



NEOPLASIA III

BUSHRA AL-TARAWNEH, MD

ANATOMICAL PATHOLOGIST

PATHOLOGY, MICROBIOLOGY AND IMMUNOLOGY
DEPARTMENT

FACULTY OF MEDICINE - MUTAH UNIVERSITY

MOLECULAR BASIS OF CANCER

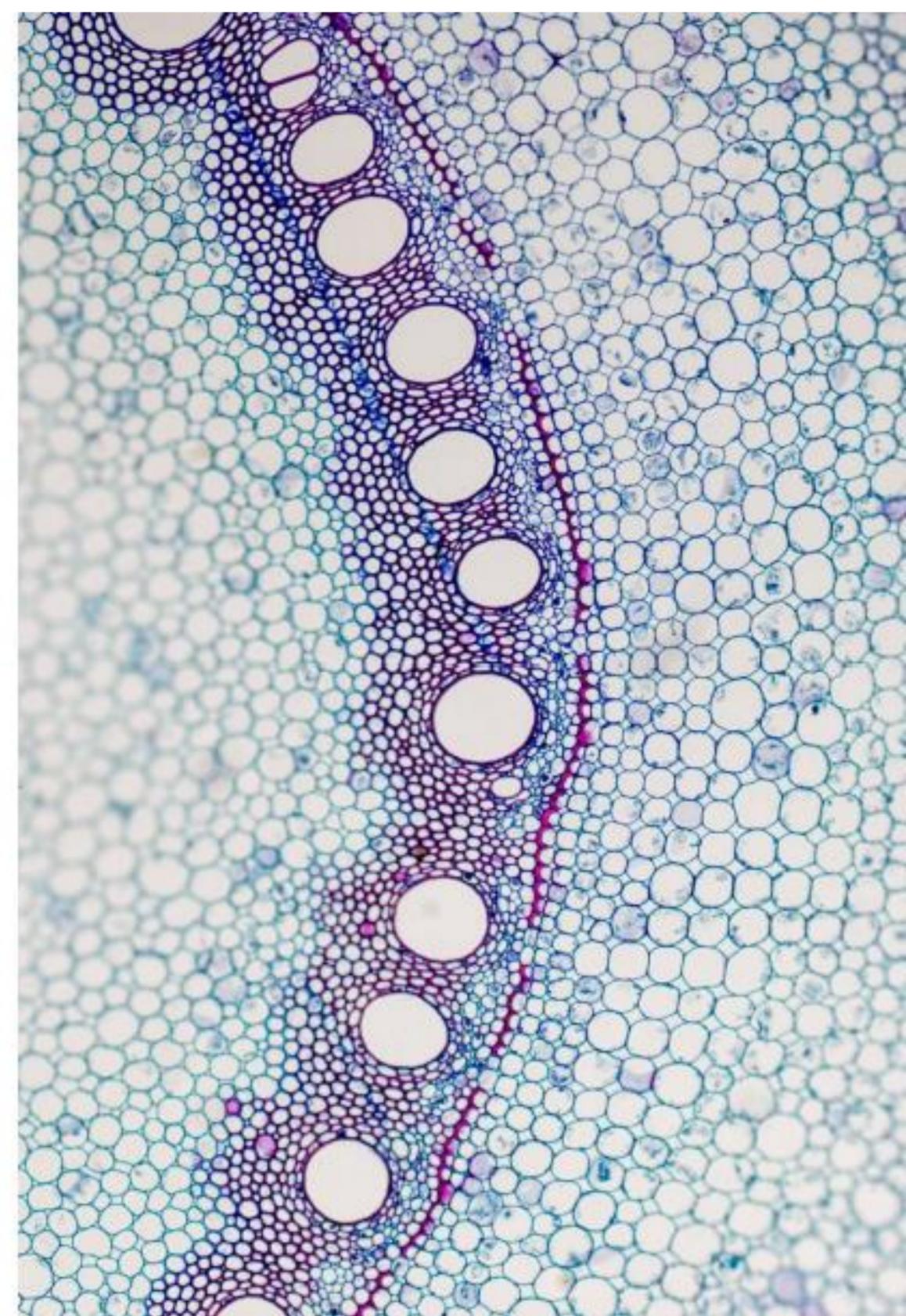
- The following are fundamental principles:
 - Non-lethal genetic damage underlies carcinogenesis; genetic injury can be inherited in the germ line or acquired in somatic cells through spontaneous mutation or environmental exposures.
 - Neoplasms are caused by nonlethal, genetic damage, which causes uncontrolled cellular proliferation.

What does a clone mean?..

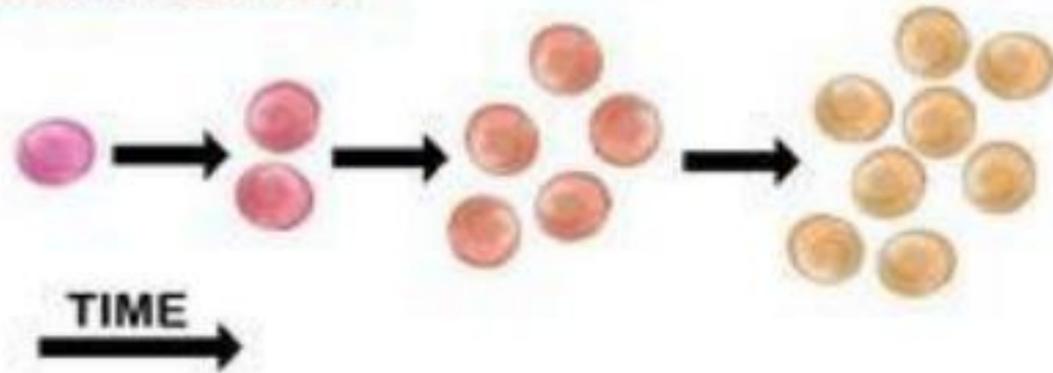
- Because tumor cells originate from one single genetically damaged *crazy* cell, they are clonal



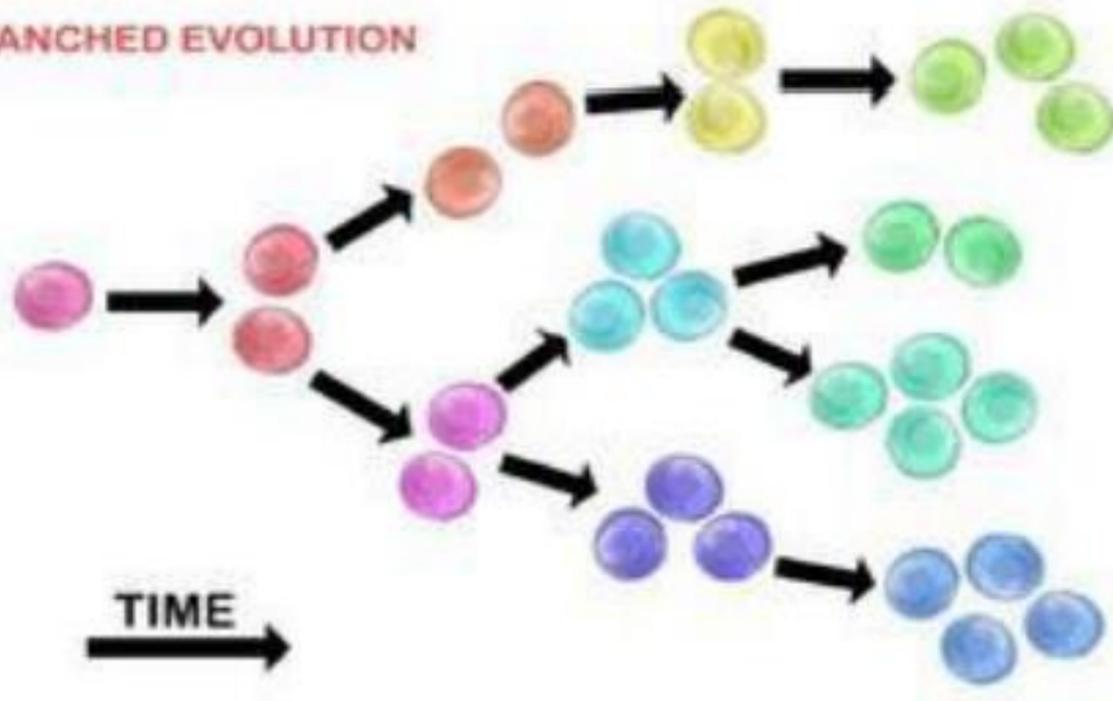
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- So: malignant cells originate from one single transformed cell that acquires a mutation allowing it to proliferate in an uncontrolled manner.
 - This cell keeps proliferating forming a clone.
 - But the proliferating cells acquire additional mutations, that help the tumor mass to grow further or to avoid death, or to metastasize ..etc.
 - Each cell with a new mutation proliferates forming a **sub-clone**.
 - This is referred to as: branched evolution (see pic on next slide)



LINEAR EVOLUTION



BRANCHED EVOLUTION



LINEAR VERSUS BRANCHED EVOLUTION. TUMORS GROW VIA THE BRANCHED ROUTE. NOTE THAT ALTHOUGH HETEROGENEOUS; THEY ALL ORIGINATED FROM ONE CELL, SO THEY ARE STILL CLONAL.



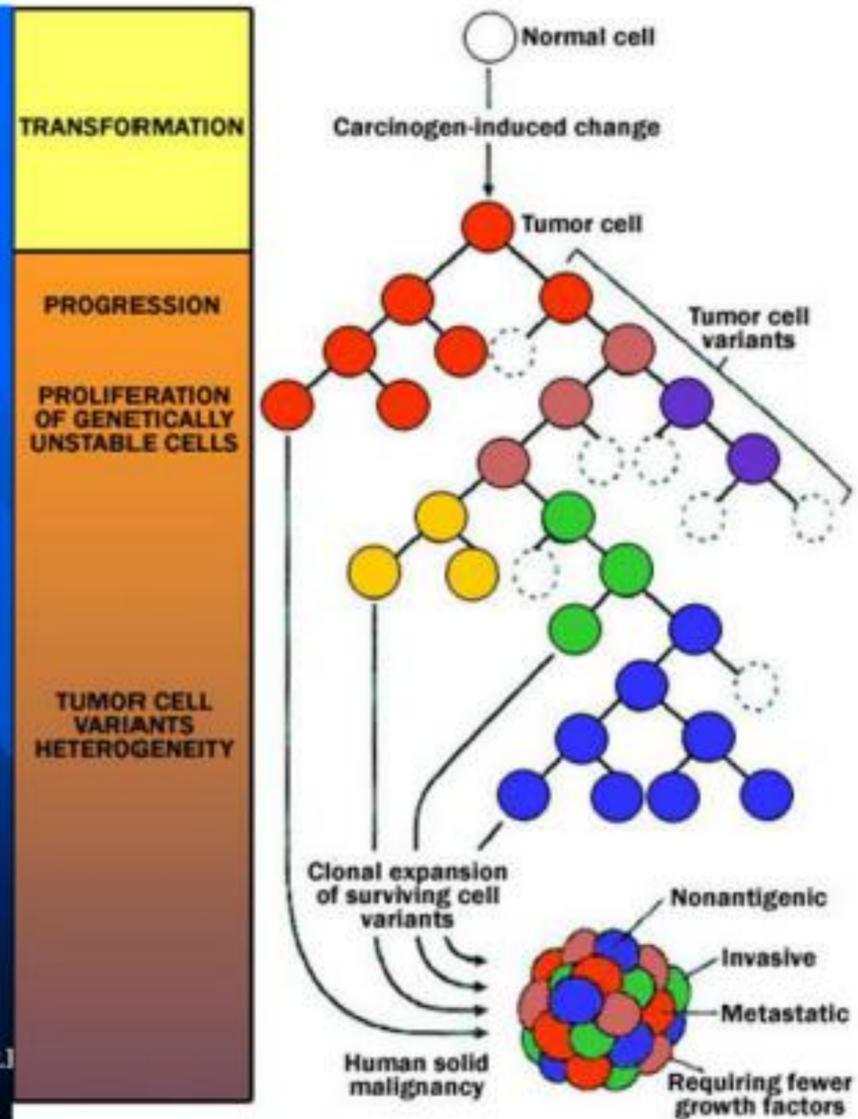
- Carcinogenesis is a multistep process. The attributes of malignancy (e.g., invasiveness, excessive growth, escape from the immune system, etc.) are acquired, a process called tumor progression. At the genetic level, progression results from accumulation of successive mutations



- Although tumors begin as monoclonal proliferations, by the time they are clinically evident, they are extremely heterogeneous

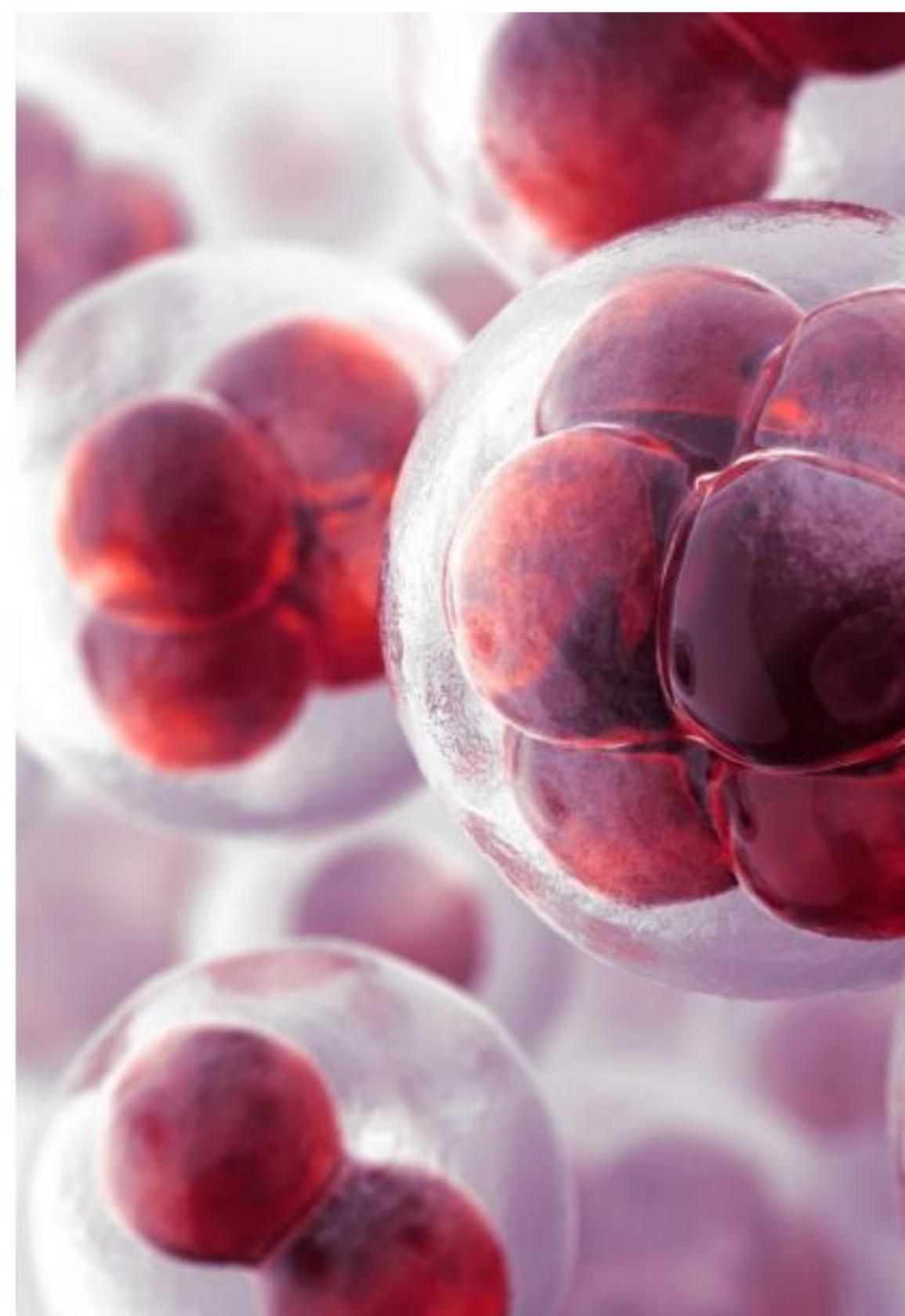
- At the molecular level, tumor progression and heterogeneity result from **multiple mutations generating subclones** with varying abilities to grow, invade, metastasize, and resist therapy .
- During progression, tumor cells are subjected to immune and nonimmune selection pressures.

