# Cell Biology
# Lecture 10

Cell Signaling - principles

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22.11.2023

Alberts • Johnson • Lewis • Morgan • Raff • Roberts • Walter

# *Molecular Biology of the Cell*

**Sixth Edition**

## Chapter 17
## The Cell Cycle Regulation

Pages: 963-974, 1010-1015

# Course overview – Tentative schedule

| Date | Lecture | | Chapters & Topics | Assignments |
|---|---|---|---|---|
| 25.10. | 1 | Part 1 | *Course overview,*<br>DNA, Chromosomes, Genome, Ch. 4 | |
| 27.10. | 2 -G | | Replication, Repair, Recombination, Ch. 5 | |
| 1.11. | 3 | | From DNA to protein, Ch. 6 | |
| 3.11. | 4 | | Control of gene expression, Ch. 7 | |
| 8.11. | 5 | Part 2 | Membrane structures, Ch. 10<br>Membrane transport, Ch. 11 | **Assignment I (Essay) Draft I** (8.11.) |
| 10.11. | 6 -G | | Intracellular compartments and protein sorting, Ch. 12 | |
| 15.11. | 7 | | Intracellular compartments and protein sorting, Ch. 12<br>**Susanna Mäkinen, Solar Foods** | **Assignment II – Draft I (15.11.)** |
| 17.11. | 8 | | Membrane Traffic, Ch. 13<br>**iGEM team 2023** | *+iGEM intro* |
| 22.11. | 9 | Part 3 | Cell signalling, Ch. 15 | **Assignment II – Peer review** (22.11.) |
| 24.11. | 10 -G | | Cell signalling, Ch. 15 | **Assignment I (Essay) Draft II** (24.11.) |
| 29.11. | 11 | | Cell cycle, Ch. 17<br>**Jere Weltner, Folkhälsan** | |
| 1.12. | 12 | | Apoptosis, Ch. 18 + About exam | **Assignment II – final version** (1.12.) |
| 7.12. | EXAM | | December 7th | |
| 8.12. | Final version essay | | December 8th | **Assignment I (Essay) Final version** (8.12.)<br>Aim at finishing before exam date. Use last days for polishing. |

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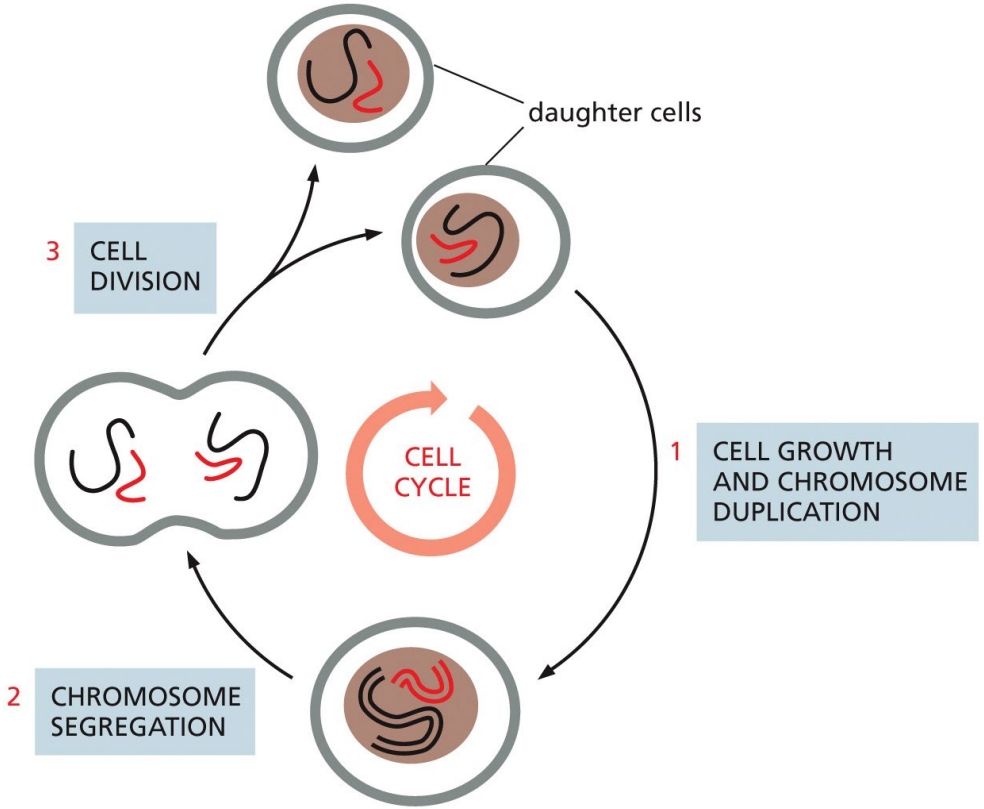
- OVERVIEW OF THE CELL CYCLE
- THE CELL-CYCLE CONTROL SYSTEM
- CONTROL OF CELL DIVISION AND CELL GROWTH



# FOUR (*FIVE*) PHASES OF THE CELL CYCLE

G1: PRIMARY GROWTH PHASE

S: DNA REPLICATION

G2: SECONDARY GROWTH PHASE

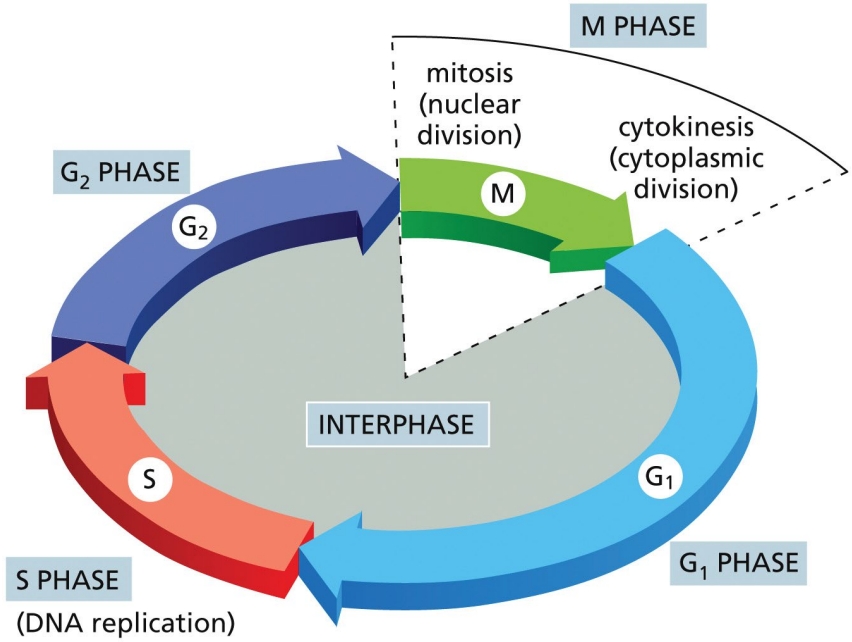
- collectively these 3 stages are called interphase

M: CELL DIVISION

- Mitosis
- Cytokinesis

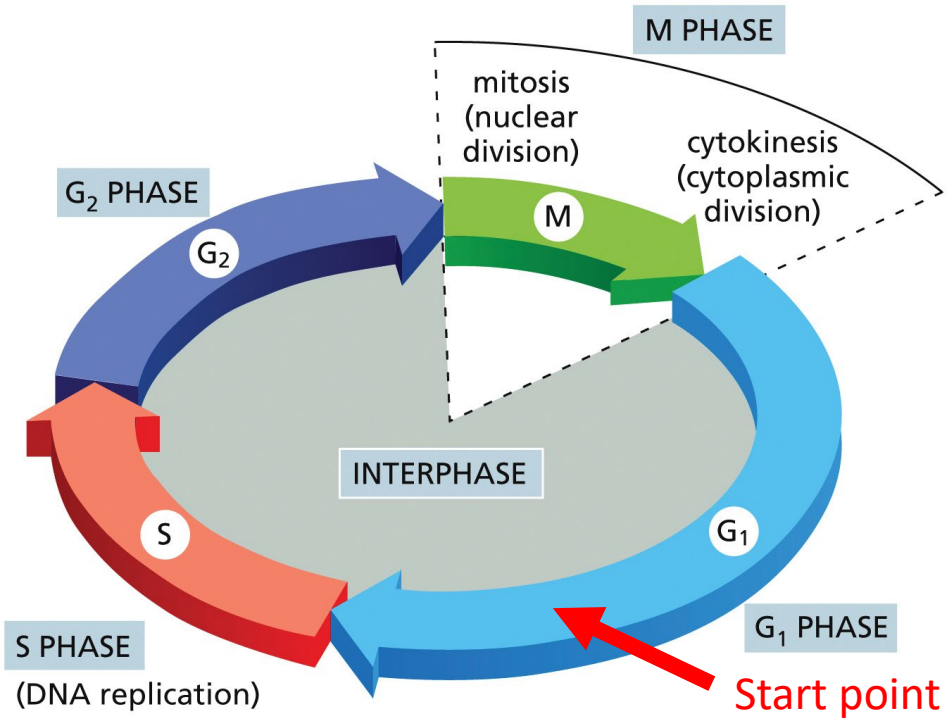
**$G_0$ PHASE ($5^{TH}$ PHASE)**

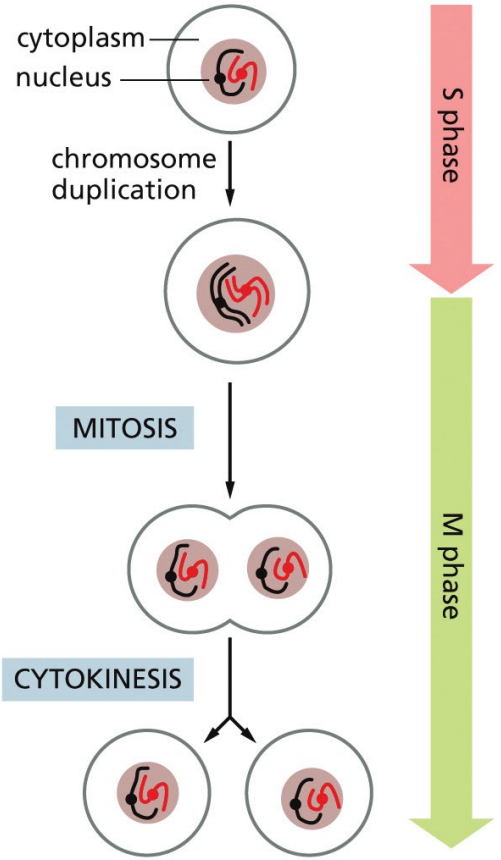
- Resting cells
- Terminally differentiated cells



# OVERVIEW OF THE CELL CYCLE

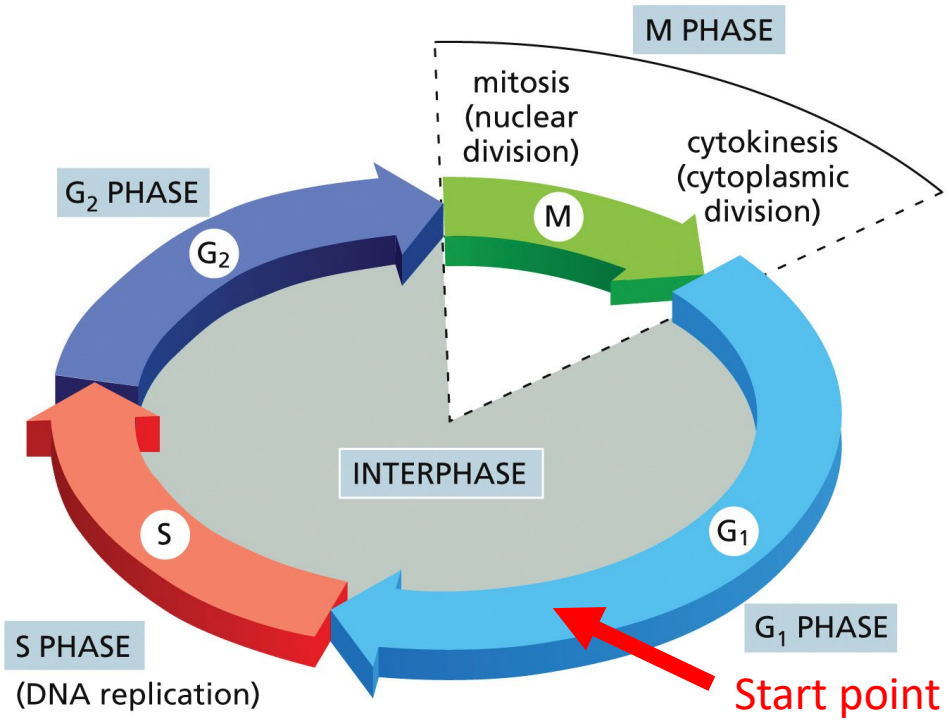
- The eukaryotic cell cycle usually consists of four phases
- G1 and G2 phases are “gap” phases
- G1+S+G2 = interphase

