Advanced Cell Biology. Lecture 16

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Outline

DNA replication (end)

DNA reparation

Outline

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DNA reparation

Previous final question: the answer

Why cells use RNA as DNA replication primers?

Previous final question: the answer

Why cells use RNA as DNA replication primers?

- Primers should be used for starting nucleotide chain, therefore they should be nucleic acids
- Primers contain errors and should be removed, therefore they should be distinguishable from the rest of chain, therefore, they should be RNA instead of DNA

Lab 4 preparation

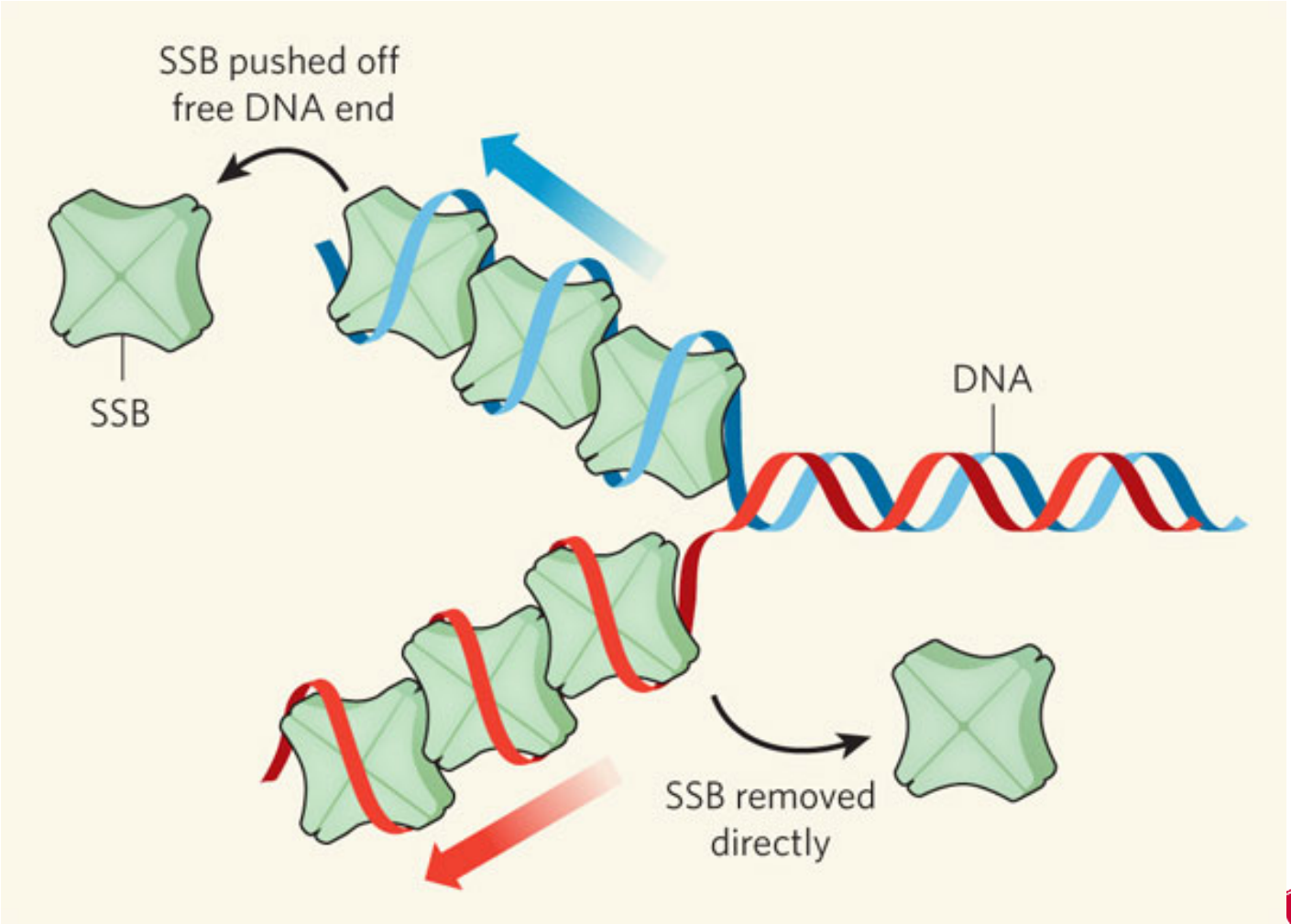
Please write you name and name of paper you will defend, also write “presenter” if you the person who will actually make presentation

