Cell Cycle Regulation

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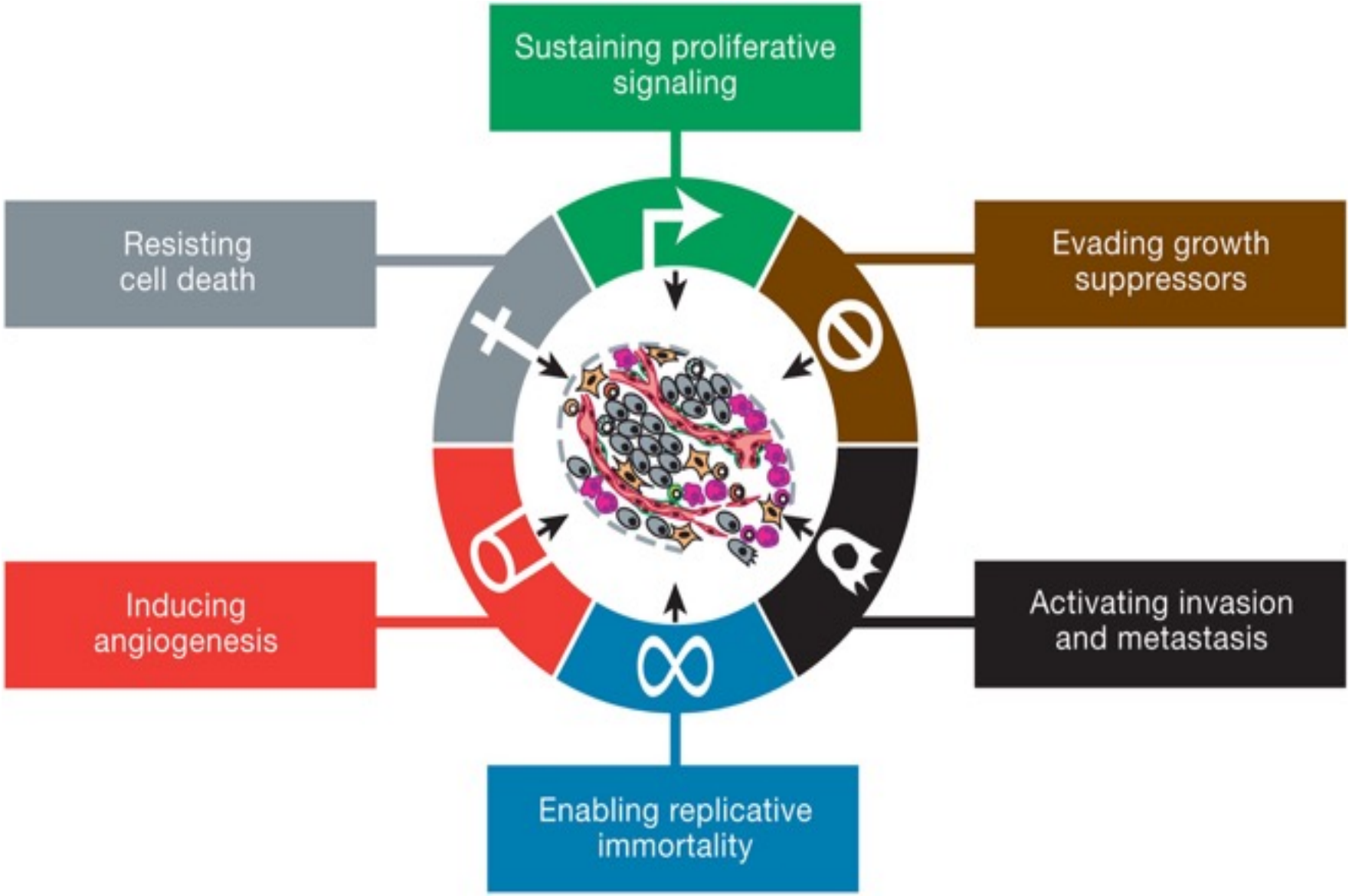
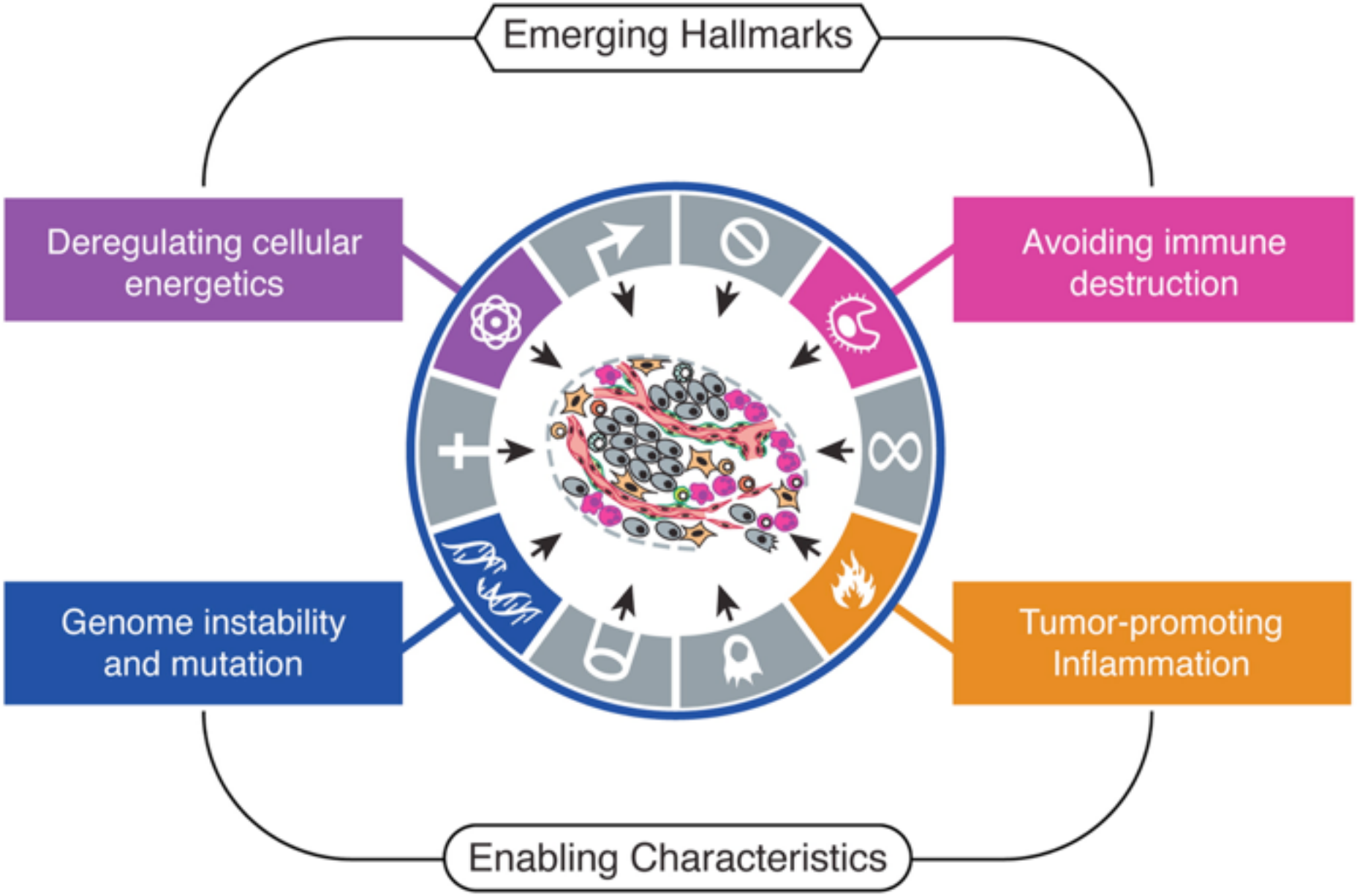
Outlines

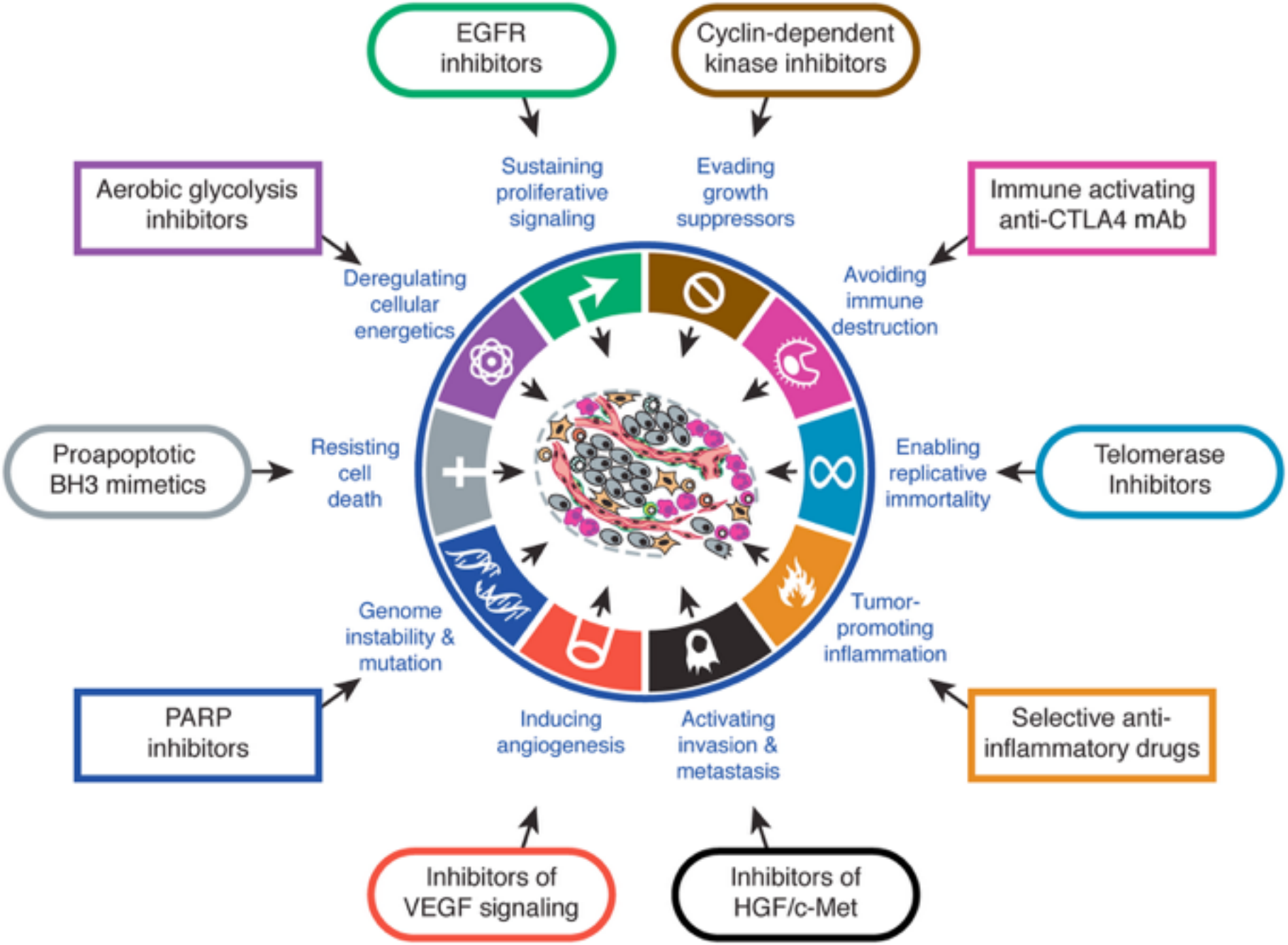
- **Phases of cell cycle**
- **Regulation of cell cycle by cyclins**
- **Regulation of cell cycle by cyclin-dependent kinases (CDKs)**
- **Regulation of cell cycle by inhibitors of cyclin-dependent kinases (CDKIs)**
- **Regulation of cyclins and CDKs functions by E2F-pRB pathway**



Hanahan and Weinberg Cell 2011



Hanahan and Weinberg Cell 2011



Hanahan and Weinberg Cell 2011

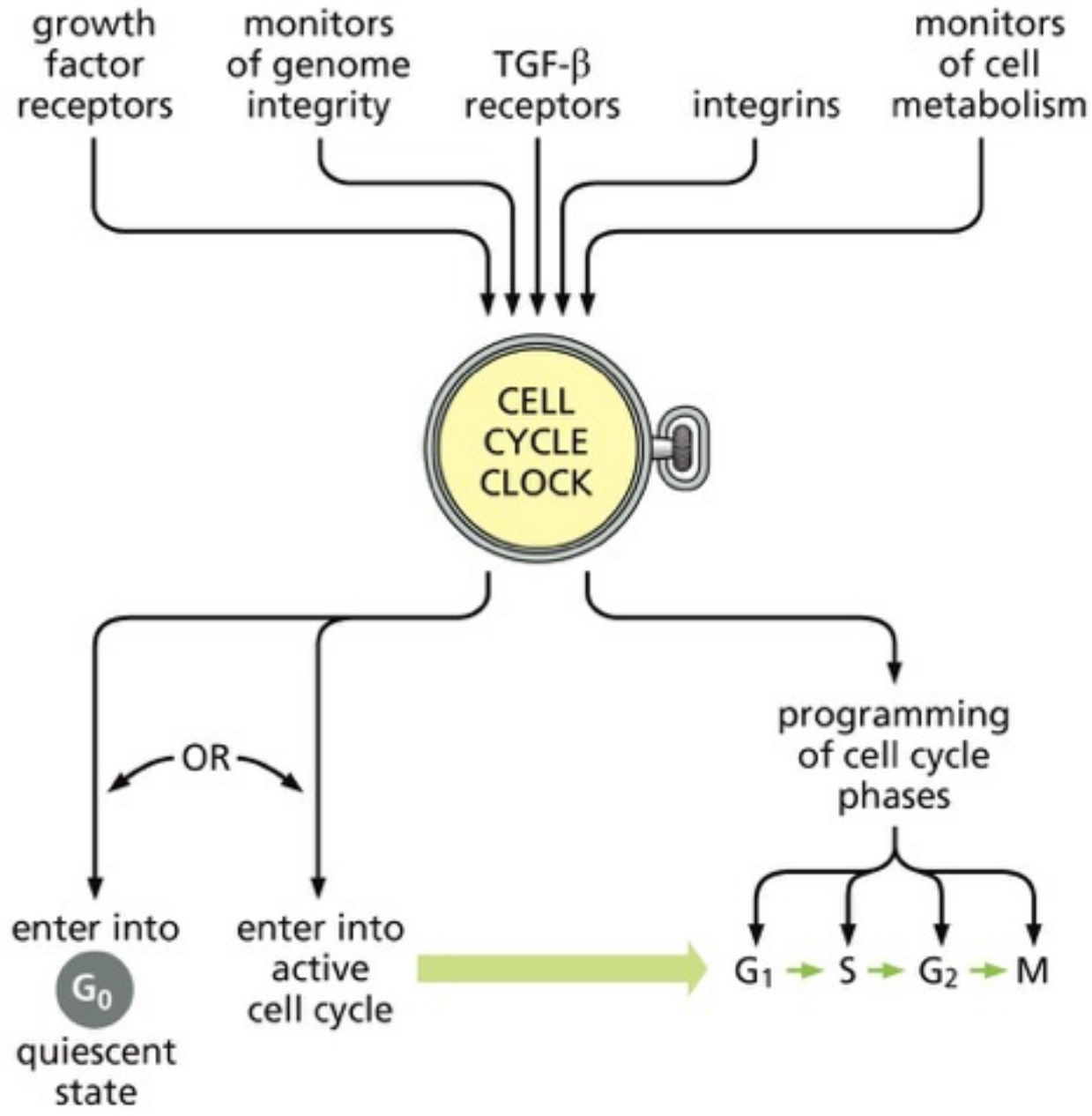


Figure 8.1 The Biology of Cancer (© Garland Science 2014)

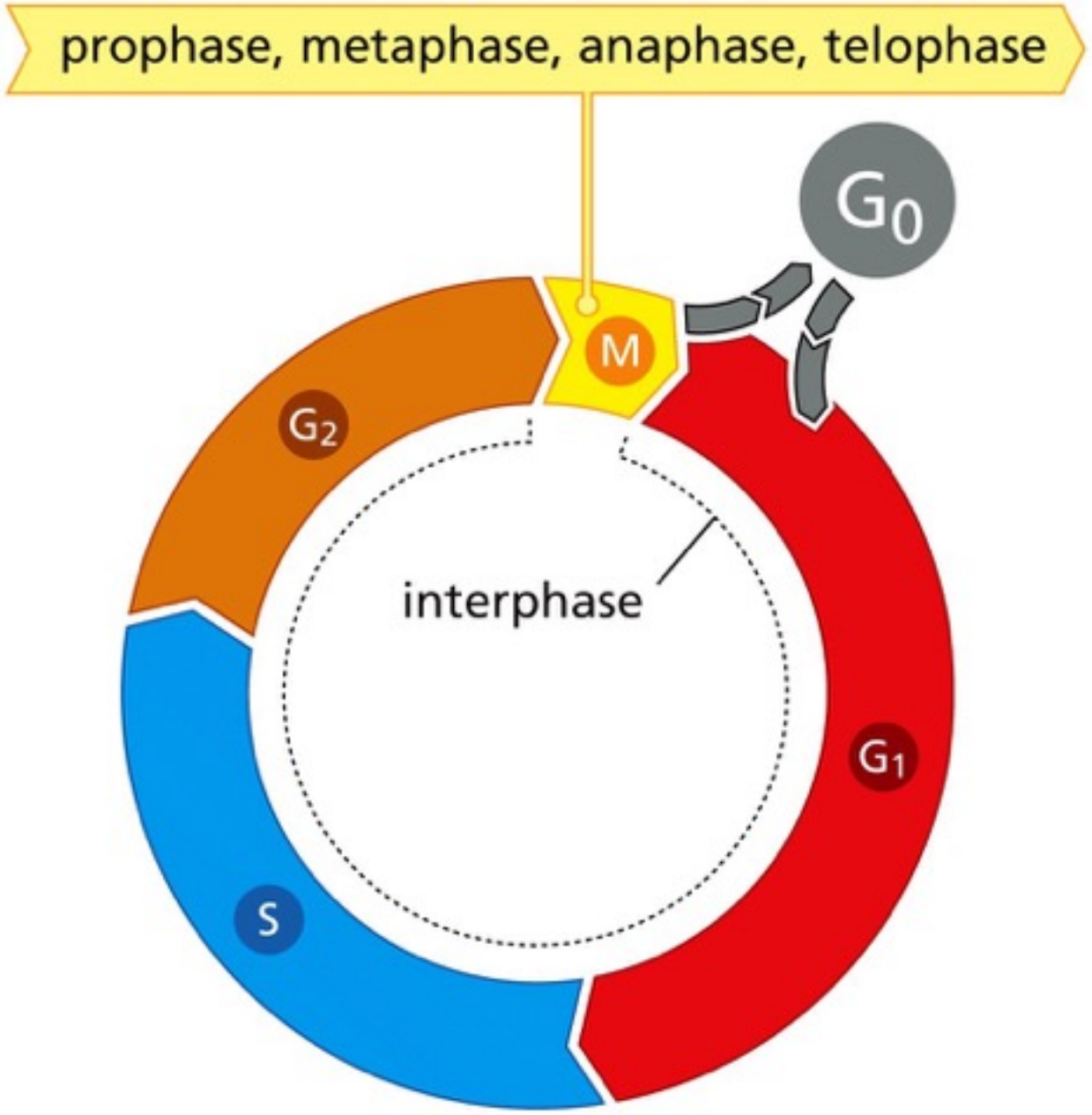
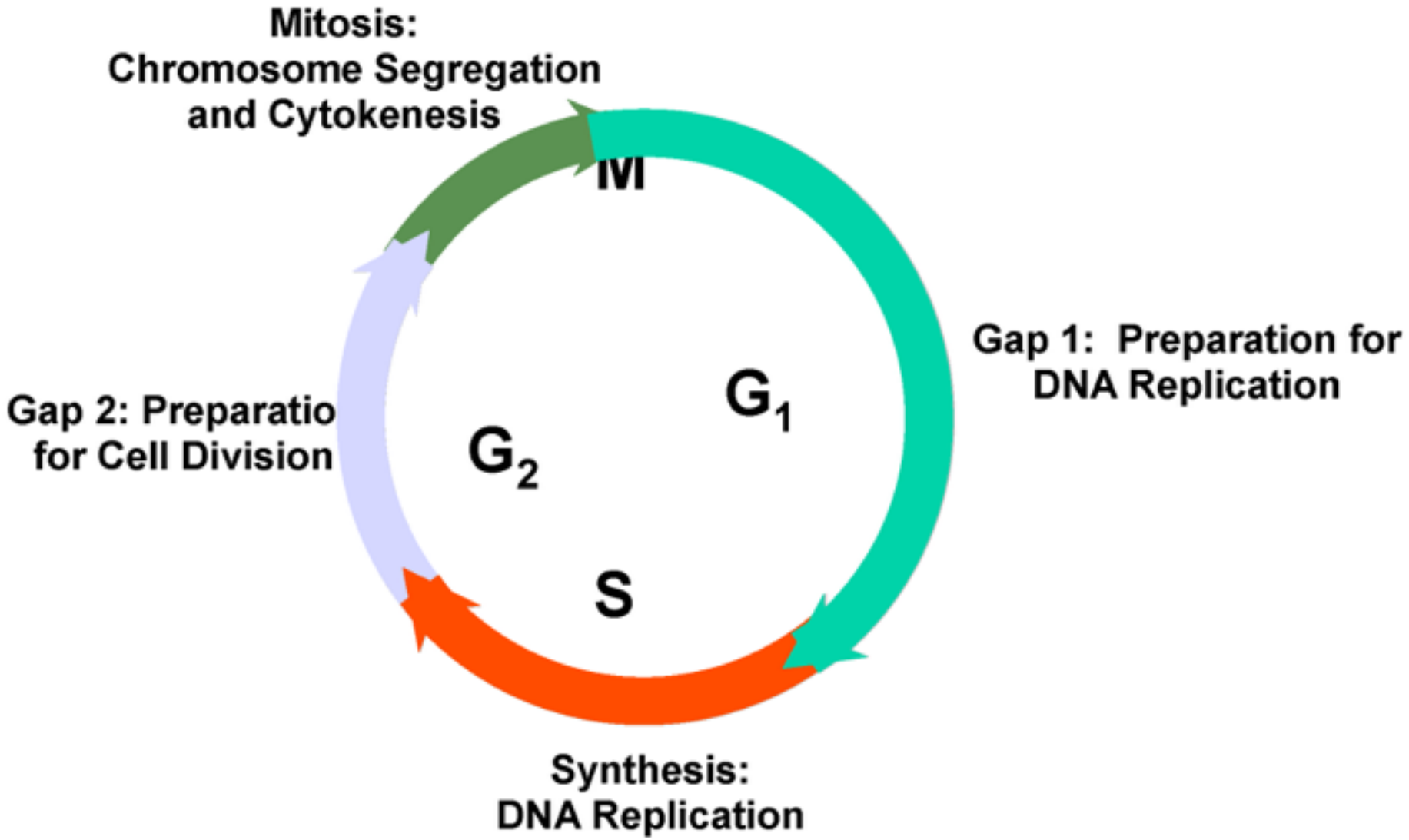
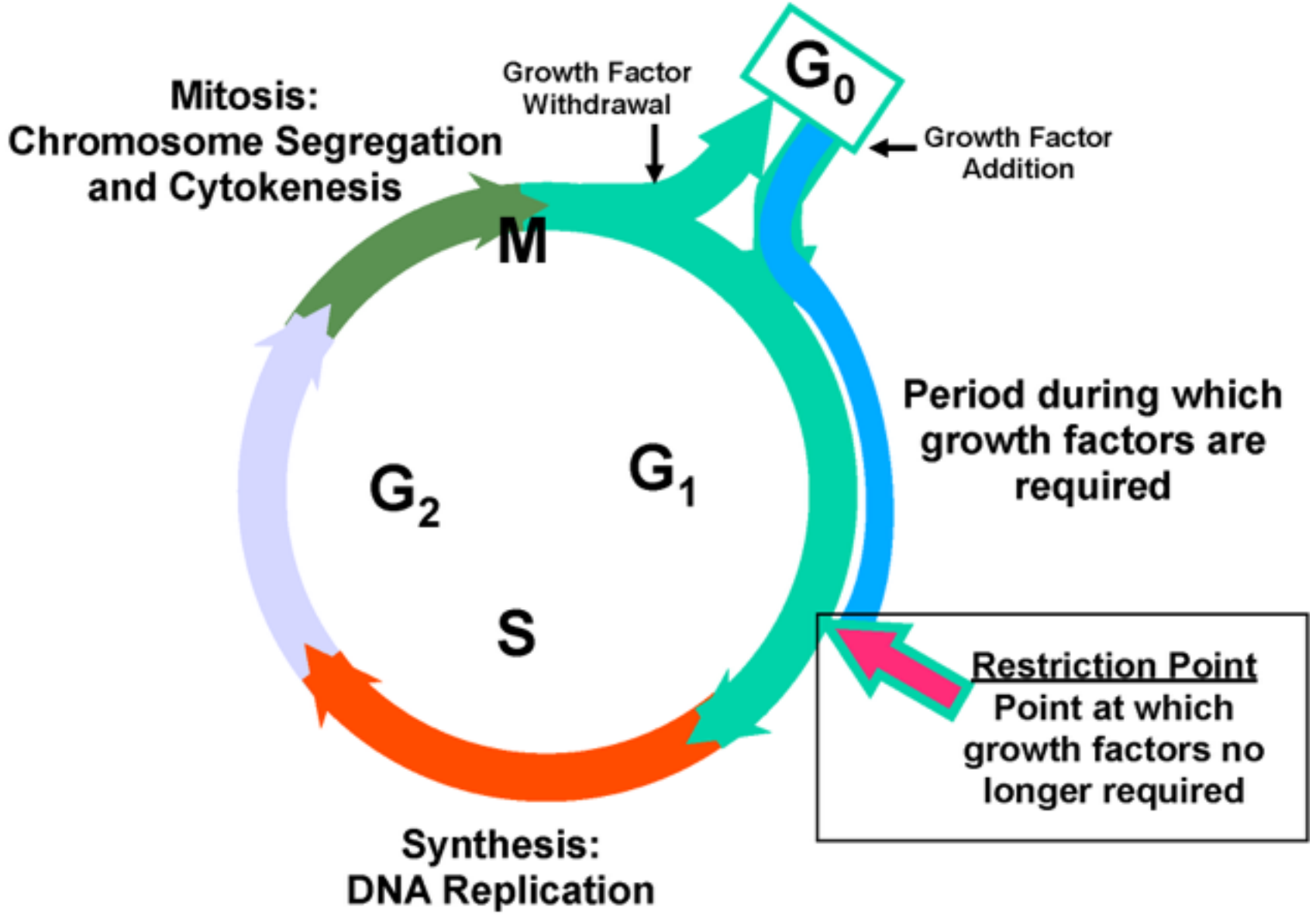