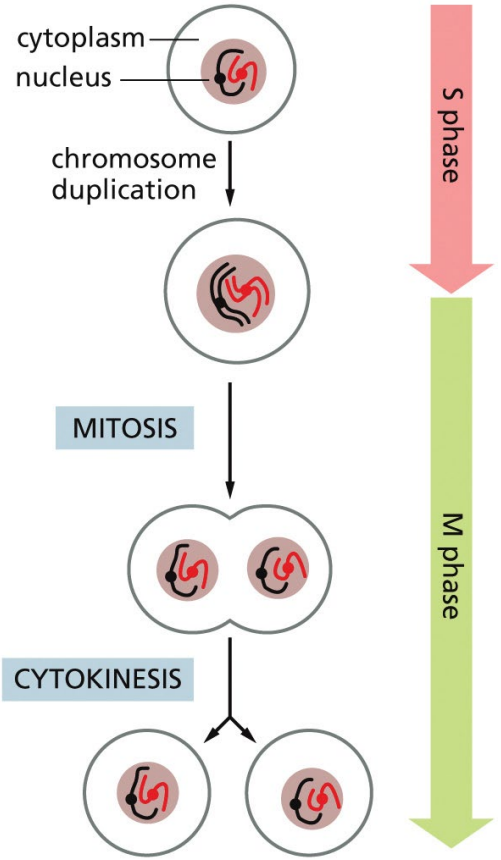


# OVERVIEW OF THE CELL CYCLE

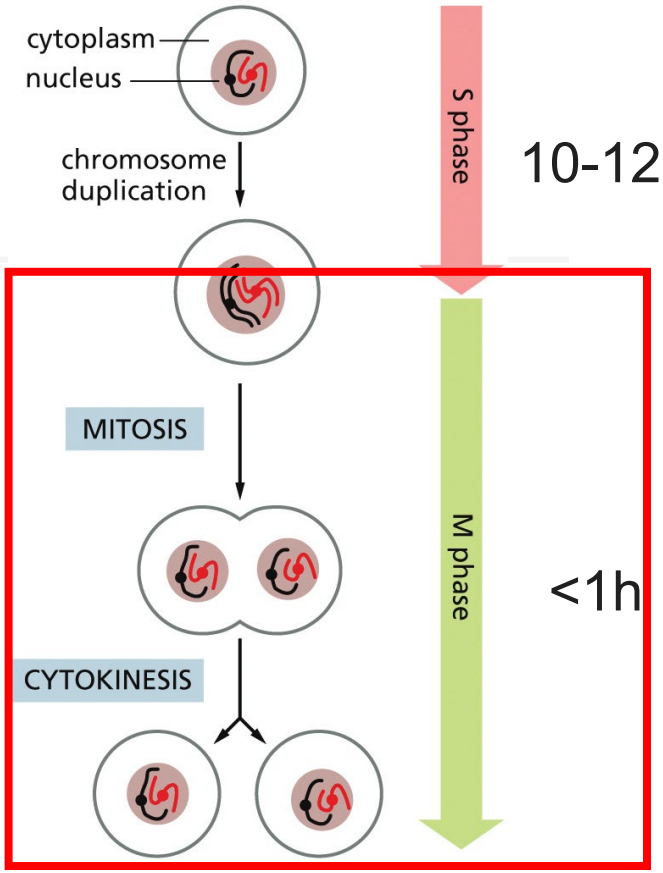
- **G0 = resting phase**
  - E.g. bone and nerve cells permanently in G0
  - Can also be temporary, e.g. in liver or lymphocytes

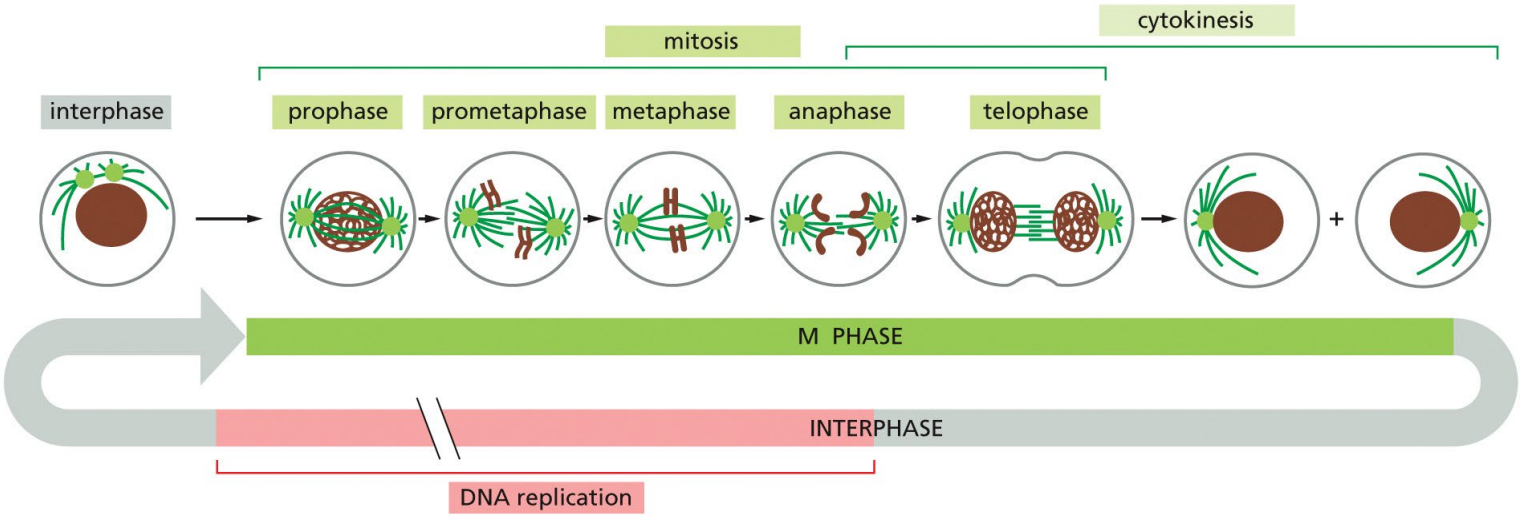


# OVERVIEW OF THE CELL CYCLE

- Cell-cycle control is similar in all eukaryotes

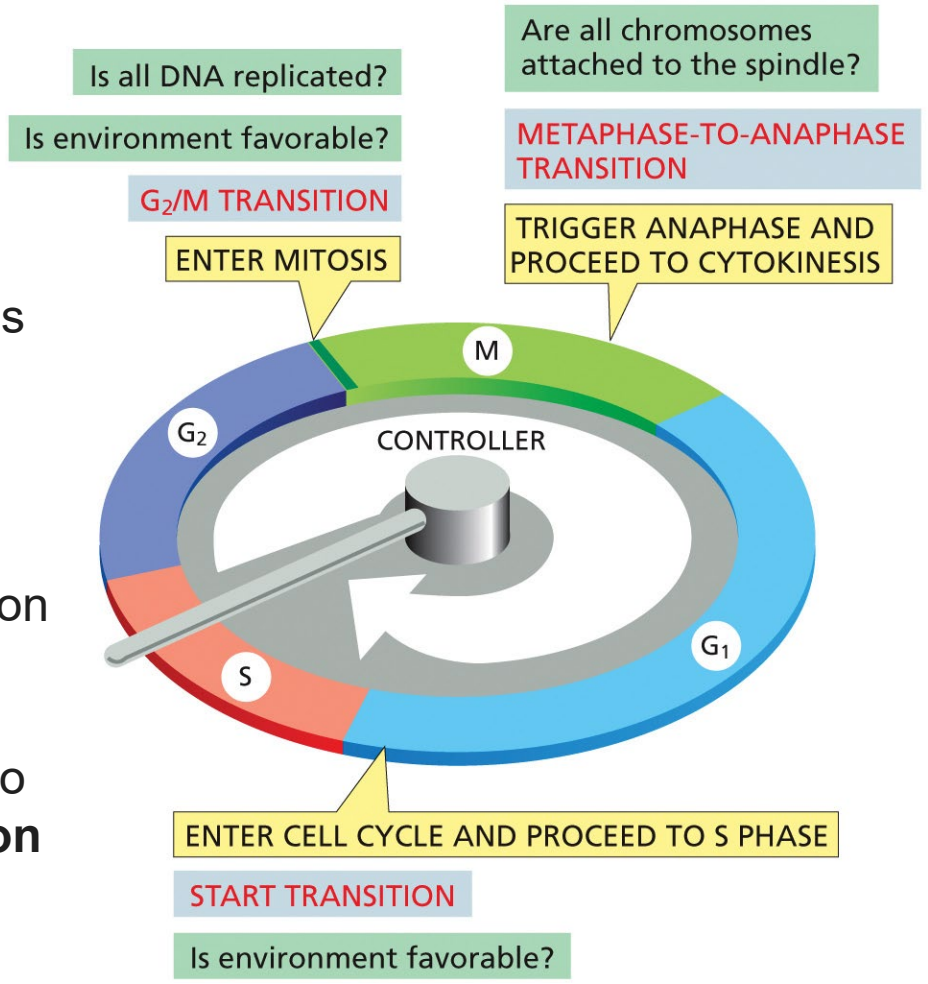


# THE CELL-CYCLE CONTROL SYSTEM

- The cell-cycle control system triggers the major events of the cell cycle

- Cell-cycle control system triggers the essential processes of the cycle:
  - DNA replication
  - mitosis
  - cytokinesis
- Information about the completion of cell-cycle events, as well as signals from the environment, can cause the control system to arrest the cycle at the **transition points**

