MOLECULAR BIOLOGY

MUTATIONS

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Any sudden ,stable, heritable change occurring in hereditary material (genetic make up) is called as **mutation**

They may be harmful, beneficial or neutral

May occur in somatic cells (aren't passed to offspring)
May occur in gametes (eggs & sperm) and be passed to offspring

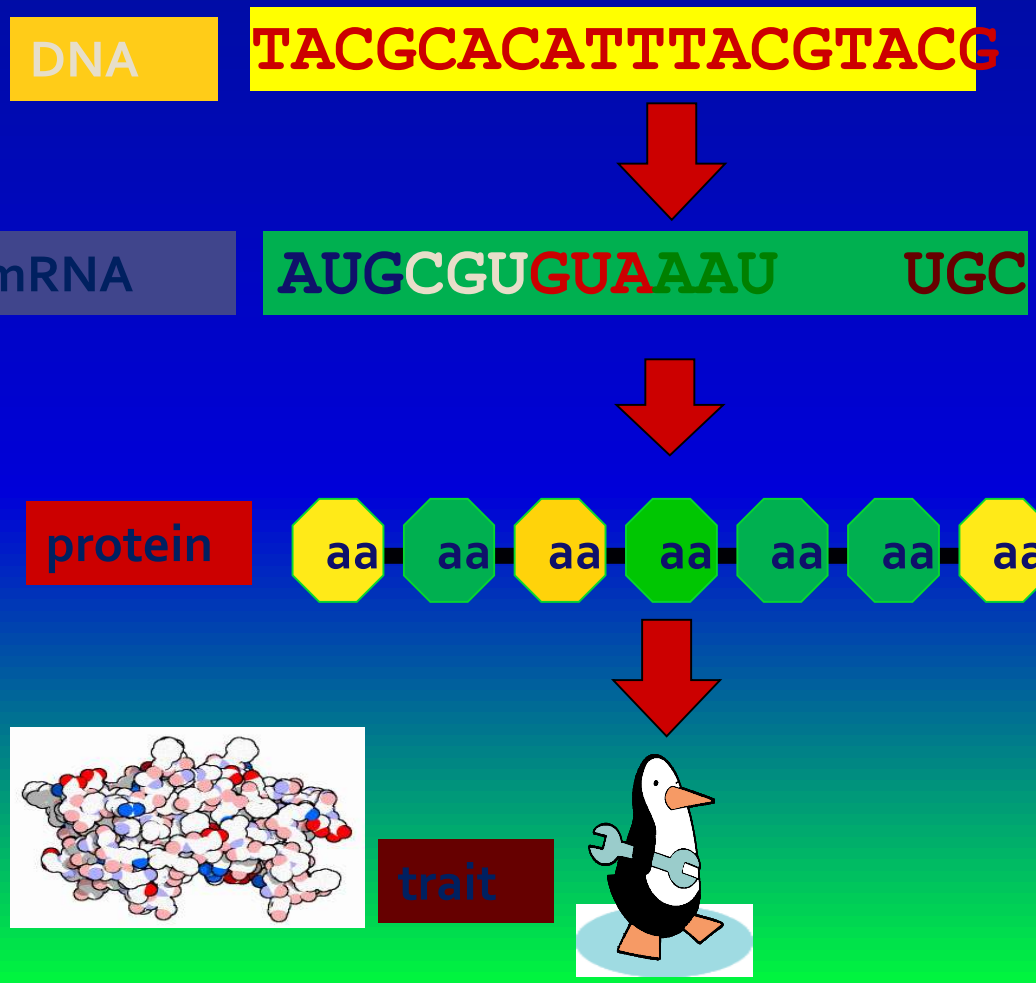
Changes to DNA are called mutations

change the DNA

changes the mRNA

may change protein

may change trait



- 1791-Seth Wright-male lamb with short leg
- Ancon breed of lamb

- Term *mutation*-Hugo de Vries 1901
- *Muttation theory of Evolution*
- *Oenothera lamarckiana*

- **Morgan-Drosophila**

Characteristics of natural mutations

- Spontaneous
- Advantageous or disadvantageous
- Neutral mutations
- Persistent
- Usually at low frequency
- Mutable gene, mutator gene, anti mutator gene
- Hot spots
- Forward and backward mutations
- Mutant alleles are pleiotropic
- Plays a role in evolution

Classification of mutations

- Somatic and germinal mutations
- Gametic and zygotic mutations
- Chromosomal and gene mutations
  - Chromosomal mutations

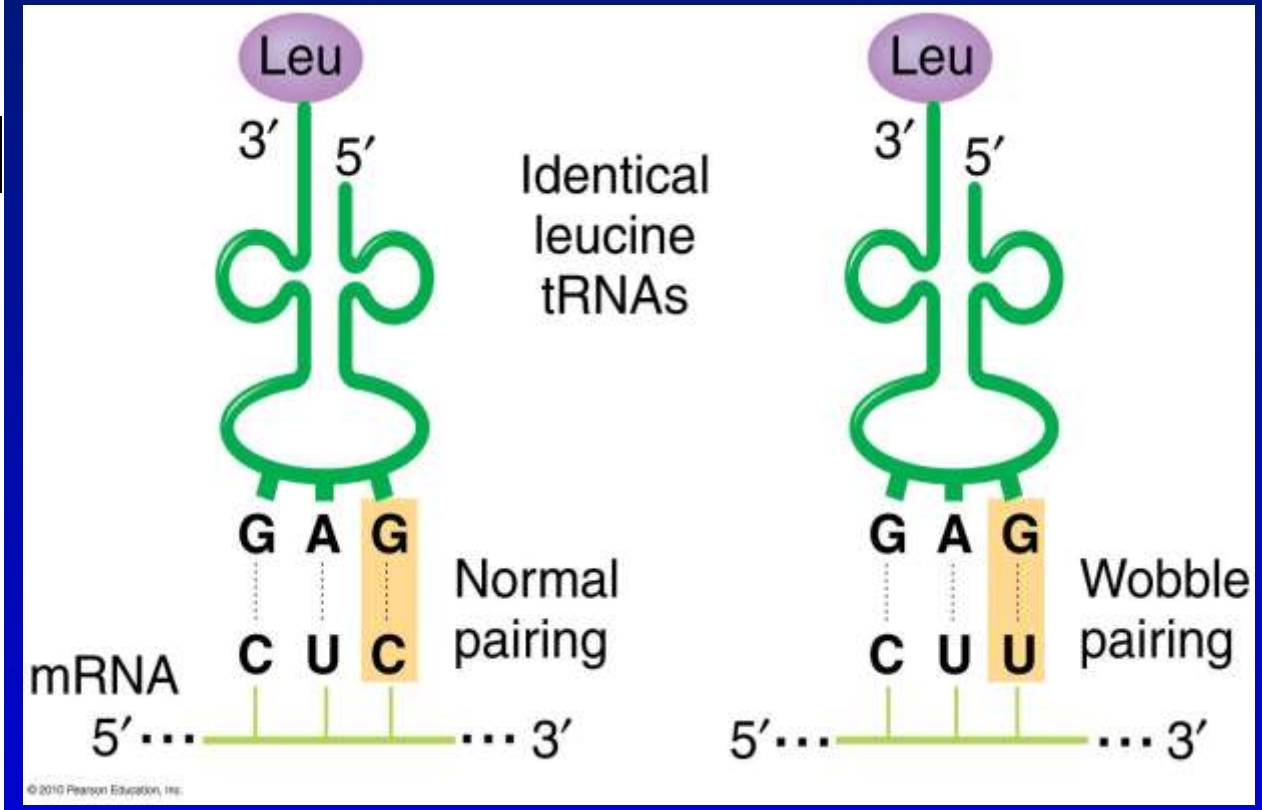
    Structural(Chromosomal abberations)

    Autosomal and sex linked

    X linked and Y linked
  - Numerical(Ploidy changes)

The yellow coloured part of this tulip petal is the result of a somatic mutation. Only a part of the organism is affected as not all the cells have inherited the mutation. This mutation most likely occurred during the flower's development in a single cell that grew and divided to form the now yellow part of the flower.

- Micro and macro mutations
- Biochemical mutations
- Lethal mutations
- Dominant and recessive mutations
- Silent and neutral mutations
- Spontaneous and induced mutations



Wobble hypothesis

Pairing of the tRNA anticodon with the mRNA codon proceeds from the 5' end of the codon. Once the first two positions are paired, exact base pairing of the third position is less critical. The third (5') base of the anticodon can typically pair with either member of the purine or pyrimidine pair in the codon as appropriate: it "*wobbles*". In this example, the double-ringed G can pair with either a single-ringed U or C.

Gene mutations
Intragenic mutations
Point mutations

- Change in the base sequence of individual genes
- Defective DNA repair system
- Defective DNA replication system
- Defective DNA polymerase and DNA ligase
- Production of aberrant DNA precursors

- Muton
- -the smallest mutable fragment of the genetic material which serves as the basic unit of gene mutation
- Muton is an intragenic region. It is the unit of mutation in a gene. In fact, it is a small unit. It can be either a single base or few bases since a mutation can also occur due to one base replacement or several bases. Hence, a single nucleotide is the smallest muton.

