# Viral Carcinogenesis – Molecular Basis of Cancer
# (MBChB III - 21May2019)

Dufton Mwaengo, PhD
Dept Medical Microbiology
University of Nairobi

# Major Types of Cancer

**Carcinoma**

- Origins: skin, lungs, breasts, pancreas, other organs/glands

**Sarcoma**

- Arise in bone, muscle, fat, or cartilage

– Rare

**Lymphoma**

- Cancer of lymphoctyes

**Leukemia**

- Cancer of the blood

# Definitions

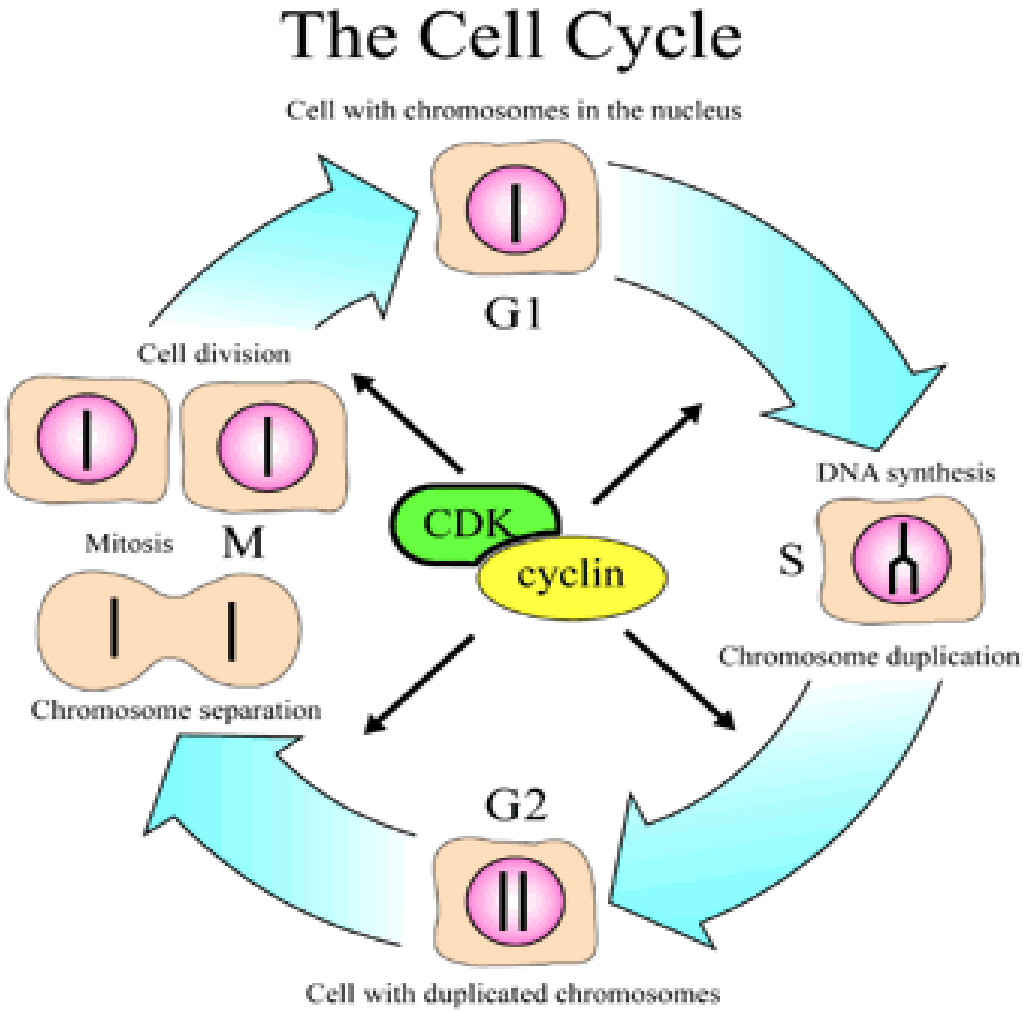
- Primary cancers
  – Cancers originating from any organ/tissue
  - Spread -> metastatic
- Most cancers form tumors, but not all tumors are cancerous.
- Tumor
  - Mass/cluster of abnormal cells

  **Benign** (non cancerous) tumors
  – Stop growing (hence no new tumors)

  **Malignant** (cancerous) tumors
  – Tumors crowd out healthy cells (interfere with body functions)
  - Draw nutrients from body tissues

- All cancer cells have one thing in common: A loss of cell cycle control

# Cell cycle




- M- mitosis
- G1 - cells grow
- S - DNA synthesis
- G2 - growth and preparation for mitosis
- G1/S decision point for going to dividing state

# Loss of Cell Cycle Regulation

A. The Cell Cycle Control system:

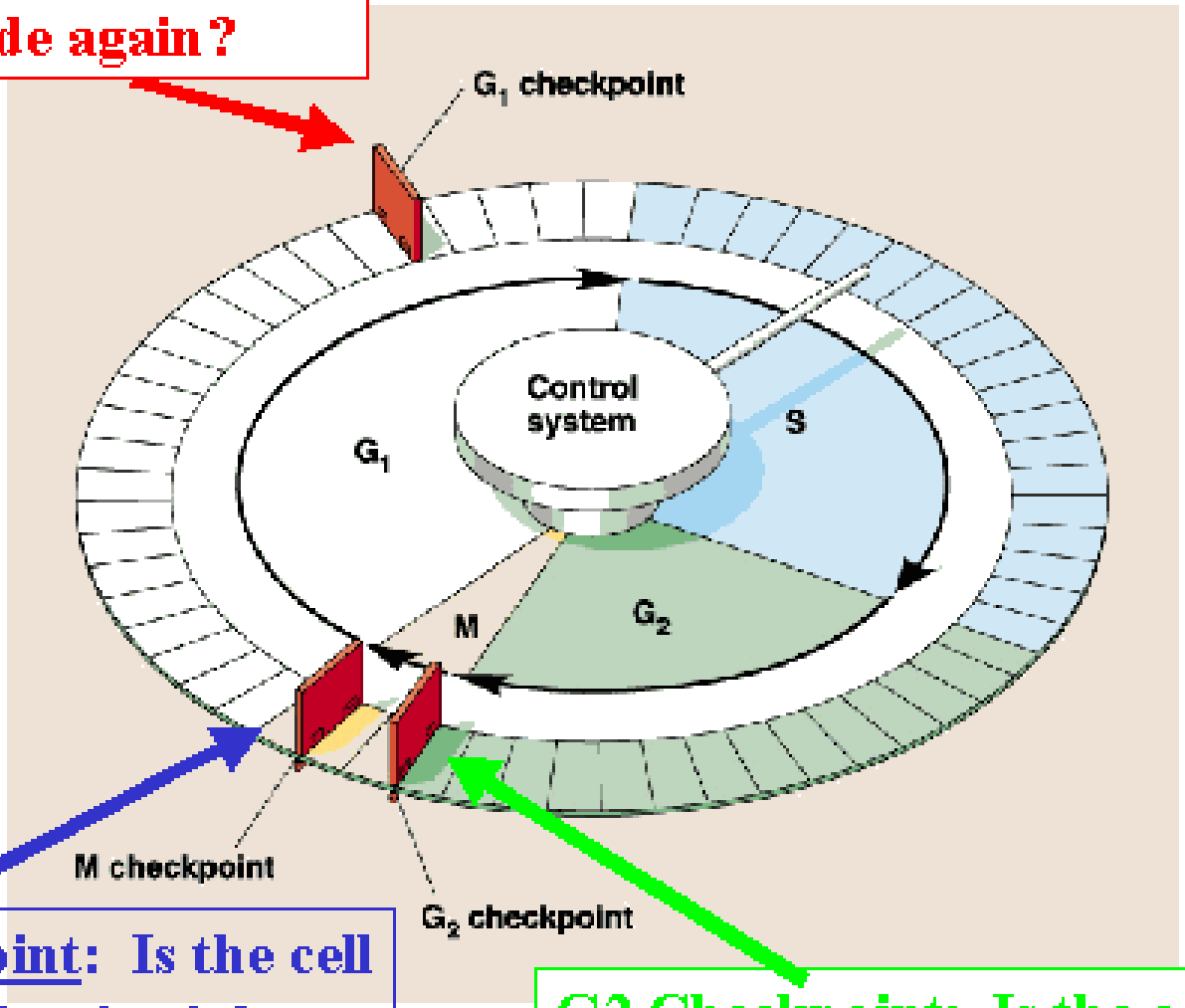
**Checkpoints** during G1, G2, and M phases

B. Checkpoint signals: report cells status -

a. Is the cell big enough?
b. Is environment favorable?
c. Is DNA damaged?
d. Is DNA replicated?
e. Are chromosomes attached to opposite poles?

Cell cycle and cancer

**G1 Checkpoint: Is the cell ready to divide again?**




**M Checkpoint: Is the cell ready to exit mitosis?**

**G2 Checkpoint: Is the cell ready to enter mitosis?**

# Cancer and Genes

**Two types of genes control cell cycling/division**

1. **Proto-oncogenes** (cellular)
   - c-onc (e.g. c-myc)
   - Promote cell proliferation/division only at appropriate times
   - **Oncogenes** – cancer-causing genes (v-onc)
   - Promote cell division continuously
2. **Tumor suppressor genes** (anti-oncogenes)
   - Repress cell division.

# 1. PROTO-ONCOGENES

# Examples of proto-oncogene (c-onc) proteins

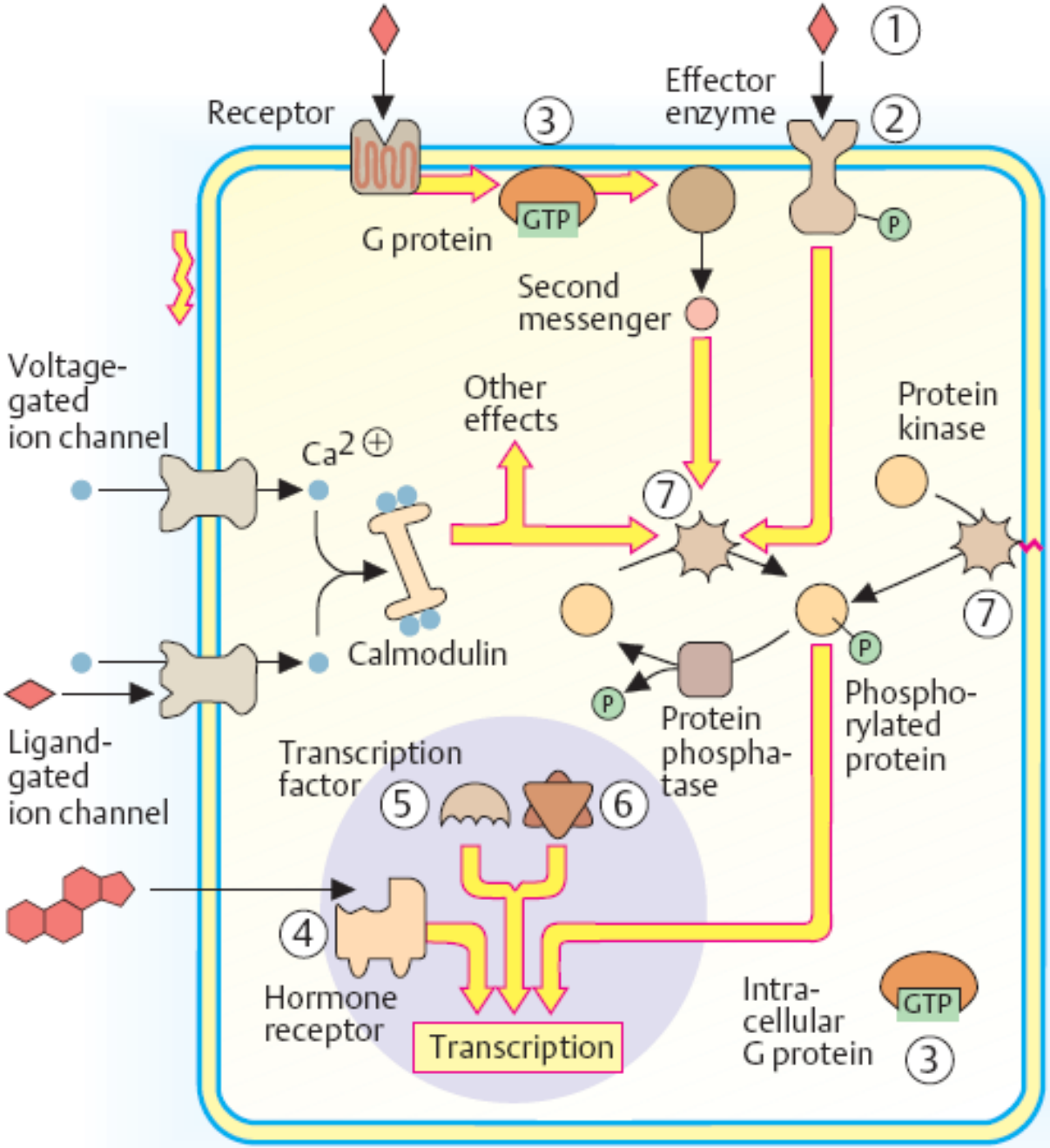
**Class I: Growth Factors**

**Class II: Receptors for Growth Factors and Hormones**

**Class III: Intracellular Signal Transducers**

**Class IV: Nuclear Transcription Factors**

**Class V: Cell-Cycle Control Proteins**




Oncogene products
(examples)

Gene

① Ligands
*sis, hst, int-2, wnt-1*

② Receptors
*fms, trk, trkB, ros, kit, mas, neu, erbB*

③ GTP-binding proteins
*Ha-ras, Ki-ras, N-ras*

④ Nuclear hormone receptors
*erbA, NGF1-B*

⑤ Nuclear tumor suppressors
*Rb, p53, wt1, DCC, APC*

⑥ DNA-binding proteins
*jun, fos, myc, N-myc, myb, fra1, egr-1, rel*

⑦ Protein kinases
*src, yes, fps, abl, met, mos, raf*

# Mechanisms of oncogene Activation

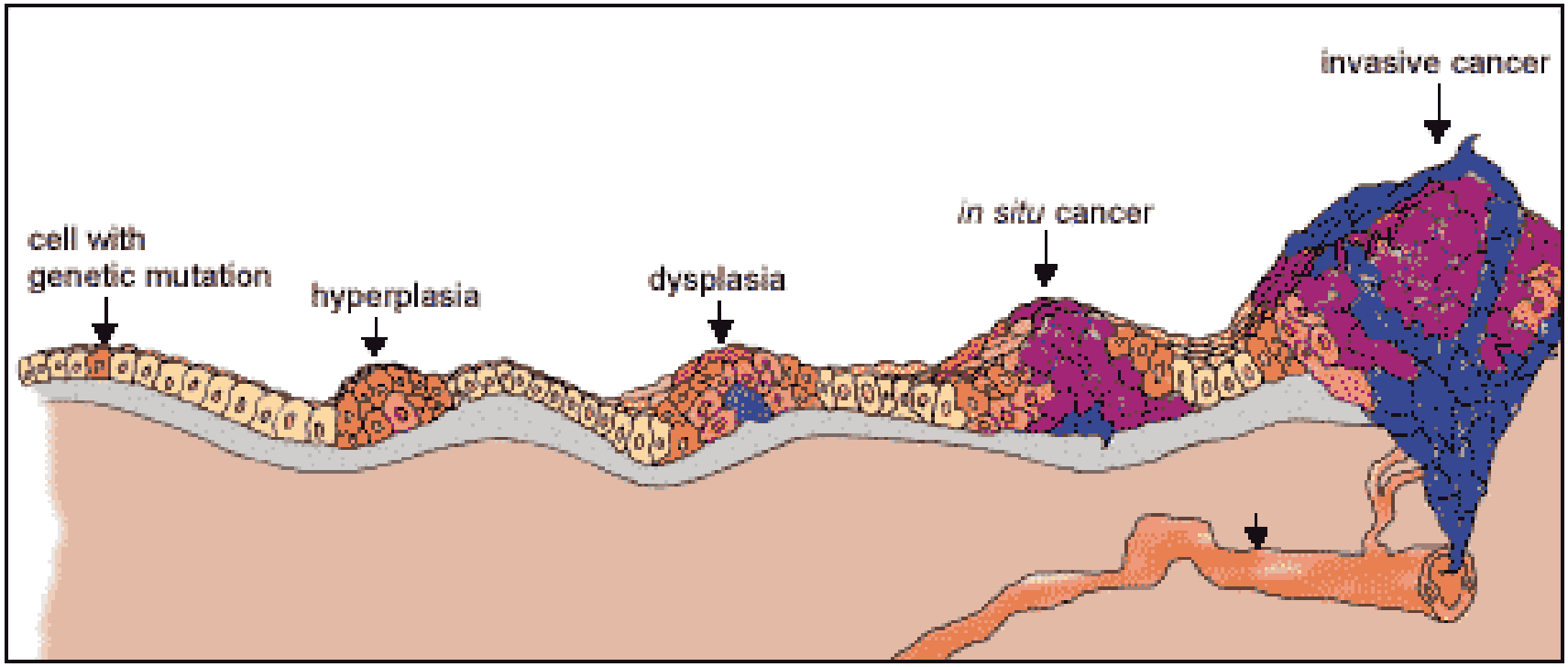
1. Obtaining a strong promoter or enhancer
2. Group translocation or chromosome rearrangements
3. Proto-oncogene amplification
4. Gene mutation