1. Avery et al. (1944) “Studies on the chemical nature of the substance inducing transformation of Pneumococcal types”
2. Wilkins et al. (1953) “Molecular Structure of Deoxypentose Nucleic Acids”
3. Watson & Crick (1953) “A Structure for Deoxyribose Nucleic Acid”, and (if possible) also “Genetical Implications of the structure of Deoxyribonucleic Acid”

DNA helicases

- DNA helicases are natural zippers
- They use energy of ATP to untangle the double helix
- *Single-strand binding protein* (SSBP) associates with DNA strand to prevent re-forming base pairs

Single strand binding protein, SSBP

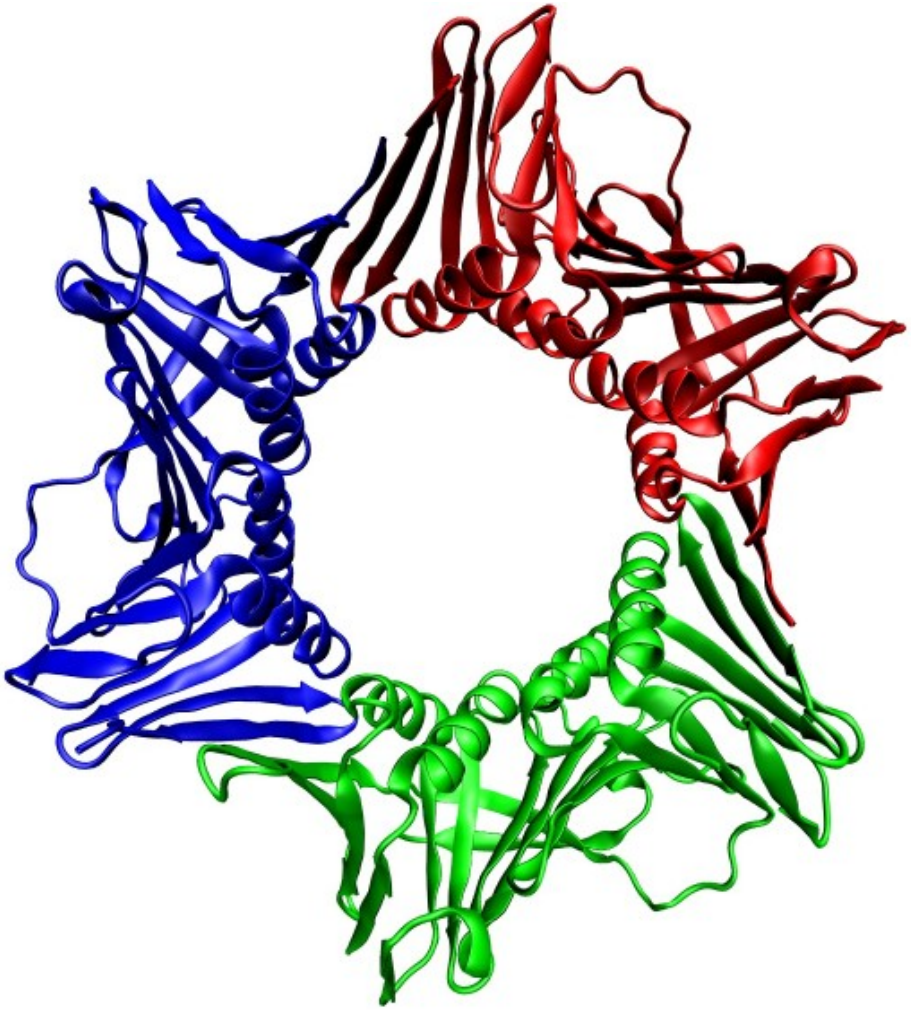


DNA helicase movie

Sliding clamp (DNA clamp)

