatin** forms a series of discrete clumps (不连续的团块), and is most often found at the **nuclear periphery** (边缘), and at the **nucleolus**. **Euchromatin** has a relatively dispersed appearance in the nucleus and occupies most of the nuclear region.



- **Heterochromatin is transcriptionally inactive. In contrast, euchromatin showed higher levels of gene expression.**

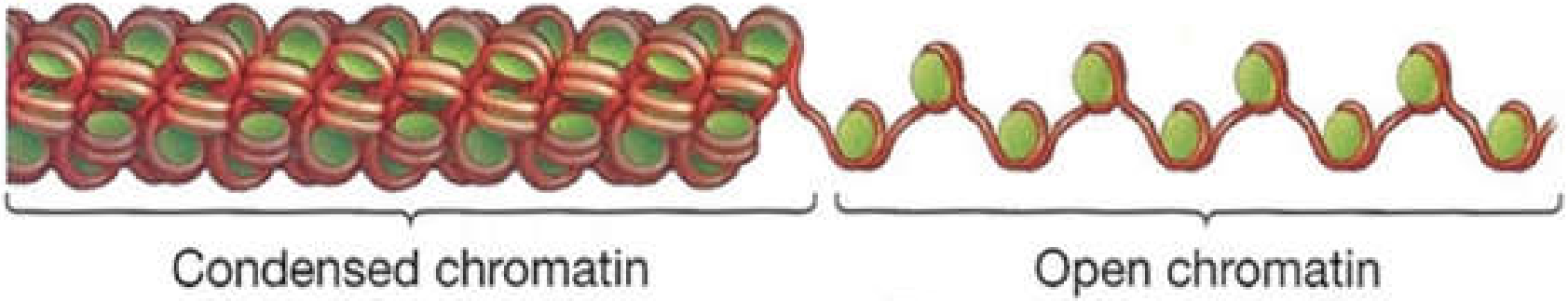
  - Ribosomal DNA in the nucleolus has the general compacted appearance and behavior of heterochromatin (such as late replication), yet is engaged in very active transcription.
  - Active genes are contained within euchromatin, but only a minority of the sequences in euchromatin are transcribed at any time.

Euchromatin is not homogeneous (均一的).

- Relative inactive regions: Consisting of chromosomal loops compacted in 30 nm fibers
- Active regions (~10%): 'Beads on a string' structure; Be depleted of nucleosomes

Loose structure

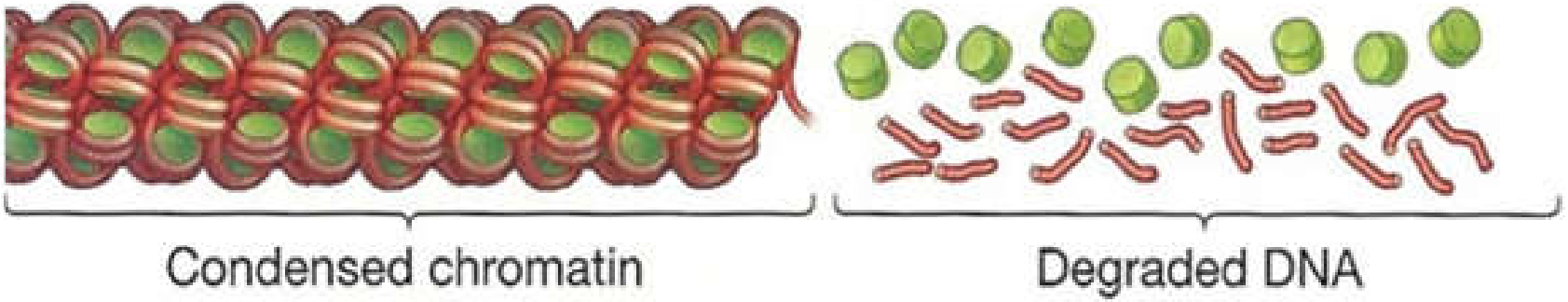
3.5 DNase Ⅰ hypersensitivity



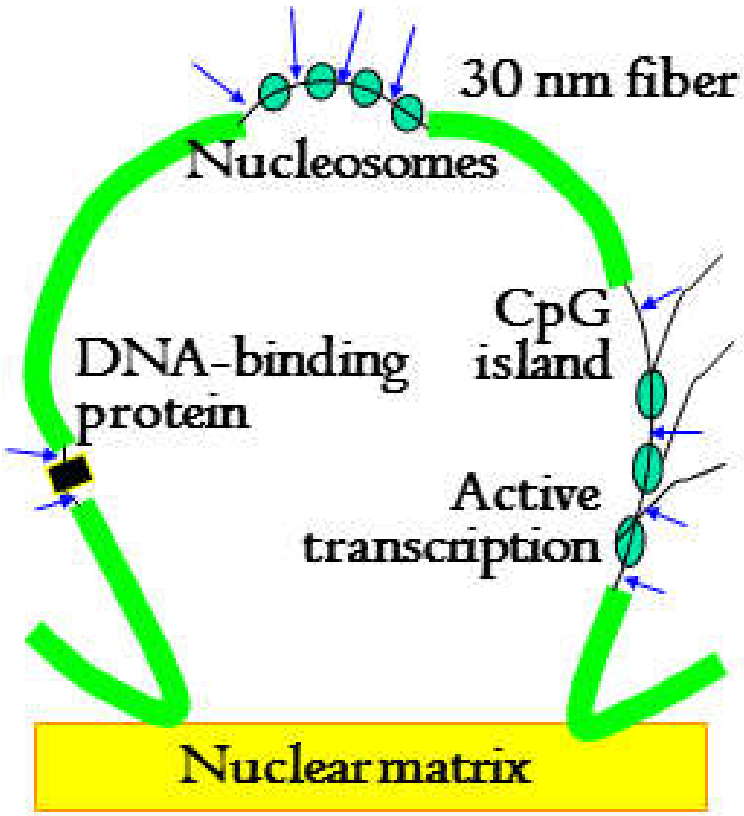
- **Deoxyribonuclease（脱氧核糖核酸酶）Ⅰ（DNase Ⅰ）cuts the backbone of DNA unless the DNA is protected by bound proteins.**

Naked(裸露的) DNA is easily cut by DNase Ⅰ.

