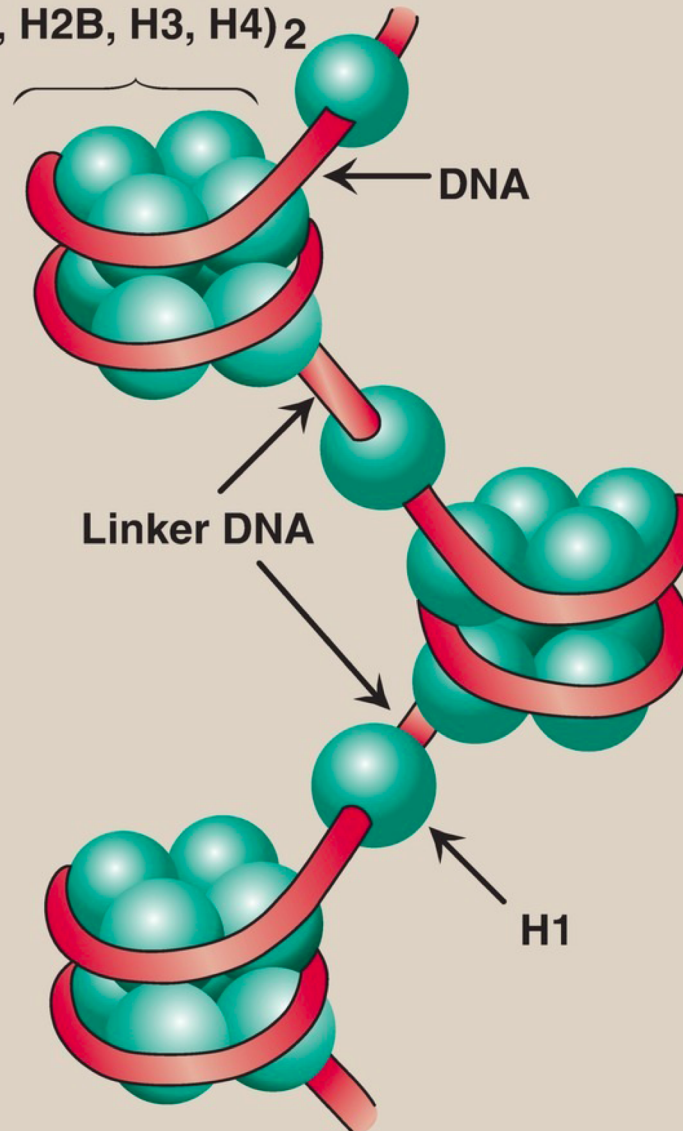


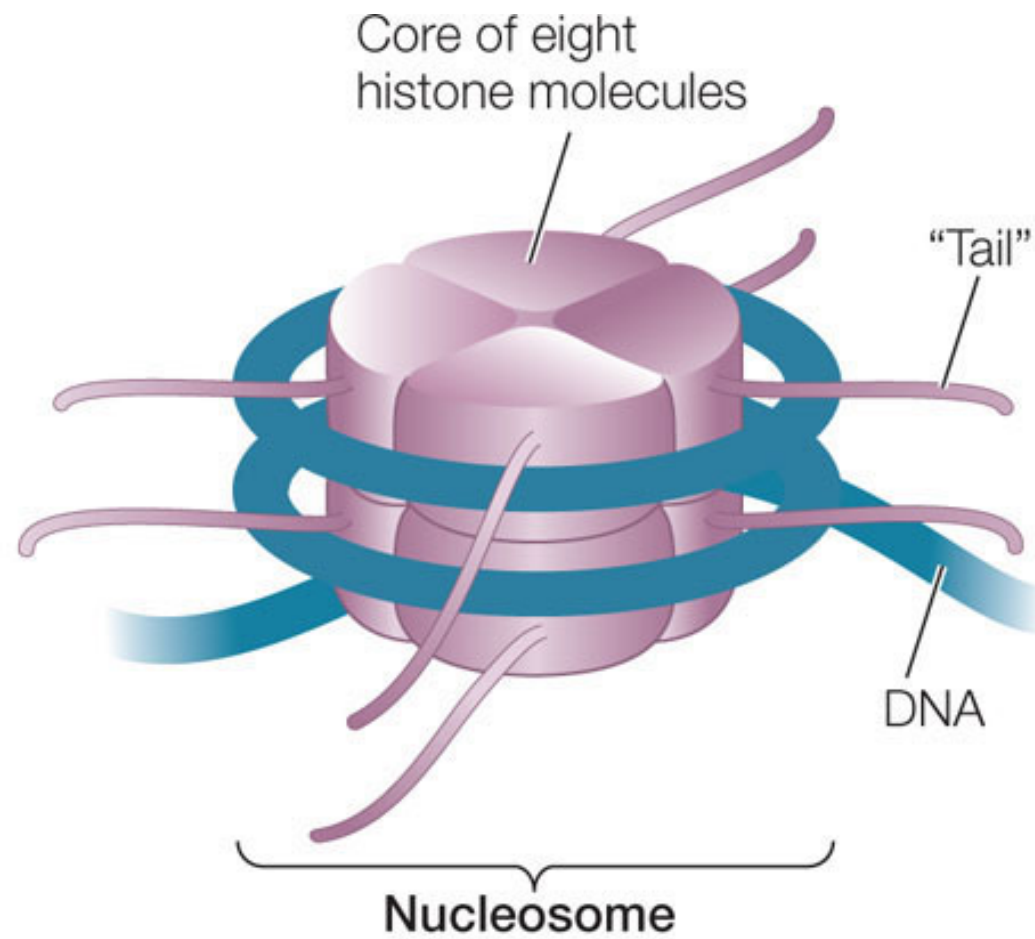
Nucleosome core
(H2A, H2B, H3, H4)₂

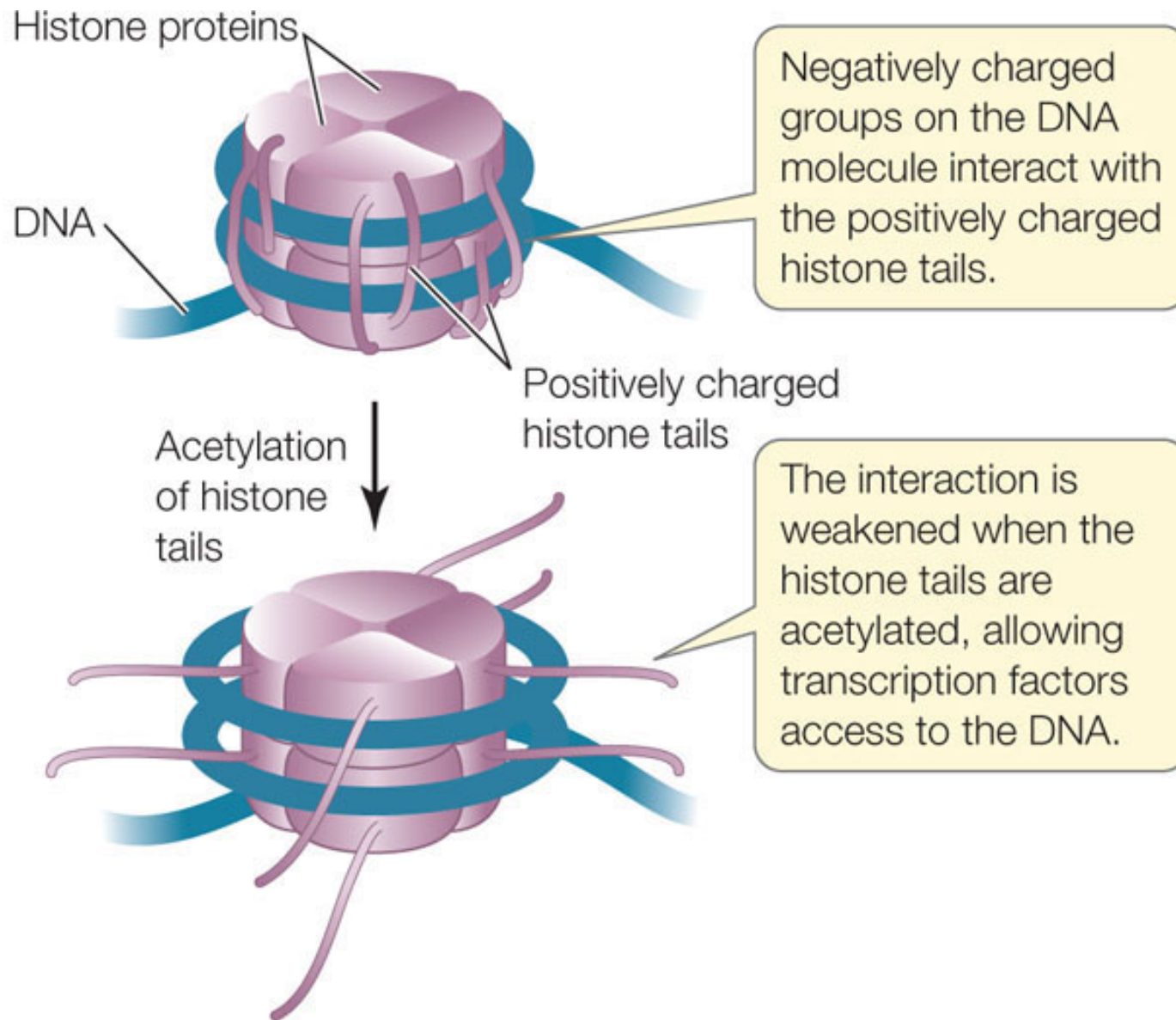


DNA

Linker DNA

H1





Ac—	<u>Ser</u>	Gly	Arg	Gly	<u>Lys</u>	Gly	Gly	<u>Lys</u>	Gly	Leu	10
	Gly	<u>Lys</u>	Gly	Gly	<u>Ala</u>	<u>Lys</u>	Arg	His	Arg	<u>Lys</u>	20
	Val	Leu	Arg	Asp	Asn	Ile	Gln	Gly	Ile	Thr	30
	Lys	Pro	Ala	Ile	Arg	Arg	Leu	Ala	Arg	Arg	40
	Gly	Gly	Val	Lys	Arg	Ile	Ser	Gly	Leu	Ile	50
	Tyr	Glu	Glu	Thr	Arg	Gly	Val	Leu	Lys	Val	60
	Phe	Leu	Glu	Asn	Val	Ile	Arg	Asp	Ala	Val	70
	Thr	Tyr	Thr	Glu	His	Ala	Lys	Arg	Lys	Thr	80
	Val	Thr	Ala	Met	Asp	Val	Val	Tyr	Ala	Leu	90
	Lys	Arg	Gln	Gly	Arg	Thr	Leu	Tyr	Gly	Phe	100
	Gly	Gly									102

Figure 34-3 The amino acid sequence of calf thymus histone H4.