# Mutations that cause cancer

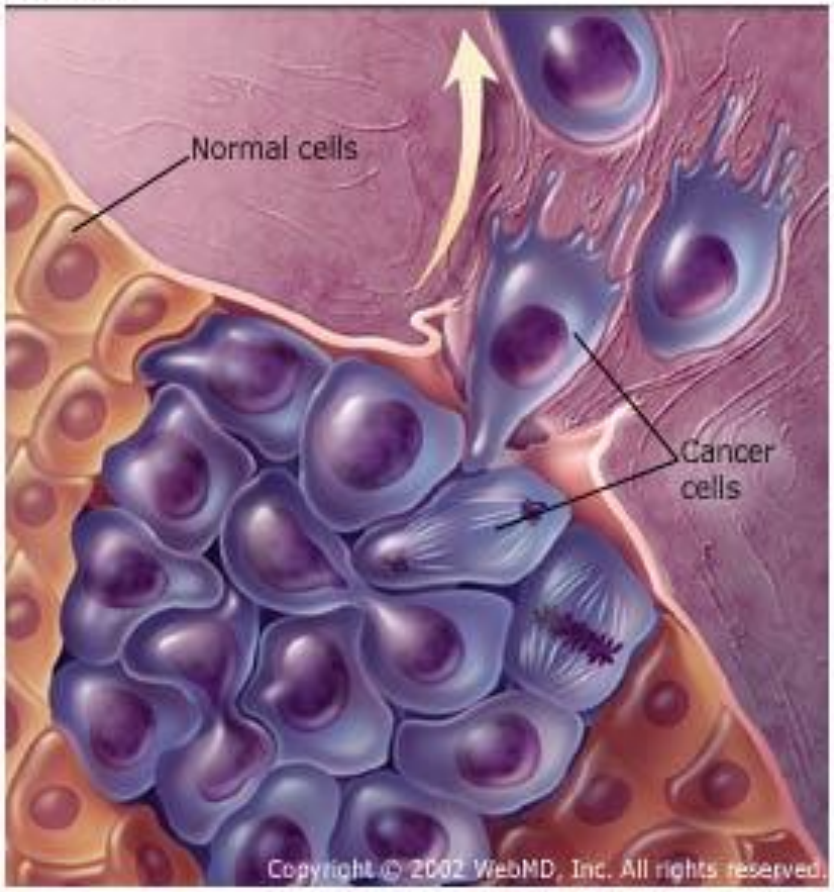
1. Increased activity of a gene whose protein causes cells to divide

2. Decreased activity of a gene whose protein blocks cell division

# Genetic mutation (c-onc)



Cancer


Normal cells
Cancer cells
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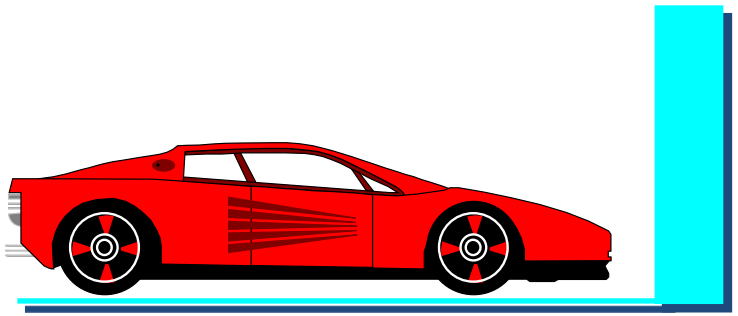
# 2. TUMOR SUPPRESSOR GENES (ANTI-ONCOGENES)

# Tumor suppressor gene (anti-oncogene)

1. Protein product -> inhibits cell division (hence prevents uncontrolled cell growth).
2. Induced occurrence of tumors when gene
   a. Deleted
   b. Mutated
3. Examples
   a. Rb gene
   b. P53 gene.

# Tumour suppressor genes

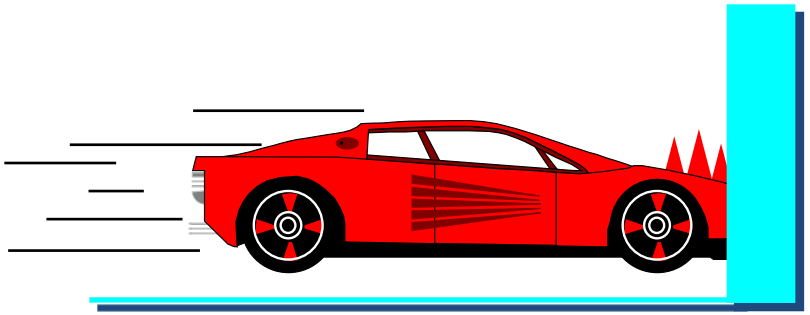
Act as a brake for cell division



**“Guardian of the genome”**

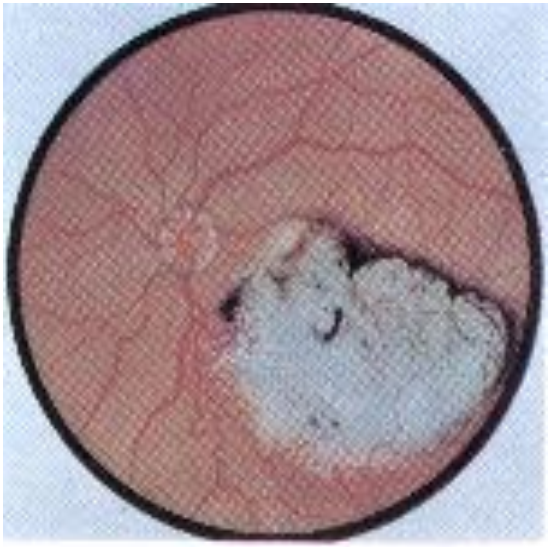
**PROBLEM:**

**Mutation in tumour suppressor genes = brakes don’t work, or there is an accumulations of mutations (DNA repair enzymes)**

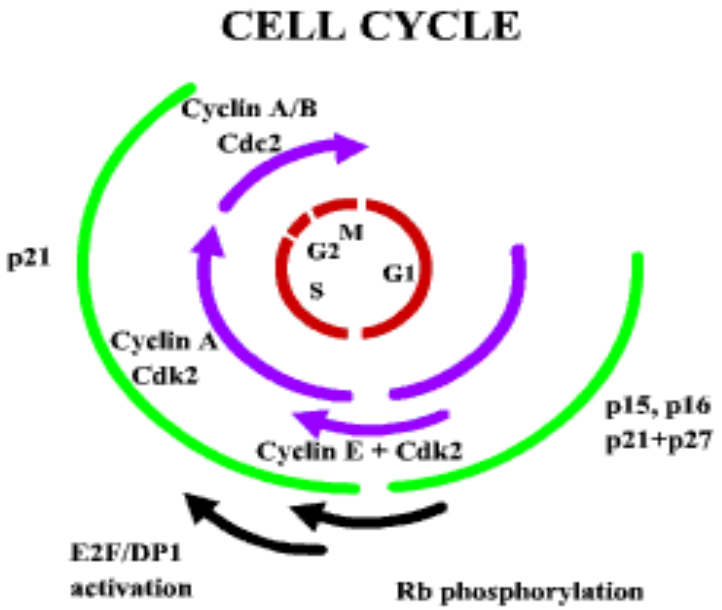


# Retinoblastoma (*Rb* gene)

- Diagnosis: "Cat's eye" reflection in affected eye.
- Common cancer of infants and children
- Individuals at greater risk of developing other cancers.



# Tumor suppressor Rb




- Rb binds to transcription factor E2F and prevents gene expression of proteins needed to go to S phase

# Mechanism of action of *Rb* gene

$G_0\,G_1$ phase

Rb protein

E-2F

**Rb = product of Retinoblastoma gene, inhibits action of E2F until chemically modified**

**E2F = transcription factor**

DNA

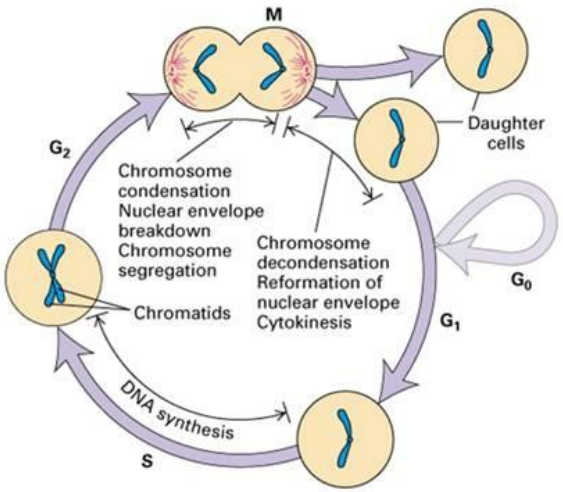
S phase

Rb protein — P

E-2F

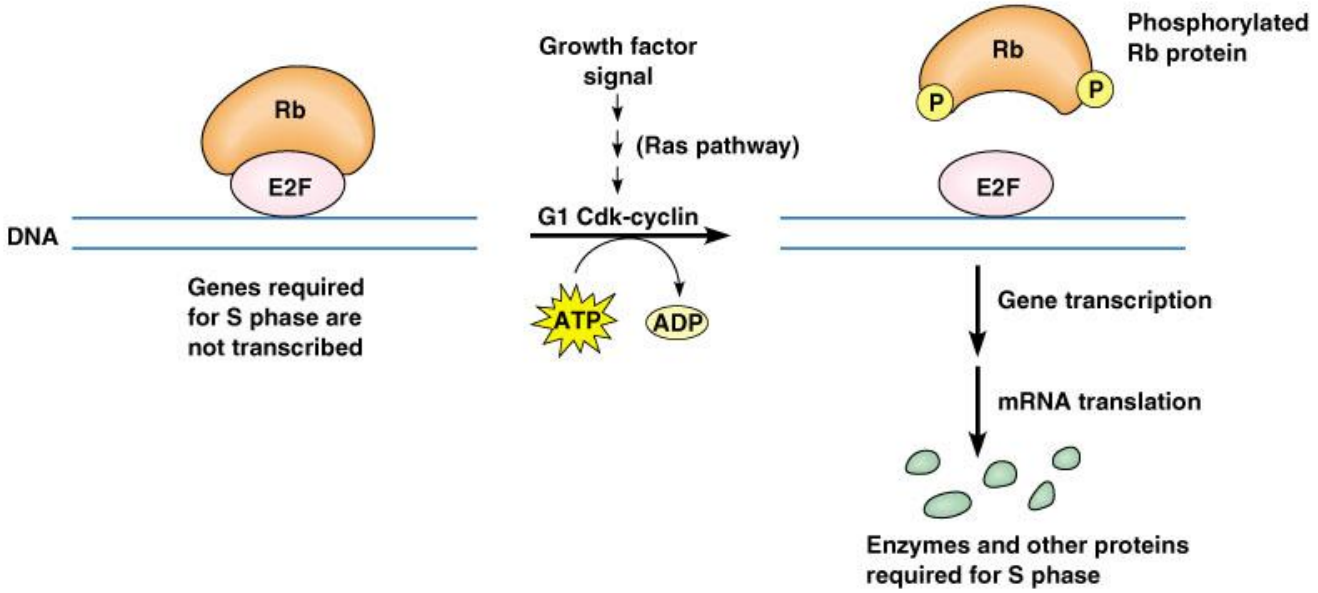
DNA

mRNA

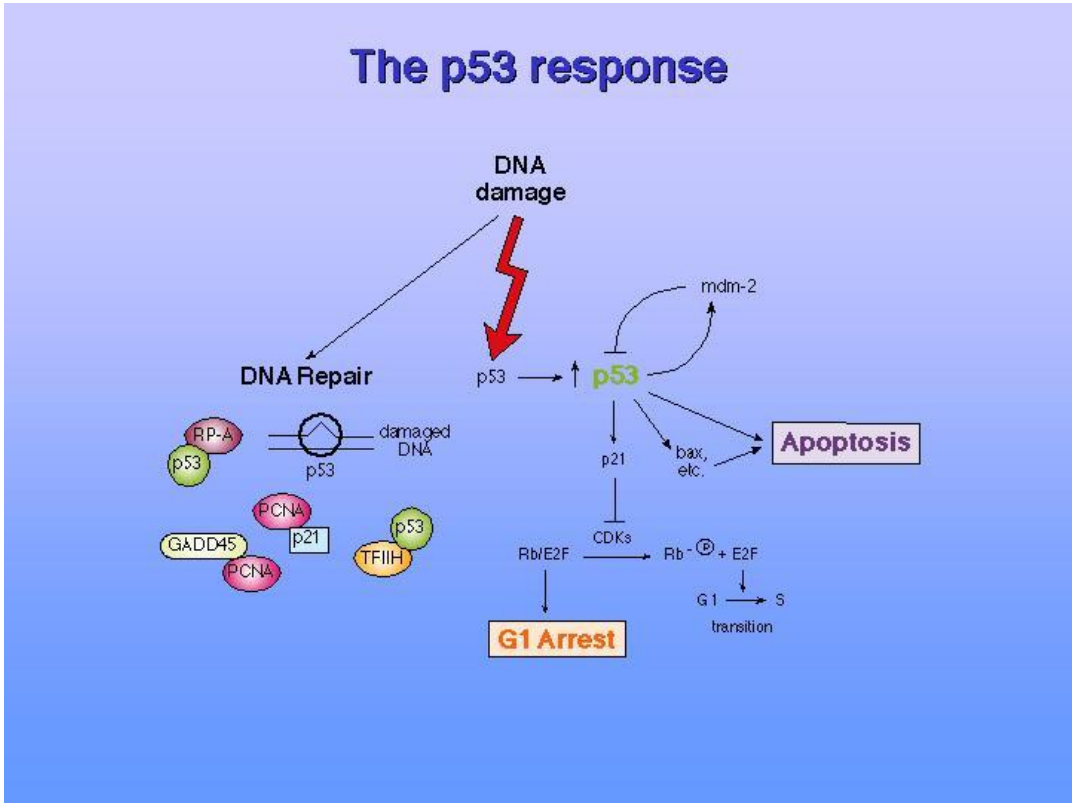


# *Rb* gene

- 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
  --> uncontrolled cell proliferation --> cancer



# Tumor suppressor p53




- P53 halts progression when DNA damaged
  - to give cell time to repair or
  - triggers apoptosis of damaged cell
- If damaged (mutated) cell moves to S phase then it may replicate

# p53

- Encodes protein of molecular wt 53KD
- The “Last Gatekeeper” gene
  - Malignant state not attained despite other cancer-causing mutations
  - Malignancy requires p53 mutations (general observation)

