BLOOD PHYSIOLOGY

Composition and Functions of blood

Erythropoeisis

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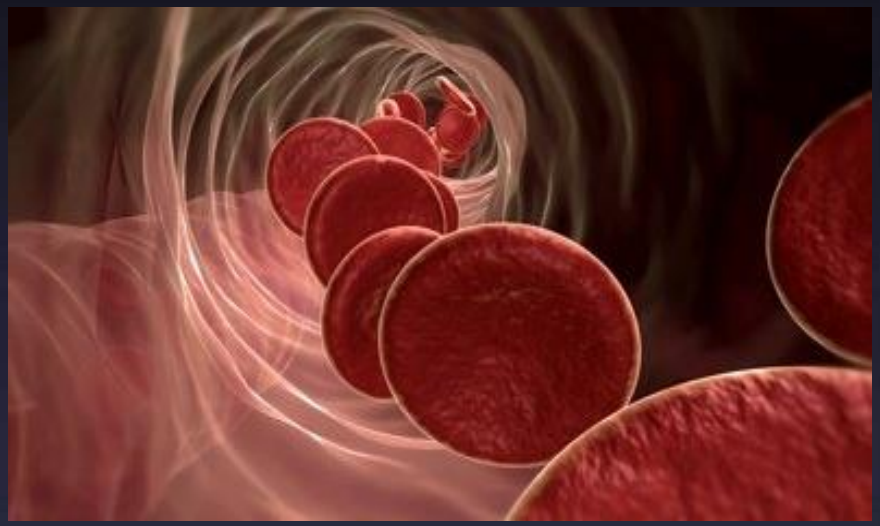


Objectives

At the end of this lecture you should be able to:

1. **Understand functions and composition of blood**
2. **Describe essential elements needed for RBC formation.**
3. **Describe the process of Vit B12 absorption and its malabsorption.**
4. **Discuss iron metabolism (absorption, storage and transport)**
5. **Recognize haemaglobin structure and its functions**
6. **Describe the fate of old RBC.**
7. **Describe anemia & polycythemiaand its causes.**

Blood is a specialized type of liquid connective tissue.

Functions Of the blood

1. ***Transport*** *(O2, nutrients, CO2, waste products, hormones)*
2. ***Protecting*** *the body against infections (White Blood Cells, Antibodies) & Hemostasis (preventing blood loss)*
3. ***Homoeostasis*** *(Regulation of body temperature, Regulation of ECF pH)*

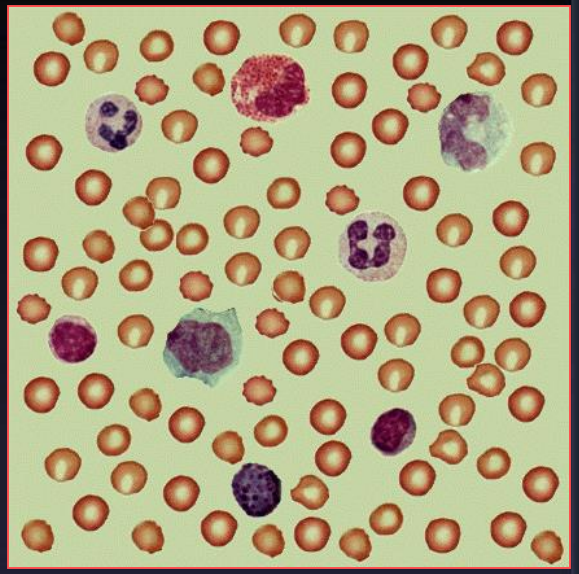
Blood: Sub-functions

- **Respiration : oxygen and carbon dioxide are transported**
- **Trophic : nutrients are delivered to the tissues**
- **Excretive : metabolites are delivered from tissues to excretory organs**
- **Regulative : hormones and BAS are transported**
- **Homeostatic : maintenance of water content and acid-base balance**
- **Protective : immunity and non-specific resistance; blood coagulation**
- **Maintenance of body temperature : as a result of a redistribution of blood volume between skin and the internal organs at high and low temperature of external environment.**