Molecular basis of gene mutations

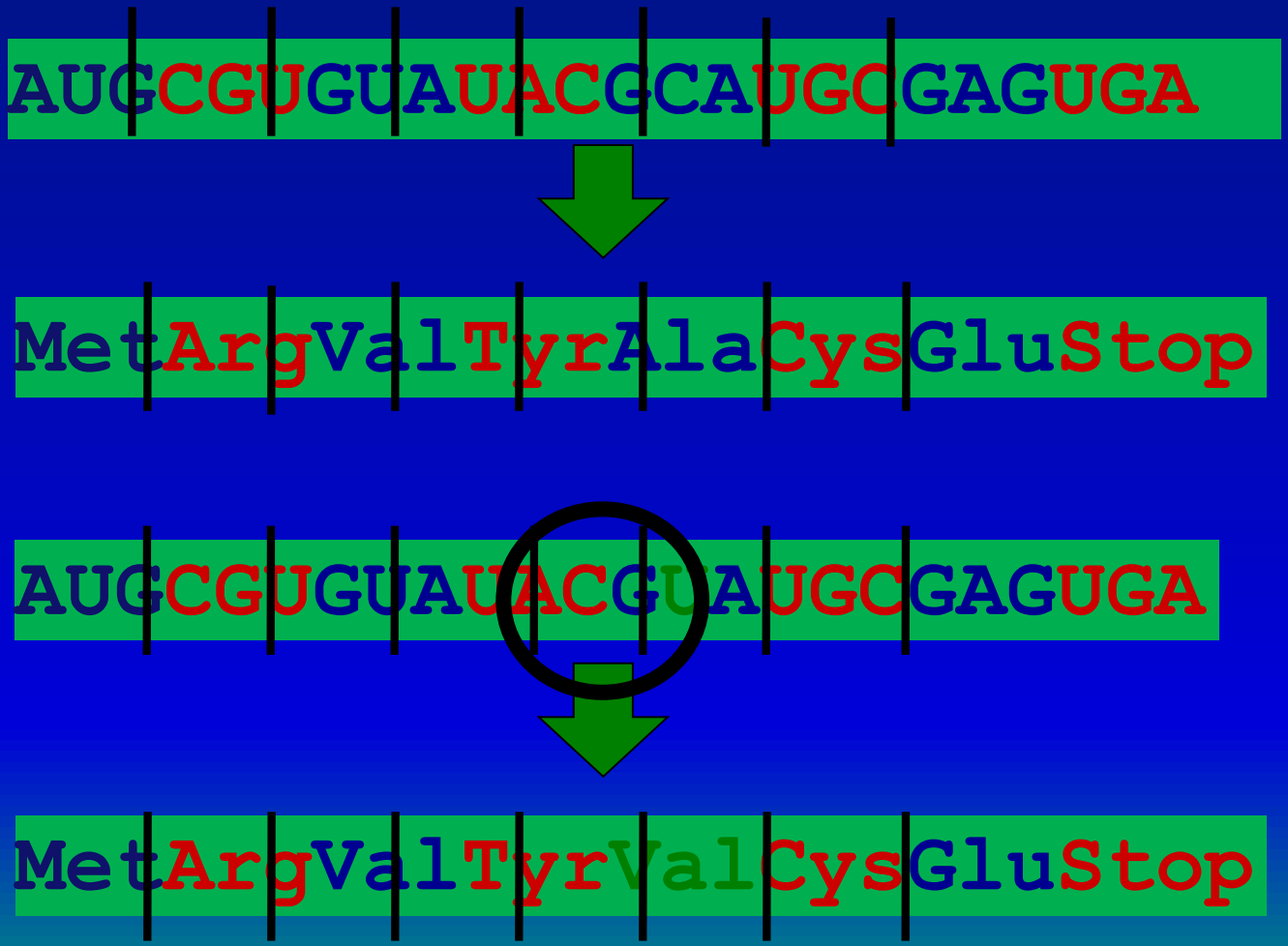
Copy error mechanism

Addition

Deletion

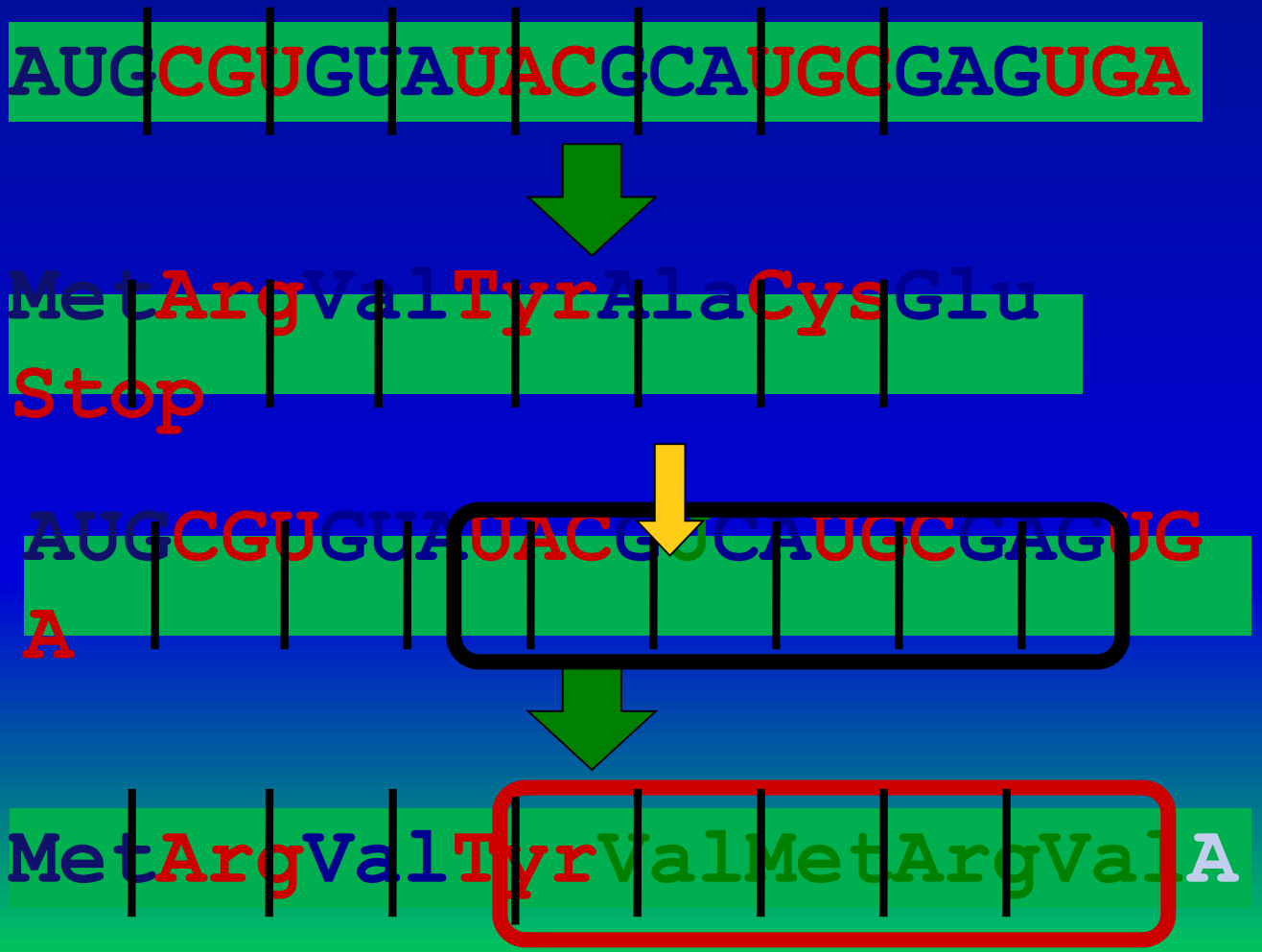
Substitution

Point mutations

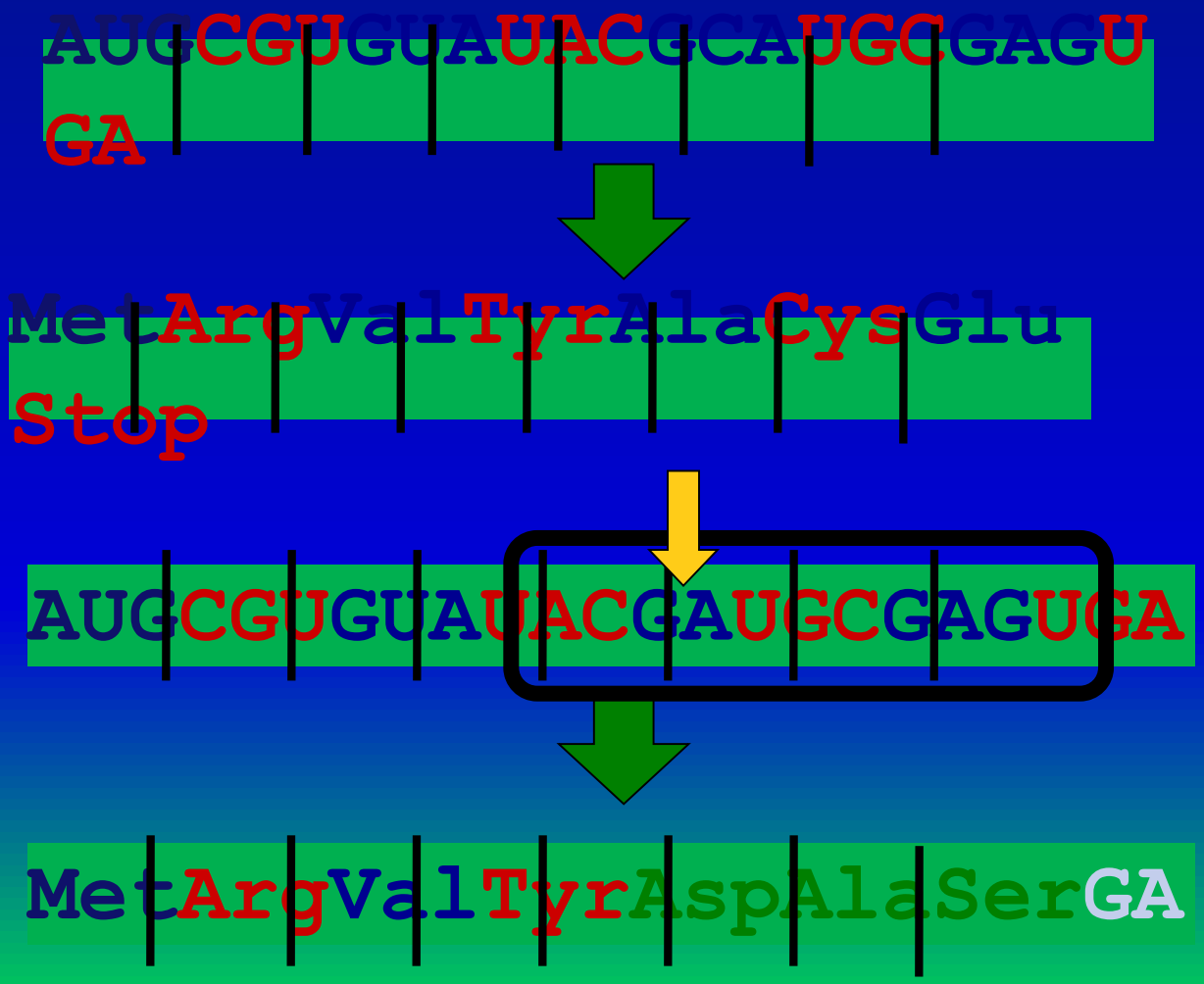


Frameshift Mutations

Addition /insertion= add one or more bases



Deletion = lose one or more bases



Substitution mutation

GGG AGT GTA GAT CGT

CCC TCA CAT CTA GCA

GGG AGT GCA GAT CGT

CCC TCA CAT CTA GCA

A base substitution

First cycle of DNA replication

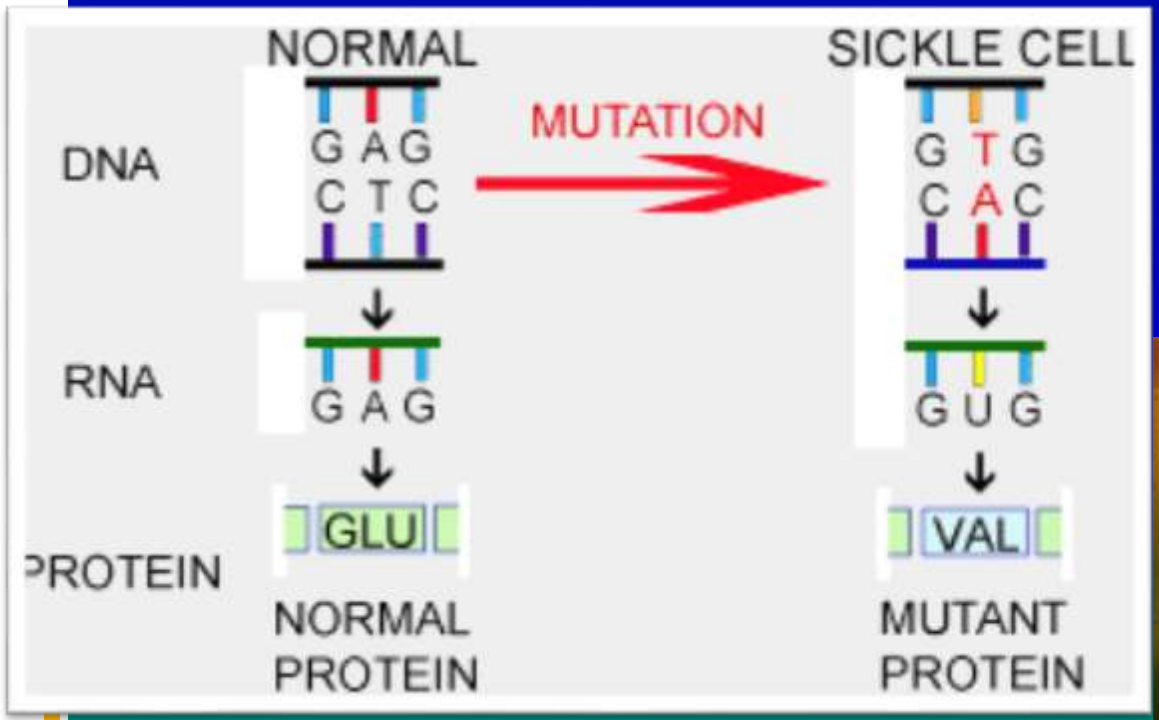
GGG AGT GCA GAT CGT

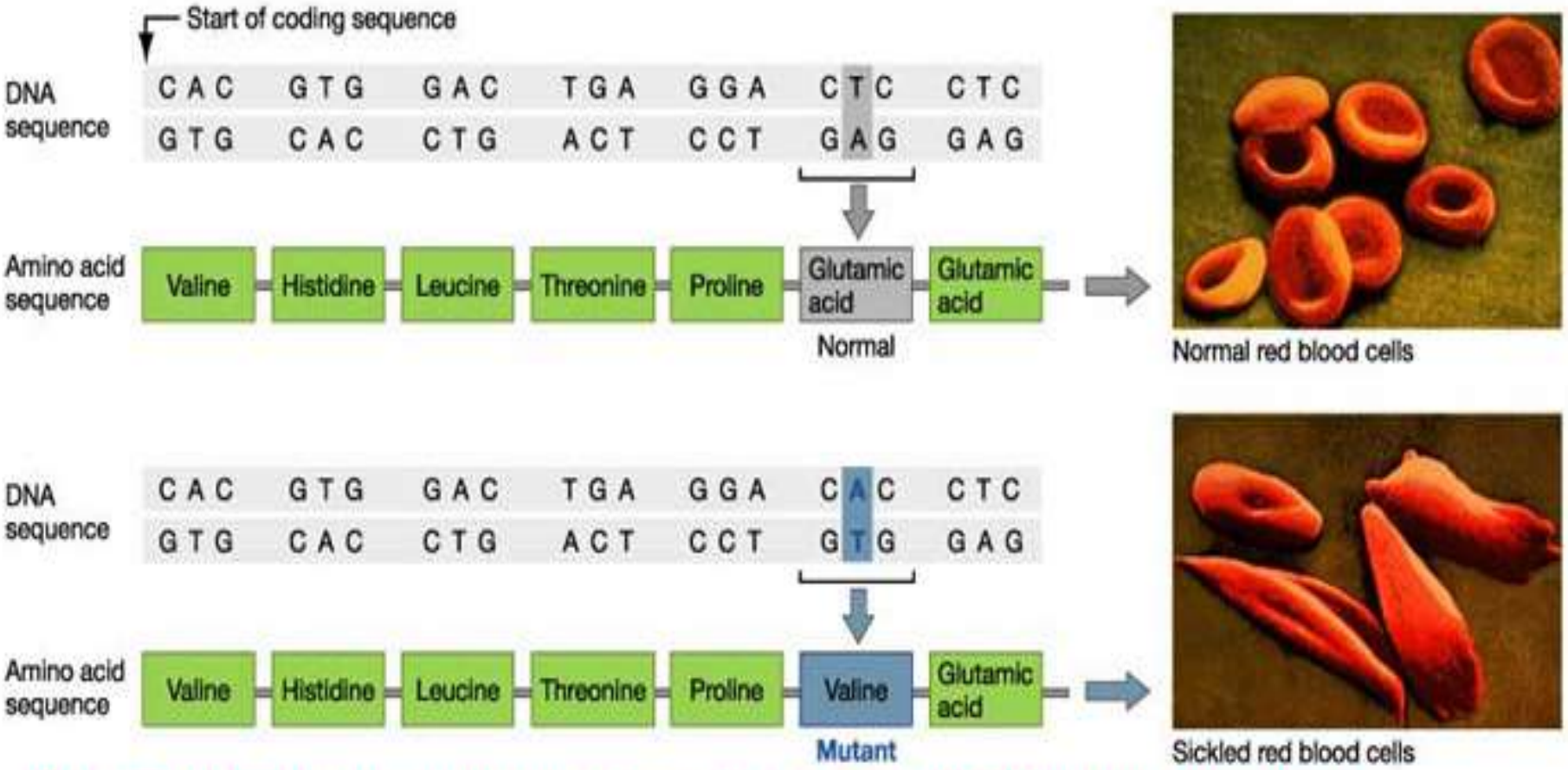
CCC TCA CGT CTA GCA

CCC TCA CAT CTA GCA

GGG AGT GTA GAT CGT

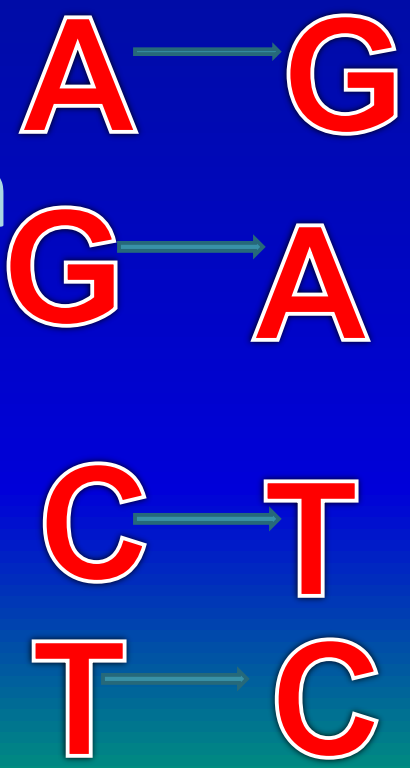
Sickle Cell disease is the result of one nucleotide substitution
Occurs in the **hemoglobin gene**