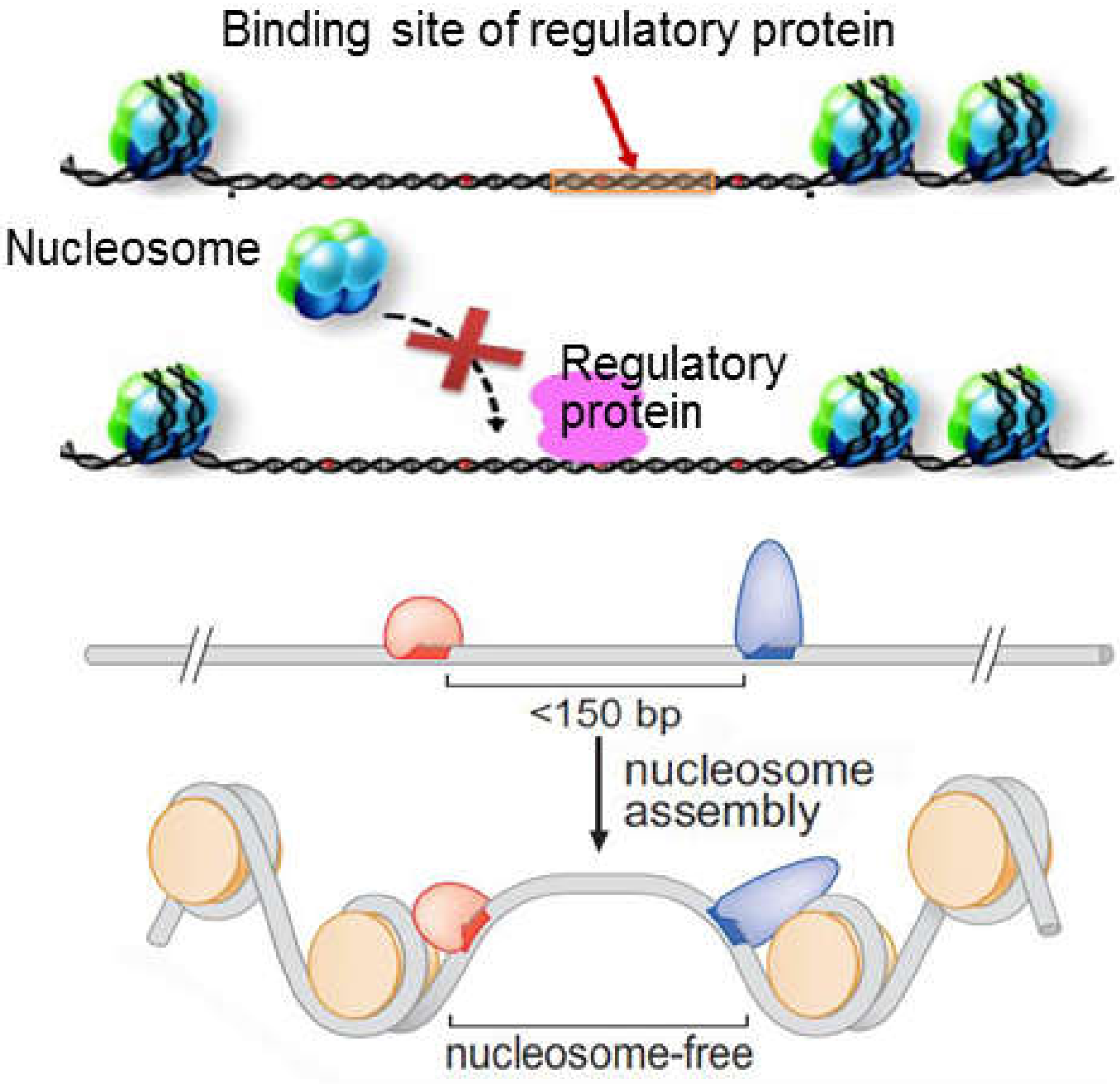
Treatment with DNase I



- **DNase I hypersensitivity has been used to map the regions of transcriptionally active chromatin:**
  - Short regions: 30 nm fiber is interrupted by the binding of a sequence-specific regulatory protein.
  - Longer regions: where transcription is taking place.

