Chapter 19

Hemostasis
Hemostasis

- Hemostasis is the cessation of bleeding
  - stopping potentially fatal leaks
  - important in small blood vessels
  - not effective in hemorrhage – excessive bleeding from large blood vessels
  - initiated by platelets and/or tissue thromboplastin

- Three homeostatic mechanisms must be present to achieve hemostasis
  - vascular spasm
  - platelet plug formation
  - blood clotting (coagulation)

- Platelets play an important role in all three of these mechanisms!
Platelets – Major Role in Hemostasis

- Platelets (also called thrombocytes) are small fragments of another cell called a megakaryocyte.
  - 2-4 μm diameter; contain “granules”
  - Complex internal structure and open canalicular system
  - Amoeboid movement and phagocytosis
- Normal platelet count = 130,000 to 400,000 platelets/μL
Platelet Functions

- secrete vasoconstrictors that help reduce blood loss
- stick together to form **platelet plugs** to seal small breaks
- secrete **procoagulants** or clotting factors to promote clotting
- initiate formation of **clot-dissolving enzyme**
- chemically attract neutrophils and monocytes to sites of inflammation
- **phagocytize and destroy bacteria**
- secrete **growth factors** that stimulate mitosis to repair blood vessels
Platelet Production = Thrombopoiesis

- stem cells develop receptors for thrombopoietin // these cells become megakaryoblasts

- megakaryoblasts // repeatedly replicate DNA without dividing // forms gigantic cell called megakaryocyte with a multilobed nucleus // 100 μm in diameter, remains in bone marrow

- megakaryocytes
  - live in bone marrow adjacent to blood sinusoids
  - long tendrils of cytoplasm (proplatelets) protrude into the blood sinusoids – blood flow splits off fragments called platelets

- platelets circulate freely for 10 days // 40% are stored in spleen
Hemostasis

(a) Vascular spasm

(b) Platelet plug formation

(c) Coagulation

all stages involve platelet function
Hemostasis - Vascular Spasm

- vascular spasm = prompt constriction of a broken vessel // most immediate protection against blood loss // the first spasm is independent of platelets
  - Initiated by // smooth muscle injury response // pain receptors // some directly innervate blood vessels to constrict
- after initial response // platelets release serotonin (vasoconstrictor) to augment vascular spasm! // second vascular spasm event

- Overall effect:
  - prompt constriction of a broken vessel // pain receptors - short duration (minutes) // smooth muscle injury - longer duration
  - This then provides time for other two clotting events to develop
Hemostasis - Platelet Plug Formation

- Under normal conditions endothelium surface needs to be smooth to inhibit activation of platelets.

- However, after vessel damage mechanism to make inner lining of blood vessels “sticky” must be activated:
  - endothelium coated with prostacyclin // a platelet repellant
  - protects against spontaneous formation of blood clots
  - this mechanism must be reversed by thromboxane (makes surface endothelium sticky)
Hemostasis - Platelet Plug Formation

- Platelet plug formation
  - broken vessel exposes collagen (this becomes a trigger)
  - platelet pseudopods stick to damaged vessel and platelets pseudopods contract to draw walls of vessel together forming a platelet plug
  - platelets degranulate releasing a variety of substances
    - serotonin vasoconstrictor // ADP attracts and degranulates more platelets // thromboxane A₂, an eicosanoid, promotes platelet aggregation, degranulation and more vasoconstriction
  - positive feedback cycle is active until break in small vessel is sealed
Hemostasis - Coagulation

• Coagulation (clotting) = last and most effective defense against bleeding

• Conversion of plasma protein fibrinogen into insoluble fibrin threads to form framework of clot // conversion needs an enzyme = thrombin

• Multiple step sequence involving several clotting factors and enzymes

• procoagulants (clotting factors) // produced by the liver // factors normally present in plasma
  
  – activate one factor and it will activate the next to form a reaction cascade
Hemostasis - Coagulation

- Coagulation can be initiated by activating two different pathways
  - extrinsic pathway // factors released by damaged tissues begin cascade // 15 sec
  - intrinsic pathway // factors found in blood begin cascade (platelet degranulation) // 3 to 6 minutes
Coagulation Pathways

- **extrinsic pathway**
  - initiated by release of tissue thromboplastin (factor III) from damaged tissue
  - cascade to factor VII, V and X (fewer steps)
  - Clot forms in 15 seconds

- **intrinsic pathway**
  - initiated by platelets releasing **Hageman factor** (factor XII)
  - cascade to factor XI to IX to VIII to X
  - Clot forms in 3 to 6 minutes

- **calcium** required for both pathways
1 Platelet adhesion

2 Platelet release reaction

3 Platelet aggregation
(b) Intermediate stage
(c) Late stage showing red blood cells trapped in fibrin threads
SEM of Blood Clot
Enzyme Amplification in Clotting

Rapid clotting - each activated cofactor activates many more molecules in the next step of the sequence / positive feedback

Reaction cascade (time)

Factors involved:
- Factor XII
- Factor XI
- Factor IX
- Factor VIII
- Factor X
- Prothrombin activator
- Thrombin
- Fibrin
Key Step in Completion of Coagulation

- activation of factor X leads to production of prothrombin activator, key step!
- prothrombin activator converts prothrombin to thrombin
- thrombin converts fibrinogen into fibrin
- positive feedback - thrombin speeds up formation of prothrombin activator
Fate of Blood Clots -1

- **Clot retraction** occurs within 30 minutes

- **Platelet-derived growth factor** secreted by platelets and endothelial cells
  - mitotic stimulant // stimulates fibroblasts and smooth muscle to multiply // together they help to repair damaged vessel
• **Fibrinolysis** // enzyme that breaks apart the blood clot

  – factor XII speeds up formation of **kallikrein** enzyme (also initiates clot formation!!!)