Dr.]



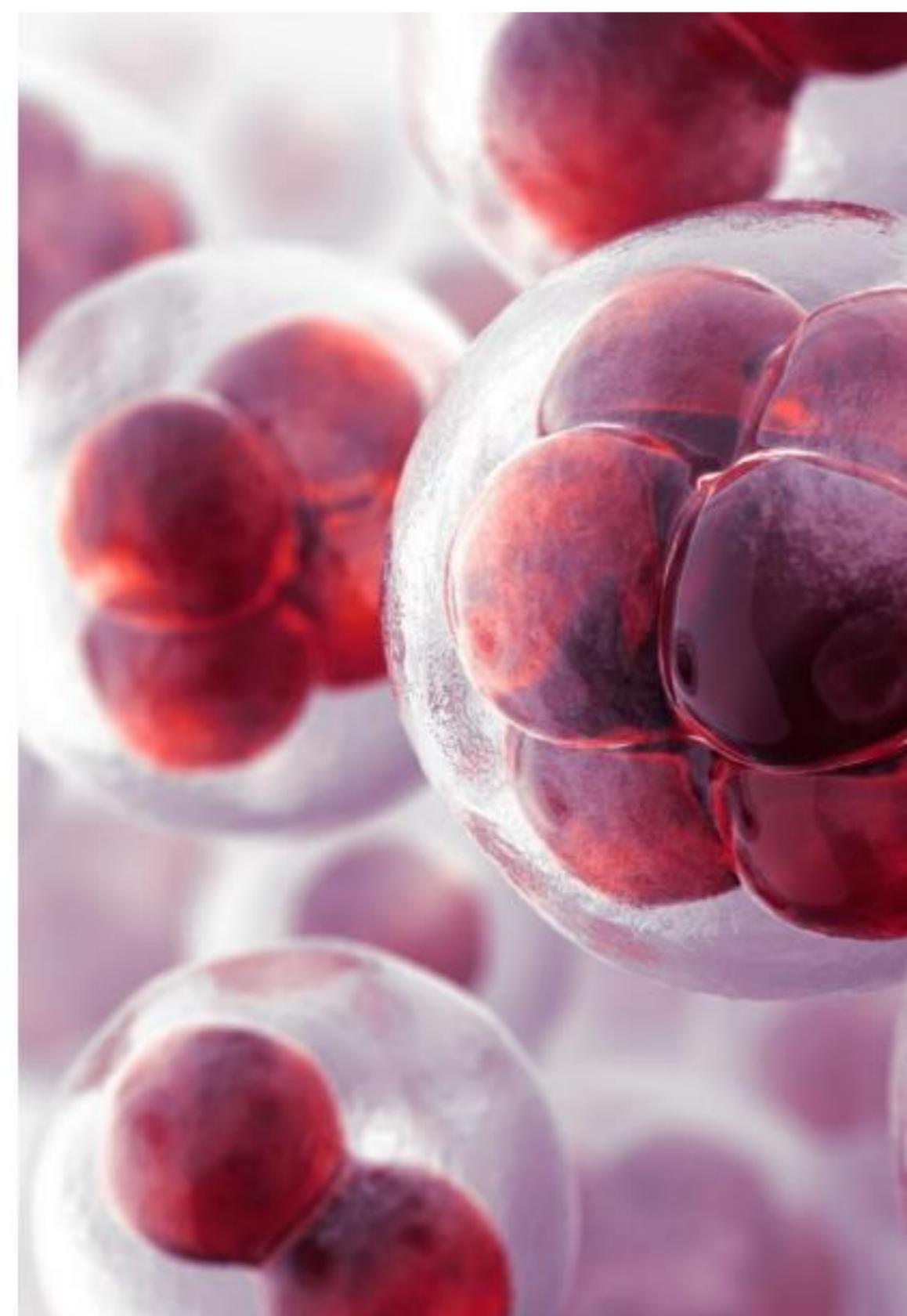
ESSENTIAL ALTERATIONS FOR MALIGNANT TRANSFORMATION

- ❑ Certain fundamental changes in cell physiology contribute to development of the malignant phenotype:
- Self-sufficiency in growth signals (proliferation without external stimuli).
- Insensitivity to growth-inhibitory signals.
- Evasion of apoptosis.
- Defects in DNA repair.
- Limitless replicative potential (related to telomere maintenance).
- Sustained angiogenesis to provide adequate nutrition and waste removal.
- Ability to invade and metastasize.
- Ability to escape immune recognition and regulation.



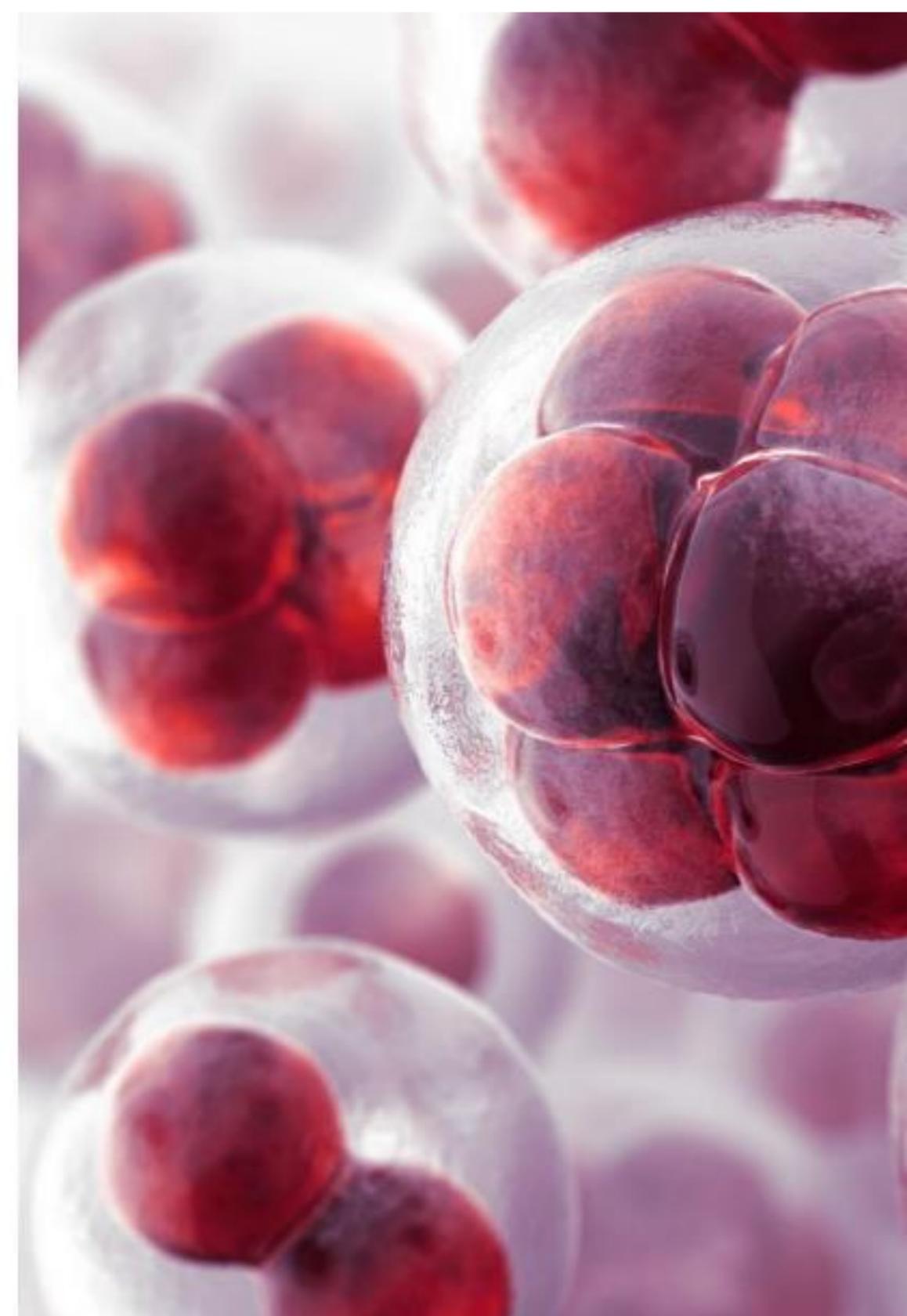
WHAT ARE THE GENETIC DAMAGES THAT CAN TRANSFORM CELLS?

- For a genetic damage to transform a cell, it has to cause uncontrolled proliferation.
- The majority of our cells proliferate continuously. This proliferation is regulated by certain genes. There is a balance between genes that stimulate growth and those inhibiting it.
Loosing this balance can cause uncontrolled proliferation.
- So : for cancer to occur there is stimulation of genes that cause cell proliferation, or downregulation of genes that inhibit proliferation.



ONCOGENES

- Normally: our cells have proto-oncogenes. These cause cell proliferation in a regulated manner
- If the proto-oncogenes are mutated or overexpressed: they are called oncogenes
- Proto-oncogenes encode for proteins: proto-oncoproteins, or oncoproteins
- These oncoproteins include: transcription factors, growth regulating proteins, proteins involved in cell survival.



ONCOGENES

- Oncogenes cause overexpression of proteins involved in cell growth.
- If one allele is mutated or overexpressed: there will be increase in the growth proteins, which is enough to increase cell growth
- So oncogenes are dominant .
- Important oncogenes : RAS and ABL



HOW ONCOGENES OVEREXPRESSED??

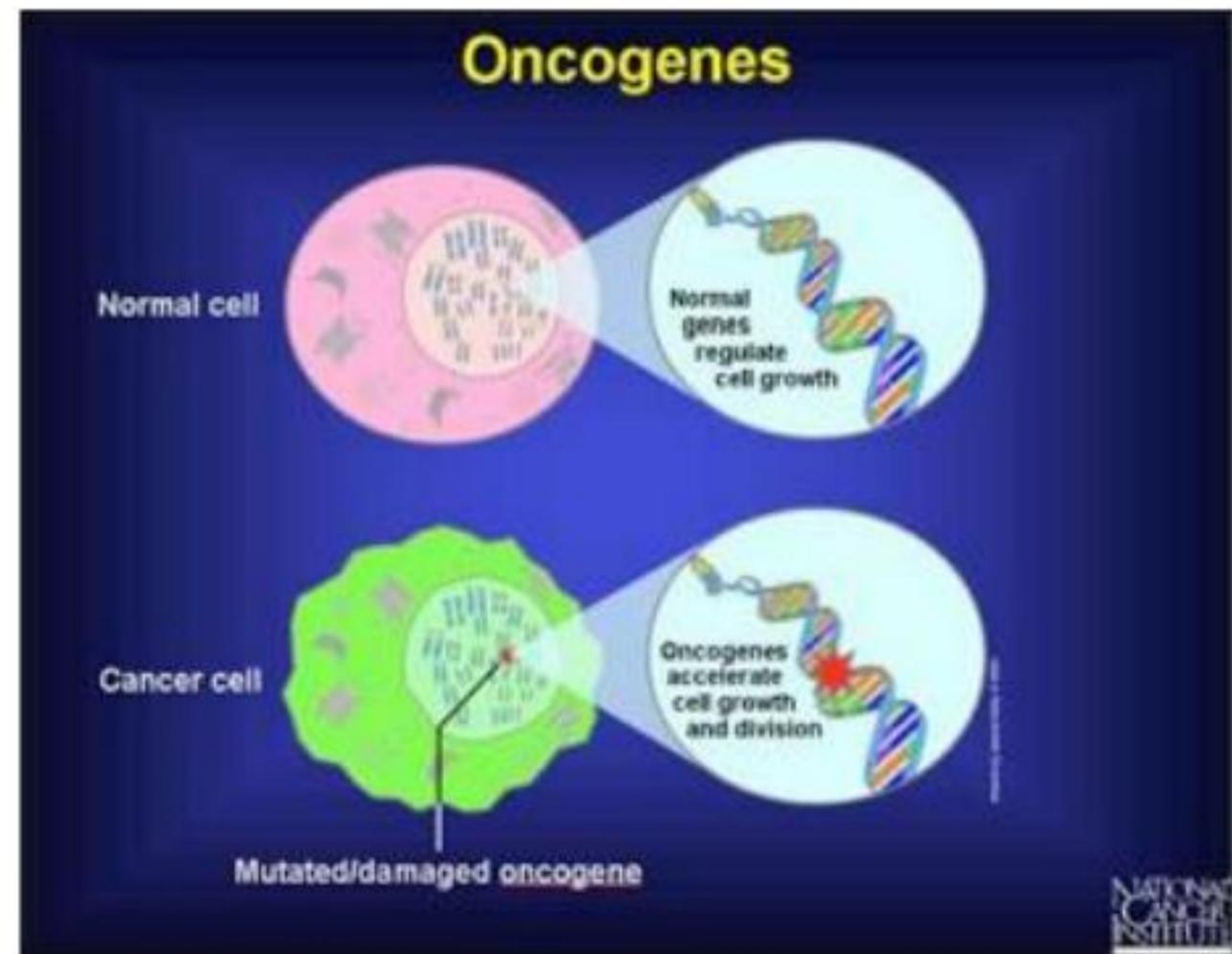
1. point mutation resulting in activation
2. amplification : increased number of copies of the oncogenes
3. translocations
4. Epigenetic modification



