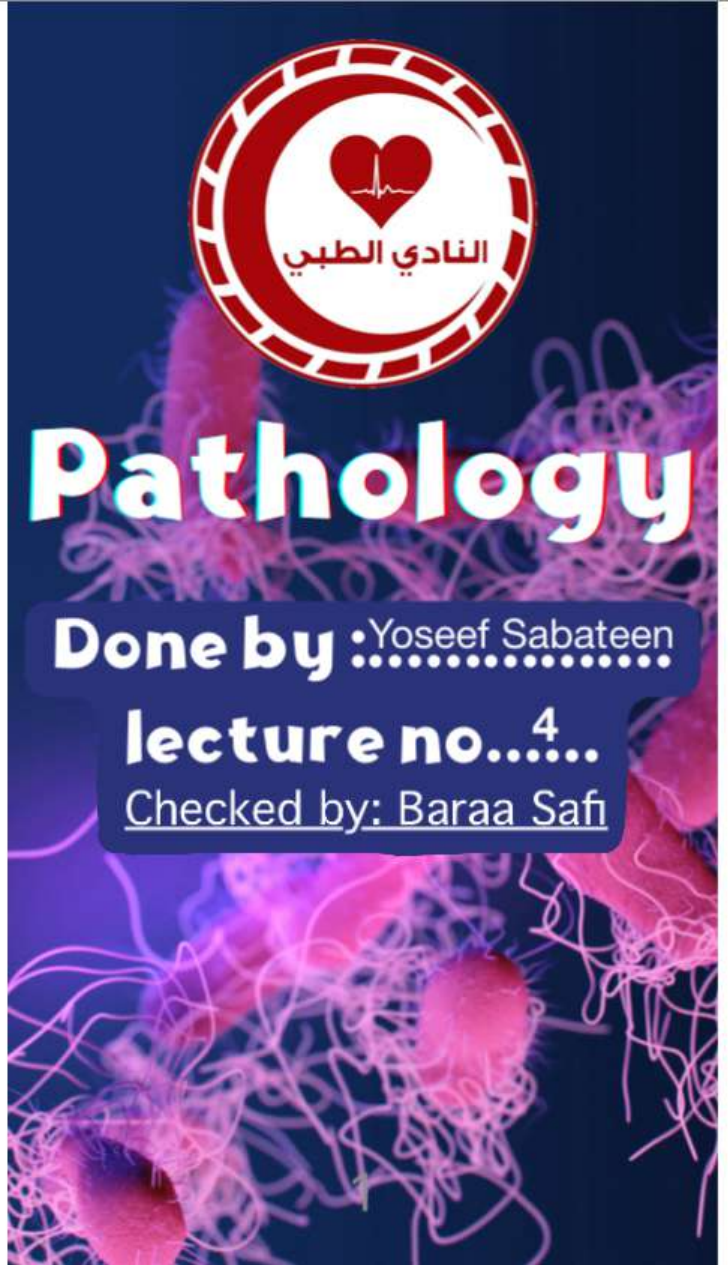

النادي الطبي
Pathology
Done by :Yoseef Sabateen
lecture no...4...
Checked by: Baraa Safi

NEOPLASIA

Dr. Ola Abu Al Karsaneh

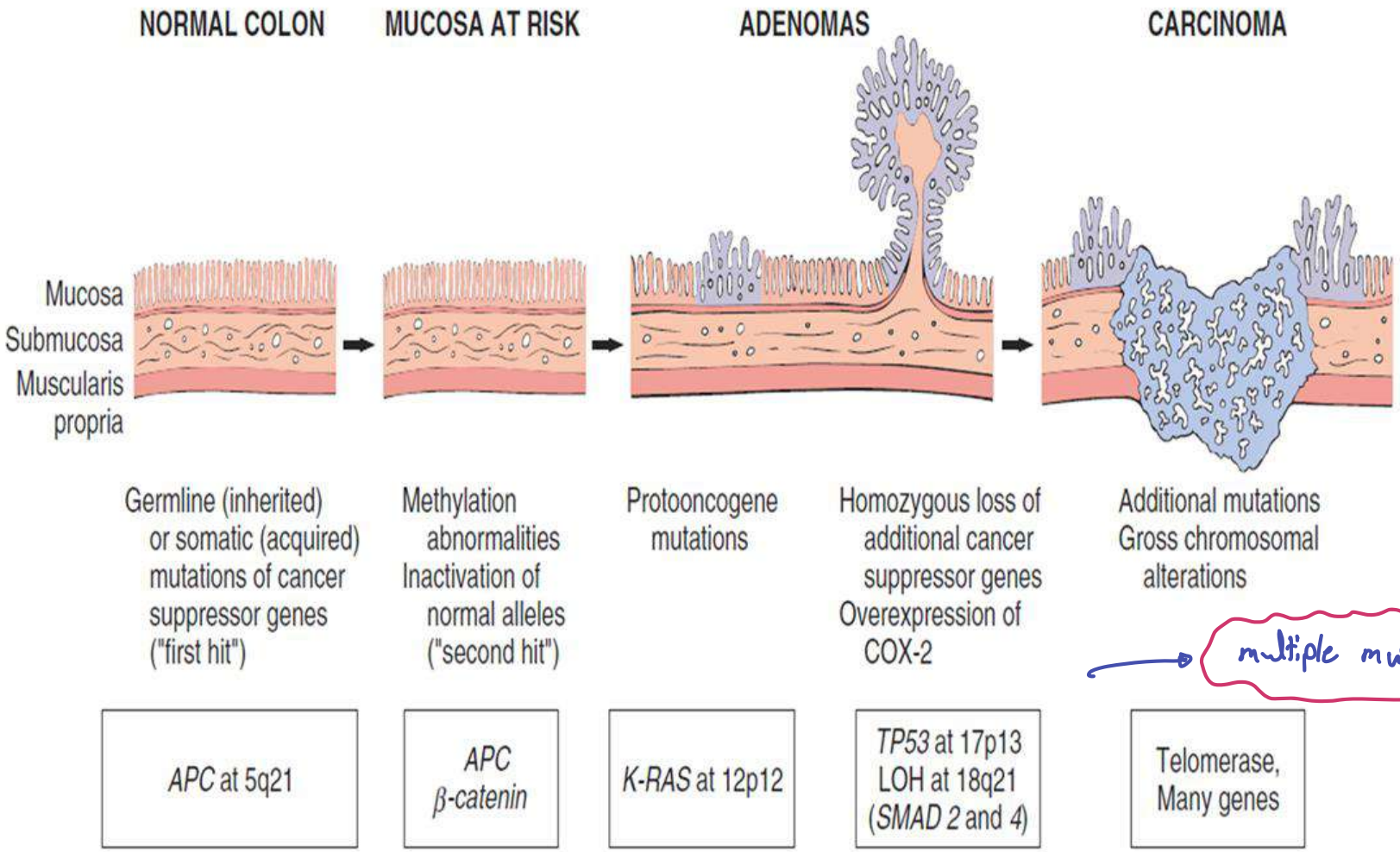
Hashemite University

Carcinogenesis

always

Carcinogenesis is a multistep process resulting from the accumulation of multiple genetic alterations that collectively give rise to the transformed phenotype and all of its associated hallmarks.

In most Cases , no single mutation is sufficient to transform a normal cell into a cancer cell.


