Genomic Variations II
Type of variants

Sep 3, 2022

Objectives

By the end of the session, the students should be able to:

1. Define genomic variations
2. Describe the changes in genome using the standard terminologies
3. Distinguish between chromosomal abnormalities and gene abnormalities
4. Recognize types of single nucleotide variant
5. Explain the process of mutagenesis

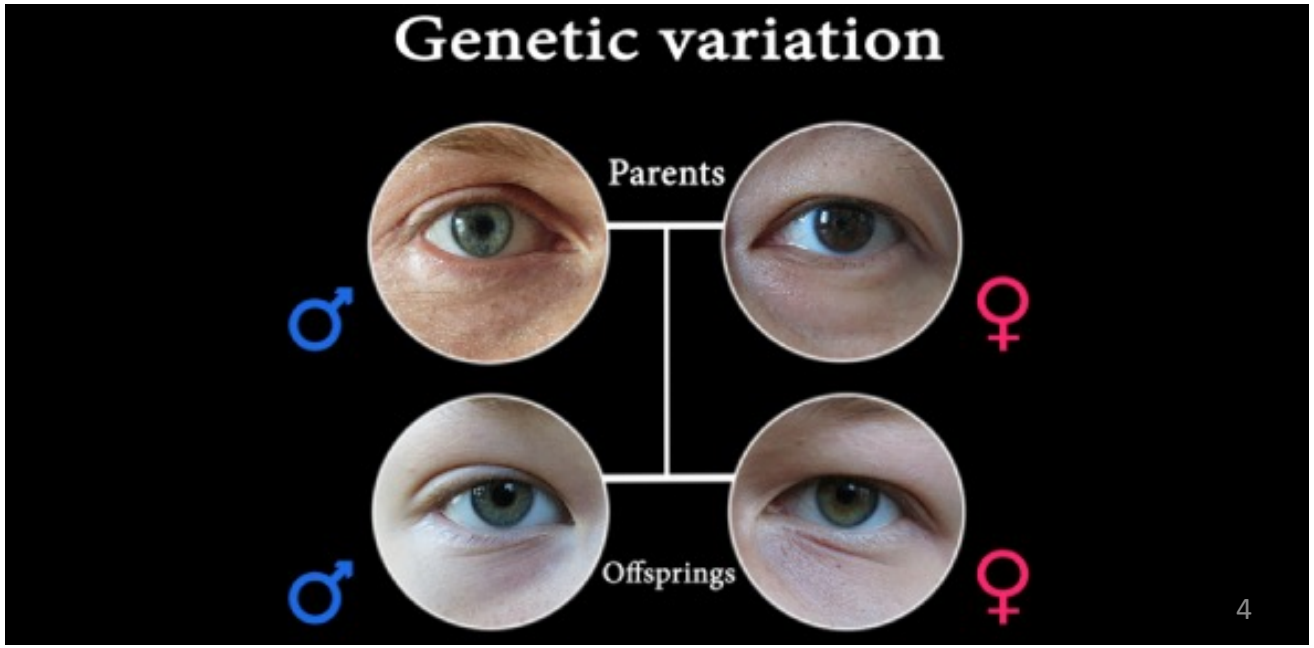
Outline

- Genomic variation
- Spectrum of variants
- Type of pathogenic variants
- Mutagenesis

What is genomic variation?

- Variation= Differences between organisms
- Genomic variations= Differences between individuals caused by alterations in their genomes

Example:



Why genomic variation?

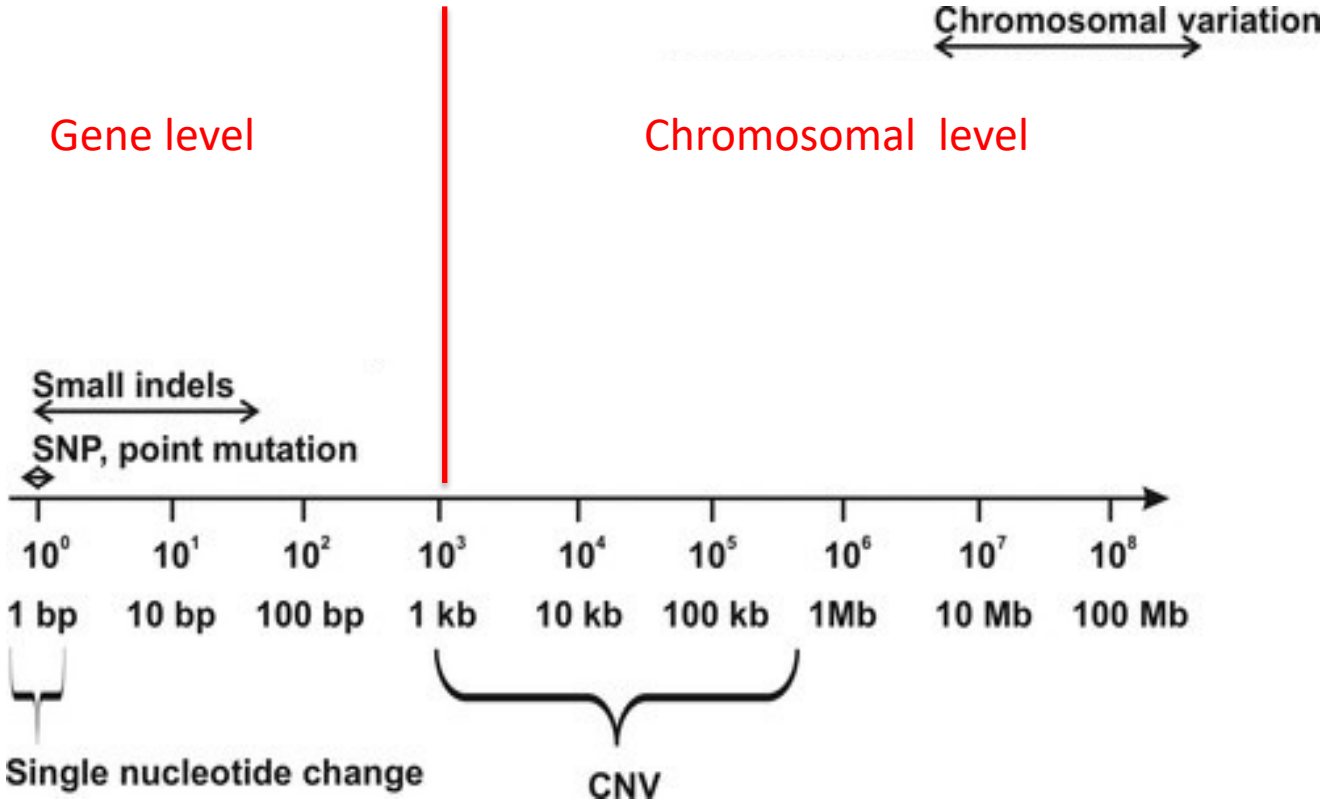
Normal genomic variations needed for:

- Better survival and reproduction
- Adaptation to live in a new environment

Example:

Beta haemoglobin gene alteration resulting in HbS i.e sickle cell trait (carrier) in African population→ built resistance to Malaria→ survival of African population during Malaria pandemic

Types of variants according to the size:

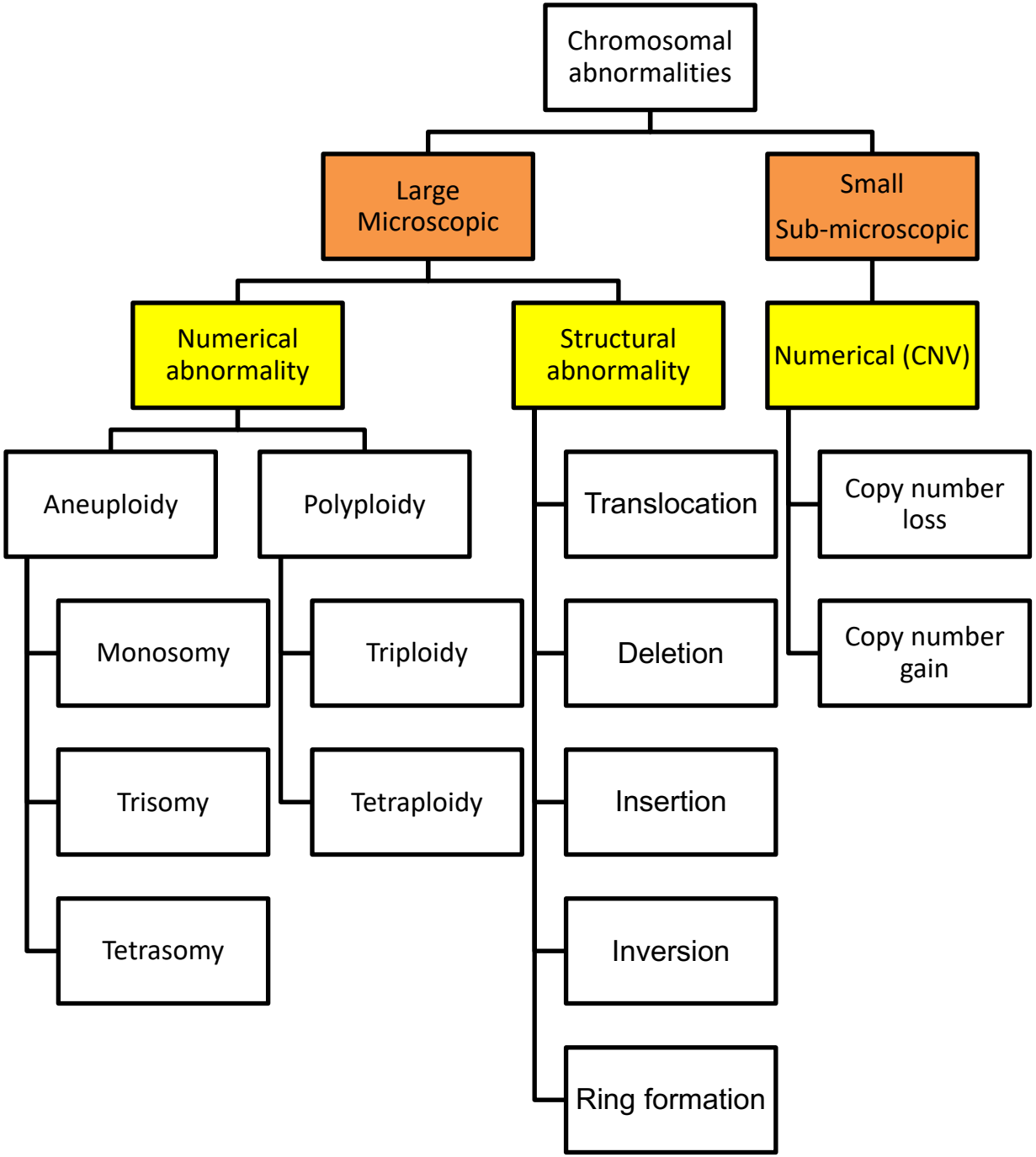


The spectrum of variation in the human genome. A logarithmic *x*-axis measures the number of nucleotides, from 1 bp to ≥100 Mb. Above the axis, types of genetic variation are shown, with their size range depicted below by a double-headed arrow. Size ranges are not definitive. SNP indicates single-nucleotide polymorphism; indels, insertions and deletions; STR, short tandem repeat; bp, base pair; kb, kilobase; Mb, megabase; and CNV, copy number variation.

