Grundlagen der Zellbiologie

DNA-Replikation und Reparatur

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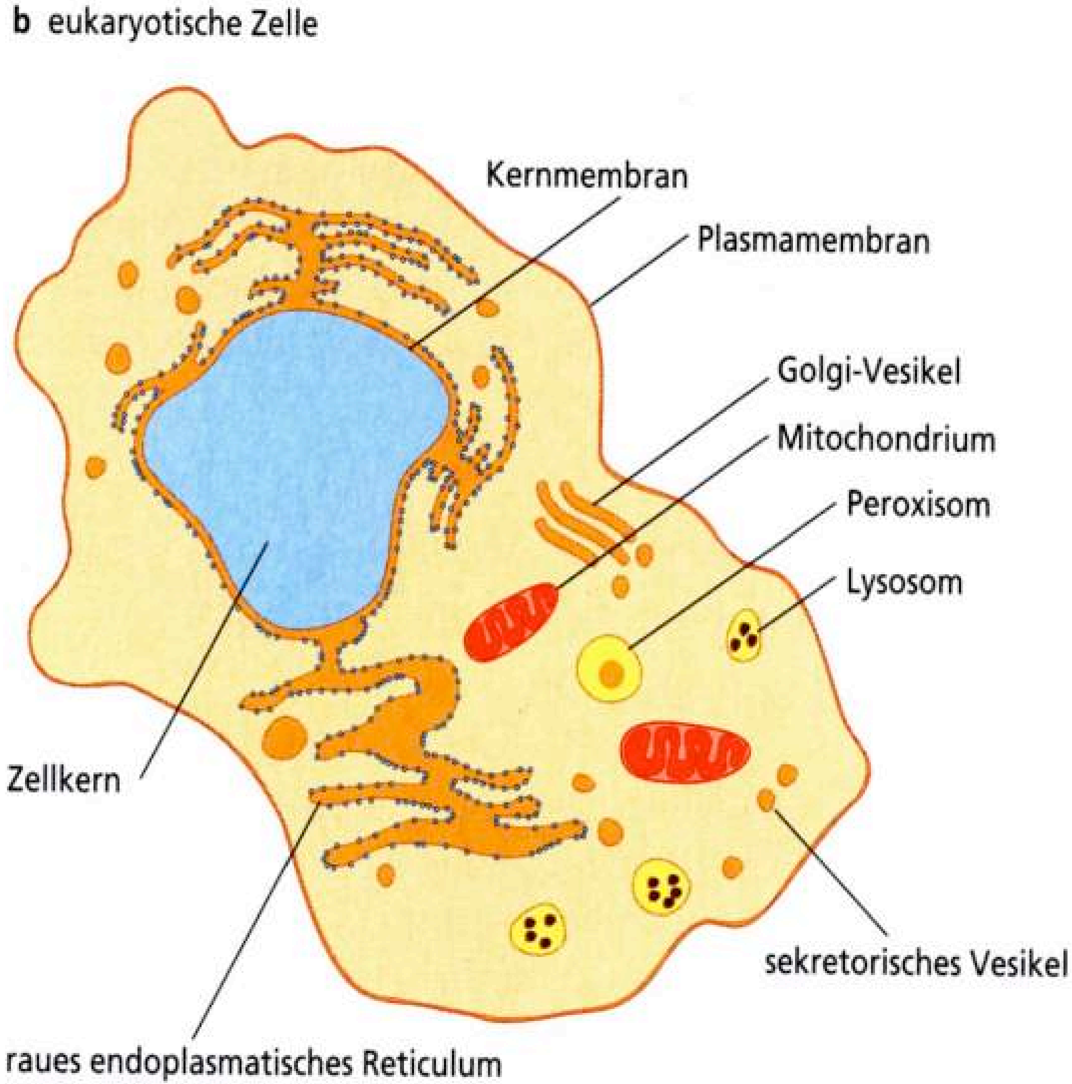
25. Oktober 2005

Überblick

- Primär-, Sekundär- und Tertiärstruktur der DNA
- DNA-Bausteine
- Biochemie der DNA-Synthese
- Initiation und Orientierung der Replikation
- Replikationsgabel
- Chromosomenreplikation
- Proofreading
- Mutagenese
- DNA-Reparatur

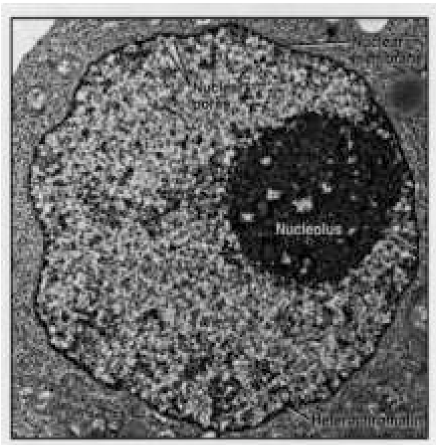
Organelle der eukaryontischen Zelle

1. **Plasmamembran**
2. **Cytoplasma**
3. **Zellkern / Nukleus**
4. **Endoplasmatisches Retikulum (ER)**
5. **Golgi-Apparat**
6. **Vesikel (sekretorisch)**
7. **Peroxysom, Lysosom**
8. **Mitochondrien**

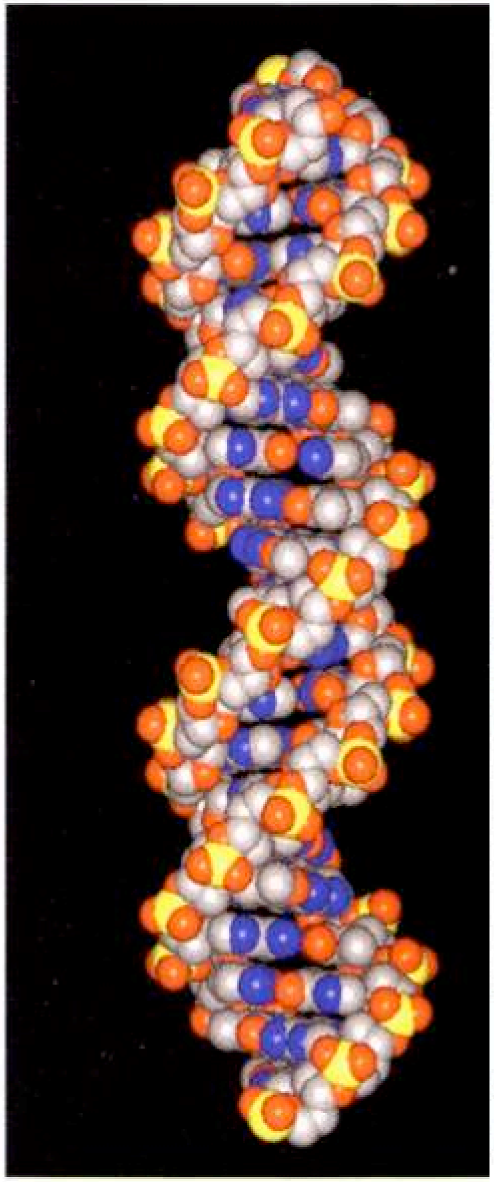


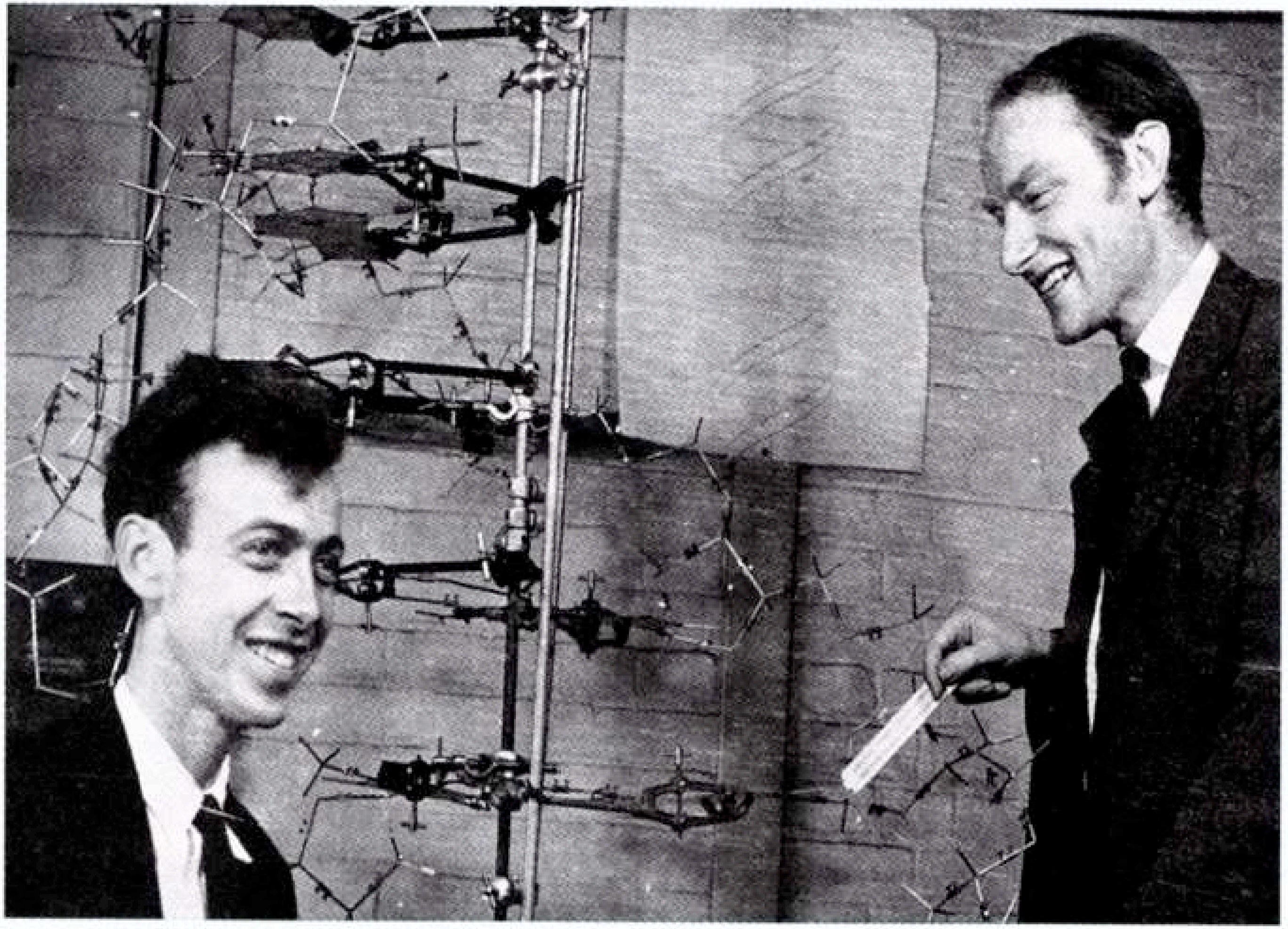
Friedrich Miescher, 1869
Tübingen
(Schloss Hohentübingen)

Isolation von „Nuklein“
aus dem Nukleus von Zellen



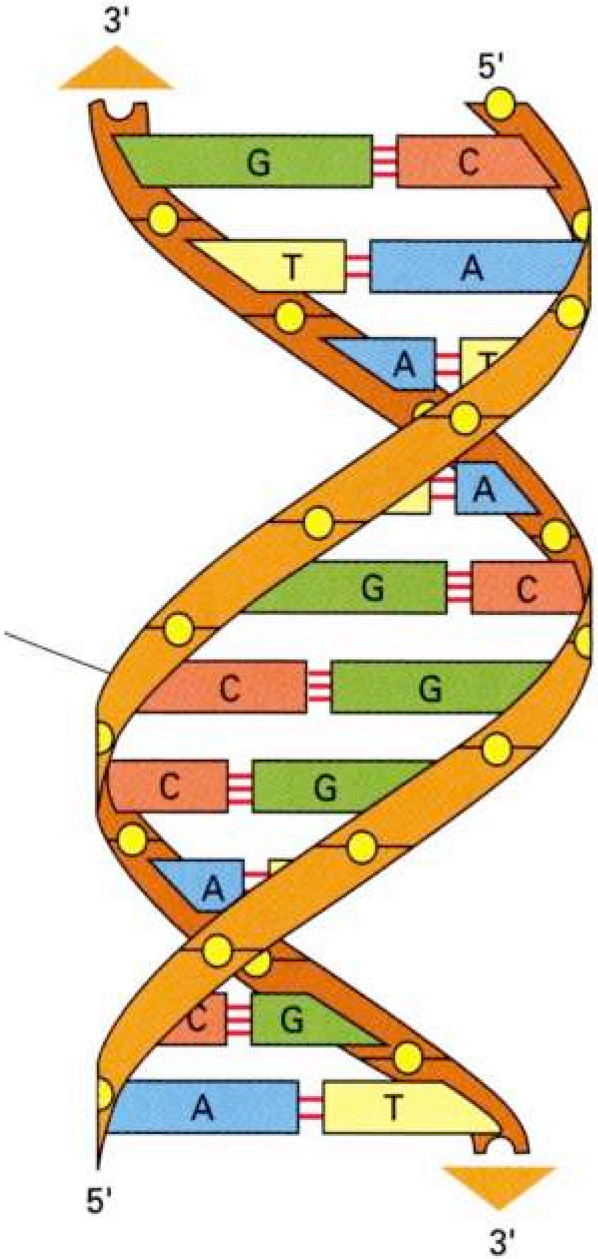
Die DNA Doppelhelix: 1953 - 2003





James Watson and Francis Crick with their DNA model.
(Camera Press.)

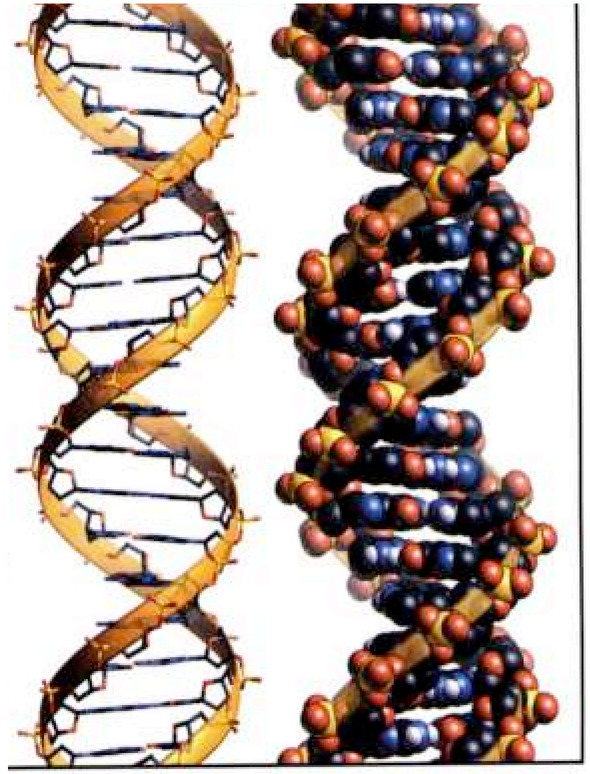
DNA Doppelhelix Struktur



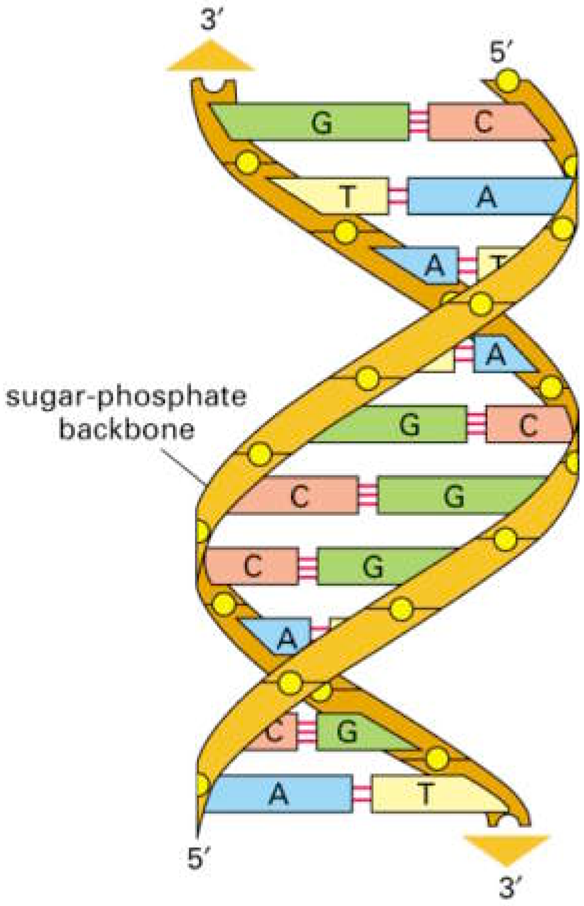
-komplementäre Einzelstränge -

(Hinweis: E. Chargaff)

