Bleeding Disorders

Homostasis

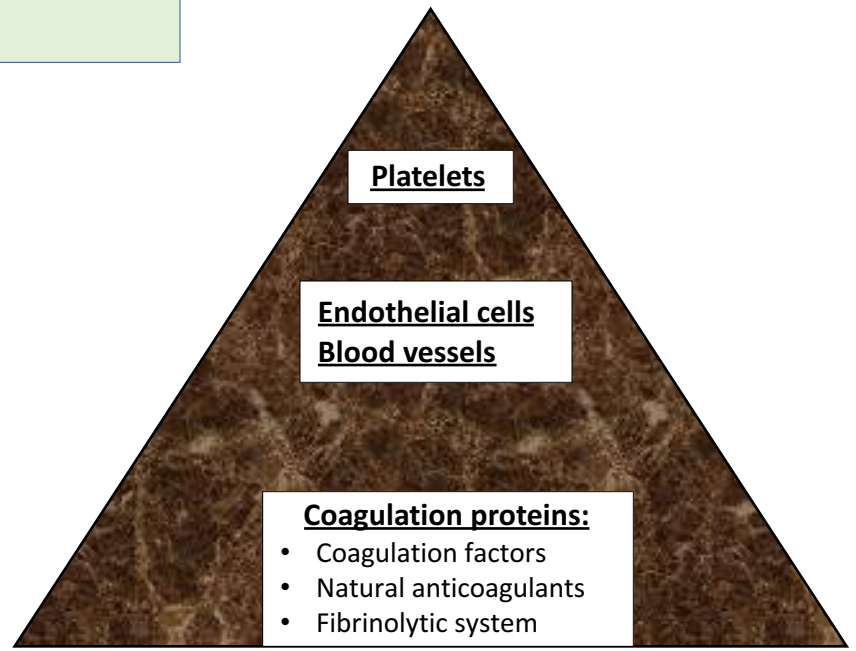
Definition

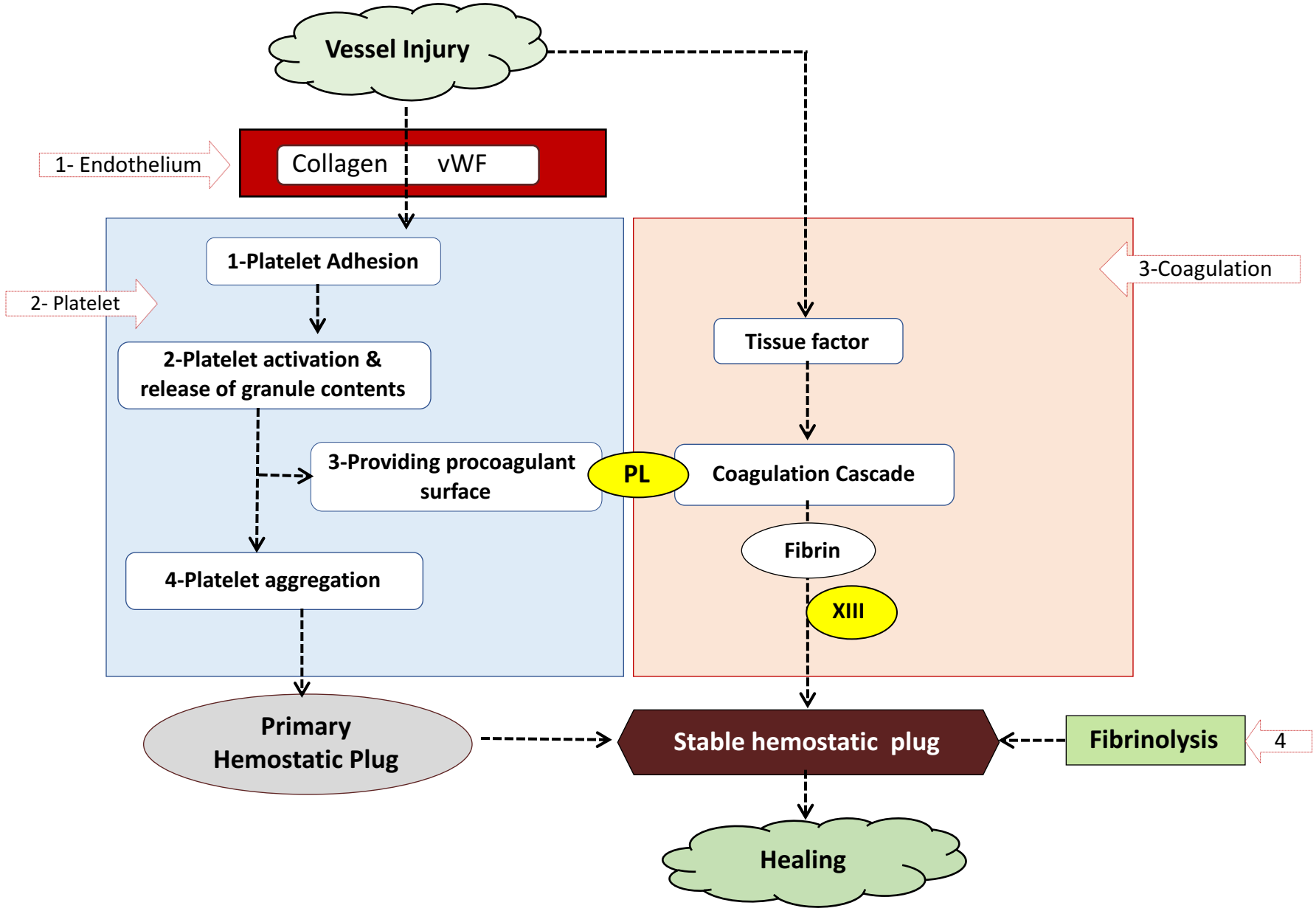
Hemostasis is a dynamic process whereby blood coagulation is initiated and terminated in a rapid and tightly regulated fashion which is followed by fibrinolysis and tissue remodeling (healing).

Functions of homeostasis

1- maintain blood in a fluid and clot-free state .
2- Induce rapid &localized plug at the site of injury.
3- Rapid removal of the plug once hemostasis is maintained

Component of homeostasis



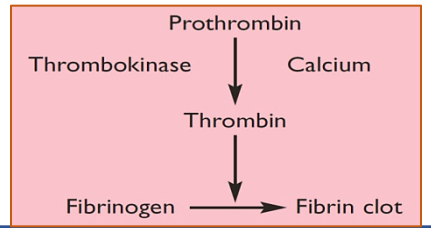

Vessel Injury
1- Endothelium
Collagen
vWF
1-Platelet Adhesion
2- Platelet
2-Platelet activation & release of granule contents
3-Providing procoagulant surface
PL
4-Platelet aggregation
3-Coagulation
Tissue factor
Coagulation Cascade
Fibrin
XIII
Primary Hemostatic Plug
Stable hemostatic plug
Fibrinolysis
4
Healing

Coagulation factors

- Plasma proteins undergo a series of progressive stepwise reactions (coagulation cascade) in order to form coagulation clot. They are synthesized in liver as:
  - Serine proteases :(the majority)
  - Glycoproteins :(FII, FV and FVIII)
  - Transglutaminase: (FXIII)

Coagulation theories

- The classic theory of coagulation was described by Paul Morawitz in 1905. He assembled 4 coagulation factors in which the prothrombin, by calcium activation, yielded thrombin converting fibrinogen to fibrin.



