Medical Genetics

Sheet: Cancer Genetics-16

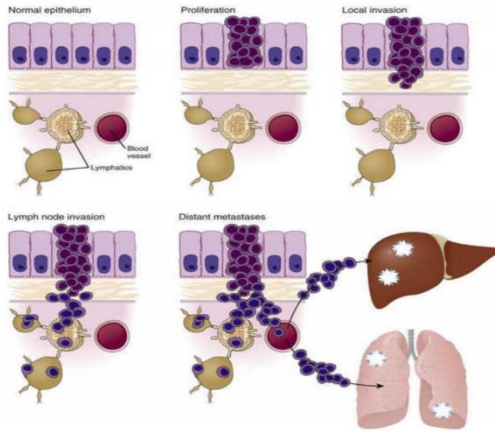
Writer: Sarah Basel

Scientific: Ameen Alsaras

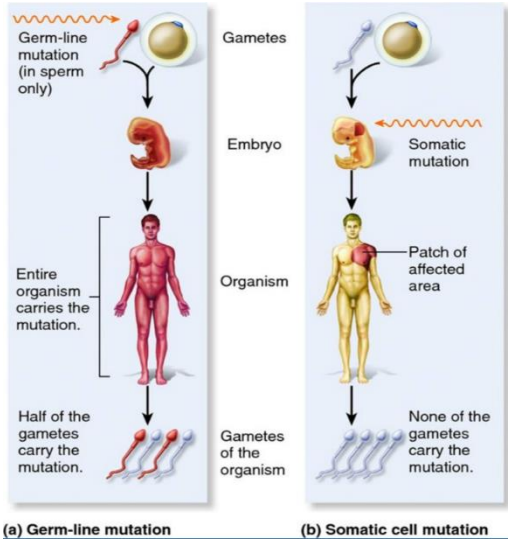
Final: Mothana Mahes

Doctor: Bilal Azab

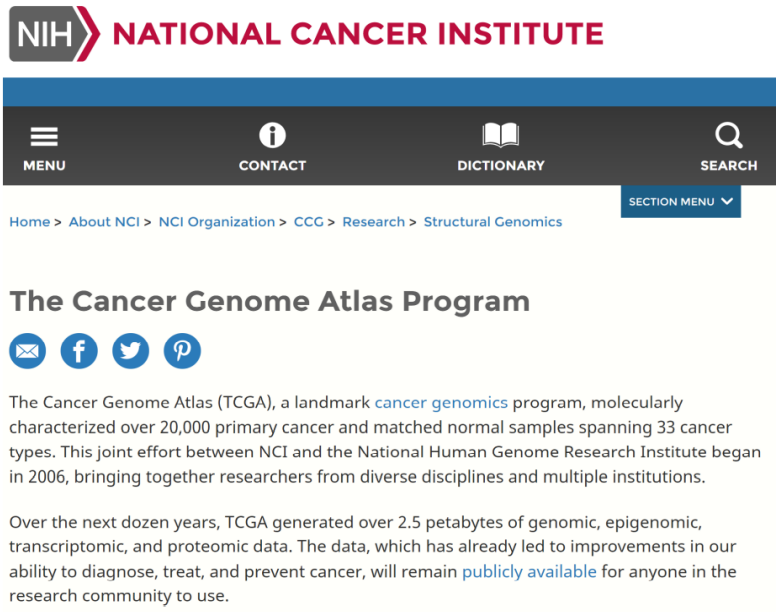
- **Neoplasia:** is a disease process characterized by uncontrolled cellular proliferation leading to a mass or tumor (neoplasm).
- **Cancer:** is the name used to describe the more virulent forms of neoplasia. Accumulation of cells in a neoplasm occurs because of an ***imbalance*** between the normal processes of cellular proliferation and cellular attrition. For a neoplasm to be a cancer, however, it must also be ***malignant***, which means that not only is its growth uncontrolled, it is also capable of **invading** neighboring tissues that surround the original site (the primary site) and can **spread (metastasize)** to more distant sites. Tumors that do not invade or metastasize are not cancerous but are referred to as benign tumors, although their abnormal function, size or location may make them anything but benign to the patient, for example most of brain tumors are benign yet they are considered dangerous because there's only small room inside the skull and the brain occupies most of it. Even if a brain tumor is benign and growing slowly, eventually the brain won't be able to tolerate that, and symptoms will develop which can be life-threatening.
- General scheme for development of a carcinoma in an epithelial tissue such as colonic epithelium. The diagram shows progression from normal epithelium to local proliferation, invasion across the lamina propria, spread to local lymph nodes, and final distant metastases to liver and lung.



