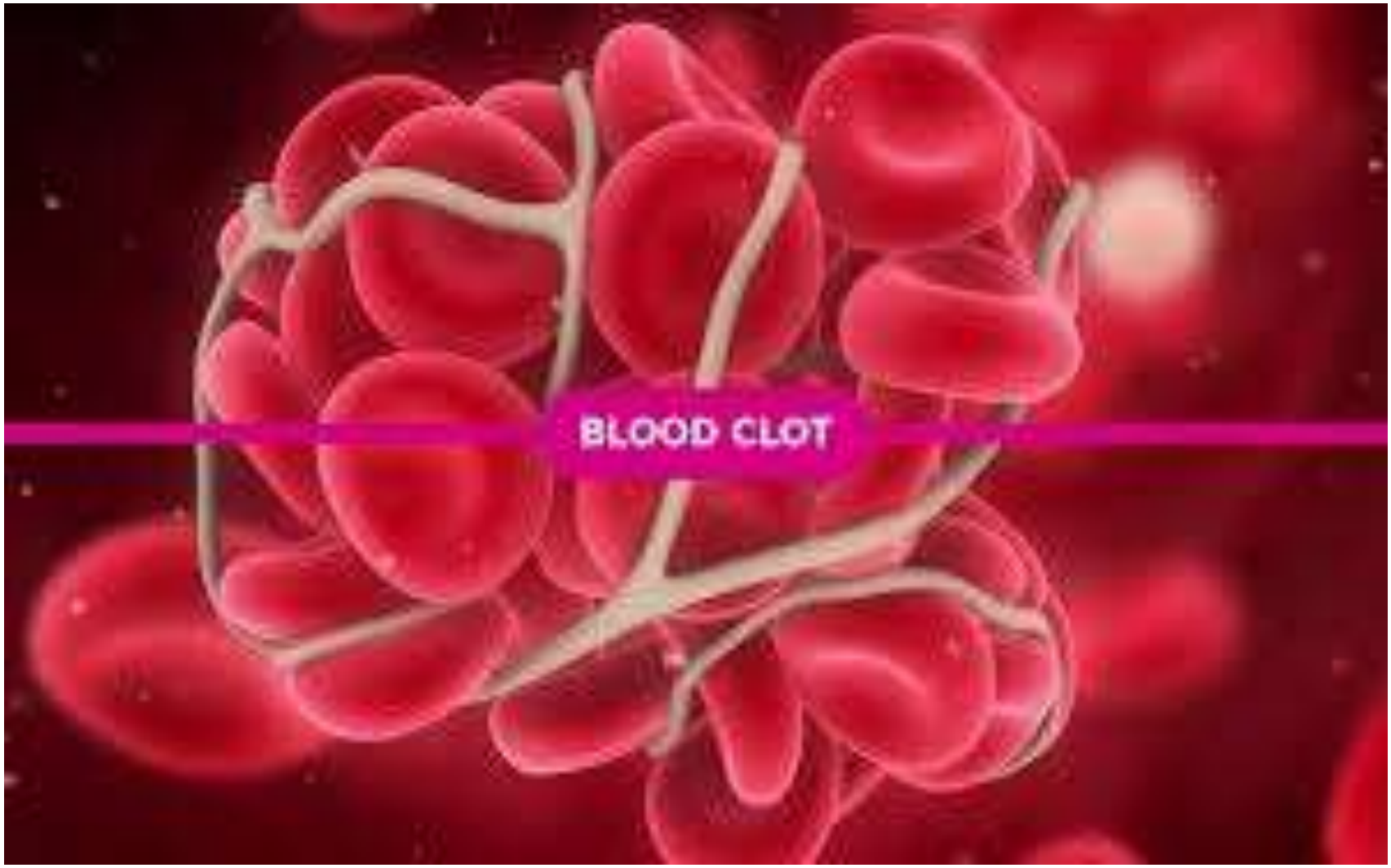
HEMOSTASIS for first year students by DR RIFFAT