This 102-residue protein's 25 Arg and Lys residues are indicated in red. Pea seedling H4 differs from that of calf thymus by conservative changes at the two shaded residues: Val 60 → Ile and Lys 77 → Arg.

The underlined residues are subject to **post-translational modification**:

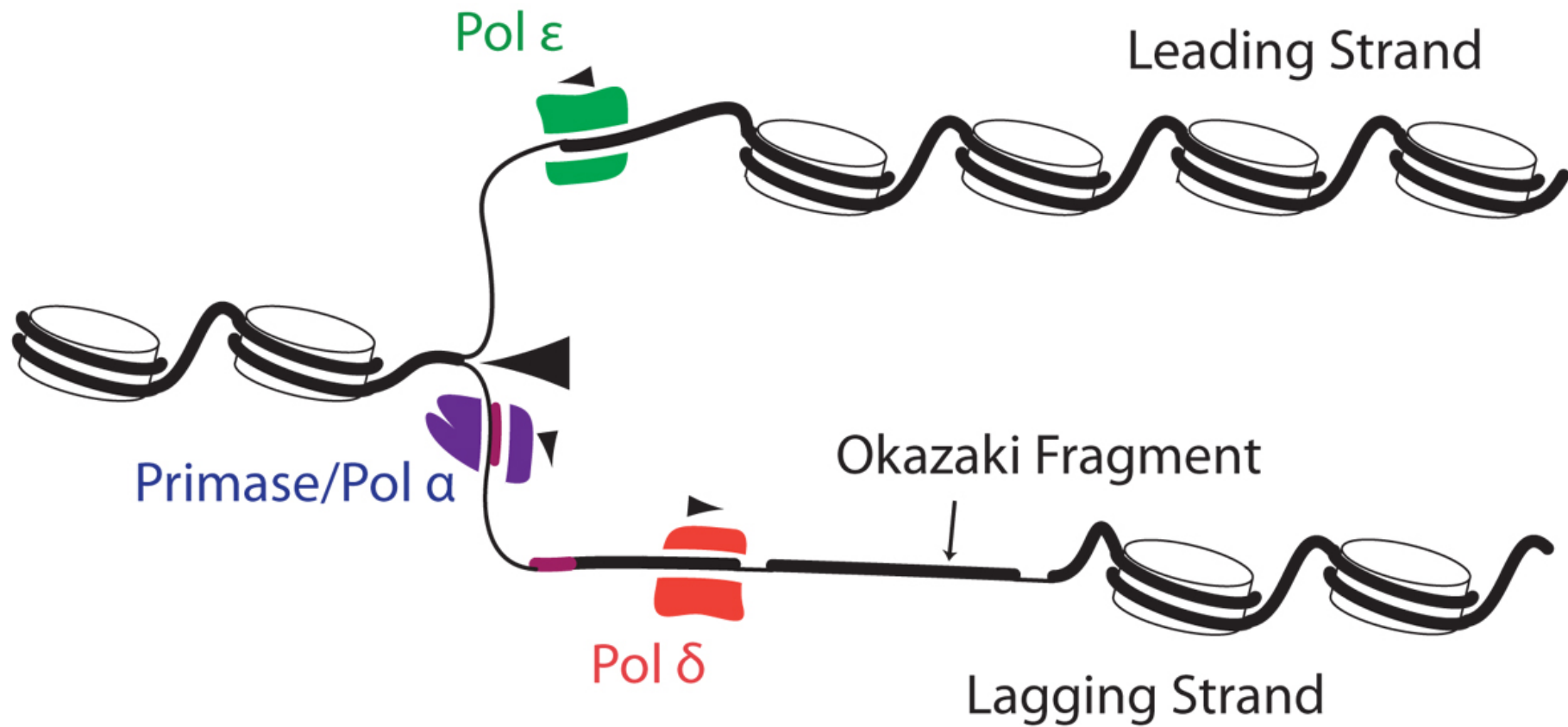
Ser 1 is invariably *N*-acetylated and may also be *O*-phosphorylated;

Lys residues 5, 8, 12, and 16 may be *N*-acetylated;

and Lys 20 may be mono- or di-*N*-methylated.

[After DeLange, R.J., Fambrough, D.M., Smith, E.L., and Bonner, J., *J. Biol. Chem.* **244**, 5678 (1969).]

DNA replication in eukaryotic cells



<i>POLY-MERASE</i>	FUNCTION	PROOF-READING*
<i>Pol</i> α (alpha)	● Contains <i>primase</i> ● Initiates DNA synthesis	—
<i>Pol</i> β (beta)	● Repair	—
<i>Pol</i> δ (delta)	● Elongates Okazaki fragments of the lagging strand	+
<i>Pol</i> ϵ (epsilon)	● Elongates the leading strand	+
<i>Pol</i> γ (gamma)	● Replicates mitochondrial DNA	+

Nucleosome structure

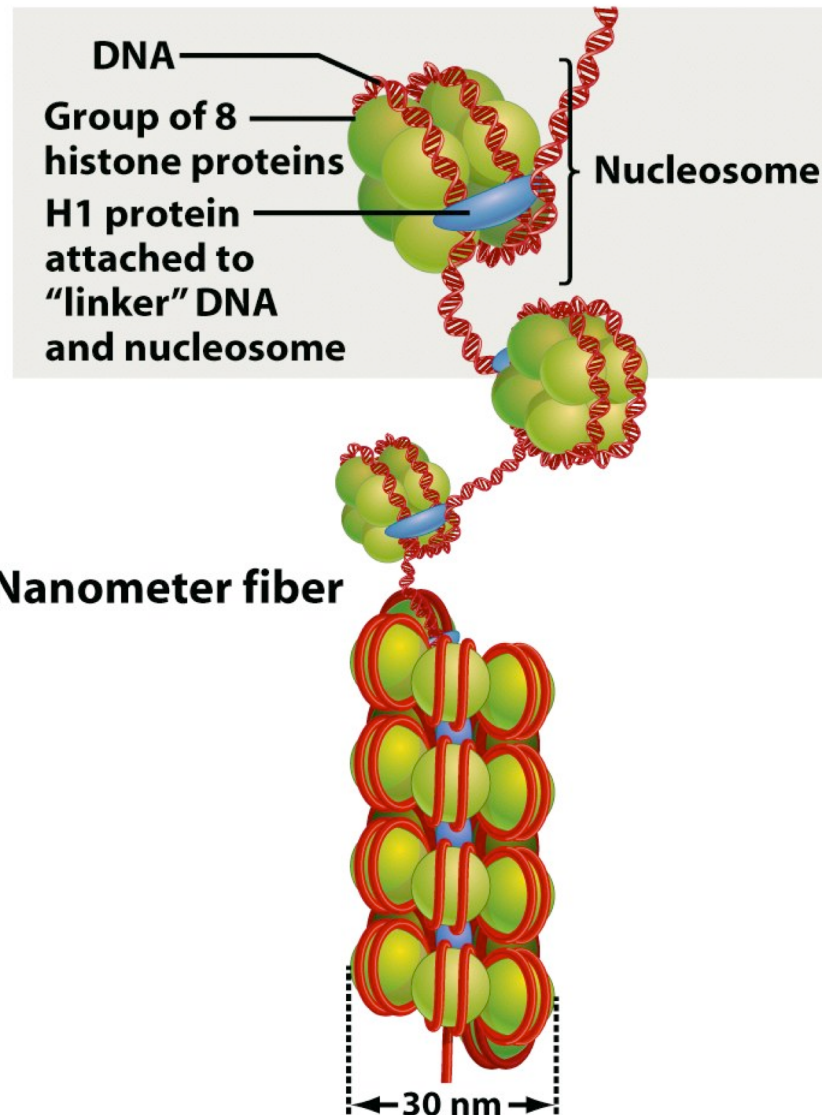
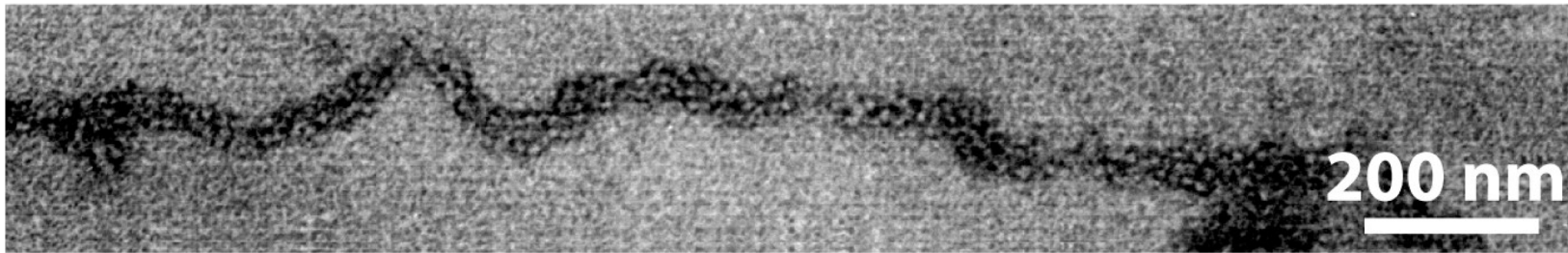