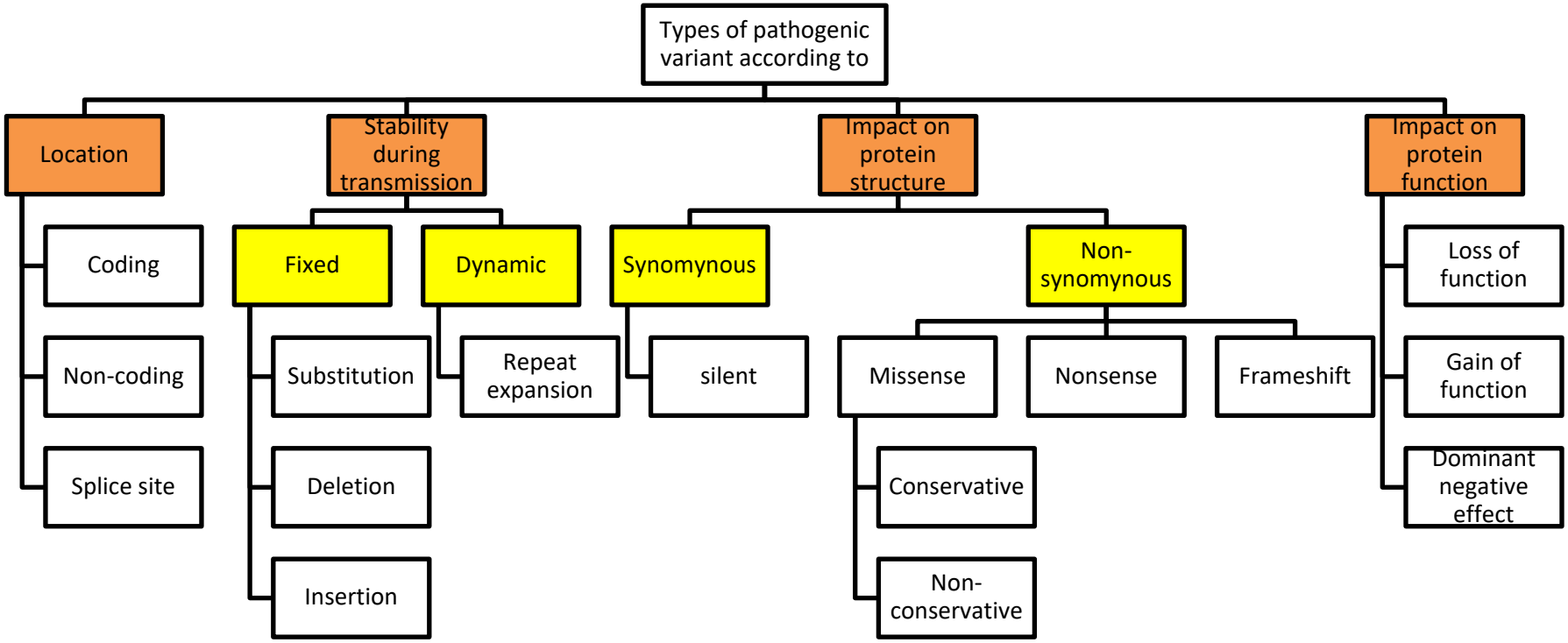
Rebecca L. Pollex. Circulation. Copy Number Variation in the Human Genome and Its Implications for Cardiovascular Disease, Volume: 115, Issue: 24, Pages: 3130-3138, DOI: (10.1161/CIRCULATIONAHA.106.677591)

What are the abnormal genomic variants?

- Abnormal genomic variation causes unfavourable outcome or disorder.
- These abnormalities could be:
  - At Chromosomal level
  - At gene level


Chromosomal abnormalities
Large Microscopic
Small Sub-microscopic
Numerical abnormality
Structural abnormality
Numerical (CNV)
Aneuploidy
Polyploidy
Translocation
Copy number loss
Monosomy
Triploidy
Deletion
Copy number gain
Trisomy
Tetraploidy
Insertion
Tetrasomy
Inversion
Ring formation

Variants at gene level



Variants at gene level

- Variant classes according to its significance (variant classification):
  - Pathogenic
  - Likely pathogenic
  - Variant of unknown significance
  - Likely benign
  - Benign

Mutation

Polymorphism (SNP)

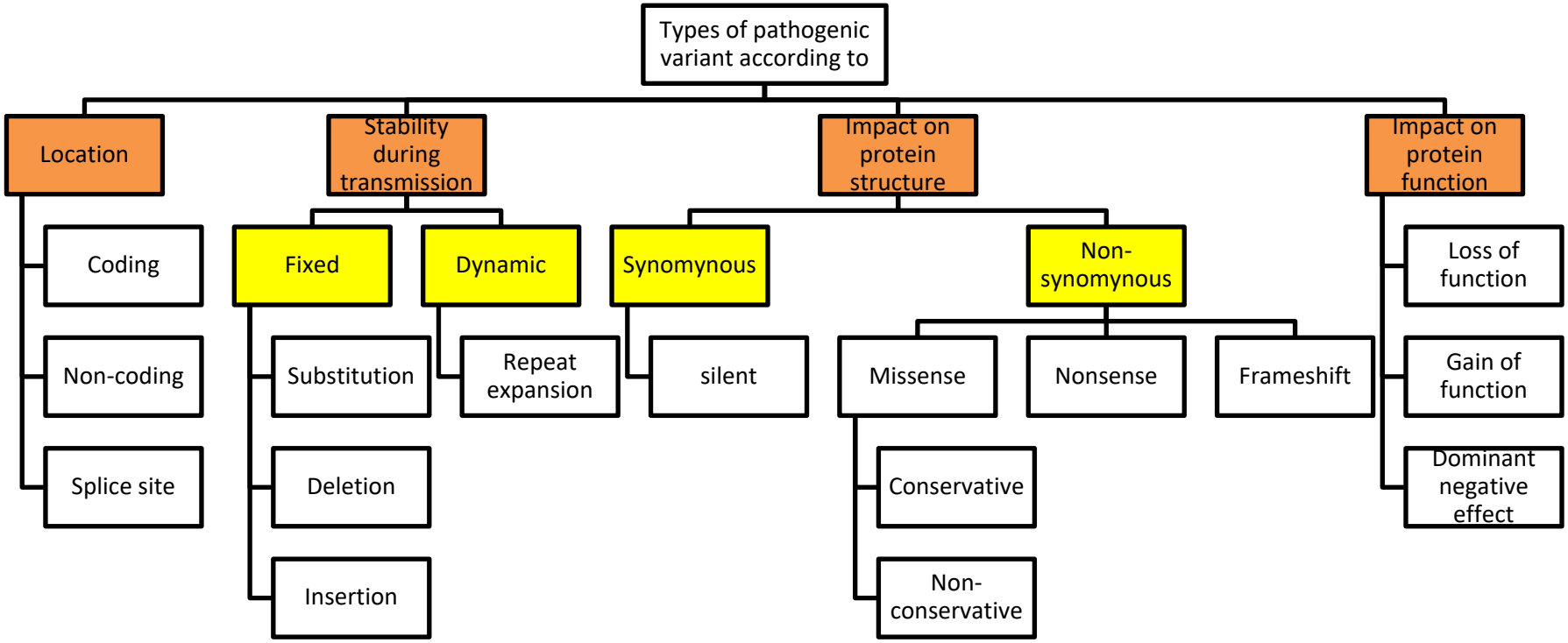
Definitions

- **Mutation** is a change in DNA sequence that leads to damage or change in gene product resulting to change in phenotype
- It is caused by:
  - Exposure to environmental factors (mutagens)
  - Errors during DNA replication or defect in DNA repair.
- **Polymorphism** is a change in DNA sequence without effect on phenotype
- It is part of normal genetic variation (how people are different)

Mutation vs polymorphism

Mutation	• A change in DNA sequence that leads to damage in gene product resulting to change in phenotype • Rare Genetic change (**Less than 1%**) • Severe effect and alter the function **of the protein or the Enzyme**.
Polymorphism	• A change in DNA sequence without effect on phenotype • Common variation (**greater than 1%**) • **No change in function** or small effect and occur on average one every **200-1000 base Pairs** .

Variants at gene level



A-Location of mutations

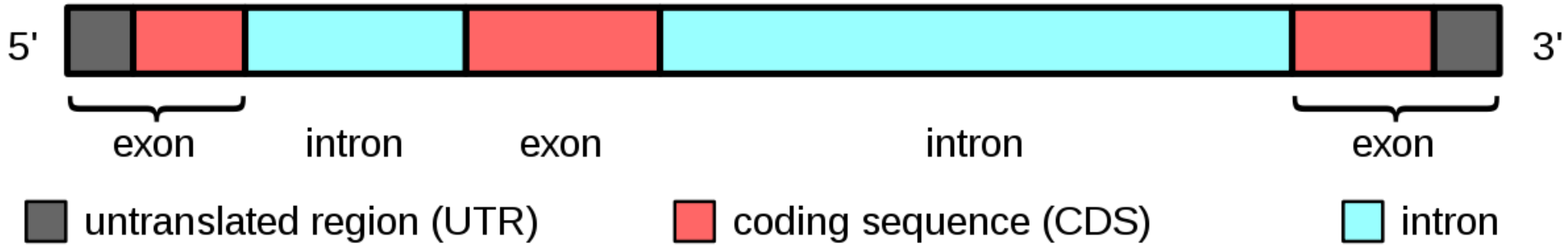
- Mutation could be in somatic cells or germline cells.
- A mutation arising in a somatic cell cannot be transmitted to offspring, whereas if it occurs in gonadal tissue or a gamete it can be transmitted to future generations.