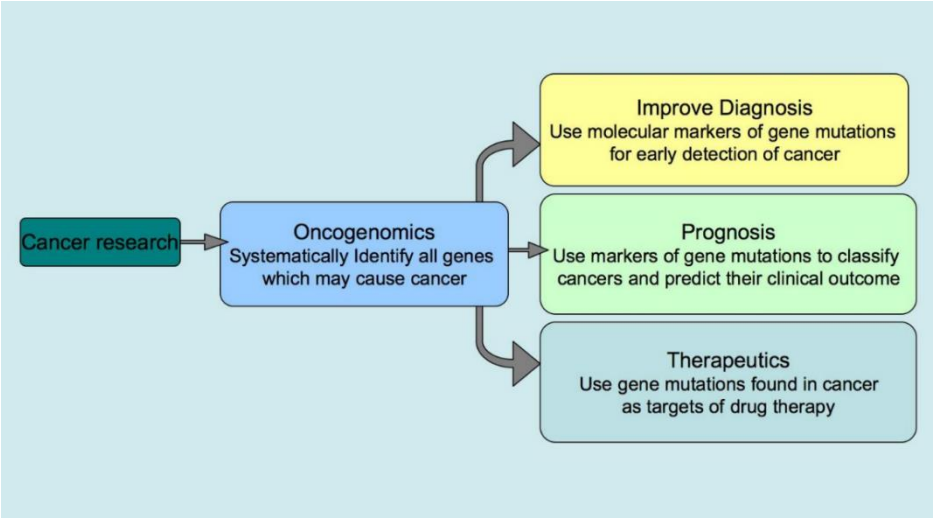
- Germline mutations are found in gametes only, whereby the entire organism carries the mutation. Somatic mutations only occur in a specific area of the body (i.e. in the tumor), are not carried in the gametes, and are not hereditary.



- There are three main classes of cancer:
  - Carcinomas (most common), which originate in epithelial tissue, such as the cells lining the intestine, bronchi, or mammary ducts.
  - Sarcomas, in which the tumor has arisen in mesenchymal tissue, such as bone, muscle, or connective tissue, or in nervous system tissue.
  - Hematopoietic and lymphoid malignant neoplasms, such as leukemia and lymphoma, which form from (and spread throughout) the bone marrow, lymphatic system, and peripheral blood.
- Cancer genomics:
  - Cancer genomics is the study of the total DNA sequence and gene expression differences between tumor cells and normal host cells. It aims to understand the genetic basis of tumor cell proliferation and the evolution of the cancer genome under mutation and selection by the body environment, the immune system and therapeutic interventions.



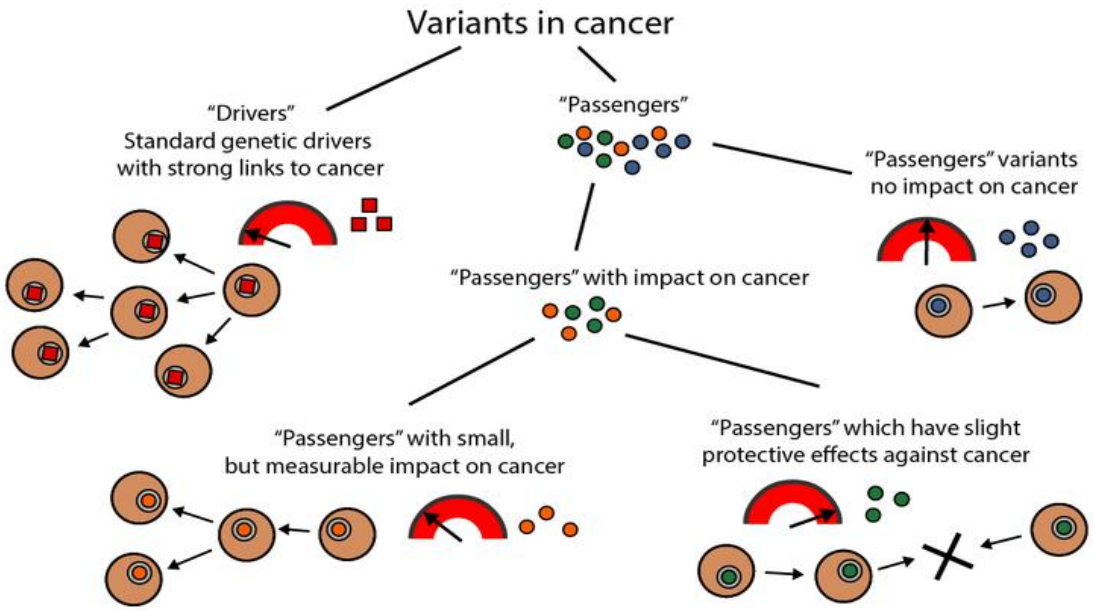
  - Genomics—in particular the identification of **mutations**, altered **epigenomic modifications**, and abnormal **gene expression** in cancer cells—is vastly expanding our knowledge of why cancer develops and is truly changing cancer diagnosis and treatment.
  - Within each of the major groups, tumors are classified by site, tissue type, histological appearance, degree of malignancy, chromosomal aneuploidy, and, increasingly, by which **gene mutations** and abnormalities in gene expression are found within the tumor.



- The goal of oncogenomics is to identify new oncogenes or tumor suppressor genes that may provide new insights into cancer diagnosis, predicting clinical outcome of cancers and new targets for cancer therapies. Oncogenomics also target personalized cancer treatment. Cancer develops due to DNA mutations and epigenetic alterations that accumulate randomly. Identifying and targeting the molecular signature in an individual patient -as early as possible- may lead to increased treatment efficacy.

Driver vs passenger genes:

- Most mutations found through sequencing of tumor tissue appear to be random, are not recurrent in particular cancer types, and probably **occurred as the cancer developed**, rather than directly causing the neoplasia to develop or progress. Such mutations are referred to as **"passenger"** mutations However, a subset of a few hundred genes has been repeatedly found to be mutated at high frequency in many samples of the same type of cancer or even in multiple different types of cancers, mutated in fact far too frequently to simply be passenger mutations. These genes are thus presumed to be involved in the development or progression of the cancer itself and are therefore referred to as **"driver"** genes, that is, they harbor mutations (so-called driver gene mutations). Although many driver genes are specific to particular tumor types, some, such as those in the **TP53** gene encoding the p53 protein, are found in the vast majority of cancers of many different types. Although the most common driver genes are now known, it is likely that additional, less abundant driver genes will be identified as The Cancer Genome Atlas continues to grow.