Cascade and Waterfall hypotheses

- In 1964, the Cascade and Waterfall hypotheses was introduced by Macfarlane and Ratnoff respectively
- They suggested that the coagulation could be initiated via :
  - **Intrinsic pathway** : so named because all the components were present in blood.
  - **Extrinsic pathway** : in which the tissue factor (TF) was required in addition to circulating components.
  - The initiation of either pathway resulted in activation of FX and the eventual generation of a fibrin clot through a **common pathway**

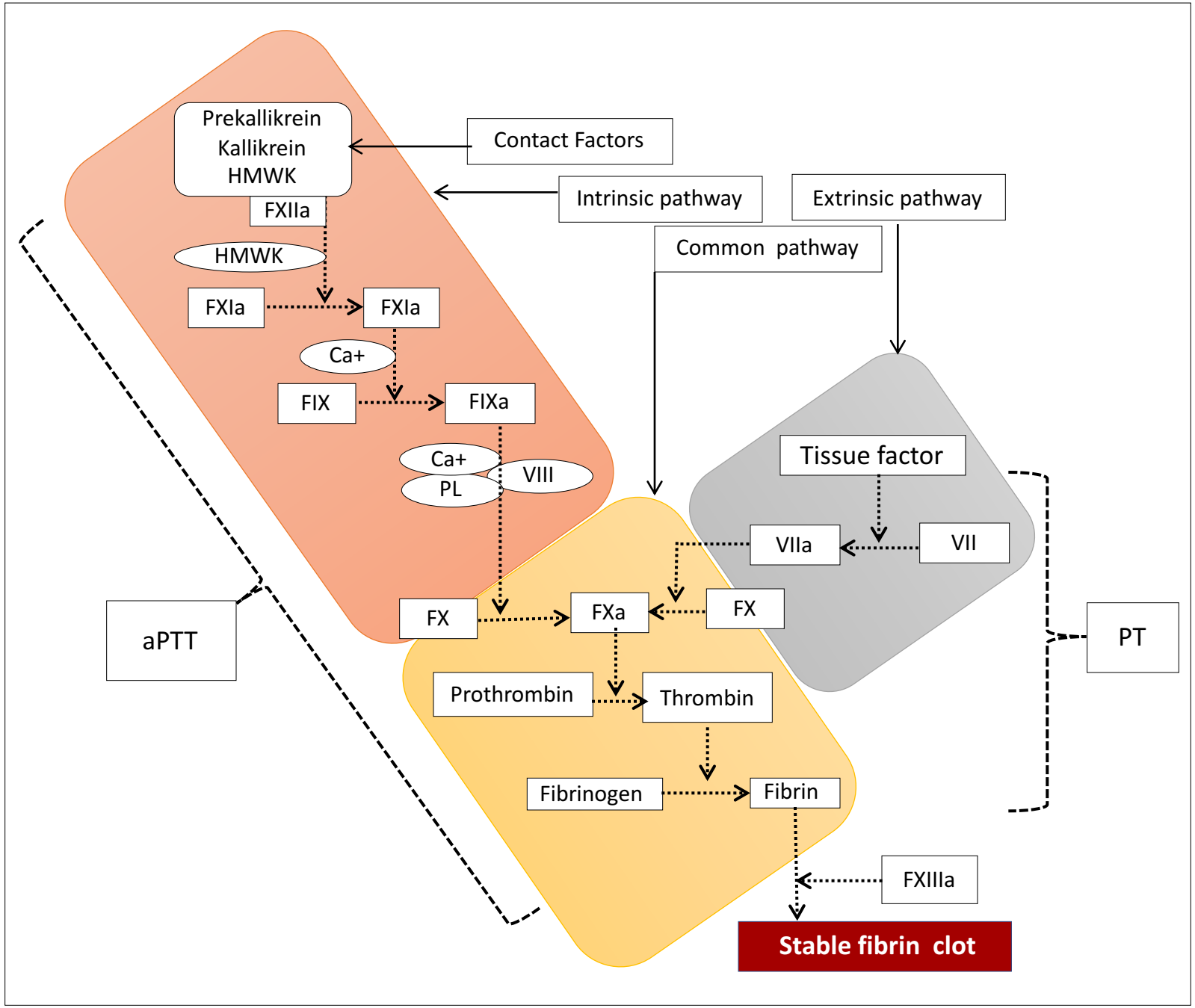
Pitfalls of Waterfall hypothesis

- Coagulation cascade has been widely accepted by physicians and coagulation specialists . Prothrombin time (PT) and activated partial thrombin test (aPTT) were used to test for the adequacy of the extrinsic pathway and intrinsic pathway respectively and are often treated as though they are predictive of clinical bleeding.
- As researches evolved, many unanswered questions has emerged :

1-Why FVIII &FIX deficiencies are associated with severe bleeding while others not?
2- Why an intact extrinsic pathway could not compensate for the lack of FIX or VIII in hemophilia?
3- What is the role of TF expressing cell & Platelets?
4- TF-VIIa is able to activate FIX which is an intrinsic factor !!

The new concept on hemostasis

- Thrombin generation is a complex net work of interactions with positive and negative feed back loops in order to ensure that fibrin clot is limited and localized .
- In vivo, the whole system of hemostasis is highly integrated and the subsystem of hemostasis do not operate independently.
- Normal hemostasis is not possible in the absence of platelets (as procoagulant surface and source of phospholipids) and tissue factor (TF) as the sole initiator for coagulation.
- TF is an integral membrane protein expressed on smooth muscle cells and fibroblasts and released constitutively at biological boundaries such as skin, organ surfaces and vascular adventitia where it functions as a haemostatic envelope.
- The primary control of haemostasis is the anatomical segregation of cells expressing functional TF from other components of the coagulation network present in blood.


Prekallikrein
Kallikrein
HMWK
Contact Factors
Intrinsic pathway
Extrinsic pathway
Common pathway
FXIIa
HMWK
FXIa
FXIa
Ca+
FIX
FIXa
Ca+
PL
VIII
Tissue factor
VIIa
VII
FX
FXa
FX
aPTT
PT
Prothrombin
Thrombin
Fibrinogen
Fibrin
FXIIIa
Stable fibrin clot

The cell based coagulation model

Step 1: Initiation of Coagulation on TF-bearing Cells:

- ❑ Following vascular damage blood coagulation is initiated by exposure of FVII to TF expressing cell . Once bound to TF, zymogen FVII is rapidly converted to FVIIa forming TF-FVIIa complex which is able to activate FX and FIX.
- ❑ In the absence of its activated cofactor FVa, FXa generates only trace amounts of thrombin. Although insufficient to clot fibrinogen, this amount of thrombin is able to activate the amplification phase through activation of FV,VIII and FIX.
- ❑ The initiation phase is rapidly inhibited by tissue factor pathway inhibitor (TFPI) released from endothelial cells or activated platelets.

