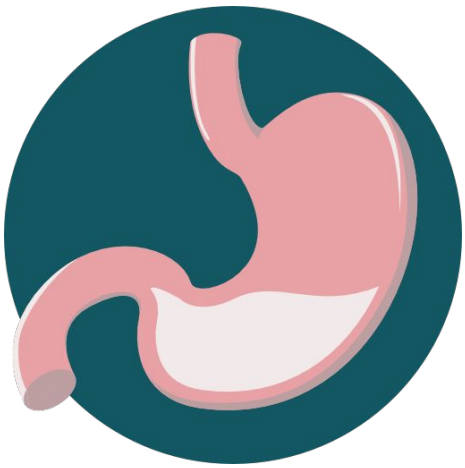


Coagulation

Objectives :

- Recognize different stages of haemostasis.
- Explain the role of platelets in haemostasis.
- Recognize different clotting factors & cascade of clotting.
- Describe the intrinsic, extrinsic and common pathway.
- Recognize the role of thrombin in coagulation.
- Explain process of fibrinolysis and function of plasmin.

Done by :

- Team leader:Abdurhman AlHayssoni, Rahaf AlShammari
- Done by:
  - Abdurhman AlHayssoni
  - Abdullah AlZaid

Colour index:
- Important
- Numbers
- Extra

وَأَن لَّيْسَ لِلْإِنسَانِ إِلَّا مَا سَعَىٰ

HAEMOSTASIS

The spontaneous arrest of bleeding from ruptured blood vessels

The doctor mentioned that I can ask you in SAQ, enumerate the steps of haemostasis in sequence.

FOUR STEPS OF HEMOSTASIS

Haeme: Blood , Stasis: to stop
Haemostasis : Stoppage of bleeding
Do not confuse it with homeostasis.
Homeostasis: A property of cells, tissues, and organisms that allows the maintenance and regulation of the stability and constancy needed to function properly..

1-Vascular spasm (Vascular Constriction)

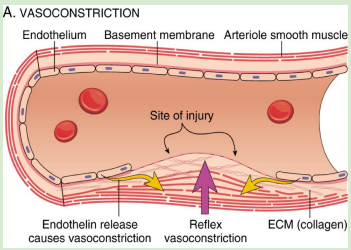
Causative Factors are three:

1. Nervous reflexes "the nerves are irritated that will send sensory signals to initiate a reflex"
2. Local myogenic spasm "the smooth muscles of injured blood vessels are irritated that will cause contraction even an inhibitory nerve is acting , spasm will proceed"
3. Local humoral factors....Platelets → Thromboxane A [TXA2] (Vasoconstrictor)

Importance:

Crushing injuries → Intense spasm → No lethal loss of blood.

TXA2 is inhibited by aspirin



The more area is injured, the stronger is the vascular spasm.
ومثال عليه اذا واحد جرح اصبعه بسكين وواحد ثاني يلعب كورة و وتزحلق واحتكت ركبته ورجله بالارض ، ايهم بينزف اكثر؟؟ الي جرح اصبعه بينزف اكثر لان الجرح تضمن منطقة اقل مقارنة بالاحتكاك.

If the first step is enough, our body will not proceed to the second step and this applies for other steps.

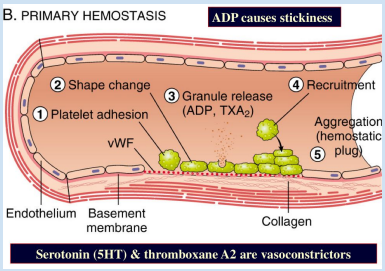
2-Formation of platelet plug (Primary hemostasis)

Production, activation and formation of platelet plug.

Platelets adhesion either directly to collagen fibers or to Von Willebrand factor.

