p53 Inhibition for Cancer Therapy

- The inhibition of p53 can protect normal cells during genotoxic chemotherapy or radiation therapy. The side effects of genotoxic therapy for cancer are largely caused by p53-mediated apoptosis.
- The small molecule pifithrin-alpha can block p53-dependent transcriptional activity and protect mice from the lethal side effects associated with anticancer treatment.

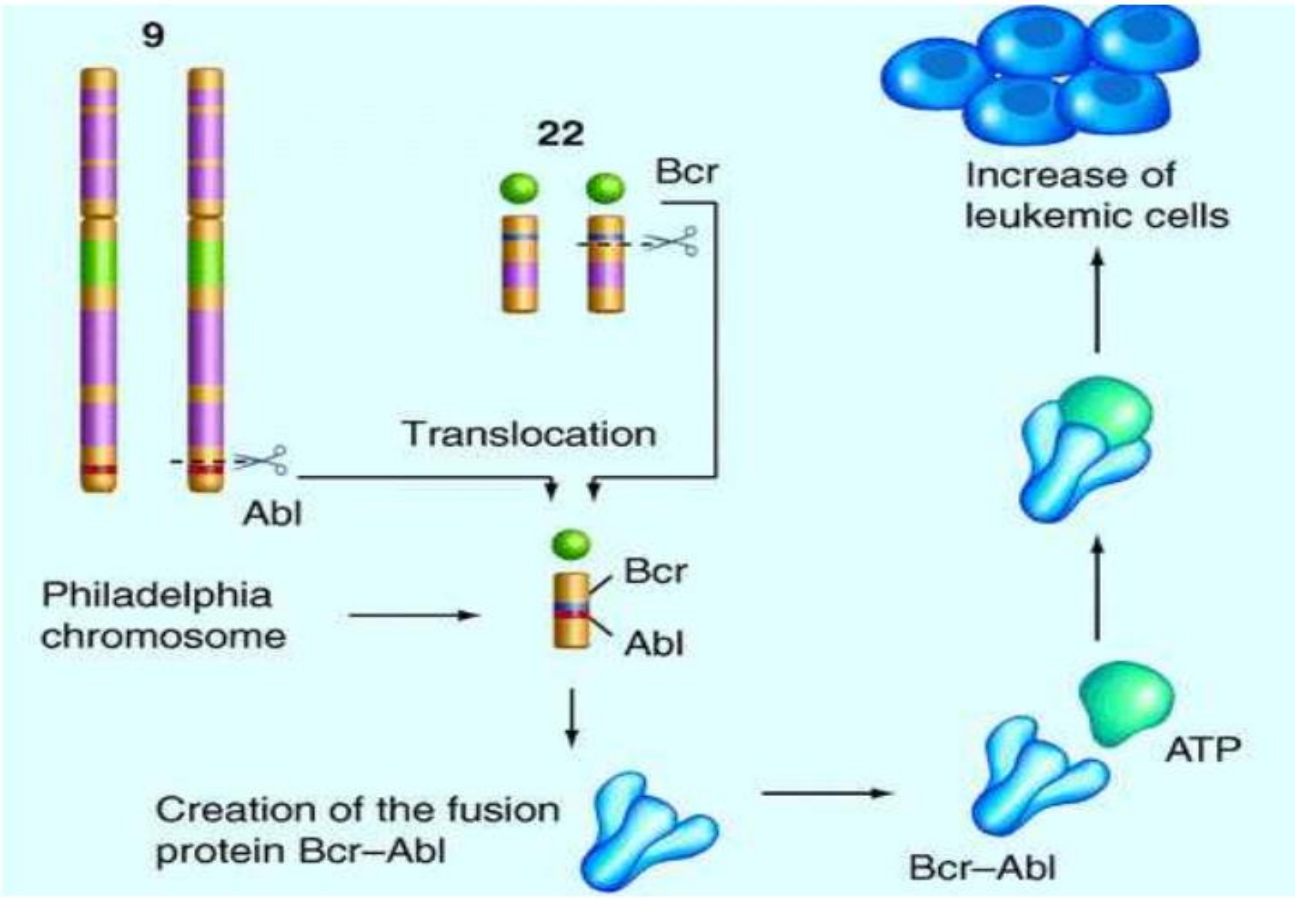
p53 Gene Therapy

- The first **p53-based gene therapy** was reported in 1996. A retroviral vector containing the wild-type p53 gene under the control of an actin promoter was injected directly into tumors of non small cell lung cancer patients .
- After development of a **replication-defective recombinant p53 virus (Ad5CMV-p53), many clinical trials have been performed, including one in esophageal cancer patients** .
- A **few trials reached phase III**, but final approval from the FDA has not yet been granted .
- Recently, **p53-based gene therapy has been developing in China**

Oncogene activation can also arise through chromosomal translocation

The Philadelphia chromosome, discovered in 1960 in the Philadelphia laboratories of Peter Nowell and David Hungerford, is the best-known example of an oncogenic chromosomal translocation.

- In this case, one end of chromosome 9 is exchanged with one end of chromosome 22. At the broken end of chromosome 22 lies **the *BCR* gene,** which fuses with a fragment of chromosome 9 that carries the ***ABL1* gene**; this fused **chromosome is called the Philadelphia chromosome.** When the **chromosome ends fuse, the two genes also fuse with each other to become *BCR-ABL*.**



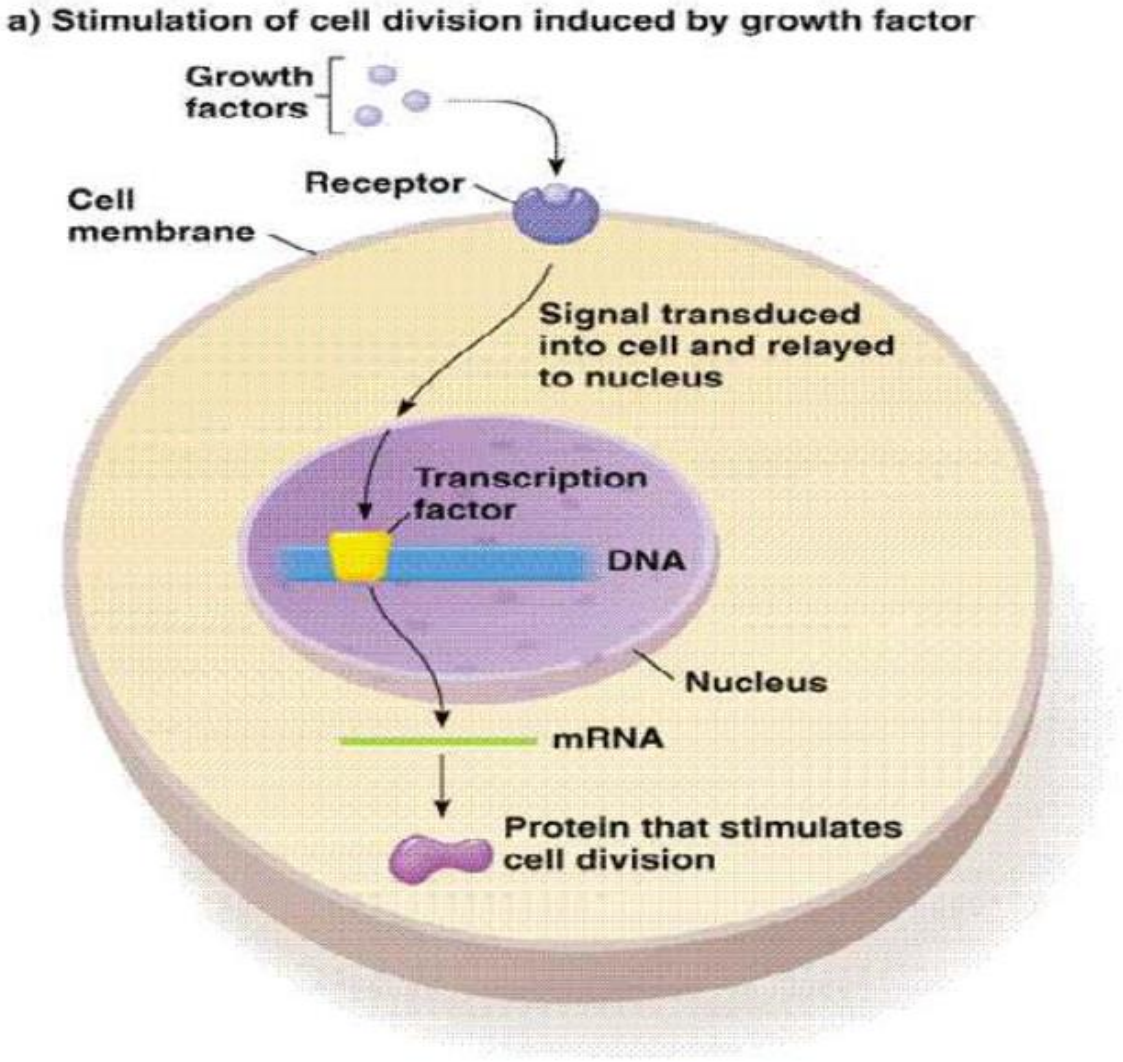
The **fused gene is expressed, and it encodes a protein that exhibits high protein tyrosine kinase activity**, **courtesy of the *ABL1* half of the protein**. The unregulated expression of this protein activates a repertoire of other proteins that are involved in cell cycle regulation and stimulation of cell division. As a result, the **Philadelphia chromosome is associated with chronic myelogenous leukemia (CML) and several other forms of leukemia**.

- Evidence that cells expressing the BCR-ABL fusion protein suffer from oncogene addiction comes from studies using **imatinib**, an ABL kinase inhibitor, to treat CML patients. This drug, which **targets the tyrosine kinase activity of ABL**.However, more long-**term clinical studies using imatinib to treat CML patients are showing that some patients may begin to show signs of drug resistance over time.**

Tumor Suppressor Gene

- Tumor suppressor genes often function to restrain inappropriate cell growth and division, as well as to stimulate cell death to keep our cells in proper balance.
- In addition, some of these genes are involved in DNA repair processes, which help prevent the accumulation of mutations in cancer-related genes.