Step 2: Amplification of the initial stimulus

❑ The small amount of thrombin generated during the initiation phase activates the amplification phase through activation of :

1. Platelets :exposing receptors for activated clotting factors and releasing FV form alpha granules onto platelet surfaces
2. Factor XI : which activates factor IX subsequently.
3. Tenase complex (VIIIa-FIXa) : that activate sufficient amount of FXa
4. Prothrombinase complex (FXa– FVa) : this result **in explosive generation of thrombin** that ultimately leads to generation of fibrin clot.

❑ This step is tightly regulated by:

1. Anti-thrombin (AT): serine protease inhibitor synthesized in the liver. AT forms a stable 1 : 1 complex with several coagulation factors, predominantly thrombin but also to some extent FIXa, FXIa and FXIIIa. Heparin induces > 2000 folds increase in the action of AT.
2. Protein C (PC) and Protein S (PS) : vitamin K–dependent inhibitors that inactivate factors Va and VIIIa. PC is activated by the binding of thrombin to thrombomodulin on endothelial cell receptors for PC (EPCR).Protein S is a cofactor that enhances the action of PC.
3. Fibrinolysis: hemostatic system that generate plasmin at the site of injury in order to limit the extent of the evolving thrombus. It includes plasminogen, plasmin and several activators and inhibitors

Remember!

Congenital deficiency of natural anticoagulants (TFPI, AT, PC or PS) predispose the patients to risk of thrombosis

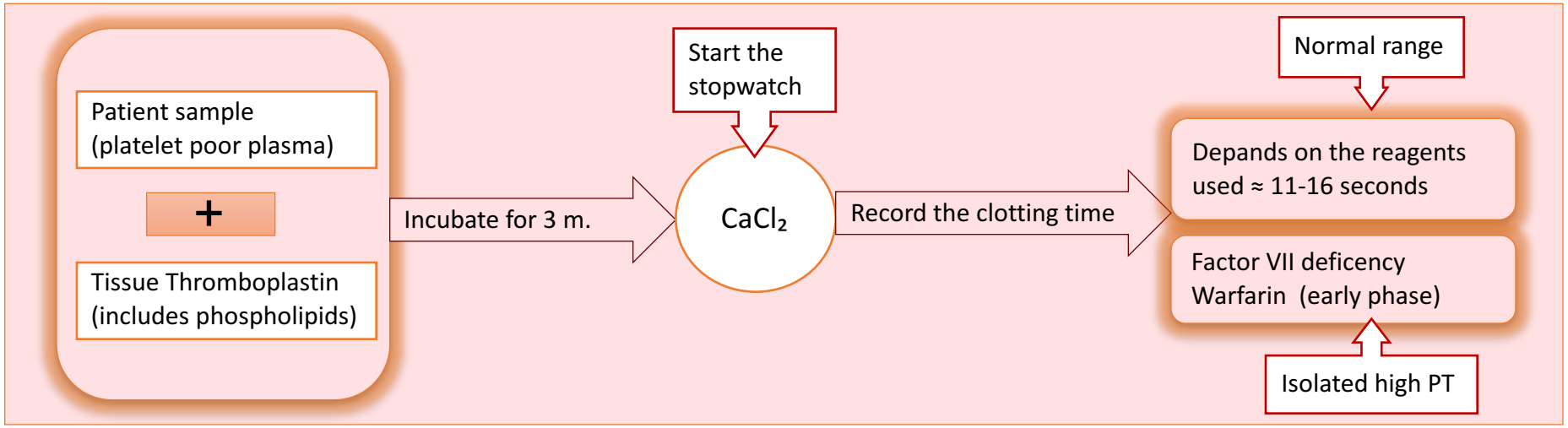
Laboratory investigation of hemostasis

Blood Sample

- Patient should be relaxed and not on physical or emotional stress .The sample should not be taken from a running line, especially if heparin is being used to keep the line open.
- The only specimen acceptable for routine coagulation testing is blood anticoagulated with sodium citrate. the tube must be properly filled (not overfilled and not underfilled) .When hematocrit is abnormal with either sever anemia (<25) or polycythemia (>55), the blood citrate ratio should be adjusted.
- Samples should be delivered as quickly as possible to prevent deterioration of the labile coagulation factors (FV and VIII). The testing should preferably completed within 2hours. However, If the sample is not urgent , It can be stored at -40˚C after proper centrifugation in order to remove platelets (platelet-poor plasma).