Figure 18-2bc Biological Science, 2/e



Condensed chromatin

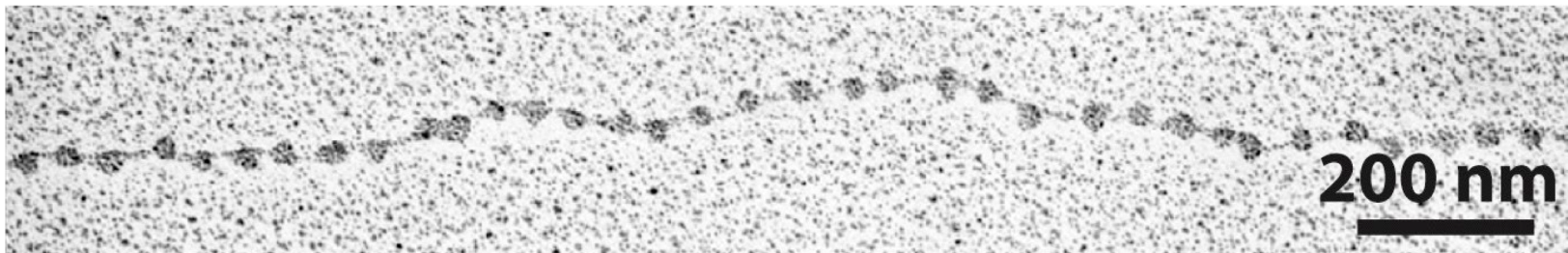
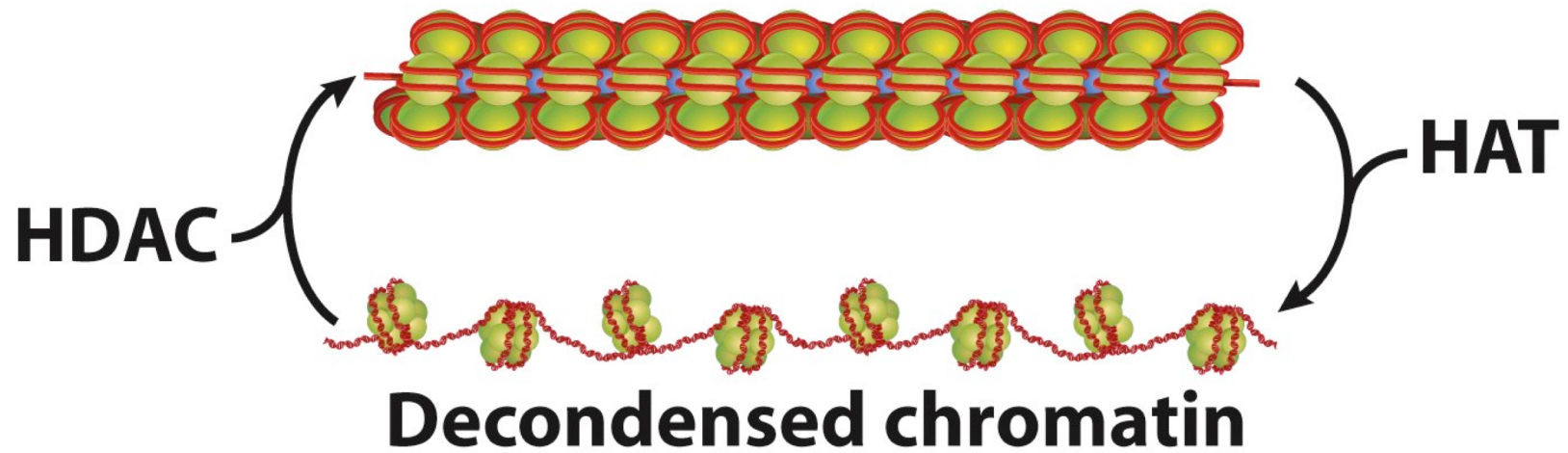
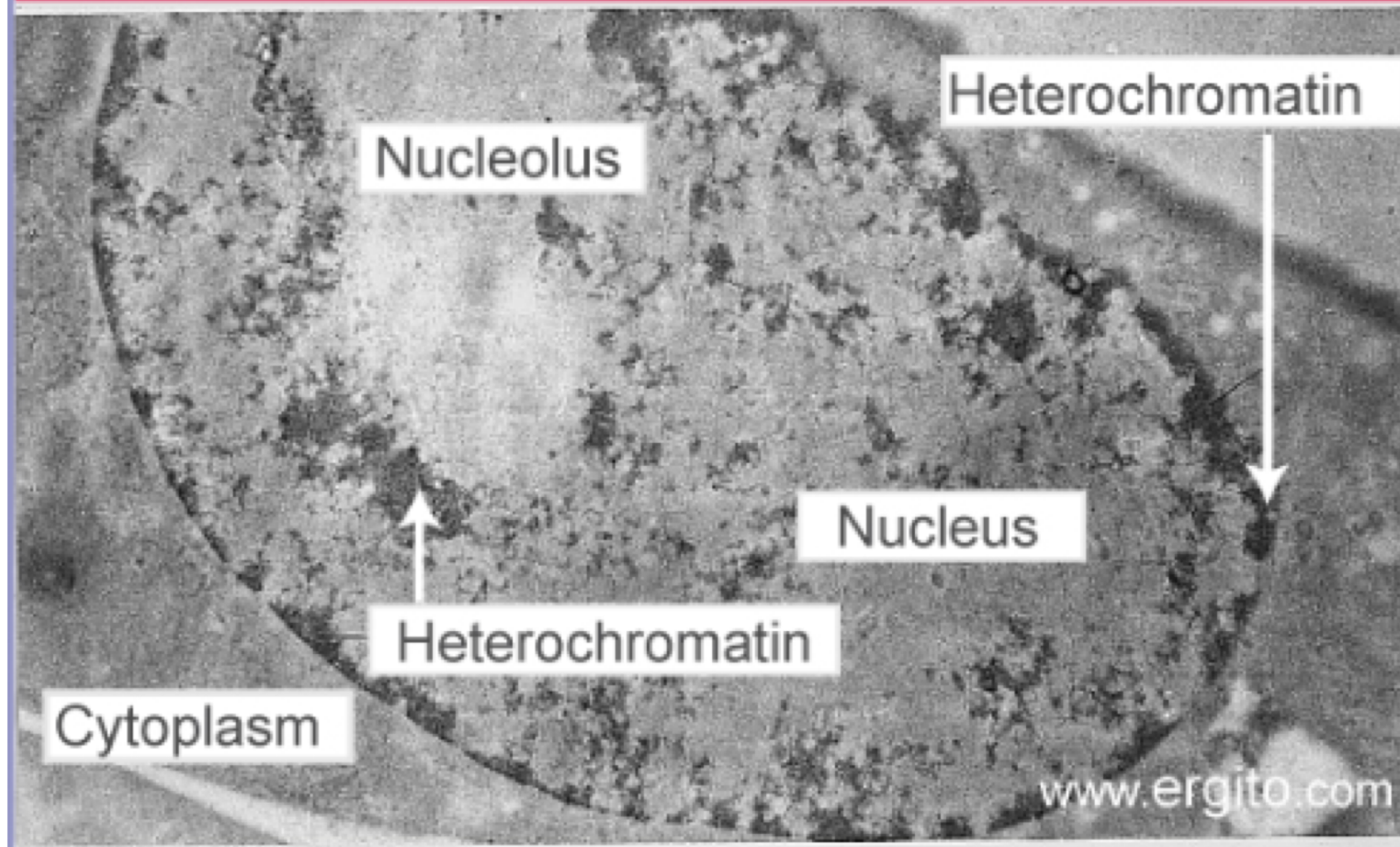
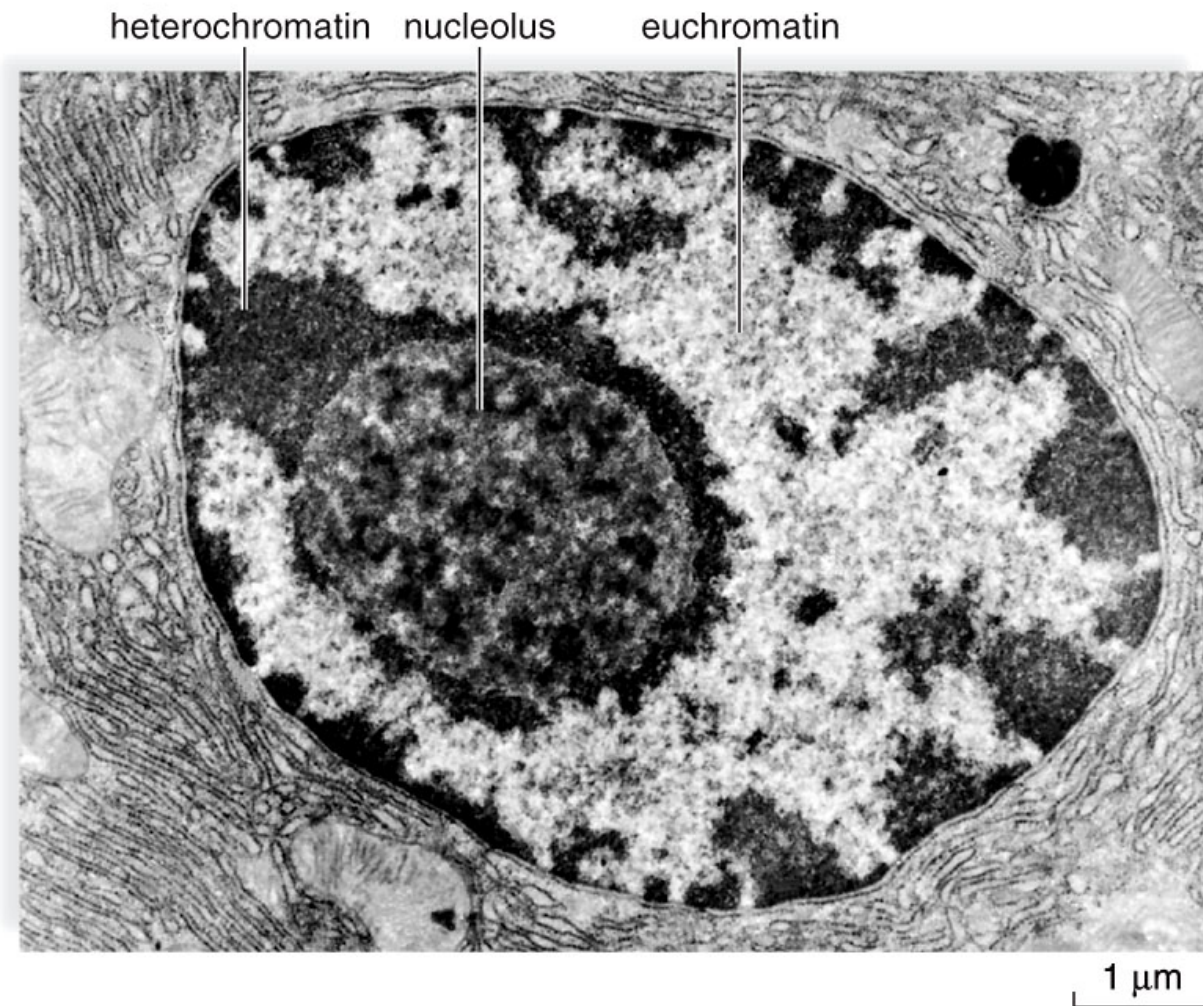