Figure 8.3b The Biology of Cancer (© Garland Science 2014)

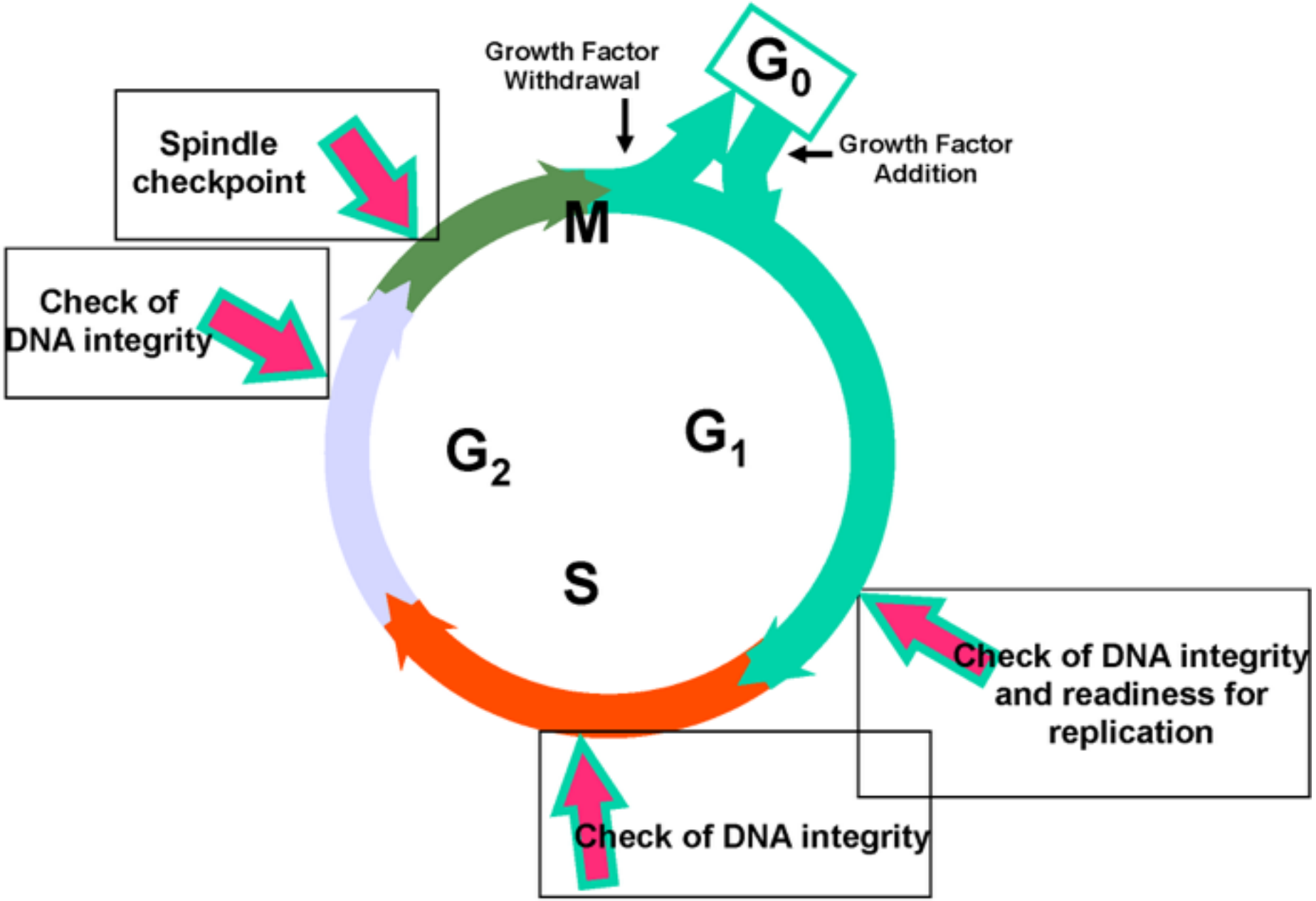
Phases of the cell cycle



Phases of the cell cycle



Cell cycle checkpoints



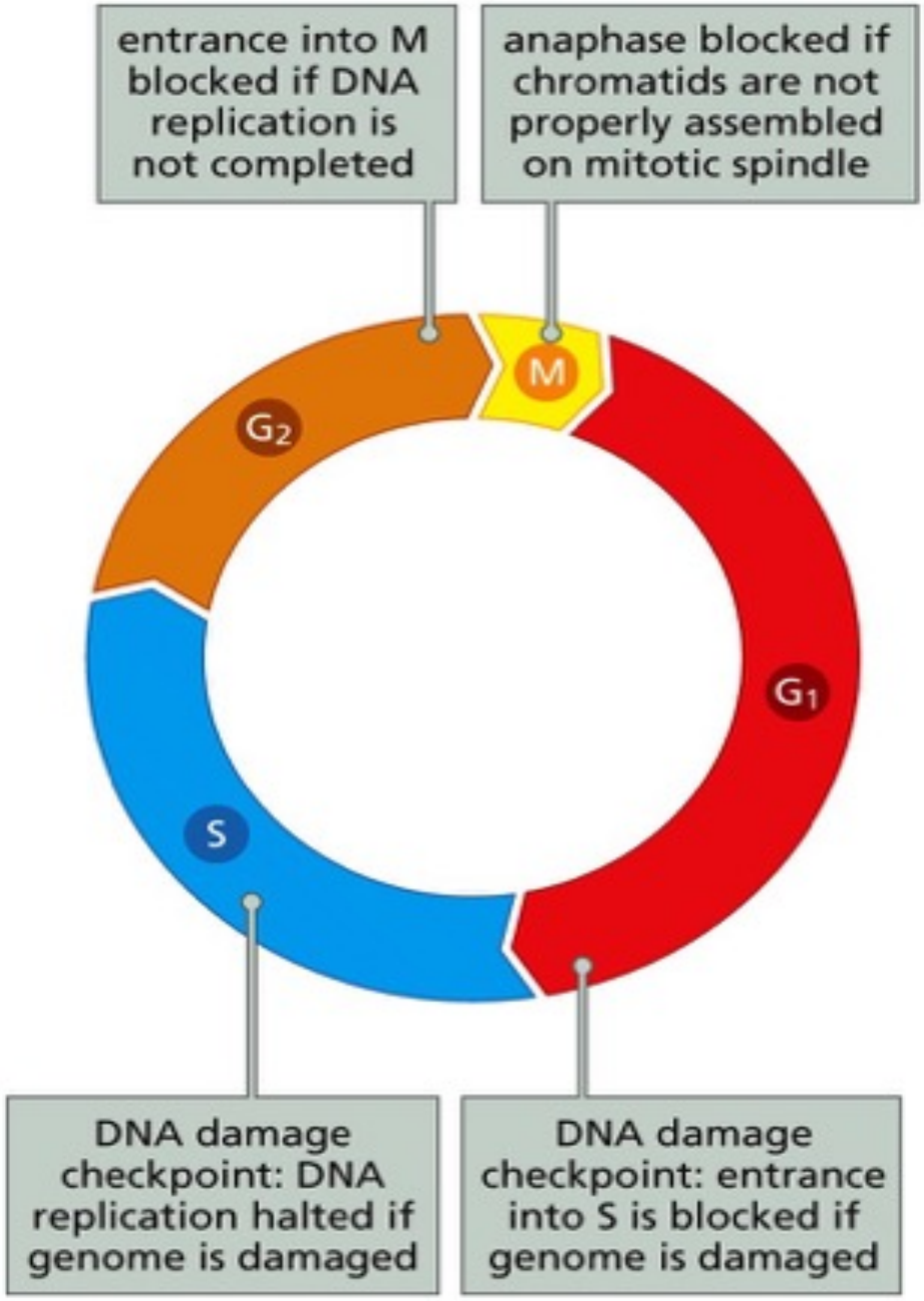
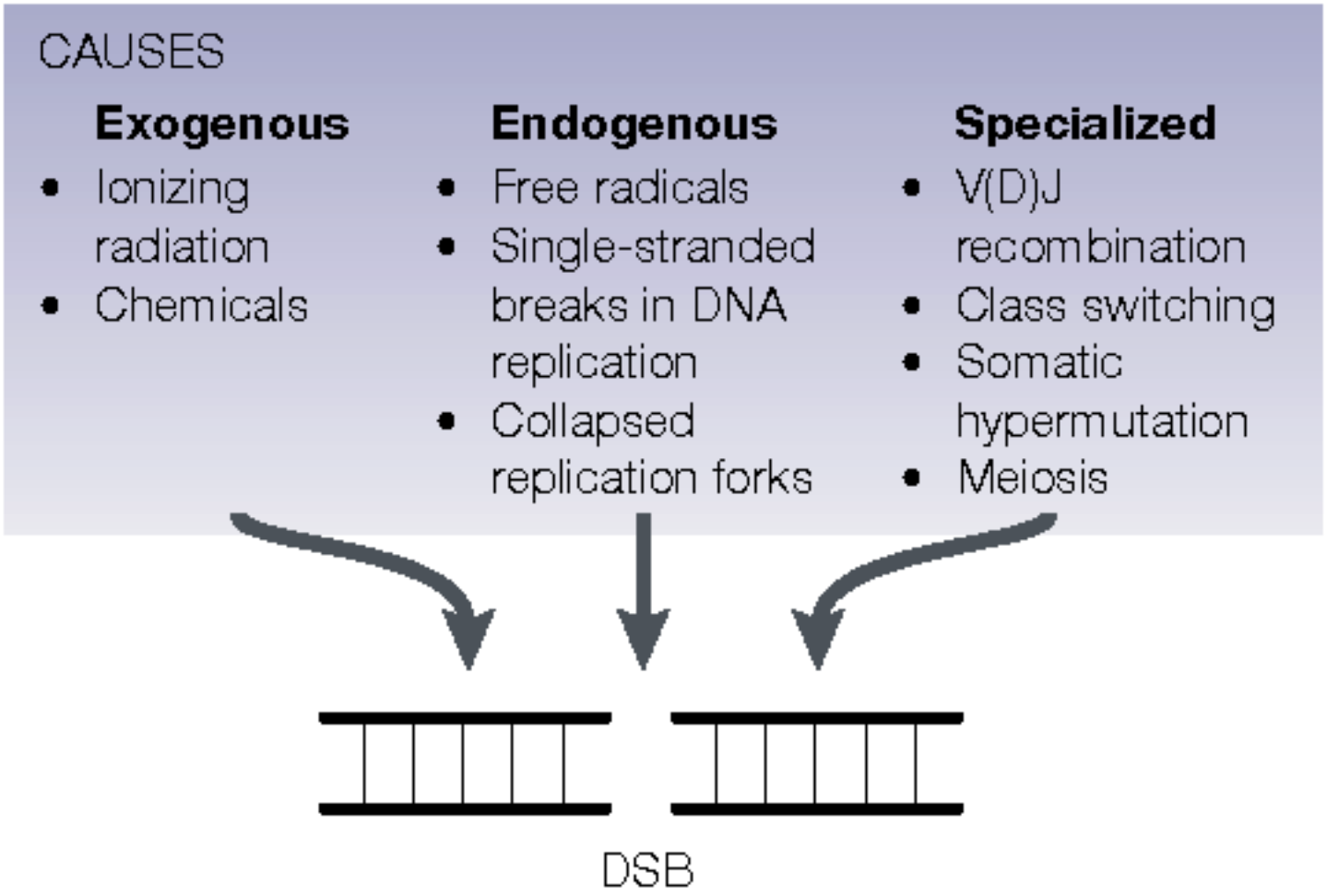


Figure 8.4 The Biology of Cancer (© Garland Science 2014)

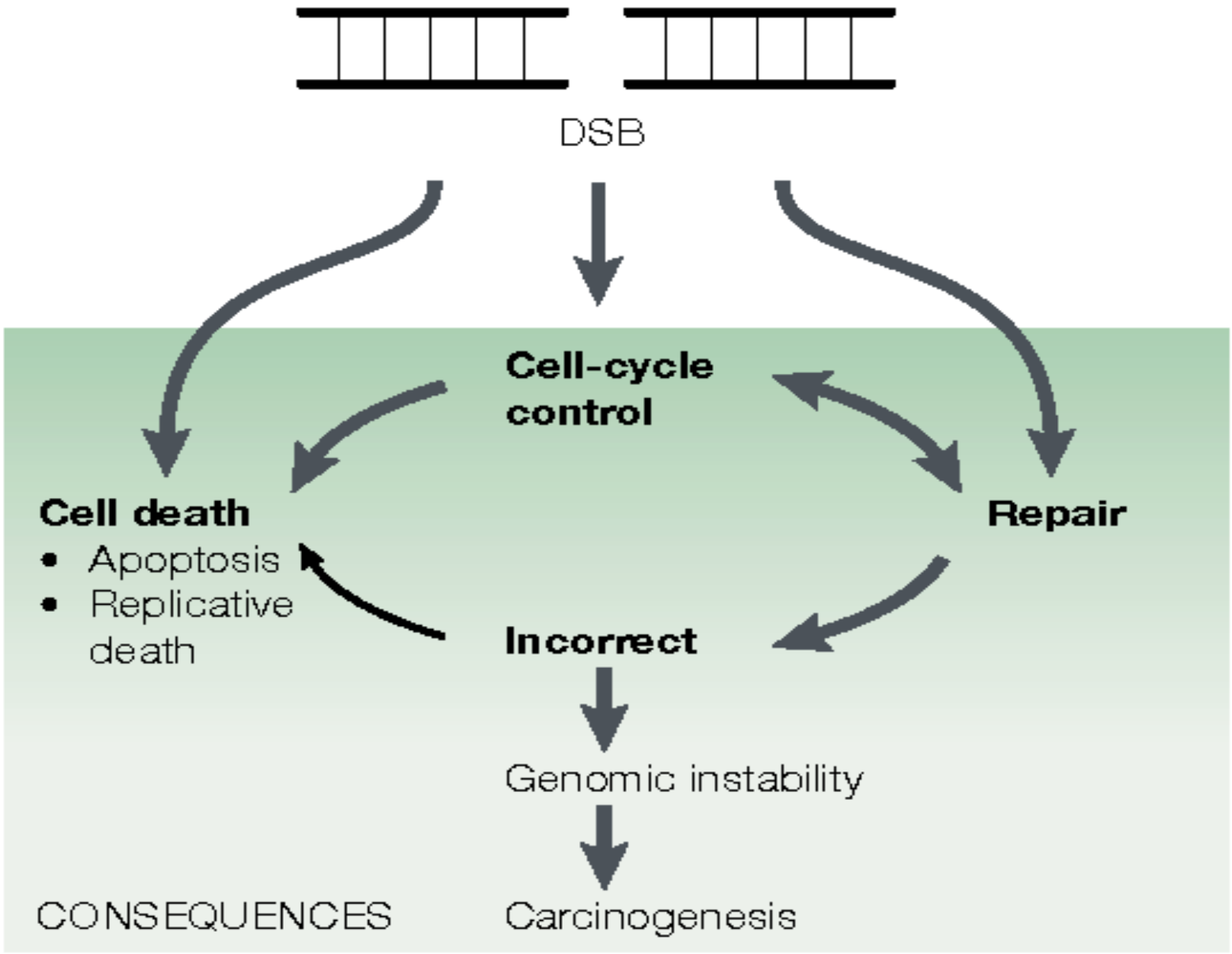
Cell cycle checkpoints

Causes of double strand breaks



Dik C. van Gent et al, 2001 Nature Reviews

Cellular response and consequence of DSBs



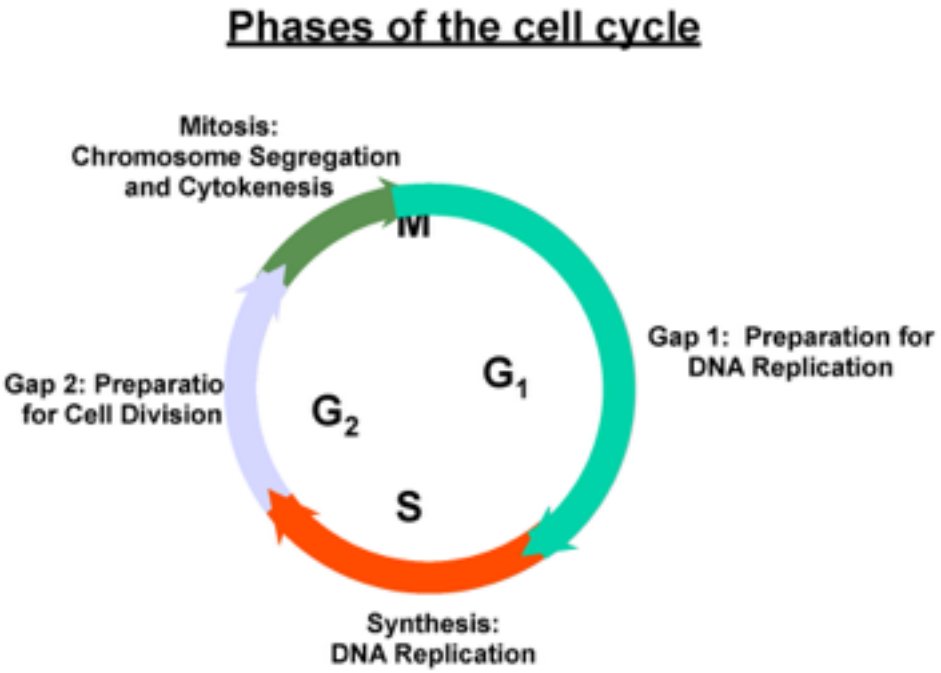
Radiation-induced chromosomal aberrations

Mitosis:

- -Prophase
- -Metaphase
- -Anaphase
- -Telophase

X-rays or ionizing radiation induces DSBs in the chromosomes. DSBs causes sticky ends, which can join with any other sticky ends.

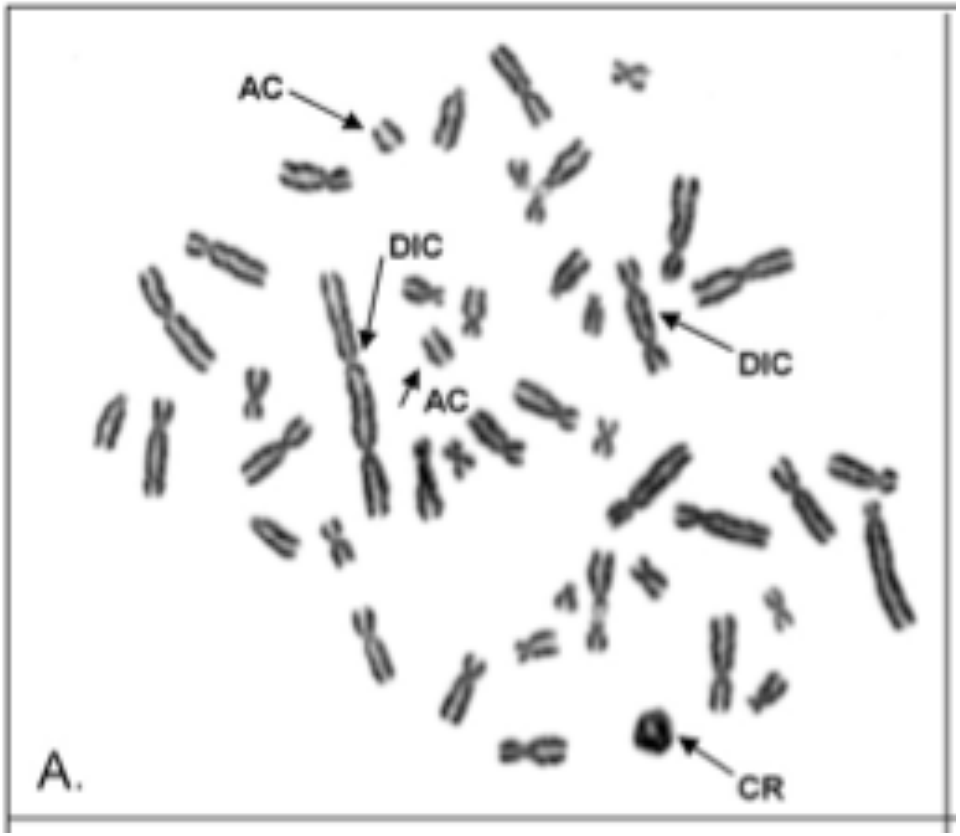
1) Rejoin to original configurations
2) The breaks fails to rejoin causing deletion
3) Broken ends may join other sticky ends



Example of radiation-induced chromosomal aberrations

DNA damage induced in different cell cycle stages leads to the formation of different types of chromosomal aberrations.

-For example: Damage during G0/G1 chromosomal aberrations.



G.I. Terzoudi et al., Mutation Research 711 (2011) 174–186

Cyclins regulate the cell cycle

- **Expression Oscillates During the Cell Cycle**

- **Main Cell Cycle Regulatory Cyclins:**
  cyclin D1-3, cyclin E, cyclin A and cyclin B

Cyclin expression during cell cycle

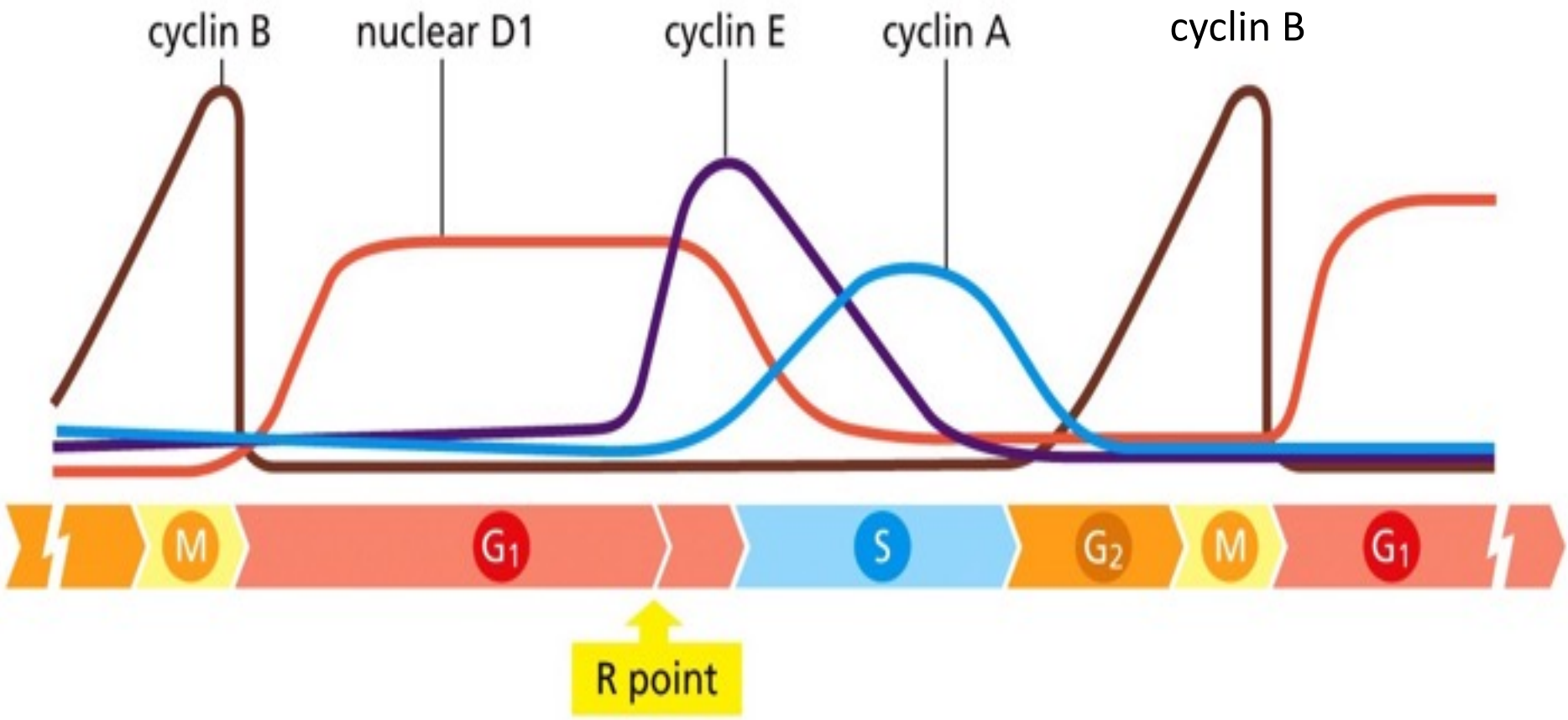


Figure 8.10 The Biology of Cancer (© Garland Science 2014)

