Session 11
Disorders of Red cells

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Red cells

- Biconcave cells
- carry oxygen to the peripheral tissues
- red colour- haemoglobin
- Haemoglobin + oxygen-more efficient delivery of oxygen to peripheral tissues.
- No nucleus- maximize the space to carry haemoglobin and oxygen
- Biconcave disc; it has a central area of pallor
Normal red cell morphology
• Red cell production regulation- erythropoietin (a hormone)
• Erythropoietin- produced in kidney in response to hypoxia.
• Red cells are produced in the bone marrow.
• The bone marrow is confined to the skull, sternum and the pelvic bones in adult life.
• Average red cells survive for 120 days.
• At the end of its lifespan the cells are lysed in the reticuloendothelial system which comprises of the spleen, liver and lymph nodes.
Haemoglobin

• A metallo-protein (a protein that contains a bound metal ion as part of its structure)
• Metal – iron, ferrous state (Fe$^{2+}$): red colour
• 4 globins
• 2 types of globins
  alpha (α)
  beta (β)
Haemoglobin molecule has 2 alpha globin chains and 2 beta globin chains.
Each globin molecule has a small pocket in which it holds onto a molecule of “haem”.
Haem molecule binds to globin and keeps the ferrous (Fe^{2+}) ion in a reduced state.
• Maintain iron in the ferrous form is important as the ferric form or the oxidized form does not carry oxygen.
Functions of Hemoglobin

• Delivery of O2 from lungs to the tissues
• Transport of CO2 & protons from tissues to lungs for excretion
• Heme is present in Myoglobin, Cytochromes, Peroxidase, Catalase, Tryptophan pyrrolase & Nitric oxide synthase
• In cytochromes, oxidation & reduction of iron is essential for their biological function in ETC
Anaemia

• Reduction in oxygen carrying capacity of the blood.
• Reduction in the circulating red cell mass in the body.
• The age and the sex of the patient are important in determining anaemia.
• Anaemia may be classified by
  ▪ Morphology
  ▪ Aetiology
Morphological classification

- normal in size and haemoglobin content - normocytic and normochromic.
- Smaller RBC and reduced haemoglobin - hypochromic and microcytic.
- Cells are larger al red cells – macrocytic.
Aetiological classification

• 3 categories.
  1. Blood loss
  2. Increased red cell destruction
  3. Impaired red cell production
1. Blood loss

- Blood loss - acute with large volumes lost as in a victim of trauma.
- In such a situation loss of blood volume and shock is more of a concern than the effects of anaemia.
- Small amounts of blood lost over a long period of time as in hook worm infestations deplete iron stores and give rise to mainly a deficiency type of anaemia.
- The patient will rarely complain in anaemia caused by chronic blood loss as the body readily adapts to low haemoglobin over time.
B. Increased red cell destruction

- Any reduction in the life span is classified as increased red cell destruction or haemolytic anaemia.
- Red cells destruction results the haemoglobin degrade into bilirubin.
- Bilirubin is a yellow pigment which stains mucosal surfaces.
- This yellowish discolouration is called jaundice.
• Causes of red cell destruction- genetically inherited which gives rise to inherent red cell defects.
• They can be broadly classified as follows:

![Classification of haemolytic anaemia diagram]

- Inherited
  - Intra corpuscular or intrinsic defects
- Acquired
  - Extra corpuscular or extrinsic defects
• Classification- according to the site of red cell destruction.
• Within the vascular system- intra vascular haemolysis, release the haemoglobin pigment directly into the plasma.
• Extravascular haemolysis- cells are destroyed in the spleen, liver and lymph nodes
• This is in fact an exaggeration of the normal removal of cells when they are aged.
• The bilirubin produced by red cell lysis is bound to albumin and transported to the liver for further degradation and excretion by bile.
Causes of Anemia

