Notes NO. DNA - Repair Lec 5

→ DNA is relatively stable molecule, but in the absence of repair systems, the cumulative effect of many infrequent but damaging reactions would make life impossible.

→ Repair systems present inside the body can provide protections against DNA damage.

Q What are the factors that affect rate of DNA repair?

1) Cell type
2) Age of the cell
3) Ext environment

Q what will happen to the all that has ① accumulated a large amount of DNA damage ② No longer effectively repairs damaged incured to its DNA ?

1) Senescence → irreversible state of dormancy
2) Apoptosis / programmed cell death
3) formation of cancerous tumors as a result of unregulated cell division.

Q Scientists who recieved nobel prize in chemistry for their work on molecular mechanisms of DNA repair processes :—

1) Aziz Sancer
2) Tomas Lindahl
3) Paul Modrich.

Scanned by CamScanner

NO.

→ 2 types of DNA repair processes

nucleotide excission repair (1)

base excission repair (2)

Mismatch Repair system during DNA replication

1) by exonucleolytic activity of DNA polymerases. This exonucleolytic activity usually occurring simultaneosly with DNA replication

Pol-1 has 5`-3` polymerization activity, in addition to 3`-5` exonuclease and 5`-3` exonuclease activities which are very important to correct the mismatches in the newly synthesized DNA, the process that we call proofreading.

Remember that DNA polymerase has 1) polymerase active site
1) addition of nucleotides
2) formation of phosphodiester bonds
2) Exonuclease active site / Exonuclease catalytic site

Removes unpaired residues at the terminus and replace them by the correct base pair

Scanned by CamScanner

NO.

exonuclear act. لل alternative بس replication برضو خلال ال

2) A system that recognizes and repairs major insertion ① ② deletion and mis incorporation of bases. ③

Collectively, mismatch repair system carries out the following corrections.

- Removes replication errors which aren't recognized by the replication machine
- Detects structural changes in the DNA helix.

always remember, Newly synthesized strand of DNA is preferentially nicked to be distinguished from the parental strand:

Parental → methyl group فيه

New/daughter strand → methyl group ما فيه

Repair after maturation

- Things that cause DNA damage after its synthesis:-
  1) Chemical polutants
  2) Radiation.

Types of chemicals
- Industrial chemicals
  - Vinyl chloride
  - H_2O_2
- Environmental chemicals
  - ↳ Polycyclic Hydrocarbon in smoke and tar

Their effect?

Large chemical modifications in DNA.
- oxidized base
- alkylated base.

Scanned by CamScanner

⇒ Thermal disruption

what effect does elevated T have on DNA?

1) ↑ rate of depurination
2) ↑ rate of single strand breaks

الـ Thermophilic b تستخدم كمصدر للحصول على الـ DNA polymerase

↓

for polymerase chain reaction in DNA fingerprinting

Collectively, Damage produced by these factors:-

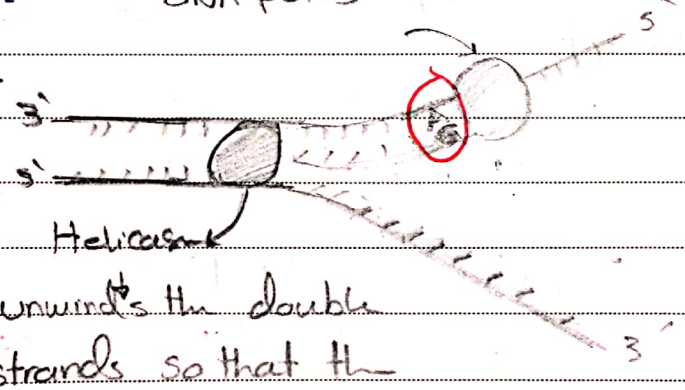
1) Base loss 2) Base modification
3) strand breakage.

Types of DNA Repair Sys

1) Base Excision Repair

يعتبر single strand repair mechanism.