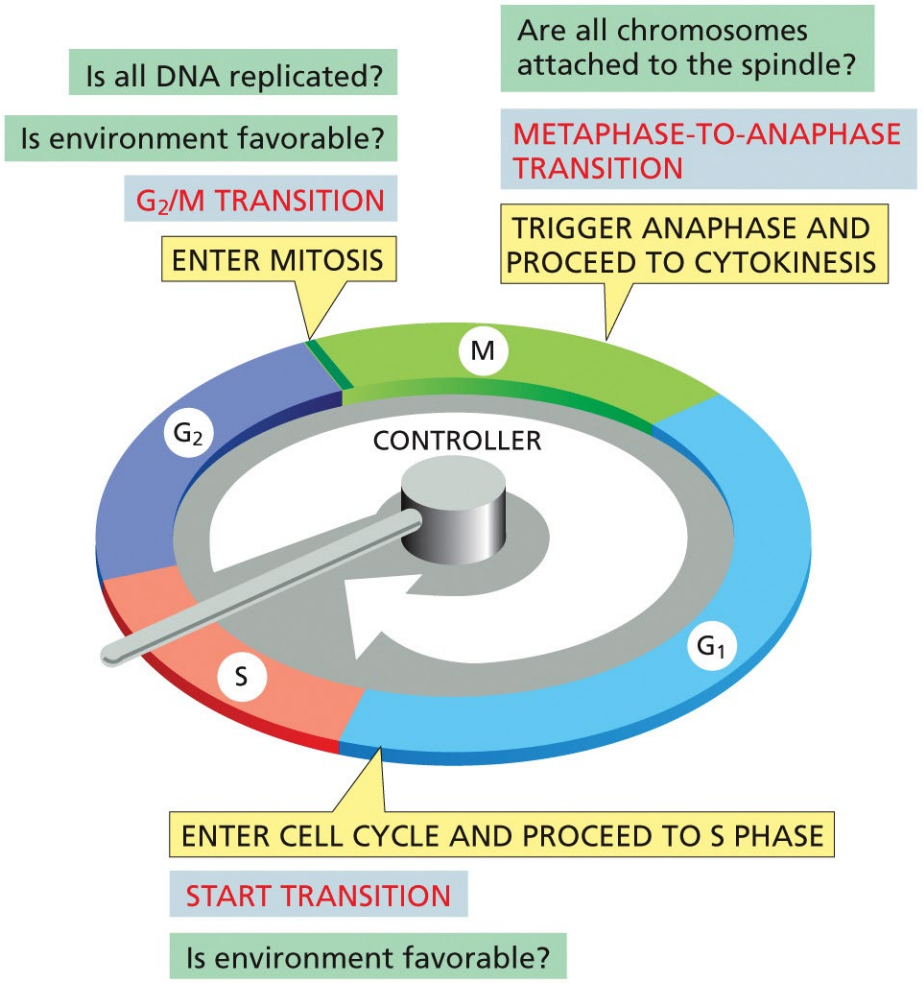


# THE CELL-CYCLE CONTROL SYSTEM

- The cell-cycle control system triggers the major events of the cell cycle

- Control system is based on **molecular switches**
- Typically, **on/off + irreversible**
- **Robust + adaptive**

- 3 major transitions



# THE CELL-CYCLE CONTROL SYSTEM

- The cell-cycle control system depends on *cyclically activated* **cyclin-dependent protein kinases**

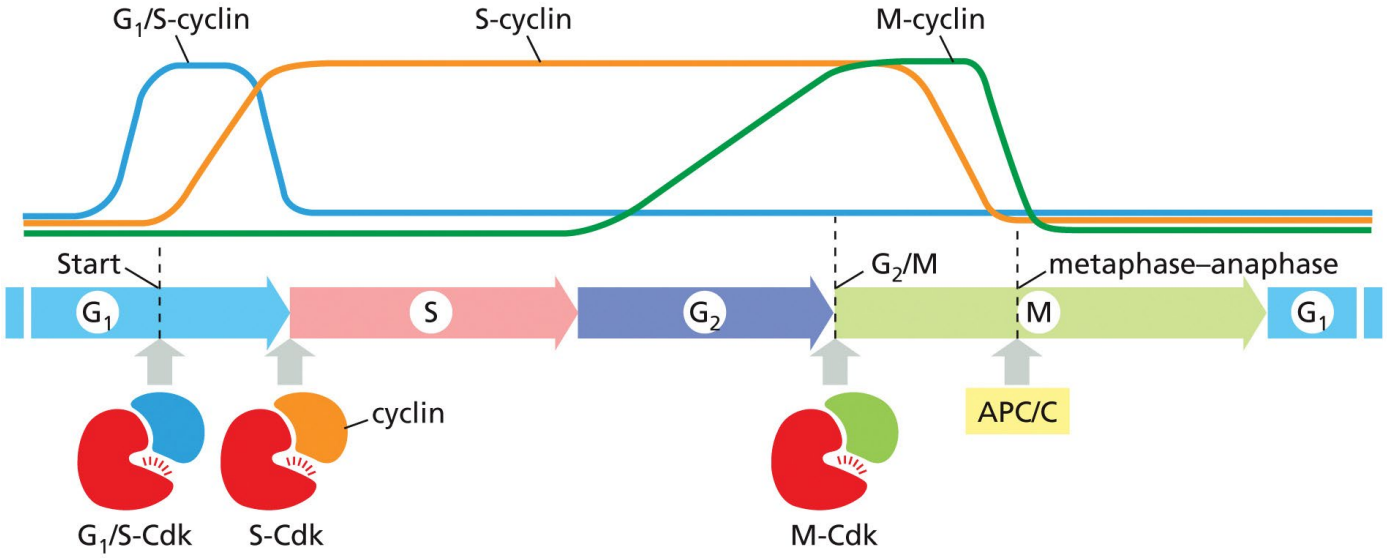

- **Cyclin** forms a **complex with Cdk** → the **protein kinase is activated** to trigger specific cell-cycle events.
- Without cyclin, Cdk is inactive.

- **Cdk:**
  - Constant levels
  - Phosphorylate proteins that mediate cell cycle
- **Cyclin:**
  - Regulate Cdk activity + direct to correct target
  - Cycles of synthesis and degradation in each cell cycle

# CYCLIN–CDK COMPLEXES OF THE CELL-CYCLE CONTROL SYSTEM

*The concentrations of the three major cyclin types oscillate during the cell cycle*

*The concentrations of Cdks do not change.*

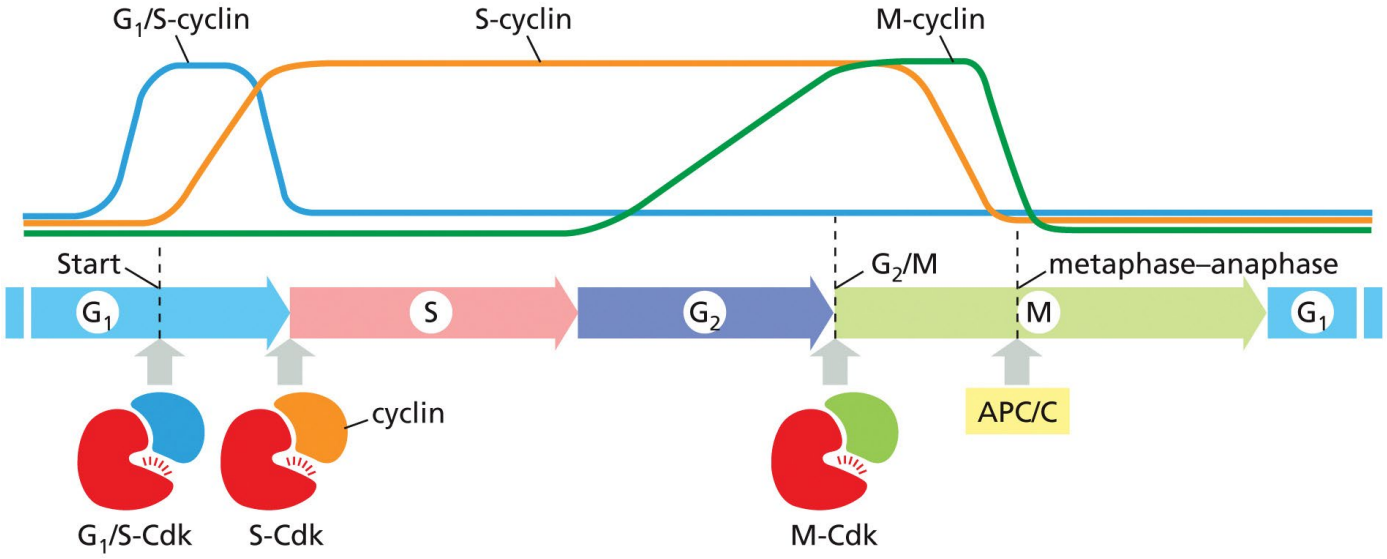



- Each Cdk-cyclin complex acts on specific target proteins
- Cdk-cyclin complex activity and levels regulated by:
    - Phosphorylation & dephosphorylation
    - Degradation of cyclins by proteasome

# CYCLIN–CDK COMPLEXES OF THE CELL-CYCLE CONTROL SYSTEM

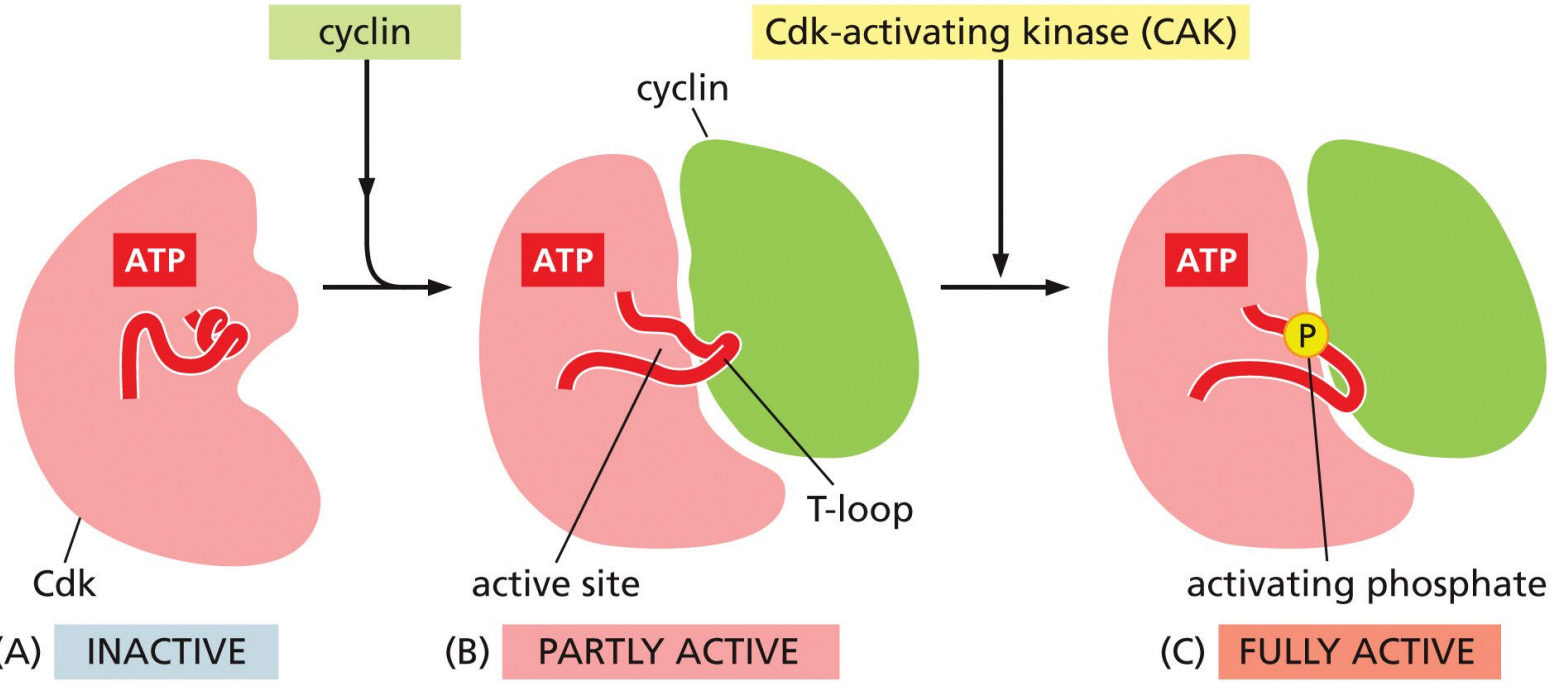
*The concentrations of the three major cyclin types oscillate during the cell cycle*

*The concentrations of Cdks do not change.*




- **$G_1$/S-cyclin:** levels rise in late $G_1$ → $G_1$/S-Cdk complexes → **Start transition**
- **S-cyclins**: S-Cdk complexes in $G_1$ after **start**→ trigger **DNA replication** + **early mitotic events**
- **M-cyclins:** M-Cdk complexes form during $G_2$ but are held in an inactive state → activated at the end of $G_2$ → trigger **entry into mitosis** at the **$G_2$/M transition**
- (APC/C, initiates the metaphase-to-anaphase transition)