# p53

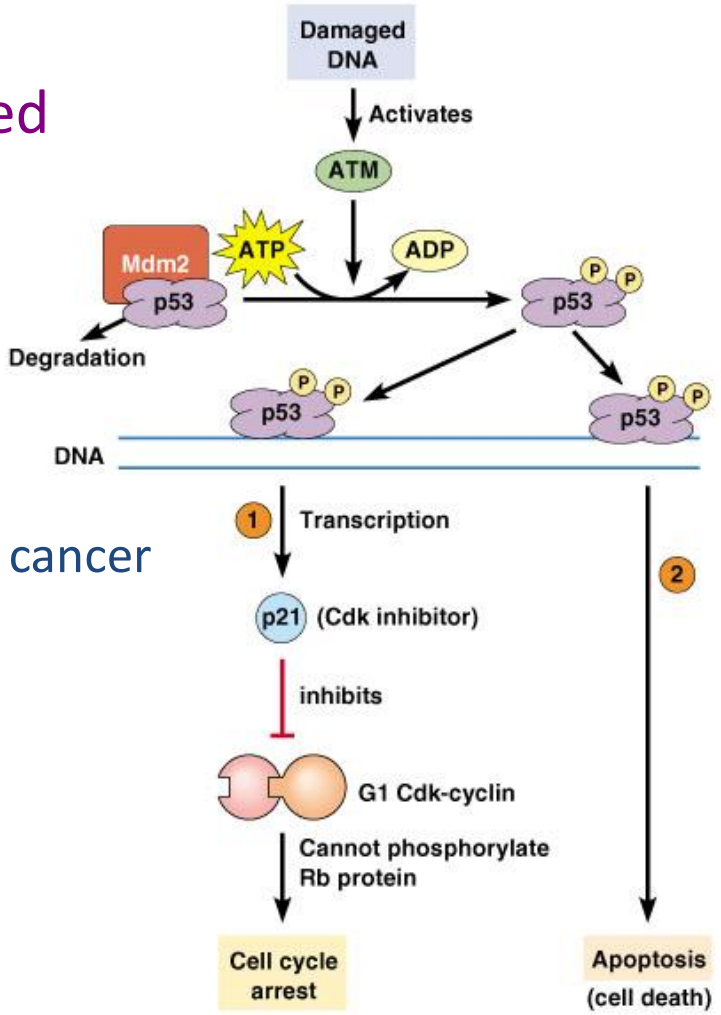
**If DNA damaged**

- 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

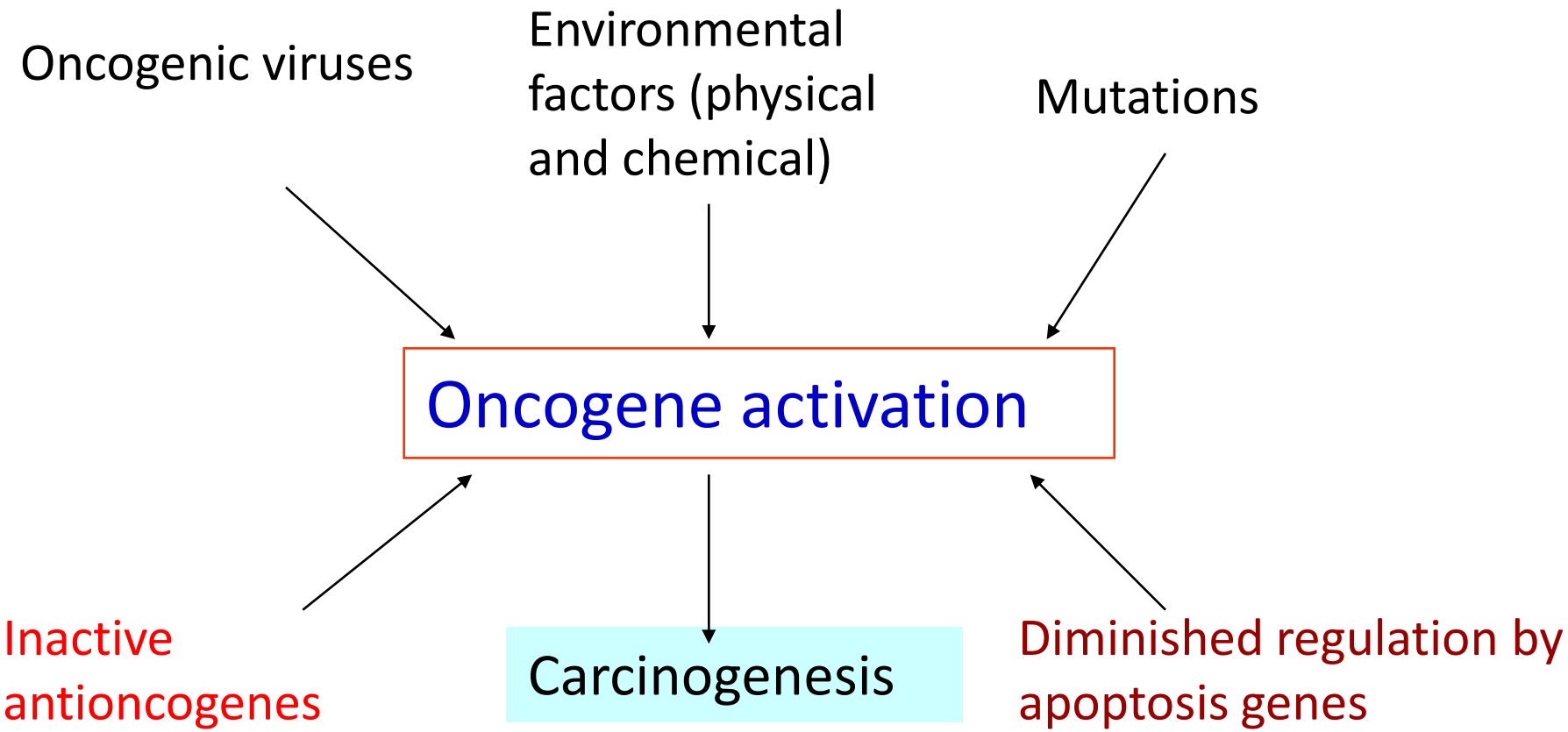


# Biological functions of p53

- Suppressing cell cycle
- Suppressing transformation functions of some oncogenes
- Monitoring cell DNA damage
- Inducing the cell apoptosis

# Human cancers involving p53

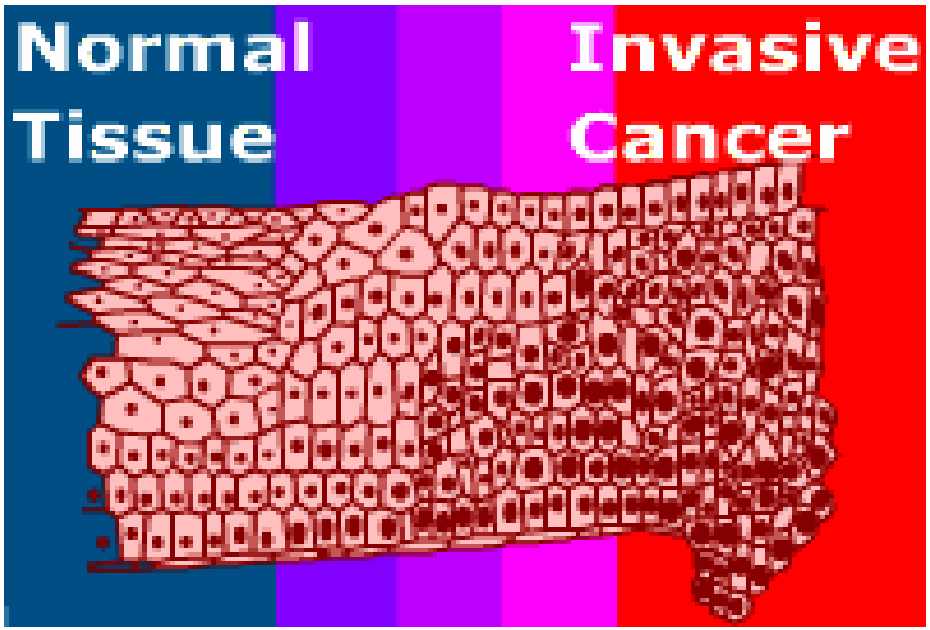
- Cervix
- Breast
- Bladder
- Prostate
- Liver
- Lung
- Skin
- Colon




A simplified hypothesis for the development of cancer

# VIRAL ONCOGENESIS

# 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 Human Cancers

| Family | Virus | Cancer |
|---|---|---|
| *Papillomaviridae* | Human papillomaviruses | Genital tumors<br>Squamous cell carcinomas<br>Oropharyngeal carcinomas |
| *Herpesviridae* | Epstein-Barr virus | Nasopharyngeal carcinoma<br>African Burkitt's lymphoma<br>B cell lymphoma |
| *Hepadnaviridae* | Hepatitis B virus | Hepatocellular carcinoma |
| *Retroviridae* | Human T lymphotrophic viruses<br>Human immunodeficiency viruses | Adult T cell leukemias<br>AIDS-associated tumors (due to impaired T cell responses |
| *Flaviviridae* | Hepatitis C virus | Hepatocellular carcinoma |

# DNA-viral oncogenes

DNA Virus- derived oncogenes:

- Adenovirus:

  E1A pRB

  E1B p19 Apoptosis (anti)

  p55 p53

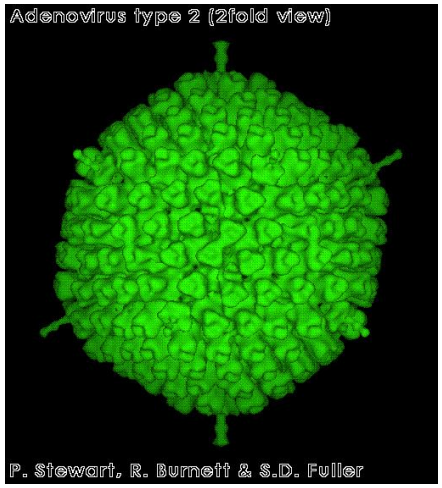
- Polyoma virus (SV40):

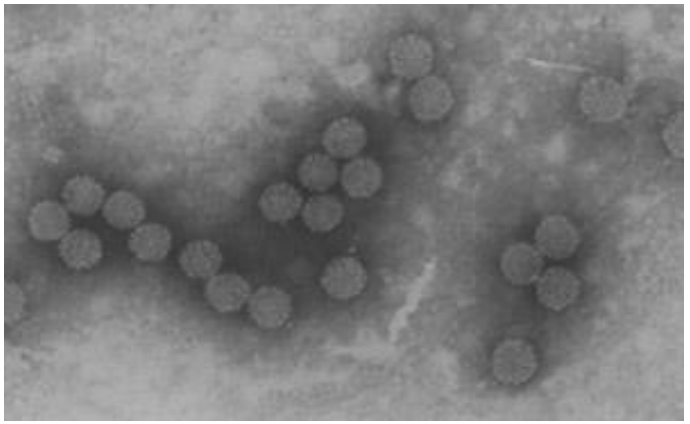
  T antigen pRB / p53

- Papilloma virus:

  E6 p53

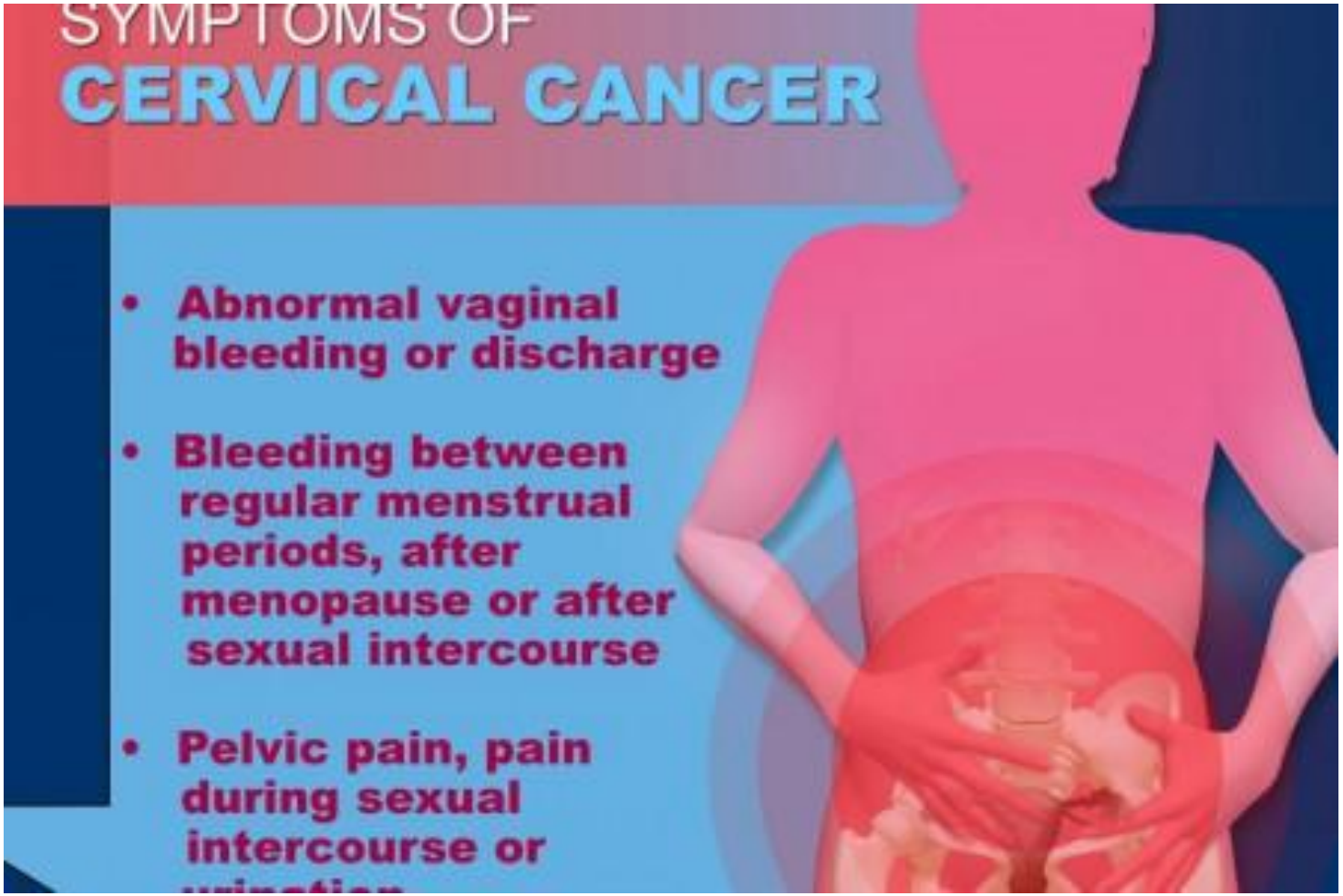
  E7 pRB (approx **75** HPVs)