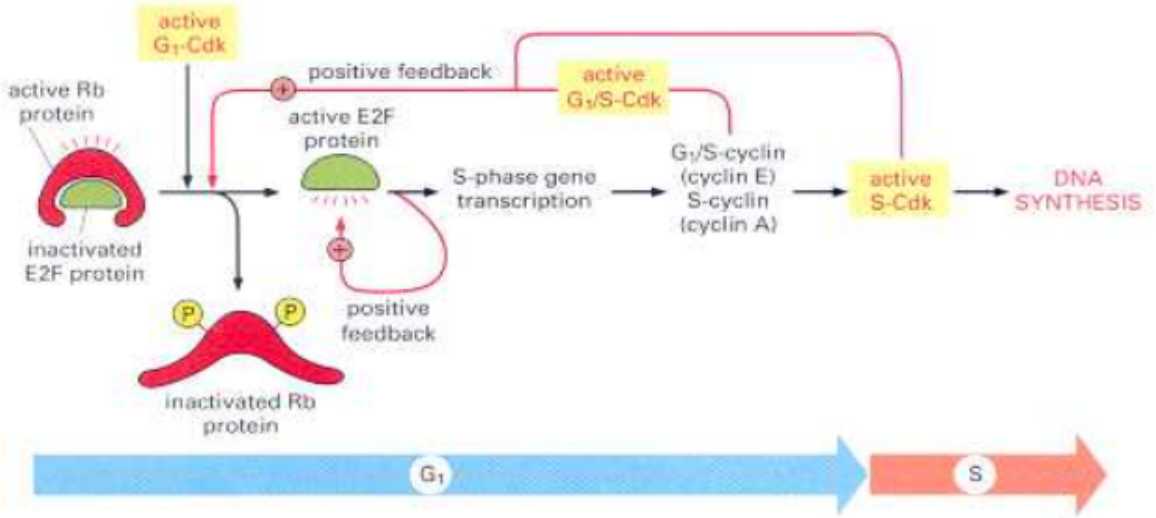
- In this way, tumor suppressor genes act as "brakes" to stop cells in their tracks before they can take the road to cancer.



signal transduction pathway

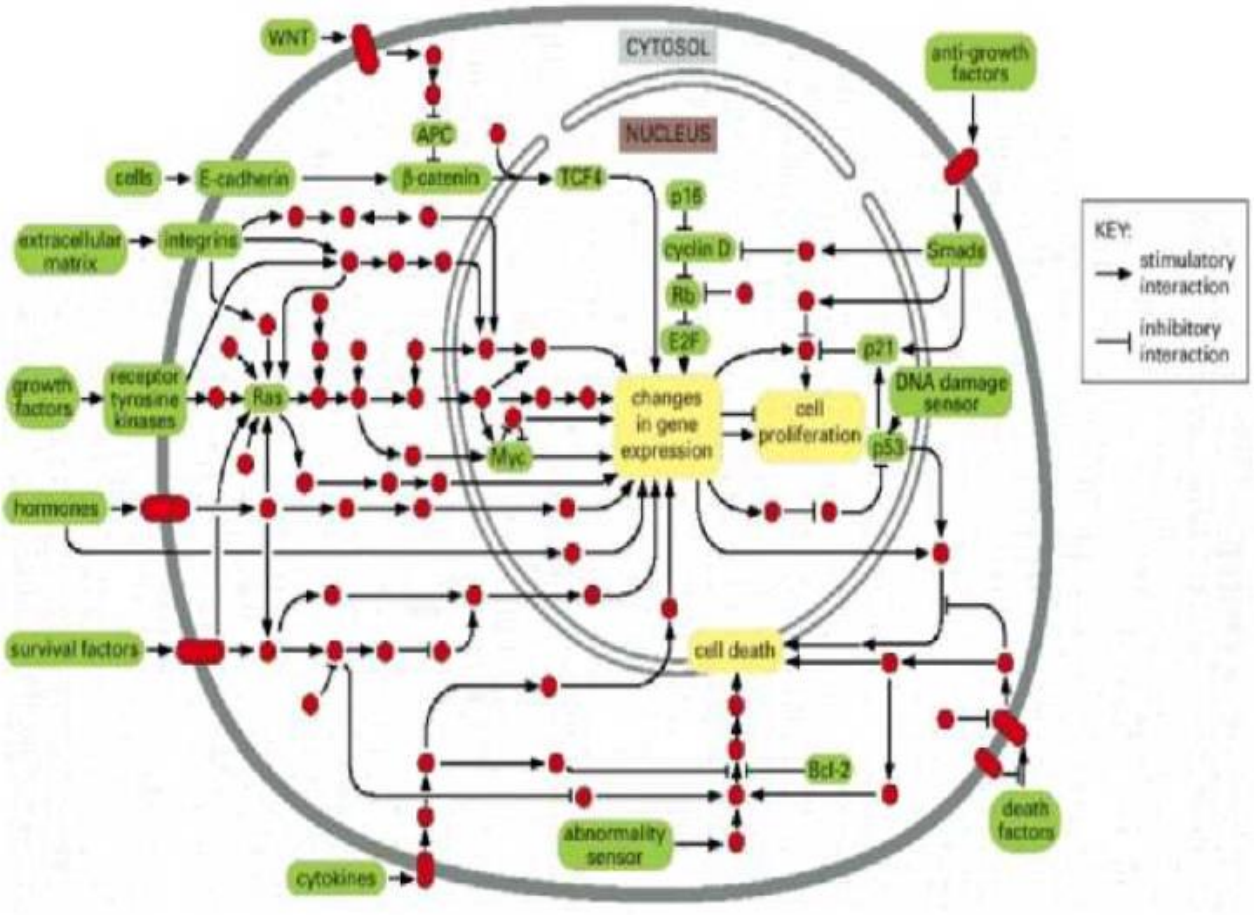
- While the Retinoblastoma protein differs in its function, it acts in a similar manner to typical signal transduction proteins.
- The RB1 gene is widely expressed, encoding a 110-kd (4.7 kb) nuclear protein, **pRb.**
- In normal cells pRb is **inactivated by phosphorylation** and **activated by dephosphorylation**.
- Active (dephosphorylated) pRb binds and inactivates the **cellular transcription factor E2F1,** the function of which is required for cell cycle progression.

- **E2F binds to specific DNA sequences** in the promoters of many genes that encode proteins required for S-phase entry, including G1/S-cyclins and S-cyclins.
- **E2F's function is controlled primarily by an interaction with the retinoblastoma protein.**
- During G1, Rb binds to E2F and blocks the transcription of S-phase genes.
- When cells are stimulated to divide by extracellular signals, active G1-Cdk accumulates and phosphorylates Rb, reducing its affinity for E2F.
- Phosphorylation is governed by a cascade of cyclins, cyclin-dependent kinases and cyclin kinase inhibitors .
- The Rb then dissociates, allowing E2F to activate S-phase gene expression.
- This G1-S checkpoint seems to be the most crucial in the cell cycle, and thus the major source of cancer



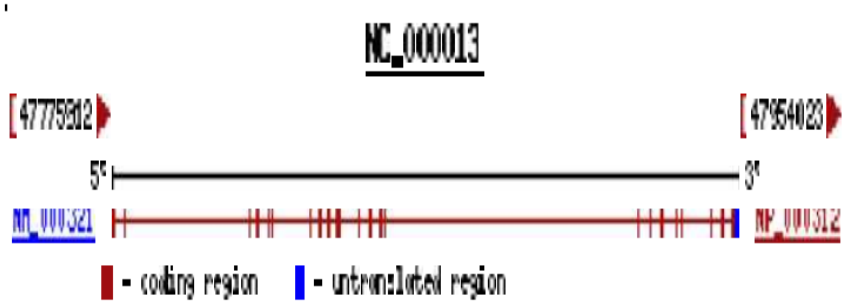
The complete loss of Rb does not immediately cause **increased proliferation of other cell types, in part because Hct1 and p27 (two other proteins) provide assistance in G1 control, and in part because other cell types contain Rb-related proteins that provide backup support in the absence of Rb** .

It is also likely that other proteins, unrelated to Rb, help to regulate the activity of E2F.



The Size and Structure of Rb Protein

- A 928 Amino Acid Protein.
- It contains a **putative "leucine-zipper" motif** that is exclusively **encoded by exon-20**.
- Rb A = **Comprising 268 amino acids**, this is the Retinoblastoma-associated protein A domain. **This domain has the cyclin fold as predicted** .
- Rb B = **Comprising 173 amino acids**, this is the Retinoblastoma-associated protein B domain.
- The crystal **structure of the Rb pocket bound to a nine-residue E7 peptide containing the LxCxE motif, shared by other Rb-binding viral and cellular proteins**, shows that the **LxCxE peptide binds a highly conserved groove on the B domain.** The **B domain has a cyclin fold** .