INTRODUCTION

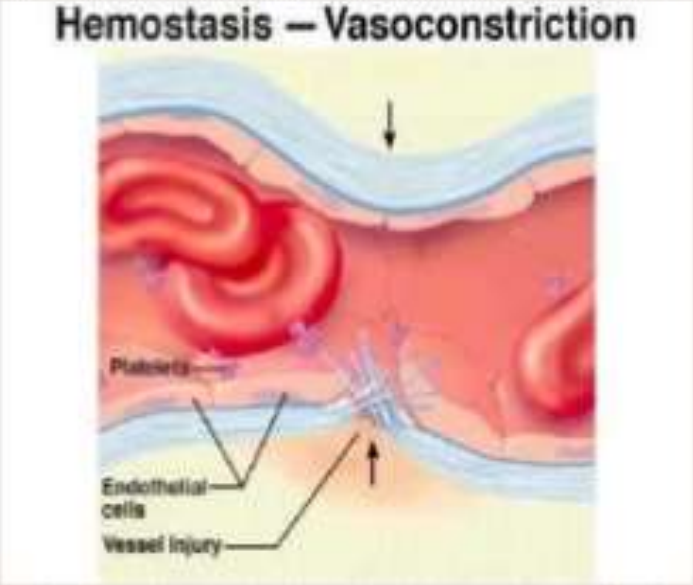
Hemostasis is the process of forming clots in the walls of damaged blood vessels and preventing blood loss while maintaining blood in the fluid state within the vascular system.

EVENTS IN HAEMOSTASIS

Haemostasis means prevention of '**Blood Loss**'. Haemostasis is achieved by several mechanism:-

- Vascular constriction
- Formation of platelet plug
- Formation of blood clot
- Growth of fibrous tissue into the clot.

VASCULAR CONSTRICTION



The contraction results from:-

- Local myogenic spasms
- Local autacoid factors
- Nervous reflexes

Platelets release, ***thromboxane-A_2 which*** is responsible for vasoconstriction of smaller vessels.

The more severely a vessel is traumatized, the greater the degree of vascular spasm.

Mechanism

Vasoconstriction is as a result of increased Ca ion conc. in smooth muscles.

Hormonal components :

Circulating epinephrine & activation of sympathetic nervous system.

Interact with cell surface adrenergic receptors

↓

Signal transduction

↓

Increased intercellular Ca from sarcoplasmic reticulum

↓

Ca-calmodulin complex

↓

Activates myosin light- chain kinase

↓

stimulate cross bridge cycle

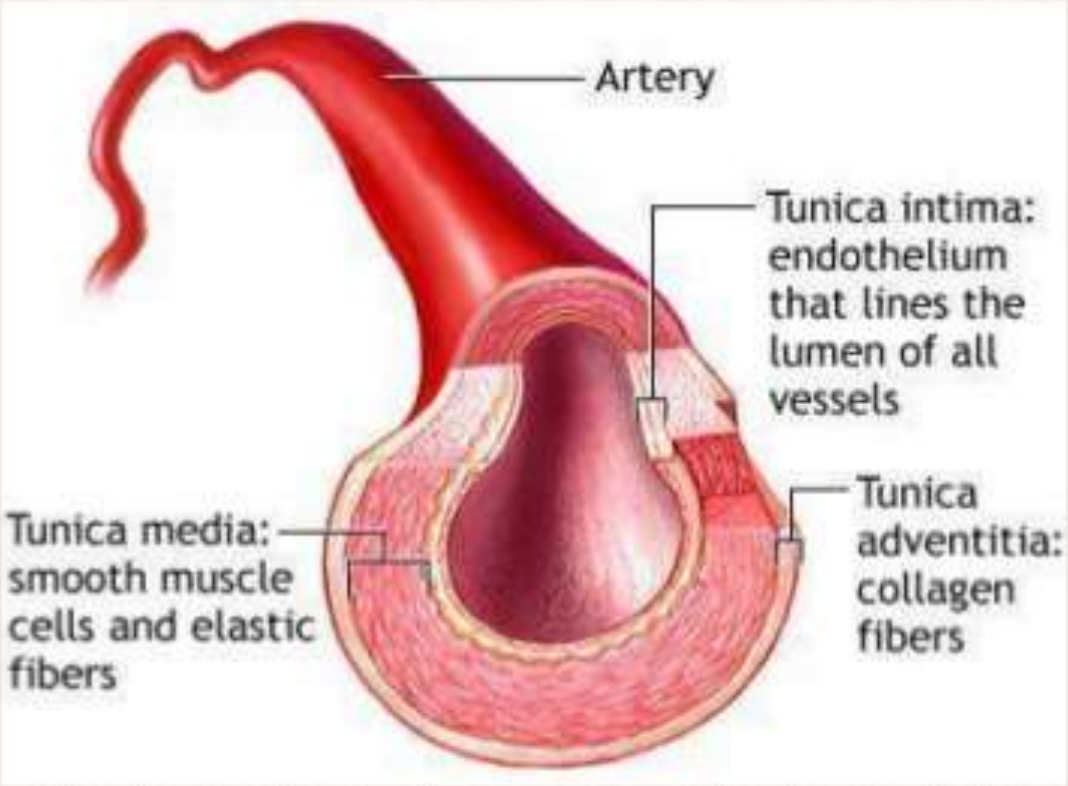
- Factors :
- -Exogenous – temperature (cold)
- -Endogenous-Autonomic nervous system, hormones and mechanisms inherent to vasculature (myogenic response)

