

DNA REPAIR AND CANCER

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DNA REPAIR IN CANCER

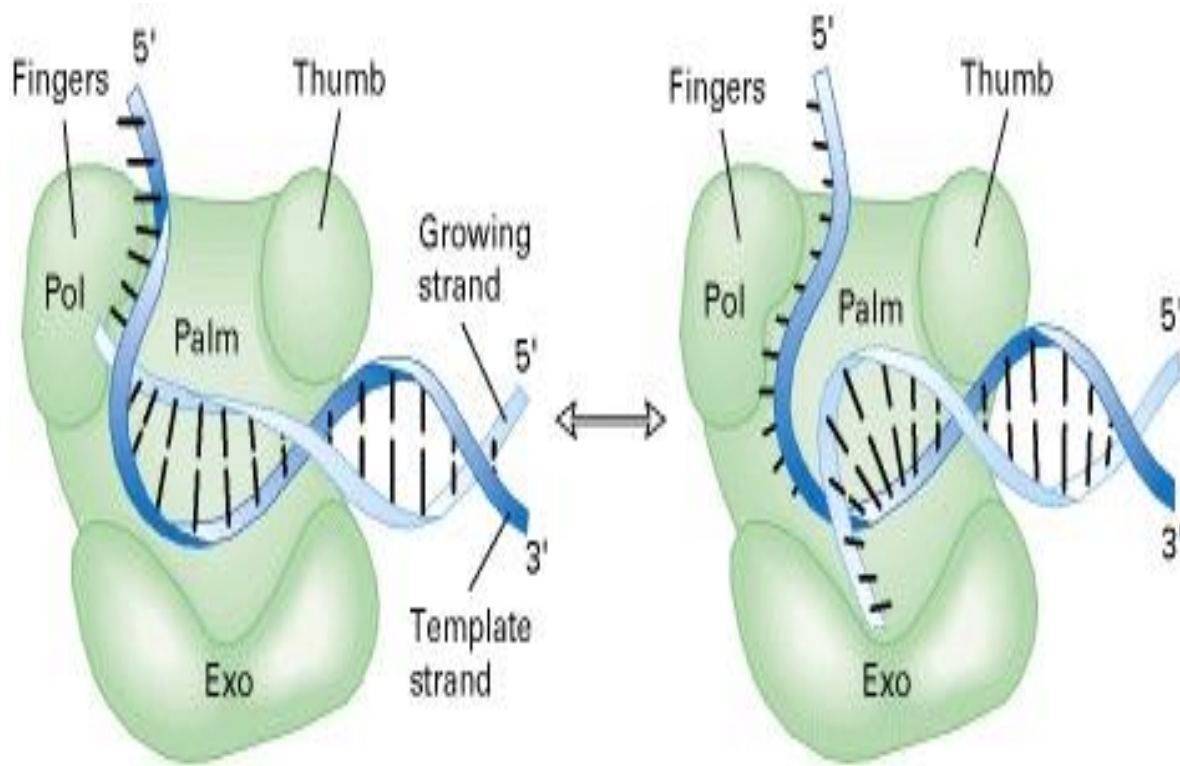
- The DNA sequence can be changed as the result of **copying errors** introduced by DNA polymerases during **replication** and by environmental agents such as mutagenic chemicals and certain types of radiation.
- If DNA sequence changes, whatever their cause, are left uncorrected, both somatic cells and germ cells might **accumulate** so many mutations that they could no longer function.
- Thus the correction of DNA sequence errors in all types of cells is **important for survival**.

TYPES OF REPAIR

- **PROOFREADING**
- **MISMATCH REPAIR**
- **BASE EXCISION REPAIR**
- **NUCLEOTIDE EXCISION REPAIR**
- **DOUBLE-STRANDED BREAK REPAIR**

PROOFREADING BY DNA POLYMERASE CORRECTS COPYING ERRORS

- Because the specificity of nucleotide addition by DNA polymerases is determined by **Watson-Crick base pairing**, a wrong base (e.g., A instead of G) occasionally is inserted during DNA synthesis.
- *E. coli* DNA polymerase III introduces about 1 incorrect base in 10^4 internucleotide linkages during replication and would cause a potentially harmful mutation during each replication
- DNA polymerase I can remove a mismatched base at the 3' growing end of a primer-template complex.