Figure 18-4 Biological Science, 2/e

Heterochromatin forms localized clusters

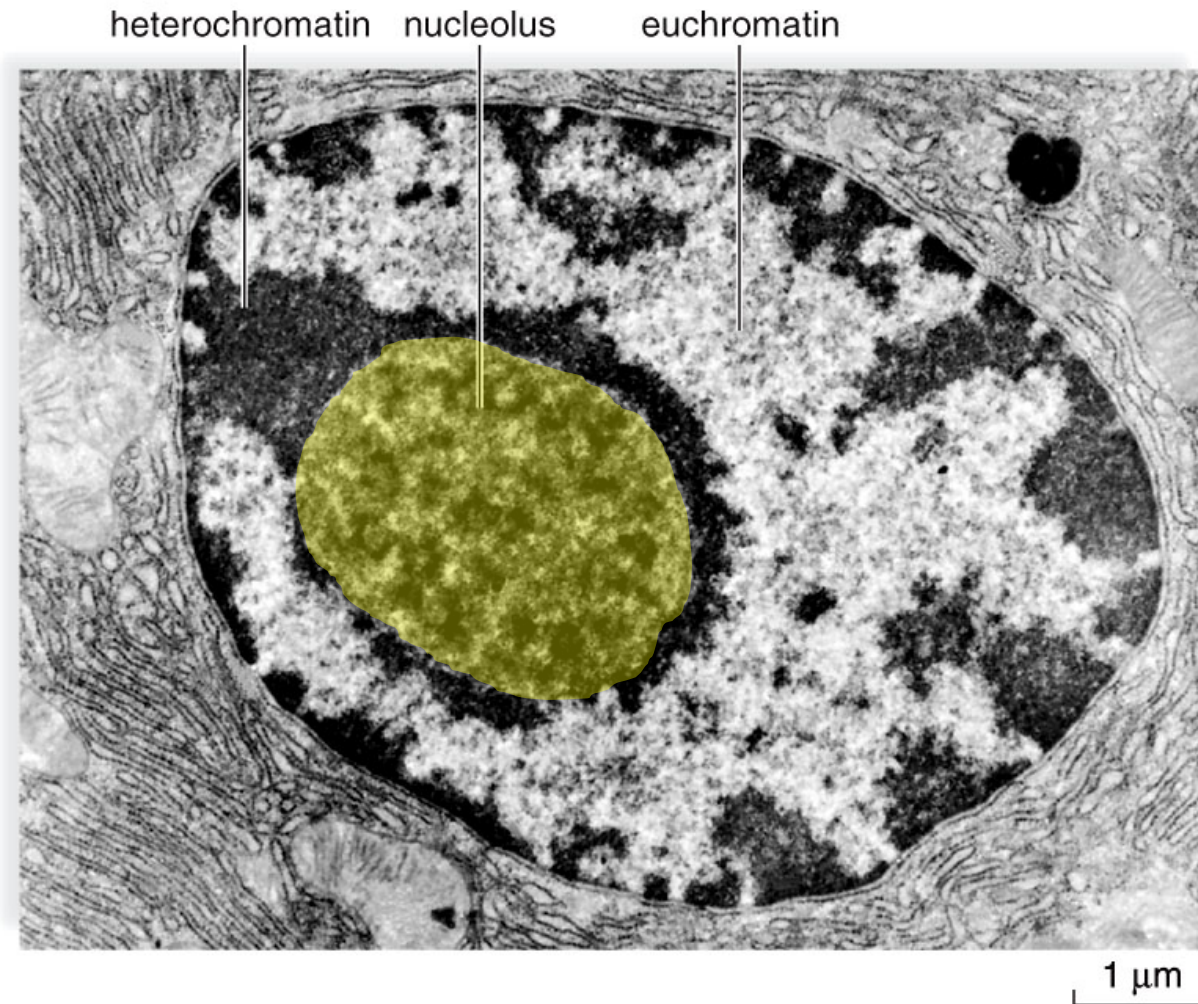


Nuclear content appears heterogenous :



b. Darkly-stained heterochromatin and lightly-stained euchromatin

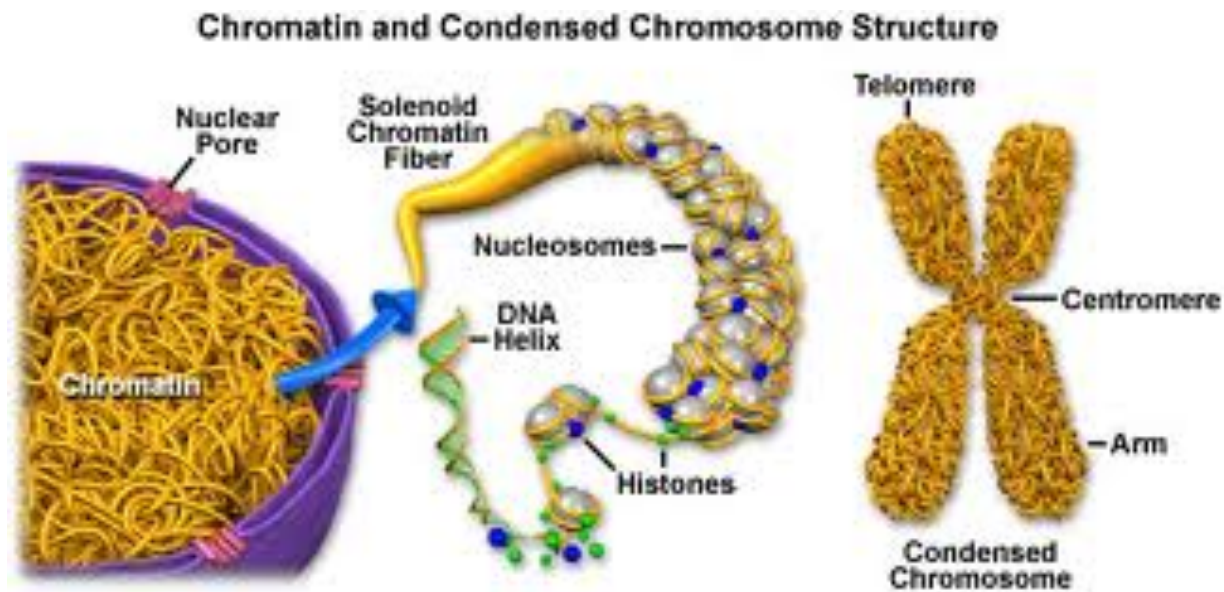
Nuclear content appears hétérogenous :



Nucleolus :
ribosomes assembly

b. Darkly-stained heterochromatin and lightly-stained euchromatin

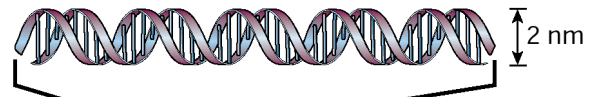
Compaction de l'ADN au cours du cycle cellulaire



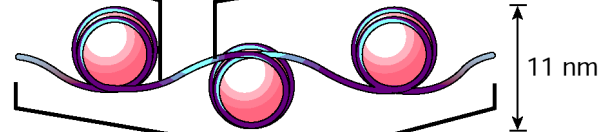
interphase

metaphase
(mitosis)

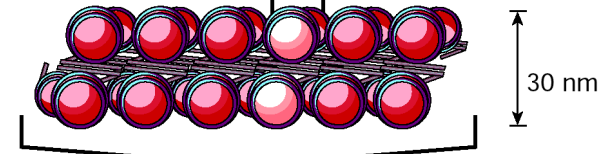
Short region of
DNA double helix



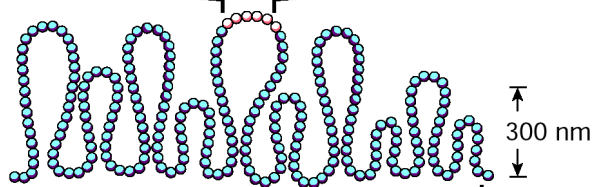
"Beads on a string"
form of chromatin



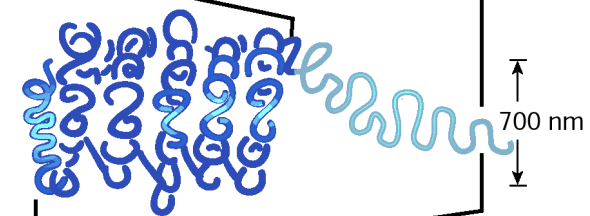
30-nm chromatin
fibre of packed
nucleosomes



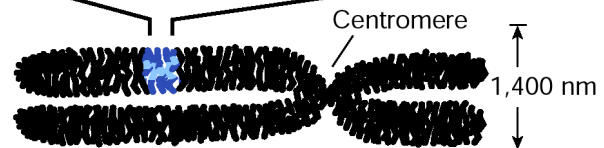
Section of
chromosome in an
extended form