- Mutations can occur in:

Non-coding (e.g intronic mutation)

Coding sequences (exonic mutation)

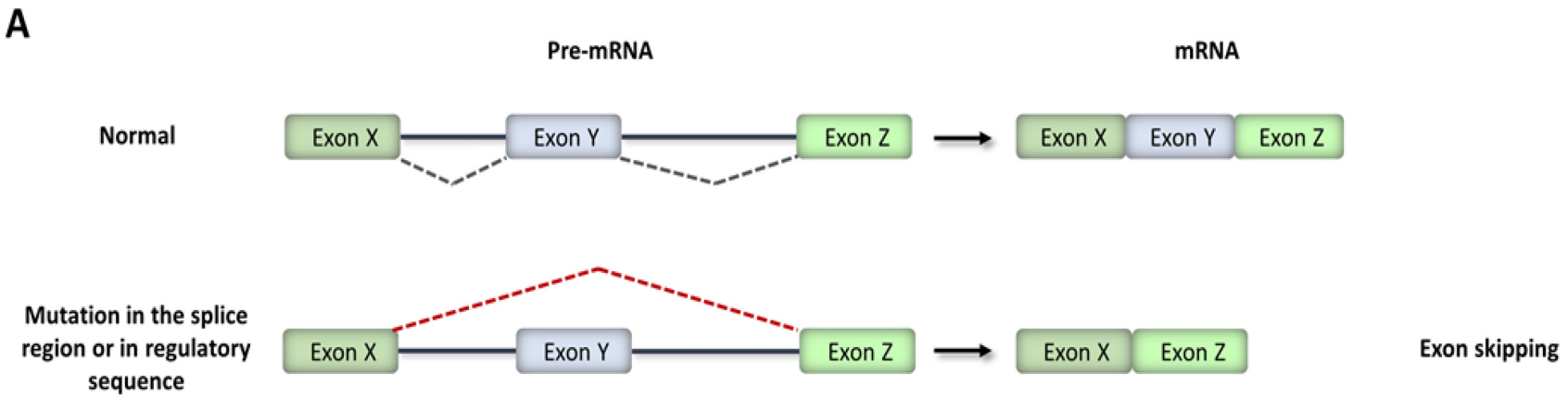
At boundaries between exons and introns (Splice site mutation)


5'
3'
exon
intron
exon
intron
exon
untranslated region (UTR)
coding sequence (CDS)
intron

SPLICE SITE MUTATION

- A genetic alteration in the DNA sequence that occurs at the boundary between an exon and an intron (splice site).
- This change can disrupt RNA splicing of precursor mRNA into mature mRNA

- This mutation leads to loss of exons or inclusion of introns in mature RNA resulting in an altered protein-coding sequence i.e production of abnormal protein



B-Stability during transmission

Fixed/Stable mutations: mutation which is transmitted unchanged (unaltered .)

Dynamic Or Unstable Mutations: This is new class of mutation which undergo alteration as they are transmitted in families .

FIXED/STABLE MUTATION

These mutations are at gene level and could be:

1. **Substitutions**
2. **Insertions**
3. **Deletions**

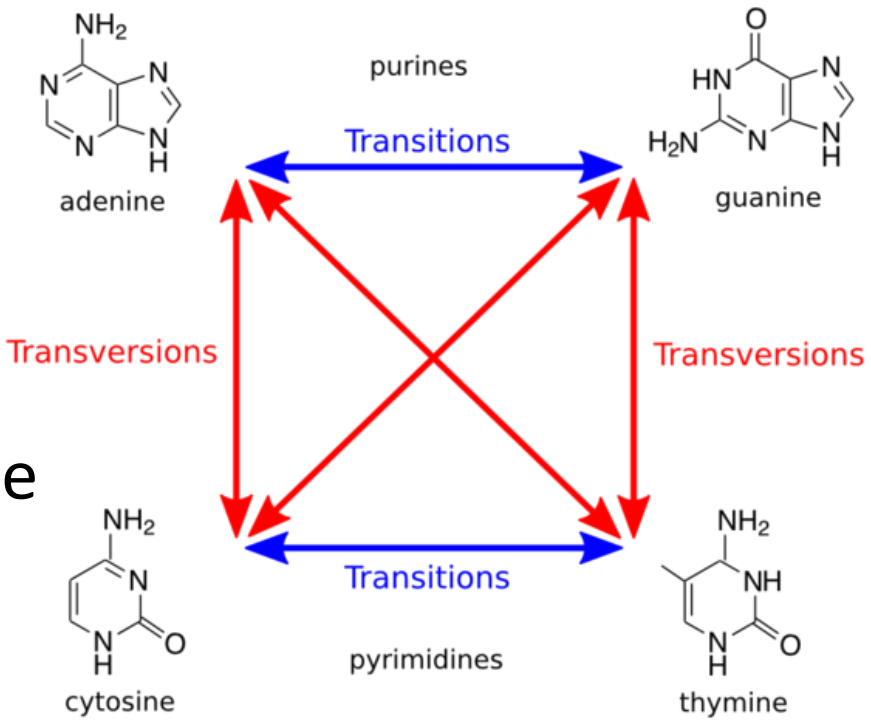
1. SUBSTITUTION

Definition: substitution is the replacement of a single nucleotide by another.

<u>Two type</u> of substitution:

- **Transition**: If the substitution involves replacement by the same type of nucleotide

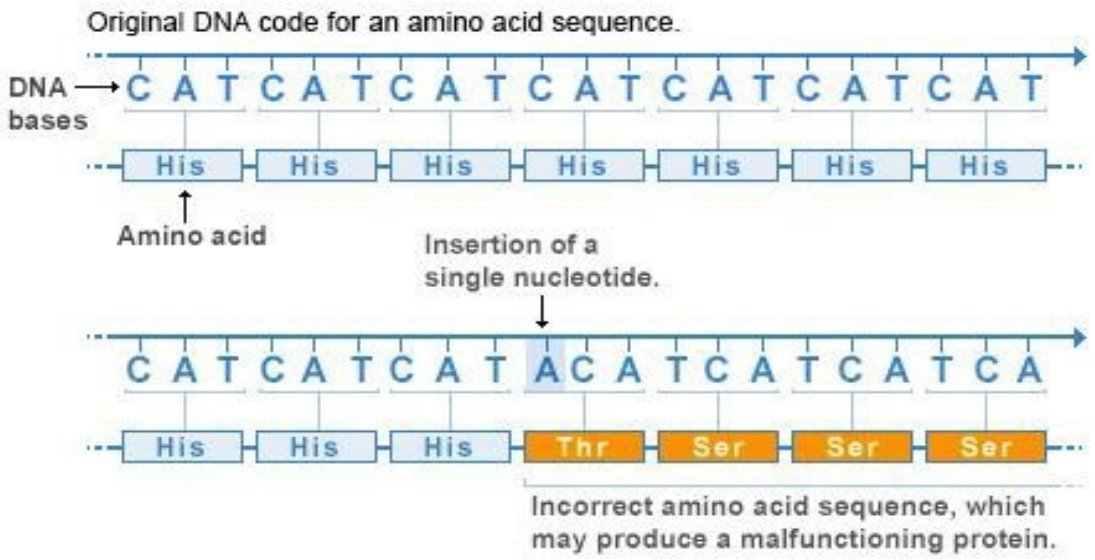
- **Transversion**: Substitution of a pyrimidine by a purine or (vice versa)



2. INSERTION

Definition: An insertion involves the **addition** **of one or more nucleotides into a gene**.

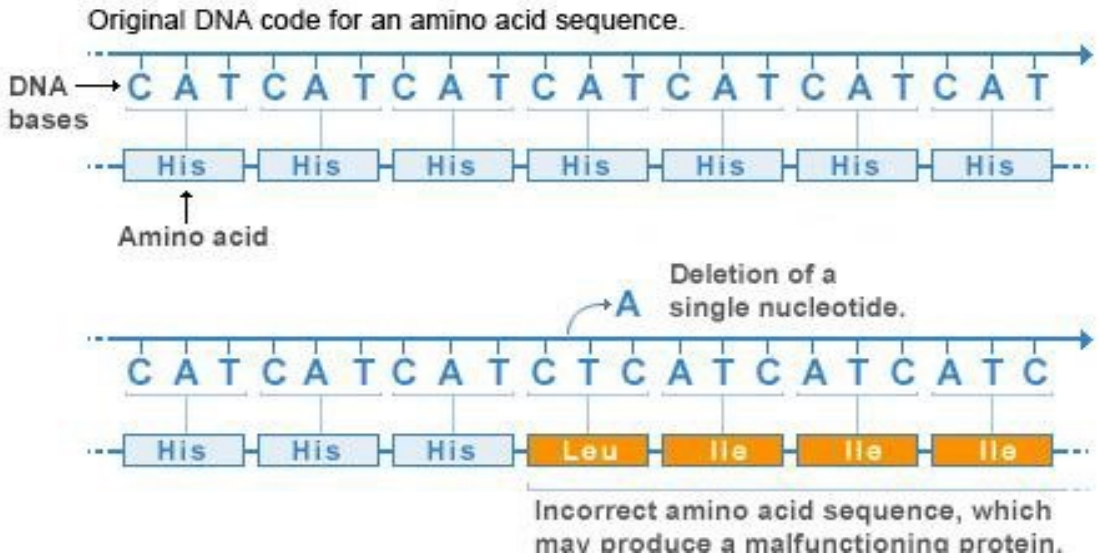
If an insertion occurs in a coding sequence and involves one, two or more nucleotides which are not a multiple of three, **it will disrupt the reading frame**.



3. DELETION

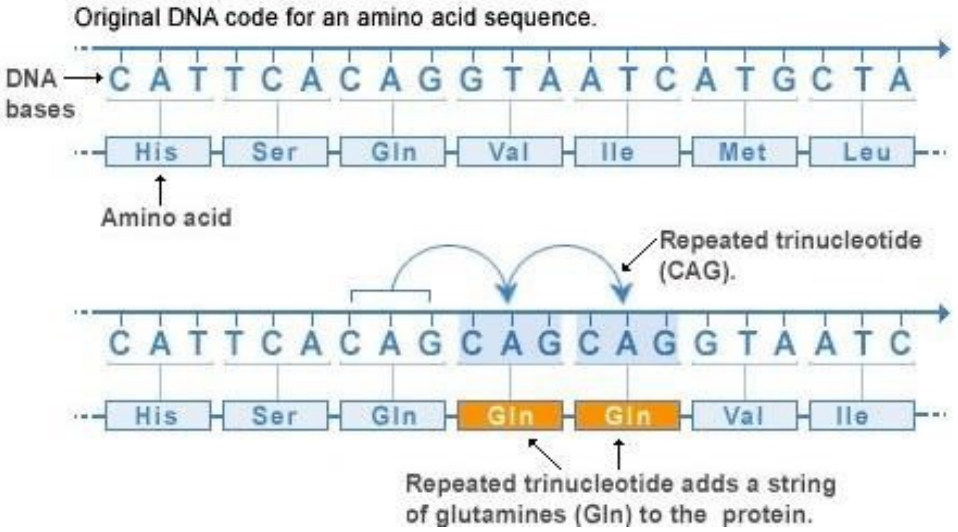
Definition: Deletion involves the **loss** of one or more nucleotides.