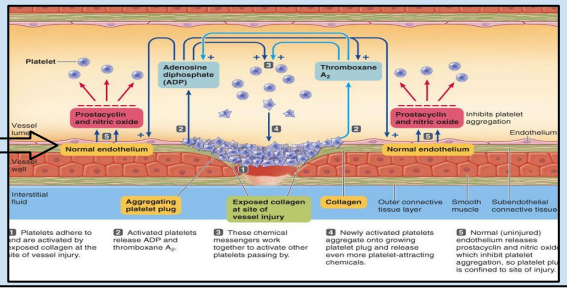
Importance:

Enough to stop bleeding from small vascular damage



From NE to prevent the plug formation

Secrete
- prostacyclin (PGI2)
- NO
- ADP phosphatase "ADP antagonist"



To understand better:

Platelet repair of vascular openings is based on several important functions of the platelet. When platelets come in contact with a damaged vascular surface, especially with collagen fibers in the vascular wall, the platelets rapidly change their own characteristics drastically. They begin to swell; they assume irregular forms with numerous irradiating pseudopods protruding from their surfaces; their contractile proteins contract forcefully and cause the release of granules that contain multiple active factors; they become sticky so that they adhere to collagen in the tissues and to a protein called von Willebrand factor that leaks into the traumatized tissue from the plasma; they secrete large quantities of ADP; and their enzymes form thromboxane A2. The ADP and thromboxane in turn act on nearby platelets to activate them as well, and the stickiness of these additional platelets causes them to adhere to the original activated platelets.
Therefore, at the site of a puncture in a blood vessel wall, the damaged vascular wall activates successively increasing numbers of platelets that attract more and more additional platelets, thus forming a platelet plug. This plug is loose at first, but it is usually successful in blocking blood loss if the vascular opening is small. Then, during the subsequent process of blood coagulation, fibrin threads form. These threads attach tightly to the platelets, thus constructing an unyielding plug.

When we put blood in a glass tube, the blood will clot, this clot is mediated by intrinsic pathway not extrinsic. That's why factor 12 is named glass factor.

3-Blood coagulation (secondary hemostasis)

Strong and complete

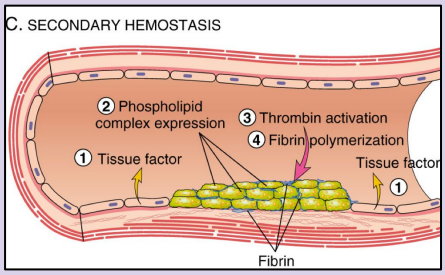
Blood clotting is the transformation of blood (soluble fibrinogen) from a liquid into a solid gel form (insoluble fibrin strands)

Pathways :

It is useful that we have two pathways because if one pathway is blocked by any means the another one will function to form clot.

- Intrinsic
- Extrinsic

CLOT is a meshwork of fibrin fibres running in all directions entrapping blood cells, platelets and plasma. Clot contents



Begins to develop in :

- 1-2 min → Minor trauma
- 15-20 sec→ Severe trauma

To understand better:

The third mechanism for hemostasis is formation of the blood clot. The clot begins to develop in 15 to 20 seconds if the trauma to the vascular wall has been severe and in 1 to 2 minutes if the trauma has been minor. Activator substances from the traumatized vascular wall, from platelets, and from blood proteins adhering to the traumatized vascular wall initiate the clotting process.

SAQ

Mechanism of clotting

The purpose of intrinsic and extrinsic pathways is to form prothrombin activator complex.

Formation of Prothrombin activator complex (**Xa+Ca+PF-3+V**) by Extrinsic & Intrinsic Pathways leading to Common Pathway

↓

Conversion of prothrombin into thrombin

↓

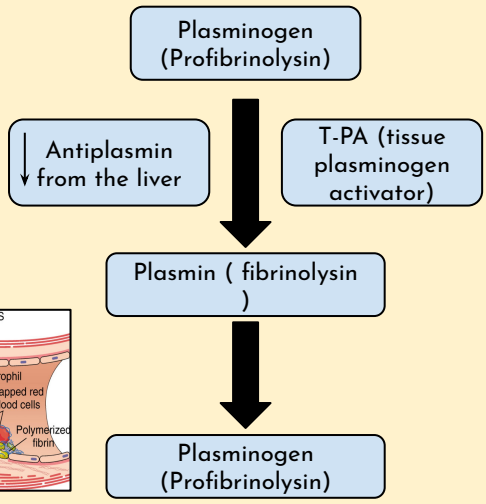
Conversion of fibrinogen into fibrin

↓

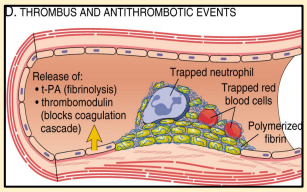
Fibrin converts to stable fibrin polymer

4- lysis of the clot by plasmin

- Formed blood clot can either become fibrous or dissolve.
- Fibrinolysis (dissolving) = Break down of fibrin by naturally occurring enzyme plasmin therefore prevent intravascular blocking.