NORMAL COLON
MUCOSA AT RISK
ADENOMAS
CARCINOMA
Mucosa
Submucosa
Muscularis propria
Germline (inherited) or somatic (acquired) mutations of cancer suppressor genes ("first hit")
Methylation abnormalities
Inactivation of normal alleles ("second hit")
Protooncogene mutations
Homozygous loss of additional cancer suppressor genes
Overexpression of COX-2
Additional mutations
Gross chromosomal alterations
multiple mutations
APC at 5q21
APC
β-catenin
K-RAS at 12p12
TP53 at 17p13
LOH at 18q21
(SMAD 2 and 4)
Telomerase,
Many genes

Tumor Progression: → after cells are transformed

This is the stepwise accumulation of mutations resulting in increasing features of malignancy:

- More Aggressive
- Less responsive to therapy

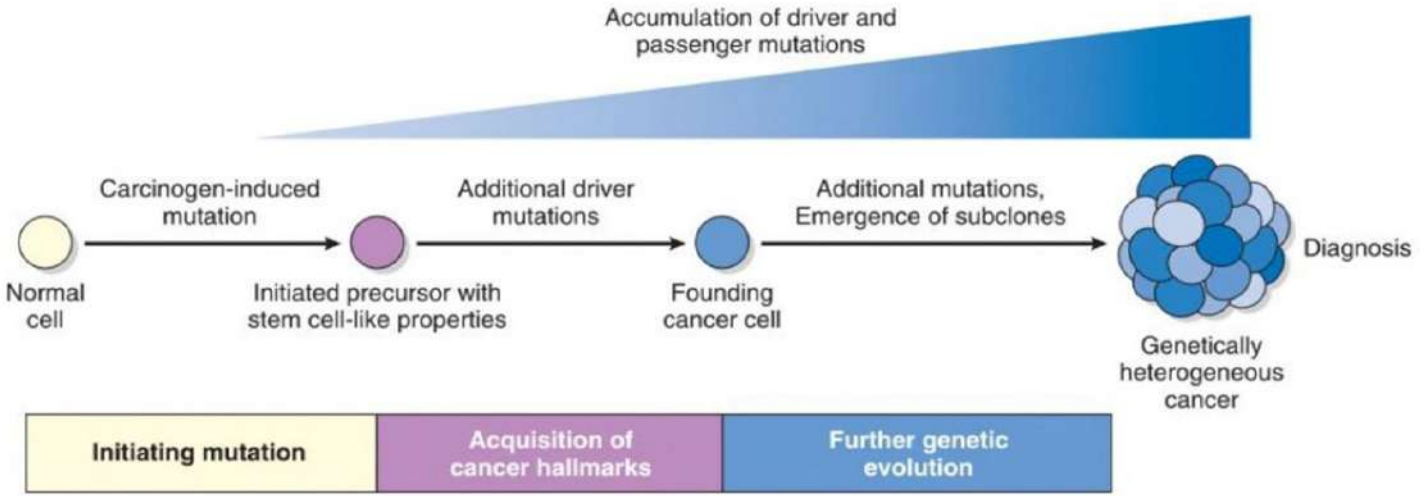
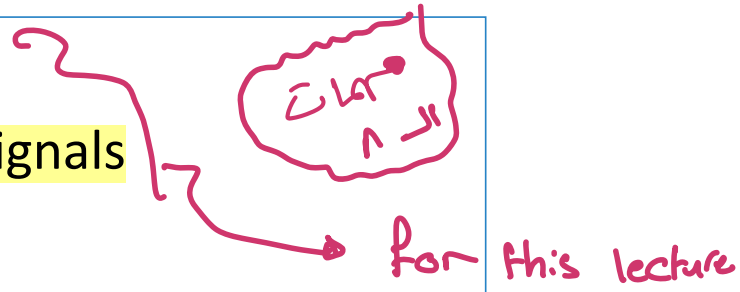


Fig. 6.16 Development of cancer through stepwise accumulation of complementary driver mutations. The order in which various driver mutations occur is usually unknown and may vary from tumor to tumor.

Hallmarks of Cancer

- All cancers appear to display eight fundamental changes in cell physiology and two enabling factors (genomic instability and tumor-promoting inflammation) that promote cellular transformation and subsequent tumor progression.

1. Self-sufficiency in growth signals
2. Insensitivity to growth-inhibitory signals
3. Altered cellular metabolism
4. Evasion of apoptosis
5. Limitless replicative potential (Immortality)
6. Sustained angiogenesis
7. Ability to invade & metastasize
8. Evasion of immune surveillance



1. Self-sufficiency in growth signals

↳ mutation → gain function

- Gain of function mutation in Genes coding for growth: Classified by the site of action

↳ oncogenes ↑

- Proto-oncogenes: Normal.
- Oncogenes: Mutant/overexpressed
- oncogenes ⟹ oncoproteins (promote cell growth, even without normal growth-promoting Signals).
- They include genes coding:

1. Growth factors
2. Cell surface receptors
3. Signal transduction proteins
4. Nuclear transcription factors
5. Cell cycle proteins
6. Inhibitors of apoptosis

activate receptors

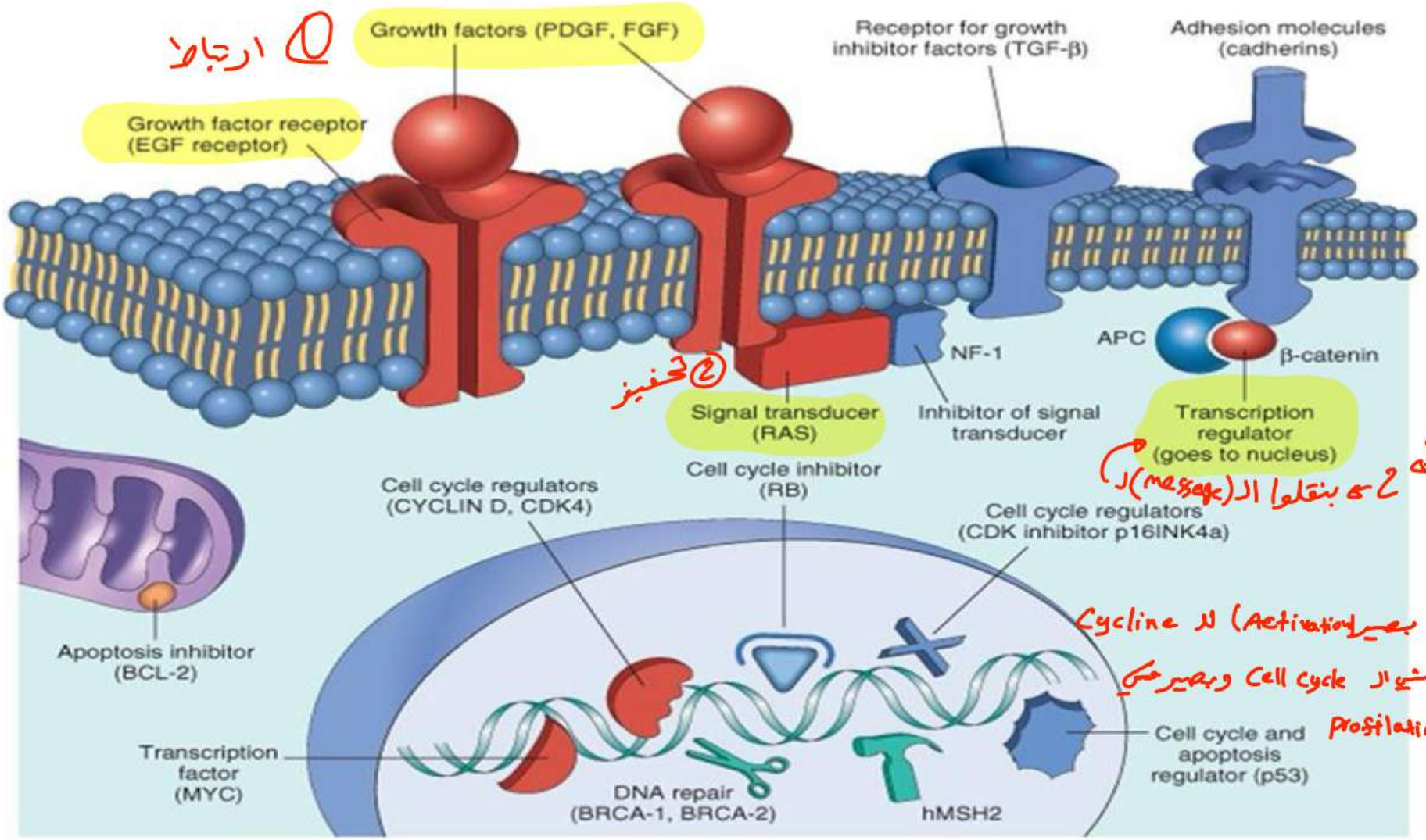
sends signals inside cells

into the nucleus to activate the transcription of the genes

expression of proteins

can either → inhibit apoptosis / tumor suppressor genes

↳ over expression of proto-oncogenes


Growth factors (PDGF, FGF)
Receptor for growth inhibitor factors (TGF-β)
Adhesion molecules (cadherins)
Growth factor receptor (EGF receptor)
NF-1
APC
β-catenin
Signal transducer (RAS)
Inhibitor of signal transducer
Transcription regulator (goes to nucleus)
Cell cycle regulators (CYCLIN D, CDK4)
Cell cycle inhibitor (RB)
Cell cycle regulators (CDK inhibitor p16INK4a)
Apoptosis inhibitor (BCL-2)
Transcription factor (MYC)
DNA repair (BRCA-1, BRCA-2)
hMSH2
Cell cycle and apoptosis regulator (p53)
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1- Oncogenes coding Growth Factors