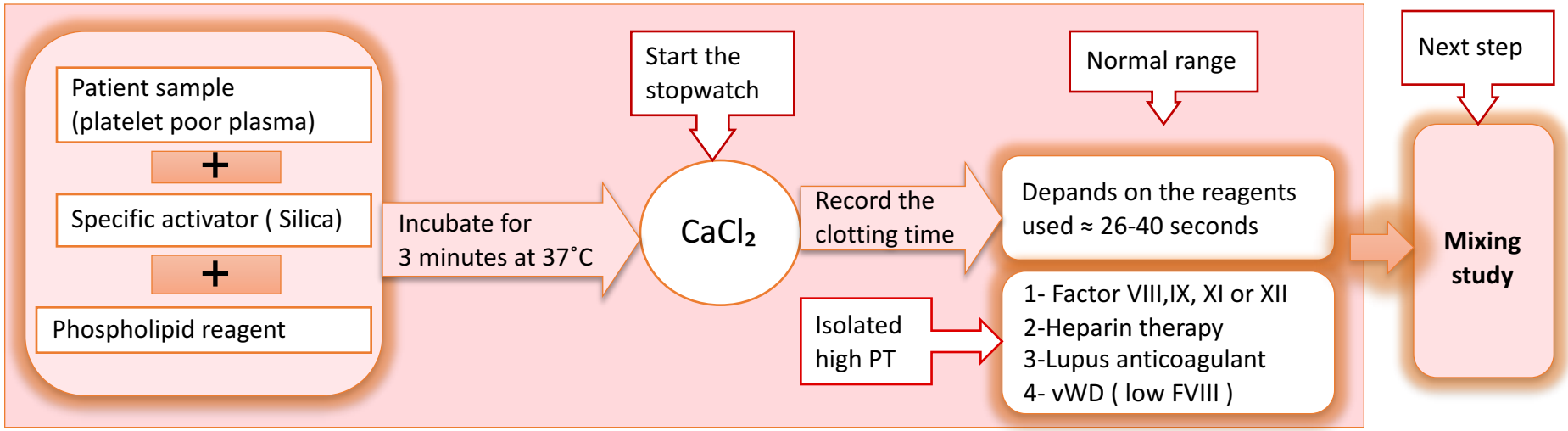
Prothrombin Time PT

An essential coagulation study that tests the extrinsic and common pathways

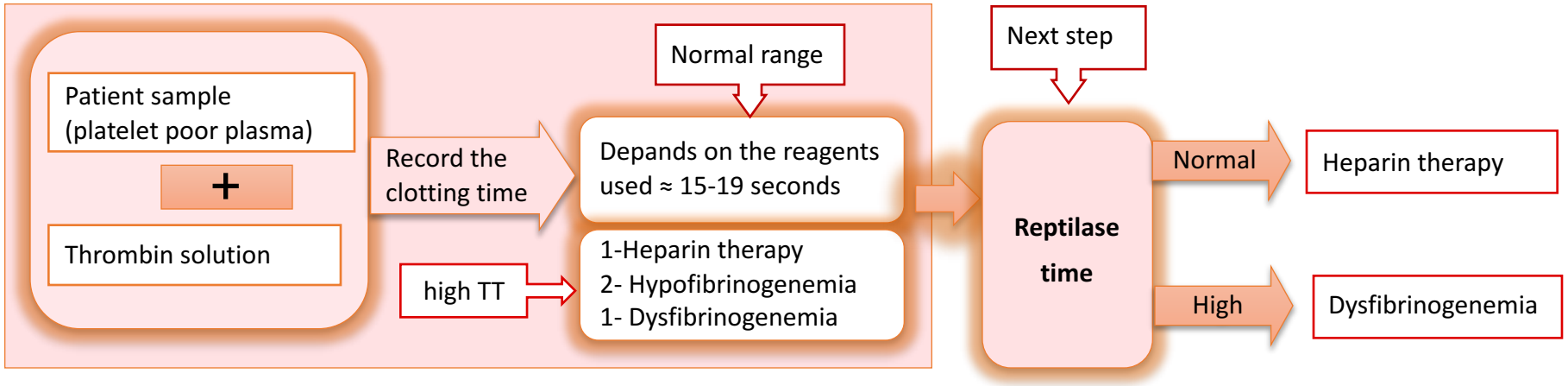


Activated Partial Thromboplastin time (aPTT)

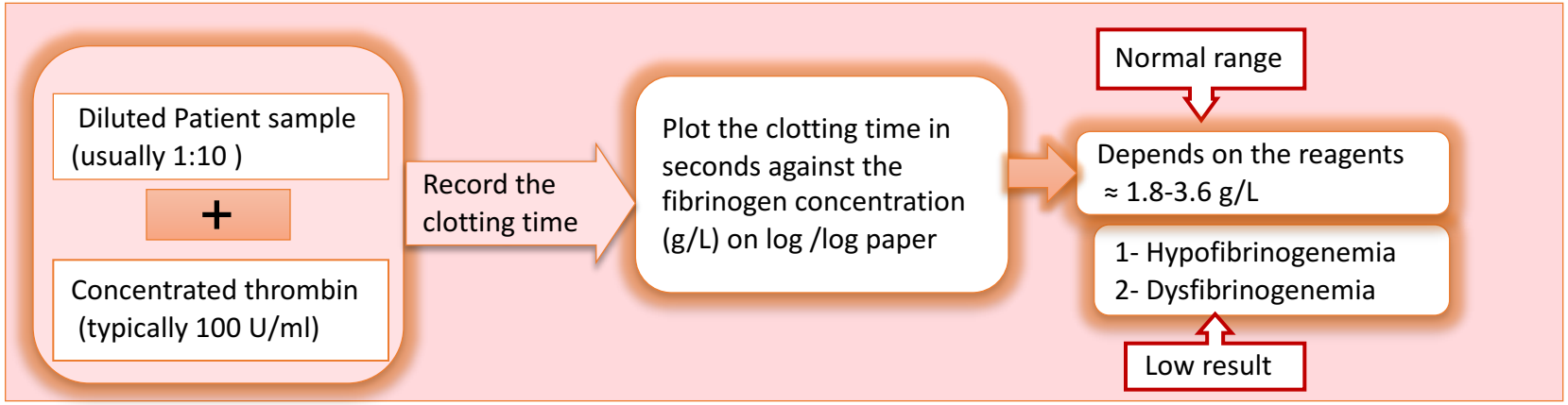
An essential coagulation study that tests the intrinsic and common pathways



Thrombin Time (TT)

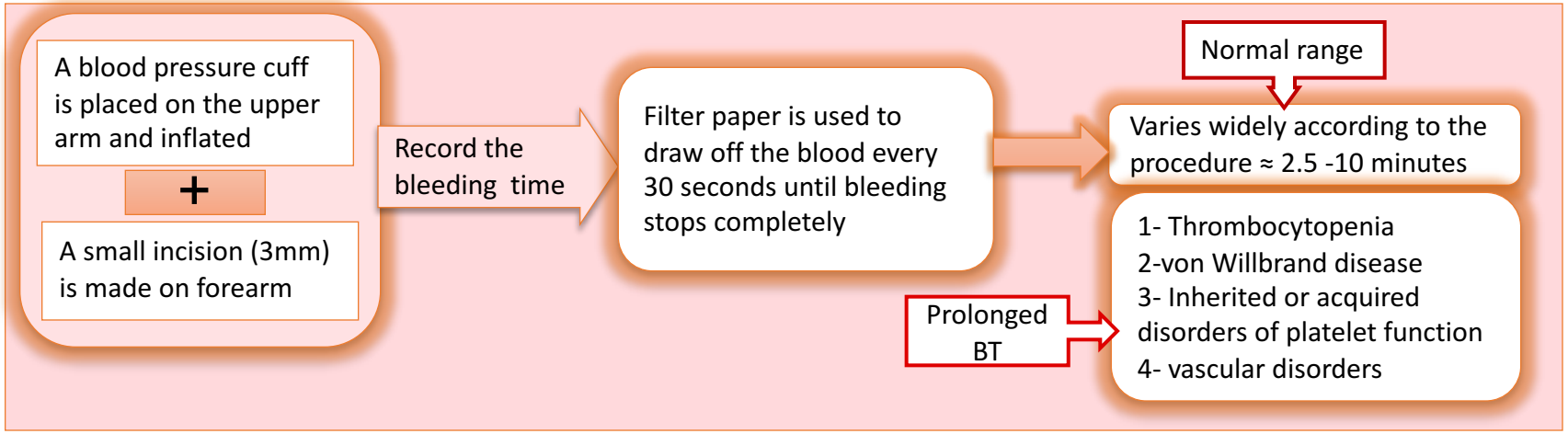


Fibrinogen (Claus method)



Bleeding Time (BT)

Bleeding time is a crude test of hemostasis. It indicates how well platelets interact with blood vessel walls and vWD to form blood clots. The BT tests the function of platelets , vWF, the integrity of skin and blood vessel walls. It is usually normal or minimally prolonged with coagulation factor deficiencies . In general, BT has poor sensitivity and reproducibility and rarely used .



Bleeding Disorders

Bleeding disorders result from:

1 Vascular disorders.

2 Thrombocytopenia.

3 Defective platelet function.

4 Defective coagulation.

Vascular bleeding disorders

- Heterogeneous group of conditions characterized by easy bruising and spontaneous bleeding from the small vessels.
- The underlying abnormality is either in the vessels themselves or in the perivascular connective tissues.
- Coagulation tests are normal.

Inherited vascular disorders

- **Hereditary haemorrhagic telangiectasia**

AD, Rare.