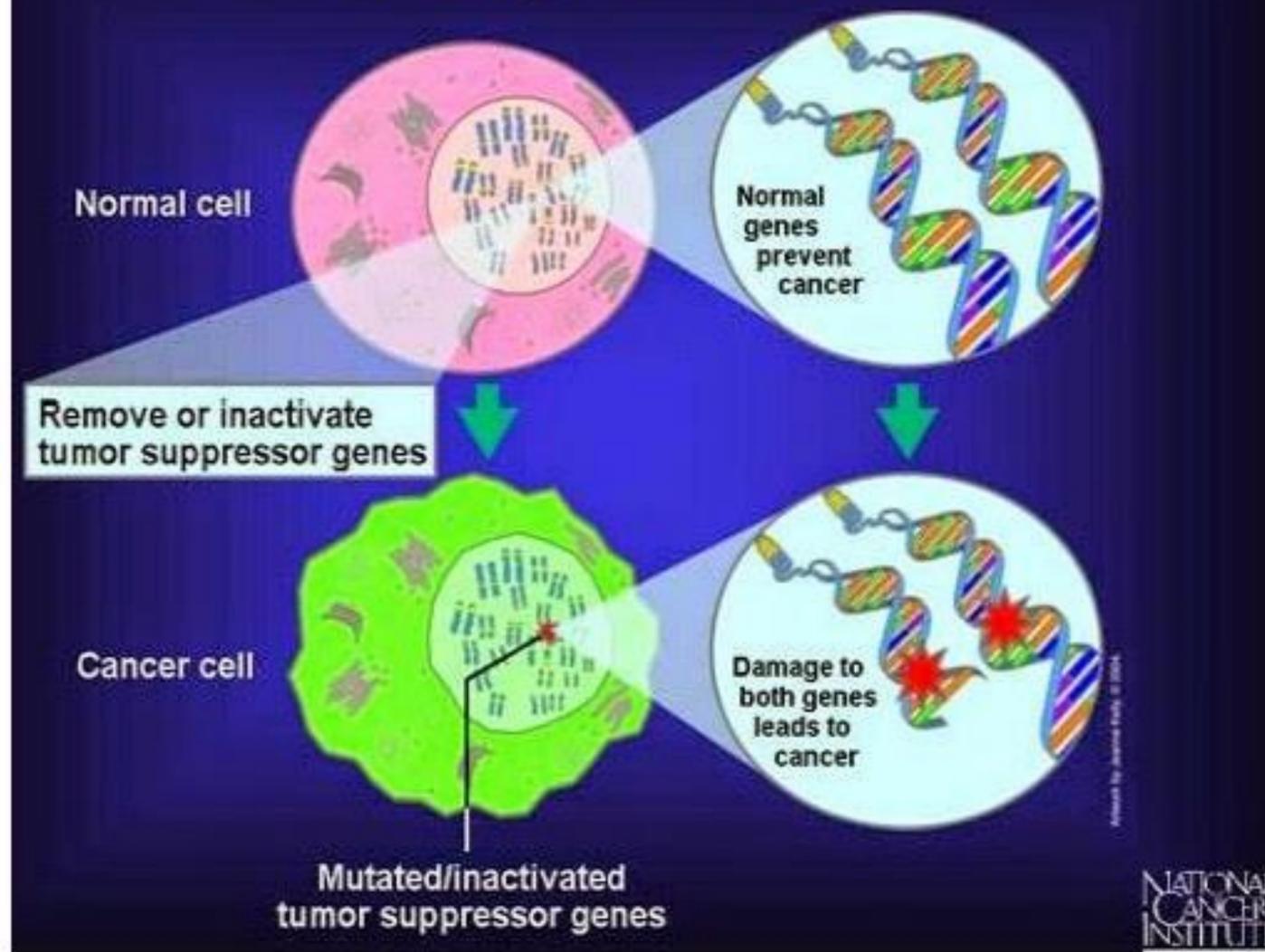
PROTO-ONCOGENES

Proto-oncogenes normally stimulate growth in a controlled manner. If they are mutated they cause uncontrolled growth (cancer)

Mutations convert proto-oncogenes into constitutively active oncogenes that endow the cell with growth self-sufficiency. These can be grouped in the following categories

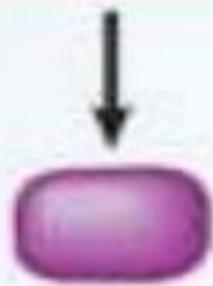
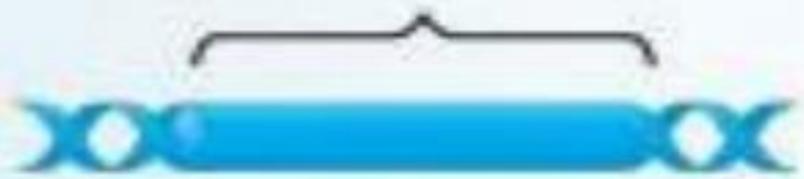


Tumor Suppressor Genes



TUMOR SUPPRESSOR GENES COUNTERACT THE FUNCTION OF THE ONCOGENES. IF THEY ARE INHIBITED BY A MUTATION THEN CELLS CAN PROLIFERATE WITHOUT THIS "BRAKING" EFFECT OF THE TUMOR SUPPRESSOR GENES.

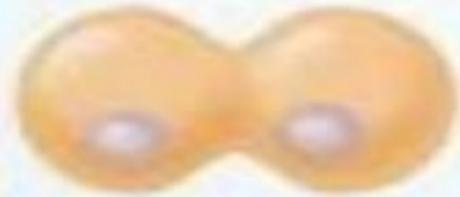
Tumor-suppressor gene



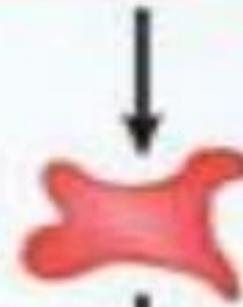
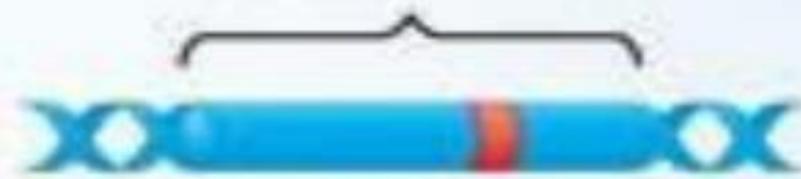
Normal growth - inhibiting protein



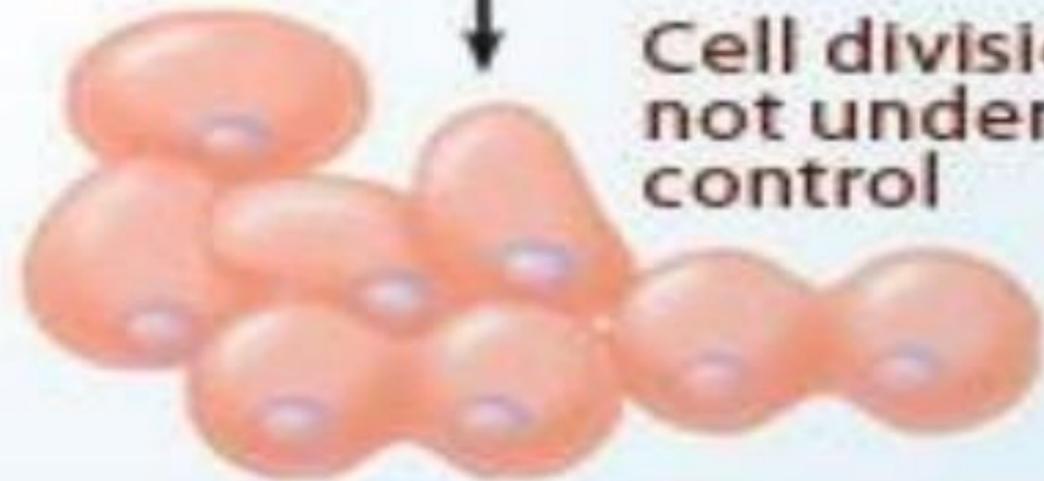
Cell division under control



Mutated tumor - suppressor gene



Defective, non functioning protein



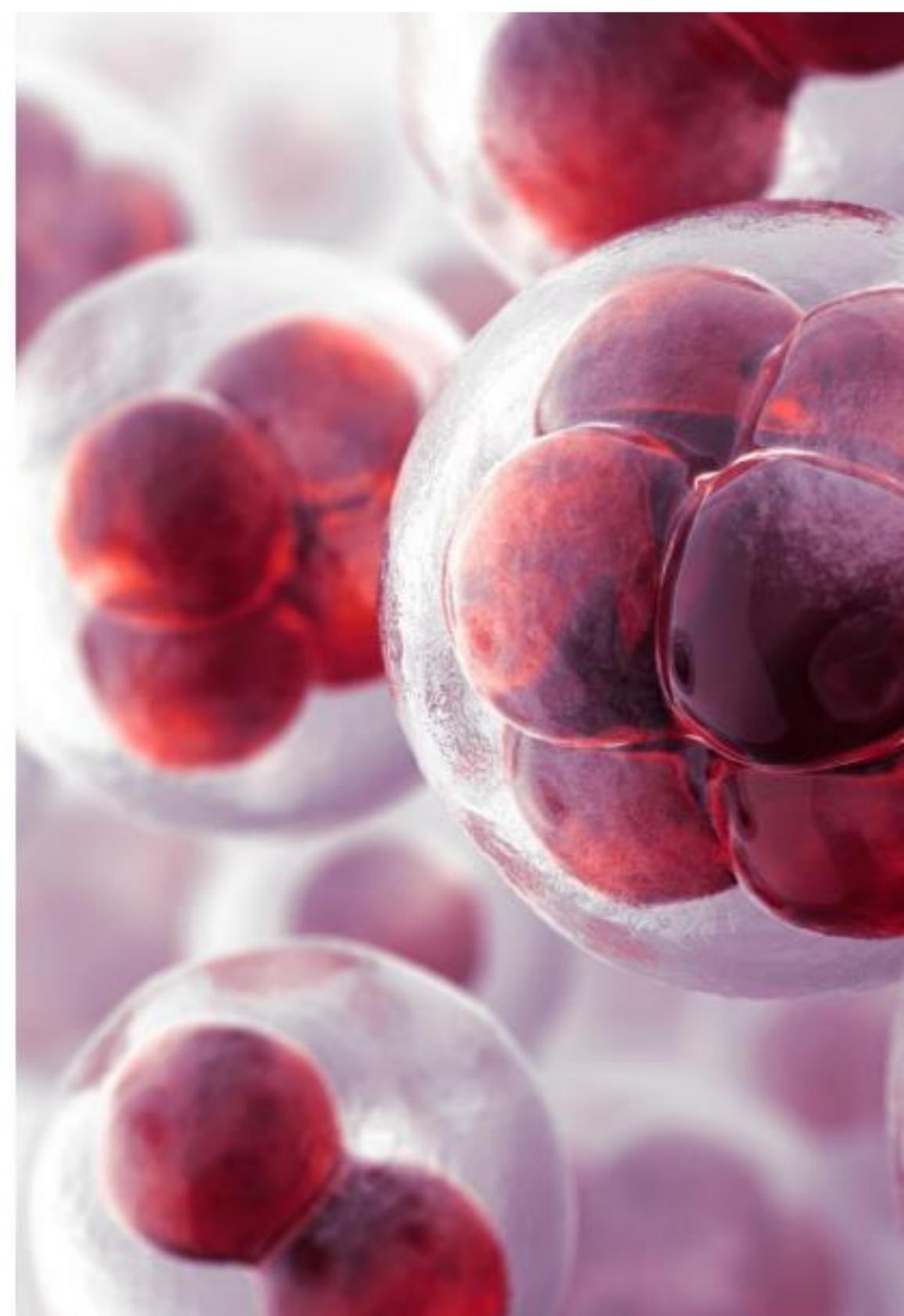
Cell division not under control

GENETIC DAMAGES IN NEOPLASMS

So: five types of regulatory genes are mainly affected:

- **1. growth promoting proto-oncogenes**
- **2. growth inhibiting tumor suppressor genes**
- **3. genes that regulate apoptosis**
- **4. genes involved in DNA repair.**
- **5. genes that regulate interactions between tumor cells and host cells .**

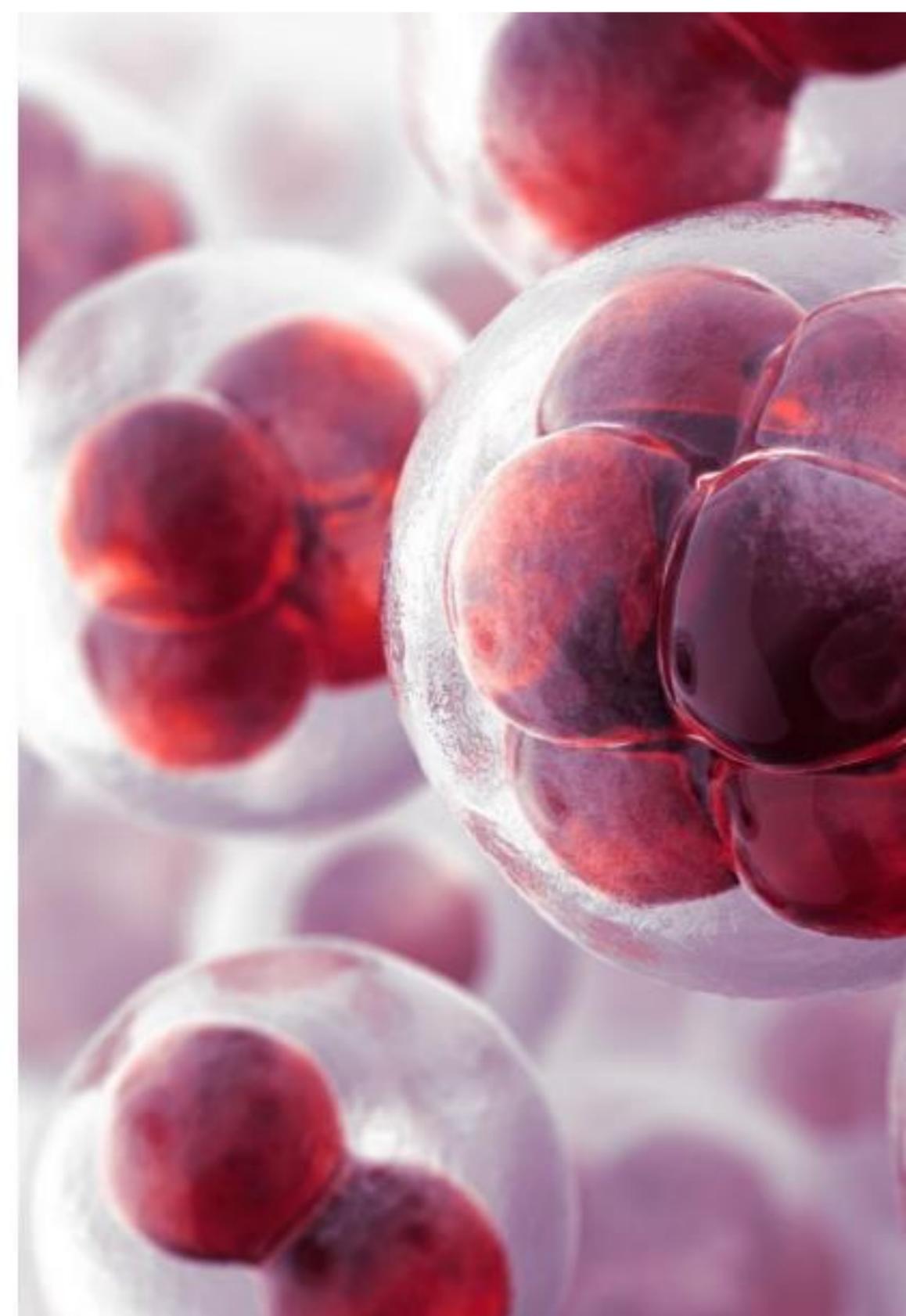
Particularly important are genes that enhance or inhibit recognition of tumors cells by the host immune system.