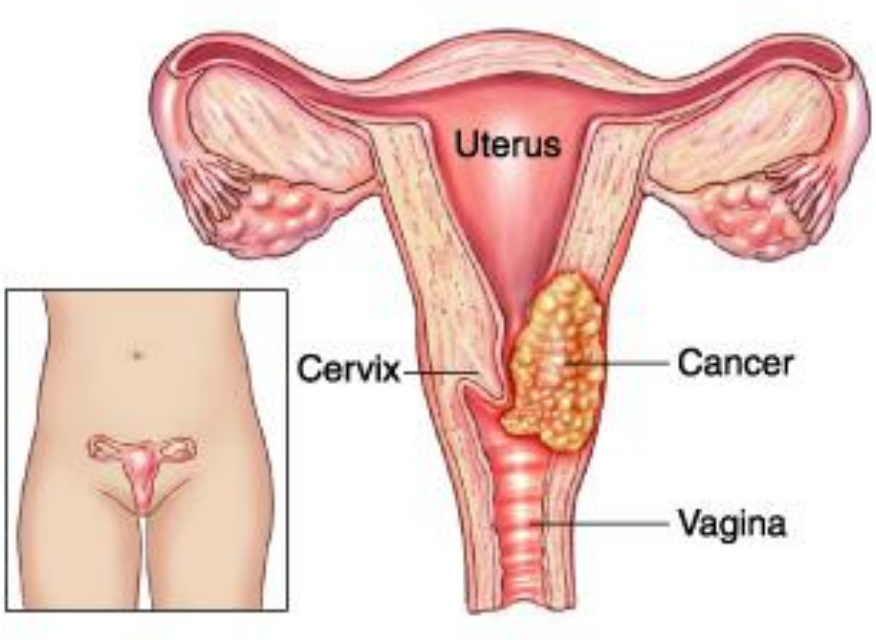
# CERVICAL CANCER

# Cervical Cancer



# Cervical Cancer

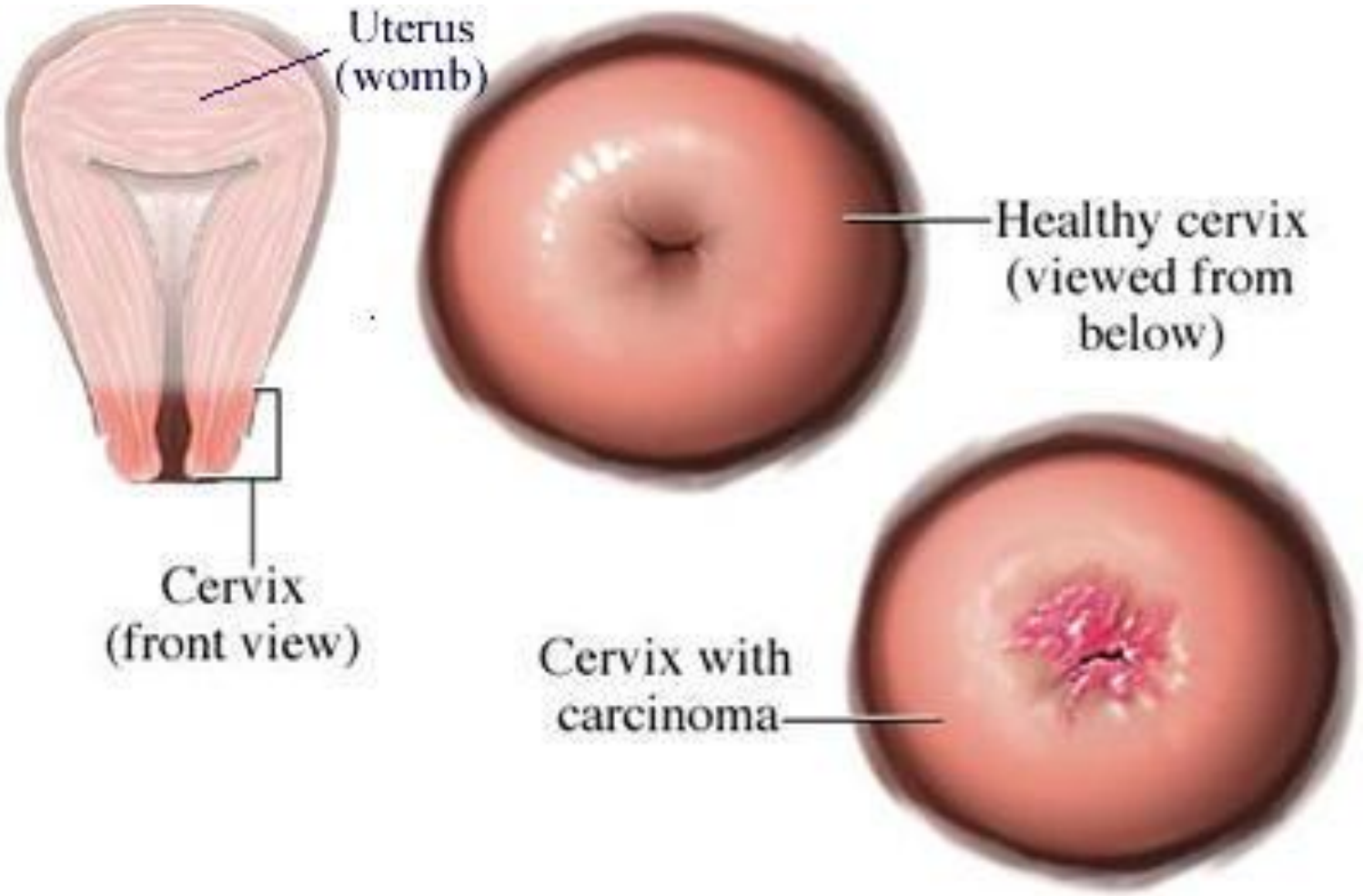
Cancer of the narrowed entry to the uterus (cervix)




**Categorized into stages 0 through IV.**

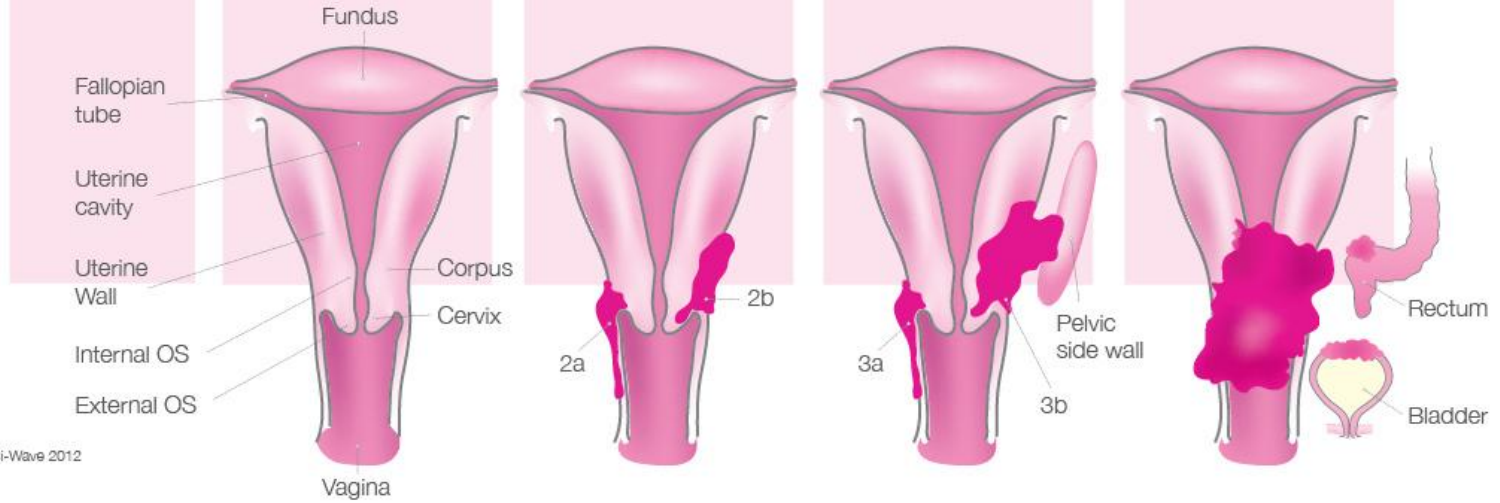
a. Stage 0
   - Limited to cells on surface layer
   - Carcinoma in situ, or pre-invasive cancer

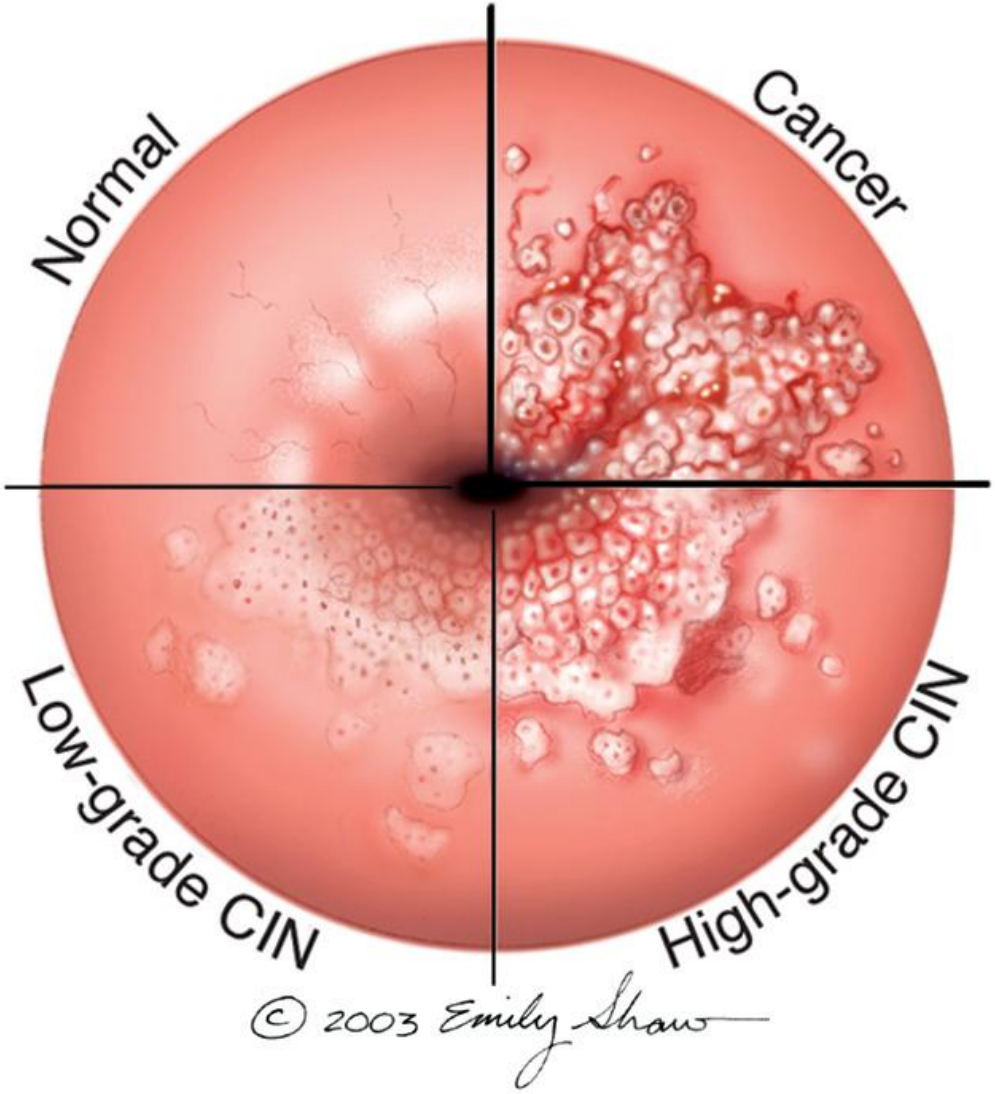
a. Stage IV
   - Spread beyond pelvis - involves the bladder, rectum, or distant organs (liver, lung, or bone).


Uterus
(womb)
Cervix
(front view)
Healthy cervix
(viewed from
below)
Cervix with
carcinoma

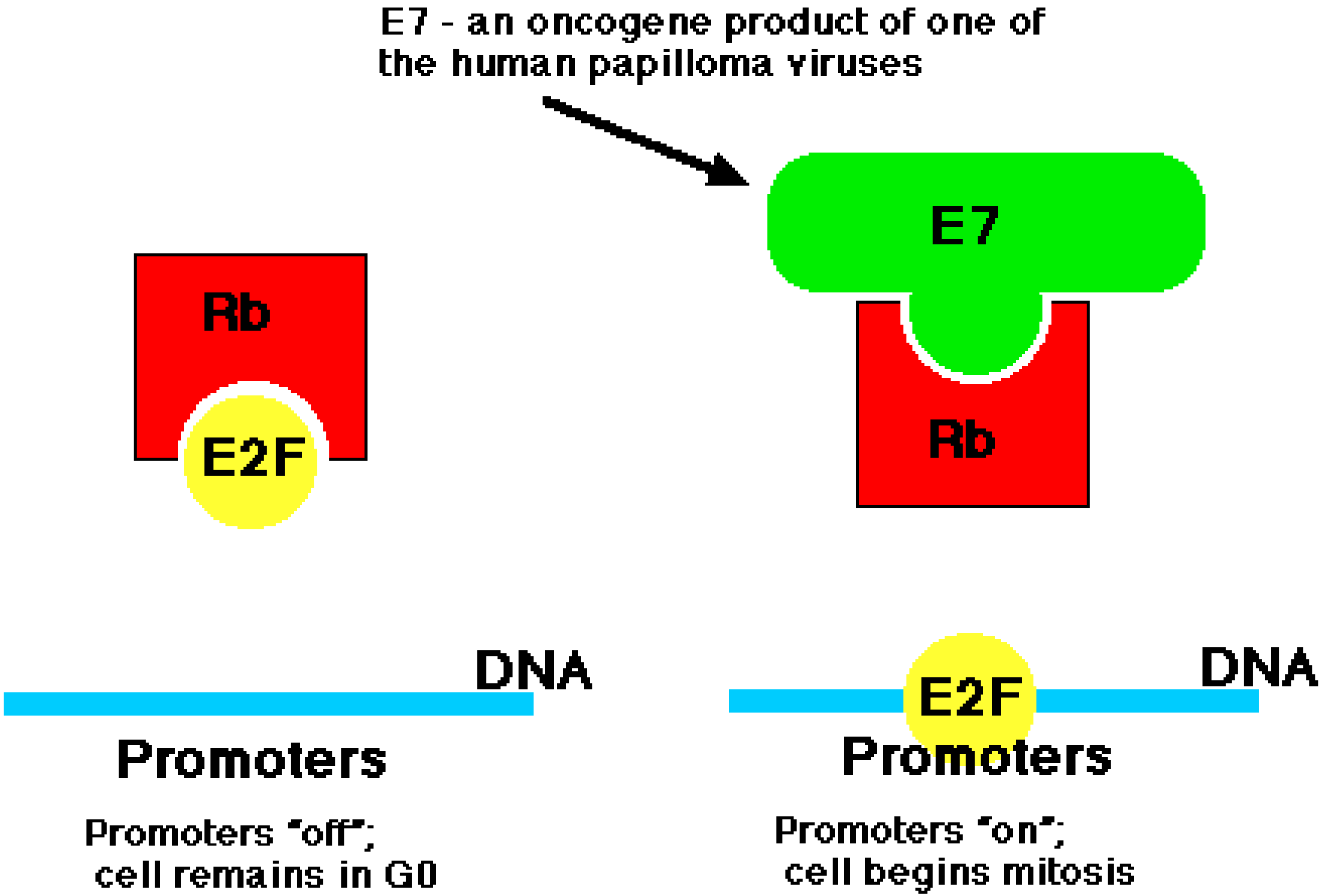
# Staging of Cervical Cancer

| | Stage 0 | Stage 1 | Stage 2 | Stage 3 | Stage 4 |
|---|---|---|---|---|---|
| Extent of tumor | Carcinoma in-situ | Confined to Cervix | Disease beyond cervix but not to pelvic wall or lower 1/3 of vagina | Disease to pelvic wall or lower 1/3 of vagina | Invades bladder, rectum or metastasis |
| 5-year survival | 100% | 85% | 65% | 35% | 7% |
| Stage at presentation | | 47% | 28% | 21% | 4% |