- Nutritional deficiencies
- Hemolytic disorders
- Blood loss
- Bone marrow (hypoproliferative)
- Infection
- Toxicity
- Hemopoietic stem cell damage (maturation disorder)
- Heredity or acquired defect
- Unknown
Morphological Classification of Anemias

- Morphological based on sizes and color of RBCs
  - Normochromic Normocytic
  - Hypochromic Microcytic
  - Normochromic Microcytic
  - Normochromic Macrocytic
Inherited or intra corpuscular defects

• These may be broadly classified as
1. Membrane defects
2. Enzyme defects
3. Haemoglobin defects
Membrane defects

• Membrane - Made up of a double phospholipid layer with a few proteins in between.
• Skeleton - made up of several integral proteins, called as the cytoskeleton.
• Cytoskeletal defects due to inherited deficiencies of certain proteins
• Most common - hereditary spherocytosis.
Diagram of a cross-section of the red cell membrane.
Hereditary Spherocytosis

- Inherited in an autosomal dominant- one third of the patients
- Autosomal recessive in a third of the patients
- The red cell produced in this condition are inherently spherical.
- Deficiency of production of membrane proteins such as spectrin and ankyrin.
• Spleen is the major site of red cell destruction
  * RBCs retained for long time in the splenic pulp as a result of decreased deformability (the ability of cells to change shape as they pass through narrow spaces)
  
  + Unfavorable environmental conditions in the splenic pulp (acid pH & decreased glucose)

  ↓

  Failure of the cation pump

  ↓

  Loss of water

  ↓

  Loss of RBCs discoid shape

  ↓

  Vicious circle
Spherocytes
• The spherical red cells are trapped within the micro circulation as they have more rigid cell membrane and therefore lyse.

• Lysis is mainly in the spleen as the microcirculation in the spleen tries to remove defective red cells.
Hereditary elliptocytosis

- Red cells are oval in shape.
HEREDITARY ELLIPTOCYTOSIS (HE)
Enzyme defects

• Red cells do not have a nucleus or mitochondria.
• They depend solely on glycolysis for energy, where they utilize glucose in the blood as fuel.
• To maintain haemoglobin and the iron in it in a reduced ferrous state, it has a pentose monophosphate pathway.
• The pentose monophosphate pathway produces vital NADPH that is necessary to combat oxidant induced injury and to maintain the integrity of the red cell membrane.
• In the absence of NADPH, the haemoglobin and red cell membrane will be damaged by oxidants.

• As the red cell does not have a nucleus or mitochondria, synthesis of new structures does not take place.

• If the red cell is irreversibly damaged, it undergoes lysis prematurely.
G6PD deficiency

• The pentose phosphate pathway contains several enzymes; one of which is Glucose 6 phosphate dehydrogenase (G6PD).

• Inherited as sex linked recessive. i.e. the males display the definitive characteristics with females being carriers.

• G6PD deficiency leads to increased susceptibility of red cells to oxidant stress which will leads to
  – denaturing of the red cell membrane
  – oxidization of the ferrous atom of the haem molecule to ferric which does not bind oxygen readily
• Hb loses its main purpose as an oxygen carrier molecule.
• Denatured cell membrane results cell less flexible and more prone to be collected in the splenic circulation.
• Oxidized haemoglobin precipitates within the cell and damages the cell membrane.
• This leads to premature destruction of red cells.
Haemoglobin defects

• 2 main types.