- Keeps DNA polymerase attached to the template
- Form a ring around different DNA polymerases
- Most are trimers of PCNA proteins

Human DNA clamp protein (trimer of PCNA)



DNA sliding clamp movie

DNA replication complex

1. DNA: old helix, leading strand, lagging strand
2. DNA polymerases
3. DNA helicases
4. SSBP
5. DNA clamp
6. Primase
7. DNA ligase

DNA replication machine (simplified)

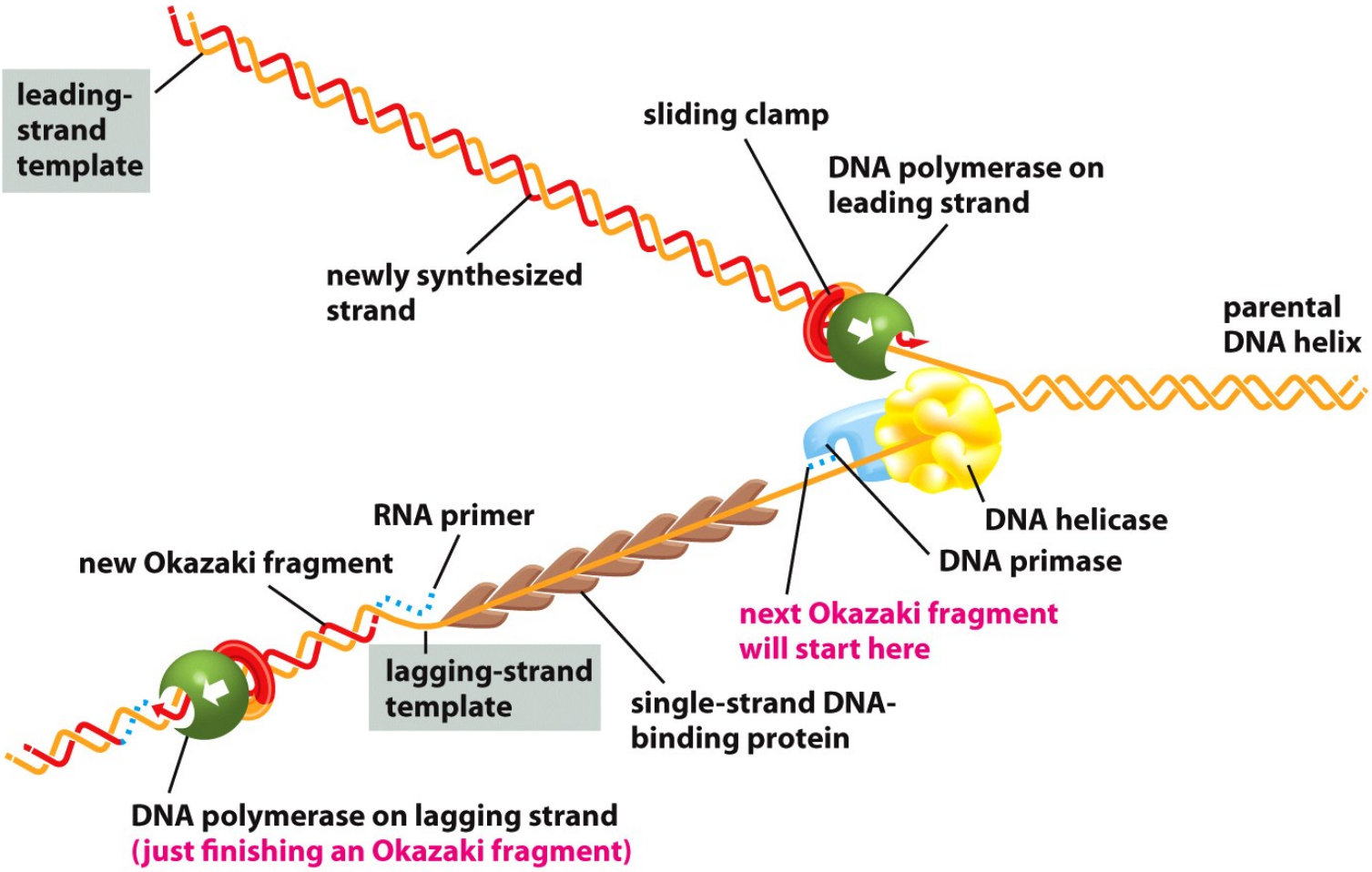


Figure 6-17a Essential Cell Biology 3/e (© Garland Science 2010)

DNA replication machine (more realistic)