Die Basenpaarung:
A-T, G-C



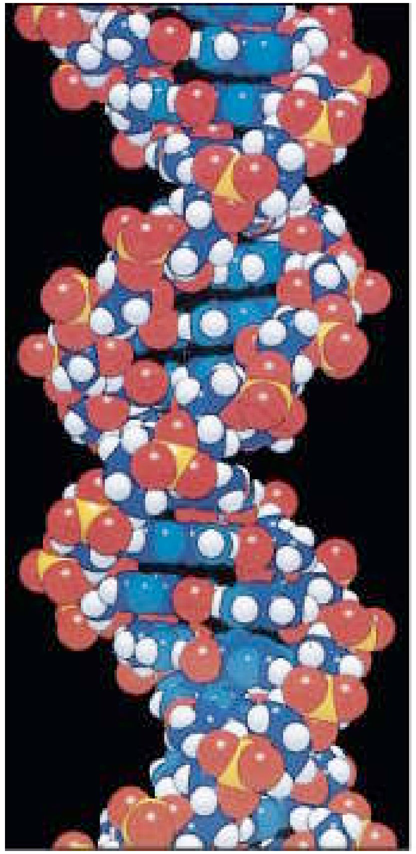
Replikationsahnungen

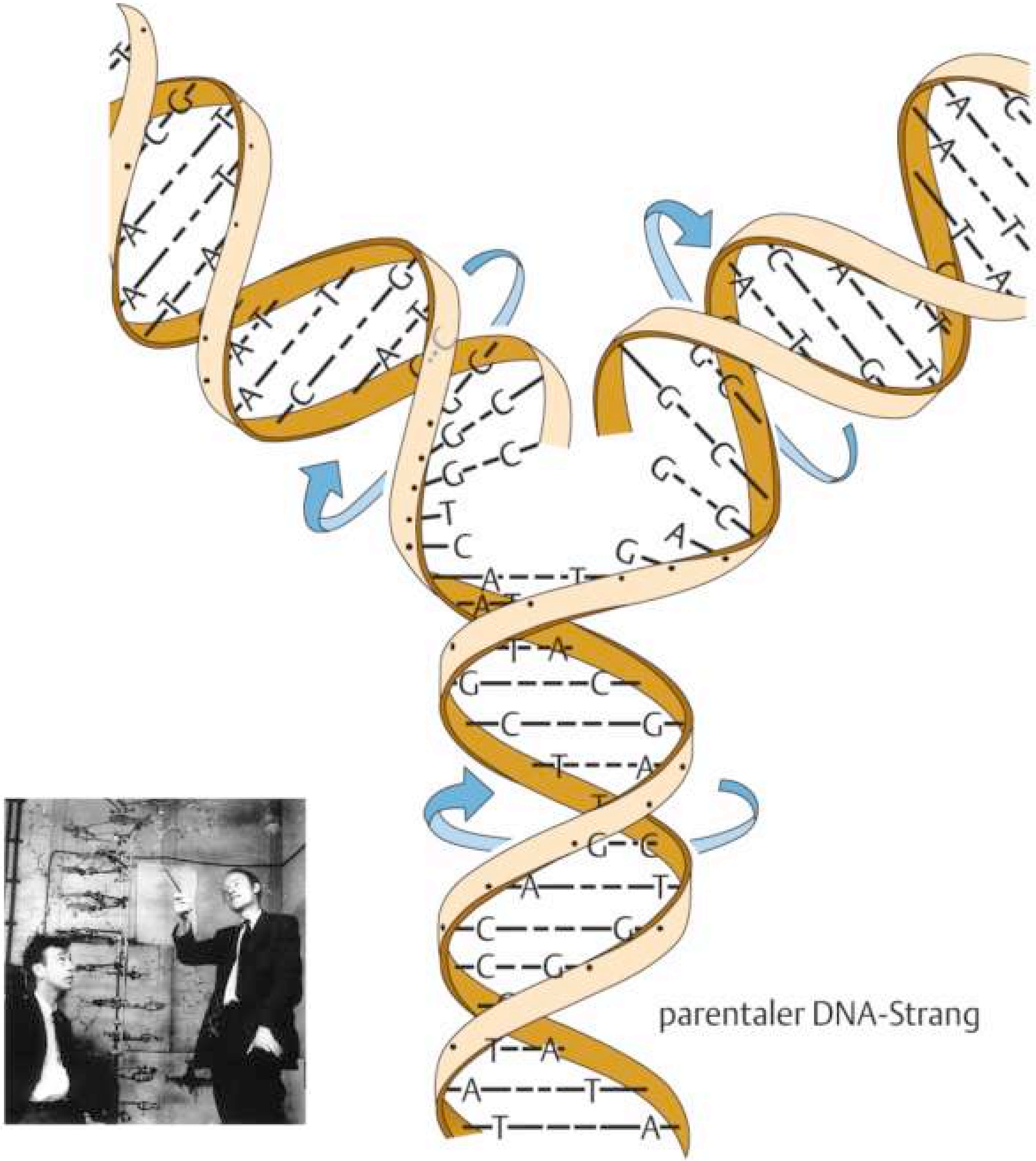


!. Molecular Biology of the Cell, 4th Edition.

“It has not escaped our notice that the specific pairing we have postulated immediately suggest a possible copying mechanism for the genetic material.”

Watson & Crick 1953



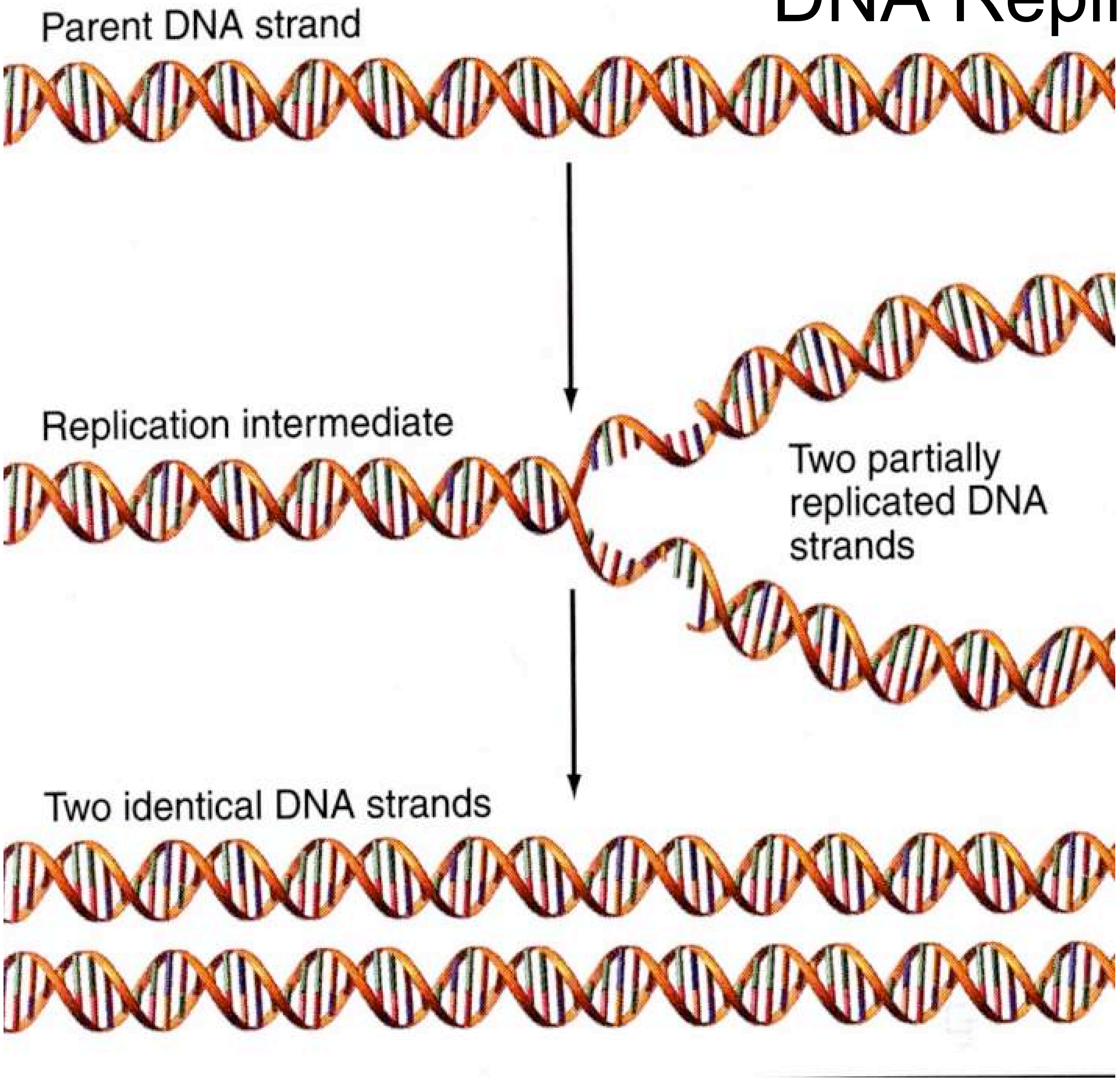


Sprache der DNA:
4 Buchstaben
(G, A, T, C)

DNA Doppelhelix
hat komplementäre
Einzelstränge

Doppelhelikale Struktur
der DNA ermöglicht
identische Verdoppelung
der Erbinformation.

DNA Replikation



Die DNA-Replikation ist semikonservativ

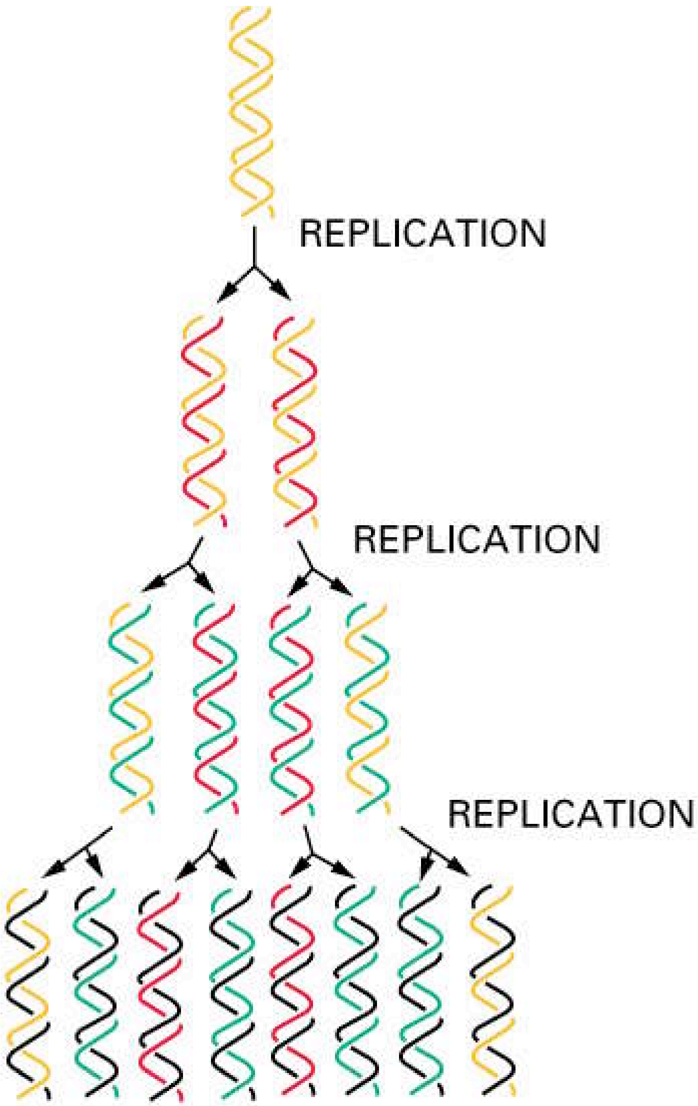
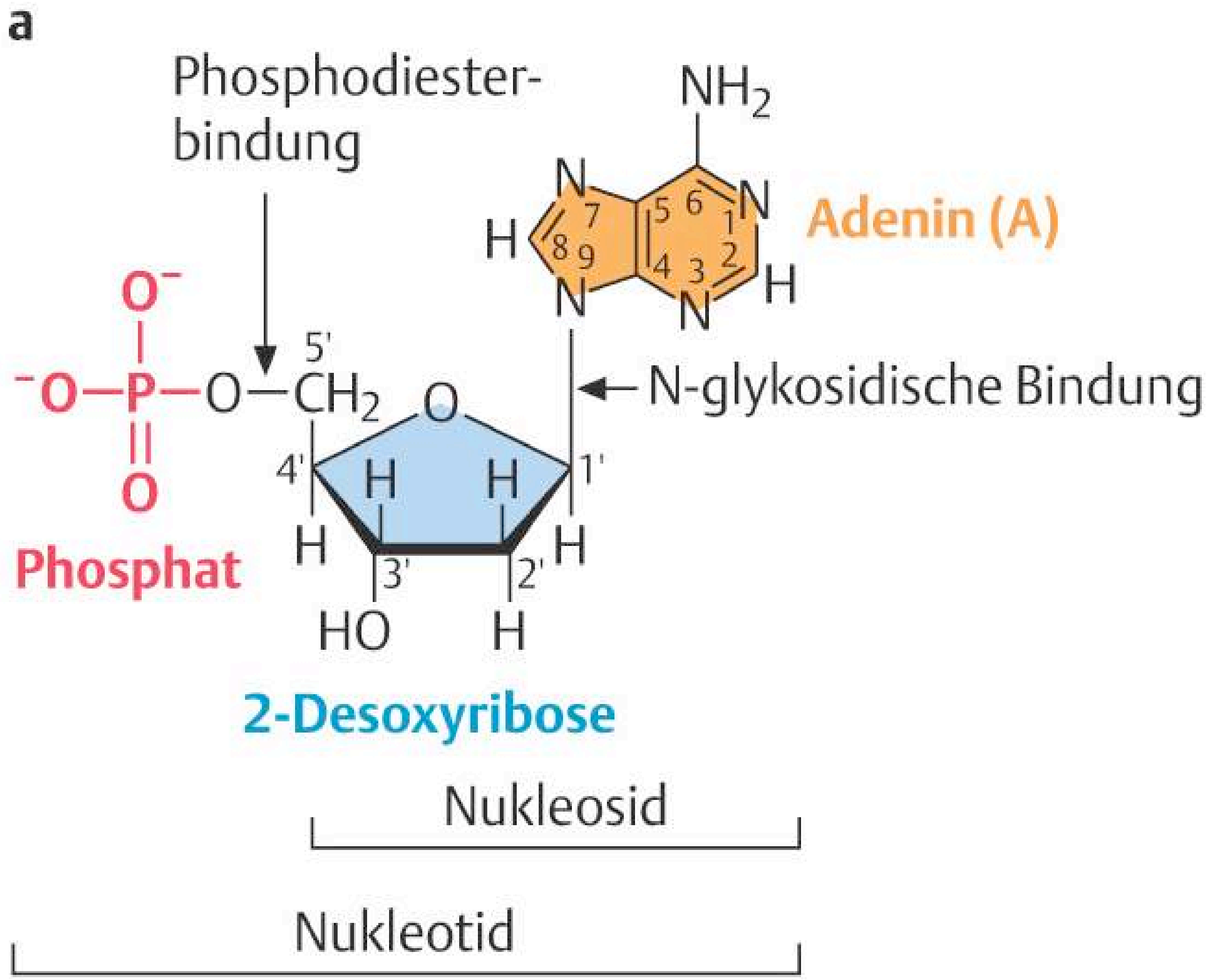
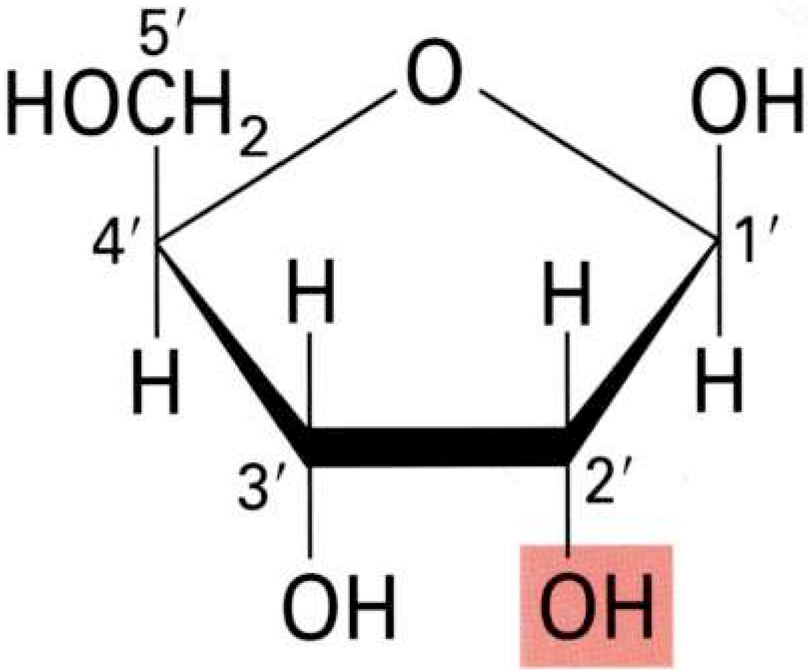


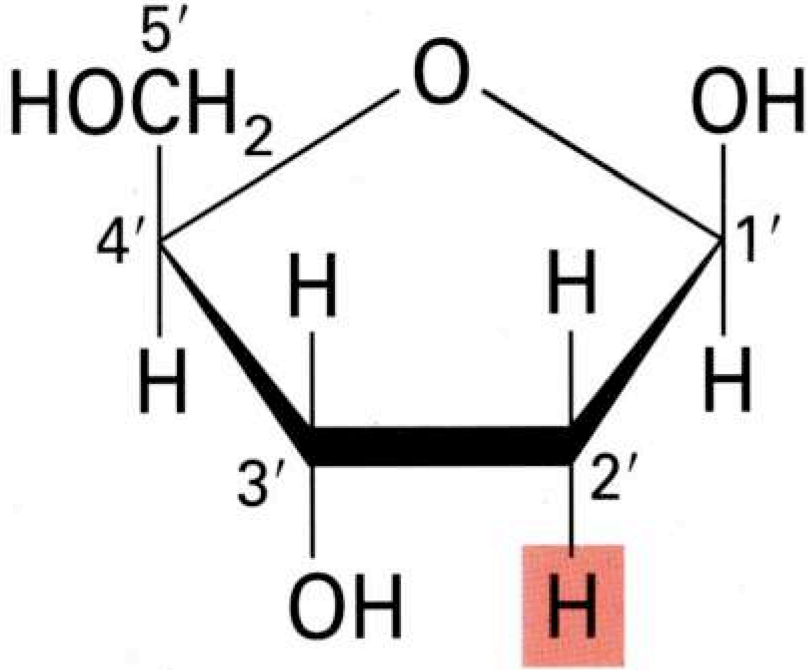
Figure 5–5. Molecular Biology of the Cell, 4th Edition.