Normal
Cancer
Low-grade CIN
High-grade CIN
© 2003 Emily Shaw

# Human Papilloma virus and cancer



# HPV E7 sequences differ in low and high risk strains

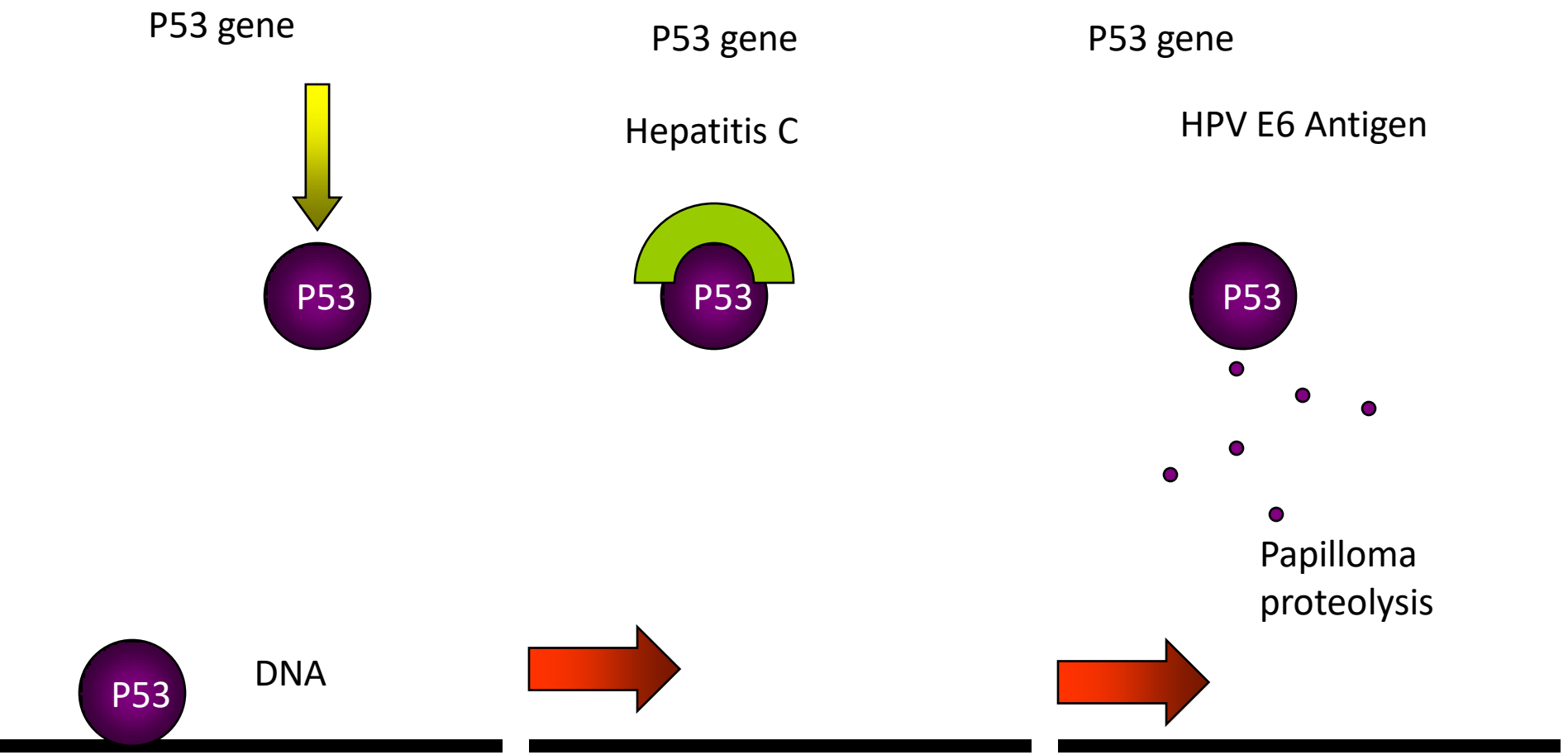
| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 6/11 | P | V | G | L | H | C | Y | E | Q | L | N | D |
| 16 | T | T | D | L | Y | C | Y | E | Q | L | N | D |
| 18 | P | V | D | L | L | C | H | E | Q | L | S | D |
| 31 | A | T | D | L | H | C | Y | E | Q | L | P | S |
| 33 | P | T | D | L | Y | C | Y | E | Q | L | S | D |

**Amino acid sequences in HPV E7 protein affects binding affinity to Rb.**

1. **High risk strains of HPV interact strongly with Rb.**
2. **Low risk HPVs have low affinity for Rb**

# Hepatitis C virus & HPV E6 Antigen

## p53



# Cervical Cancer

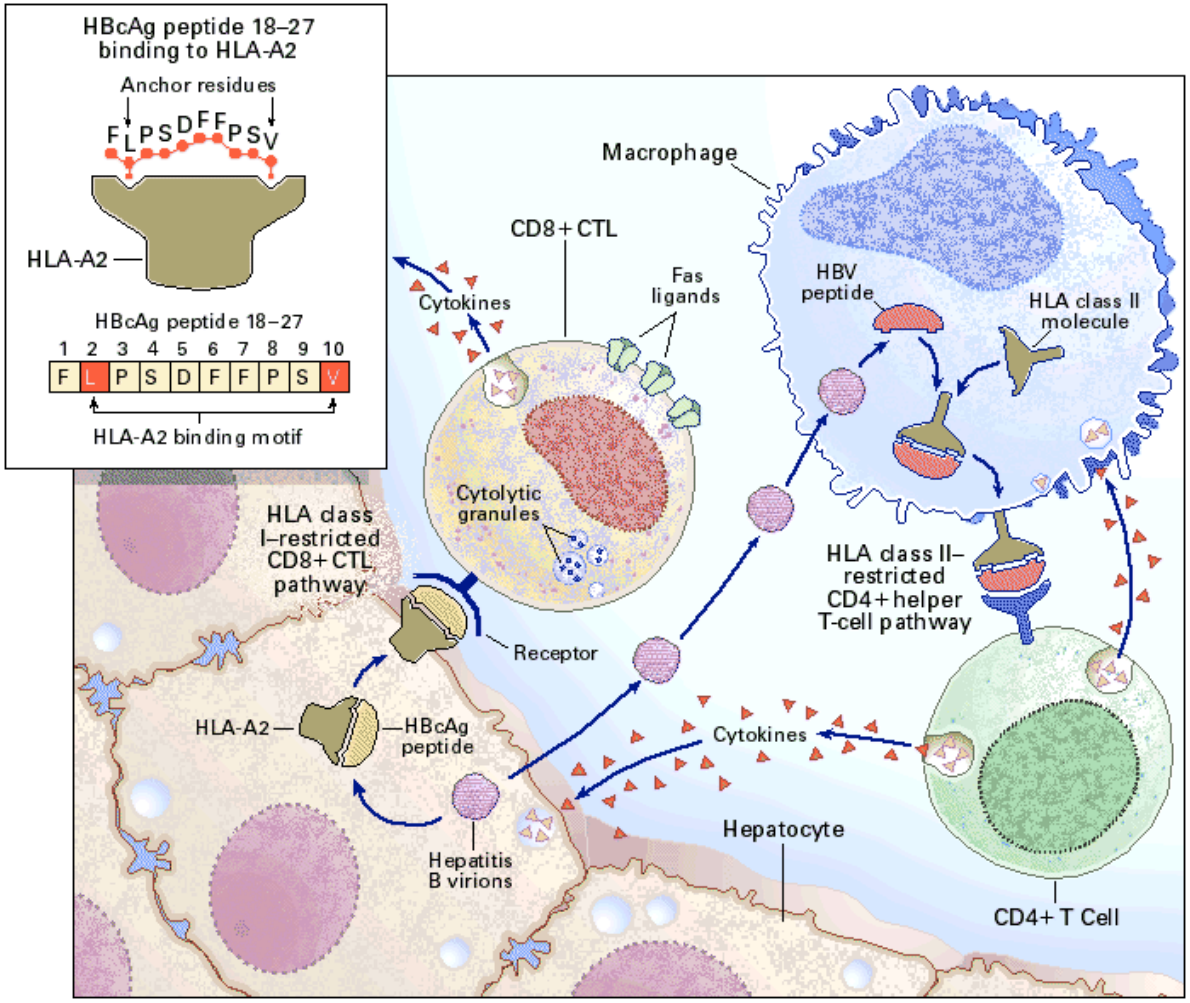
1. Associated with sexually transmitted HPV
2. Types of HPV:
   -High-risk -> e.g. HPV16 & HPV18
   - Low risk

**Risk factors**

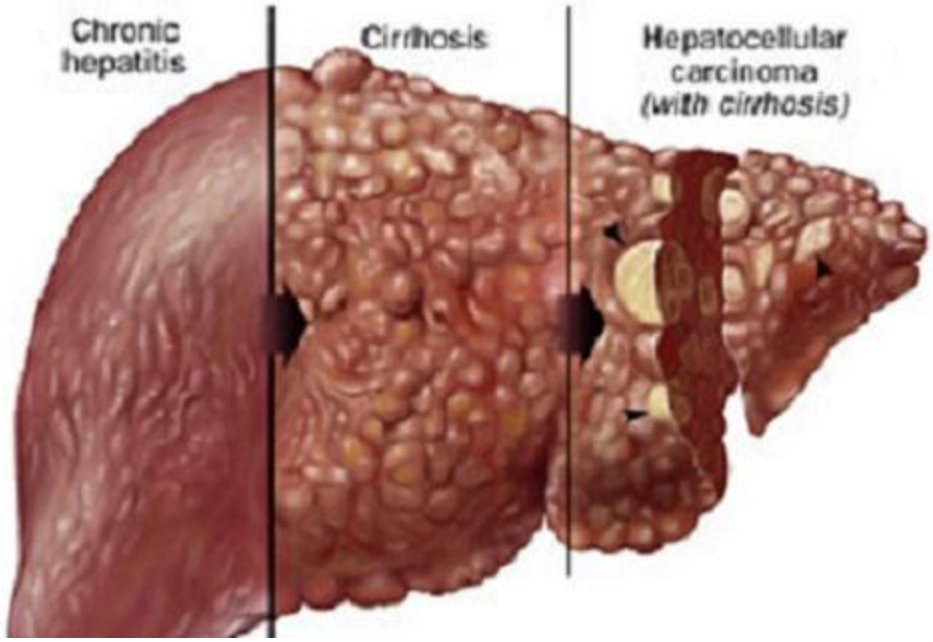
- Multiple partners (promiscuous )
- Presence of genital HPV infections
- Presence of other STDs (e.g. Chlamydia, HIV)
- Prolonged use of oral contraceptives (>5 yrs) – increased risk
- Smoking etc

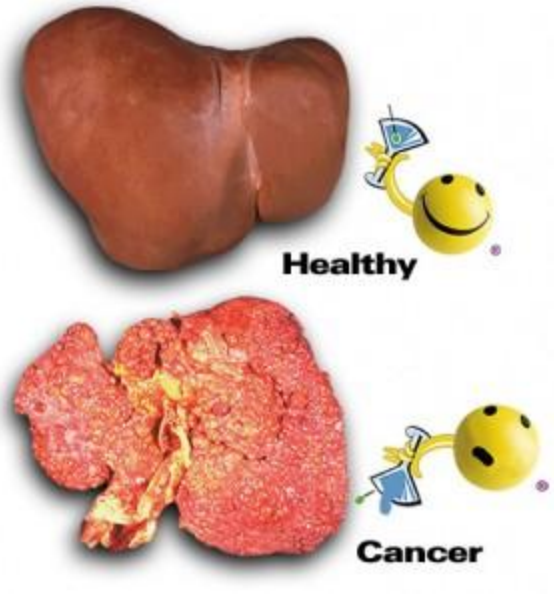
# LIVER CANCER

# HBV & Liver Cancer

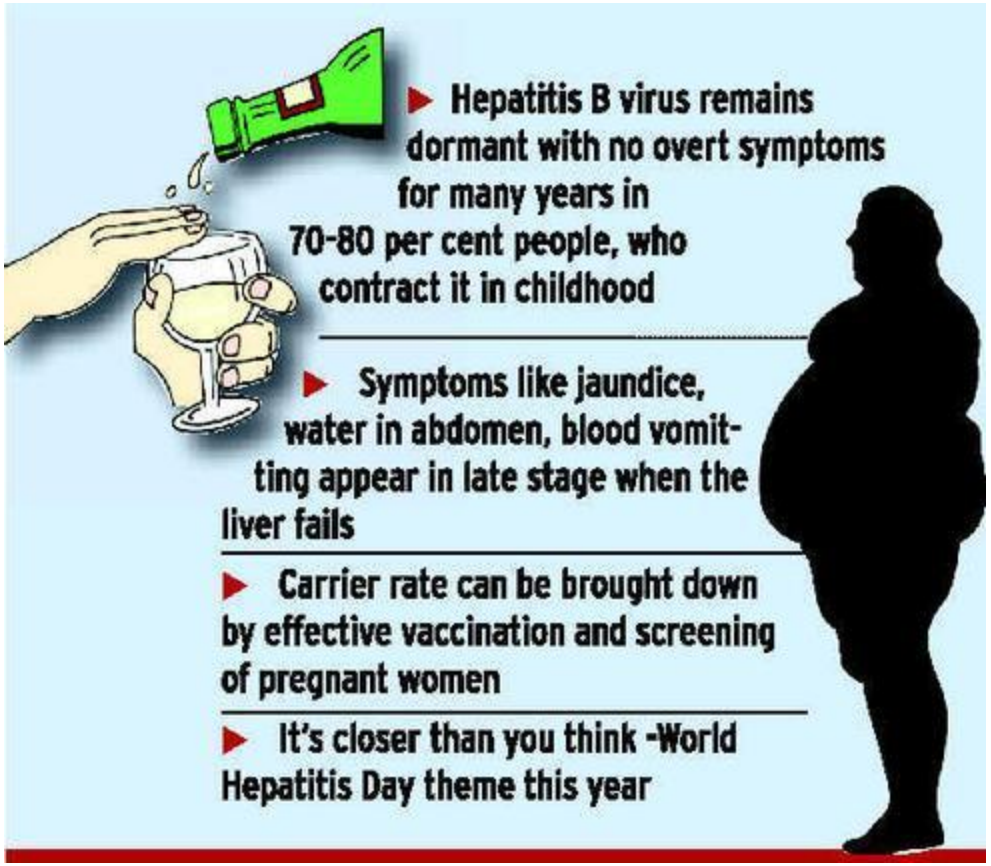


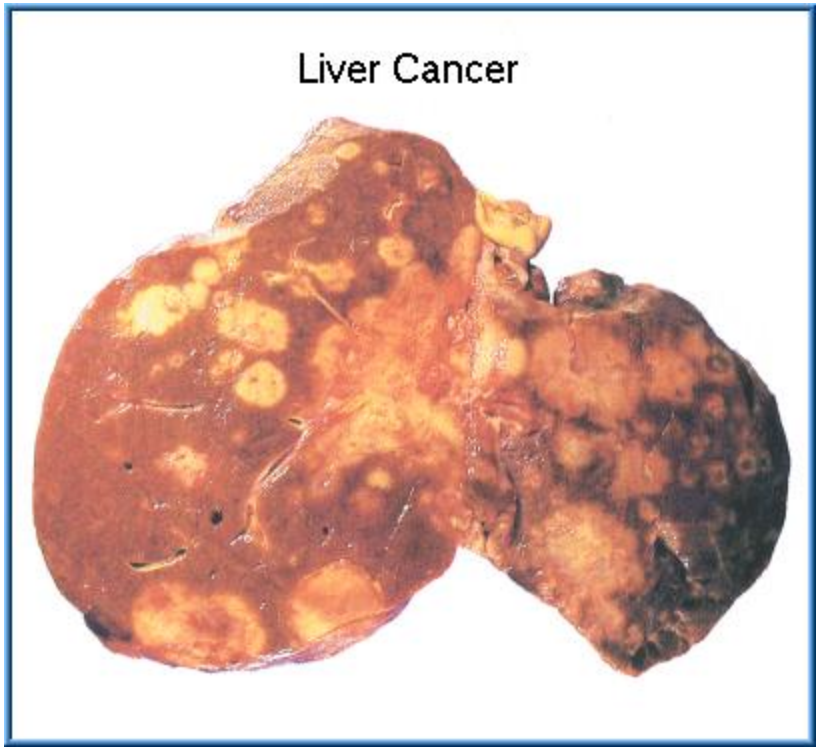

HBV
-95% acute infections (liver inflammation, jaundice, liver failure) -> autoimmune
- 5% chronic -> HCC
- Integrated viral DNA common


Chronic hepatitis
Cirrhosis
Hepatocellular carcinoma (with cirrhosis)