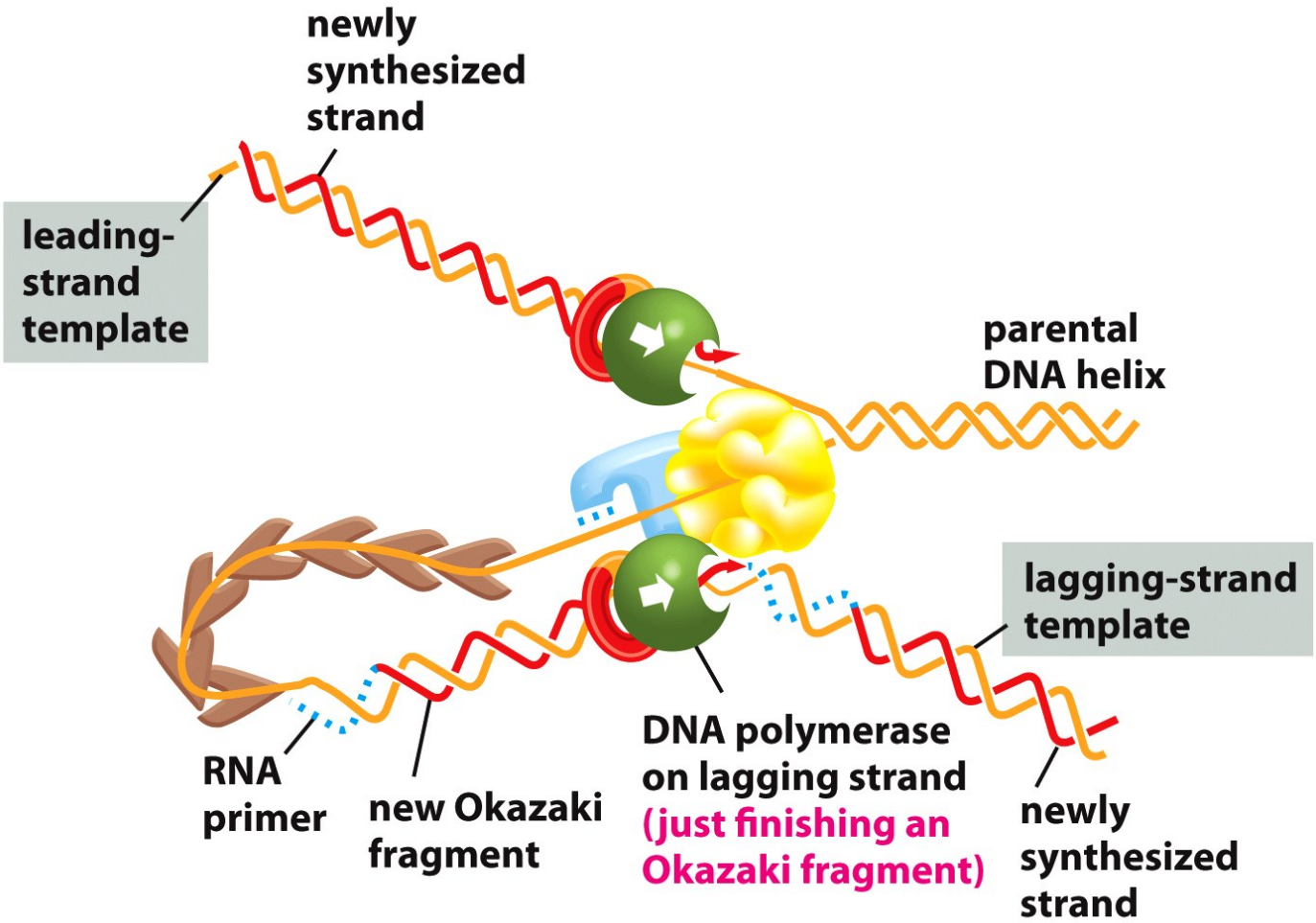


Figure 6-17b Essential Cell Biology 3/e (© Garland Science 2010)

