Molecular basis of Cancer

➢Cancer is an unregulated proliferation of cells due to loss of normal controls, resulting in unregulated growth, lack of differentiation, local tissue invasion and metastasis.

➢Cancer can develop in any tissue or organ at any age.

➢Carcinogenesis is a multistep process at both the phenotypic and the genetic levels

➢Normal body cells grow, divide, and die in an orderly fashion. During the early years of a person's life, normal cells divide more rapidly until the person becomes an adult. After that, cells in most parts of the body divide only to replace worn-out or dying cells and to repair injuries.

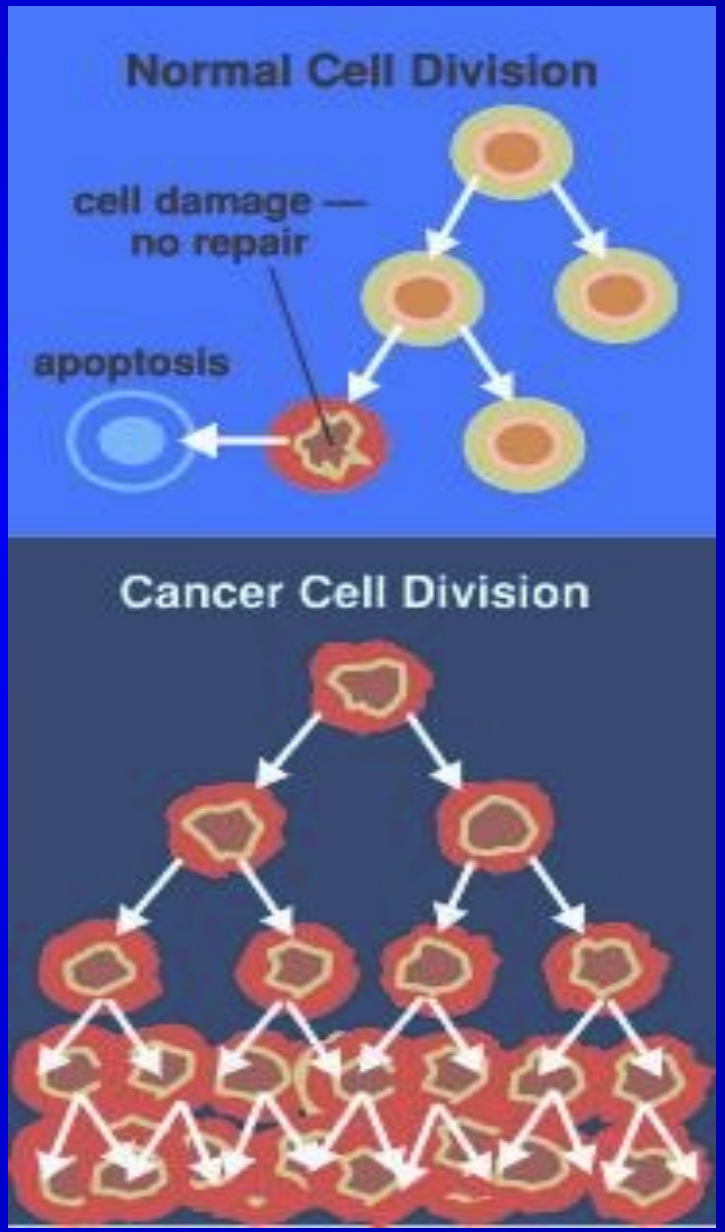
- Cancer cells develop because of damage to DNA. Most of the time when DNA becomes damaged the body is able to repair it.

- In cancer cells, the damaged DNA is not repaired. People can inherit damaged DNA, which accounts for inherited cancers. Many times though, a person's DNA becomes damaged by exposure to something in the environment, like smoking

- Because cancer cells continue to grow and divide, they are different from normal cells. Instead of dying, they outlive normal cells and continue to form new abnormal cells.

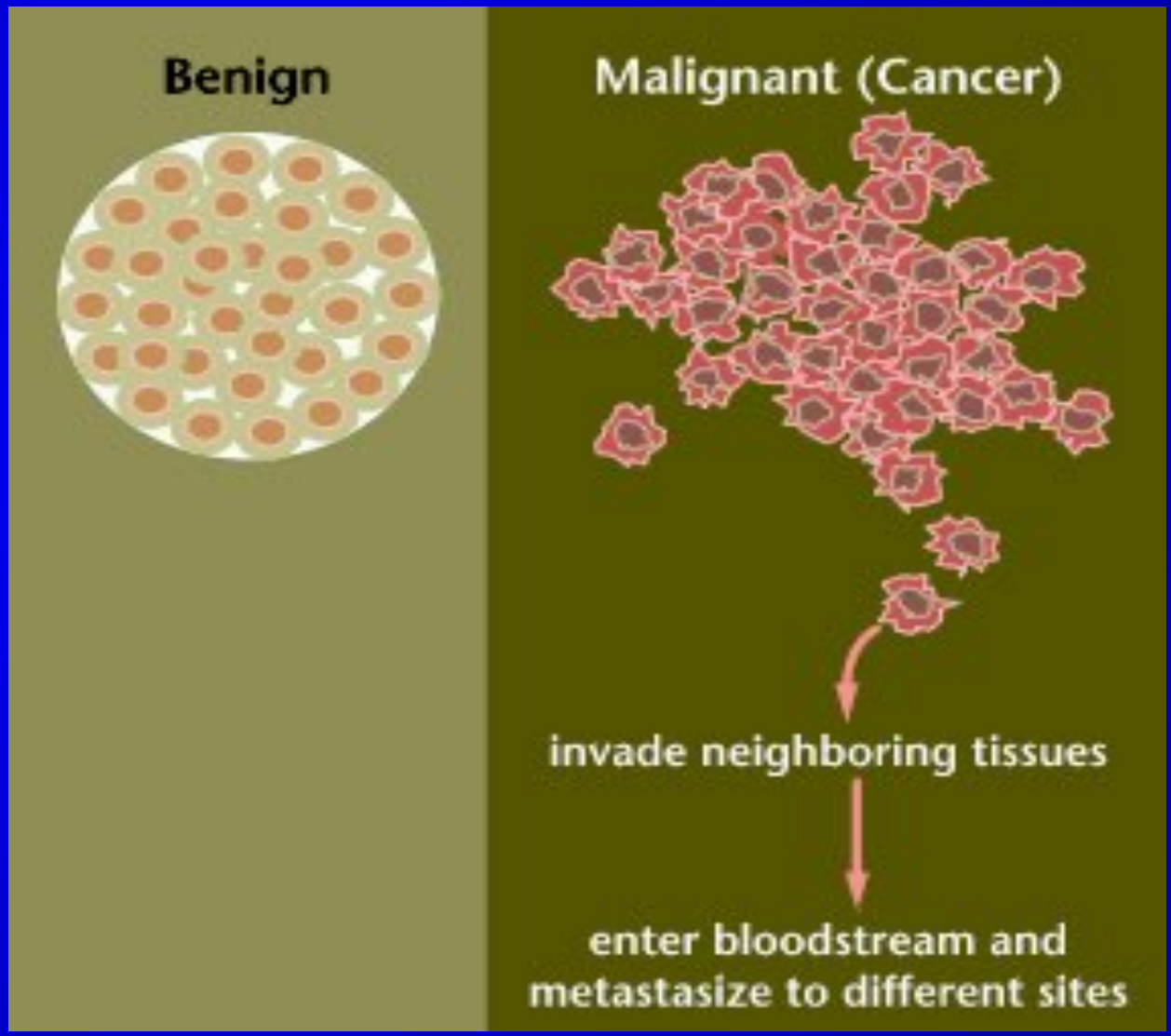
Cellular Basis of Cancer

- Cancer is a collection of diseases characterized by abnormal and uncontrolled growth
- Cancer arises from a loss of normal growth control
- In normal tissues, the rates of new cell growth and old cell death are kept in balance
- In cancer, this balance is disrupted
- This disruption can result from
  1) uncontrolled cell growth or
  2) loss of a cell's ability to undergo apoptosis

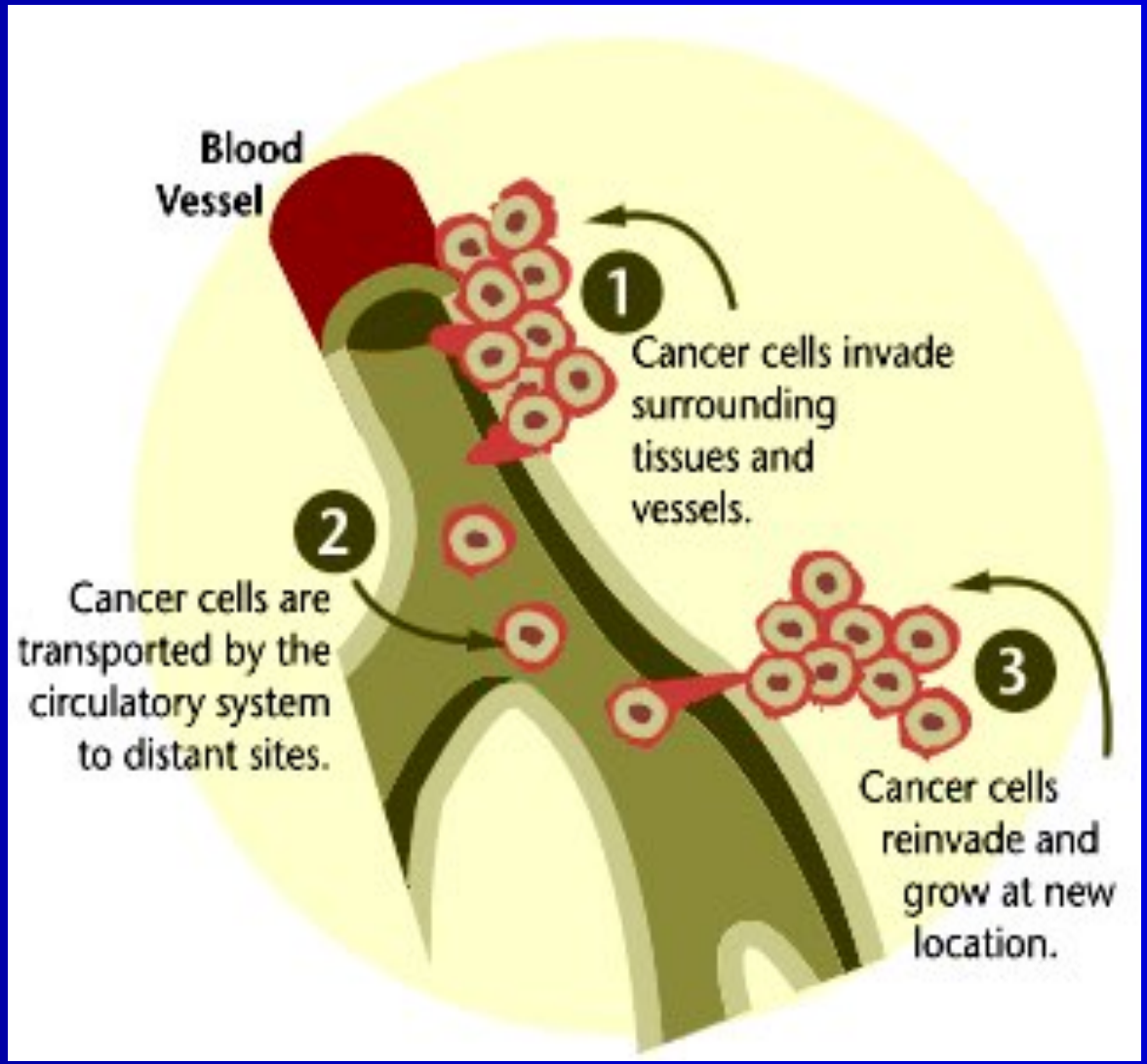


Malignant versus Benign Tumors

- Benign tumors generally do not spread by invasion or metastasis
- Malignant tumors are capable of spreading by invasion and metastasis