NOTE

- Normal genes that cause cell proliferation are traditionally called: proto-oncogenes.

- When they are mutated, they are called oncogenes.



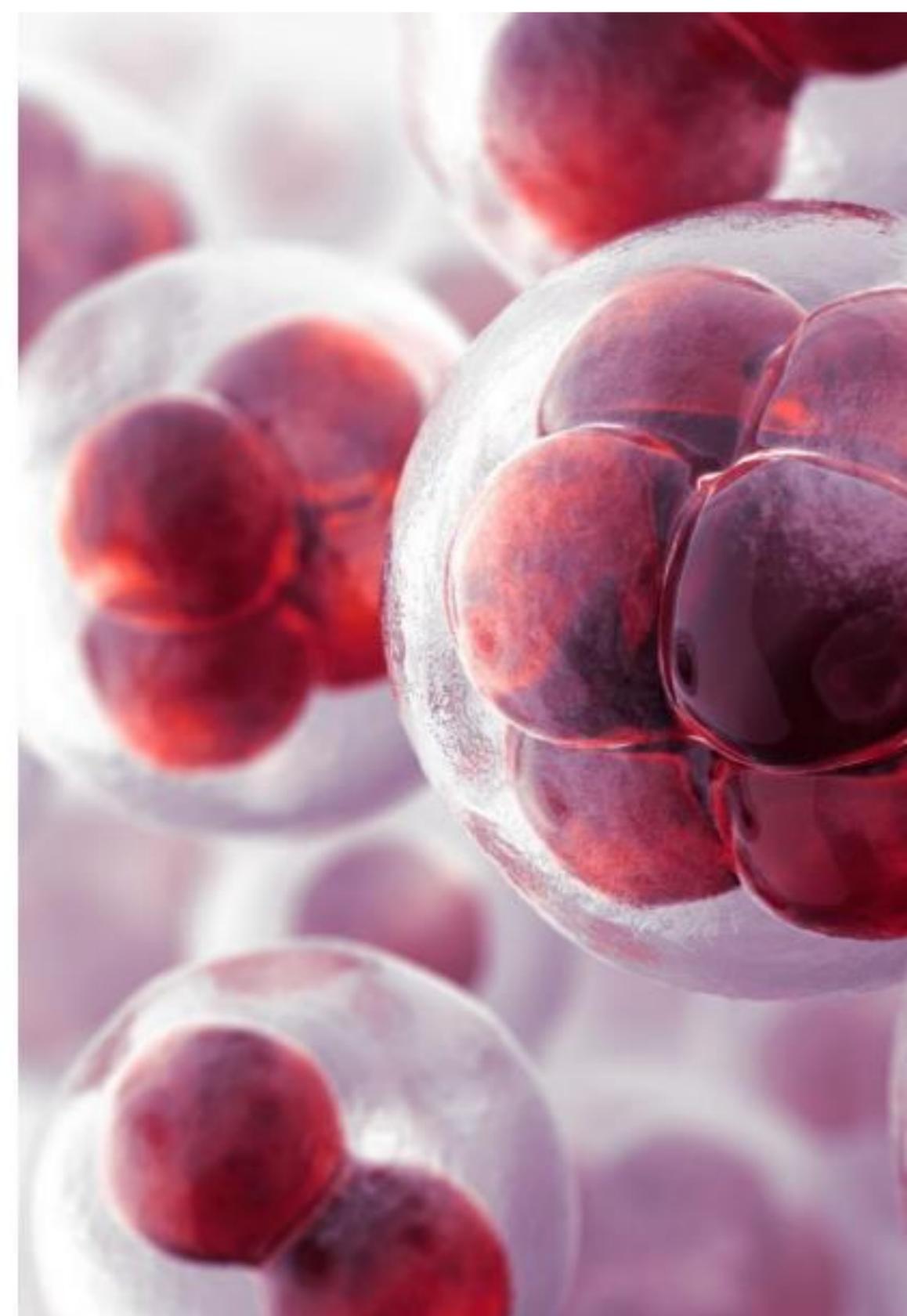
TUMOR SUPPRESSOR GENES

- They normally inhibit cell growth
- If mutated or lost: loss of growth inhibition : so tumors occur.
- *Both alleles need to be lost or mutated for the tumors to develop....*

Because if only one allele is lost , the other can compensate!

- So they are **recessive genes**
- In some cases loss of one allele is enough for transformation...

haploinsufficiency (see next slide for definition)



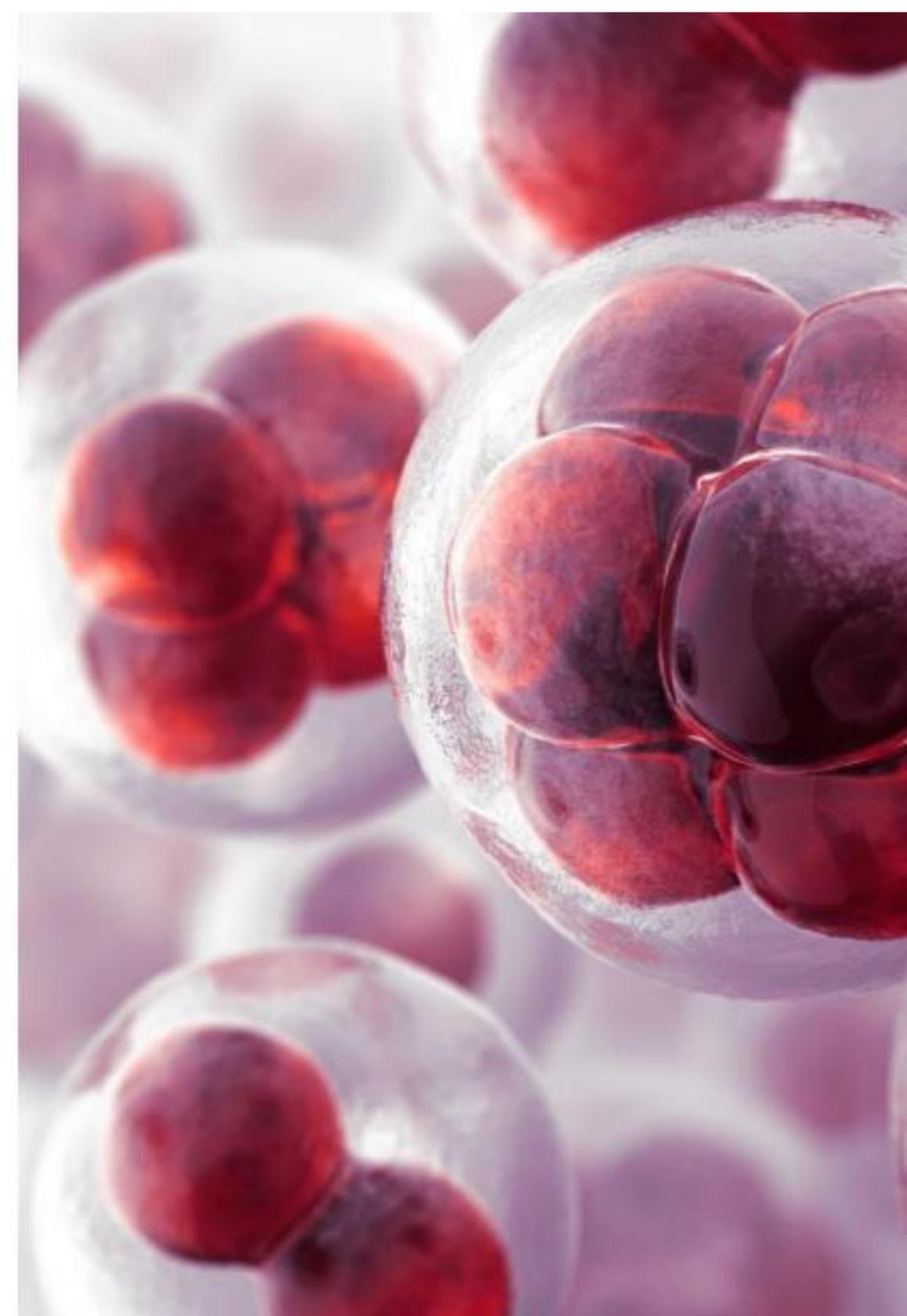
HAPLOINSUFFICIENCY

- **Haploinsufficiency:** A situation in which the total level of a gene product (a particular protein) produced by the cell is about half of the normal level and that is not sufficient to permit the cell to function normally.
- Another way to define haploinsufficiency is as a condition that arises when the normal phenotype requires the protein product of both alleles, and reduction of 50% of gene function results in an abnormal phenotype.



TUMOR SUPPRESSOR GENES

- Most important examples:
 1. RB gene (retinoblastoma gene) .. Governor: controls growth and puts a brake in cellular proliferation
 2. TP53 gene ... guardian of the genome... sense genetic damage. So if there is damage it causes cessation of proliferation or if the damage cannot be repaired it causes apoptosis.



TP53

- Mutations in TP53 do not directly transform cells. They permit and accelerate acquisition of mutations in oncogenes or tumor suppressor genes.
- Same rule applies for mutations in genes that are responsible for DNA repair.

Note:

- Genes that regulate apoptosis or DNA repair may be dominant or recessive.



POINT MUTATIONS

- These are single changes in nucleotides

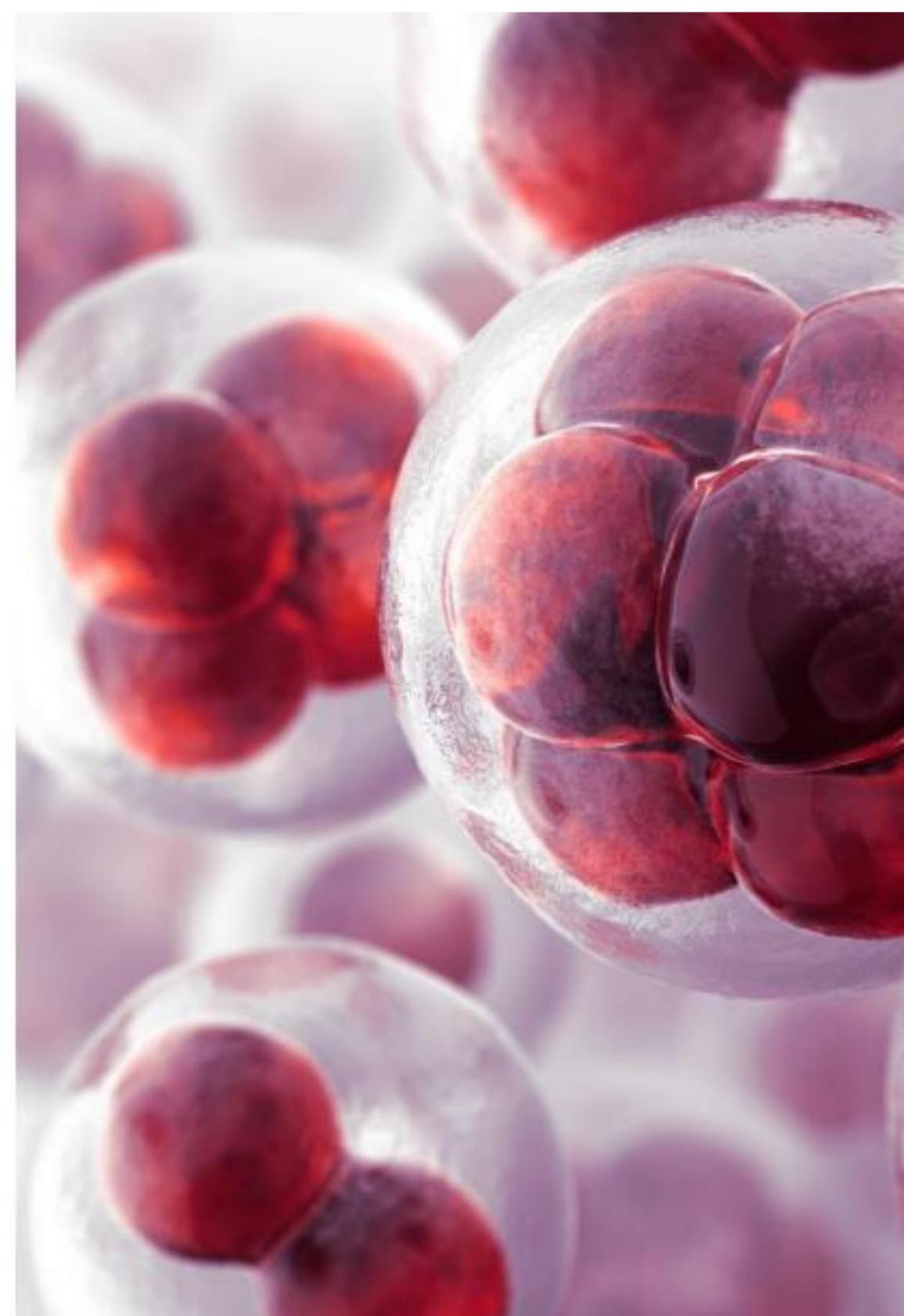
- Point mutations that stimulate an oncogene or inhibit both alleles of a tumor suppressor gene can result in cancer.