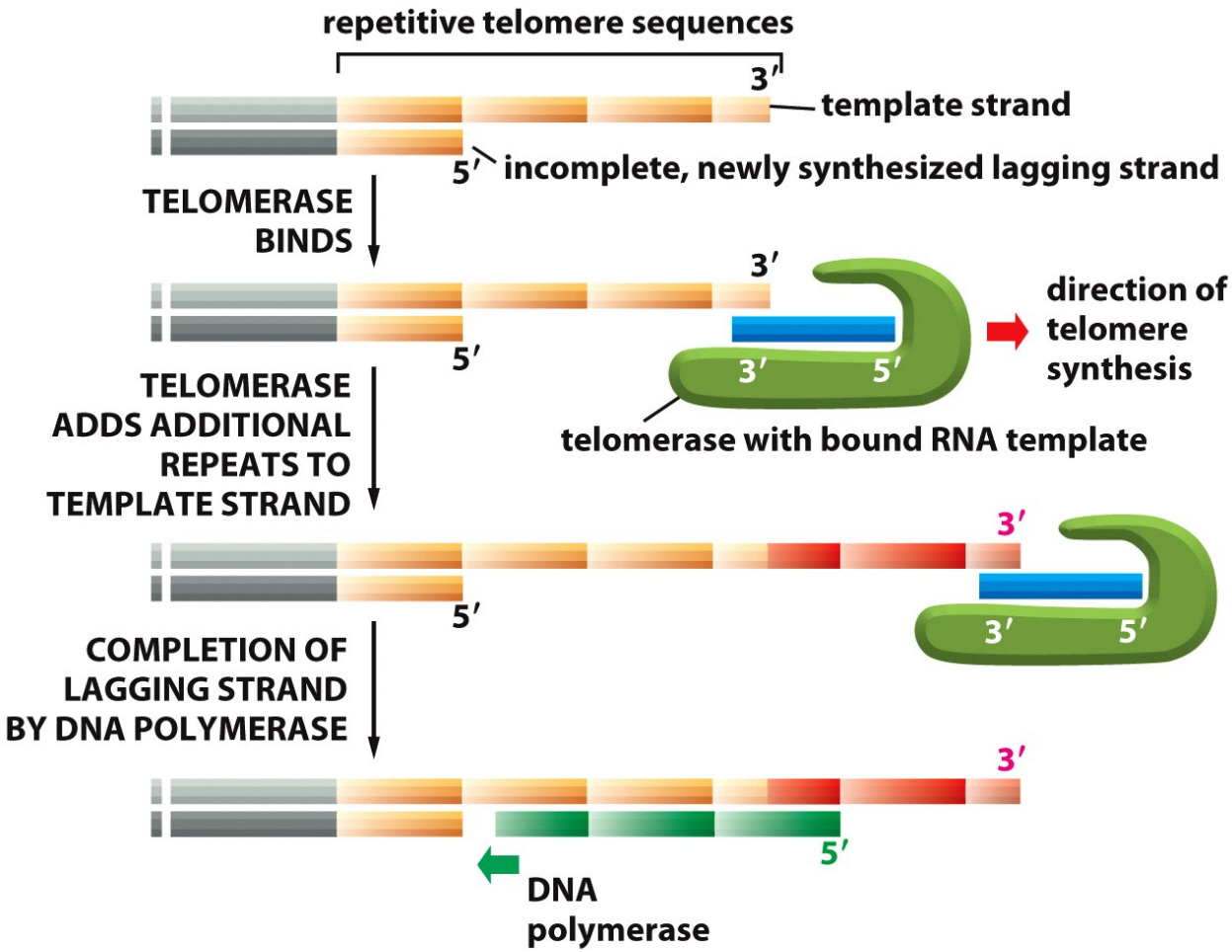
DNA replication sewing machine movie

DNA replication in general movie

Telomerase

- Lagging strand cannot reach the end of DNA molecule
- Every replication cycle chromosome lost parts of telomeres from its ends
- To prevent a loss on meaningful DNA fragments, telomerase extends chromosome with new telomere sequences
- In humans, telomeres are several thousands of TTAGGG sequences
- Telomeres are also recognizable ends of chromosomes

Telomerase and telomere