❖ Variants of cancer include both driver and passenger:

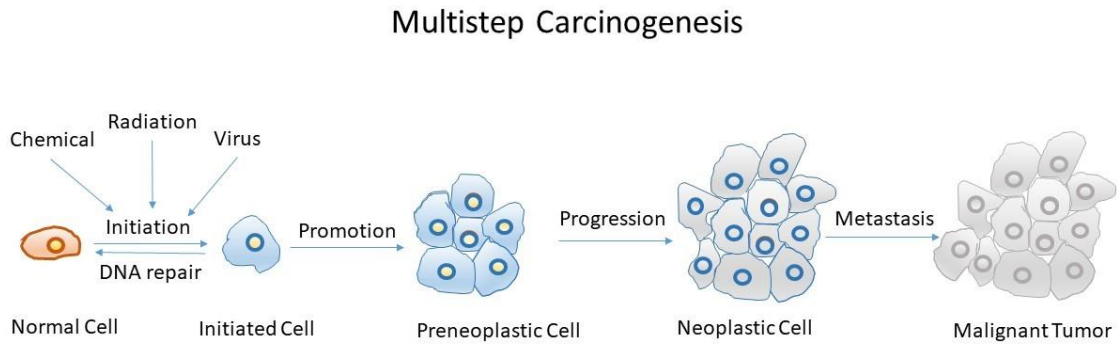
- Mutations that provide a selective growth advantage, and thus promote cancer development, are termed **driver mutations**, and those that do not are termed **passenger mutations**.
- Passenger variants are classified into variants that have no impact on cancer and others which do have impact on cancer progression.
- Some passenger mutations promote tumor growth, while others might help hinder cancer development. Those variants have protective effects against tumor growth.
- Even within the same tumor type, like colon cancer, the specific genes mutated can vary from person to person making cancer a unique disease for each individual. Although a small number of mutations are commonly shared between multiple cancer patients, the majority of passenger mutations are unique to individual cases.

❖ Spectrum of Driver Gene Mutations:

- What causes these mutations to occur?
  Replication errors, environmental agents and failure of DNA repair could occur to dividing and arrested cells, this will increase the rate of variants around the genome. When normal repair processes fail, and when cellular apoptosis does not occur, irreversible DNA damage may occur. This can eventually lead to malignant tumors.
- If, by chance, mutations occur in critical driver genes in a particular cell, then the oncogenic process may be initiated.

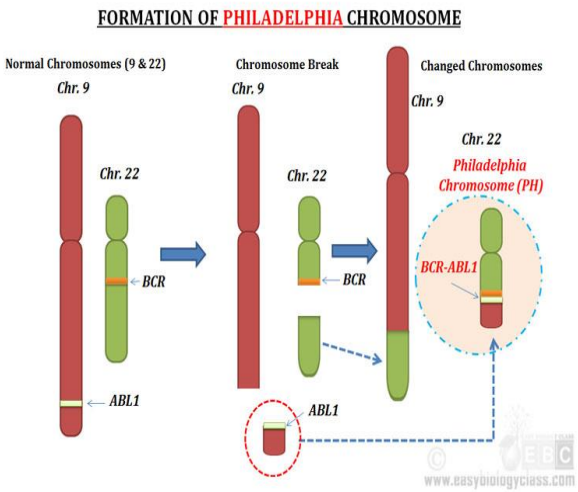
❖ Multistep carcinogenesis:

- The transformation of a normal cell into a cancer cell is a multi-step process that involves initiation, promotion, progression and finally malignancy. This process takes years and starts with a single cell in which the right genes are mutated and escape repair, the cell does not appropriately die and begins to proliferate abnormally. Then, additional mutations occur that select more rapidly growing cells within this population leading to a tumor with rapid growth and malignancy. By the time the cells are cancerous, proto-oncogenes have been activated and tumor suppressor genes inactivated. Neoplastic cells can invade nearby tissues or blood vessels and establish metastasis.



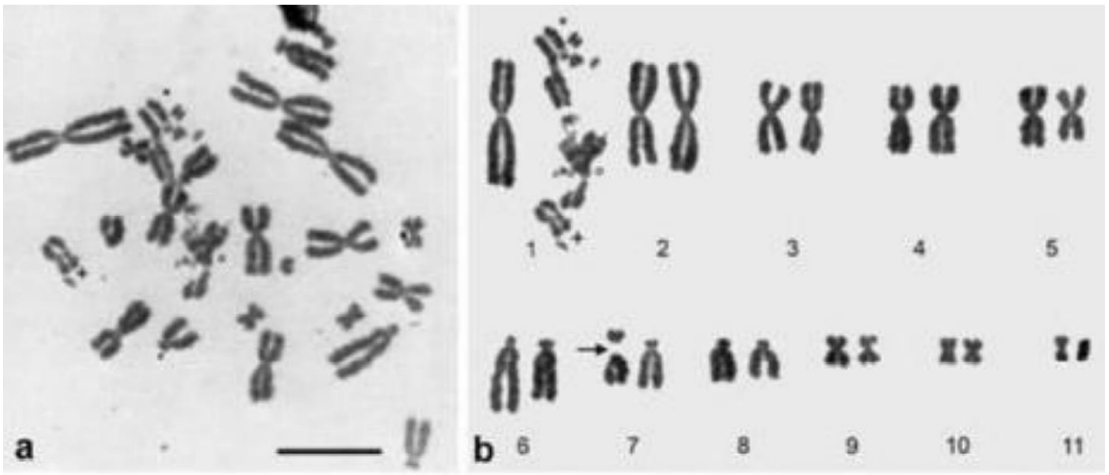
❖ Driver mutations could occur on the chromosomal level:

- Driver mutations could occur on the chromosomal level and sub-chromosomal variants can also serve as driver mutations. Particular translocations are sometimes highly specific for certain types of cancer and involve specific genes e.g., the BCR - ABL translocation in chronic myelogenous leukemia



- Because of reciprocal translocation of genetic material between chromosome 9 and 22, some gene sequences from chromosome 9 migrates on chromosome 22 and vice versa. The translocation results in the fusion of the BCR gene of chromosome 22 and ABL gene of chromosome 9. Consequently, BCR-ABL oncogene is formed. The uncontrolled expression of this oncogene results in the activation of tyrosine kinase protein which causes uncontrolled and uninterrupted cell division (especially in the bone marrow and blood cell) → chronic myeloid leukemia (CML).

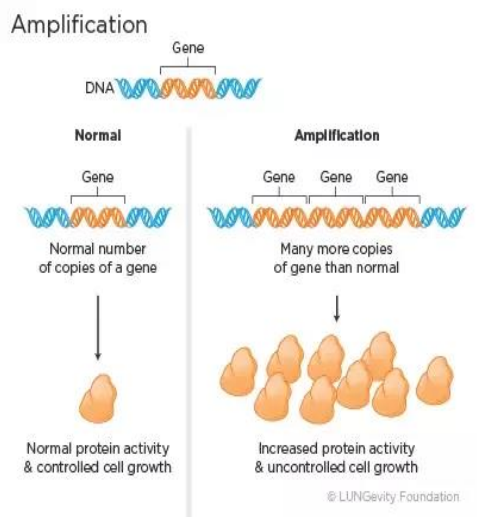
- Other cancers can show complex rearrangements in which chromosomes break into numerous pieces and rejoin, forming novel and complex combinations (a process known as **"chromosome shattering"**).
  - Chromosomal shattering occurs when genomic repair mechanisms fail resulting in inappropriate recombination of chromosomal fragments. During this process fragments of the chromosome may be lost or remain and reassemble incorrectly. Shattering may be directly induced by ionizing irradiation. When a portion of the nuclei is irradiated, multiple chromosomes sitting in the irradiated area are shattered into pieces.



Metaphase spreads with damaged chromosomes obtained after laser UV microirradiation of nuclei in living Chinese hamster cells. Nuclei in living Chinese hamster cells were microirradiated (k = 257 nm) at a single nuclear site comprising about 5% of the total nuclear area. Microirradiated cells were followed to the next mitosis (about 3-15 h) in medium with 1 mM caffeine. a, b Metaphase spread (a) and the corresponding karyogram (b) from a diploid, fibroblastoid Chinese hamster cell reveal a shattered chromosome 1 and a break in a chromosome 7.

Gene amplification:

- Another mechanism for activation of oncogenes is DNA amplification, leading to overexpression of the gene product (protein). An increased amount of the products of certain oncogenes produced by amplification play a role in the progression of tumors.
- Large genomic alterations include deletions of a segment of a chromosome or multiplication of a chromosomal segment to produce regions with many copies of the same gene (gene amplification).



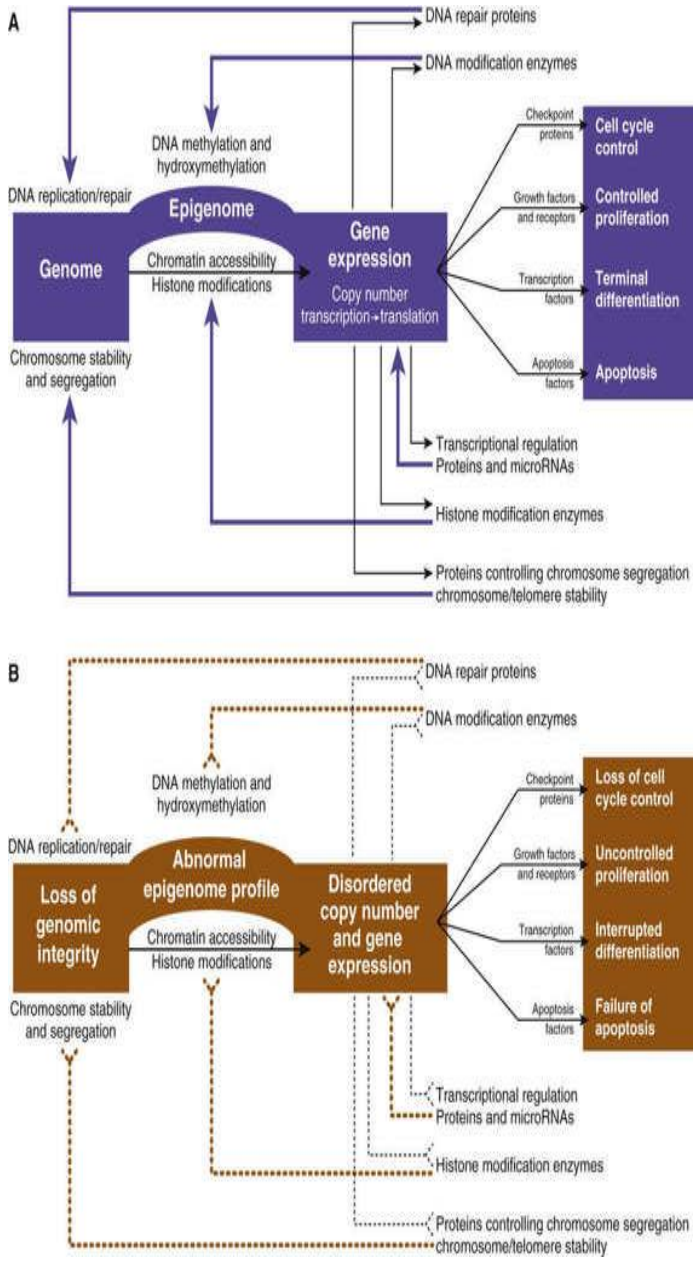
❖ The Cellular Functions of Driver Genes:

- The nature of some driver gene mutations comes as no surprise: the mutations directly affect specific genes that regulate processes that are readily understood to be important in oncogenesis.
- These processes include cell-cycle regulation, cellular proliferation, differentiation and exit from the cell cycle, growth inhibition by cell-cell contacts, and programmed cell death (apoptosis).

❖ Classes of driver genes:

1. Genes with specific effects on cellular proliferation or Survival.
2. Genes with global effects on genome or DNA integrity.

- Overview of normal genetic pathways controlling normal tissue homeostasis. The information encoded in the genome (*black arrows*) results in normal gene expression, as modulated by the epigenomic state. Many genes provide negative feedback (*purple arrows*) to ensure normal homeostasis.
- Epigenetic modifications alter chromatin accessibility which affect gene expression.
- Noncoding microRNA play a role in transcriptional regulation of proteins.
- Perturbations in neoplasia. Abnormalities in gene expression (*dotted black arrows*) lead to a vicious cycle of positive feedback (*brown dotted lines*) of progressively more disordered gene expression and genome integrity.
- leading to a vicious cycle causing loss of cell cycle control, uncontrolled proliferation, interrupted differentiation, and defects in apoptosis.



❖ Activated Oncogenes and Tumor Suppressor Genes:

- Both classes of driver genes—those with specific effects on cellular proliferation or survival and those with global effects on genome or DNA integrity —can be further subdivided into one of two functional categories depending on how, if mutated, they drive oncogenesis. The first category includes **proto-oncogenes**. These are normal genes that promote growth and survival of cells. When mutated in very particular ways, become driver genes through alterations that lead to excessive levels of activity. Once mutated in this way, driver genes of this type are referred to as activated oncogenes. Only a single mutation at **one allele** can be sufficient for activation. The mutations that activate a proto-oncogene can range from highly specific point mutations causing dysregulation or hyperactivity of a protein, to chromosome translocations that drive overexpression of a gene, to gene amplification events that create an overabundance of the encoded mRNA and protein product.

❖ Oncogenes encode proteins such as the following:

1. Proteins in signaling pathways for cell proliferation.
2. Transcription factors that control the expression of growth-promoting genes.
3. Inhibitors of programmed cell death machinery.

- The second, and more common, category of driver genes includes **tumor suppressor genes (TSGs)**, variants in which cause a loss of expression of proteins necessary to control the development of cancers. To drive oncogenesis, loss of function of a TSG typically requires mutations **at both alleles.**
- Loss-of-function mechanisms can range from missense, nonsense, or frame-shift mutations to gene deletions or loss of a part or even an entire chromosome.

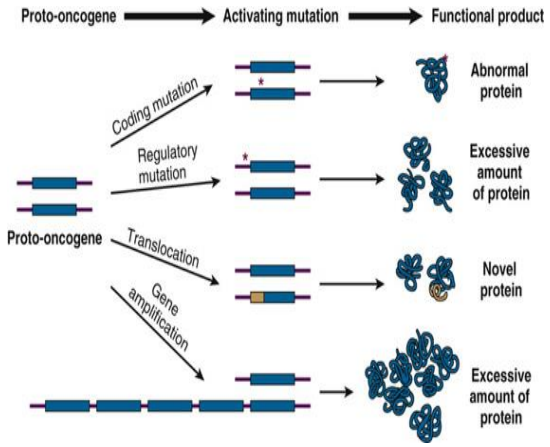
❖ Loss of function of TSGs can also result from epigenomic transcriptional silencing due to:

1. altered chromatin conformation.
2. promoter methylation.
3. translational silencing by miRNAs or disturbances in other components of the translational machinery.

❖ TSGs encode proteins involved in many aspects of cellular function, including but not limited to:

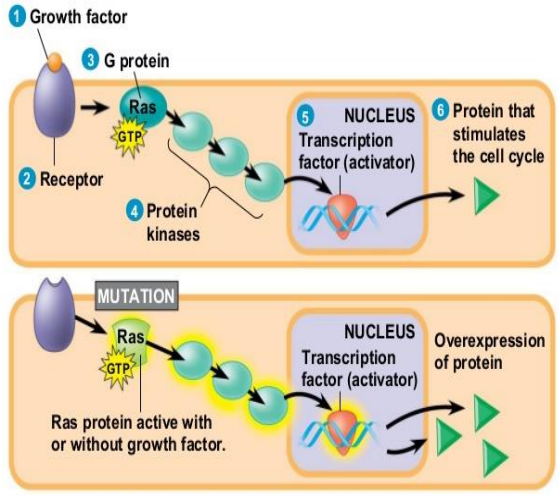
1. maintenance of correct chromosome number and structure.
2. DNA repair proteins.
3. proteins involved in regulating the cell cycle, cellular proliferation, or contact inhibition.