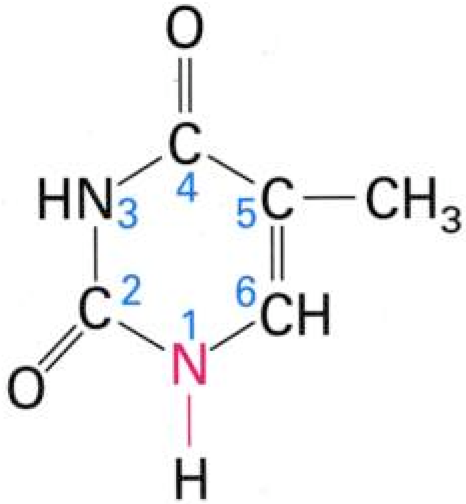
a
Phosphodiester-
bindung
NH2
N
N
5 6
7 1
H 8 9 2
4 3
N N H
Adenin (A)
O⁻
5'
⁻O–P–O–CH2
O
N-glykosidische Bindung
4'
H H
1'
O
H
H
3'
2'
HO H
Phosphat
2-Desoxyribose
Nukleosid
Nukleotid



Ribose

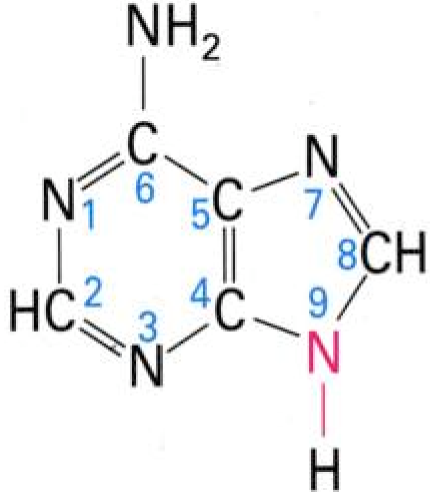


2-Deoxyribose

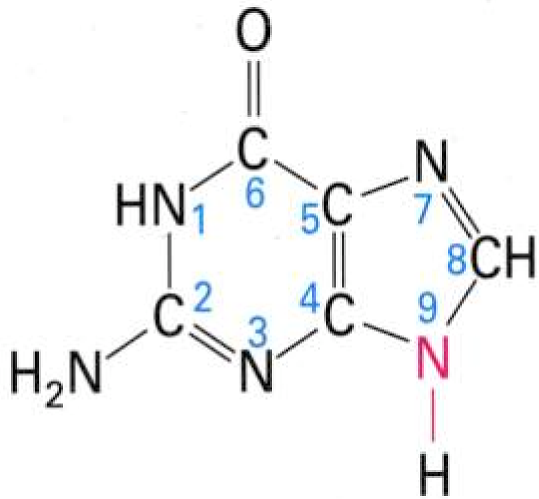


Thymine (T)

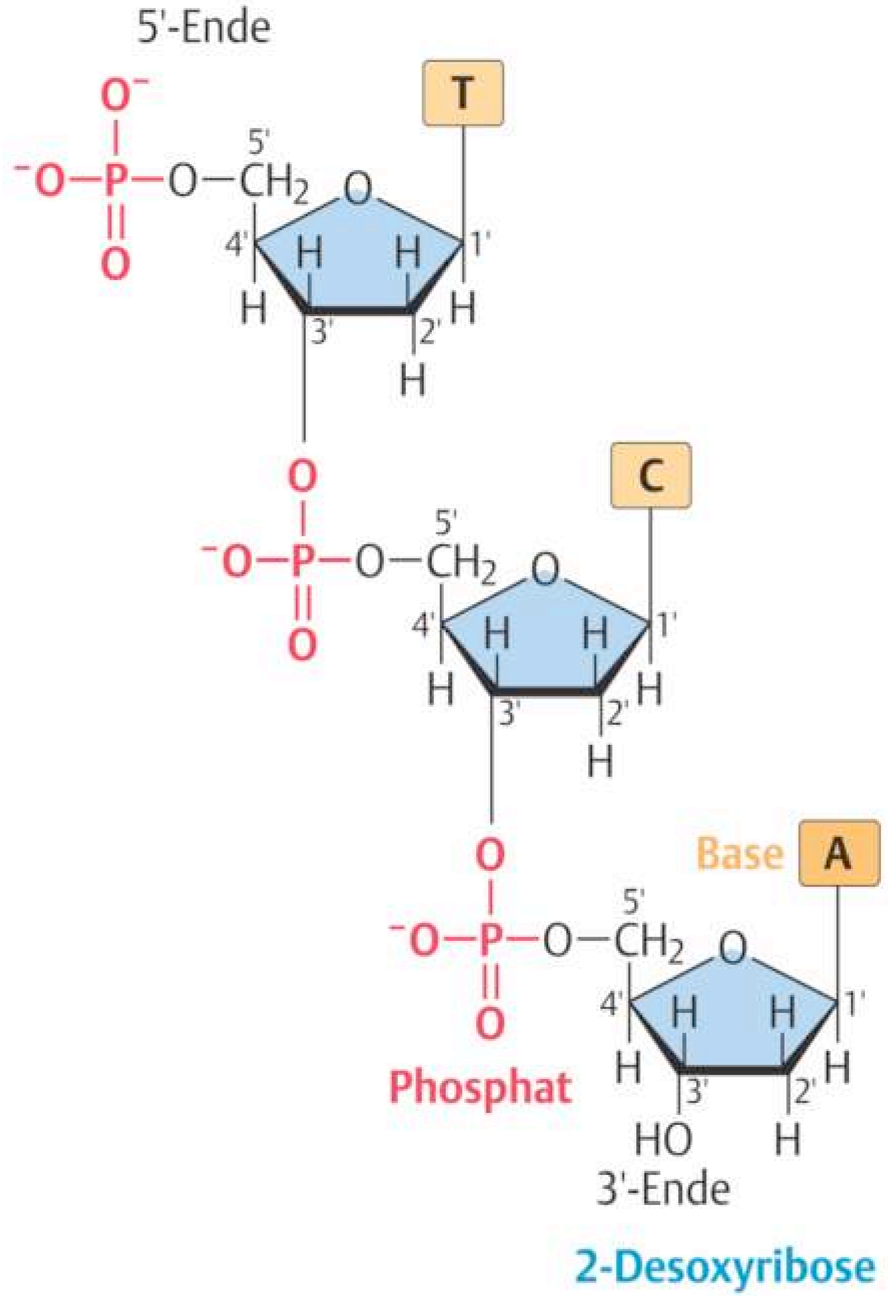
Cytosine (C)

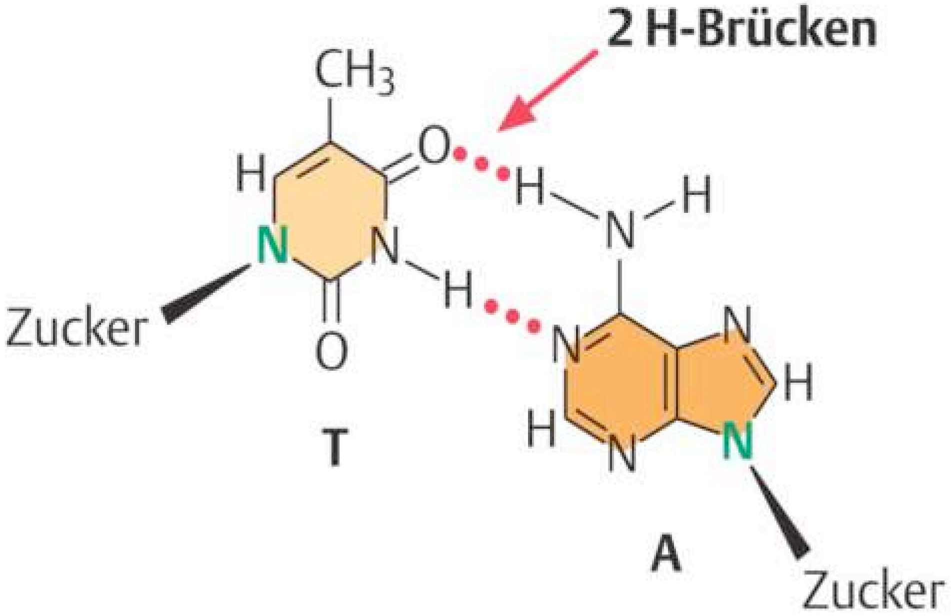


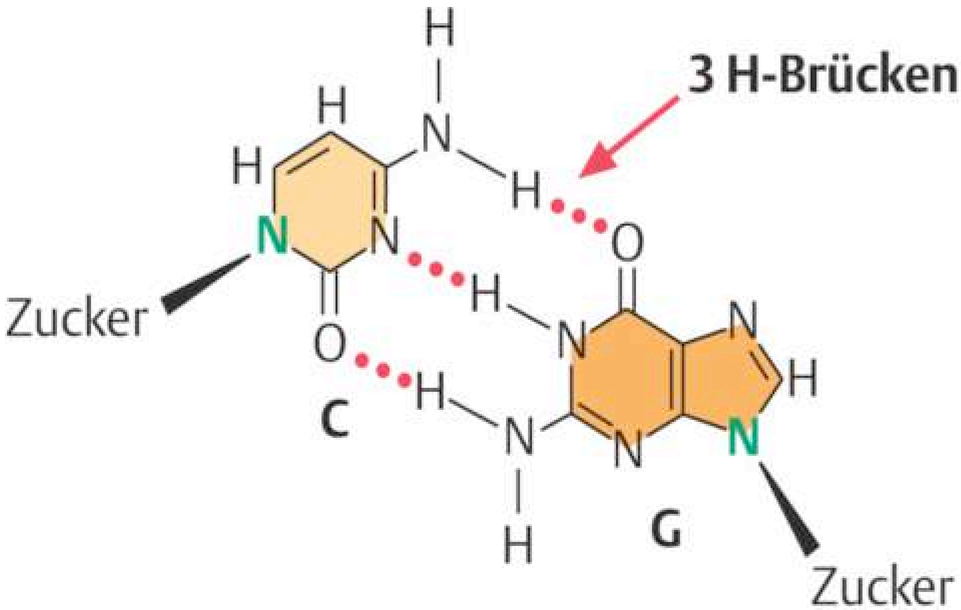
Adenine (A)

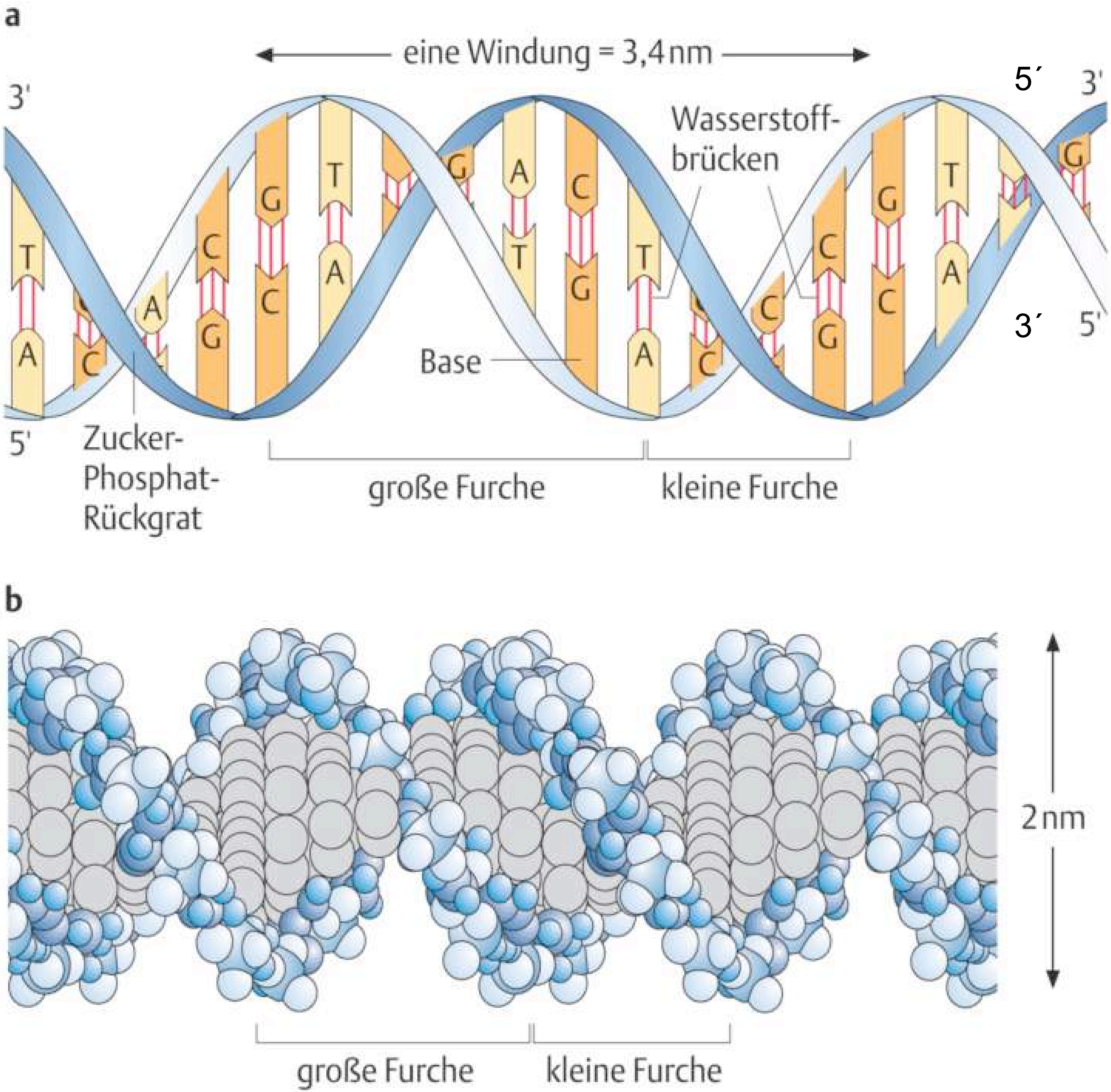


Guanine (G)

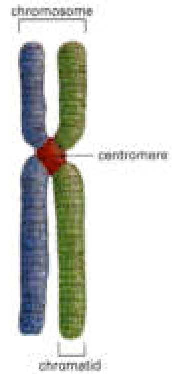

5'-Ende
T
C
Base
A
Phosphat
3'-Ende
2-Desoxyribose


2 H-Brücken
CH3
Zucker
T
A
Zucker


3 H-Brücken
Zucker
C
G
Zucker


a
eine Windung = 3,4 nm
3'
5'
5´
3'
3´
5'
Wasserstoff-
brücken
Base
Zucker-
Phosphat-
Rückgrat
große Furche
kleine Furche
b
2 nm
große Furche
kleine Furche

Genome und deren Größen



Species	Grösse (Bp)	Anz. Chrom.	
E. coli Bakterien	4.72 x 10^6	1	*
S. cerevisiae (Hefe)	14 x 10^6	16	*
C. elegans (Fadenw.)	80 x 10^6	4	*
D. melanogaster	165 x 10^6	4	*
X. laevis	3 x 10^9 (!)	18	
M. musculus	3 x 10^9 (!)	20	*
H. sapiens	3 x 10^9 (!)	23	*
A. cepa (Zwiebel)	15 x 10^9 (!)	8	

Die Chromosomen im Zellzyklus

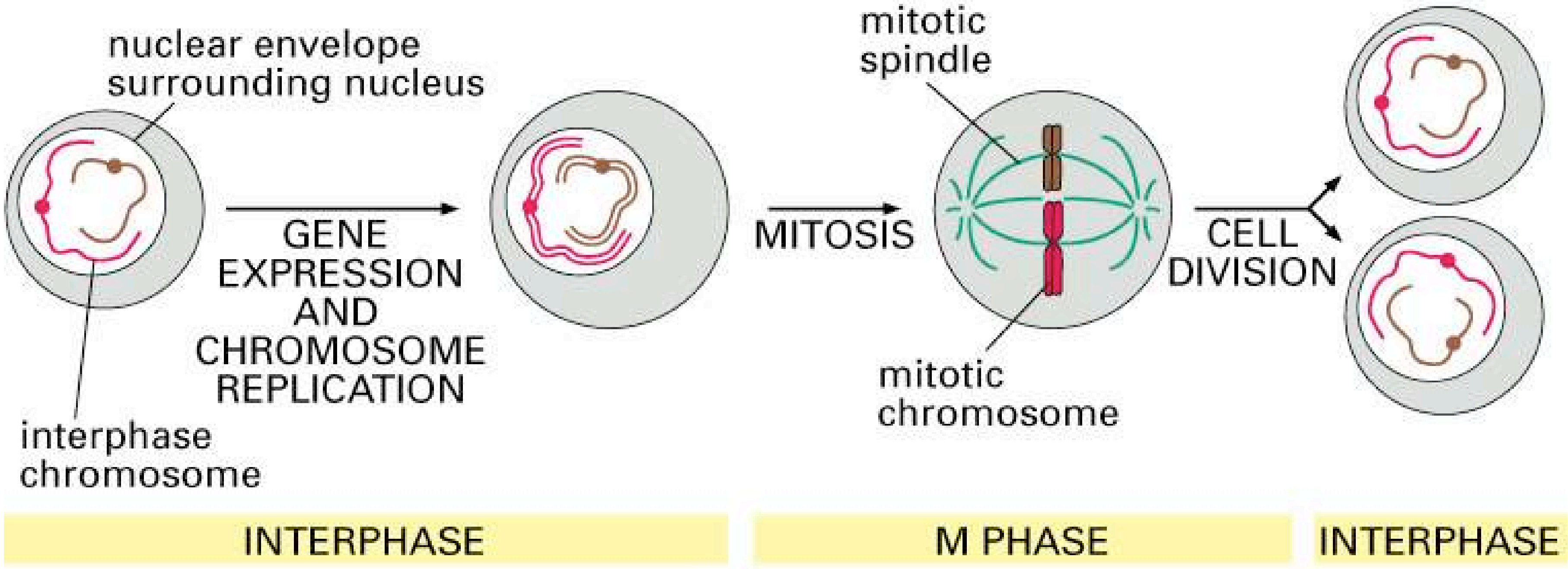
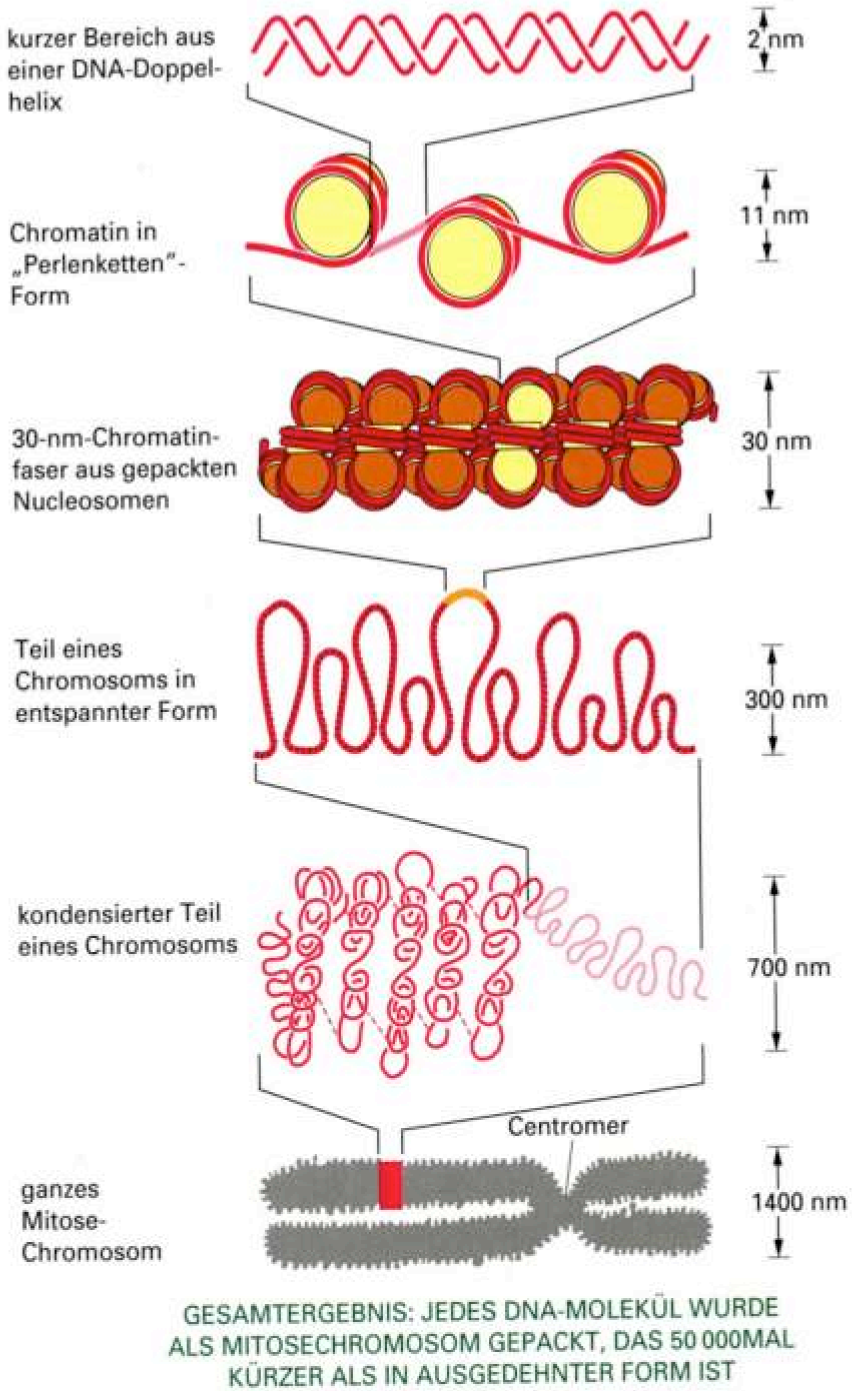