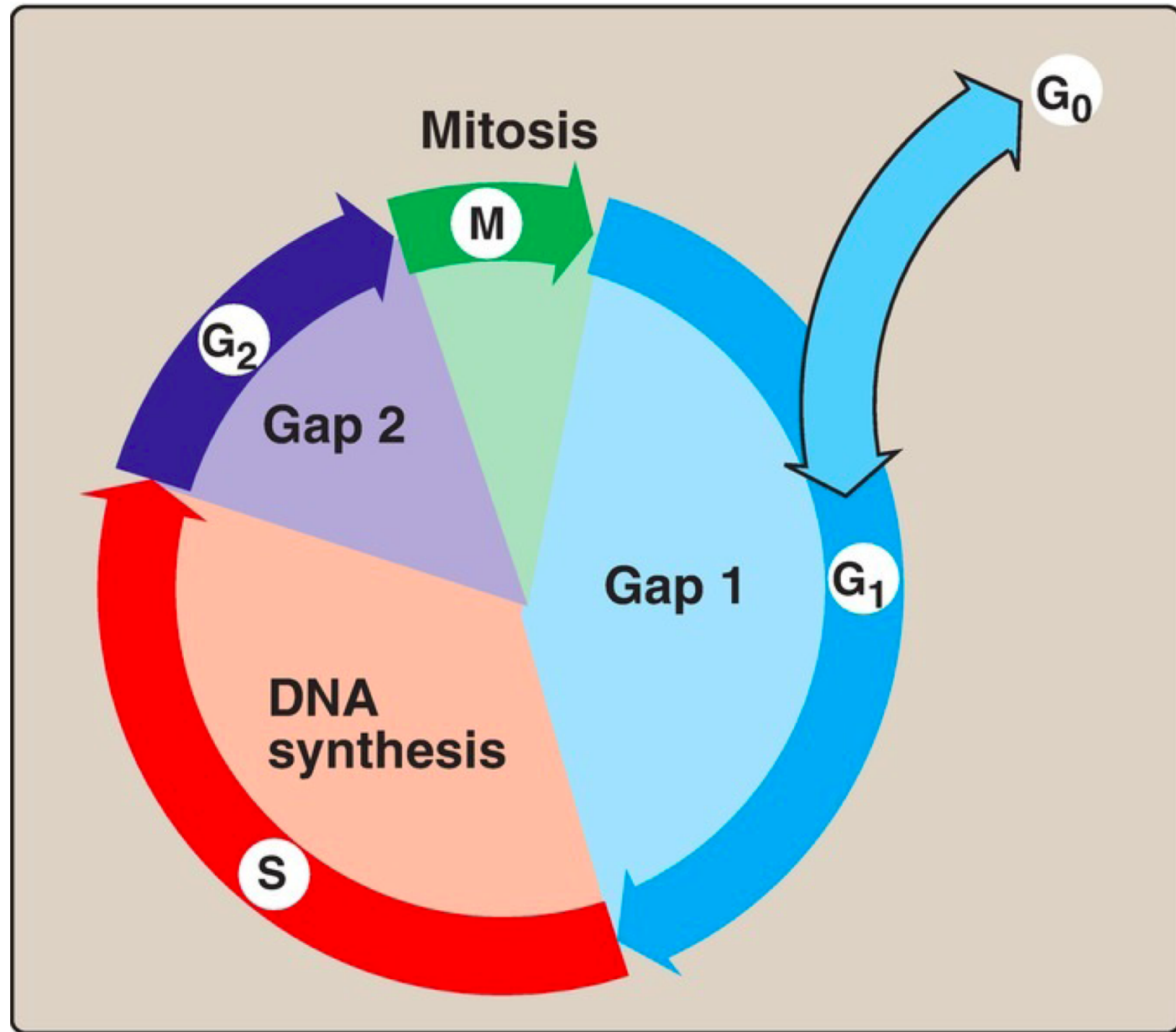
Condensed section
of chromosome



Entire mitotic
chromosome



Cell cycle :



Cell cycle :

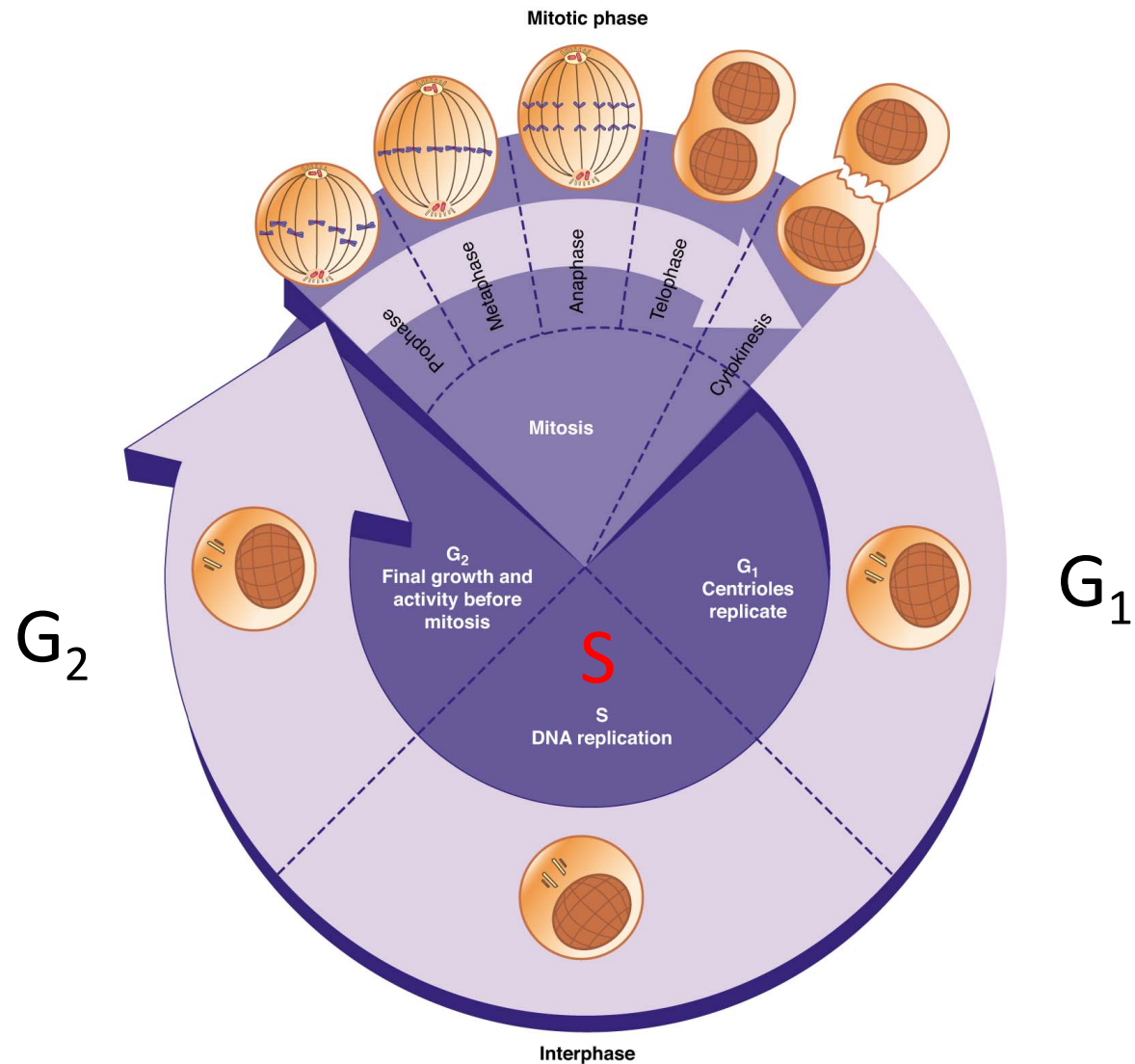


FIGURE 1-13 Sequence of events during the cell cycle. Immediately following mitosis (M) the cell enters a gap phase (G₁) before a DNA synthesis phase (S) a second gap phase (G₂) and back to mitosis. Collectively G₁, S, and G₂ phases are referred to as interphase (I).