BALANCED TRANSLOCATIONS

- Translocations can cause cancer if they increase expression of a protooncogene.
- This can happen by two mechanisms:
 1. Removing the proto-oncogene from its normal, regulated locus to a new position where it becomes under influence of a highly active promoter.
 2. Translocation forms a new fusion gene that encodes a novel protein.

TRANSLOCATIONS

- Occur mainly in haematogenous neoplasms ; why ??
- Because lymphoid cells make DNA breaks during antibody or T cell receptor recombination. (loads of cutting and rearrangements of the genes... so there is more chance that a gene that was cut will be “pasted “ in a new locus!



TRANSLOCATIONS

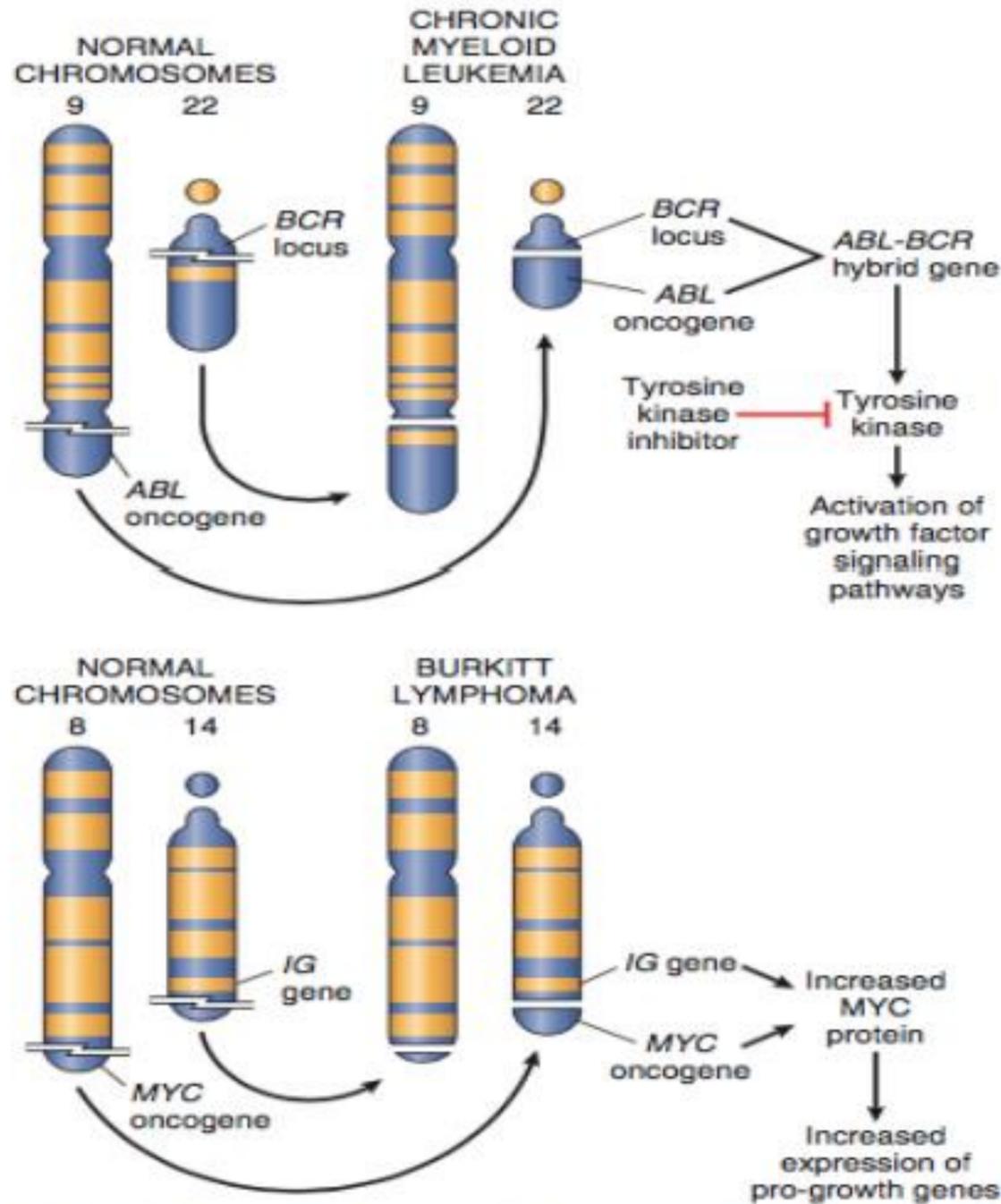
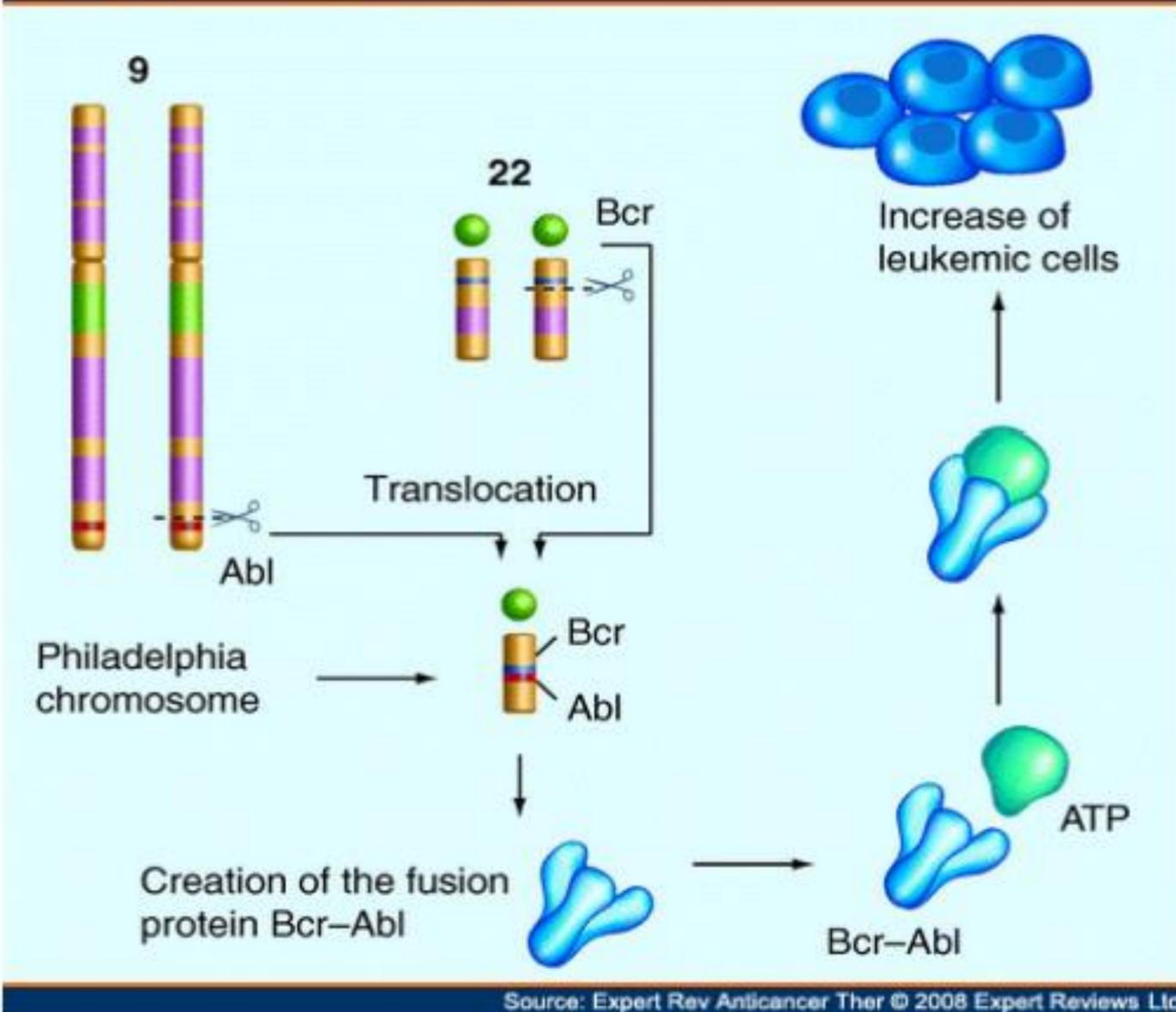


Fig. 6.14 The chromosomal translocations and associated oncogenes in chronic myelogenous leukemia and Burkitt lymphoma.

- In the upper example, the translocation created a new gene *ABL-BCR* from fusion of two genes (*ABL* and *BCR*). This created a new tyrosine kinase that can activate cell proliferation resulting in leukemia.
- in the other example in the pic, the translocation moved the *MYC* oncogene to a new locus (near the *IG* gene) that increased expression of the *MYC* gene resulting in increased cell proliferation



PHILADELPHIA CHROMOSOME:
AN EXAMPLE OF A
TRANSLOCATION
CAUSING A NEW PROTEIN (A
KINASE) THAT INCREASES CELL
PROLIFERATION.

GENE AMPLIFICATIONS

- Proto-oncogenes can be amplified and overexpressed .. Converted to oncogenes.
- This is seen in karyotyping as two patterns :
 - 1.homogenously stained region (HSR) = increased copies of the gene present within the chromosome
 - 2.Double minutes: extra copies of the gene separated from the chromosome.

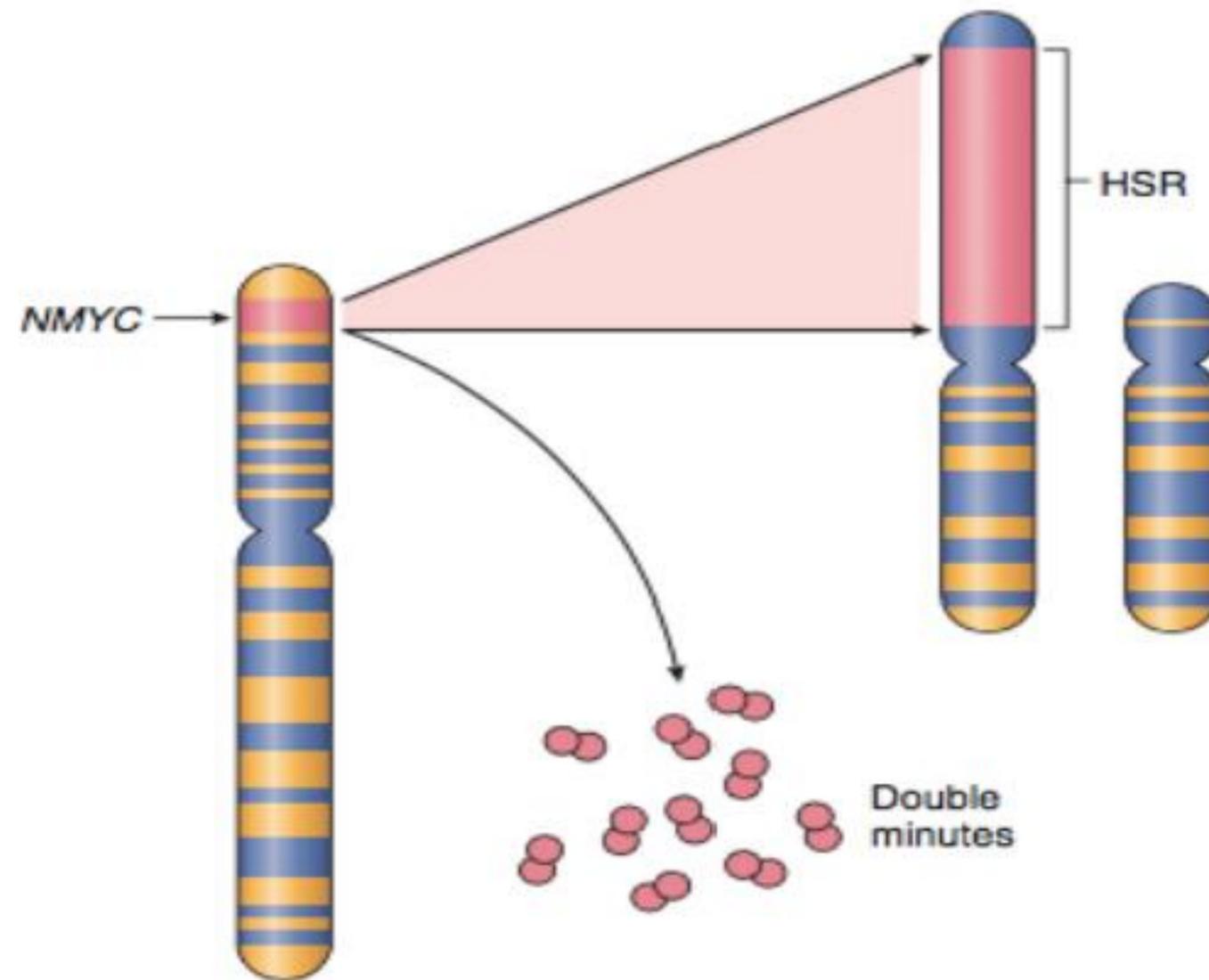
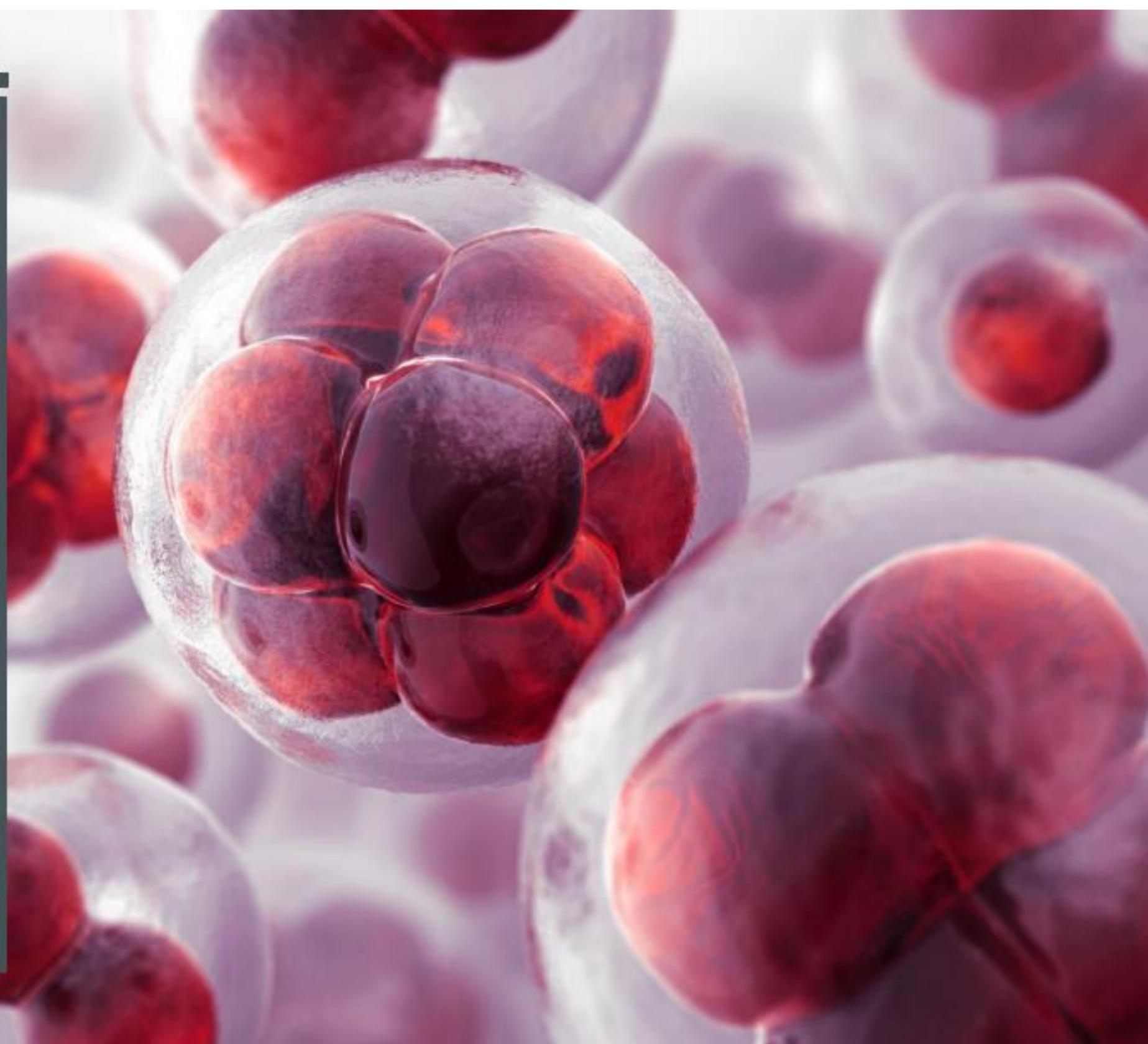