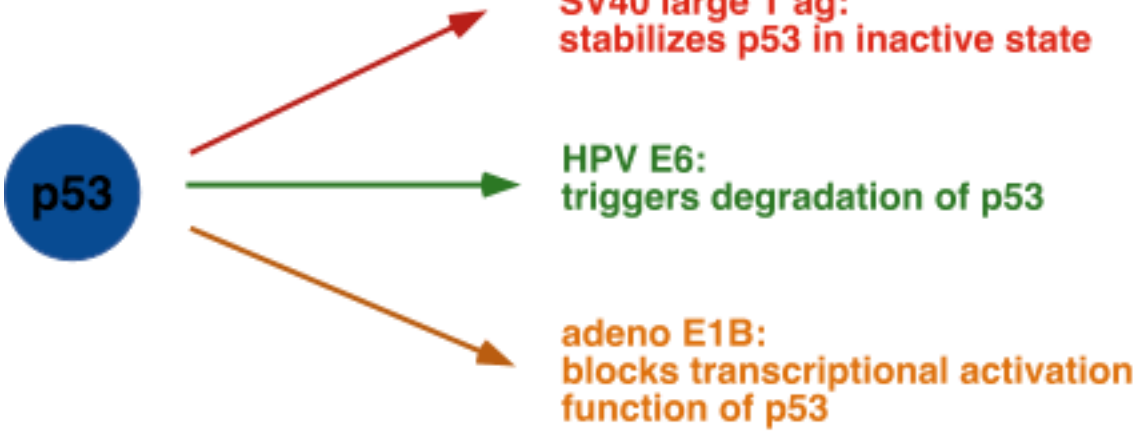

Healthy
Cancer

# Liver cancer

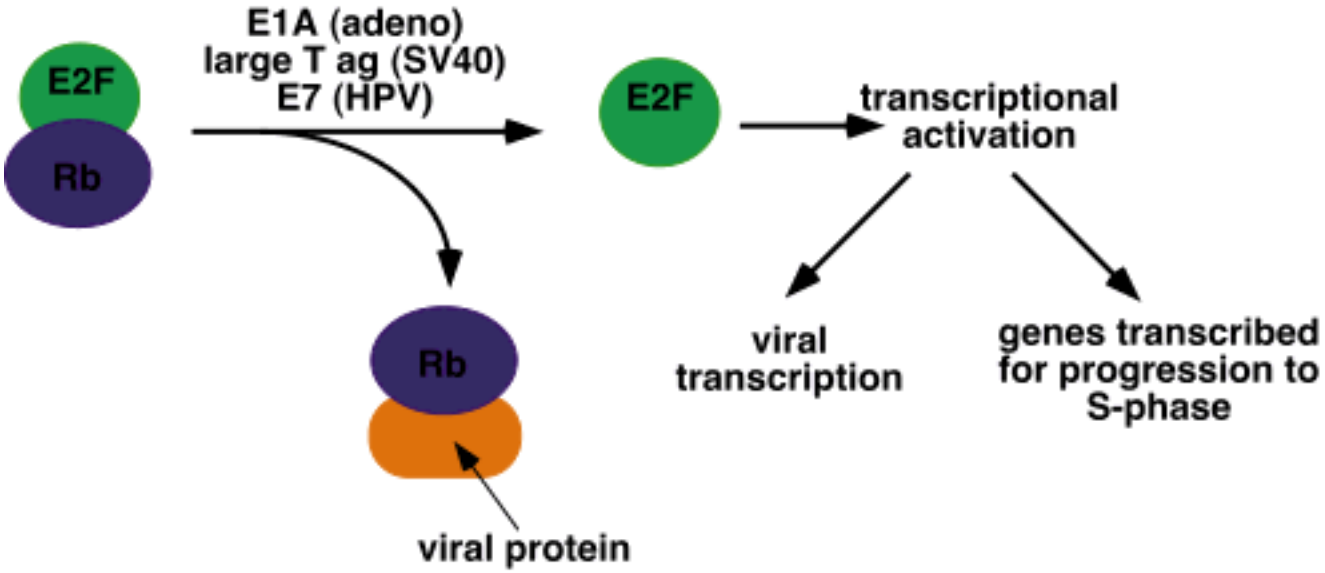


## Viral Inactivation of p53 Function



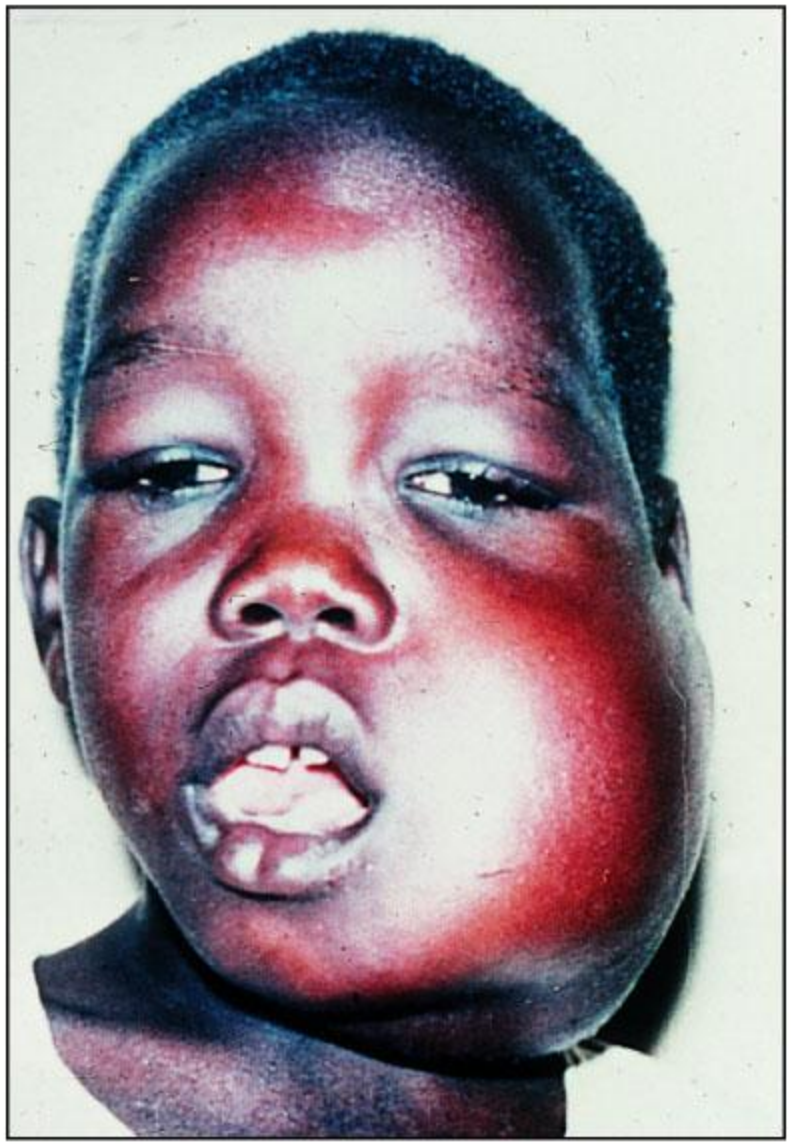

## DNA Virus Inactivation of Rb Protein Function




How should these proteins be similar?

# BURKITT'S LYMPHOMA (BL)

# Burkitt's Lymphoma (BL)

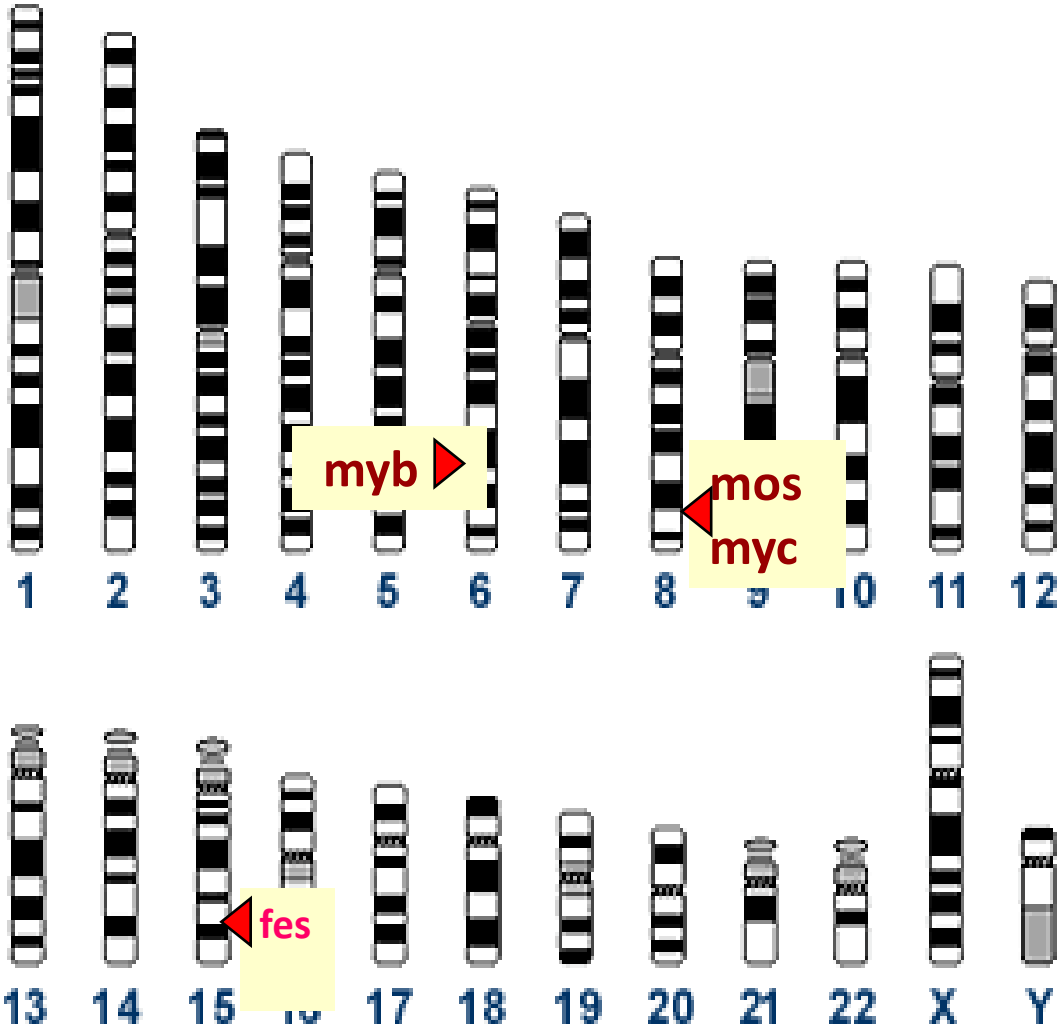




Causative agent = EBV
Two cancers
1. BL
2. Nasopharyngeal Carcinoma

- EBV – no viral oncogenes
- Cellular gene (c-myc) translocation
  – chromosome 8->14

# DNA Tumor Viruses e.g. Herpesviruses can cause chromosomal breaks




**Genes can be assigned to sites on specific chromosomes**

**mos and myc : chromosome 8**

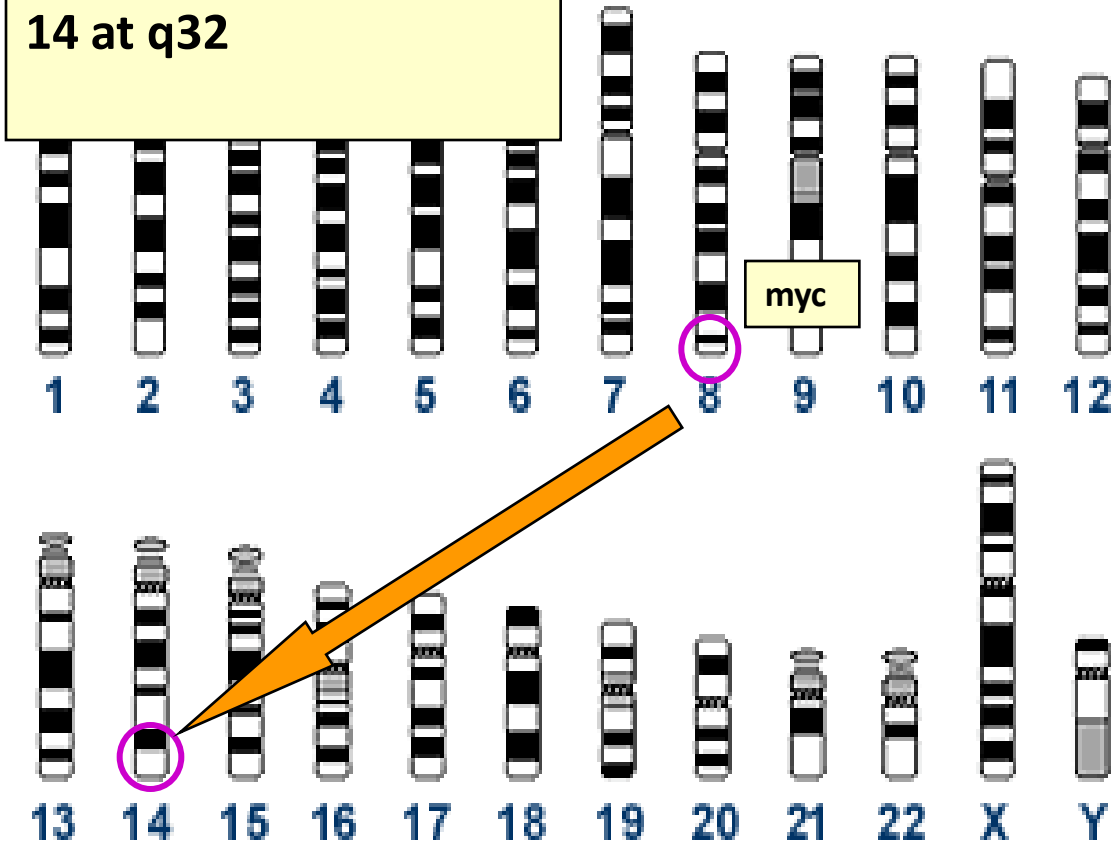
**fes: chromosome 15**

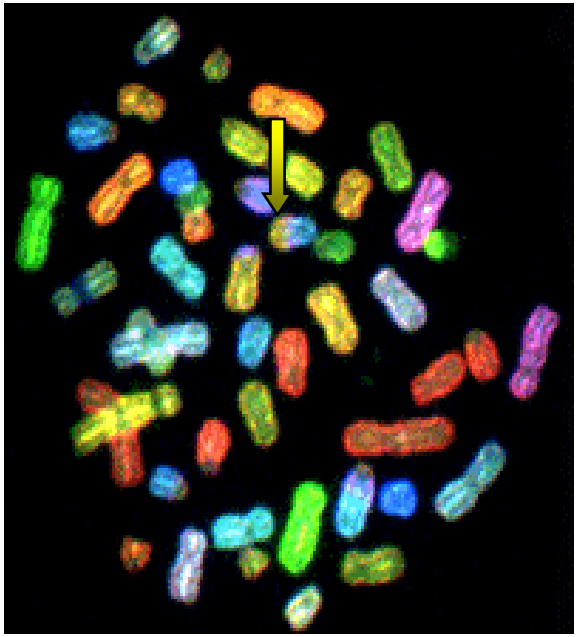
# Cancers often result from gene translocations

**Burkitt's Lymphoma**

**8:14 translocation**

**Break in chromosome 14 at q32**



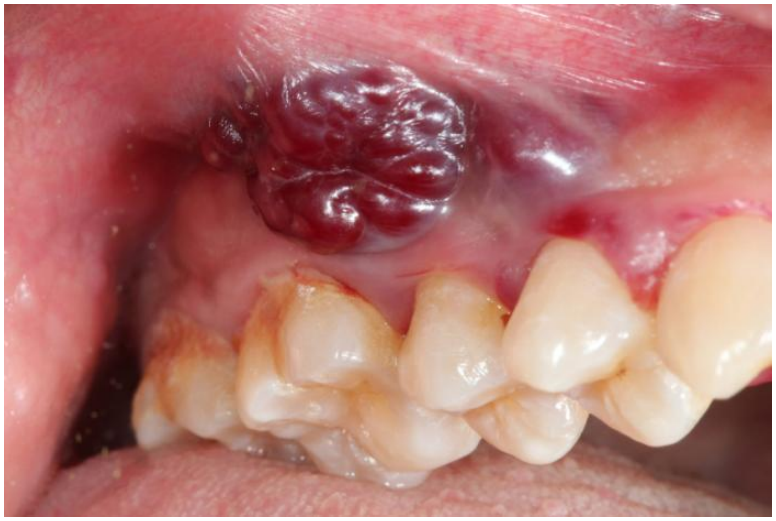
**Acute myelocytic leukemia**
**7:15**
**9:18**
**11:15:17**