https://wps.prenhall.com/wps/media/objects/1109/1135980/9_1_1_plain.html

Tutorial 9
parts 3 and 4

Principle of FISH analysis

Chromosome preparation on microscope slide



Denature DNA *in situ*

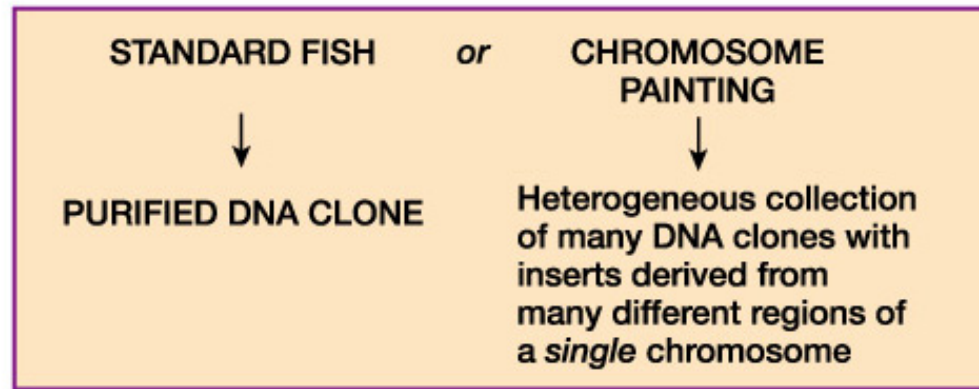


Allow to anneal, expose to UV and visualize fluorescence *in situ*



Single probe bound

Chromosome paint bound



Label by incorporating nucleotides with attached fluorophore; denature

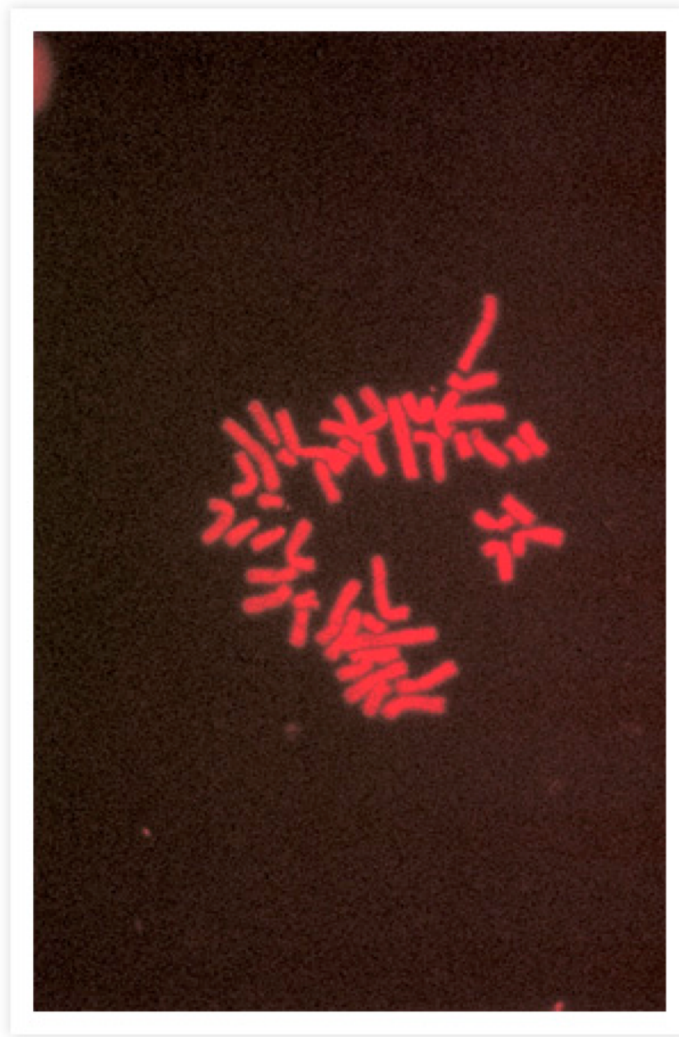
Label single probe

Chromosome paint

DNA probes are hybridized to fixed cells on microscope slides.

The hybridization to target loci is visualized by the detection of fluorescent signals

on metaphase chromosomes or interphase nuclei.



Human metaphase chromosomes stained with propidium iodide

Alkaline DNA denaturation

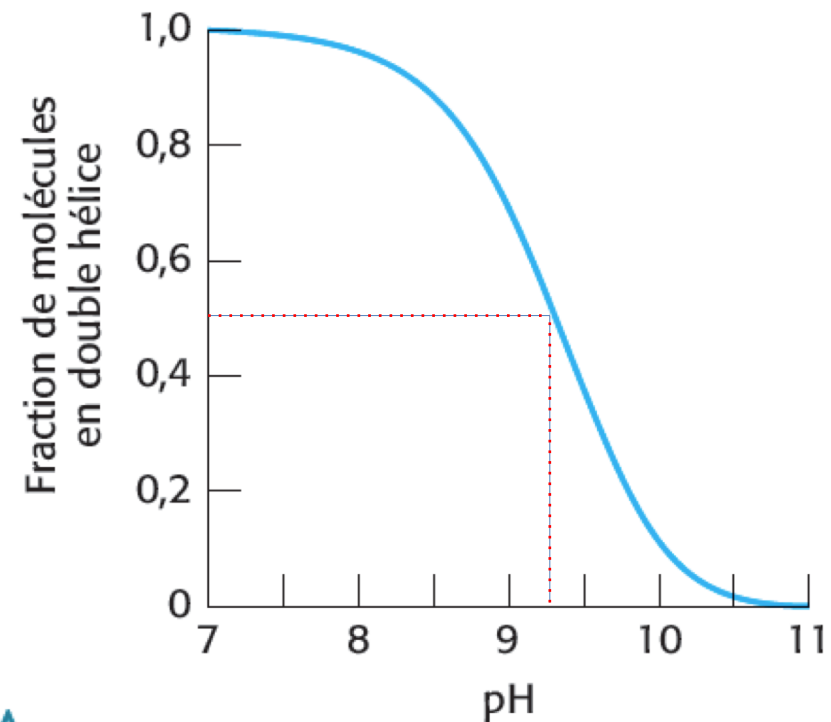
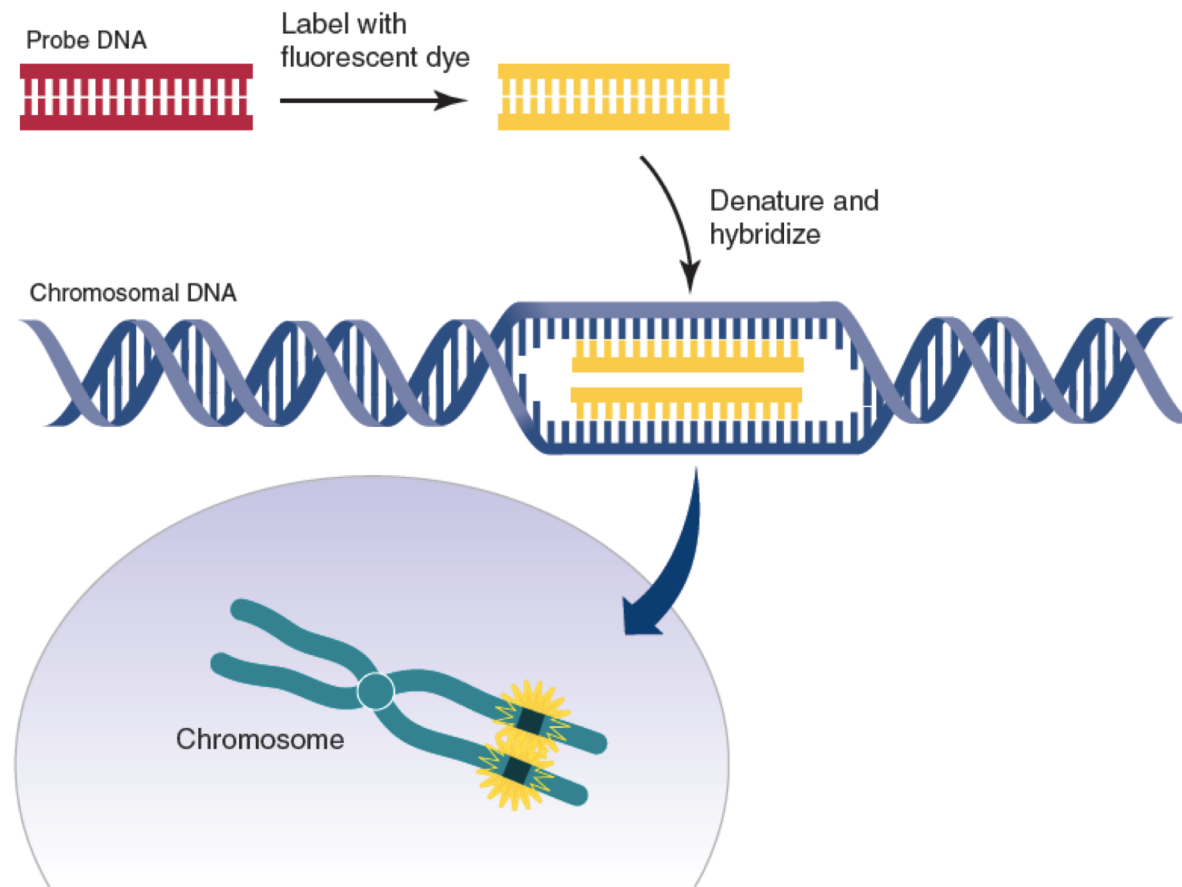
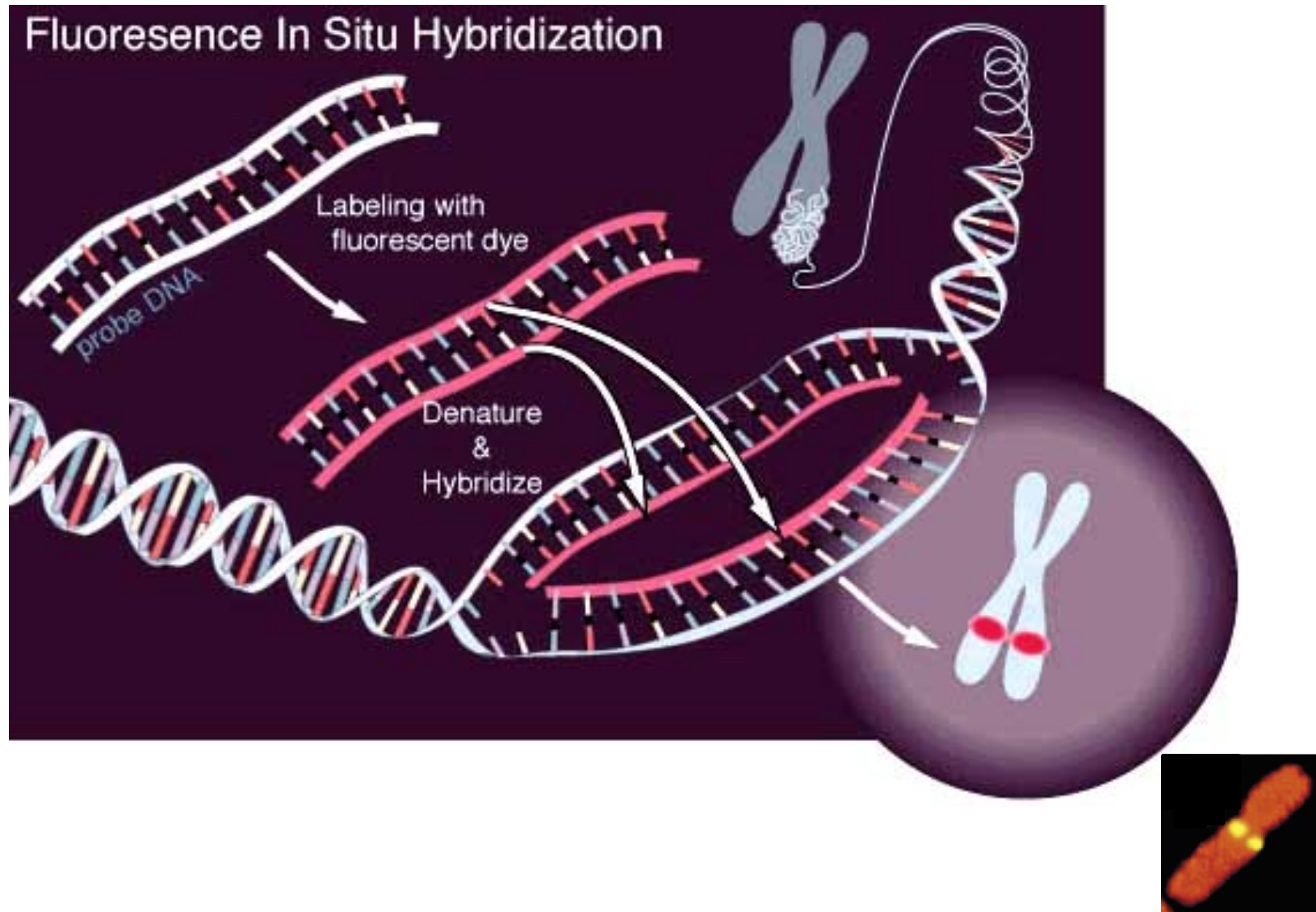