Cell cycle-dependent fluctuations in cyclin B levels

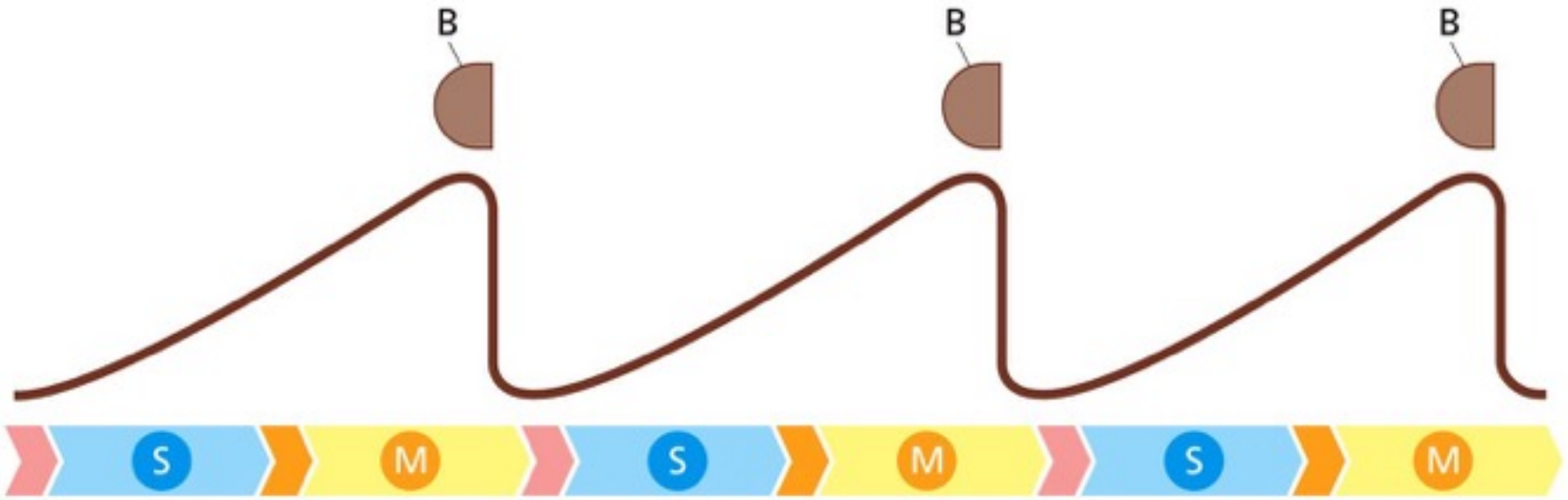
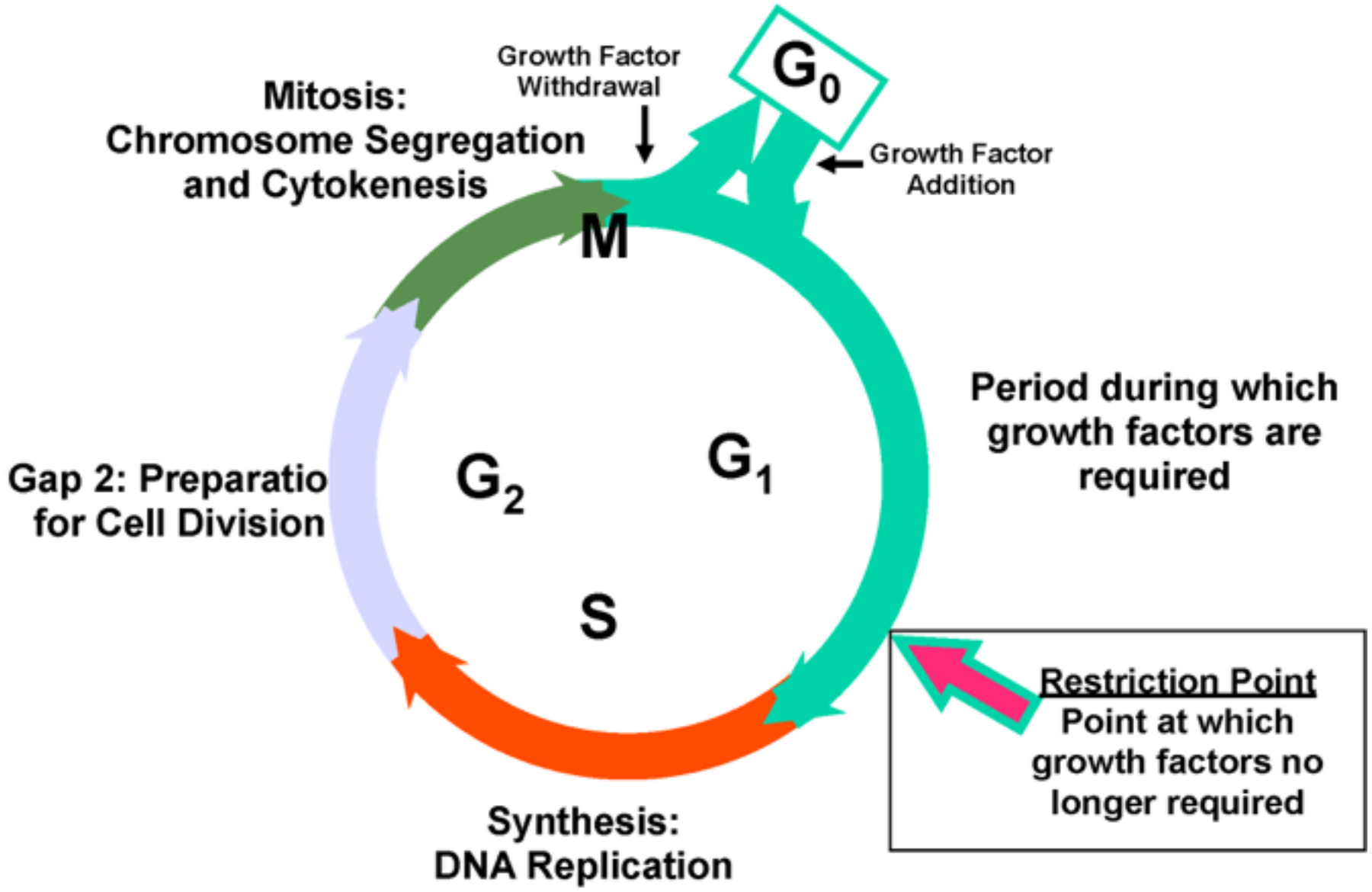


Figure 8.9 The Biology of Cancer (© Garland Science 2014)

Phases of the cell cycle



Cyclins Regulate Cyclin-Dependent Kinases

Cyclin-dependent kinases (cdks)

- Multigene Family [cdk4, cdk6, cdk2, cdk1 (cdc2)]
- Serine/Threonine Kinases
- Require Cyclin Binding for Activity
- Regulated by Phosphorylation
- Regulated by Cyclin-Dependent Kinase Inhibitors (ckis)

Cyclins

- Regulatory Subunits of Cyclin/cdk Complexes
- Main Cell Cycle Regulatory Cyclins:
  cyclin D1-3, cyclin E, cyclin A and cyclin B
- Cyclin/cdk Associations:
  cyclin D/cdk4, cyclin D/cdk6, cyclin E/cdk2,
  cyclin A/cdk2, cyclin A/cdk1 and cyclin B/cdk1
- Expression Oscillates During the Cell Cycle

Pairing of cyclins with cyclin-dependent kinases

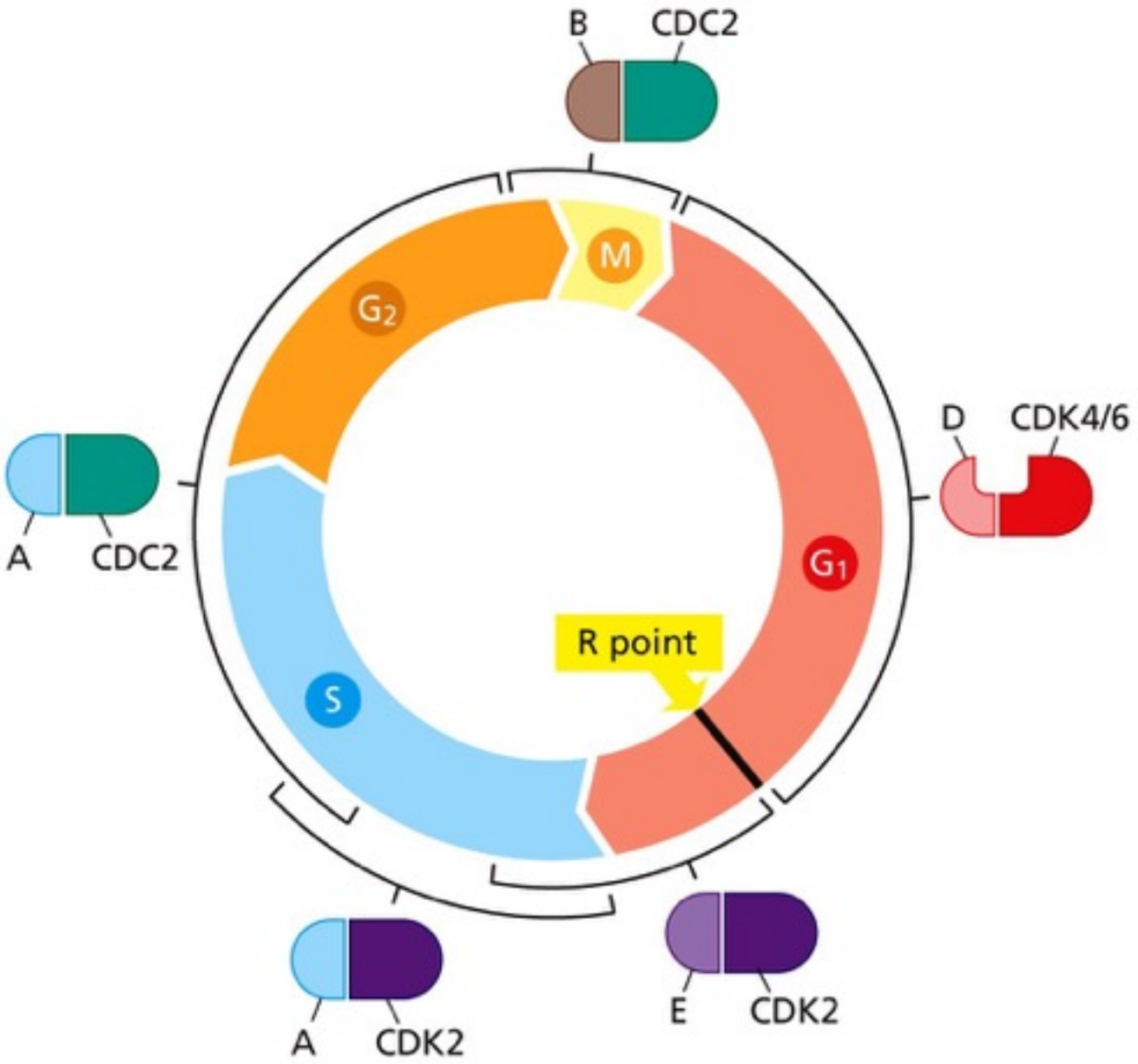


Figure 8.8 The Biology of Cancer (© Garland Science 2014)

Control of cyclin levels during the cell cycle

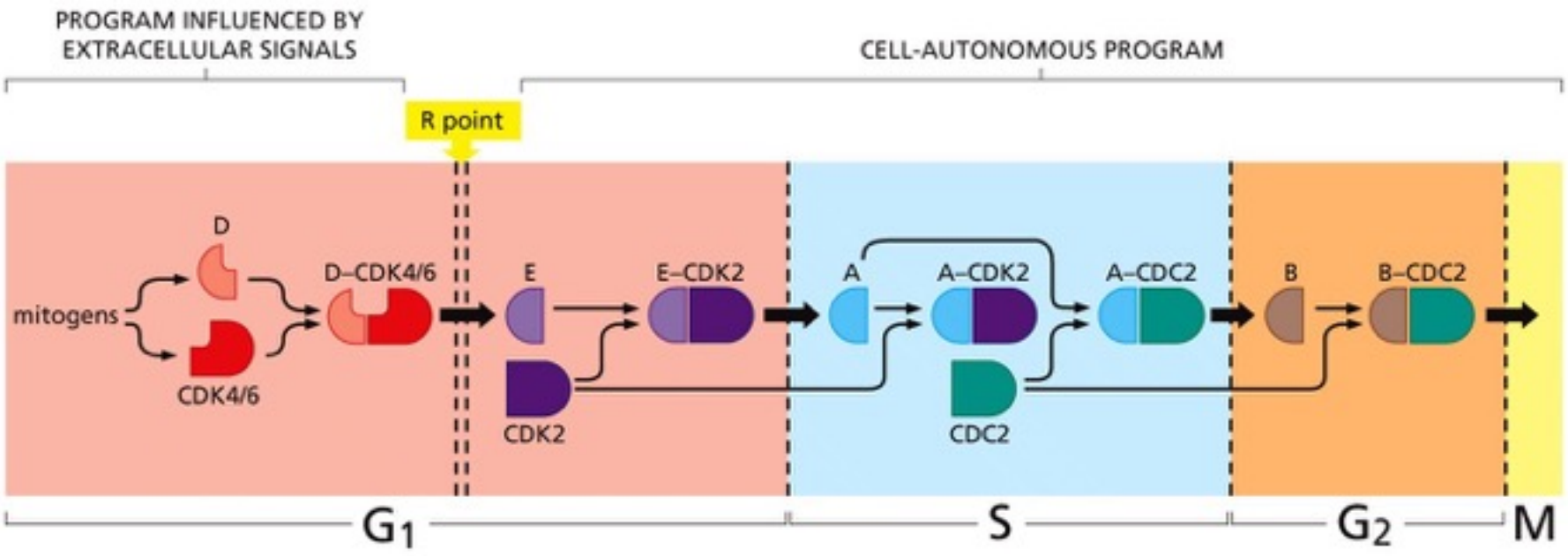


Figure 8.12 The Biology of Cancer (© Garland Science 2014)

Cyclin Dependent-Kinase Inhibitors (ckis)

INK Family

- $p15^{INK4b}$, $p16^{INK4a}$, $p18^{INK4c}$, $p19^{INK4d}$
- Bind cyclin D-associating cdks (cdk4 and cdk6)
- Block interaction of cdks with cyclins

CIP/KIP Family

- $p21^{WAF1/CIP1}$, $p27^{KIP1}$, $p57^{KIP2}$
- Inhibit Cyclin A/cdk2 and Cyclin E/cdk2
- Required for assembly of Cyclin D/cdk complexes
- Do not inhibit cyclin D/cdk complexes at physiological levels

Interaction of cdki's with cdks

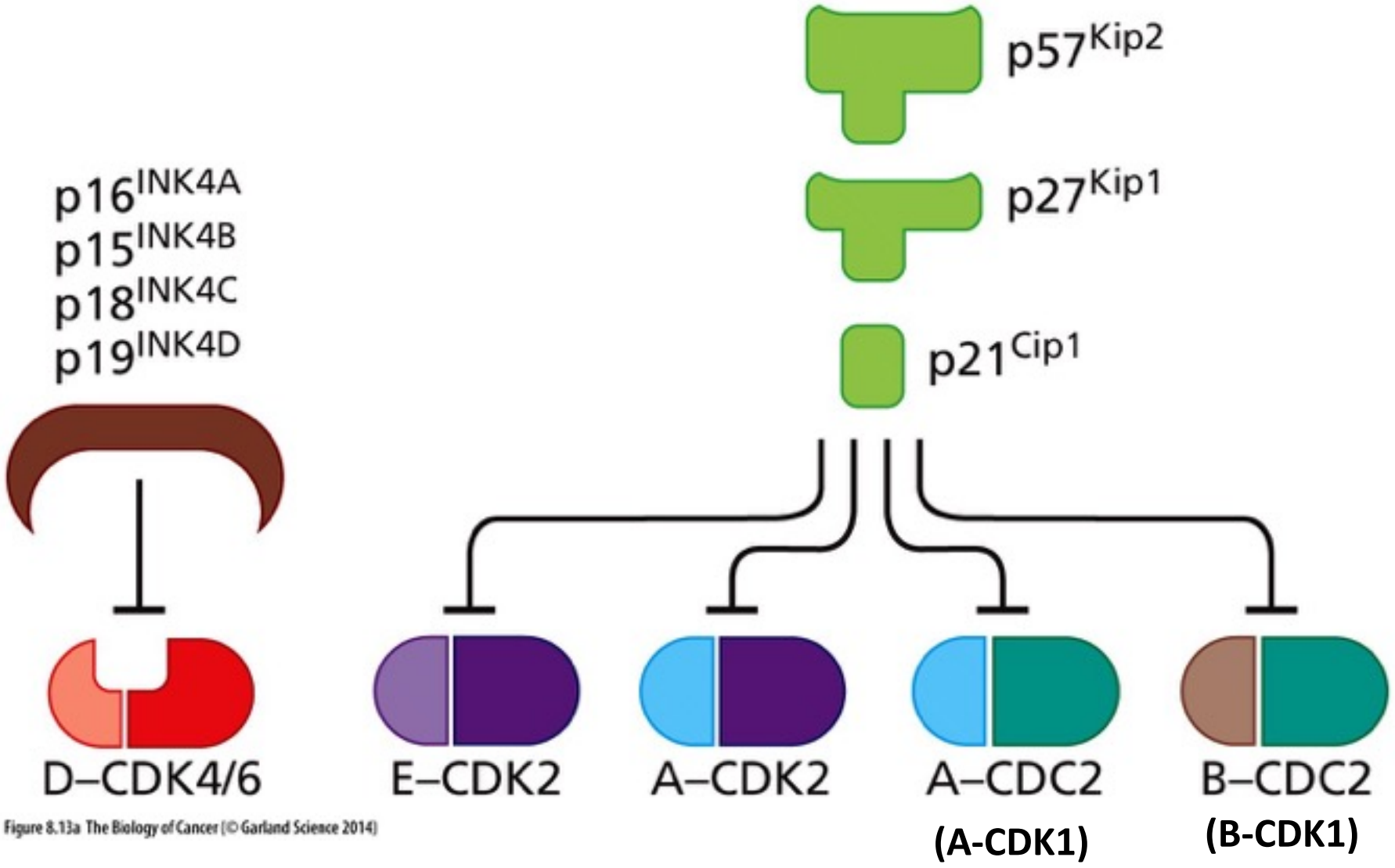


Figure 8.13a The Biology of Cancer (© Garland Science 2014)

Cip/Kip cdki's stimulate cyclin D/cdk association and activity

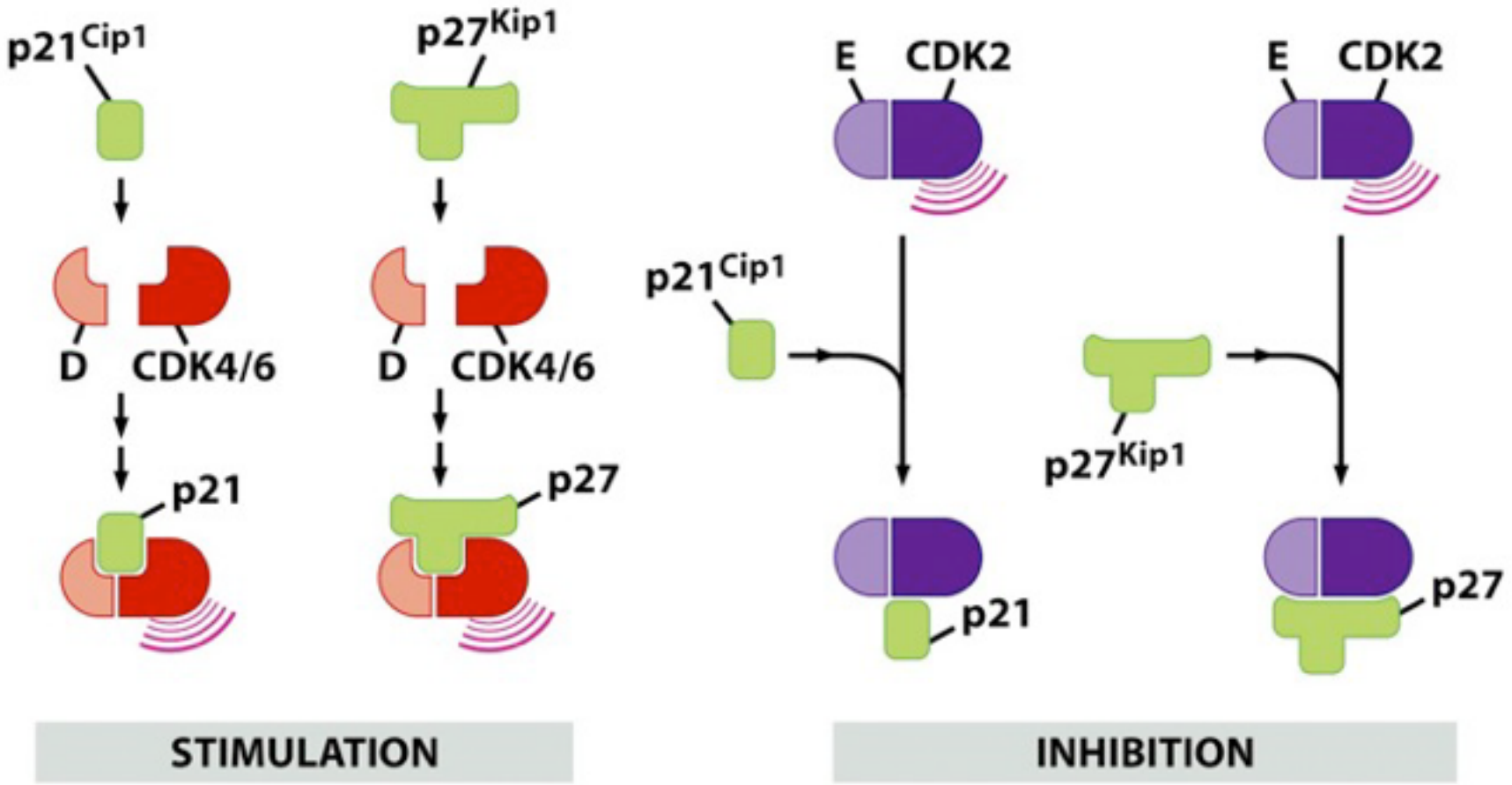


Figure 8.17a *The Biology of Cancer* (© Garland Science 2007)

Cyclin D/cdk complexes sequester cip/kip proteins form cyclin E/cdk2

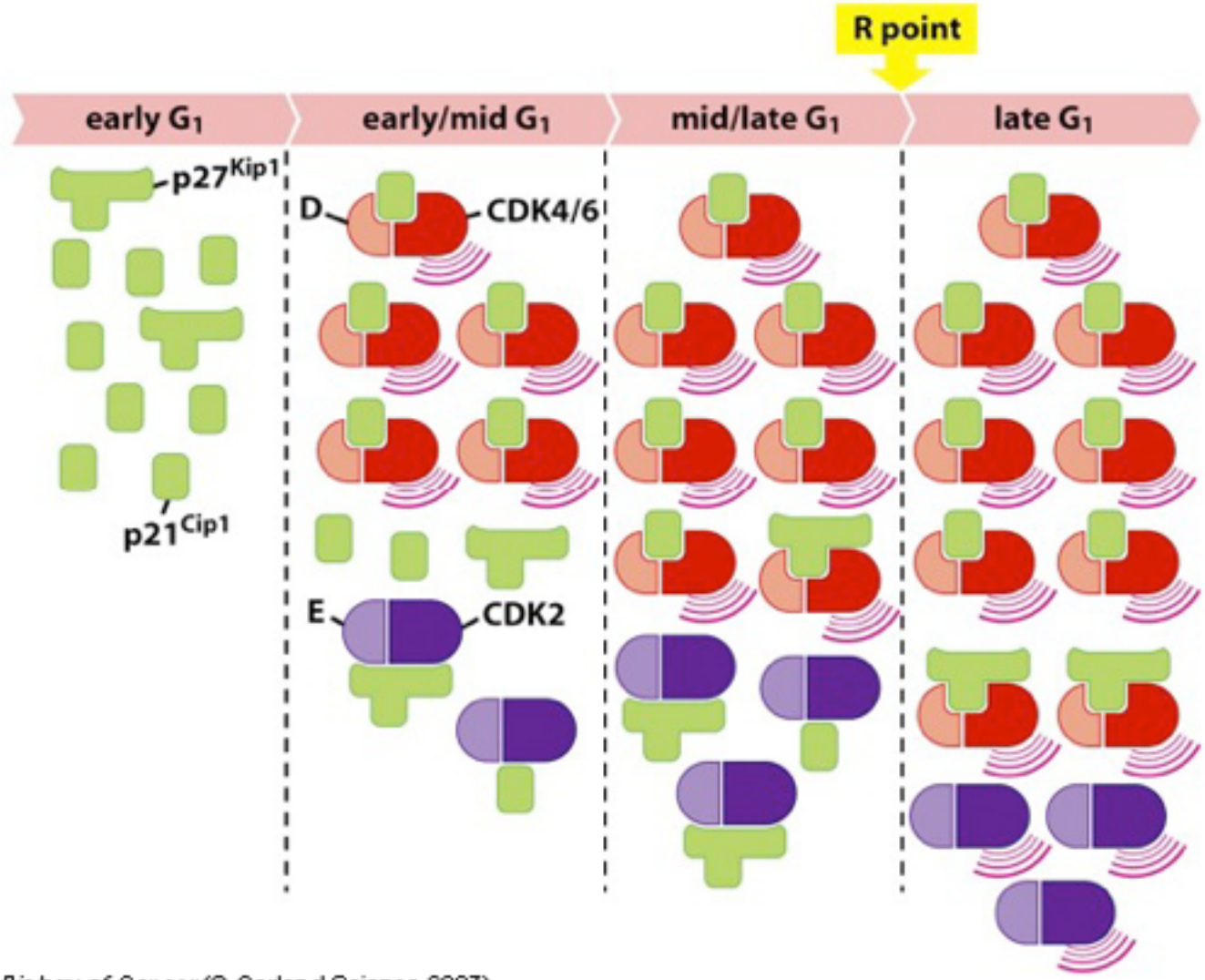
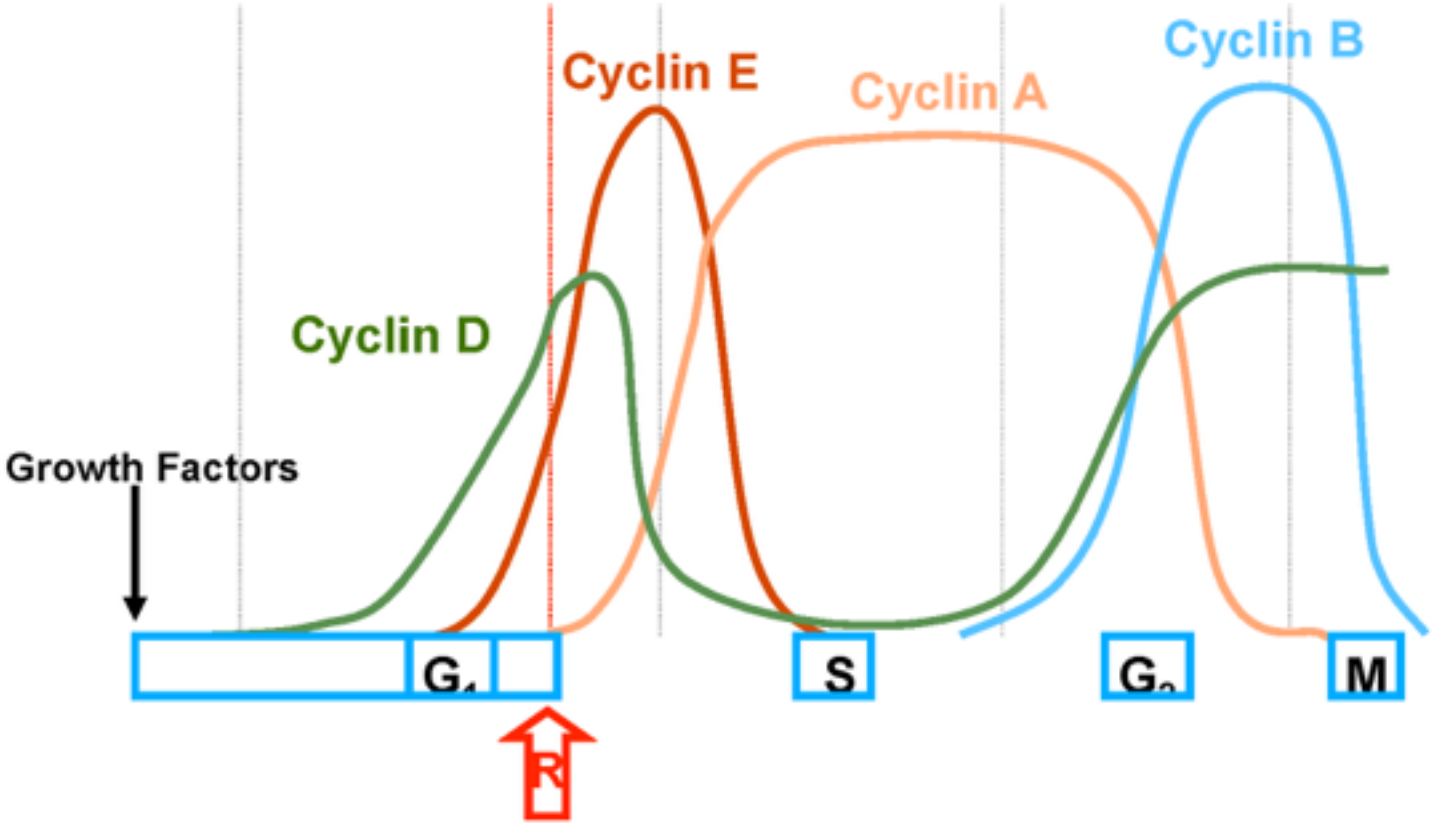