- Different mutational mechanisms leading to proto-oncogene activation. These include a single point mutation leading to an amino acid change that alters protein function, mutations in promoter region or untranslated region (UTR), or translocations that increase expression of an oncogene, a chromosome translocation that produces a novel product with oncogenic properties, and gene amplification leading to excessive amounts of the gene product.



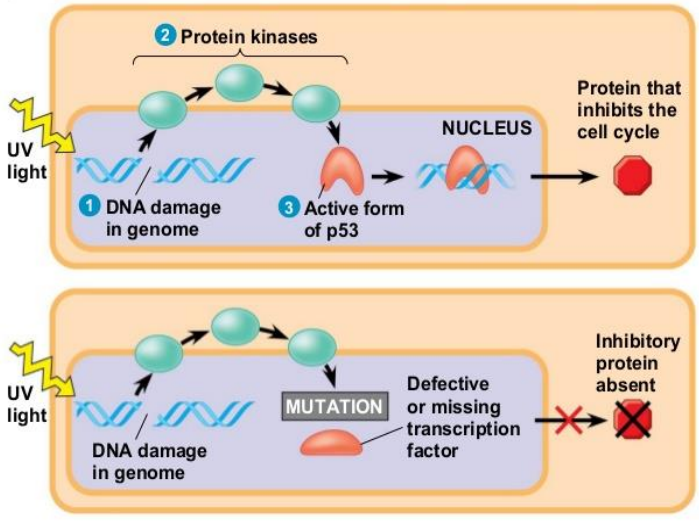
- Ras belongs to the family of small G proteins that govern various cellular signal transduction pathways. RAS is stimulated when a growth factor binds to its receptor, this binding causes phosphorylation of RAS. This results in a series of downstream signaling cascades, which initiate cell growth, differentiation, proliferation and cell survival. Certain point mutations within the Ras gene lock the protein into a constitutively active state (autoactivation), which leads to cell signaling even in the absence of external signals leading to cancer.

Figure 16.17



- **Activation of RAS is considered a gain of function.**
- Suppression of cell cycle can be important in the case of damage to cell's DNA, p53 prevents a cell from passing on mutations due to DNA damage (tumor suppressor gene).
- DNA damage induces p53 activation through the protein kinase pathway, activated p53 translocates to the nucleus and inhibits the progression of cell cycle.
- Mutations in the p53 gene prevent suppression of cell cycle when needed. Consequently, cell cycle proceeds unchecked.

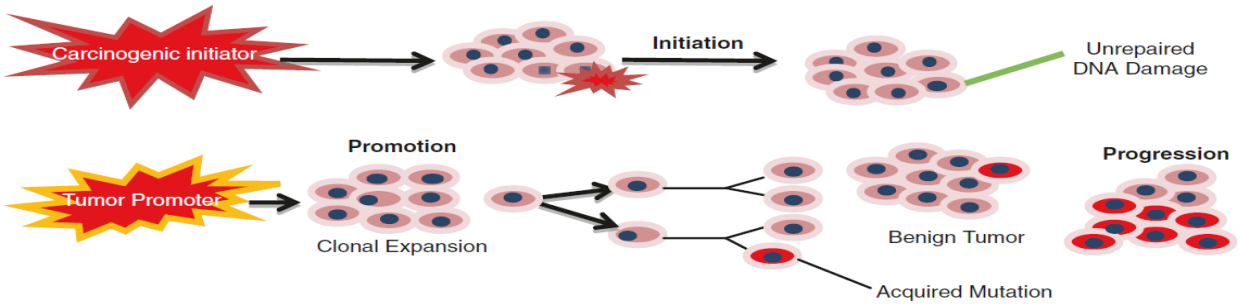
Figure 16.18



❖ Cellular Heterogeneity within Individual Tumors:

- ➲ Tumor heterogeneity refers to the difference between genotype and phenotype of tumor cells in different parts of the same malignancy or in different parts of the same patient.

❖ The accumulation of driver gene mutations does not occur synchronously, in lockstep, in every cell of a tumor. To the contrary, cancer evolves along multiple lineages within a tumor. Mutational and epigenetic events in different cells activate proto-oncogenes and cripple the machinery for maintaining genome integrity, leading to more genetic changes in a vicious cycle of more mutations and worsening growth control. The lineages that experience an enhancement of growth, survival, invasion, and distant spread will come to predominate as the cancer evolves and progresses.



- ➲ Multiple somatic mutations are generally needed for full-fledged cancer. These changes generally include at least one active oncogene and the loss of several tumor suppressor genes.

❖ The evolutionary process of cancer begins with a single cell. At each division, a cell acquires a few mutations to its genetic code. If the mutations occur in driver genes, the cell lineage follow a path of rapid growth. If these mutations survive, cells continue to divide at a rate faster than normal, and the result is a tumor. With each additional division, the cell continues to acquire mutations. The result is that a single tumor can consist of a variety of unique cell populations. As tumors metastasize, or spread to other locations throughout the body, the possibility for diversity grows.

❖ The original clone of neoplastic cells evolves and gives rise to multiple sub-lineages each carrying a set of mutations and epigenomic alterations that are different from but overlap with what is carried in other sub-lineages.

❖ The profile of mutations and epigenomic changes can differ:

A. Between the primary and its metastases.
B. Between different metastases.
C. Between the cells of the original tumor or within a single metastasis.