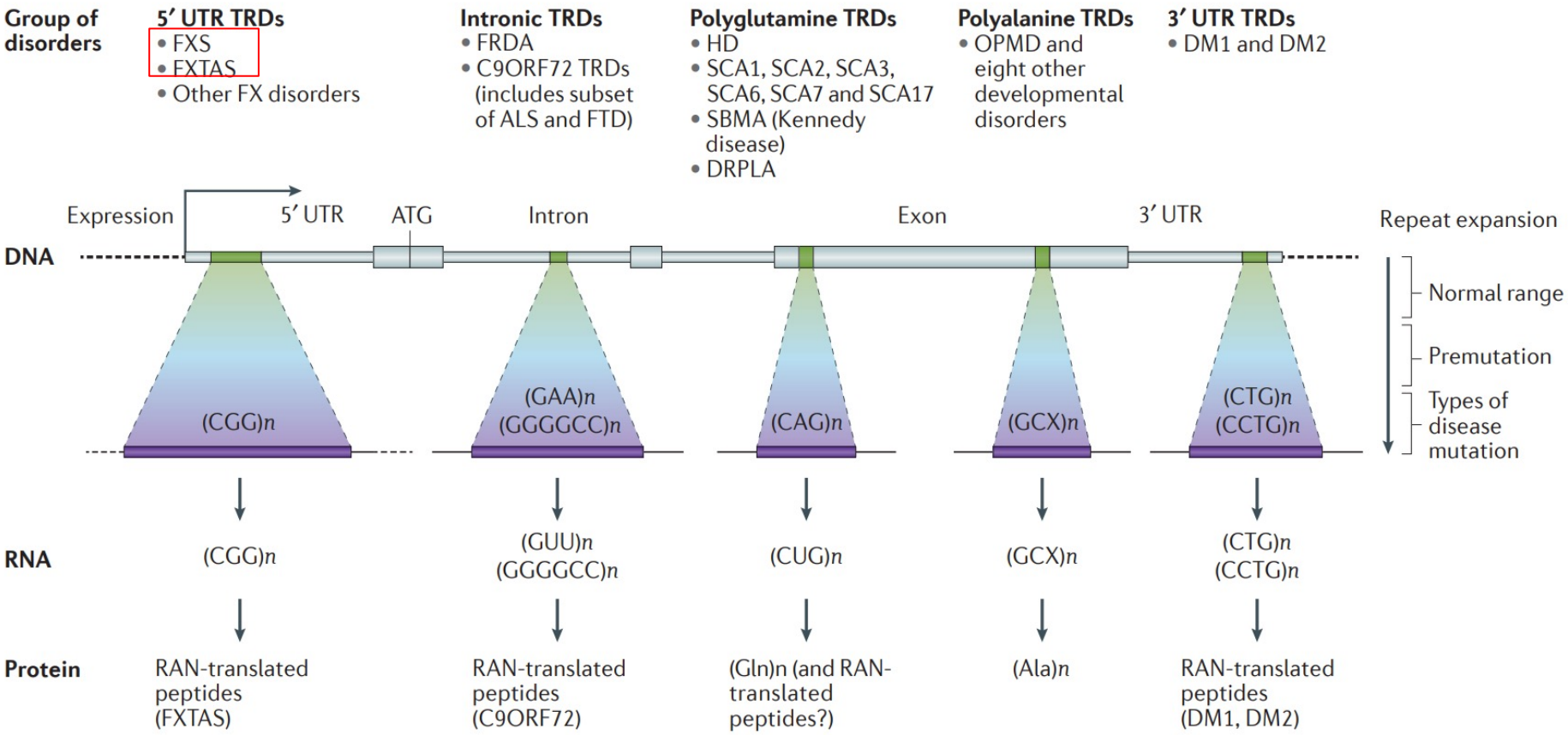
If it occurs in coding sequences and involves one, two or more nucleotides which are not a multiple of three, **it will disrupt the reading frame .**



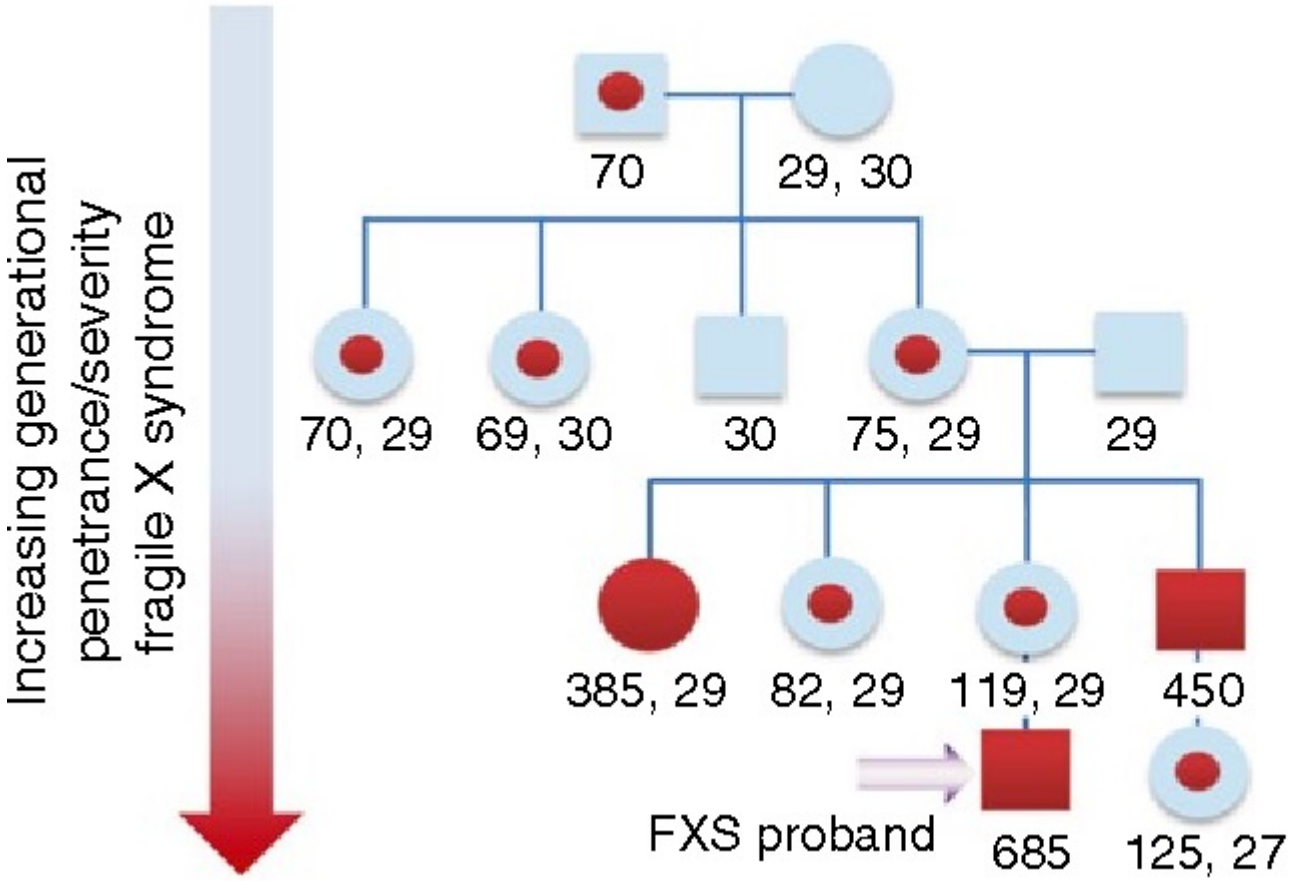
DYNAMIC/UNSTABLE MUTATION

- **Tandem repeat** is a sequence of 1-20 bp that is repeated in such a way that the repeats lie adjacent to each other on the chromosome
- **Tandem repeat expansion mutation** results in increase in copy numbers until they cross a threshold above which they become unstable




Group of disorders
5′ UTR TRDs
• FXS
• FXTAS
• Other FX disorders
Intronic TRDs
• FRDA
• C9ORF72 TRDs (includes subset of ALS and FTD)
Polyglutamine TRDs
• HD
• SCA1, SCA2, SCA3, SCA6, SCA7 and SCA17
• SBMA (Kennedy disease)
• DRPLA
Polyalanine TRDs
• OPMD and eight other developmental disorders
3′ UTR TRDs
• DM1 and DM2
Expression
5′ UTR
ATG
Intron
Exon
3′ UTR
Repeat expansion
DNA
(CGG)n
(GAA)n (GGGGCC)n
(CAG)n
(GCX)n
(CTG)n (CCTG)n
Normal range
Premutation
Types of disease mutation
RNA
(CGG)n
(GUU)n (GGGGCC)n
(CUG)n
(GCX)n
(CTG)n (CCTG)n
Protein
RAN-translated peptides (FXTAS)
RAN-translated peptides (C9ORF72)
(Gln)n (and RAN-translated peptides?)
(Ala)n
RAN-translated peptides (DM1, DM2)

Genetic anticipation:



Human disorder	Gene	Tandem repeat motif (amino acid repeat)	Range of tandem repeat length Normal (expanded)
Fragile X disorders			
FXS	*FMR1*	CGG (not translated)	5–44 (>200)
FXTAS	*FMR1*	CGG (RAN translation)	5–44 (55–200)

C-STRUCTURAL EFFECTS OF MUTATIONS ON THE PROTEIN

Mutations can also be subdivided into two main groups according to the effect on the polypeptide sequence of the encoded protein:

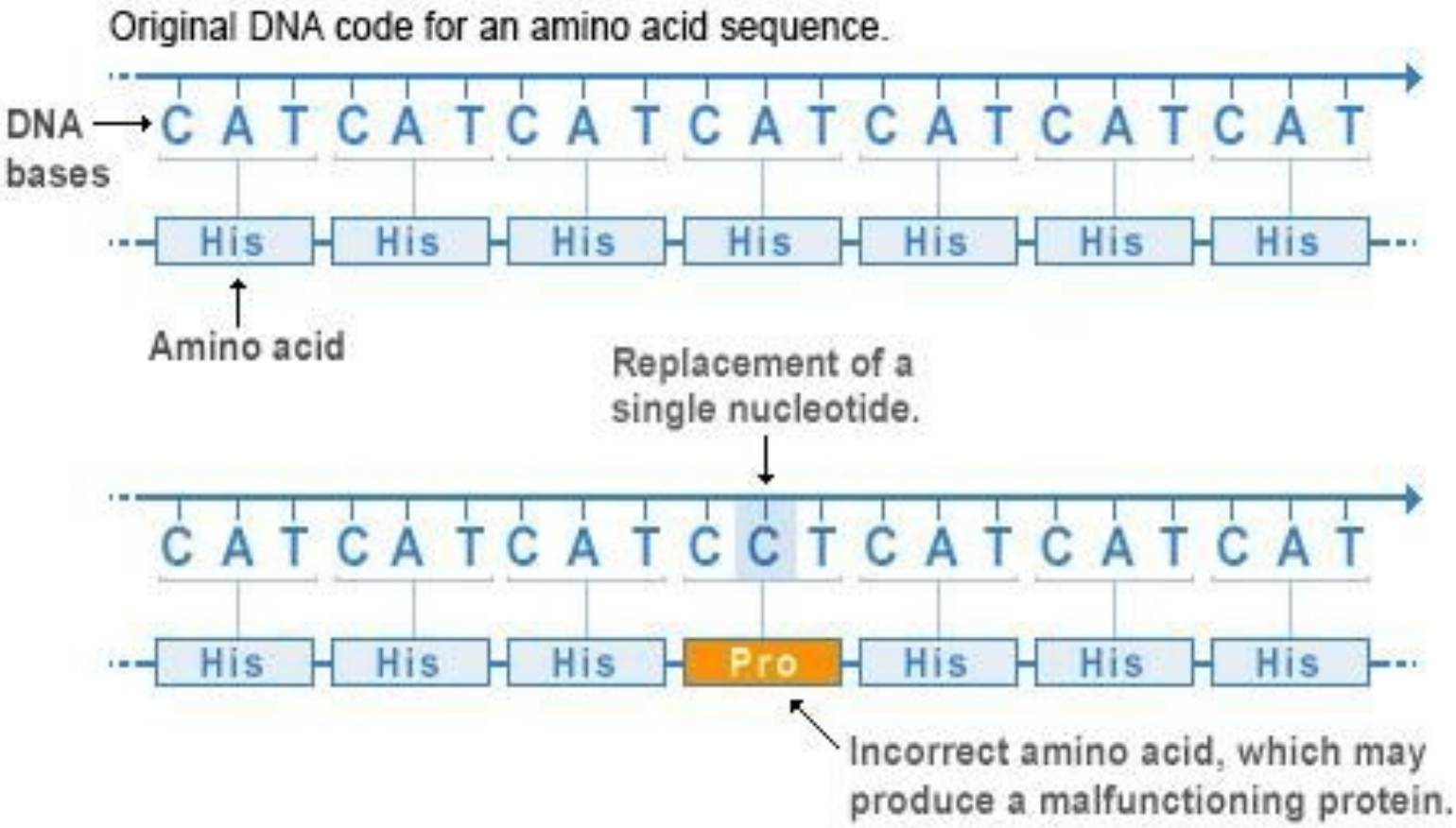
1. **Synonymous**
   - Silent
2. **Non- synonymous**
   - Missense
   - Nonsense
   - Frameshift

1. SYNONYMOUS/SILENT MUTATIONS

- If a mutation does not alter the polypeptide product of the gene, this is termed a synonymous or silent mutation.
- A single base pair substitution, particularly if it occurs in the third position of a codon, will often result in another triplet which codes for the **same amino acid** with no alteration in the properties of the resulting protein.

2. NON-SYNONYMOUS MUTATIONS

- If a mutation leads to an alteration in the encoded polypeptide, it is known as a non- synonymous mutation.
- Alteration of the amino acid sequence of the protein product of a gene is likely to result in abnormal function
- Types:
  - Missense
  - Nonsense
  - Frameshift



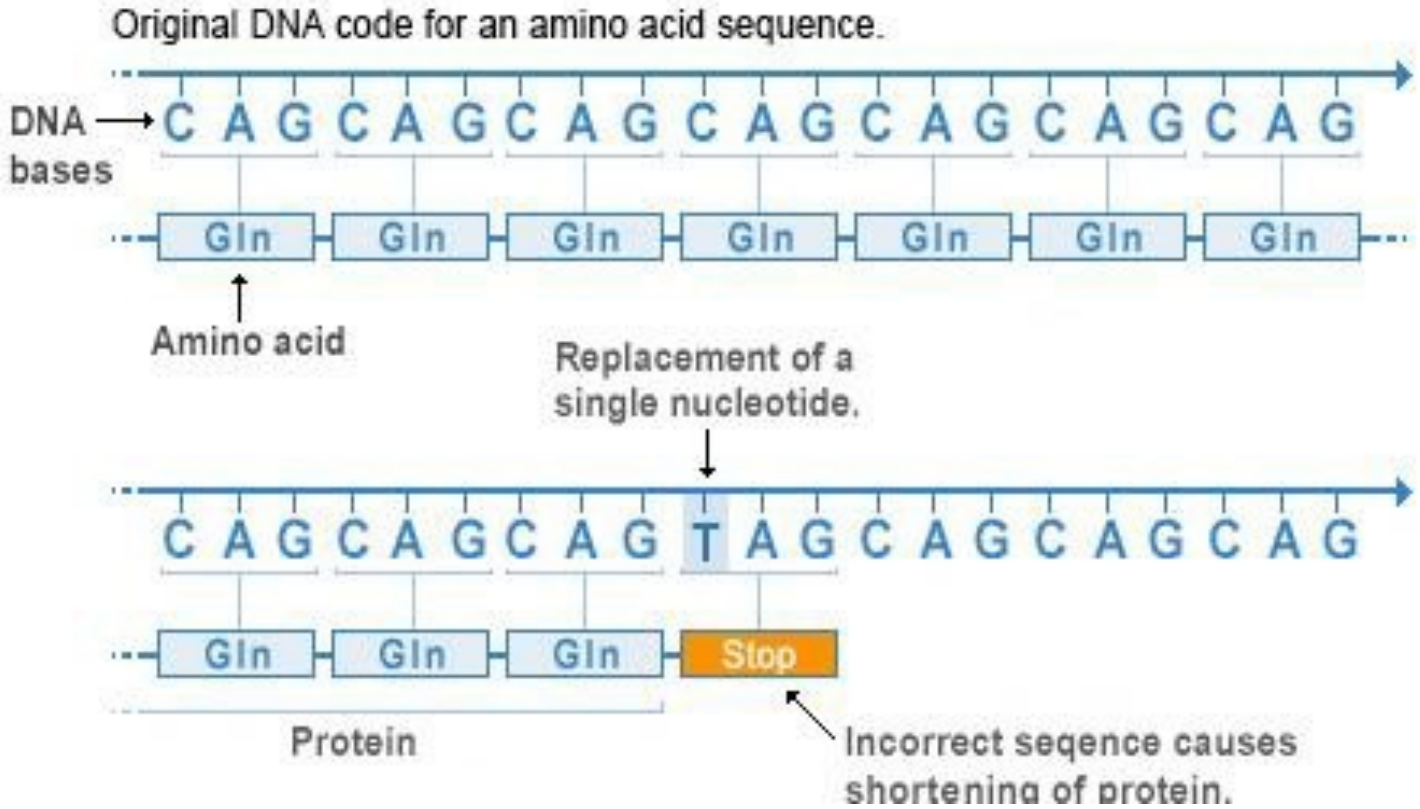
MISSENSE MUTATION

A single base pair substitution can result in coding for a different amino acid and the synthesis of an altered protein, a so-called missense mutation

MISSENSE Mutation

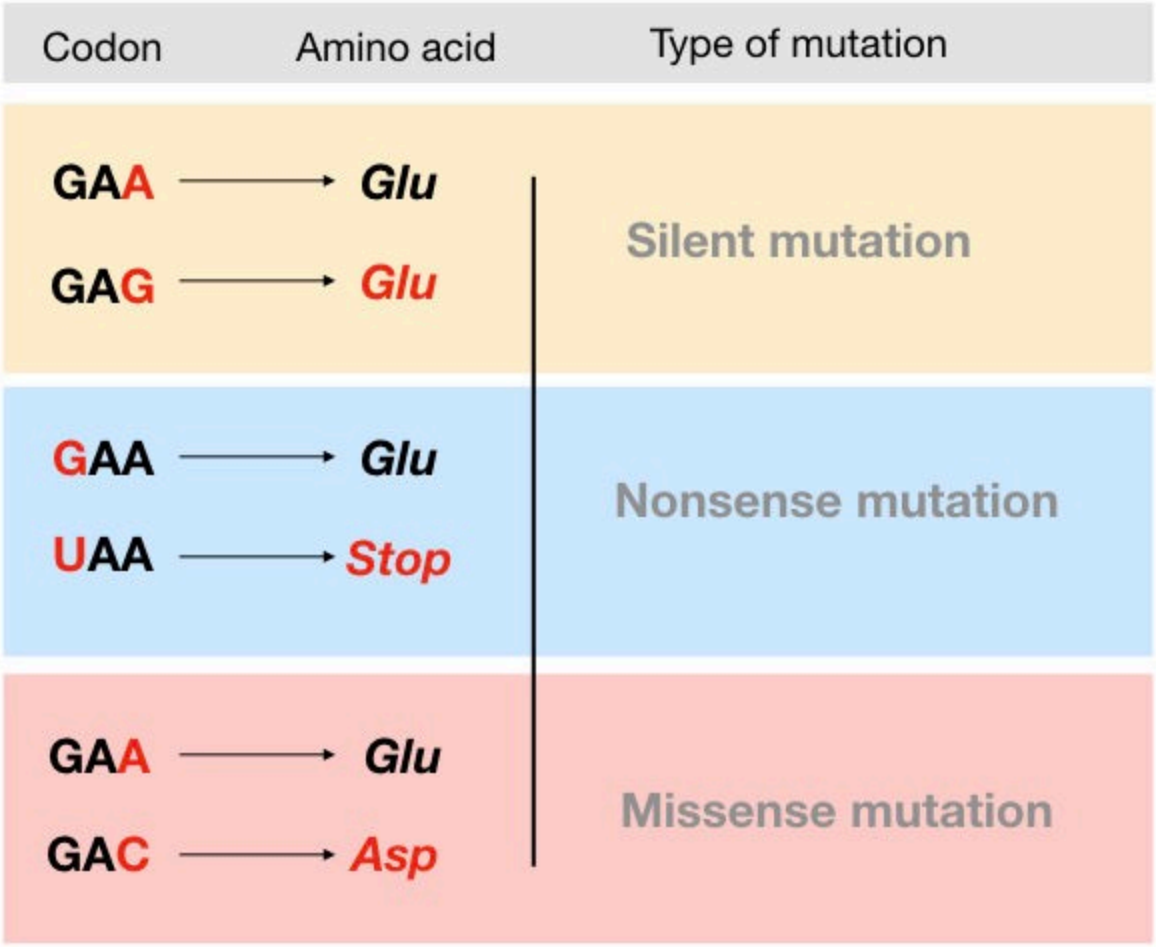
- **Non-conservative substitution**: If mutation coding for an amino acid which is chemically dissimilar such different charge of protein or structure of protein. Non-conservative substitution will result in complete loss or gross reduction of biological activity of the resulting protein.