- Some cancers acquire the ability to synthesize the same growth factors (GF) to which they are responsive (autocrine) or send signals to activate normal cells in the supporting stroma to synthesize the same GF to which they are responsive (paracrine).
  - Platelet-derived growth factor (PDGF) seen in glioblastomas
  - Transforming Growth Factor (TGF- α) in sarcomas
- Products of other oncogenes (e.g. RAS) may cause overexpression of GF.

* Auto crine → parenchymal cells will stimulate itself → by a GF

* para crine → parenchymal cells will stimulate the stromal cells
→ stromal cells will stimulate the parenchymal cells → by the same GF

2- Oncogenes coding Growth Factor Receptors

- **Many of the growth factor receptors function as oncoproteins when they:**

- Mutant receptor ⟶ continuous signals even in the absence of GF…

Or Normal but overexpressed ⟹ hypersensitive to GF…

- Epidermal GF receptor family:

ERBB1 overexpressed in sq. CA lung (1) & Glioblastoma (2)

ERBB2 (Her2) amplified in breast Ca (1), less lung (2), ovary (3), stomach ca (4) → Can use Treatment that inhibits it

- Increase = POOR PROGNOSIS

3- Oncogenes in signal transduction

- The signals are transmitted to the nucleus through various signal transduction molecules.

- Two important oncoproteins in the category of signaling molecules:

1. RAS

2.ABL

g protein Activity

1. RAS

- RAS proteins **are inactive when bound to GDP**
- Stimulation of cells by growth factors: exchange of GDP for GTP and generate active RAS.

to stop the stimulase

- Intrinsic guanosine triphosphatase (GTPase) of RAS hydrolyzes GTP to GDP, releasing a phosphate group and returning RAS to its quiescent GDP-bound state.
- The GTPase protein is magnified dramatically by a family of GTPase-activating proteins (GAPs).

activates GTpase

لازم ترجعه لل (inactive form)

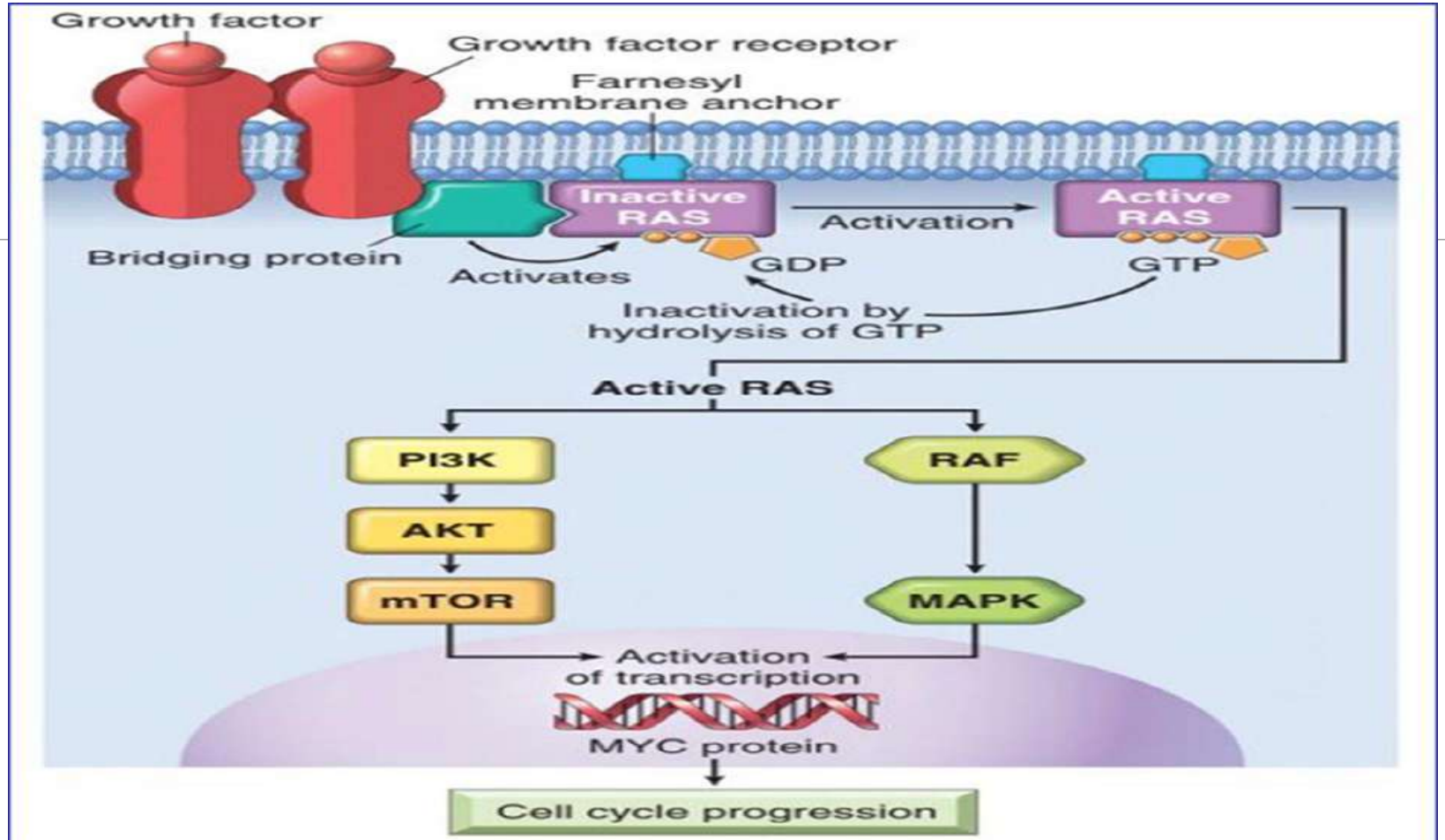


Fig. 6.18 Model for action of RAS. When a normal cell is stimulated through a growth factor receptor, inactive (GDP-bound) RAS is activated to a GTP-bound state. Activated RAS transduces proliferative signals to the nucleus along two pathways: the so-called "RAF/ERK/MAP kinase pathway" and the *PI3 kinase/AKT pathway*. *GDP*, Guanosine diphosphate; *GTP*, guanosine triphosphate; *MAP*, mitogen-activated protein; *PI3*, phosphatidylinositol-3.

- Active RAS → Signal transduction (RAF/MAP-K or PI3-K/AKT pathways) → transcription activation → ↑ proliferation

- RAS most commonly is activated by point mutations in amino acid residues that are either within the GTP binding pocket or in the enzymatic region that carries out GTP hydrolysis

(GTP لازم يضل يحس أنه مرتبط فيه اذا) (GTPase لازم يحس أنه مرتبط فيه اذا)

-Activated RAS stimulates downstream regulators of proliferation by several interconnected pathways.