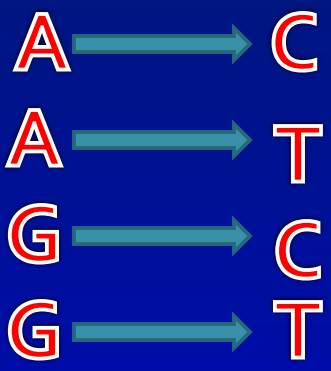
The change in amino acid sequence causes hemoglobin molecules to crystallize when oxygen levels in the blood are low. As a result, red blood cells sickle and get stuck in small blood vessels.

- *Transition:*
- Purine is replaced with a purine
- Pyrimidine is replaced with a pyrimidine

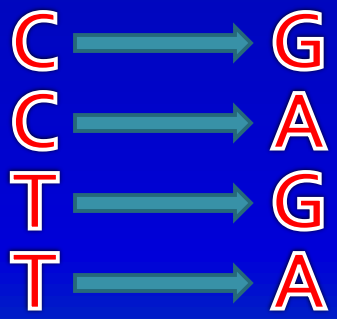


Transversions:

- A purine is replaced by a pyrimidine

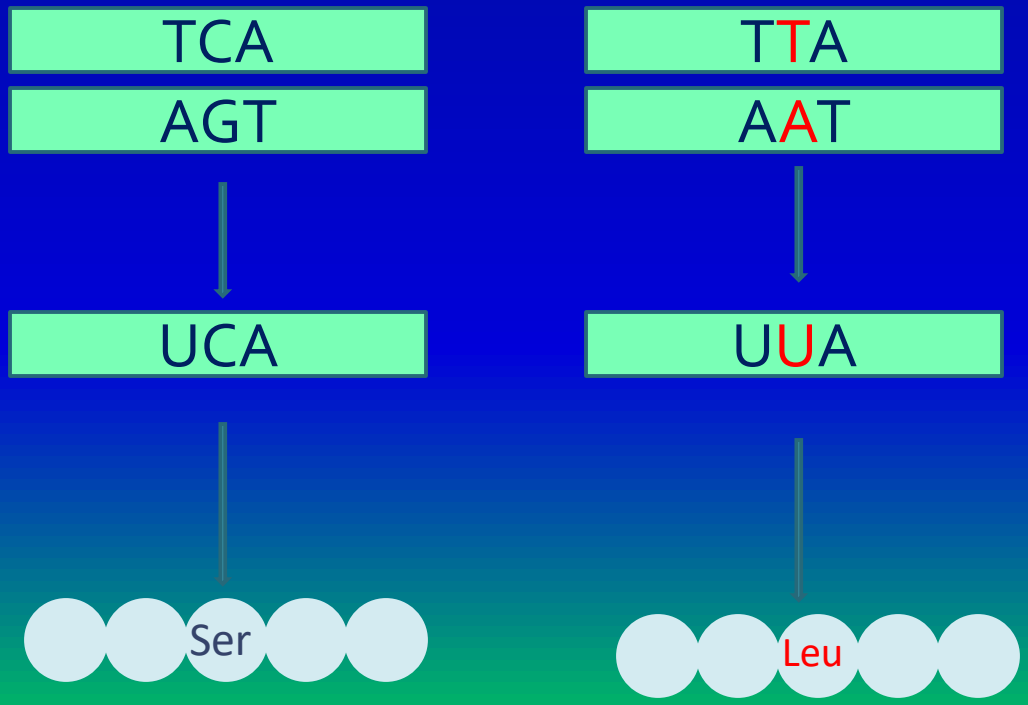


- or a pyrimidine is replaced by a purine

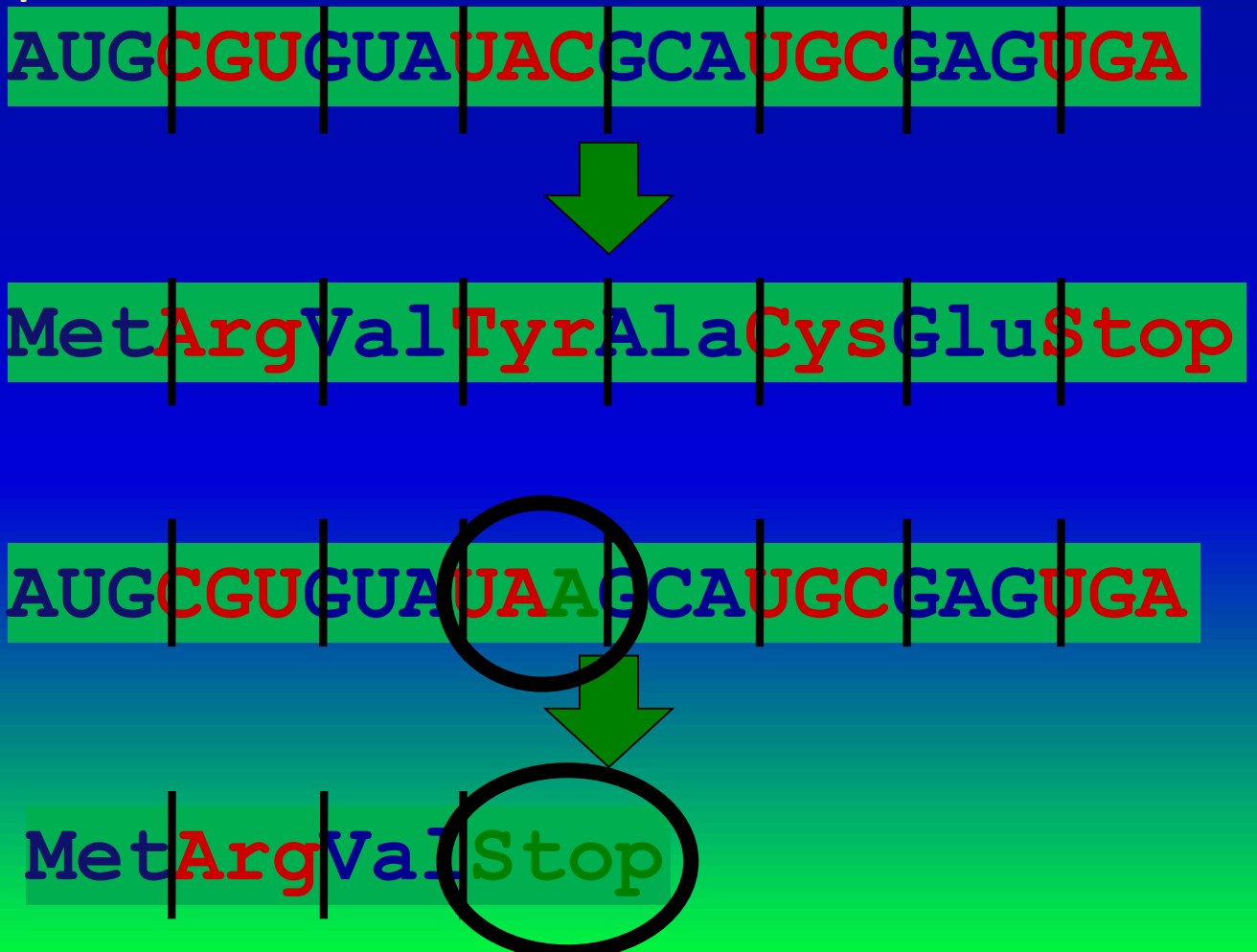


Transitions are usually more frequent *in vivo*

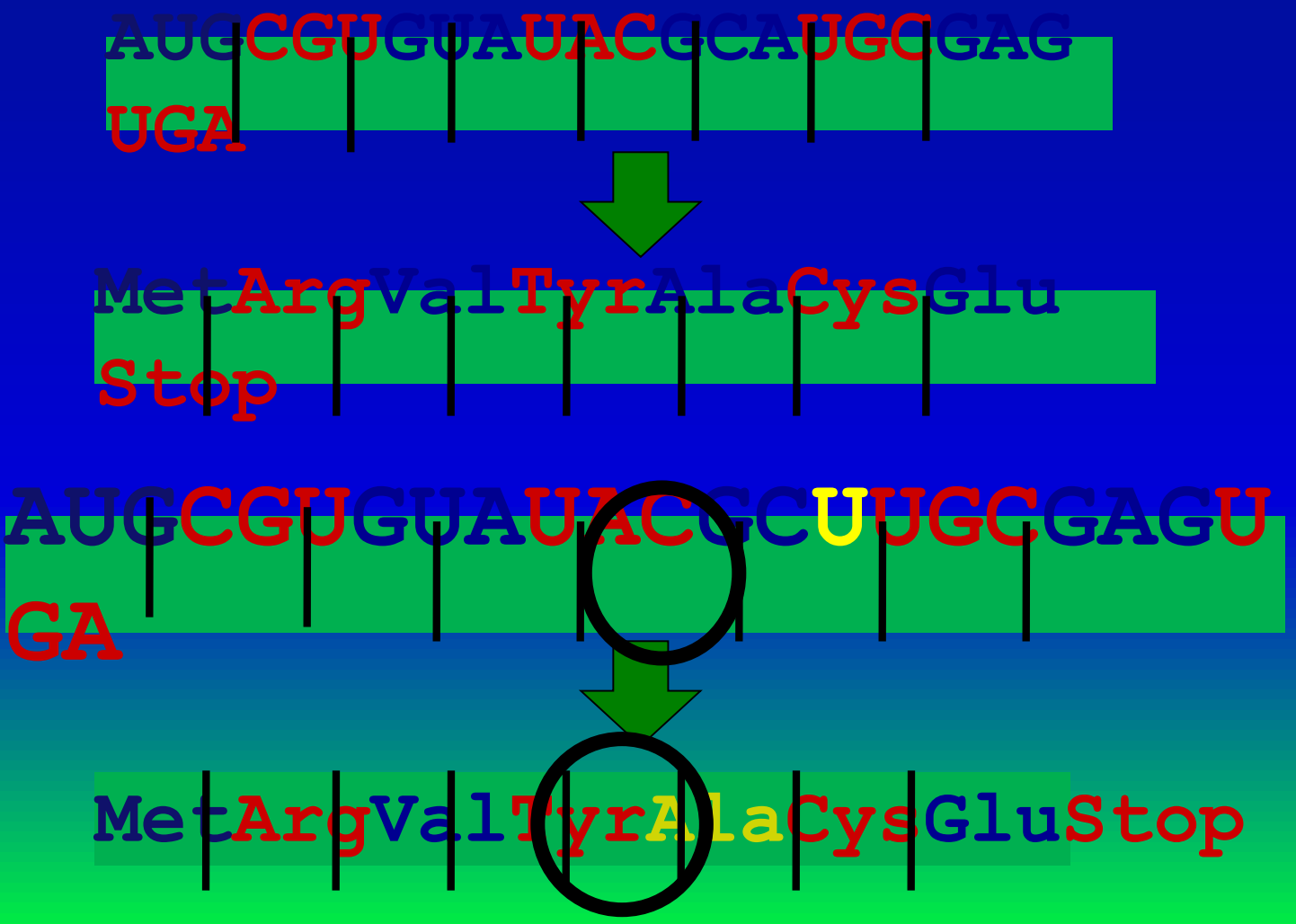
Missense mutation: a base is substituted that alters a codon in the mRNA resulting in a different amino acid in the protein product