Figure 8.17b *The Biology of Cancer* (© Garland Science 2007)

Cyclin Expression During Cell Cycle



Cip/Kip cdki's stimulate cyclin D/cdk association and activity

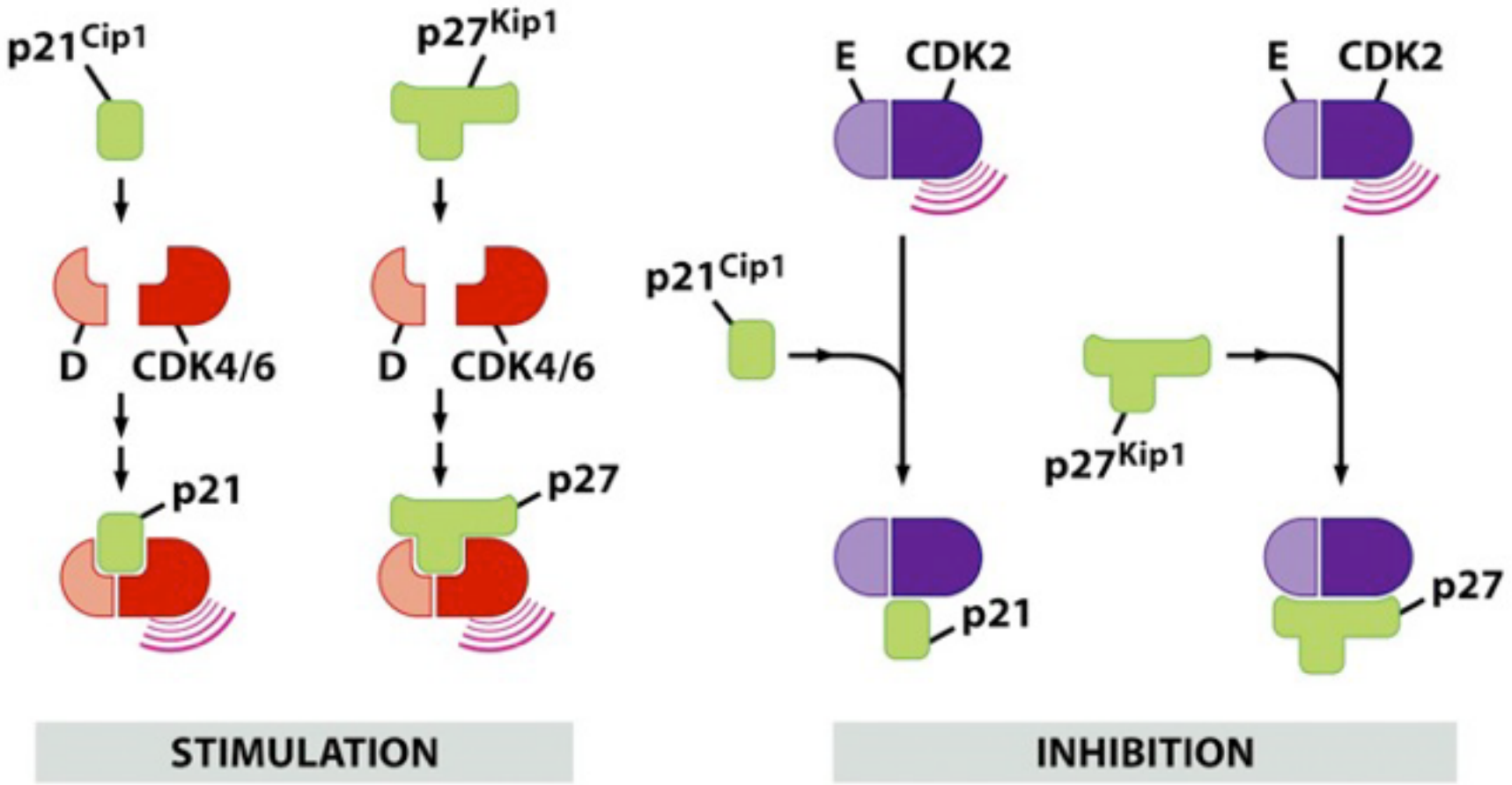


Figure 8.17a *The Biology of Cancer* (© Garland Science 2007)

Interaction of cdki's with cdks

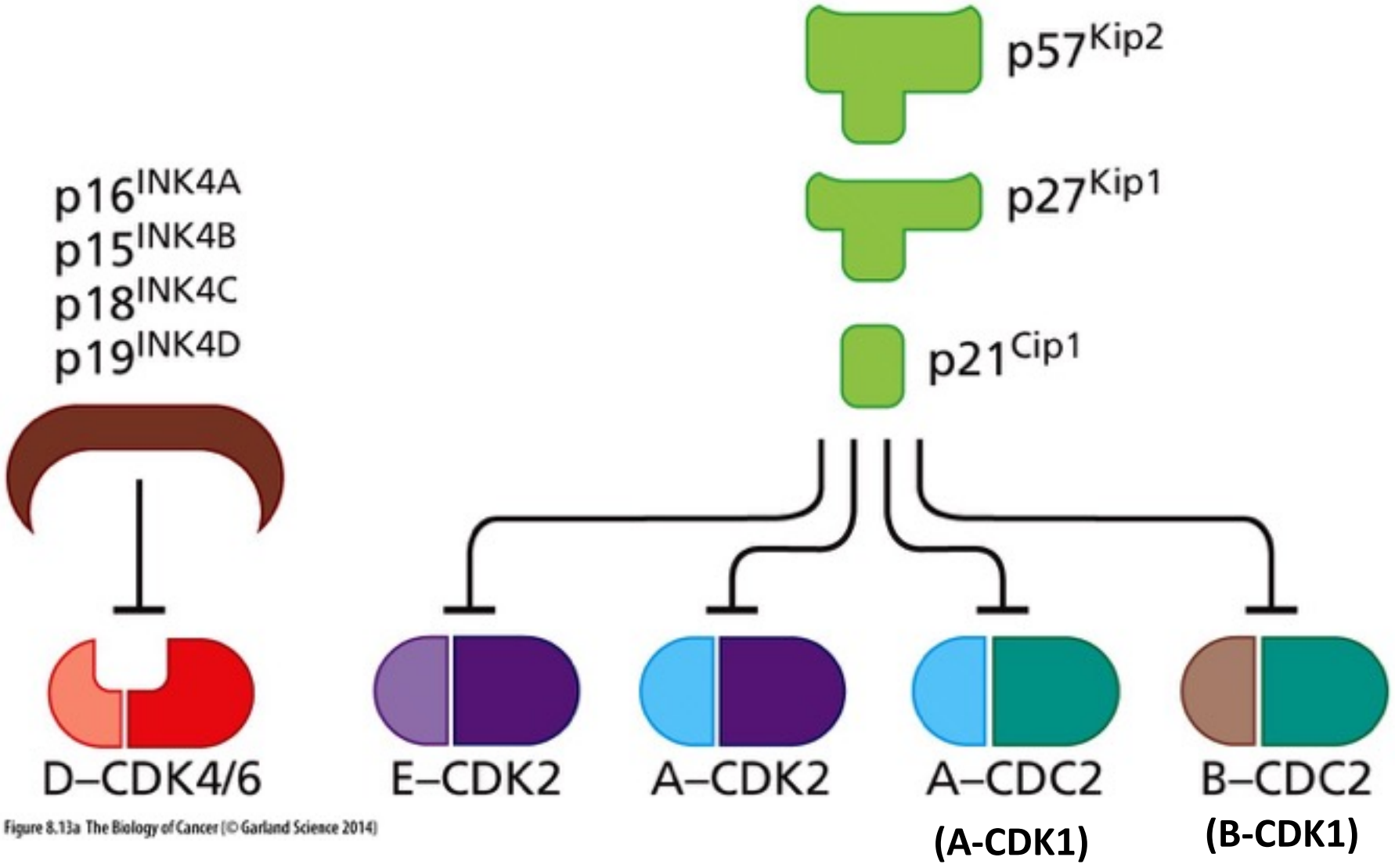


Figure 8.13a The Biology of Cancer (© Garland Science 2014)

Control of cyclin level progression by TGF-beta

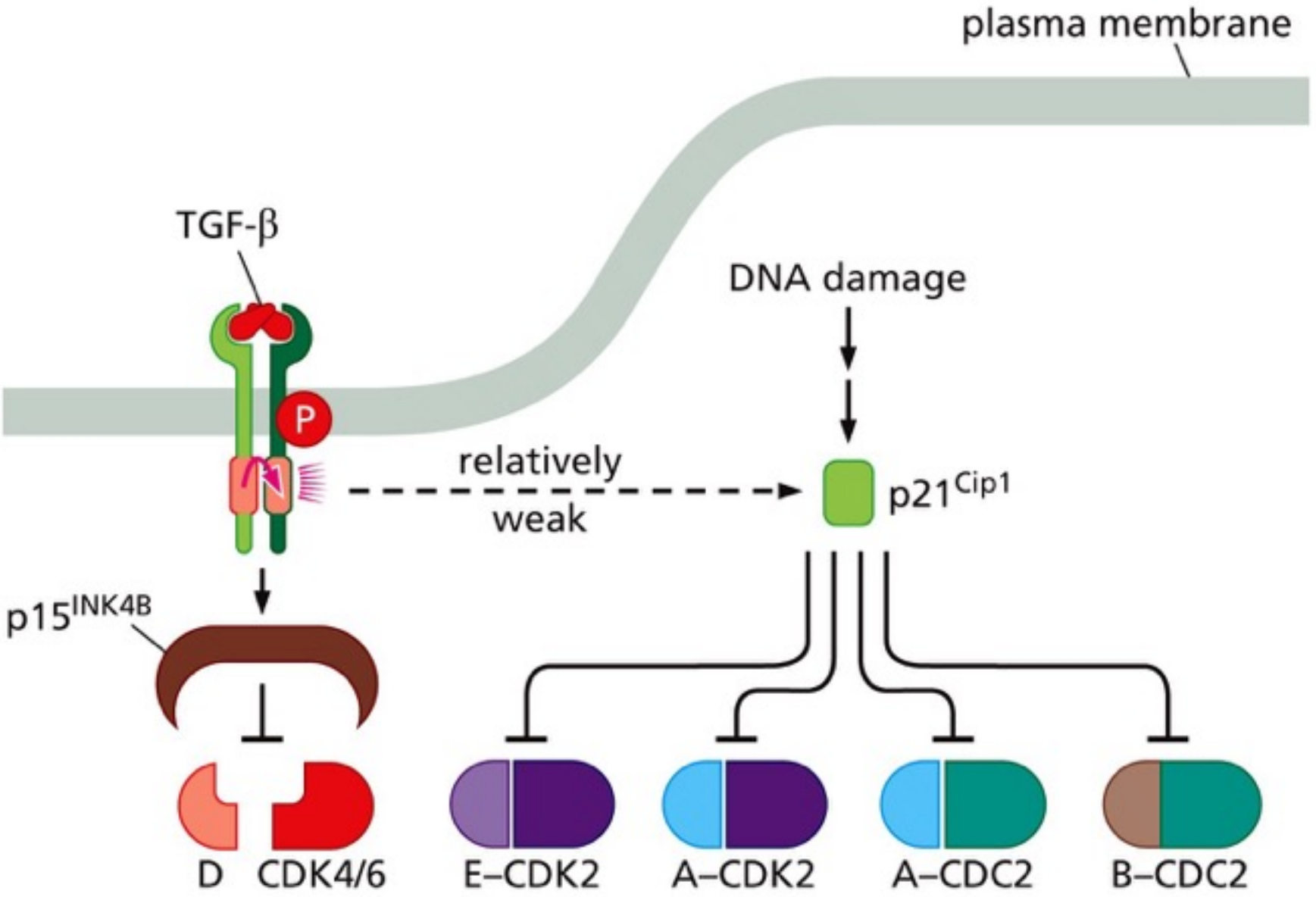


Figure 8.14a The Biology of Cancer (© Garland Science 2014)

Control of cyclin advance by extracellular signals

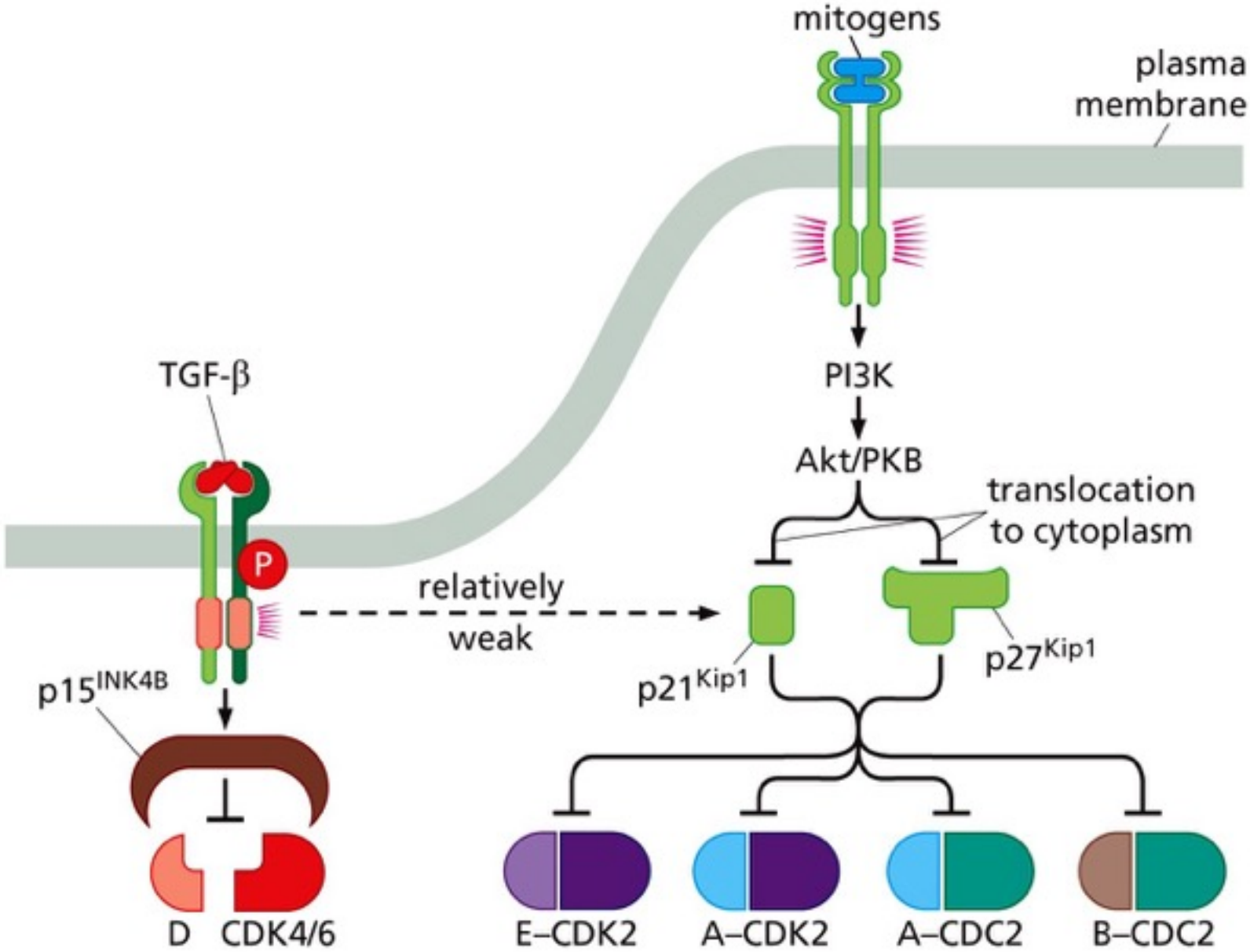
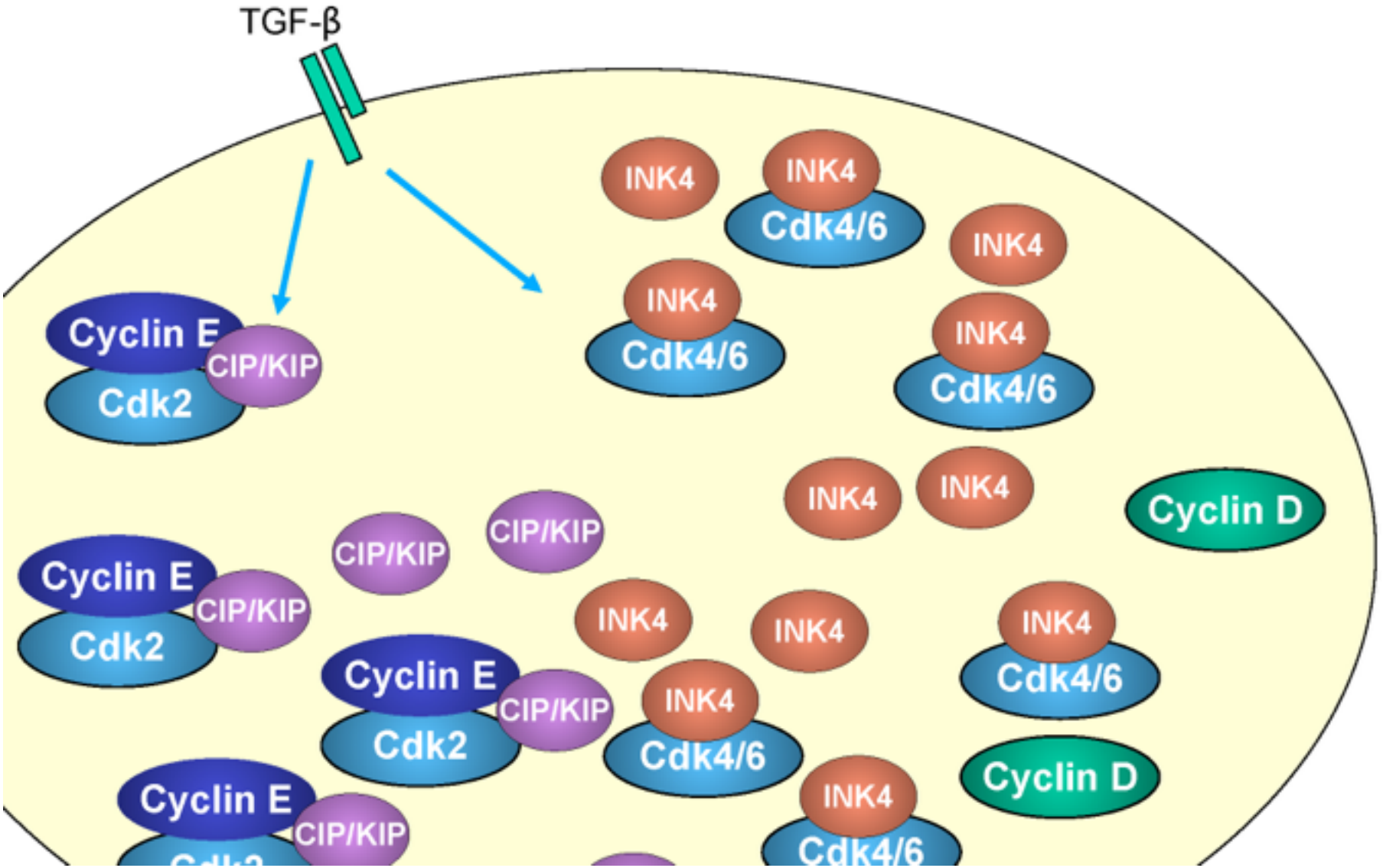
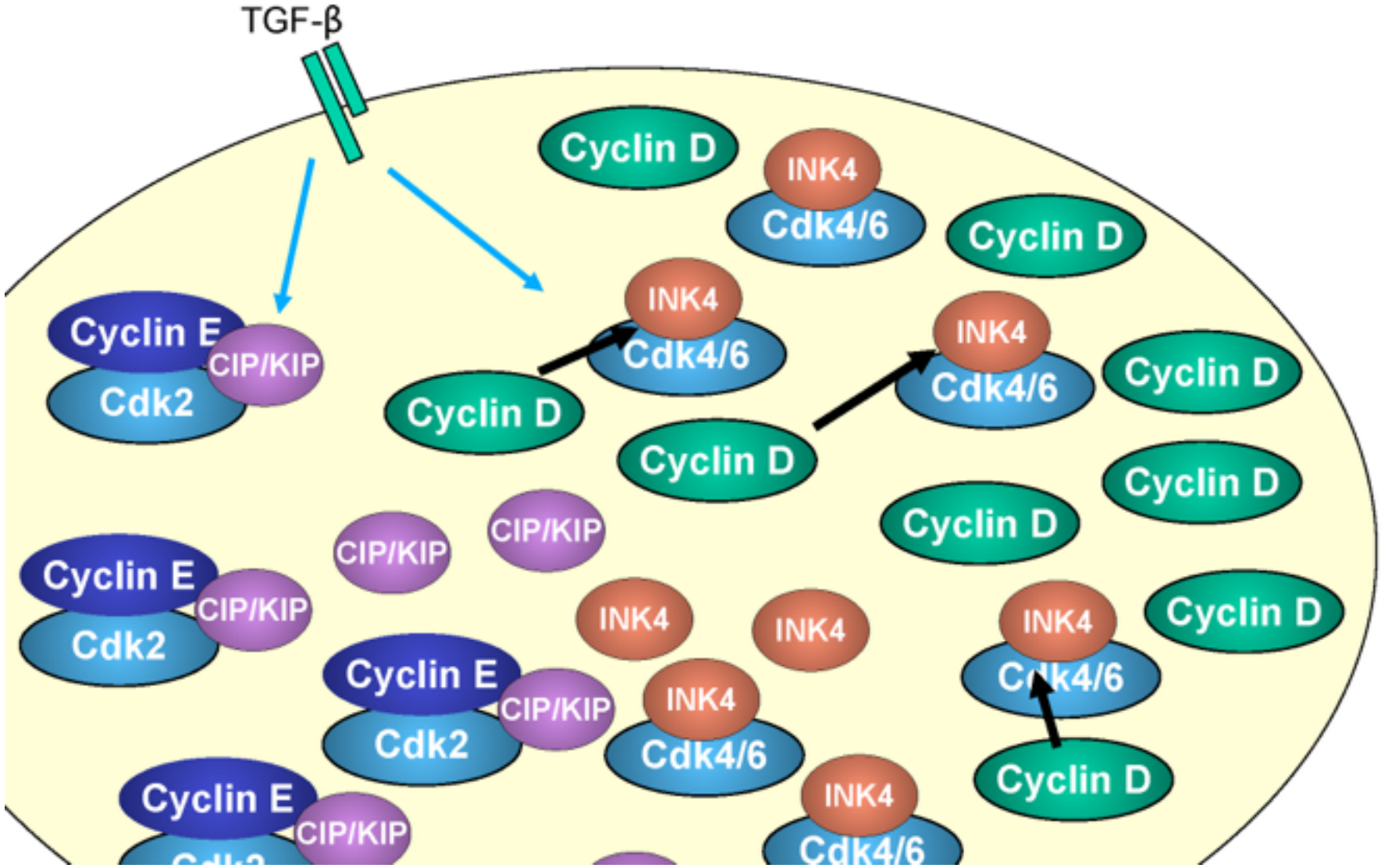