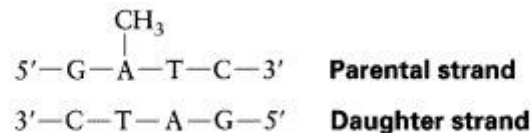


DNA polymerases add the correct nucleotides are added to the 3' end of the growing strand. Incorporation of an incorrect base at the 3' end causes a melting of the end of the duplex.

As a result, the polymerase pauses and the 3' end of the growing strand is transferred to the 3' → 5' exonuclease site (Exo) where the mispaired base and probably other bases are removed. Subsequently, the 3' end flips back into the polymerase site and elongation resumes.

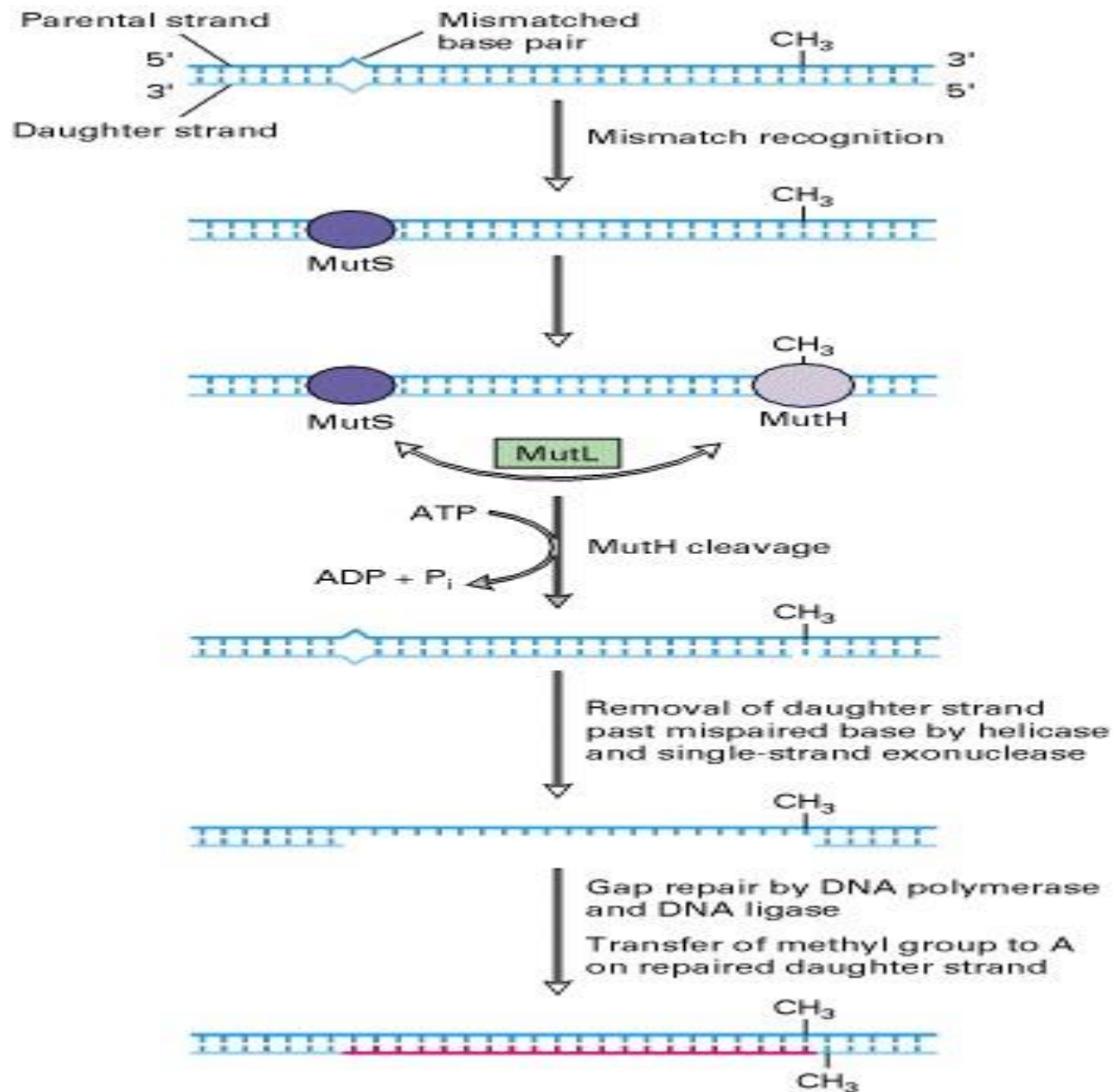
MISMATCH REPAIR

- Bacterial and eukaryotic cells have a mismatch-repair system that recognizes and repairs all single-base mispairs except C·C, as well as small insertions and deletions.
- The conceptual problem with mismatch repair is determining which is the normal and which is the mutant DNA strand,
- In *E. coli* DNA, adenine residues in a GATC sequence are methylated at the 6 position. Since DNA polymerases incorporate adenine, **not methyl-adenine**, into DNA,
- methylated only on the parental strand. methylated by a specific enzyme, called *Dam methyltransferase*, only after a lag of several minutes.
- During this lag period, the newly replicated DNA contains hemimethylated GATC sequences.



MODEL OF MISMATCH REPAIR

- This repair system operates soon after incorporation of a **wrong base**, before the newly synthesized daughter strand becomes **methylated**.
- **MutH** binds specifically to a hemimethylated GATC sequence, and **MutS** binds to the site of a mismatch.
- Binding of **MutL protein** simultaneously to MutS and to a nearby MutH activates the **endonuclease activity of MutH**, which then cuts the unmethylated (daughter) strand in the GATC sequence.
- A stretch of the daughter strand containing the mispaired base is excised
- Then gap repair and ligation and then **methylation** of the daughter strand.(DNA ligase and polymerase)