Figure 4–20. Molecular Biology of the Cell, 4th Edition.



proteinfreie DNA

nukleosomale DNA

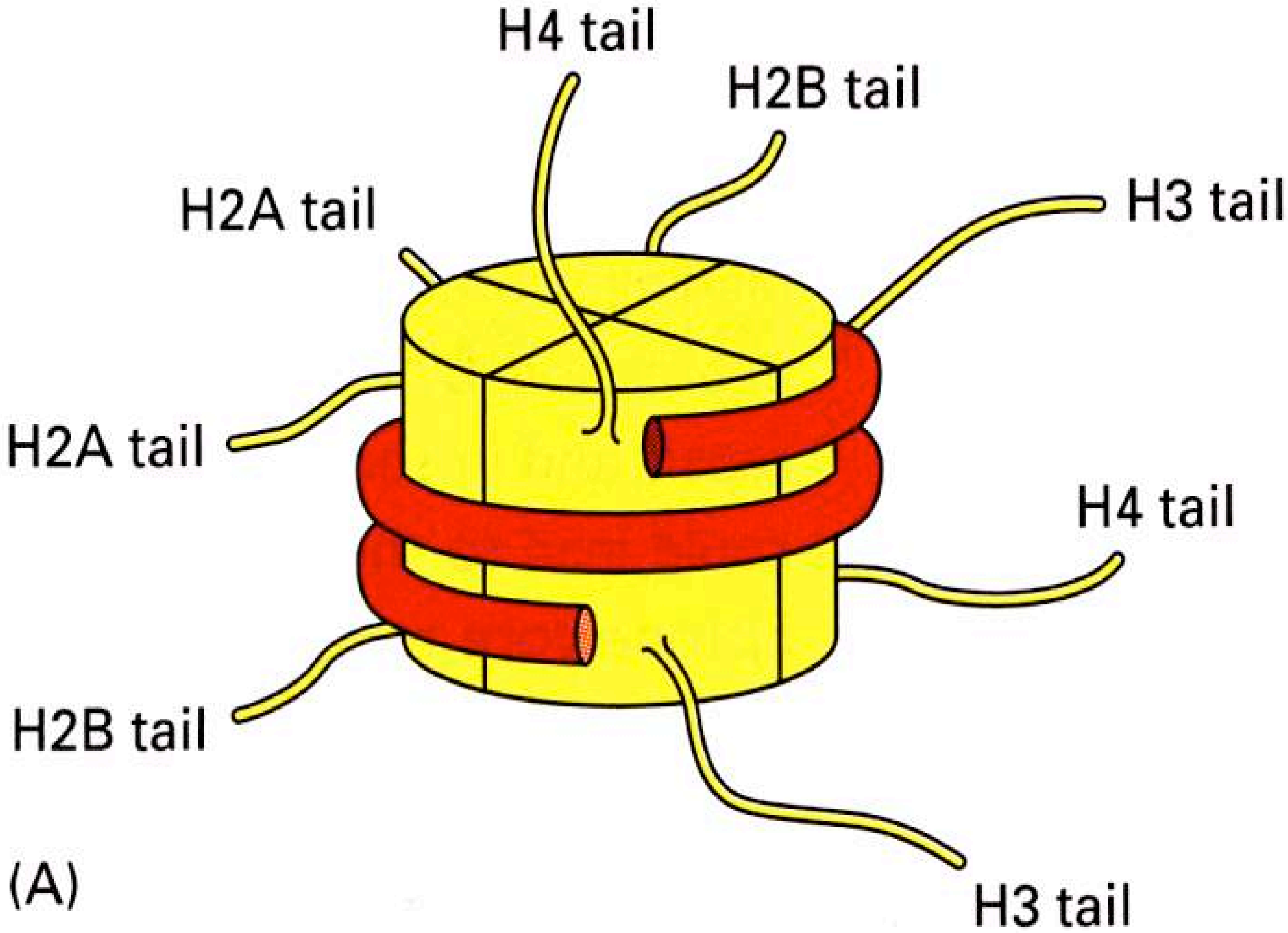
Superhelix nukleosomaler DNA („Solenoid")

chromosomale Schleifen

kondensierte Schleifen

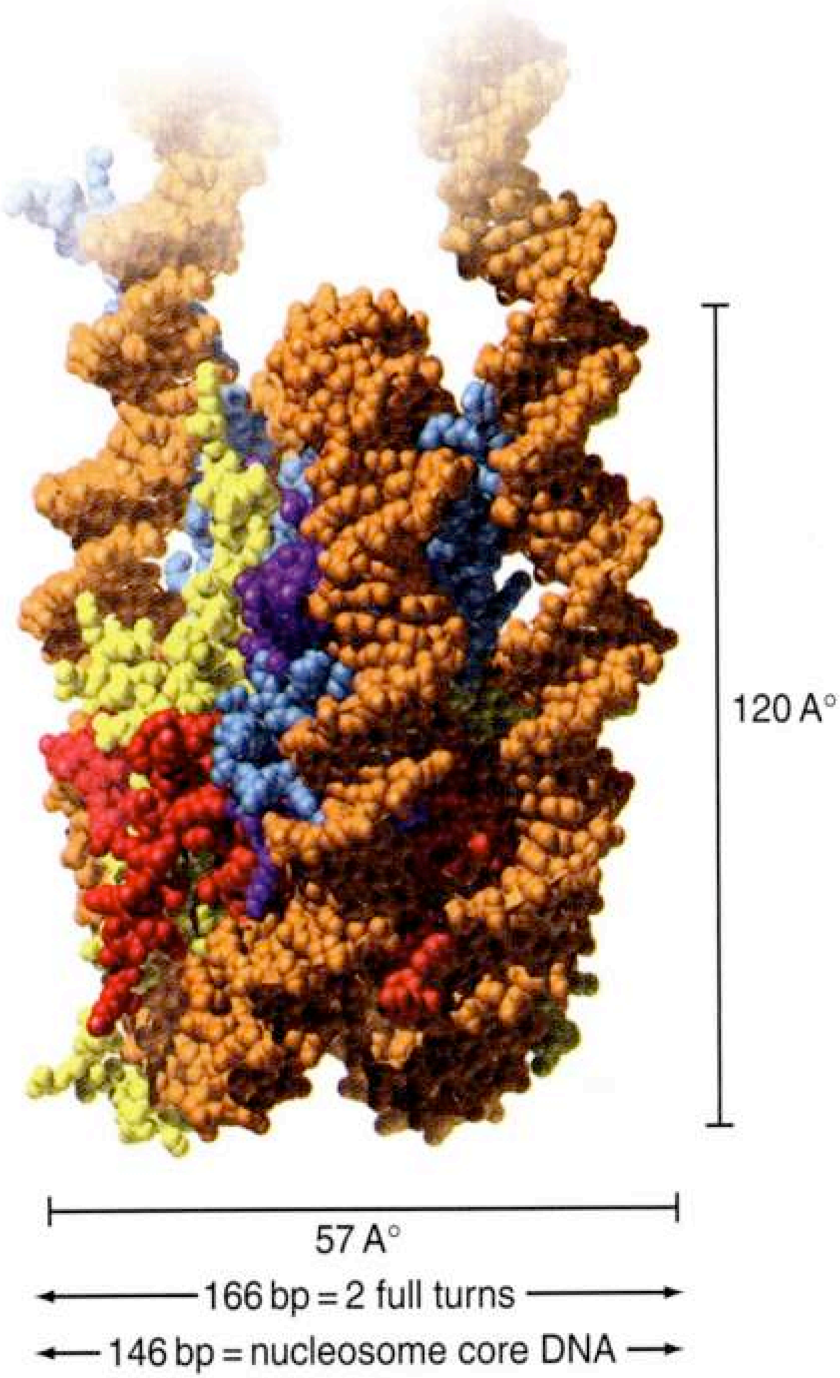
Metaphase Chromatide

Das Nukleosom



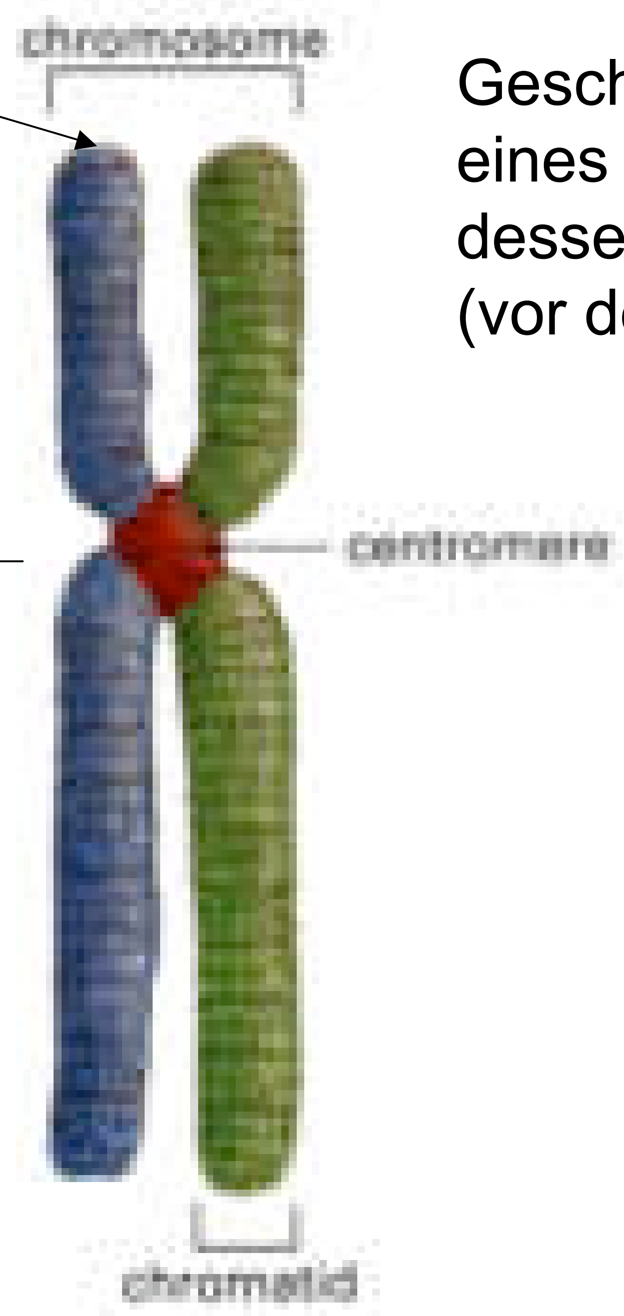
Die Kristallstruktur des Nukleosoms

(Roentgen-Strahlung Beugungsdaten)



Telomer

p-Arm



q-Arm

Geschwister-Chromatide eines Chromosomes nach dessen Replikation (vor der Zytokinese)

Der humane Chromosomensatz

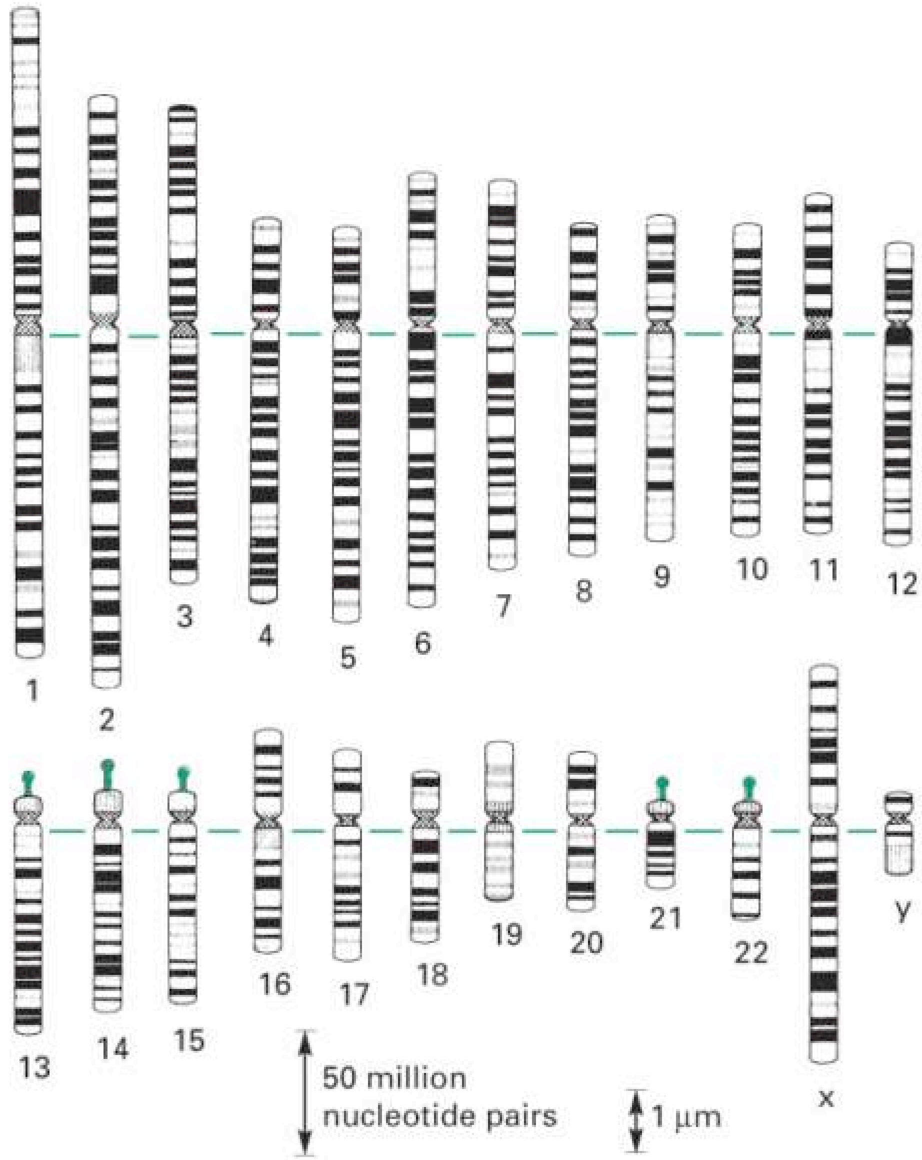


Figure 4–11. Molecular Biology of the Cell, 4th Edition.

Chromatidentrennung in der Anaphase

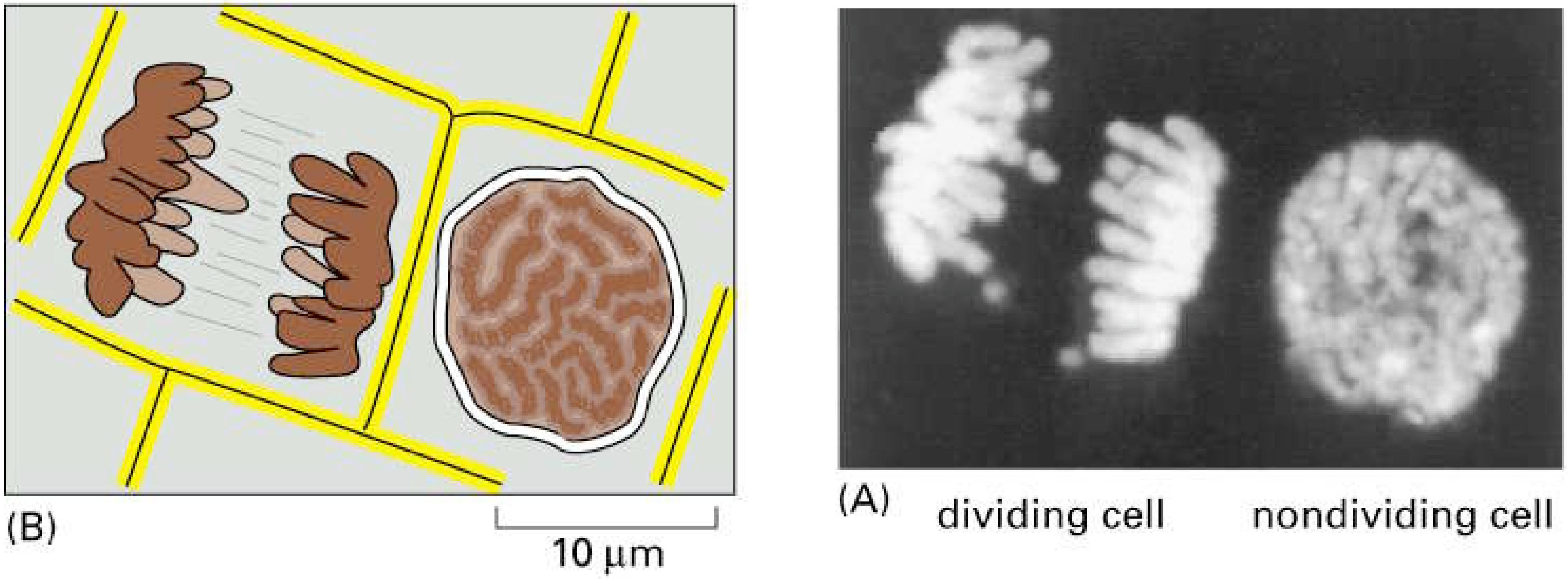


Figure 4–1. Molecular Biology of the Cell, 4th Edition.