Figure 8.15a The Biology of Cancer (© Garland Science 2014)

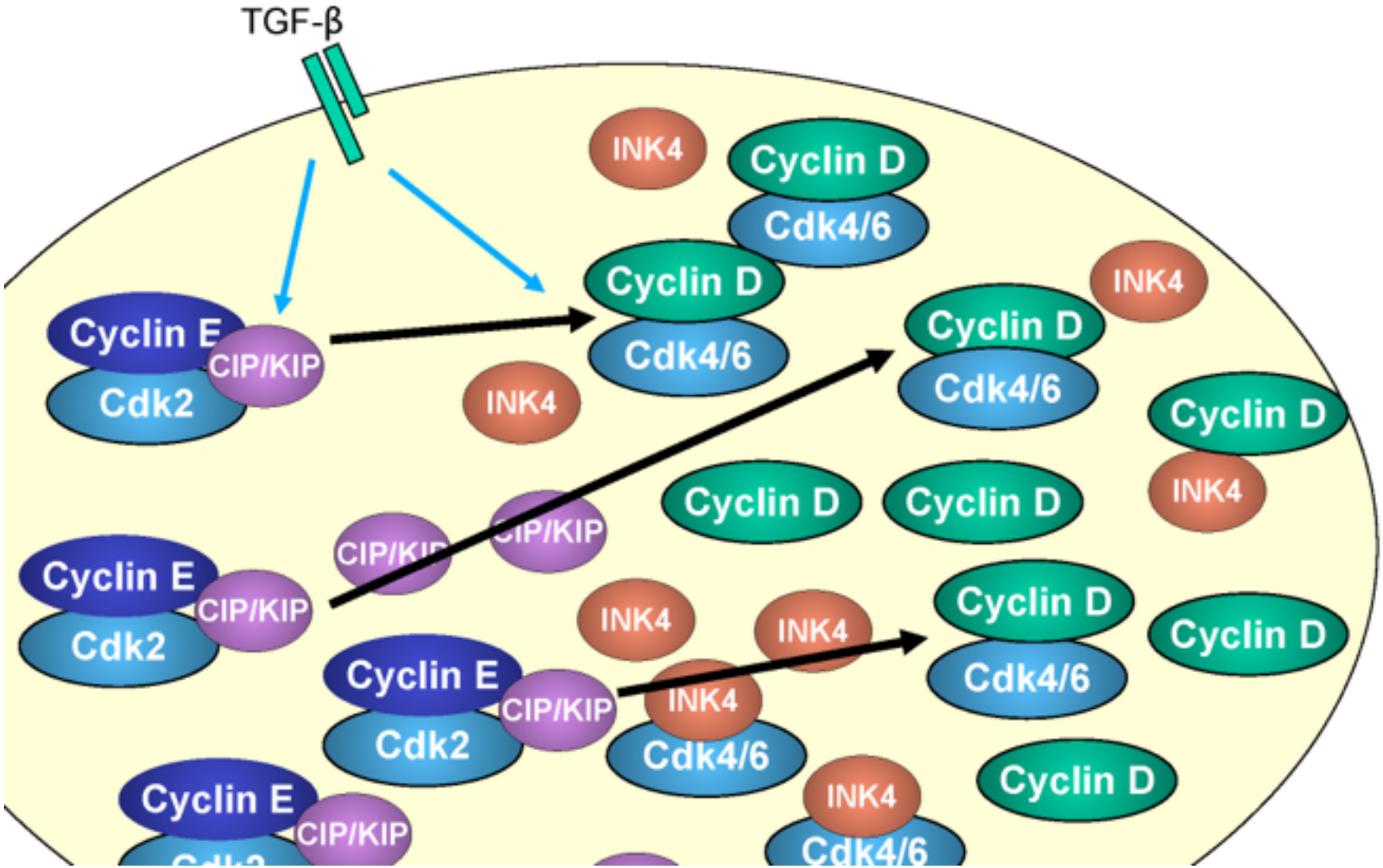
Cyclin D1 and TGF-β



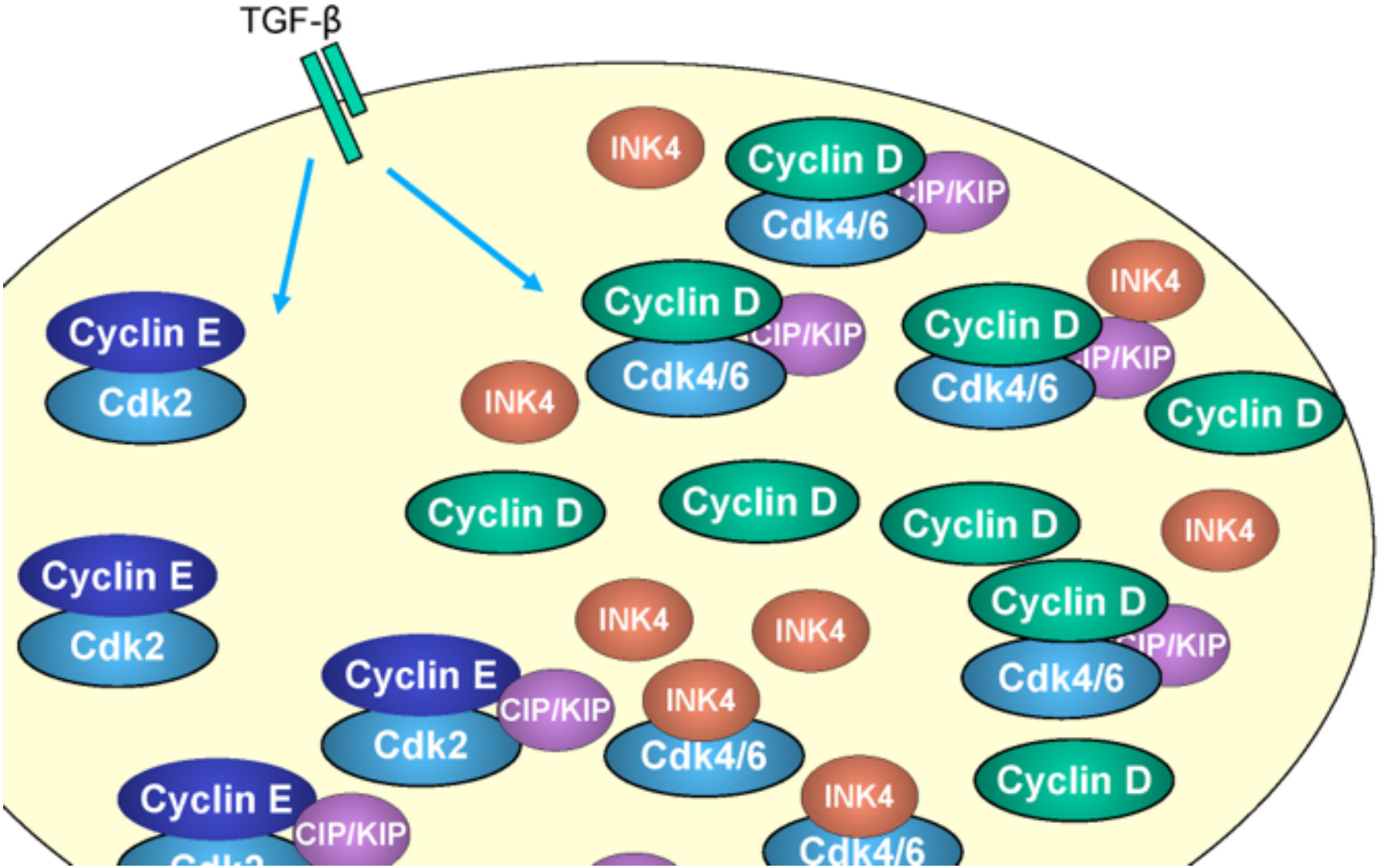
Cyclin D1 and TGF-β



Cyclin D1 and TGF-β



Cyclin D1 and TGF-β



How do cyclin/cdk complexes regulate the cell cycle?

The E2F-pRB pathway

- Disrupted in most if not all cancers
- A major regulator of cell cycle progression
- Involved in regulation of apoptosis

E2F-pRB pathway

Major Players:

E2F Family of Transcription Factors

Retinoblastoma Family of Proteins (Pocket Proteins)

Cyclins and Cyclin Dependent Kinases (cdks)

Cyclin Dependent Kinase Inhibitors (ckis)

E2F-Regulated Genes

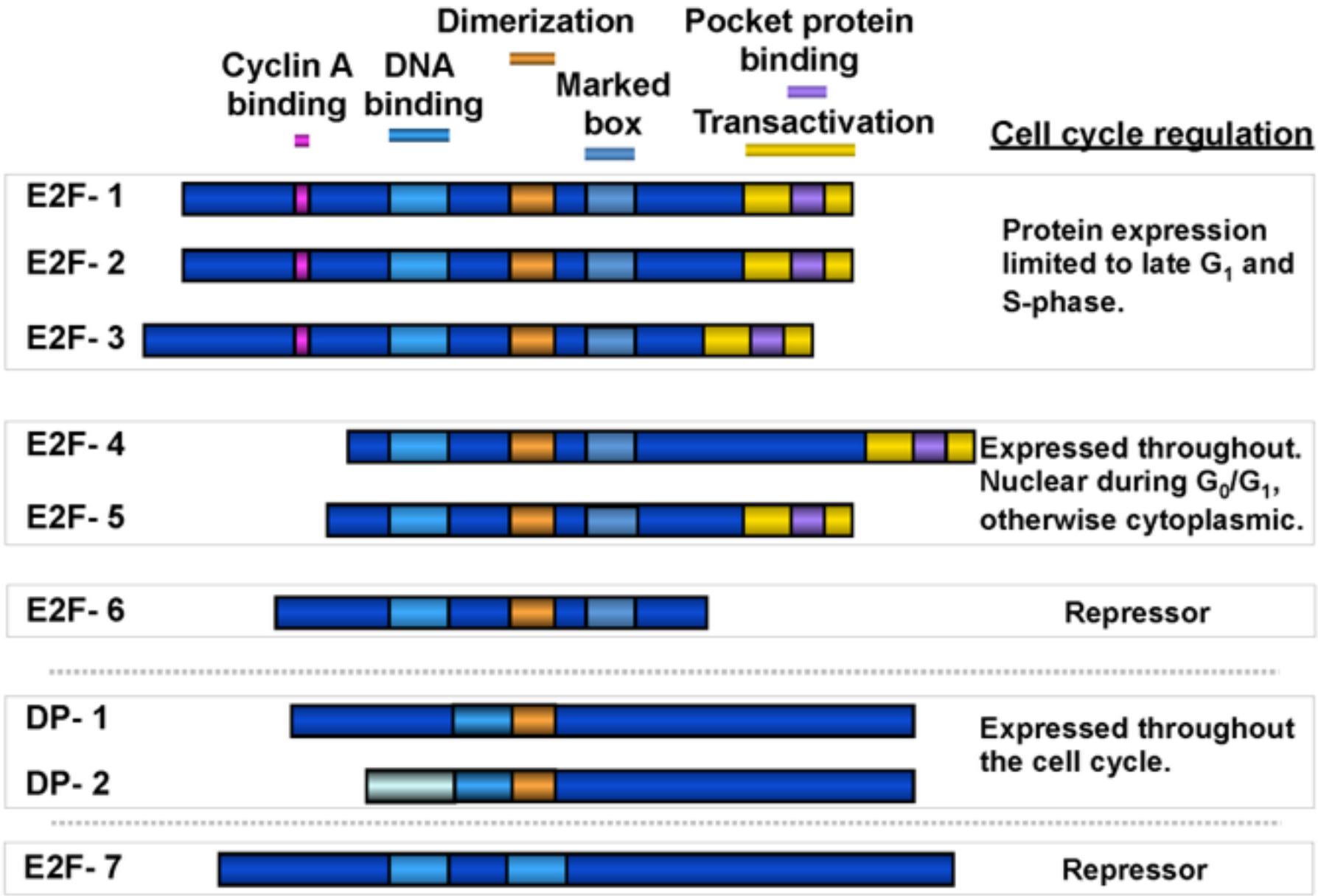
DNA Synthesis Genes

dihydrofolate reductase (DHFR)
DNA polymerase α
thymidylate synthase (TS)
thymidine kinase (TK)
proliferating cell nuclear antigen (PCNA)
licensing factors (e.g. cdc6, mcm's)

Growth Regulatory Genes

c-myc
B-myb
pRB
p107
cyclin E
cyclin A
E2F1
E2F2
cdk1
$p21^{WAF1/CIP1}$
$p27^{KIP1}$
$p19^{ARF}$

The E2F Family of Transcription Factors



Pocket Proteins

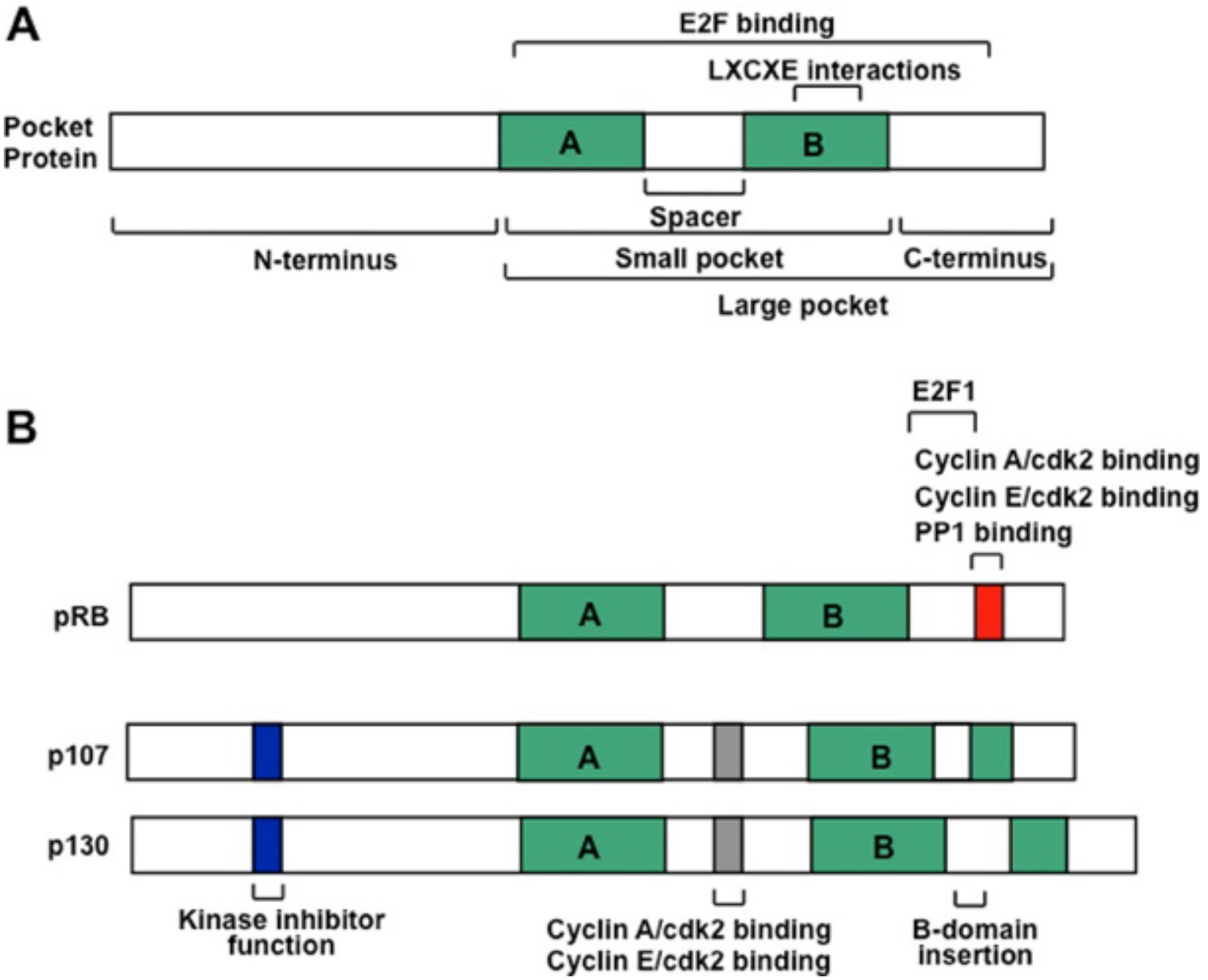
- Founding member is the Retinoblastoma Gene Product (pRB)
  mutated in familial retinoblastoma

- Other members are p107 and p130, named for molecular weight

- Bind to transactivation domain of E2F's with dual effect
  a) Blocks transactivation function
  b) Recruitment of histone deacetylase 1 (HDAC1),
  causing general repression of promoters

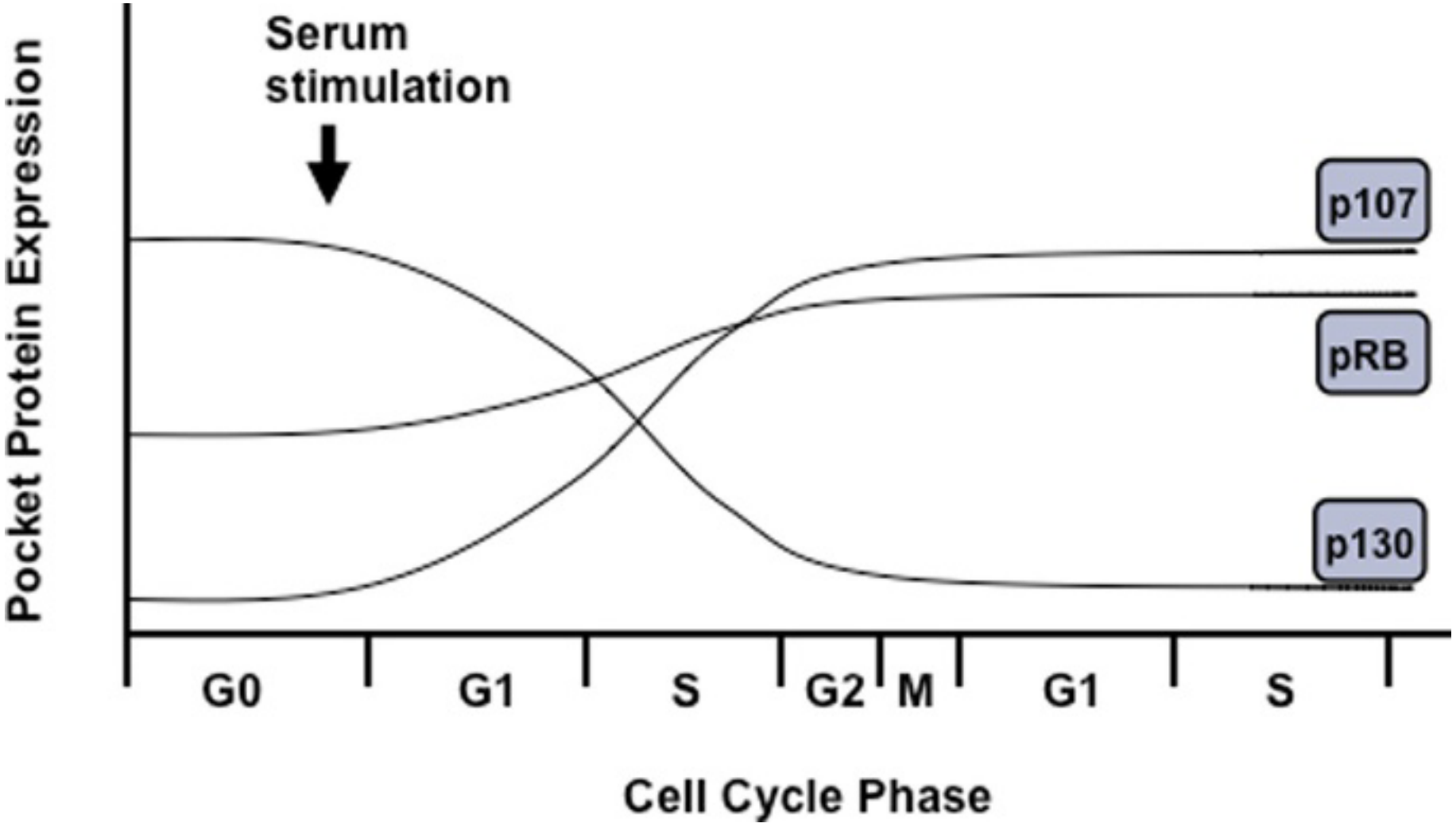
- Regulated by phosphorylation

  a) Hypophosphorylated forms bind E2F's and HDAC1
  b) Hyperphosphorylated forms do not bind E2F's or HDAC1
  c) Phosphorylated by cyclin/cdk complexes



