HEMATOPOIETIC & LYMPHATIC SYSTEM

-HAYAT BATCH-

SUBJECT : Pathology

LEC NO. : 5

DONE BY : Anas zakarneh

وَقُل رَّبِّ زِدْنِي عِلْمًا

أقرو دعاء السفر حبايبي😘

- G6PD deficiency اليوم راح نحكي عن
Immune-mediated hemolytic anemia
Hereditary spherocytosis
Paroxysmal nocturnal hemoglobuinuria
MAHA معظمهم بالمحاضرة الخامسة

Intravascular

I. Microangiopathy (MAHA)

II. Acute hemolytic transfusion reaction (ABO mismatch)

III. Paroxysmal nocturnal hemoglobinuria (PNH)

IV. Paroxysmal cold hemoglobinuria (PCH)

V. Infections

VI. Snake bites/venoms

Extravascular

I. Intrinsic RBC defects
- A. Hemoglobinopathies
  - i. Sickle cell
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- C. Enzyme deficiencies
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- A. Immune-mediated hemolytic anemia
  - i. Autoimmune
  - ii. Drug-induced
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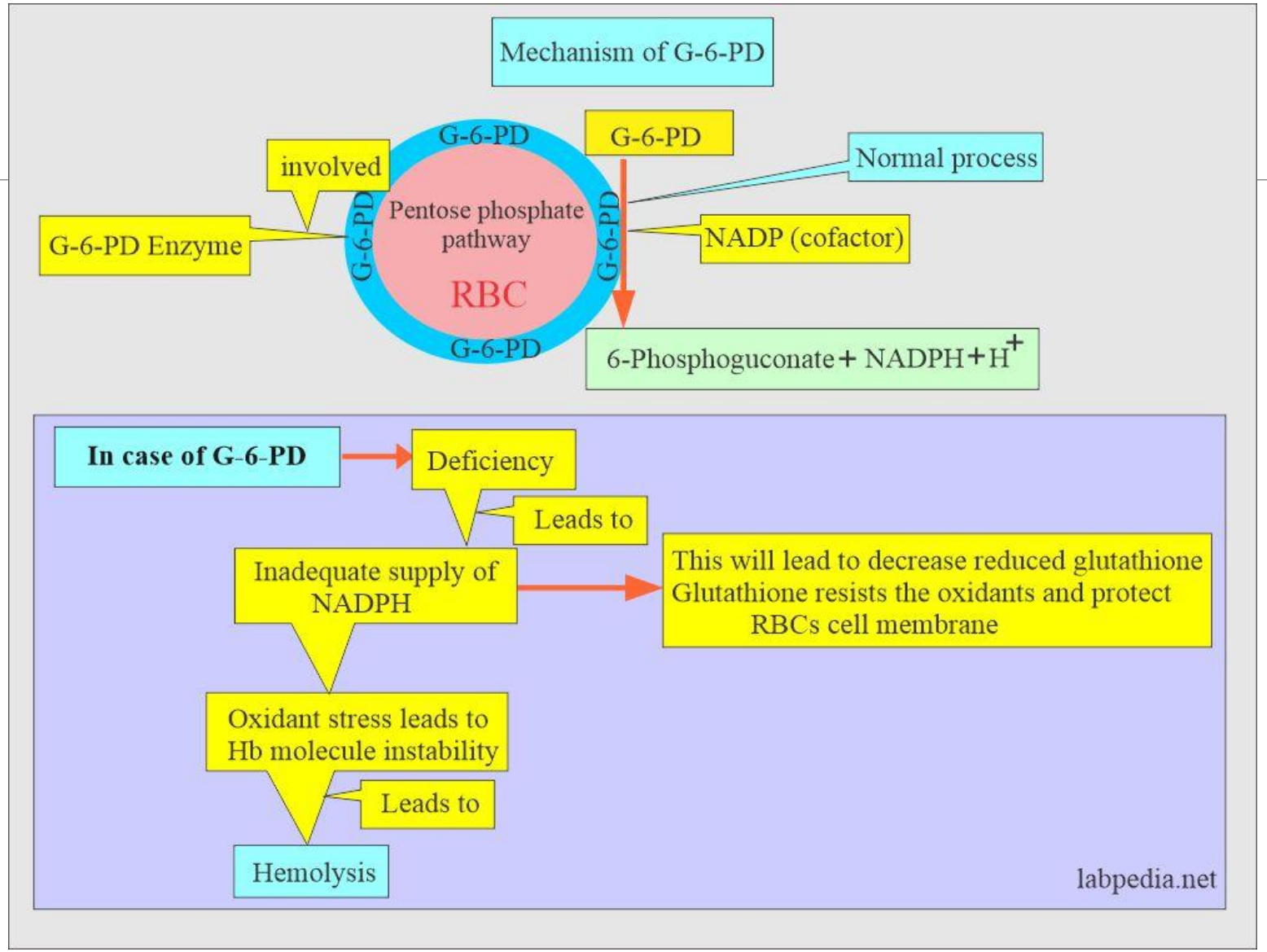
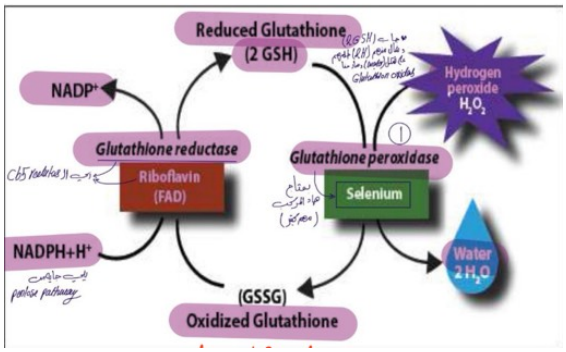
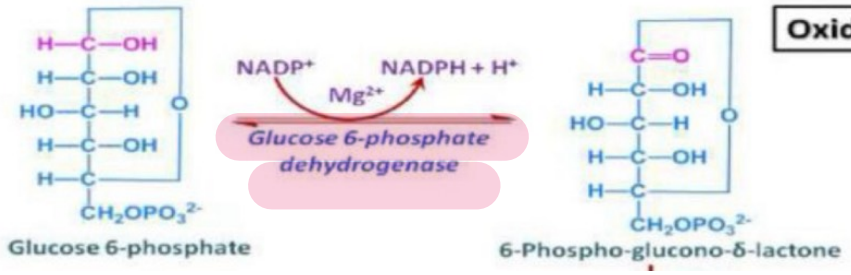
الشرح تحت


Mechanism of G-6-PD
G-6-PD
G-6-PD
involved
Normal process
G-6-PD Enzyme
Pentose phosphate pathway
NADP (cofactor)
RBC
G-6-PD
G-6-PD
G-6-PD
6-Phosphoguconate + NADPH + H+
In case of G-6-PD
Deficiency
Leads to
Inadequate supply of NADPH
This will lead to decrease reduced glutathione
Glutathione resists the oxidants and protect RBCs cell membrane
Oxidant stress leads to Hb molecule instability
Leads to
Hemolysis
labpedia.net

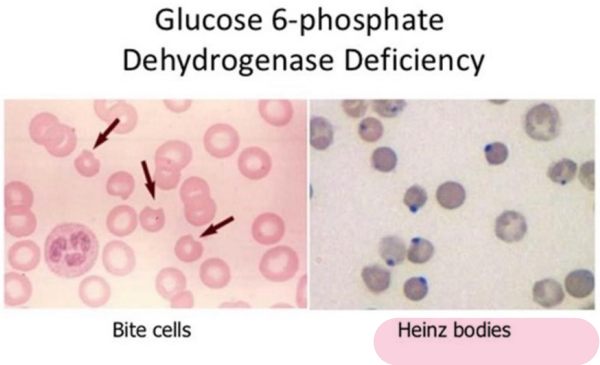
اول اشي الRBC عرضة للoxidative reaction و هاض الاشئ عرفناه بالبيو كيم بسبب وظيفتها في نقل O_2
فمثلا H_2O_2 عامل مؤكسد قوي ...كيف بقدر احمي الRBC منه؟
بكل بساطة عن طريق reduced glutathione
ذاكرين هاض التفاعل ؟
يهال التفاعل بحمي الRBC من H_2O_2 عن طريق
الreduced glutathione اللي بحول الH2O2
إلى water و بعدين هول بتحول إلى الoxidized form
طب هسا فعليا أنا بحاجة إلى الglutathione بالreduced form
و عشان اخليه بالreduced form بحتاج NADPH
طب من وين جبناها ؟
جبناها من pentose phosphate shunt
طيب هاي الshunt بشو بتبلش
يا عيني عليكم داسين بيو كيم منيح
بتبلش بG-6-p
طب لنفرض انو ما عندي G-6-pD(الإنزيم اللي بخليه يكمل بالshunt)
معناته الshunt مش حتكمل
طيب وين المشكلة؟
المشكله انو هاي الshunt بتعطيني NADPh المهم عشان الglutathione
(بحافظ عليه بالreduce form)
طب إذا هاض الإنزيم مش موجود معناته حيقل انتاج الNADPH معناته بطل في اشي يحمي الRBC
و هيك بتروح فيها الحزينة





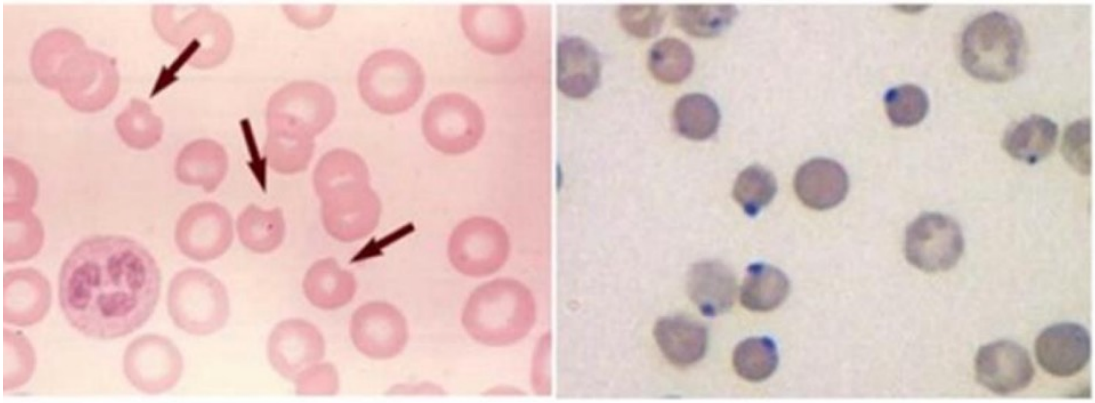
etc.