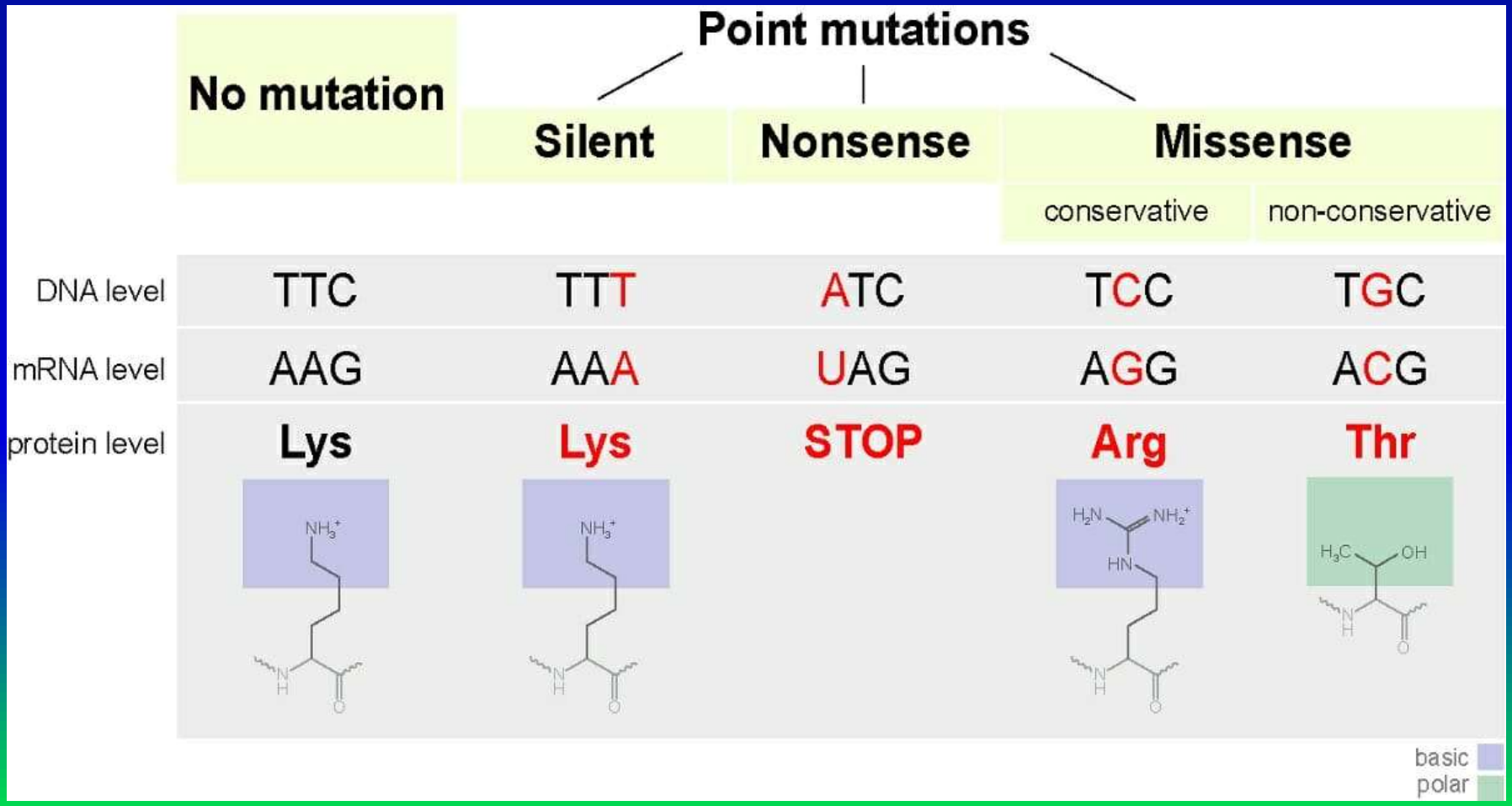
Nonsense mutation: changes a sense codon into a nonsense codon. Nonsense mutation early in the mRNA sequence produces a greatly shortened & usually nonfunctional protein.



Silent mutation: alters a codon but due to degeneracy of the codon, same amino acid is specified

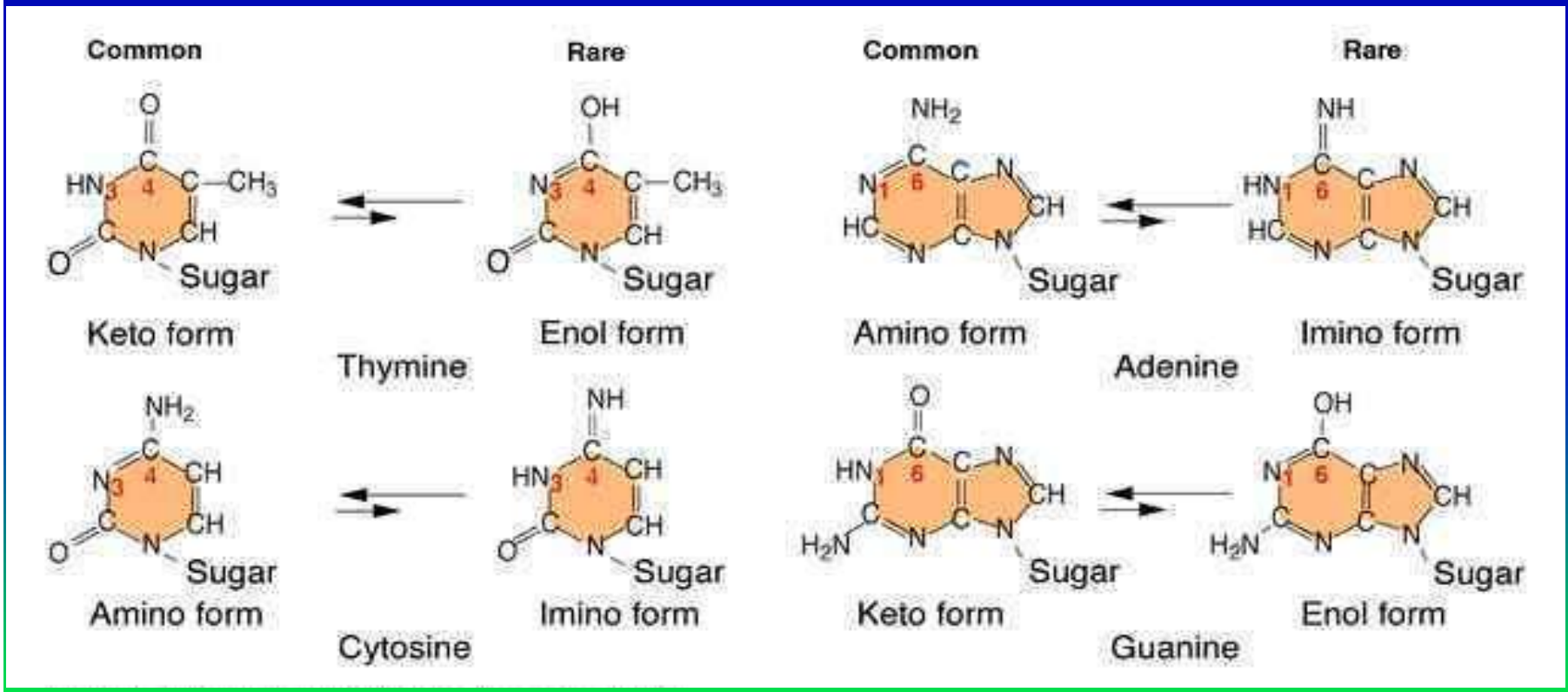


Non sense and mis-sense mutations



Reasons for transition mutations

- **1.Tautomerization**:Tautomers are the alternate forms of bases and are produced by rearrangements of electrons and protons in the molecules

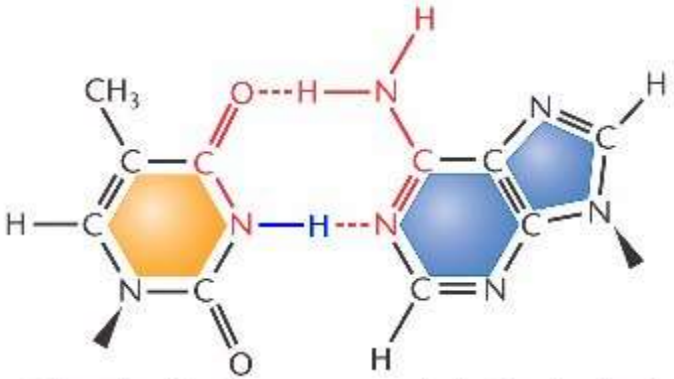


Nucleotides can exist in tautomeric forms (structural isomers) by a position change of a proton (tautomeric shift).

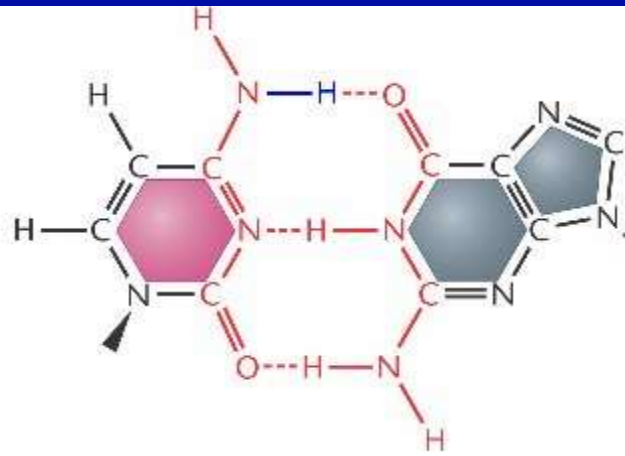
The less common, transient tautomers can form hydrogen bonds with noncomplementary bases.

The anomalous pairing is always between a pyrimidine and a purine, as shown in the T - G and C - A pairs.

(a) Standard base-pairing arrangements



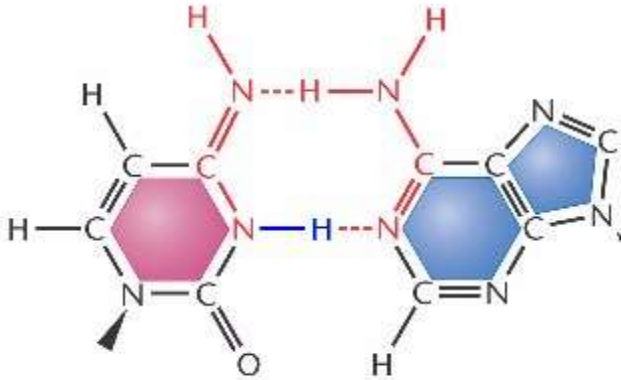
Thymine (keto) Adenine (amino)



Cytosine (amino) Guanine (ke

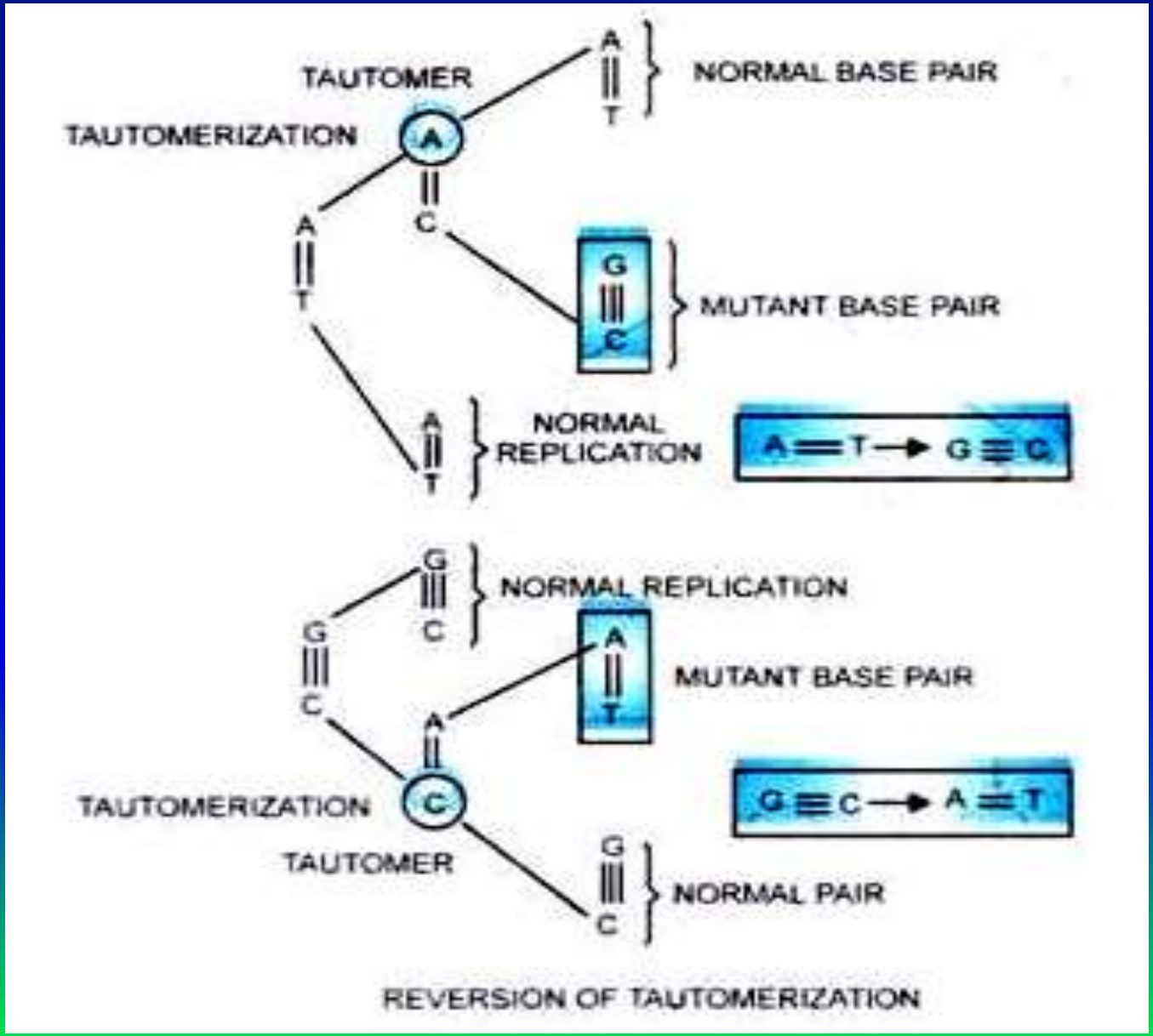
(b) Anomalous base-pairing arrangements

Thymine (enol) Guanine (keto)