Blood Composition

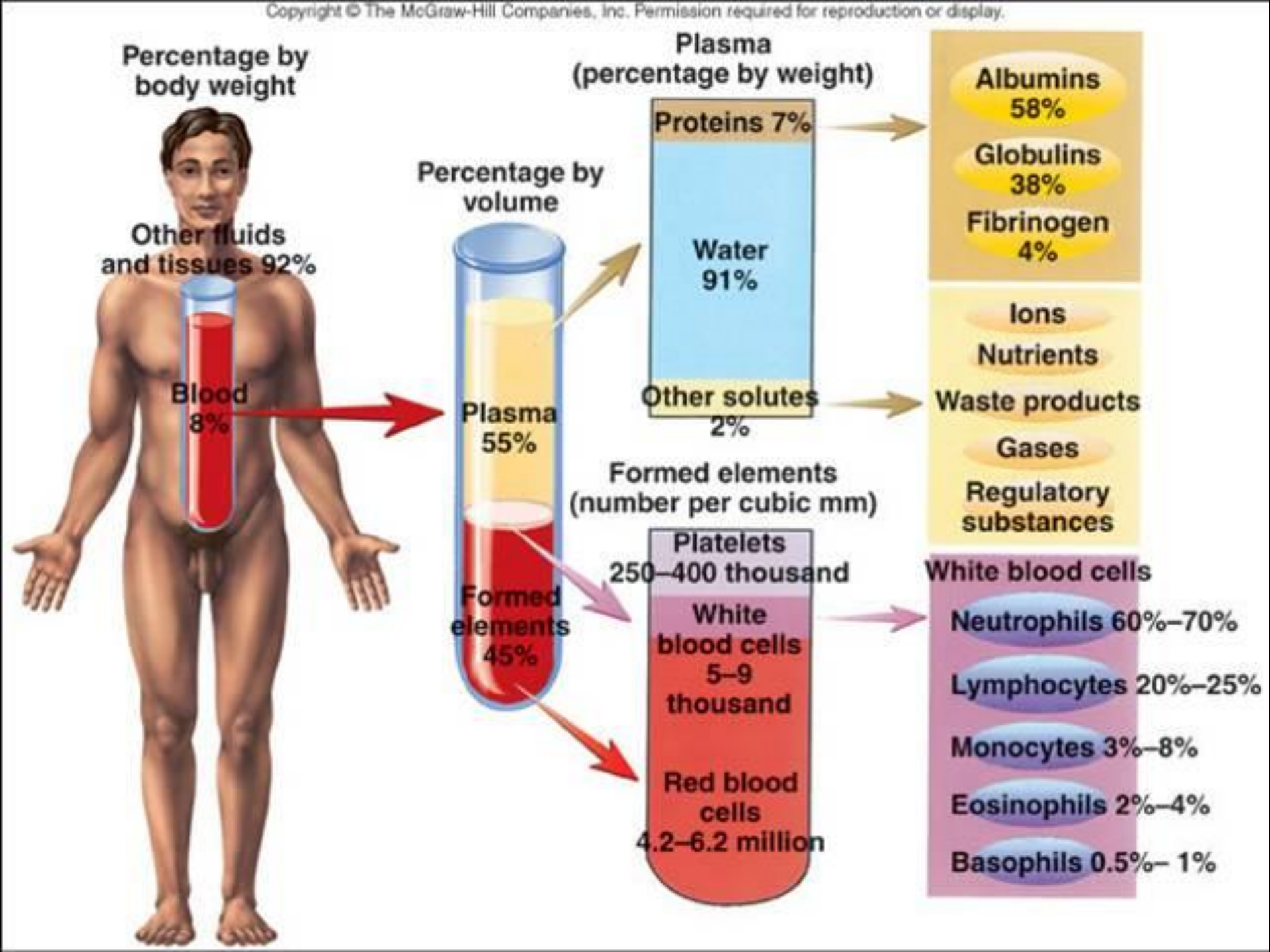
1. Cellular components

- *Red Blood Cells 5.2 million/ul-4.7 million/ul*
- *White Blood Cells 4000-11000/ul*
- *Platelets 150000-400000/ul*

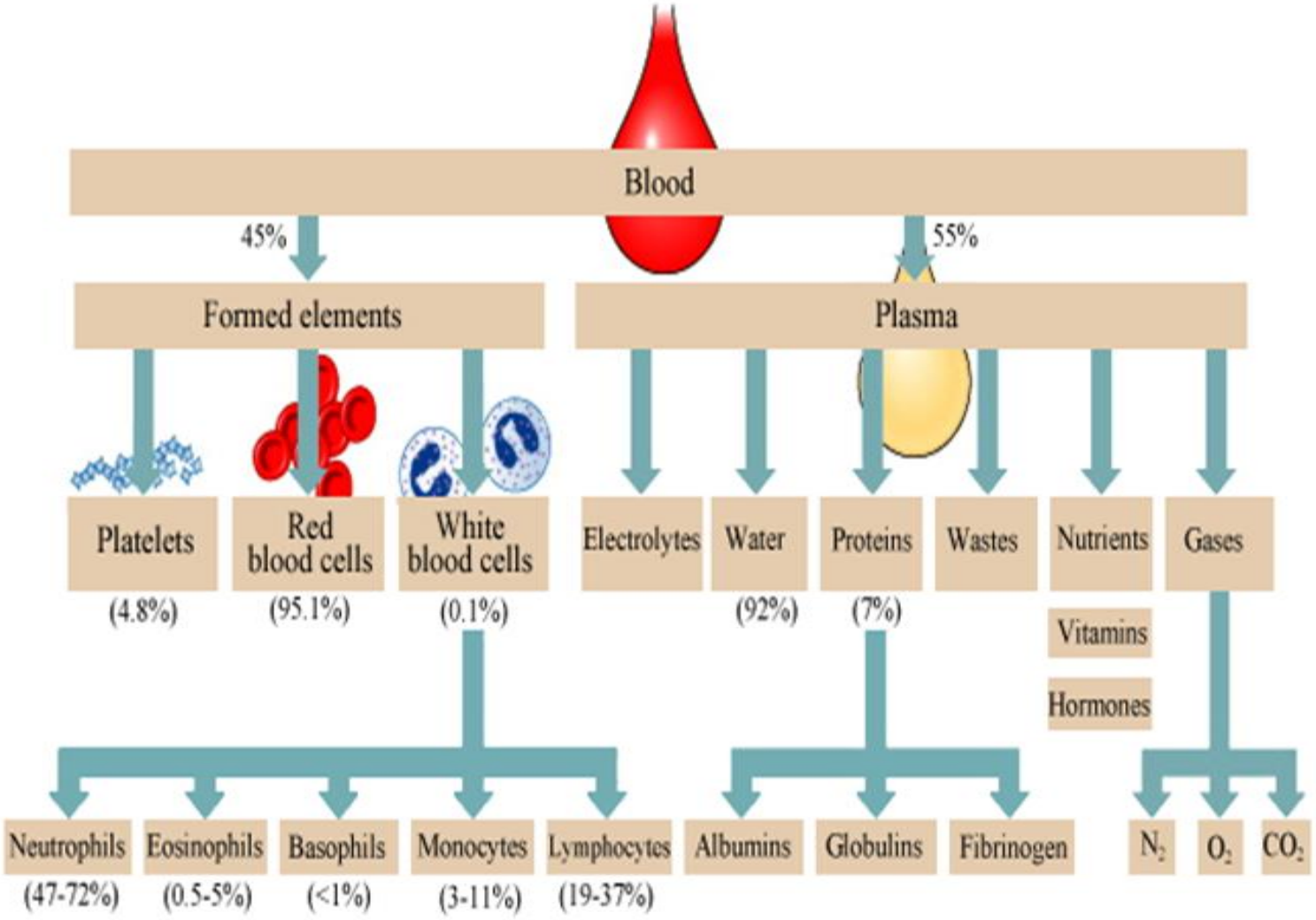


2. Plasma consist of:

- *Water: 98%*
- *Ions: Na, K, HCO3, PO4 ..etc*
- *Plasma proteins (Albumin, globulin, Fibrinogen)*
- *Same ionic composition as interstitial fluid*



Blood Composition



Characteristics of Blood

- *Quantity 5-6 Liters*
- *Temperatute 37 ^{o} C*
- *Viscosity 3-4 times than Water*
- *Hemoglobin 15 gm/dl (13-16 females, 14-18 males)*
- *O2 Carrying Capacity of Blood 1.39 ml/gm of Hb*

Physical and chemical characteristics

- **Specific gravity:**

  Total blood (1.050-1.060) more influenced by red blood cells; **plasma** (1.025-1.030) more influenced by plasma protein; **RBC** (1.090-1.092) more influenced by Hb.