Fig. 6.15 Amplification of the *NMYC* gene in human neuroblastoma. The *NMYC* gene, present normally on chromosome 2p, becomes amplified and is seen either as extrachromosomal double minutes or as a chromosomally integrated homogeneous-staining region (HSR). The integration involves other autosomes, such as 4, 9, or 13. (Modified from Brodeur GM, Seeger RC,

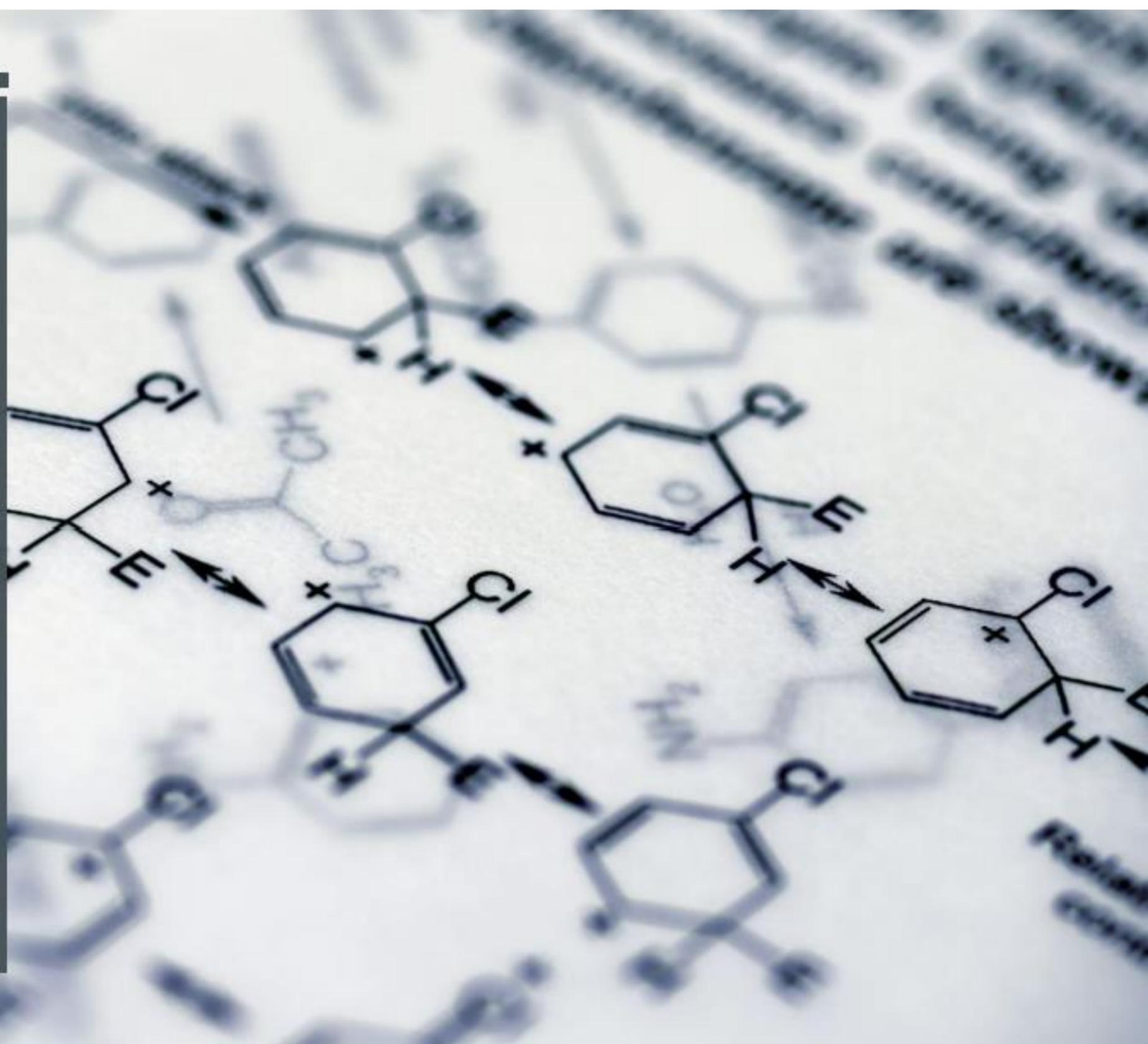
DELETIONS

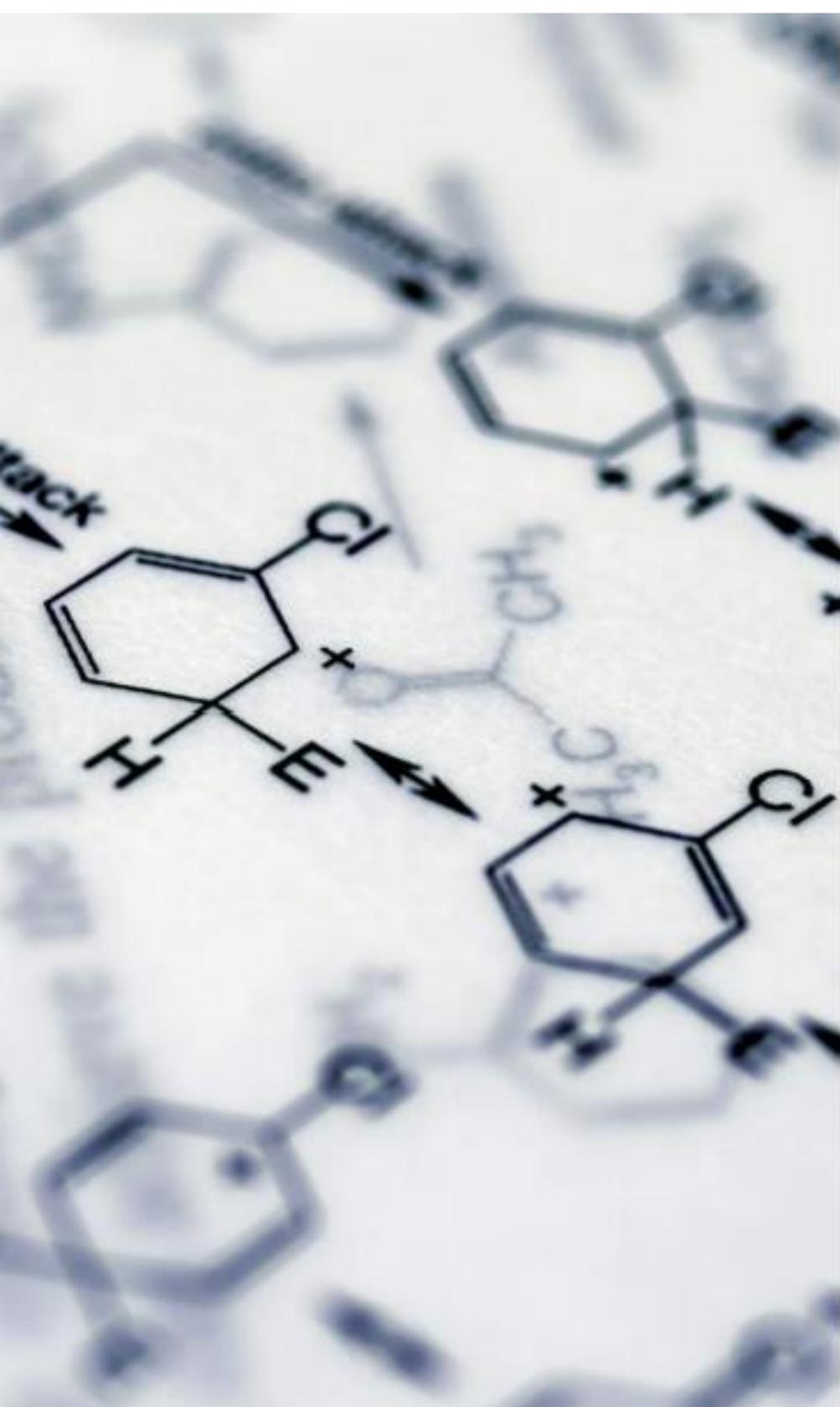
- More in non-hematopoietic solid tumors
- Second most common karyotypic abnormality.
- Result in loss of tumor suppressor genes
- 2 copies of the tumor suppressor gene need to be lost, usually one by point mutation and another by deletion



ANEUPLOIDY

- = number of chromosomes not multiple of the haploid state (23).
- Result from errors of the mitotic checkpoint



