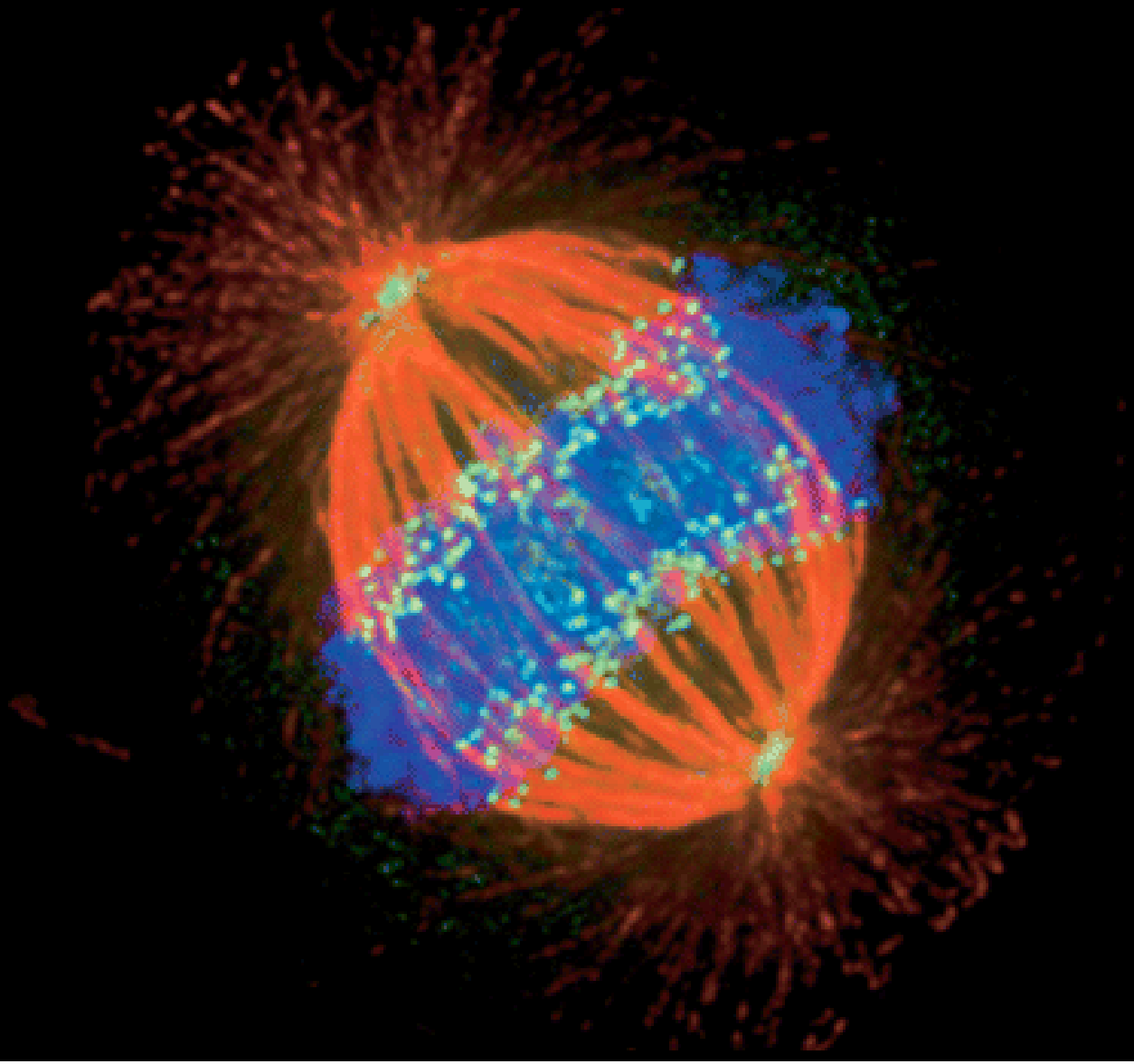
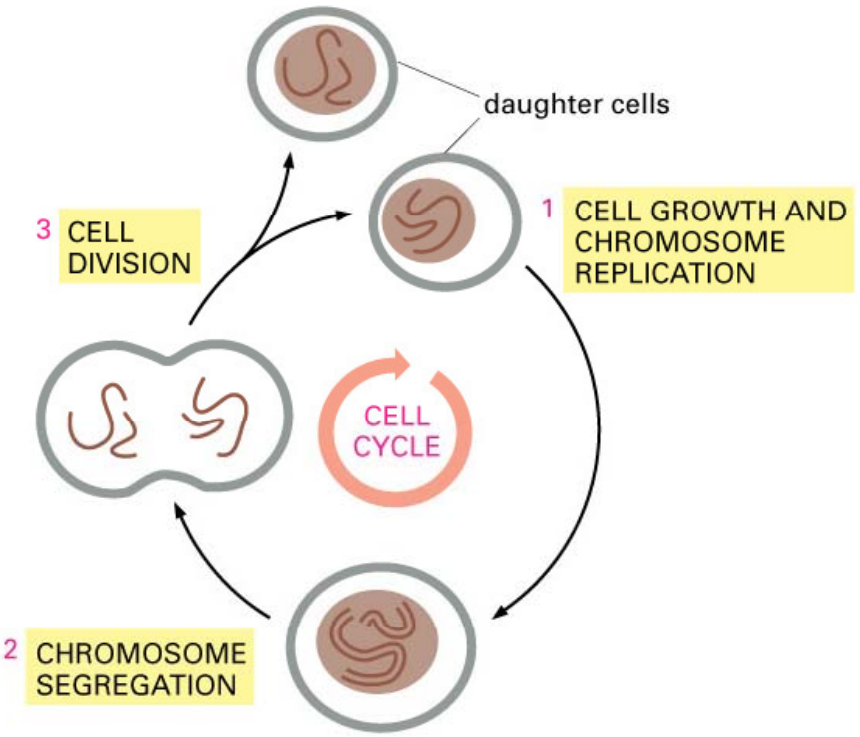
# Cell division and mitosis

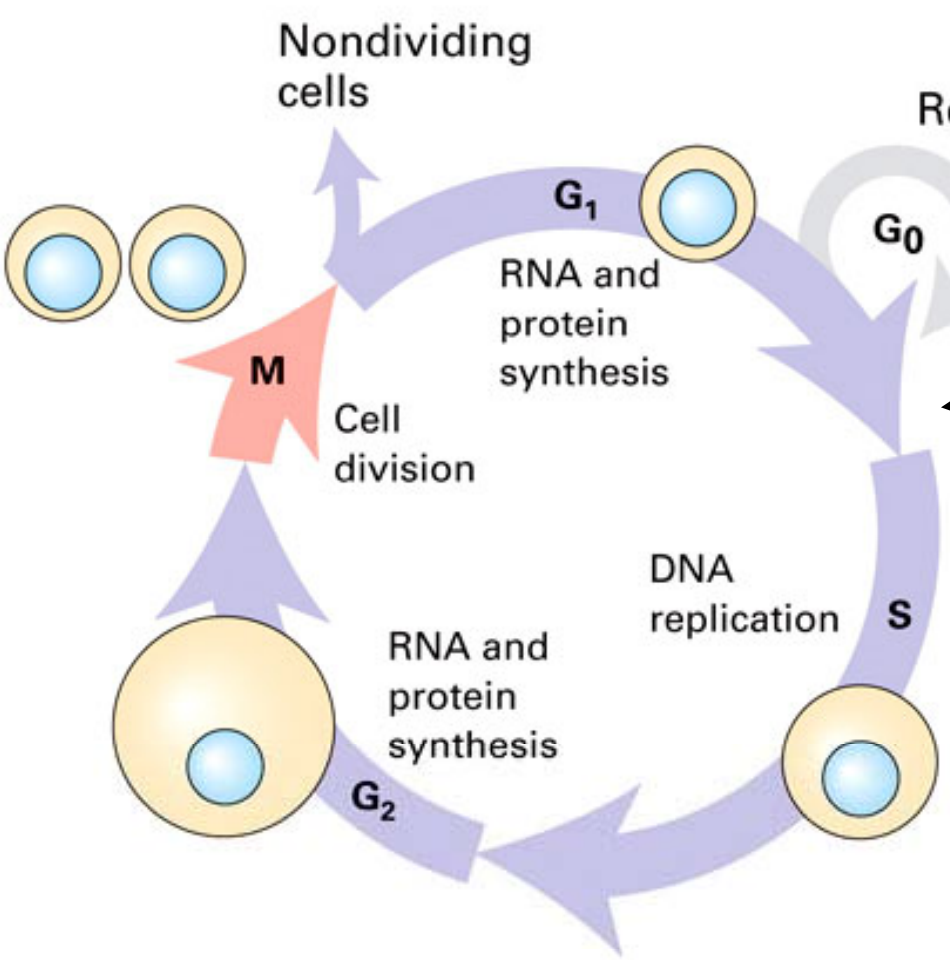


# Universal functions of the cell cycle

- Replicate the DNA.
- **Segregate it into daughter cells.**
- **Replicate and/or distribute organelles into daughter cells.**
- Grow.



# The Cell Cycle



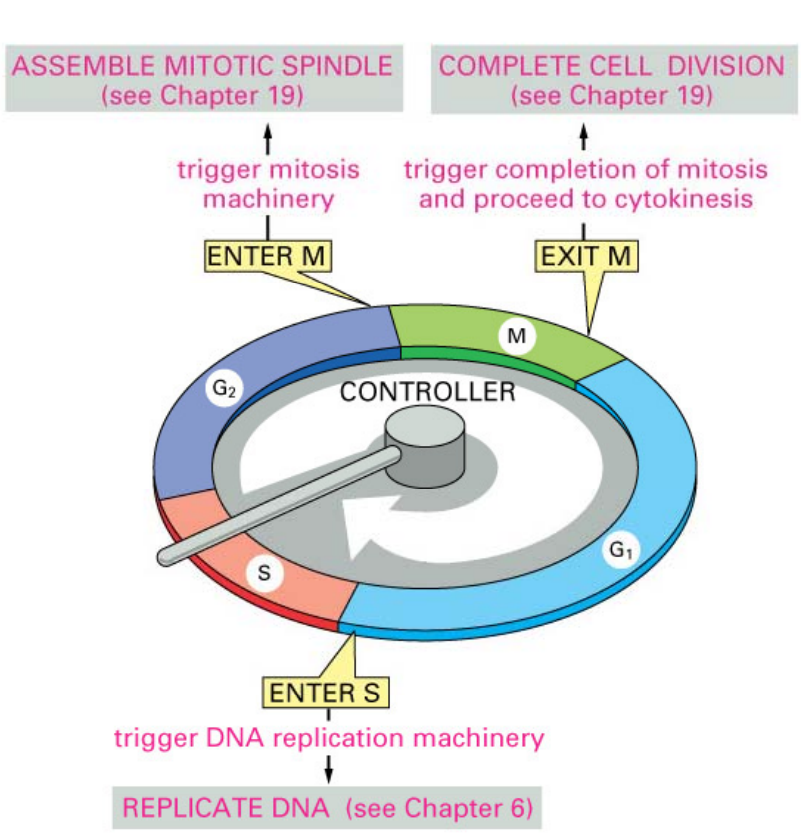

**Growth factors**

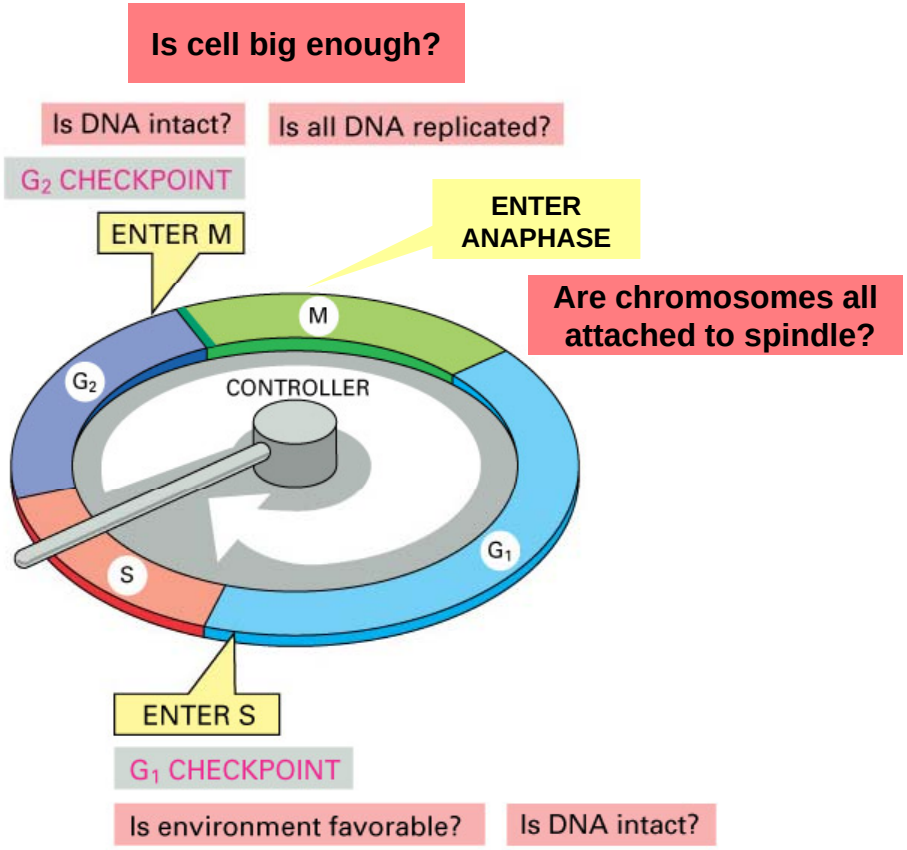
**Mitogens release "brakes"**

**-Cyclin-dependent kinases WITHIN cells**
**-Signals from OTHER cells – communication!**
- **-mitogens**
- **-growth factors**
- **-survival factors**
- **-(avoid apoptosis/suicide)**

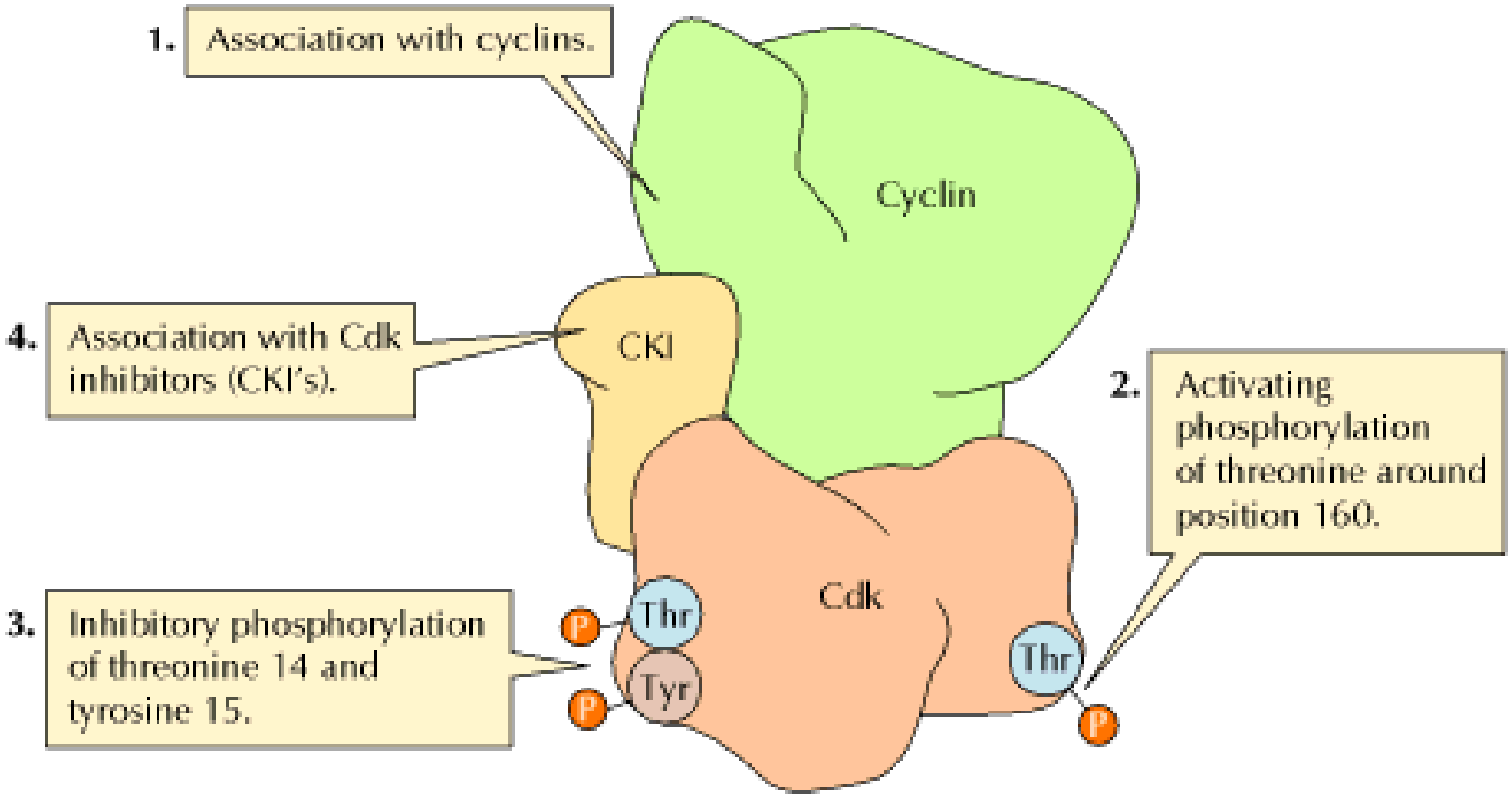
G1 or G0➔ hours (epithelial cells) to years (liver, nerve, bone, muscle cells)
Rest of cycle:➔ 12-24 hours

# Checkpoints



# Cyclin dependent kinases




…these go on to phosphorylate a team of regulatory proteins that can then activate other proteins in a signalling cascade.