# CDK ACTIVATION AND SPESIFICITY




1. **Active site is blocked** by a region of the protein called the T-loop

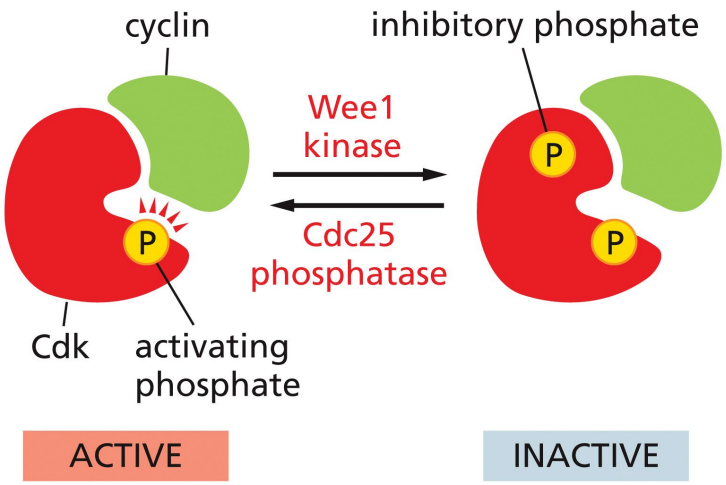
2. The **binding of cyclin** causes the T-loop to move out of the active site, resulting in *partial activation* of Cdk2

3. **Phosphorylation of Cdk2** (by CAK) at a threonine residue in the T-loop *further activates the enzyme* by changing the shape of the T-loop

- Activity of cyclin/Cdk complexes targeted by cyclins and by availability of substrates (different at different states of cell cycle)

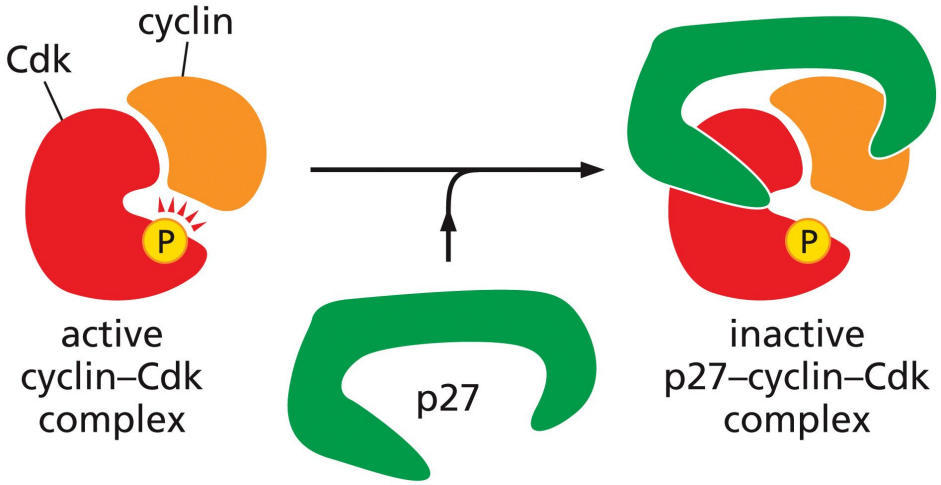
# The regulation of Cdk activity by inhibitory phosphorylation and Cdk inhibitor proteins

Wee1 kinase inactivates



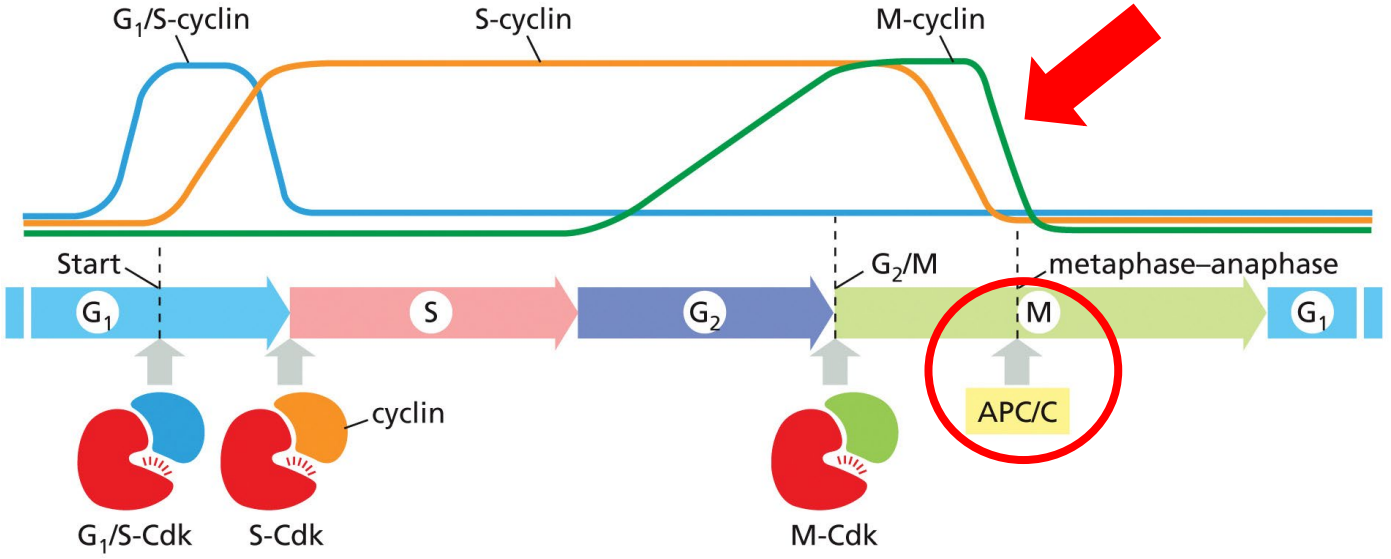

Cdc25 phosphatase activates

p27 binds to both the cyclin and Cdk in the complex, distorting the active site of the Cdk




It also inserts into the ATP-binding site, further inhibiting the enzyme activity

# CYCLIN–CDK COMPLEXES OF THE CELL-CYCLE CONTROL SYSTEM

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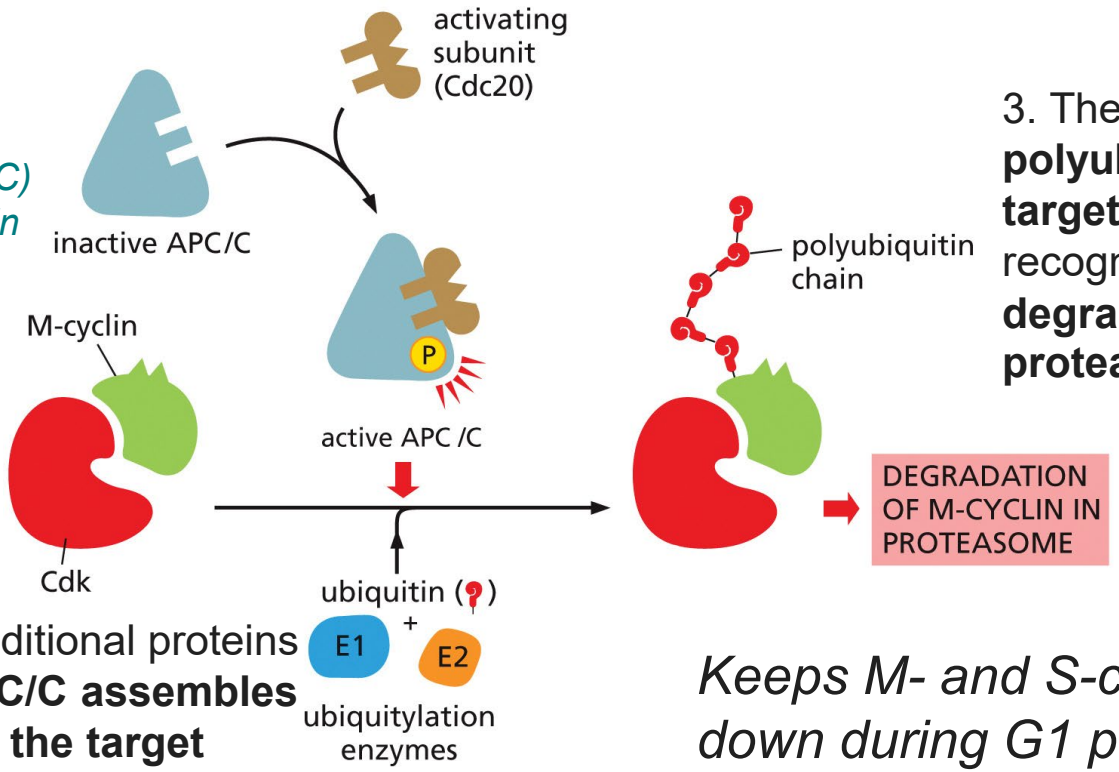
- **APC/C, initiates the metaphase-to-anaphase transition**

# THE CONTROL OF PROTEOLYSIS BY THE APC/C

- The anaphase-promoting complex/cyclosome (APC/C) triggers the metaphase-to-anaphase transition

1. The **APC/C is activated in metaphase** by association with Cdc20, which recognizes specific amino acid sequences on **M-cyclin, S-cyclin**, and other target proteins.

*Anaphase-Promoting Complex (APC/C) is an E3 ubiquitin ligase*




3. The **polyubiquitylated target** is then recognized and **degraded in a proteasome.**

2. With the help of two additional proteins called E1 and E2, the **APC/C assembles polyubiquitin chains on the target protein**.