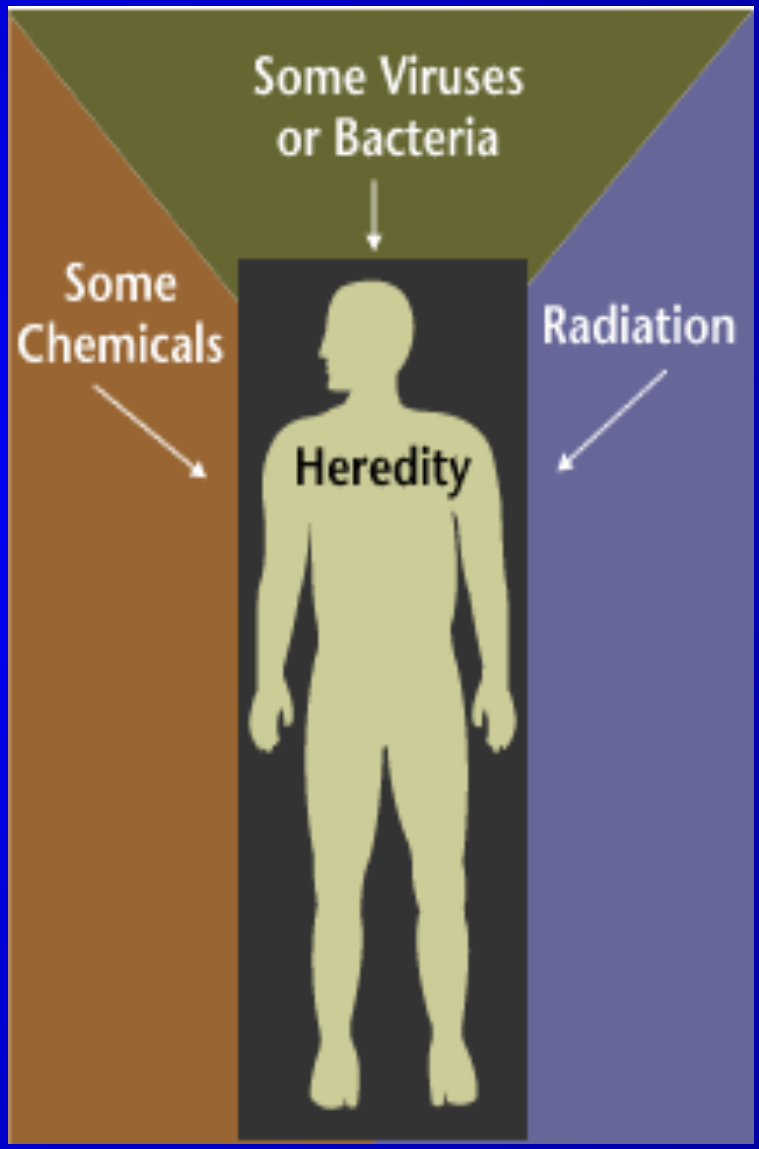
Invasion and Metastasis



- **Abnormal cells proliferate and spread (metastasize) to other parts of the body**
- **Invasion - direct migration and penetration into neighboring tissues**
- **Metastasis - cancer cells penetrate into lymphatic system and blood vessels**

What causes Cancer?

- Cancer is caused by alterations or mutations in the genetic code
- Can be induced in somatic cells by:
  - Carcinogenic chemicals
  - Radiation
  - Some viruses
- Heredity - 5%



Six acquired capabilities of cancer cells:

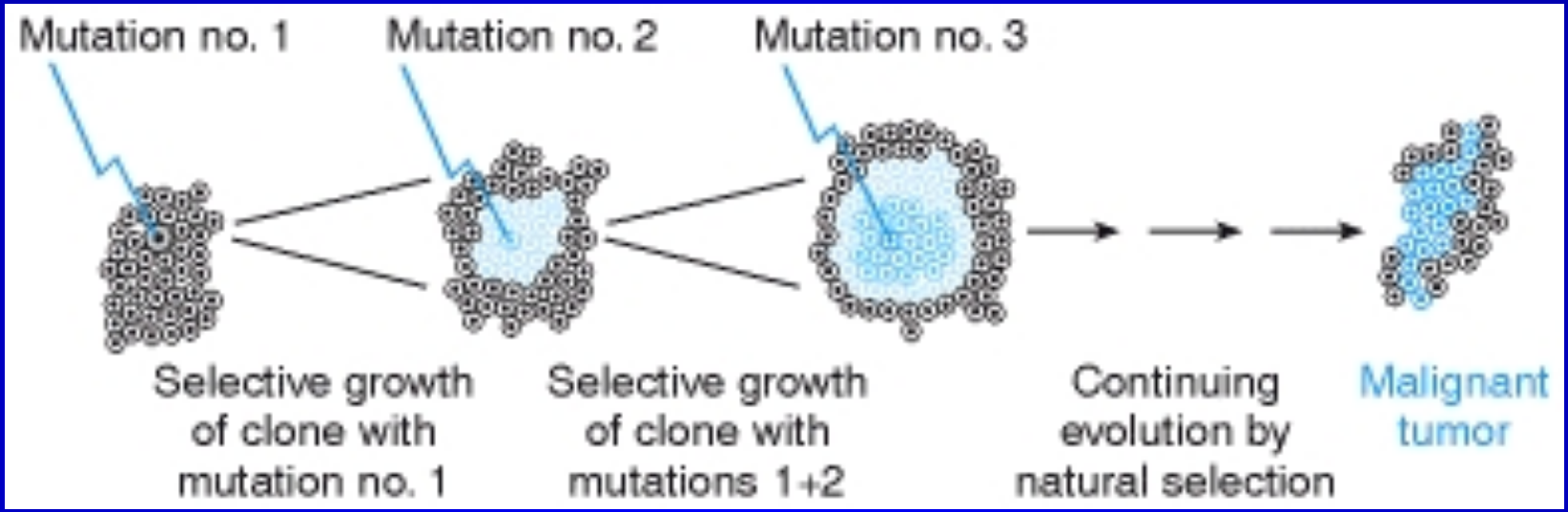
1. Self-sufficiency in growth signals.
2. Insensitivity to growth-inhibitory signals.
3. Resistant to apoptosis.
4. Limitless replicative potential (e.g. over-coming cellular senescence, no ageing).
5. Sustained angiogenesis.
6. Ability to invade and metastasize.

- When genes that normally sense and repair DNA damage are lost (enabler genes), the resultant genomic instability favors mutations in genes that regulate the six acquired capabilities of cancer cells.

- ***What is the molecular basis of cancer?***
- Cancer is a genetic disease.
    - Mutations in genes result in altered proteins
        - During cell division
        - External agents
        - Random event
    - Most cancers result from mutations in somatic cells
    - Some cancers are caused by mutations in germline cells

- **Genes involved in Cancer**

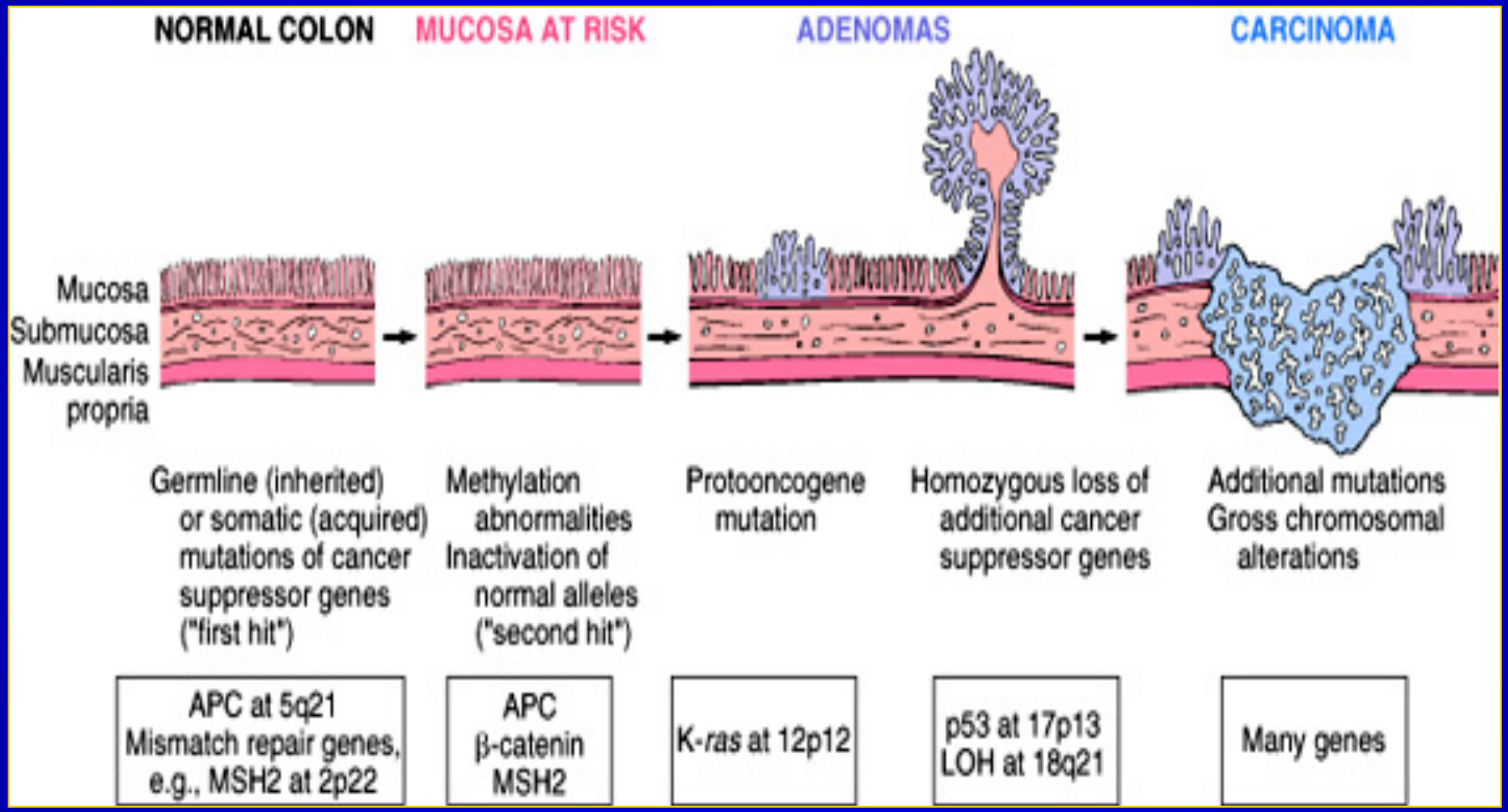
- Genes promote growth eg. RAS
- Genes inhibit growth eg. P53
- Genes control apoptosis eg. Bcl-2
- Genes of DNA repair
- And others….