Die Replikation der Chromosomen im Zellzyklus

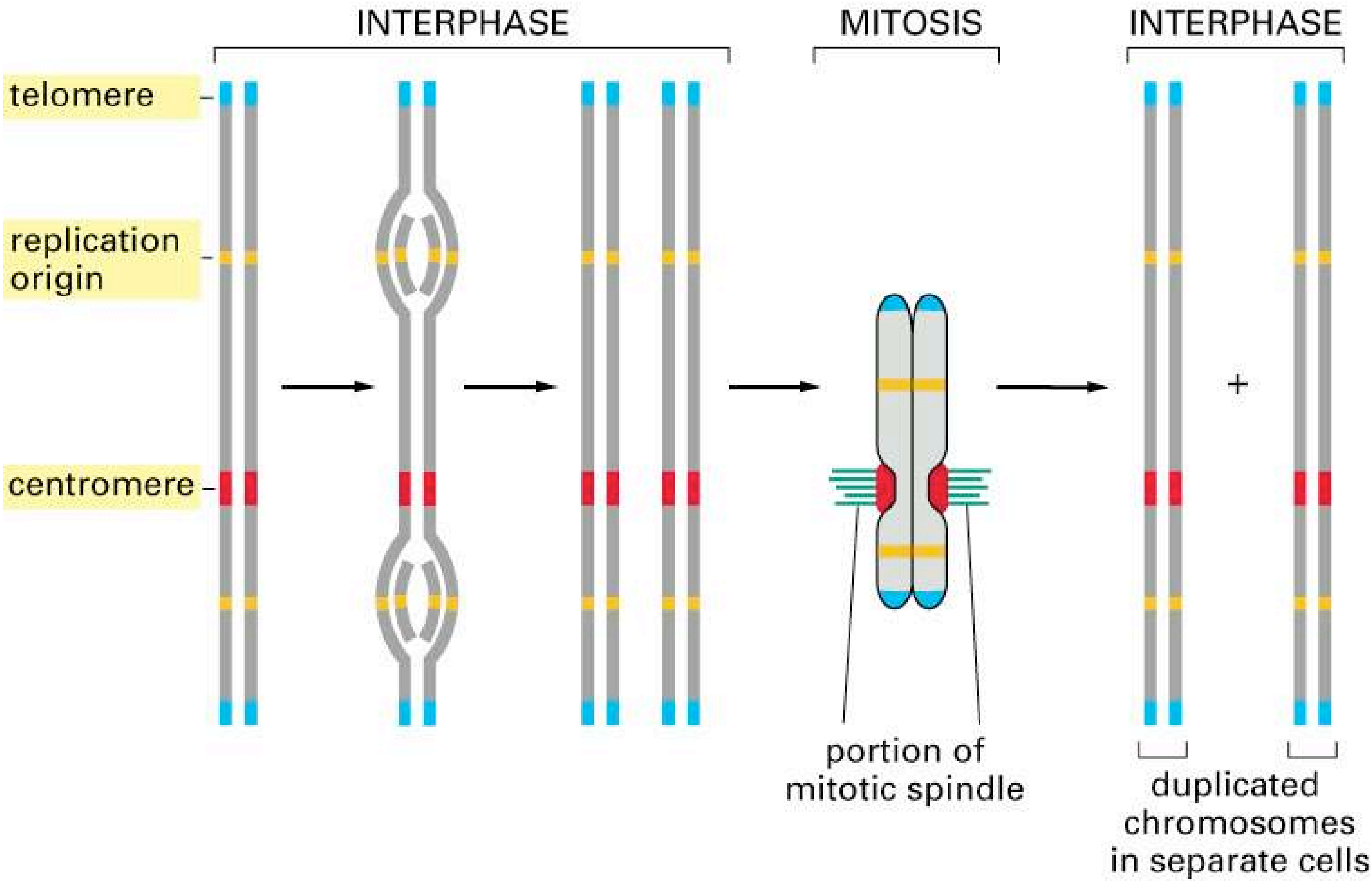


Figure 4–22. Molecular Biology of the Cell, 4th Edition.

Die Verdoppelung der DNA-Doppelhelix

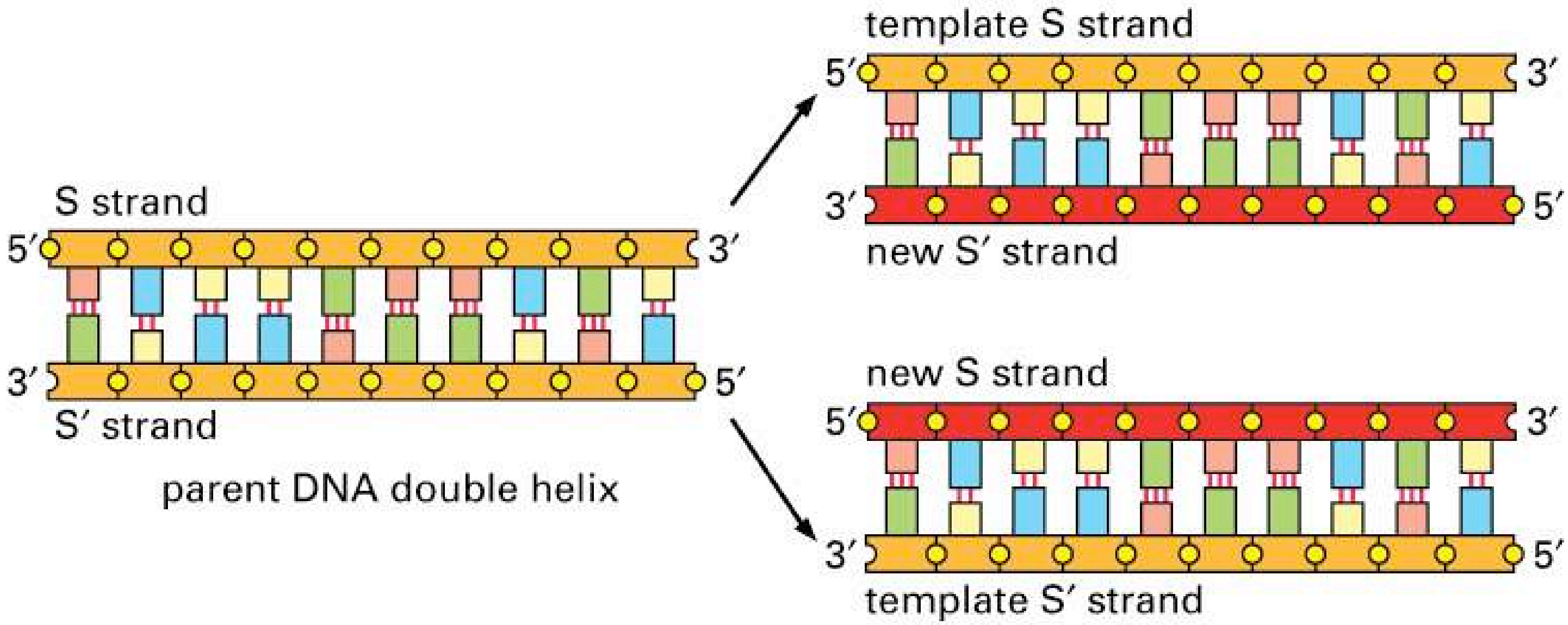


Figure 4–8. Molecular Biology of the Cell, 4th Edition.

Richtung der DNA-Polymerisierung

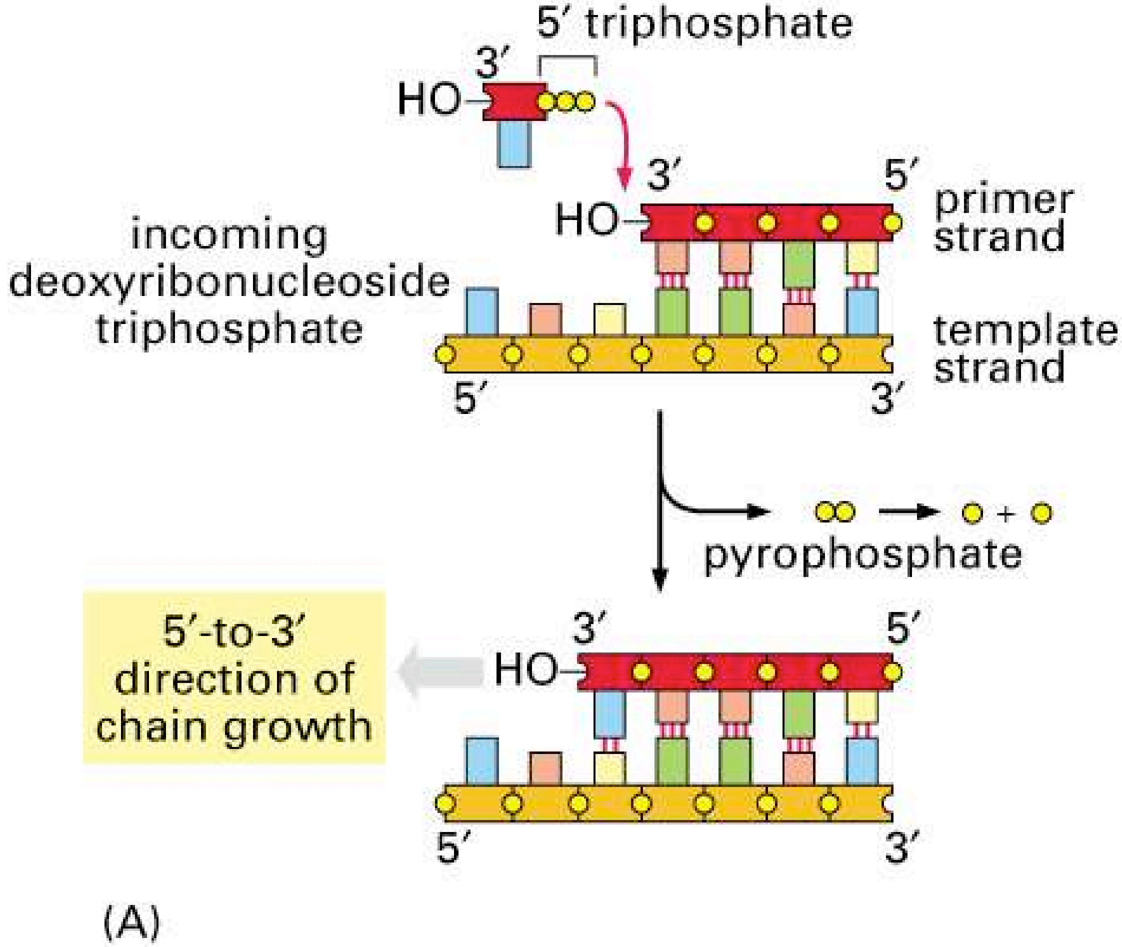


Figure 4–5 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

DNA-Synthese

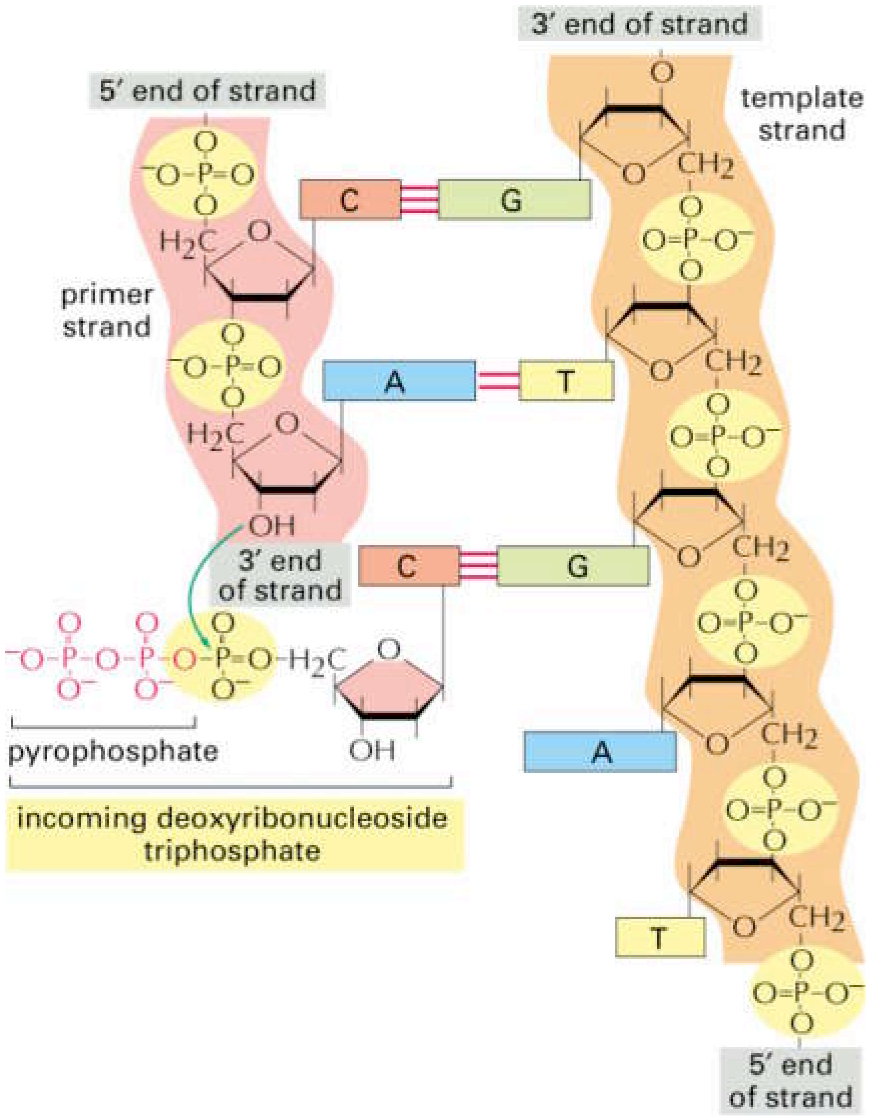


Figure 5–3. Molecular Biology of the Cell, 4th Edition.

Modell der enzymatischen DNA-Polymerisierung

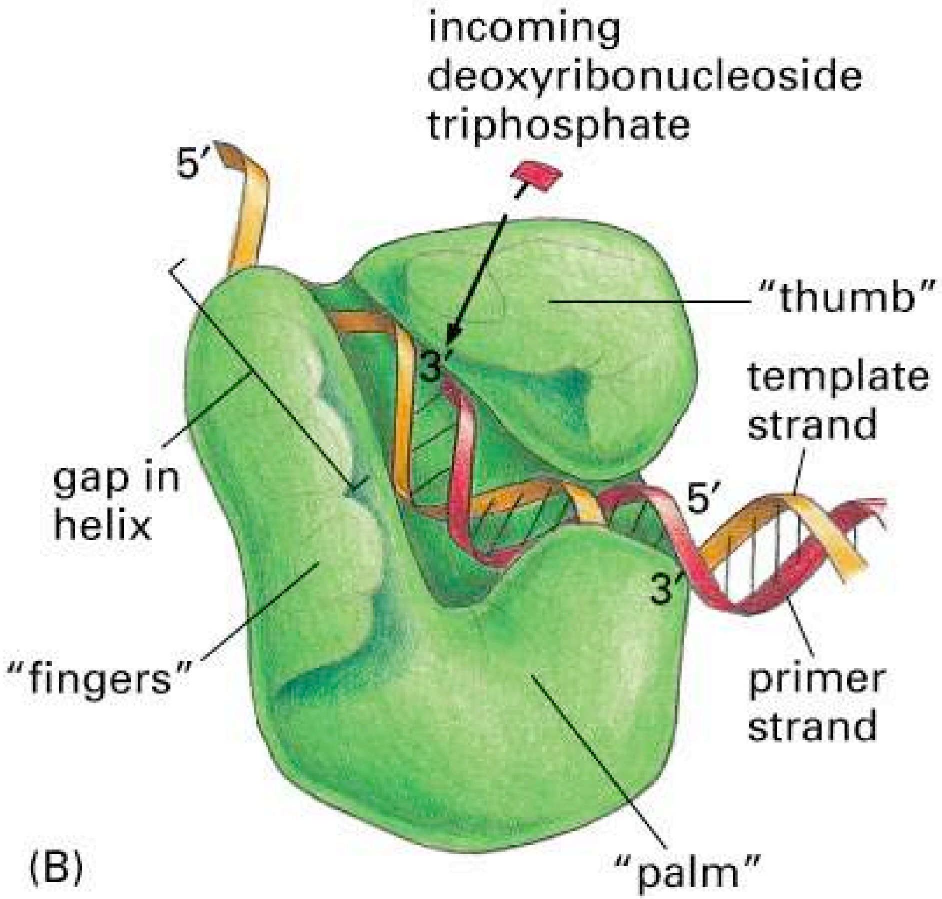


Figure 5–4 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

“Replikationsblase”

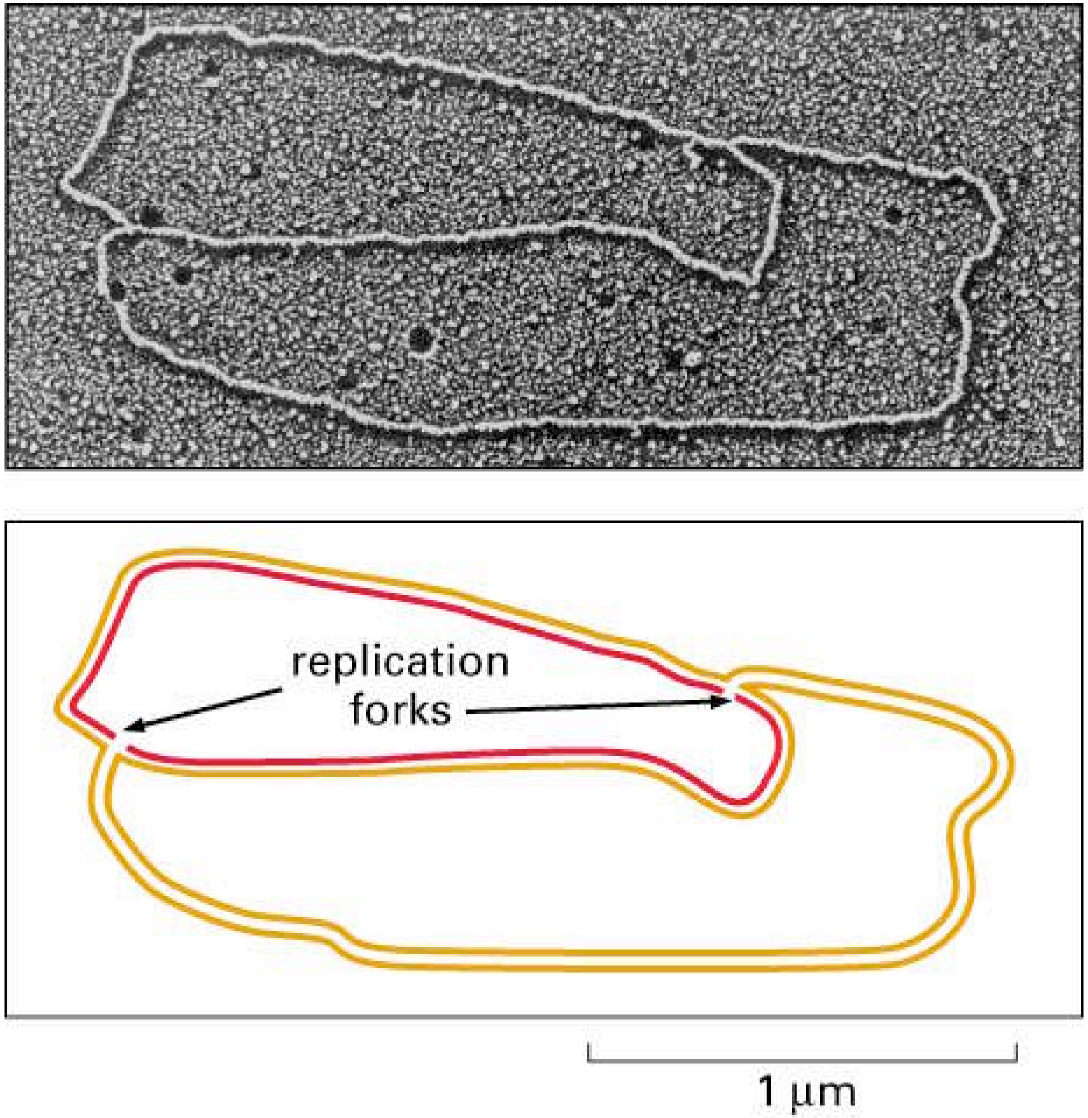


Figure 5–6. Molecular Biology of the Cell, 4th Edition.