- **Viscosity:**

  Blood relative viscosity (4~5) mainly depends on the numbers of red blood cells.

  Plasma relative viscosity (1.6~2.4) is mainly involved in plasma protein

Physical and chemical characteristics

- **Plasma osmotic pressure** is 300 mmol/L or 770kPa

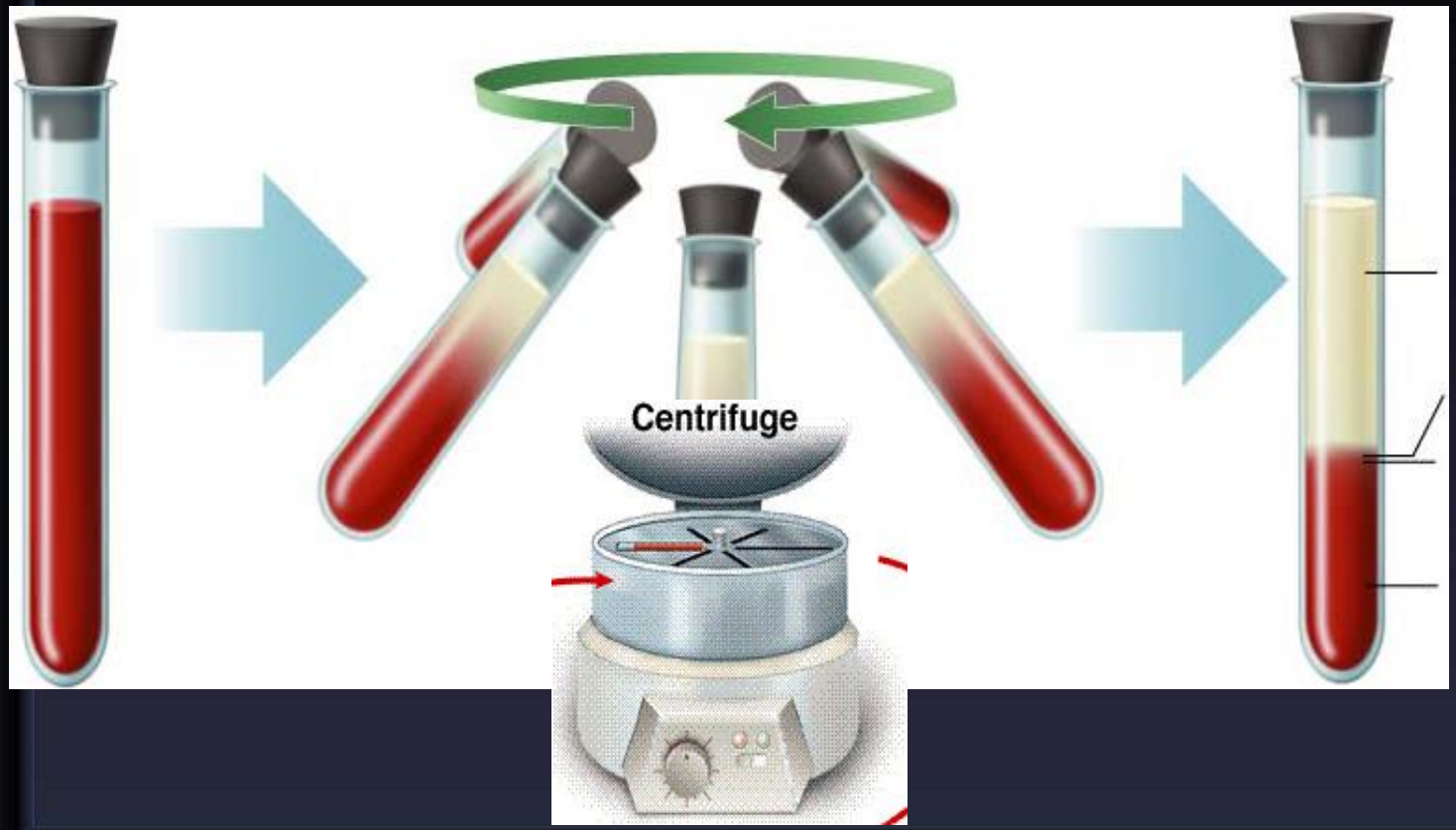
  Crystal osmotic pressure results from NaCl and modulates water distribution between inside and outside of cells.

  Colloid osmotic pressure results from albumin and regulates water distribution between inside and outside of capillary.
- **Plasma pH** value is about 7.35~7.45 (Hb acts as blood buffer)

Whole Blood Sample

Sample Placed in Centrifuge

Blood Sample That Has Been Centrifuged



Plasma (55%)

Platelets

White blood cells < 1%)

Red blood cells (45%)

Composition of Blood

Cells

45 %

RBCs
WBCs
Platelets

Plasma

55%

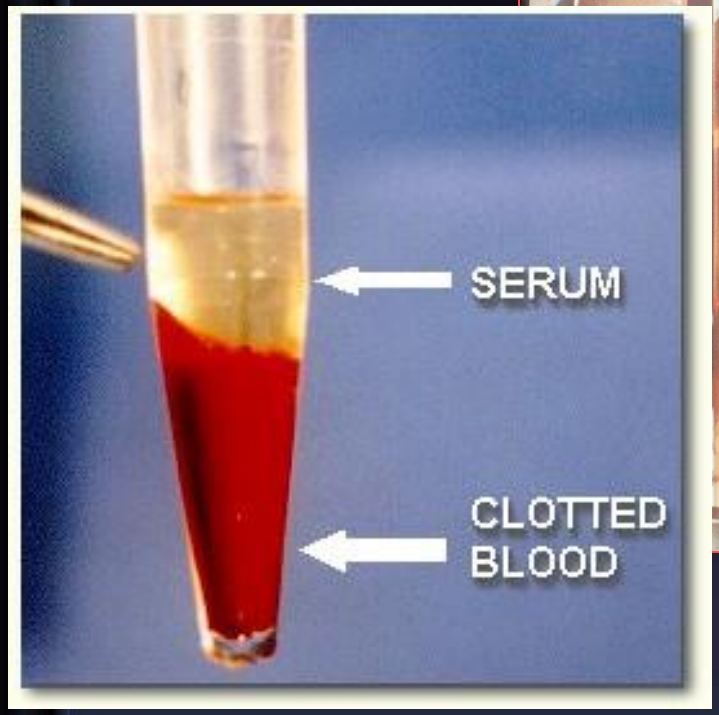
Electrolytes
Clotting Factors
Antibodies
Blood Gases
Nutrients
Wastes

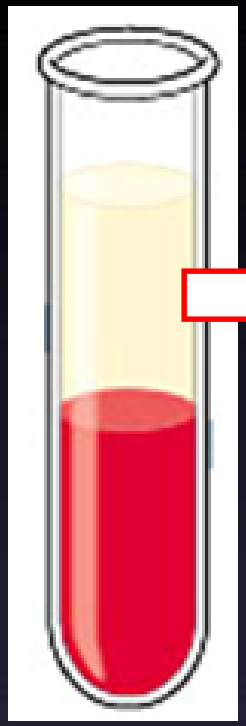
The Plasma is a straw colored liquid (90-92% Water). It serves as a transport medium for blood cells and platelets.