هسا بهاي الحاله بسبب التعرض إلى oxidative stress و ما عندي
glutathione كافي الHB باخ يصير اله denaturation
و يتجمع على شكل Heinz bodies



متى بصير عندي hemolysis ؟
إذا تجمع عندي RBC بهاض الشكل (فيهم Heinz bodies)
بروحو يمشو بال blood حتى يوصلو ال spleen
هسا هونني عندي احتماليين
١)
ممكن إنها تأكل ال whole RBC فبصير extra vascular hemolysis
2)
ممكن إذا كانت ال Heinz bodies ع الطرف تأكل جزء من ال RBC فبتصير
Bite cell
هاي ال bite cell بترجع لل circulation و بتنفجر فبصير عندي element of intravascular hemolysis
فهون عندي a vascular hemolysis (intra+extr)

Glucose 6-phosphate Dehydrogenase Deficiency



Bite cells

Heinz bodies

Note : we can see Heinz bodies and bite cells in blood smear

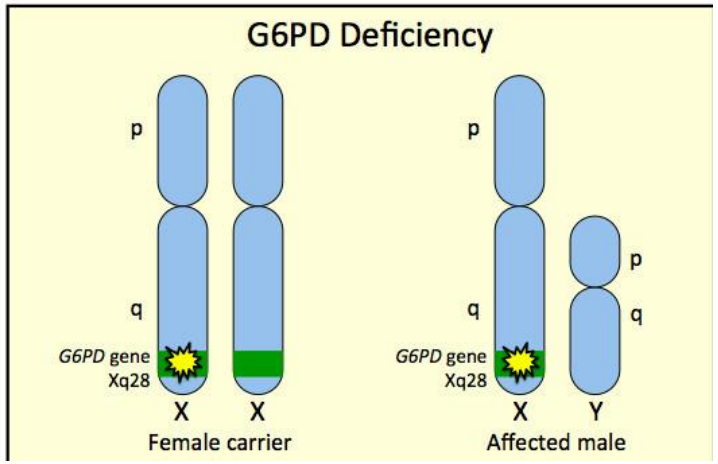
التفول

GDPD Deficiency aka “favism”

- X-linked recessive disorder

So mainly in males

- G6PD is first enzyme in pentose phosphate pathway and is required to make NADPH. NADPH is important to reduce oxidative stress.
- **G6PD deficiency presents as increased oxidative stress including hemolytic anemia.**
- The majority of patients are asymptomatic most of the time and go through life without ever being aware of their genetic trait.
- Hemolysis occurs after a lag of 2-3 days
- Males more vulnerable than female (heterozygous)



GDPD Deficiency aka favism

Hemolysis due to oxidant stress:

- **Drugs: eg. Antimalarials, sulfonamides, furantoins,…etc.**
- **Favism: chickpeas , green peas , all types of beans should be avoided**
- **Infections: produces free radicals**

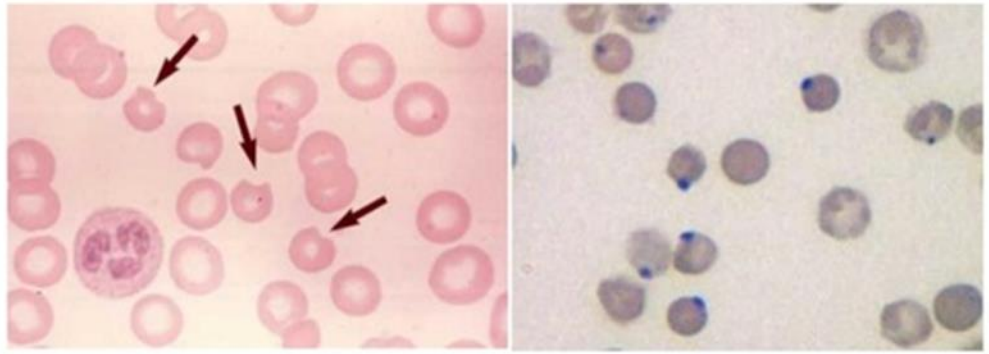
من اسمها.(الفول) بتكون بالأشياء الخضره بالذات

Oxidation leads to denaturation of globin chains, and precipitation at membranes forming **Heinz bodies.**

RBCs: Bite cells and Heinz bodies

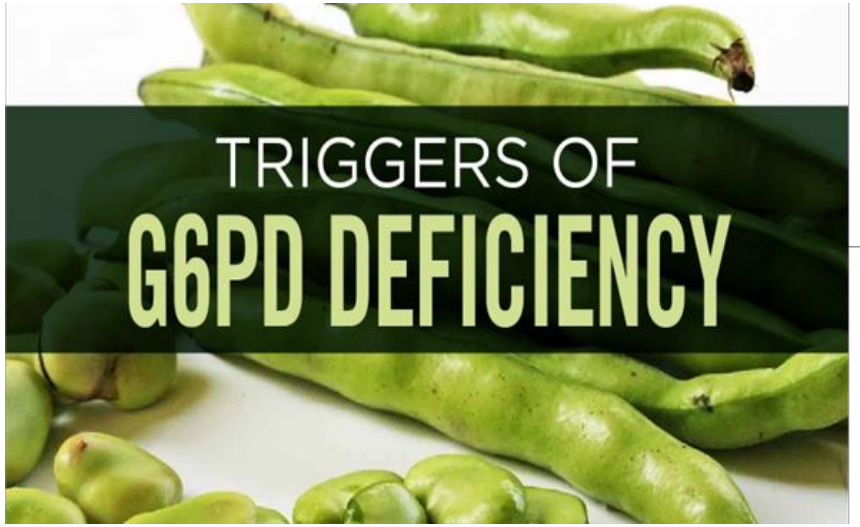
Features of Extra/Intravascular hemolysis

Glucose 6-phosphate Dehydrogenase Deficiency



Bite cells

Heinz bodies



GDPD Deficiency

Diagnosis of G6PD deficiency:

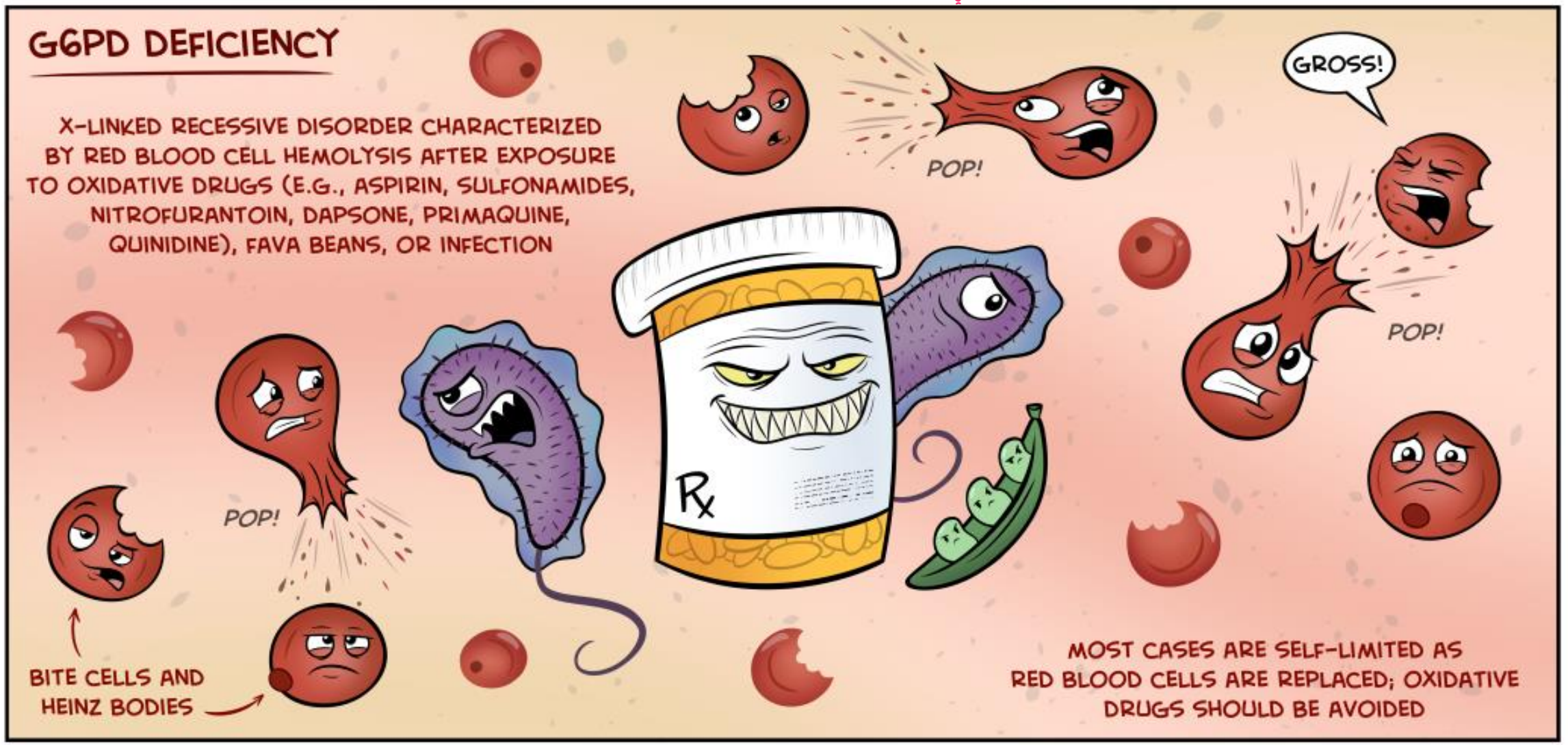
blood smear slide بصبغ فيه الـ

- Screening - Heinz preparation – Blood smear will show Heinz bodies

- Confirm - enzymatic studies (however; in the acute phase, RBCs lacking G6PD are hemolyzed and dead, so they cannot be detected)

ما أجي اعمل detection or confirm ما بعمله بحاله الـ acute phase بكل بساطة
لنفرض مثلا عندي مجموعة من RBCs في منهم normal G-6-PH enzyme و مجموعة عندهم deficiency
كل المجموعة اللي عندهم deficiency باخ تتكسر فهيك بالفحص بيبنو كلهم طبيعية و بيبن انو ما عندي نقص بالإنزيم
لأنو الخلايا اللي فيها نقص تدمرت