Tissue Plasminogen Activator (TPA) is used clinically to activate plasminogen to dissolve coronary and cerebral clots.



Coagulation factors

You **should** memorize them all and focus on the first four factors , hemophilia related factors(8,9,11) and factors 10 and 13.

Mnemonics:
"Fresher's Party Tonight, Come Let's Sing And Call Seniors, Please Have Fun"

I Fibrinogen. (Fresher's)
II Prothrombin. (party)
III Tissue factor. (tonight)
IV Calcium. (come)
V Proaccelerin, Labile factor. (Let's)
VII Stable factor, proconvertin. (Sing)
VIII Antihemophilic factor A. (And)
IX Antihemophilic factor B or Christmas factor. (call)
X Stuart-Prower factor. (seniors)
XI Plasma thromboplastin antecedent. (please)
XII Hageman factor. (have)
XIII Fibrin-stabilizing factor. (fun)

Factor VI is missing because they found out that is not related to clotting mechanism.

Factor[a]	Names
I	Fibrinogen
II	Prothrombin
III	Thromboplastin
IV	Calcium
V	Proaccelerin, labile factor, accelerator globulin
VII	Proconvertin, SPCA, stable factor
VIII	Antihemophilic factor (AHF), antihemophilic factor A, antihemophilic globulin (AHG)
IX	Plasma thromboplastic component (PTC), Christmas factor, antihemophilic factor B
X	Stuart–Prower factor
XI	Plasma thromboplastin antecedent (PTA), antihemophilic factor C
XII	Hageman factor, glass factor
XIII	Fibrin-stabilizing factor, Laki–Lorand factor
HMW-K	High-molecular-weight kininogen, Fitzgerald factor
Pre-Ka	Prekallikrein, Fletcher factor
Ka	Kallikrein
PL	Platelet phospholipid

Prothrombin	- Plasma protein (Alpha globulin) - Mol. Wt. - 68,700 - Plasma conc. - 15 mg/dl - Unstable protein - Synthesized by liver - Vitamin-K is required for synthesis
Fibrinogen	- Mol. Wt. – 340,000 - Plasma conc. – 100 – 700 mg/dl - Synthesized in liver

Most abundant in the blood, so it has to have number one.