When clotting factors specially fibrinogen is removed from plasma as a result of coagulation, it is called serum.

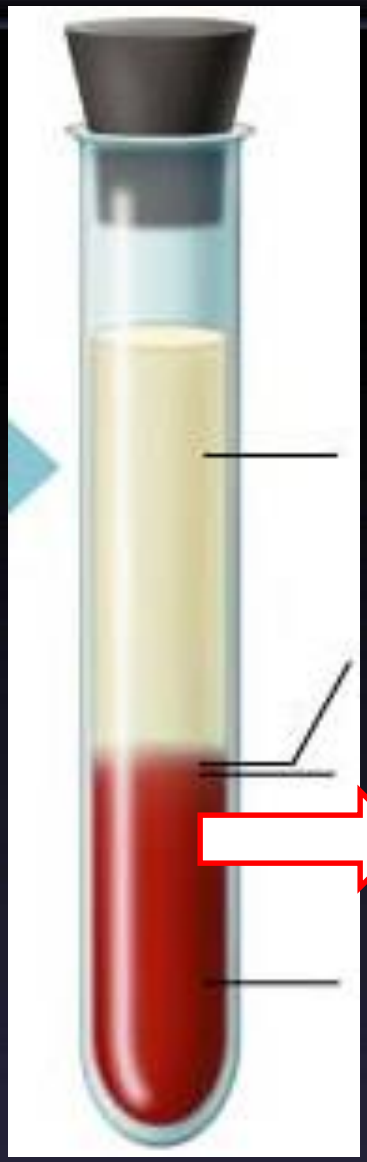

Baxter


129169J 6
Fresh Frozen Plasma (CPD-A1)
Human
Infuse using an in-line filter
Lot
87K04B67
DO NOT VENT
Sterile, nonpyrogenic
ABO Blood Group
Rh Type
O
Rh POSITIVE


SERUM
CLOTTED BLOOD



Constituents of PLASMA	Major Functions
Water	Solvent for carrying other substances
Salts Sodium Potassium Calcium Magnesium Chloride Hydrogen carbonate	Osmotic balance, pH buffering, Regulation of membrane permeability
Plasma Proteins Albumin Fibrinogen Antibodies	Osmotic balance, pH buffering, Clotting, Immunity Transportation (Binding Prot)
Substances transported by blood Nutrients (e.g. glucose, vitamins etc) Waste products of metabolism (Urea, Uric acid) Respiratory gases (O2 & CO2) Hormones (Many)	



CELLULAR ELEMENTS 45%

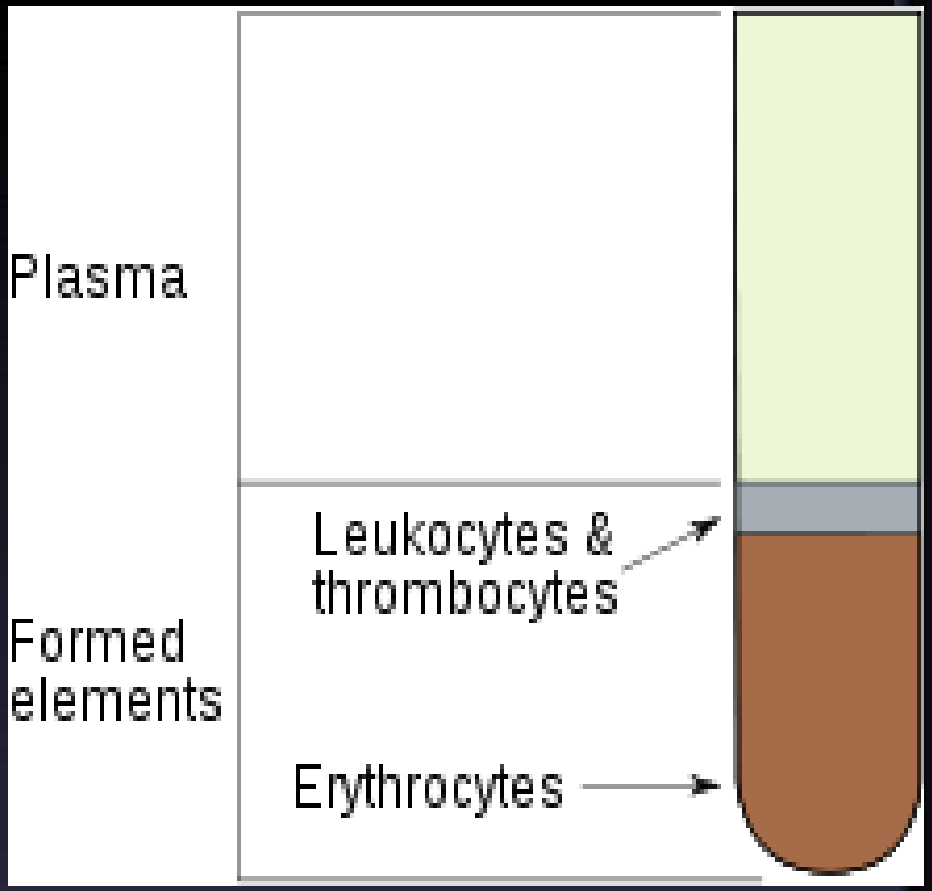
CELL TYPE	NUMBER (per mm^3 of blood)	FUNCTIONS
Erythrocytes (red blood cells)	5–6 million	Transport of oxygen (and carbon dioxide)
Leucocytes (white blood cells)	5,000–10,000	Defence and immunity
Basophil		
Eosinophil		
Lymphocyte		
Neutrophil		
Monocyte		
Platelets	250,000–400,000	Blood clotting