*Keeps M- and S-cyclin levels down during G1 phase*

# The marking of proteins with ubiquitin

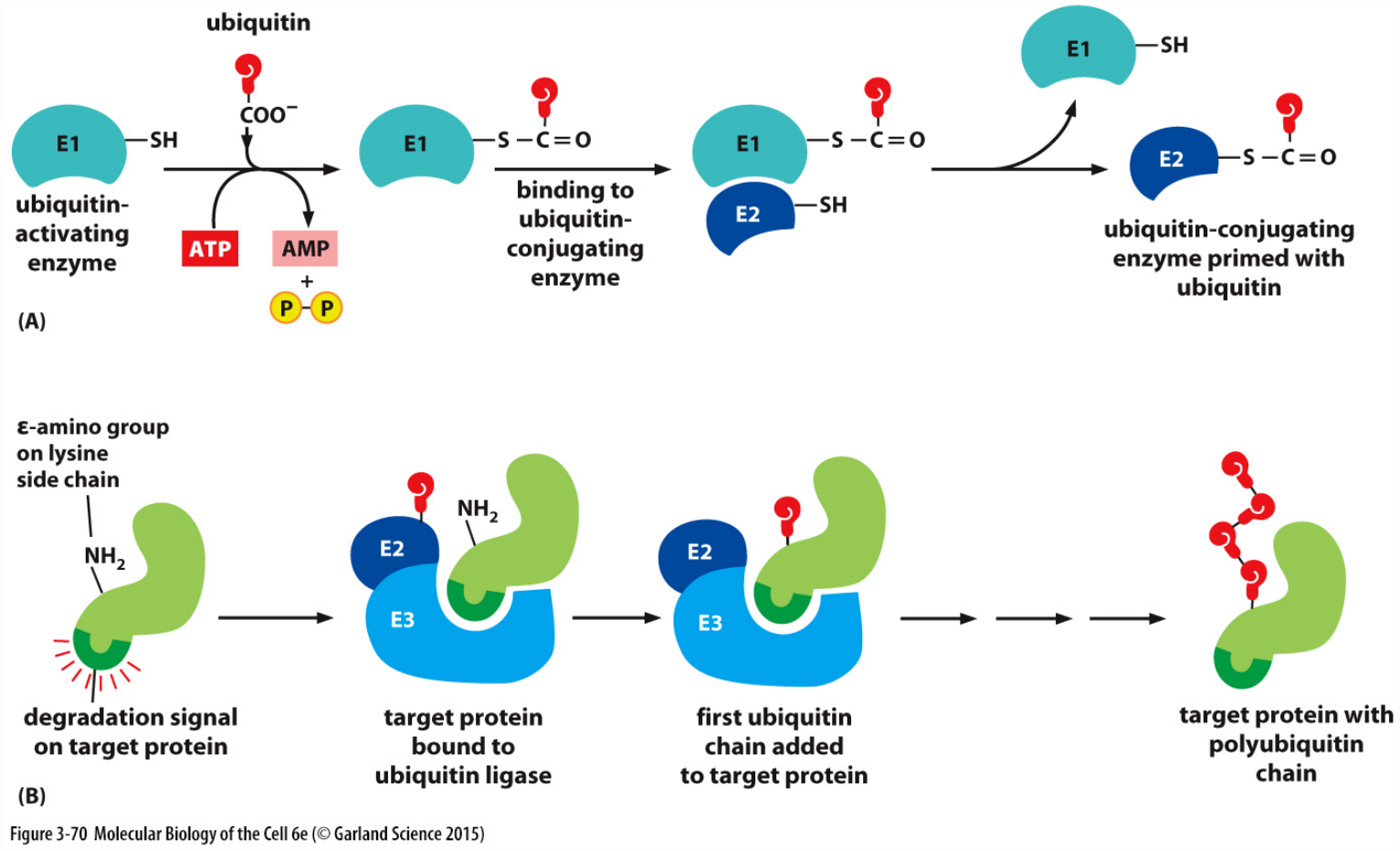
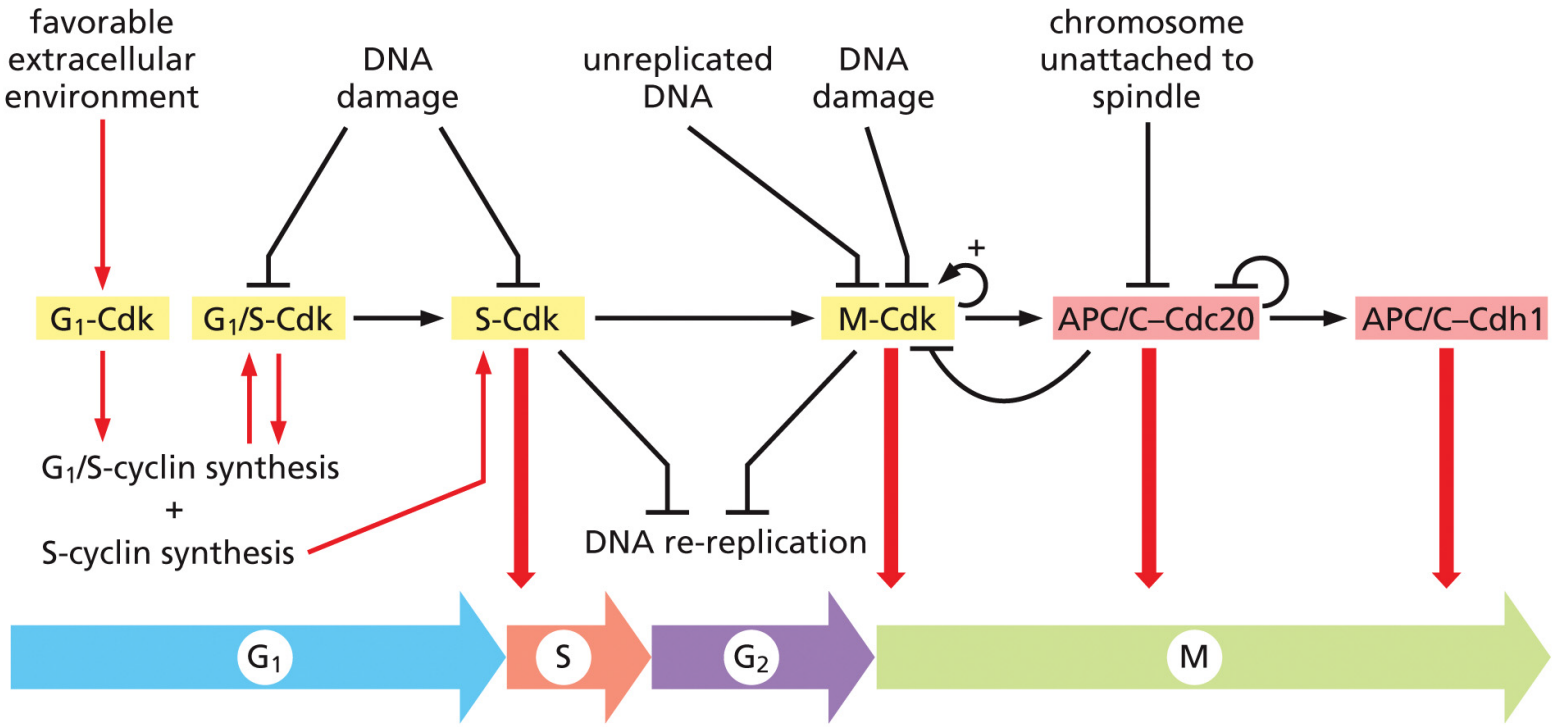



Figure 3-70 Molecular Biology of the Cell 6e (© Garland Science 2015)

E3: ubiquitin ligase

# SEQUENTIAL ACTIVATION OF CDKS DURING THE CELL CYCLE

- The cell-cycle control system functions as a linked series of biochemical switches



# CONTROL OF CELL DIVISION AND CELL GROWTH

- Extracellular signal molecules regulate cell growth, division and survival
- Signals are generally soluble secreted proteins

- **Mitogens** stimulate cell division via stimulating G1/S-Cdk activity -> more cells
- **Growth factors** stimulate cell growth by promoting synthesis of proteins, etc. -> bigger cells
- **Survival factors** promote cell survival by suppressing apoptosis

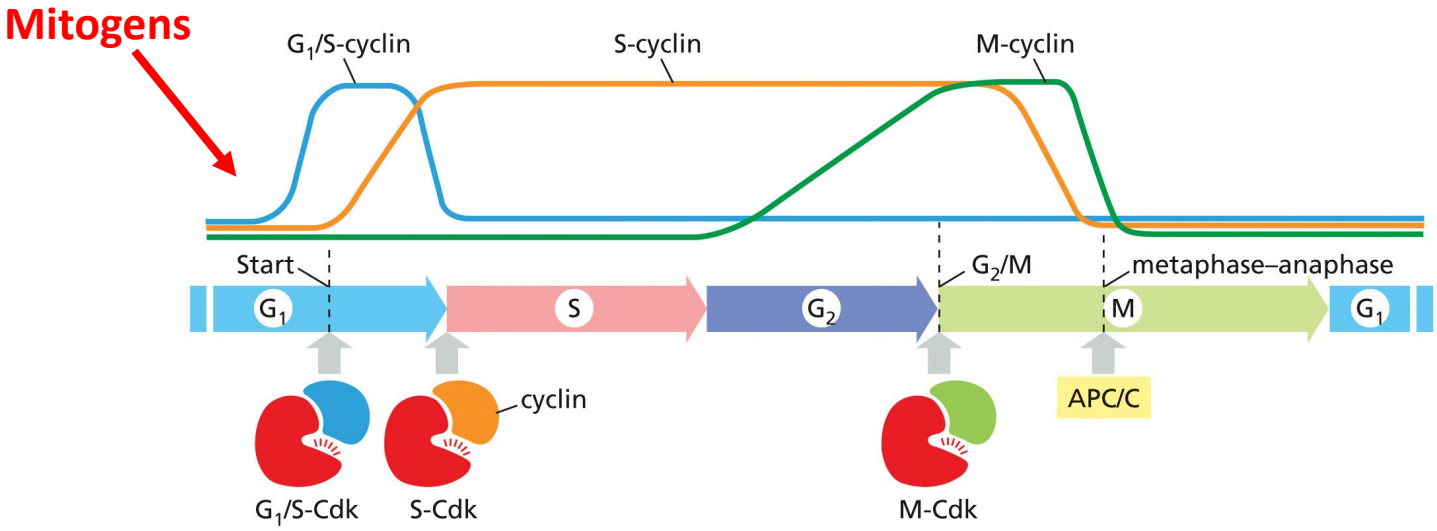
# CONTROL OF CELL DIVISION AND CELL GROWTH

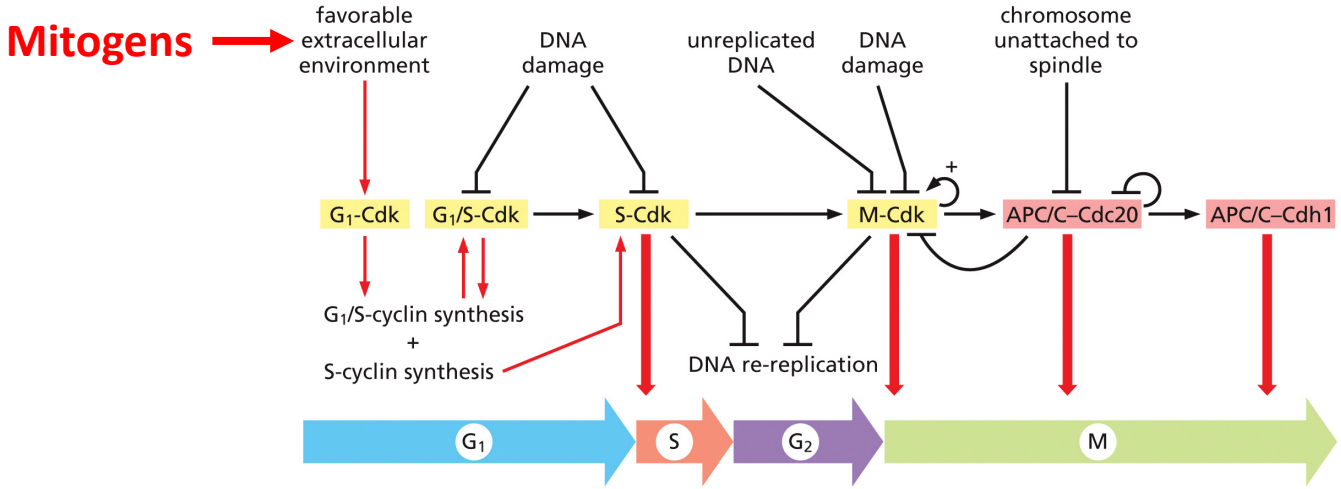
- **Mitogens stimulate cell division**
- Growth factors stimulate cell growth
- Survival factors suppress apoptosis

>50 mitogens in human, including *Platelet-derived growth factor* (*PDGF*) and epidermal growth factor (EGF)

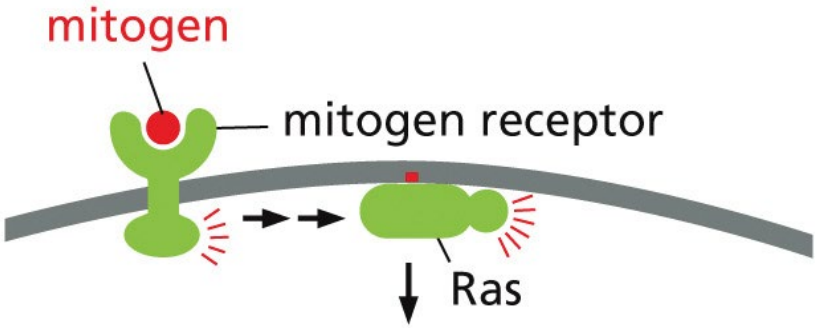
# Mitogens release the brakes on Cdk activity in G1 phase