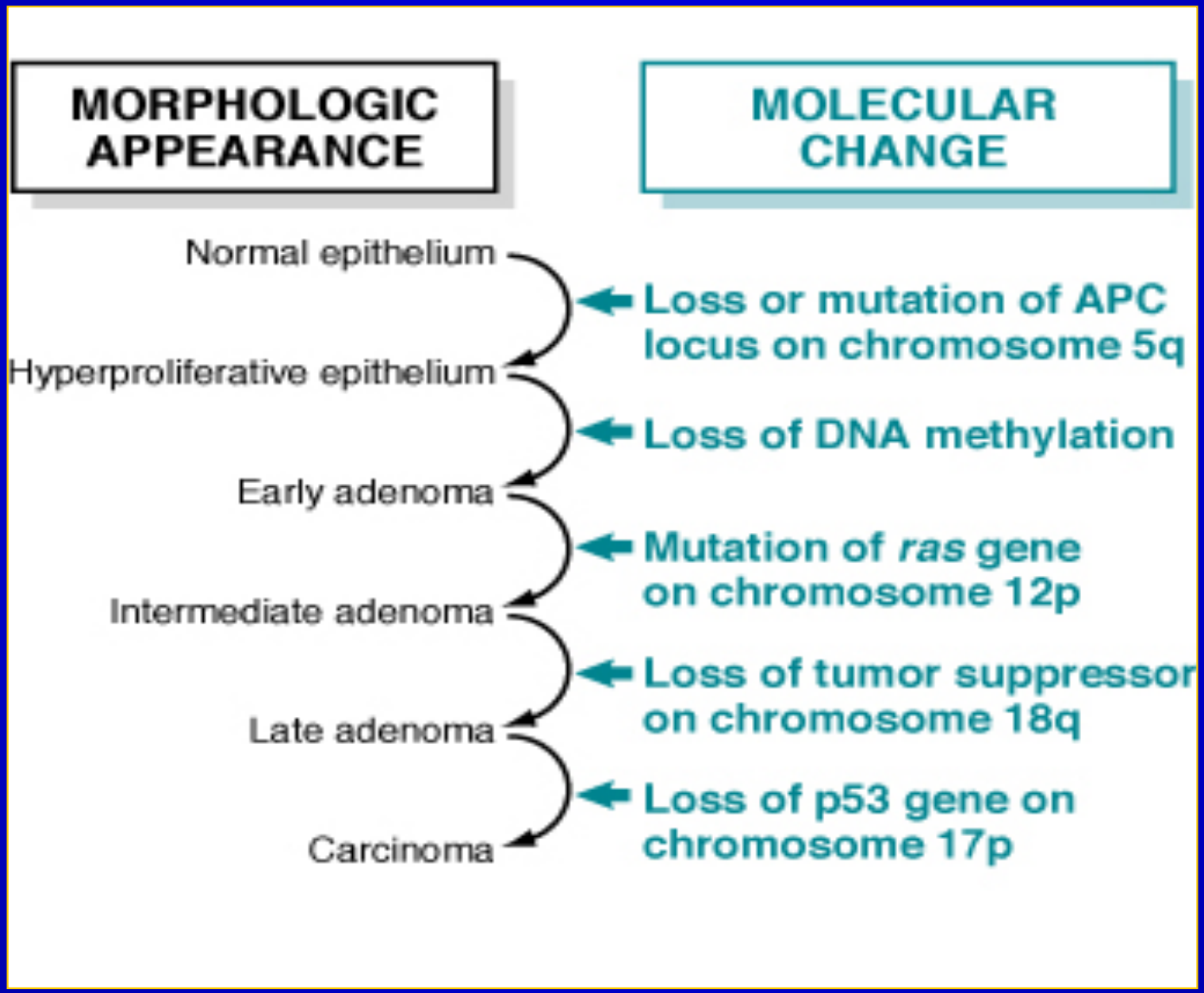
Multistage evolution of cancer

Each successive mutation gives the cell a growth advantage, so that it forms an expanded clone, thus presenting a larger target for the next mutation.

Dysplasia neoplasia sequence in colonic carcinoma



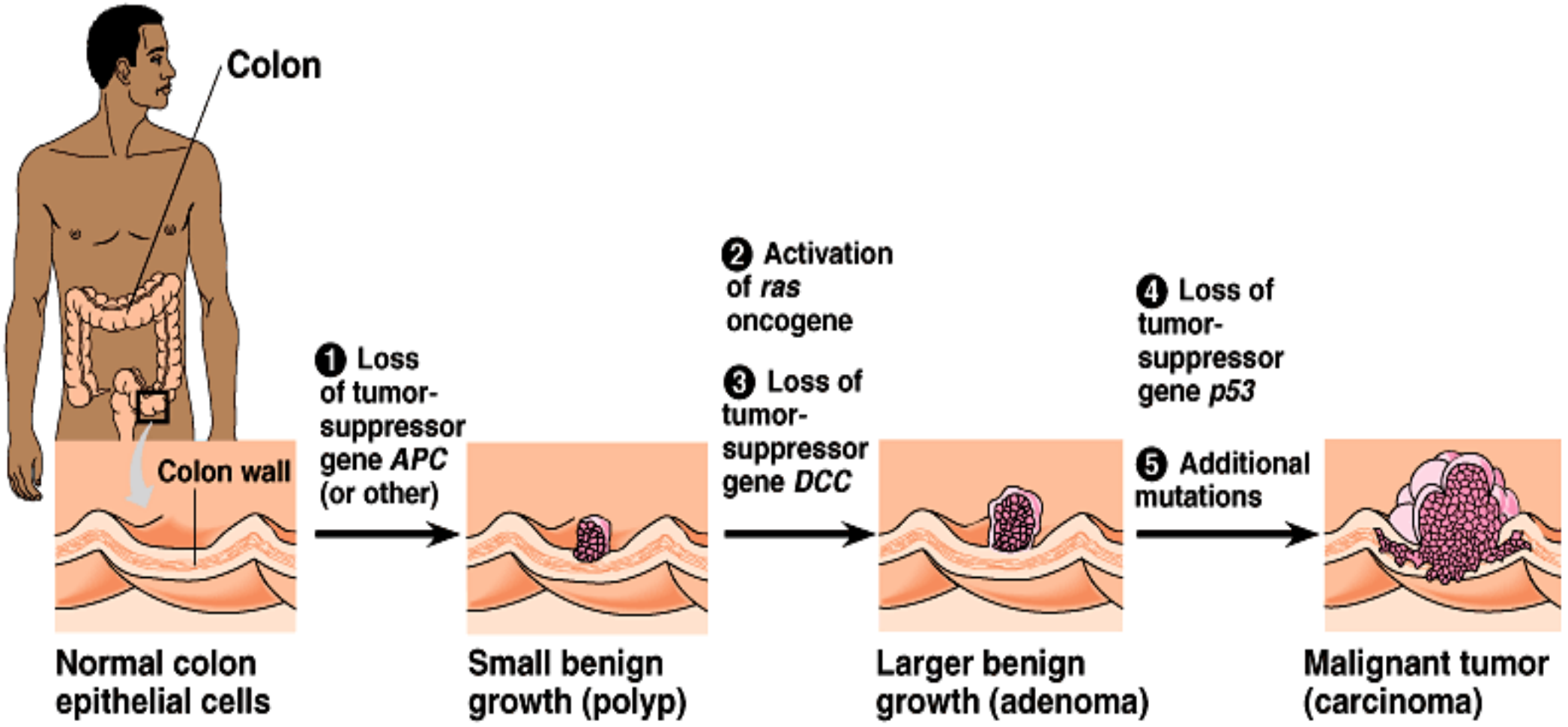
Dysplasia neoplasia sequence in colonic carcinoma



Tumor Progression

Multiple mutations lead to colon cancer

Genetic changes --> tumor changes



Genetic Instability in Tumors

- **Activation of Oncogenes**
- **Inactivation of Tumor Suppressor Genes**
- Mismatch repair (MMR) Genes
- Chromosomal Instability
- **Telomere Shortening**
- Microsatellite (variable tandem repeats, VTRs) Instability
- Apoptosis

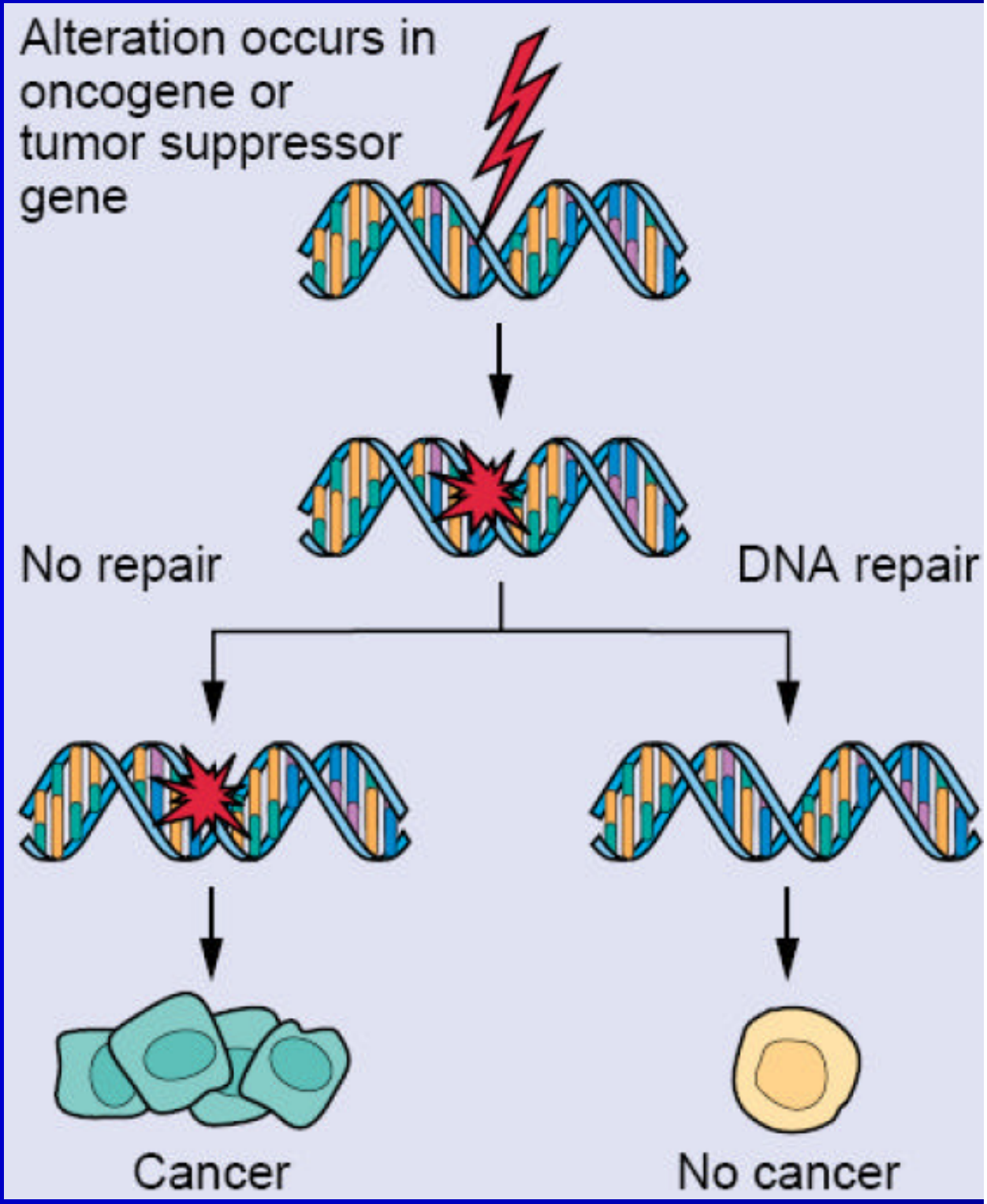
Possible Causes of Tumor Progression

GENES PLAYING ROLE IN CANCER DEVELOPMENT

- **Oncogenes**
- **Tumor suppressor genes**
- **DNA repair genes**

Genes and Tumor Development

- Inherited or acquired alterations in genes
  - Oncogenes – growth promoting genes
  - Tumor suppressor genes – brakes for cell division