Hematocrit

The hematocrit , also known as packed cell volume (PCV) or erythrocyte volume fraction (EVF), is the volume percentage (%) of red blood cells in the blood. It is normally about

40-48% for men and

36-42% for women



Summary

pH of blood	• The normal pH range of blood is 7.35 to 7.45,which is slightly alkaline. The venous blood normally has a lower pH than the arterial blood because of presence of more Carbon dioxide.
Temperature	• The temperature of the blood is 38°C(100.4°F), about 1°Chigher than oral or rectal body temperature.
Viscosity	• 'Viscosity' means thickness or resistance to flow. Blood is about 3-5 times denser & more viscous(thicker) than water & feels slightly sticky. Viscosity is increased by the presences of blood cells & plasma proteins. This thickness contributes to normal blood pressure.
Colour	• The colour of blood varies with its oxygen content. Arterial blood is bright red due to it's high level of oxygen. Venous blood has given up much of it's oxygen in tissues & thus has a darker, dull red colour.
Amount	• Blood constitutes about 20% of extracellular fluid, amounting to 8% of total body mass. The blood volume is 5L to 6L(1.5gal) in average sized adult male & 4L to 5L(1.2gal) in an average sized adult female.

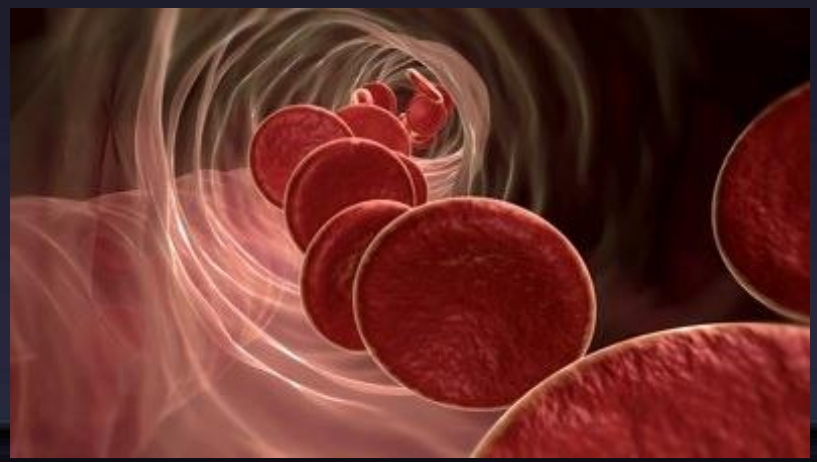
Red Blood Cells

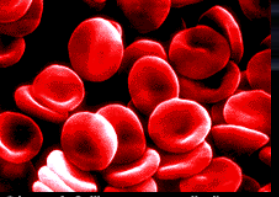
Biconcave Discs: (7.5X2X1um)
Negative Charge
Life Span=120 days

ERYTHROS MEANS : RED
KYTOS MEANS : CELL

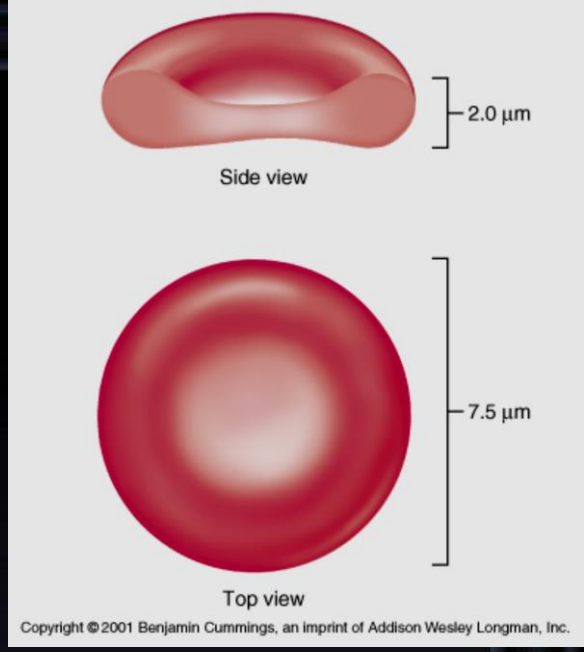
Composition: **60% is water & 40% solids**
(90% of solids content is Hb while 10% is stromatin)

Count: **Males 4.8-5.8 million cells/mm3**
Females 4.2-5.2 million cells/mm3





Red Blood Cells



- **Structure** (7.5X2X1um)
  - Biconcave Discs
  - Non-nucleated
  - framework of protein (stromatin) + hamoglobin
- **Phospholipid semi-permeable membrane**
- **Composition**
  - 60% water
  - 40% solids (90% of solids content is Hb, 10% stromatin)

A Mature RBC

DOES NOT HAVE

- Nucleus
- Mitochondria
- Ribosomes
- Endoplasmic Reticulum
- Golgi Apparatus

IT CONTAINS

- Haemoglobin
- Enzymes For Glucose Metabolism (Carbonic Anhydrase & 2,3 DPG Synthesis)
- Structural Proteins (cytoskeleton)

ENERGY METABOLISM OF RBC:

(1) GLYCOLYSIS 90%

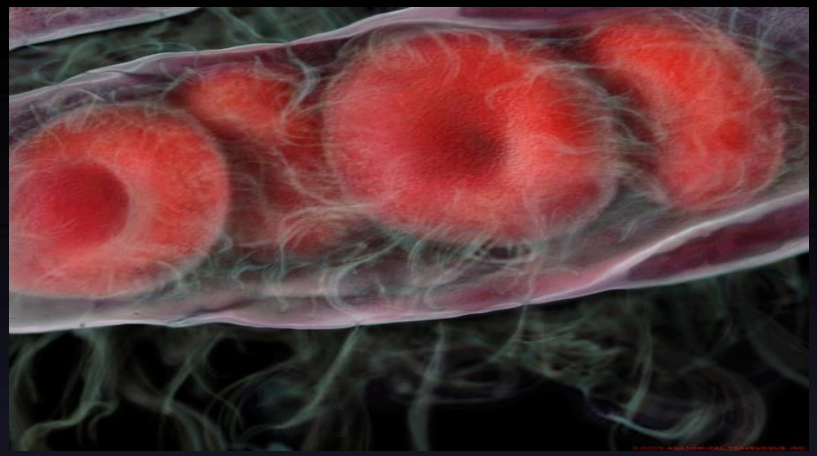
(2) HMP SHUNT 10%

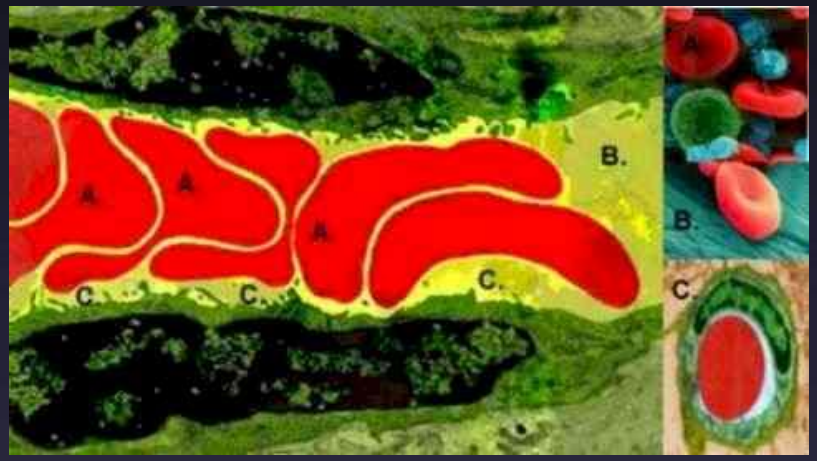
(GLUCOSE ENTERS INTO RBC BY CARRIER MEDIATED DIFFUSION)

Red blood cells

- Contain **haemoglobin** which carry (reversible reaction)
- They **don’t have a nucleus** so there is more room for haemoglobin
- Are a **biconcave disc**, this increases surface area to volume ratio to increase rate of oxygen uptake.
- They **bend** as they pass through capillaries (Thimble / Parachute Shaped)

Red Blood cells in capillaries





- Capillaries are very narrow.
- Red blood cells bend as they pass through them
- This is an advantage, it keeps them in very close contact with the capillary walls; this reduces the diffusion distance for gas exchange with the surrounding tissues

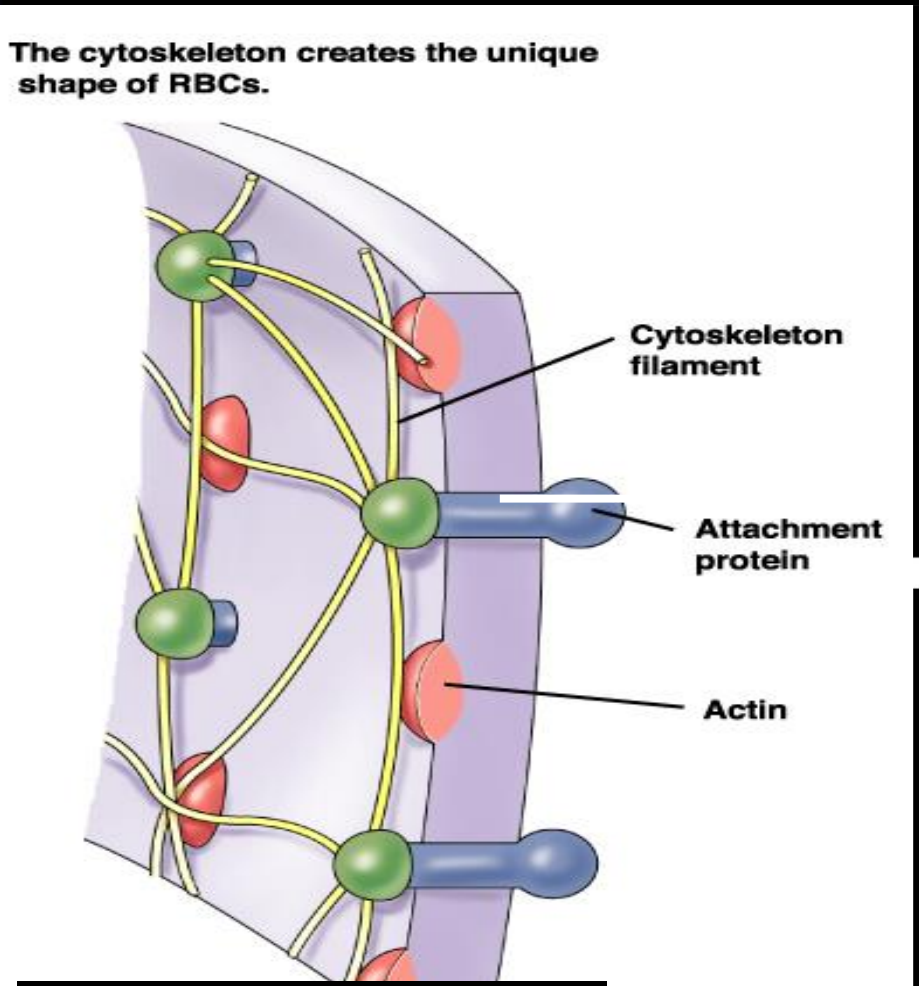
RBC are flexible & elastic:

- **to squeeze through narrow capillaries**

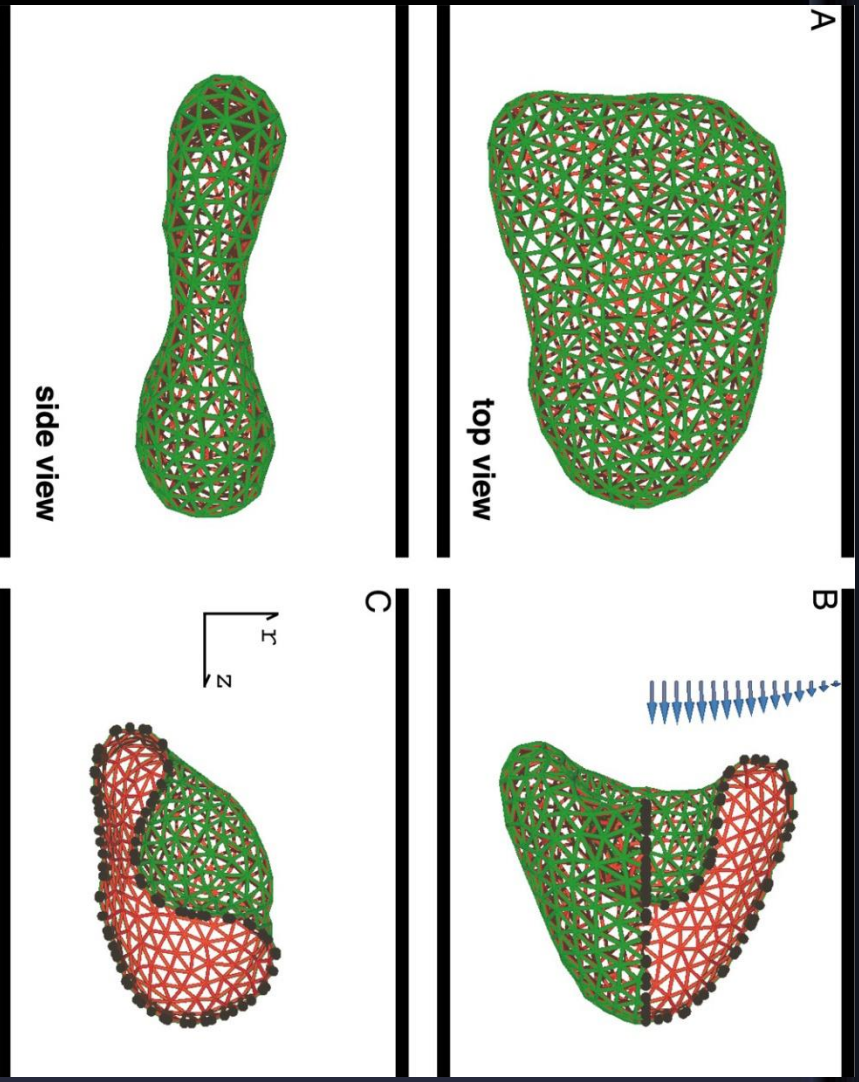
Cardiac muscle and capillary

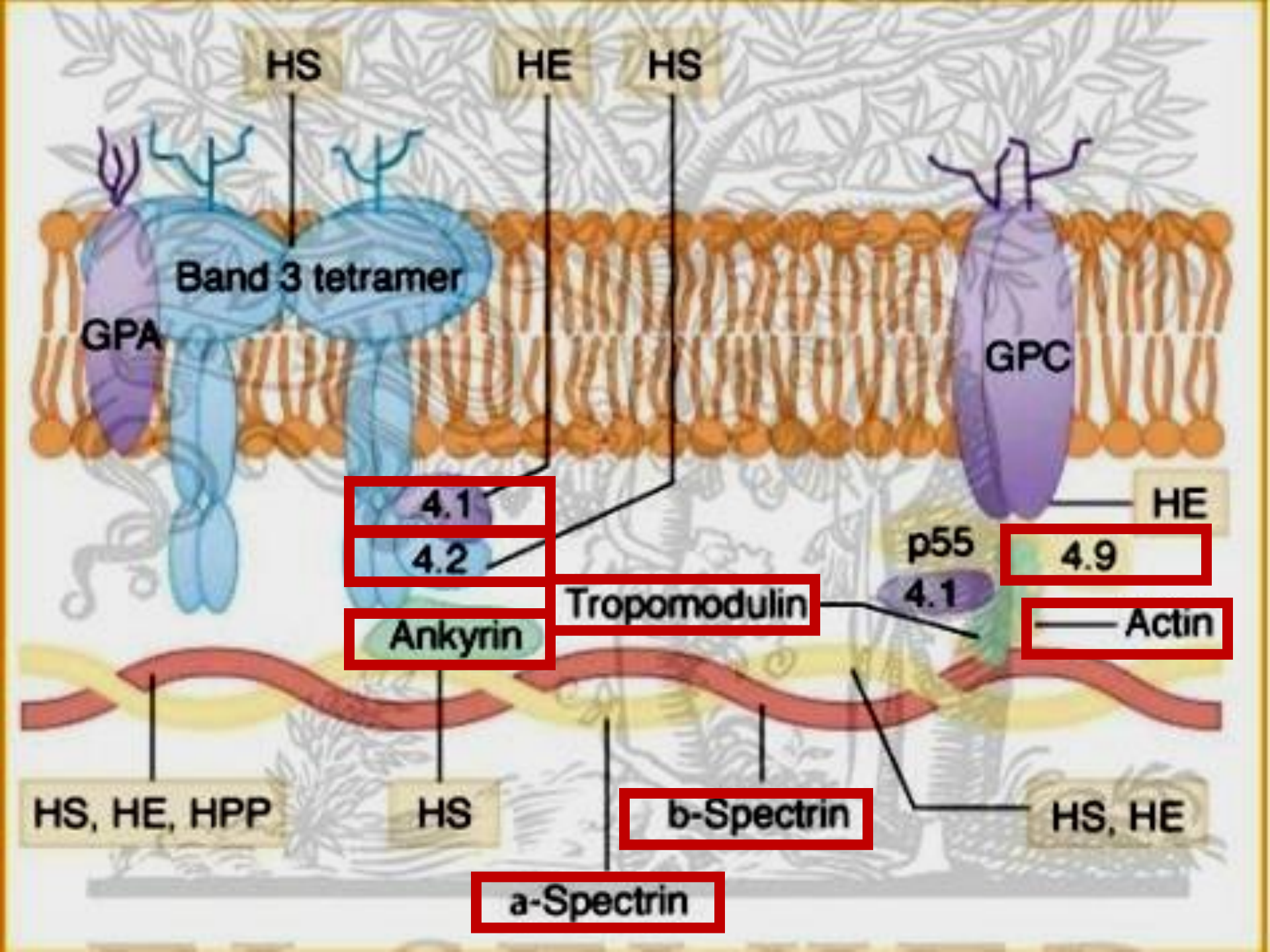
Erythrocytes in single file – capillary is so narrow

Reason for shape of RBC



biconcave disc like




HS
HE
HS
Band 3 tetramer
GPA
GPC
4.1
4.2
HE
p55
4.1
4.9
Tropomodulin
Ankyrin
Actin
HS, HE, HPP
HS
b-Spectrin
HS, HE
a-Spectrin