الأدوية المطالب حفظهم هي اللي فوق
إذا حاب تحفظهم أنت حر
و أنا مالي



هسا يلا ع ثاني نووووع

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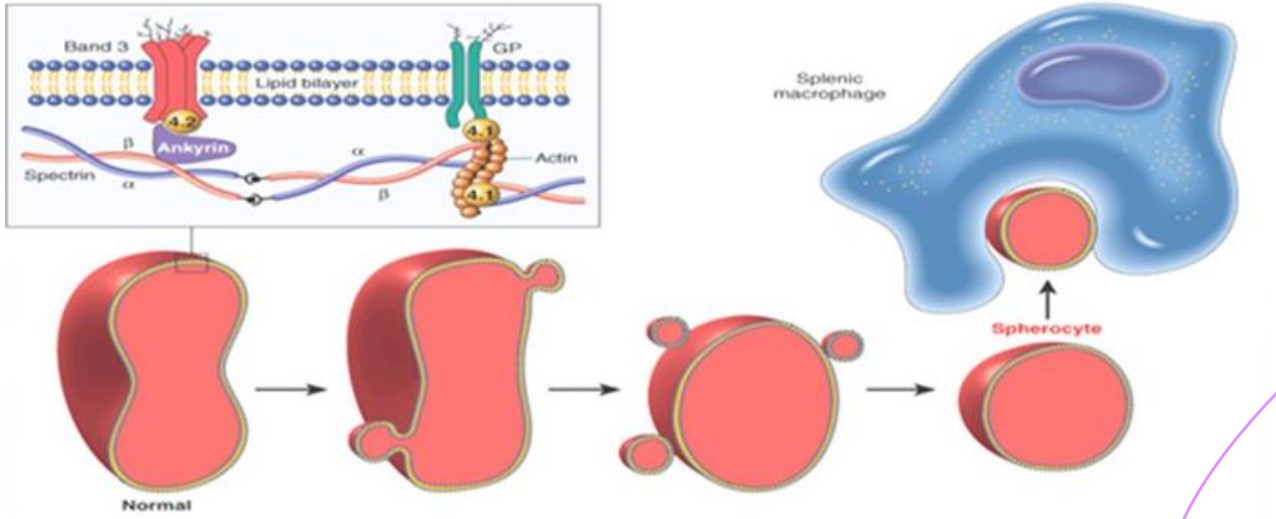
المشكله هون بالبروتين اللي بتحافظ على شكل membrane
فعندي cytoskeleton protein بتربط الRBC membrane بالcytoskeleton جوا الRBC
عندي أكثر من بروتين منهم (ankyrin/spectrin/band 3)
هون بكون عندي abnormal ankyrin and spectrin
فما بقدر أحافظ على شكل الRBC للbiconcave

HEREDITARY SPHEROCYTOSIS



Spectrin and **ankyrin** are tethering proteins that attach RBC cytoskeleton

- ✓**Congenital hemolytic anemia (AD)** due to genetically determined **abnormal spectrin and ankyrin molecules**, leading to defects in red blood cell membrane
- ✓RBC membrane blebs and are lost over time. RBC becomes more spherical.
- ✓Red blood cells become trapped within spleen and have less than usual 120-day lifespan

هسا هون بيصير يطلع عندي blebs وكل مره الspleen بتشيلهم
فبصير شكلها كروي

HEREDITARY SPHEROCYTOSIS

✓Spherocytes are phagocytosed by splenic macrophages , leading to extravascular hemolysis characterized by anemia, jaundice, increased reticulocytes & splenomegaly.

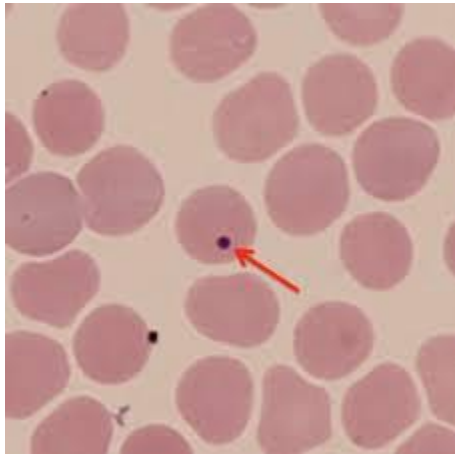
CBC and blood smear findings:

- RBC becomes round instead of disc shaped (loss of central pallor)
- High MCHC - high concentration of hemoglobin as cells are getting small
- Howell-Jolley bodies in peripheral blood RBCs . The Howell-Jolley body is anuclear DNA remnant

لأنو ما صار أي مشكله على HB اكل المشكلة بال membrane

Howell-jolley bodies لأنو بنعمل للمرضى splenectomy باخ نشوف

Howell-Jolly bodies are little fragments of the red cell nucleus. You see them most commonly in patients with splenectomies (normally, the spleen just bites them out). You can see them without a special stain – they look like dark, round dots. Heinz bodies are seen in G6PD deficiency

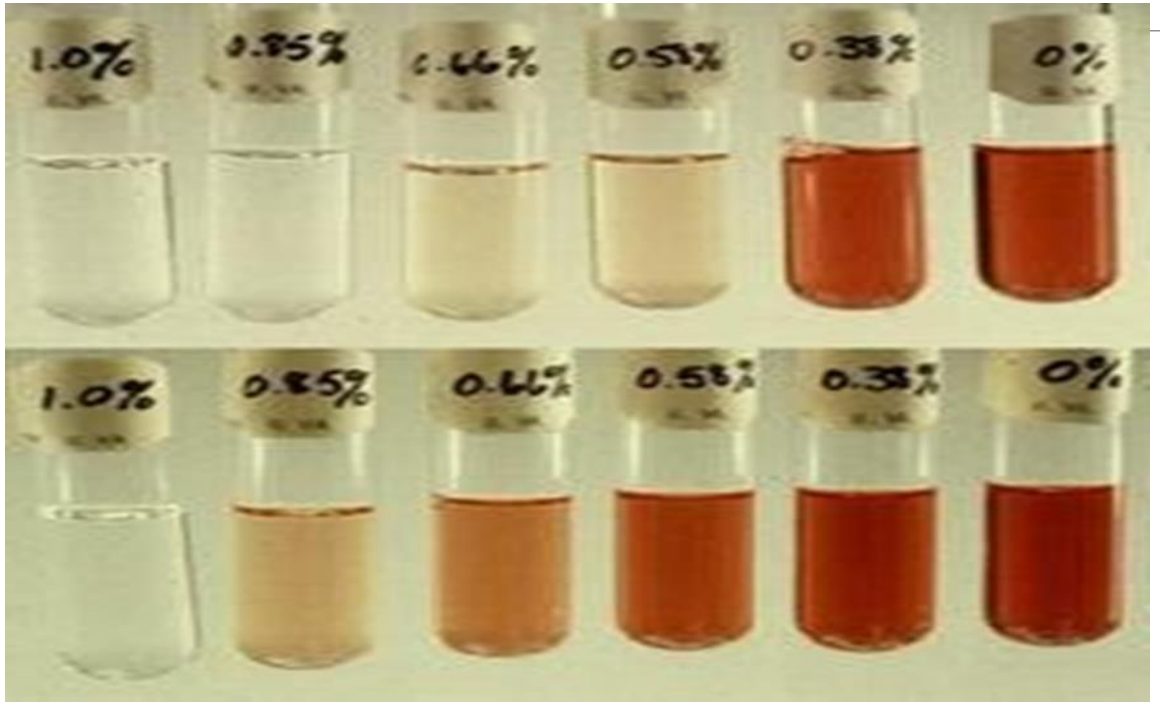


Howell-Jolley body

Treatment: Splenectomy (prolongs survival of red blood cells, although they still have membrane defects)

The osmotic fragility test (OFT) is used to measure erythrocyte resistance to hemolysis while being exposed to varying levels of dilution of a saline solution. When erythrocytes are exposed to a hypotonic environment, water enters the cell and causes swelling and eventual lysis

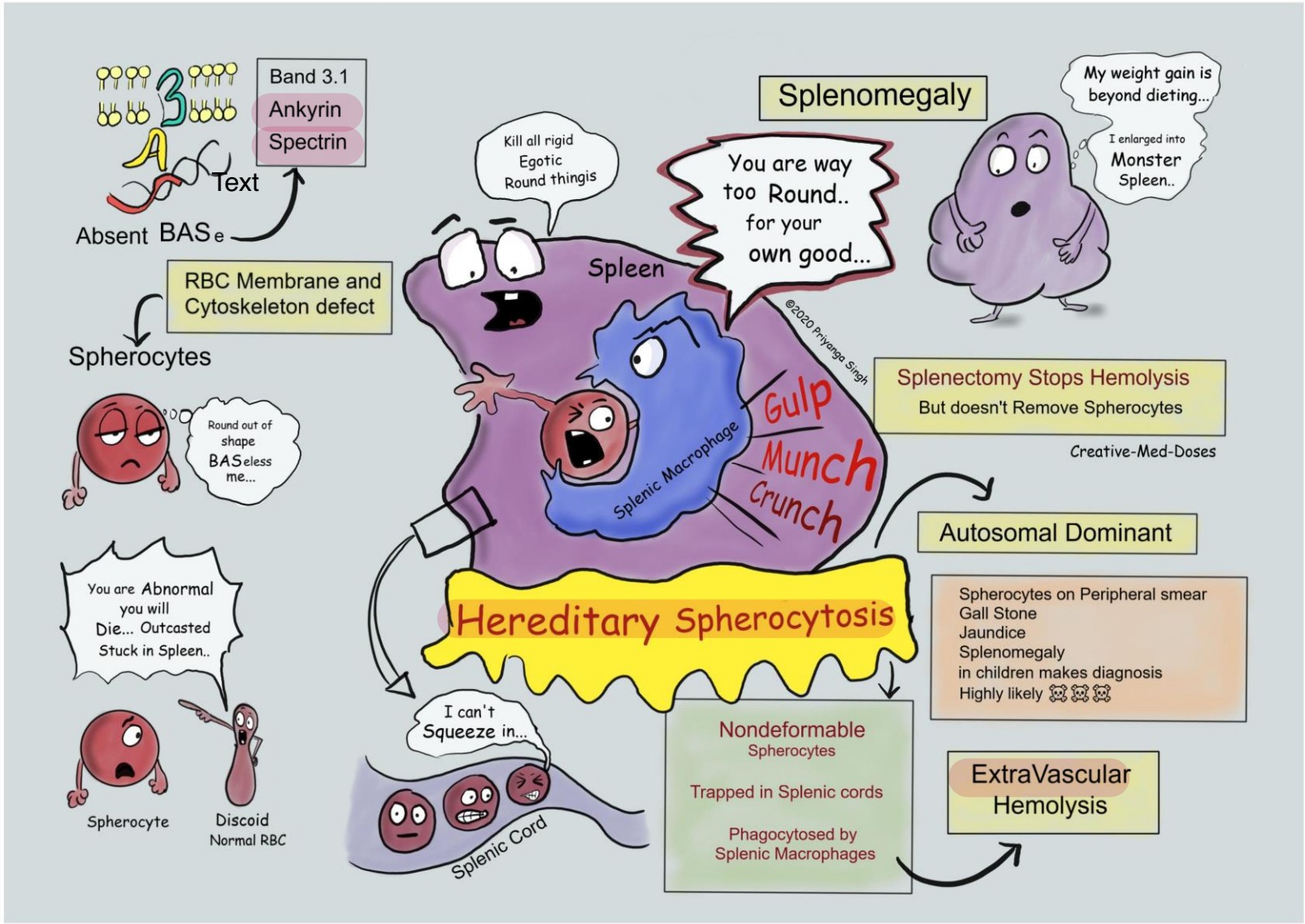
HEREDITARY SPHEROCYTOSIS



كل ما يصير اللون غامق معناته زاد. الhemolysis

Osmotic fragility: increased; basis for diagnostic testing

- The osmotic fragility of red cells is increased i.e . the RBCs are easily hemolysed when kept in a hypotonic saline solution.
- The test consists of exposing RBC to varying strengths of hypotonic saline solutions and measuring the degree of hemolysis colorimetrically at room temperature .