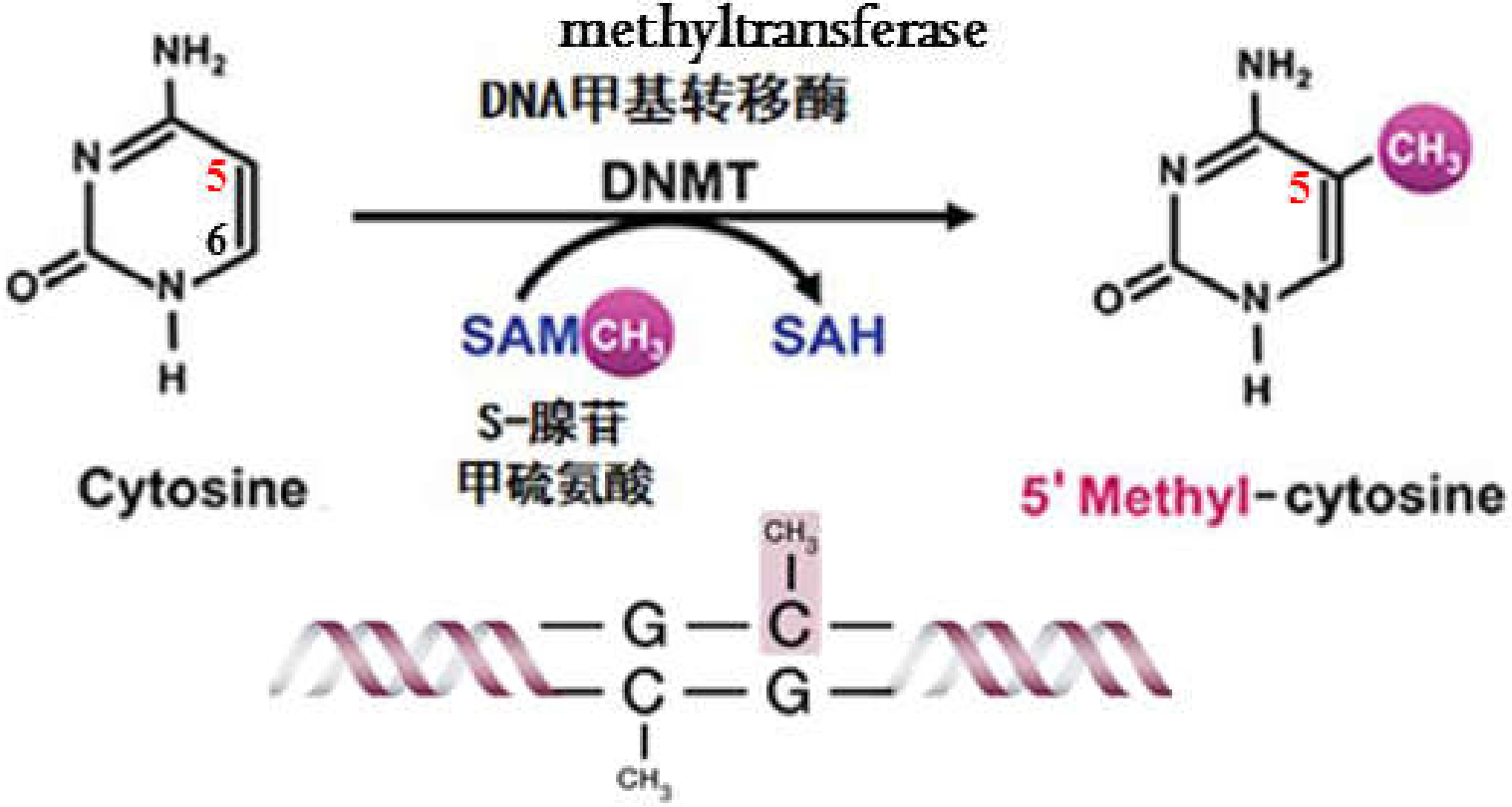


- **These regions vary between different cell types and different phases during cell development.**


Binding site of regulatory protein
Nucleosome
Regulatory protein
<150 bp
nucleosome assembly
nucleosome-free

3.6 CpG methylation (甲基化)

- **CpG methylation is the methylation of C-5 in the cytosine (胞嘧啶) base of 5'-CG-3' sequences in mammalian cells.**