Commonly inherited cancers and associated tumor suppressor genes

Inherited Cancer	Mutated Tumor Suppressor Gene(s)	Gene Function(s)	Associated Noninherited Cancers
Retinoblastoma	RB1	Cell division, DNA replication, cell death	Many different cancers
Li-Fraumeni syndrome (brain tumors, sarcomas, leukemia)	TP53	Cell division, DNA repair, cell death	Many different cancers
Melanoma	CDKN2A (INK4A)	Cell division, cell death	Many different cancers
Colorectal cancer (due to familial polyposis)	APC	Cell division, DNA damage, cell migration, cell adhesion, cell death	Most colorectal cancers
Colorectal cancer (without polyposis)	MLH1, MSH2, MSH6	DNA mismatch repair, cell cycle regulation	Colorectal, gastric, endometrial cancers
Breast and/or ovarian cancer	BRCA1, BRCA2	Repair of double-stranded DNA breaks, cell division, cell death	Only rare ovarian cancers
Wilms' tumor	WT1, WT2	Cell division, transcriptional regulation	Wilms' tumors
Nerve tumors (including brain)	NF1, NF2	RAS-mediated signal transduction, cell differentiation, cell division, developmental processes	Small numbers of colon cancers, melanomas, neuroblastoma
Kidney cancer	VHL	Cell division, cell death, cell differentiation, response to cell stress	Certain types of kidney cancer

Include the notes about p53 related function ALSO AS EXAMPLE

Viruses and Cancer

Viruses Associated With The Development Of Human Neoplasia

VIRUSES	NEOPLASMS
RNA VIRUSES	
Human T-cell leukemia virus I	Some T-cell leukemia, Lymphoma
Human T-cell leukemia virus II	Some cases of hairy cell leukemia
Human immunodeficiency virus	Lymphoma; Kaposi's sarcoma

Promote

Changes in cell that are at the roots of cancer

Genetic and epigenetic alterations:

- Mutations
- Deletions
- Recombination's
- Transpositions
- Epigenetic alterations (DNA methylation, imprinting)
- Acquisition of viral genetic material

How do Viruses contribute to cancer?

- **Integrations** that cause activation or inactivation of oncogenes or tumor suppressors (e.g. RNA viruses)

- **Expression of genes** that alter key signal transduction pathways - this is our focus

- **Chronic activation of inflammatory responses**

How virus causes Cancers:

- The viral agents causing cancer in eukaryotic animals by integrating in host genome
  *A virus associated with malignancies in natural host, experimental animals or cell cultures.
  *viruses which modified proto-oncogene, obligatory host specific, with the ability immortalization, possess genes which stimulate growth and cause cancer.

Oncovirus

- An **oncovirus** is a virus that can cause cancer. This term originated from studies of acutely-transforming retroviruses in the 1950–60s, often called oncornaviruses to denote their RNA virus origin. It now refers to any virus with a DNA or RNA genome causing cancer and is synonymous with "tumor virus" or "cancer virus".

by transforming cells → cancer

- When a virus infects a cell, it expresses proteins that cause the cell to proliferate and/or block apoptosis
- Cancer is multi-factorial: Oncogenic viruses are very common, only a small % of people infected actually get cancer

Major viral cancers

Viruses are involved in about 15% of human cancers:

- Cancer of the cervix
- Cancer of the liver
- Certain leukemia's & lymphomas
- Kaposi's sarcoma

Major human Oncogenic Viruses

DNA Viruses

Small DNA tumor viruses

- Adenovirus
- SV40
- Human Papilloma virus (HPV)

Herpesviruses (large)

- Epstein Barr virus (EBV)
- Kaposi's Sarcoma Herpesvirus (KSHV)

Other

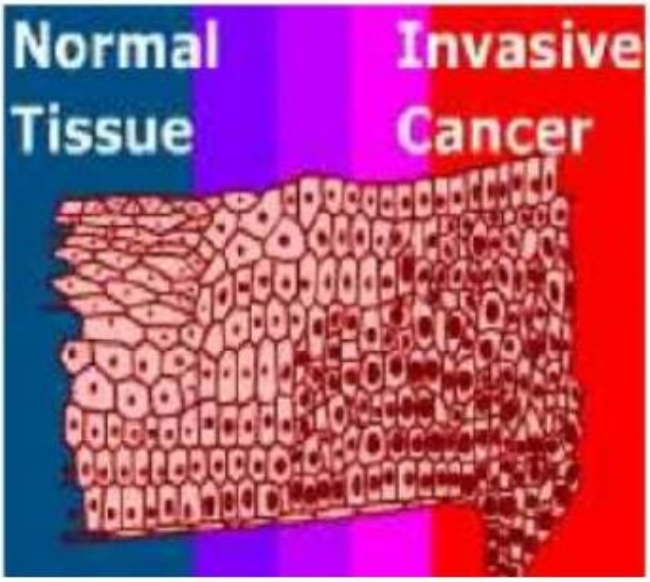
- Hepatitis virus B

RNA viruses

Human T-cell Leukemia Virus 1 (HTLV1)

Hepatitis virus C

Oncogenic viruses may be RNA or DNA



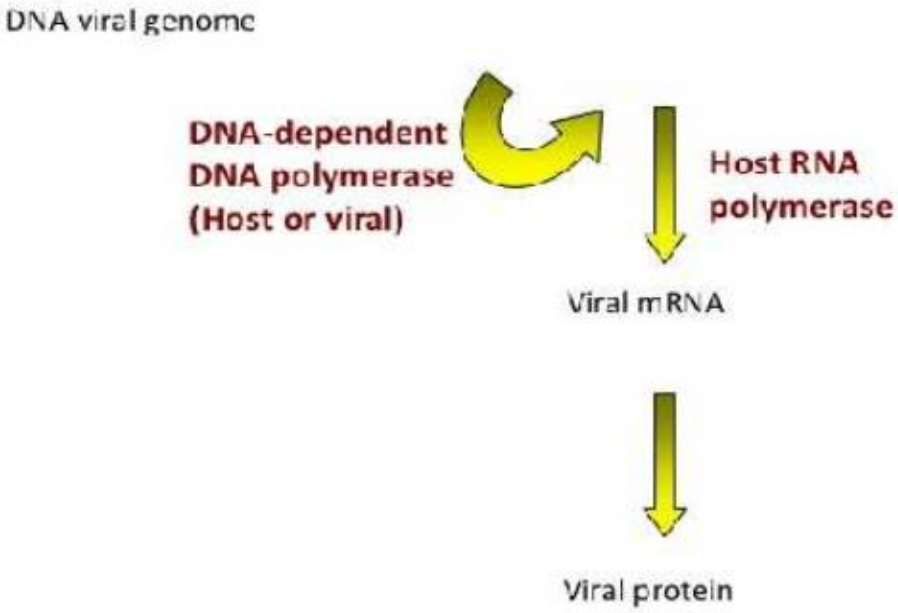
- 20% of human cancers believed to be of viral origin
- These include:
  - Cervical cancer
  - Burkitt's lymphoma
  - Hepatocarcinoma
  - Kaposi's sarcoma
- Virus is not only factor

Viruses Associated With The Development Of Human Neoplasia

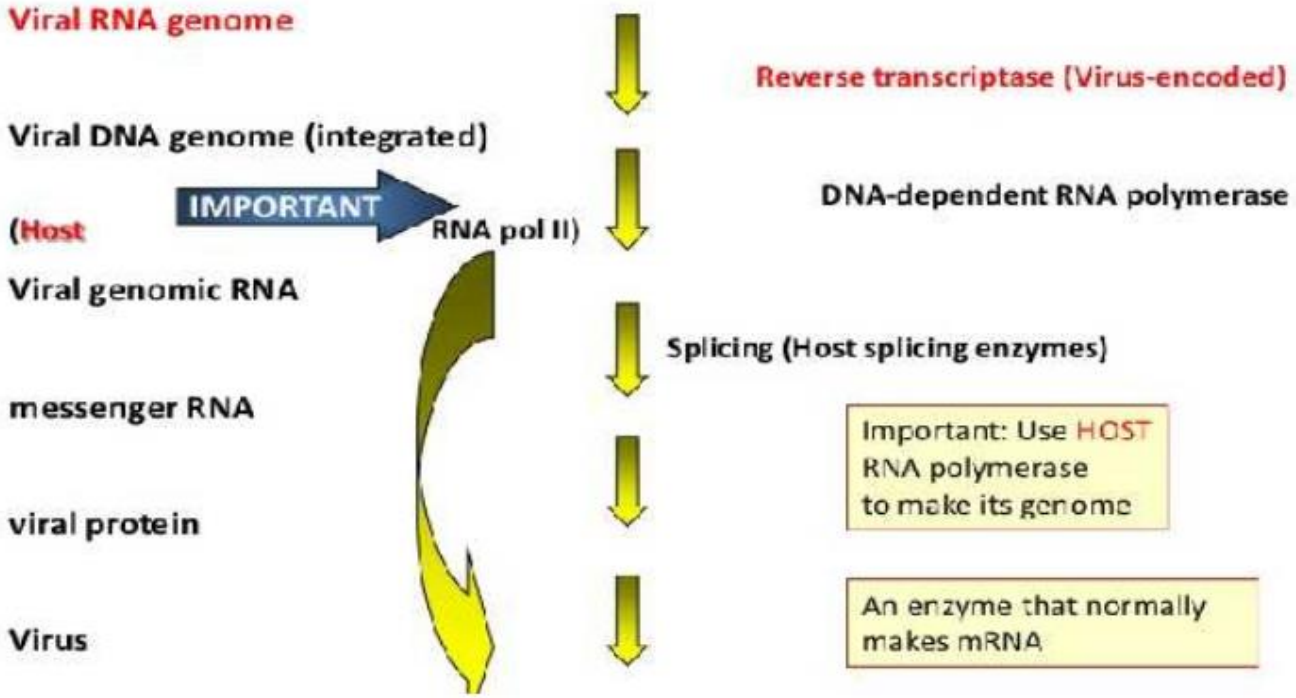
VIRUSES	NEOPLASMS
DNA VIRUSES	
Human papilloma virus	Cervical Ca, warts, ano-genital carcinoma
Herpes simplex virus II	Cervical carcinoma
Epstein-Barr virus	NPCa, African Burkitt's
Human Herpes virus 8	Kaposi's sarcoma
Hepatitis B virus	Hepatocellular Ca
Herpes simplex virus 6 (HBLV)	Certain B cell lymphomas