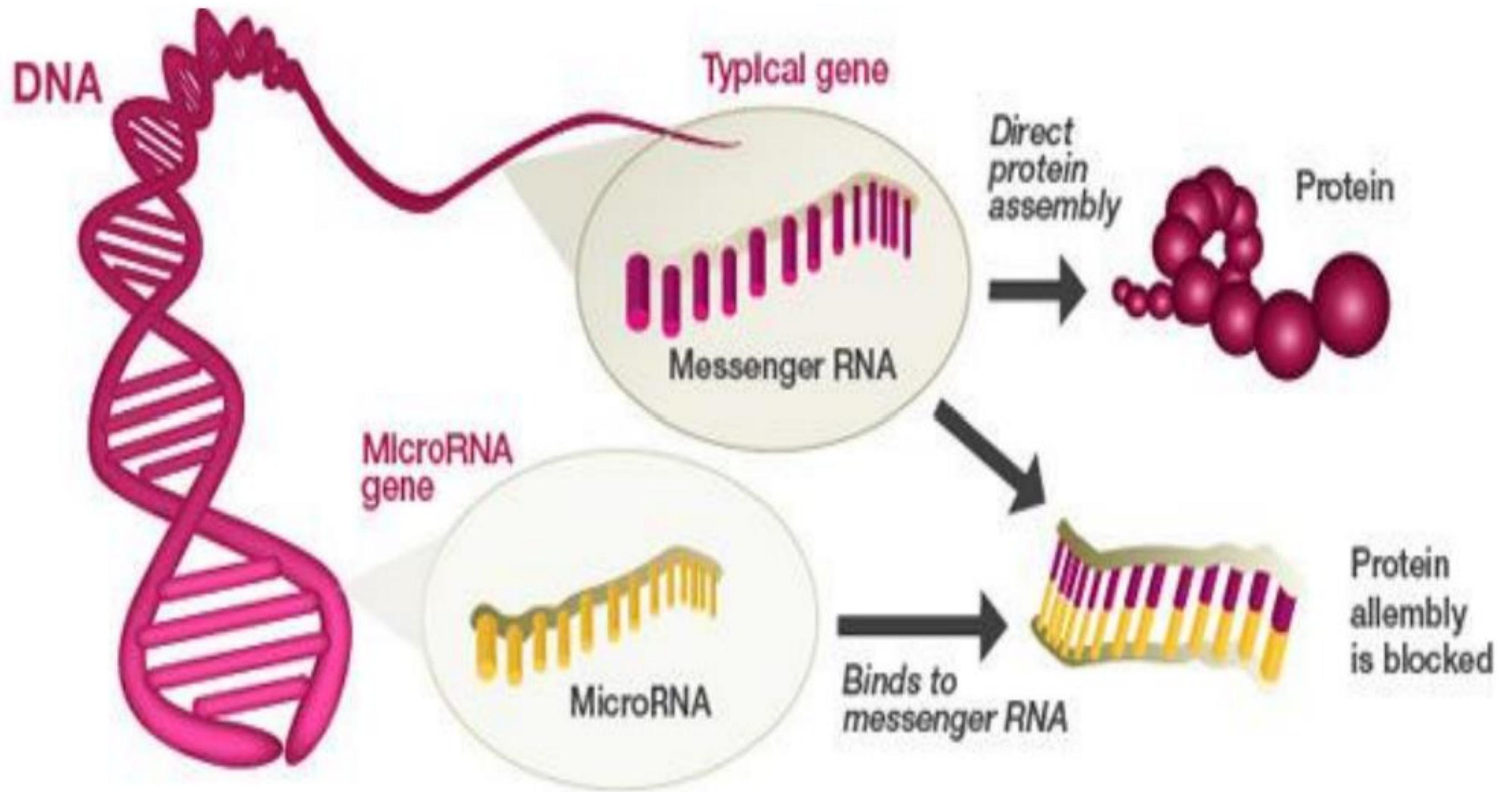


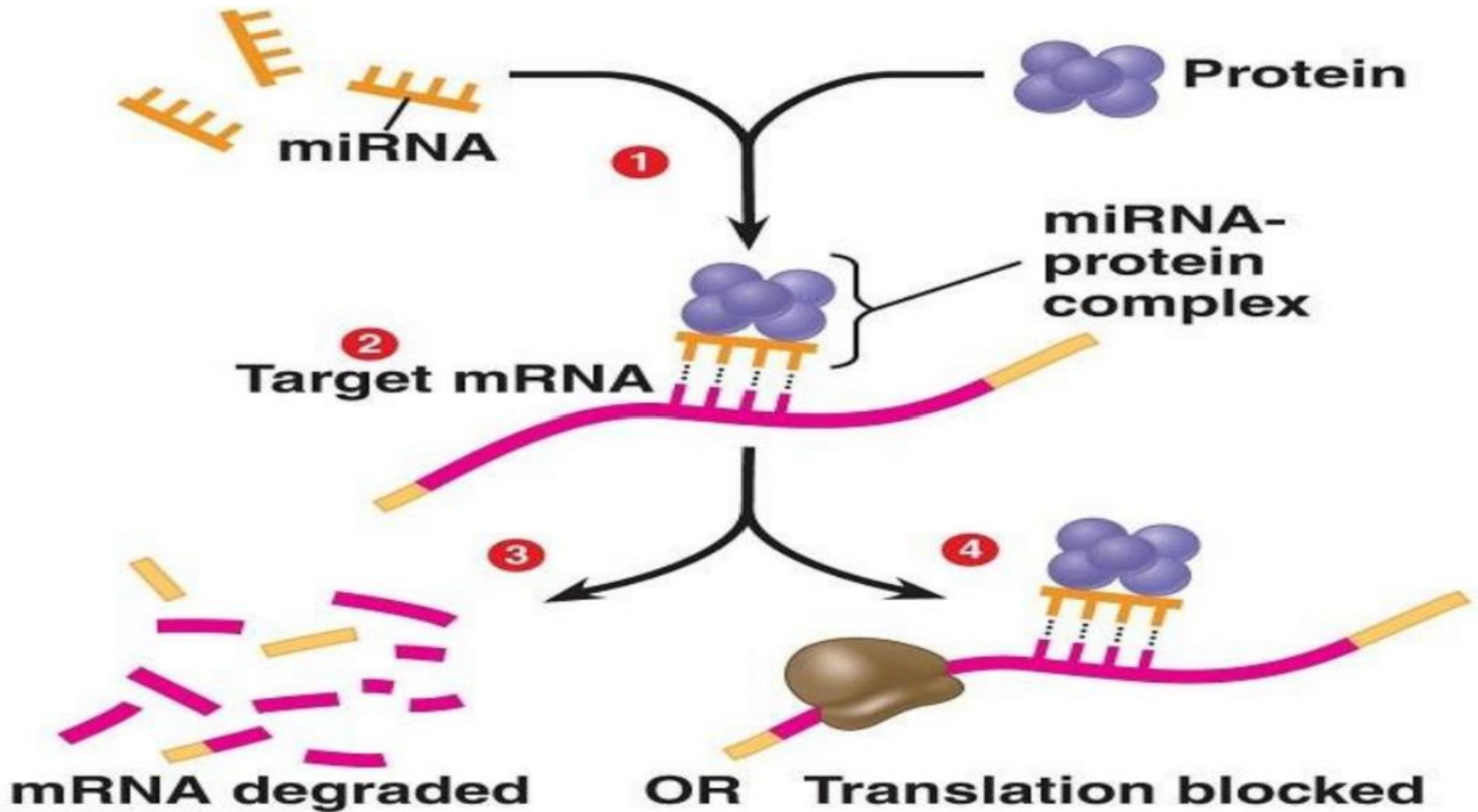
MICRORNAS (MIRNAS)

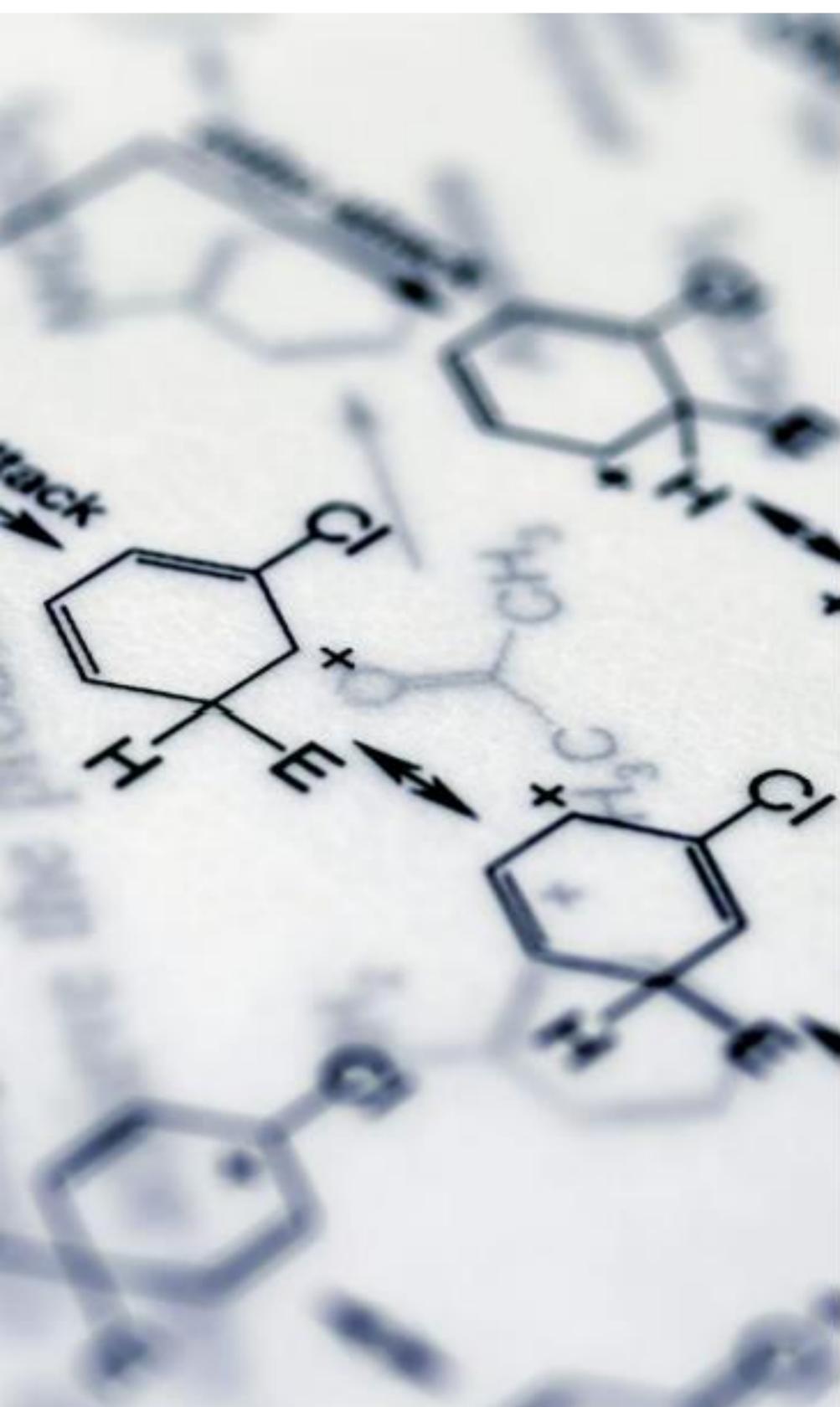
- **Noncoding**, micro RNA segments (22 nucleotides) that are **negative regulators** of the genes.
- They inhibit gene expression **post-transcriptionally** = repress translation or cleave mRNA.
- SO: transcription occurs = messenger RNA formed.. But mRNA is not translated to a protein.
- microRNA can inhibit translation or cleave the messenger (tears the message before it is read)

MIRNA

- Cause cancer by increasing oncogene expression or decreasing tumor suppressor gene expression.
- miRNAs that target oncogenes.... If reduced, then inhibition caused by microRNA is lost causing overexpression of oncogenes.
- miRNAs that target tumor suppressor genes... if increased they cause downregulation of tumor suppressor genes, resulting in cancer (as if we are functionally reducing the tumor suppressor genes)





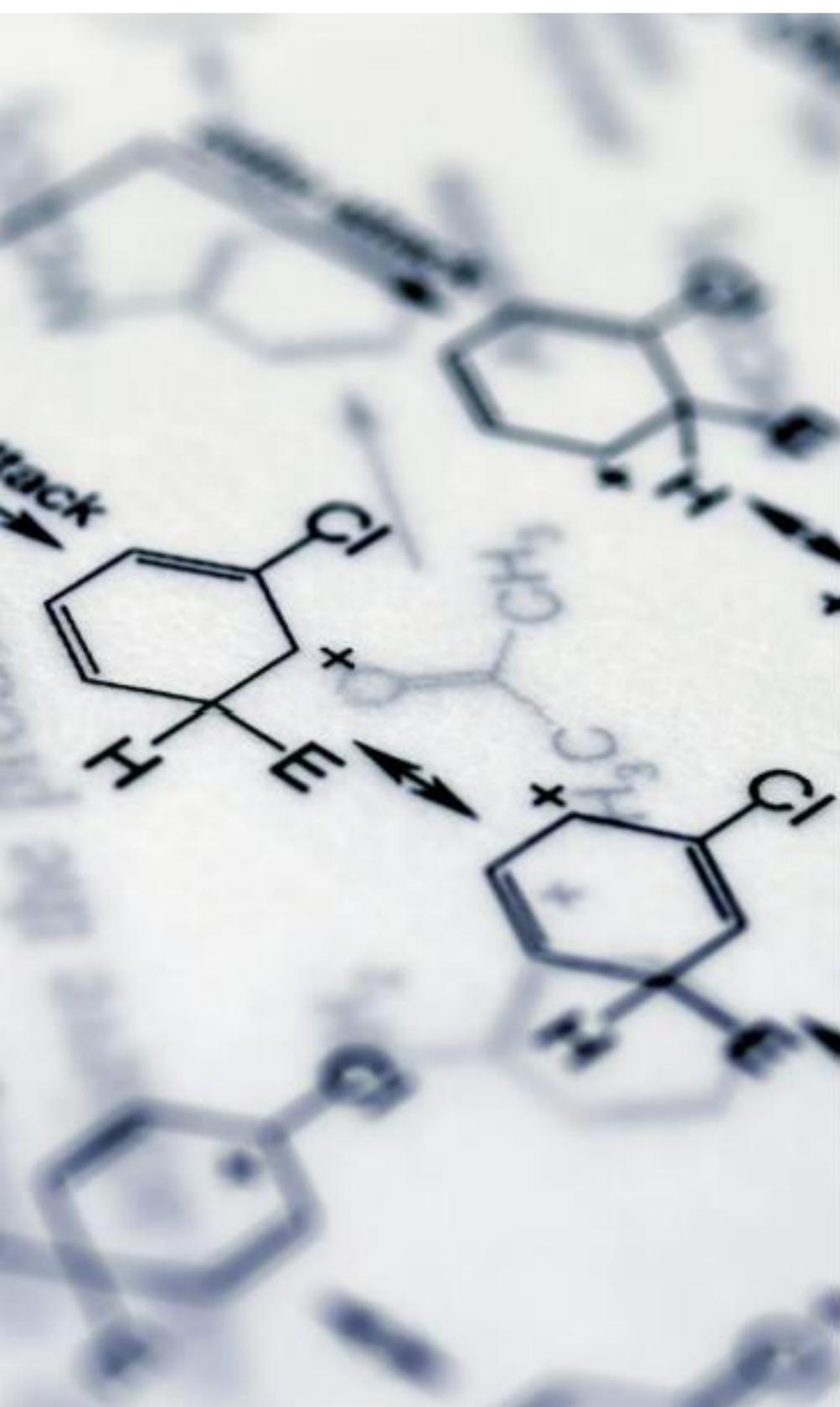


EPIGENETICS

- Epigenetics are reversible, heritable changes in gene expression that occur without mutation.

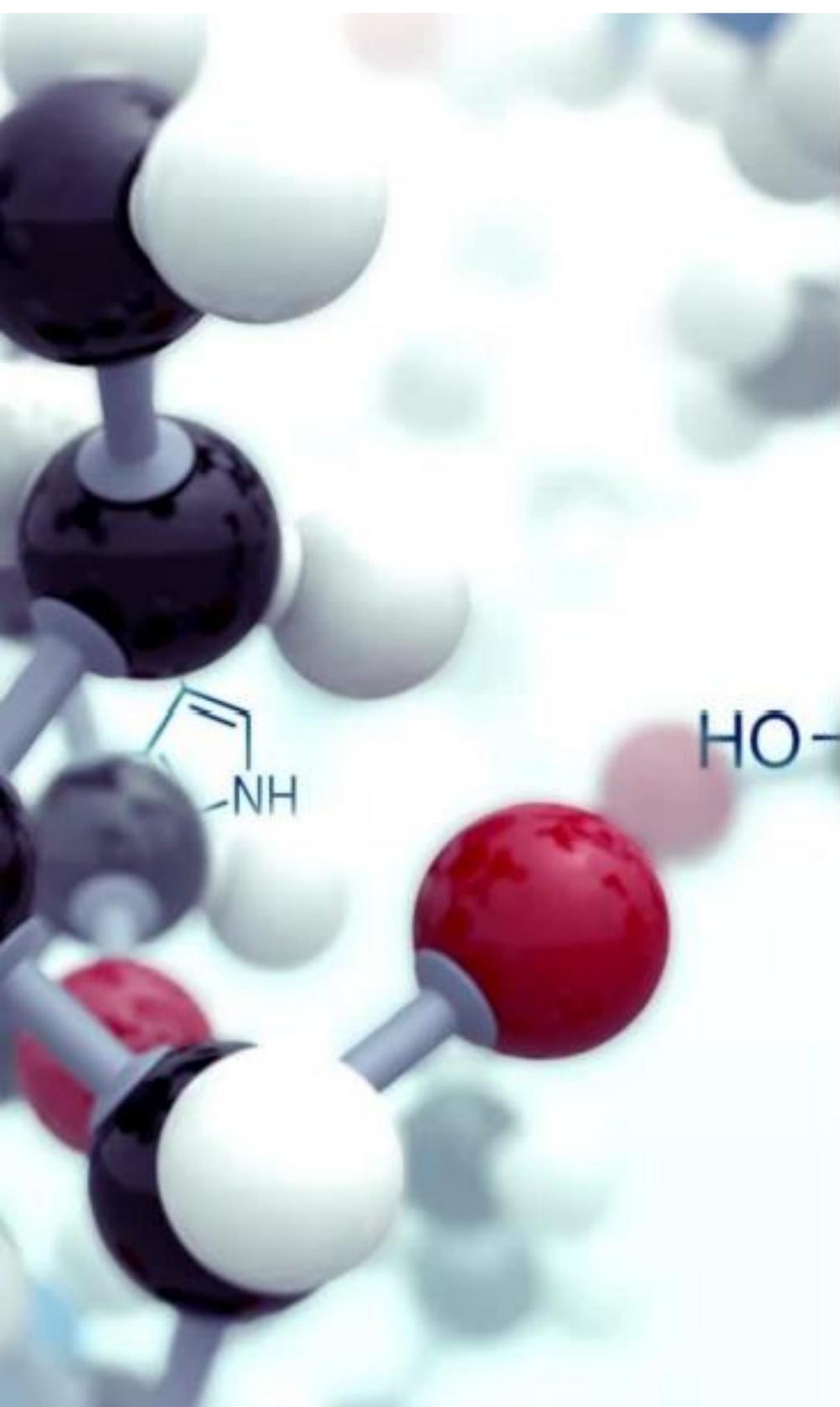
Epigenetic mutations: functionally relevant changes to the genome that do not involve a change in the nucleotide sequence.