Band 3.1
Ankyrin
Spectrin
Text
Absent BASe
RBC Membrane and Cytoskeleton defect
Spherocytes
Round out of shape BASeless me...
You are Abnormal you will Die... Outcasted Stuck in Spleen..
Spherocyte
Discoid Normal RBC
Kill all rigid Egotic Round thingis
You are way too Round.. for your own good...
Spleen
Splenic Macrophage
Gulp
Munch
Crunch
©2020 Priyanga Singh
Splenomegaly
My weight gain is beyond dieting...
I enlarged into Monster Spleen..
Splenectomy Stops Hemolysis
But doesn't Remove Spherocytes
Creative-Med-Doses
Autosomal Dominant
Hereditary Spherocytosis
Spherocytes on Peripheral smear
Gall Stone
Jaundice
Splenomegaly
in children makes diagnosis
Highly likely
I can't Squeeze in...
Splenic Cord
Nondeformable
Spherocytes
Trapped in Splenic cords
Phagocytosed by Splenic Macrophages
ExtraVascular Hemolysis

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Immune Hemolytic Anemia

AIHA

Antibodies that’s attack RBC

Three broad categories:

Alloimmune: The patient produces alloantibodies to foreign red cell antigens (transfusion, pregnancy, or organ transplant)

مش موضوعنا

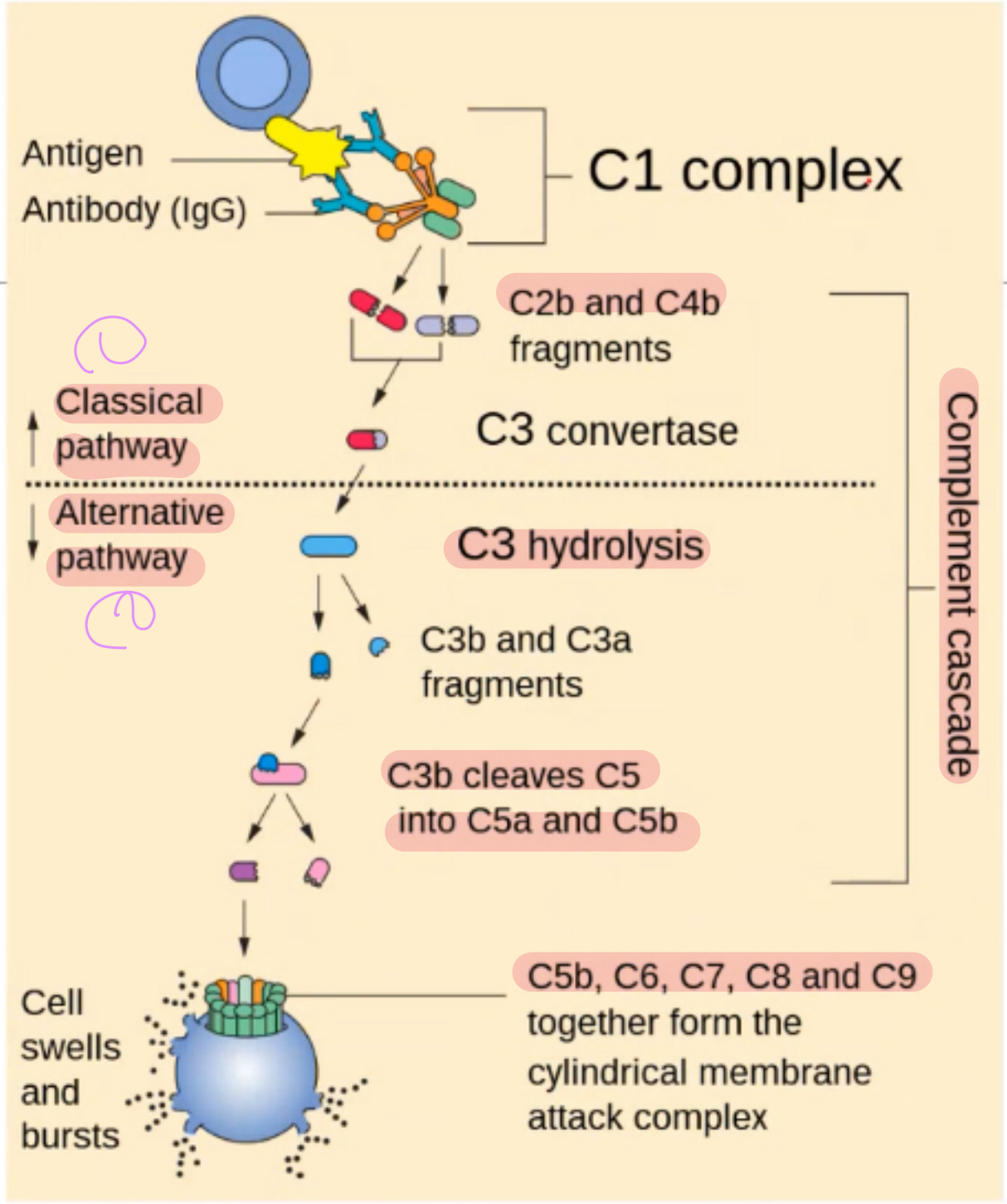
Autoimmune: Autoreactive antibodies (loss of self recognition of individual's own red cell antigen)

happens when the body's natural defense system can't tell the difference between your own cells and foreign cells, causing the body to mistakenly attack normal cells.

Drug-induced: Antibodies against red cells coated with drug or it is metabolites.

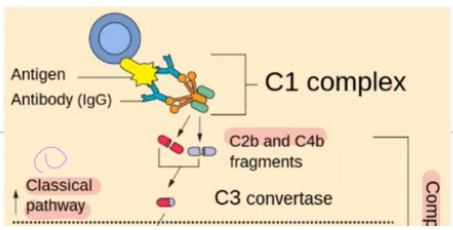
Drug or it’s metabolites that’s covers the RBC
فهيك بتخلي الantibodies تهاجمها

مراجعة قبل ما نبلش بالسلايد الجاي مهم نعرف أمور بسيطة



Complement pathway runs from c1–c9

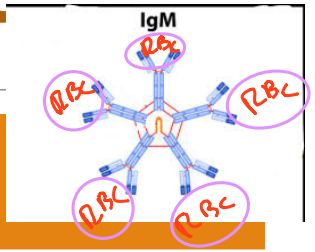
Classic pathway :activation c1 complex result in c2b/c4b fragment and the main thing that's I have antigen-antibody (IgG) connect to cell and this cell go to spleen
And the spleen break this cell



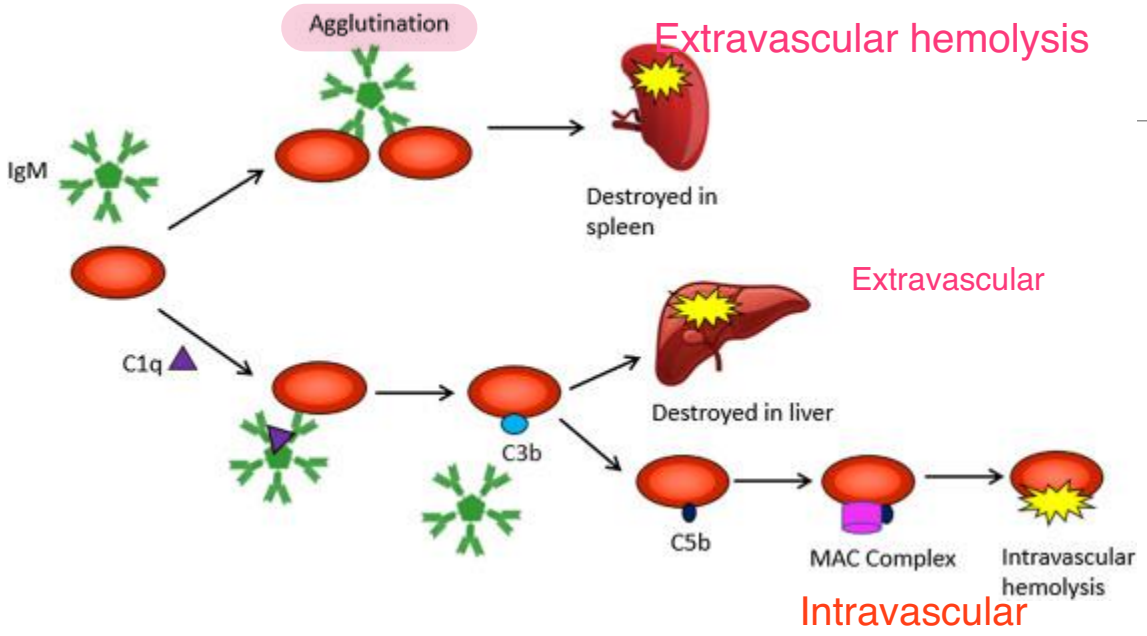
Alternative pathway: Haneen in the serum (blood) not in spleen
When tha activation happen from c3-c9 form something called
MAC(membrane attack complex)
وظيفته يعمل فتحة بالجسم الغريب عشان يقتله
تخيل يا سيدي انو هاي العملية تصير ع RBC
فراح تدمرها

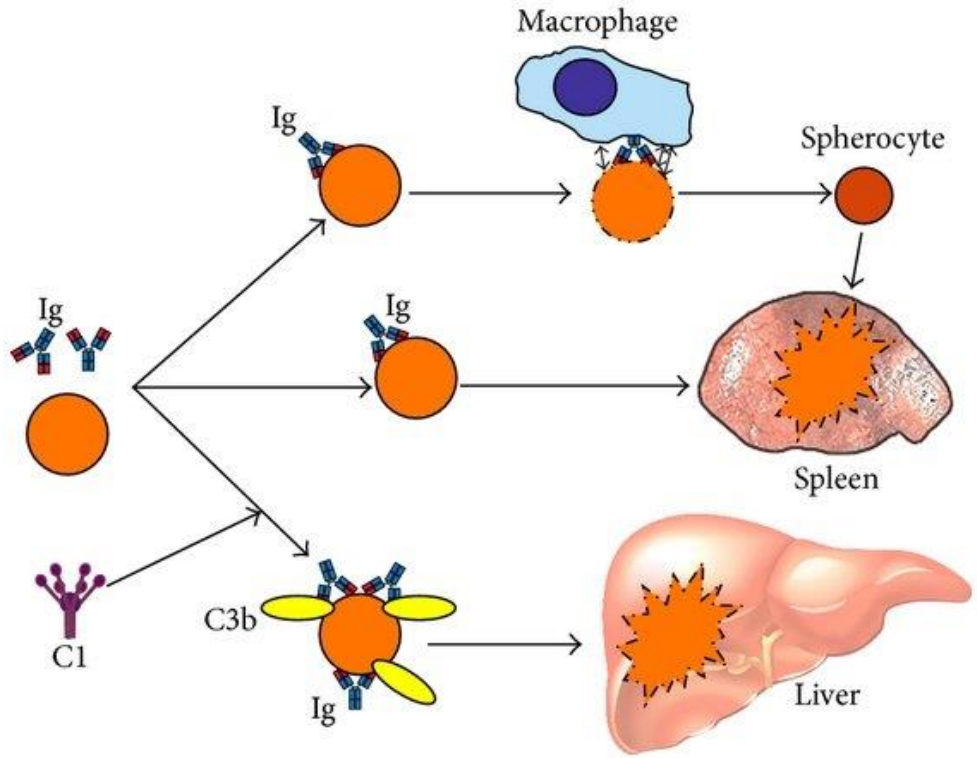
Causes of autoimmune-IHA
IgG or IgM mediated destruction of RBC