Two Major Classes of Tumor Viruses

DNA Tumor Viruses



RNA Tumor Viruses

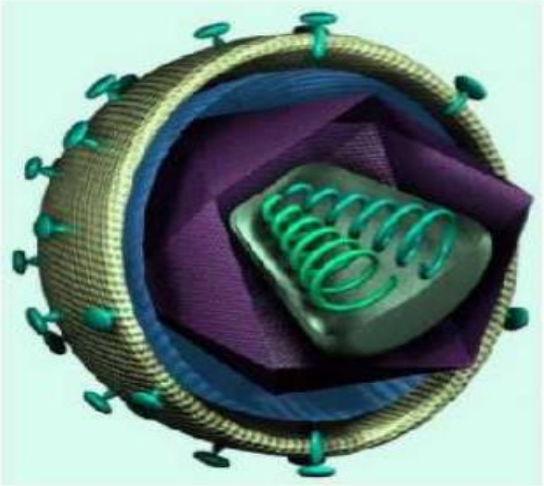


Oncogenic Retroviruses

- More than 40 different highly oncogenic retroviruses have been isolated from a variety of animals, including chickens, turkeys, mice, rats, cats, and monkeys. All of these viruses, like RSV, contain at least one oncogene In some cases, different viruses contain the same oncogenes, but more than two dozen distinct oncogenes have been identified among this group of viruses

Retroviruses:

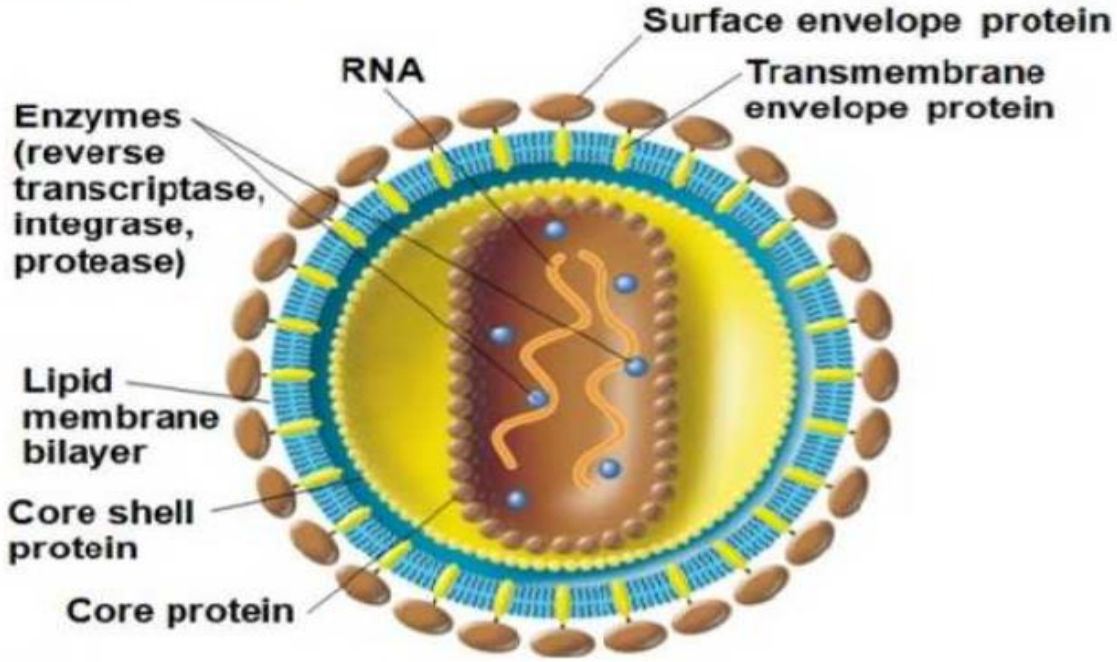
- 1.Avian leukosis viruses
- 2.Murine leukosis viruses
- 3.Murine mammary tumor virus
- 4.Leukosis-sarcoma viruses
- 5.Human T cell leukemia virus

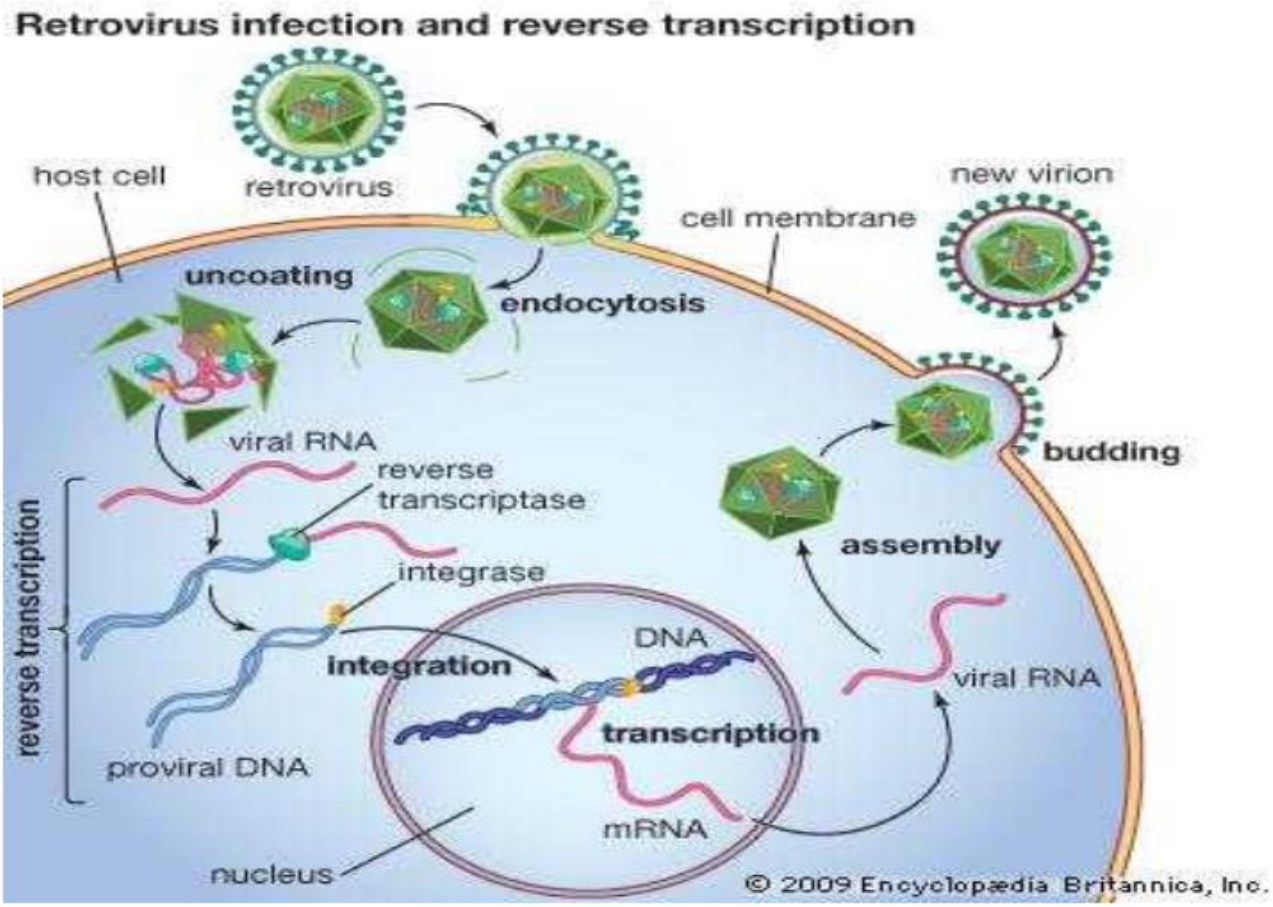


Retroviruses and cancer

Retroviruses employ the RNA-dependent DNA polymerase (reverse transcriptase) within their capsid to replicate their RNA genome into a DNA intermediate, which can be incorporated into the host cell's DNA by an *integrase* enzyme.