In each of your organs, your cells are dividing as often as they are dying. **True or false?**

All cells in your body are approximately the same size. **True or false?**

Organ and body size are determined by:

A. Cell growth
B. Cell division
C. Cell death
D. B & C
E. A, B, & C

A cyclin is:

A. A kinase that binds its Cdk partner
B. A regulatory subunit that binds its Cdk partner
C. An extracellular signal

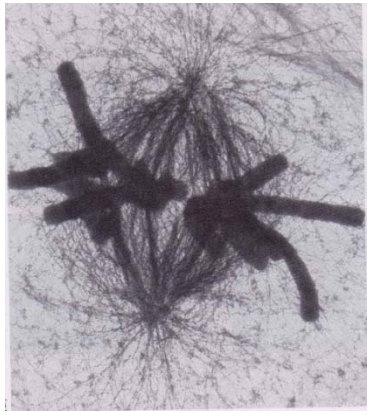
Apoptosis is:

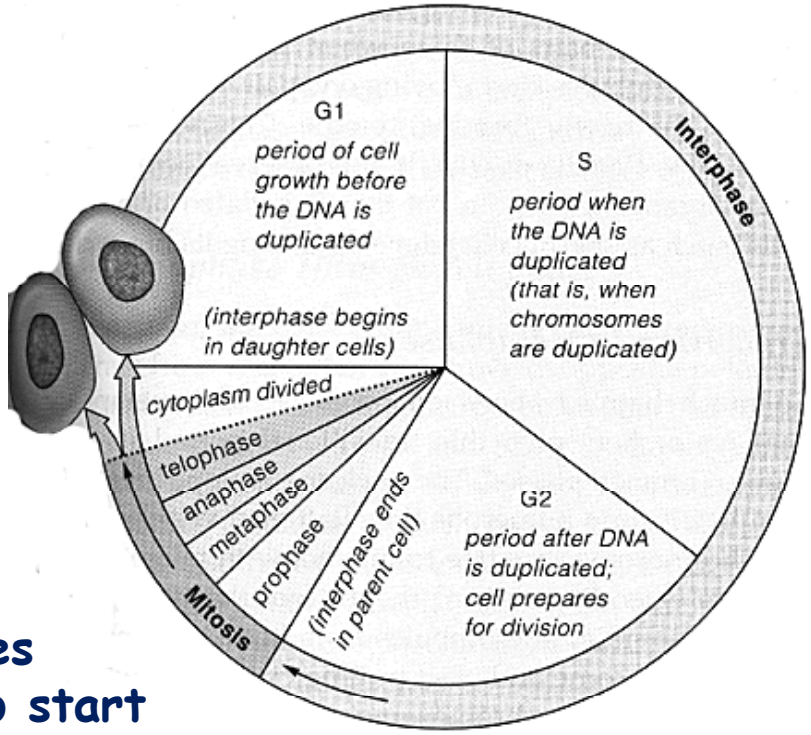
A. A default state inhibited by extracellular signals
B. Mediated by caspases
C. An explosive, uncontrolled event

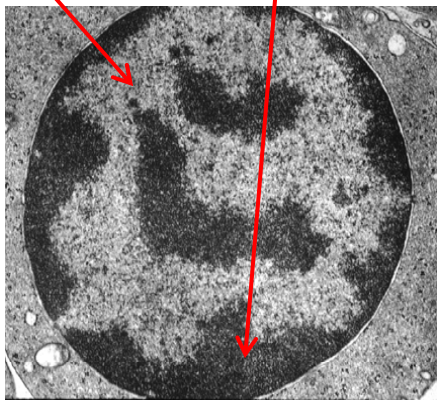
# Mitosis

Heterochromatin: more tightly packed chromatin

Euchromatin: less tightly packed chromatin (most of the genes are here)



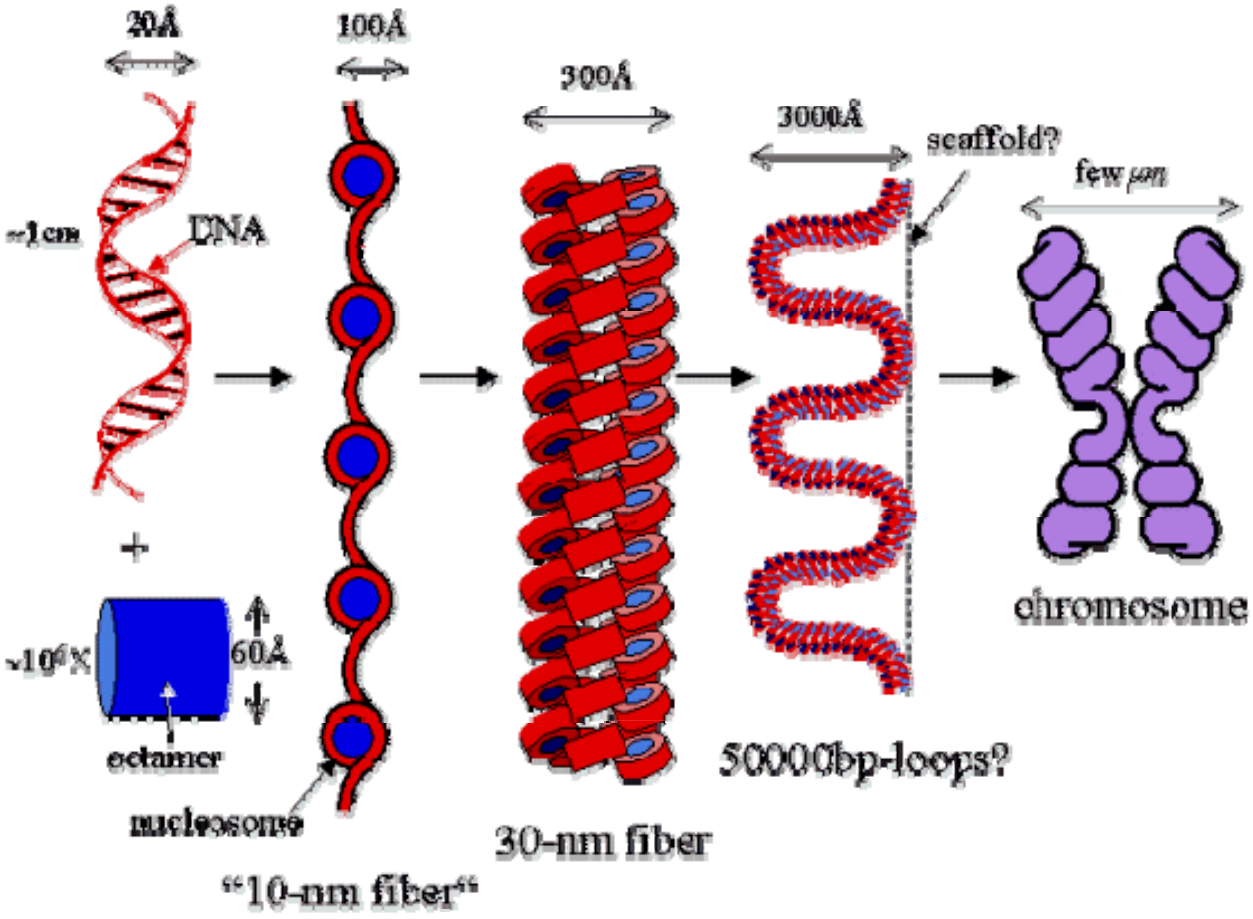


**Mitosis:**
**M-Cdk phosphorylates condensin subunits to start chromosome condensation**

**Interphase:**
**Chromosomes indistinct because decondensed**

# Chromosome condensation



***What essential process happens in S phase????***

# What essential process happens in S phase????

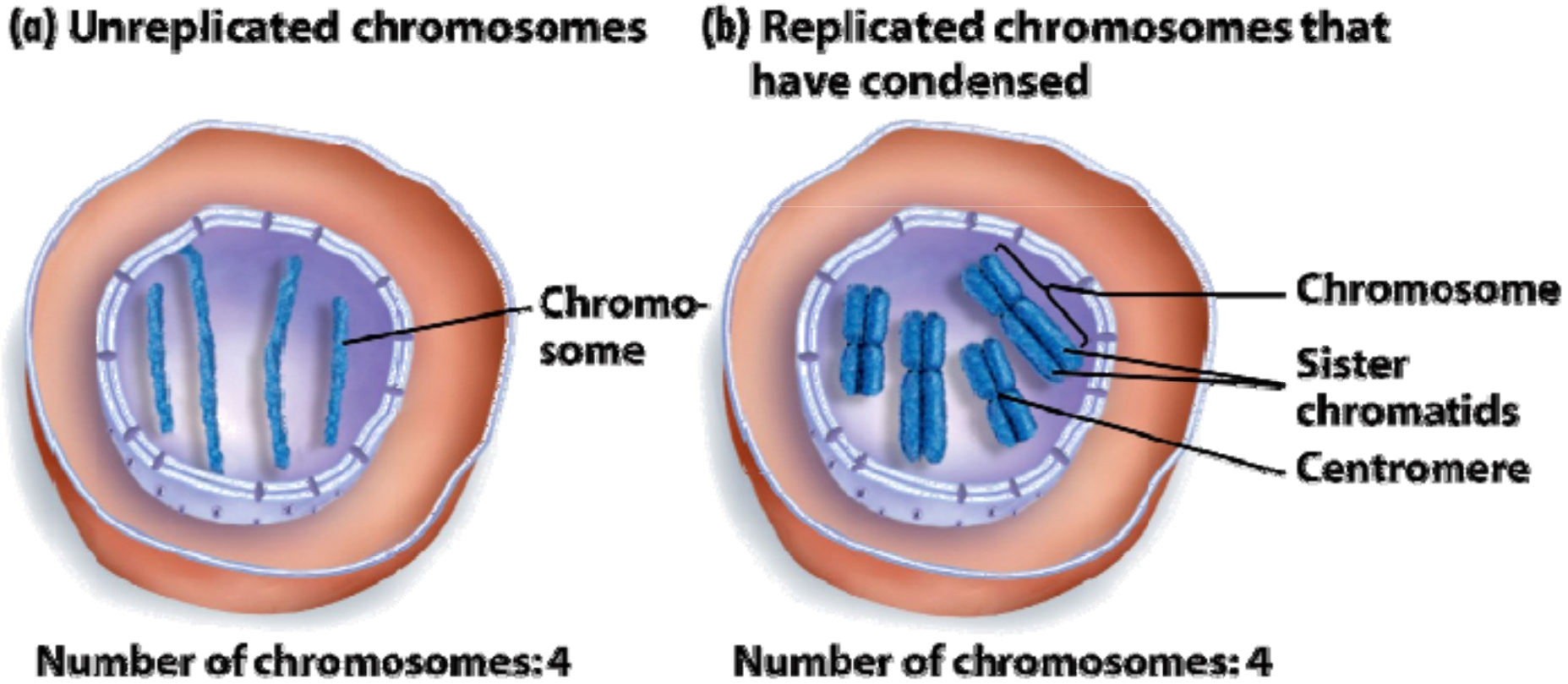
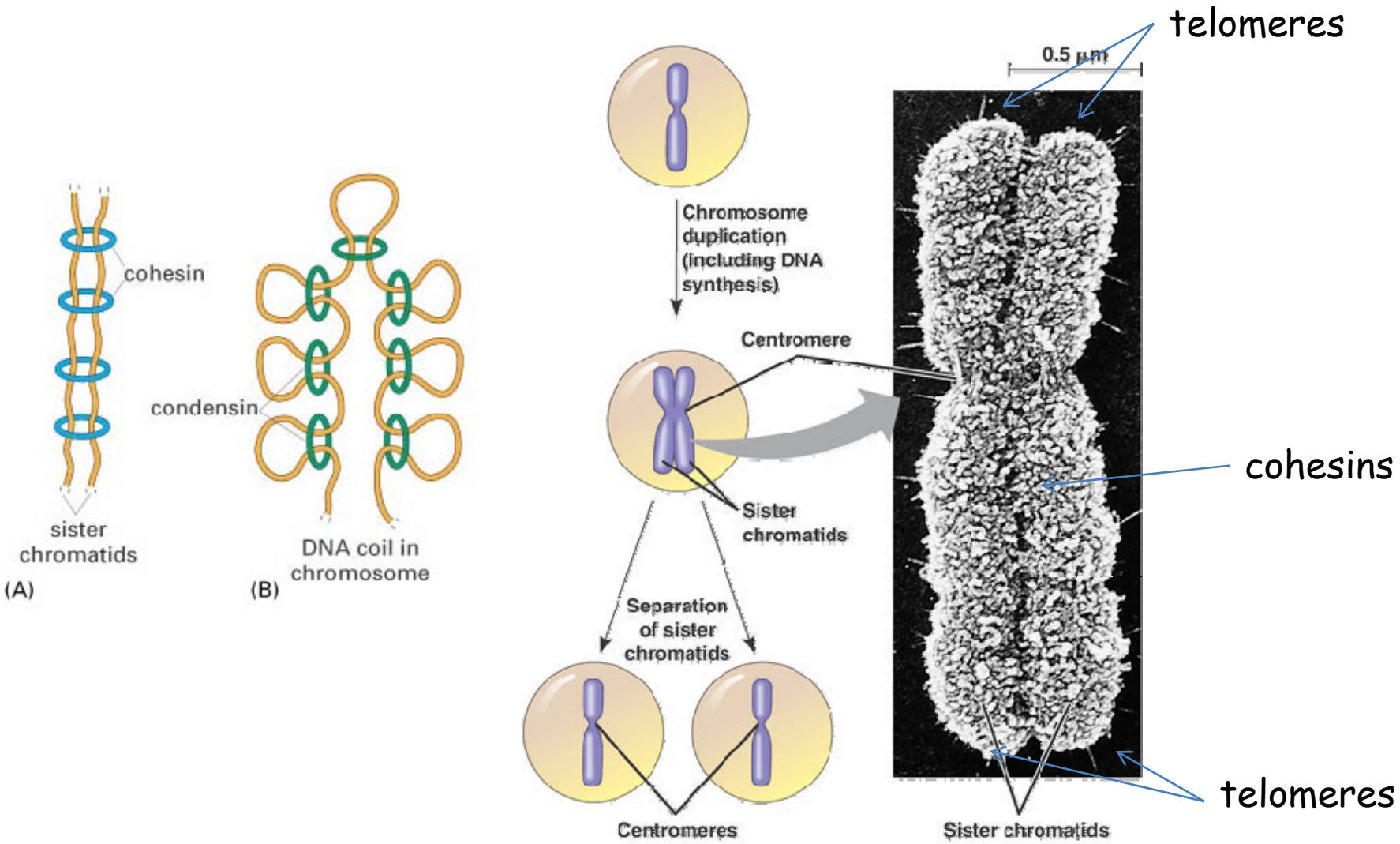



Figure 11-7 Biological Science, 2/e

© 2005 Pearson Prentice Hall, Inc.