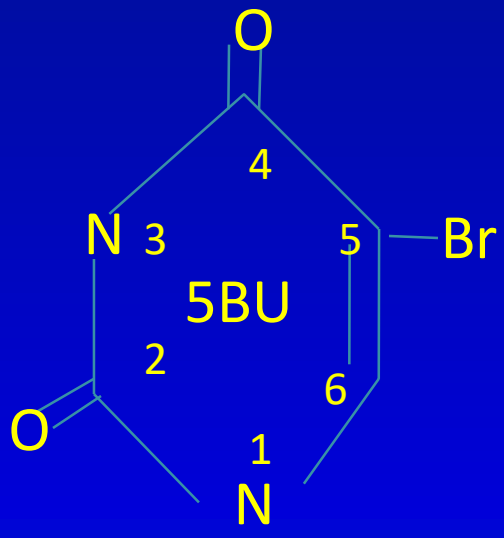
Cytosine (imino) Adenine (ami

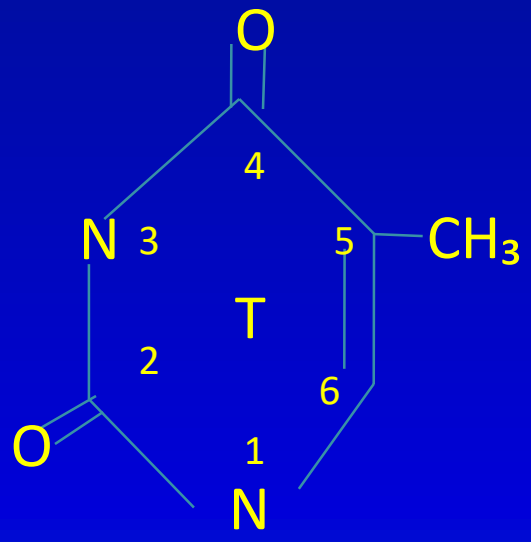
Unusual base pairing or Forbidden base-pair



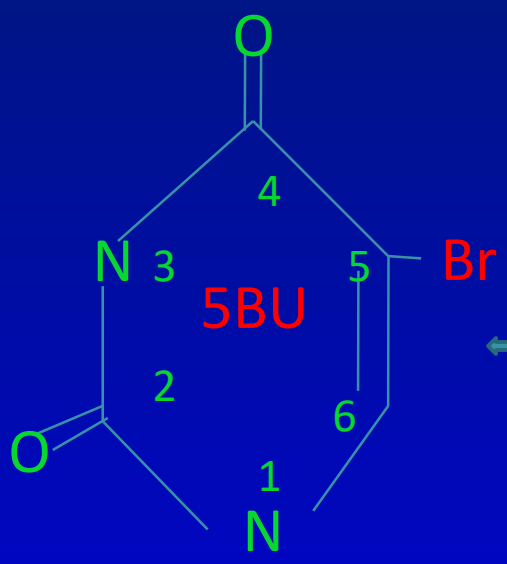
- 2.Ionization
- Ionization causes the loss of hydrogen atoms from the base and such deionised bases involve forbidden base pairing

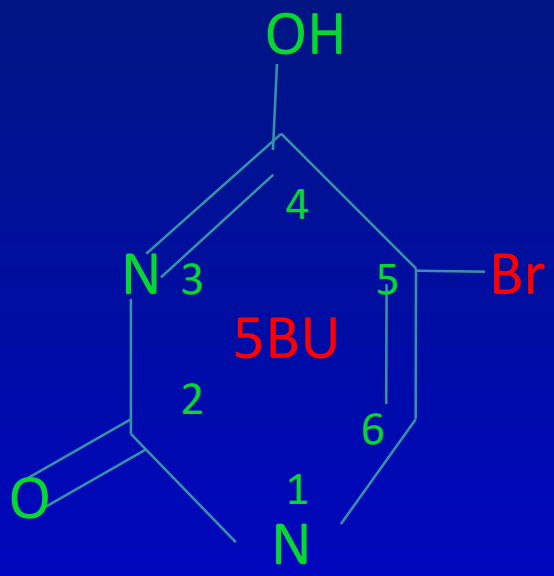
3. Base analogs—substances that are similar to and can substitute for standard bases





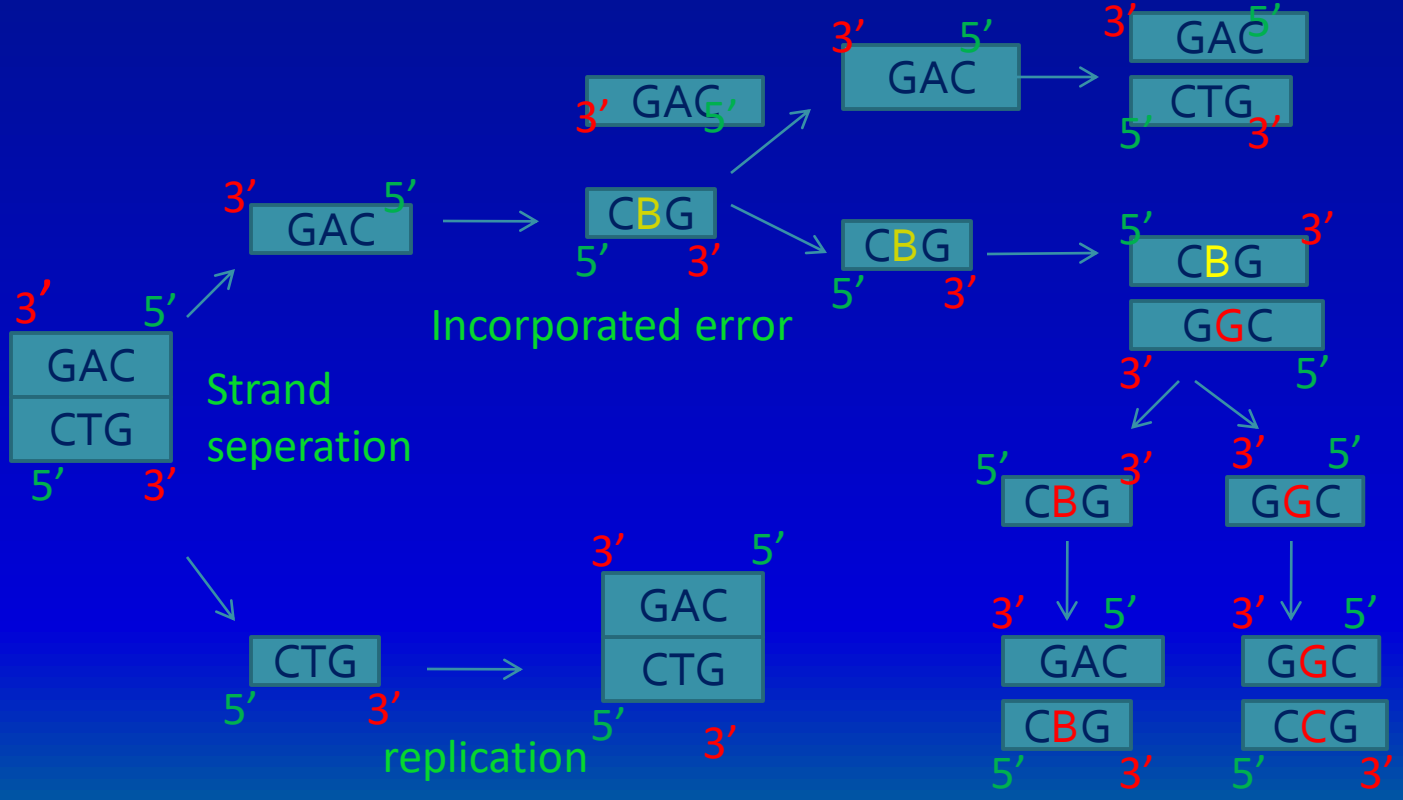
5-bromouracil **an analog of thymine**





Keto
pairs with A

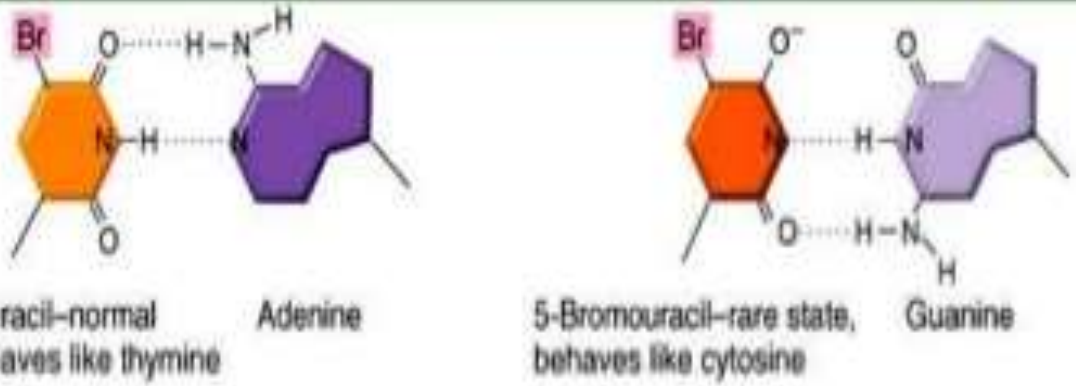
Enol
mispair with G


3′ GAC 5′
CTG
5′ 3′
Strand seperation
3′ GAC 5′
CTG
5′ 3′
3′ GAC 5′
CTG
5′ 3′
Incorporated error
3′ GAC 5′
CBG
5′ 3′
3′ GAC 5′
CBG
5′ 3′
5′ CBG 3′
GGC
3′ 5′
5′ CBG 3′
3′ GGC 5′
3′ GAC 5′
CBG
5′ 3′
3′ GGC 5′
CCG
5′ 3′

Chemical Mutagens

Type of mutagen	Chemical action of mutagen

(a) **Replace a base:** Base analogs have a chemical structure almost identical to that of a DNA base.

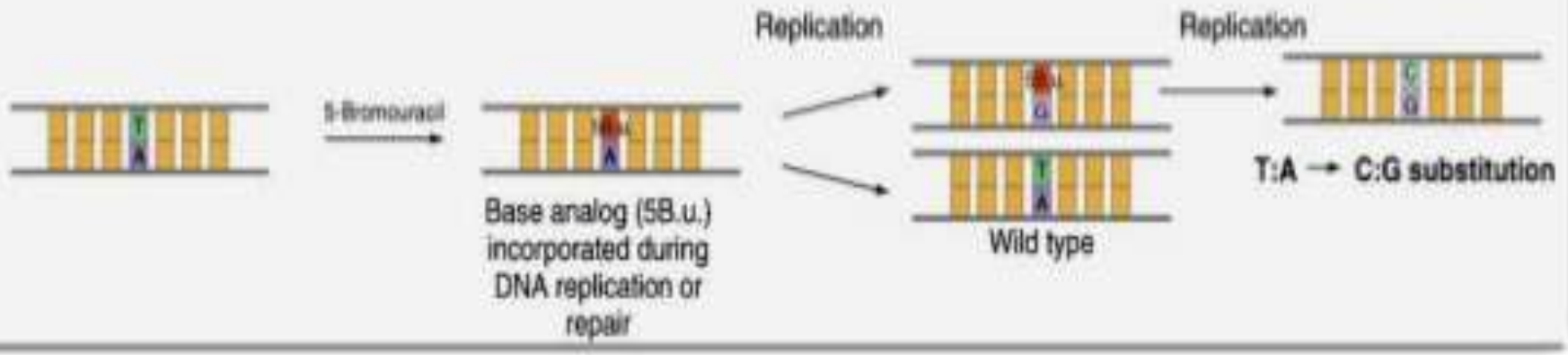


5-Bromouracil–normal state, behaves like thymine — Adenine

5-Bromouracil–rare state, behaves like cytosine — Guanine

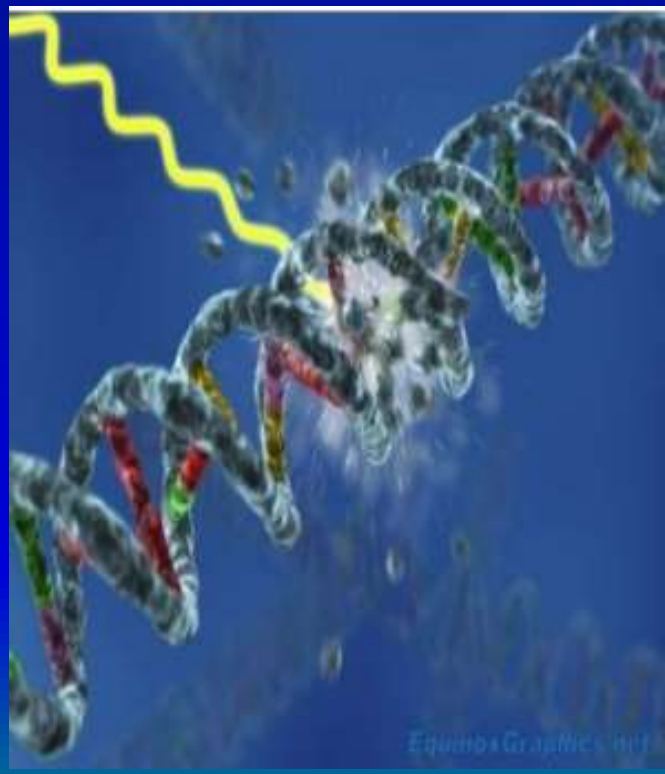
5-Bromouracil: almost identical to thymine. Normally pairs with A; in transient state, pairs with G.