Retrovirus structure and function

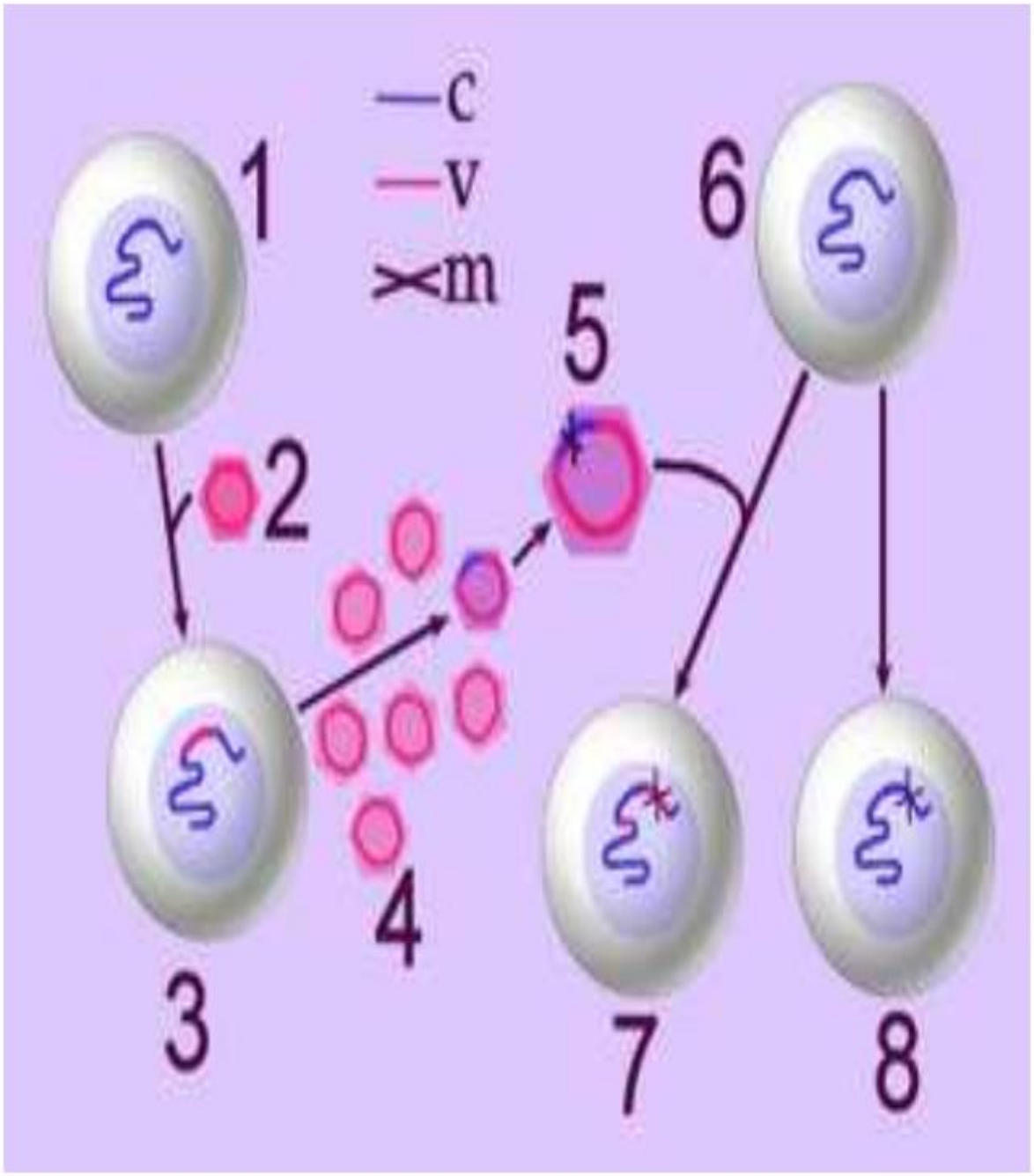


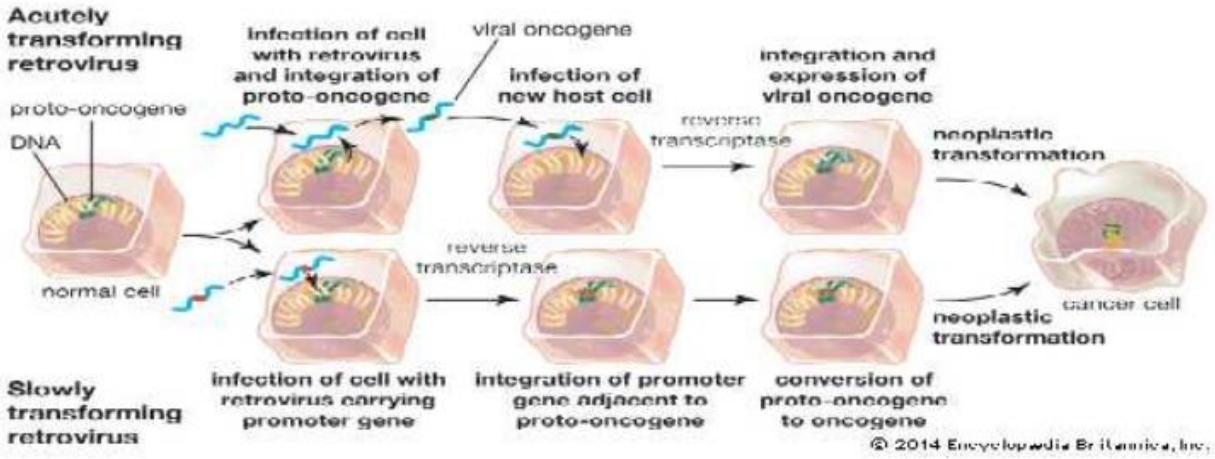


- **RETROVIRAL INFECTION AND ONCOGENESIS.**

- When a normal cell (1) is infected by a retrovirus (2), the viral reverse transcriptase reverse-transcribes the viral RNA into 'viral' DNA (v), which an integrase randomly integrates (inserts) into the host cell's genome (3-c-v).
- New viral particles are produced and shed by the infected cell (4) and some of these may contain proto-oncogene fragments of the host's genome (purple virion).
- Occasionally, the transducted sequence undergoes mutation (m) into an oncogene (5) that is subsequently integrated into the genome of a second normal cell (6), which becomes transformed into a tumorigenic line (7).

- Under the influence of other carcinogens, normal cells may suffer mutation (m) of a proto-oncogene to an oncogene (8).





- Retroviral insertion can convert a proto-oncogene, integral to the control of cell division, into an oncogene, the agent responsible for transforming a healthy cell into a cancer cell.
- **An acutely transforming retrovirus (shown at top),** which produces **tumours within weeks of infection**, **incorporates genetic material from a host cell into its own genome upon infection**, forming **a viral oncogene**. When the viral oncogene infects another cell, an **enzyme called reverse transcriptase copies the single-stranded genetic material into double-stranded DNA, which is then integrated into the cellular genome.**
- A slowly transforming retrovirus (shown at bottom), **which requires months to elicit tumour growth, does not disrupt cellular function through the insertion of a viral oncogene**. Rather, it carries a promoter gene that is integrated into the cellular genome of the host cell next to or within a proto-oncogene, allowing conversion of the proto-oncogene to an oncogene.

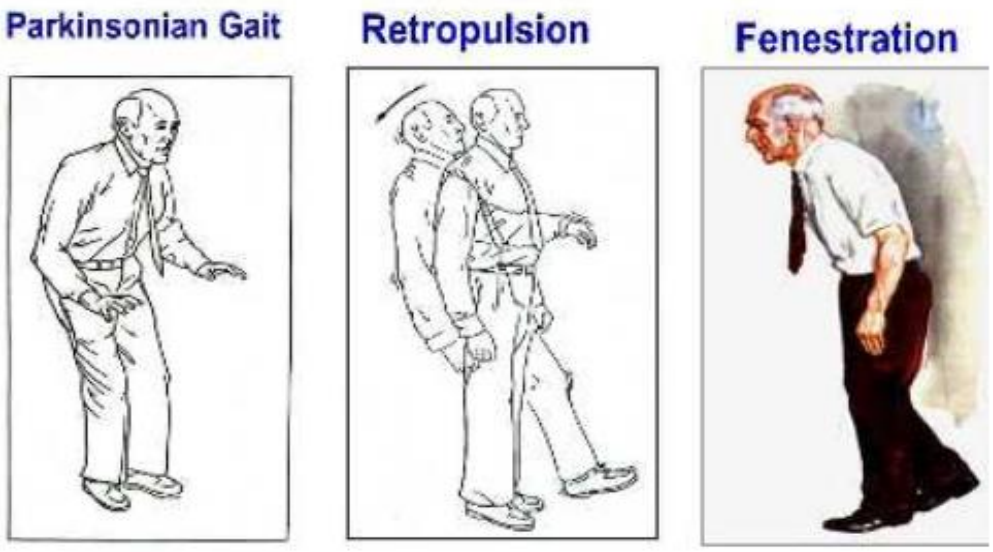
NEURODEGENERATIVE DISORDERS

PARKINSON DISEASE

- ✓ **Parkinson's disease (PD, also known as idiopathic or primary parkinsonism, hypokinetic rigid syndrome (HRS), or paralysis agitans) is a degenerative disorder of the central nervous system.**
- ✓ **Parkinson disease is a progressive disorder of the nervous system. The disorder affects several regions of the brain, especially an area called the substantia nigra that controls balance and movement.**
- ✓ **Often the first symptom of Parkinson disease is trembling or shaking (tremor) of a limb, especially when the body is at rest.**
- ✓ **Typically, the tremor begins on one side of the body, usually in one hand.**
- ✓ **Tremors can also affect the arms, legs, feet, and face.**
- ✓ ***TREMOR***
- ✓ Initial complaint ..... in some patients
- ✓ Resting Tremor ..... at Rest ..... Low - frequency movement
- ✓ Pill – rolling tremor ..... 'thumb & fore' finger ..... are involved
- ✓ Action Tremor ..... 'During activity '

LIMB - Rigidity

1. *Muscles …… Tight & Rigid*
2. *Muscle spasm … Slows Movement*
3. *Muscle ….. Aches*
4. *Posture …… Stooped*
5. *Movement ….. Slow*
6. *Walking …. Short - Shuffling Steps*

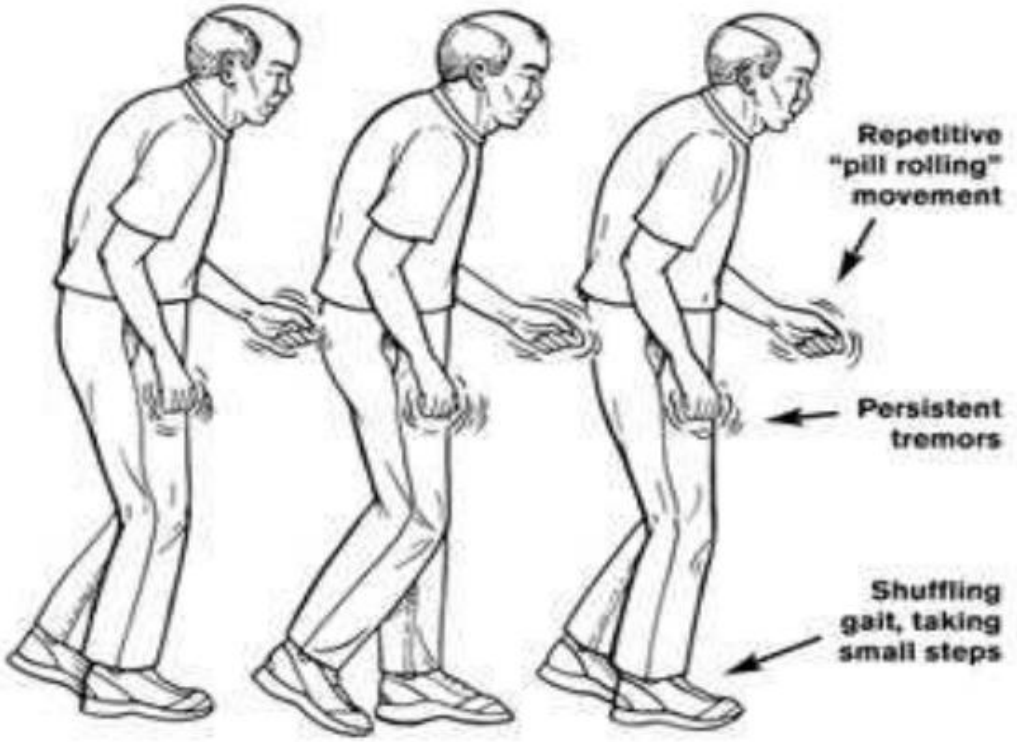


✓ Other characteristic symptoms of Parkinson disease include rigidity or stiffness of the limbs and torso, slow movement (bradykinesia) or

✓ an inability to move (akinesia).