1. Abnormal quantity of structurally normal haemoglobin production- thalassaemia.

2. Structurally abnormal haemoglobin production- haemoglobinopathies
Hereditary disorders of Haemoglobin

- **Haemoglobinopathies:** Production of structurally defective haemoglobin due to abnormalities in the formation of the globin moiety of the molecule

- **Thalassaemias:** Due to reduced rate of production of normal haemoglobin due to absent or decreased synthesis of one or more types of globin polypeptide chains
Thalassaemia

• Thalassaemia is an autosomal, inherited disorder.
• Due to a mutation or deletion in the genes that code for the globin chains.
• Results an imbalance in the alpha and beta globin chain production.
• Adult haemoglobin molecule- 2 molecules of alpha chains and 2 molecules of beta chains.
• Impaired synthesis of beta chains- beta thalassaemia
• Impaired synthesis of alpha chains- alpha thalassaemia
• In Sri Lanka beta thalassaemia is the predominant form present.
• Excess chains precipitate out and give rise to small inclusions within the red cells.
• These inclusions make the red cell more fragile and therefore undergo haemolysis.
• Two beta globin genes are inherited one each from each parent.
• The presence of one defective beta globin gene usually gives rise to a mild form of anaemia which is called beta thalassaemia trait.
• If both beta genes inherited from the parents are defective, and the baby cannot synthesize any beta globin chains, a transfusion dependent anaemia results.
Beta Thalassaemia minor

Genetic carrier for thalassaemia (mother)

Non-carrier (father)

2 out of 4 chances 50%

Beta Thalassaemia minor
Haemoglobinopathy

- Structurally abnormal haemoglobins are produced due to mutations in the DNA sequence.
- The commonest haemoglobinopathy in the world - sickle cell anaemia where the red cells take the shape of a sickle.
- This too like thalassaemia is an autosomal, inherited disorder.
- However, unlike thalassaemia, they are not transfusion dependent for survival.
- Sickle cells are formed when haemoglobin releases its oxygen in the peripheral tissues and assumes a deoxygenated state.
• The haemoglobin molecules in this state become sticky and adhere to one another forming sickles.

• These sticky adherent cells block small capillaries causing hypoxia
Acquired or extracorpuscular causes of red cell destruction

• Red cells may be prematurely destroyed by certain conditions which are acquired later in life. They may be divided into 2 types.
  1. Immune causes of red cell destruction
  2. Non immune causes of red cell destruction
Immune causes of red cell destruction

- Immunoglobulins produced against antigens on the surface of the red cell membrane.

- **Auto immune haemolytic anaemia** - immunoglobulins produced within the person itself. These antibodies are usually produced with other auto immune antibodies directed against a variety of other tissues.

- Examples:
  - Systemic lupus erythematosus
  - rheumatoid arthritis.
• Rarely- Infectious mononucleosis, mycoplasma may get the host to produce antibodies which are very similar to the antigens on the red cell membrane which cross react to destroy the red cells.
• Certain drugs for e.g. chlorambucil may give rise to auto antibodies that destroy red cells.
• Depending on the temperature that antibodies are most active- 2 types of immune haemolytic anaemia.
• Warm antibodies- Antibodies that show optimal activity at body temperature- warm immune haemolytic anaemia.
• Cold antibodies- Antibodies that show optimal activity at room temperature or lower temperatures than the body temperature - cold antibody immune haemolytic anaemia.
• Once the red cells are coated with antibodies, complement activation occurs.
• Complement activation gives rise to the membrane attack complex which bores small holes in the membranes of the red cells causing haemolysis.
Red cell destruction

Antibody coated in the activation of the red cell

Complement attack on the red cell membrane

Membrane attack complex bores holes
Non immune causes of red cell destruction

- Destroyed mechanically by traversing through the vascular system.
- Causes are –
  - Burns
  - CRF
  - Infections
  - Liver disease
  - Drugs
  - Pb-Lead
  - Chemicals
  - Malignancy
Impaired red cell production

- Due to a defect in the bone marrow itself or the lack of vital nutrients.
- The commonest cause in the world- iron deficiency.
- There are 3 important substances that cause anaemia. They are
  - Iron
  - Vitamin B12
  - Folic acid
Iron deficiency anaemia

• Essential for formation of **haemoglobin**.

• Iron deficiency occurs mainly due to
  – loss of blood
  – inadequate intake in the diet.

• The most common cause of blood loss is due to hookworm infestation.