How mutagens induce mutations



Induced mutations

- Mutations induced by non biologic agents
- Process of induction of mutation is called mutagenesis
- Agents which cause mutations are called mutagens

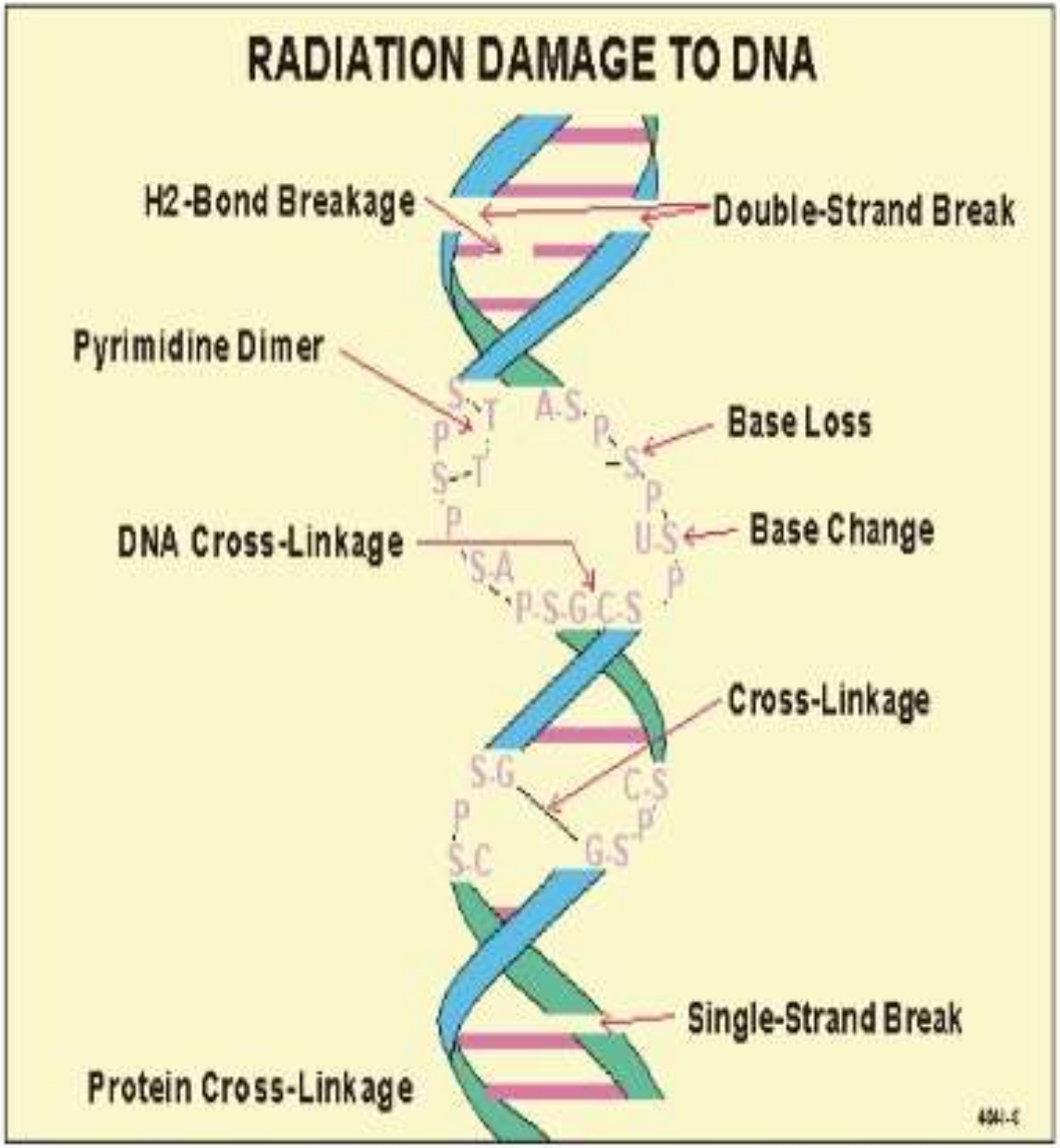


- Physical mutagens
- Chemical mutagens

Physical mutagens

- Ionizing radiations
- Non ionizing radiations

- Ionizing radiation breaks covalent bonds including those in DNA and is the leading cause of chromosome mutations.
- Ionizing radiation has a cumulative effect and kills cells at high doses.
- UV (254-260 nm) causes purines and pyrimidines to form abnormal dimer bonds and bulges in the DNA strands.



Physical Mutagens

Radiation was the first mutagenic agent known; its effects on genes were first reported in the 1920's.

Radiations are of two types.

I. EM radiations

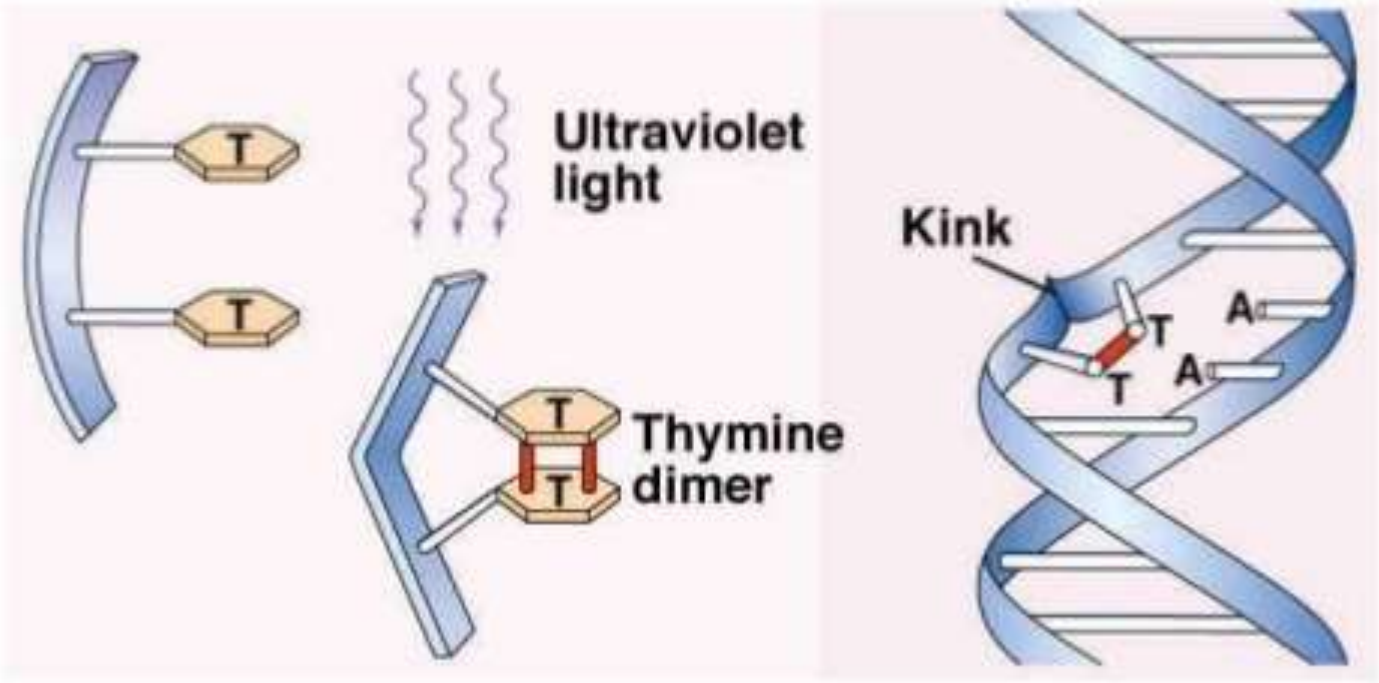
II. Ionizing radiations

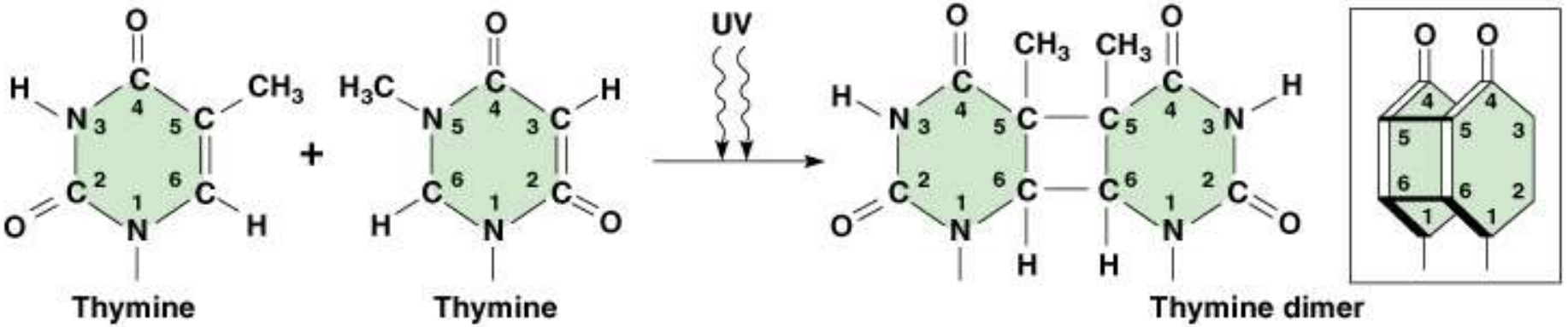
Physical Mutagen

- 1.Electromagnetic Radiations(Non ionizing):
- Visible light and other forms of radiation are all types of electromagnetic radiation (consists of electric and magnetic waves).
- The portion of light which is biologically significant is UV and higher energy radiation.
- UV radiation is not ionizing but can react with DNA and other biological molecules
- UV radiation: Cyclobutane pyrimidine dimers, Thymidine dimers (T-T)

Physical Mutagen

Pyrimidine Dimer




UV
Thymine
Thymine
Thymine dimer

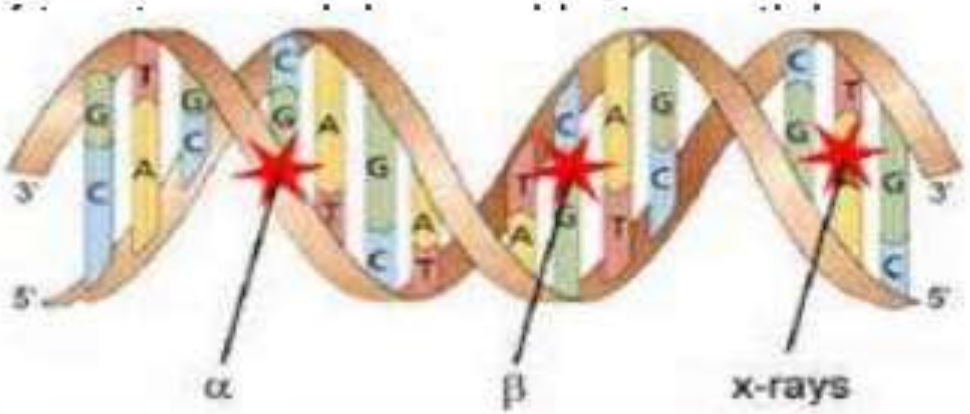
Physical Mutagen

- The rapidly dividing cell types (blood cell-forming areas of bone marrow, gastrointestinal tract lining) are the most affected by ionizing radiation.
- The severity of the effects depends upon the dose received.
- **Ionizing** radiation produces a range of effects on DNA both through free radical effects and direct action:
  - -breaks in one or both
  - -damage to/loss of bases (mutations)
  - -crosslinking of DNA to itself or proteins