-Medications: antihistamines , amphetamines , cocaine

FORMATION OF PLATELET PLUG

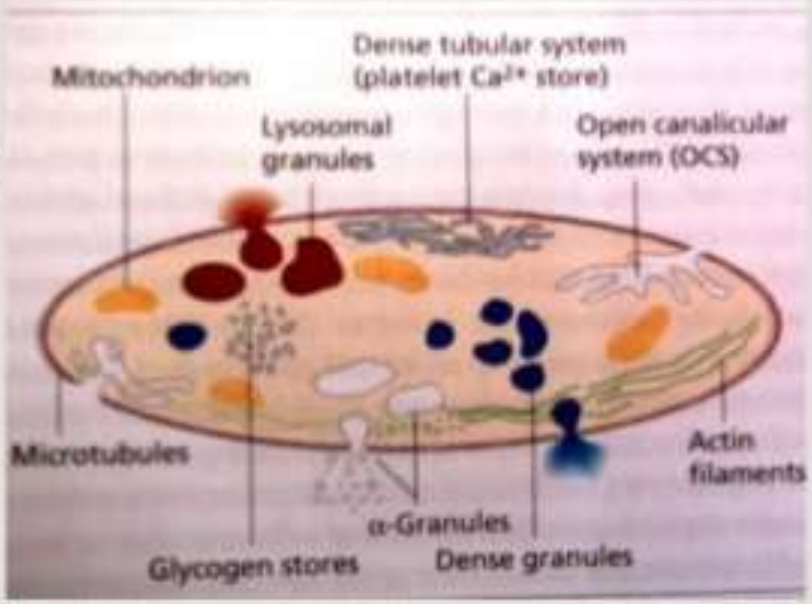
- PHYSICAL AND CHEMICAL CHARACTERISTICS OF PLATELETS
- MECHANISM OF PLATELET PLUG
  - Platelet adhesion
  - Platelet activation
  - Platelet aggregation
  - Formation of temporary haemostatic plug

NORMAL ARCHITECTURE OF A BLOOD VESSEL



PLATELETS

- Platelets are enucleate cells
- 1-4 micro meters in size
- Normal blood concentration of platelets is 1.5L-3L/micro liters
- Formed in bone marrow from *megakaryocytes*
- T-1/2 is 8-12 days
- Eliminated from the circulation by tissue macrophages.



CONTD…

Platelet cytoplasm contains active factors such as:-

- Actin and myosin molecules

  Thrombosthenin is another contractile protein which can cause platelets to contract.
- ER and the Golgi apparatus
- Mitochindria and enzyme system.

PLATELET-CELL MEMBRAN

- Surface coat of ***glycoproteins***

  a) repulse adherence to normal endothelium

  b) but adherence to injured vessel wall i.e., endothelial cells, and even more to the exposed collagen from deep with in the vessel wall
- Large amounts of ***phospholipids*** – activates multiple stages in blood-clotting process.

PLATELET AGGREGATION

- The activated sticky platelets stick to each other form Platelet aggregation
- TXA2 powerful vasoconsrictor &mobilisation Ca from dense tubular system –activates myosin -actin cause release reaction –Platelet aggregation
- This involves a series of self sustaining events
- Leads to formation of platelet plug

BLOOD COAGULATION IN RUPTURE VESSEL

- Third mechanism for hemostasis is formation of blood clot
- Clot begins to develop-

  severe trauma-15 to 20 sec

  minor trauma-1 to 2 min

MECHANISM OF BLOOD COAGULATION

BASIC THEORY

GENERAL MECHANISM

GENERAL MECHANISM

- In response to rupture of the vessel or damage to the blood itself-formation of ***prothrombin activator***
- Prothrombin activator catalyzes conversion of prothrombin to thrombin
- Thrombin catalyzes fibrinogen into fibrin fibers.