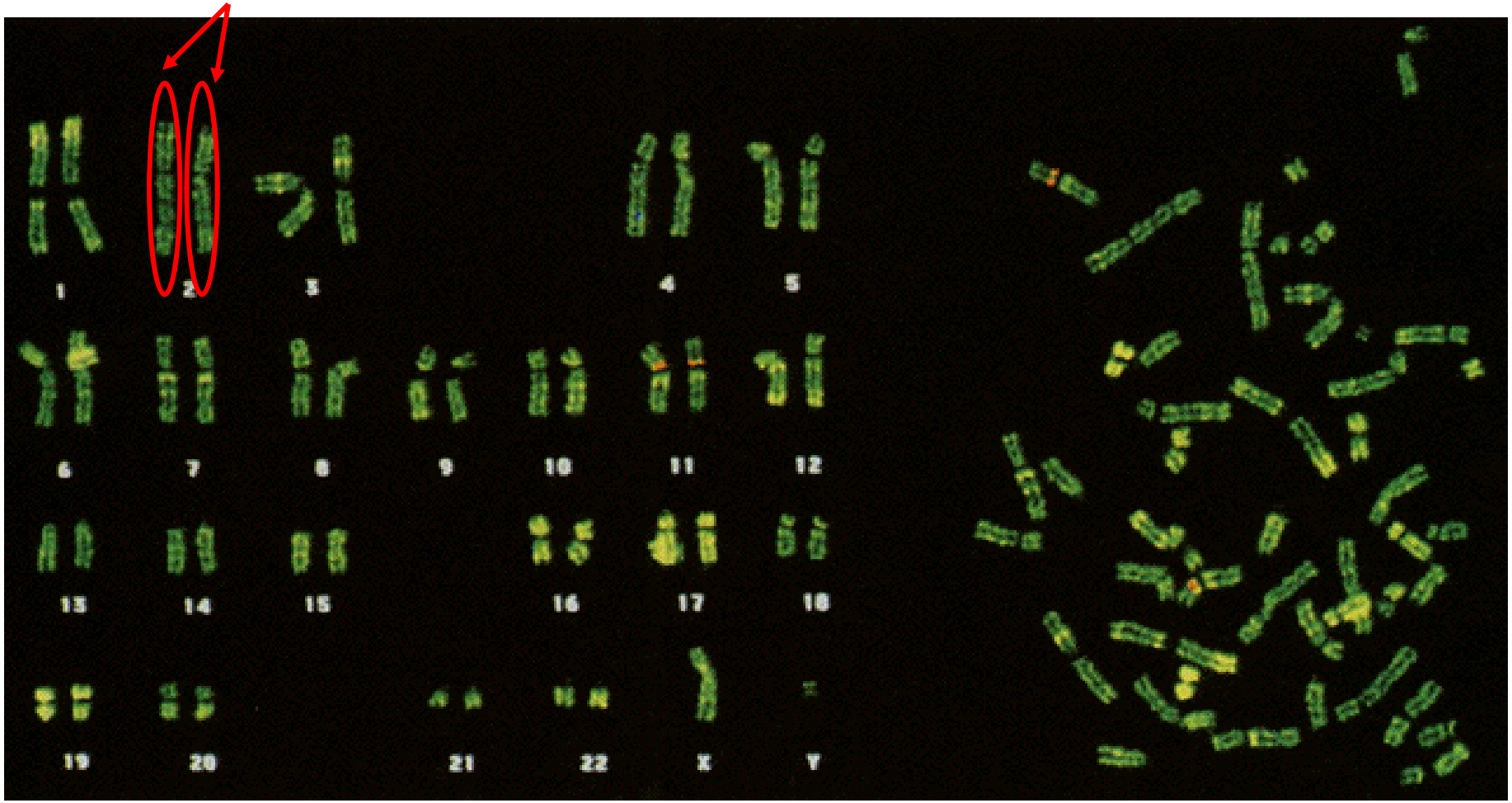
# Terminology



# humans are *diploid*

## two of each chromosome –*homologous pairs*

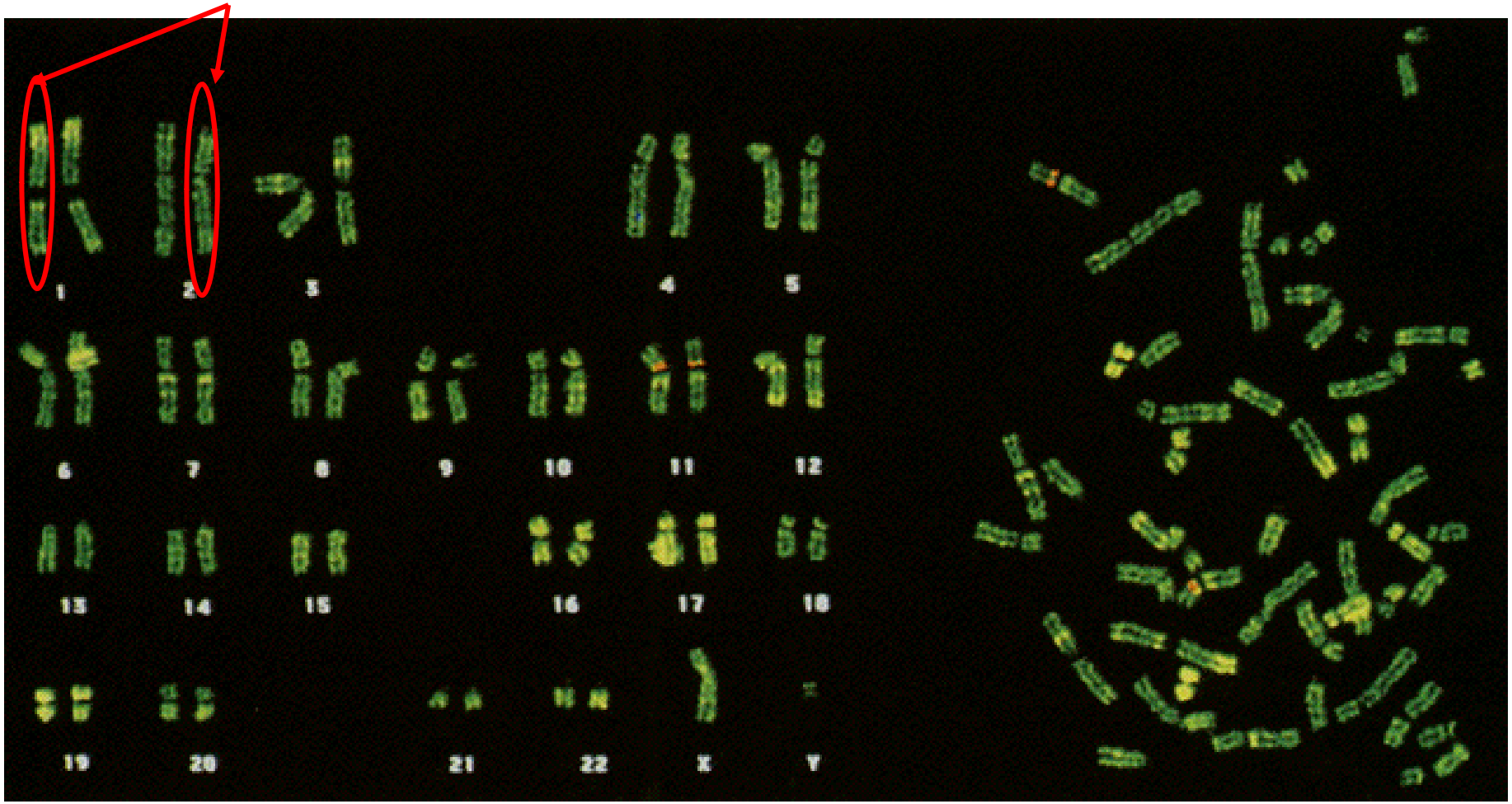
Homologous pair of chromosomes



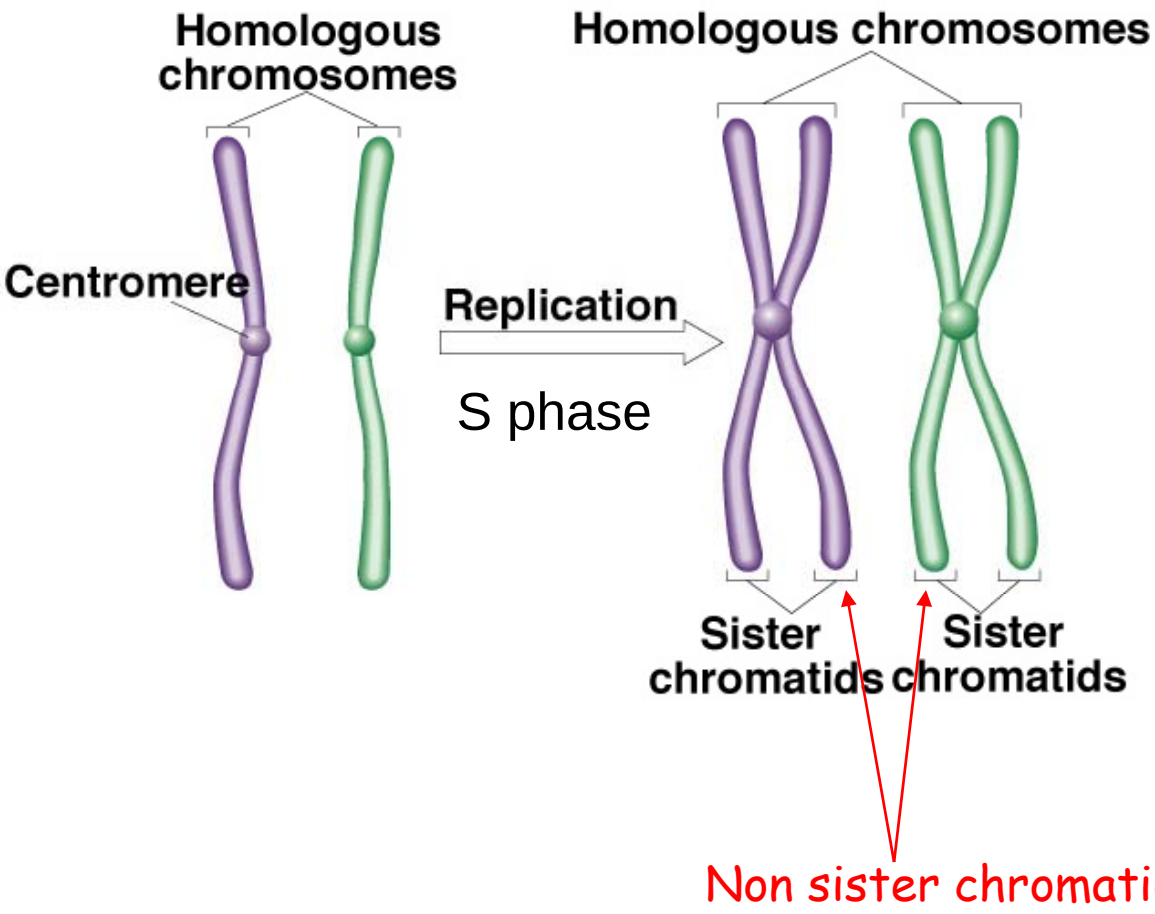
# humans are *diploid*

## two of each chromosome –*homologous pairs*

Non-homologous pair of chromosomes

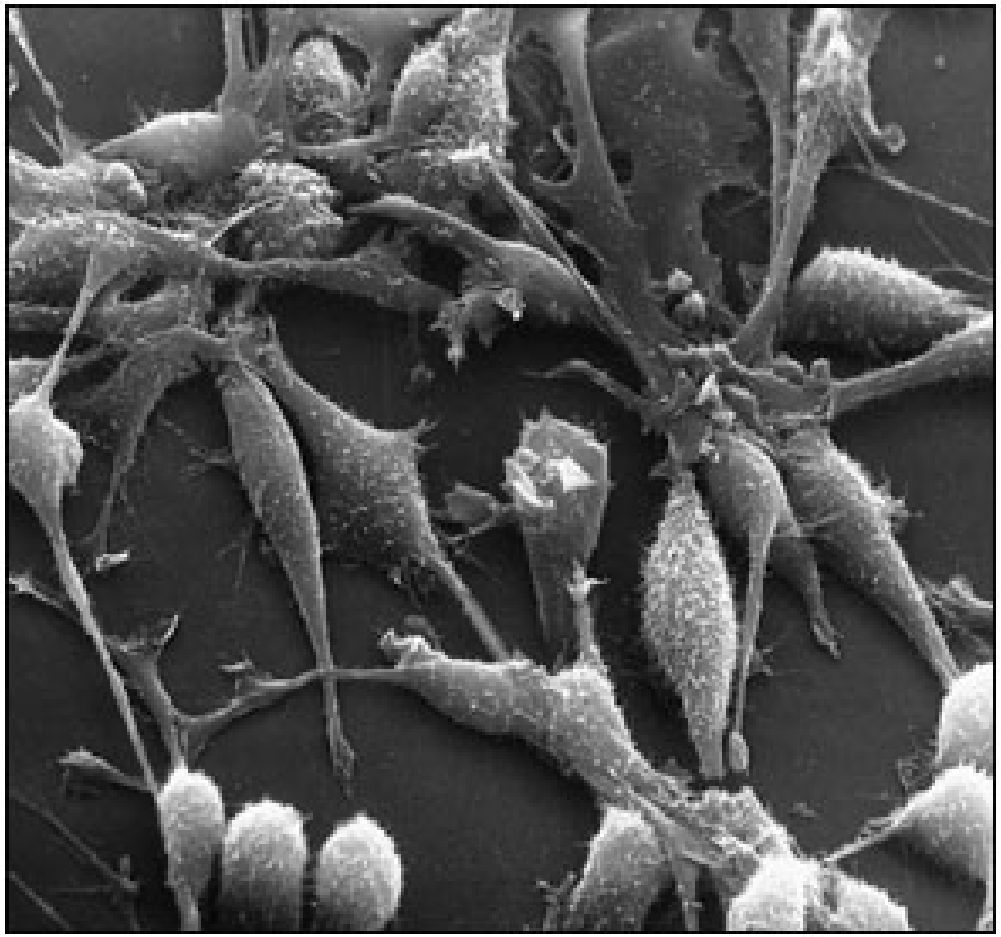


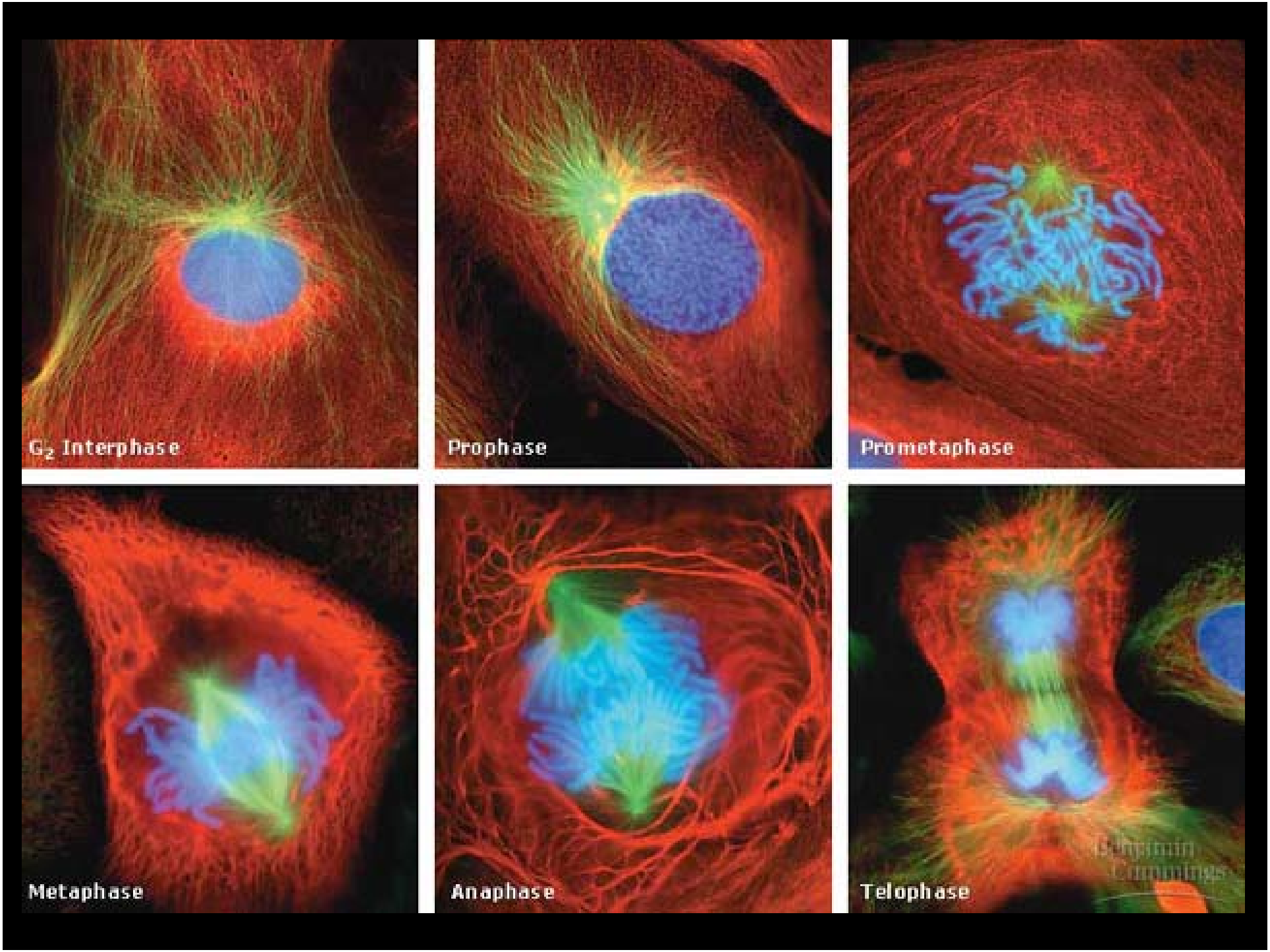
# Chromosomes and chromatids

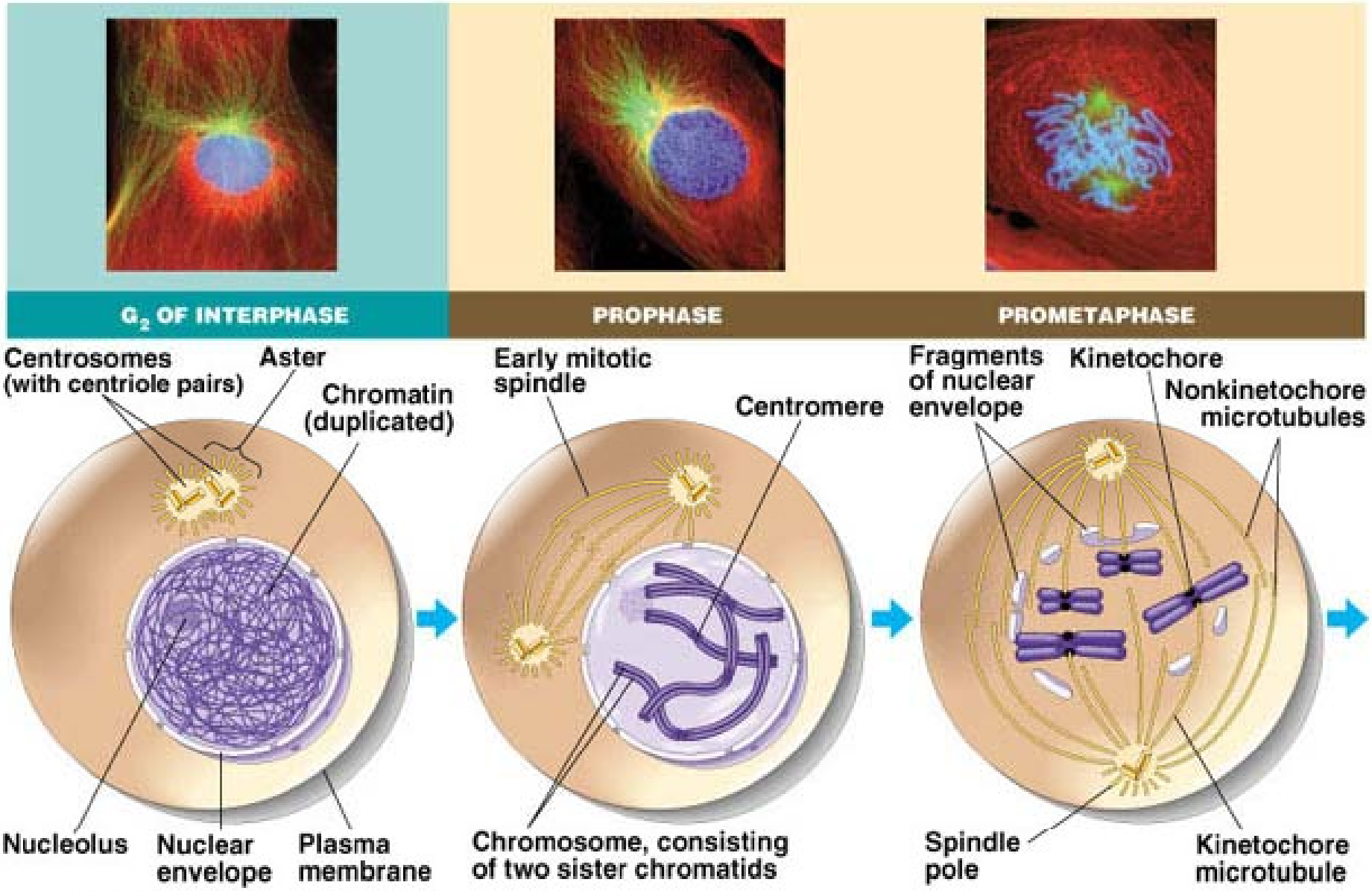