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Telomere movie

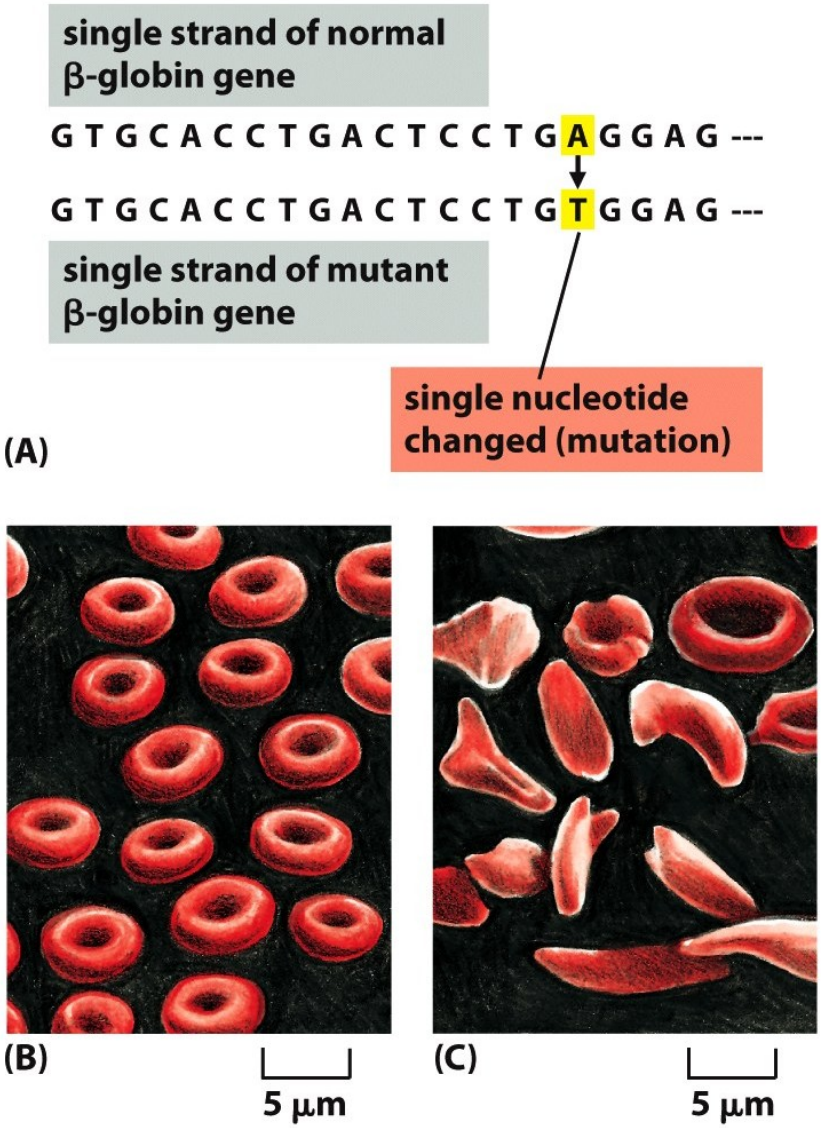
Telomere theory of aging

- In humans, telomerase is only active in germ cells, stem cells and certain white blood cells
- Olovnikov (1971) postulated that lost of DNA ends will eventually stop division of cells and may stimulate senescence of cells
- There is a strong support of this hypothesis with some cell types (e.g., blood vessels wall cells); however, mice with knocked-out telomerase gene do not show significantly less lifespan