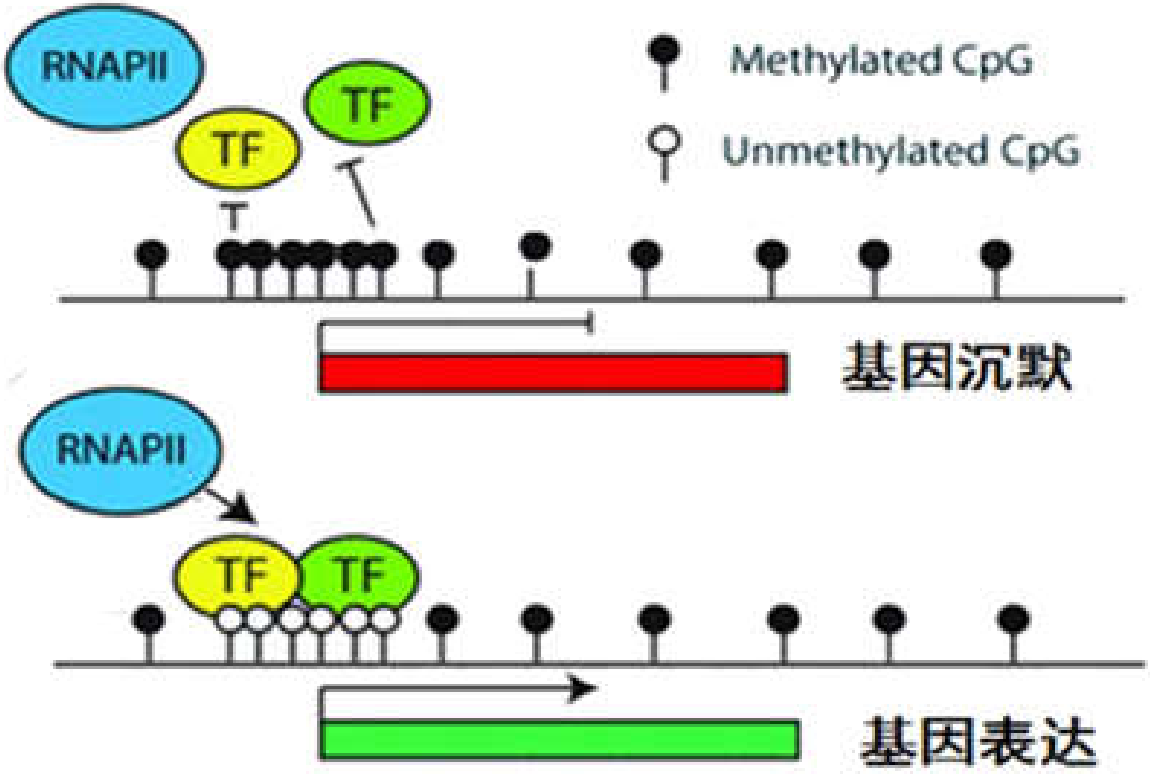
Functions of CpG methylation

(1) Involved in keeping the appropriate level of chromosomal packing

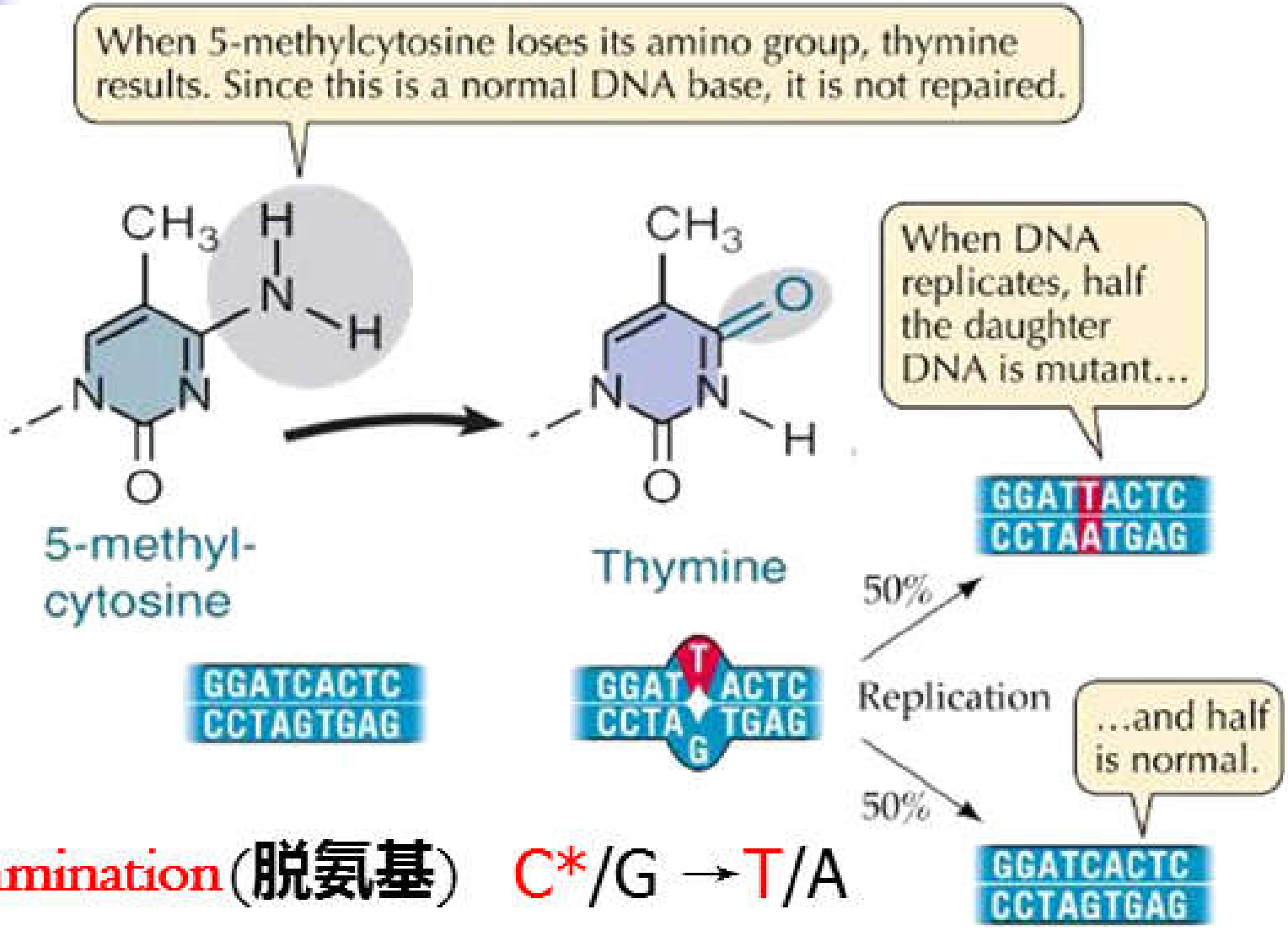
- CpG methylation induces tighter wrapping of DNA around the histone core.
- Unmethylated CpG islands may be largely free of nucleosomes, and are coincident with regions of particular sensitivity to DNase I.

(2) Regulate gene expression

- The methylation of CpG is associated with transcriptionally inactive regions of chromatin.
- Unmethylated CpG islands surround the promoter regions of active genes (e.g. housekeeping genes 管家基因)