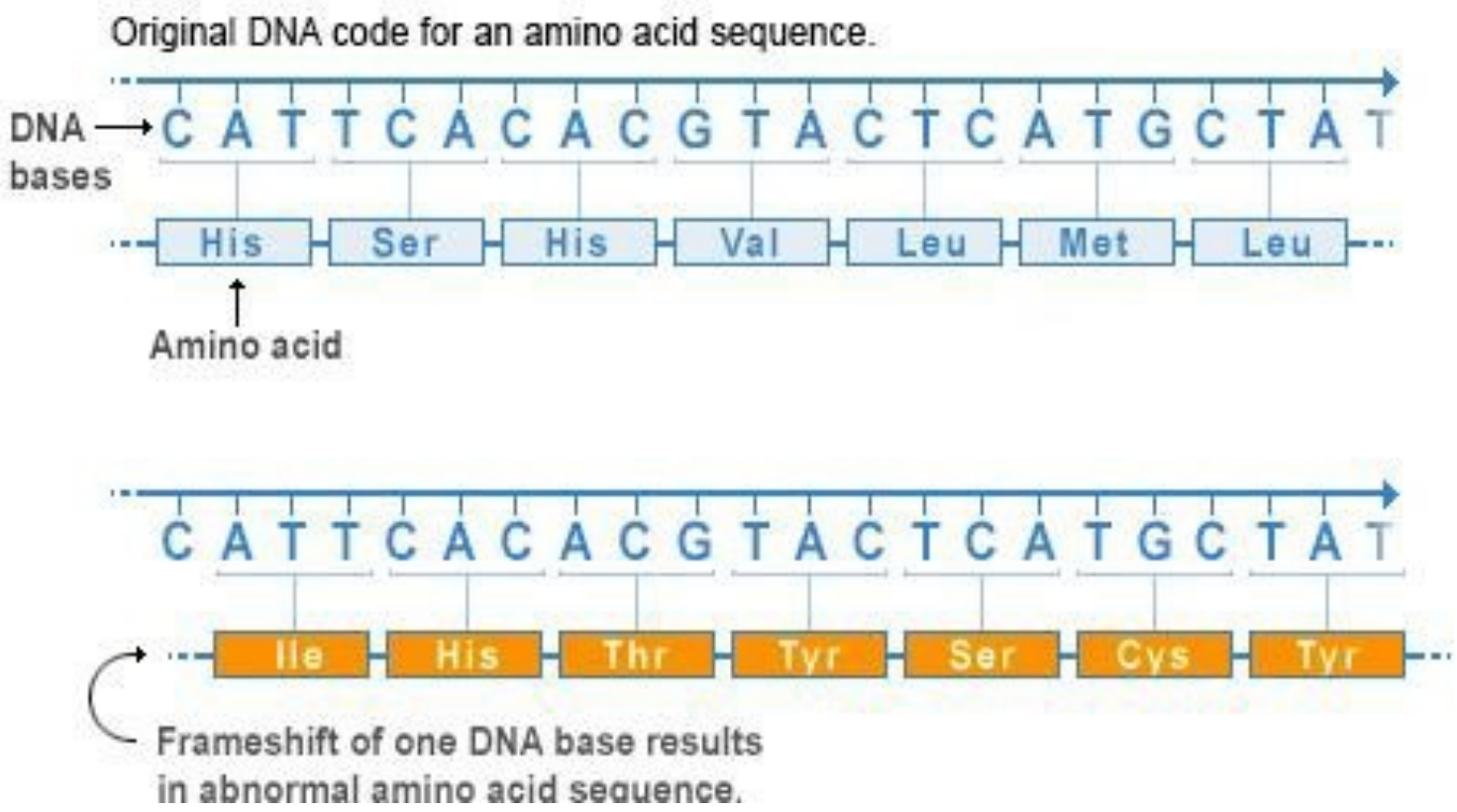
- **Conservative substitution**: If mutation coding for an amino acid which is chemically similar, have no functional effect.



NONSENSE MUTATION

A substitution of base pair which leads to the generation of one of the stop codons will result in premature termination of translation of a peptide chain.


Codon
Amino acid
Type of mutation
GAA → Glu
GAG → Glu
Silent mutation
GAA → Glu
UAA → Stop
Nonsense mutation
GAA → Glu
GAC → Asp
Missense mutation

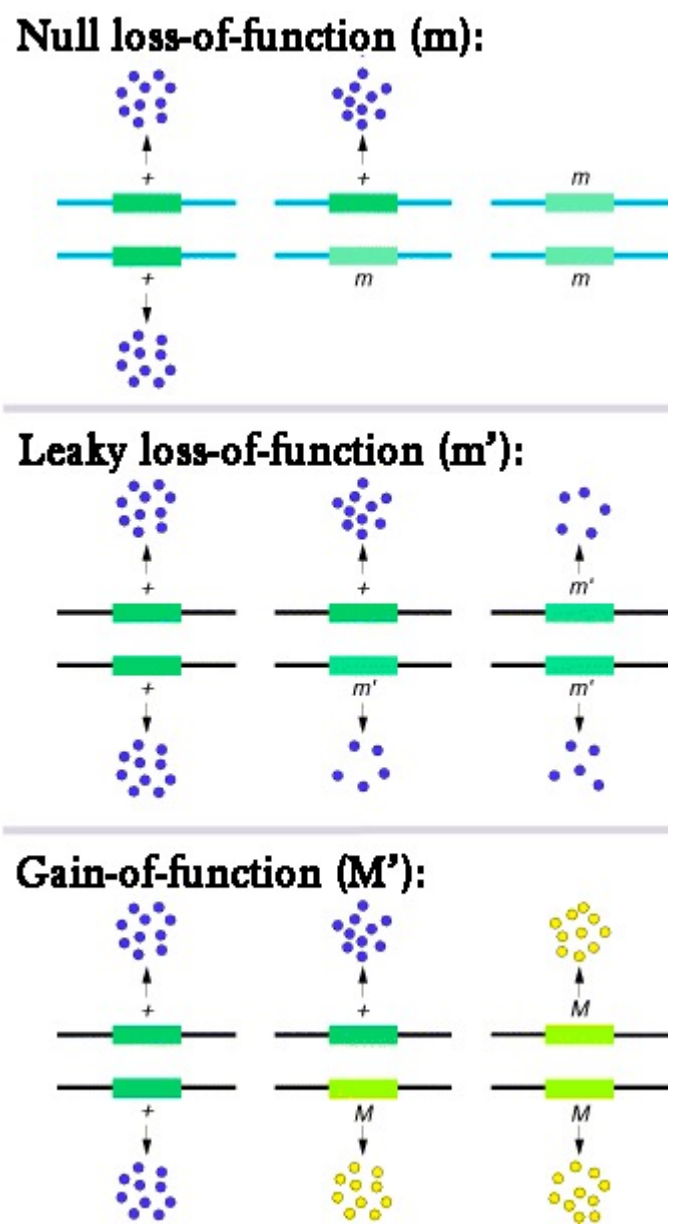


FRAMESHIFT MUTATION

If a mutation involves the insertion or deletion of nucleotides which are not a multiple of three, it will disrupt the reading frame. Most of these mutation result in premature
stop codon

D-FUNCTIONAL EFFECTS OF MUTATIONS ON THE PROTEIN

- **Loss of function** mutation eliminates or reduces the function of a wild type gene product
- **Gain of function** mutation results in either increased levels of gene expression or the development of a new function(s) of the gene product .
- **Dominant Negative**: A mutation whose gene product interferes with the normal, wild-type gene product within the same cell



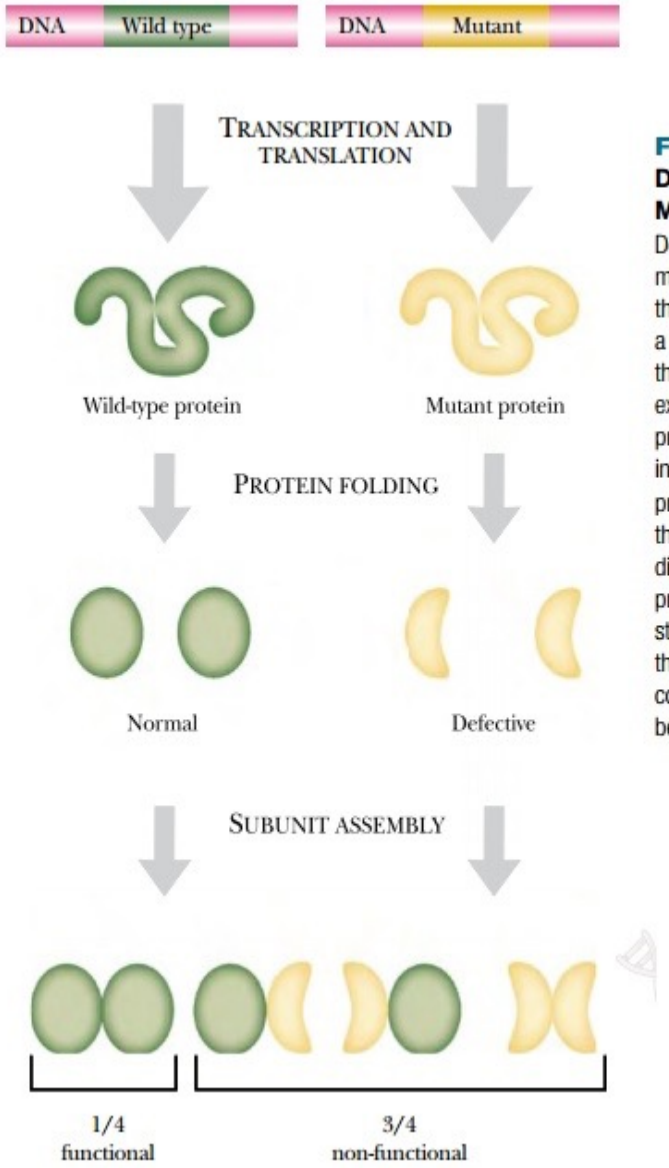


FIGURE 16.3
Dominant Negative Mutations
Dominant negative mutations occur when the defective copy of a gene interferes with the functional copy. For example, the defective protein may bind to and interfere with the normal protein. In this scenario, the proteins function as dimers. If the mutant protein is defective but still forms dimers, then three-fourths of the complexes will be defective.