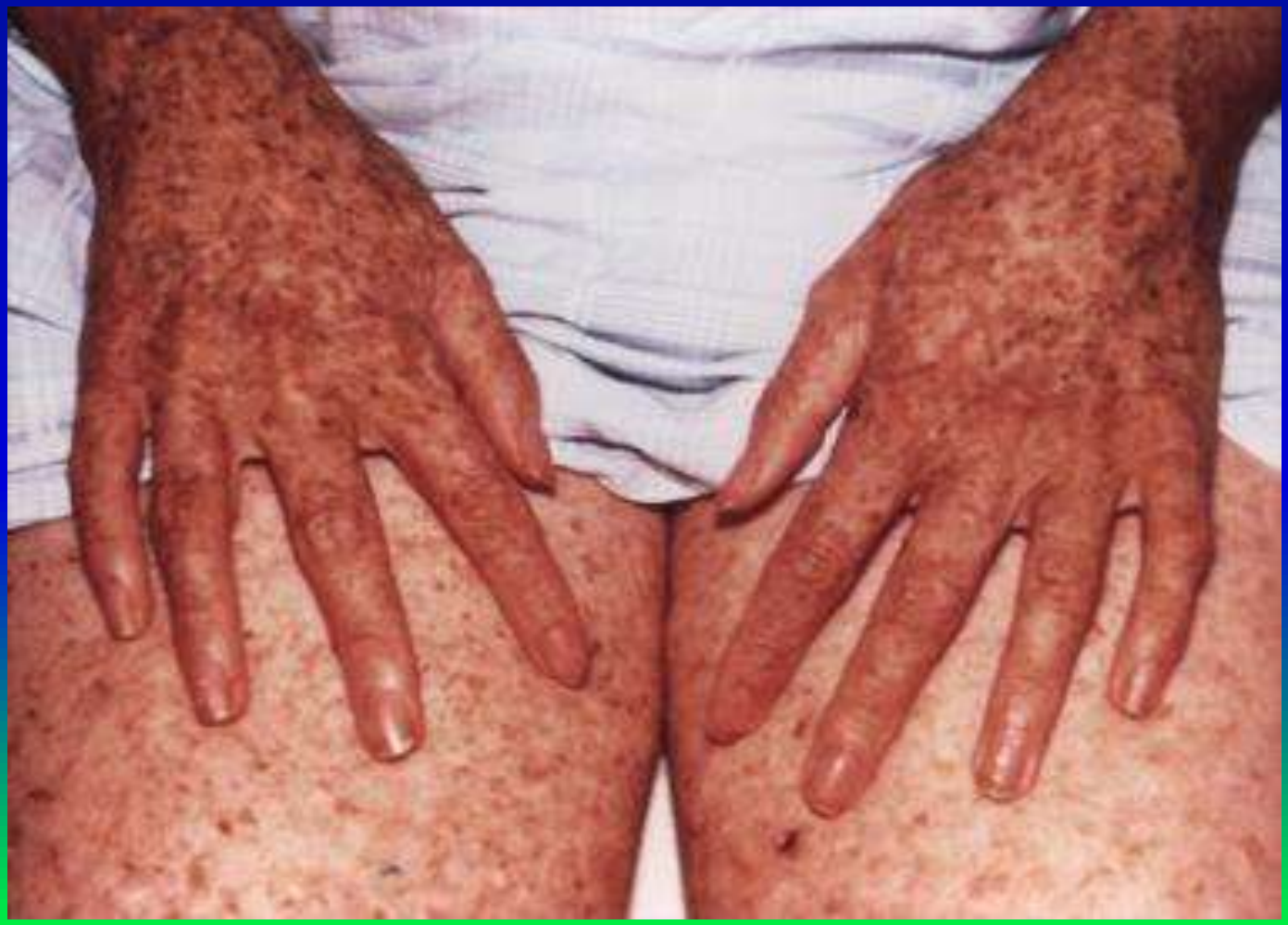
Physical Mutagen

- 2. Ionizing Radiatons:

- X- and gamma-rays.
- Produce reactive ions (charged atoms or molecules) which react with biological molecules.
- Damage base and sugar residues.
- This term also includes corpuscular radiation-
- streams of atomic and subatomic particles emitted by radioactive elements:
- these a



- Photo reactivation
- Dark repair
- Excision Repair



Chemical mutageneis

Base analogues

Alkylating agents:

Chemicals that donate alkyl groups

e.g.

Ethylmethane sulfonate(EMS)

It adds an ethyl group to guanine and produces 6-ethylguanine, which pairs with thymine and leads to CG:TA transitions

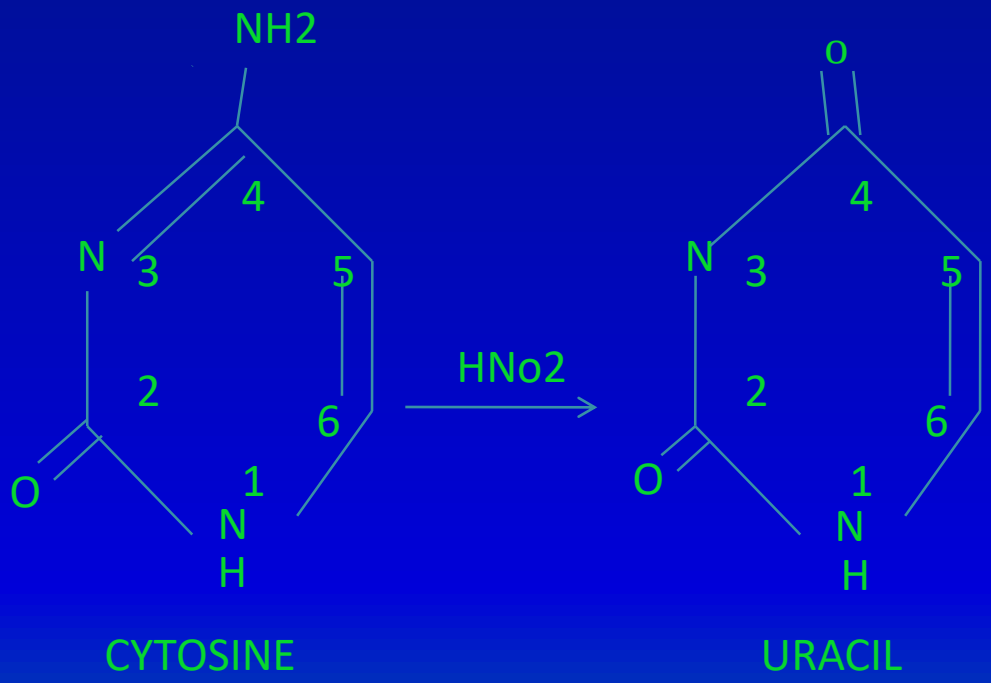
Also adds an ethyl group to thymine to produce 4~ethylthymine, which then pairs with guanine, leading to a TA:CG transition

Mutations produced by EMS can be reversed by additional treatment with EMS.

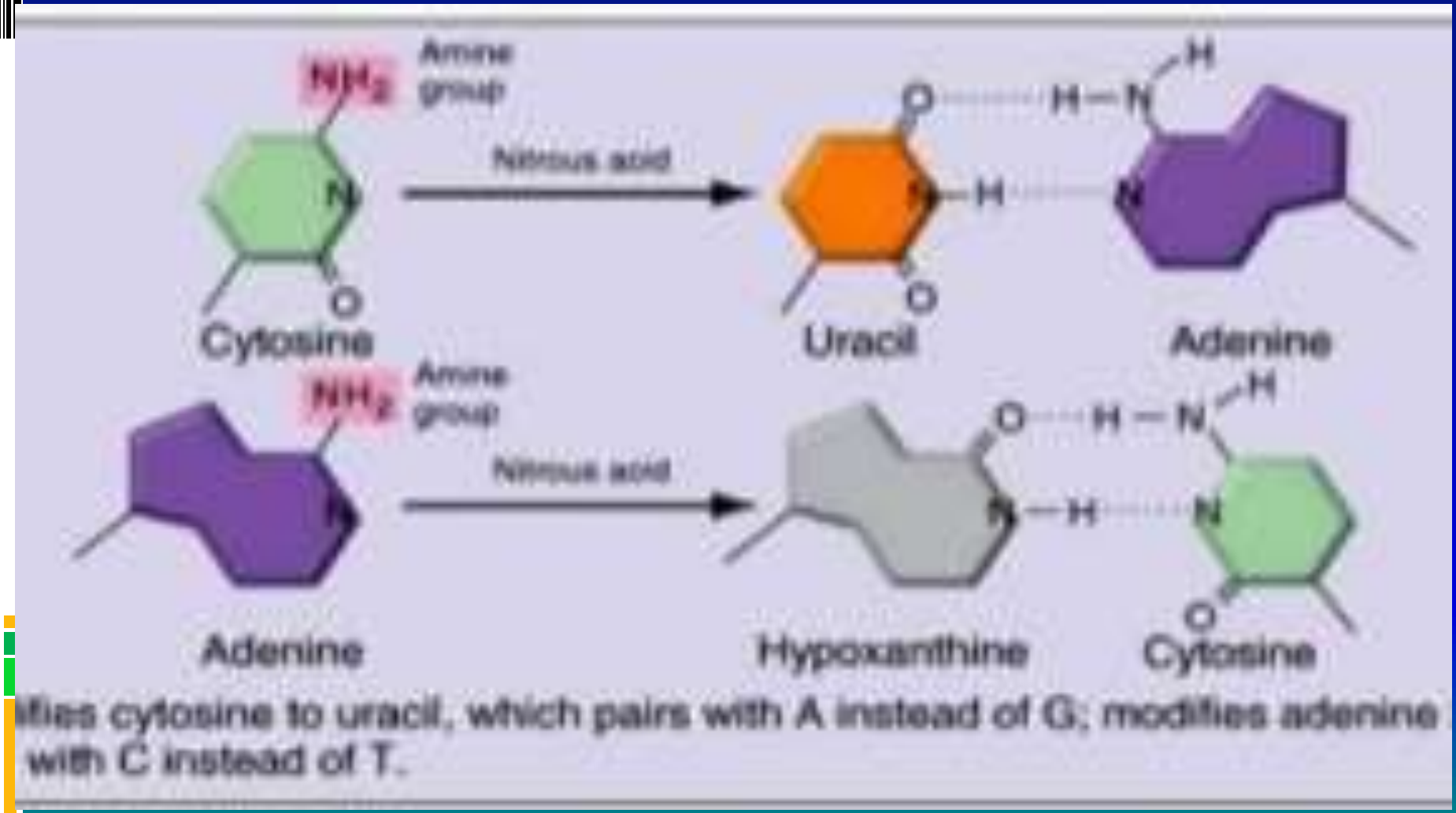
Methyl methane suphonate(MMS), Dimethyl sulphonate(DMS), Mustard gas are alkylating agent.

Deamination Agents

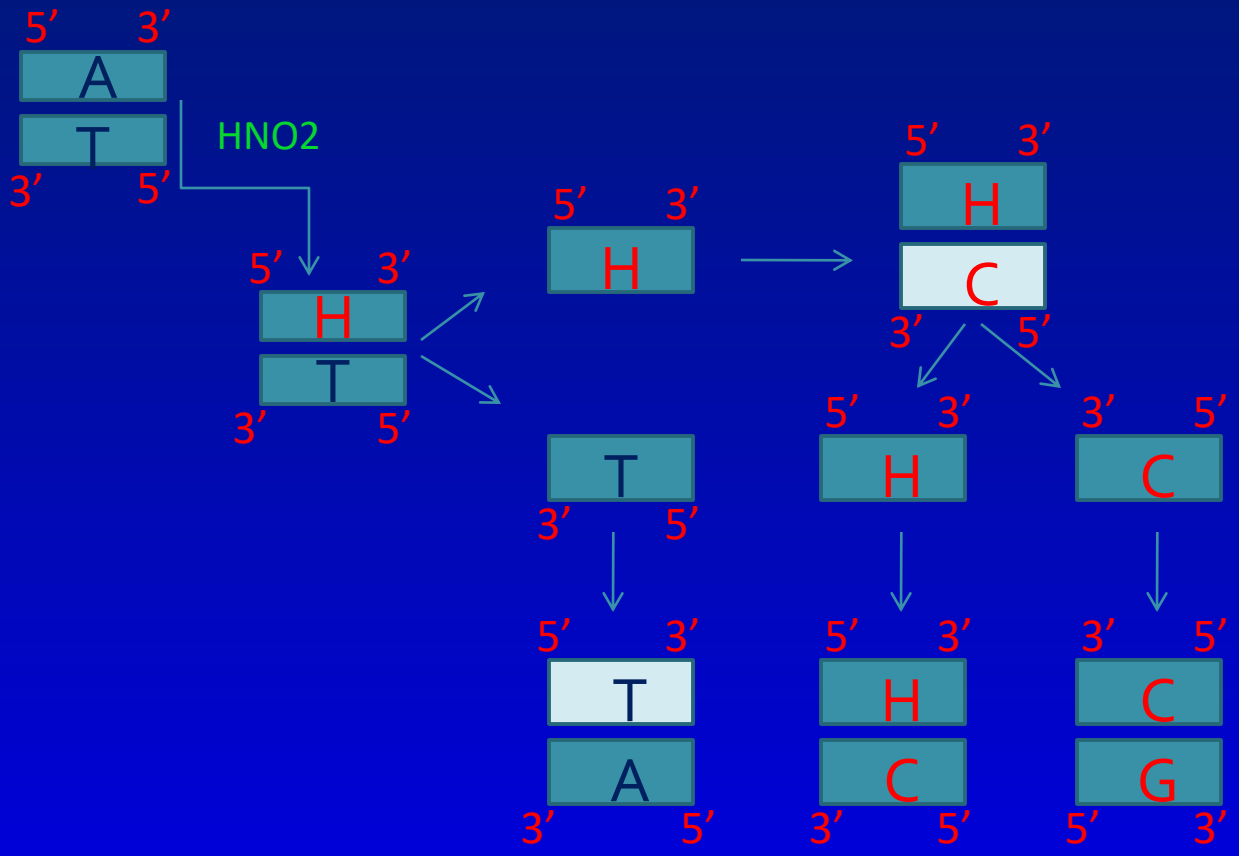
Nitrous acid: causes deamination



CYTOSINE URACIL



ifies cytosine to uracil, which pairs with A instead of G; modifies adenine with C instead of T.