Coagulation cascade

There are intrinsic and extrinsic pathways

Extrinsic

Intrinsic

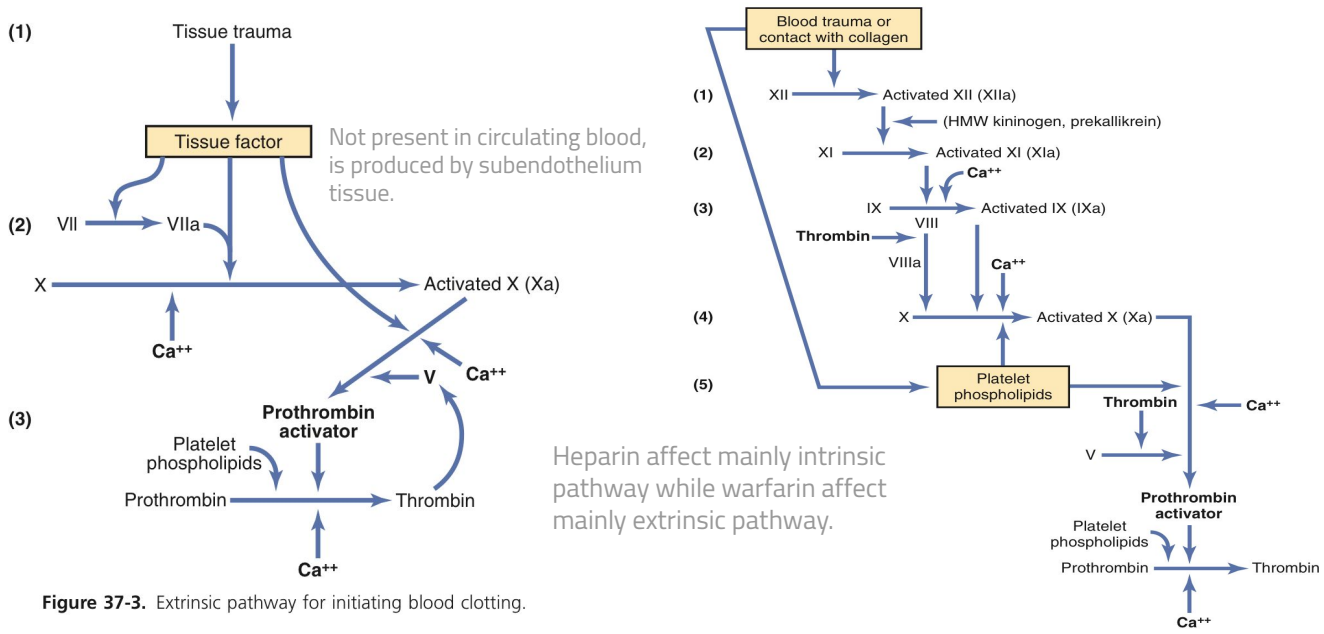


Figure 37-3. Extrinsic pathway for initiating blood clotting.

Heparin affect mainly intrinsic pathway while warfarin affect mainly extrinsic pathway.

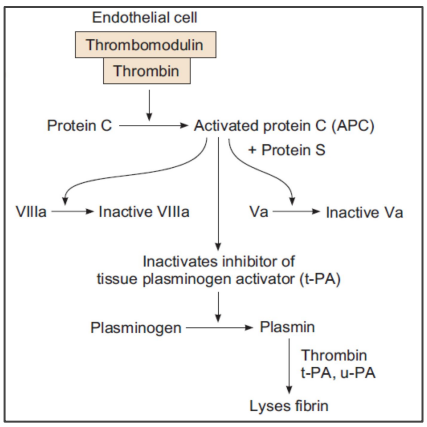
TF or tissue thromboplastin; includes phospholipids from the membranes of the tissue plus a lipoprotein complex that functions mainly as a proteolytic enzyme.

Trauma to the blood itself or exposure of the blood to collagen (from a traumatized blood vessel wall), foreign surface/glass

The fibrinolytic system

The fibrinolytic system and its regulation by **Protein C**.

Question from Dr Shahid:
What is the main factor in the blood that is activated by thrombin which will initiate fibrinolytic pathway?
The answer is Protein C.



Clot reaction

- When clot retracts (contracts), it expresses most of the fluid from the clot within 20-60 min called **Serum**
- Serum cannot clot
- Role of platelets in clot formation & retraction, they are *contractile*.

Role of calcium ion in clotting

No Ca++ → No Clotting (Needed in many steps)

Blood samples are prevented from clotting by:

1. **Citrate ions** → Deionization of Ca^{++} Ionized Ca is active, deionized Ca is inactive