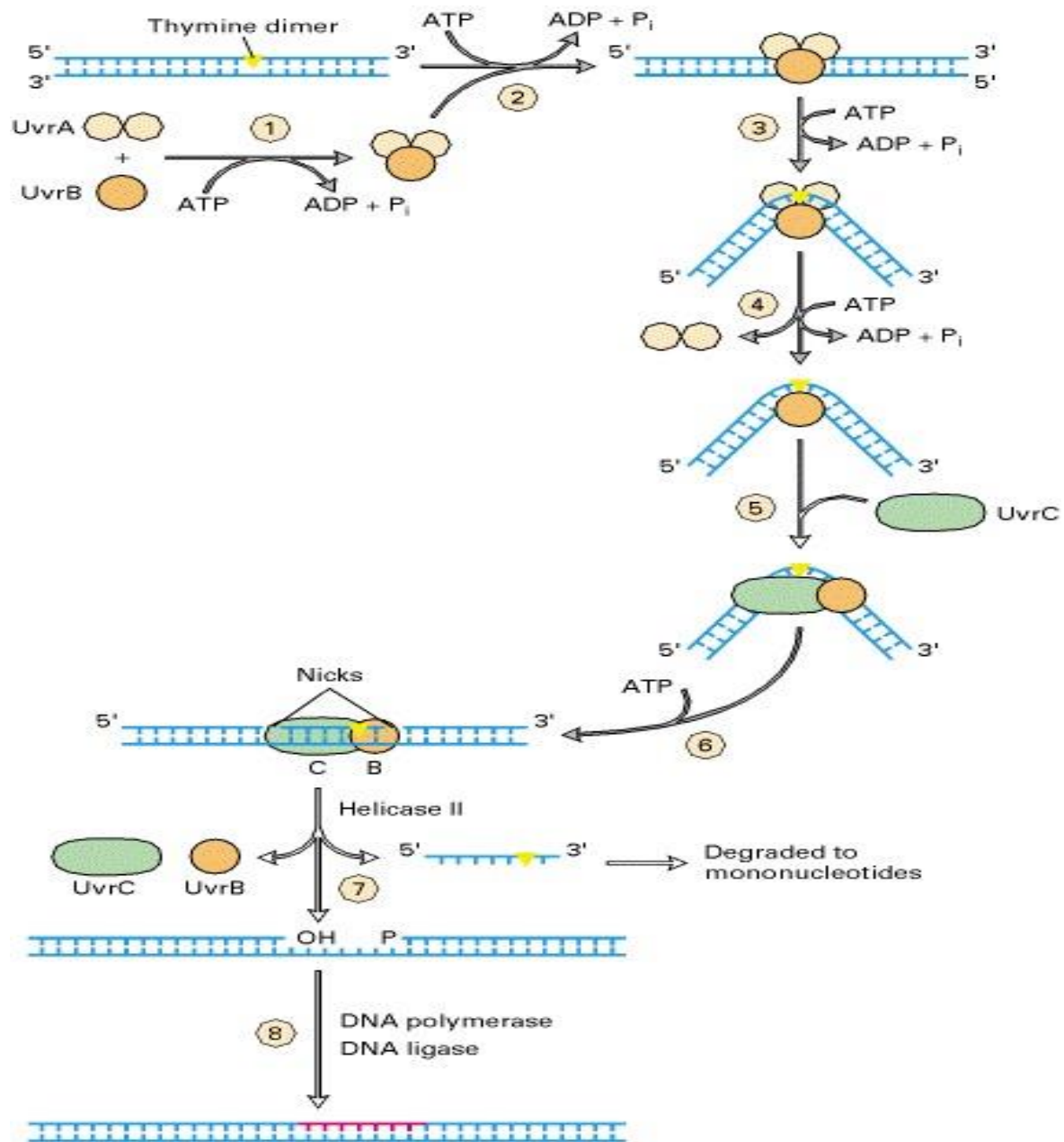


EXCISION REPAIR

- Two molecules of UvrA and one of UvrB form a complex that moves randomly along [DNA](#) (**steps 1 and 2**).
- Once the complex encounters a lesion, conformational changes in DNA, powered by ATP [hydrolysis](#), cause the helix to become locally denatured and kinked by 130° (step **3**).
- After the UvrA dimer dissociates (step **4**), the UvrC endonuclease binds and cuts the damaged strand at two sites separated by 12 or 13 bases (steps **5** and **6**). UvrB and UvrC then dissociate, and [helicase II](#) unwinds the damaged region (step **7**).
- Releasing the single-stranded fragment with the lesion, which is degraded to mononucleotides.
- The gap is filled by [DNA polymerase I](#), and the remaining nick is sealed by DNA [ligase](#).