Figure 1.16 Dénaturation du DNA par l'addition d'une base. L'addition d'une base à une solution de DNA en double hélice initialement à pH 7 provoque la séparation de la double hélice en simples brins. Le processus est à moitié complet un peu au-dessus de pH 9.

FISH

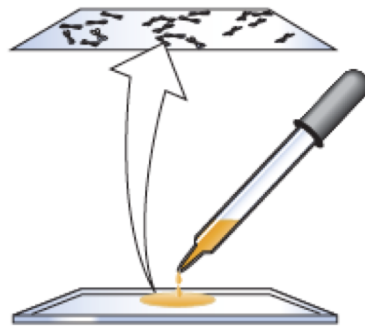


Fluorescence In Situ Hybridization

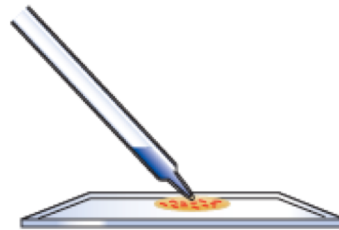


Karyotype

(a)



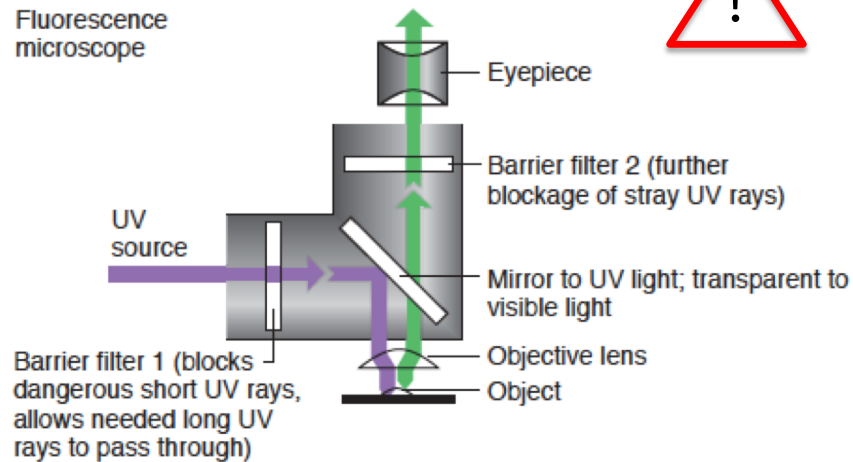
1. Drop cells onto a glass slide.



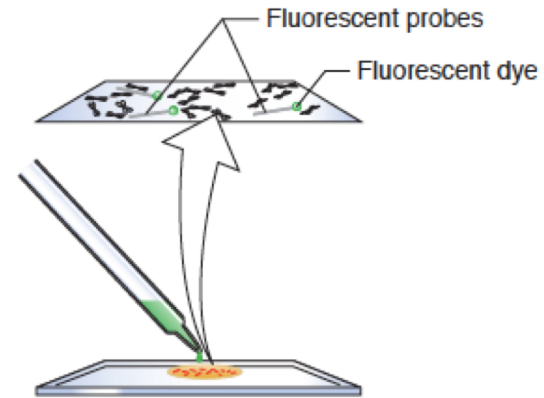
2. Gently denature DNA by treating briefly with DNase.



Fluorescence microscope

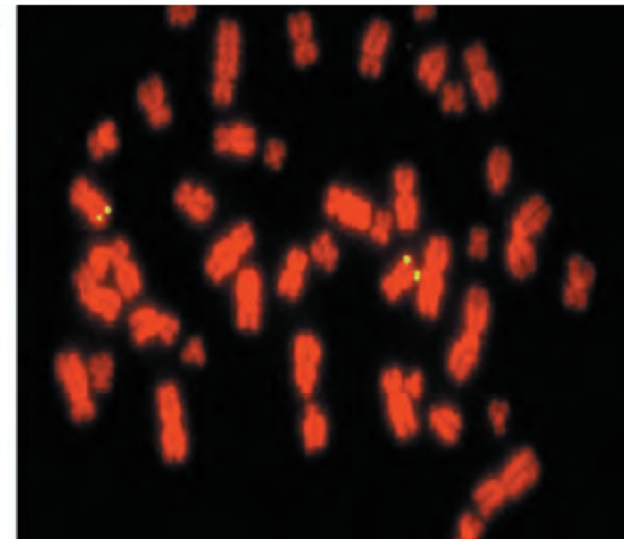


4. Expose to ultraviolet (UV) light.
Take picture of fluorescent chromosomes.



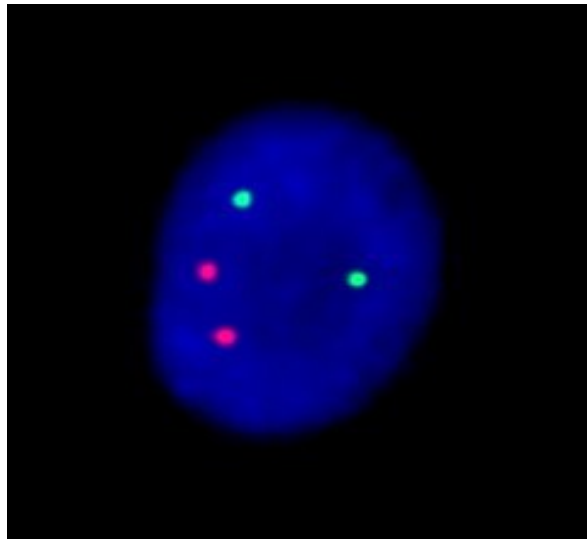
3. Add hybridization probes labeled with fluorescent dye and wash away unhybridized probe.

(b)



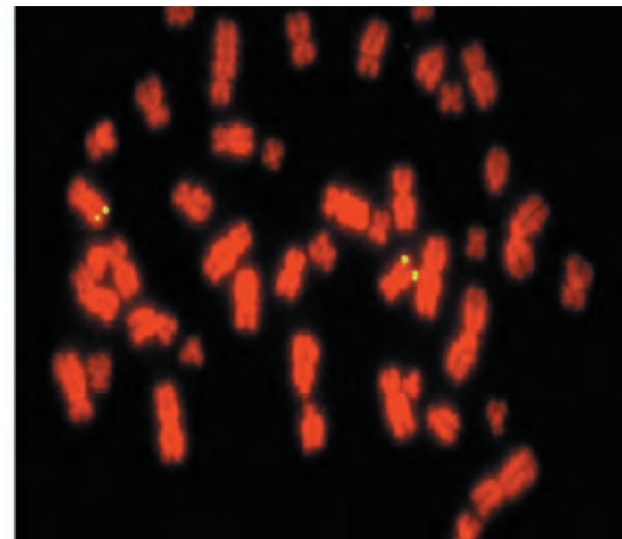
FISH : Fluorescence in situ hybridization