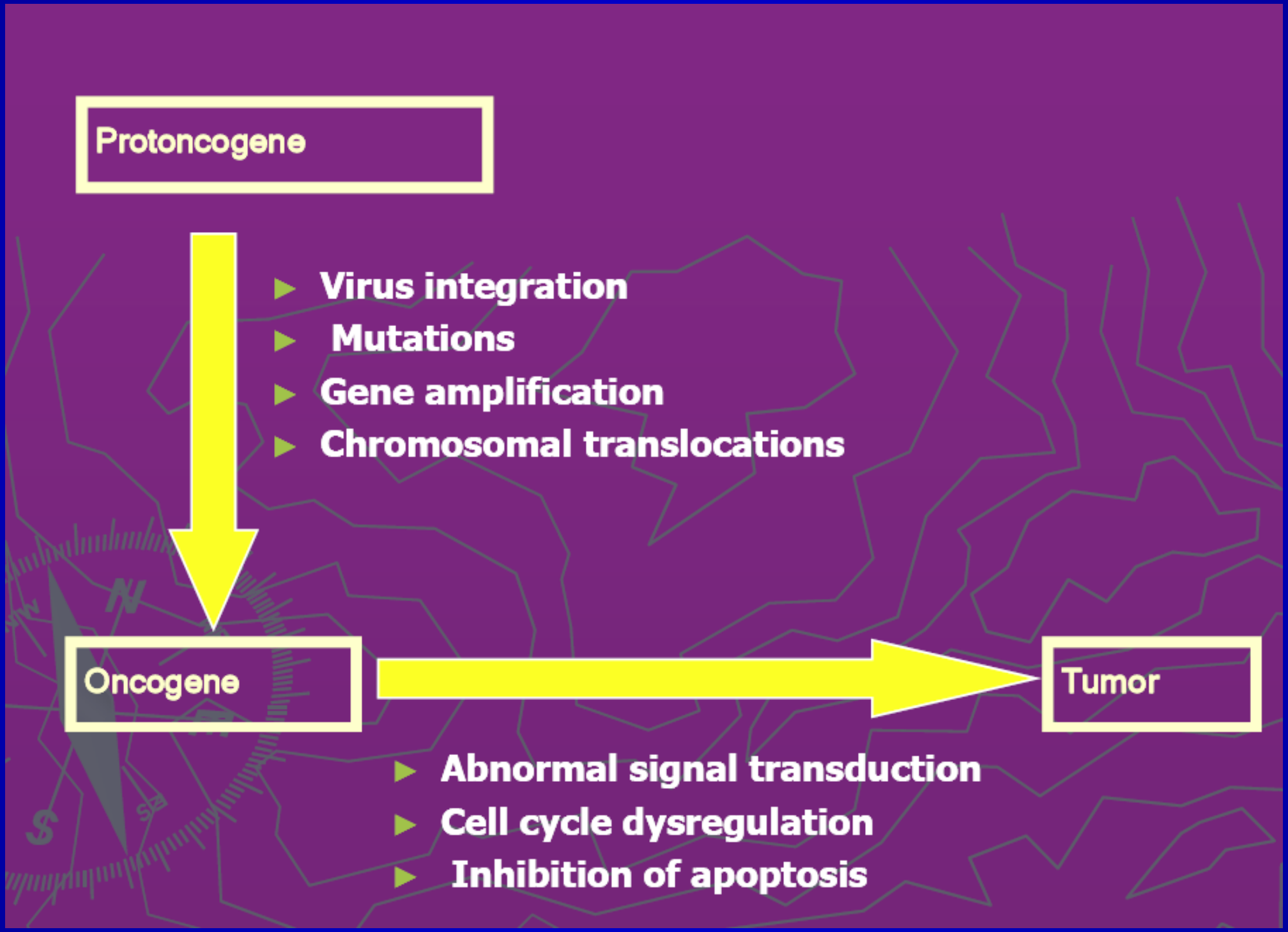


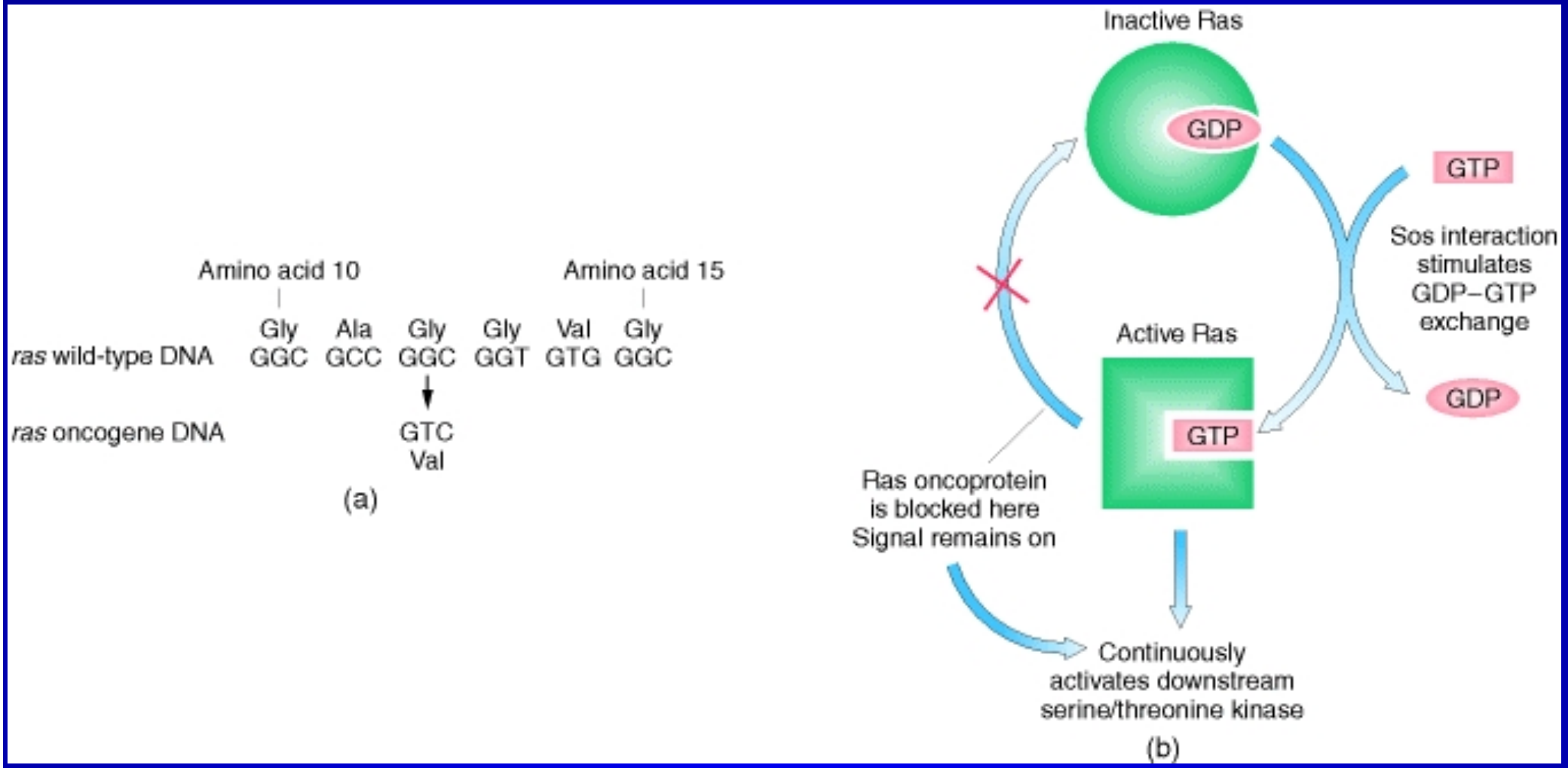
ONCOGENES

- Oncogenes are mutated forms of cellular proto-oncogenes. one or more forms of which is associated with cancer
- Proto-oncogene: A gene which may mutate to become an oncogene.
- Proto-oncogenes code for cellular proteins which regulate normal cell growth and differentiation.
- There are > 100 known oncogenes that may contribute to human neoplastic transformation.
- For example, the ras gene encodes the Ras protein, which regulates cell division. Mutations may result in the inappropriate activation of the Ras protein, leading to uncontrolled cell growth and division. The Ras protein is abnormal in about 25% of human cancers

Some well characterize oncogenes and proteins they encodes

	PROPERTIES OF PROTEIN	
Oncogene	**Location**	**Function**
Nuclear transcription regulators		
jun	Nucleus	Transcription factor
fos	Nucleus	Transcription factor
erbA	Nucleus	Member of steroid receptor family
Intracellular signal transducers		
abl	Cytoplasm	Protein tyrosine kinase
raf	Cytoplasm	Protein serine kinase
gsp	Cytoplasm	G-protein α subunit
ras	Cytoplasm	GTP/GDP-binding protein
Mitogen		
sis	Extracellular	Secreted growth factor
Mitogen receptors		
erbB	Transmembrane	Receptor tyrosine kinase
fms	Transmembrane	Receptor tyrosine kinase
Apoptosis inhibitor		
bcl2	Cytoplasm	Upstream inhibitor of caspase cascade


Protoncogene
Virus integration
Mutations
Gene amplification
Chromosomal translocations
Oncogene
Tumor
Abnormal signal transduction
Cell cycle dysregulation
Inhibition of apoptosis



The Ras oncoprotein. (a) The *ras* oncogene differs from the wild type by a single base pair, producing a Ras oncoprotein that differs from the wild type in one amino acid, at position 12 in the *ras* open reading frame. (b) The effect of this missense mutation is to create a Ras oncoprotein that cannot hydrolyze GTP to GDP. Because of this defect, the Ras oncoprotein remains in the active Ras–GTP complex and continuously activates the down-stream serine/threonine kinase.

Five types of proteins encoded by proto-oncogenes participate in control of cell growth:

Class I: Growth Factors

Class II: Receptors for Growth Factors and Hormones

Class III: Intracellular Signal Transducers

Class IV: Nuclear Transcription Factors

Class V: Cell-Cycle Control Proteins

GENE AMPLIFICATION

Oncogene	Amplification	Source of tumor
c-myc	~20-fold	leukemia and lung carcinoma
N-myc	5-1,000-fold	neuroblastoma retinoblastoma
L-myc	10-20-fold	small-cell lung cancer
c-abl	~5-fold	chronic myoloid leukemia
c-myb	5-10-fold	acute myeloid leukemia colon carcinoma
c-erbB	~30-fold	epidermoid carcinoma
K-ras	4-20-fold 30-60-fold	colon carcinoma adrenocortical carcinoma

Oncogenes are usually dominant (gain of function)

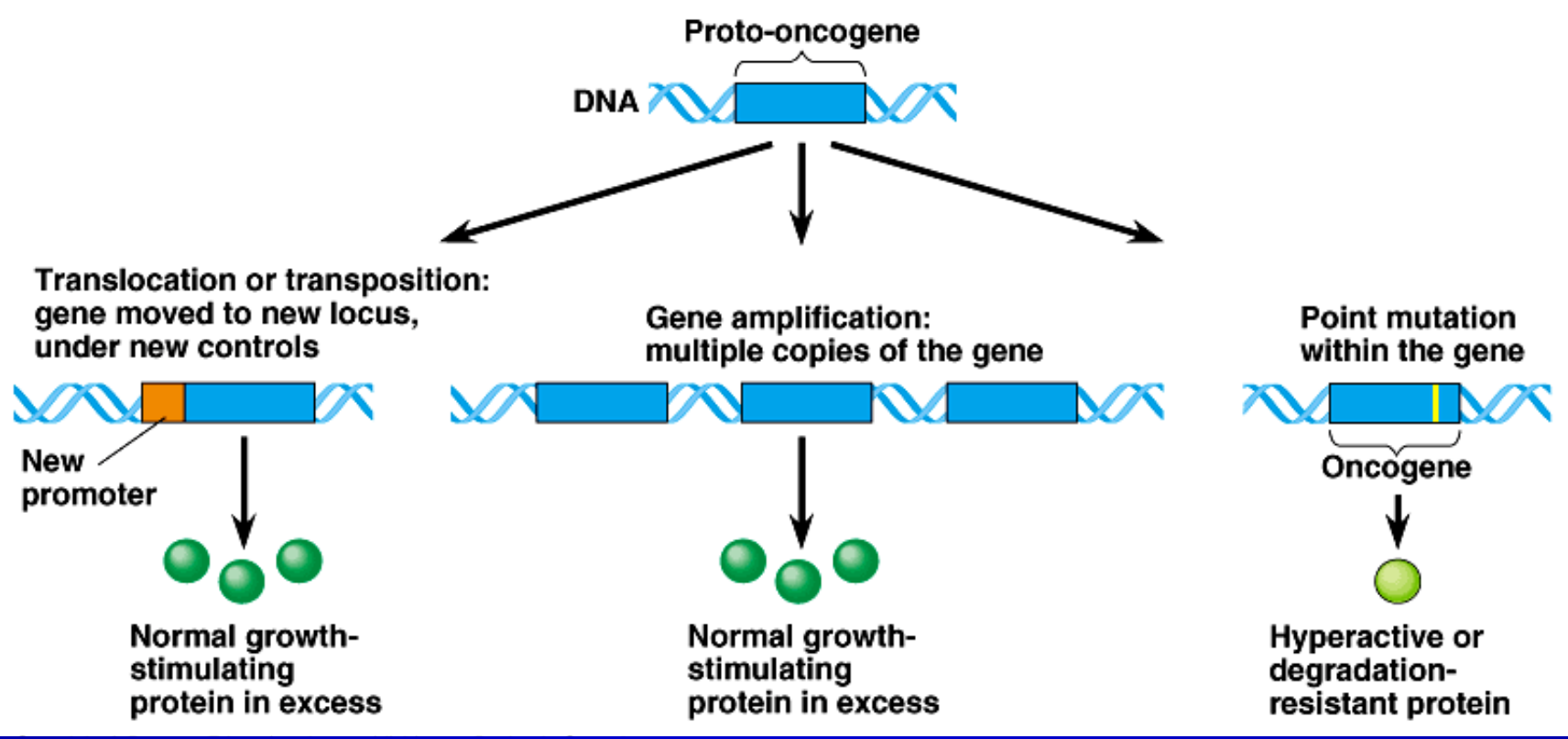
- cellular proto-oncogenes that have been mutated (and "activated")
- cellular proto-oncogenes that have been captured by retroviruses and have been mutated in the process (and "activated")
- virus-specific genes that behave like cellular proto-oncogenes that have been mutated to oncogenes (i.e., "activated")