- **Connective tissue disorders**

In the Ehlers–Danlos syndromes there are hereditary collagen abnormalities with purpura resulting from defective platelet adhesion.

Acquired vascular defects

Simple easy bruising is a common benign disorder which occurs in otherwise healthy women, especially those of child-bearing age.

Senile purpura caused by atrophy of the supporting tissues of cutaneous blood vessels

The Henoch–Schönlein syndrome is usually seen in children and often follows an acute upper respiratory tract infection

Congenital Coagulation Disorders

- haemophilia A (factor VIII)
- haemophilia B (factor IX)
- VWD

Hemophilia

Definition

A group of hereditary genetic disorders characterized by deficiency of coagulation factors leading to bleeding predisposition

Classification of hemophilia

Hemophilia

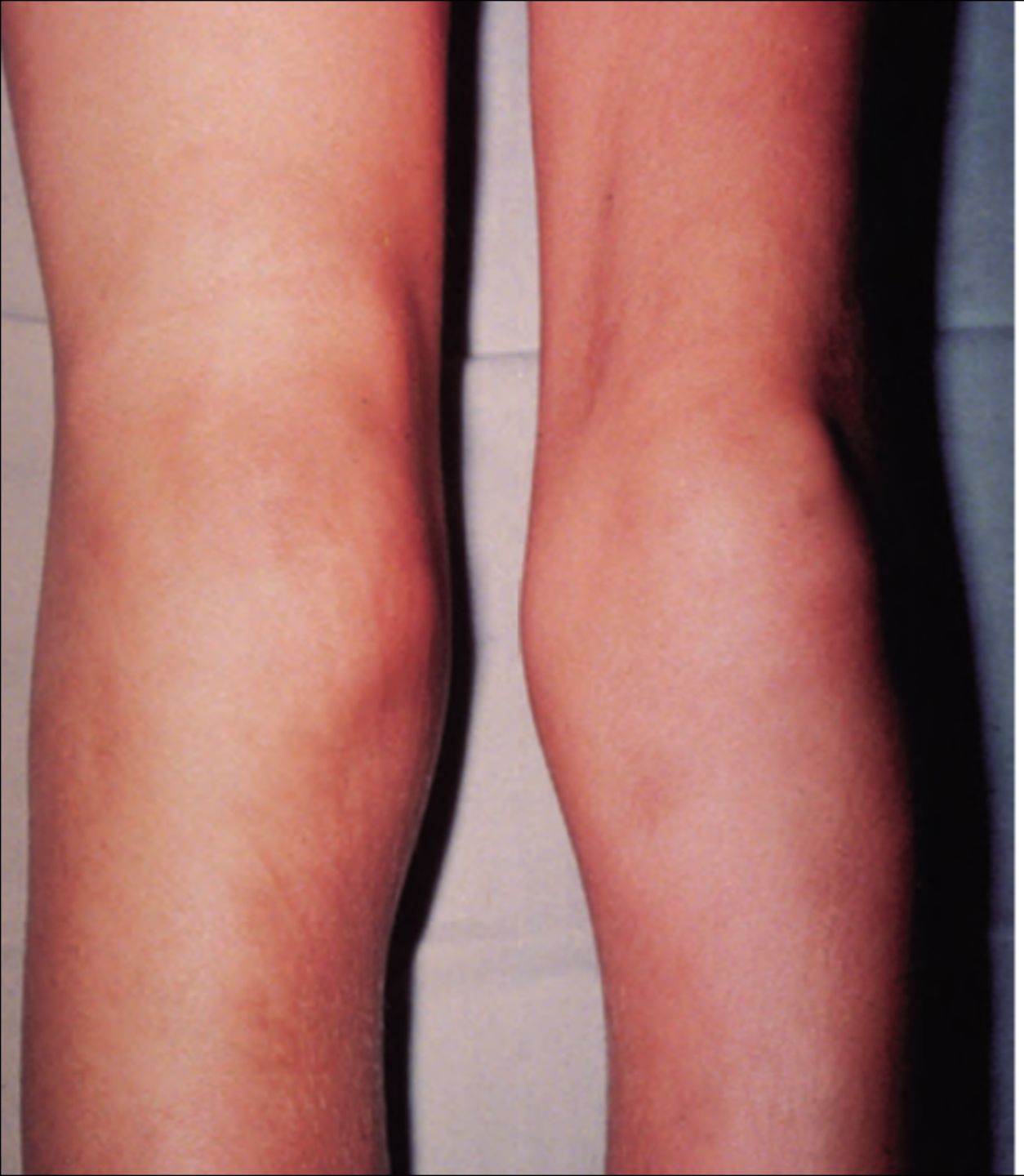
	Hemophilia A	Hemophilia B	Hemophilia C
Cause	•Factor VIII deficiency	•Factor IX deficiency	•Factor XI deficiency
Inheritance	• X-linked recessive	• X-linked recessive	• Autosomal recessive
Incidence	• 85% of hemophilia (1:5000)	• 15% of hemophilia (1:30000)	• Very rare (1:100,000)
Severity	• Severity correlate with the factor level	• Severity correlate with the factor level	• Mild bleeding tendency even with sever factor deficiency
Clinical Feature	• Hemarthrosis and muscle hematoma	• Hemarthrosis and muscle hematoma	• bleeding post injuries, epistaxis and menorrhagia
Lab investigation	• Prolonged aPTT , Corrected by mixing study and Low FVIII	• Prolonged aPTT , Corrected by mixing study and Low FIX	• Prolonged aPTT , Corrected by mixing study and Low FXI
Late complication	• Development of inhibitors and joints destruction	• Development of inhibitors and joints destruction	• Prognosis is excellent