Single nucleotide mutations

- SN mutations (deletions and substitutions) may damage severely the function of protein
- In sickle-cell anemia, substitution turns Glu to Val and modify structure of hemoglobin and then red blood cells
- However, people with partly modified red blood cells are less susceptible to malaria

Sickle-cell anemia



Mutation theory of ageing and/or cancer

- Accumulation of mutations will result in a constant loss of functions
- Cells will either degrade or start to go out of control (cancer)

DNA mismatch repair

- Normally, error rate of DNA polymerase + proofreading is $\approx 10^{-7}$
- DNA mismatch repair proteins decrease it to $\approx 10^{-9}$
- They react on DNA conformation deviations; recognize newly synthesized strand by nicks, and remove wrong fragments which are later replaced with DNA polymerase and ligase
- Some cancers are results of mutations in DNA mismatch protein genes

DNA mismatched repair system

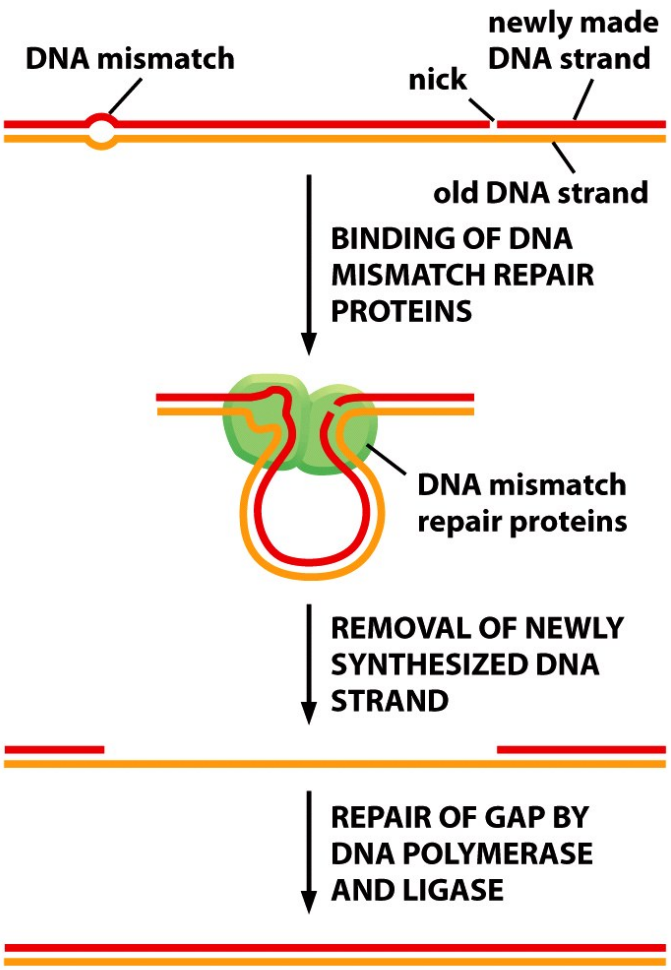