Sources of mutations

- **Inherited**
- **Induced**

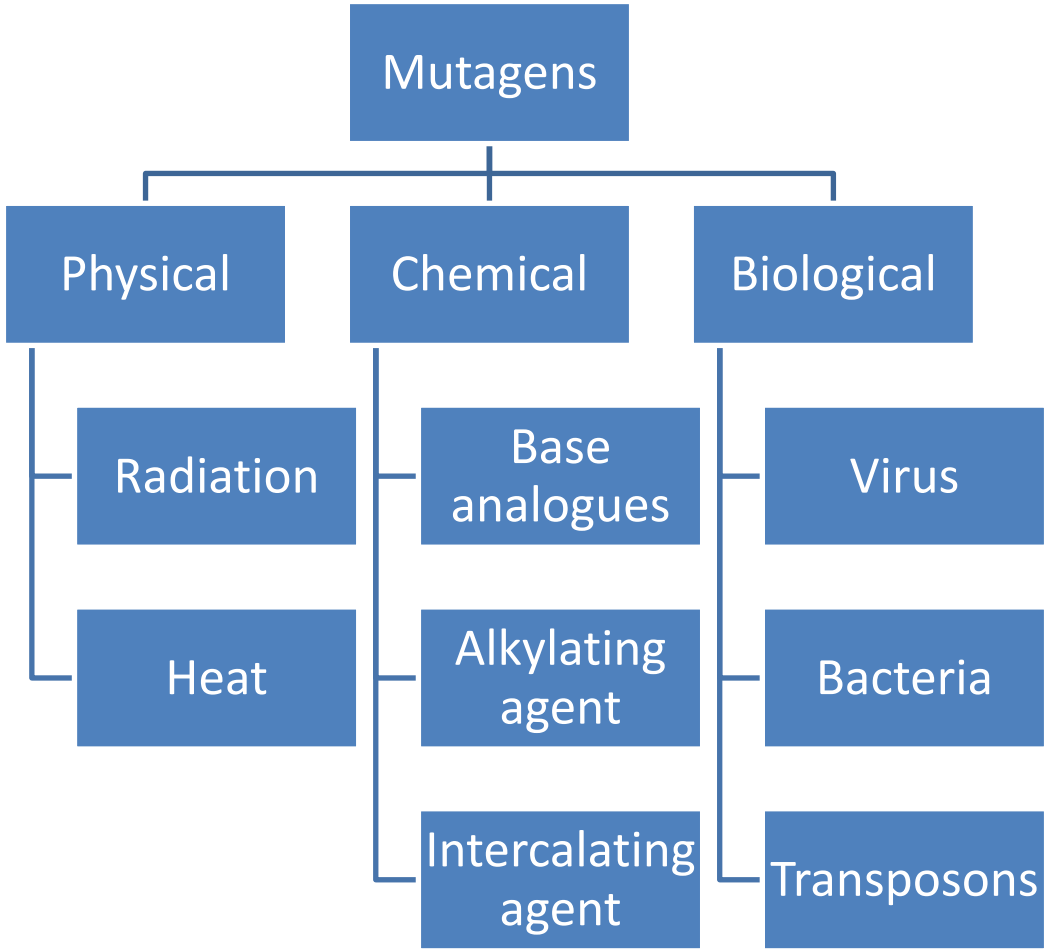
  Exposure to environmental agents increases the rate of mutation
- **Spontaneous (naturally occurring):**
  - Error in DNA replication
  - Faulty DNA repair e.g. mismatch
  - Exposure to Natural mutagens

Mutagenesis

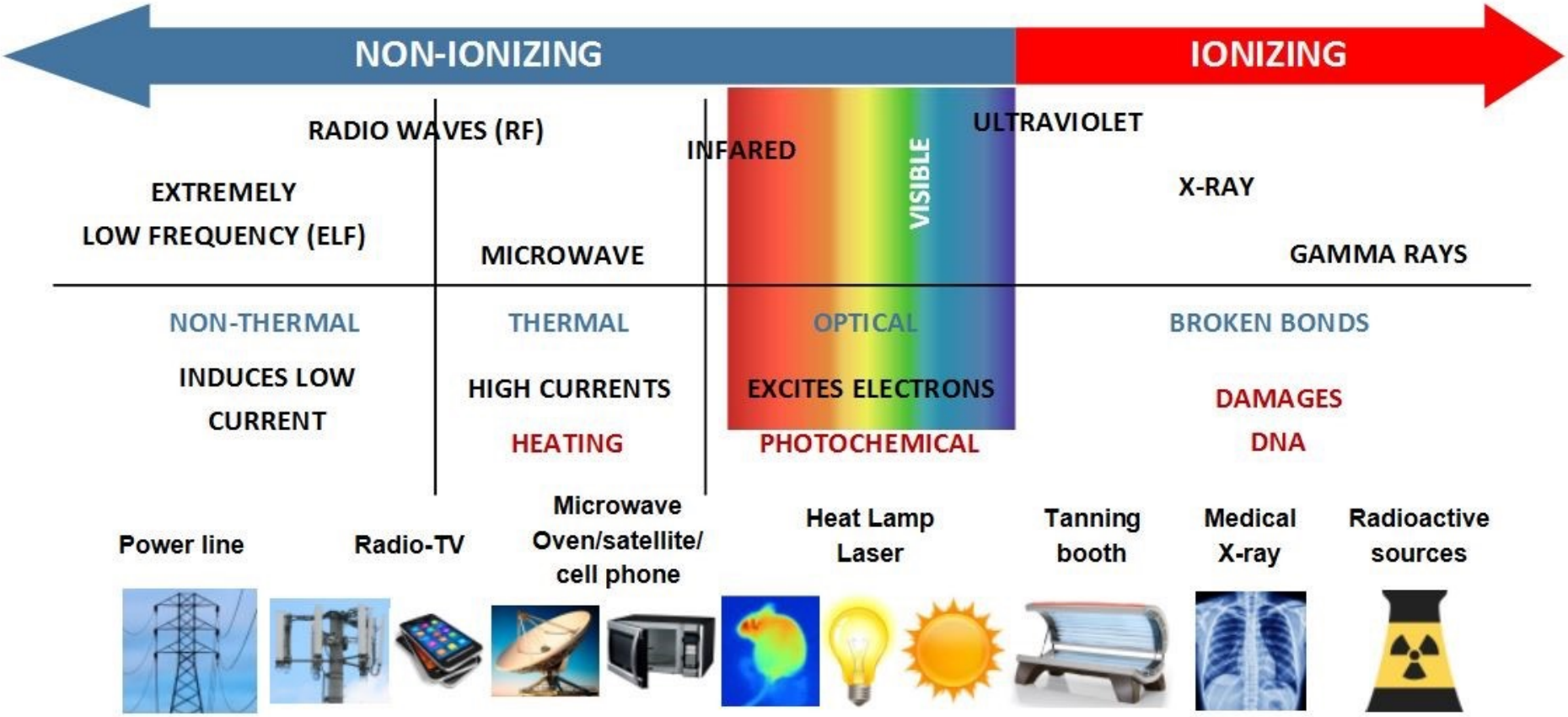
The process by which mutation occurs in organism either spontaneously or due to exposure to mutagens

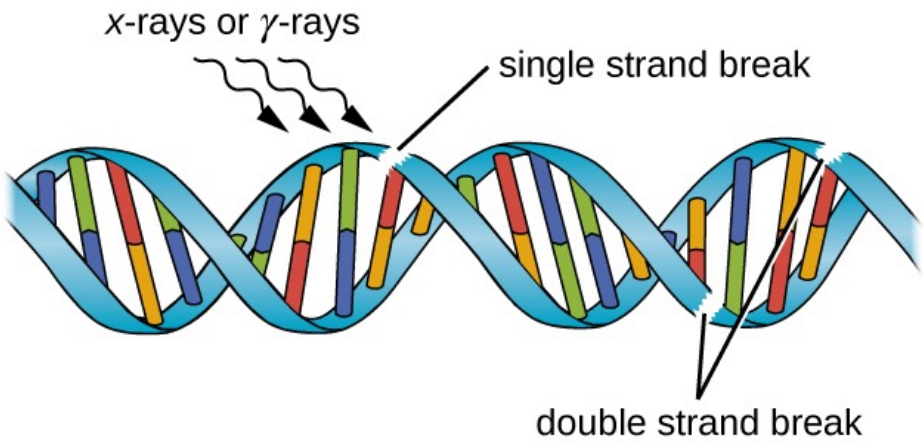
Agent	Effect
Carcinogen	Causes Cancer
Clastogen	Causes fragmentation of chromosomes
Mutagen	Causes mutations
Oncogen	Induces tumor formation
Teratogen	Results in developmental abnormalities