Contd...

- Then Fibrin Stabilizing Factor is released from platelets entrapped in the clot
- Same thrombin that causes fibrin formation activates the Fibrin Stabilizing Factor, before FSF can have effect on fibrin fibers
- Activated FSF forms strong covalent bond b/w monomer of the fibrin and multiple cross linkage b/w adjacent fibrin fibers.

BLOOD CLOT

- The clot is a meshwork
- Fibrin fibers also adhere to damaged surfaces of blood vessels.

CLOT RETRACTION-SERUM

- Contraction causes expression of fluid from clot-serum
- Platelet contractile proteins contributes greatly to the clot retraction by activating ***Platelet Thrombosthenin***
- They also helps compress fibrin mesh work into smaller mass.
- As the clot contracts, the edges are further pulled together, contributing ultimate state of ***Hemostasis.***

VISCIOUS CIRCLE OF CLOT FORMATION

- Positive feed back for clot formation
- Most important cause of this is the the proteolytic action of thrombin
- For instance, thrombin has a direct proteolytic effect on prothrombin itself, hence more of thrombin formation
- Critical amount of thrombin causes more blood clotting and hence further more production of thrombin and hence called a vicious circle of clotting.

INITIATION OF COAGUALTION

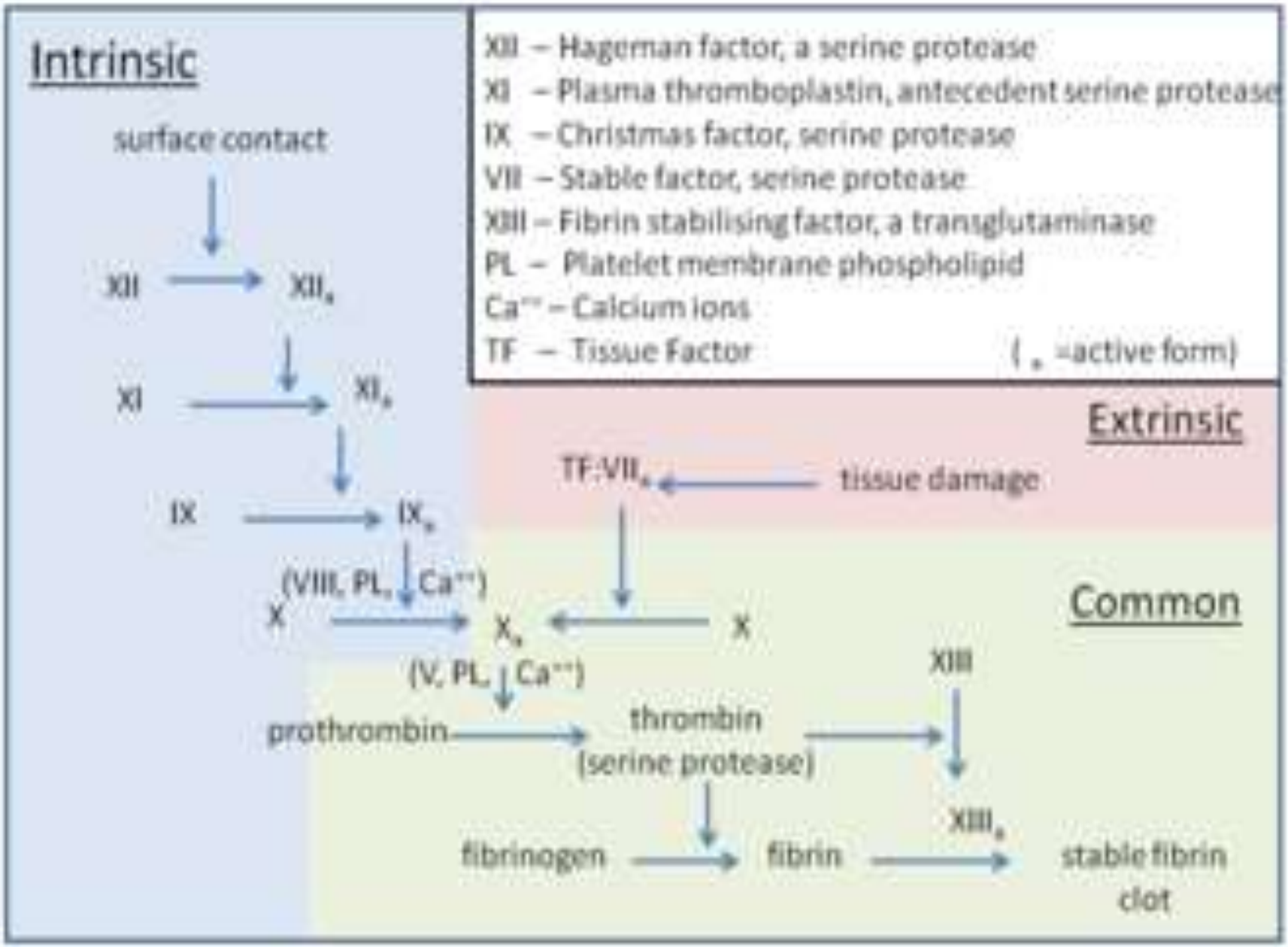
FORMATION OF PROTHROMBIN ACTIVATOR

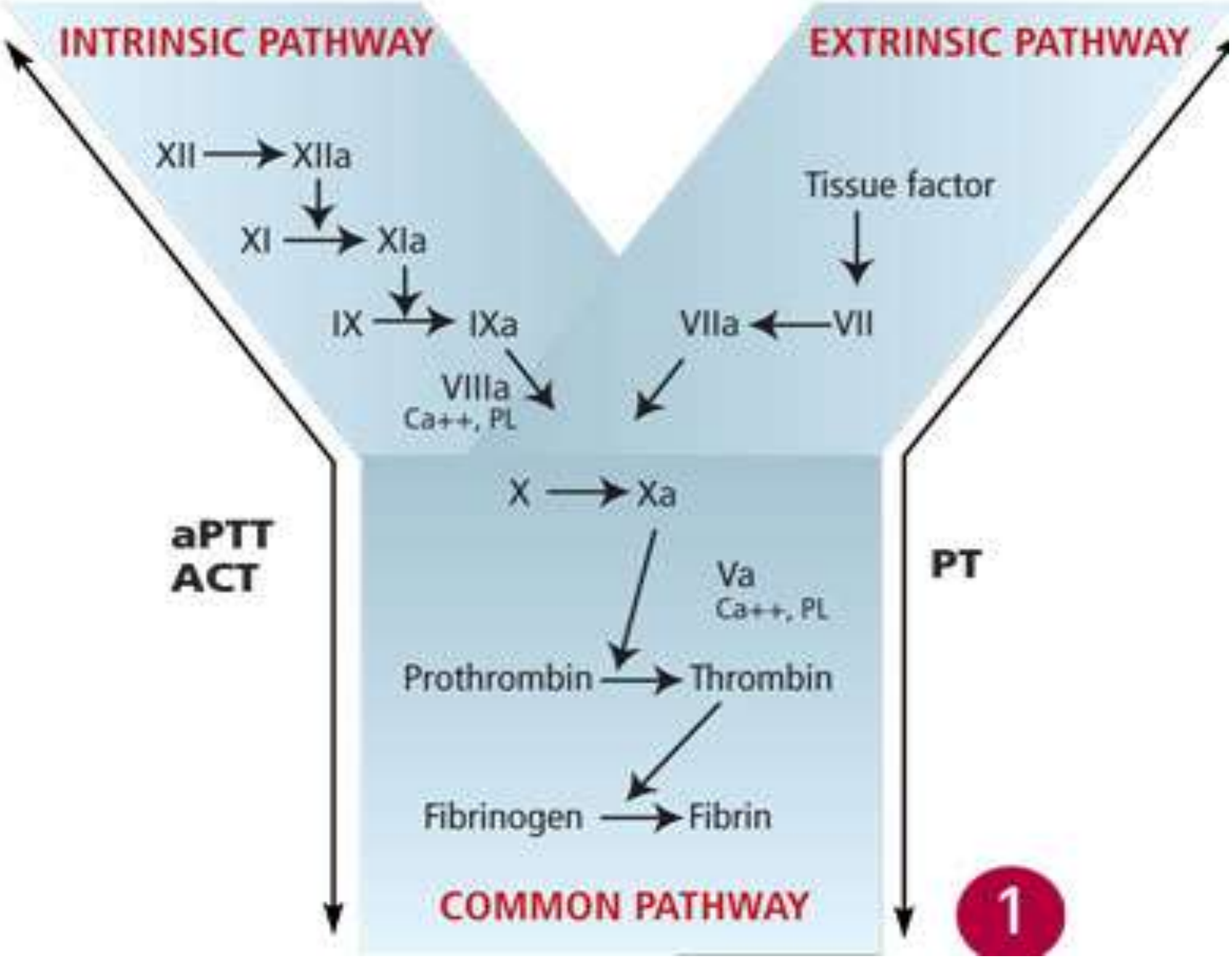
These mechanisms are set into play by:-

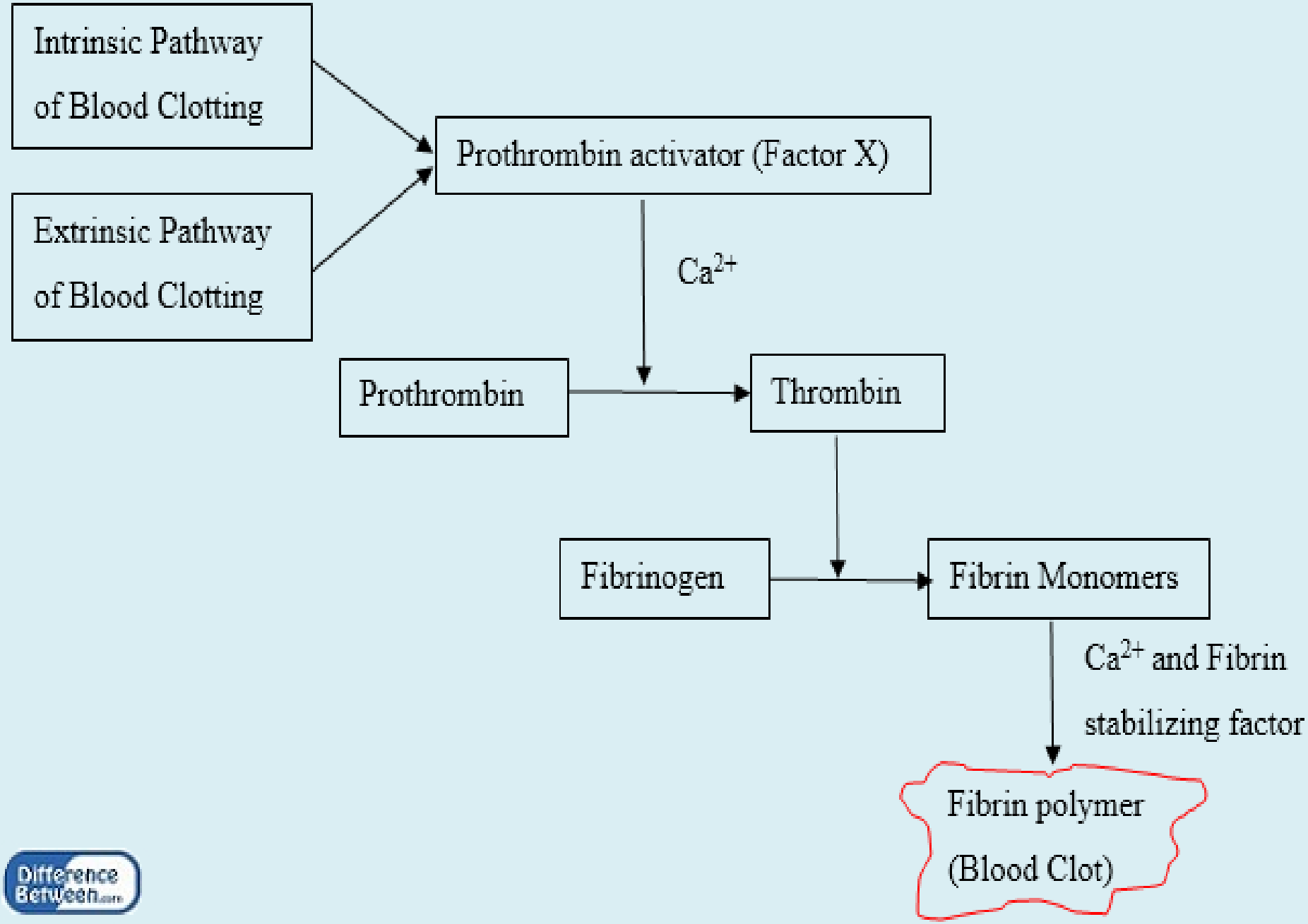
- Trauma to the vascular wall and the adjacent tissues
- Contact of the blood with damaged endothelial cells