↑ RAS → ↑ proliferation

- Commonest oncogene mutation in human tumors.

- Point mutations in codons 12, 13 are present in 30% of cancers, especially CA pancreas & Colon.

2. ABL:

normal function → negatively regulated when proliferation is inhibited

- Non-receptor tyrosine kinase function as signal transduction molecule
- The ABL proto- oncogene has tyrosine kinase activity dampened by internal negative regulatory domain

- Chronic myeloid leukemia: t(9;22) ⟹ BCR-ABL hybrid gene explained in lecture (3)
- This new gene protein is retained in the cytoplasm where it has tyrosine kinase activity activates all of the signals downstream of RAS ⟹ cell proliferation

Cell cycle ← control يعملوا

(Cyclins الـ على (control) يعملوا)

4- Nuclear Transcription Factors:

→ Signal transduced from cytosol → Nucleus

(activating expression Molecules)

- DNA transcription regulated by genes e.g. MYC, JUN,....etc. that regulate the expression of growth-promoting genes, such as cyclins ⟹ initiation of cell cycle
- MYC mutation ⟹ sustained activation
- Examples:
  - Dysregulation of *MYC* in Burkitt lymphoma
  - *MYC* amp. in Breast, lung CA.

Cyclins : بفعل شغال

Cell cycle : sustained profilation

ما حكتبهم

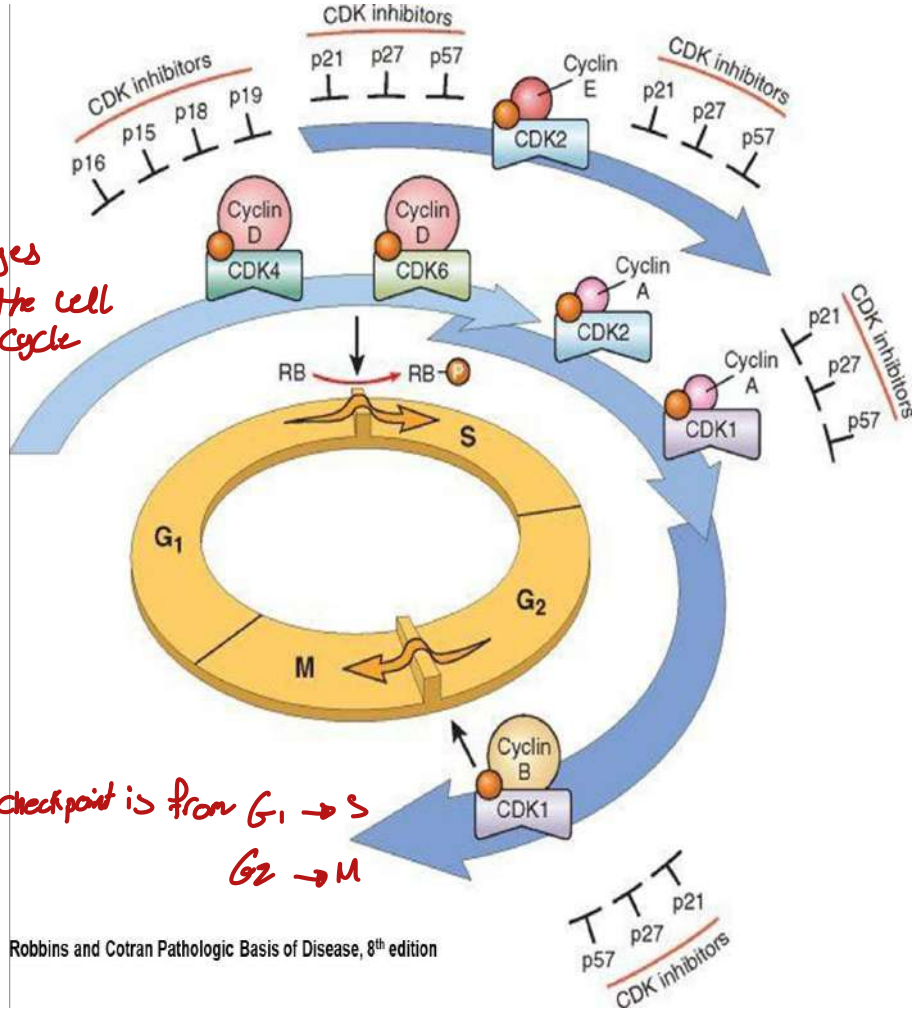
5- Cyclins & Cyclin Dependent – Kinases regulate cell cycle phases

↳ Cyclin → proteins that are at different concentration at at different stages of the cell cycle

- Family of proteins that control entry of the cells at specific phases of cell cycle (D, E, A, B….etc.)

  Cyclins

- Level of a specific cyclin increases at a specific phase, then decreases rapidly after the cell departs that phase
- Function by **phosphorylating certain proteins (e.g. RB protein)**
- Cyclins bind to CDKs, activating them



↳ negative regulators of Cyclin /CDK

- CDK inhibitors regulate the activity of CDK/ Cyclin.

- Selective or nonselective inhibition.

↳ work on a specific one

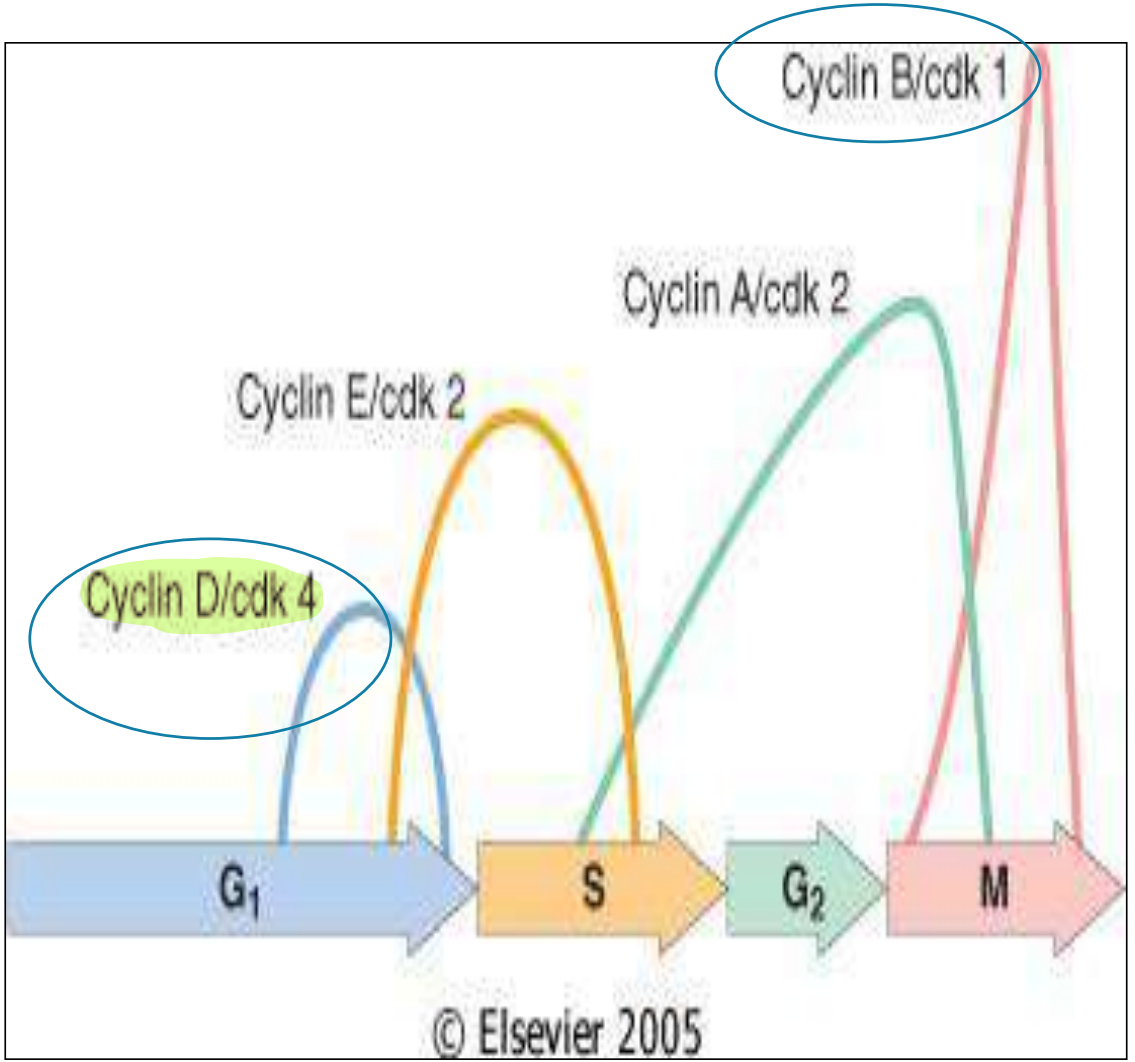
- Examples: p21, p27 & p57 inhibit all CDKs while INK4 Inhibitors (p15, p16, p18 & p19) inhibit CDK4 & CDK6.

