Intro :- Blood is a special form of C.T → Blood cells ⅓ plasma as ECM.
It's color depends on amount of O2 → more O2 : brighter red.
PH = 7.4.

Functions :- transport nutrients, gases, waste, hormones, immune cells
Regulate body temperature , Maintain osmotic pressure.

Amount :- 5-6 L, propelled thro rythemic contractions of the heart unidirectionally.

Blood Content : We centrifuge a blood sample along with anticoagulants → 45% live content (WBC, RBC, Platelets), 55% Non living : Plasma
↳ Cells : RBCs 44% + 1% buffy coat [WBCs, Platelets]
Plasma: Pale yellow, 90% water + electrolytes, nutrients, proteins, waste, gases ⅓ hormones.

Values :- RBCs : 3.5- 5.5 million /mm³ , Platelets : 150000 - 400 000 /mm³ , WBCs : 4500 - 11000 /mm³.

* plasma has many proteins mainly to create osmotic pressure and keep fluid inside capillaries
1. Albumin :- most abundant, liver, maintain O.P, transport F.A and steroid hormones.
2. Fibrinogen :- largest, liver, clotting.
3. α, β globulins :- liver, transport iron, fat soluble vitamins ⅓ lipids.
4. γ globulins :- antibodies → Plasma cells.

Blood Smear :- drop of blood on slide, spread with another slide, let it dry, add stain and fixative then cover it.
↳ staining :- neutral stain / polychromatic stain (leishman) dissolved in methyl alcohol as a fixative.
leishman stain contains eosin dye and methylene blue.

Red Blood Cells :-

small biconcave discs → 20% - 30% more S.A relative to spherical shape / V → facilitates gas exchange.
→ shape + membrane fluidity (50% proteins) → bend in small diameters.

Transport O2 (lungs to peripheral tissue) , CO2.
Cytoplasm full of hemoglobin (basophilic) → appears eosinophilic w/ pale central region. [normochromic RBC]
No nuclei nor organelles. 6-9 in diameter.
Survive for 120 days, macrophages remove dead ones in spleen, liver and B.M.
Cellular features : outer plasma membrane enclosing hemoglobin
some enzyme for gas exchange and membrane integrity.
submembranous meshwork of proteins (spectrin and Ankyrin) maintains cell shape and provide elasticity.
Appearance under microscope :- LM → leishman stain, rounded eosinophilic w/ central pallor forming ⅓rd of it's size
EM → Electron dense and homogenous.
Rouleaux formation :- RBCs stack together loosely like a pile of coins. Occurs in slow circulation due to surface tension of their biconcave surface
Hereditary spherocytosis :- mutation in genes related to membrane protein so the RBC won't hold it's shape anymore
Abnormalities in erythrocytes :-
1. Abnormal size :- microcyte < 6 um, macrocyte >9 um, Anisocyte : different sizes.
2. Abnormal staining :- hypochromia : less stain → less hemoglobin, usually accompanies microcytosis.
3. Abnormal shape :- spherocyte, sickle cell (abnormal hemoglobin), ovalocyte / elliptocyte, dacrocyte
↳ Erythropoiesis in B.M is stimulated by erythropoietin from kidneys
during blood loss, hypoxemia is sensed by kidneys → ↑erythropoietin → ↑ erythropoiesis.
consequently :- People living in higher altitudes
Athletes w/ high O2 demand] ↑ RBCs count.
Anemia :- decrease in total RBCs count and / or hemoglobin
Polycythemia : increase in total RBCs count.
ABO blood typing :- according to glycoproteins on their surface.
↳ glycophorin A → integral membrane protein.
→ glycosylated extracellular domain → antigenic site → basis for ABO typing system.

White blood Cells :-

Originate in bone marrow, travel in the blood and function in loose CT mainly to protect the body from damage by pathogens ⅓ tumor cells.

Leukocytes are nucleated cells that are devided according to presence of specific granules into :-

1. Granulocytes: Neutrophils, Eosinophils, Basophils
2. Agranulocytes: Monocytes, Lymphocytes.

→ Granulocytes:

single multi-lobulated nucleus, phagocytic ability

cytoplasmic granules: grainy cytoplasm, enzymes / chemicals.

↳ non specific / primary / azurophilic / lysosomes ⅓ specific / secondary.

General features: spherical in blood ⅓ irregular in C.T.

highly motile → amoeboid movement.

leave blood stream between endothelial cells thro diapedesis.

types: eosinophils : large red granules, Basophils : large blue granules, Neutrophils : small baby pink granules. [no affinity to either type + dye]

1. Neutrophils : polymorphs, Pus cells, myelocytes, microphages, cells + A.I.

most common leukocyte, 2-5 lobes in nucleus connected by threads.

light pink cytoplasm = takes up pink ⅓ blue stain equally.

short lived, 6-8 h in blood ⅓ 1-4 d in C.T

responds to bacterial acute infections (pyrogenic infections)

↳ 1st cell to leave the blood in large numbers to site of inflammation → abundant, motile, neutrophil chemotactins released first.

↳ ability to survive in anaerobic environments → kill bacteria and clean up debris in necrotic tissue.

