Introduction to Haematology, Platelets and Haemostasis (W11)

Introduction to Haematology (L1)

Blood: Hippocrates of Kos

- Greek physician (460 – c370BC)
- **Father of Western medicine**
- **Rejected idea of disease as affliction of gods**
- Body composed of 4 humours ('humourism')
  - Blood: sanguine
  - Phlegm: phlegmatic
  - Yellow bile: choleric
  - Black bile: melancholic
- Imbalance of the humours: the cause of disease

The circulation

- William Harvey (1578 – 1657): English physician
- Revolutionised concepts of disease causation
- A severe blow to the humoural theory
- **1613: Described circulation of the blood**
- **Functioning** & **pumping of the heart**
- Observed cardiac pathology:
  - Ventricular rupture

Blood Transfusion

- Jean-Baptiste Denis **1667**; Physician to Louis XIV
- Gave the **1st reported blood transfusion** to Antoine Mauroy, a madman
- **Direct transfusion of calf (baby cow) blood**
- Beginnings of blood transfusion

First successful blood transfusion

- James Blundell (1790-1878): London
- "the patient might very probably have been saved by transfusion"
- **1818**: **1st human-to-human transfusion**
- 10 transfusions given over 5-years
- 5 were successful
- Determined **only human blood should be used**; transfusions **only to replace blood** (not to treat madness or change character)

Stains: Paul Ehrlich (1854-1915)

- "**Father of haematology**"
- 1878 Doctoral thesis: compared textile dyes and cell staining (acidic and basic dyes)
- **Haematoxylin: 1865**
- Able to **identify leukaemia and subtypes**

Blood Groups: Landsteiner

- Karl Landsteiner (1868 -1943)
- **1901: discovered ABO blood groups**
- Mixed **red cells** and **plasma agglutination** and **saw three patterns**
  - Identification of **blood groups A, B and C (O)**
- 1930: won the Nobel Prize
- "**Father of transfusion medicine**"
- Safe blood transfusions

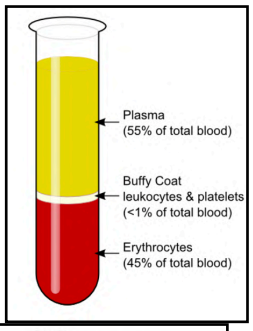
Modern History of Haematology

- Cell immunology 1980s
  - Monoclonal antibodies, Immunocytochemistry, Flow cytometry
- Cell genetics 1990s
  - Cytogenetics; karyotype; FISH,
- Molecular genetics: 2000s,

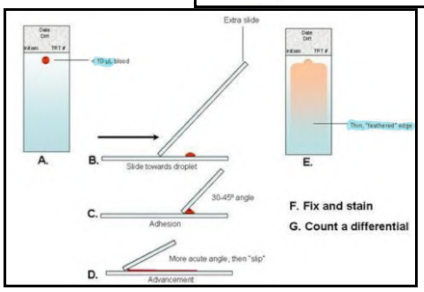
- DNA, RNA, Massively parallel sequencing

What is haematology?
- **Blood** and **bone marrow** assessment
- **Coagulation** and **bleeding** disorders
- **Blood transfusion**
- Diagnosis of:
  - **Inherited** conditions
  - **Acquired**: reactive; secondary; malignant disorders
- **Management** of patients with blood disorders
- Provide **advice to other clinicians on blood disorders**

Blood: plasma and cells
- Suspension of **cells in water, proteins and electrolytes** (**average vol = 5 L**)
- **Plasma** contains **coagulation proteins** (55%)
- Cellular components:
  - **Red blood cells** (erythrocytes): Hb (45%)
  - Buffy coat (<1%):
    - **White blood cells** (leucocytes): neutrophils, lymphocytes, monocytes, eosinophils, basophils
    - **Platelets**

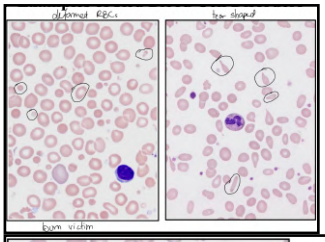


The Blood Film
- 10µL of blood is placed on a slide
- Extra slide is used to spread the blood to create a gradient
  - Thin, feathered edge
- Fix cells and stain, count a differential



Review of the blood film
- RBCs: do they look normal?
- Leukocytes: number and morphology
- Platelets: number and morphology

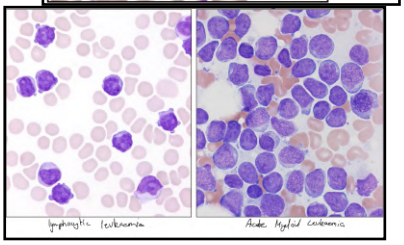
Examples of RBC disorders:
- Deformed RBCs (burn victim)
- Tear shaped RBCs