  – kallikrein converts **plasminogen** into **plasmin**

  – Plasmin = a fibrin-dissolving enzyme that breaks up the clot
Blood Clot Dissolution

- This is also a positive feedback event!
Factors to Prevent Inappropriate Clotting

- platelet repulsion // platelets do not adhere to prostacyclin // this coats inside of endothelium // note thromboxane is an antagonist to prostacyclin

- thrombin dilution // by rapidly flowing blood // heart slowing in shock can result in clot formation

- natural anticoagulants
  - heparin (from basophils and mast cells) interferes with formation of prothrombin activator
  - antithrombin (from liver) deactivates thrombin before it can act on fibrinogen
Terminology

- **thrombosis** - abnormal clotting in unbroken vessel
- **thrombus** = clot // most likely to occur in leg veins of inactive people
- **pulmonary embolism** - clot may break free, travel from veins to lungs
- **embolus** – anything that can travel in the blood and block blood vessels
- **infarction** (tissue death) may occur if clot blocks blood supply to an organ (MI or stroke) // 650,000 Americans die annually of thromboembolism – traveling blood clots
- **Thrombocytosis** // increase number of platelets
- **Thrombocytopenia** // decrease number of platelets
Clinical Management of Clotting

• goal to prevent formation of clots or dissolve existing clots

• preventing clots
  – Vitamin K is required for formation of clotting factors // coumarin (Coumadin) is a vitamin K antagonist
  – aspirin suppresses thromboxane $A_2$
  – other anticoagulants discovered in animal research
    • medicinal leeches used since 1884 (hirudin)
    • snake venom from vipers (Arvin)
Clinical Management of Clotting

- dissolving clots that have already formed
  - streptokinase – enzyme make by streptococci bacteria
    - used to dissolve clots in coronary vessels
    - digests almost any protein
  - tissue plasminogen activator (TPA) – works faster, is more specific, and now made by transgenic bacteria
  - hementin – produced by giant Amazon leech
Clotting Disorders - Hemophilia

- deficiency of any clotting factor can shut down the coagulation cascade

- **hemophilia** – family of hereditary diseases characterized by deficiencies of one factor or another
  - sex-linked recessive (on X chromosome)
    - **hemophilia A** missing factor VIII (83% of cases)
    - **hemophilia B** missing factor IX (15% of cases)

- physical exertion causes bleeding and excruciating pain
  - transfusion of plasma or purified clotting factors
  - factor VIII produced by transgenic bacteria

- **hematomas** – masses of clotted blood in the tissues
Disseminated Intravascular Coagulation

- Involves both excessive bleeding and clotting

- First: Excessive clotting in circulation // Thrombi and infarcts occur.

- Secondly: Clotting factors are reduced to a dangerous level. // Widespread, uncontrollable hemorrhage results.

- Very poor prognosis, with high fatality rate

- Complication of many primary problems
  - Obstetrical complications, such as abruptio placentae
  - Infections
  - Carcinomas
  - Major trauma
Disseminated Intravascular Coagulation

A primary condition such as septicemia, obstetric complication, severe burns, or trauma causes

EXTENSIVE ENDOTHELIAL DAMAGE or RELEASE OF TISSUE THROMBOPLASTIN

INITIATE THE CLOTTING PROCESS

Many thrombi form
Platelets collect

Throughout the microcirculation
Use up clotting factors

DECREASED SERUM FIBRINOGEN

THROMBOCYTOPENIA

ISCHEMIA AND MULTIPLE INFARCTIONS

EXCESSIVE BLEEDING AND HEMORRHAGE

FIBRINOLYSIS

Activate plasmin

stimulated

ORGAN FAILURE

Normal Physiologic Condition

[Diagram showing the balance of Coagulation + Fibrinolysis]

- Homeostasis
- Coagulation + Fibrinolysis
  - Vascular Injury
  - Tissue Injury
  - Coagulation Cascade
    - Prothrombin
    - Fibrinogen
    - Fibrin ... Stable Clot Formation
  - FDPs Excreted
    - Fibrin Degradation Products (FDPs)
    - Fibrinogen + Fibrin Clot Breakdown
    - Plasmin
    - Plasminogen
Abnormal Pathway - DIC

Diagram: Flowchart showing the pathway of DIC (Disseminated Intravascular Coagulation).

- Massive tissue destruction → Release of tissue factor
- Sepsis → Release of tissue factor
- Endothelial injury → Platelet aggregation

Widespread microvascular thrombosis leads to:

- Activation of plasmin
- Microangiopathic hemolytic anemia
- Vascular occlusion

Fibrinolysis:
- Fibrin split products → Inhibition of thrombin, platelet aggregation, and fibrin polymerization

Proteolysis of clotting factors:
- Ischemic tissue damage

Consumption of clotting factors and platelets:
- Bleeding
DIC is associated with three conditions: fever, hypotension (i.e. shock), and intravascular coagulation. We can usually control two out of the three conditions, however. Intravascular coagulation is the greatest risk to life.
DIC – Clinical Presentation
DIC – Clinical Presentation
DIC - Spleen

Fig. 2 — This pictures show the spleen after it has been removed due to splenic infarction. The consistency of the spleen normally is fairly spongy while in splenic infarction the spleen feels very “wooden”.

Area of splenic infarction at the pedicle of the spleen and greatly enlarged spleen.