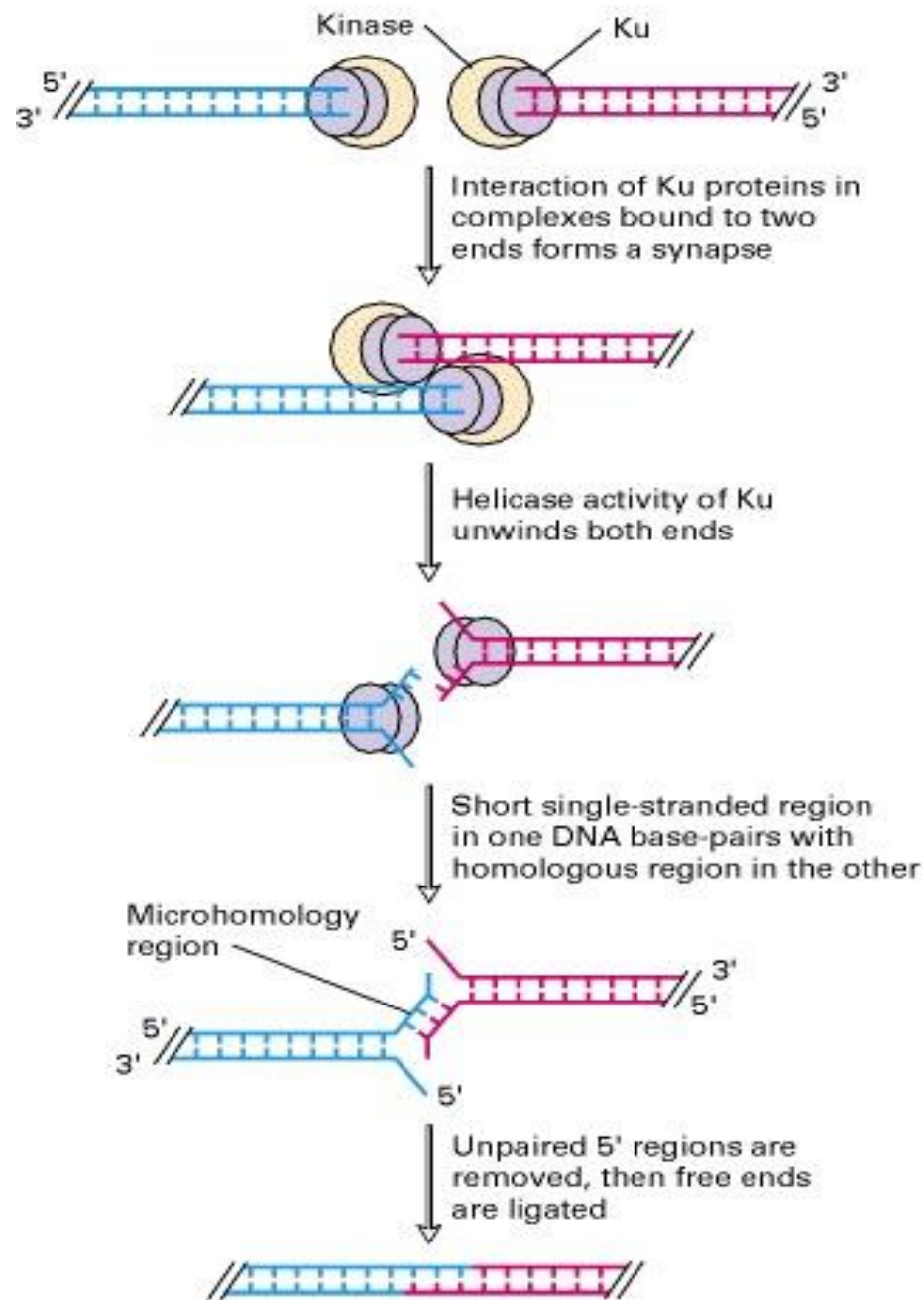
EXCISION REPAIR

- Cells use excision repair to fix [DNA](#) regions containing **chemically modified bases**, often called **chemical adducts**, that distort the normal shape of DNA locally.
- It has the ability of certain proteins to slide along the surface of a double-stranded DNA molecule looking for bulges or other irregularities in the shape of the double helix.
- For example, this mechanism repairs *thymine-thymine dimers*, the most common type of damage caused by UV light and these dimers interfere with both replication and [transcription](#) of DNA. Excision repair also can correct DNA regions containing bases. carcinogens such as benzo(*a*)pyrene



Repair of double-strand breaks by end-joining of nonhomologous DNAs. DNAs with dissimilar sequences at their ends

- These DNAs could be cut fragments from a single gene, or DNAs cut from different chromosomes.
- A complex of two proteins, **Ku and DNA dependant protein kinase**, binds to the ends of a double-strand break.
- After formation of synapse in which the broken ends overlap, **Ku unwinds** the ends, by chance revealing short homologous sequences in the two DNAs, which base-pair to yield a region of microhomology.
- The unpaired single-stranded 5' ends are removed
- The two double-stranded molecules ligated together. As a result, the double-strand break is repaired.