Classification of Mutagens

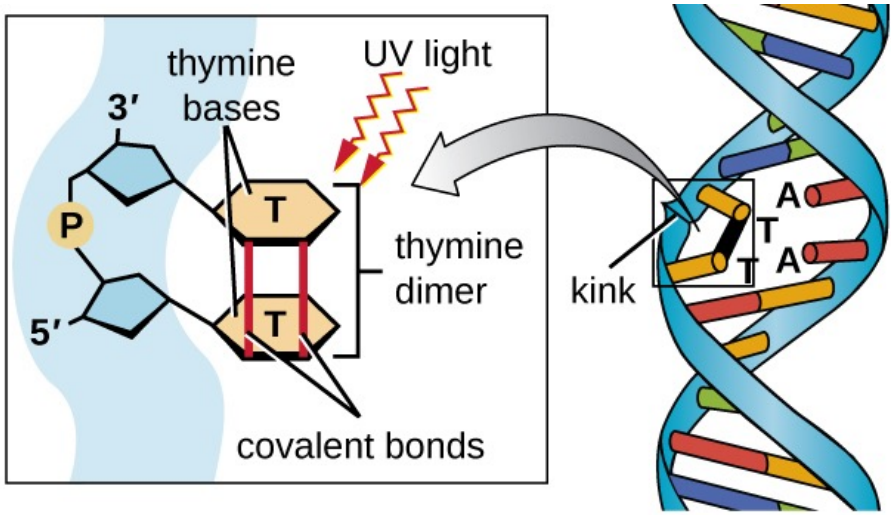


Physical agents



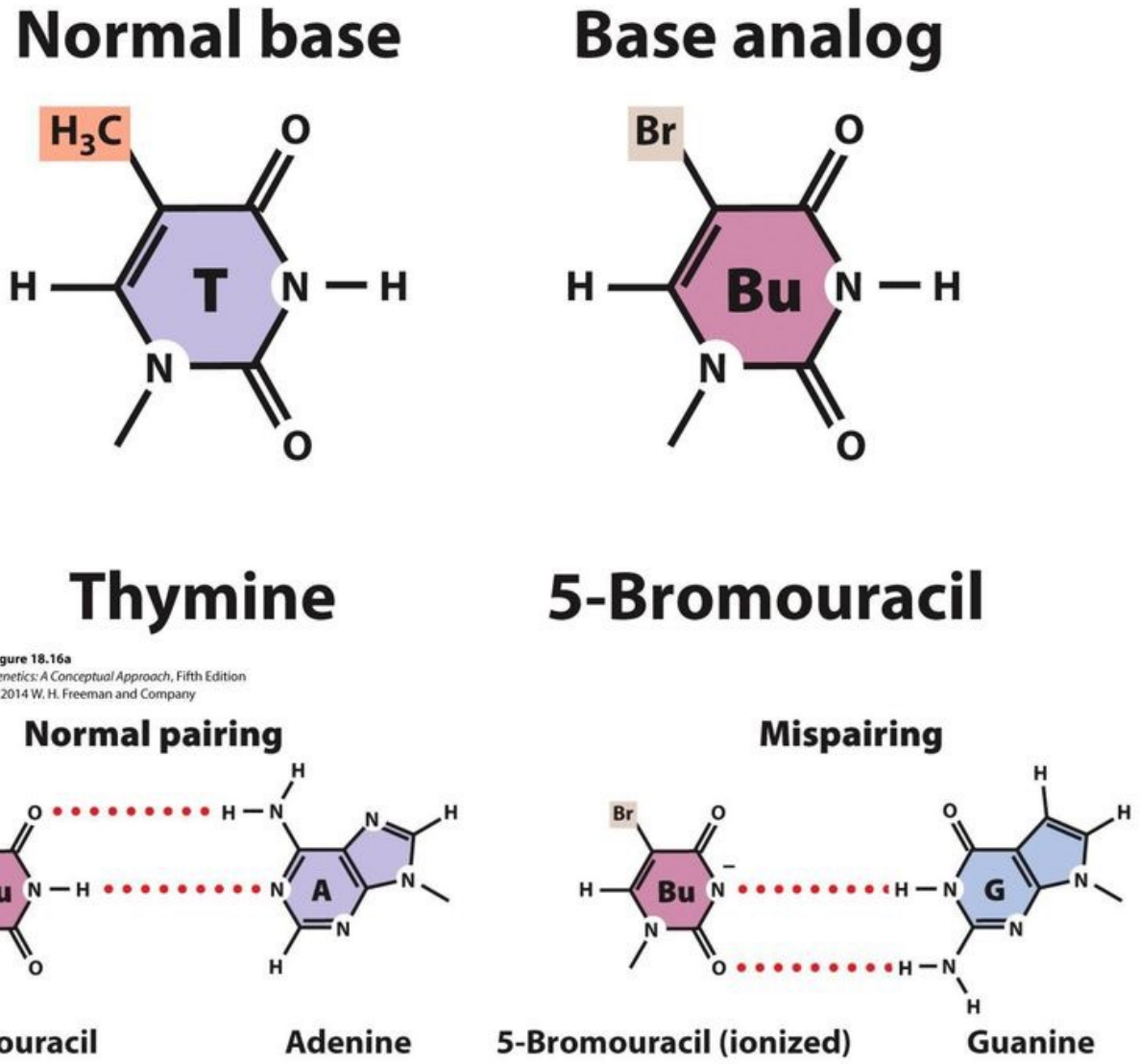


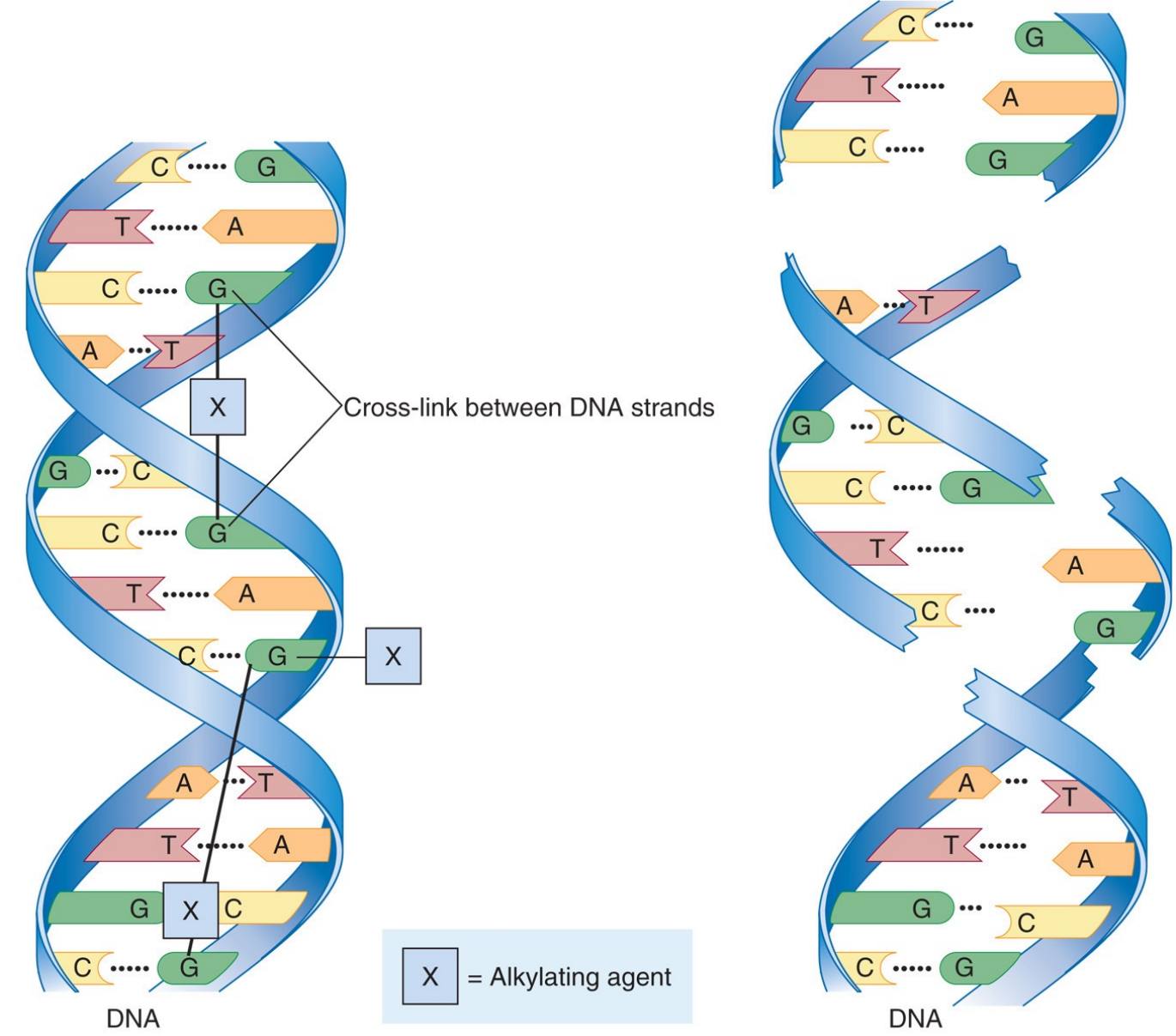
(a) Ionizing radiation



(b) Non-ionizing radiation

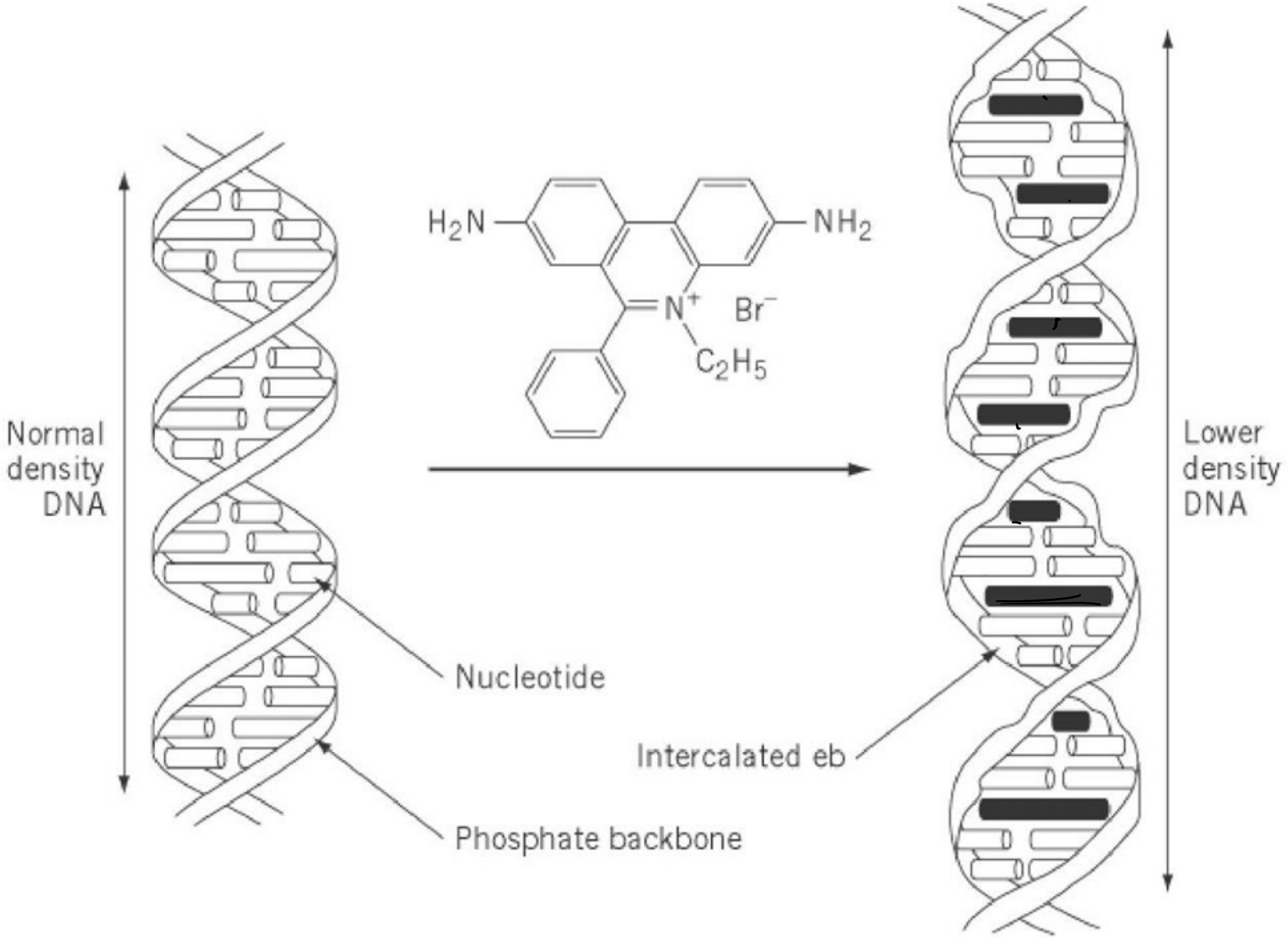
Chemical agents





(a) Alkylation occurring during G_0 (resting) phase of cell cycle

(b) Strand breaks occurring when DNA replicates during S phase of cell cycle


Normal density DNA
H_2N
NH_2
N^+
Br^-
C_2H_5
Nucleotide
Phosphate backbone
Intercalated eb
Lower density DNA

References

- Genetic Variation, Comparative Genomics, and the Diagnosis of Disease. Evan E. Eichler. N Engl J Med. 2019 July 04; 381(1): 64–7
- HGVS Recommendations for the Description of Sequence Variants: 2016 Update. Johan T. den Dunnen et al. HUMAN MUTATION, Vol. 37, No. 6, 564–569, 2016
- Tandem repeats mediating genetic plasticity in health and disease. Anthony J. Hannan. Nat Gen 2018

THANK YOU