✓ impaired balance and coordination (postural instability). These symptoms worsen slowly over time.

Gait & Postural Difficulties



Patients walk with

1. ***Stooped ... Flexed Posture***
2. ***Short ... Shuffling Stride***
3. ***Diminished ... arm swing***

- ✓ Parkinson disease can also **affect emotions and thinking ability (cognition).**
- ✓ Some affected individuals develop psychiatric conditions such as **depression and visual hallucinations**.
- ✓ People with Parkinson disease also have an increased risk of **developing dementia**, which is a decline in intellectual functions including judgment and memory.
- ✓ Generally, Parkinson disease that begins after age 50 is called **late-onset disease.**

- ✓ The condition is described as **early-onset** disease if signs and symptoms begin **before age 50.**
- ✓ Early-onset cases that begin **before age 20** are sometimes referred to as **juvenile-onset Parkinson disease**.

Classification

The term parkinsonism is used for a motor syndrome whose main symptoms are tremor at rest, stiffness, slowing of movement and postural instability.

Parkinsonian syndromes can be divided into four subtypes according to their origin:

1. Primary or idiopathic
2. Secondary or acquired
3. Hereditary parkinsonism, and
4. Parkinson plus syndromes or multiple system degeneration.

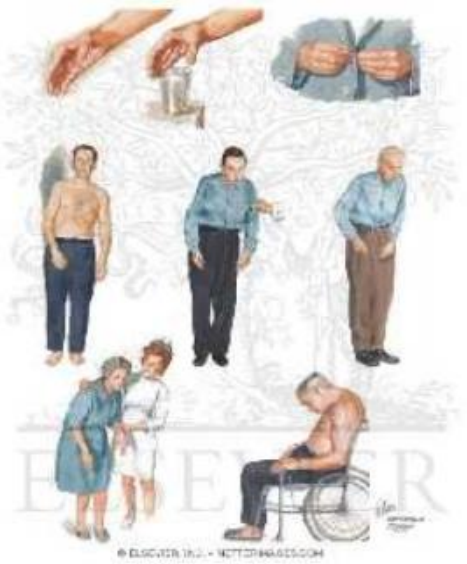
- Parkinson's disease is the most common form of parkinsonism and is usually defined as **"primary" parkinsonism**, meaning parkinsonism with **no external identifiable cause**.
- In recent years several genes that are directly related to some cases of Parkinson's disease have been discovered. As much as this conflicts with the definition of Parkinson's disease as **an idiopathic illness, genetic parkinsonism disorders with a similar clinical course to PD are generally included under the Parkinson's disease label**.
- The terms **"familial Parkinson's disease" and "sporadic Parkinson's disease"** can be used to differentiate genetic from truly idiopathic forms of the disease.

Unified - Parkinson Disease

Rating Scale

(UPDRS)

1. ***Evaluate - clinical efficacy - antiparkinson drugs***
2. ***Monitor - Disease Progression***



Stages - Parkinson Disease

STAGE	***CHARACTERISTICS***
0	***No sign …… disease***
I ***Unilateral Disease***	➢ ***Only on one side*** ➢ ***Do not cause disability*** ➢ ***Tremor of one limb***
II ***Bilateral Disease***	➢ ***Without impairment of balance*** ➢ ***Minimal disability*** ➢ ***Posture & gait have been affected***

III ***Mild to Moderate Bilateral Disease***	➢ ***Postural instability ..... physically independent*** ➢ ***Early instability in gait or posture*** ➢ ***Generalized moderate dysfunction***
IV ***Severe Disability***	➢ ***Can still walk to a limited extent*** ➢ ***Rigidity & bradykinesia*** ➢ ***No longer able to live alone***
V ***Wheelchair - bound or Bedridden***	➢ ***Cannot stand or walk*** ➢ ***Requires constant nursing care***

Diagnosis

- ***PET – Scan - Positron - Emission Tomography - visualize dopamine uptake in the substantia nigra & basal ganglia.***
- ***PET scan - measures the extent of neuronal loss.***
- ***SPECT – Single - Photon Emission – Computed - Tomography***

Causes

Parkinson's disease in most people is idiopathic (having no specific known cause). However, a small proportion of cases can be attributed to known genetic factors. Other factors have been associated with the risk of developing PD, but no causal relationships have been proven.

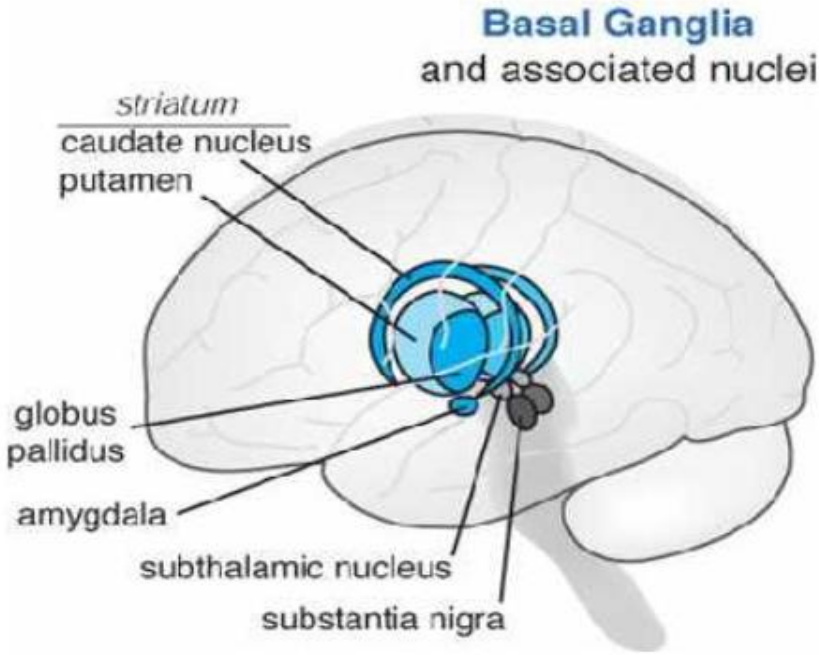
Environmental factors

- ✓ U.S. Army helicopter spraying Agent Orange (**Herbicide Orange** (HO) — is one of the herbicides and defoliants used by the U.S.) over Vietnamese agricultural land during the Vietnam war
- ✓ A number of environmental factors have been associated with an increased risk of Parkinson's including: pesticide exposure, head injuries, and living in the country or farming.
- ✓ **Rural environments and the drinking of well water** may be risks as they are indirect measures of exposure to pesticides.
- ✓ Implicated agents include insecticides, primarily chlorpyrifos and organochlorinesand pesticides, such as rotenone or paraquat, and herbicides, such as Agent Orange.
- ✓ **Heavy metals exposure has been proposed to be a risk factor**, through possible accumulation in the substantia nigra; however, studies on the issue have been inconclusive.

What genes are related to Parkinson disease?

- ✓ Approximately ***15 percent*** *of people with Parkinson disease have* ***a family history of this disorder***. Familial cases of Parkinson disease can be caused by mutations in the ***LRRK2***, ***PARK2***, ***PARK7***, ***PINK1***, **or*SNCA* gene,** or by alterations in genes that have not been identified.
- ✓ Mutations in some of these genes may also play a role in cases that appear to be sporadic (not inherited).
- ✓ Alterations in certain genes, including ***GBA*** **and** ***UCHL1*****, do not cause Parkinson disease but appear to modify the risk of developing the condition in some families**.
- ✓ It is not fully understood how genetic changes cause Parkinson disease or influence the risk of developing the disorder.
- ✓ Many Parkinson disease symptoms occur when nerve cells (neurons) in **the substantia nigra die or become impaired**.

- ✓ Normally, these cells **produce a chemical messenger called dopamine,** which transmits signals within the brain to produce smooth physical movements.
- ✓ When these dopamine-producing neurons are damaged or die, communication between the brain and muscles weakens. Eventually, the brain becomes unable to control muscle movement.



- ✓ Some gene mutations appear to **disturb the cell machinery that breaks down (degrades) unwanted proteins in dopamine-producing neurons.**
- ✓ As a result, **undegraded proteins accumulate, leading to the impairment or death of these cells.**
- ✓ Other mutations **may affect the function of mitochondria,** the energy-producing structures within cells.
- ✓ As a byproduct of energy production, mitochondria **make unstable molecules called free radicals that can damage cells.**
- ✓ Cells normally counteract the effects of free radicals before they cause damage, but mutations can disrupt this process. As a result, **free radicals may accumulate and impair or kill dopamine-producing neurons**

- ✓ In most cases of Parkinson disease, protein deposits called **Lewy bodies** appear in dead or dying dopamine-producing neurons.
- ✓ (When Lewy bodies are not present, the condition is sometimes referred to as parkinsonism)
- ✓ It is **unclear whether Lewy bodies play a role in killing nerve cells** or if they are part of the cells' response to the disease.

How are changes in the *LRRK2(*"leucine-rich repeat kinase 2.") gene related to PD

What is the normal function of the *LRRK2* gene?