(Non-selective الباقي)

- The tumor suppressor protein p53 controls expression of p21.

Two important checkpoints, each of which is tightly regulated by a balance of growth-promoting and growth-suppressing factors, as well as by sensors of DNA damage:

- **G1 - S phase checkpoint:** Cyclin D family---CDK4 & CDK6
- **G2 - M transition:** Cyclin B-CDK1 activate



* reason why these are the most imp. checkpoints

- Once cells pass through the G1/S checkpoint, they are committed to undergo cell division.

- Then, defects in the G1/S checkpoint are particularly important in cancer since these lead directly to increased cell division.

- All cancers appear to have genetic lesions that disable the G1/S checkpoint, causing cells to continually reenter the S phase.

معنى موجودة بسلايد اتنا خليت هاد السلايد حتيته
مهم كشرح الفكرة

Mutations that dysregulate activity of cyclins & CDKs → Gain-of-function mutations and cell proliferation:

e.g.: Cyclin D is overexpressed in breast → passes G1 → S checkpoint

Loss-of-function mutations involving CDKIs ⟹ cell proliferation:

- Disabling mutations of CDKN2A (encoding p16): germline (in melanoma)
- Acquired deletion or inactivation of *CDKN2A* is seen in pancreatic carcinomas, glioblastomas, esophageal cancers

↳ p16

as we said → Multi step process → oncogene ↑
tumor suppressor gene ↓

A final consideration :

- The increased production of oncoproteins does not by itself lead to a sustained proliferation of cancer cells.

- There are two built-in mechanisms, **cell senescence and apoptosis**, that oppose oncogene-mediated cell growth.

- Therefore, genes that regulate these two braking mechanisms must be disabled to allow the action of oncogenes to proceed unopposed.

2. Insensitivity to growth-inhibitory signals

- **Disruption in Cancer Suppressor Genes**

- Growth inhibitory pathway by:

RB gene: Regulate cell cycle

TP53 gene: Regulate cycle & apoptosis

TGF- β: Block GF signals → Anti proliferative action

APC gene: regulates β –catenin

* 2 Alleles must be Mutated / deleted for inhibition of tumor suppressor gene

لحتى أي (Tumer suppressor gene) يبطل يشتغل لازم الـ (two copies) للجينه يكونوا معه شغالين سواء كانه المرض وراثي أو لا

لو الـ (copy) الأول خرب الثاني بشتغل بداله

1- *RB* gene (Governor of cell cycle):

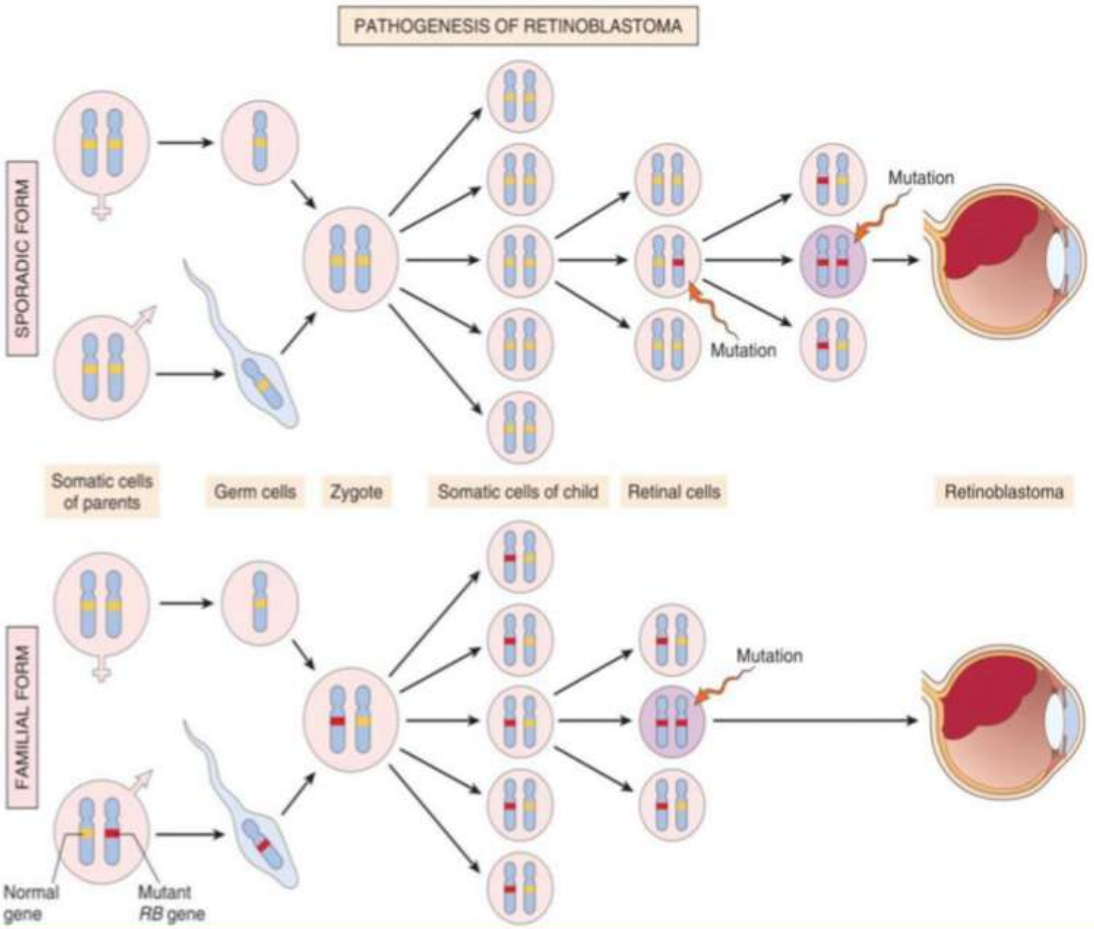
- The first tumor suppressor gene to be discovered
- First studied in Retinoblastoma
- *RB* gene ⟹ RB protein
- Both copies of gene must be lost for neoplastic transformation to occur

→ both Mutated

- This is called loss of heterozygosity
- Familial (*RB* ➡ *RB*) or
- Sporadic (*RB* ➡ *RB* ➡ *RB*)

red → normal

blue → Mutated / defected



Mode of action of *RB* gene:

- The function of the RB protein is to regulate the G1/S checkpoint, the portal through which cells must pass before DNA replication commences.