- Prothrombin activator is generally considered to be formed in these ways-
  - a) Extrinsic pathway for initiating blood clotting
  - b) Intrinsic pathway for initiating blood clotting

The three pathways that makeup the classical blood coagulation pathway




INTRINSIC PATHWAY
EXTRINSIC PATHWAY
XII → XIIa
XI → XIa
IX → IXa
VIIIa
Ca++, PL
Tissue factor
VIIa ← VII
X → Xa
Va
Ca++, PL
Prothrombin → Thrombin
Fibrinogen → Fibrin
aPTT
ACT
PT
COMMON PATHWAY
1


Intrinsic Pathway of Blood Clotting
Extrinsic Pathway of Blood Clotting
Prothrombin activator (Factor X)
Ca^{2+}
Prothrombin
Thrombin
Fibrinogen
Fibrin Monomers
Ca^{2+} and Fibrin stabilizing factor
Fibrin polymer (Blood Clot)

CLOTTING FACTORS

- **Factor I (fibrinogen)-**

  Fibrinogen is a soluble plasma protein (MW 330000) which is acted upon by thrombin to form insoluble fibrin clot.

- **Factor II (prothrombin)-**

  Inactive precursor of thrombin is formed in liver.

- **Factor III (tissue factor, tissue extract, thromboplastin)-**

  This converts prothrombin in the presence of factors V, VII, and Xa, Calcium, and phospholipid.

- **Factor IV (calcium)-**
- **Factor V (labile factor, thrombogene or proaccelerin)-**

  This factor is reqd. for the conversion of prothrombin to thrombin by tissue extract and plasma factors.
- **Factor VII (stable factor, autoprothrombin I)-**

  Factor VII is reqd. for the formation of prothrombin activator by tissue extract.
- **Factor VIII (antihemophilic globulin [AHG], antihemophilic factor)-**

Factor VIII is reqd. for the formation of prothrombin activator from blood constituents; it's consumed during clotting and hence absent from serum. In vivo the half life of factor VIII is 10-20h.

CONTD..

- **Factor IX (Christmas factor, plasma thromboplastin component, autoprothrombin II)-**

  It's needed for the formation of prothrombin activator from blood constituents..

- **Factor X (stuart-prower factor)-**

  Converted to factor Xa either by factors IXa & VIII or factor VII & tissue factor. Factor X can be activated by other proteases such as trypsin or Russel's viper venom.

- **Factor XI (plasma thromboplastin acntecedent-PTA)-**

  Also a $beta_2$ globulin present both in serum & alumina treated plasma. It's thermolabile. Unlike factor XII, its activity increases when stored frozen.

CONTD..

- **Factor XII (Hageman factor or contact factor)-**

  It takes part in the formation of prothrombin activator from blood constituents. It's present in both serum & plasma.

- **Factor XIII (fibrin stabilizing factor)-**

  This is plasma protein which causes polymerization of soluble fibrin to produce insoluble fibrin.

- **Fletcher factor-**

  Described by Hathway (1965). Deficiency resembles factor XII deficiency. It's a prekallikerin. Evidence indicating that prekallikerin is activated by limited proteolysis.

- **Fitzgerald factor-**

  Its heat stable. This appears to act after the activation of Hageman factor & Fletcher factor but before the activation of factor XI. It's necessary for conversion of factor XI by Kaolin activated factor XII. It's reqd. for normal fibrinolysis & kinin formation.

ROLE OF CALCIUM IONS IN BOTH PATHWAYS

- Except for first two steps in the intrinsic pathway Ca^{2+} ions are required for the promotion/acceleration of all the blood-clotting reactions
- In the living body Ca^{2+} ion concentration seldom falls low enough to significantly affect the kinetics

ACTIVATION OF PLASMINOGEN TO FORM PLASMIN: THEN LYSIS OF CLOT

- Injured tissues and vascular endothelium gradually, after the clot has stopped the bleeding, release powerful activator, t-PA (tissue plasminogen activator)
- t-PA converts plasminogen to plasmin, which in turn removes the remaining unnecessary blood clot.
- Infact, many small blood vessels in which blood flow has been blocked are reopened by this mechanism.