DNA pol-3



Helicase

unwinds the double strands so that the polymerase can perform its function

DNA - Pol 3

↓ synthesize DNA also has the ability to proofread

NO.

if Adenine, insted of being paired up with the proper base which is thymine, it accidently paired up with Guanine → This is Mistake

هلأ ال pol DNA فعلياً sensus لما يصير غلط في التصنيع ، then هاد ال pol
بيعملها ، incorrect base ال it removes ، شوي goes backward
exonuclear activity بنحكيه هاد يلي بنحكيه replacment بالصح

only from the end of DNA strand

ability to remove nucleotide

BER: removing damaged bases that could otherwise cause mutations by mispairing or lead to breaks in DNA during replication.

① Glycosylase enzyme → generally specific for one type of lesion.

- Recognizes a specific type of incorrect base
- generate AP site
- Cleaves the N-glycosyl blond to remove the deliction N-base.

② The generated AP site is recognized by AP endonuclear

↓

Cleaves the phosphodiester backbone immediately 5' to the AP site

↓

Leaving 3'OH and 5'-deoxyribose Phosphate terminus

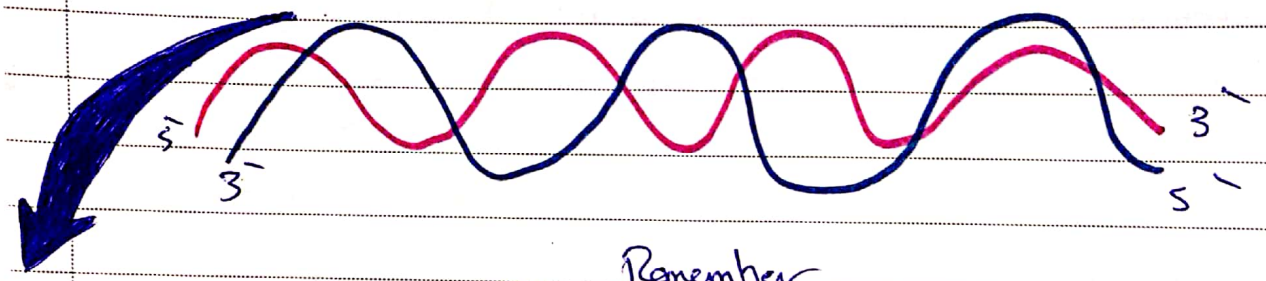
هاد الخطوة هي incision step

FIVE APPLE

Scanned by CamScanner

Uracil DNA glycosylase: Removes the Uracil that results from hydrolytic deamination of cytosin.

NO.



[Damage to single Base
A, T, C, G]

Remember
Single base damage is mediated by?
1) Hydrolytic Deamination
2) Oxidation
3) Alkylation
[Cytosin → Uracil] etc.

BER
Enzymes involved:—
1) DNA glycosylase → Recognizes the lesion and remove the damaged base (by cleavage of N-glycosyl bond)
2) AP endonuclease → Recognizes the AP site (missing base site) (1) by glycosylase
(2) Creats nick in phosphodiester backbone
mediated by?
3) DNA deoxyribo-phosphodiesterase (dRpase)
↳ Removes the 5'-deoxyribose phosphate leaving a gap → nick
exonuclease enzyme
4) DNA polymerase → Insertion of base at AP site, using the 2nd strand as template.
5) DNA Ligase → Seals the nick.

FIVE APPLE

Scanned by CamScanner

NO.

Nucleotide Excision Repair NER

- BER can recognize specific non-bulky lesions in DNA and it can correct only damaged bases that are removed by DNA glycosylase.

Cyclobutane Pyrimidine dimers result from UV light are pre-mutagenic lesions that alter the structure and base-pairing.

When NER?

Removes DNA damage induced by UV light, dna damage results in bulky DNA adducts

↓ mostly

((Thymine dimers))

Effect of UV-light (UV-B particularly) :-

- Causes covalent cross linking b/w adjacent thymine ~~bases dimers~~

⇒ Cyclobutane thymine dimers.

Such ionization radiations are created by
1) Radioactive rays
2) X-rays.