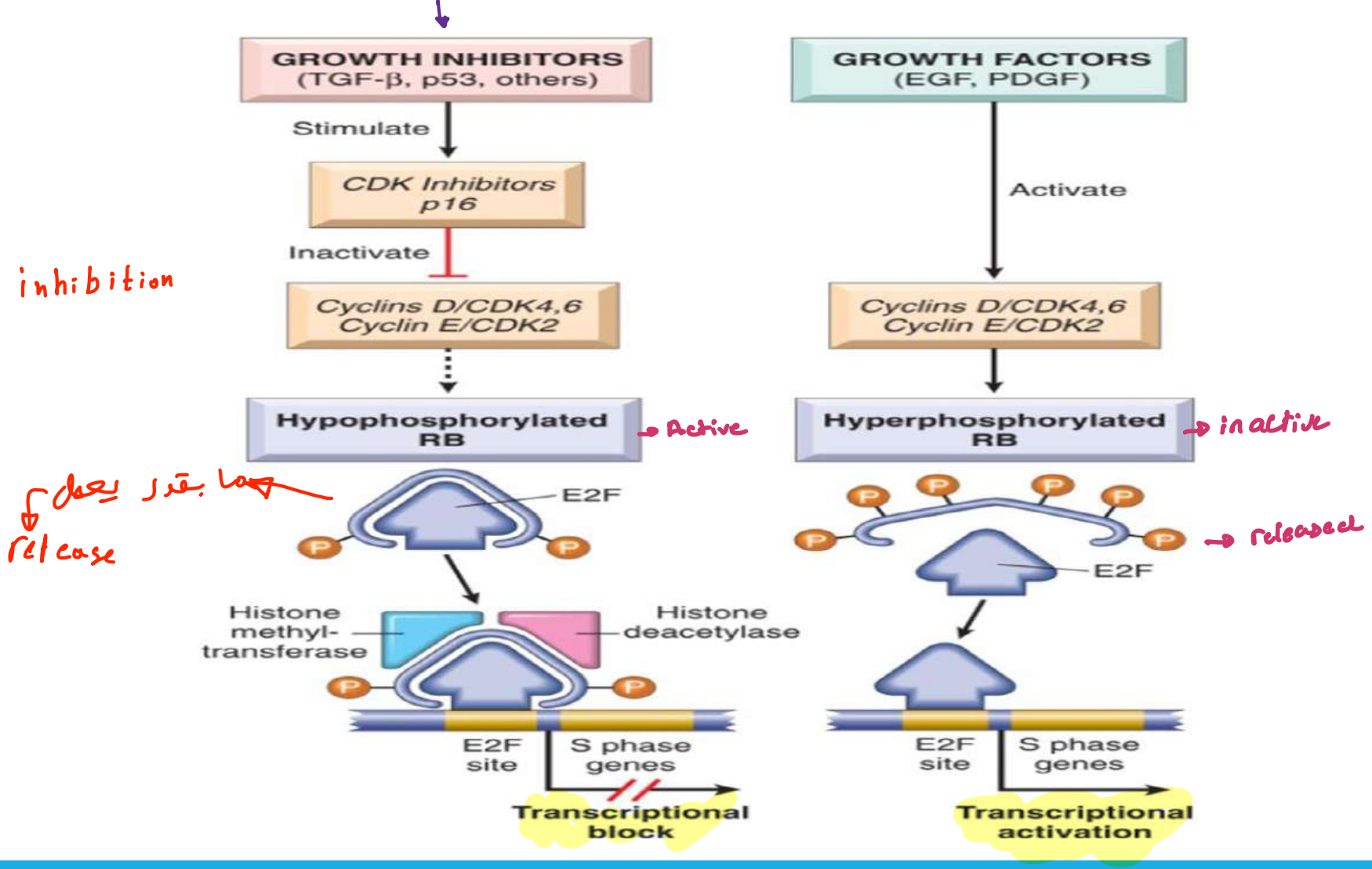
- G1 – S transition and DNA replication require the activity of cyclin E/CDK2

- Cyclin E is dependent on the E2F family of transcription factors

- Active hypophosphorylated RB binds to & inhibits the E2F family of transcription factors --- NO TRANSCRIPTION of cyclin E

Growth factor signaling leads to cyclin D expression and activation of cyclin D- CDK4/6 complexes, which phosphorylate RB, inactivating the protein and releasing E2F ⟶ induce target genes such as cyclin E. ⟶ TRANSCRIPTION (G1 -- S phase)

DNA DAMage


GROWTH INHIBITORS
(TGF-β, p53, others)
Stimulate
CDK Inhibitors
p16
Inactivate
Cyclins D/CDK4,6
Cyclin E/CDK2
Hypophosphorylated
RB
Active
E2F
Histone
methyl-
transferase
Histone
deacetylase
E2F
site
S phase
genes
Transcriptional
block
GROWTH FACTORS
(EGF, PDGF)
Activate
Cyclins D/CDK4,6
Cyclin E/CDK2
Hyperphosphorylated
RB
inactive
released
E2F
E2F
site
S phase
genes
Transcriptional
activation

Retinoblastoma:

- Sporadic in 60% of cases
- Familial (40%), AD
- In familial form, patients carry one mutation in their genome, followed by a second mutation in retinal cells
- No tumor develops unless two alleles in chr. 13q14 become mutant (two hit theory)
- Familial form: ↑ incidence of bilateral ret., osteosarcoma, and other tumors

↳ only one mutation is needed

police man

2- TP53 (Guardian of Genome)

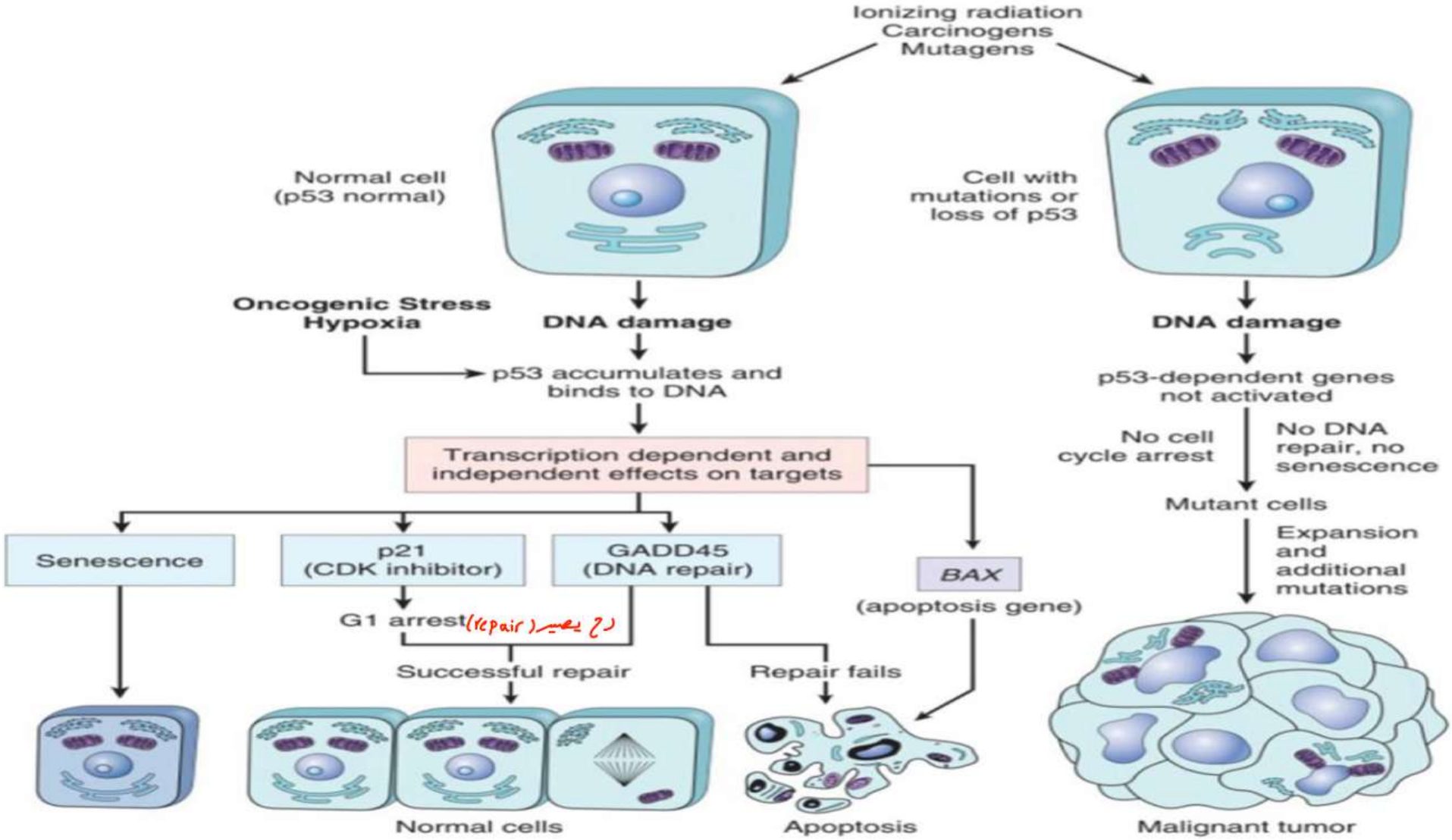
- **The most commonly mutated gene (and suppressor gene) in human cancer.**
- Homozygous loss in 70% of cancers
- *TP53* is a negative regulator of the cell cycle (protein product is p53).
- 'Guardian of the Genome' OR (Policeman) preventing genetically damaged cells from progressing through a new cycle.
- p53 is inactivated by MDM2.
- Upon DNA damage or other stresses, various pathways will dissociate the p53 and MDM2 complex.