Agglutination =RBCs+ IgM



Warm Antibody: IgG/IgA type	Cold Antibody: IgM type
Activated at body temp. (37 c) warm عشان هيك اسمها	Active at 0-4°C IgM binds to RBC in cold temp (extremities)
IgG-coated RBC lysis in spleen (predominantly extravascular) بيروح IgG يرتبط بـ antigen على سطح RBC بوديها ع الـ spleen حتى تتكسر	Clumping and complement fixation causes lysis in blood vessels and liver (intra- and extravascular)
Morphology: spherocytes (splenic macrophage phagocytose tagged RBC leading to formation of spherocytes) هسا بس تروح RBC على الـ spleen IgG -antigen يلتهمها من جهة الـ spleen فكل شوي يلتهمها من جهة حتى يصير شكلها كروي	IgM agglutination (hemolysis occurs in the hands & feet in cold weather)
We don't know the causes 80% of immune hemolytic anemias: Primary (50-70%) Secondary: - Lymphoproliferative disorders We know the cause - Autoimmune diseases (SLE) بتعمل coat بعدين بيجي IgG يرتبط - Drugs (penicillin and cephalosporins)	• Infectious mononucleosis (EBV) • Mycoplasma infection • Lymphoproliferative disorders

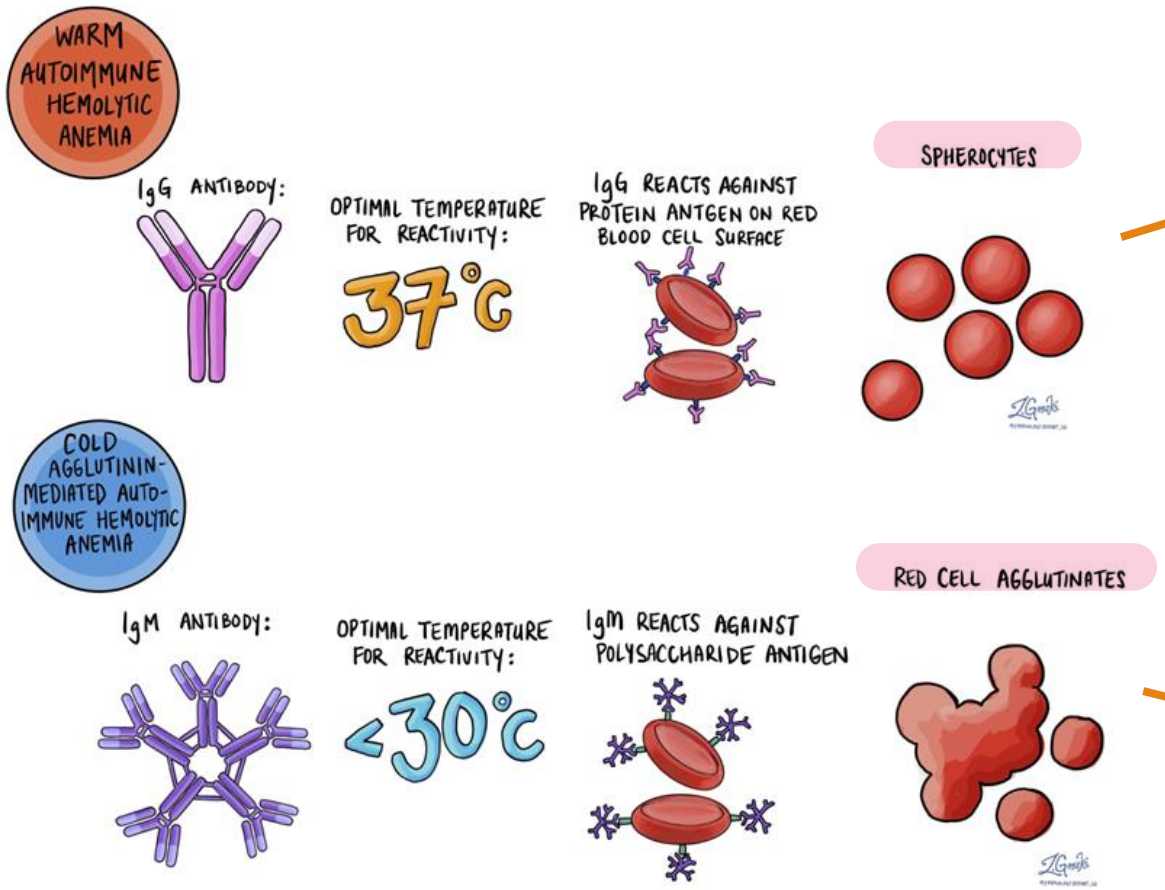


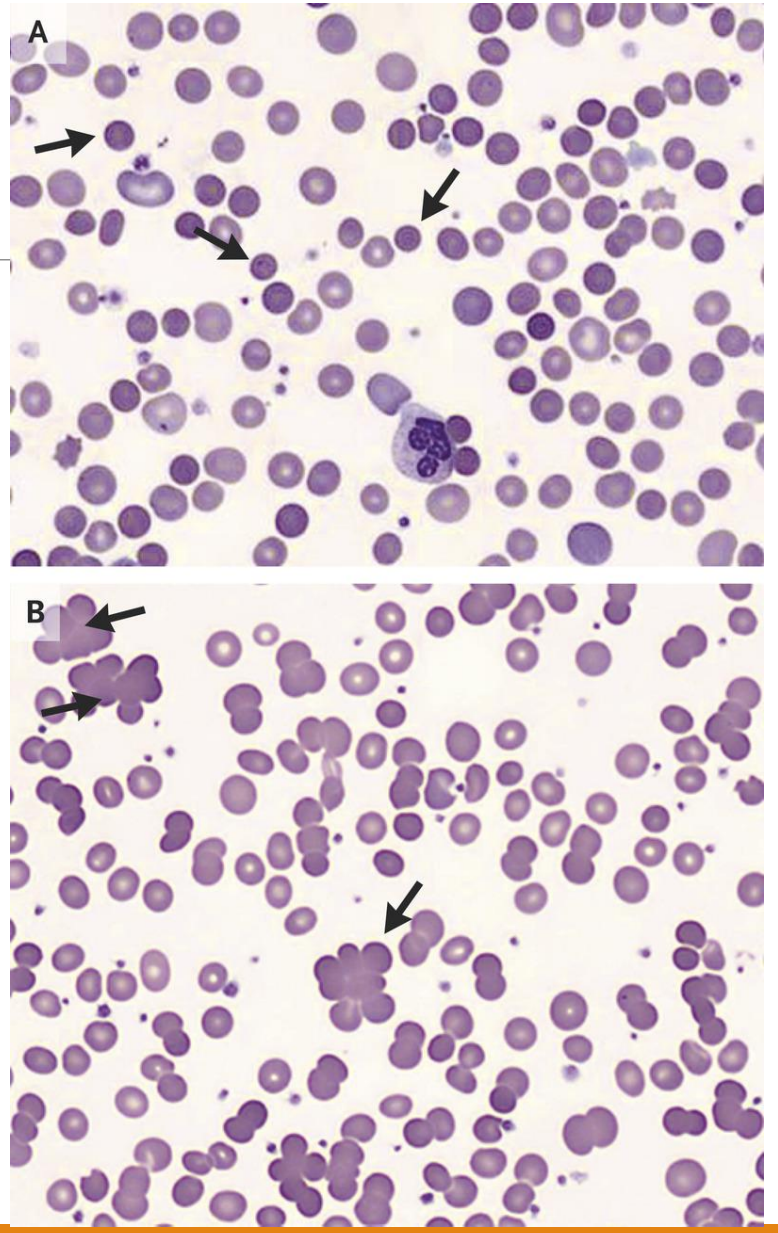


Cold Antibody: Clumping and complement fixation causes lysis in blood vessels and liver

Extravascular hemolysis

Warm antibody: Opsonization, phagocytosis and spherocytosis


WARM AUTOIMMUNE HEMOLYTIC ANEMIA
IgG ANTIBODY:
OPTIMAL TEMPERATURE FOR REACTIVITY:
37°C
IgG REACTS AGAINST PROTEIN ANTGEN ON RED BLOOD CELL SURFACE
SPHEROCYTES
COLD AGGLUTININ-MEDIATED AUTO-IMMUNE HEMOLYTIC ANEMIA
IgM ANTIBODY:
OPTIMAL TEMPERATURE FOR REACTIVITY:
<30°C
IgM REACTS AGAINST POLYSACCHARIDE ANTIGEN
RED CELL AGGLUTINATES


A
B

Intravascular hemolysis

Paroxysmal nocturnal Hemoglobinuria(PNH)

Not inherited

- PNH is caused by an acquired somatic (non-germline) mutation in the X-linked phosphatidylinositol glycan class A (*PIGA*) gene;

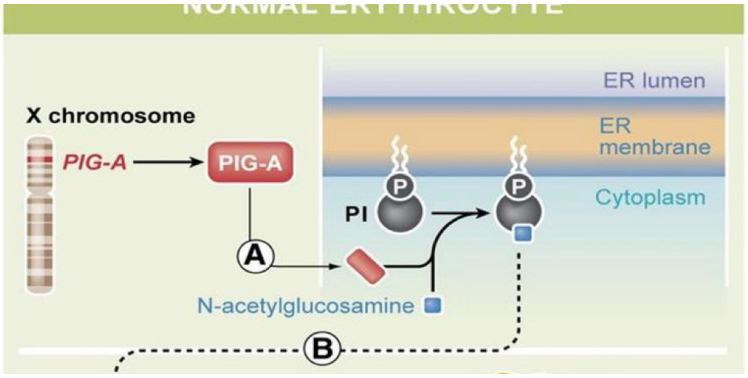
- (*PIGA*) gene produces the glycosylphosphatidylinositol (GPI) anchor proteins (GPI-APs), that links cell surface proteins to cell membranes

- Hematopoietic cells containing *PIGA* mutations lack GPI anchored cell surface markers, including complement inhibitors (such as CD59 and CD55)