Examples of mechanisms that produce such changes are DNA methylation and histone modification, each of which *alters how genes are expressed without altering the underlying DNA sequence.*



EPIGENETIC MODIFICATIONS

- Reversible , heritable changes in gene expression without mutation.
- Two types: Histone modifications and DNA methylation.



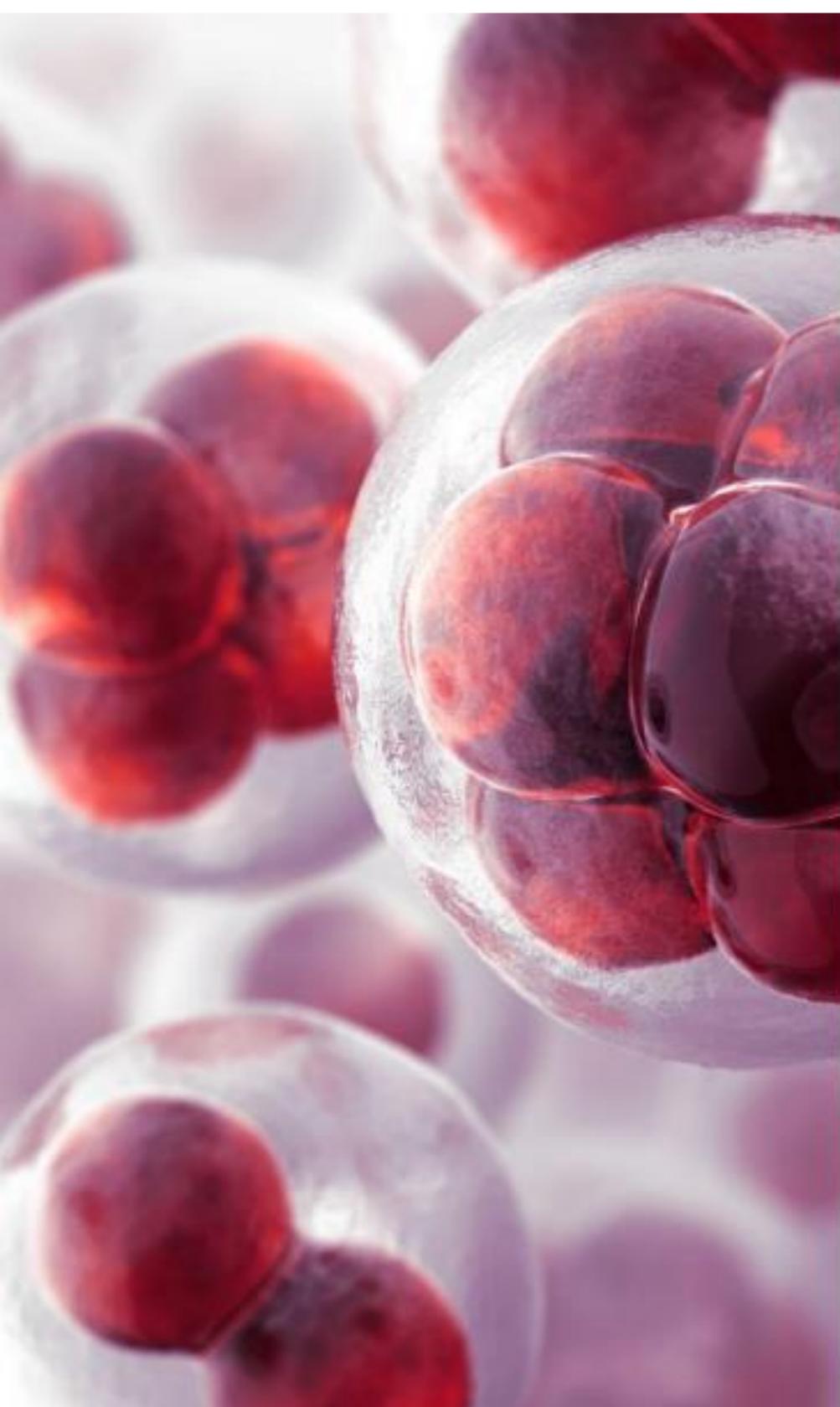
EPIGENETICS AND CANCER

- Gene expression is silenced by DNA methylation= more methyl groups lead to more silencing.

In cancer cells:

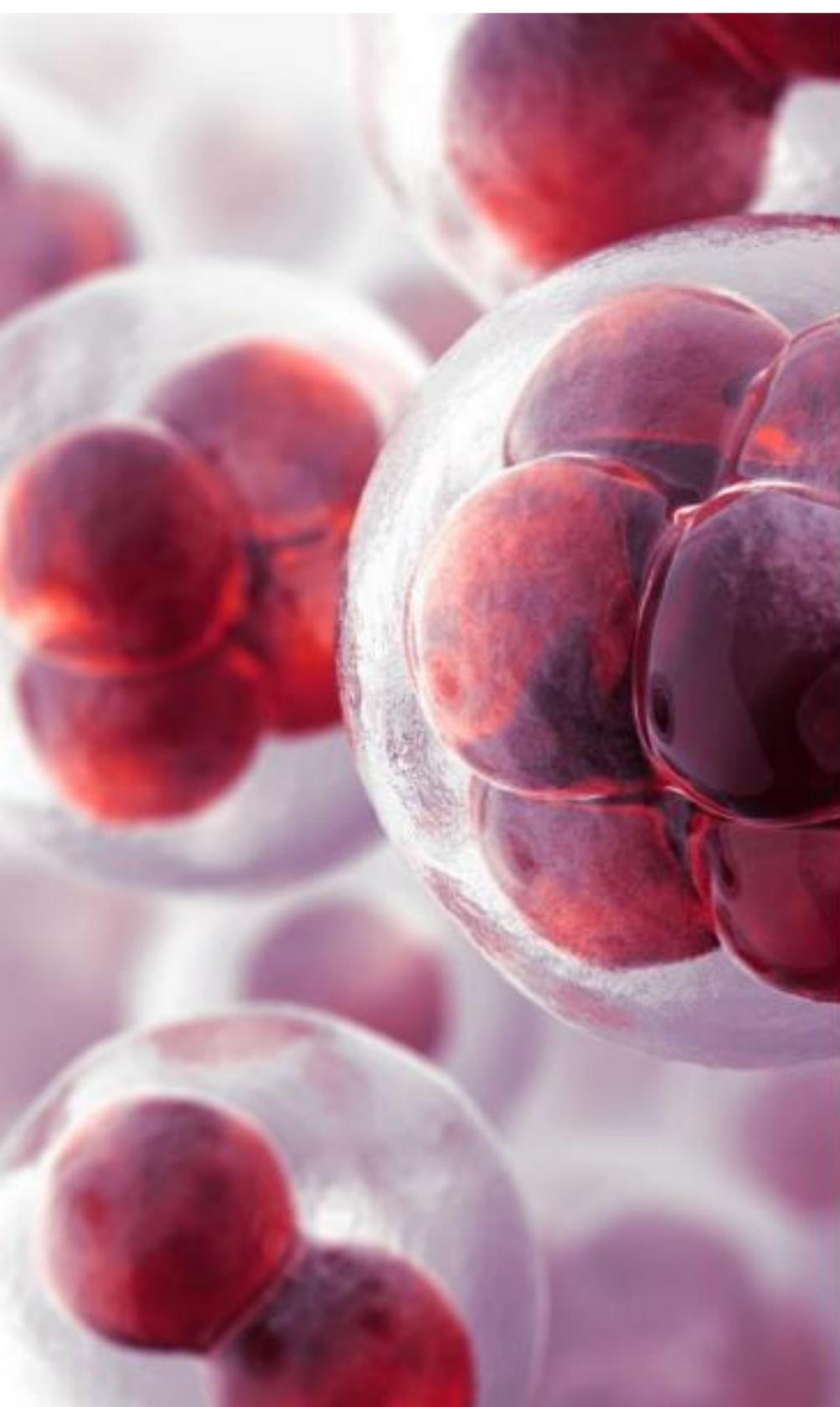
1. Global DNA hypo methylation : increases expression of genes.
Also causes chromosomal instability

2. Selective promoter hyper methylation of tumor suppressor genes: silenced

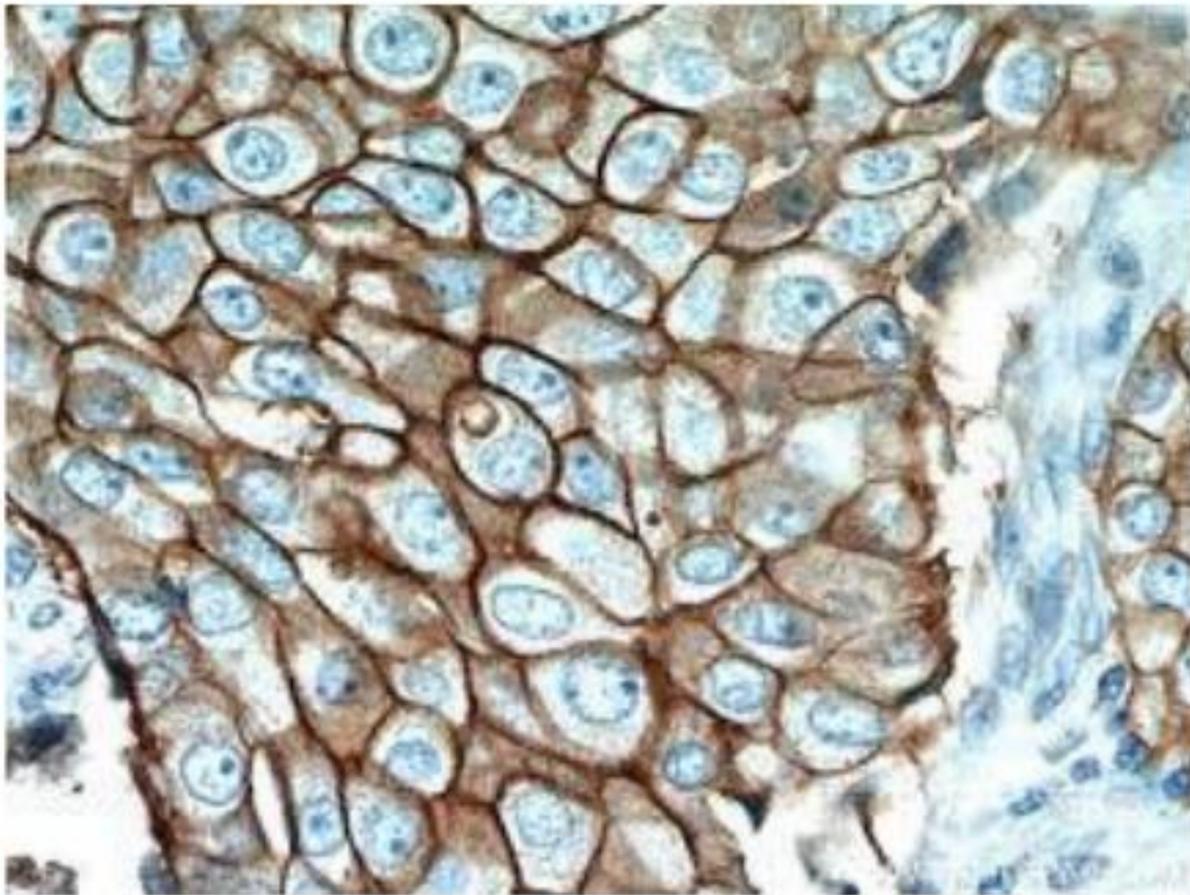


CASE STUDY: APPLICATION OF TODAY'S LECTURE

- A 66 year old lady had a breast lump.
- A biopsy was taken and examined histologically.
- The biopsy reported as follows: the breast biopsy shows infiltration by glandular structures lined by epithelial cells showing large hyperchromatic nuclei.
- Question: Does this description indicate a benign or malignant tumor?
- Answer: definitely malignant..The presence of infiltration and the anaplastic features described mean its malignant.
- Question: can you indicate the type of this malignancy from the description??



-
- Answer: the report describes glandular structures, so this is an adenocarcinoma.
 - Note: adenocarcinoma in the breast is called invasive ductal carcinoma.
 - Ok: the pathologist then mentions that they did a stain for EGFR(epidermal growth factor receptor) which is an epidermal growth receptor. And in this patient the EGFR was positive.
 - What does this mean????



- EGFR is a receptor, so it is expressed on the cell membrane
- In the pic below you see brown color around the cells (membrane staining). This means this tumor has high level of this receptor.
- How this increase happened?

- 
- 
- EGFR is a protein.. So in this tumor its production is increased.
 - This increase was found to be due to amplification of the gene encoding this protein (HER2/neu)
 - This is an example of an increased oncoprotein due to amplification of an oncogene.
 - Patients with this mutation can be treated by a drug that targets and inhibits this gene which will decrease the EGFR production.This will deprive the tumor cells from the receptor which increases the proliferation.
 - This is an example of why we need to know the genetic mutations in cancers..We can develop specific treatments that target the mutation.