- After the chromosomes replicate during S phase, two sister chromatids are joined together by the centromere
- Sister chromatids are held together by ***cohesins*** until anaphase
- When sister chromatids separate during mitosis, they can still be called chromatids but more accurately they are daughter chromosomes

# Cell shape changes prior to mitosis

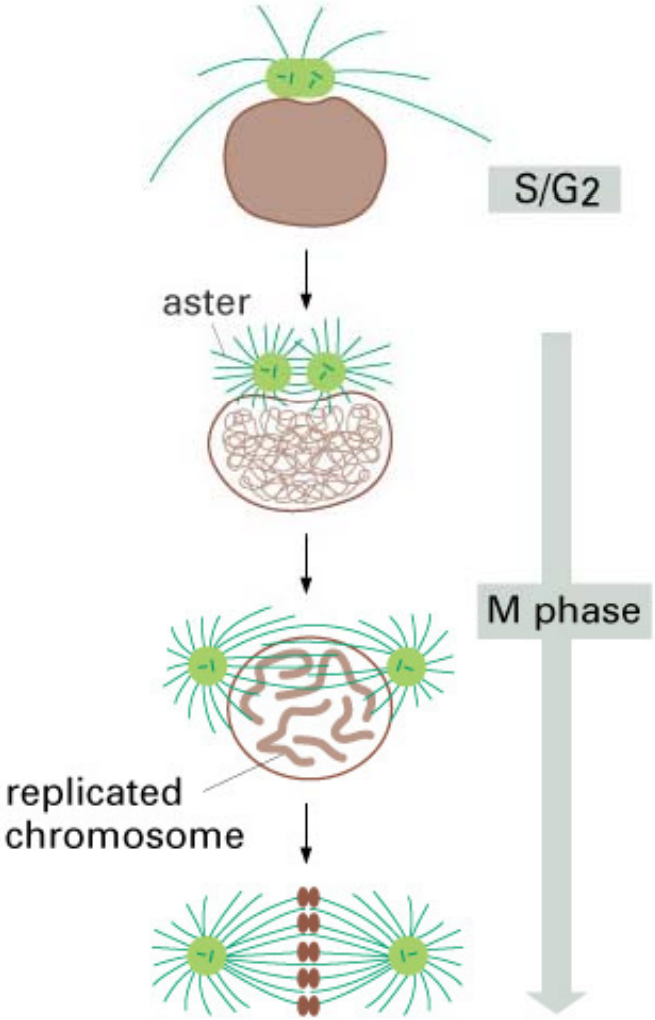



$G_2$ Interphase
Prophase
Prometaphase
Metaphase
Anaphase
Telophase

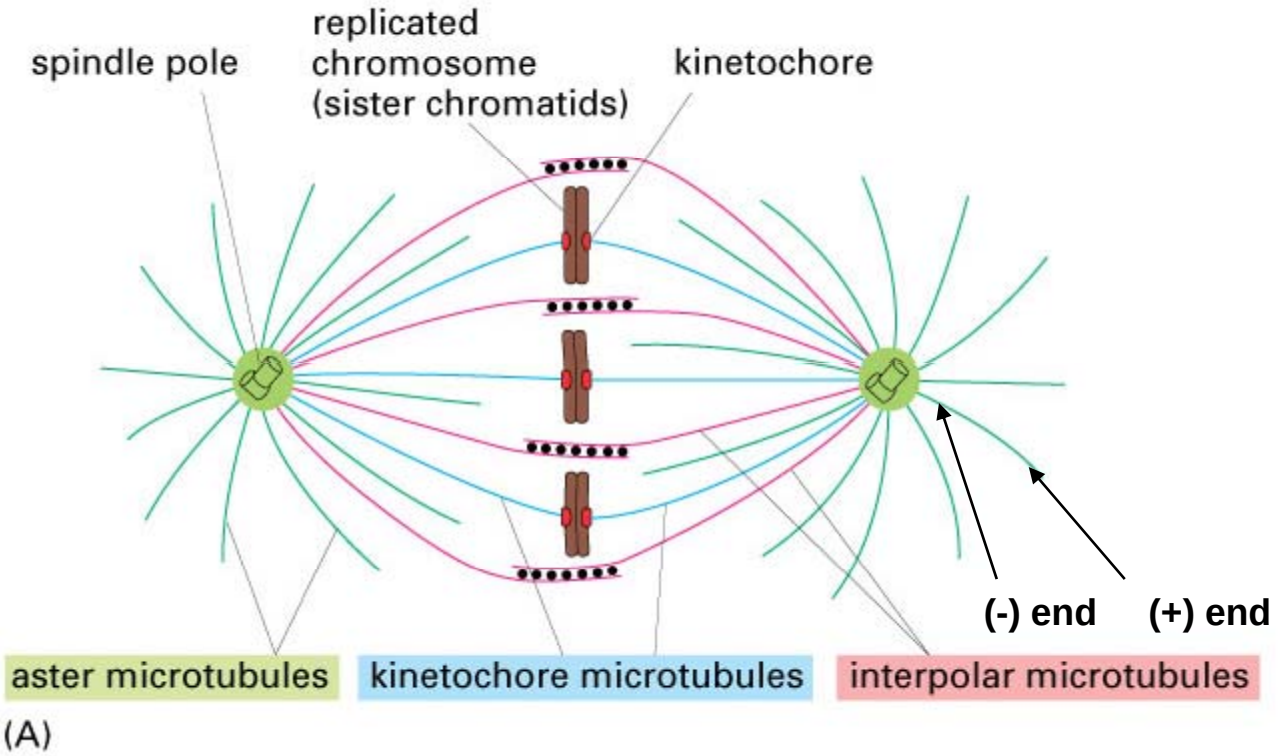



Copyright © Pearson Education, Inc., publishing as Benjamin Cummings.