- ✓ The *LRRK2* gene provides instructions for making a protein called **dardarin.** The *LRRK2* gene is active in the brain and other tissues throughout the body.
- ✓ One segment of the **dardarin protein is called a leucine-rich region** because it contains a large amount of a protein building block (amino acid) known as **leucine**.
- ✓ Proteins with **leucine-rich regions appear to play a role in activities that require interactions with other proteins, such as transmitting signals or helping to assemble the cell's structural framework (cytoskeleton)**. Other parts of the dardarin protein are also thought to be involved **in protein-protein interactions**.
- ✓ Additional studies indicate that **dardarin has an enzyme function known as kinase activity.**
- ✓ Proteins with kinase activity assist in the transfer of a phosphate group (a cluster of oxygen and phosphorus atoms) from the energy molecule ATP to amino acids in certain proteins. This phosphate transfer is called **phosphorylation**, and it is an essential step in turning on and off many cell activities.

- ✓ Dardarin also has **a second enzyme function referred to as a GTPase activity**. This activity is associated with a region of the protein called the **ROC domain**. The **ROC domain may help control the overall shape of the dardarin protein.**

- ✓ **How are changes in the *LRRK2* gene related to PD**

- ✓ Researchers have identified more than **100 *LRRK2*** gene mutations in families with **late-onset Parkinson disease** (the most common form of the disorder, which appears after age 50).

- ✓ These mutations replace single **amino acids in the dardarin protein**, which affects the protein's structure and function.

- ✓ It is unclear how *LRRK2* gene mutations lead to the movement and balance problems characteristic of Parkinson disease.

- ✓ A mutation that replaces the amino acid arginine with the amino acid glycine at protein position **1441 (written as Arg1441Gly or R1441G)** is a relatively common cause of Parkinson disease in the Basque region between **France and Spain.**

- ✓ The protein name dardarin comes from the Basque word "dardara," which means tremor, a characteristic feature of Parkinson disease.

- ✓ Studies of several different populations from around the world revealed a common ***LRRK2* gene mutation in 3 to 7 percent of familial Parkinson disease cases**.

- ✓ This mutation replaces the amino acid glycine with the amino acid serine at protein position **2019 (written as Gly2019Ser or G2019S).**

- ✓ The incidence of the Gly2019Ser mutation in familial cases is highest among Arabs from North Africa and people of Ashkenazi (eastern and central European) Jewish ancestry, and it is lowest in Asian and northern European populations.

- ✓ This particular mutation has also been reported in 1 to **3 percent of sporadic Parkinson disease cases**, in which there is no family history of the disease.

- ✓ Studies in Chinese and Japanese populations have identified an *LRRK2* gene mutation that occurs more frequently in people with Parkinson disease than in people without the disease.

This mutation replaces the amino acid glycine with the amino acid **arginine at protein position 2385 (written as Gly2385Arg or G2385R).** This mutation appears to increase the risk of Parkinson disease among people in these populations

How are changes in the *PARK2* gene related to PD

What is the normal function of the *PARK2* gene?("parkin RBR E3 ubiquitin protein ligase.")

- ✓ The *PARK2* gene, one of the largest human genes, provides instructions for making a protein called **parkin**.
- ✓ Parkin plays a role in the cell machinery that **breaks down (degrades) unneeded proteins** damaged and excess proteins by **tagging** with molecules called **ubiquitin**.
- ✓ Ubiquitin serves as a **signal to move unneeded proteins into specialized cell structures known as proteasomes**, where the proteins are degraded.
- ✓ The ubiquitin-proteasome system acts as the cell's quality control system by disposing of damaged, misshapen, and excess proteins.
- ✓ This system also regulates the availability of proteins that are involved in several critical cell activities, such as the **timing of cell division and growth.**
- ✓ **Because of its activity in the ubiquitin-proteasome system**, parkin belongs to a group of proteins called **E3 ubiquitin ligases.**
- ✓ Parkin appears to be involved in the maintenance of mitochondria, the energy-producing centers in cells. However, little is known about its role in mitochondrial function. **Research suggests that parkin may**

help trigger the destruction of mitochondria that are not working properly.

- ✓ Studies of the **structure and activity of parkin have led researchers to propose several additional activities for this protein**.
- ✓ Parkin may act as a **tumor suppressor protein**, which means it prevents **cells from growing and dividing too rapidly or in an uncontrolled way**.
- ✓ Parkin may also **regulate the supply and release of sacs called synaptic vesicles from nerve cells**.
- ✓ **Synaptic vesicles** contain chemical messengers that transmit signals from one nerve cell to another.

- ✓ **How are changes in the *PARK2* gene related to PD**

- ✓ Researchers have identified more than **200 *PARK2* gene mutations that cause Parkinson disease,** a condition characterized by progressive problems with movement and balance.
- ✓ Mutations in this gene are associated with the **juvenile form of Parkinson disease, which appears before age 20,** and some cases of the more common, **late-onset form that begins after age 50**.
- ✓ Some ***PARK2* gene mutations lead to an abnormally small parkin protein that is nonfunctional and is rapidly broken down (degraded) within cells**.
- ✓ Other mutations insert, **delete, or change DNA building blocks (nucleotides) in the *PARK2* gene,** leading to a **defective version of the parkin protein or preventing the production of this protein**.
- ✓ The *PARK2* gene mutations associated with Parkinson disease usually lead to a **loss of parkin activity.**
- ✓ It is unclear how *PARK2* gene mutations cause Parkinson disease.
- ✓ The loss of parkin activity probably disturbs the ubiquitin-proteasome system, which **allows unneeded proteins to accumulate**.

- ✓ A buildup of these proteins could disrupt normal cell activities such as the **supply and release of synaptic vesicles, particularly those that contain a chemical messenger called dopamine**.
- ✓ As parkin is **normally abundant in the brain**, its loss could lead to the impairment or death of nerve cells, including those that produce dopamine.
- ✓ Loss of dopamine-producing nerve cells is a characteristic feature of Parkinson disease.
- ✓ Mutations in the *PARK2* gene may **also disrupt the regulation of mitochondria.** Researchers speculate that mitochondrial dysfunction in dopamine-producing nerve cells may play an important role in causing the signs and symptoms of Parkinson disease.
- ✓ **How are changes in the *PARK7* gene related to PD? "parkinson protein 7."**

What is the normal function of the *PARK7* gene?

- ✓ The *PARK7* gene provides instructions for making the DJ-1 protein.
- ✓ This protein is found in many tissues and organs, including the brain.
- ✓ Studies indicate that the DJ-1 protein has several functions, although none are fully understood.
- ✓ One of the protein's functions may be to **help protect cells, particularly brain cells, from oxidative stress.**
- ✓ Oxidative stress occurs when unstable **molecules called free radicals accumulate to levels that can damage or kill cells.**
- ✓ Additionally, the **DJ-1 protein may serve as a chaperone molecule t**hat helps fold **newly produced proteins into the proper 3-dimensional shape** and **helps refold damaged proteins**.
- ✓ Like other chaperone molecules, the DJ-1 protein may assist in delivering selected proteins to proteasomes, which are structures within cells that break down unneeded molecules.

- ✓ Researchers suggest that the DJ-1 protein may also play a role in activities that produce and process RNA, a chemical cousin of DNA.

- ✓ **How are changes in the *PARK7* gene related to PD?**

- ✓ Researchers have identified **more than 25 *PARK7* gene** mutations that can cause Parkinson disease, a condition characterized by progressive problems with movement and balance.

- ✓ These mutations are associated with the **early-onset form of the disorder, which begins before age 50**.

- ✓ Some *PARK7*gene mutations lead to **an abnormally small DJ-1 protein or change the building blocks** (amino acids) used to make the protein.

- ✓ The altered protein is unstable and does not function properly, if at all.

- ✓ Other mutations delete a large portion of the *PARK7* gene, preventing the production of any functional DJ-1 protein.

- ✓ It is unclear how loss of functional DJ-1 protein leads to Parkinson disease.

- ✓ Some studies suggest that *PARK7* gene mutations disrupt the **protein's chaperone function**, which leads to a **toxic buildup of misfolded** or **damaged proteins** and eventually to cell death.

- ✓ Another possibility is that *PARK7*gene mutations **impair the protein's ability to protect cells from destructive oxidative stress**.

- ✓ Nerve cells that make the chemical messenger **dopamine are particularly vulnerable to oxidative stress.** With diminished protection, **free radicals may cause enough damage to kill these nerve cells.** Progressive loss of dopamine-producing nerve cells is a characteristic feature of Parkinson disease. The death of these cells weakens communication between the brain and muscles, and ultimately the brain becomes unable to control muscle movement.

- ✓ **How are changes in the *PINK1(*** "PTEN induced putative kinase 1." **) gene related to PD**

✓ What is the normal function of the *PINK1* gene?

- ✓ The *PINK1* gene provides instructions for making a protein called **PTEN induced putative kinase 1.** This protein is found in cells throughout the body, with highest levels in the heart, muscles, and testes.
- ✓ Within cells, the **protein is located in the mitochondria,** the energy-producing centers that provide power for cellular activities. The function of PTEN induced putative kinase 1 is not fully understood. It appears to help protect mitochondria from malfunctioning during periods of cellular stress, such as unusually high energy demands.
- ✓ Researchers believe that two specialized regions of PTEN induced putative kinase 1 are essential for the **protein to function properly**.
- ✓ One region, called the mitochondrial-targeting motif, serves as a delivery address: after the protein is made, this motif helps ensure that it is delivered to the mitochondria.
- ✓ Another region, called the **kinase domain,** probably carries out the **protein's protective function.**

How are changes in the *PINK1 (* "PTEN induced putative kinase 1.")

gene related to PD

- ✓ Researchers have identified more than **70 mutations in the *PINK1* gene** that can cause Parkinson disease, a condition characterized by progressive problems with movement and balance. *PINK1*gene mutations are **associated with the early-onset form of the disorder, which typically begins before age 50.**
- ✓ Many *PINK1* **gene mutations alter or eliminate the kinase domain**, leading to a loss of protein function. At least one **mutation affects the mitochondrial-targeting motif** and may disrupt delivery of the **protein to mitochondria**.
- ✓ With **reduced or absent PTEN induced putative kinase 1 activity**, mitochondria may malfunction, particularly when cells are stressed.