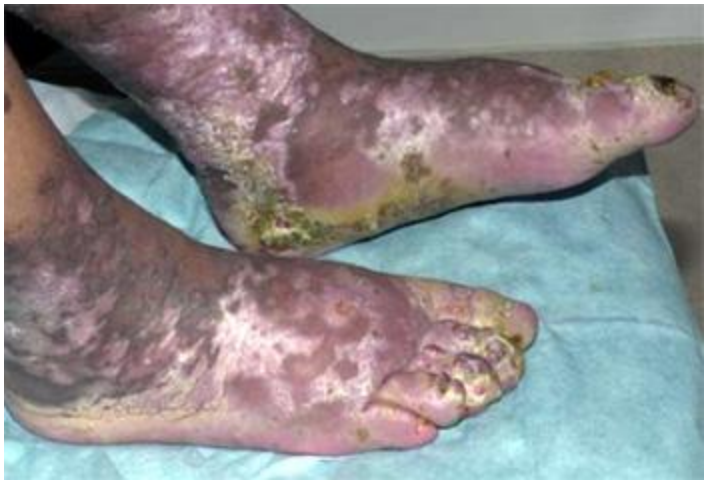
# Oncogenesis by rearrangement

| Tumor | c-onc | new promotor | |
|---|---|---|---|
| Burkitt's lymphoma | myc | (8) | Ig heavy (8 to 14) |
| B-cell chronic lymphocytic | bcl-1 | | Ig heavy (11 to 14) |
| leukemia | bcl-2 | | Ig heavy (18 to 14) |

# KAPOSI’S SARCOMA (KS)

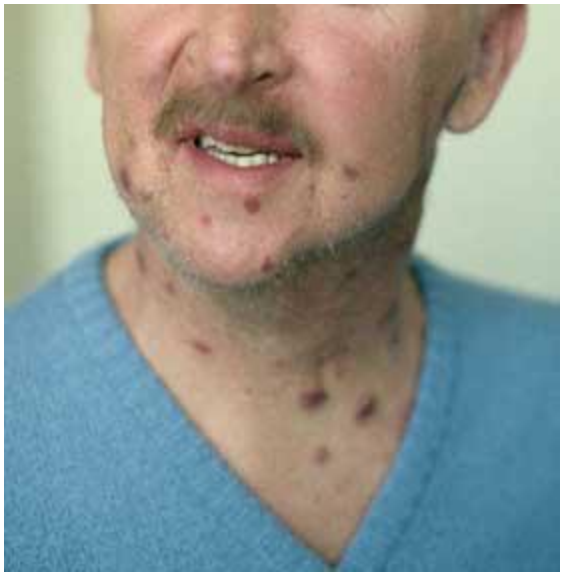
# Kaposi's Sarcoma (KS)





Lesions can occur in several parts of the body

- KSHV associated (not proven) with KS
- Herpesviridae family (gamma subfamily)
- KS common in context of HIV/AIDS



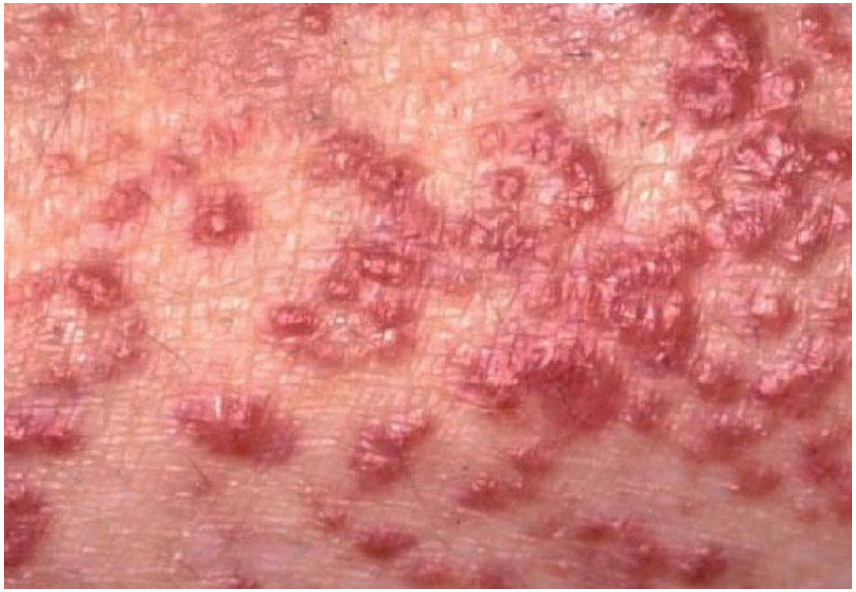
# Kaposi’s Sarcoma (KS)

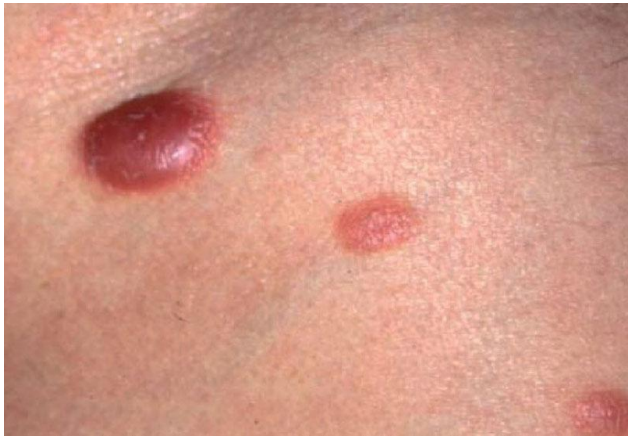
1. Associated with HHV-8
   - Virus: Kaposi's Sarcoma-Associated Herpesvirus (KSHV)
   - Preferentially infects a type of endothelial cells (spindle cells) from which KS develops
2. Inflamed, angioproliferative lesions
3. Can spread both locally & systemically

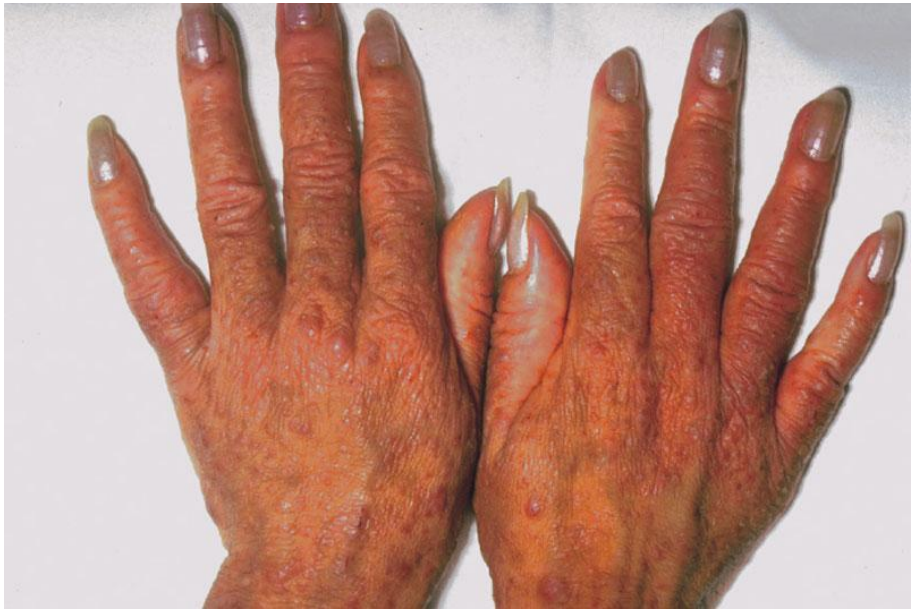
# RETROVIRUSES

# Retroviruses

- Epidemiology
  - Typical infectious viruses (exogenous)
    - Sexual transmission
    - IV drug abusers
    - Other, unknown transmission mechanisms
- Classification
  - Leukemia viruses
    - *Alpharetrovirus*
    - *Gammaretrovirus*
  - Nontransforming retroviruses
    - *Deltaretrovirus*
    - *Lentivirus*

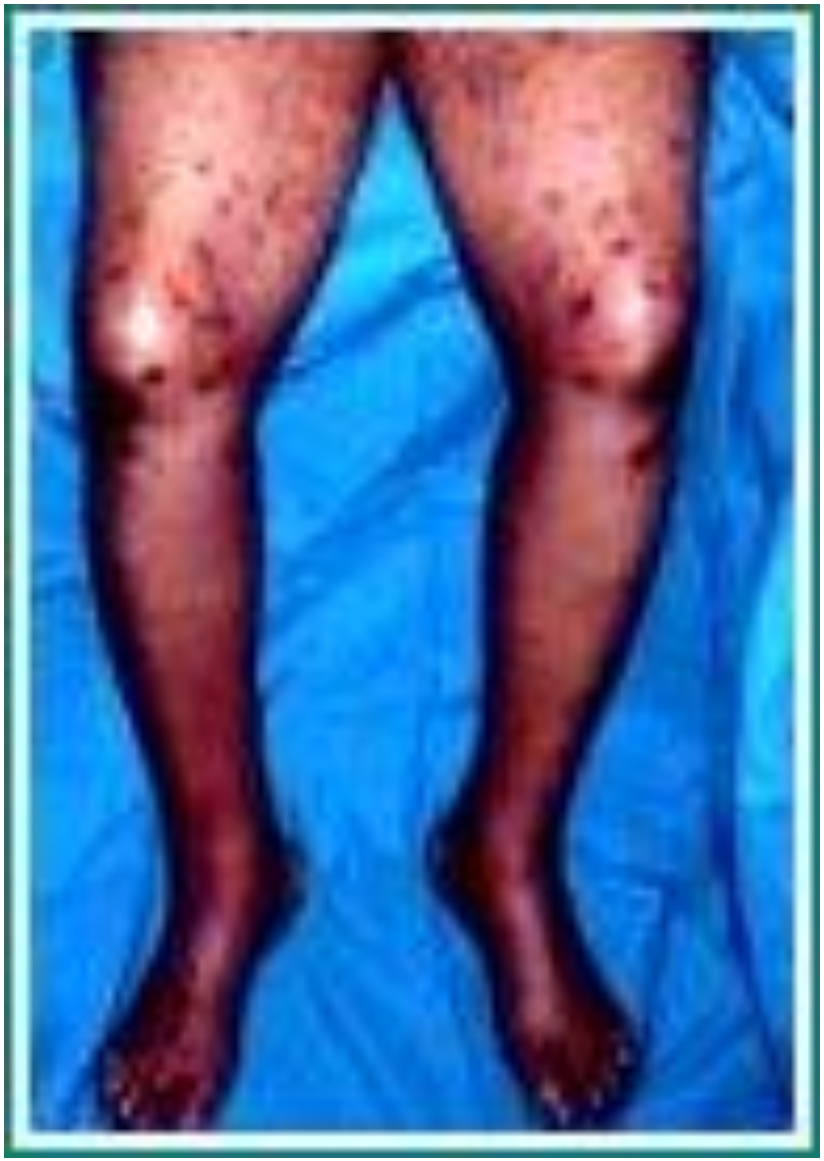






**Adult T-cell leukaemia/lymphoma (ATLL)**

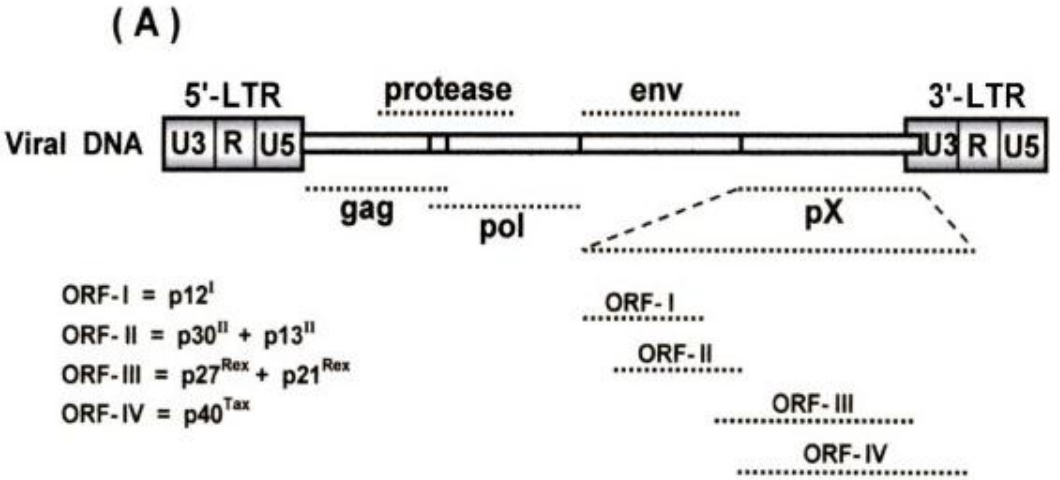
1. Caused by HTLV-1
2. Patients frequently have skin or pulmonary lesions

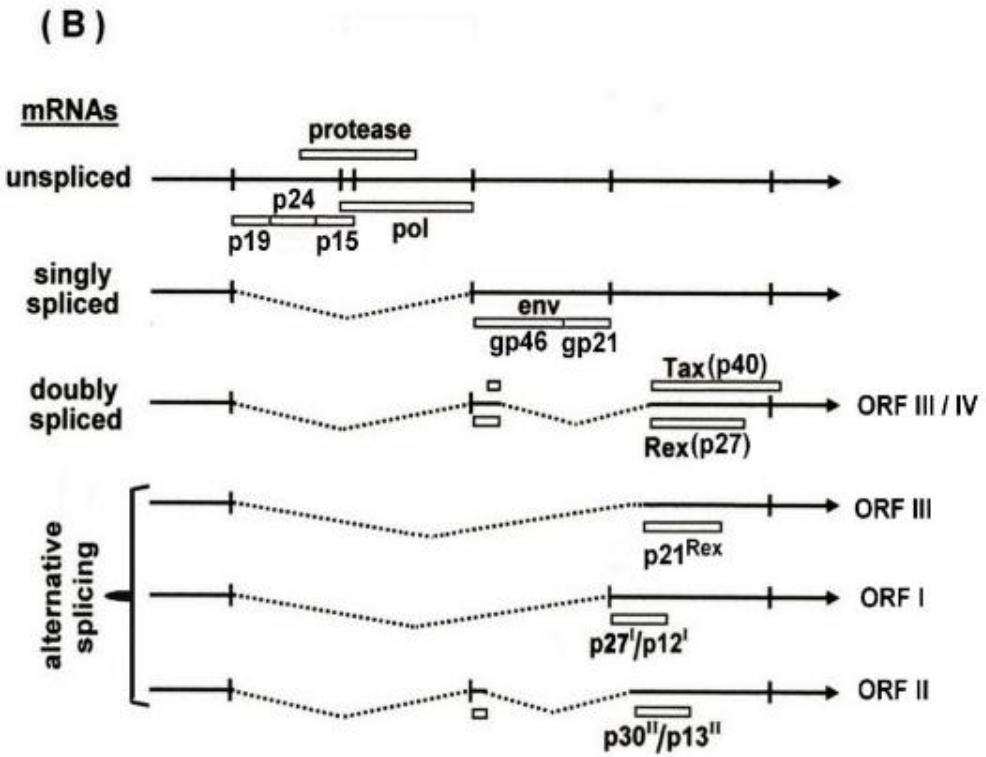


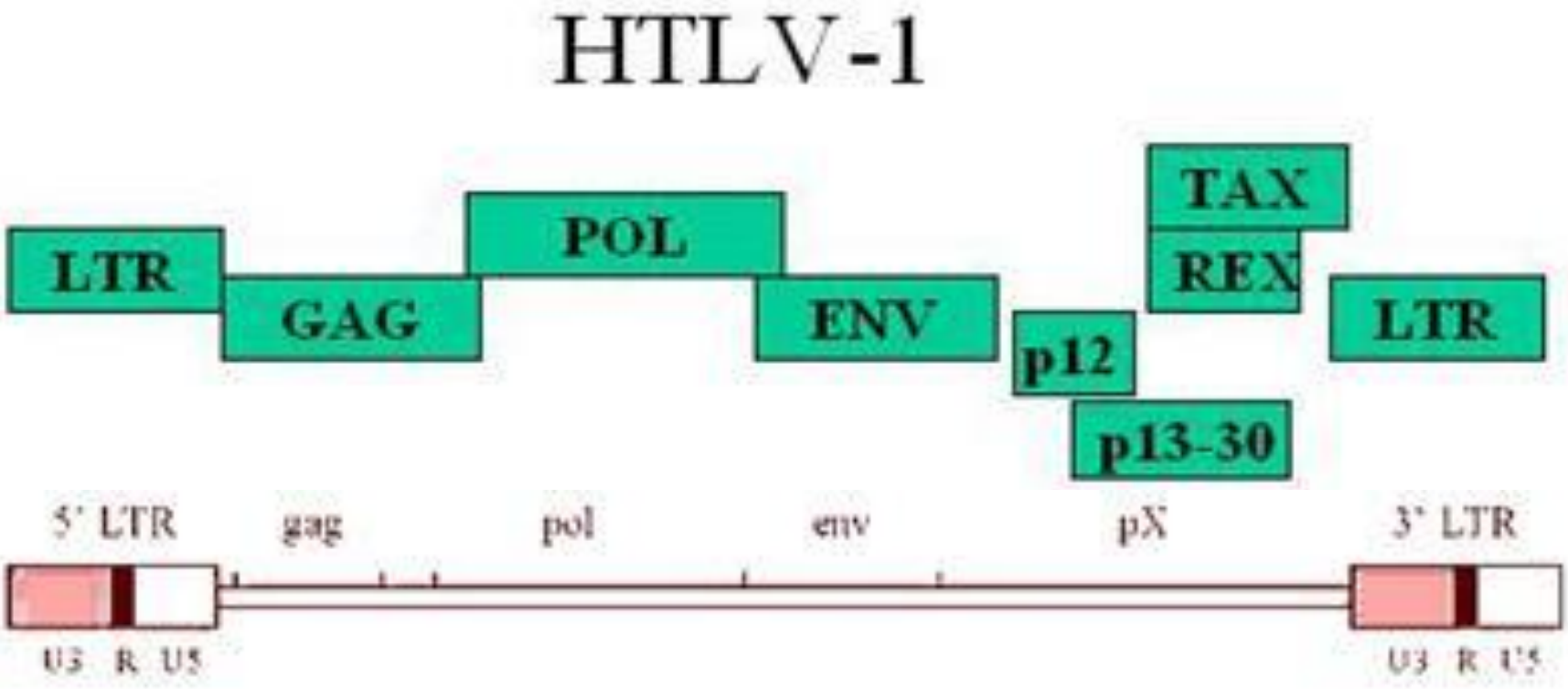
## Human T-cell Lymphotropic Virus (HTLV)