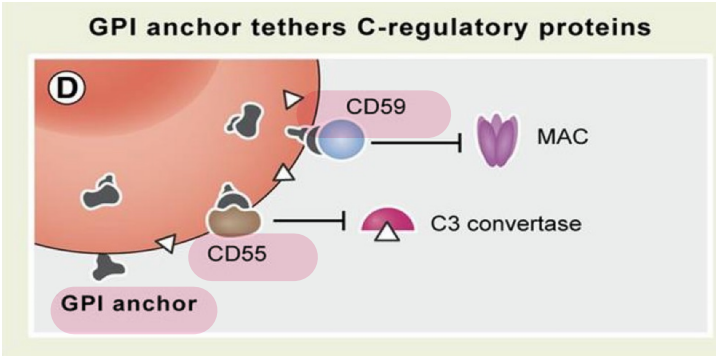
- **So, mature erythrocytes lacking GPI-APs are unprotected from the membrane attack complex (MAC or C5b9), leading to paroxysmal hemolysis**

اقرأ الشرح تحت

هسا في complement بتتمشى جوا الblood جنب الRBC ممكن يعملو membrane attacks complex (MAC)
و ممكن يعملو activation للalternative/classic pathways
هسا بالوضع الطبيعي الRBC محمية بسبب وجود surface proteins (CD55/CD59)
هدول يحموا RBC من الcomplement اللي حكيت عنهم فوق
هسا الCD55/CD59 محمولين على سطح الRBC عن طريق proteins اسمهم GPI anchor. proteins
اللي بصير بالparoxysmal nocturnal hemoglobinuria
هو عباره عن طفره في PIG-A اللي يعطيني GPI anchor proteins
معناته هيك ما بتم انتاج GPI anchor. proteins اللي بتحمل CD55/59
معناته ما في اشي يحمل CD55/59
فبتقدرش تمسك بسطح الRBC و تحميها من الcomplements
فبقدرو يدمرو الRBC بسهولة

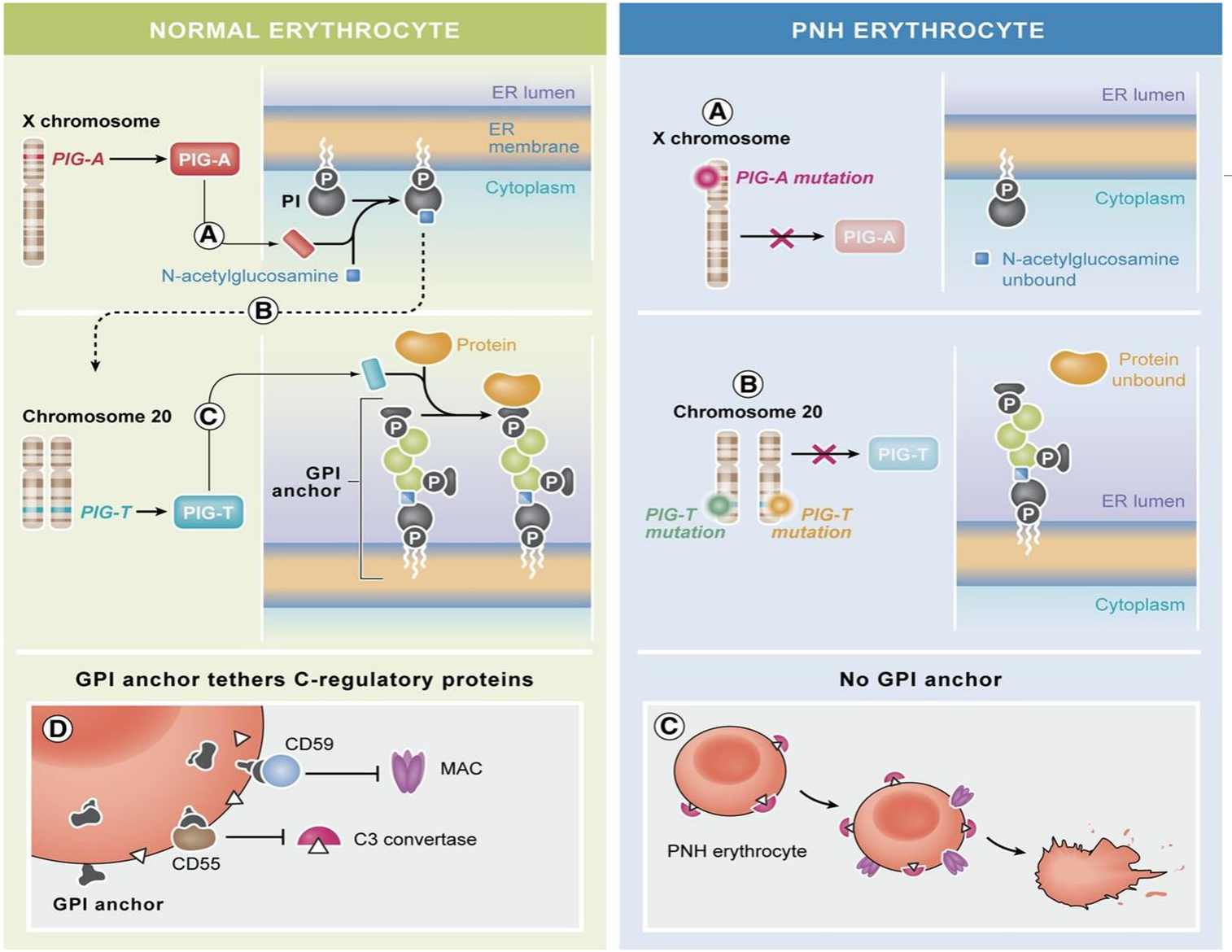


هون بتصير الطفره



GpI anchor protein and surface proteins CD55/59

ما الواحد يتنفس باليل النفس تبعه بكون very shallow
و ما يكون shallow معناته بتراكم عندي Co2
ما بتراكم الـ blood ph بقل
و بصير عندي acidosis
هسا الـ complements اللي قرفتكم فيهم بصير النهار activation
في حالة الـ acidosis



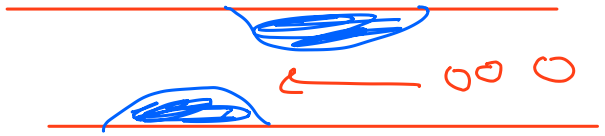
Hemolysis occurs mostly at night when there is fixation of complement which is enhanced by decrease of blood PH during sleep

- ✓ Chronic intravascular hemolysis with hemoglobinemia, hemosiderinuria -/+ hemoglobinuria
- ✓ Reticulocytosis
- ✓ Venous thrombosis (hypercoagulability due to free Hb in blood)

Hemolytic anemias due to mechanical trauma to RBCs

شبه المطبات اللي بالشارع لما السياره تمرق عليهم بتطبش و هيك بصير بال RBC
بتصير على شكل helmet cells
Or burr cells

Red cells are disrupted by **physical trauma**:



I. Cardiac valve prostheses

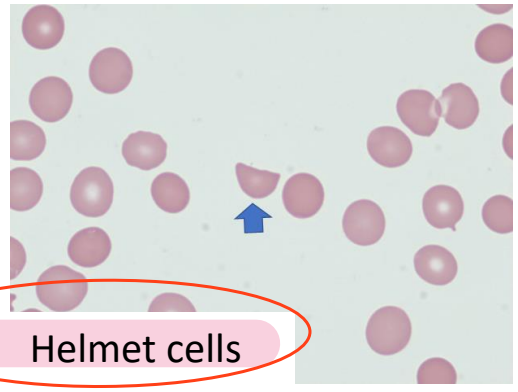
II. Microangiopathic hemolytic anemia as in DIC , malignant hypertension, thrombotic thrombocytopenic purpura (TTP), hemolytic uremic syndrome (HUS).

(Disseminated intravascular coagulation

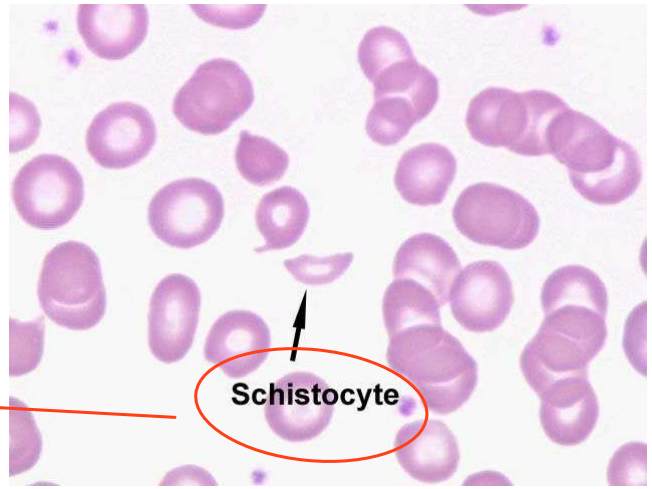
احفظوه كمثال فقط

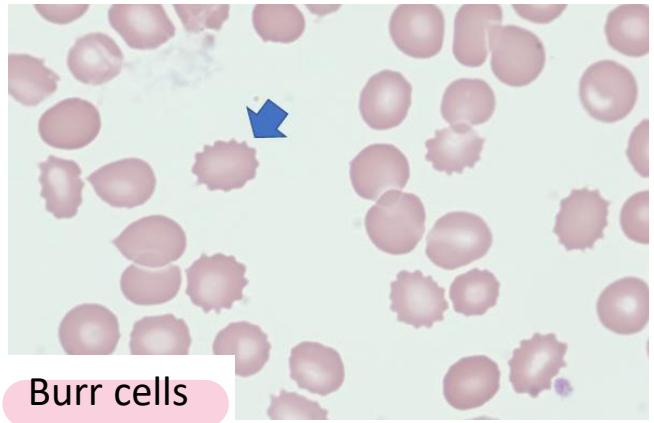
We will discuss them

In all these conditions the circulating RBCs are mechanically traumatized , get the appearance of **Schistocytes , burr cells or helmet cells**