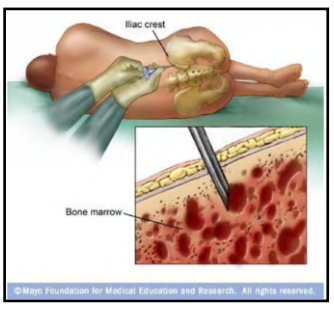
Respones to Infections
- Increase in WBC
- More granules
- Top photo: bacterial infection
- Bottom photo: activated to viral infection



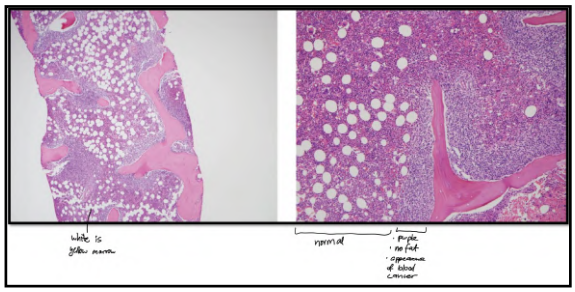
Blood cancers: leukaemia
- Too many WBCs
- Lymphocytic leukaemia (left) vs acute myeloid leukaemia (right)



Bone marrow examination
- When born - all bones have marrow; Grown - axial skeleton only
- Performed to investigate blood abnormalities
- Aspiration



- **Trephine biopsy (hole saw used in surgery to remove a circle of tissue or bone)**
- Assess cell number, type and morphology
- Make a diagnosis
- May need additional tests



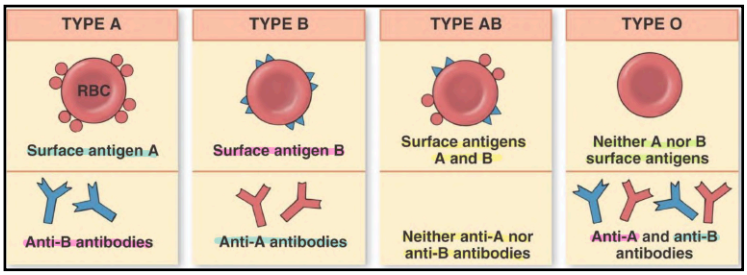
An abnormal bone marrow trephine
- Lymphoma involving bone marrow (Stage 4)
- White is yellow marrow (fat)
- Blood cancer: purple, no fat

Blood clotting: coagulation
- 3 components (**Virchow's Triad**)
  1. **Clotting factors** - **circulating** proteins
  2. **Platelets** - derived from **megakaryocytes** in BM
  3. **Endothelium** - **blood vessel lining**
- Clotting disorders with abnormalities
- Laboratory tests for clotting

Bleeding and clotting disorders
- **Unexplained bruising**
- **Unexplained bleeding**
- **Unexplained clotting**
- **Inherited**: haemophilia A
- **Acquired**: medications; liver disease
- Diagnosis
- Management

Blood transfusion: donor blood is collected, prepared and transfused to a patient
- Blood donation:
  1. **Collection** of blood
  2. **Testing** of donated blood
  3. **Blood group**
  4. Screening for **infectious agents**
  5. **Preparing** blood for transfusion
- Blood products
  1. **Packed red blood cells**
  2. **Platelets**
  3. Fresh frozen **plasma**
  4. Cryoprecipitate
  5. **Immunoglobulins**
  6. **Clotting factors**

Blood grouping and products
- ABO blood groups: A, B, AB and O
- Type A: **surface antigen A**
  - Anti-B antibodies
- Type B: **surface antigen B**
  - Anti-A antibodies
- Type AB: **Surface antigens A and B**
  - Neither Anti-A and Anti-B antibodies
- Type O: **neither A nor B surface antigens**
  - Anti-A and Anti-B antibodies



Haematologist Role in Blood Transfusion

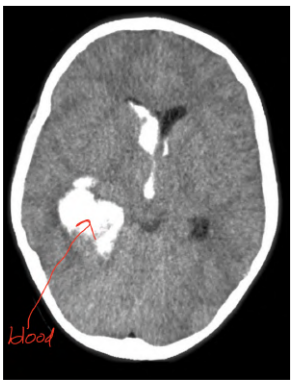
- **Advice** on indications for blood transfusion
- Which product?
- How much?
- The blood is **incompatible**
- **Management** of uncontrolled haemorrhage
- No suitable blood is available
- Management of transfusion reactions
- **Liaising** with **clinicians, surgeons, anaesthetists, obstetricians in emergencies**

What does a haematologist do?