→ *Cell-cycle entry*



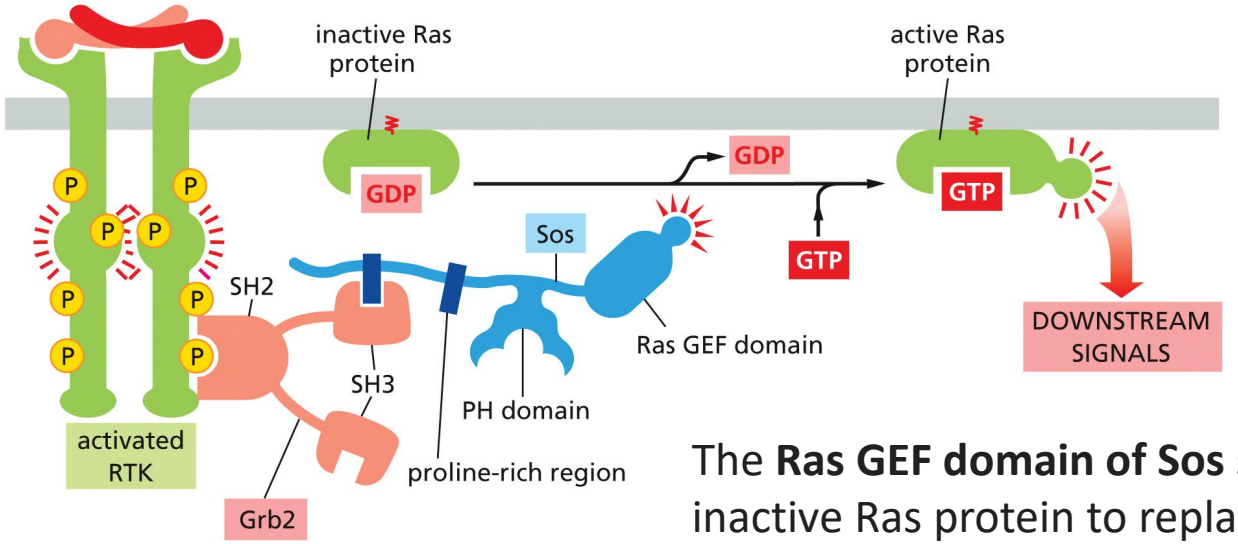



# MITOGEN STIMULATION OF CELL-CYCLE ENTRY




- Mitogens Stimulate $G_1$-Cdk and $G_1$/S-Cdk Activities

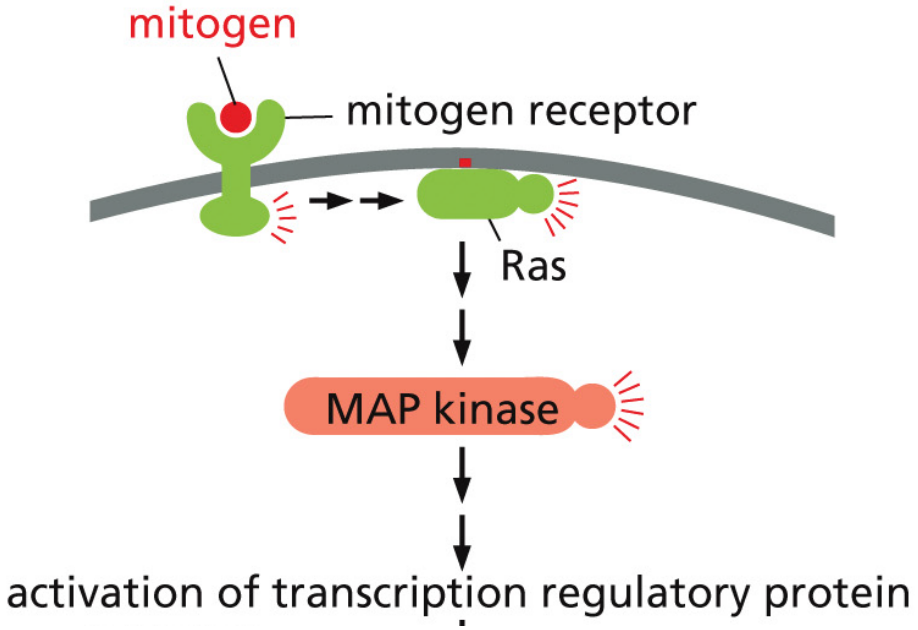
The Ras GAP increases hydrolysis of bound GTP in Ras -> inactivates

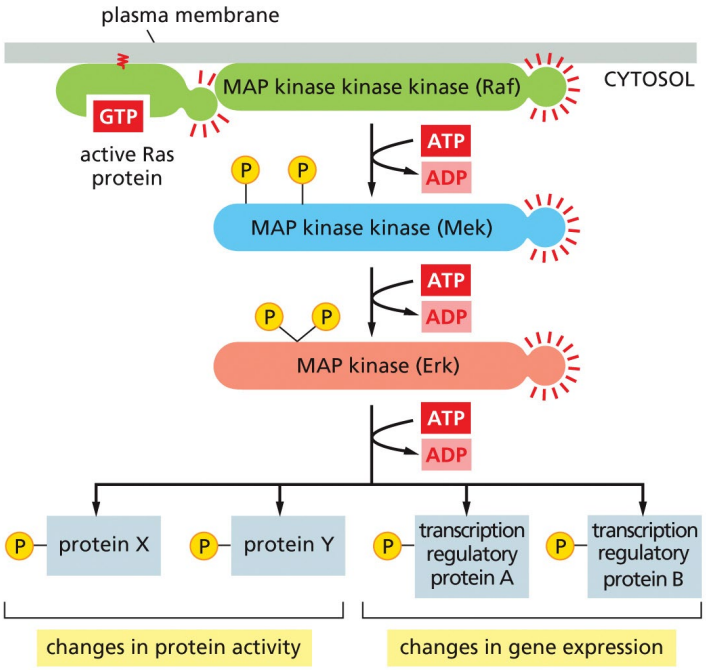



The **Ras GEF domain of Sos** stimulates the inactive Ras protein to replace its bound GDP by GTP, which **activates Ras** to relay the signal downstream.

# MITOGEN STIMULATION OF CELL-CYCLE ENTRY

- Mitogens Stimulate $G_1$-Cdk and $G_1$/S-Cdk Activities

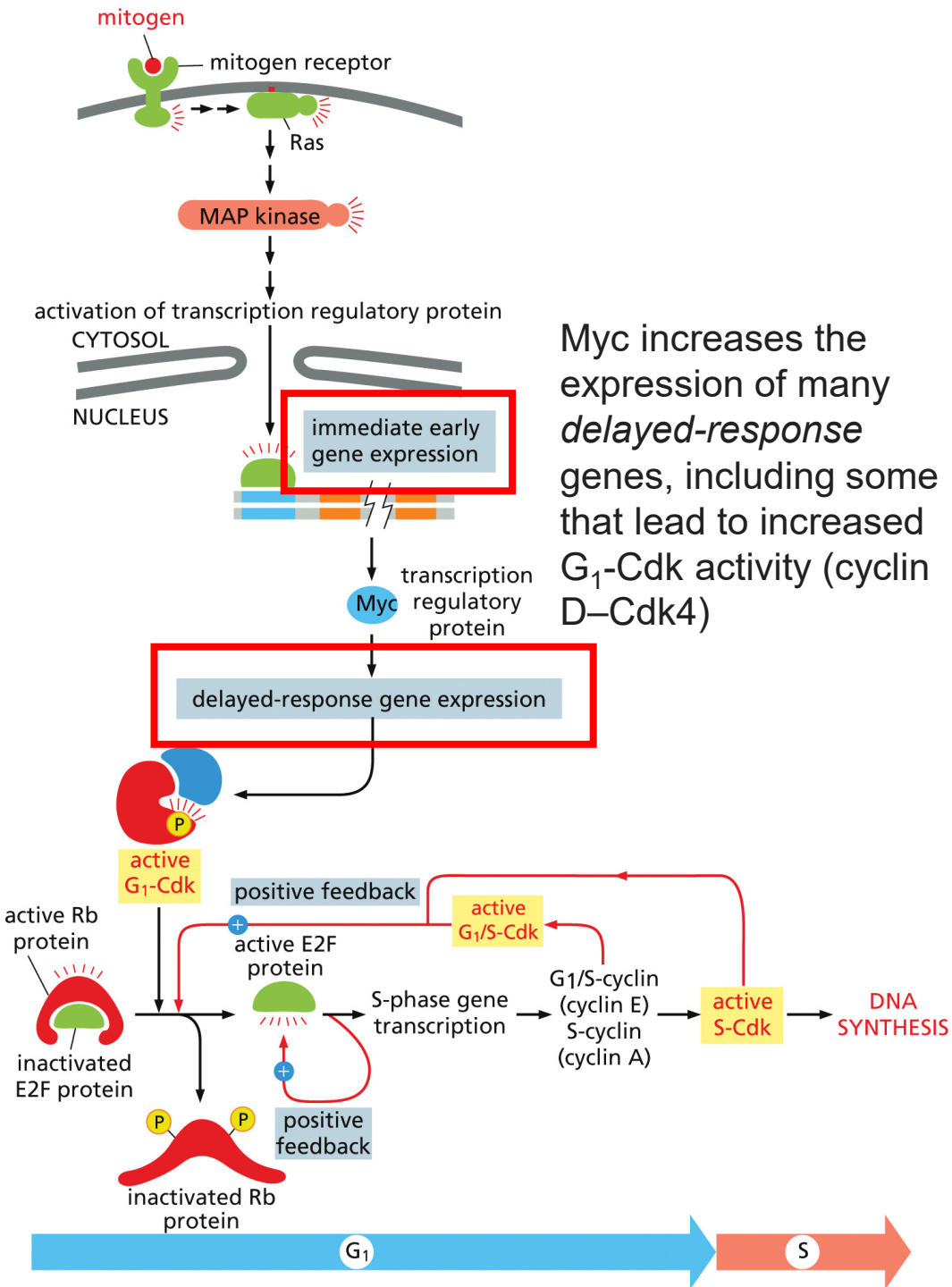


# MITOGEN STIMULATION OF CELL-CYCLE ENTRY

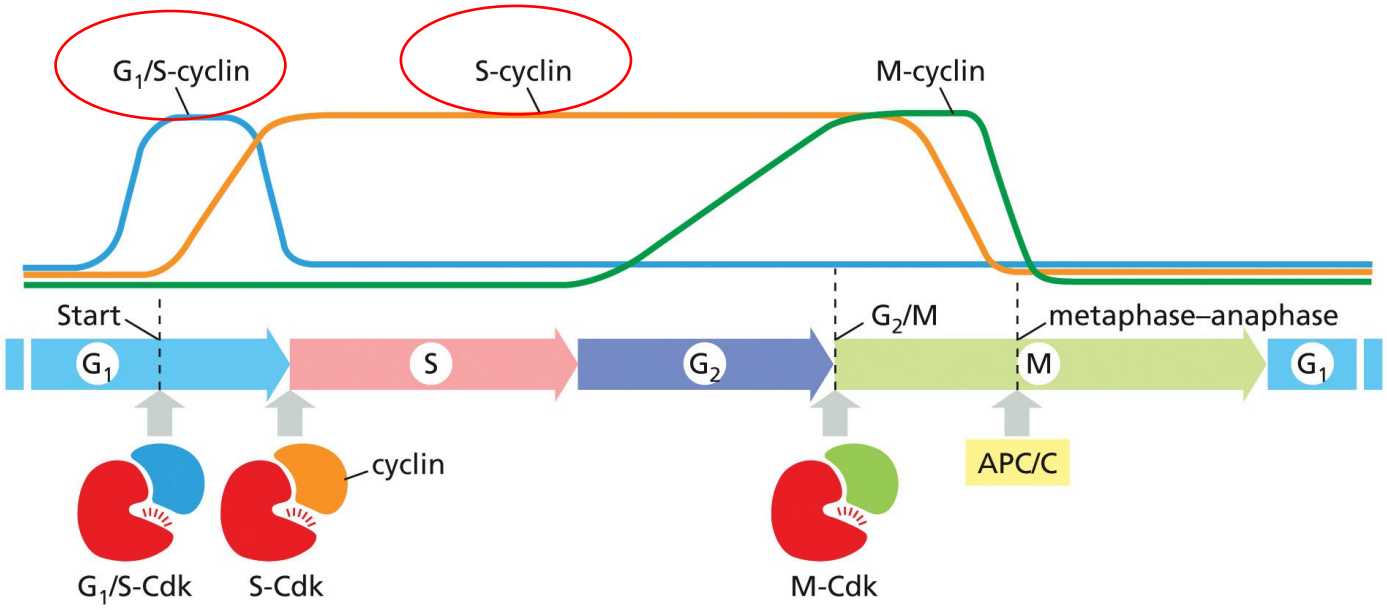
- Mitogens Stimulate $G_1$-Cdk and $G_1$/S-Cdk Activities