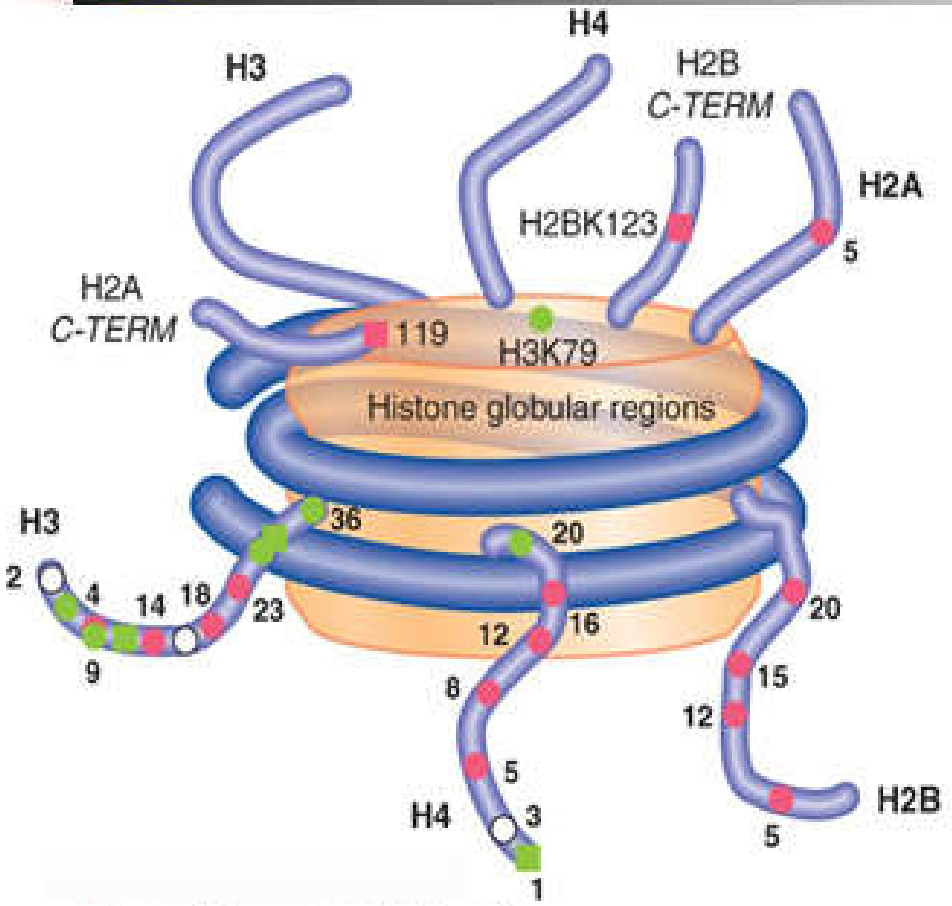


CpG methylation mutation



Deamination(脱氨基) C*/G →T/A

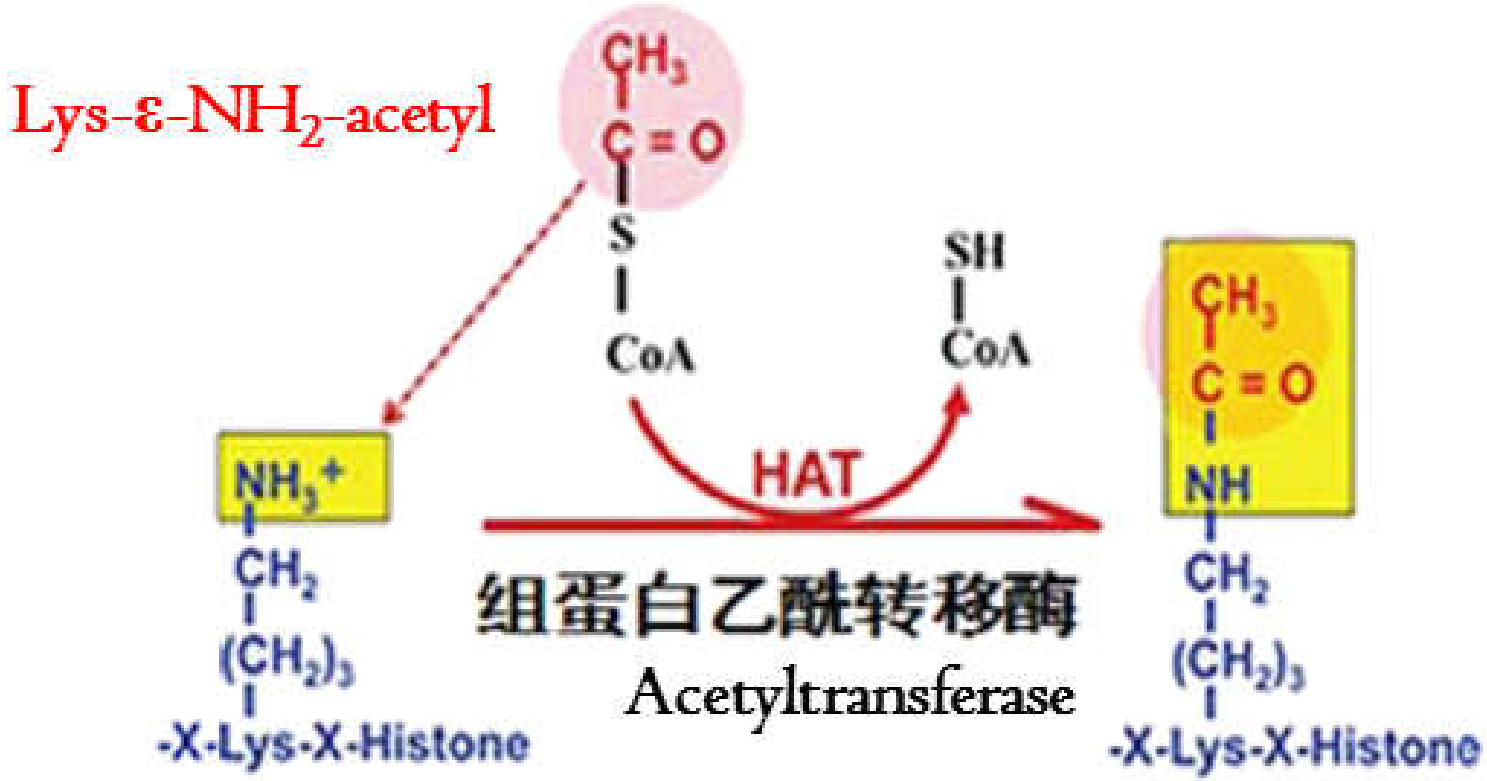
3.7 Histone modification



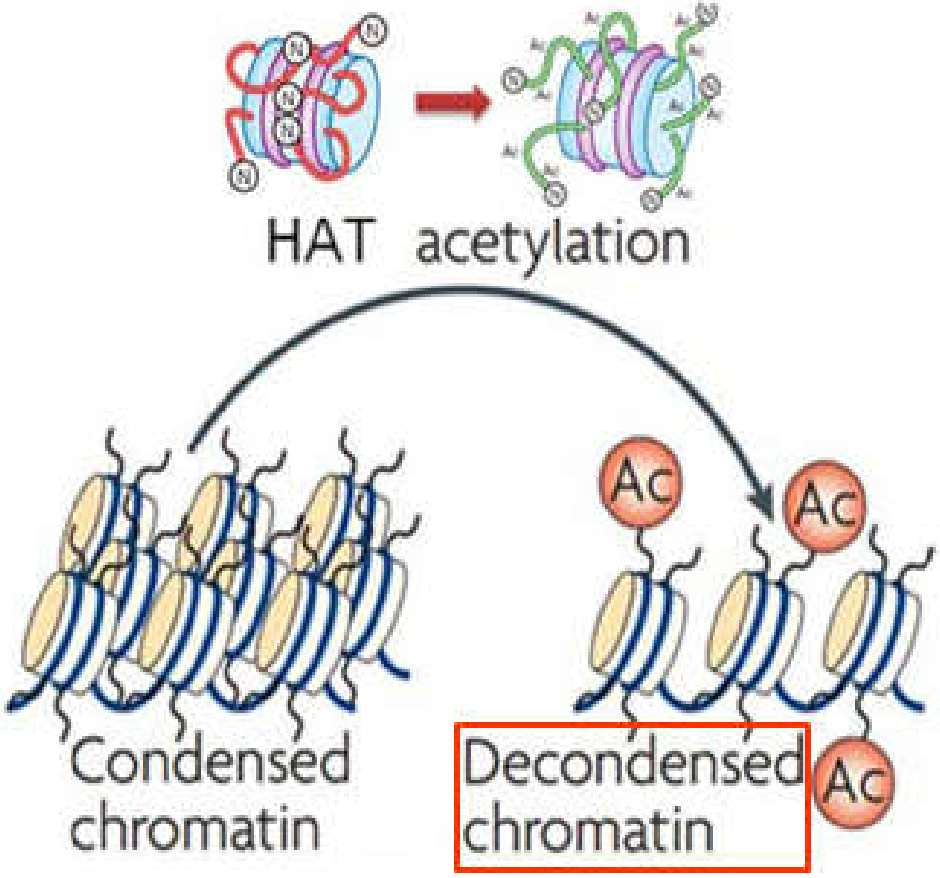
- Acetylation (乙酰化)
- Phosphorylation (磷酸化)
- Methylation (甲基化)
- Mono-ubiquitination (单泛素化)

- **Chemical modification of histone proteins is believed to controls the degree of chromatin condensation.**

(1) Acetylation (乙酰化)