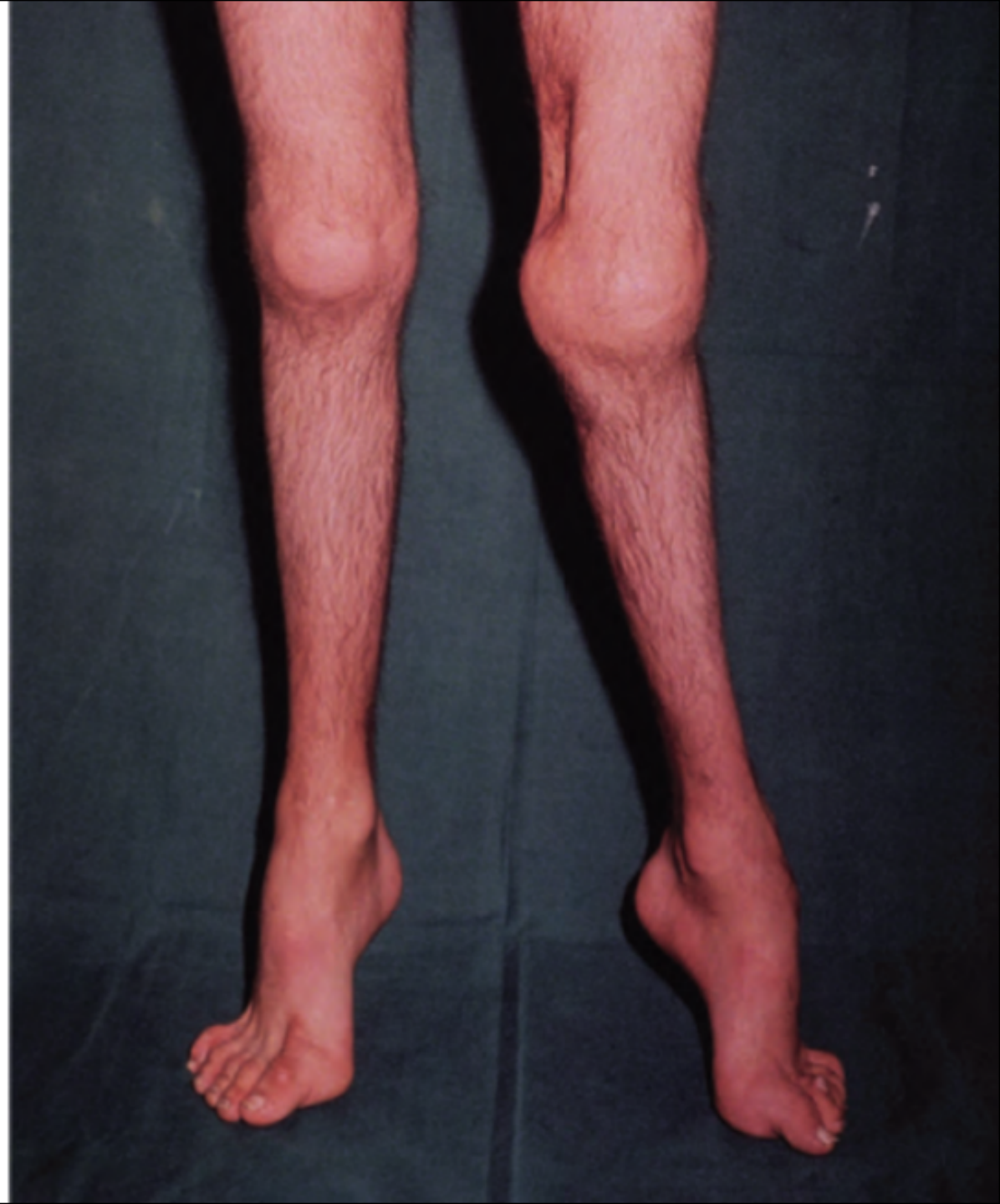
Hemophilia A

- Deficiency of factor VIII results from an abnormality in the factor VIII gene, which lies at the the long arm of the X-chromosome.
- Ranging form single-point mutations to large deletions.
- The prevalence of this disorder is about one per 10 000 males.
- Females with haemophilia have been observed extremely rarely and these are either homozygotes for the abnormal gene or are heterozygotes in whom the normal X-chromosome has not produced sufficient quantities of factor VIII due to lyonization.
- In the plasma, factor VIII is only found complexed with VWF, which acts as a carrier and prolongs its plasma half-life.

Clinical features

- Infants may develop profuse post-circumcision haemorrhage or joint and soft tissue bleeds and excessive bruising.
- Recurrent painful haemarthroses and muscle haematomas dominate the clinical course of severely affected patients
- If inadequately treated, lead to progressive joint deformity and disability.
- Intracranial bleeding is the most common cause of death from the disease itself.





Diagnosis

- The possibility of haemophilia is suggested by the finding of a normal PT and a prolonged APTT.
- Confirmation is by a specific assay of factor VIII coagulant activity with normal VWF.

Correlation of coagulation factor activity and disease severity in haemophilia A or B.

Coagulation factor activity (percentage of normal)	Clinical manifestations
<1	Severe disease Frequent spontaneous bleeding into joints, muscles, internal organs from early life Joint deformity and crippling if not adequately prevented or treated
1–5	Moderate disease Bleeding after minor trauma Occasional spontaneous episodes
>5	Mild disease Bleeding only after significant trauma, surgery

Treatment

- Treatment should be given at the earliest sign of spontaneous or post-traumatic bleeding.
- Treatment consists of intravenous injections of factor VIII concentrate.
- Guidelines exist for the plasma level to be achieved for different types of haemorrhage.
- A controlled trial has proven that regular prophylaxis is far superior to on-demand treatment.
- Approximately 25% of patients with haemophilia, usually after treatment with factor VIII on 10-20 occasions, develop antibodies that inhibit its functional activity.

Treatment

- Haemorrhage in patients with high-titre inhibitors may require treatment with ‘bypassing agents’ such as recombinant factor VIIa or FEIBA (factor eight inhibitor bypassing activity; that is, a plasma-derived activated prothrombin complex concentrate), which activate the coagulation cascade below the level of factor VIII.
- The administration of factor VIII may be avoided in mild to moderate haemophilia by using the vasopressin analogue desmopressin (DDAVP), which causes a temporary increase in factor VIII and VWF by provoking the release of these factors from endothelial cells. DDAVP is used intravenously, subcutaneously or intranasally.

Haemophilia B (Factor IX deficiency, Christmas disease)

- The clinical features and inheritance of factor IX deficiency are identical to those in factor VIII deficiency.
- Factor IX deficiency affects about 1 in every 50 000 males.
- The factor IX gene is located on the long arm of the X-chromosome.
- The APTT is prolonged and the PT normal. The diagnosis can be made by assay of the factor IX level.
- Plasma-derived factor IX concentrate or recombinant factor IX is available and should be administered intravenously as soon as spontaneous or post-traumatic bleeding starts.
- Factor IX has a longer half-life in the plasma (18-24 hours) than factor VIII and hence can be given at less frequent intervals.

Von Willebrand disease

- It is an autosomal disorder characterized by mild, moderate or severe bleeding.

❖VWF has two function:

  - binds platelets to subendothelial tissues.
  - It acts as a carrier for factor VIII.

- The most common inherited bleeding disorder withprevalence of up to 1%
- Most mild cases are undiagnosed.
- The bleeding results from either a qualitative abnormality or a quantitative deficiency of VWF.
- The reduction in VWF results in a reduction in factor VIII concentration (can be misdiagnosed as hemophilia A).

VWD

❖VWD has been divided into three types:

- Types 1 (most frequent) partial reduction, AD
- Type 3 there is nearly complete absence of VWF molecules, AR
- Type 2 there are qualitative abnormalities, AD or AR

- Spontaneous bleeding is usually confined to mucous membranes and skin most commonly epistaxes and ecchymoses.

- Bleeding into joints and muscles is rare except in type 3 disease.

Diagnosis

- The laboratory findings include:
  - Prolonged PFA closure time.
  - Usually a prolonged APTT.
  - Reduced factor VIII clotting activity
  - Reduced levels of VWF antigen or activity.
  - Impaired ristocetin-induced platelet aggregation.

Treatment

- For type 1 disease, desmopressin (DDAVP) is the first line treatment.
- DDAVP increases plasma levels of both VWF and factor VIII.
- Very high purity VWF concentrate may be used.
- The antifibrinolytic drug (tranexamic acid) may be used for treating epistaxis or menorrhagia.