A paradigm for the development of cancer

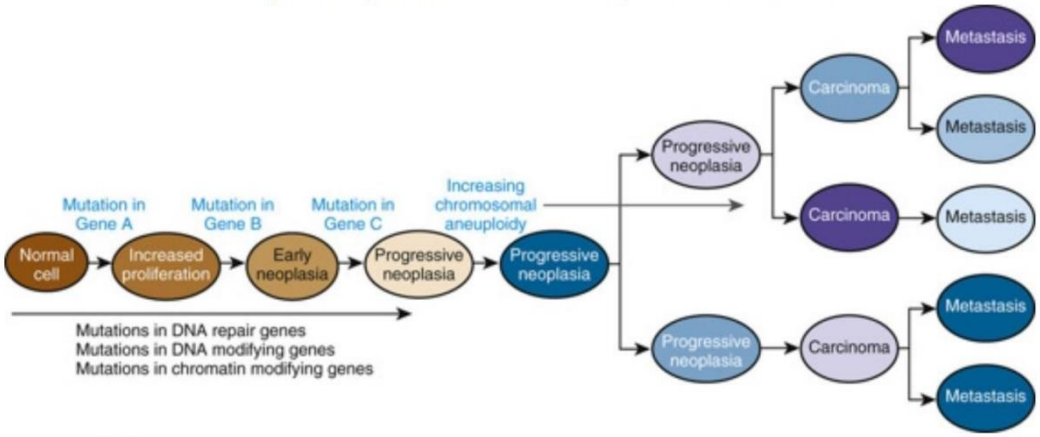


FIGURE 15-4 Stages in the evolution of cancer. Increasing degrees of abnormality are associated with sequential loss of tumor suppressor genes from several chromosomes and activation of proto-oncogenes, with or without a concomitant defect in DNA repair. Multiple lineages, carrying different mutations and epigenomic profiles, occur within the primary tumor itself, between the primary and metastases and between different metastases.

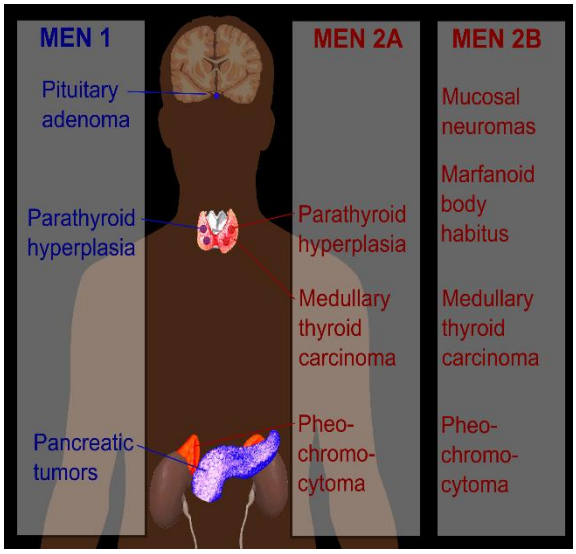
- Heterogeneity between primary tumors and different metastases represents a challenge. If treatment plans are made based on biopsy of the primary tumor but the metastases differ from each other and from the primary tumor, the efficacy of treatment will be greatly limited.

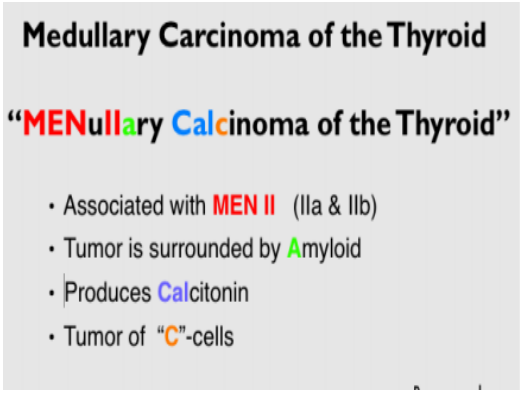
❖ Cancer in Families:

- hereditary cancer syndromes follow mendelian patterns of inheritance, where increased incidence is due primarily to inheritance of a single mutant gene with high penetrance.
- Approximately 100 different genes in which deleterious mutations increase the risk for cancer many-fold higher than in the general population.
- There are also many dozens of additional genetic disorders that are not usually considered to be hereditary cancer syndromes and yet include some increased predisposition to cancer (for example, the ten- to twenty-fold increased lifetime risk for leukemia in Down syndrome).
- Not all families with an apparently increased incidence of cancer can be explained by known mendelian or clearly recognized genetic disorders. These families likely represent the effects of both shared environment and one or more genetic variants that increase susceptibility and are therefore classified as **multifactorial**, with complex inheritance. Although individuals with a hereditary cancer syndrome represent ~ 5% of all patients with cancer, identification of a genetic basis for their disease has great importance both for clinical management of these families and for understanding cancer in general.