1. Discovery from a Afro American with a T-cell lymphoma infiltrating his skin
2. HTLV-1 (1980)
3. Related virus HTLV-II discovered in 1982
4. Tax (viral oncogene) a transcriptional activator
5. Tax protein known to immortalize cells

# Retroviruses






- tax gene encodes Tax protein
- Tax is a transcription factor
- Promotes cell division

# Mechanisms of Retroviral Carcinogenesis

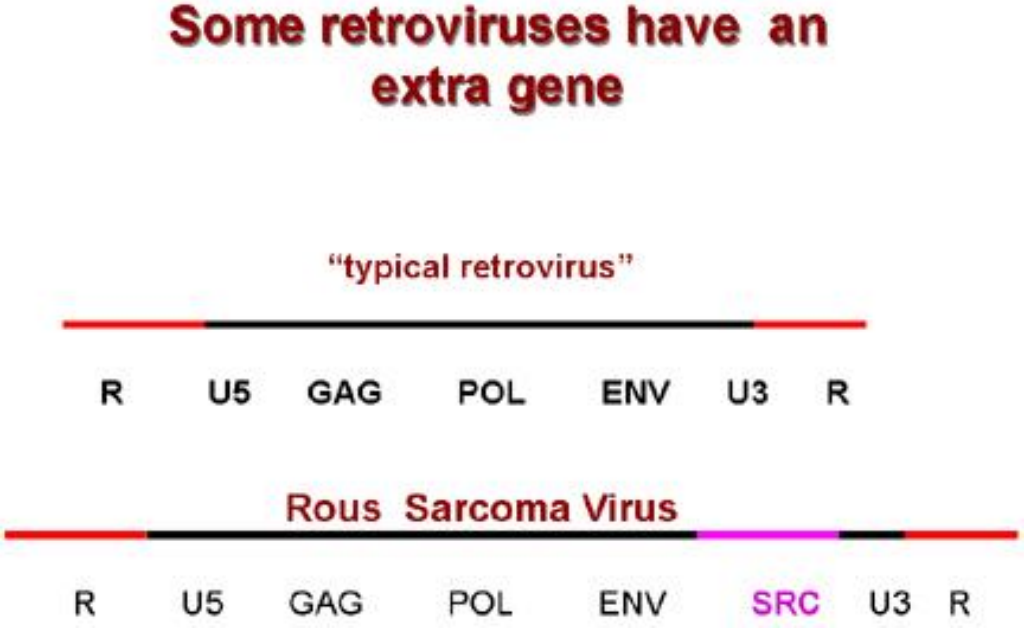
- Infection -> uncoating (cytoplasm)
- Reverse transcriptase makes a dsDNA copy
- dsDNA integration (provirus)
- Tax (e.g. HTLV-1)
  - Viral oncogene (v-onc)
  - Transcription factor (Tax) combines with cellular activating transcription factor-4 (ATF4)
  - The dimer binds to HTLV proviral and cellular promoters to drive cell division
  - Eventually: Leukemia (blood cancer)

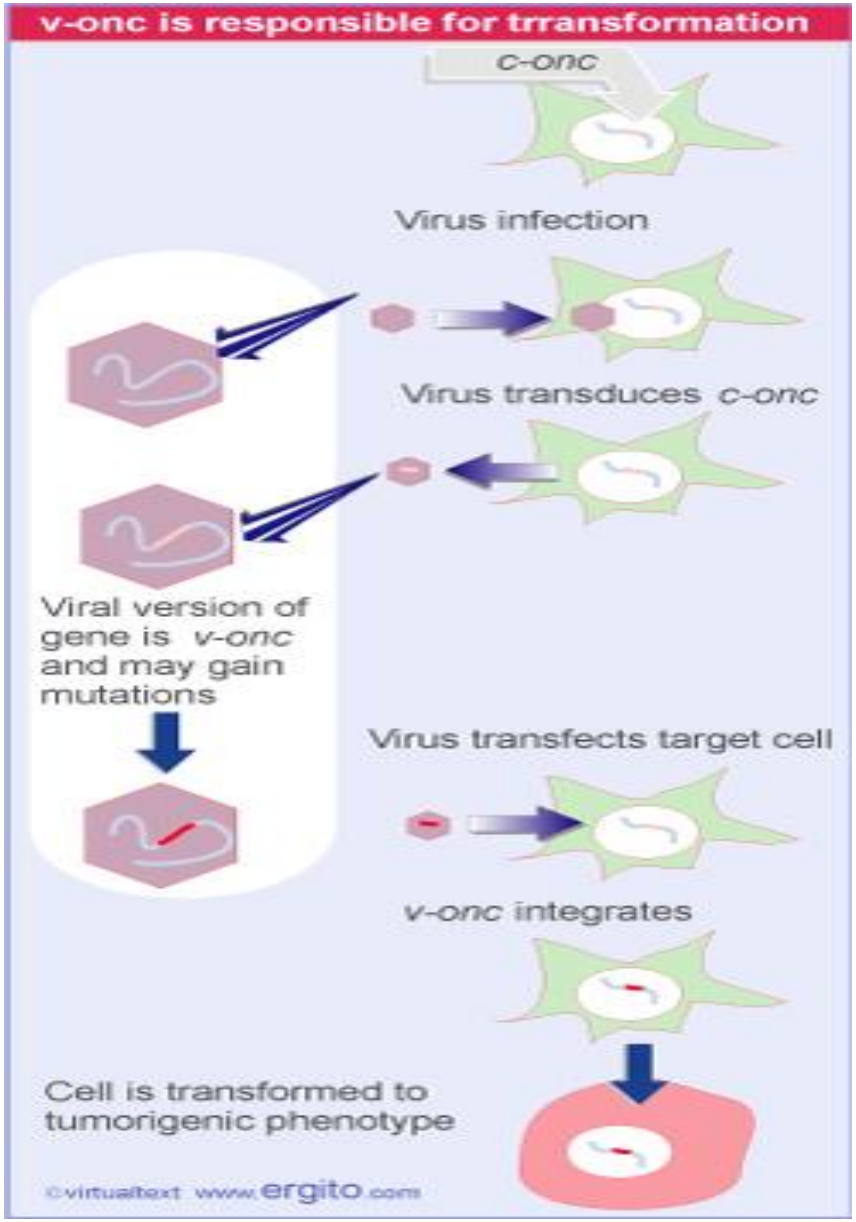
# RNA Tumor Viruses

## Retroviruses - Two types

1. **Acutely transforming (rapid onset of neoplasia)**

   **– have an extra gene e.g. RSV**

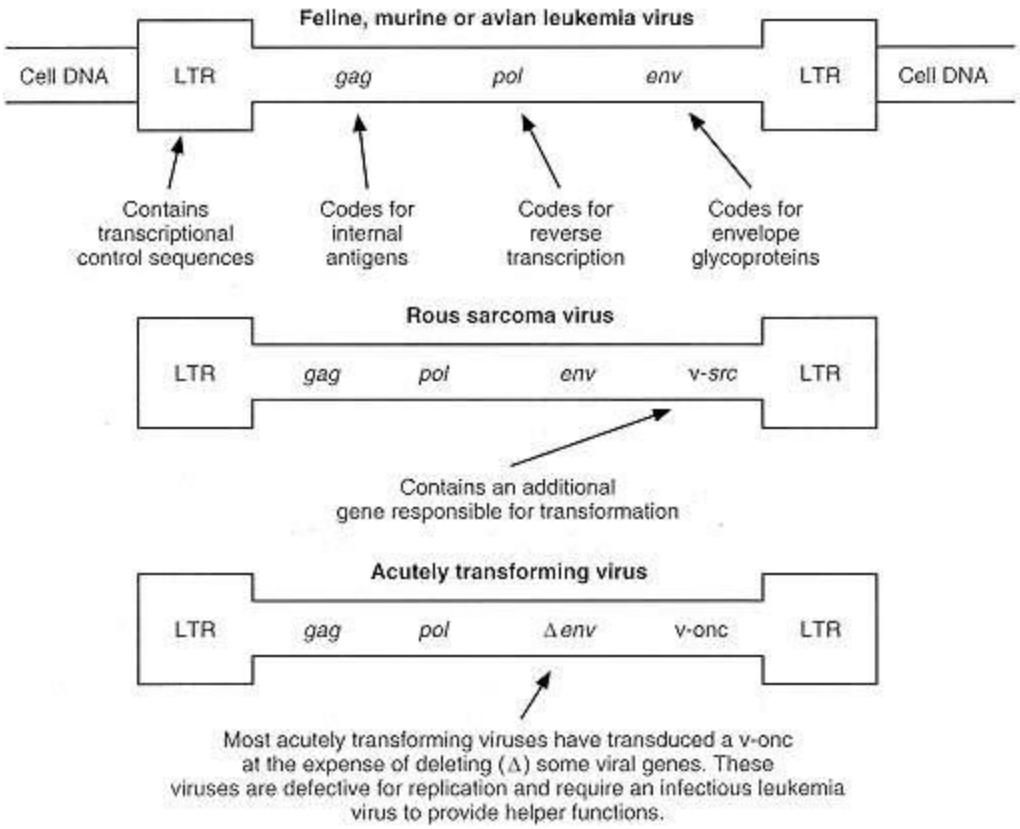





Normal gene
Cell-oncogene
(c-onc)

**Replication-defective virus**

# Acutely transforming retroviruses



# Comparison of oncogenic and non-oncogenic retroviruses

Fields Virology 4th edition, 2002, Chapter 10, Lippincott, Williams and Wilkins, 2002 Fig. 10-2

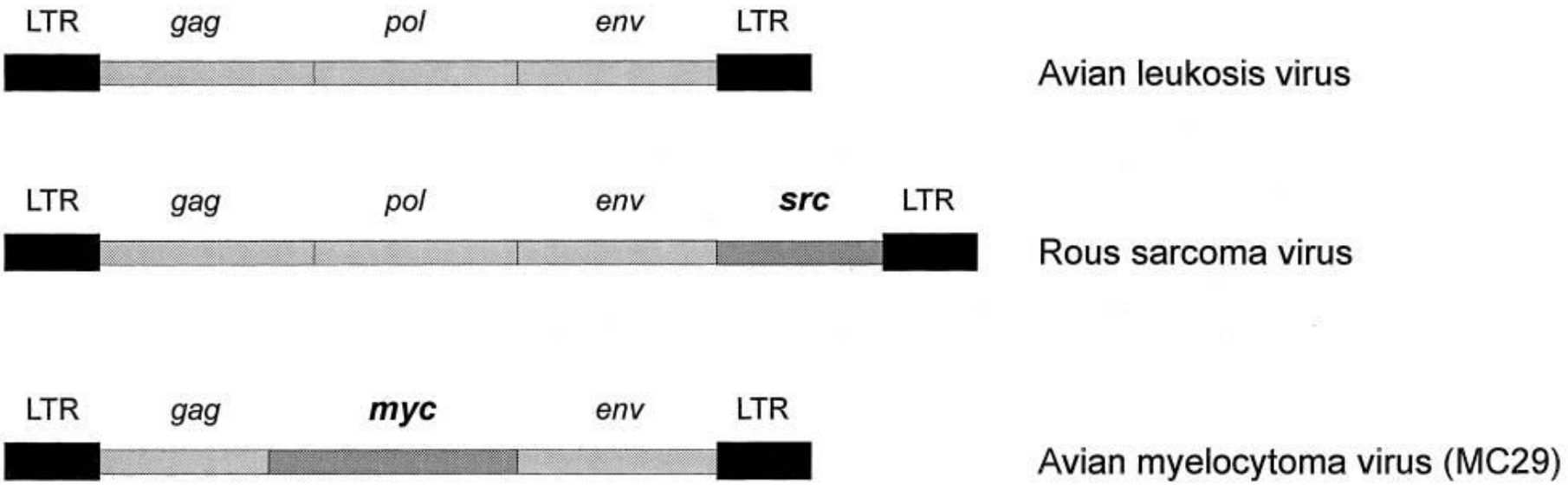



**Figure 10-2** Genomic structure of avian leukosis virus (ALV) and two transducing retroviruses. In addition to the long terminal repeat (LTR) sequences that provide transcriptional regulatory elements, the normal genome of ALV contains three major coding regions including gag, pol, and env. Gag encodes structural proteins of the virus, pol encodes enzymes involved in reverse transcription and integration, and env encodes the virion surface glycoproteins. In Rous sarcoma virus, the cellular src sequences are added to an otherwise intact retroviral genome. In contrast, in the MC29 virus, the addition of myc sequence is at the expense of the entire pol gene and parts of both gag and env.

# Examples of some viral oncogenes (v-onc)

| Virus | Oncogene |
|---|---|
| Rous sarcoma virus | v-src |
| Simian sarcoma virus | v-sis |
| Avian erythroblastosis virus | v-erbA or v-erbB |
| Kirsten murine sarcoma virus | v-*k*ras |
| Moloney murine sarcoma virus | v-mos |
| MC29 avian myelocytoma virus | v-myc |

# RNA Tumor Viruses

**2. Chronically transforming retroviruses**

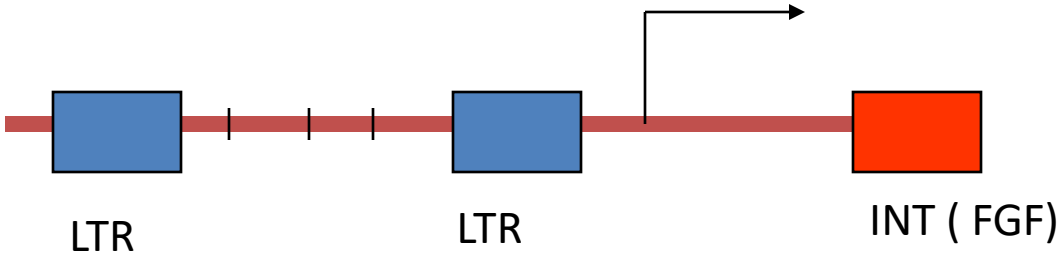
**- e.g. Avian leukemia virus (ALV)**

**- No viral oncogene**

**Avian Leukosis Virus (causes lymphomas)**


**No oncogene! – How does it cause a tumor?**

# Insertional Mutagenesis




MMTV

mammalian

# Insertional mutagenesis