Acetylation of histones neutralizes the positive charge of histones.



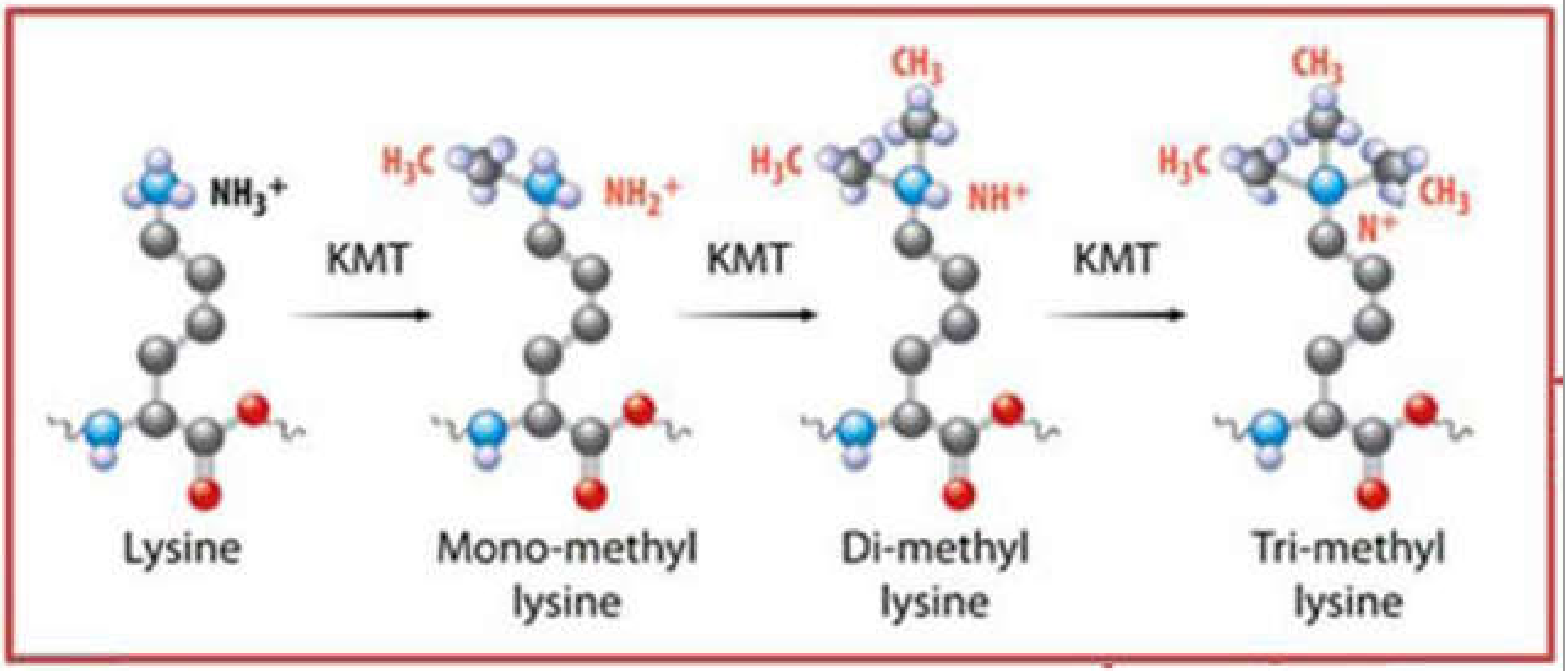
- The loss of positive charge reduces the affinity of the tails for the negatively charged backbone of the DNA.
- Histone amino-terminal tails are required to form the 30-nm fiber, and modification of the tails modulates this function.
- Histone acetylation is generally associated with gene activation.