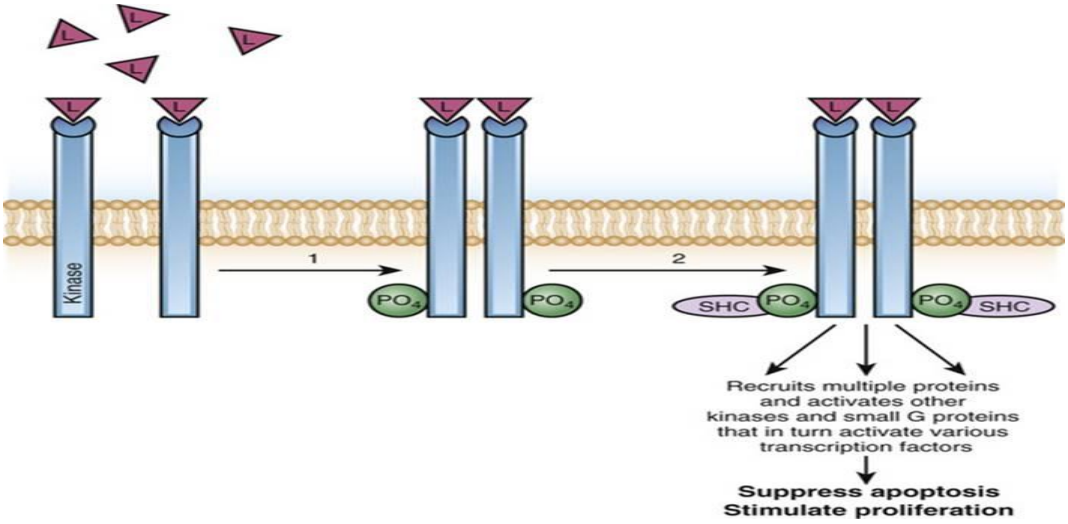
❖ Activated Oncogenes in Hereditary Cancer Syndromes:

➲ Multiple Endocrine Adenomatosis, Type 2

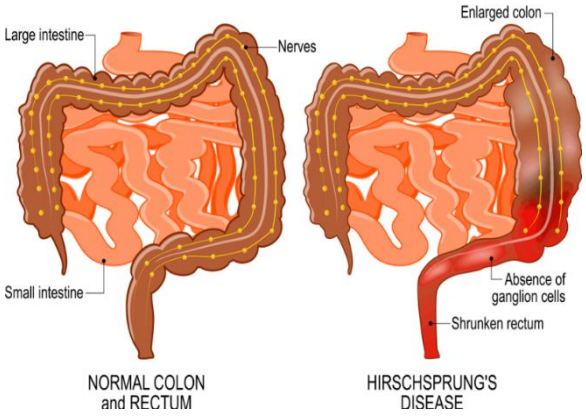




- Adenomatosis: An abnormal overgrowth of, or TUMOUR formation in, two or more of the ENDOCRINE glands.
- MEN2-A is an autosomal dominant disorder characterized by: high incidence of medullary carcinoma of the thyroid that is often but not always associated with pheochromocytoma, benign parathyroid adenomas, or both.
- Pheochromocytoma: is a rare, usually noncancerous (benign) tumor that develops in an adrenal gland.
- Patients with the rarer type B variant, MEN2B, have, in addition to the tumors seen in patients with MEN2A, thickening of nerves and the development of benign neural tumors, known as neuromas, on the mucosal surface of the mouth and lips and along the GI tract.
- The gene responsible for MEN2 is a proto-oncogene called **RET** which codes for the tyrosine kinase RET protein subunit of a cell surface receptor (gain of function). The mutation of the RET gene leads to activation of the receptor, with uncontrolled growth and overactivity of the target cells, and subsequent tumor formation.
- Individuals who inherit an activating mutation in RET have a greater than 60% chance of developing a particular type of thyroid carcinoma (medullary). More sensitive tests, such as blood tests for thyrocalcitonin or urinary catecholamines synthesized by pheochromocytomas, are abnormal in above 90% of heterozygotes for MEN2.
- RET encodes a cell-surface protein that contains an extracellular domain that can bind signaling molecules and a cytoplasmic tyrosine kinase domain. Tyrosine kinases are a class of enzymes that phosphorylate tyrosines in proteins. Tyrosine phosphorylation initiates a signaling cascade of changes in protein-protein and DNA-protein interactions and in the enzymatic activity of many proteins.
- Normally, tyrosine kinase receptors must bind specific signaling molecules in order to undergo the conformational change that makes them enzymatically active and able to phosphorylate other cellular proteins.
- Mutations in RET that cause MEN2A increase its kinase activity even in the absence of its ligand (a state referred to as constitutive activation).



- The *RET* gene is expressed in many tissues of the body and is required for normal embryonic development of autonomic ganglia and kidney. It is unclear why germline activating mutations in this proto-oncogene result in a particular cancer of distinct histological types restricted to specific tissues, whereas other tissues in which the oncogene is expressed do not develop tumors.



- Interestingly, *RET* is the same gene implicated in Hirschsprung disease, although those mutations are usually loss-of-function, not activating mutations like the ones encountered in MEN syndrome.
- There are, however, some families in which the same mutation in RET can act as an activated oncogene in some tissues (such as thyroid) and cause MEN2A, while not having sufficient function in other tissues (loss of function), such as the developing enteric neurons of the gastrointestinal tract, resulting in Hirschsprung disease. Thus, even the identical mutation can have different effects on different tissues.
- In these cases, patients suffer from both MEN syndrome and Hirschsprung disease.
  - MEN → as a result of activating mutation of RET gene in endocrine tissues.
  - Hirschsprung disease → as a result of inactivating mutations of RET gene in enteric neurons of the GI tract.

- **HSCR:** Hirschsprung disease (inactivation → loss of RET function).
- **MEN:** Multiple endocrine neoplasia (Activation → gain of RET function).
- **FMTC:** Familial medullary thyroid cancer (Activation → gain of RET function).
- **SMTC:** Sporadic medullary thyroid cancer (Activation → gain of RET function).