- **Interpret blood test results** (“detective”)
- **Interpret morphology** (microscopy)
- Make diagnoses
- Provide advice to clinicians on diagnosis, management and blood transfusion
- Laboratory management & quality
- Most diseases have blood abnormalities
- Care for patients with blood diseases

Make a diagnosis: acute leukaemia

- Blood count
- Blood film
- Bone marrow
- Cell phenotype
- Genetics
- Mutations
- Rx: chemotherapy; transplantation



Make a diagnosis: haemorrhage

- Patient presents with severe headache
- History of previous cardiac surgery
- On **Warfarin** treatment (results in haemorrhage)
- **Prothrombin Time (PT)**: 45 (11-14s)
- **APPT**: 33 (23-35s)
- **Thrombin Time**: 12 (10-13s)

The language of haematology

- Prefix haem- or haemo- or haemato- refer to blood
- From Greek (haimo-) & Latin (**haemo**) for **blood**
- Some examples:
  - **Haematology**: the **study** of **blood and blood diseases**
  - **Haemoglobin**: iron-containing protein in red blood cells that transport oxygen
  - **Haematocrit**: part of **blood that is cellular**
  - **Haemopoiesis**: **process of making blood cells**
    - “-poiesis” = process of making

The practice of haematology

- **It is capable of throwing light upon the diagnosis in a wide range of pathological conditions**
- It is difficult to single out any disease in which it may not be of some utility
- Haematology findings may be:
  - Diagnostic (pathognomonic)
  - Essential
  - Supportive
  - Exclude a diagnosis

Normal Haemostasis and Testing (L2)

Clot formation

- **Primary haemostasis** - **platelet** mediated
- **Secondary haemostasis** - **coagulation factors**
  - Coagulation cascade
  - Inhibitors
- Fibrinolysis

Testing

- Screening - platelet dysfunction, coagulation profile - APTT/PT
- Specialised tests - platelet function tests, factor testing

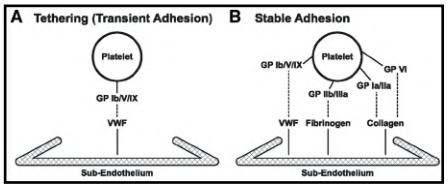
Normal Haemostasis - Overview

- Requires:
  - **Amplification/cascade system** —> small changes make big clots
  - **Control/brakes** - **inhibitors** and **fibrinolysis**, restrict to site of injury
- Components
  - **Vessel wall initiation**
  - **Primary haemostasis** - **platelet plug**
    - Adhesion
    - Activation
    - Aggregation
  - **Secondary haemostasis** - **coagulation factors**
    - **Intrinsic** and **extrinsic** pathway
    - Initiation-amplification-propagation
  - **Fibrinolysis**
- Result
  - Platelet-fibrin thrombus

Primary Haemostasis

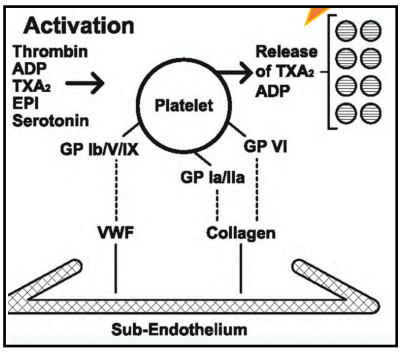
- Primary haemostasis - **platelets**
- **Megakaryocytes** - multinucleate platelet precursors
  - Growth and maturation stimulated by Thrombopoietin (TPO) from liver

Primary Haemostasis - Adhesion



- **Platelet adhesion** (1)
  - **Vessel wall damage**
  - Exposure of **subendothelium** to blood - **platelets bind to subendothelial collagen**
  - Collagen adheres and **activate platelets**
  - Platelets adhere to subendotheium by:
    - **Von willebrand factor** and **GP (glycoprotein) Ib/V/IX receptor (on platelets)**
      - **VWF binds** to **exposed collagen** and recruits platelets
    - **GP VI receptor (on platelets)** —> binds to **collagen**
    - **GP Ia/IIa** (integrin alpga-2-beta-1) —> binds to **collagen**
    - **GP IIb/IIIa** —> binds to **fibrinogen**

Primary Haemostasis - Activation



- **Activation**
  - **Thromboxane A2 (TXA2) production** – **activates new platelets**, **increases aggregation**
  - Granule release
  - **Amplifies** thrombus formation
- Granules:
  - **Alpha granules** (proteins, clotting factors) - P-selectin, **fibrinogen**, fibronectin, **factor V, factor VIII, platelet factor IV**, platelet-derived growth factor and TGF-α