Deficiency of other clotting factors

Single deficiencies of factors other than VIII and IX are rare.

All factors deficiency except contact factor (e.g. factor XII) give rise to bleeding disorders of varying degrees of severity.

Acquired bleeding disorders

- Liver disease.
- Vitamin K deficiency
- DIC
- Acquired hemophilia.
- Drugs (heparin, warfarin, tPA, rivaroxaban, dabigatran)

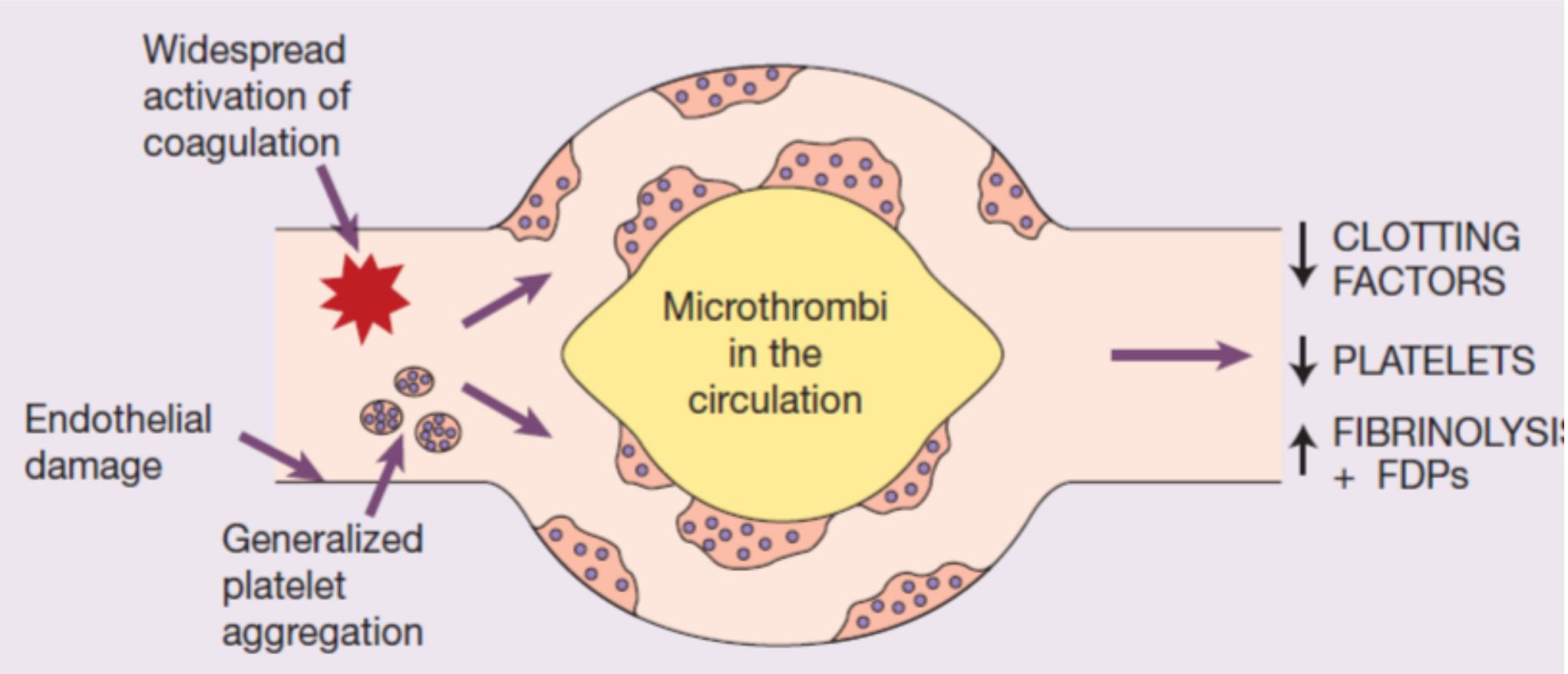
Vitamin K Deficiency

- Fat-soluble obtained from green vegetables and bacterial synthesis in the gut.
- Hemorrhagic disease of the newborn:
  - Caused by liver cell immaturity, lack of gut bacterial synthesis of the vitamin and low quantities in breast milk.
  - usually on the second to fourth day of life, but occasionally during the first 2 months.
  - PT and APTT are both prolonged.

Disseminated intravascular coagulation (DIC)

- Generalized activation of the clotting system followed by marked activation of the fibrinolytic system.
- Acute DIC may be associated many serious/lifethreatening diseases.
- Clotting cascade is activated in various ways(tissue damage, collagen exposure, release of TF and other procoagulants)
- Activation of the cascade leads to the generation and dissemination of large amounts of thrombin in the circulation, the activation of platelets and the formation of intravascular microthrombi.
- As a consequence of the fibrin formation, the fibrinolytic mechanism is activated, resulting in high concentrations of FDPs, including D-dimers.