Initiation der Replikation

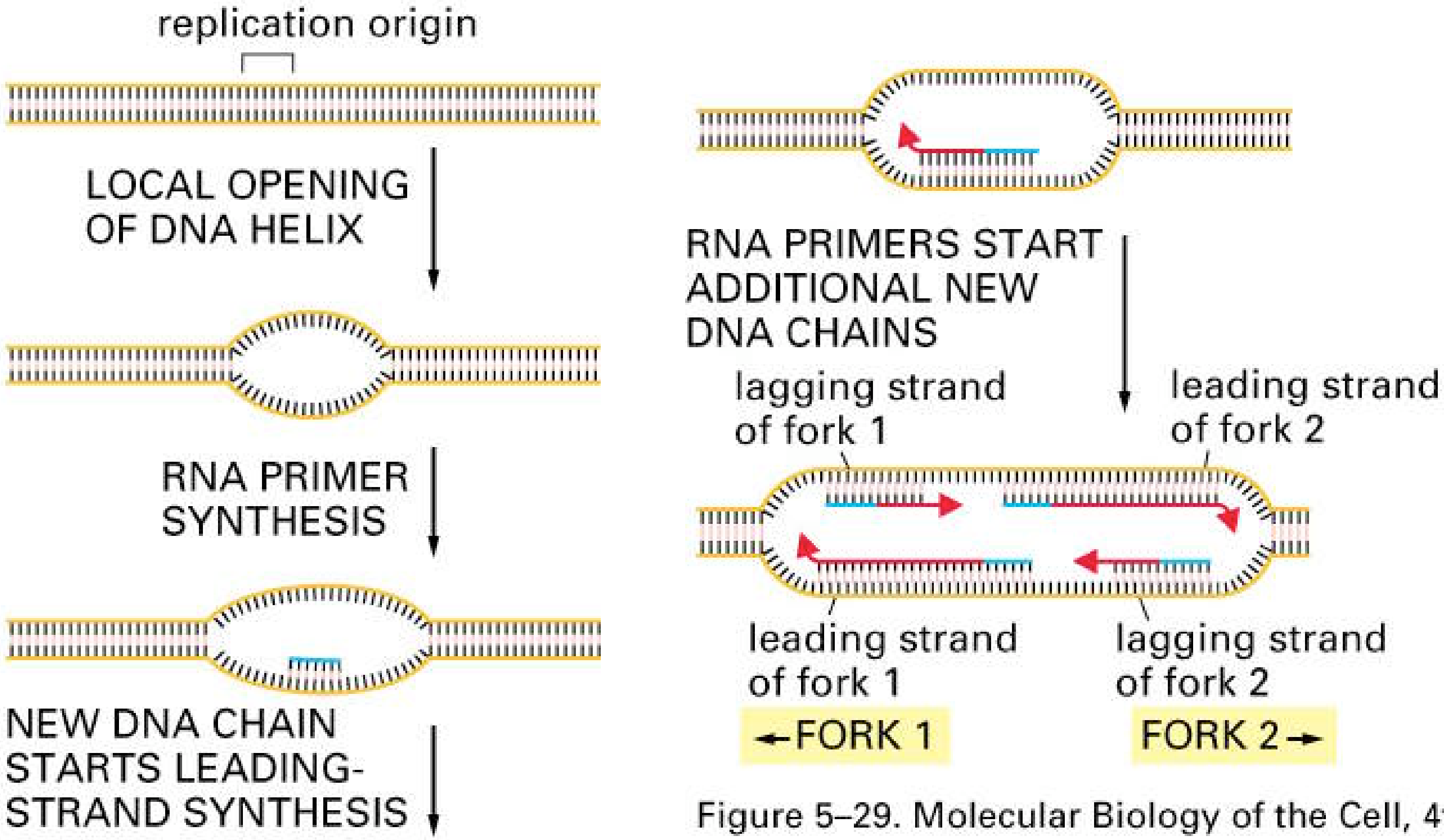


Figure 5–29. Molecular Biology of the Cell, 4th Edition.

Die Replikationsgabel

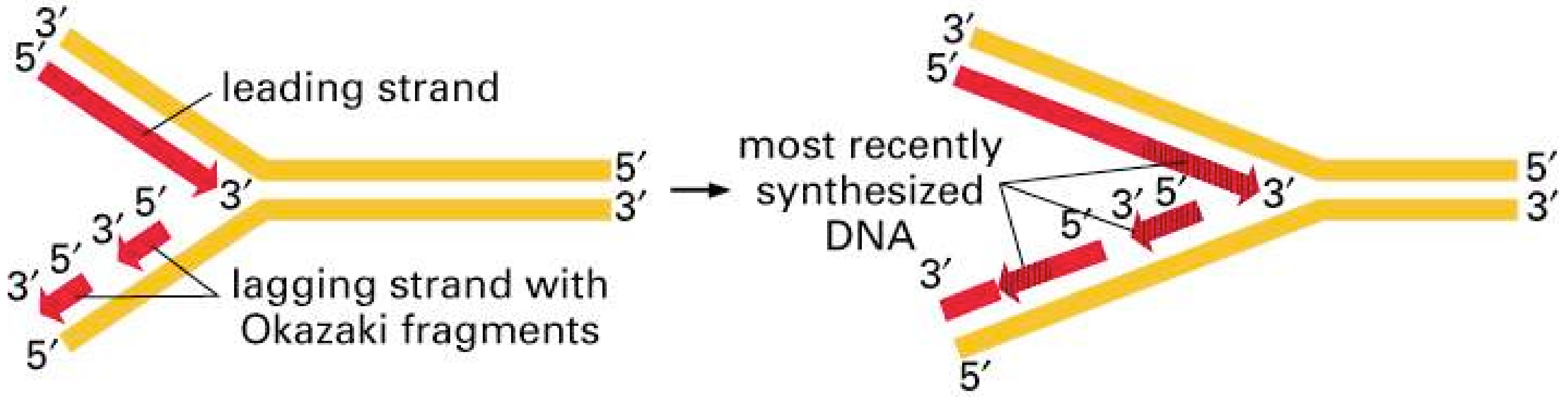
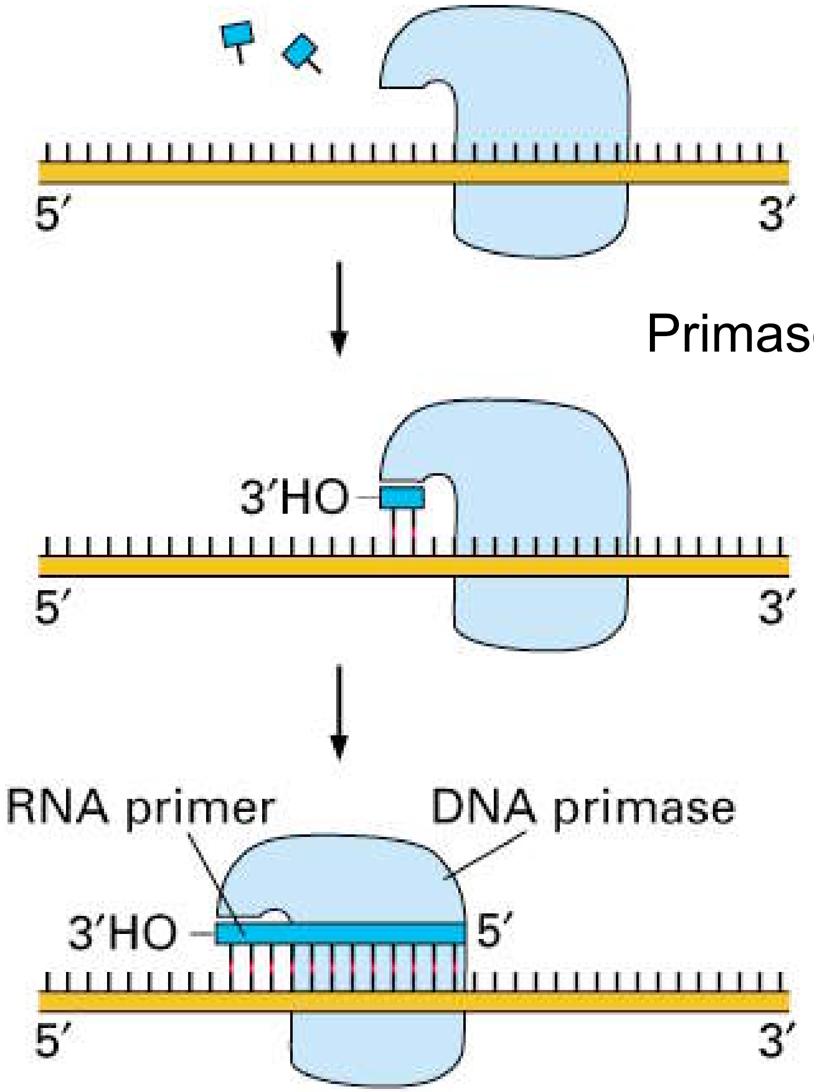


Figure 5–8. Molecular Biology of the Cell, 4th Edition.

RNA-Primer-Synthese



Primase ist eine RNA-Polymerase.

Figure 5–12. Molecular Biology of the Cell, 4th Edition.

DNA-Polymerisierung am "lagging" Strang

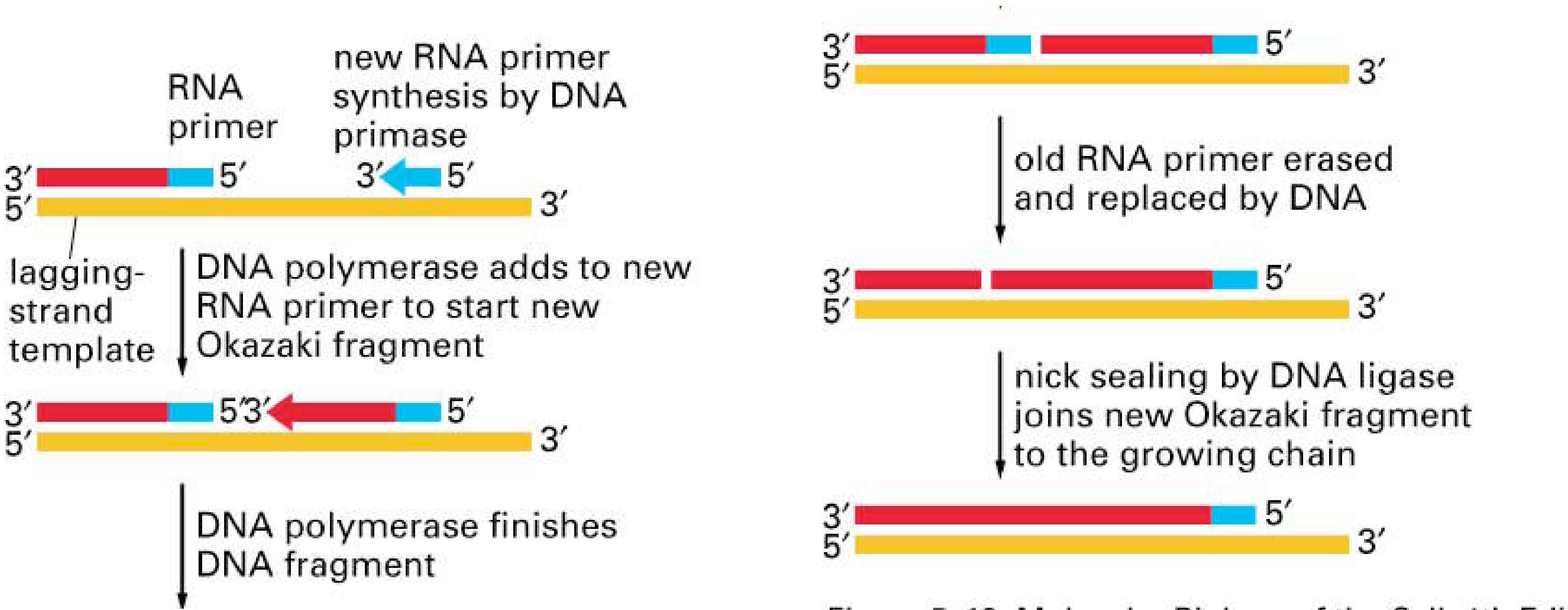


Figure 5–13. Molecular Biology of the Cell, 4th Edition.

Die Struktur der Telomerase

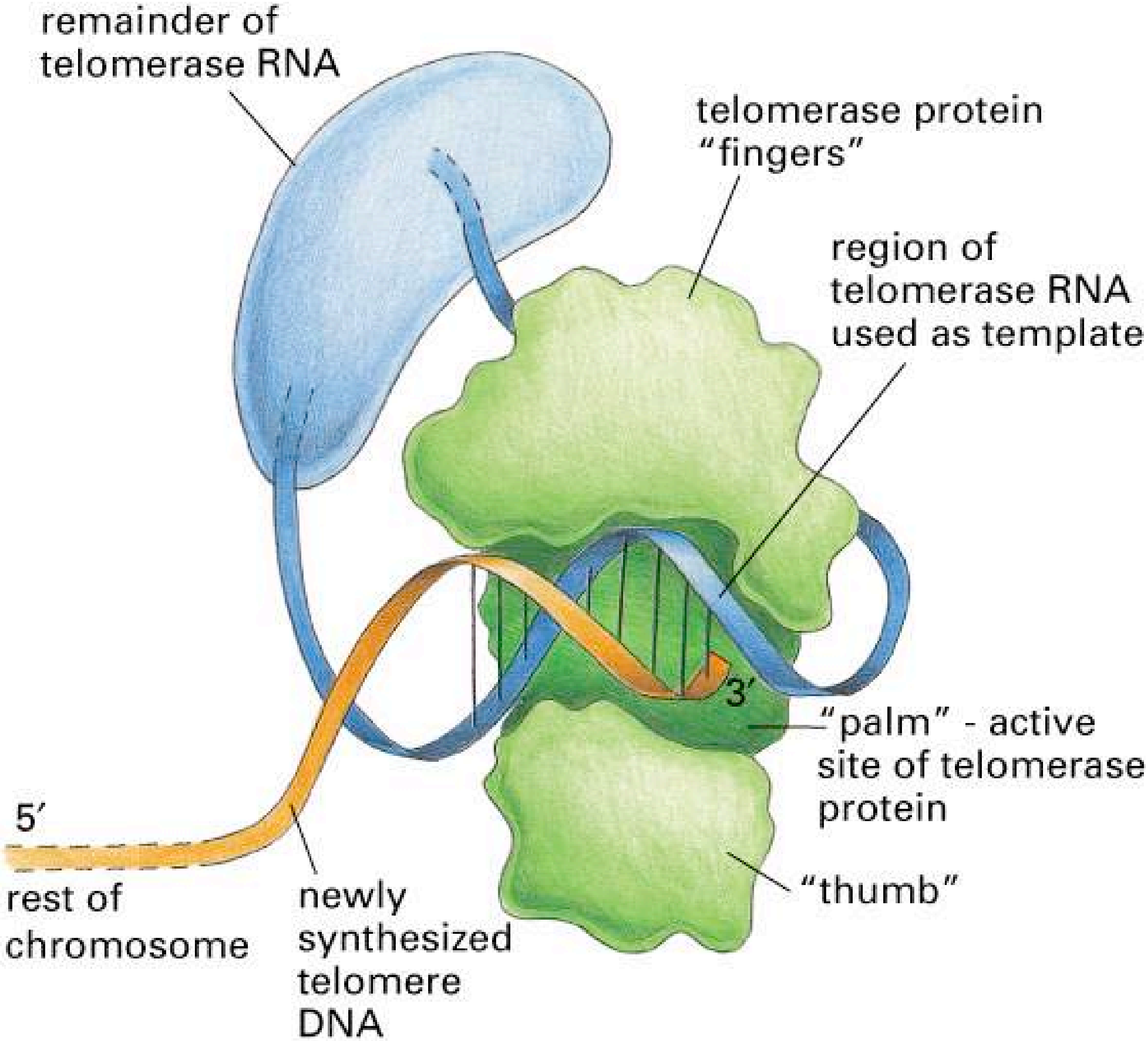


Figure 5–42. Molecular Biology of the Cell, 4th Edition.