**Before M phase begins, the DNA and centrosomes must both be replicated**



# Three classes of mitotic microtubules

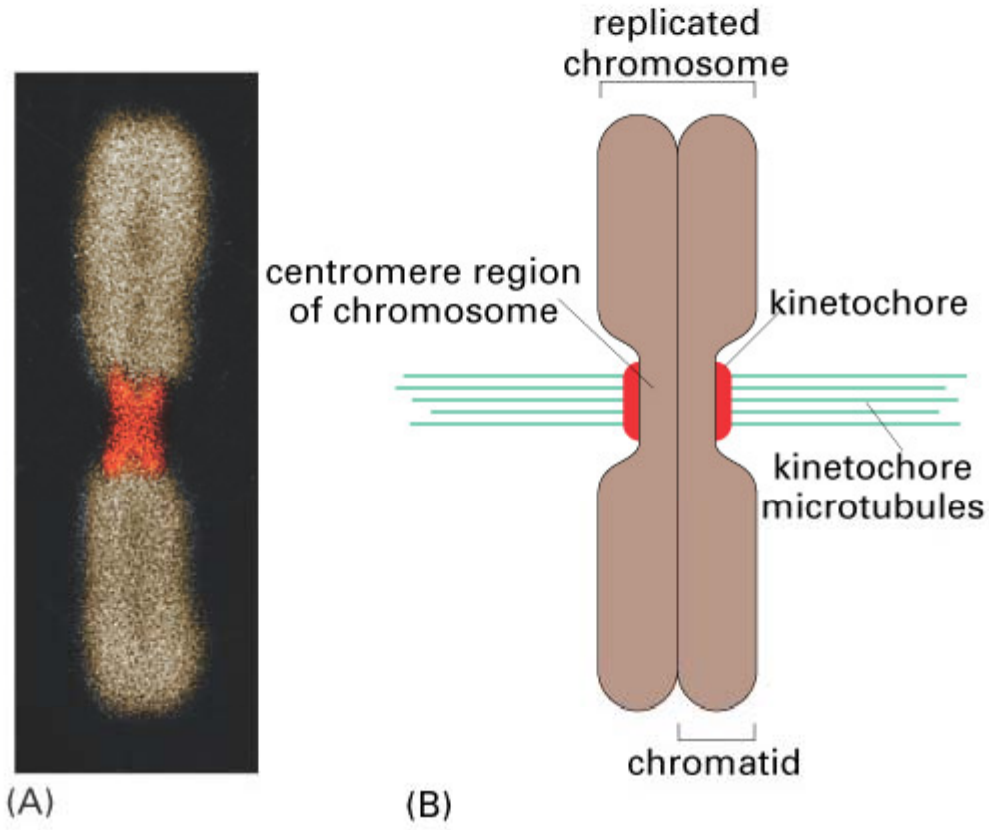



**20X increase in dynamic instability: change in ratio of stabilizing:destabilizing microtubule-associated proteins.**

1. **stablizing "microtubule-associated proteins" are phosphorylated**
2. **destabilizing "catastrophins" show up.**

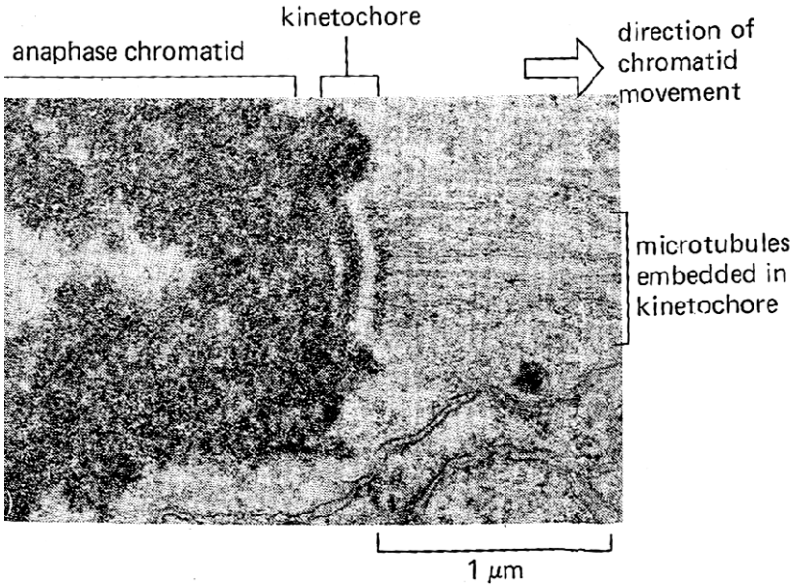
**Which Cdk triggers this change?**

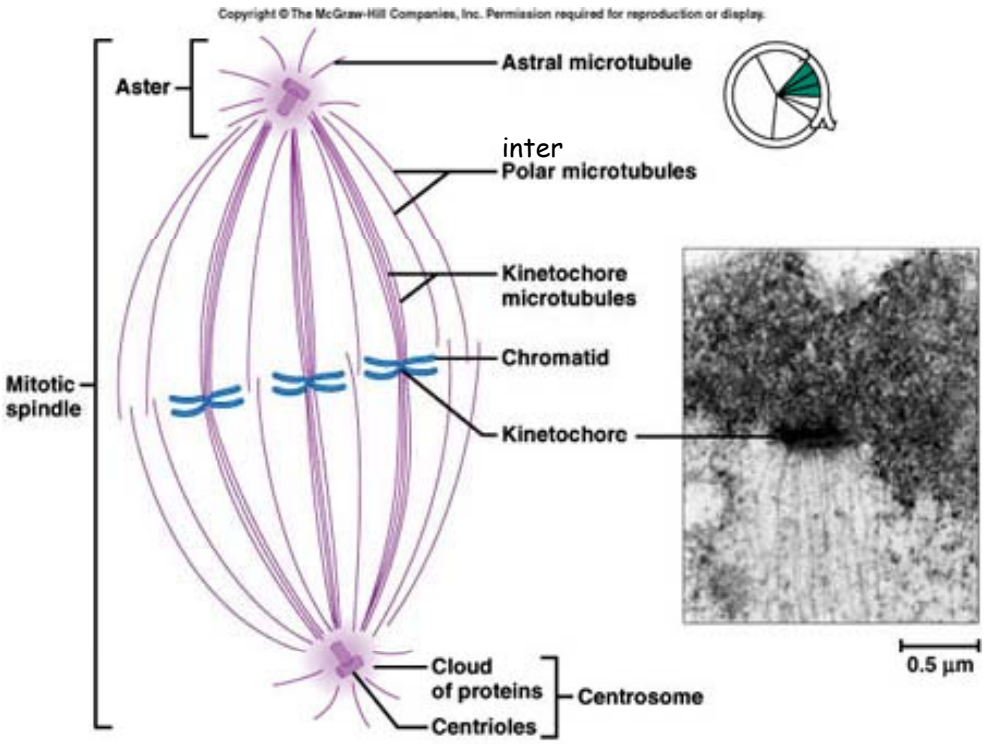
# The enigmatic kinetochore

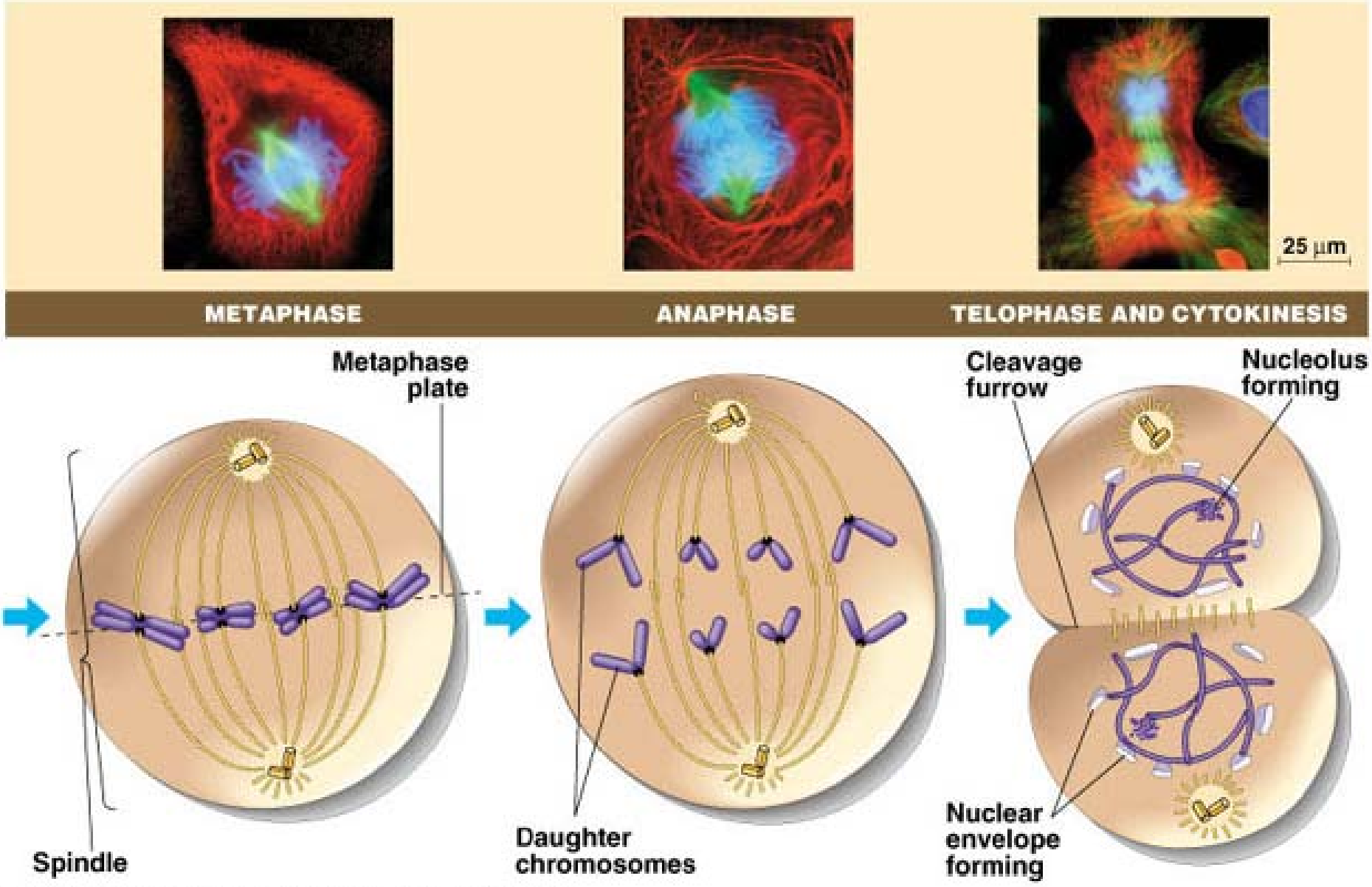



**Kinetochore = specialized proteins that bind centromere-specific DNA sequences**

# The enigmatic kinetochore



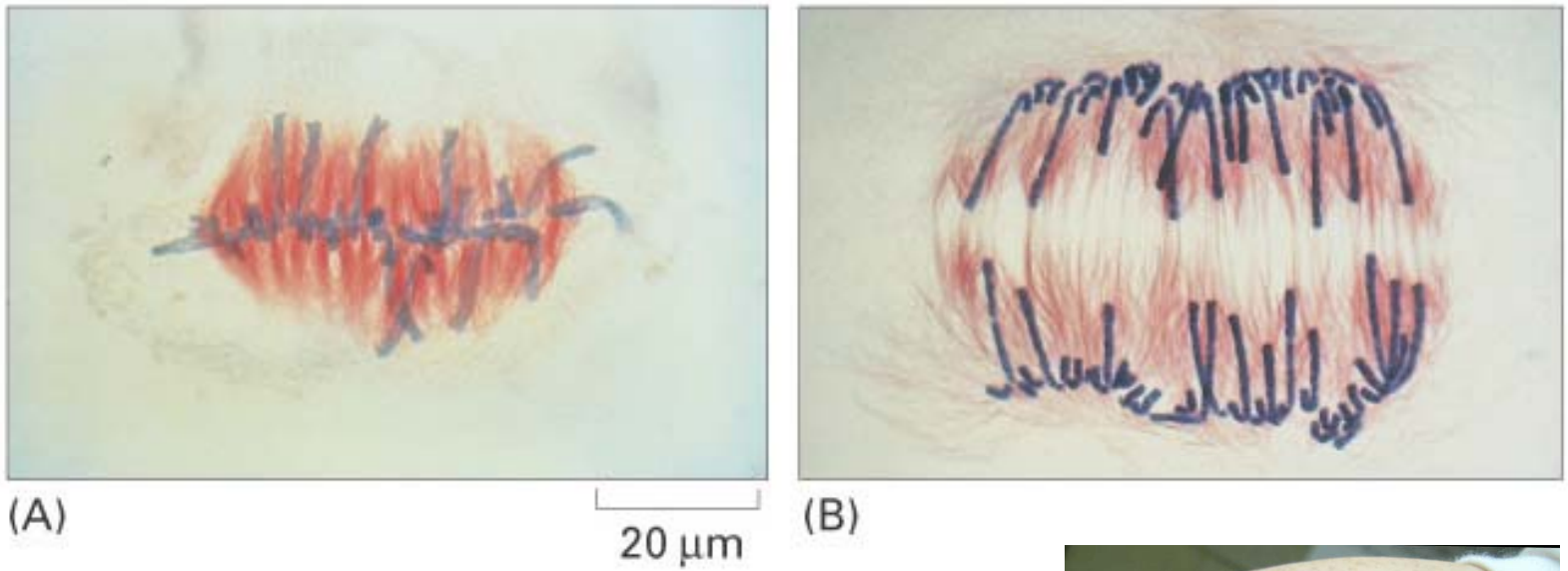


Copyright © Pearson Education, Inc., publishing as Benjamin Cummings.

Equal tension on each sister chromatid's kinetochore

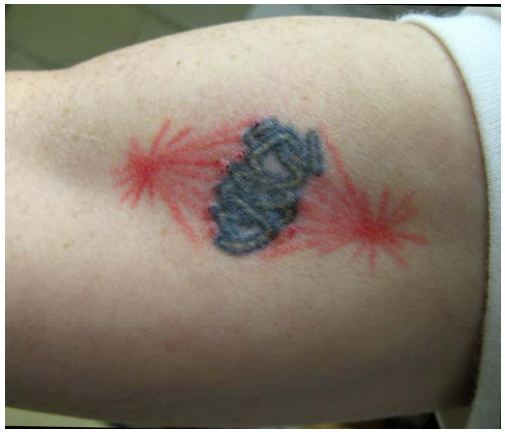
# Anaphase Promoting Complex (APC)