***The result*:**

- Overproduction of growth factors
- Flooding of the cell with replication signals
- Uncontrolled stimulation in the intermediary pathways
- Cell growth by elevated levels of transcription factors

Activation mechanisms of proto-oncogenes

proto-oncogene --> oncogene



Tumor suppressor genes

- Normal function - inhibit cell proliferation
- Absence/inactivation of inhibitor --> cancer
- Both gene copies must be defective

TUMOR SUPPRESSOR GENES

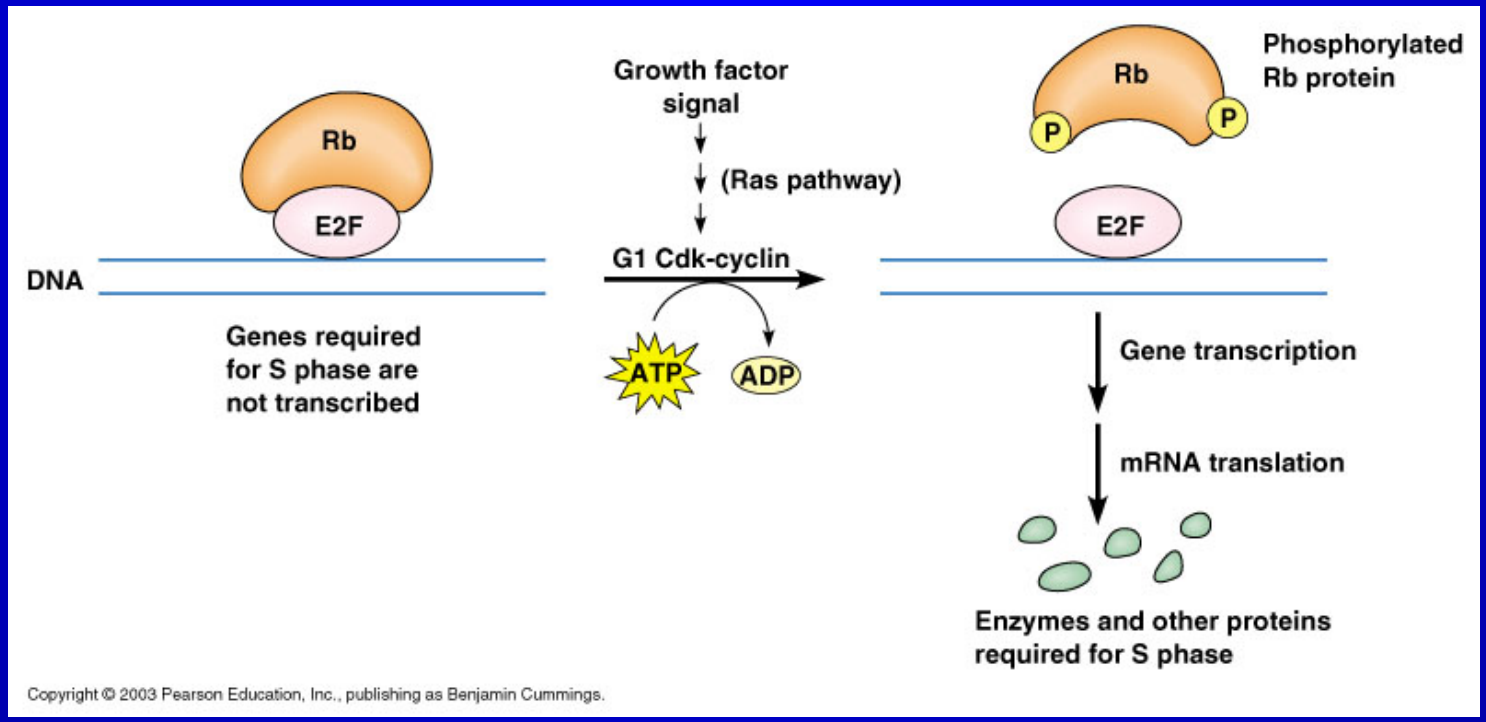
		Disorders in which gene is affected	
Gene (locus)	***Function***	***Familial***	***Sporadic***
DCC (18q)	cell surface interactions	unknown	colorectal cancer
WT1 (11p)	transcription	Wilm's tumor	lung cancer
Rb1 (13q)	transcription	retinoblastoma	small-cell lung carcinoma
p53 (17p)	transcription	Li-Fraumeni syndrome	breast, colon, & lung cancer
BRCA1(17q)	transcriptional	breast cancer	breast/ovarian tumors
BRCA2 (13q)	regulator/DNA repair		

- The active p53 protein is a transcriptional regulator that is activated in response to DNA damage.
- Activated wild-type p53 serves double duty, preventing progression of the cell cycle until the DNA damage is repaired and, under some circumstances, inducing apoptosis.
- In the absence of a functional p53 gene, the p53 apoptosis pathway does not become activated, and the cell cycle progresses even in the absence of DNA repair. This progression elevates the overall frequency of mutations
- In retinoblastoma, the gene encoding the Rb protein, considered in the regulation of the cell cycle, is mutated

Rb gene

- Rb protein controls cell cycle moving past G1 checkpoint
- Rb protein binds regulatory transcription factor E2F
- E2F required for synthesis of replication enzymes
- E2F - Rb bound = no transcription/replication
- Growth factor --> Ras pathway
  --> G1Cdk-cyclin synthesized
- Active G1 Cdk-cyclin kinase phosphorylates Rb
- Phosphorylated Rb cannot bind E2F --> S phase
  - Disruption/deletion of *Rb* gene
  - Inactivation of Rb protein

  --> uncontrolled cell proliferation --> cancer



p53

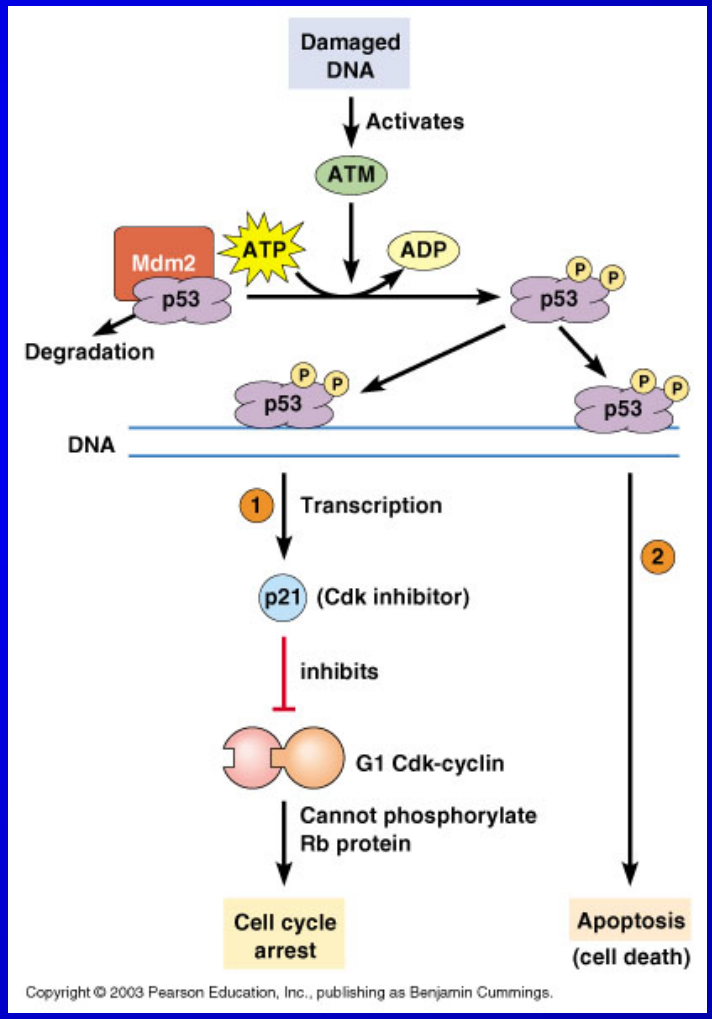
- Phosphyorylated p53 activates transcription of *p21* gene
- p21 Cdk inhibitor (binds Cdk-cyclin complex --> inhibits kinase activity)
- Cell cycle arrested to allow DNA to be repaired
- If damage cannot be repaired

--> cell death (apoptosis)

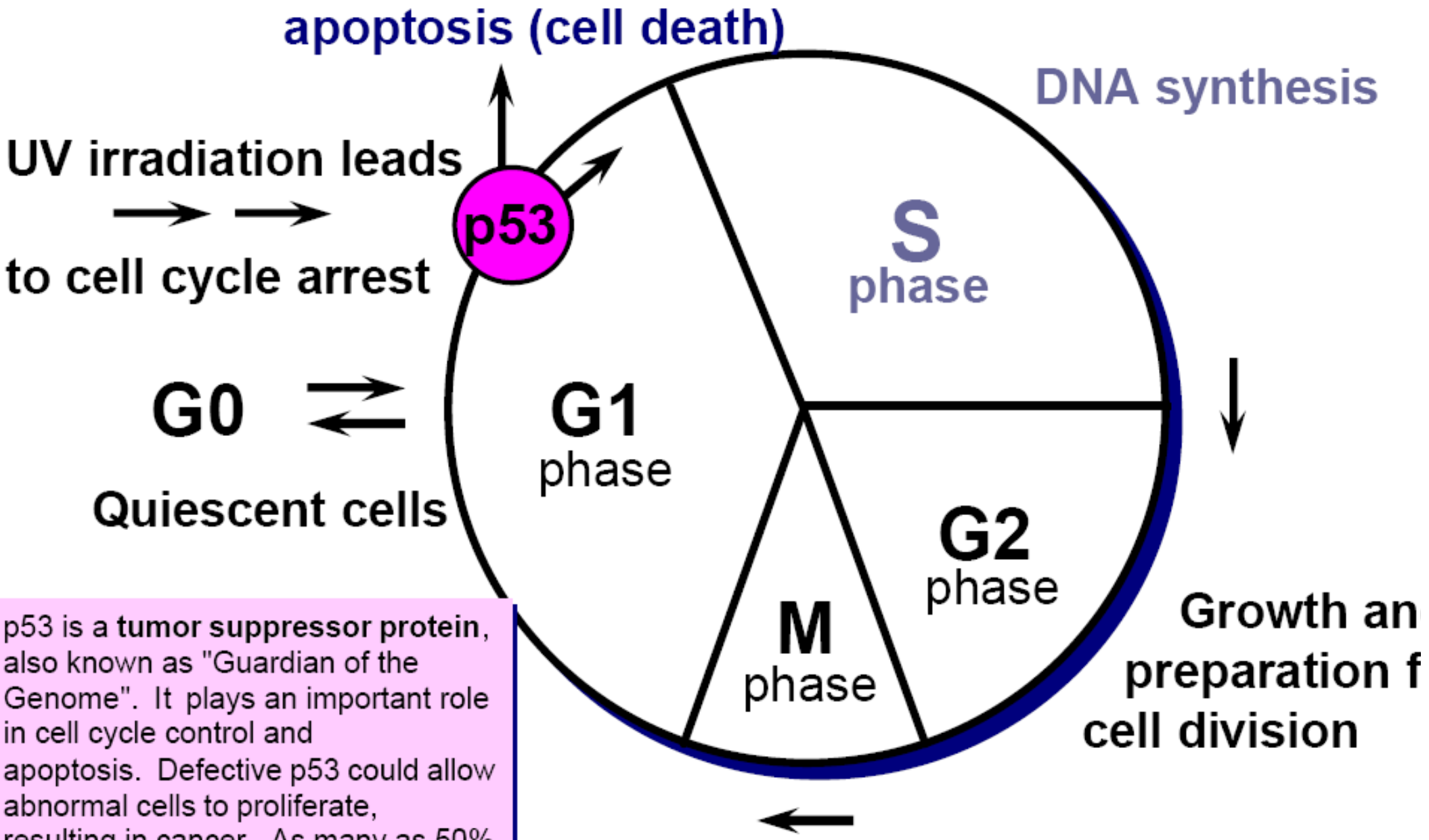
- Disruption/deletion of *p53* gene
- Inactivation of p53 protein