Mode of activation & action:

- p53 senses DNA damage or other stresses through various sensors, like protein kinases e.g. Ataxia telangiectasia mutated (ATM) protein → phosphorylates p53 → prevent degradation by MDM_2
- p53 released from MDM2 & activated with longer half-life →
  - Transcription of CDKI gene *CDKN1A* (p21) → cell cycle arrest at G1 (Quiescence) --- Result: more time for repair --- Normal (inactive)

OR → GADD45 / PCNA → for repair

- If repair fails ----Senescence (permanent cell cycle arrest) or Apoptosis(p53 is a positive regulator of apoptosis (*BAX* & *PUMA*). (BAK)
- **OR** *Fixed mutation* --- ***NEOPLASIA***


Ionizing radiation
Carcinogens
Mutagens
Normal cell
(p53 normal)
Cell with
mutations or
loss of p53
Oncogenic Stress
Hypoxia
DNA damage
DNA damage
p53 accumulates and
binds to DNA
p53-dependent genes
not activated
No cell
cycle arrest
No DNA
repair, no
senescence
Transcription dependent and
independent effects on targets
Mutant cells
Expansion
and
additional
mutations
Senescence
p21
(CDK inhibitor)
GADD45
(DNA repair)
BAX
(apoptosis gene)
G1 arrest(repair) راح يصير
Successful repair
Repair fails
Normal cells
Apoptosis
Malignant tumor

70% of Cancers

Most common mutant of tumor suppressor gene for cancer

Significance of TP53 mutation:

- Acquired mutation in many cancers

  e.g. colon, breast, lung, …etc

- Inherited mutation in one allele---

  Li-Fraumeni syndrome – 25-fold ---malignancy: sarcoma, breast carcinoma, brain tumors ….. etc

↳ Compared to general population

3- Transforming Growth Factor-β (TGF-B):

A potent inhibitor of proliferation (Antiproliferative activity): -

Act by binding to a complex composed of TGF-β receptors I and II resulting in the transcriptional activation of **CDKIs** with growth-suppressing activity and **repression** of growth-promoting genes.

- Mutations may alter the **type II TGF-β receptor.**

-Mutational inactivation of TGF-β components seen in a very high percent of pancreatic carcinoma & the majority of colonic CA

4- Contact Inhibition, APC:

all junctions attached by cadherins

- Cell–cell contacts in many tissues are mediated by homodimeric interactions between transmembrane proteins called **cadherins**.

junctions

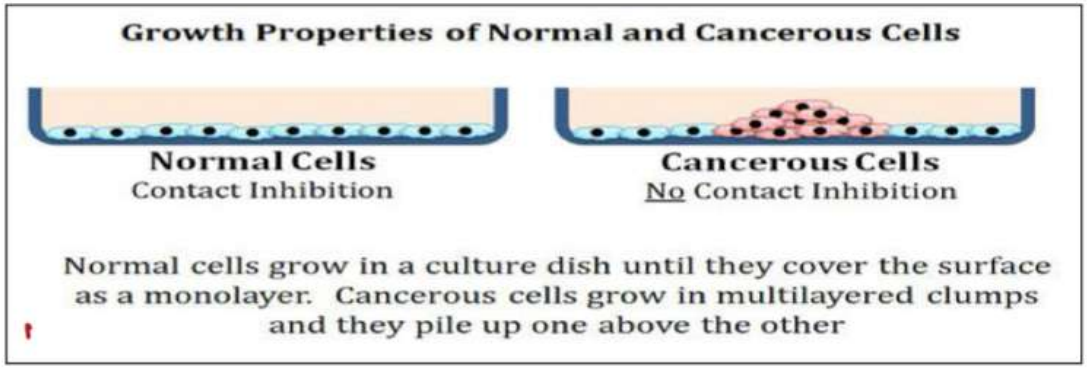
- **E-cadherin** (E for epithelial) mediates cell–cell contact in epithelial layers.

- **Contact inhibition** enables noncancerous cells to cease (وقف) proliferation and growth when they form confluent monolayers and **contact** each other.

- **Two mechanisms have been proposed to explain how E-cadherin maintains contact inhibition:**

1-Tumor suppressor gene *NF2*

2- APC gene and β-catenin (**a key component of the WNT signaling pathway**).



-This characteristic is lost when cells undergo malignant transformation, leading to uncontrolled proliferation and solid tumor formation.

By:

-E-cadherins are reduced in many cancers.

all explained on the picture below

- Mutant APC.

- β-catenin overfunctioning

ACTION OF APC GENE:

- Gene product is a cytoplasmic protein that acts in adhesion by regulating the destruction of **β-catenin** in the cytoplasm.

- In quiescent cells that have not been exposed to WNT, cytoplasmic β-catenin is degraded by a *destruction complex, so no proliferation of cells occur.*