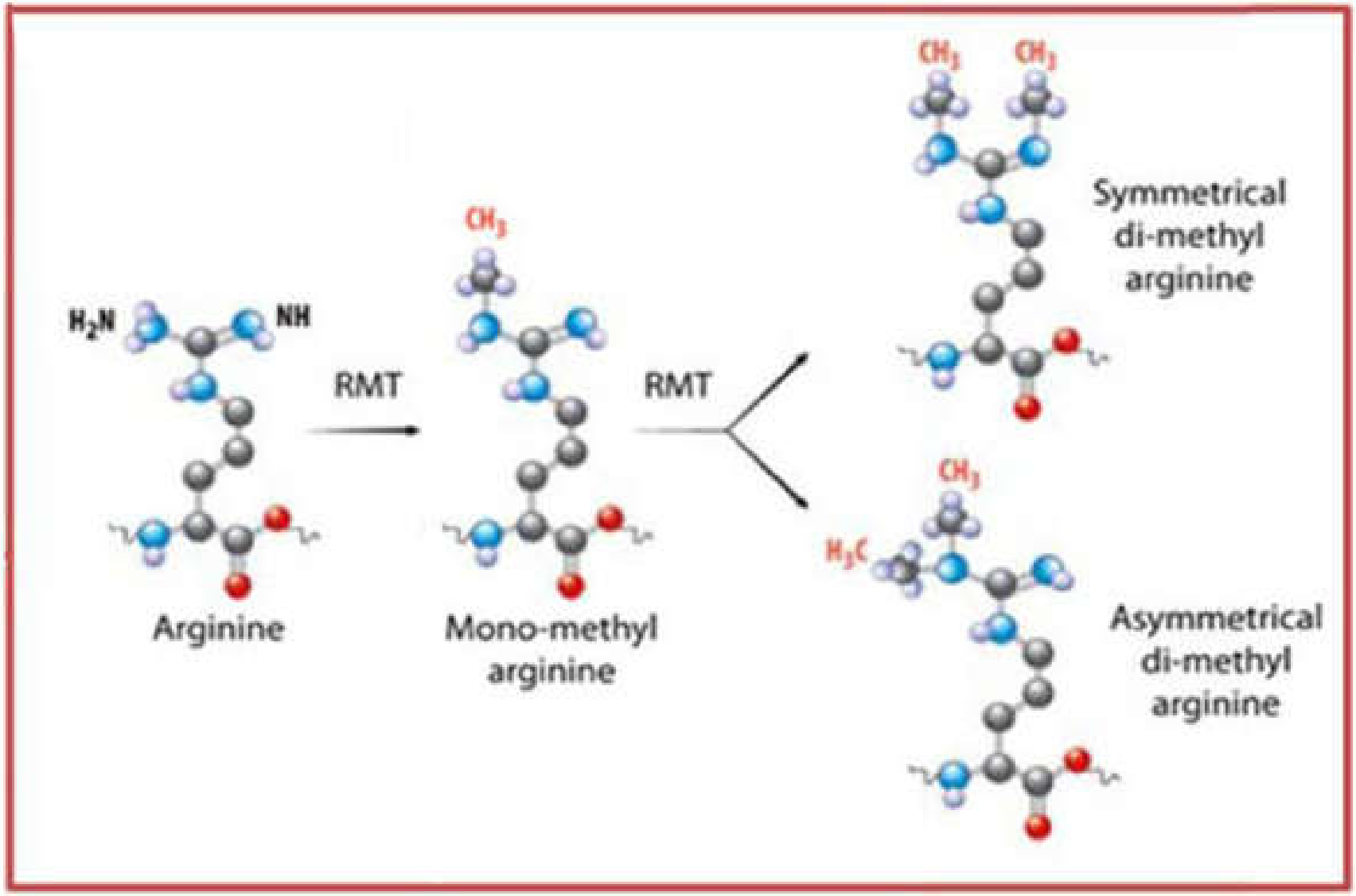
(2) Phosphorylation (磷酸化)

- Phosphorylation occurs on the hydroxyl group (羟基) of serine (Ser), threonine (Thr) and tyrosine (Tyr).
- Phosphorylation introduces a negative charge in the form of the phosphate group. → Decondensed chromatin
- Histone phosphorylation is generally associated with gene activation.
- The condensation of chromosomes at mitosis is accompanied by the phosphorylation of histone H1.

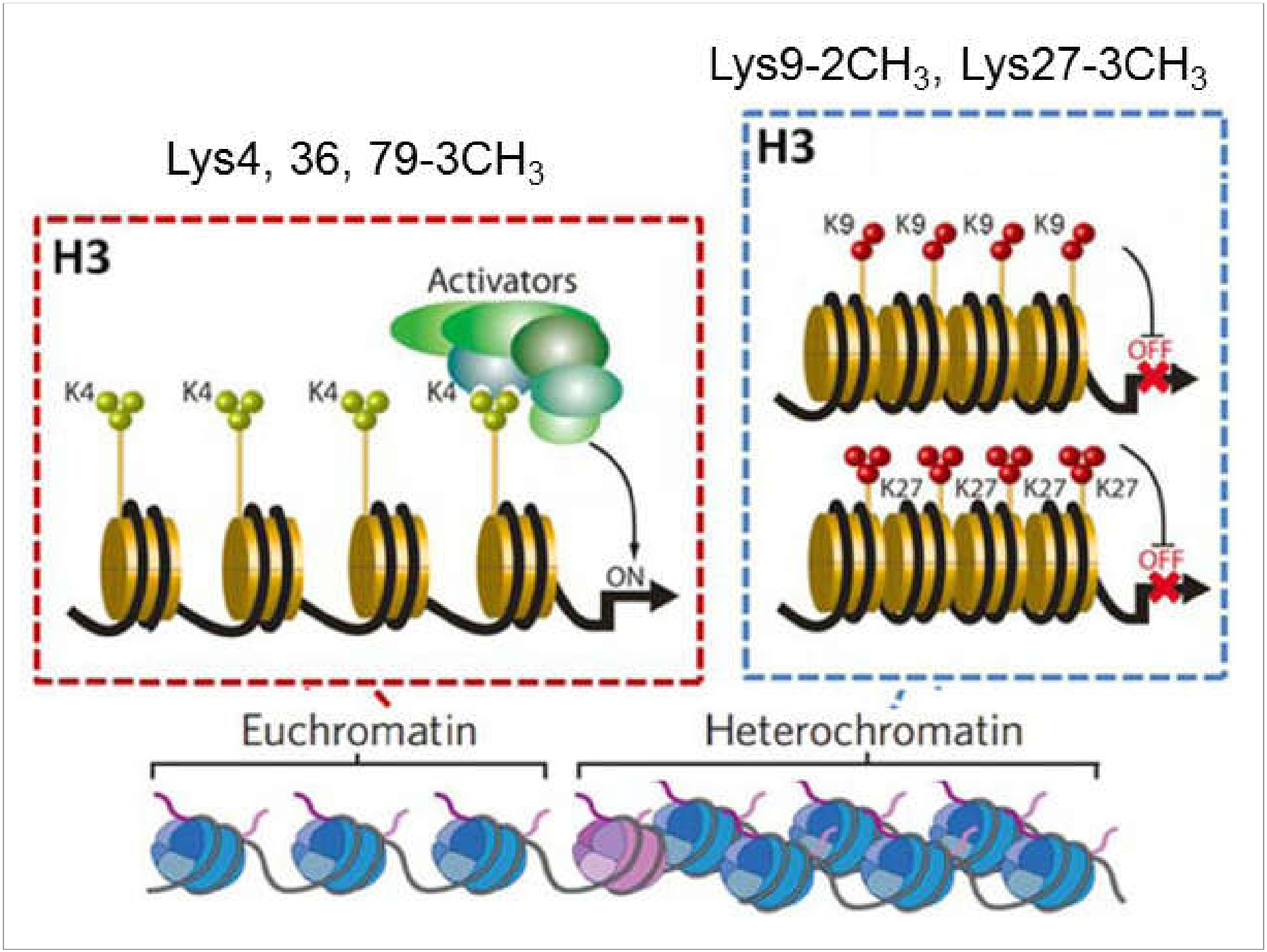
(3) Methylation (甲基化)