*Which Cdk triggers the APC?*

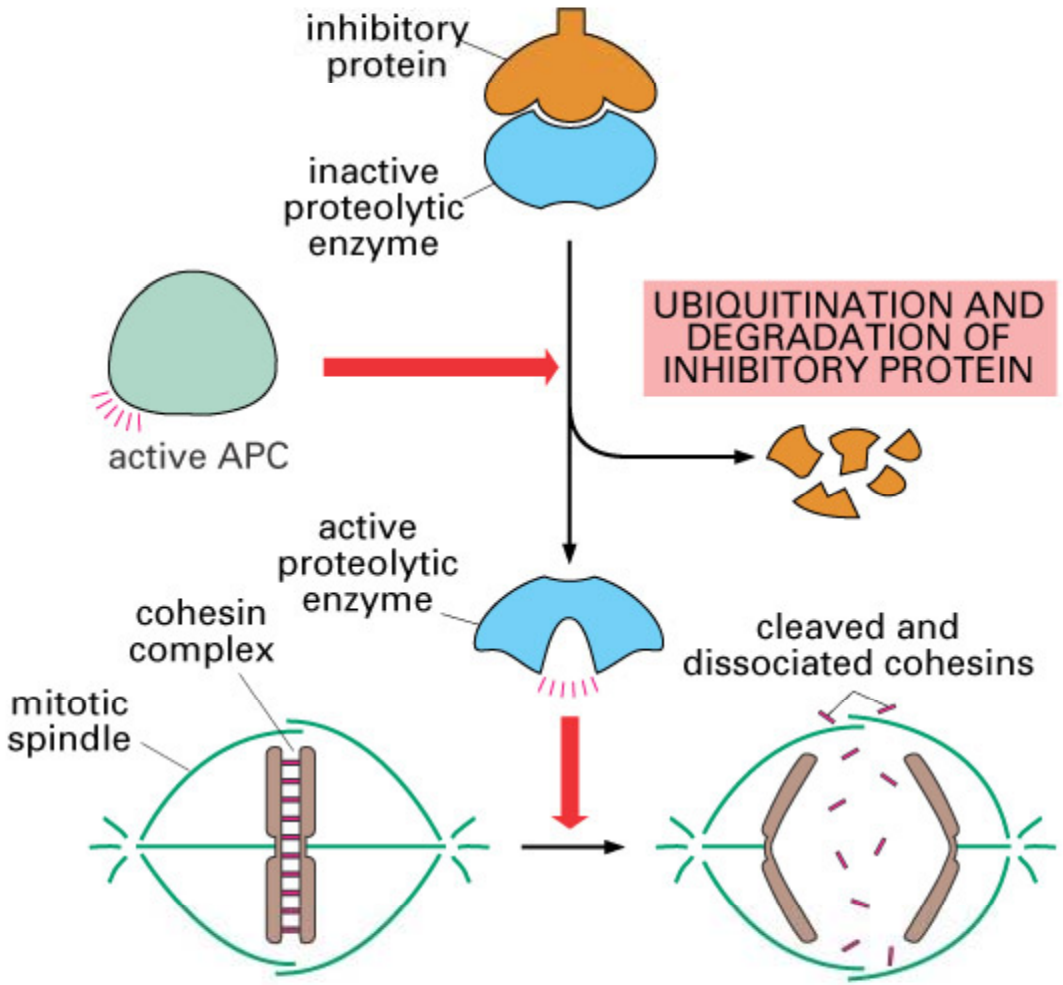



**Jobs of the APC**

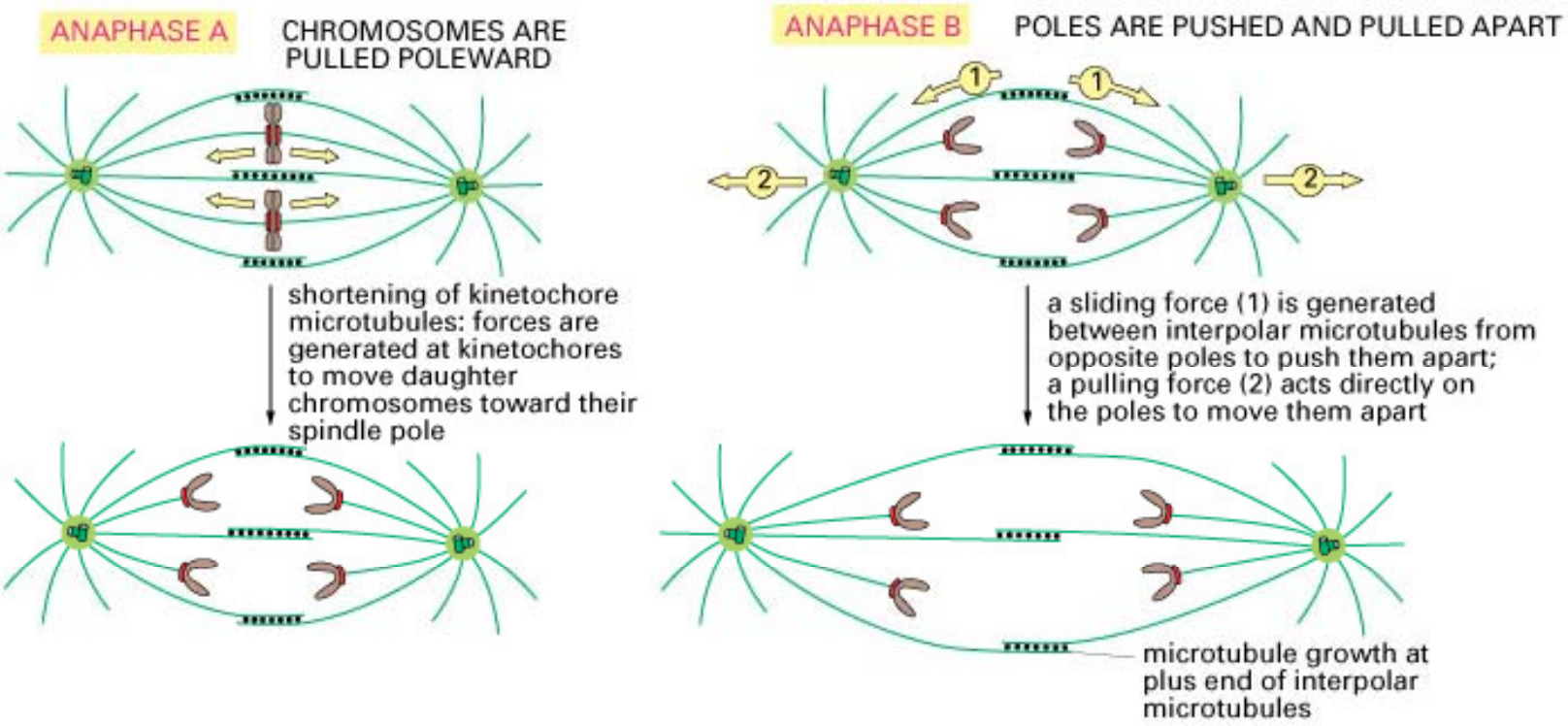
1. **Ubiquinates M-cyclin**
2. **Triggers proteolysis of cohesins that held sister chromatids together**



# Anaphase Promoting Complex (APC)




# What drives the movements at anaphase?




**Kinesin and dynein MT motors attached at:**

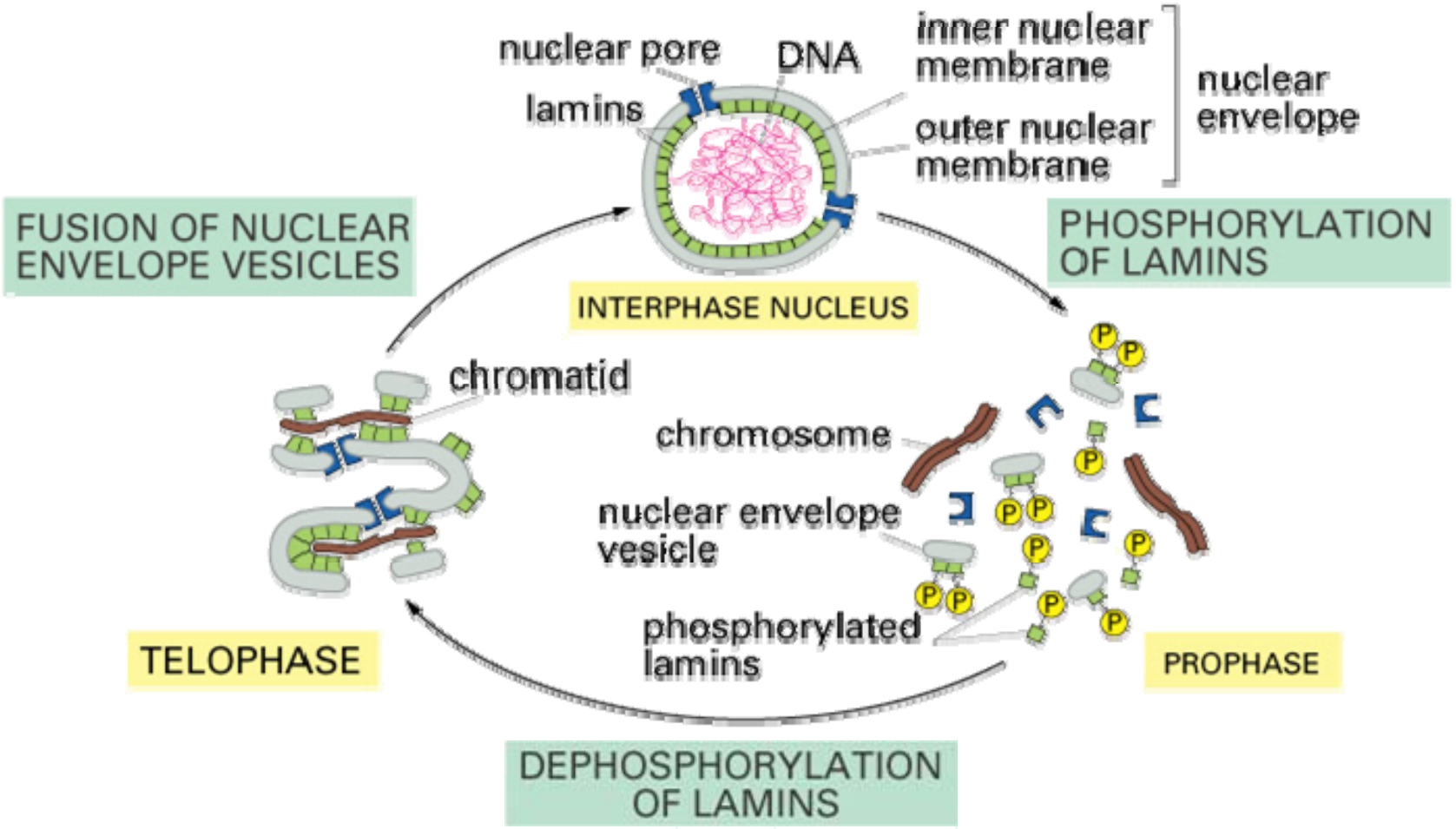
1. **interpolor microtubules (push apart poles)**
2. **kinetochore (releasing tension to permit destabilization and shortening)**
3. **cell cortex (beneath cell membrane), pulling centrosomes poleward**

# Telophase

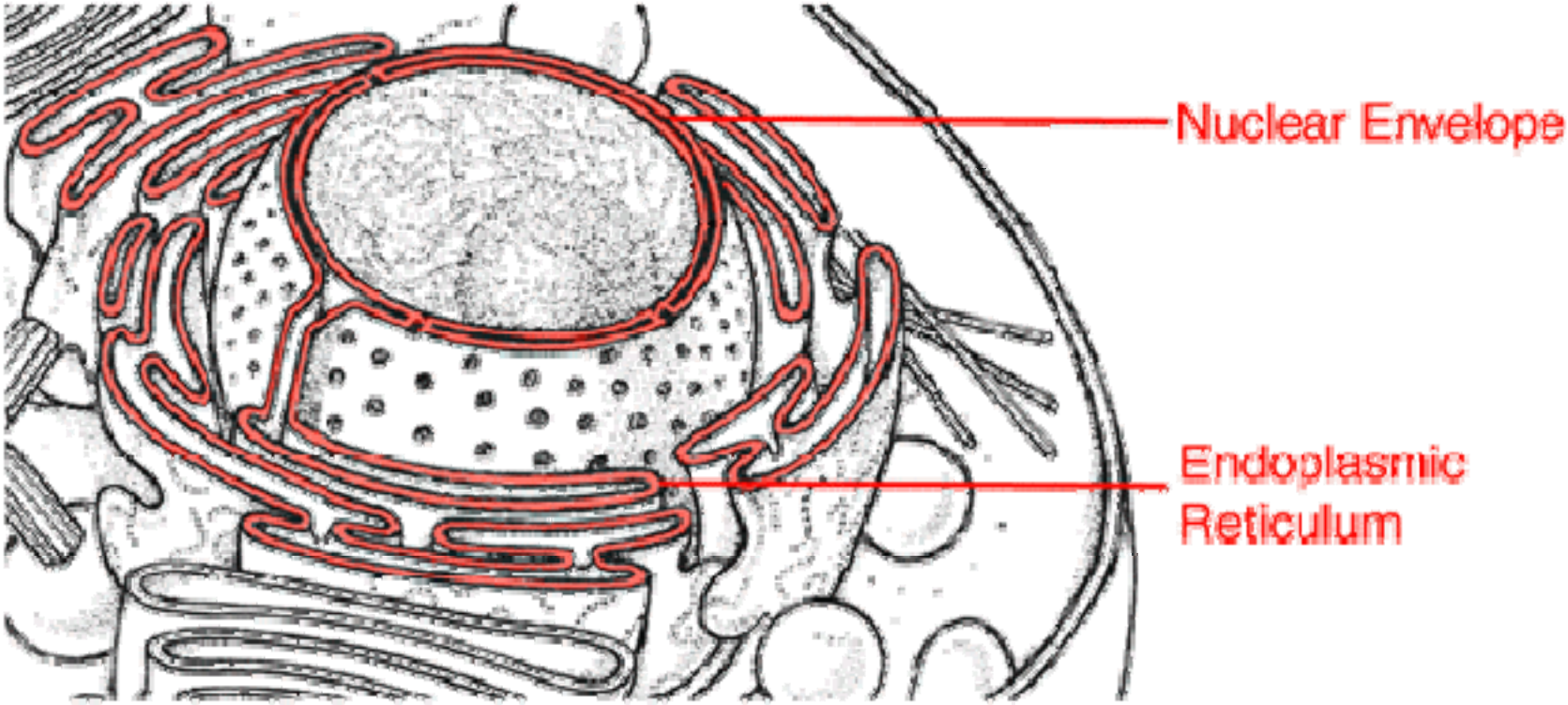
During telophase, the chromosomes become less condensed.

The nuclear envelopes and nucleoli re-form, producing two nuclei whose chromosomes are identical to each other and to those of the cell that began the cycle.

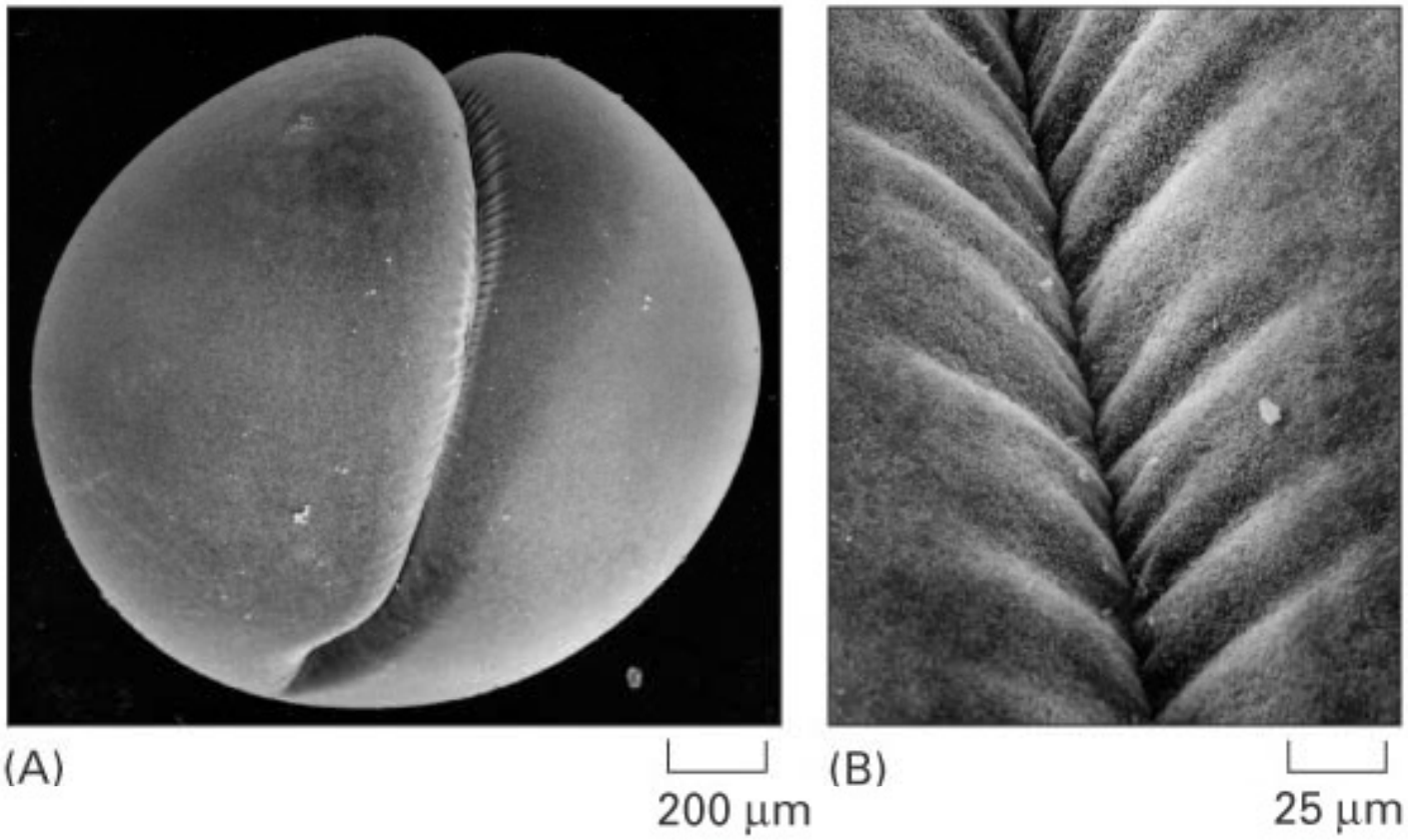
# Nuclear envelope reassembly



# Nuclear envelope reassembly



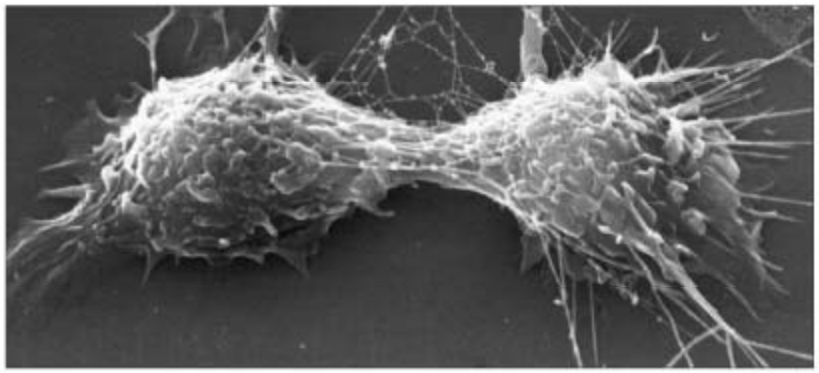
# Cytokinesis




**In animals, location of furrow is determined by aster MTs: midway between them, even if no mitotic spindle in between! Specifics still a mystery. In plants, midpoint marked by actin and MTs.**