↳ Dead PMNs + bacteria + lysed ECM → Pus (yellow viscous fluid) → Pyogenic.

pyrogenic → bacterial infection causing a fever.

Inactive chromosome X (barr body) appears as a drumstick appendage on one of the lobes. (3%).

Specific G.	Primary G.
- lysozyme.	- Myeloperoxidase.
- phagocytic (bactericidal)	- Acid hydrolase.
- lactoferrin (bacteriostatic).	- Defensins
- collagenase.	

2. Basophils

Rarest leukocyte, bi-lobed / S-shaped nucleus obscured by large dark blue granules.

Granules contain histamine, heparin ⅔ eosinophilic chemotactin → Allergies & parasitic Infections.

Basophils ⅓ most cells have receptors for IgE ⅔ secrete their contents in response to certain antigens and allergens.

↳ 1st exposure to allergen → produced IgE bind to mast cells ⅔ basophils → 2nd exposure the allergen → binds to IgE on cells → Degranulation → Inflammatory mediators → different forms of type I hypersensitivity

* sometimes 2nd exposure to strong antigens → strong systemic response → drop in BP → Anaphylaxis. [degranulation in more than one site]

Basophils are rather abundant in allergic dermatitis (15%) and skin allograft rejection.

3. Eosinophils :-

Bilobed nucleus w/ large red granules.

Fights Allergic reactions and parasitic Infections

Phagocytic affinity for antigen - antibody complex.

Specific Granules :- Oval w/ flat crystalloid cores.

↳ externum : pale, histaminase and sulfatase.

Internum : dark, basic protein to kill parasites

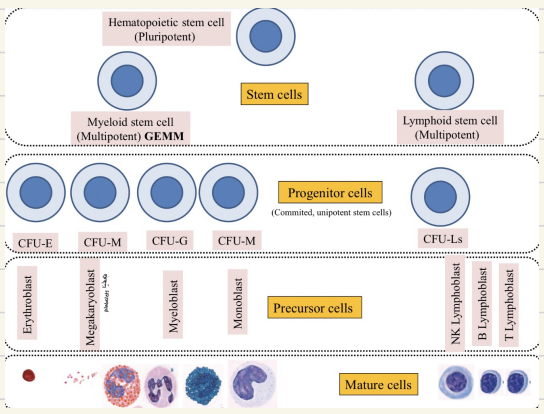
* Hematopoiesis :-

- Blood cells have short life spans, thats why they need to be constantly replaced from precusors thro hematopoiesis.
- Hemotopoiesis takes place: early embryo : yolk sac, mesoderm.
  2nd trimester : developing liver and spleen.
  3rd trimester and so on : BM.

↳ Bone marrow:

Red bone marrow is a highly cellular structure located in medullary cavities of bones.
consists of : hemopoietic stem cells, surrounded by macrophages, sinusoidal capillaries and supported by reticular tissue

* as we age, red BM is constricted to axial skeleton [skull, rib cage, vertebrae, pelvic bone].
  remaining bones accumilate fat cells and BM turns yellow.
  ↳ in conditions such as hypoxia and bleeding, yellow BM can convert back to red BM.
* Extramedullary hematopoiesis : hematopoiesis in organs other than BM. such as infetal developement, normal immune response, pathologies.
* Sinusoidal capillaries → discontinuous basement membrane & intercellular gaps between endothelial cells thro which newly formed blood cells leave to enter the circulation.
  → Macrophages extend their processes thro these gaps to engulf old blood cells and reprocesses heme bound iron for delivery to maturing blood cells.
* Reticular tissue forms the stroma of Bone Marrow.



- All blood cells arise from a single type of stem cells → pluripotent stem cells.
  they proliferate into 2 cell lineages → myeloid and lymphoid.
- Stem cells are capable of asymmetrical devision and self renewal.
  ↳ each time a SC devides it gives 2 cells, one turns into a mature blood cell and the other adds to original population this way SC maintain original population.
  ↳ progenitor cells are capable of self renewal but less capacity than stem cells.
  ↳ precursor cells produce only mature blood cells. and no precursor cells are left after devision.
- Rate of cell devision : stem cells → slow Progenitor and precusor → rapid.
- Morphology : SC & progenitor cells : large lymphocytes, precusor cells gradually resemble the morphology of mature cells.
- Progenitor cells are called CFUs : give rise to colonies of one cell type when cultured or injected in spleen.
  cell type in the colony is the cell type the CFU is committed to produce [CFU-E → colony of erythrocytes]

→ lymphopoiesis occurs in BM and in other lymph organs where precusor cells migrate to mature and differentiate [pre-T cell → thymus].