Die Replikation der Telomere

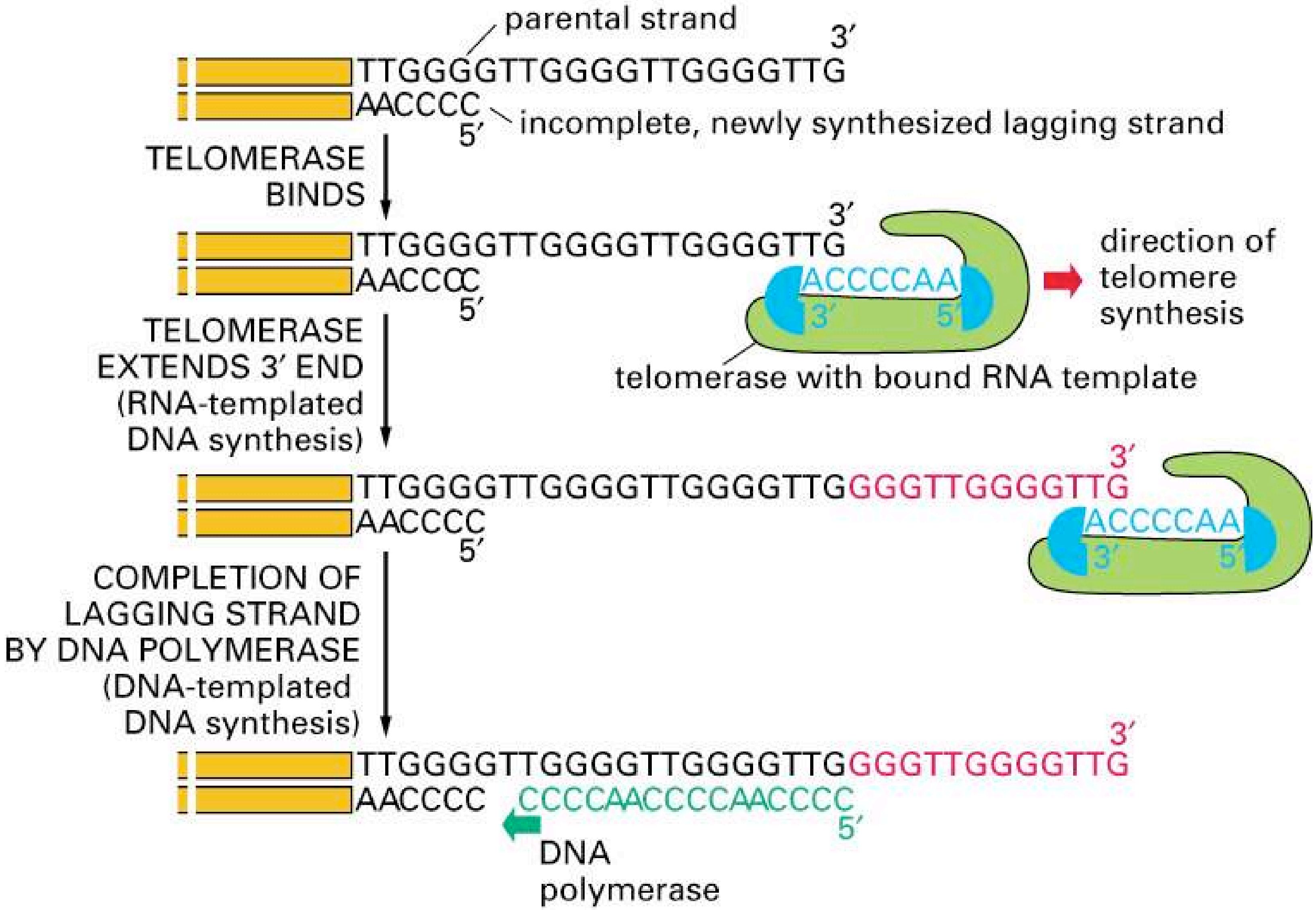


Figure 5–43. Molecular Biology of the Cell, 4th Edition.

Warum DNA-Reparatur?

- Genauigkeit der DNA-Polymerisierung: 10^5
- Proofreading: 10^2
- Strang-gerichtete Mismatch-Reparatur: 10^2
- Gesamtgenauigkeit der Replikation: 10^9
- Humanes Genom: $3x10^9$ bp
- 5000 Depurinierungen pro Tag und humaner Zelle
- 100 Desaminierungen pro Tag und humaner Zelle
- UV-Strahlung usw. usf.

“Proofreading” der DNA-Polymerase

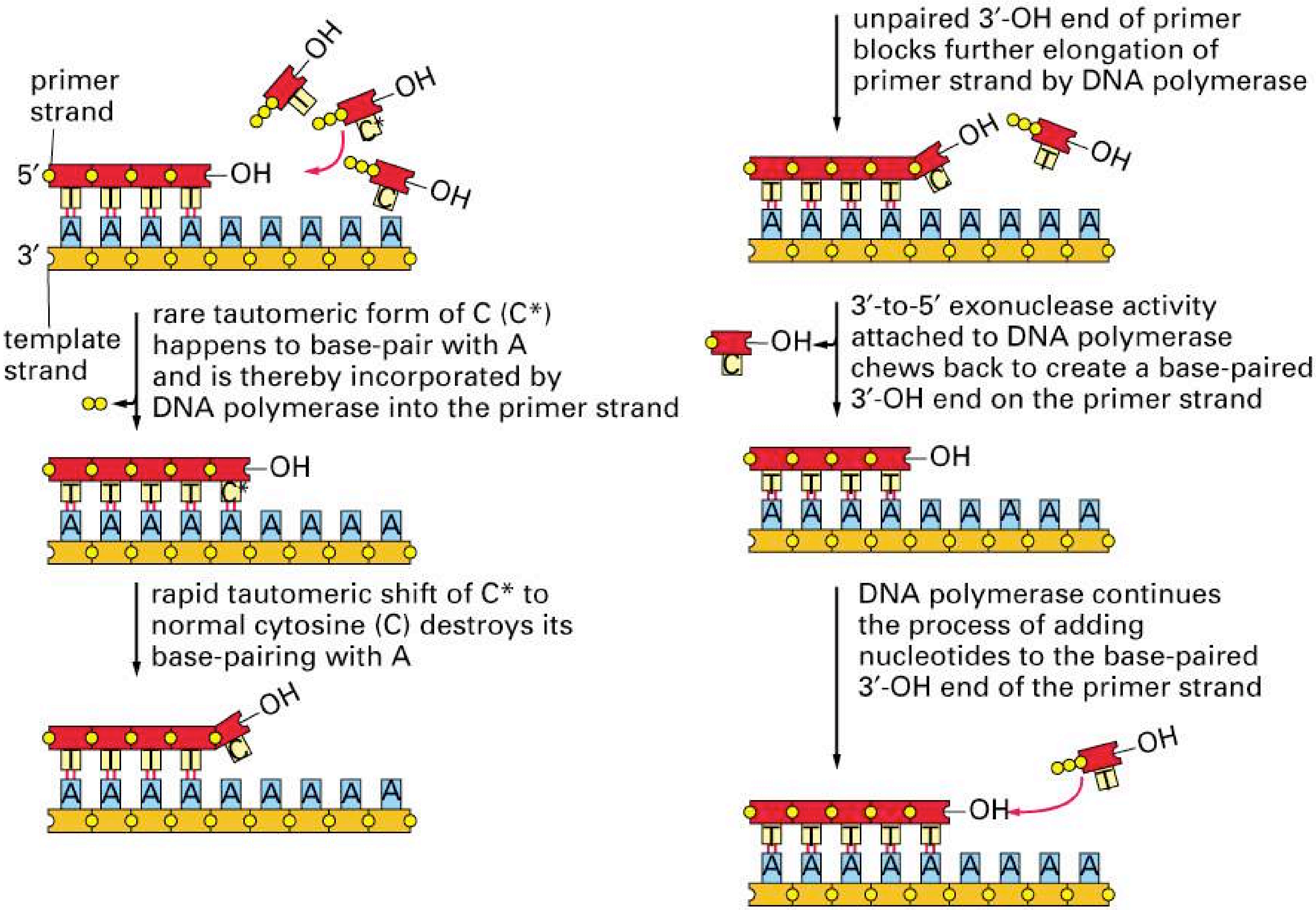


Figure 5–9 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Modell des Proofreadings

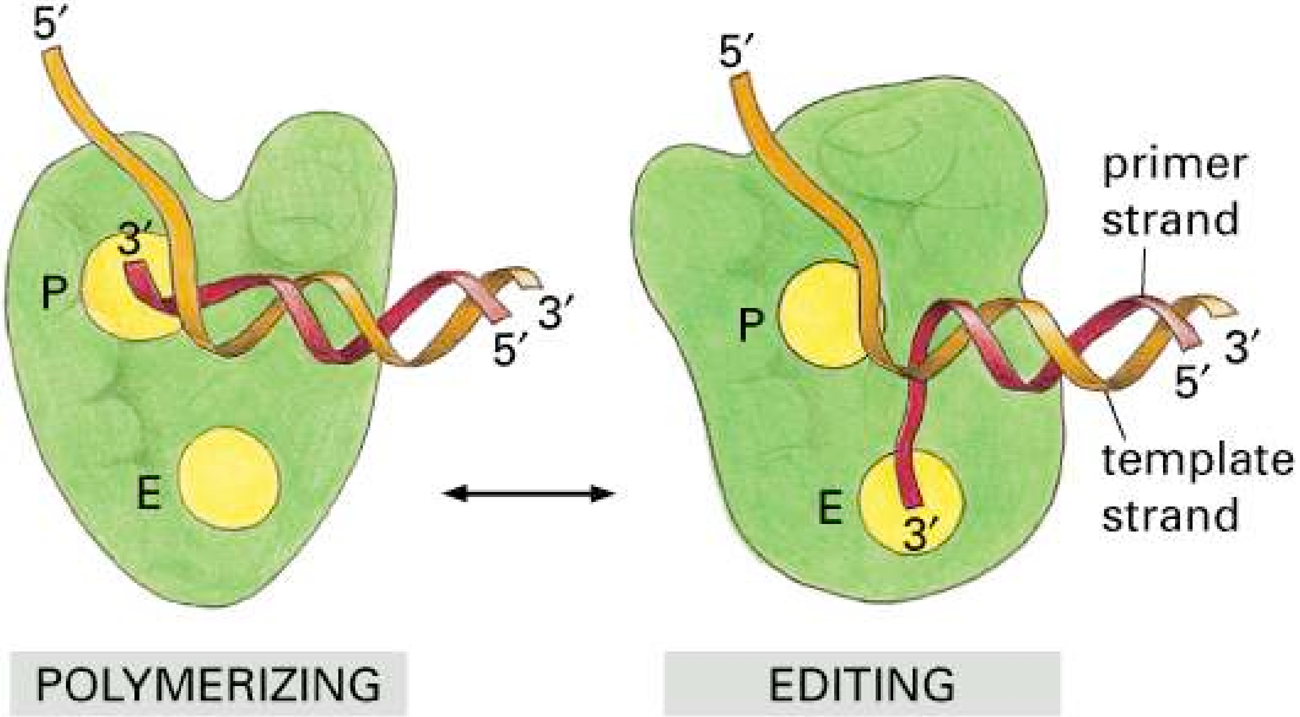


Figure 5–10. Molecular Biology of the Cell, 4th Edition.

Mögliche Angriffspunkte für DNA-schädigende Substanzen

Oxidation Hydrolyse Methylierung

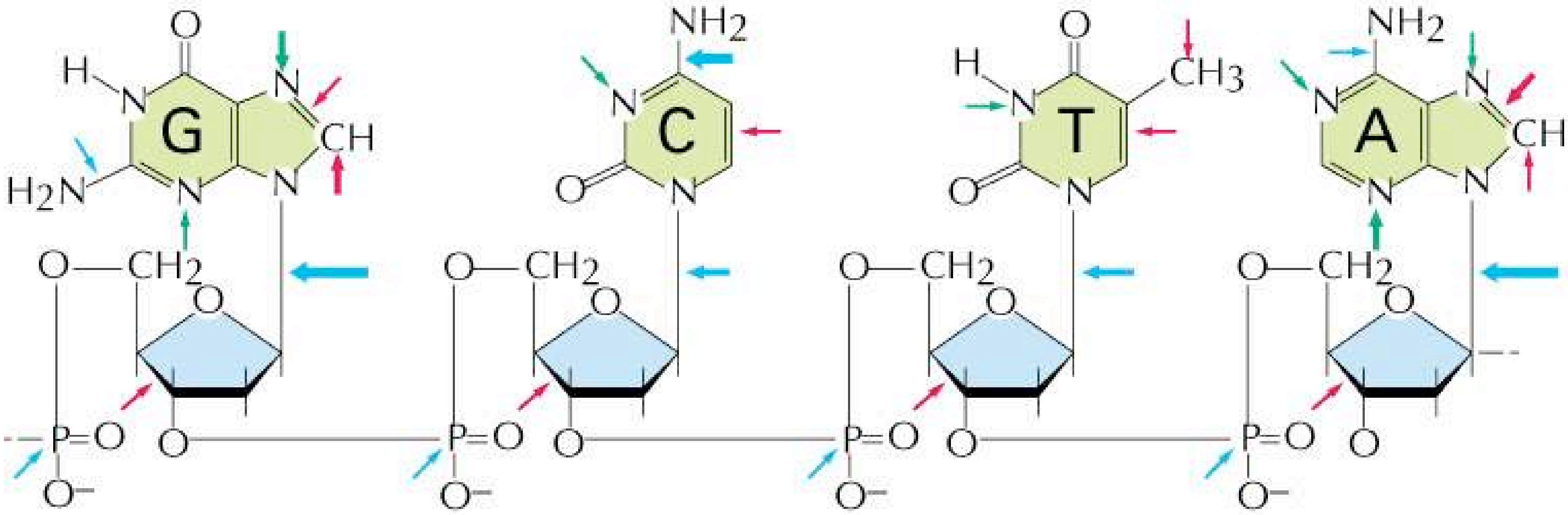


Figure 5–46. Molecular Biology of the Cell, 4th Edition.

Depurinierung und Desaminierung

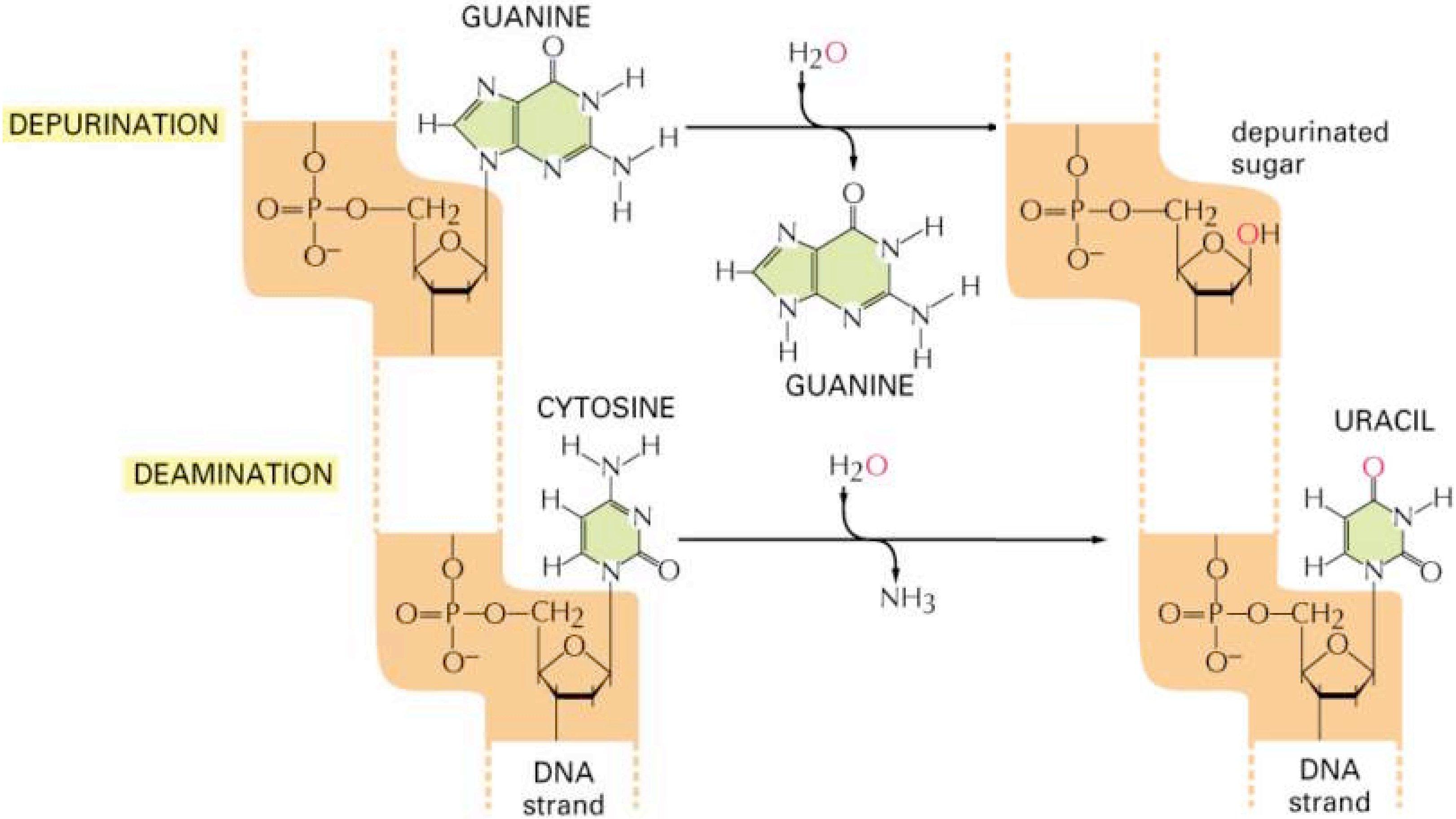
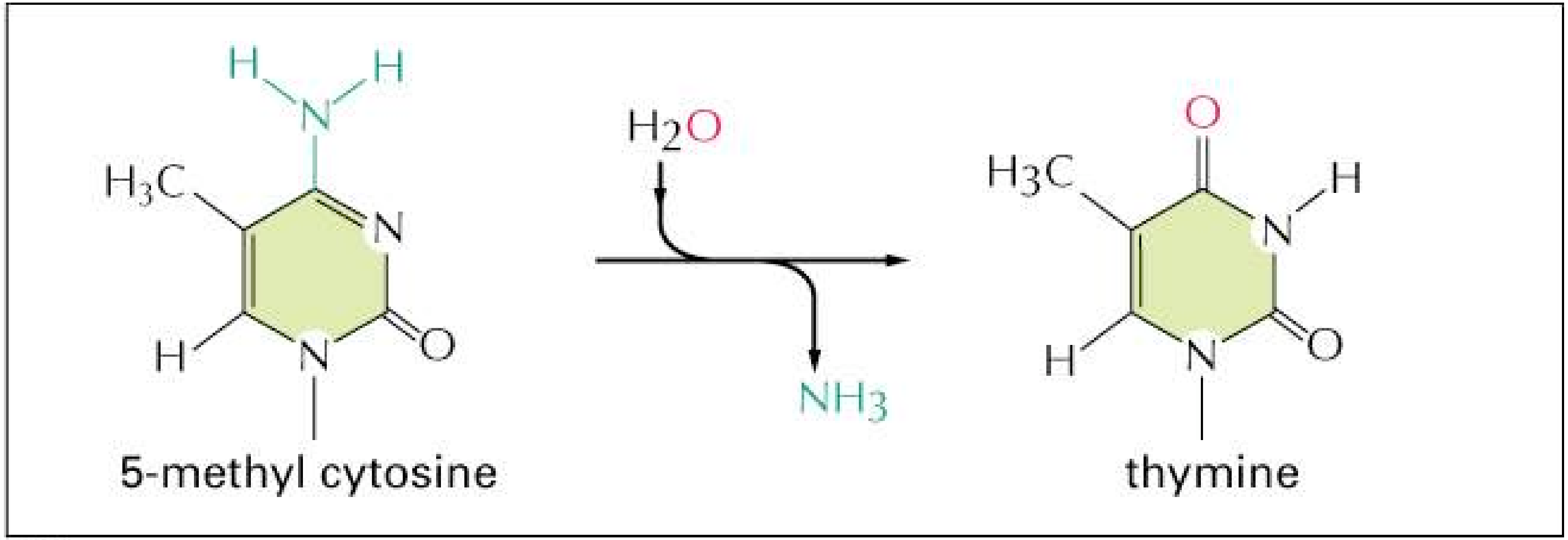