--> uncorrected DNA damage

--> uncontrolled cell proliferation --> cancer



The role of p53 in the cell cycle ("guardián del genom



p53 is a **tumor suppressor protein**, also known as "Guardian of the Genome". It plays an important role in cell cycle control and apoptosis. Defective p53 could allow abnormal cells to proliferate, resulting in cancer. As many as 50%

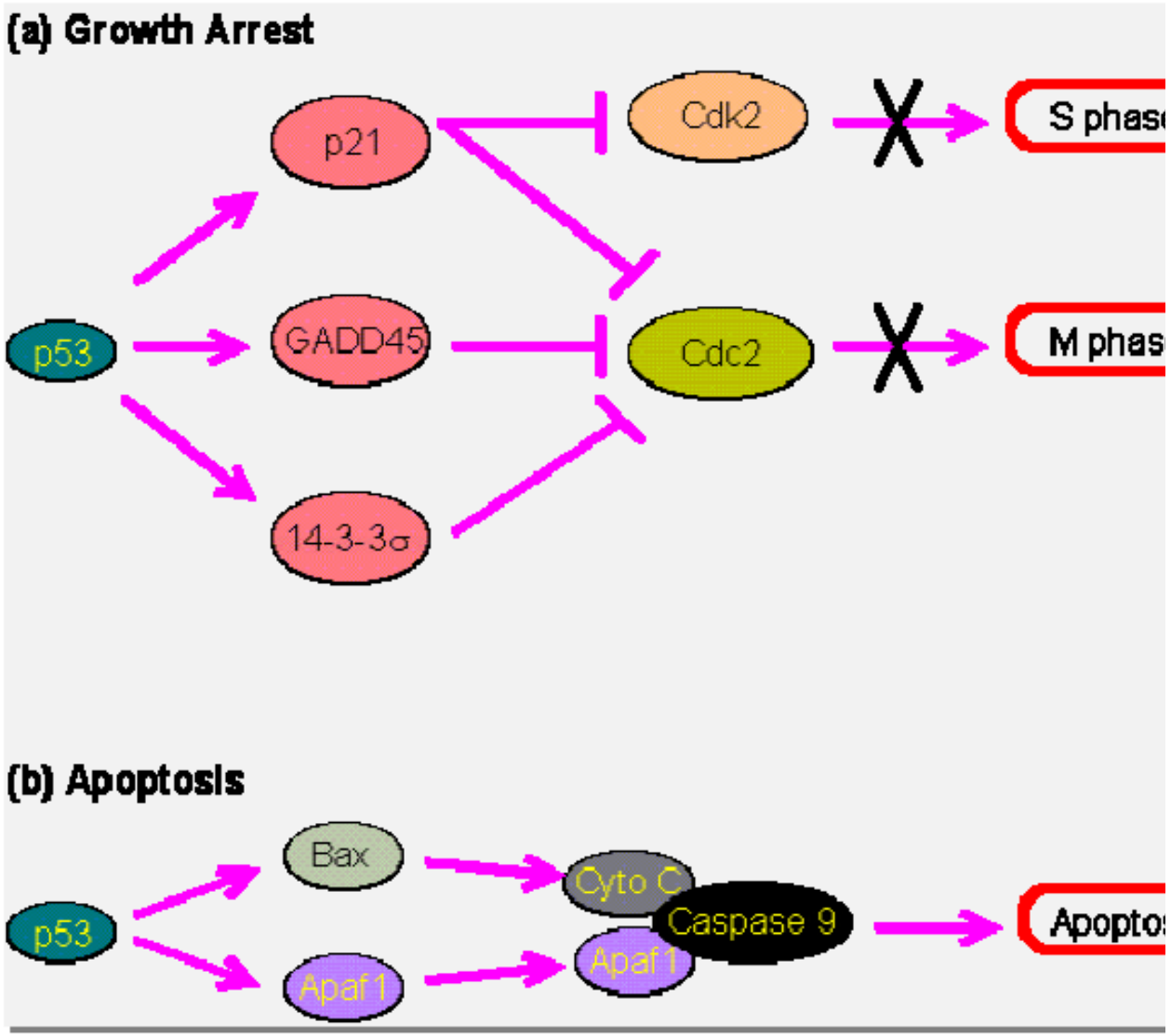
Roles of p53 in growth arrest and apoptosis

a) Growth Arrest

The cell cycle progression into the S phase requires the enzyme Cdk2, which can be inhibited by p21. The progression into the M phase requires Cdc2 which can be inhibited by p21, GADD45 or 14-3-3s. **p53 regulates the expression of these inhibitory proteins to induce growth arrest.**

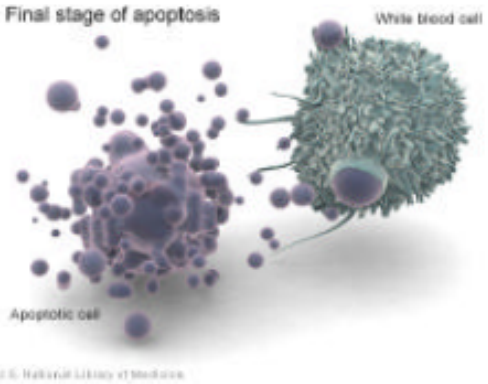
b) Apoptosis

Apoptosis can be induced by the binding of Caspase 9 to cytochrome c and Apaf1. **p53 may activate the expression of Apaf1 and Bax.** The latter can then stimulate the

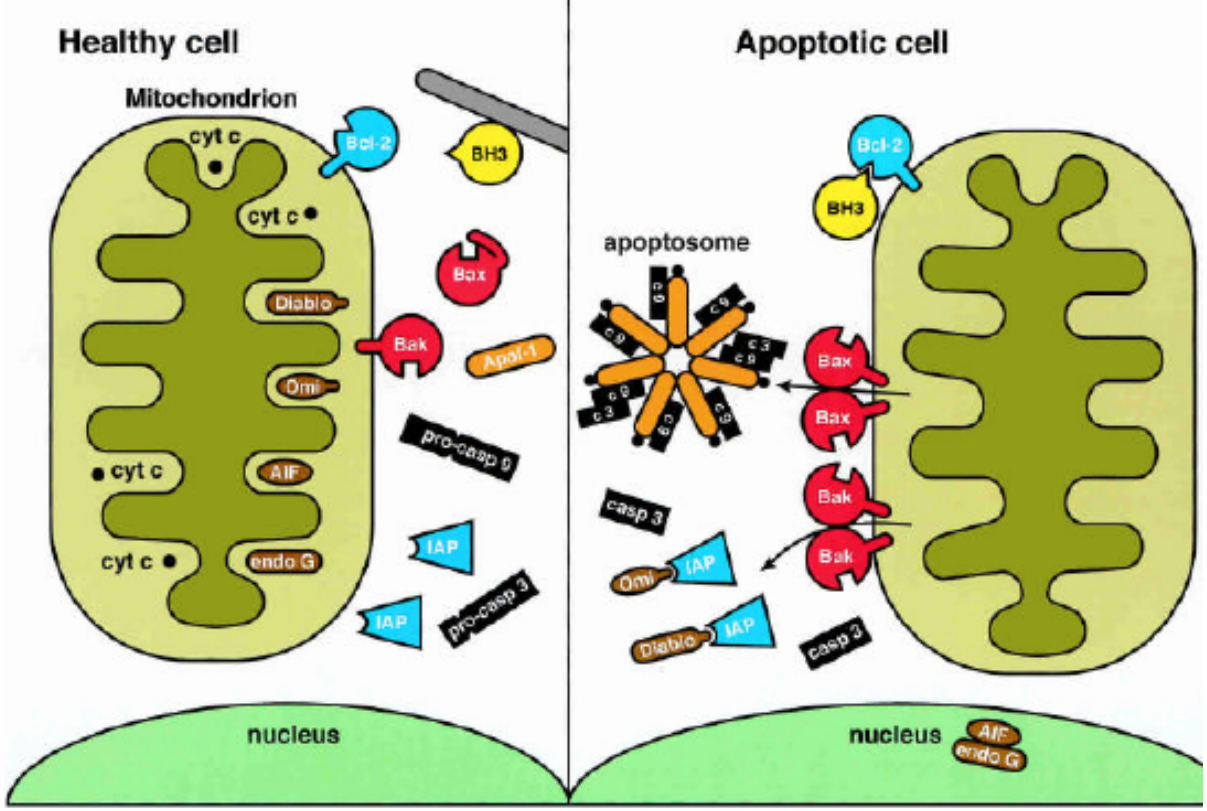


Apoptosis

- Apoptosis, or programmed cell death, is a highly regulated process that allows a cell to self-degrade in order for the body to eliminate unwanted or dysfunctional cells.



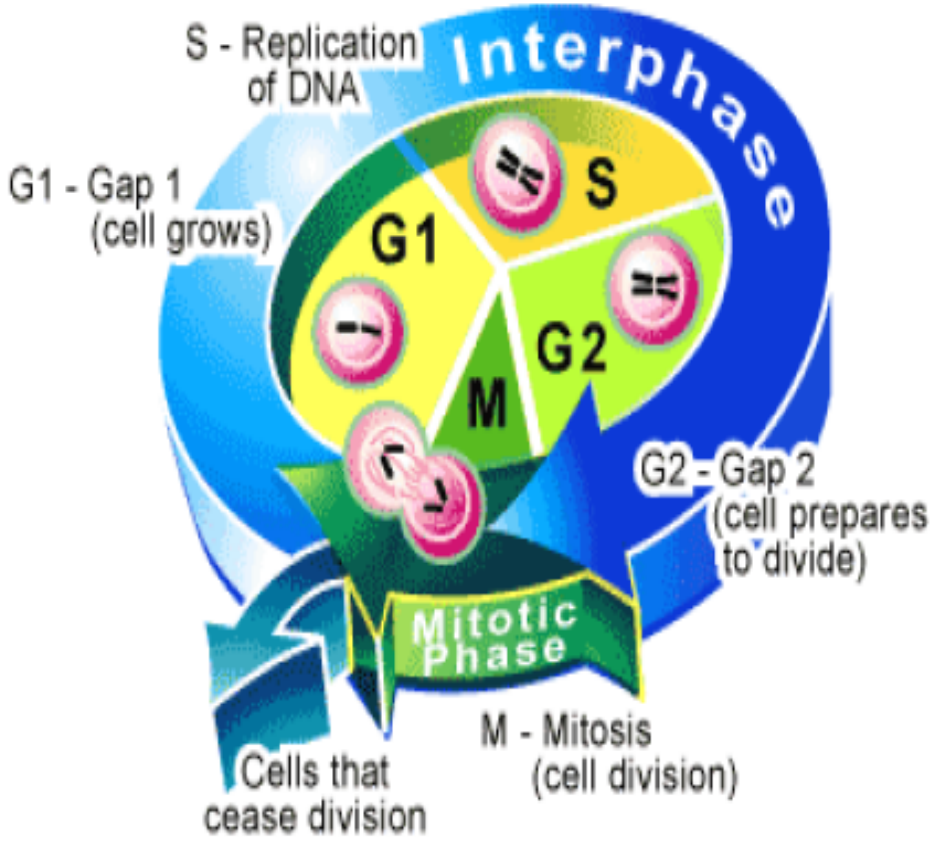
- Cancer may arise from the dysfunction in the apoptotic pathway.