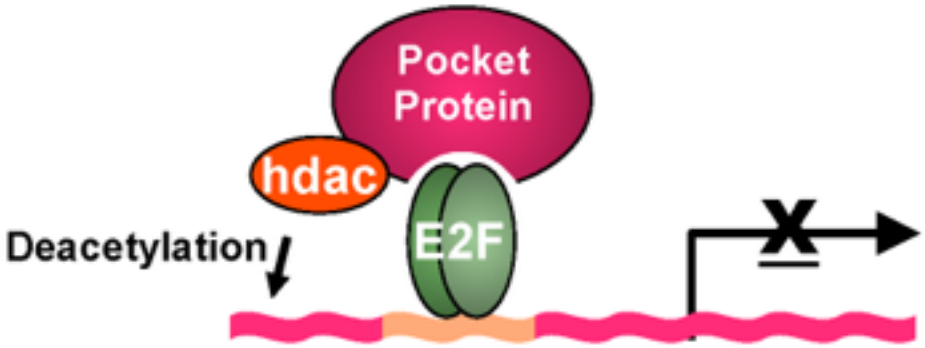
Henley and Dick, Cell Division 2012

Expression levels of pocket proteins throughout the cell cycle

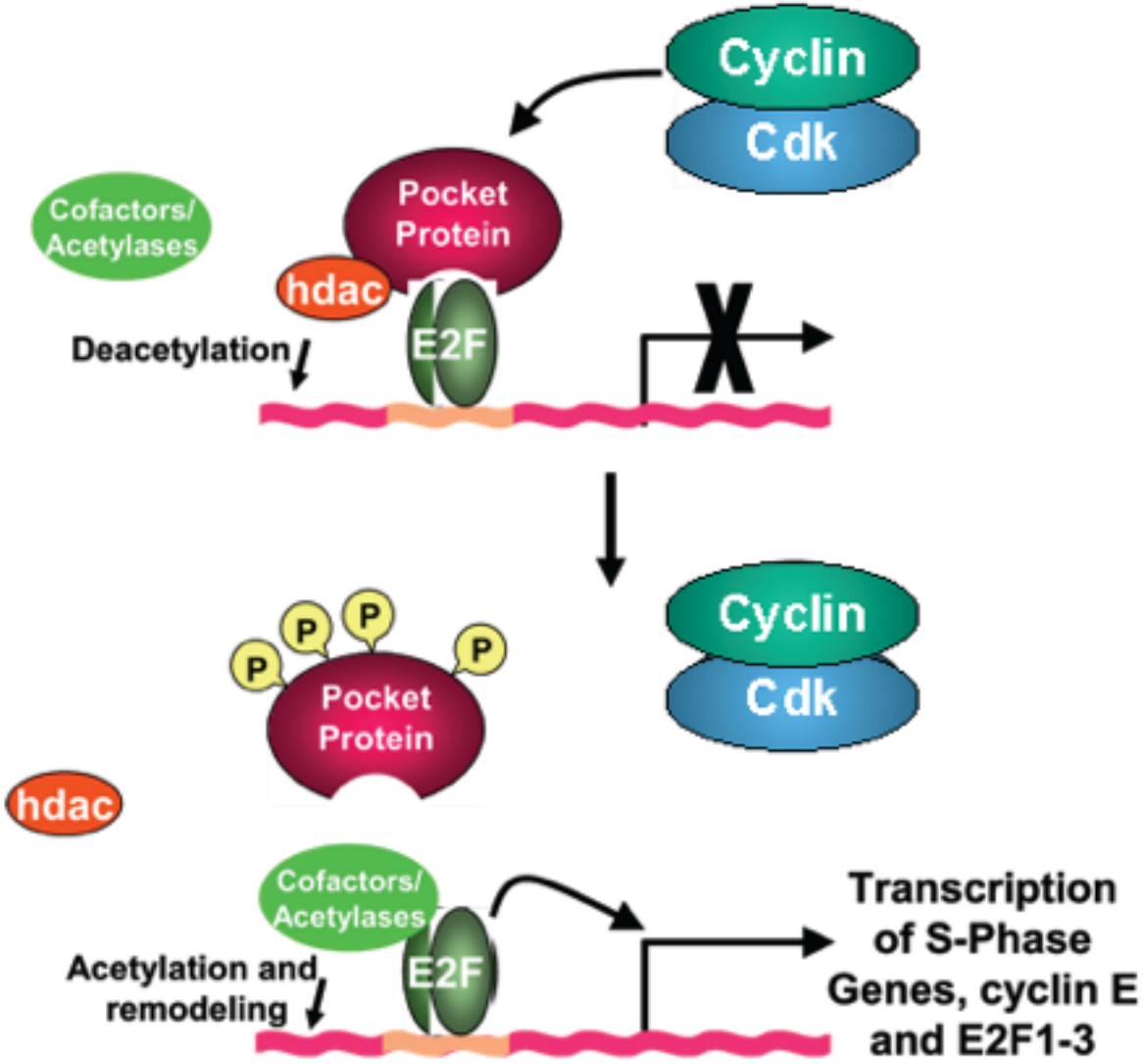


Henley and Dick, Cell Division 2012

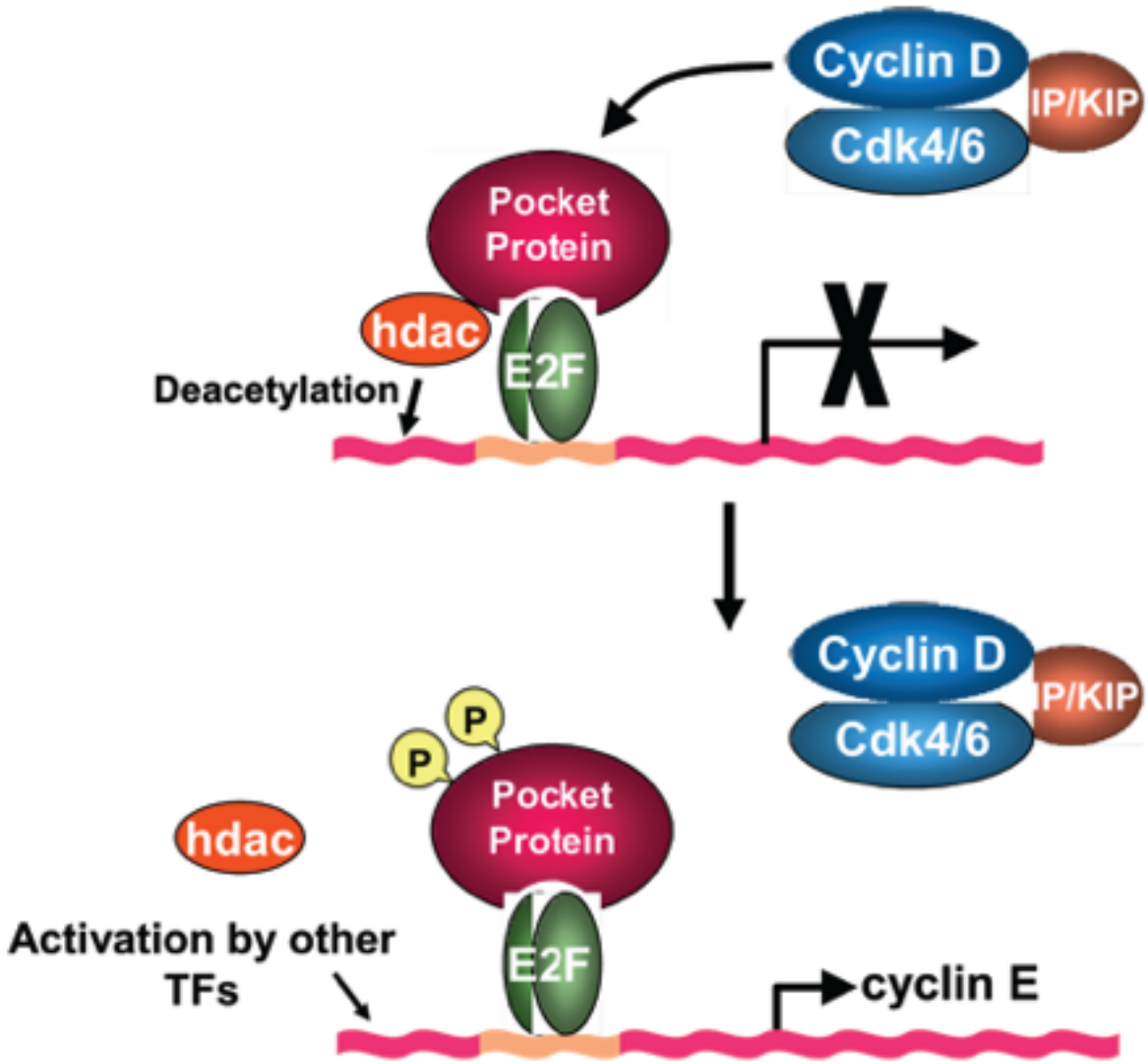
Pocket proteins bind E2F on DNA and actively repress transcription



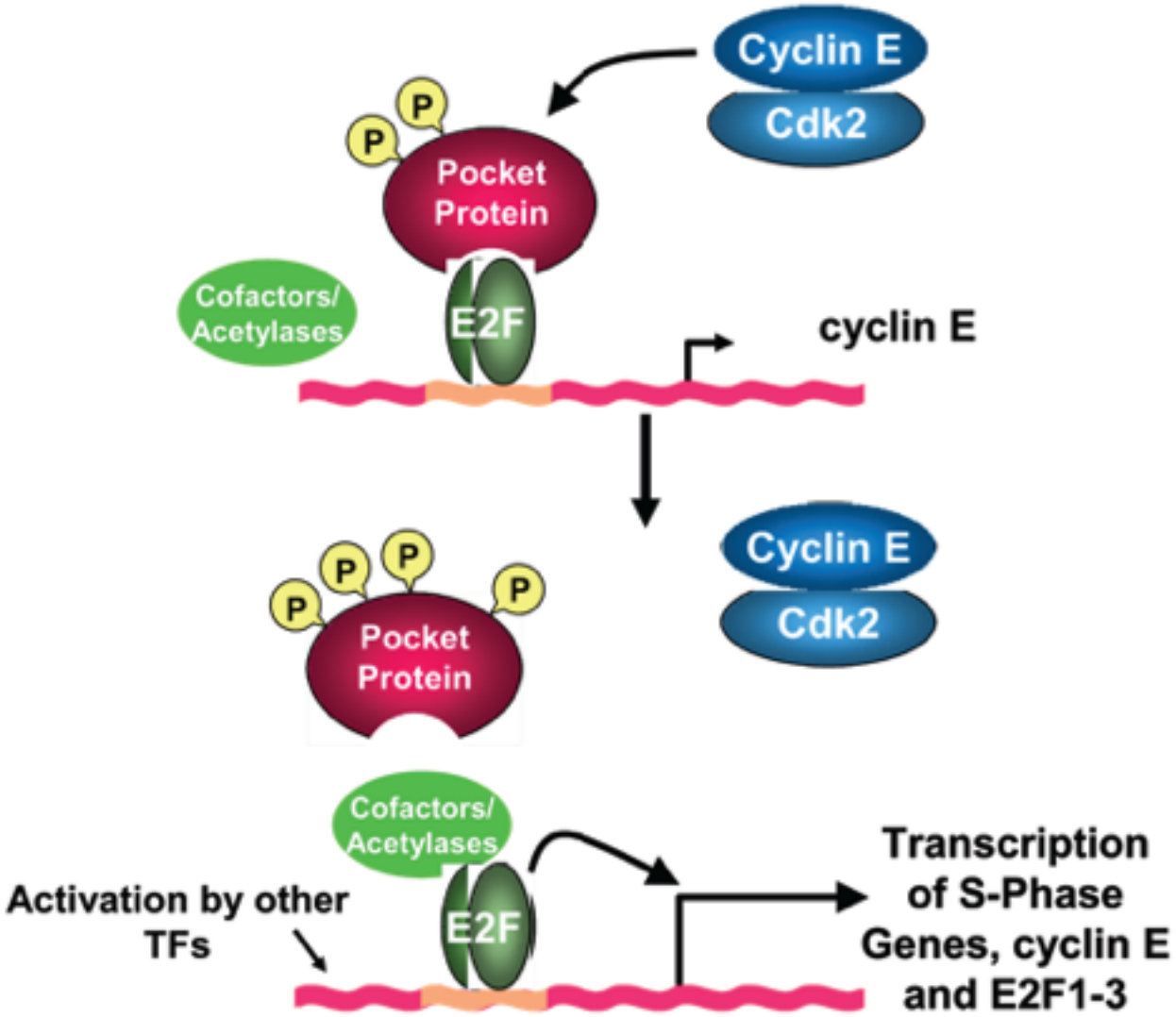
Cyclin/cdks phosphorylate and inactivate pocket proteins



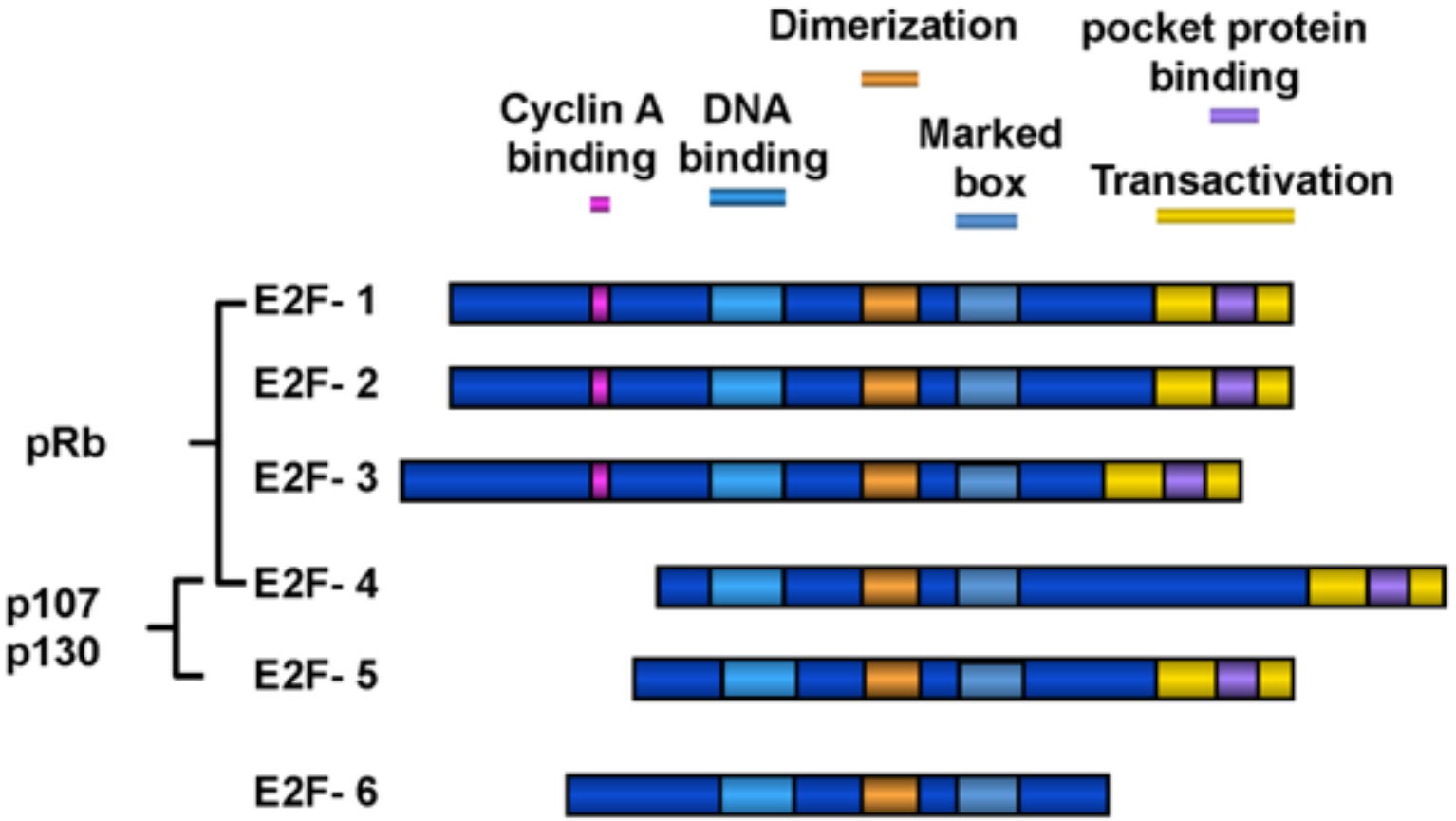
Cyclin D/cdk and cyclin E/cdk sequentially phosphorylate pocket proteins



Cyclin D/cdk and cyclin E/cdk sequentially phosphorylate pocket proteins



Pocket Protein Interactions with the E2F Family of Transcription Factors



Components of the E2F-pRB pathway disrupted in cancer

pRB.....**mutated or deleted**

p130.....**mutated (rare?)**

Cyclin D1.....**overexpressed (amplification/transcriptional)**

Cyclin E (rare).....**overexpressed**

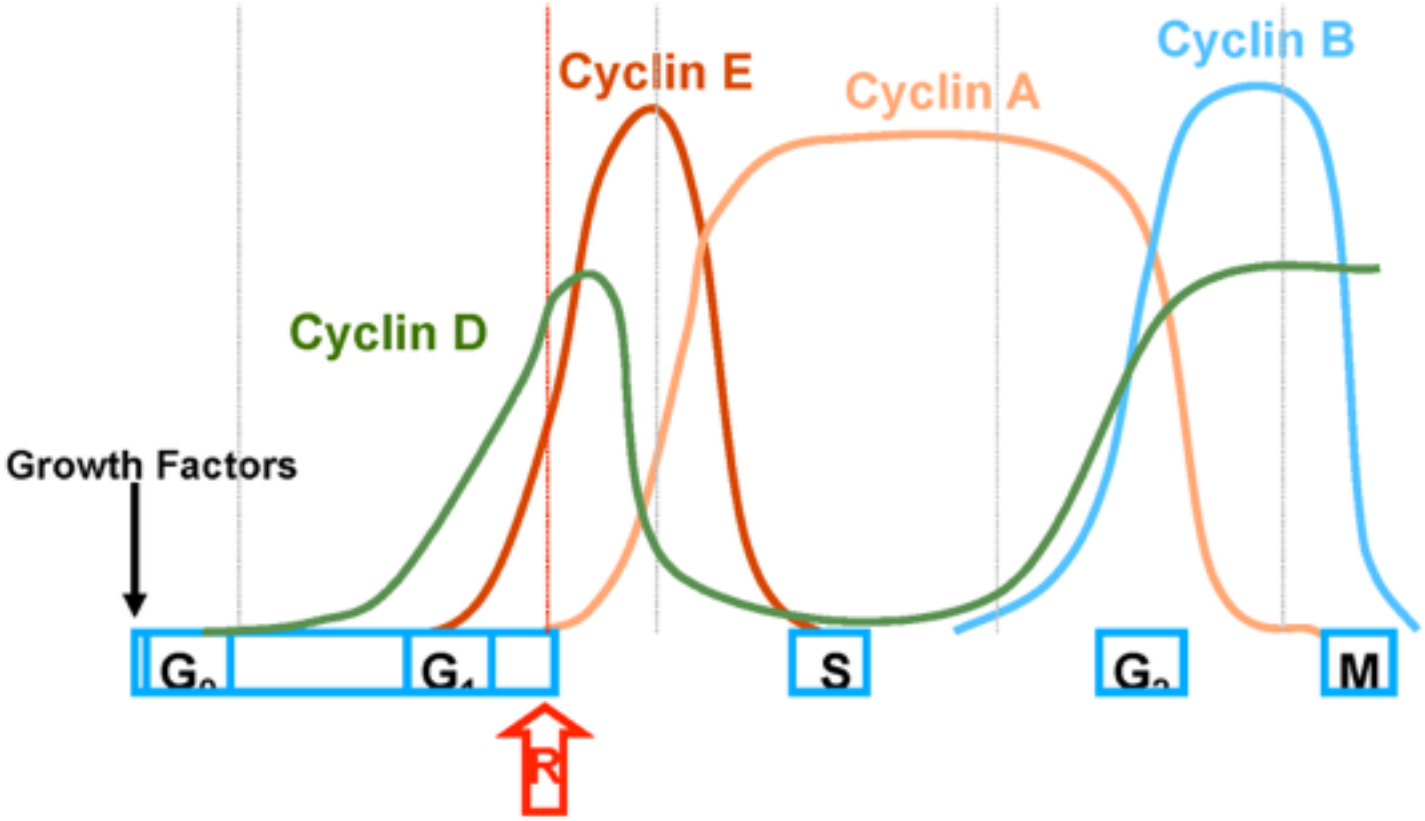
cdk4.....**overexpressed**

mutated to disrupt $p16^{INK4a}$ binding

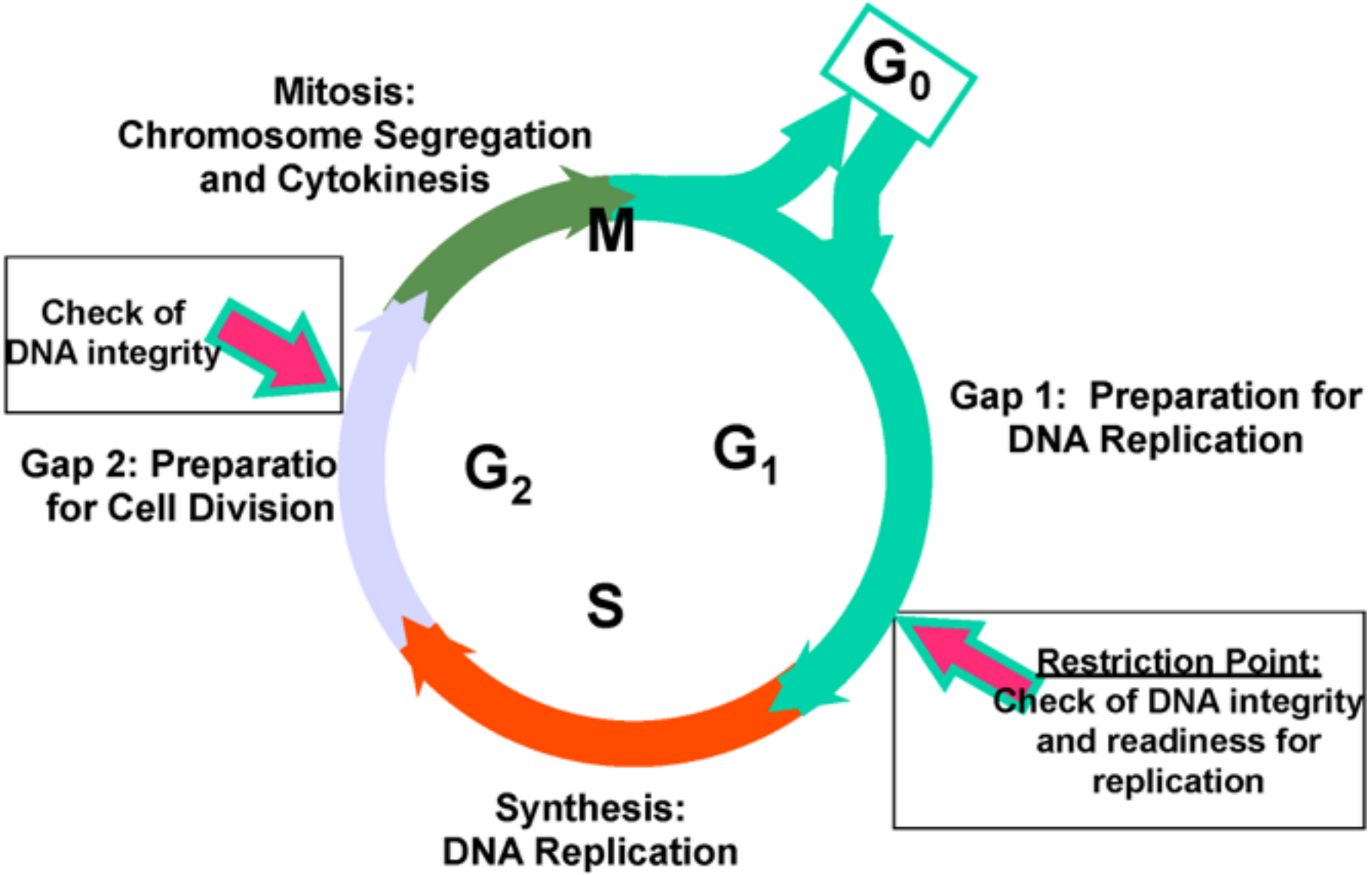
$p16^{INK4a}$..... **deleted or transcriptionally repressed**