• Rare causes- surgical resection of the distal part of the ileum where iron is absorbed from.
Folate deficiency

- Folate-Not synthesized in the human body.
- It is a vitamin that is required for DNA synthesis.
- In red cell formation, DNA synthesis which is an integral part of cell division is happen.
- DNA molecule is make up of an nucleotides with guanine, adenine, cytosine and thymine nucleotides.
- Folate helps in the conversion of uracil to thymine.
- Folate deficiency- deficiency of thymine containing nucleotides and they cannot pair up with adenine to hold the double stranded DNA together.
- At the time of cell division as the DNA strands loosen their double helical structure small breaks occur in it which halts cell division.
Vitamin B12 deficiency

- Vitamin B12 - present in animal proteins and is stored in the liver.
- Stores are adequate for 2 years and anaemia sets in long after the stores are depleted.
- Vitamin B12 - coenzyme in the formation of methionine.
- Vitamin B12 - a gate keeper to let folate from the plasma into the cell.
- Thus in vitamin B12 deficiency the cells have an absolute deficiency of folate in the intracellular compartment.
- Anaemia due to inability of the bone marrow to proliferate to produce effective red cells.
Vitamin B12 is...

- Also known as cobalamin
- Related to blood deficiency diseases
Folate and B12 in DNA Synthesis
Bone marrow failure

• Site of production of white cells, red cells and platelets.
• In instances of bone marrow failure all three types of cells that is white cells, red cells and platelets are produced in very small numbers.
• The reduction in all these three cell lines is called pancytopenia.
• Features of bone marrow failure are therefore due to the effects of reduction of these cells.
• Low level of
  – white cells, if neutrophils-recurrent bacterial infections.
  – red cells- anaemia
  – Platelets-bruising and gum bleeding.
causes of bone marrow failure

• They can be divided into the following categories.
  – Intrinsic stem cell failure
  – Toxic chemical exposure
  – Deficiencies of essential components of cell division
  – Exposure to radiation
Intrinsic stem cell failure

- Stem cell - capability of giving rise to any cell line that is produced by the marrow- pluripotent stem cell.
- Ex. 1. Aplastic anaemia - the pluripotent stem cell is defective with markedly reduced production of all cell lines of the bone marrow. The low proliferation rate of the bone marrow results in the bone marrow space being replaced by fat tissue.
- Exposed to cytotoxic drugs - the stem cell could be damaged due to the toxin
- In older people - unknown.
4. Myelodysplastic syndromes- a group of disorders that arise due to a clonal abnormality of the bone marrow stem cell, causes abnormality in proliferation and differentiation
Exposure to toxic chemicals or drugs

- Certain drugs and toxins damage the pluripotent stem cell.
- This damage has a harmful effect on the proliferation of the marrow cells.
- This results in reduction in cell production.
- Examples:
  - heavy metals
  - Drugs used in cancer therapy: anti cancer drugs halt proliferation in the cancer cells. These drugs unfortunately do not differentiate between normal cells and the malignant cells. Therefore all the cells with rapid proliferative activity in the body are affected. The cells that are continually proliferating and being replaced are mainly the cells of the gastro intestinal tract, skin cells, hair cells and the bone marrow. These systems are the worst affected in times of chemotherapy. With chemotherapy, there is a marked reduction in cell proliferation resulting in transient bone marrow failure.
Deficiency of essential nutrients for cell division

- Folate and vitamin B12
- Vitamin B12 is present only in animal proteins.
- Some very strict vegetarians may present with pancytopenia.
Exposure to radiation

• Accidental or intentional exposure to ionizing radiation as in therapy for malignancies, can irreversibly damage the bone marrow.
• Ionizing radiation damages the chromosomes which make up the nucleus.
• The damage to the chromosomes delays cell division and cells die without giving rise to new cells.
• Ionizing radiation irreversibly harm the stem cells in the bone marrow causing the entire marrow to stop cell production completely.
Thank you!