Same





استريح تقريبا عشر دقايق و بعدين قول بسم الله

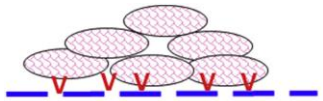
HLS pathology lecture 5

COAGULATION DISORDERS

DR. DUA ABUQUTEISH

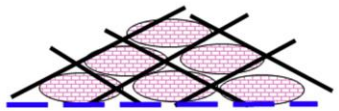
مو راح تسألنا عن الfactors

الشرح تحت كالعادة😍😍 Coagulation Cascade



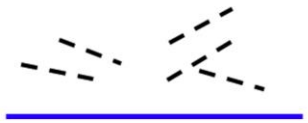
Primary Hemostasis

Vessel/platelet/VWF interactions



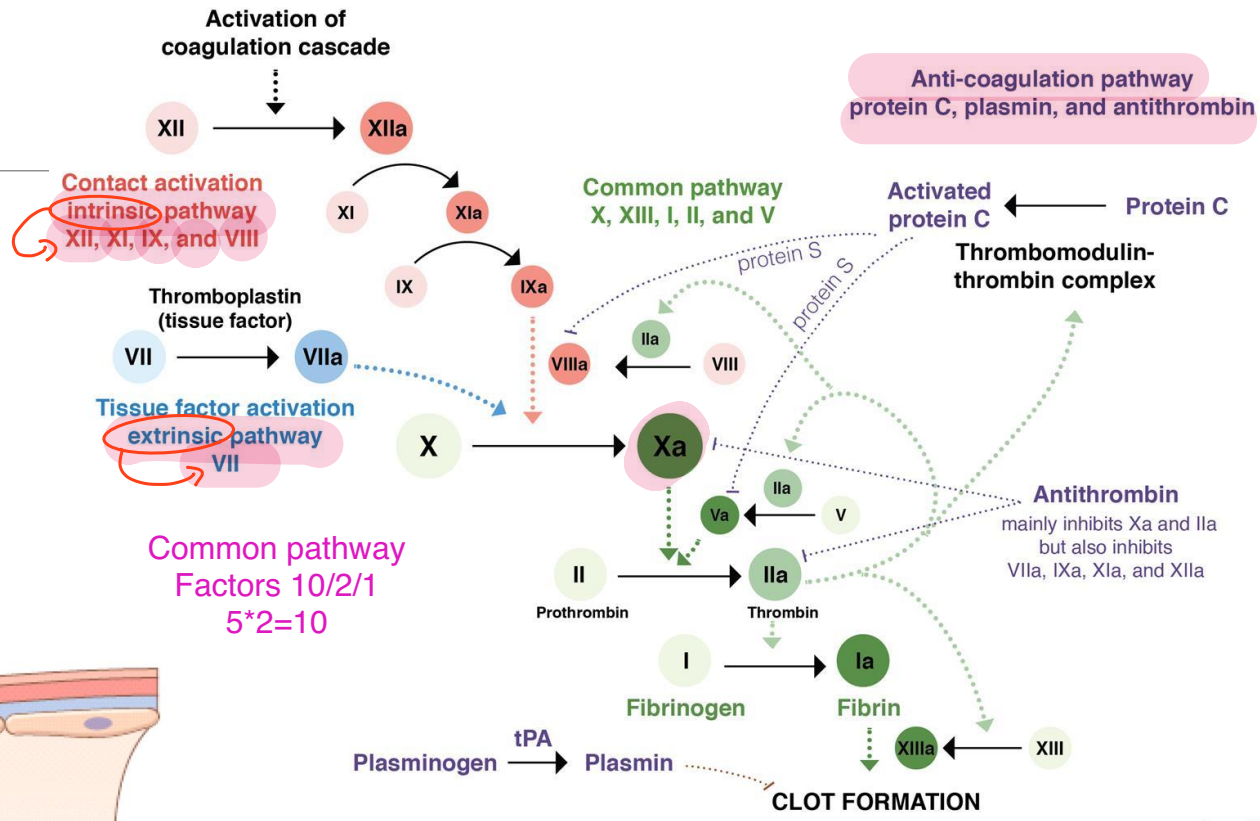
Secondary Hemostasis

Coagulation & anticoagulant factors



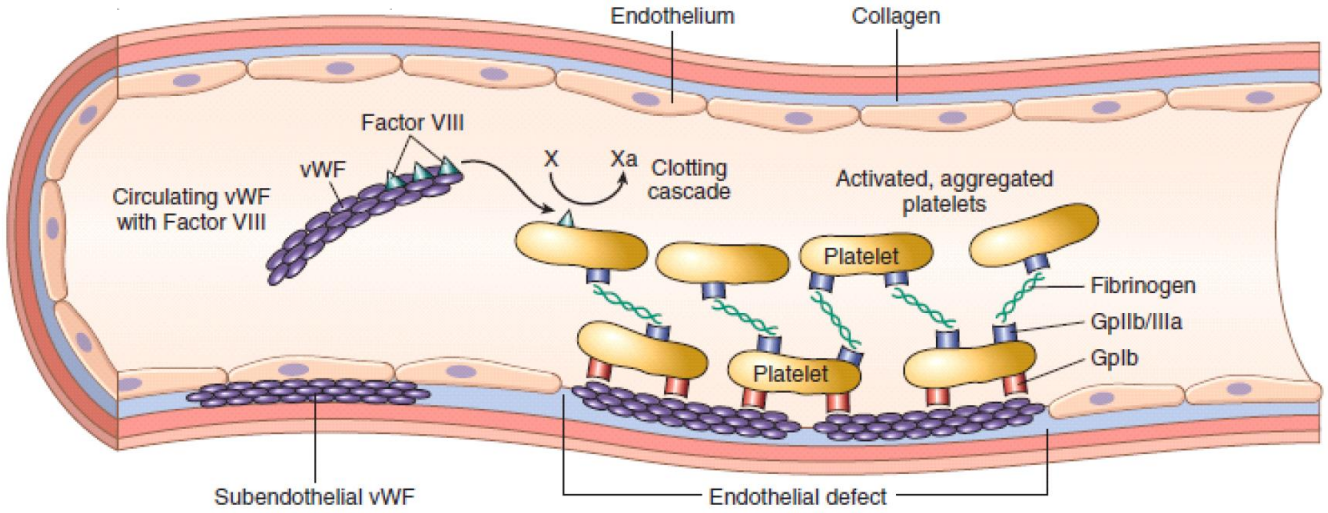
Fibrinolysis

Plasmin activation & fibrin dissolution



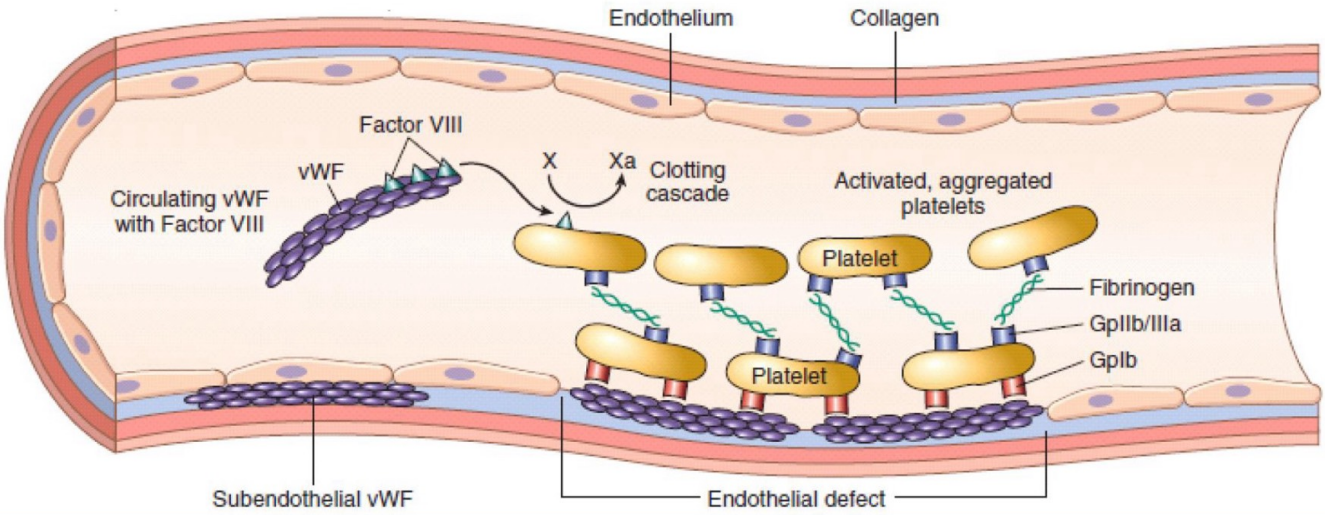
Common pathway
Factors 10/2/1
5*2=10

- Platele



Lucy Liu

- Platele



بس يصير عنا **endothelial injury** راح تعمل **expouse** لل**(VWF (von Willebrand facto** الموجوده بال**sub endothelial collagen**

فراح تكون اول مرحله **adhesion** لل**platelet** مع **sub endothelial collagen** مع **VWF**

راح يتم ال**adhesion** عن طريق **receptor GP1p**

بعدين بصير عندي **platelets aggregation** فراح يتغير شكلها و تفرز ال**granules**اللي جواتها مثل **ADP/ thrompoxin**

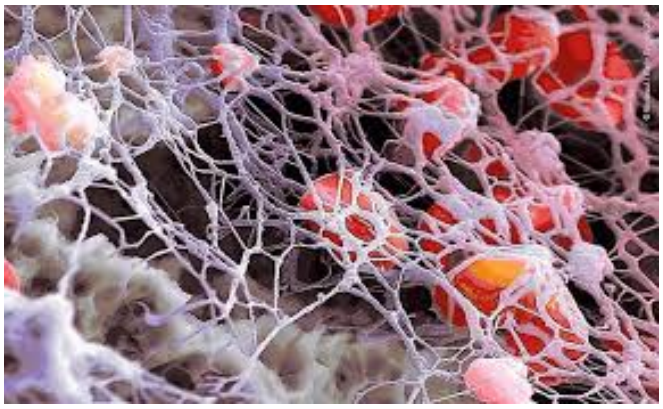
ال**aggregation** بتم عن طريق **GP23A receptor**

هسا بنلاحظ انو في **fibrinogen** بين ال**platelets** بس مشكلته انو ضعيف فلو كملت بوجود ال**fibrinogen** لحاله راح يكون الترابط ضعيف

عشان أقويها لازم اعمل **activation** لل**clotting factors**

لأنو ال**clotting factor s**بتحول **fibrinogen** إلى **fibrin** و ال**fibrin** بعمل **fibrin meshwork**و تمسك كل ال**clot**

و بتسكر الجرح

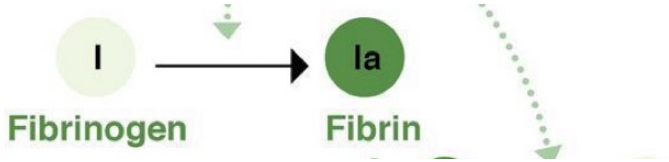


Fibrin meshwork

Initial Laboratory Tests For Bleeding Abnormalities

thrombocytopenia اقل من ١٥٠،٠٠٠

- **Platelet count** (normal is 150,000 and 400,000 per microliter of blood)
- **Bleeding time:** to assess platelet function. It involves making a patient bleed, then timing how long it takes to stop bleeding using a stopwatch.
- **Partial thromboplastin time (PTT):** measures activity of the intrinsic clotting pathway (اطول)
- **Prothrombin time (PT)/INR:** both measure activity of the extrinsic clotting pathway
- **Thrombin time (TT):** measures the final step in the clotting cascade, in which fibrinogen is converted to fibrin



Common pathway factors X, V, prothrombin (II) and fibrinogen (I): affect both PTT & PT

Clinical presentation; symptoms

بتكون في مشكلة بالplatelets/VWF

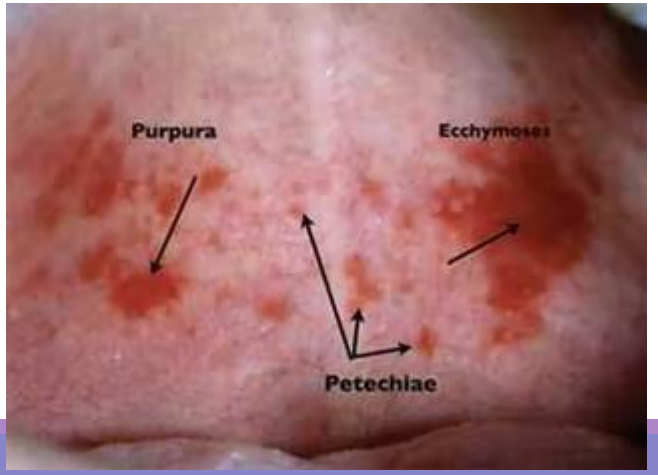
Common features of patients with problems in primary hemostasis:

- ✓Mucosal and skin bleeding; intracranial bleeding with severe thrombocytopenia
- ✓Mucosal bleeding - epistaxis (nose bleeding), hemoptysis (coughing blood), GI bleeding, hematuria, and menorrhagia (heavy menses)
- ✓Skin bleeding - petechiae (bleeding spots on skin), purpura (>3mm bleeding spots), ecchymoses, (>1cm), easy bruising.