- ✓ Cells can die if energy is not provided for essential activities. It is unclear how ***PINK1*** **gene mutations cause the selective death of nerve cells that characterizes Parkinson disease.**
- ✓ The loss of these cells weakens communication between the brain and muscles, and ultimately the brain becomes unable to control muscle movement.
- ✓ **How are changes in the *SNCA* gene related to PD**
- ✓ **What is the normal function of the *SNCA* gene?(** "synuclein, alpha (non A4 component of amyloid precursor)."
- ✓ The *SNCA* gene provides instructions for making a **small protein called alpha-synuclein**.
- ✓ **Alpha-synuclein is abundant in the brain**, and smaller amounts are found in the heart, muscles, and other tissues. In the brain, alpha-synuclein is found mainly at the tips of nerve cells (neurons) in specialized structures called **presynaptic terminals.**
- ✓ Within these structures, **alpha-synuclein interacts with fats (lipids) and proteins**.
- ✓ Presynaptic terminals release **chemical messengers, *called neurotransmitters***, from compartments known as **synaptic vesicles**. The release of neurotransmitters relays signals between neurons and is critical for normal brain function.
- ✓ Although the function of **alpha-synuclein** is not well understood, **studies suggest that it plays an important role in maintaining a supply of synaptic vesicles in presynaptic terminals**. It may also help **regulate the release of dopamine**, a type of neurotransmitter that is critical for controlling the start and stop of voluntary and involuntary movements.
- ✓ **How are changes in the *SNCA* gene related to PD**
- ✓ At least **18 mutations in the *SNCA*** gene have been found to cause Parkinson disease, a condition characterized by progressive problems with movement and balance.

- ✓ ***SNCA* gene mutations are associated with the early-onset form of the disorder, which typically appears before age 50.**
- ✓ Researchers have described **two types of alterations of the *SNCA* gene** in people with Parkinson disease.
- ✓ One type **changes a single protein building block (amino acid) used to make alpha-synuclein.** In some cases, the amino acid **alanine is replaced with the amino acid threonine at protein position 53** (written as Ala53Thr) or with the amino acid proline at position 30 (written as Ala30Pro).
- ✓ These mutations cause the alpha-synuclein **protein to take on an incorrect 3-dimensional shape (misfold**).
- ✓ In the other type of alteration, one of the two ***SNCA* genes in each cell is inappropriately duplicated or triplicated.** The extra copies of the ***SNCA* gene lead to an excess of alpha-synuclein.**
- ✓ It is unclear how alterations in the *SNCA* gene cause Parkinson disease. This **condition involves the selective death or impairment of neurons that produce dopamine**.
- ✓ Misfolded or excess alpha-synuclein proteins may cluster together (aggregate) and impair the function of these neurons in specific regions of the brain.
- ✓ Aggregated alpha-synuclein may disrupt the regulation of dopamine, which allows dopamine to accumulate to toxic levels and eventually kill neurons.
- ✓ Researchers also suspect that misfolded or excess alpha-synuclein stalls or shuts down the cell machinery that removes unneeded proteins.
- ✓ As a result, **unneeded proteins may clog neurons and impair their functions**.
- ✓ Symptoms of Parkinson disease appear when dopamine-producing neurons become impaired or die. The loss of these cells weakens

communication between the brain and muscles, and ultimately the brain becomes unable to control muscle movement.

- ✓ Misfolded alpha-synuclein **is also a major component of Lewy bodies**, abnormal deposits that appear in certain neurons in the brain in people with Parkinson disease.
- ✓ The presence of **Lewy bodies in a region of the brain called the substantia nigra**, which controls balance and movement, are a characteristic feature of Parkinson disease.
- ✓ However, it is unclear whether Lewy bodies play a role in killing nerve cells or if they are part of the cells' response to the disease.
- ✓ **How are changes in the *GBA*(**"glucosidase, beta, acid.") **gene RELATED PD?**
- ✓ **What is the normal function of the *GBA* gene?**
- ✓ The *GBA* gene provides instructions for making an enzyme called **beta-glucocerebrosidase.**
- ✓ This enzyme is **active in lysosomes**, which are structures inside cells that act as recycling centers.
- ✓ Lysosomes use digestive enzymes to **break down toxic substances, digest bacteria that invade the cell, and recycle worn-out cell components.**
- ✓ Based on these functions, enzymes in the lysosome are sometimes called housekeeping enzymes.
- ✓ Beta-glucocerebrosidase is a **housekeeping** enzyme that helps break down a large molecule called glucocerebroside into a sugar (glucose) and a simpler fat molecule (ceramide).
- ✓ Changes in the *GBA* gene are also associated with Parkinson disease and parkinsonism, which are similar disorders that affect movement and balance.

- ✓ People with Gaucher disease have mutations in both copies of the *GBA* gene in each cell, while those with a mutation in just one copy of the gene are called carriers.
- ✓ People with Gaucher disease and people who are carriers of a *GBA* gene mutation have an increased risk of developing Parkinson disease or parkinsonism.
- ✓ Symptoms of Parkinson disease and parkinsonism result from the loss of nerve cells that produce dopamine.
- ✓ Dopamine is a chemical messenger that transmits signals within the brain to produce smooth physical movements.
- ✓ It remains unclear how *GBA* gene mutations are related to these disorders.
- ✓ Studies suggest that changes in this gene **may contribute to the faulty breakdown of toxic substances in nerve cells by impairing the function of lysosomes**.
- ✓ Alternatively, **the changes may increase the formation of abnormal protein deposits. As a result, toxic substances or protein deposits could accumulate and kill dopamine-producing nerve cells**, leading to abnormal movements and balance problems.
- ✓ **How are changes in the *UCHL1(*"ubiquitin carboxyl-terminal esterase L1 (ubiquitin thiolesterase).") gene related to PD**
- ✓ **What is the normal function of the *UCHL1* gene?**
- ✓ The *UCHL1* gene provides instructions for making an enzyme called **ubiquitin carboxyl-terminal esterase L1**.
- ✓ This enzyme is **found in nerve cells throughout the brain**. Ubiquitin carboxyl-terminal esterase L1 is **probably involved in the cell machinery that breaks down (degrades) unneeded proteins.** In cells, damaged or excess proteins are **tagged with molecules called ubiquitin.**
- ✓ **Ubiquitin serves as a signal to move these unneeded proteins into specialized structures known as proteasomes**, where the proteins are

degraded. The ubiquitin-proteasome system acts as the cell's quality control system by disposing of damaged, misshapen, and excess proteins.

- ✓ Although the **exact function of ubiquitin carboxyl-terminal esterase L1 is not fully understood, it appears to have two types of enzyme activity**.
- ✓ One of these, called **hydrolase activity,** removes and recycles ubiquitin molecules from degraded proteins. This recycling step is important to sustain the degradation process.
- ✓ The other enzyme function**, known as ligase activity**, **links together ubiquitin molecules for use in tagging proteins for disposal.**
- ✓ A relatively **common variation (polymorphism) in the *UCHL1* gene** may **reduce the risk** of developing Parkinson disease, a condition characterized by progressive problems with movement and balance. This change is most common in Chinese and Japanese populations and occurs **less frequently in European populations.**
- ✓ The polymorphism **reduces the ligase activity of ubiquitin carboxyl-terminal esterase L1 but has little effect on the hydrolase** activity.
- ✓ Some studies suggest that having the **S18Y polymorphism** may **help protect against Parkinson disease,** particularly in young adults. However, other studies have not shown this effect. It remains unclear how this amino acid variation might reduce the risk of developing Parkinson disease.
- ✓ This change is most common in Chinese and Japanese populations and occurs **less frequently in European populations.**

- ✓ The polymorphism **reduces the ligase activity of ubiquitin carboxyl-terminal esterase L1 but has little effect on the hydrolase** activity.
- ✓ Some studies suggest that having the **S18Y polymorphism** may **help protect against Parkinson disease,** particularly in young adults. However, other studies have not shown this effect. It remains unclear how this amino acid variation might reduce the risk of developing Parkinson disease.

- ✓ The variation leads to a change in one of the building blocks (amino acids) used to make **ubiquitin carboxyl-terminal esterase L1**. Instead of **serine at position 18 in the enzyme's chain of amino acids,** people with the polymorphism have the amino acid tyrosine (written as Ser18Tyr or S18Y).
- ✓ A different change in the *UCHL1* gene may increase the risk of Parkinson disease.
- ✓ This mutation has been reported in two siblings with the disease.
- ✓ The mutation replaces the amino acid isoleucine with the amino acid methionine at position 93 in ubiquitin carboxyl-terminal esterase L1 **(written as Ile93Met or I93M).**
- ✓ The mutation leads to **decreased hydrolase activity, which may disrupt the ubiquitin-proteasome system.** Instead of **being degraded, unneeded proteins could accumulate to toxic levels that impair or kill nerve cells in the brain**.
- ✓ The loss of these cells weakens communication between the brain and muscles, and ultimately the brain becomes unable to control muscle movement.
- ✓ It is unclear whether this *UCHL1* gene mutation is a true risk factor for Parkinson disease, because it has been identified in only one family.

✓ Other genes reported

1. **"ATPase type 13A2."**
2. **"vacuolar protein sorting 35 homolog (S. cerevisiae)."**

✓ ***ATP13A2* gene**

- ✓ This gene encodes a member of the P5 subfamily of ATPases which transports inorganic cations as well as other substrates.
- ✓ Mutations in this gene are associated **with Kufor-Rakeb syndrome (KRS), also referred to as Parkinson disease 9**

VPS35 gene

- Gene belongs to a group of vacuolar protein sorting (VPS) genes. The encoded protein is a component of a large multimeric complex, termed the **retromer complex,** involved in retrograde **transport of proteins from endosomes to the trans-Golgi network**.
- The close **structural similarity between the yeast and human proteins that make up this complex suggests a similarity in function**.
- Expression studies in yeast and mammalian cells indicate that this **protein interacts directly with VPS35,** which serves as the **core of the retromer complex**