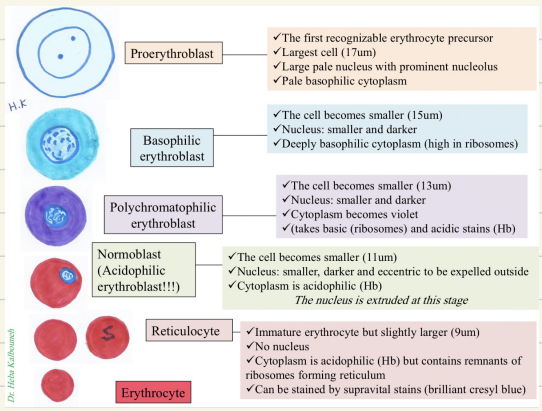
* Erythropoiesis :-

needs 1 week, controlled by EPO (from kidneys) and availability of iron, folate, vitB12 and protein precusors.
Stages of differentiation are characterized by :-

1. decreasing cell size.
2. Nuclear maturation : shrinks and extruded from cell.
3. cytoplasmic maturation : progressive loss of organelles, ribosomes at early stages → basophilia, accumilation of Hb → eosinophilia.

↳ Reticulocytes :

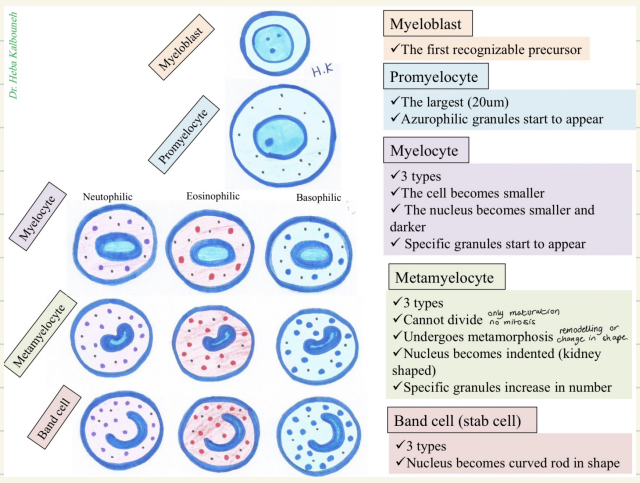
- immature RBCs (last stage), cell lost its nucleus but still capable of producing hemoglobin during the 1-2 days they circulate before becoming mature RBCs.
  Normally, 1% of all RBCs in blood are reticulocytes.
  ↑ reticulocytes → blood loss (hemorrhage).
- Supra vital dye (brilliant crystal blue) to stain precipitation of reticulum.



* Granulopoiesis

needs 2 weeks, gives neutrophils, eosinophils and basophils.
Stages of differentiation are characterized by :

1. cytoplasmic changes :- 1st formation of azurophilic granules (similar in all), 2nd formation of specific granules (different for each type).
2. condensation, indentation and segmentation of nucleus.



- If we culture myeloblast, promyelocyte and myelocyte → mitotic cells → we will get many granulocytes culturing a metamyelocyte will give one mature cell at the end.
- Band cells need further nuclear maturation (segmentation), sometimes this happens in blood stream.
  * neutrophilic band cells must not exceed 5%, if it's more it's called shift to the left implying that BM is being stimulated to release and produce more neutrophils indicating bacterial infection.

* Neutrophils (developing or mature) exist in 4 pools :-

1. granulopoietic compartment in BM
2. Stored as mature cells in BM till release.
3. circulating (interchangeable)
4. Marginating cells :- neutrophils adhere loosely and accumulate transiently on endothelial surface of small venules. can last for hours and won't necessarily end in exit of cells.

* At sites of injury neutrophils enter C.T by migrating thro intercellular junctions between endothelial cells of post capillary venules by diapedesis. thus inflammed C.T is the 5th compartment where they reside for a few days then die by apoptosis.
* Changes in neutrophil numbers in blood must be evaluated by taking all compartments into consideration
  - ↳ neutrophilia ≠ ↑ granulopoiesis.
  - intense muscular activity , administration of epi ⟶ moves neutrophils from marginating compartment to circulating compartment
  - However, glucocorticoids (cortisone) increase granulopoiesis (↑ mitotic activity) resulting in neutrophilia.

* White blood cells abnormalities.

- ↑ neutrophils in peripheral blood ⟶ bacterial infection.
  ↑ eosinophils // // ⟶ Parasitic infection and some allergies.
- Leukemia : malignant proliferation of WBC precursors in BM.
  resulting in large no. of WBCs and their precursors which spill out to the blood.
  classified according to cell lineage involved (granulocytic , monocytic , lymphocytic).
  Bone marrow biopsy or aspirate to diagnose leukemia and aplastic anemia.
  Bone marrow transplantation : hemopoietic SCs are taken from a donor and infused into the same or another person.

NOTE: 5million RBCs / mm^3 and 4500 – 11000 WBCs / mm^3.
but in BM erythroid : myeloid is 1 : 3.
⟶ Remember life spans! WBCs have short life spans (days) ⟶ we need more progenitor cells to produce enough of them.

* Hemopoietic Growth factors (CSF or cytokines)

- Glycoproteins that stimulate proliferation of progenitor and precursor cells and promote differentiation of different lineages.
  ↳ erythropoiten , Thrombopoiten , CSF-G.
- Cloning of genes of imp. CSF has advanced the study of blood formation and permitted the production of clinically useful factors for patients w/ hemopoietic disorders.

* Other forms of hemopoiesis :-