⇒ Results in breaks in DNA strands.

effect of thymine dimers:-

1) double helix is distorted (bent) → bcz thymines are pulled toward each other
2) H-bonding to adenines on the opposite strand can weakened
3) Distortion of strands → loss of template info.
→ prevent advancing replication forks
→ Inhibit process of RNA transcription

FIVE APPLE

Scanned by CamScanner

Radiation
↳ Dimerization
Oxygen. free radicals in the Presence of heavy metals (Fe^{+2})
↓
Oxygenation damage

So: Oxy radical + Fe^{+2} ⟶ sugar destruction
(ergo)
↓
breaks in DNA strand

NO.

NER, BER → Repair DNA damage that may be, missing corporation nucleotide in DNA and any wasn't properly fixed during proofreading of DNA

Briefly :-

- Recognition of the damaged leads to removal of a short single-stranded DNA segment that contains the lesion.
  ↳ Cleavage of the damaged DNA on either side of a lesion by : endonuclease.
- The unclamaged single-stranded DNA remains and DNA polymrase (Highly processive polymrase) uses it as a template to produce a short complimentary sequence
- final ligation to complete NER and form a double stranded DNA is carried out by DNA ligase

So, short checkup

1) Sources of DNA damage — exogenous
   — Endogenous → free radicals
   - Replication errors.
2) Types of DNA damage due to endogenous cellular process :-
   - Oxidation
   - alkylation (usually) methylation
   - Hydrolytic deamination (ex: C → U)
   - Mismatch of bases

FIVE APPLE

Scanned by CamScanner

NO.

3) ex on exogenous causes (agents) that can dna damage?

- UV-B light ⇒ Pyrimidin dimers. "Direct damage"
- Ionization radiation ⇒ "breaks in DNA strands"
- Thermal disruption ⇒ Depurination (Loss of purines)
- Industrial chemicals (vinyl chloride / H_2O_2) + environmental chemicals
  - ↳ Cross linking of DNA
  - oxidized bases
  - Alkylated bases
  - Conversion of 1 base to another

For infrequent lesions repair → Recombination can be used
For frequent lesions repair → Error-prone repair

Associated Hereditary DNA repair disorders:
+ Non hereditary

1) Fanconi Anemia "FA"

* Rare
* Defect → Excision repair
* Non-hereditary
* Affected ppl mostly develop cancer
  - ↳ : acute myelogenous leukemia
  - ① ↳ 90%: bone marrow failure.
  - ② ↳ Congenital defects, Commonst: Short stature.

also develope

Scanned by CamScanner

NO.

2) HNPCC "Lynch syndrome"

- Common → Type of cancer syndrom
- Defect → Deficient mismatch repair.
- Autosomal Dominant

- Affected ppl are at high risk for several Cancer types:-
  * Most often → Colon Cancer
  * Endometrial Cancer
  * Ovary, stomach, SI cancers.
  * hepatobiliary tract Cancer
  * Urinary Tract Cancer
  * Brain Cancer
  * Skin Cancer

→ Why? Due to inherited mutations that impair DNA mismatch repair.

3) Werner's Syndrome (hereditary)

- Rare — aka (adult progeria)
- Defect → Deficient Helicase
- Autosomal Recessive progeroid syndrom
- Most imp characteristic → Premature Aging

- Main Cause of death → CV disease // Cancer

Scanned by CamScanner

NO.

Thymin Dimer Excision repair system.

T^T dimers may be repaired by 2 mechanisms:—

*) Photo reactivation repair

↳ Photo reactivating enzyme is activated by visible light. Recognizes the T^T dimer and clear the cyclobutam ring.

*) Excission Repair

This system contains [UV endonuclear] ↙

↓

cleaves the damaged DNA on either side of the dimer to produce a gap of 1000 nucleotides on a single strand that can be filled by DNA pol using the other strand as template.

↓

[DNA ligase] complete the process.

*) Excission repair

Xeroderma Pigmentosum

+ Rare
+ 90% of XP patients
↘ skin Carcinomas.

+ Autosomal recessive

+ Genetic Disorder of DNA repair

↓↓

* The ability to repair damage caused by UV light is deficient.

* XP individuals also

↙ extremely sensitive to sunlight

↓ suffer from neurological disorders such as mental retardation

↘ unable to repair thymin dimers and other types of DNA damage.

FIVE APPLE

Scanned by CamScanner