- In a healthy cell, the caspases are all present as zymogens the BH3-only proteins (BH3) are sequestered away from the 2-like pro-survival proteins (Bcl-2).
- After an apoptotic signal, the freed BH3-only proteins asso with Bcl-2 on mitochondria, and the recruitment of Bax **(acti by p53)** and Bak into oligomers on the mitochondrial outer membrane then leads to its permeabilization.
- The released cytochrome c induces formation of the heptar "apoptosome" of Apaf-1 **(activated by p53)** and procaspa (c9), which activates caspase-3 (c3).
- The released Diablo/Smac and Omi/Htr2 incapacitate the I in the cytosol, whereas AIF and endonuclease G (endoG) er the nucleus, where they may aid in DNA degradation

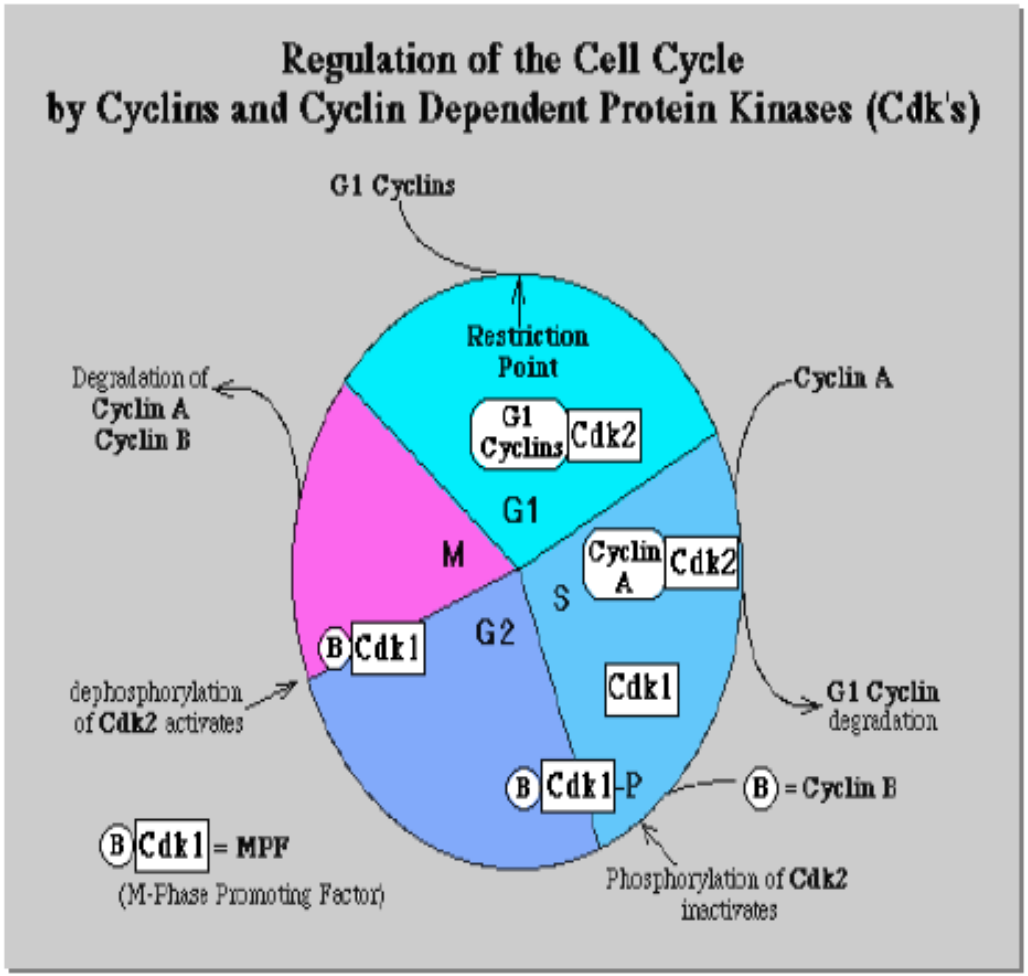
The Cell Cycle

- The cell cycle consists of:
  - **G1 =** growth and preparation of the chromosomes for replication;
  - **S =** synthesis of DNA (replication) and duplication of the centrosome;
  - **G2 =** preparation for mitotic division
  - **M =** mitosis.



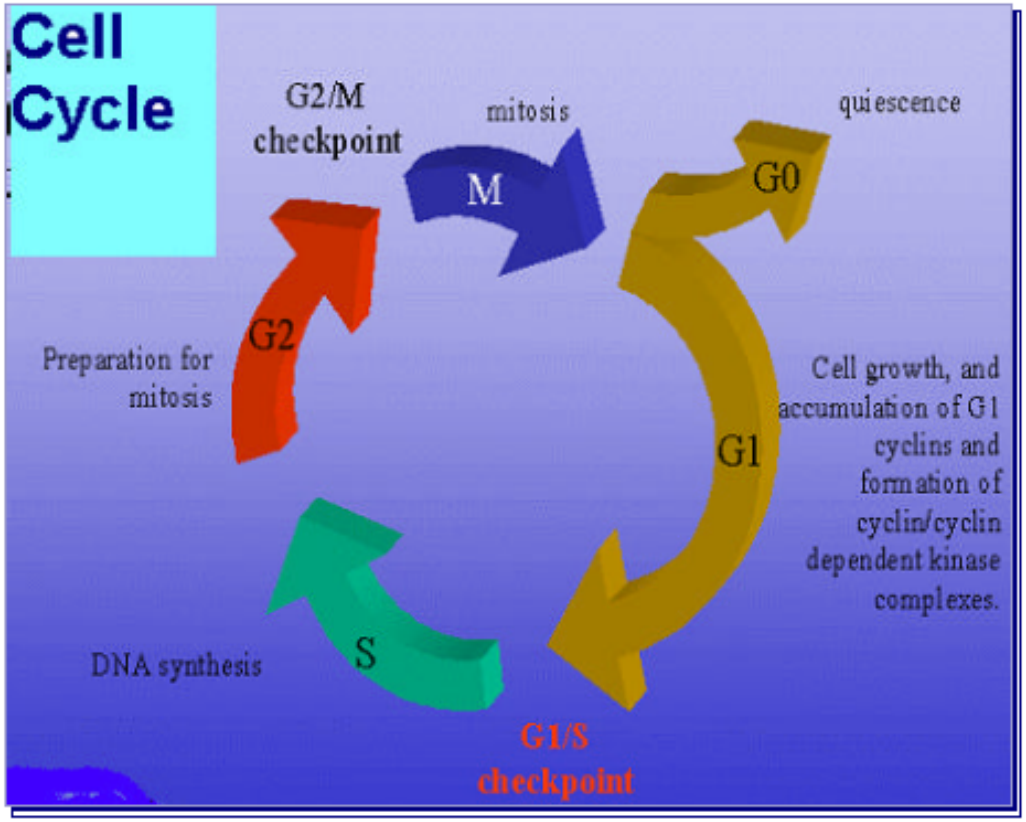
Cell Cycle Control

- The passage of a cell through the cell cycle is controlled by proteins in the cytoplasm. Among the main players in animal cells are:
  - **Cyclins** - Their levels in the cell rise and fall with the stages of the cell cycle.
  - **Cyclin-dependent kinases (Cdks)** - Their levels in the cell remain fairly stable, but each must bind the appropriate cyclin (whose levels fluctuate) in order to be activated. They add phosphate groups to a variety of protein substrates that control processes in the cell cycle.
  - The **anaphase-promoting complex (APC)**. (The APC is also called the **cyclosome.** - Triggers the events leading to the separation of sister chromatids and degrades the mitotic cyclin B.



Checkpoints: Quality Control of the Cell Cycle

- All the **checkpoints** require the services of a complex of proteins.
- Mutations in the genes encoding some of these have been associated with cancer; that is, they are **oncogenes**.
- **DNA damage** checkpoints. These sense DNA damage before the cell enters S phase (a G1 checkpoint) as well as during S phase.
- Damage to DNA inhibits the action of CDK2 thus stopping the progression of the cell cycle until the damage can be repaired (with the aid of **BRCA2**). If the damage is so severe that it cannot be repaired, the cell self-destructs by **apoptosis**.



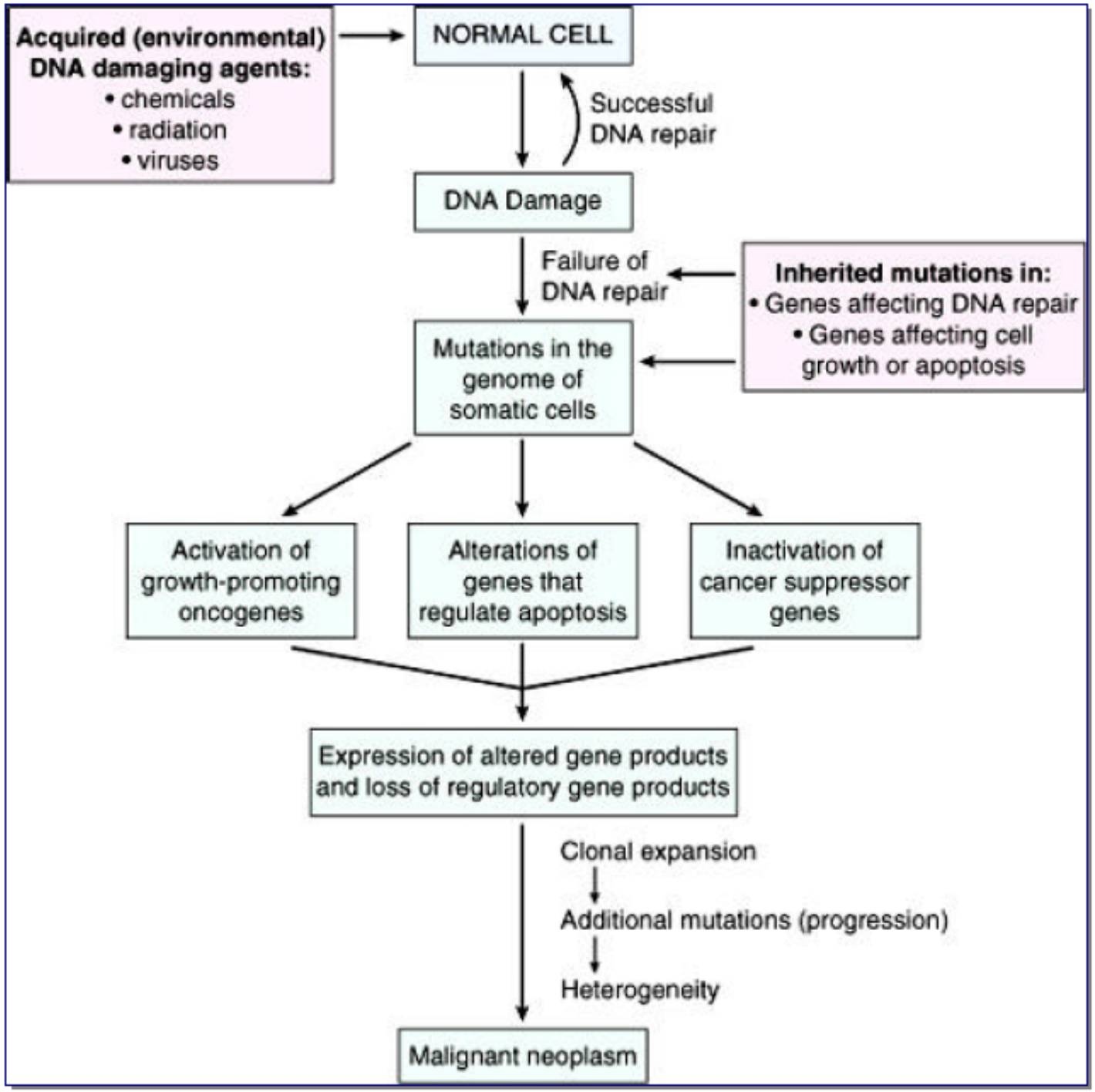
DNA REPAIR GENES

These are genes that ensure each strand of genetic information is accurately copied during cell division of the cell cycle.