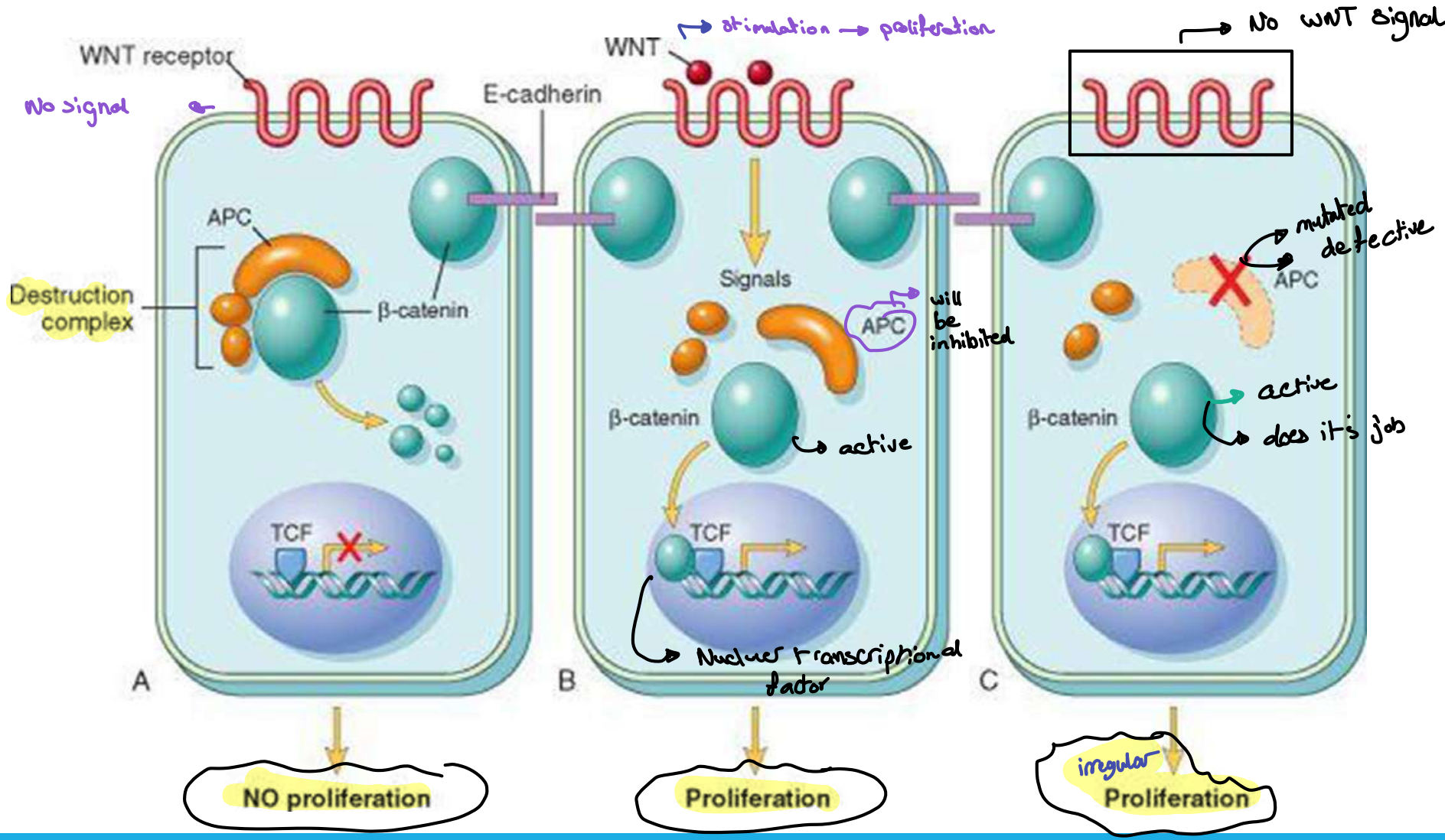
APC + β-catenin + E-Cadherin → **Destruction Complex**

WNT → activates cell proliferation

- β Catenin is also a Nuclear Transcription factor
- it goes to the Nucleus and activated gene expression during proliferation

- With the loss of APC (in malignant cells), β-catenin degradation is prevented, and the WNT signaling response is inappropriately activated in the absence of WNT ⟶ transcription of growth-promoting genes, such as cyclin D1 and MYC, as well as transcriptional regulators, such as TWIST and SLUG, that repress E-cadherin expression and thus reduce contact inhibition and proliferation.

Apc Mutation → Catinin not Degraded → cell will think that WNT signaling is present → ↑ Cyclin/CDK, ↑ Myc, Twist/slug → ↓ E cadherins → ↓ contact inhibition → ↑ cell proliferation


WNT receptor
No signal
E-cadherin
WNT
stimulation → poliferation
No WNT signal
APC
Destruction complex
β-catenin
Signals
APC
will be inhibited
mutated defective
APC
β-catenin
active
β-catenin
active
does it's job
TCF
TCF
TCF
Nuclear transcriptional factor
A
B
C
NO proliferation
Proliferation
irregular
Proliferation

chromosome 5

Clinical significance of APC

Familial Adenomatous Polyposis Coli (FAP)

* people with this mutation. Might have there colon removed as prophylaxis

- AD syndrome.
-Individuals with inherited one mutant allele of APC develop 100s to 1000s of adenomatous polyps by their teens or twenties.

↳ causing cancer/tumor

-Additional mutations → colonic carcinoma (100% ↑↑ risk in familial polyposis coli).

-70-80% of sporadic colonic carcinoma show mutant APC.

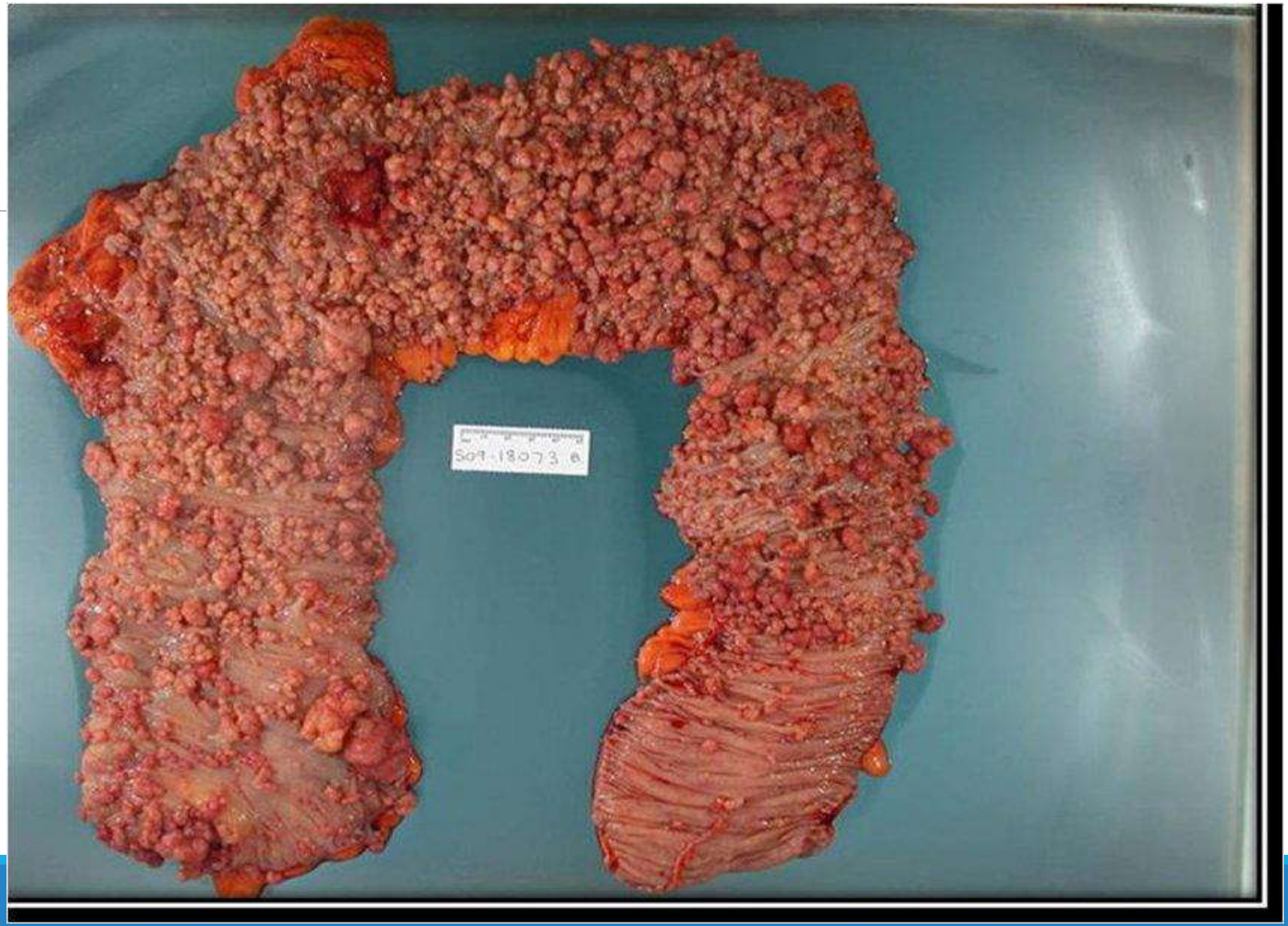
مش وراثية صارت بعد الولادة

-Colonic cancers with normal APC have activating mutations of **β-catenin** that render them refractory to the degrading action of APC.

↳ over expression

- المشكلة بهاد المرض أنه احنا متأكدين أنه المريض بعمر الـ (30) رح يصير عنده (coloncancer) فلازم نشيله

FAP




THANK
YOU