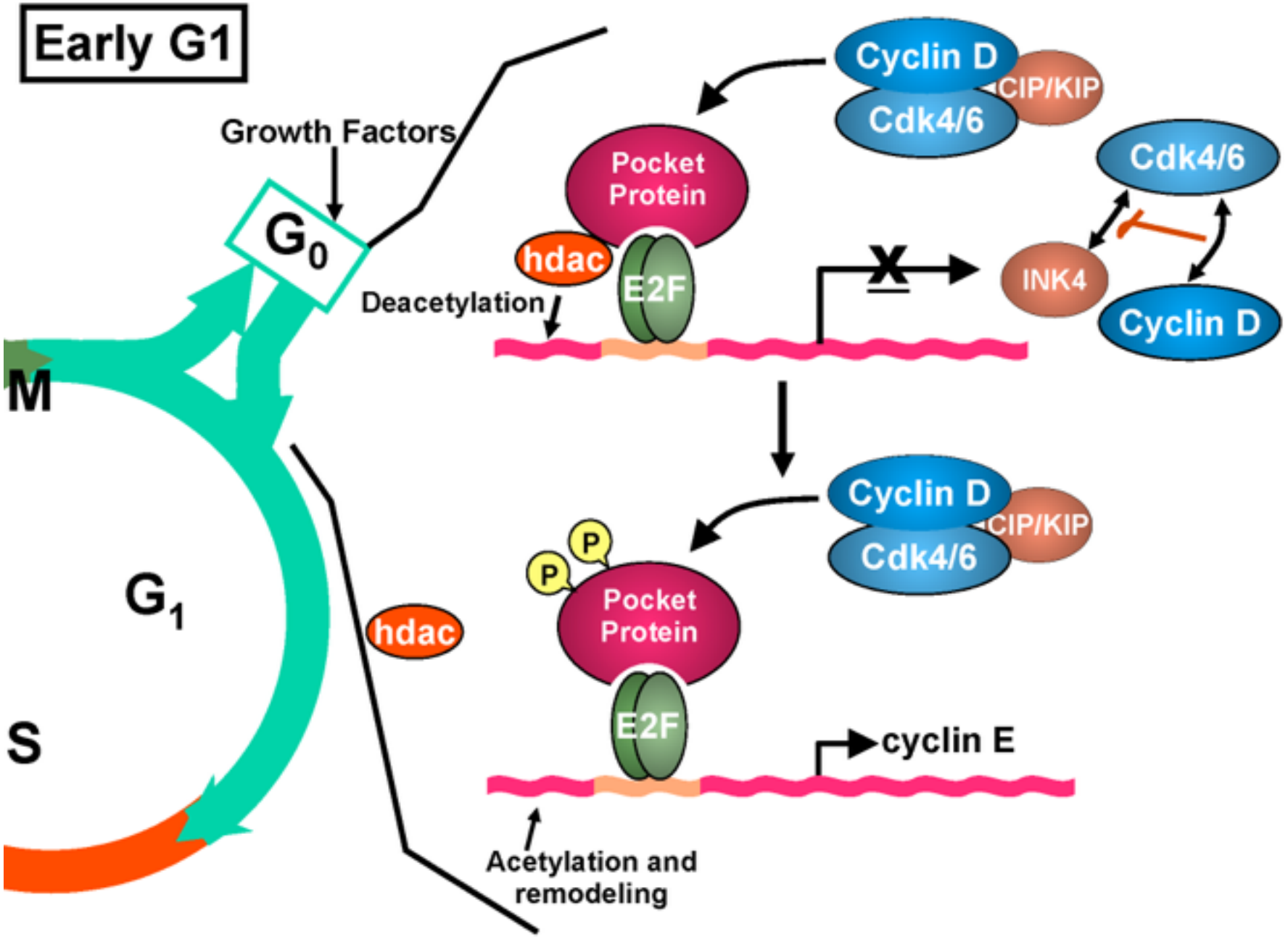
$p27^{KIP1}$..... **downregulated**

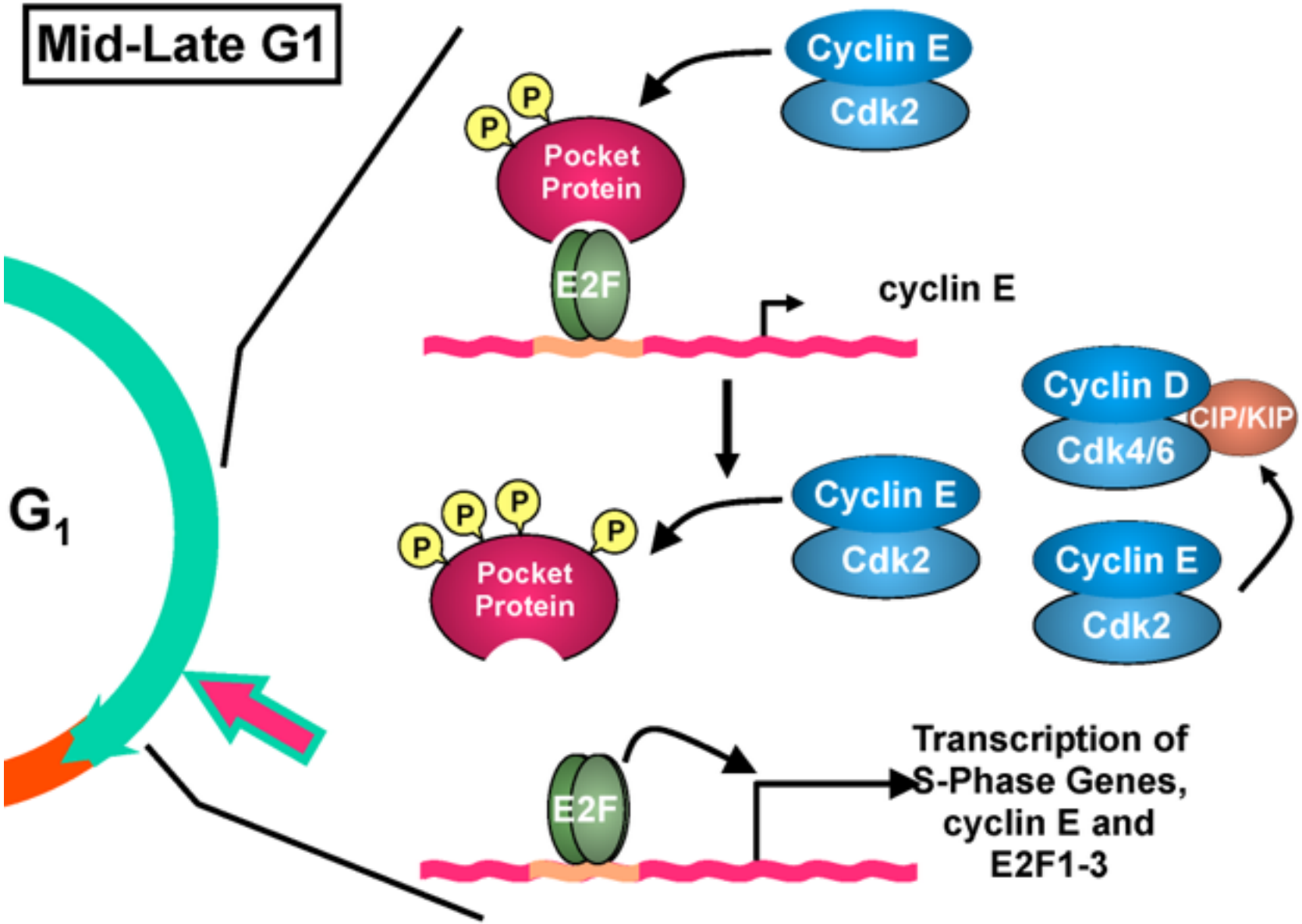
Cyclin Expression During Cell Cycle



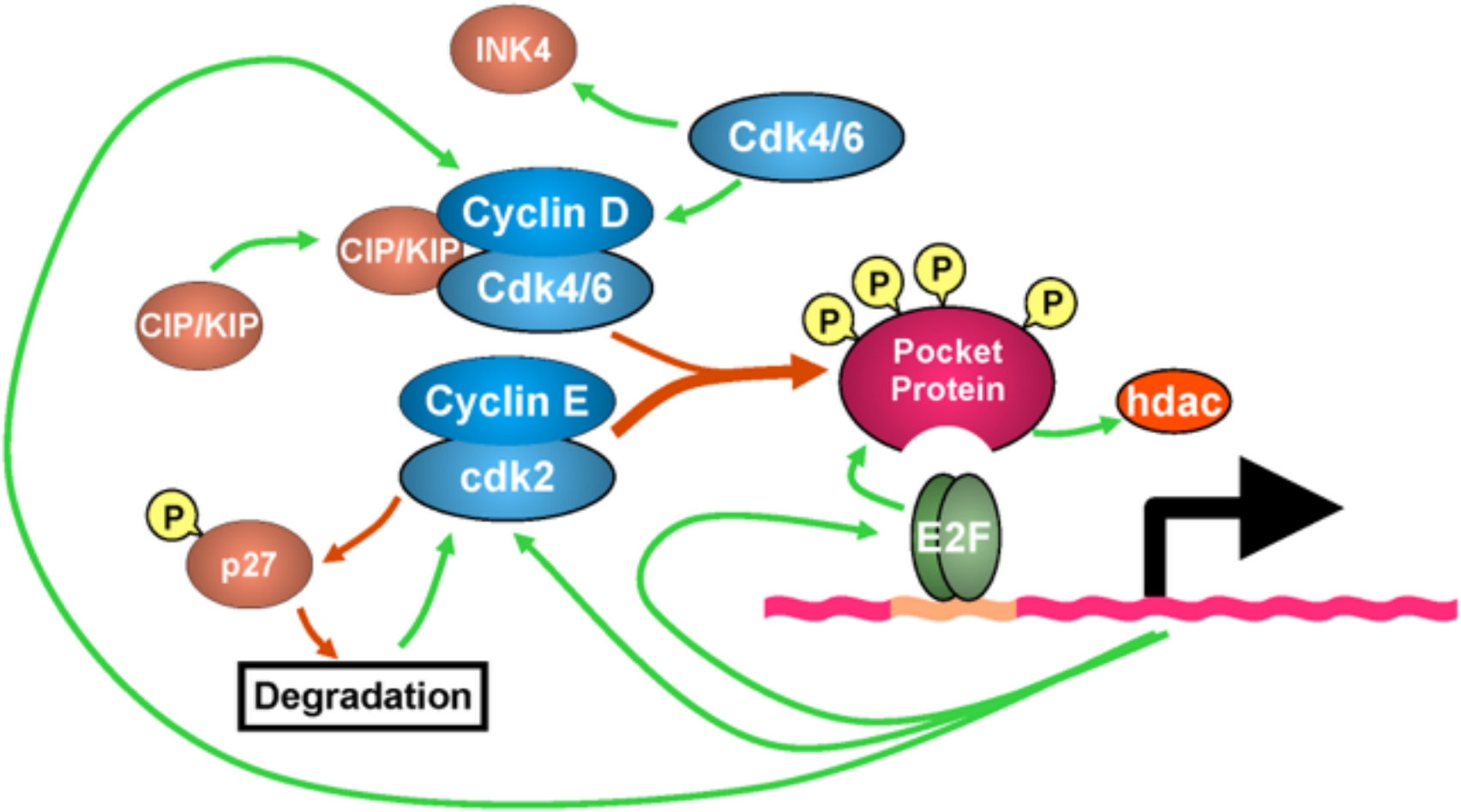
Phases of the cell cycle

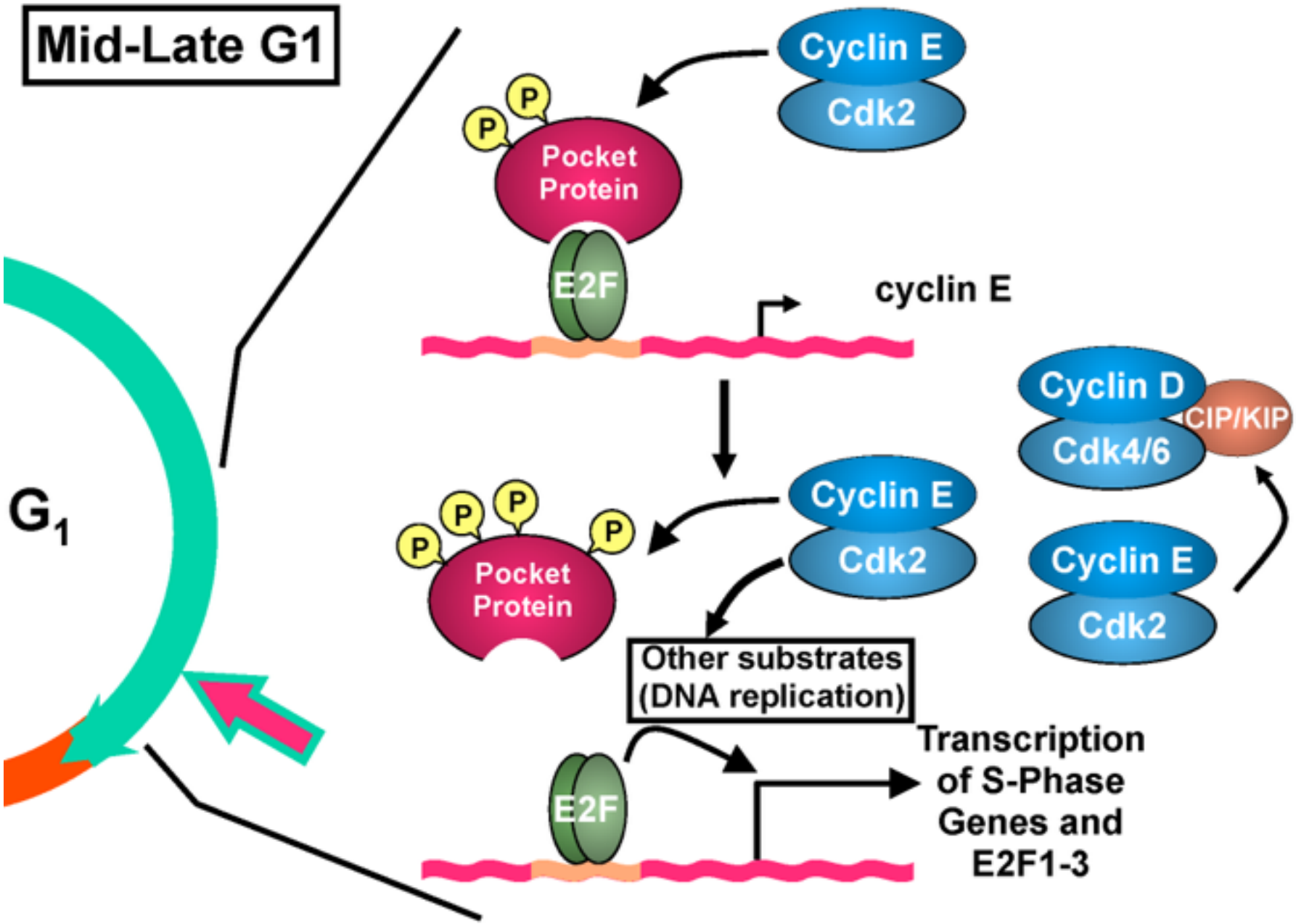



Early G1
Growth Factors
G0
M
G1
S
Cyclin D
CIP/KIP
Cdk4/6
Pocket Protein
hdac
E2F
Deacetylation
X
Cdk4/6
INK4
Cyclin D
Cyclin D
CIP/KIP
Cdk4/6
P
P
Pocket Protein
hdac
E2F
cyclin E
Acetylation and remodeling


Mid-Late G1
Cyclin E
Cdk2
P
P
Pocket Protein
E2F
cyclin E
G1
Cyclin D
CIP/KIP
Cdk4/6
Cyclin E
Cdk2
P
P
P
P
Pocket Protein
Cyclin E
Cdk2
E2F
Transcription of S-Phase Genes, cyclin E and E2F1-3

Positive Feedback Loop for E2F activation




Mid-Late G1
Cyclin E
Cdk2
P
P
Pocket Protein
E2F
cyclin E
G1
Cyclin D
CIP/KIP
Cdk4/6
Cyclin E
Cdk2
P
P
P
P
Pocket Protein
Cyclin E
Cdk2
Other substrates (DNA replication)
E2F
Transcription of S-Phase Genes and E2F1-3

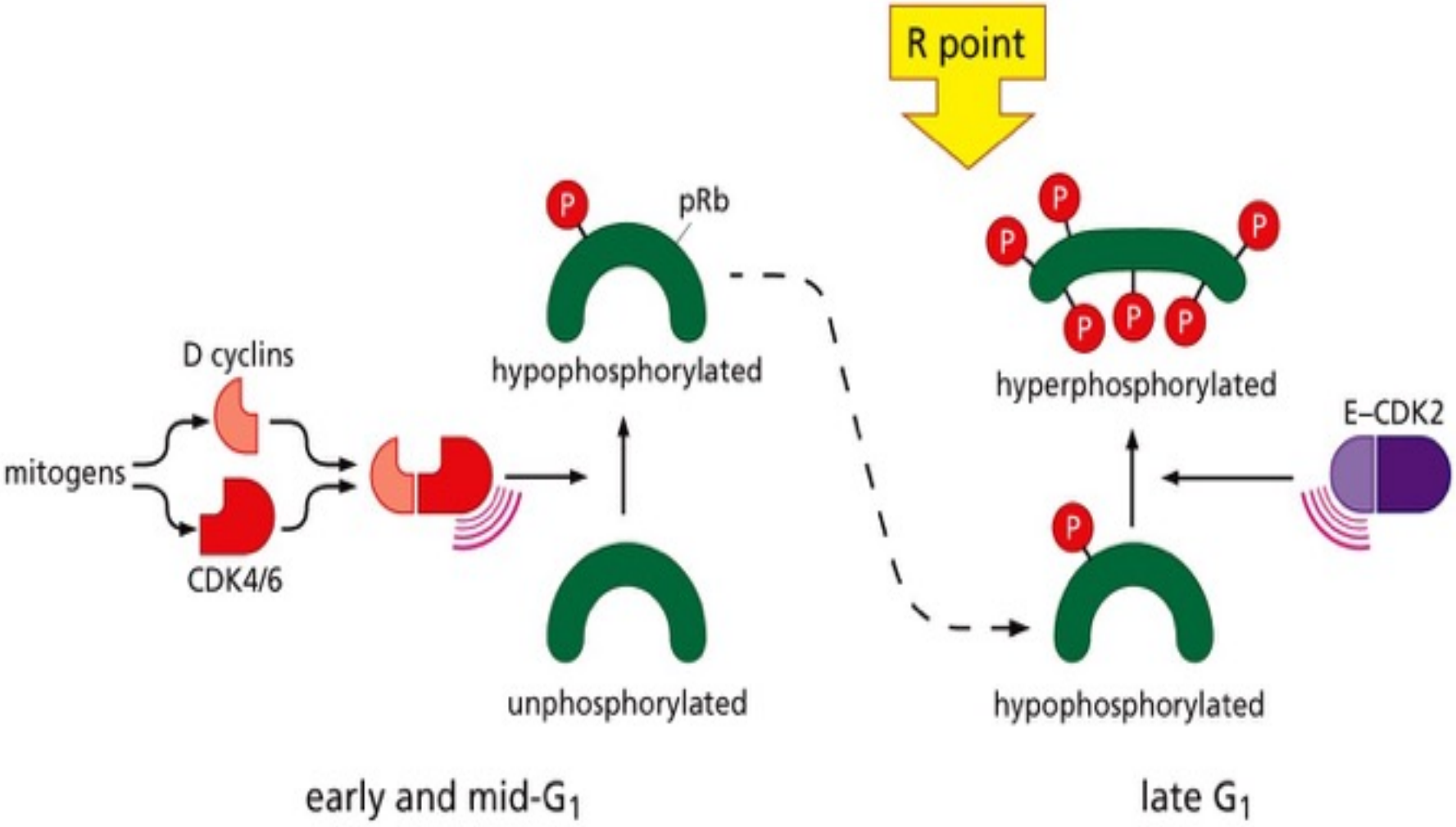
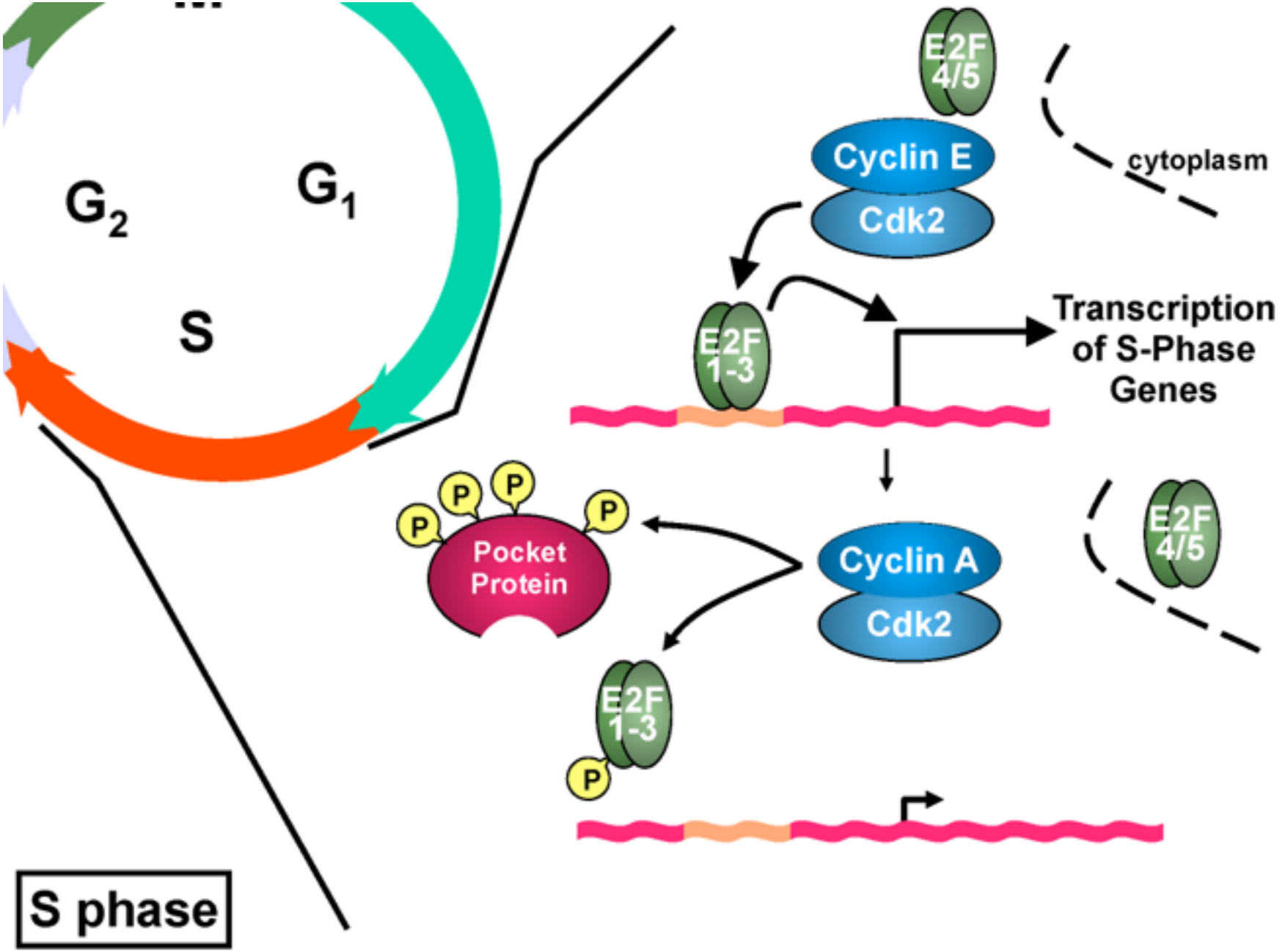


Figure 8.22 The Biology of Cancer (© Garland Science 2014)


G2
G1
S
S phase
E2F 4/5
Cyclin E
Cdk2
cytoplasm
E2F 1-3
Transcription of S-Phase Genes
P
P
P
P
Pocket Protein
Cyclin A
Cdk2
E2F 4/5
E2F 1-3
P

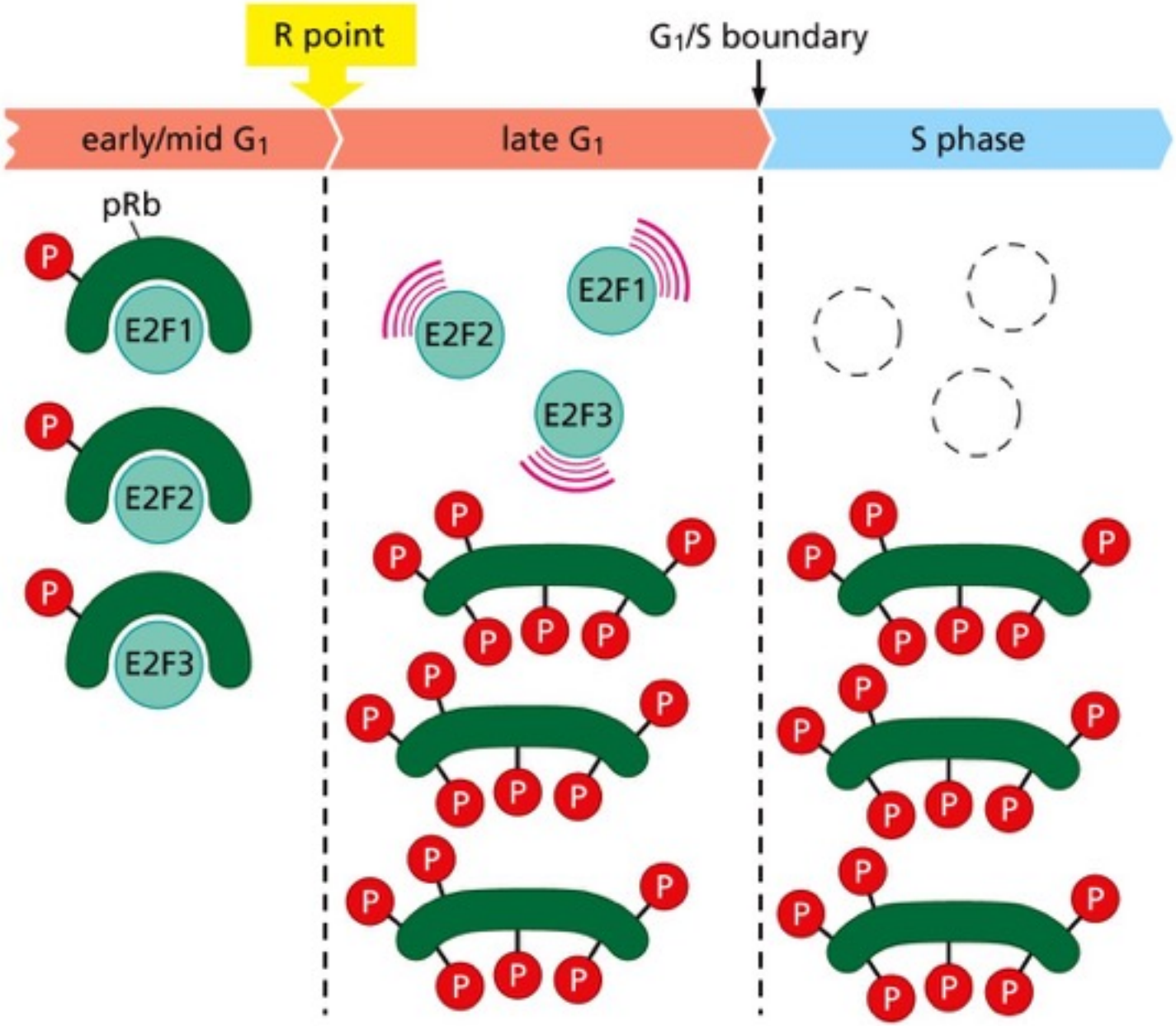
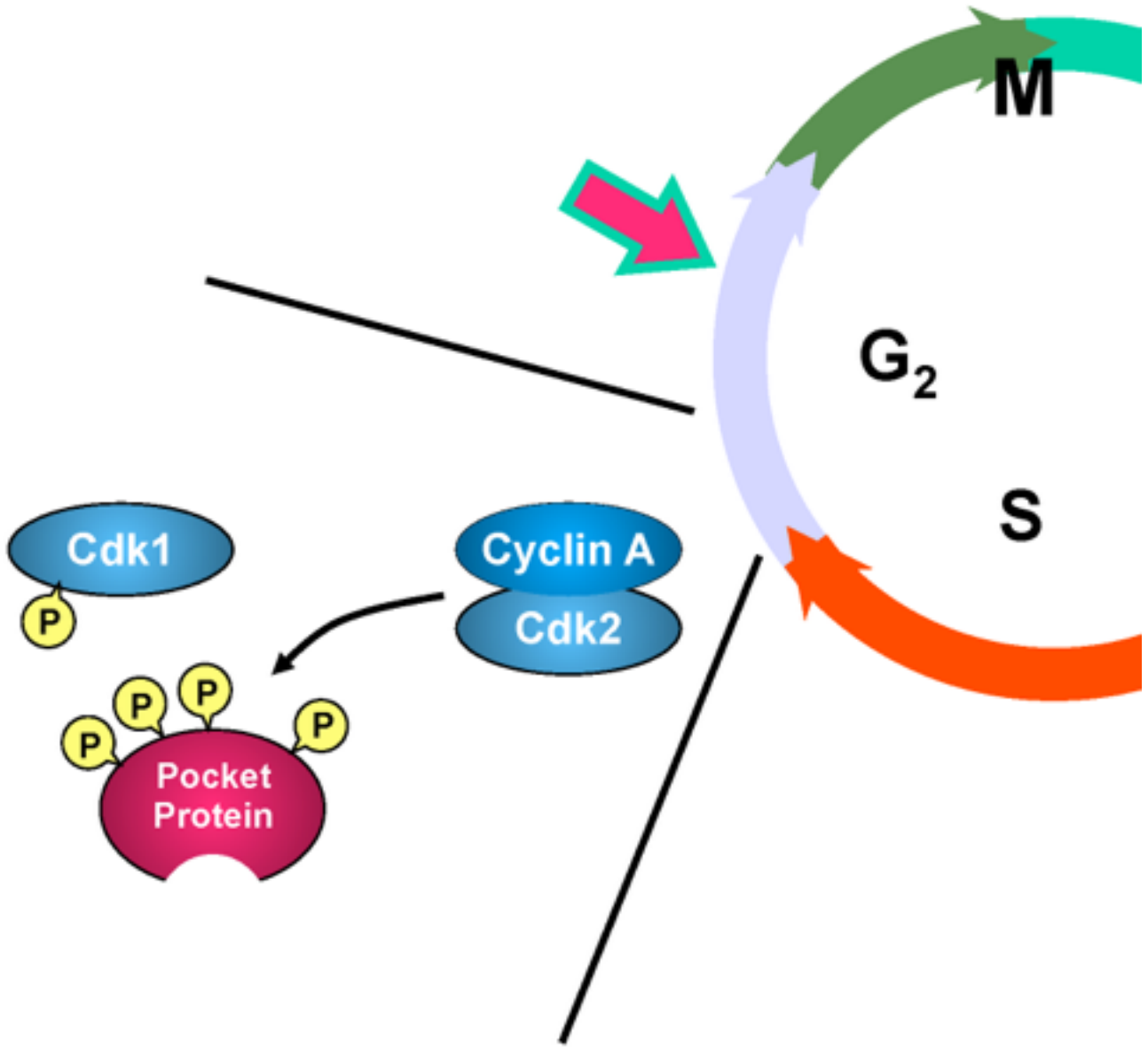
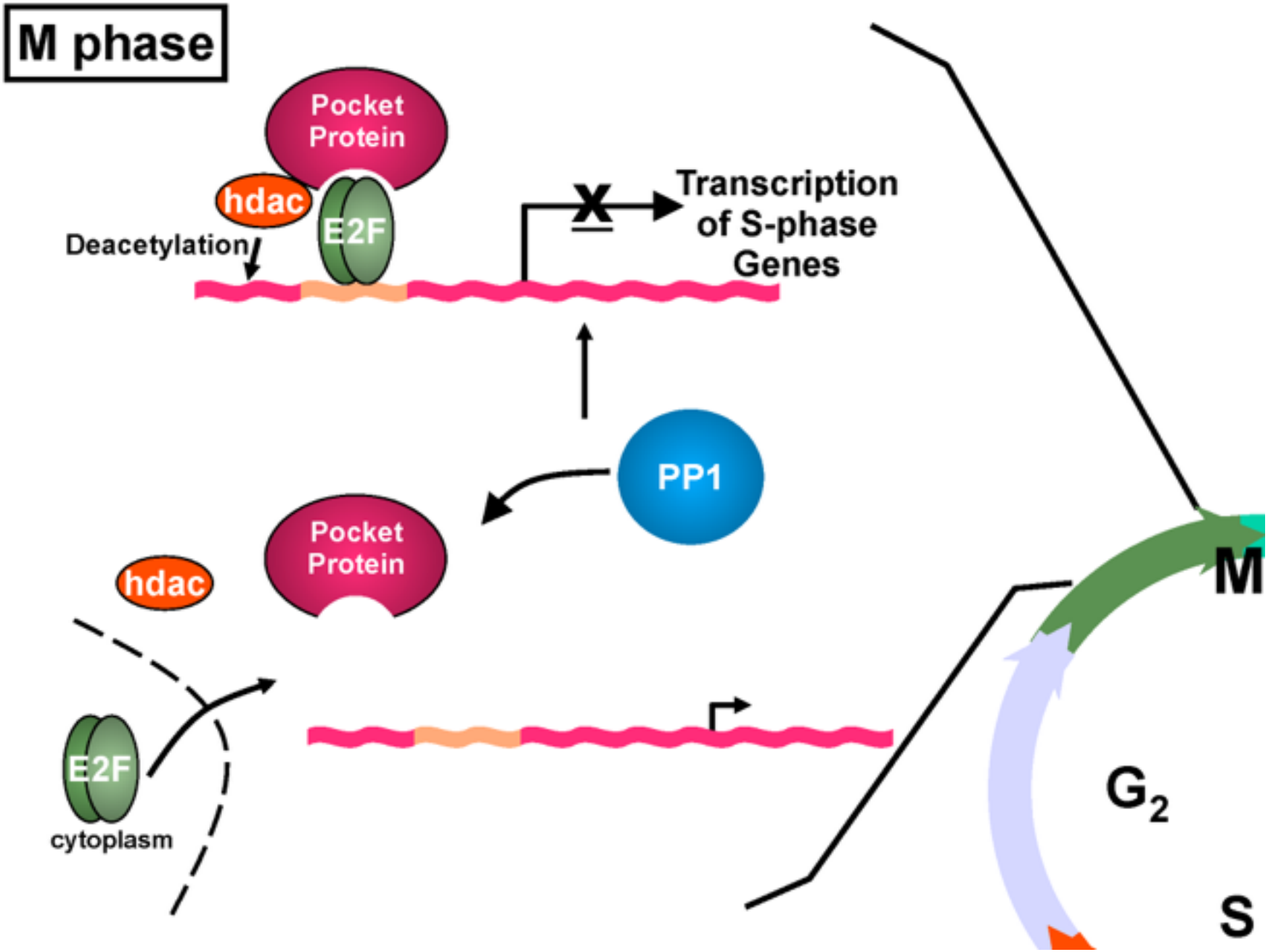
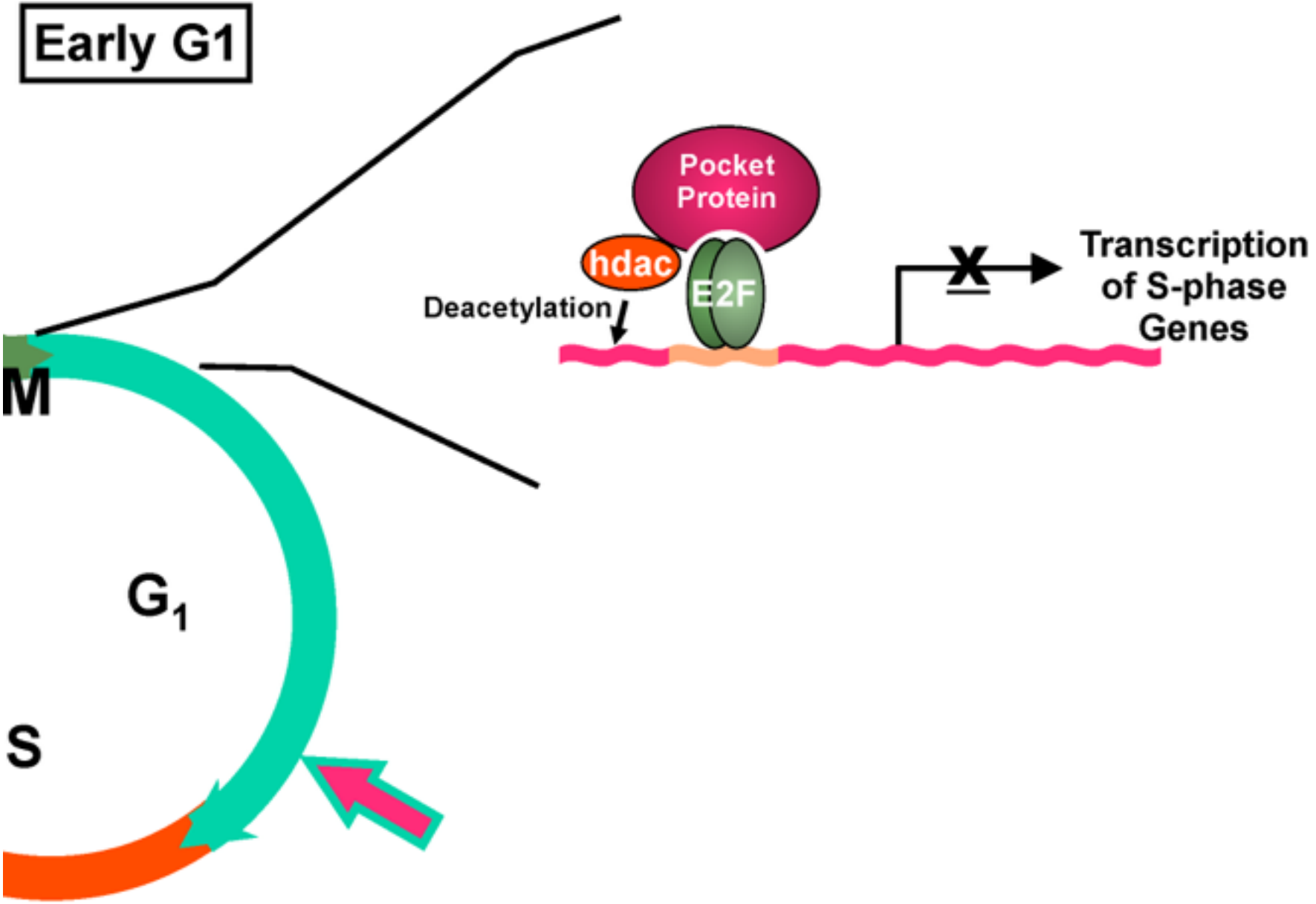