2. **Oxalate ions** → Precipitate the Ca^{++}

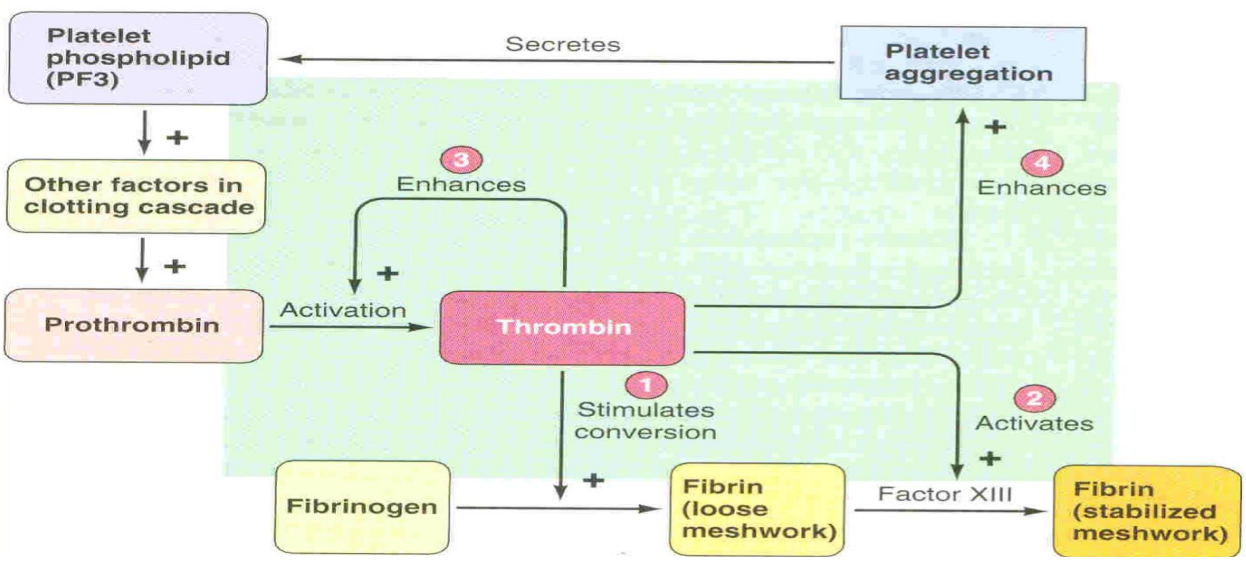
3. **Heparin** → combines with antithrombin effectiveness increases by 100-1000 fold, Also remove Factors XII, XI, X, and IX (Monitored by PTT time)

4. **Warfarin**: production of Factors VII, IX and X by liver (Monitored by PT time)

5. **EDTA** → chelates (binds) calcium ions

Roles of thrombin in hemostasis

SAQ



Natural intravascular anticoagulants

Endothelial Surface Factors

1. Smoothness of Endothelium
2. Glycocalyx Layers
3. Thrombomodulin Protein binds to thrombin and activates Protein C (with Prot. S) inactivates factors V & VIII and inactivates an inhibitor of tPA increasing the formation of plasmin.

Glycocalyx: pericellular matrix (glycoprotein and glycolipid) covering the surrounds cell membranes

Antithrombin action of Fibrin and Antithrombin III

85-90 % Thrombin binds with Fibrin
10-15 % Thrombin binds with Antithrombin III

Antithrombin III is a circulating protease blocking clot factors

Heparin

- Negatively charged conjugated polysaccharide
- Increase the effectiveness of Antithrombin III
- Produced by
  - Mast cells
  - Basophil cells
- Most widely used anticoagulant clinically e.g. in stroke

Alpha 2– Macroglobulin

Synthesized mainly in liver and acts as a binding agent for several coagulation factors and inhibits thrombin.

Dr's note: Primary hemostasis is tested by bleeding time "platelets"
Thrombocytopenia is bleeding disorder which prolongs bleeding time and also if it is very severe can prolong clotting time.
Hemophilia"clotting disorder" affect coagulation cascade, so bleeding time is normal and clotting time is prolonged.
A type of hemophilia A which is Von Willebrand disease can prolong both bleeding and clotting time.

Bleeding and clotting disorders

Hemophilia

- Genetic disorders
- Transmitted by female chromosome as recessive trait.
- Occurs exclusively in males, females are carriers.

Hemophilia A:
Classic Hemophilia
85 % cases
Def. Of factor **VIII**

Hemophilia B:
15 % cases
Def. Of factor **IX**

HEMOPHILIA C:
Def of factor **XI**
(both sexes)

- Small Comp. →Hemophilia A ▶↑PTT
- Large Comp. →Von-Willebrand's disease ▶↑PTT & BT

Clinical Features: Easy bruising, massive bleeding after trauma or operation, hemorrhages in joints

Thrombocytopenia

Count < 50,000 ul may cause spontaneous bleeding
Less than 10,000 ------ Fatal

ETIOLOGY:

Increased destruction.
- ITP
- Drugs
- Infections (HIV)

Decreased production
- Aplastic anemia
- Leukemia
- Drugs
- Infections (HIV, Measles)

Clinical Features
- Easy bruisability
- Epistaxis
- Gum bleeding
- Hemorrhage after minor trauma
- Petechiae/Ecchymosis