Nucleus in **interphase**



2 probes

Chromosomes in **metaphase**



1 probe

a



Gene-specific
probe

b



Centromeric
probe

c



Telomeric
probe

d



Chromosome-painting
probe

Repetitive-sequence probes

Examples of different types of fluorescence in situ hybridisation (FISH) probes

Expert Reviews in Molecular Medicine © 2000 Cambridge University Press



Nucleus in interphase

**Sonde
centromérique**

b



Centromeric
probe

Examples of different types of fluorescence in situ hybridisation (FISH) probes

Expert Reviews in Molecular Medicine © 2000 Cambridge University Press

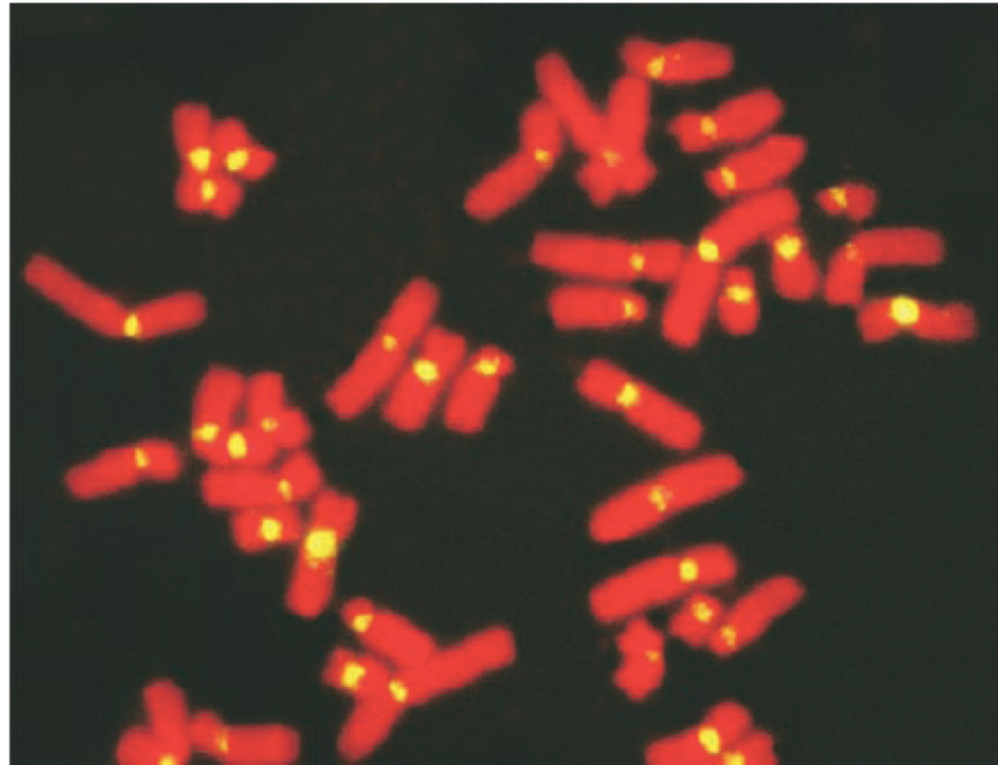


FIGURE 10-18 Fluorescent *in situ* hybridization (FISH) of human metaphase chromosomes. The probe, specific to centromeric DNA, produces a yellow fluorescence signal indicating hybridization. The red fluorescence is produced by propidium iodide counterstaining of chromosomal DNA.

c



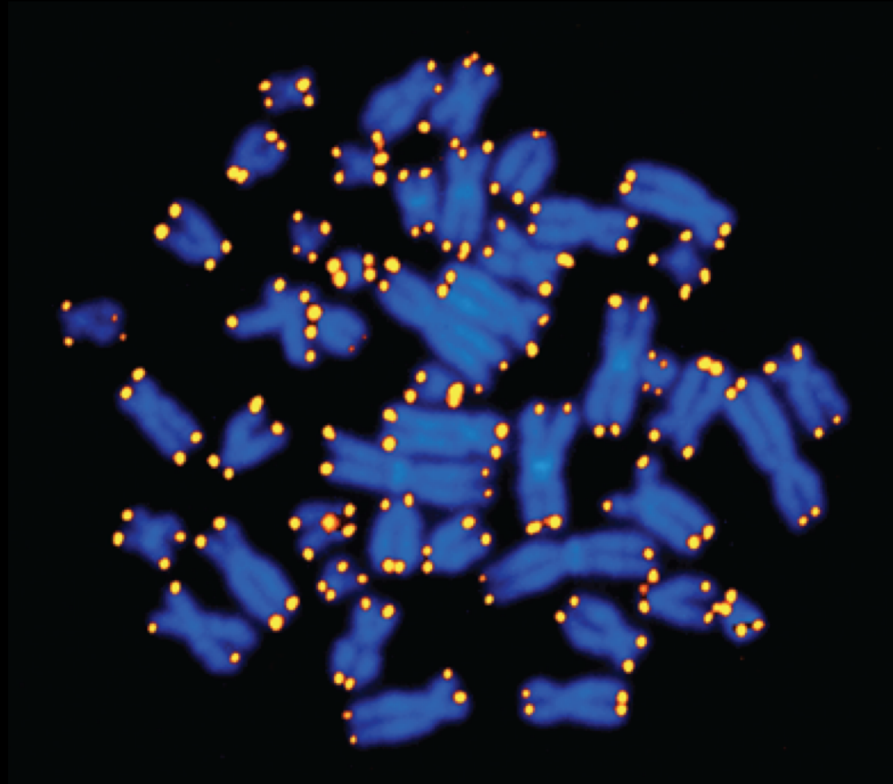
Telomeric
probe

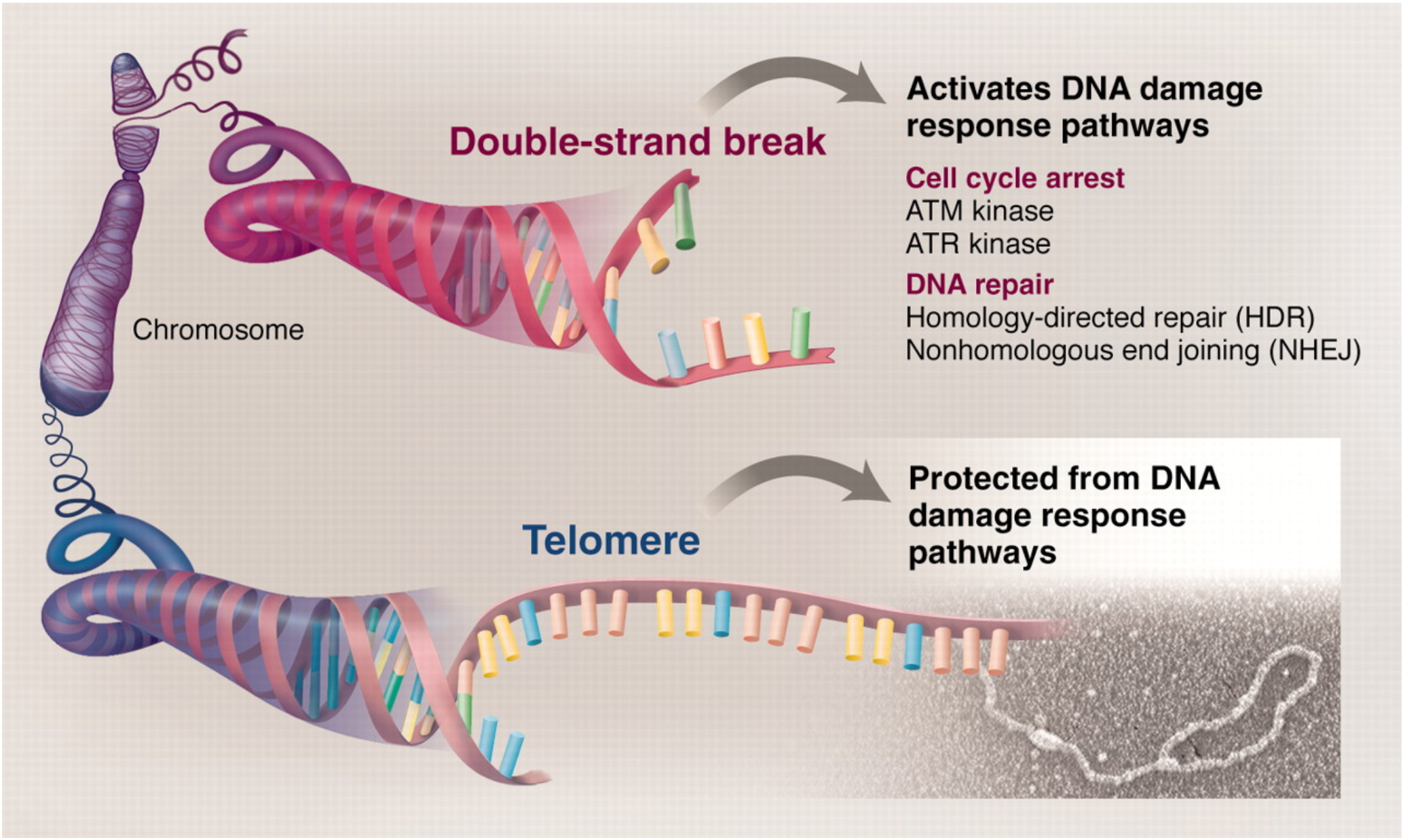
Repetitive-sequence probes

Examples of different types of fluorescence in situ hybridisation (FISH) probes

Expert Reviews in Molecular Medicine © 2000 Cambridge University Press

Telomers :





The diagram illustrates the difference in DNA damage response between a general chromosome and a telomere. On the left, a purple chromosome is shown with a red DNA double helix. A section of the red helix is broken, labeled 'Double-strand break'. An arrow points from this break to a list of cellular responses. Below this, a blue telomere is shown with a similar red DNA double helix, but it is covered by a dense layer of orange and yellow protein caps. An arrow points from the telomere to a list indicating it is protected from DNA damage response pathways. A grayscale electron micrograph of a DNA molecule is visible in the bottom right corner.

Double-strand break

Activates DNA damage response pathways

Cell cycle arrest

ATM kinase

ATR kinase

DNA repair

Homology-directed repair (HDR)

Nonhomologous end joining (NHEJ)

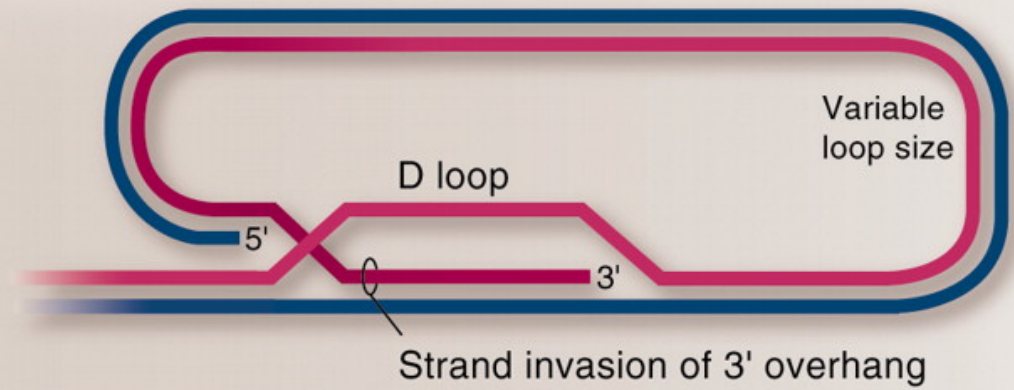
Telomere

Protected from DNA damage response pathways

Telomere



t-loop



Shelterin