- Lysine methylation retains the positive charge, and lysine can be mono-, di-, or trimethylated.



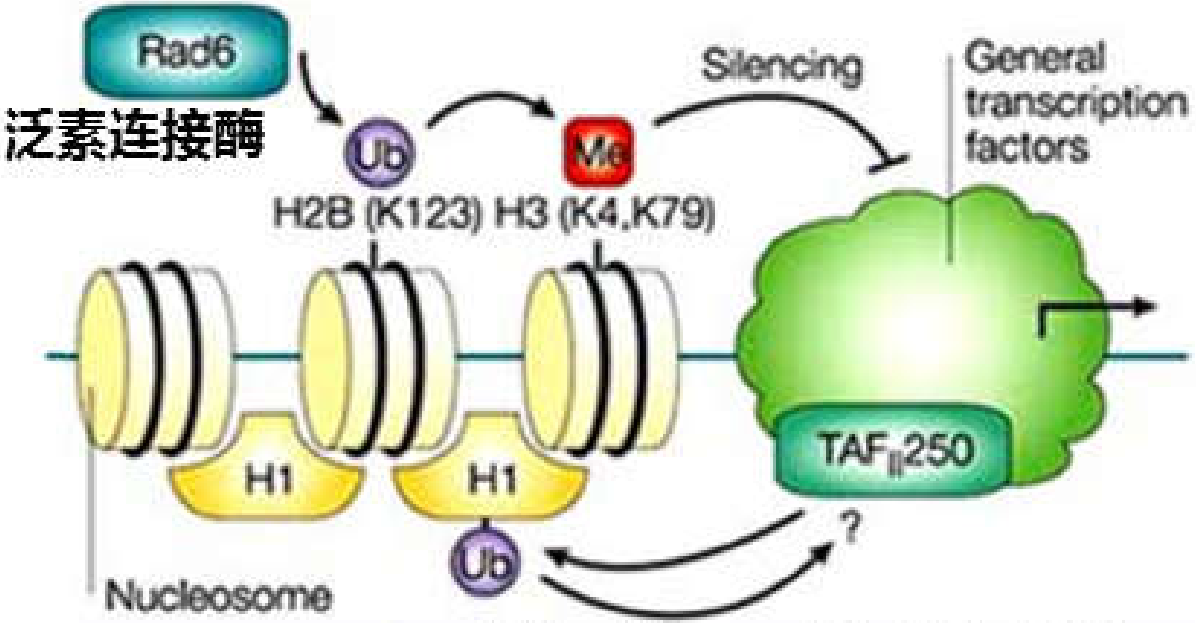


➢ Arginine can be mono- or dimethylated.

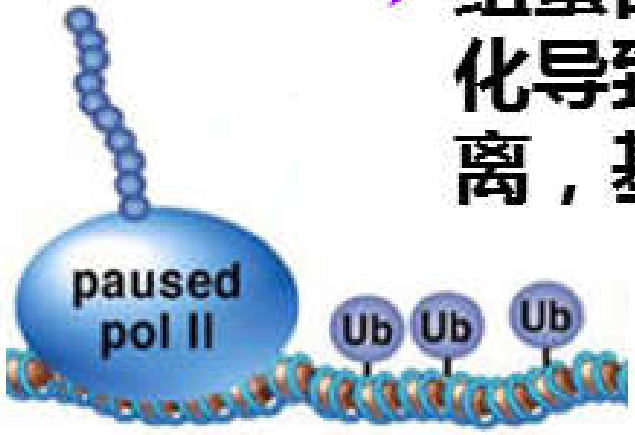

Lys9-2CH3, Lys27-3CH3
Lys4, 36, 79-3CH3
H3
Activators
K4
K4
K4
K4
ON
H3
K9
K9
K9
K9
OFF
K27
K27
K27
K27
OFF
Euchromatin
Heterochromatin

(4) Mono-ubiquitination (单泛素化)

- 泛素连接酶可催化组蛋白Lys残基与泛素之间成键。
- 组蛋白H2B的单泛素化可能导致基因沉默或者影响延伸。



- 组蛋白H1的单泛素化导致其从DNA上解离，基因被活化。



3.8 Histone variants

- **All histones except H4 are members of families of related variants.**

  (H5 replaces H1 in some very inactive chromatin, for example in avian red blood cells.)

- **Histone variants can be closely related or highly different from general histones.**

- **Different variants serve different functions in the cell.**