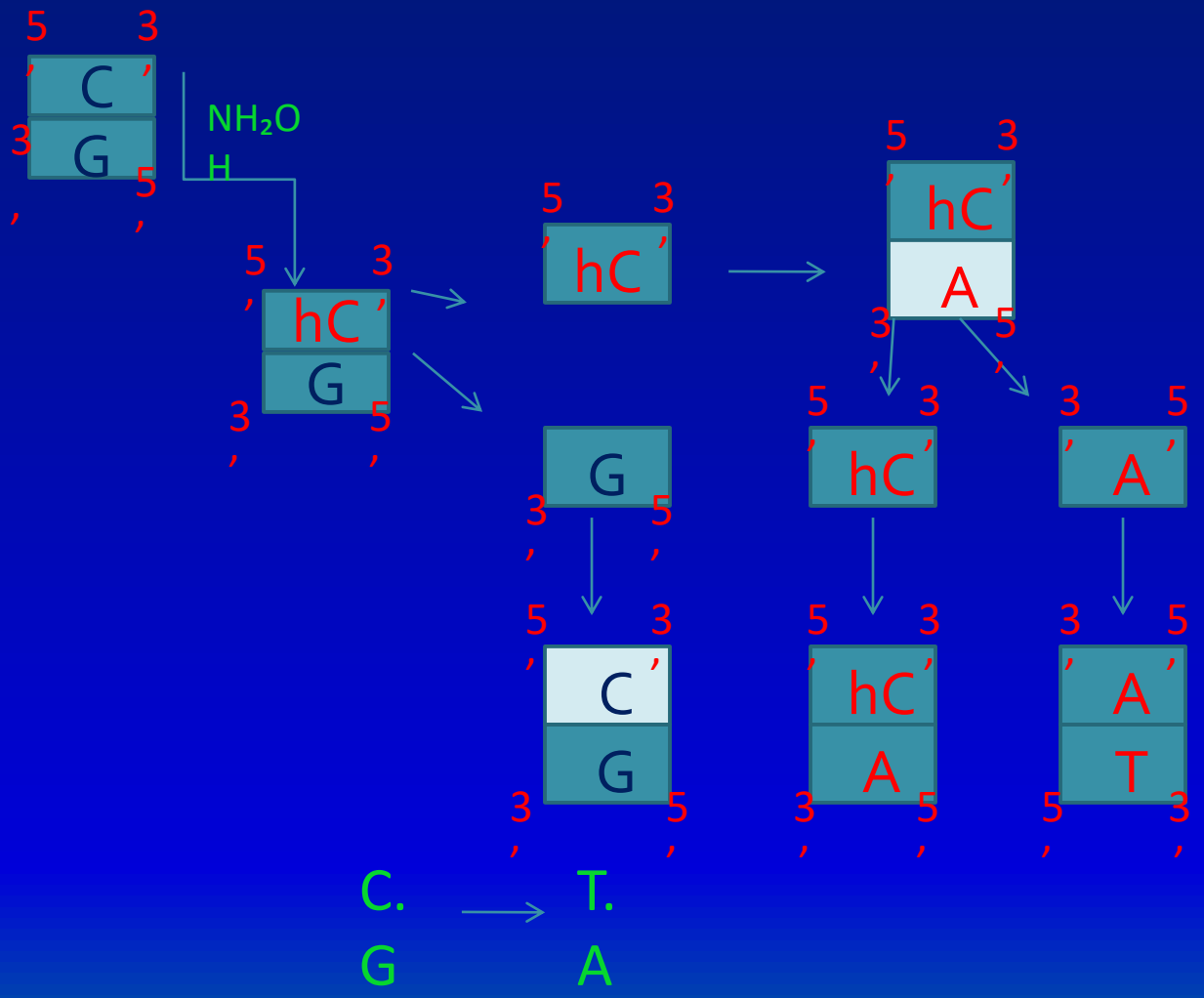
Adenine changes into Hypoxanthin which then pairs with Cytosine

Hydroxyl amine

Specific base modifying mutagen which adds a hydroxyl group to cytosine producing hydroxylamine cytosine which pairs with adenine instead of guanine

This Leads to C.G-T.A transitions

Acts only on cytosine thus can not revert the mutation produced



Cytosine changes into hydroxlamine Cytosine which pairs with Adenine instead of Guanine

Intercalating agents

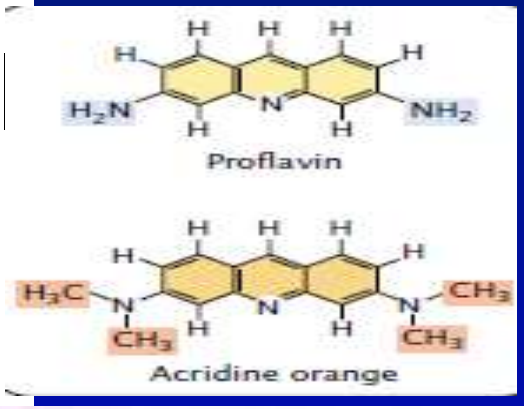
Proflavin, acridine orange, ethidium bromide, and dioxin

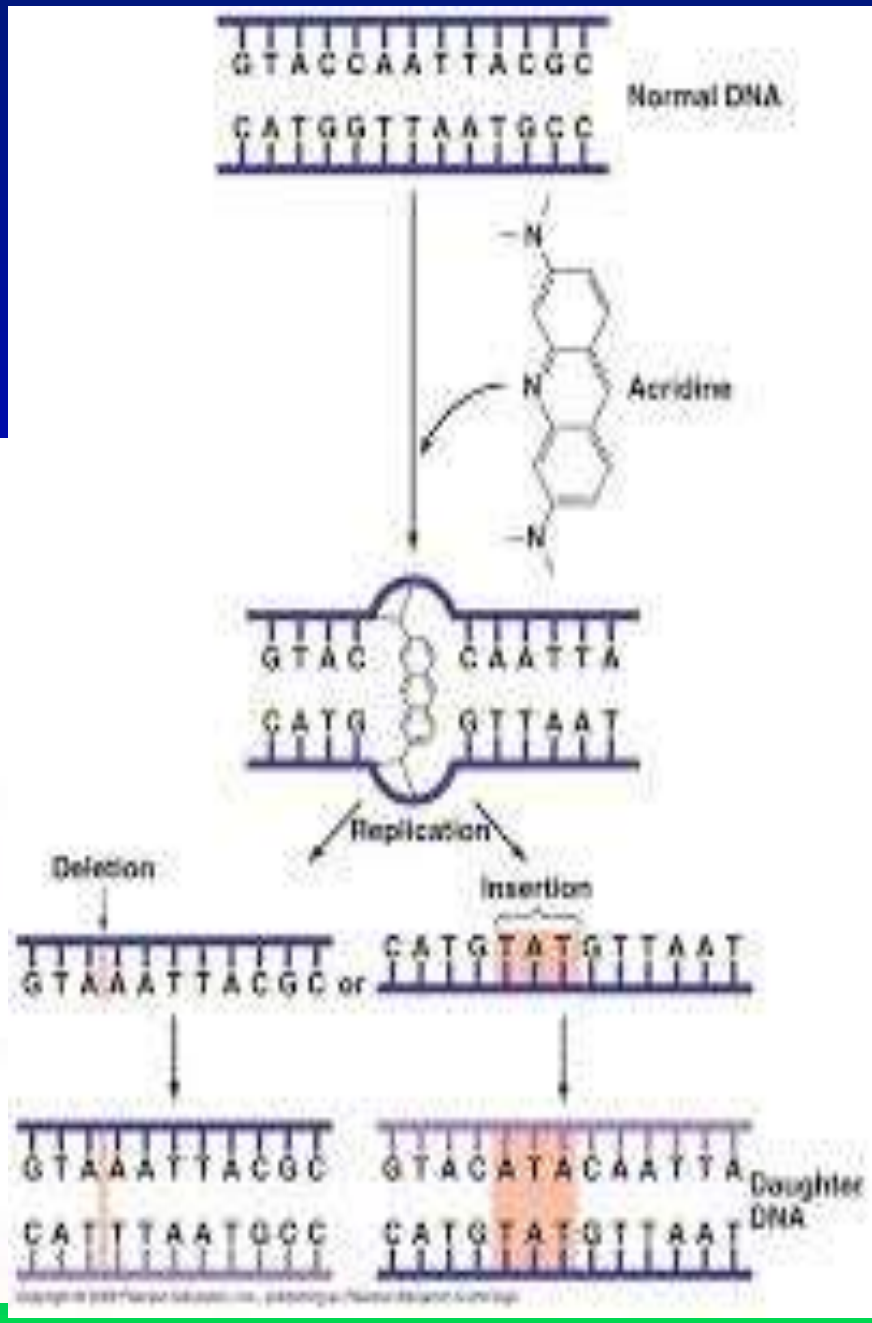
They are about the same size as a nucleotide

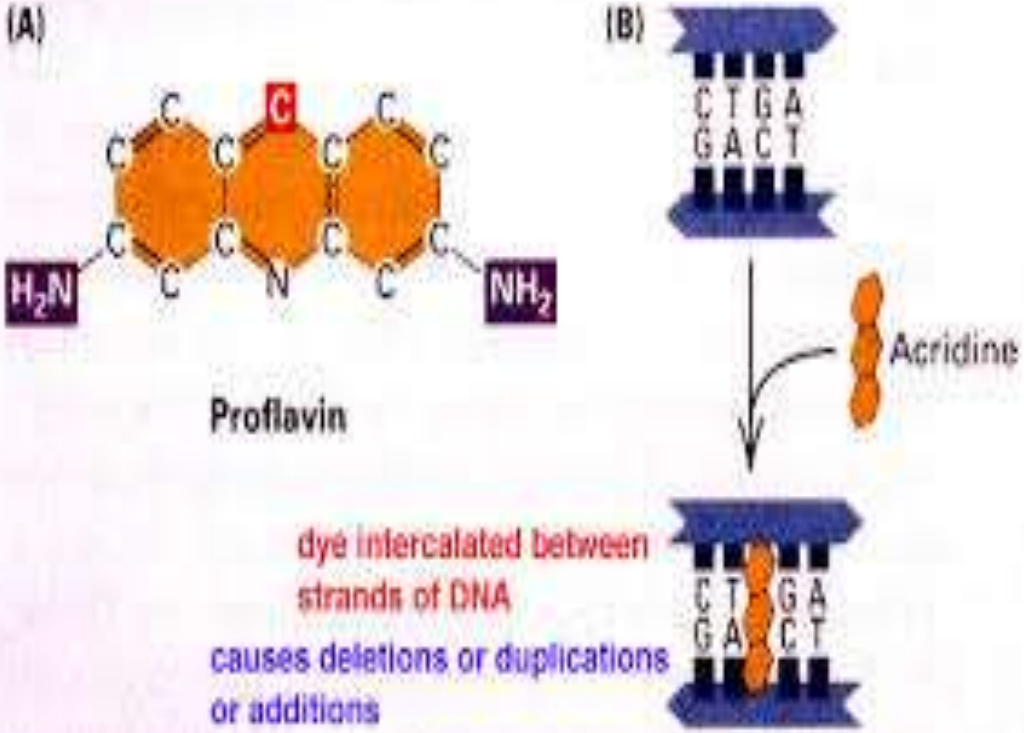
They produce mutations by sandwiching themselves (intercalating) between adjacent bases in DNA

They distort the three-dimensional structure of the helix and cause single-nucleotide insertions and deletions in replication

These insertions and deletions frequently produce frame shift mutations


Proflavin
Acridine orange


GTACCAATTACGC
CATGGTTAATGCC
Normal DNA
Acridine
GTAC
CAATTA
CATG
GTTAAT
Replication
Deletion
Insertion
GTAAATTACGC or
CATGTATGTTAAT
GTAAATTACGC
CATTTAATGCC
GTACATACAATTA
CATGTATGTTAAT
Daughter DNA


(A)
(B)
Proflavin
Acridine
dye intercalated between strands of DNA
causes deletions or duplications or additions

This class prepared for
Fifth Semester BSc Botany Students
Little Flower College, Guruvayur
Affiliated to University of Calicut

Thank You