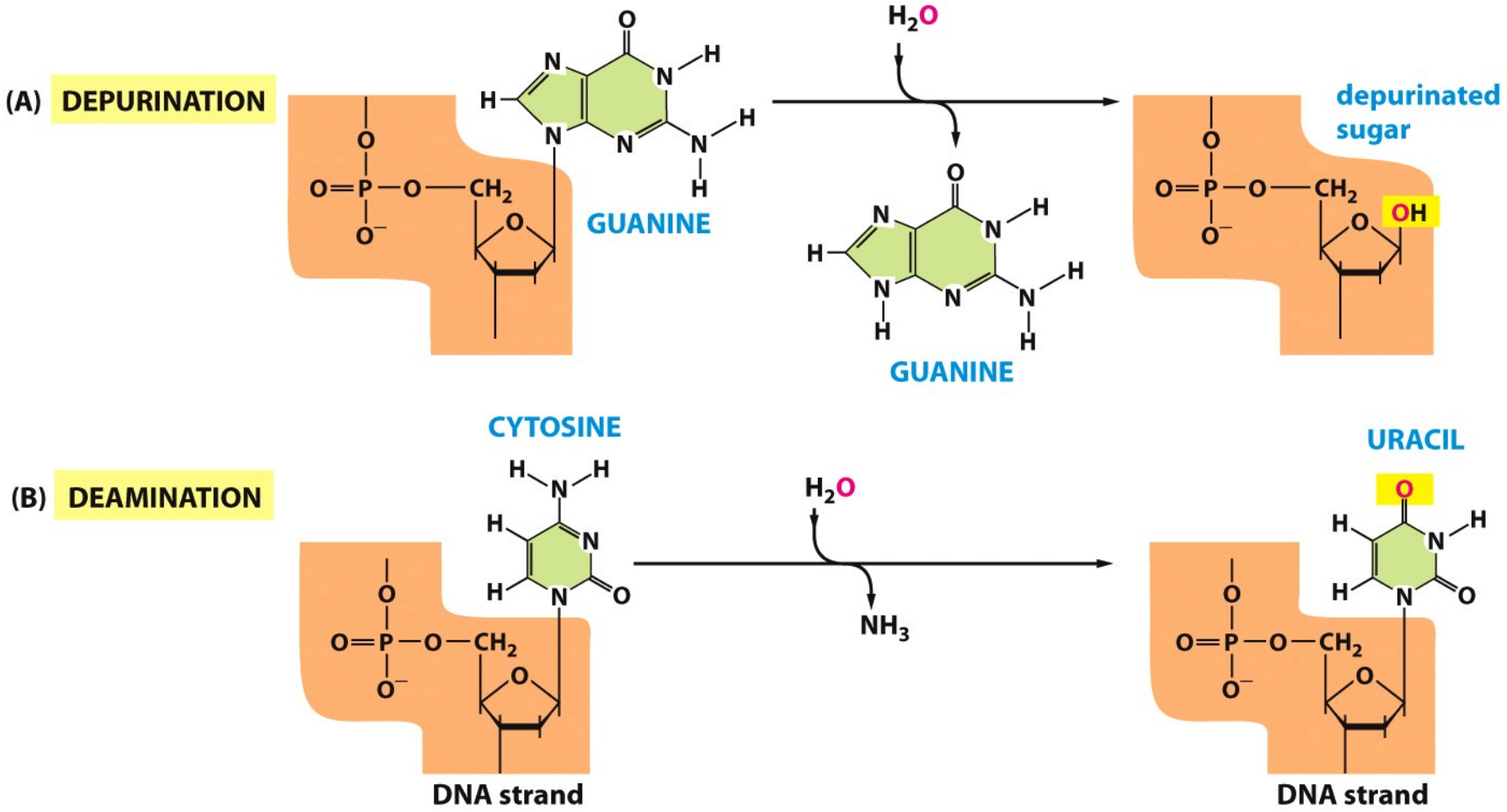


Figure 6-22 Essential Cell Biology 3/e (© Garland Science 2010)

Depurination and deamination

- Depurination is detaching A and G from its sugar; it results in deletion*
- Deamination is C to U conversion; it results in substitution*

Depurination and deamination



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Final question (1 point)

Final question (1 point)

What is the difference between deletion and substitution?

Summary

- DNA replication system is a multienzyme complex
- Ends of chromosomes are constantly shortening and extending with new telomeres
- DNA suffers from multiple damaging events; multiple reparation systems are trying to lower mutation risks

For Further Reading

A. Shipunov.
Advanced Cell Biology [Electronic resource].
2011—onwards.
Mode of access: `http://ashipunov.info/shipunov/school/biol_250`

B. Alberts et al.
Essential Cell Biology. 3rd edition.
Garland Science, 2009.
Chapter 6, pages 208–217.