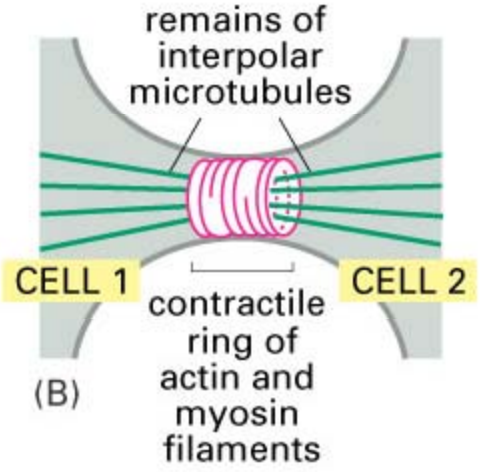
**p. 653: What happens if you move the mitotic spindle to one side with a fine glass needle?**

# Contractile ring

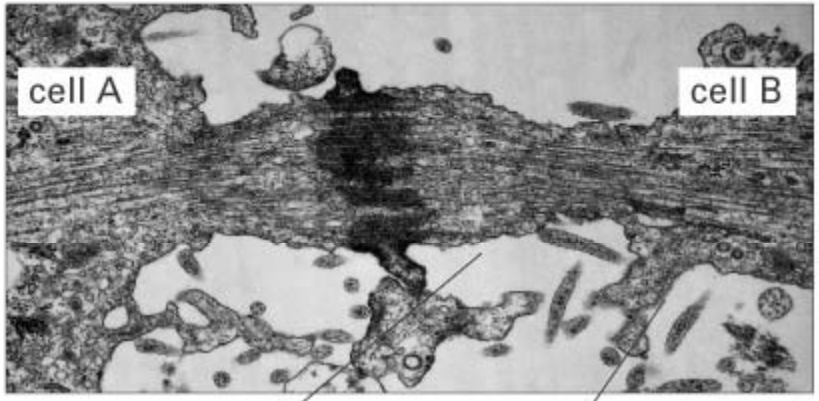


(A)

10 μm




(B)

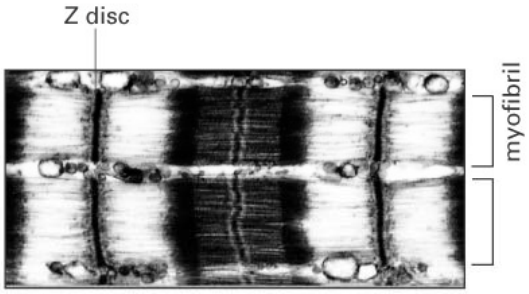



(C)

plasma membrane

1 μm

remaining interpolar microtubules




(A)

sarcomere ~2.2 μm

# Mitosis and Cell Division:

## Differences between plant and animal cells

1. **Centrosomes.**

   Plants, fungi, and diatoms have gamma tubulin clusters in the nuclear membrane rather than in exterior centrosomes. Plants also lack centrioles.

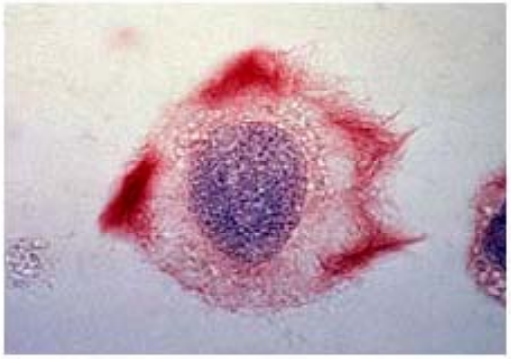
2. **Cell division.**

   Animal cell cytoplasm usually divides by plasma membrane furrowing caused by contraction of cytoplasmic actin filaments.

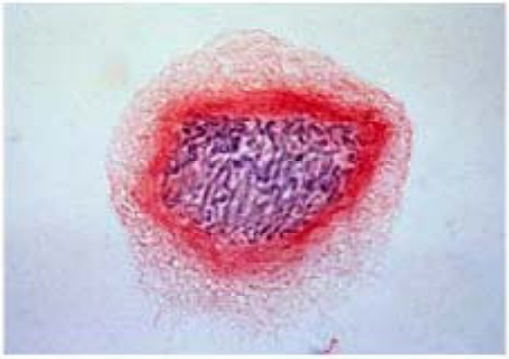
   In plant cells, cytokinesis is accomplished by vesicle fusion and the synthesis of new cell wall material by phragmoplast.

# Plant cell division

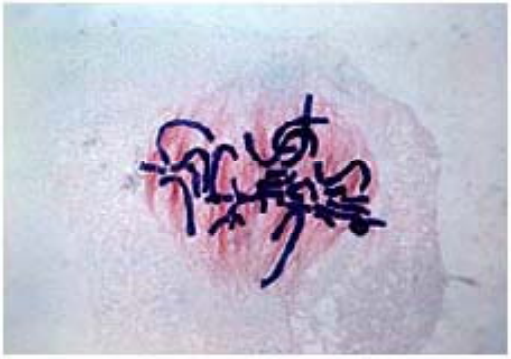
Mitosis in a plant cell (sans centrosome)



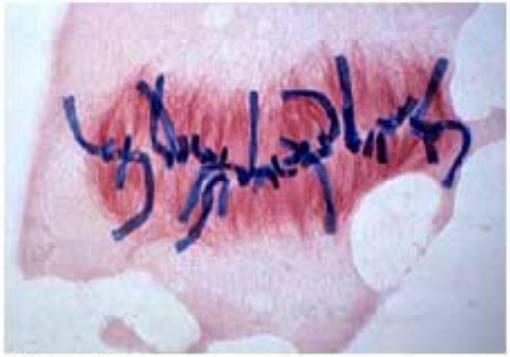
Interphase



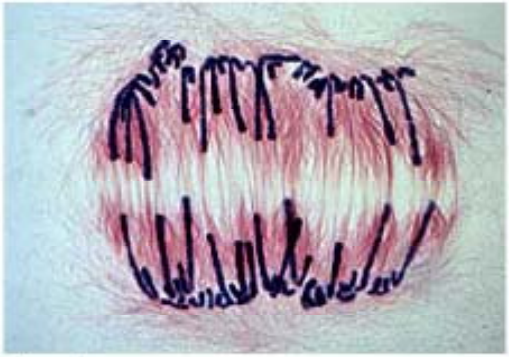
Prophase



Prometaphase



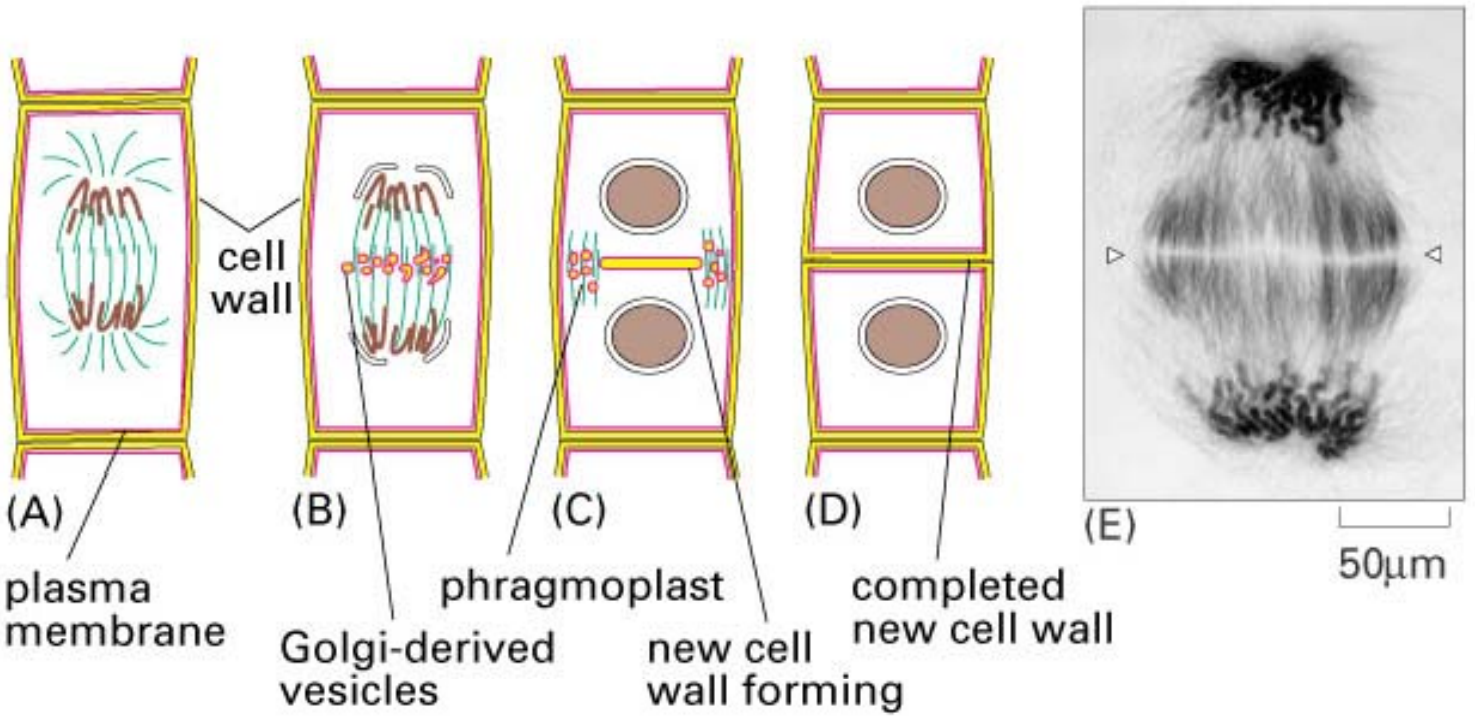
Metaphase



Anaphase

Telophase

# Plant cell division



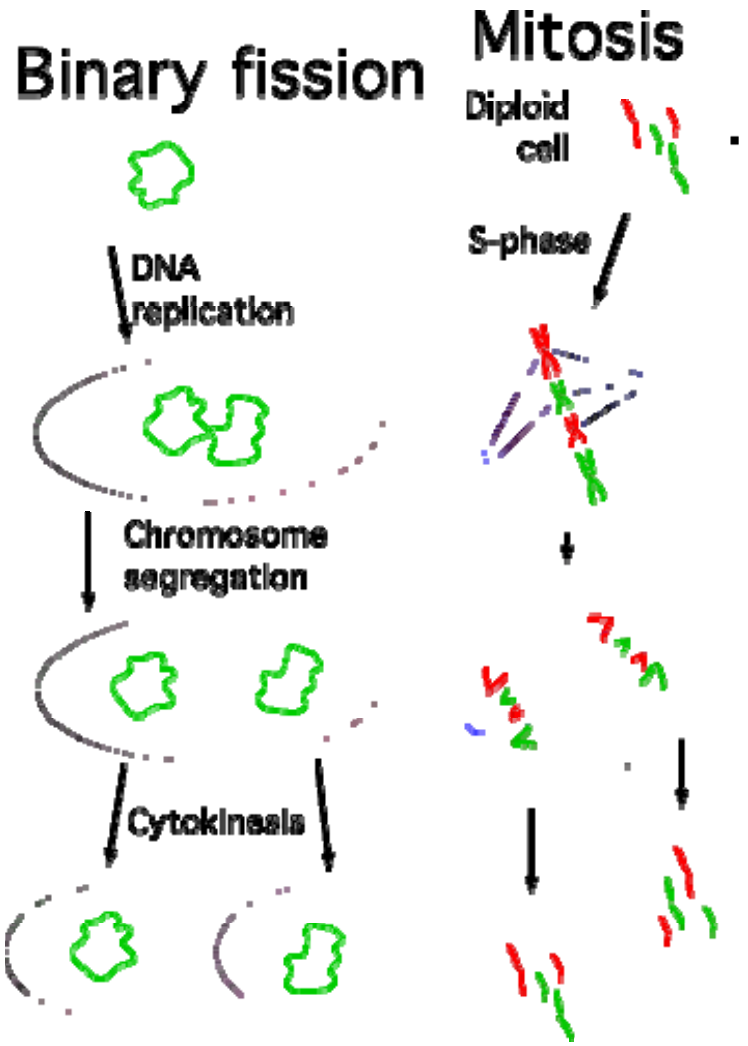

(Polysaccharides and glycoproteins)

# Cell cycle and mitosis in eukaryotes:

The cell cycle can repeat itself many times, forming a clone of genetically identical cells.

Asexual reproduction produces an organism genetically identical to the parent. Any genetic variety is the result of mutations.

# What about prokaryotes?



# Cell cycle controlled by nutrient availability: metabolism results in reproduction

- Microbial growth – an increase in a population of microbes rather than an increase in size
- On an agar plate, result of microbial growth is discrete colony – an aggregation of cells arising from single parent cell

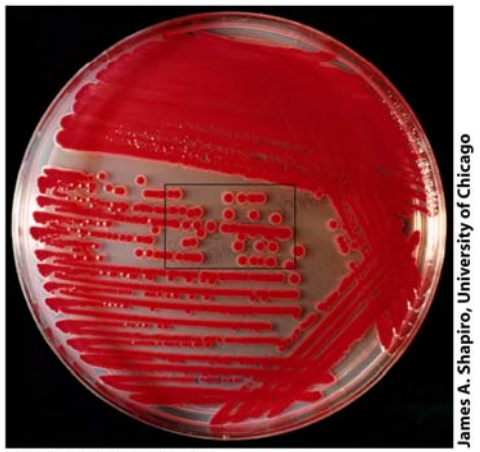



Figure 5-2a Brock Biology of Microorganisms 11/e
© 2006 Pearson Prentice Hall, Inc.