Figure 8.23a The Biology of Cancer (© Garland Science 2014)

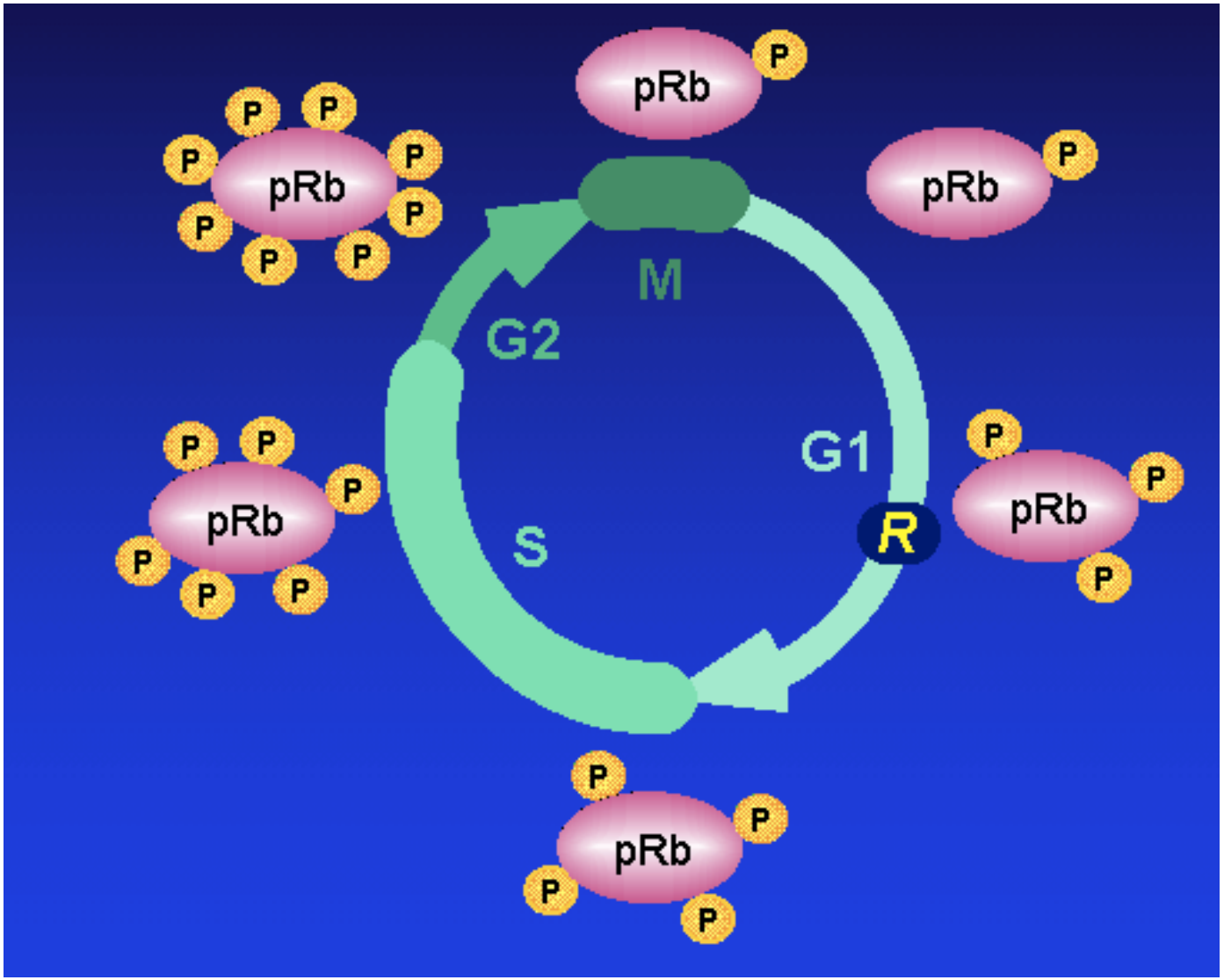


Early-mid G_2


M phase
Pocket Protein
hdac
E2F
Deacetylation
Transcription of S-phase Genes
PP1
Pocket Protein
hdac
E2F
cytoplasm
M
G2
S


Early G1
Pocket Protein
hdac
E2F
Deacetylation
Transcription of S-phase Genes
M
G1
S

Phosphorylation status of the retinoblastoma protein (pRB) during cell cycle



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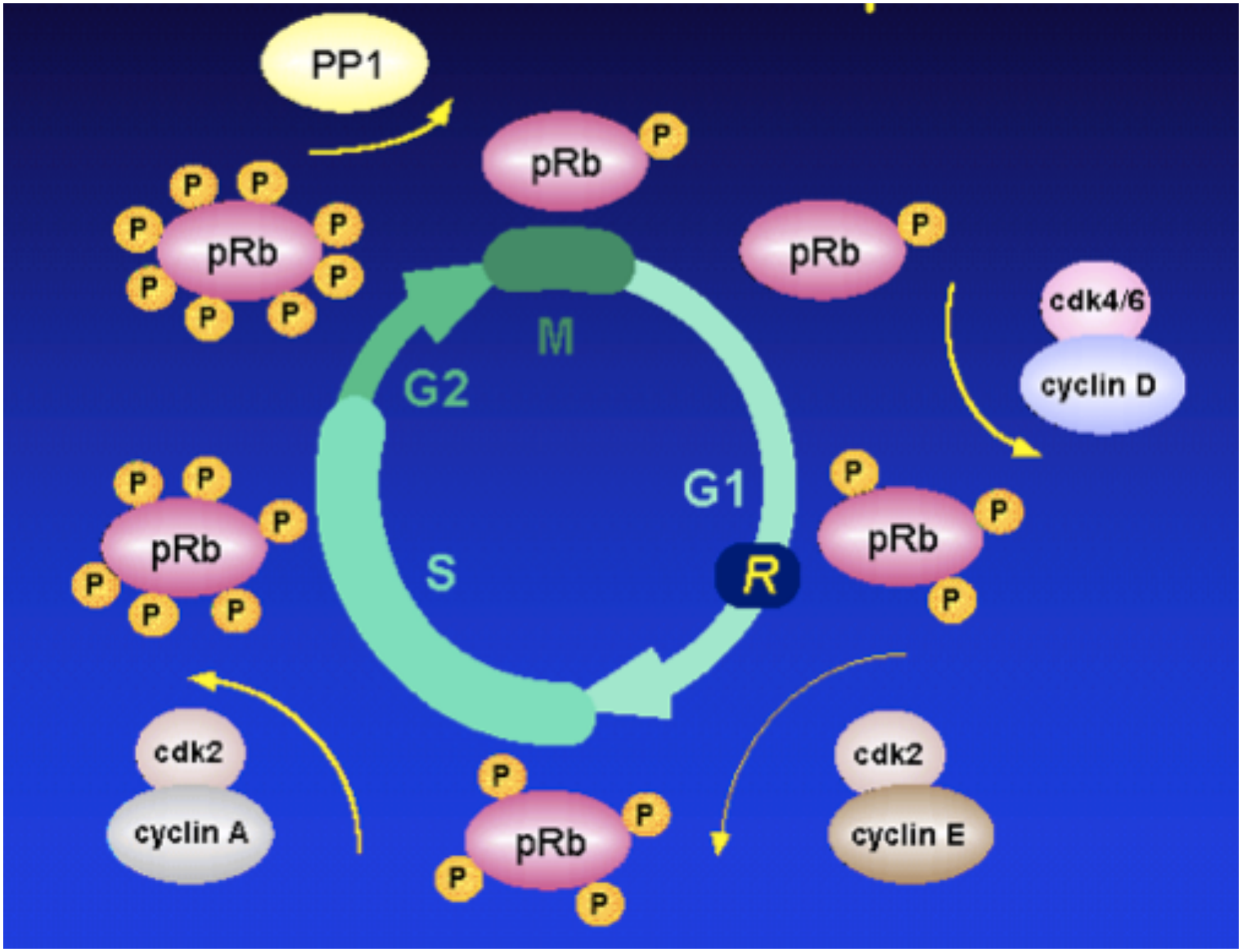
Cyclin-dependent kinases inactivate the retinoblastoma protein (pRB)



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Pairing of cyclins with cyclin-dependent kinases

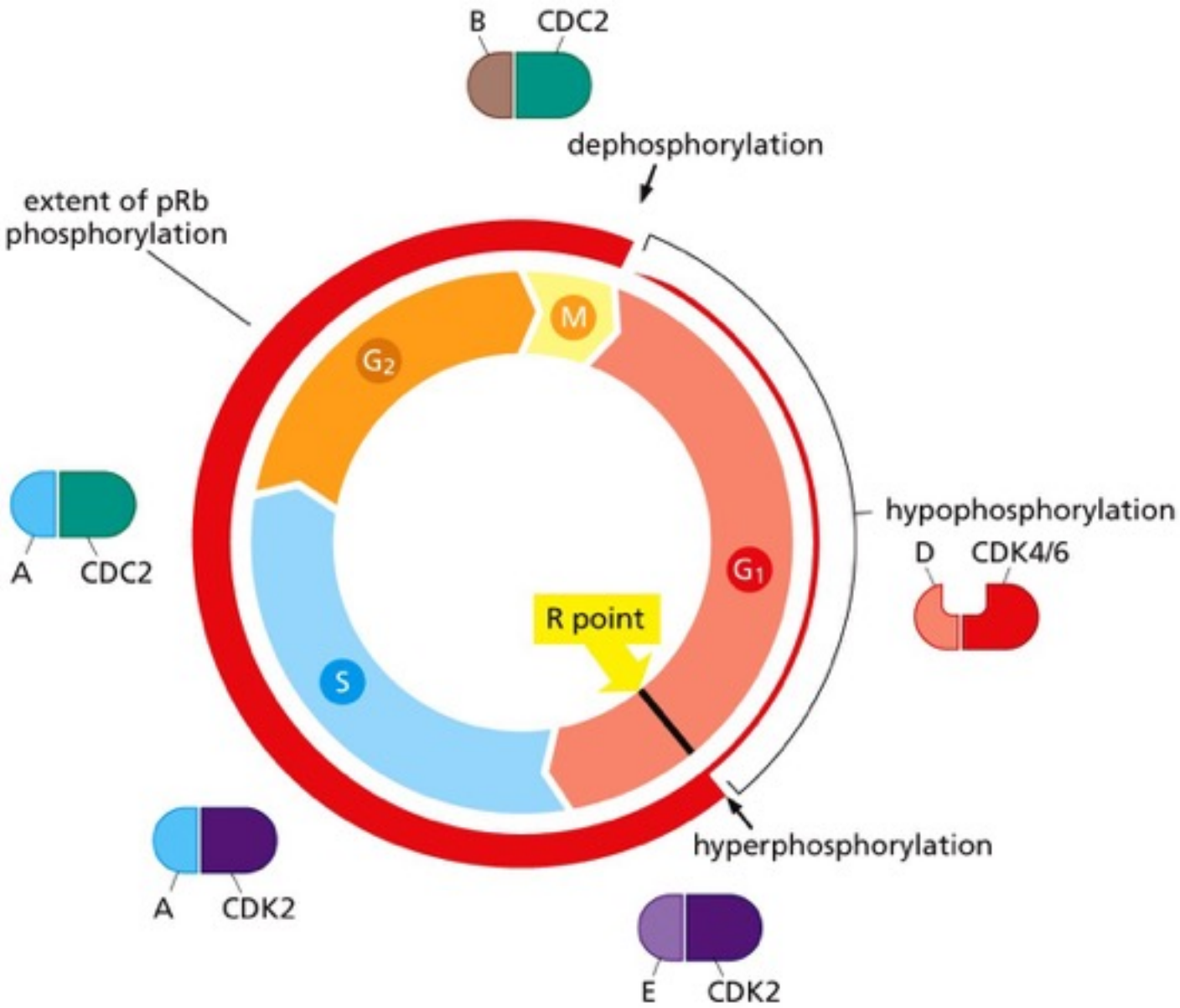
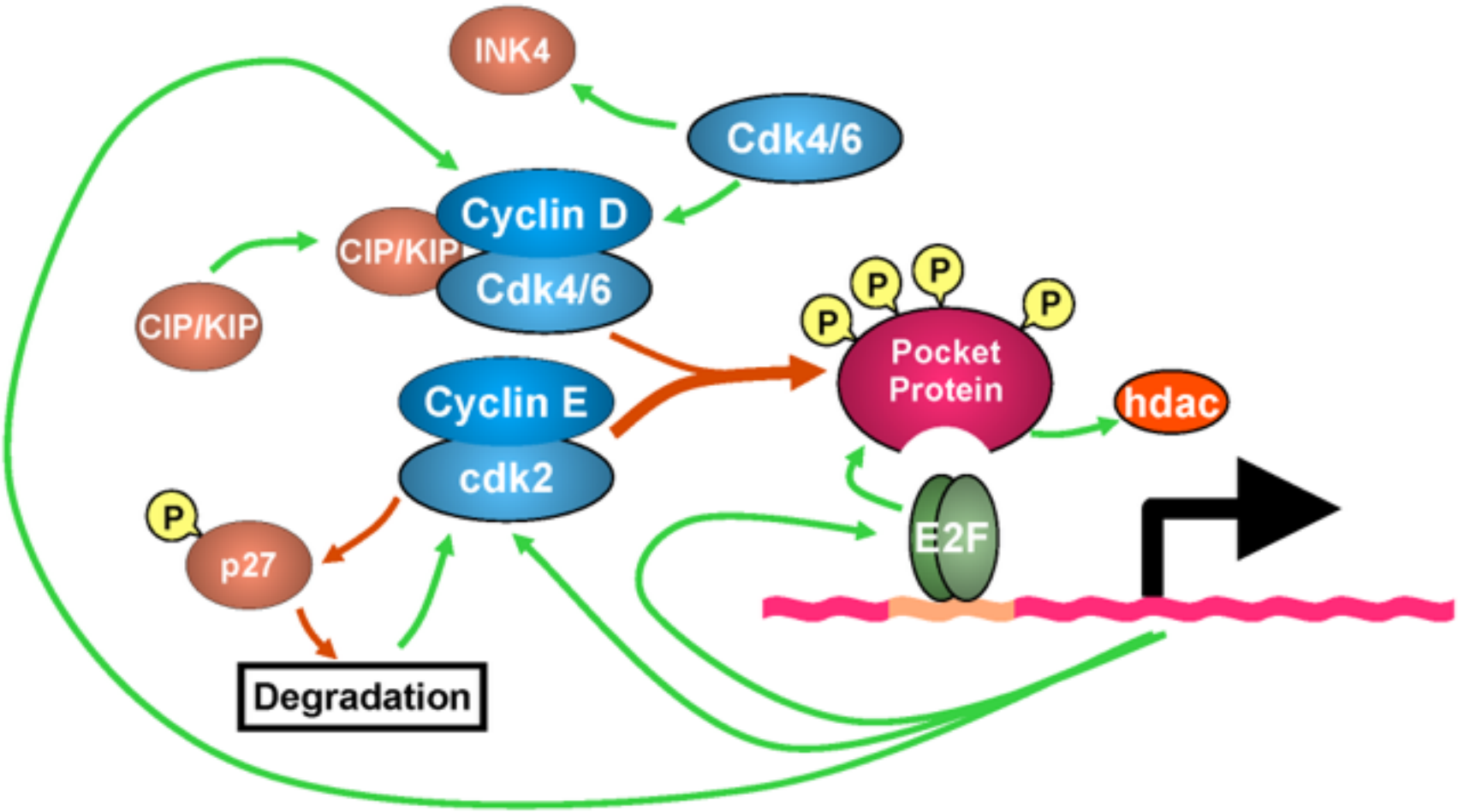
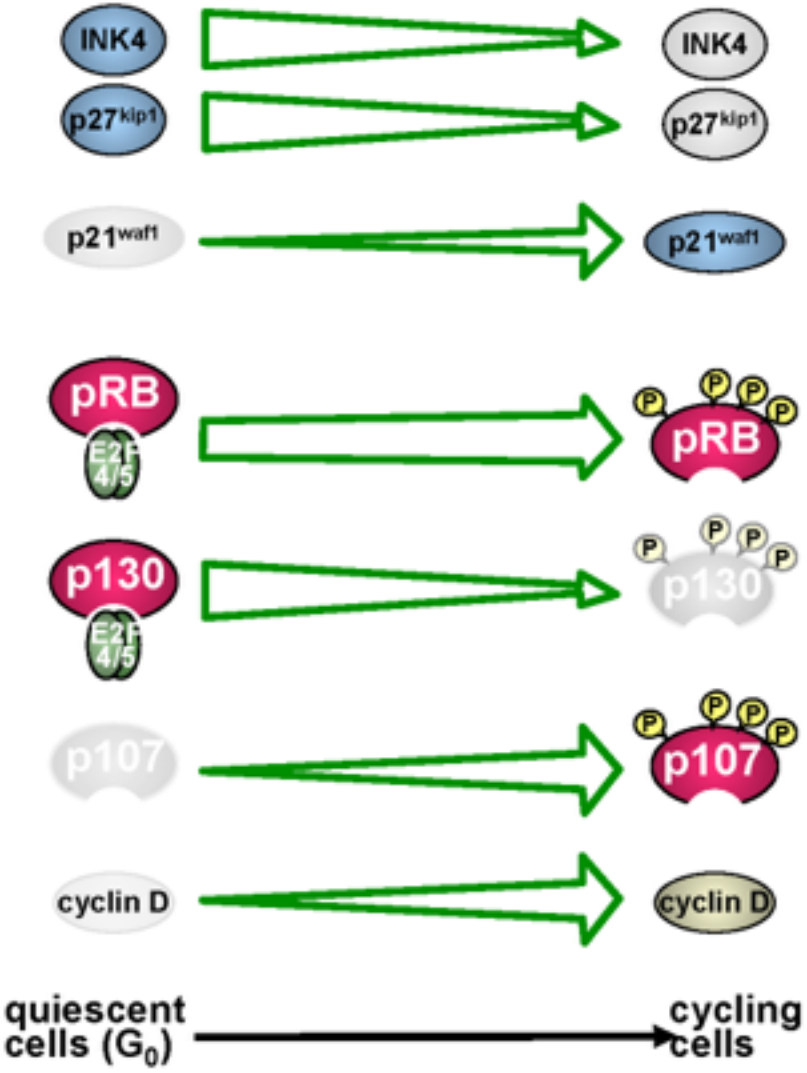


Figure 8.19 The Biology of Cancer (© Garland Science 2014)

Positive Feedback Loop for E2F activation



Transition from G_0



Transition into G_0