Liver disease

e.g. Hepatitis, Cirrhosis

- Decreased formation of clotting factors
- Increased clotting time

Vit. K deficiency

Fat soluble vitamin
Required by liver for formation 4 clotting factors (Factor II, VII, IX & X)

Sources:
- Diet
- Synthesized in the intestinal tract by bacteria

Deficiency :

- Malabsorption syndromes
- Biliary obstruction
- Broad spectrum antibiotics
- Dietary def (in Neonates)

Treat the underlying cause→Vit K injections

Haemostasis tests in hereditary coagulation disorders

	Haemophilia A	Haemophilia B	VW disease
Bleeding time	Normal	Normal	Prolonged
Prothrombin time	Normal	Normal	Normal
APTT	Prolonged	Prolonged	Prolonged
Factor VIII	Low	Normal	Low or normal
Factor IX	Normal	Low	Normal
VWF	Normal	Normal	Low

Screening tests

Very important to understand

Test	Mechanism Tested	Normal Value	Disorder
Bleeding time (BT)	Hemostasis, capillary & platelet function	3-7 min beyond neonate	Thrombocytopenia , von Willebrand disease
Platelet count	Platelet number	150 000 – 450 000 / mm^3	Thrombocytopenia
Prothrombin time (PT)	Extrinsic & common pathway	< 12 sec beyond neonate; 12-18 sec in term neonate	Defect in Vit K-dependent factor, liver disease, DIC
Activated partial thromboplastin time (APTT)	Intrinsic & common pathway	25-40 sec beyond neonate; 70 sec in term neonate	Hemophilia, von Willebrand disease, DIC

Source from : Nelson Essential of Pediatrics 5th edition

- Prothrombin time (PT) is a blood test that measures the time it takes for the liquid portion (plasma) of your blood to clot.
- Activated Partial thromboplastin time (APTT) is a blood test that looks at how long it takes for blood to clot.
- The tests are performed by taking blood samples

- If a patient is on warfarin which test is used to monitor his/her blood ? PT
- If a patient is on heparin which test is used to monitor his/her blood ? APTT
- Warfarin will prolong PT and Heparin will prolong APTT
- Because warfarin affect mainly extrinsic pathway, on the other hand heparin affect mainly intrinsic pathway.

MCQs

Q1: Which one would be inhibited by Aspirin to prevent clot formation?

A. ADP
B. Thromboxane A2
C. Serotonin
D. PGI

Q2: Bernard Soulier Syndrome is caused by:

A. A disorder of granules
B. A disorder of cytokines
C. A disorder of aggregation
D. A disorder adhesion

Q3. ADP/ATP can be found in: :

A. Dense granules
B. OCS
C. Alpha granules
D. Mitochondria

Q4. Low platelet count can be caused by:

A. Hypersplenism
B. Splenomegaly
C. Hepatomegaly
D. A&B

Q5. The coagulation pathway that begins with tissue thromboplastin is:

A. Intrinsic pathway
B. Extrinsic pathway
C. Common pathway
D. Fibrin stabilization

Q6. Why do some malnourished patients bleed excessively when injured?

A. Vitamin K deficiency
B. Platelet sequestration by fatty liver
C. Serum bilirubin raises neutralizing thrombin
D. Low serum- protein levels cause factor XIII problems

Q7. Which one of the following would best explain a prolonged bleeding time tests?

A. Hemophilia A
B. Hemophilia B
C. Thrombocytopenia
D. Coumarin use

Q8. A teenage boy with numerous nosebleeds was referred to a physician for evaluation prior to a minor surgery. His prothrombin time(PT) was 11 secs (11-15sec normal), partial thromboplastin time(PTT) was 58 secs (25-40sec normal), and bleeding time was 6.5 min (2-7 min normal). Which of the following is most likely abnormal in this young man?

A. Intrinsic pathway
B. Extrinsic pathway
C. Decreased platelet number
D. Defective platelet

SAQ

- What are the four steps for clotting?
- What is the role of thrombin in haemostasis?
- What are the Natural Intravascular anticoagulants?

Answers
1: B
2: D
3: A
4: D
5: B
6: A
7: C
8: A