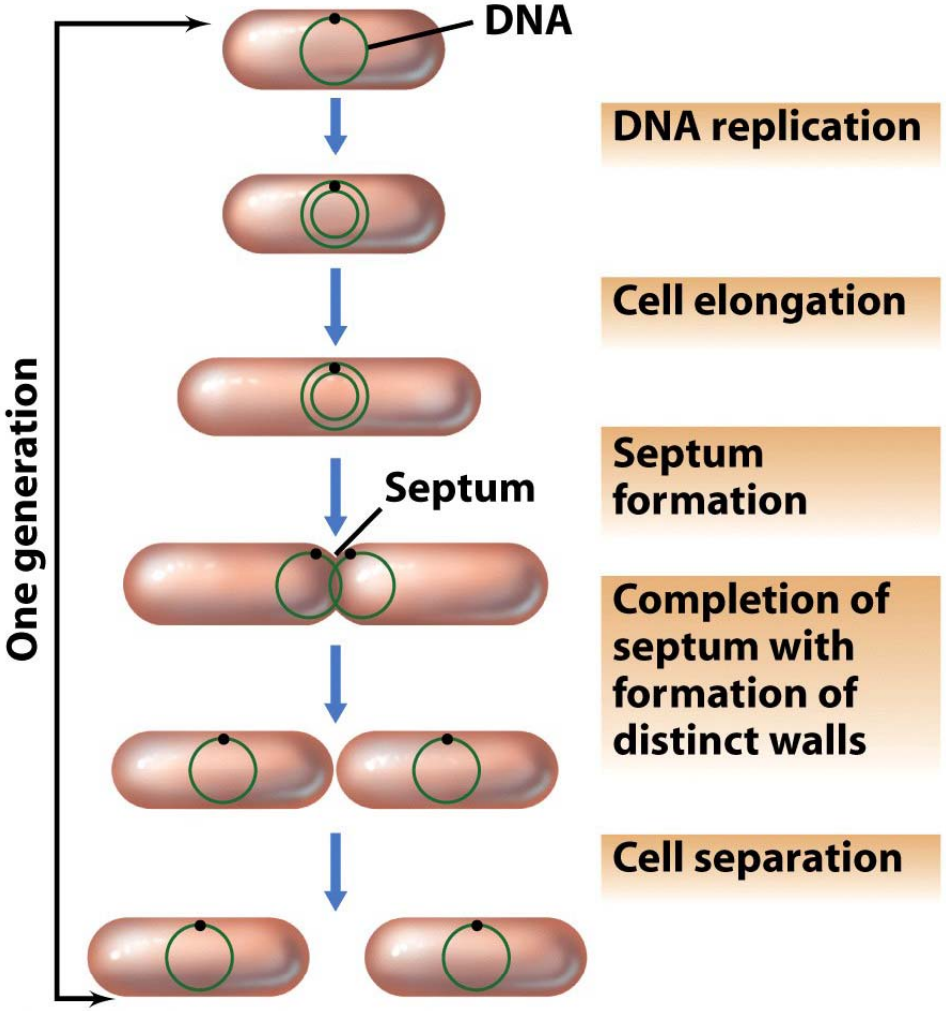
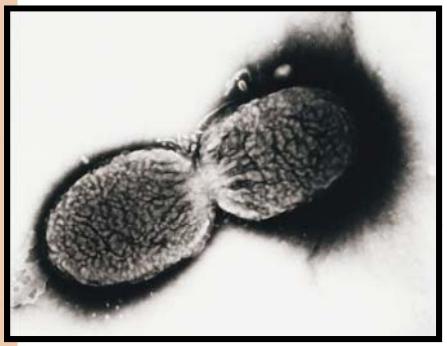



Figure 6-1 Brock Biology of Microorganisms 11/e
© 2006 Pearson Prentice Hall, Inc.

# A little food for thought about food limitation…

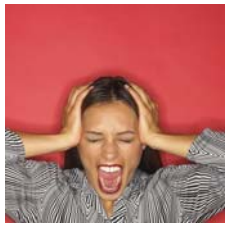
| Time (h) | Total number of cells | Time (h) | Total number of cells |
|---|---|---|---|
| 0 | 1 | 4 | 256 ($2^8$) |
| 0.5 | 2 | 4.5 | 512 ($2^9$) |
| 1 | 4 | 5 | 1,024 ($2^{10}$) |
| 1.5 | 8 | 5.5 | 2,048 ($2^{11}$) |
| 2 | 16 | 6 | 4,096 ($2^{12}$) |
| 2.5 | 32 | . | . |
| 3 | 64 | . | . |
| 3.5 | 128 | 10 | 1,048,576 ($2^{19}$) |



-***E. coli*** **cells, weighing 1 pg ($10^{-12}$) gram, have a 20 minute doubling time.**

**-If grown in unlimited nutrients for 48 hours, $10^{14}$g = $10^7$ tons of *E. coli*!**

**-Yikes!**



**Thank goodness nutrients in a single test tube (and your gut) are limited…!!**

# Taxol (Paclitaxel)

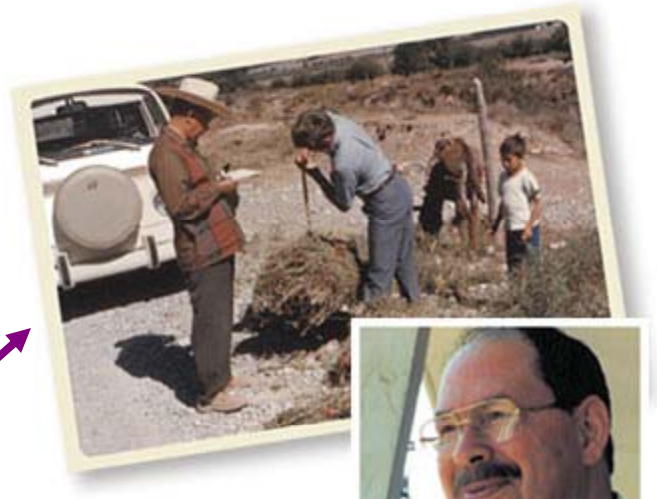
Drug discovery is exciting, but it's a long, slow process!  The taxol story started nearly 50 years ago…

1958:  As part of a natural products screening program, the National Cancer Institute (NCI) commissioned the US Department of Agriculture (USDA) to collect samples of over 30,000 plants to test for anticancer properties.

1962:  As part of this screen done in the Pacific Northwest, a team of college **students working for the USDA for the summer** collected 15 lbs of Pacific yew tree sample – needles, bark, twigs – from a site in the **Washington Cascades**.

Early 1963:  Monroe Wall (medicinal chemist) discovers that the samples are active!

-chemically fractionate *Taxus* extract
-bioassay:treat cancer cell lines (tissue culture)



**Arthur Barclay, USDA botanist in 1962 and today**

# Promotion of microtubule assembly *in vitro* by taxol

*Nature* Vol. 277 22 February 1979

PETER B. SCHIFF
JANE FANT
SUSAN B. HORWITZ

**Tubulin = building block of microbubules**

**Add taxol.**

**Wait 30 minutes…**

**Microtubule formation!**

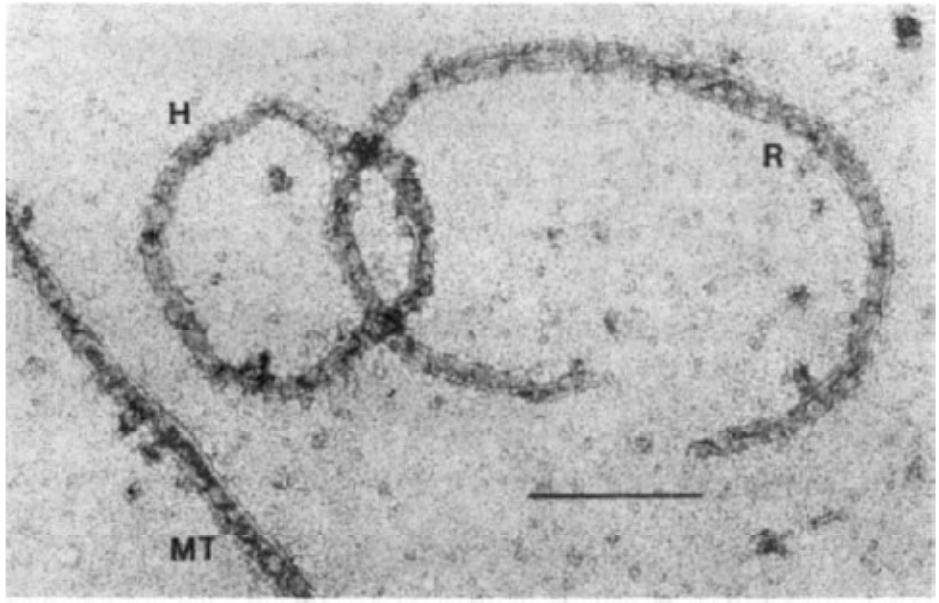
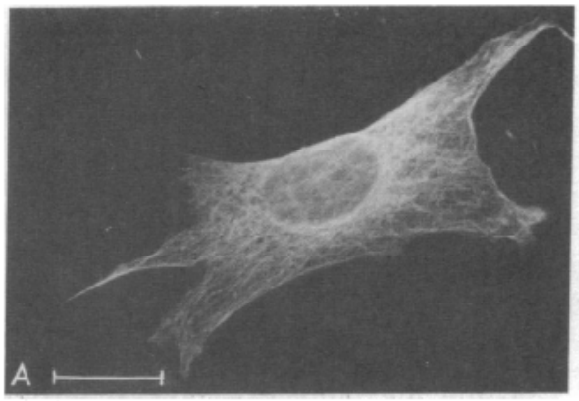
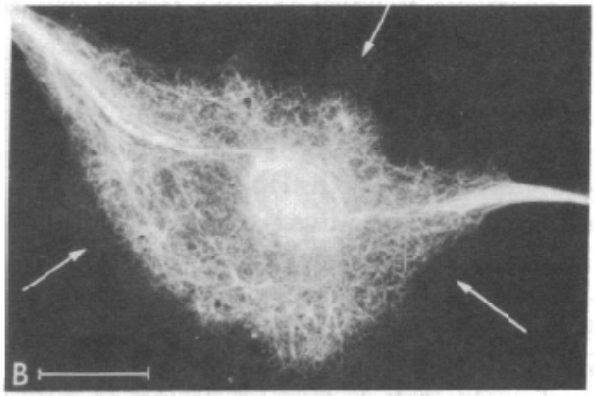



**Fig. 5** Electronmicrograph of the structures assembled in the presence of 0.1 mg ml$^{-1}$ tubulin and 5 μM taxol at 30 min in standard conditions. Structures were negatively stained with uranyl acetate[21]. MT, Microtubule; H, hoop; R, ribbon. Scale bar, 0.2 μm.

# Taxol treatment:
# cells are “so stuffed with microtubules they can’t survive”

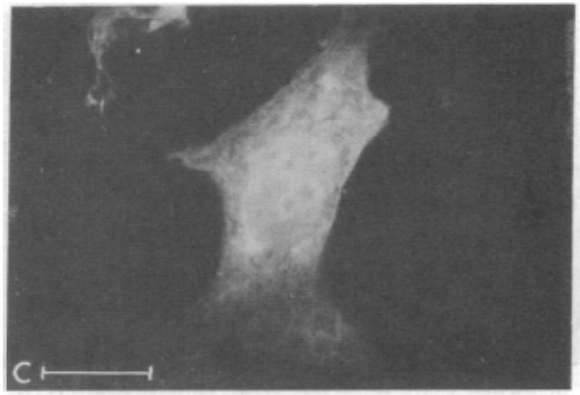
No taxol

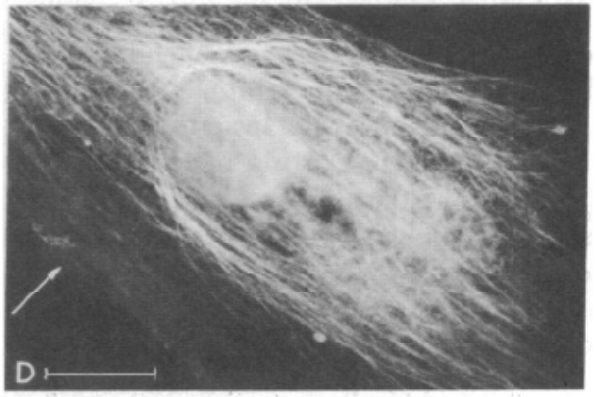



+ taxol

No taxol




+ taxol

FIG. 3. Indirect immunofluorescence of BALB/c fibroblast cells, using antibodies against tubulin. (*A*) Control cell, (*B*) cell exposed to 10 $\mu$M taxol for 22 hr, (*C*) control cell kept at 4°C for 16 hr, (*D*) cell incubated with 10 $\mu$M taxol for 22 hr, then shifted to 4°C for 16 hr. Scale bars: 20 $\mu$m. Arrows indicate the edge of the plasma membrane in the plane of focus.

**To conduct clinical trials, large quantities of taxol were required. This presented a problem:**

**Pacific yew was an environmentally protected species because it was part of the habitat for the endangered spotted owl**

**In the 1980's, cutting down Pacific yew trees was the only way to obtain taxol.**

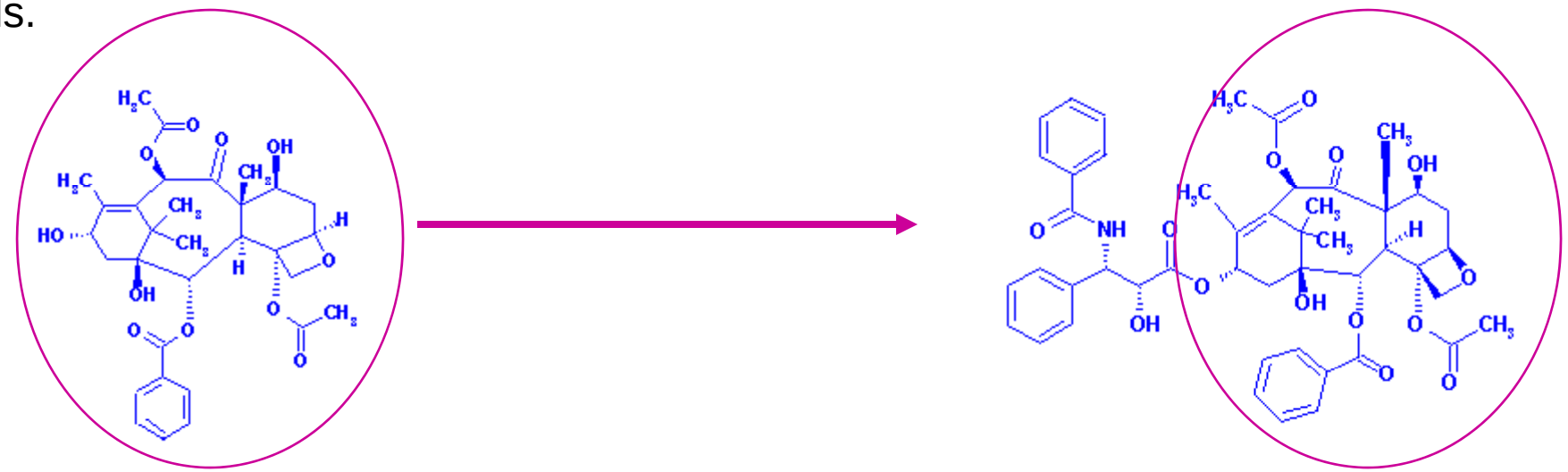
**Worse, Pacific yew is one of the slowest growing trees in the world. And there's not much taxol in the tissues (remember – 15 lbs turned out to be too small a sample to do much work with!)**

**A 40-foot Pacific yew tree, which may have taken 200 years to reach that height, yields only a half gram of taxol, and it would take three such trees to provide enough taxol to treat just one patient.**

# Taxol (Paclitaxel)

A taxol precursor, 10-deacetylbaccatin, was discovered in needles of the related ornamental shrub *Taxus baccata*. You probably have seen this shrub growing around buildings. It's not protected and does not provide habitat for spotted owls.

Semisynthesis of taxol from 10-deacetylbaccatin was very laborious, but the source was renewable, and sufficient quantities were obtained to carry out clinical trials.



Baccatin = "taxane ring"

taxol

# Clinical Trials

1989: Clinical studies demonstrated promising activity of taxol against advanced ovarian cancer.

1992: US Food and Drug Administration (FDA) approved taxol for treatment of ovarian cancer.

1994: Taxol was approved for treatment of breast cancer.


Market names: Taxol, paclitaxel