- $G_1$-Cdk activity (cyclin D–Cdk4), triggers the phosphorylation of members of the Rb family of proteins → inactivates the Rb proteins, freeing the gene regulatory protein E2F

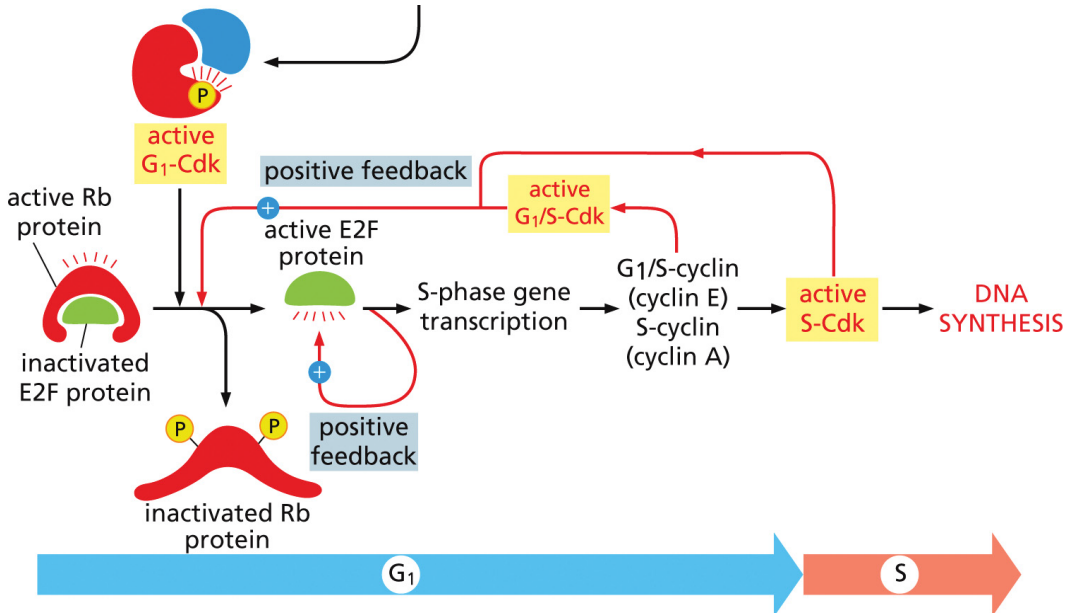



Myc increases the expression of many *delayed-response* genes, including some that lead to increased $G_1$-Cdk activity (cyclin D–Cdk4)

# MITOGEN STIMULATION OF CELL-CYCLE ENTRY




- E2F activates the transcription of $G_1$/S genes, including **the genes for a $G_1$/S-cyclin** (cyclin E) **and S-cyclin** (cyclin A)

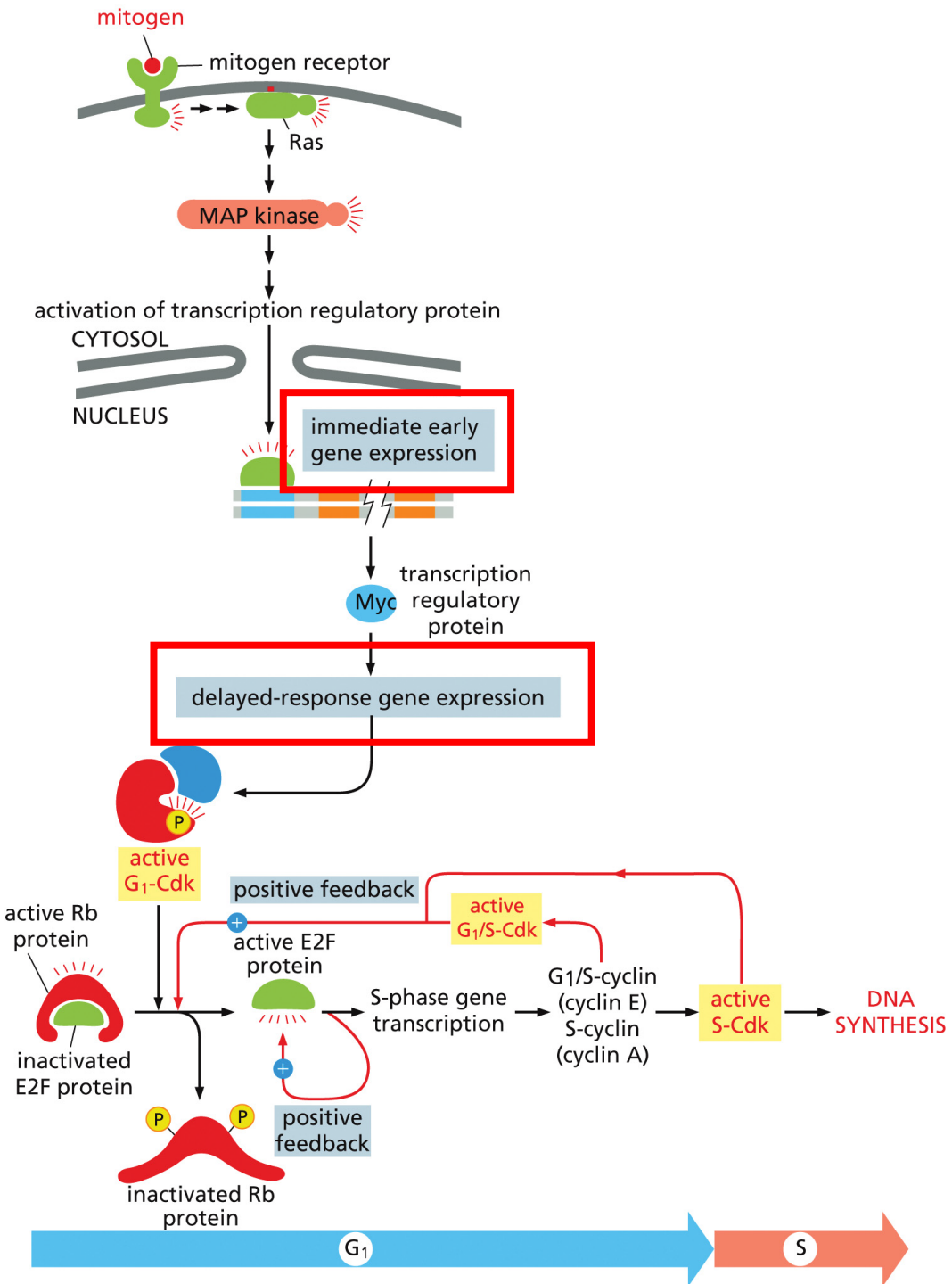


# MITOGEN STIMULATION OF CELL-CYCLE ENTRY

- Mitogens Stimulate $G_1$-Cdk and $G_1$/S-Cdk Activities

- The resulting $G_1$/S-Cdk and S-Cdk activities further enhance Rb protein phosphorylation, forming a **positive feedback loop**.
- E2F proteins also stimulate the transcription of their own genes, forming **another positive feedback loop**

→ **complete and irreversible cell cycle**



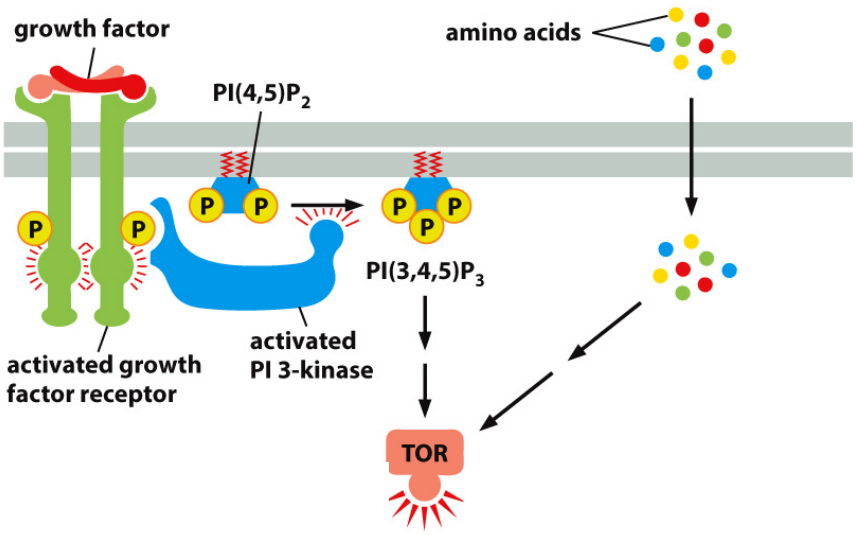
# CONTROL OF CELL DIVISION AND CELL GROWTH

- Mitogens stimulate cell division
- **Growth factors stimulate cell growth**
- Survival factors suppress apoptosis

# CELL PROLIFERATION IS ACCOMPANIED BY CELL GROWTH

- Cell division must be accompanied by cell growth
- Extracellular growth factor activate cells stimulating rate of synthesis and decreasing rate of degradation of cellular macromolecules
- Protein kinase TOR is at heart of all growth regulatory pathways in eukaryotes

# STIMULATION OF CELL GROWTH BY EXTRACELLULAR GROWTH FACTORS AND NUTRIENTS




**Growth factors** bind to cell surface receptors → activation of *PI 3-kinase*, which promotes protein synthesis through a complex signaling pathway that leads to the activation of the **protein kinase TOR**.

Extracellular nutrients such as amino acids also help activate TOR.

4E-BP: inhibitor of the translation initiation factor eIF4E
S6: ribosomal subunit
S6K: S6 kinase