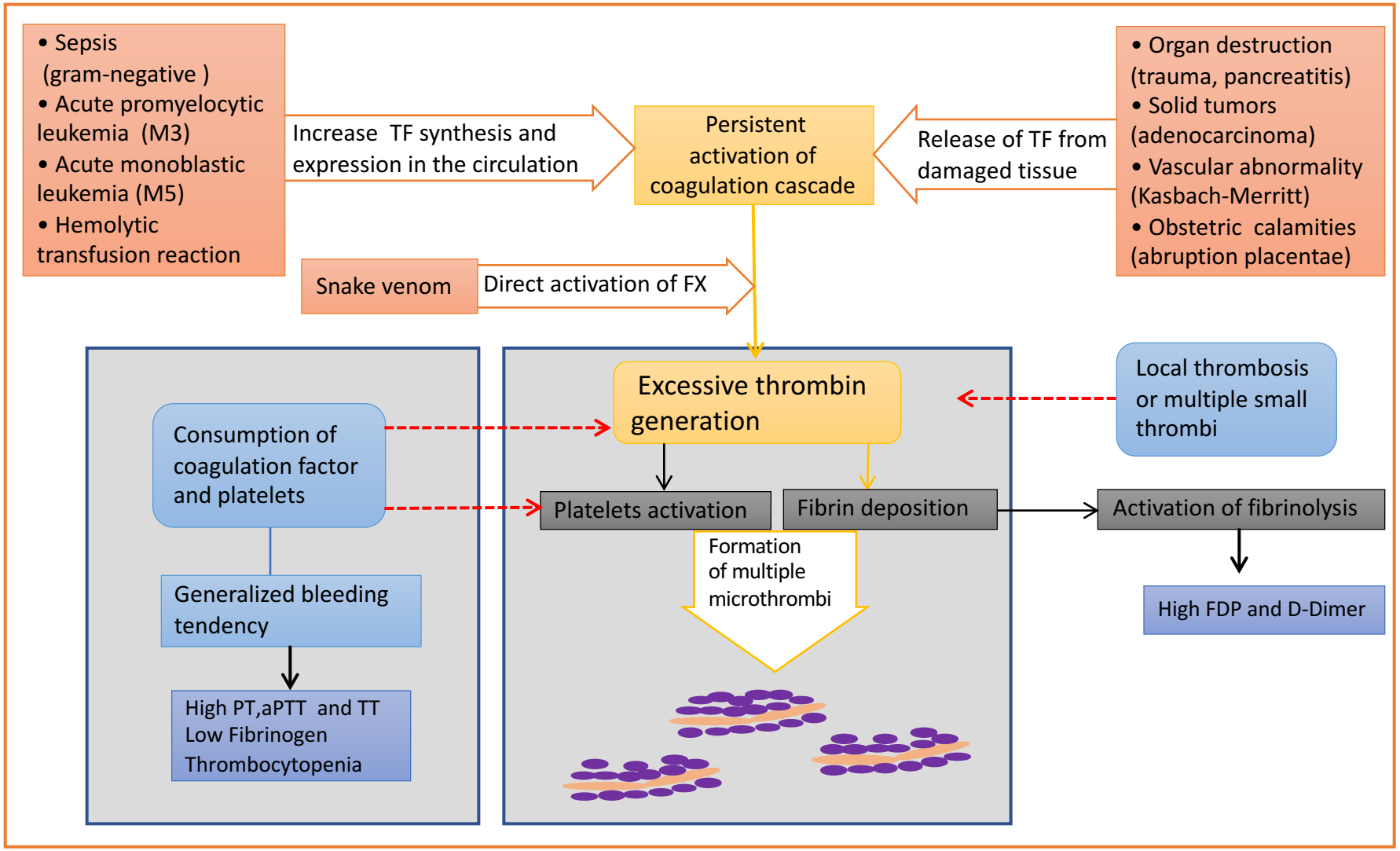
DIC


Widespread activation of coagulation
Endothelial damage
Generalized platelet aggregation
Microthrombi in the circulation
CLOTTING FACTORS
PLATELETS
FIBRINOLYSIS + FDPs

Disseminated Intravascular Coagulation (DIC)

Definition

DIC is widespread activation of coagulation system leading to intravascular fibrin deposition and consumption of platelet and coagulation factors which can be manifested as bleeding (85%) or thrombosis (15%)



Clinical features

- The picture is dominated by bleeding which can be from multiple sites especially GIT bleeding and urogenital tract .In obstetric cases ,vaginal bleeding is particularly severe . Bleeding form wounds or venipuncture sites are common as well.
- Less frequently ,microthrombi may present with skin lesions ,renal failure or fingers gangrene

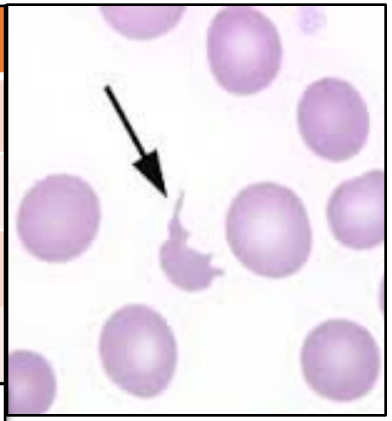
Laboratory diagnosis

- Hemolytic anemia is a common presentation in DIC associated with RBC fragmentation because of damage caused when passing through fibrin strands in small vessels .
- Sign of hemolysis are high lactate dehydrogenase (LDH) and high bilirubin .

Diagnostic score

Score	0	1	2	3
Platelet count	>100	50-100	<50	
FDP	Normal		Moderate increase	Strong increase
prolonged PT	by < 3 sec	3-6 sec	>6 sec	
Fibrinogen	>1 g/l	< 1 g/l		

Score ≥ compatible with overt DIC ; score < 5 suggestive and should be repeated after 2 days



Treatment

Treatment of the underlying cause and proper supportive therapy are the most important approachs.
For bleeding : supportive therapy with fresh frozen plasma (to compensate for factor deficiency) and platelet concentrate
Carioprecipetate provides more concentrated factors and may be required .
Thrombosis: heparin and anti-platelet drugs are used to treat patients with thrombosis. However, the benefits should outweigh the bleeding risks

Causes of DIC

Infections **Gram-negative and meningococcal septicaemia** **Clostridium welchii septicaemia** **Severe falciparum malaria** **Viral infection – varicella, HIV, hepatitis, cytomegalovirus**	**Malignancy** **Widespread mucin-secreting adenocarcinoma** **Acute promyelocytic leukaemia**	**Obstetric complications** **Amniotic fluid embolism** **Premature separation of placenta** **Eclampsia; retained placenta** **Septic abortion**
Hypersensitivity reactions Anaphylaxis Incompatible blood transfusion	Widespread tissue damage Following surgery or trauma. After severe burns	Vascular abnormalities Kasabach–Merritt syndrome Leaking prosthetic valves Cardiac bypass surgery Vascular aneurysms
Miscellaneous Liver failure Pancreatitis Snake and invertebrate venoms Hypothermia Heat stroke Acute hypoxia Massive blood loss		

DIC

- The haemorrhagic manifestations may be so severe in acute DIC as to lead to death
- In chronic DIC, the haemorrhagic tendency may be mild or moderate.
- Some patients with chronic DIC are asymptomatic because the activation of the clotting and fibrinolytic systems is finely balanced and the production of clotting factors and platelets is sufficiently increased to compensate for their increased consumption.

Diagnosis

1 The platelet count is low.

2 Fibrinogen concentration is low.

3 High levels of fibrin degradation products (D-dimers).

4 The PT and APTT are prolonged.

5 RBCs fragments in blood smear.

Compensation by the liver may render some of the coagulation

tests normal.

Treatment

- Treatment is aimed at preventing further coagulation by removal of the initiating cause.
- Supported with transfusions of blood, fresh-frozen plasma and platelet concentrates in order to restore blood volume and replace clotting factors and platelets.

Acquired haemophilia

- Acquired hemophilia is a rare but life-threatening condition
- Caused by the development of autoantibodies (inhibitors) directed against plasma coagulation factors, most frequently factor VIII (FVIII).
- Could be idiopathic or secondary to underlying condition (autoimmune disease, infection, malignancy,…)
- More common in the elderly
- Treated with ‘bypassing agents’ such as recombinant factor VIIa or

FEIBA and immune suppression.

Massive transfusion syndrome

- Blood loss results in reduced levels of platelets, coagulation factors and inhibitors.
- Further dilution of these factors occurs during replacement with red cells.
- Some protocols include 1 : 1 : 1 for red cells, platelet packs and FFP

- Congenital coagulation disorder caused by all of these factors except:

a) VWD
b) Haemophilia B
c) Haemophilia A
d) Warfarin

- The confirmation of Haemophilia B by:

a) A prolonged PT and a prolonged APTT
b) A specific assay of Factor VIII
c) A prolonged PT and normal APTT
d) A specific assay of Factor IX

- Disseminated intravascular coagulation (DIC) diagnosis can be made by:

a) The platelet count is high
b) RBC normal morphology in blood smear
c) High levels of fibrin degradation products (D-Dimers)
d) Fibrinogen concentration is high


Thank you!
TESCHIO 99