Figure 5–47 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

Desaminierung von 5-Methylcytosin



(B)

Figure 5–52 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Mutagenese durch Basenmodifizierung

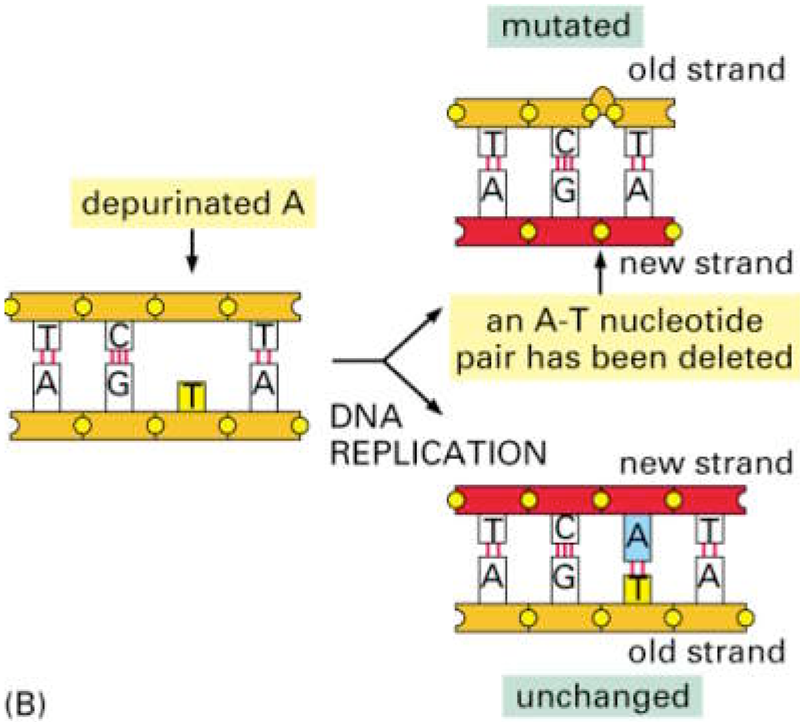


Figure 5–49 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

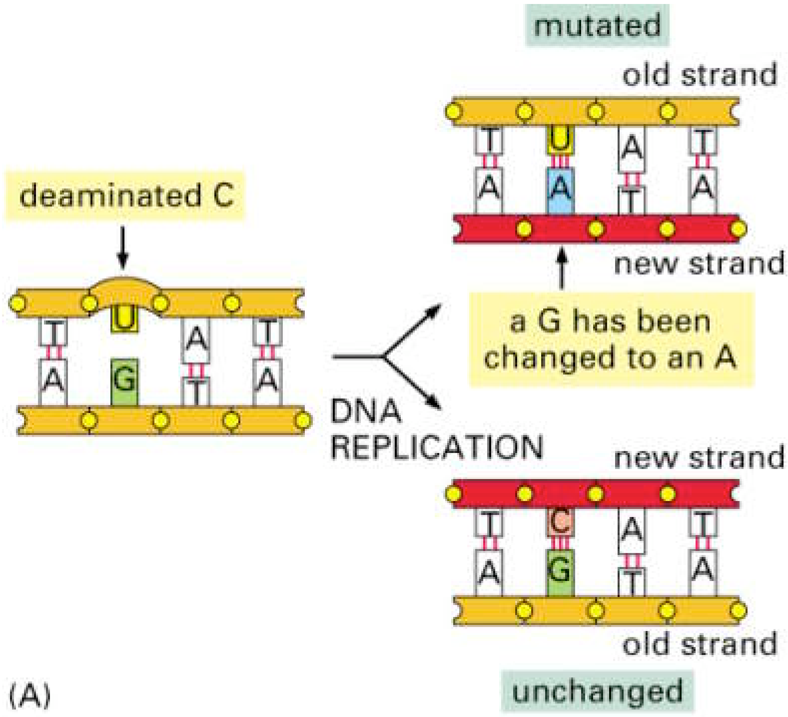


Figure 5–49 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

Thymin-Dimer

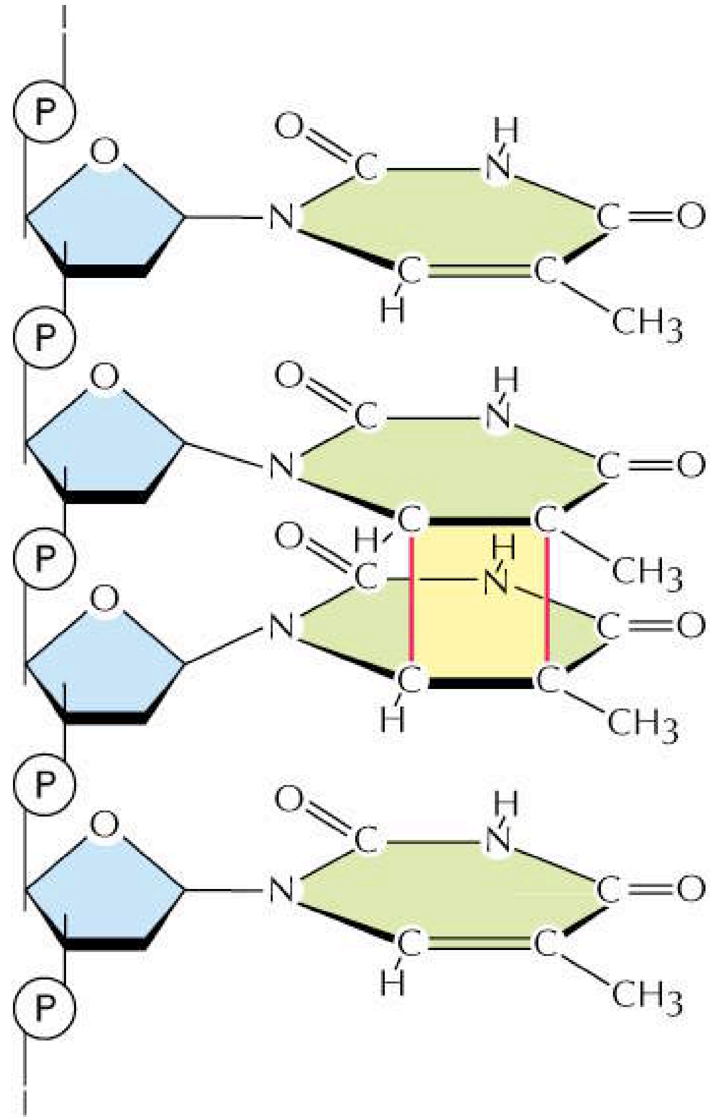


Figure 5–48. Molecular Biology of the Cell, 4th Edition.

DNA-Reparatur

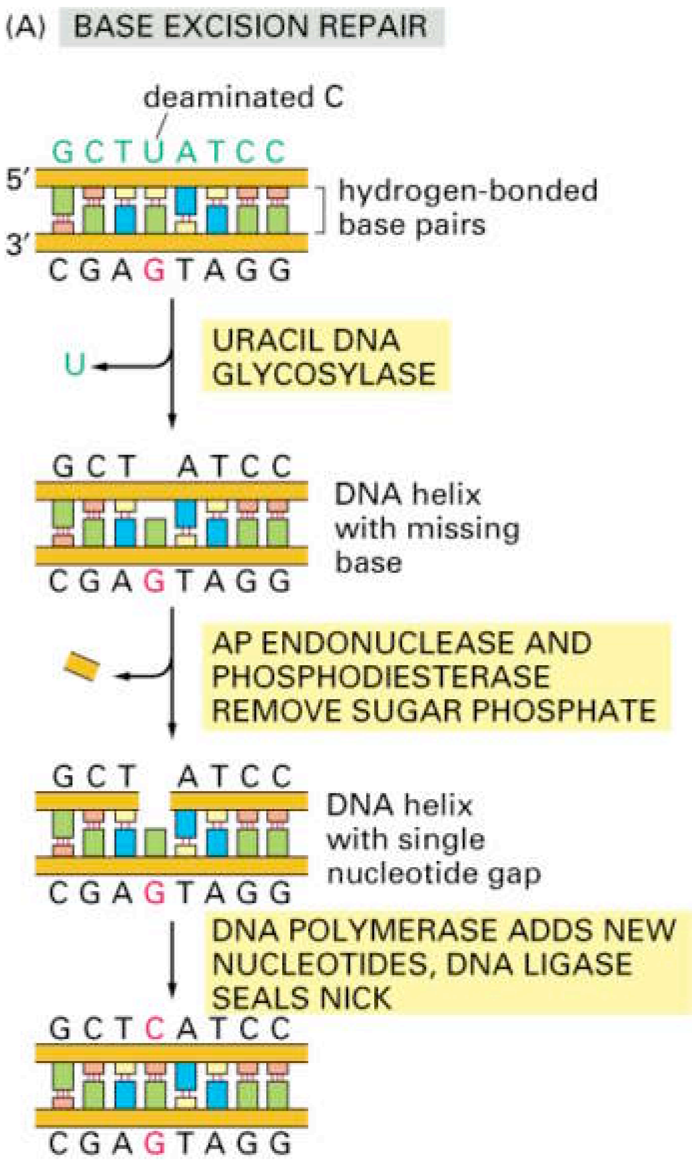


Figure 5–50 part 1 of 2. Molecular Biology of the Cell, 4th Edition.

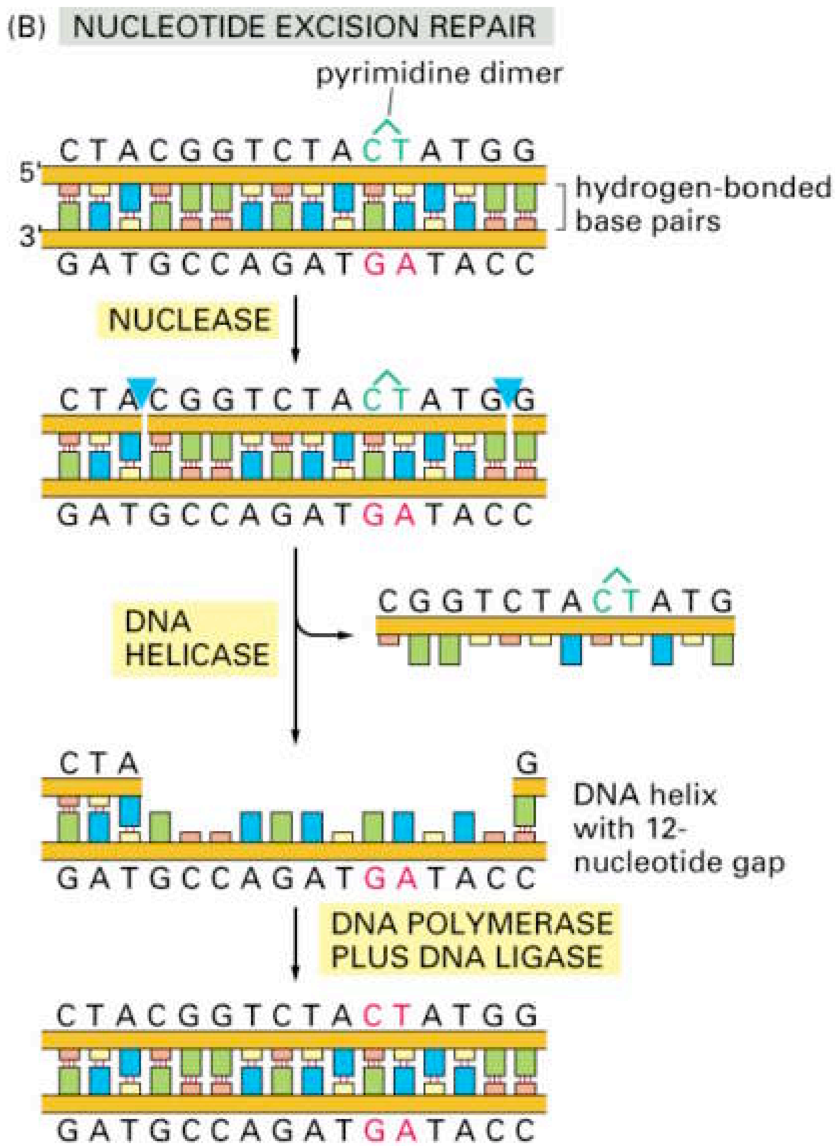


Figure 5–50 part 2 of 2. Molecular Biology of the Cell, 4th Edition.

Reparatur von Doppelstrangbrüchen

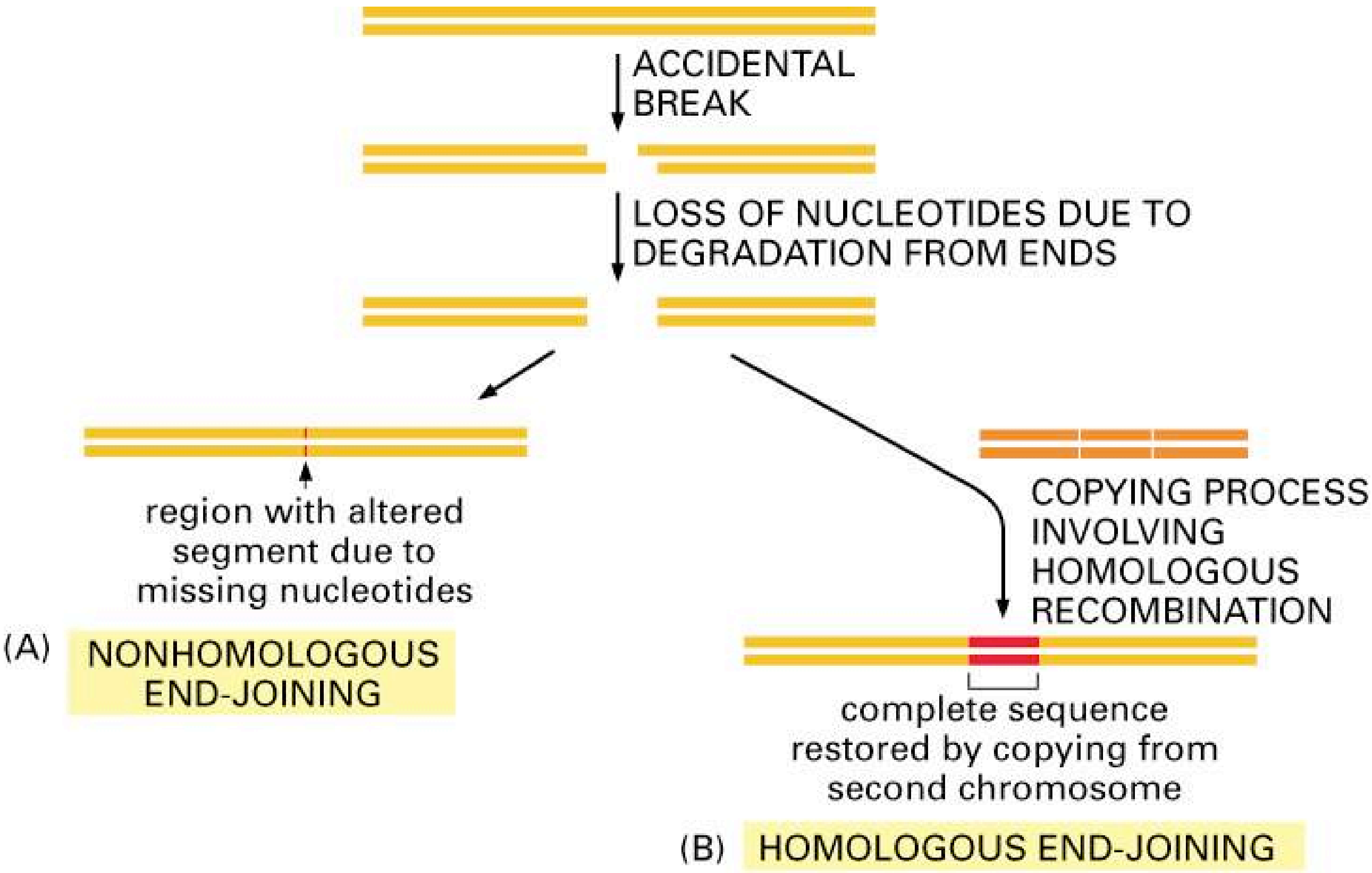


Figure 5–53. Molecular Biology of the Cell, 4th Edition.

Zusammenfassung

- Nukleotide als DNA-Bausteine
- Histone als DNA-”Spulen”
- Semikonservative DNA-Replikation
- Bidirektionale Replikation von oris ausgehend.
- “leading” und “lagging” Strang, Okazaki-Fragmente
- Chromosomale Enden werden durch Telomerase repliziert.
- Proofreading erhöht Genauigkeit der DNA-Synthese.
- DNA-Schäden werden durch unterschiedliche Mechanismen repariert (Base excision, Nucleotide excision, homologous and nonhomologous end-joining.