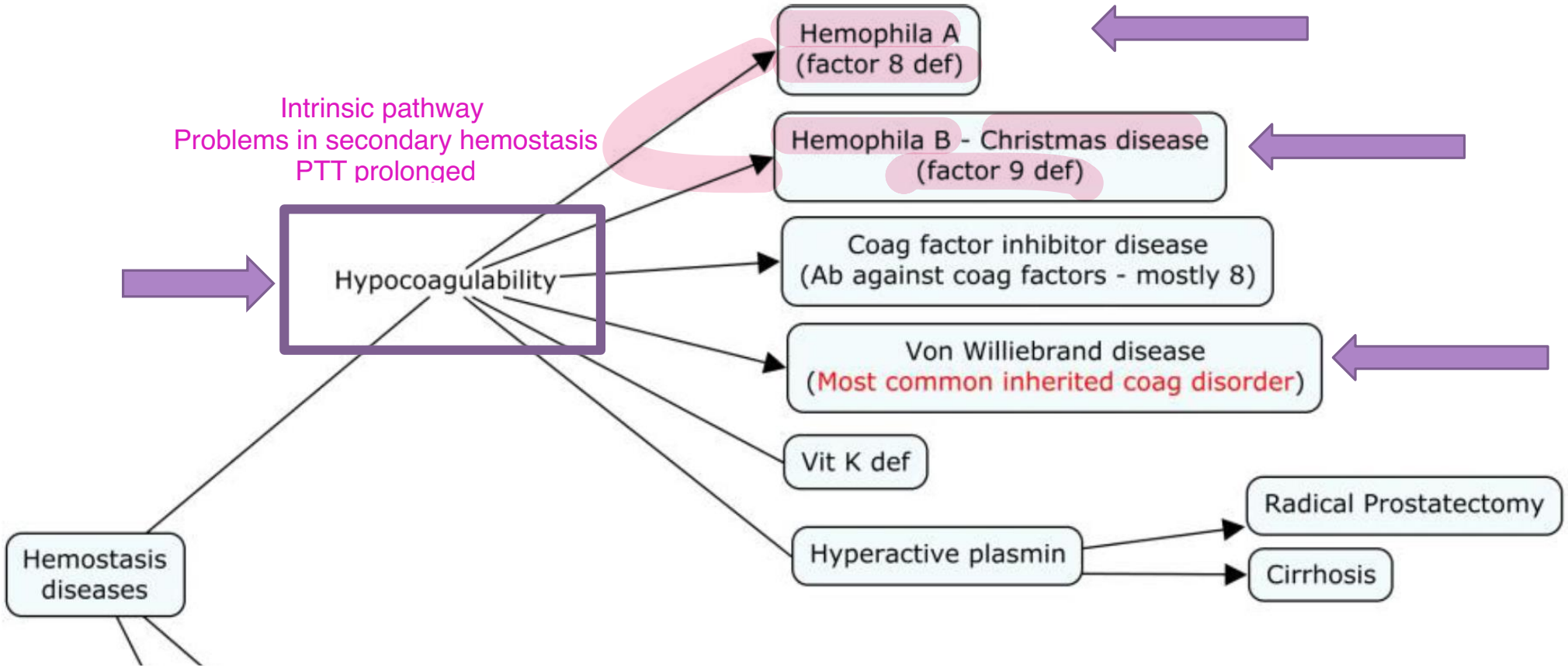
هون مشكلة بالcoagulation factors

Common features of patients with problems in secondary hemostasis:

- ✓Deep bleeding in muscles and joints
- ✓Rebleeding after surgical procedures



راح ناخذ هسا اول اثنين و بكره بنكمل



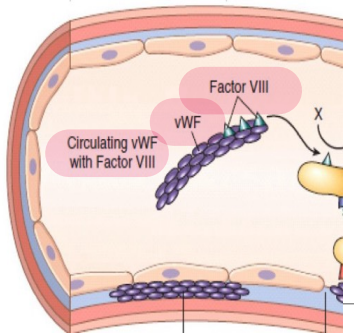
Hemophilia A

بكون عندي مشكلة بـfactor 8
(F 8 def)

- Factor VIII deficiency (hemophilia A)
- Inherited as an X-linked recessive trait
- Almost exclusively affects males
- 1 in every 10,000 births or 1 in 5000 male births
- It is characterized by mild, moderate or severe bleeding episodes (depends on factor VIII level)
- Bleeding into muscle, soft tissue or joints (hemarthrosis – 70-80%), and excessive bleeding after surgery, trauma, dental procedures or circumcision
- Petechia are characteristically absent

Because it's not primary hemostasis

- Platele



F 8/VWF
They are besties
دايما مع بعض

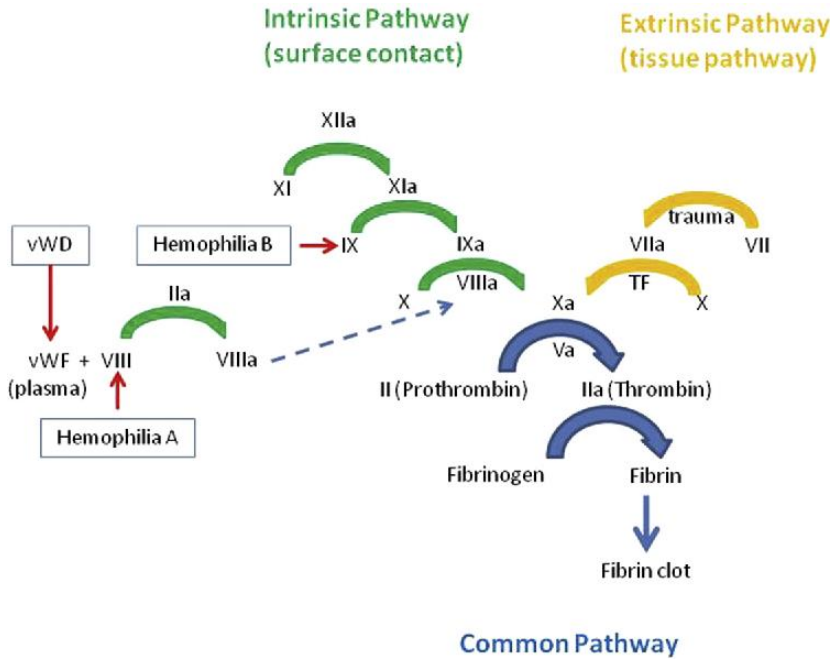
Hemophilia A

- Factor VIII in plasma circulates bound to von Willebrand factor protein. Markedly unstable in the absence of vWF.
- Factor VIII and factor IX = activate factor X, which in turn with its cofactor, factor Va, activates thrombin

Laboratory tests:

- Prolonged PTT. Normal PT and TT. Normal platelet count.

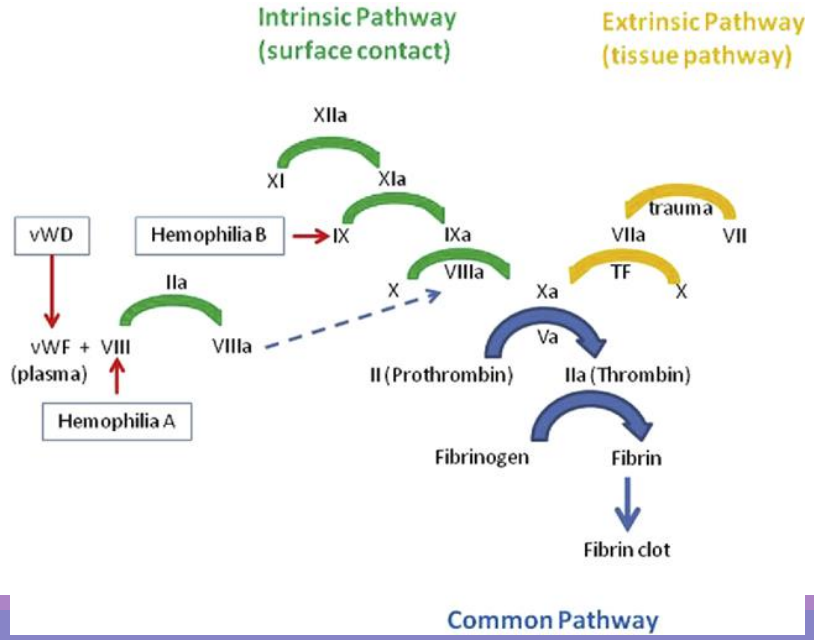
Treatment: replacement therapy (factor VIII concentrate or recombinant VIII).



factor 9ب عندي المشكلة بس haemophilia A نفس

Hemophilia B

- Factor IX deficiency (hemophilia B)
- Inherited as an X-linked recessive trait
- It is characterized by mild, moderate or severe bleeding episodes (depends on factor IX levels)
- Also known as Christmas disease
- Almost exclusively affects males
- 1 in 30,000 male births
- Bleeding sites: same as Hemophilia A



Hemophilia B

Factors 1/9/7/2 1972😘😘

- Factor IX is a vitamin K-dependent serine protease produced in the liver
- It circulates in the plasma in its inactive form
- With factor VIIIa, it catalyzes the conversion of factor X to Xa

Laboratory tests:

- Prolonged PTT. Normal PT and TT. Normal platelet count. Normal factor VIII assay.
- ***Always measure both factor VIII and IX activity, and rule out vWD***

إذا عندي مشكله ب vWD ممكن يكون عندي مشكله ب factor 8

Disease

Treatment: Plasma-derived or recombinant factor IX concentrates