# STIMULATION OF CELL GROWTH BY EXTRACELLULAR GROWTH FACTORS AND NUTRIENTS

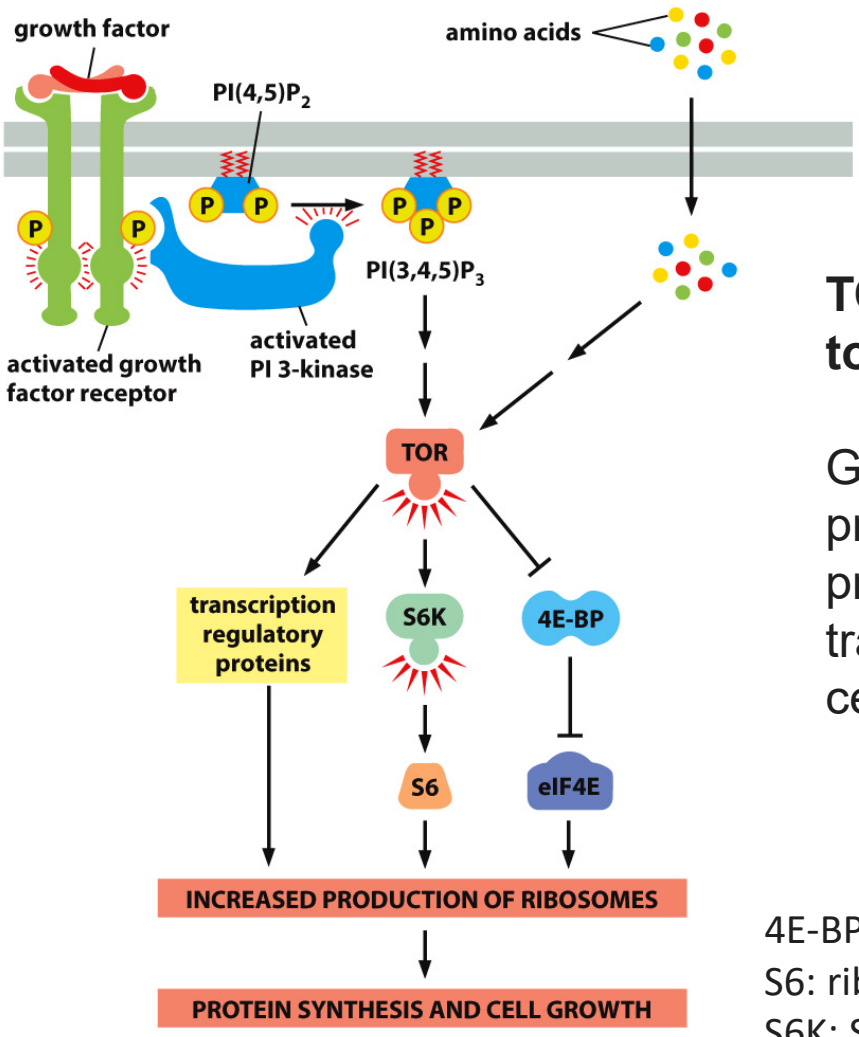


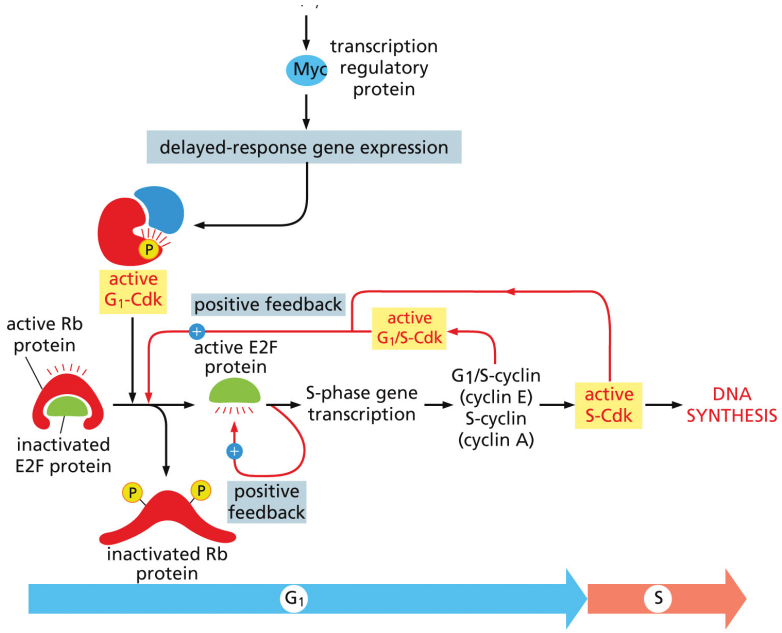

Figure 17-64 Molecular Biology of the Cell 6e (© Garland Science 2015)

**TOR phosphorylates multiple proteins to stimulate protein synthesis**

Growth factors also stimulate increased production of the transcription regulatory protein **Myc**, which activates the transcription of various genes that promote cell metabolism and growth

4E-BP: inhibitor of the translation initiation factor eIF4E
S6: ribosomal subunit
S6K: S6 kinase

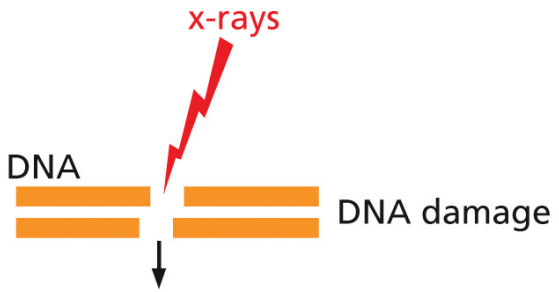
# STIMULATION OF CELL GROWTH BY EXTRACELLULAR GROWTH FACTORS AND NUTRIENTS



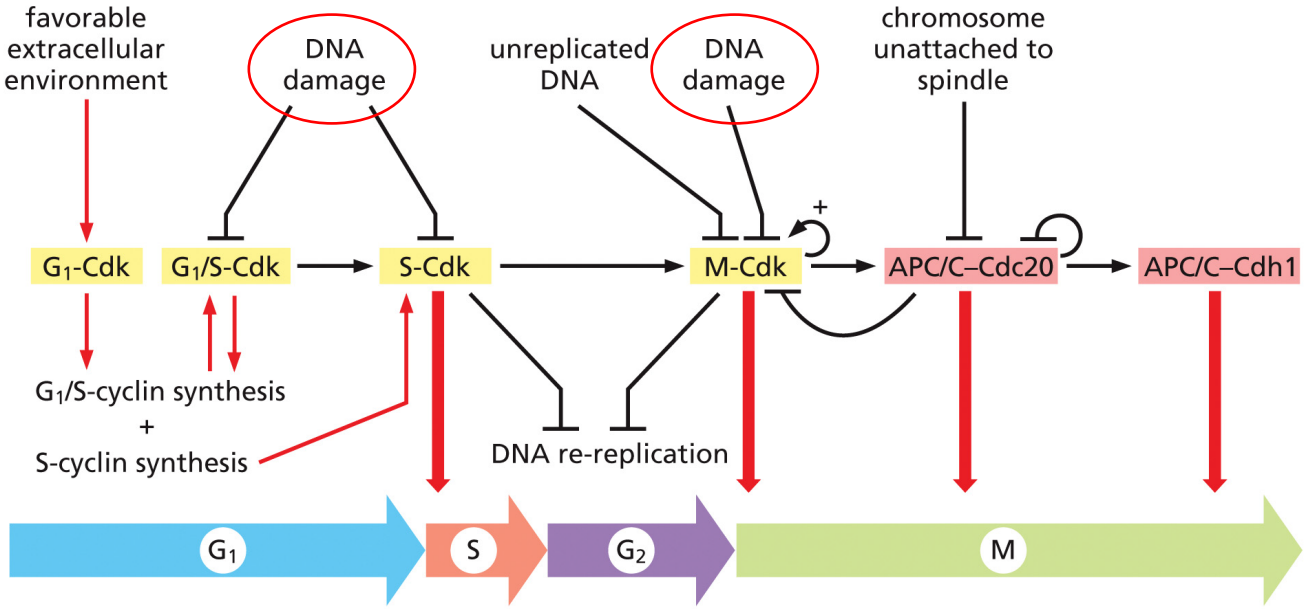

Growth factors also stimulate increased production of the transcription regulatory protein **Myc**, which activates the transcription of various genes that promote cell metabolism and growth

4E-BP: inhibitor of the translation initiation factor eIF4E
S6: ribosomal subunit
S6K: S6 kinase

# DNA DAMAGE BLOCKS CELL DIVISION

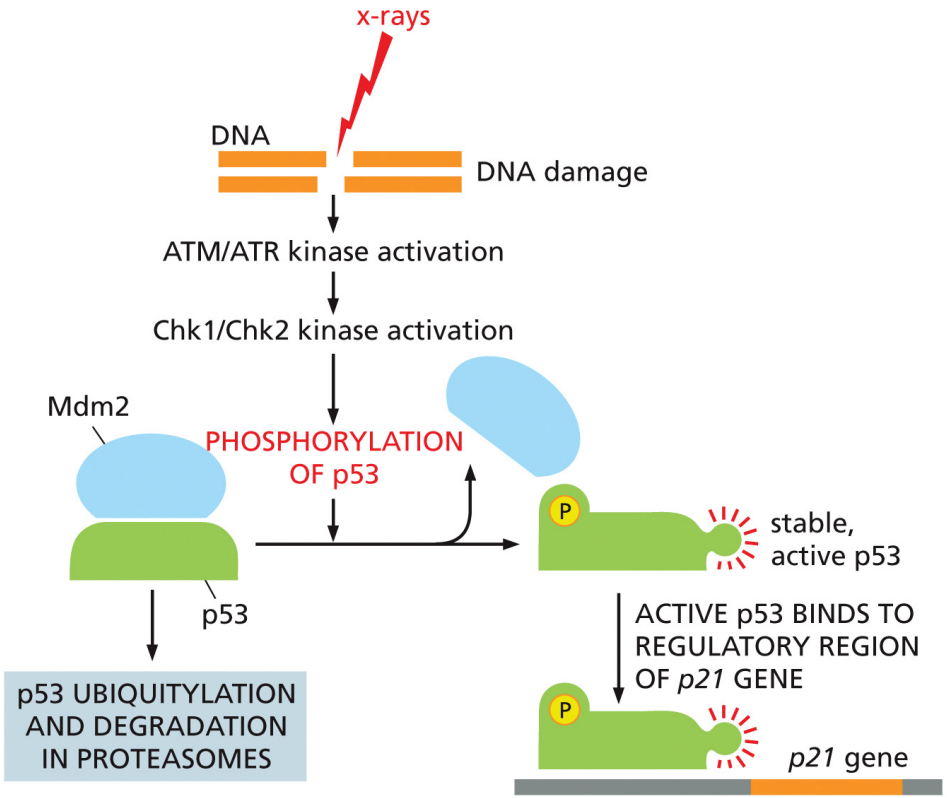


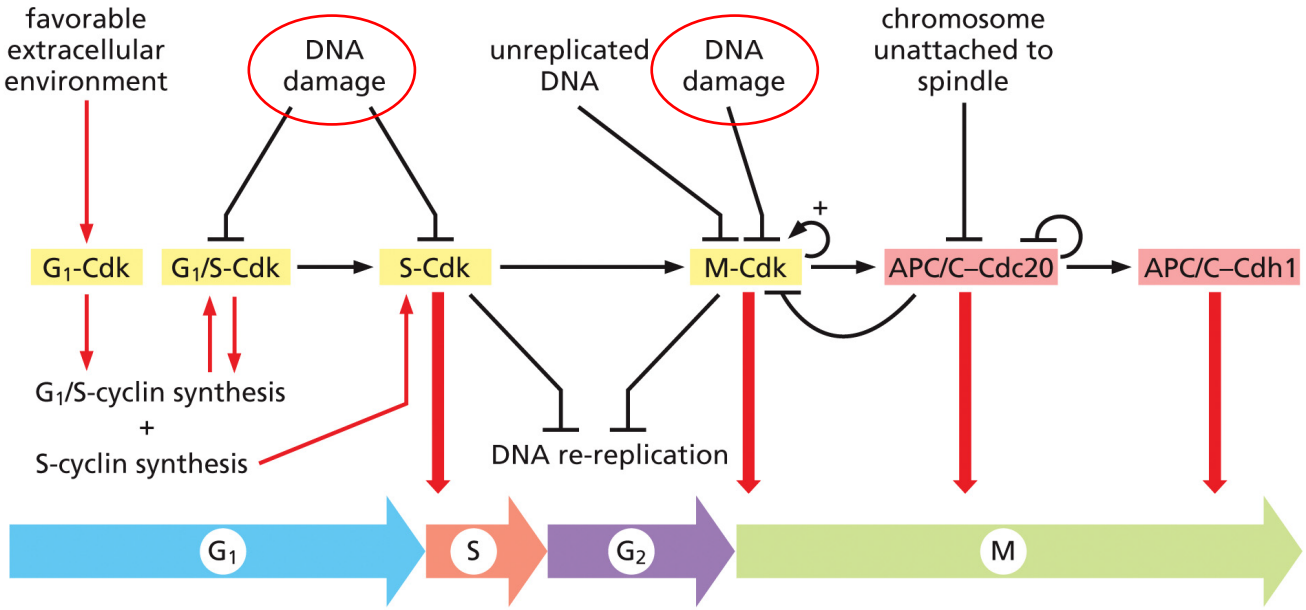

- Damaged DNA must be repaired prior to duplication



# DNA DAMAGE BLOCKS CELL DIVISION

- DNA damage leads to signaling cascade involving **p53** that activates synthesis of p21



# CONTROL OF CELL DIVISION AND CELL GROWTH

- The p21 binds and inactivates $G_1$/S-Cdk and S-Cdk complexes, arresting the cell in $G_1$