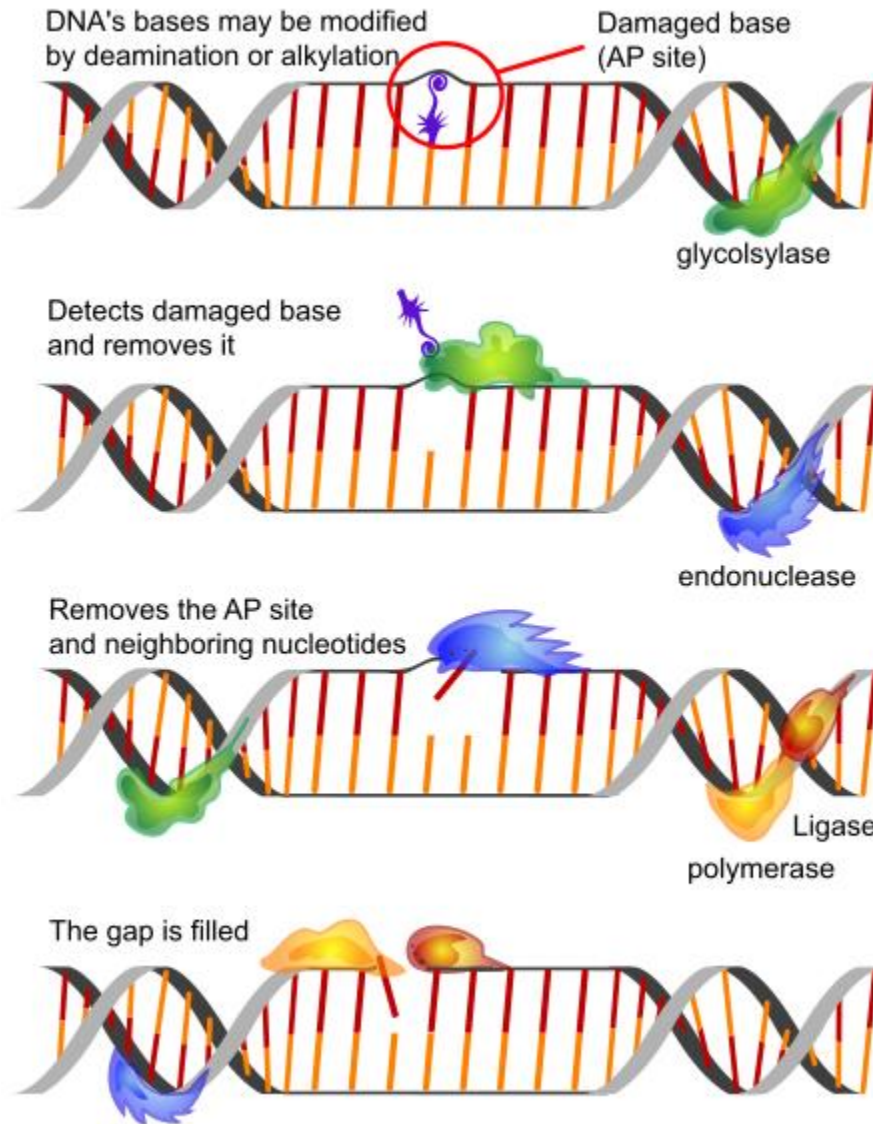
A cell that has suffered a particular double-strand break usually contains other breaks; such breaks can be repaired by joining the free DNA ends.

The joining of broken ends from different chromosomes, however, leads to translocation of pieces of DNA from **one chromosome to another**.

Double-strand breaks are caused by ionizing radiation and by **anticancer drugs**, such as **bleomycin**, which is why these drugs are used to kill rapidly growing cells.

BASE-EXCISION REPAIR

- Every cell has a class of enzymes called **DNA glycosylases** that recognize particularly common DNA lesions and remove the affected base by cleaving the *N*-glycosyl bond. This cleavage creates an **AP site** or **abasic site**.
- DNA glycosylases recognize and remove a variety of damaged bases, including formamidopyrimidine and 8-hydroxyguanine (both arising from purine oxidation), hypoxanthine (arising from adenine deamination), and alkylated bases such as 3-methyladenine and 7-methylguanine.
- Glycosylases that recognize other lesions, including **pyrimidine dimers**, have also been identified in some classes of organisms.
- Once an AP site has formed, another group of enzymes must repair it. This process begins with **AP endonucleases**, enzymes that cut the DNA strand containing the AP site.
- A segment of DNA including the AP site is then removed, **DNA polymerase I** replaces the DNA, and **DNA ligase** seals the remaining nick. In eukaryotes, nucleotide replacement is carried out by specialized polymerases.



BASE-EXCISION REPAIR

- Cellular DNA-repair processes have been implicated both in **protecting** against and contributing to the development of cancer.

THANK
YOU