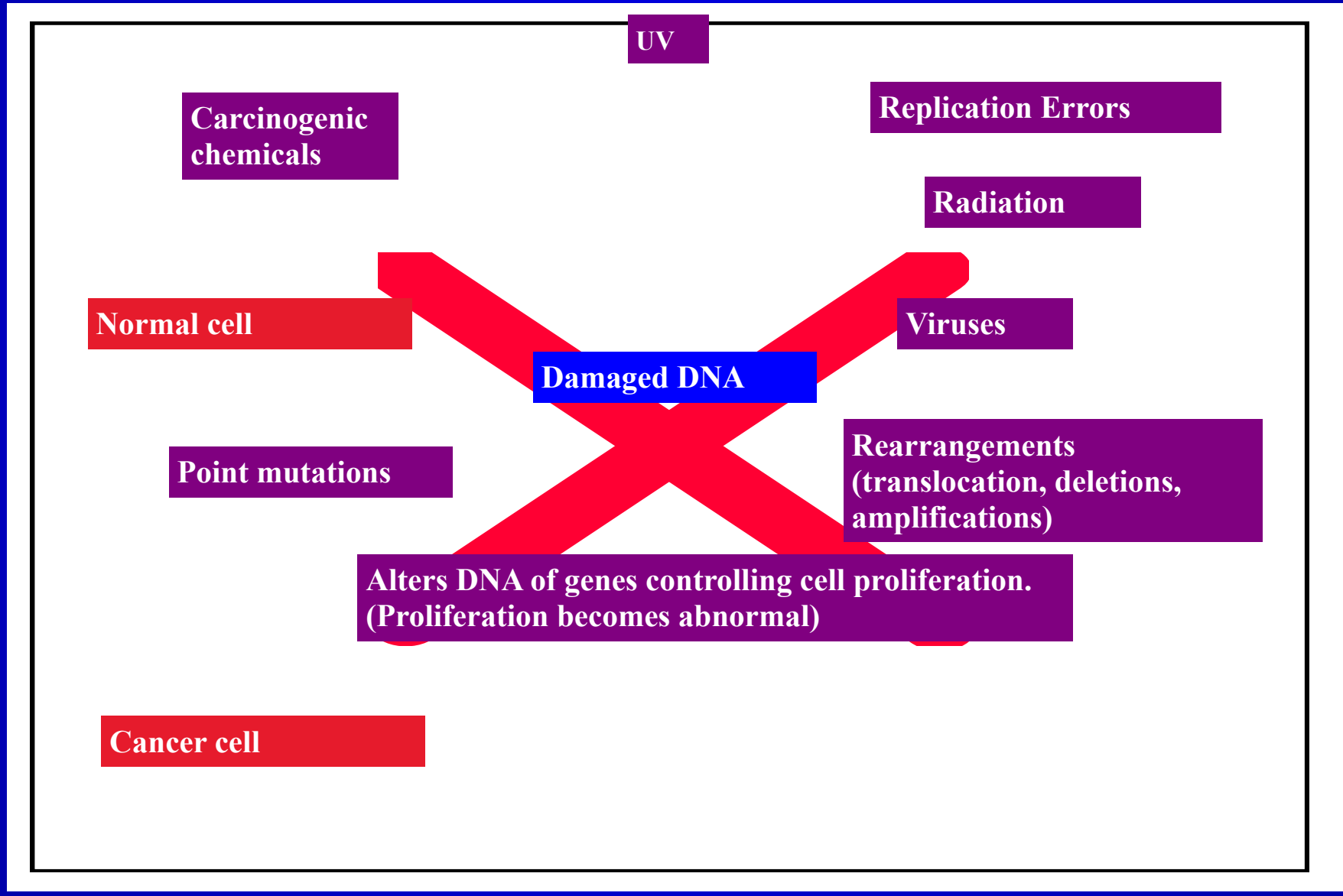
Mutations in DNA repair genes lead to an increase in the frequency of mutations in other genes, such as proto-oncogenes and tumor suppressor genes.

i.e. Breast cancer susceptibility genes (BRCA1 and BRCA2)
Hereditary non-polyposis colon cancer susceptibility genes (MSH2, MLH1, PMS1, PMS2) have DNA repair functions. Their mutation will cause tumorigenesis.

Cancer: General Etiology and Pathogenesis



THE CAUSES OF GENOMIC CHANGES IN CANCER



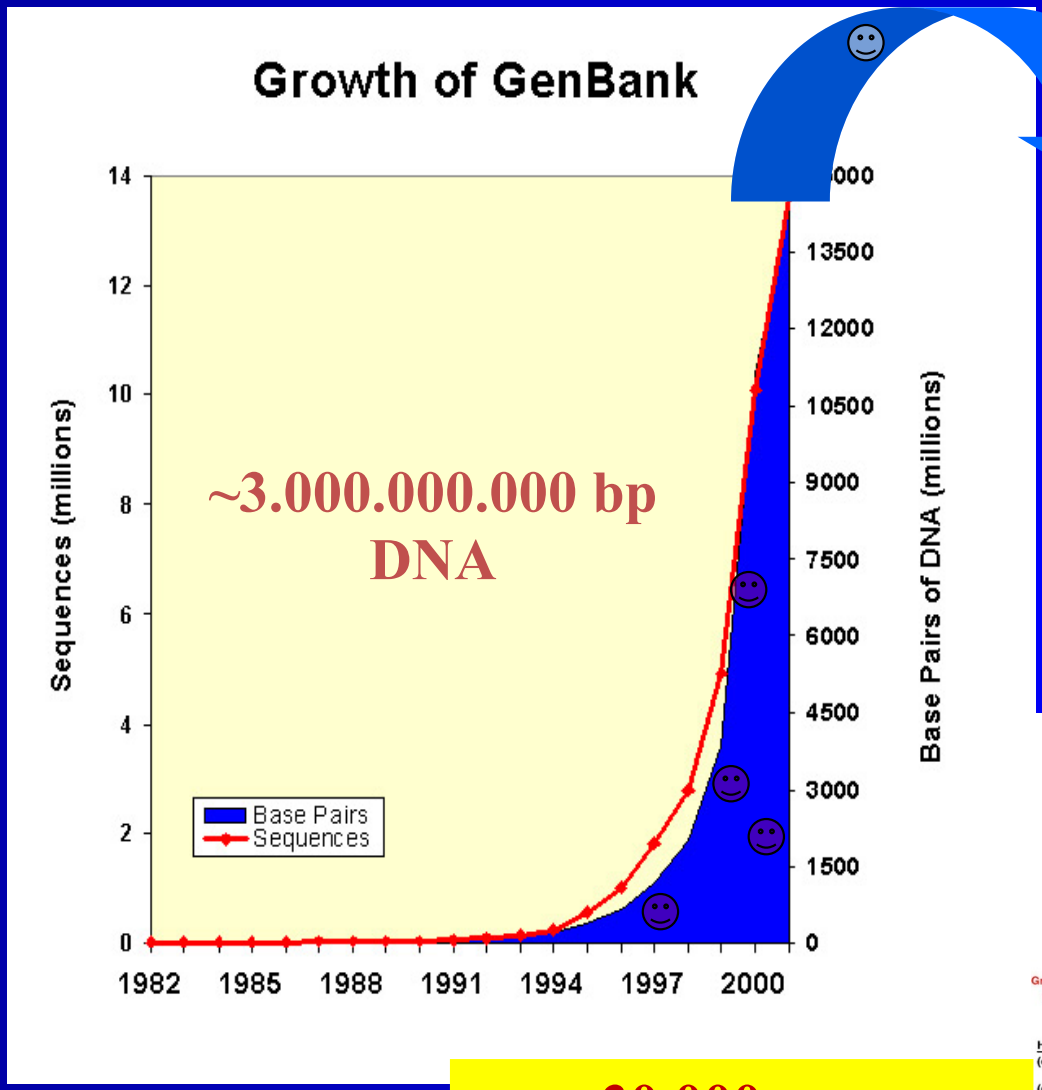
Causes of Cancer: Risk Factors

- Radiation Exposure
- UV light
- Tobacco Smoke
- Food Contamination
- Tumor Viruses
- Family History (Genetic Background)
- Lifestyle

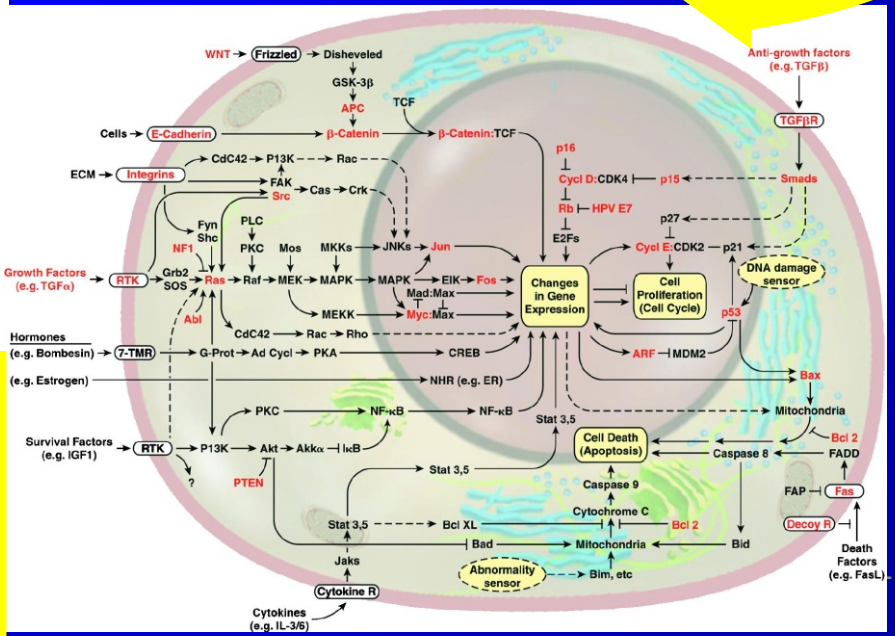
Substances in the environment known to cause cancer

- Tobacco
- Diet/weight/physical inactivity
- Alcoholic drinks
- Ultraviolet radiation
- Virus and bacteria
- Ionizing radiation
  - Special case: Radon
- Pesticides
- Medical drugs
- Solvents
- Dioxins
- Fibers, fine particles, and dust
- Polycyclic aromatic hydrocarbons
- Metals
- Diesel exhaust particles
- Toxins from fungi
- Benzidine
- Vinyl chloride

MOLECULAR BIOLOGY & INFORMATICS



Bioinformatics



~30.000 genes
~300.000 protein
~3.000.000 interaction
1 human cell