RBCs Size, Color, Indices

- **<u>According to size</u> :**

  Normocytes - Normal sized RBCs

  Microcytes - Small sized RBCs

  Macrocytes - Large sized RBCs

- **<u>According to colour</u> :**

  Normochromia - Normal coloured RBCs

  Hyperchromia - Darker,due to increased hemoglobin

  Hypochromia - Paler, due to decreased hemoglobin

- **They are determined by measuring the indices:**

  Mean corpuscular Volume

  (MCV= 78 TO 94 fl OR 83 Cubic um**)**

  Mean corpuscular hemoglobin

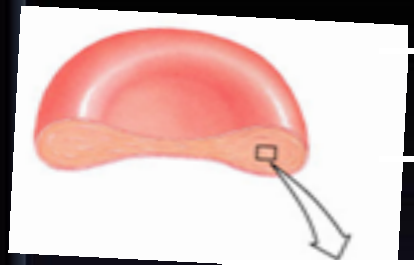
  (MCH= 27 – 32 picogram**)**

  Mean corpuscular hemoglobin concentration

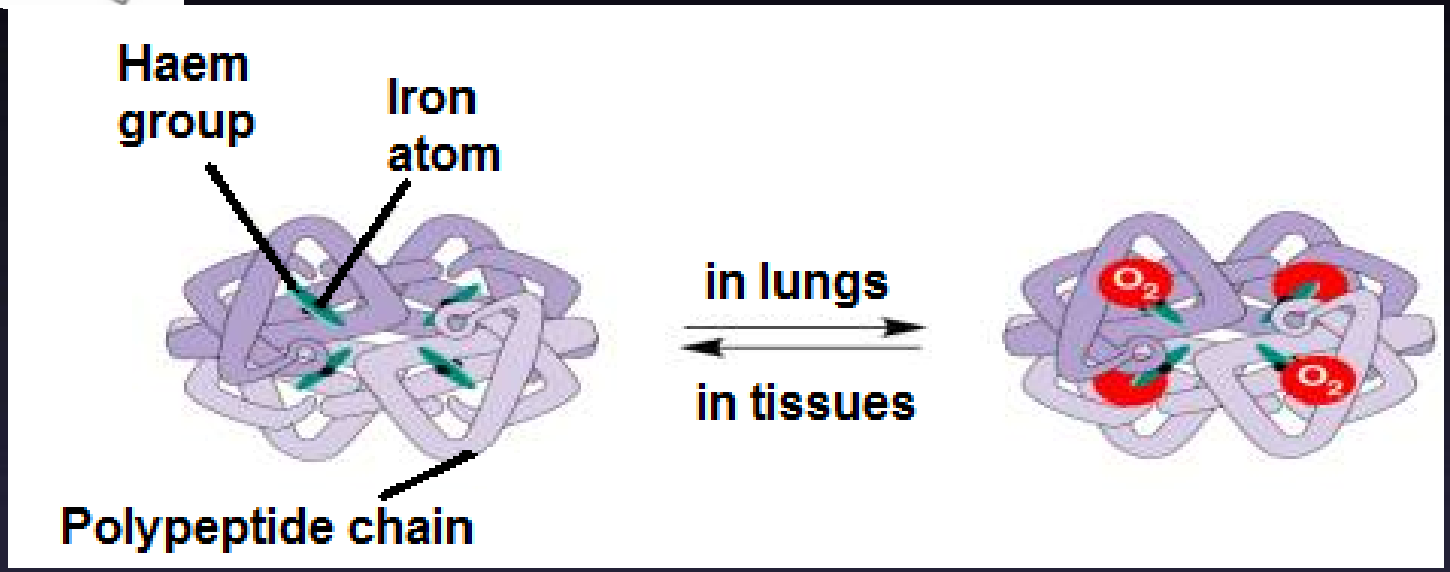
  (MCHC= 30 – 36 gm/dl**)**

RBC: packed with haemoglobin

- Hemoglobin:



- the oxygen-carrying protein pigment
- combines reversibly with O_2

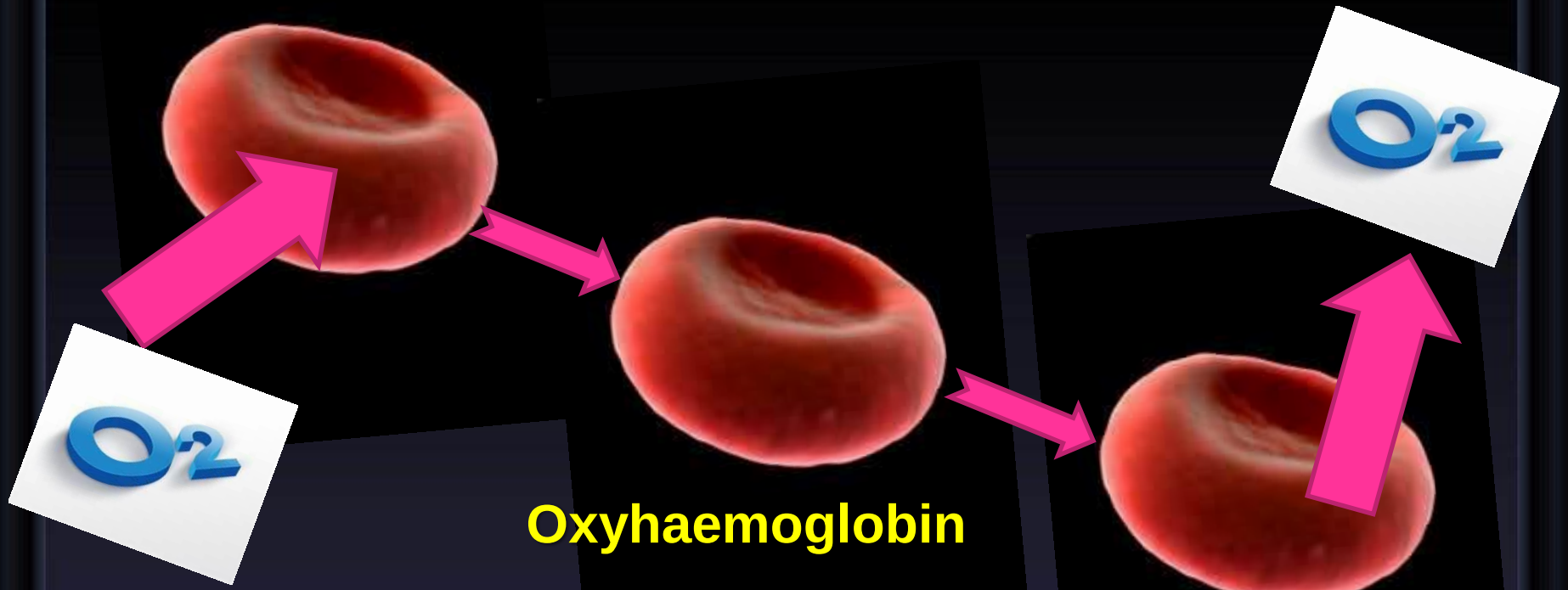


Haemoglobin

Hb + O_2 :

- in areas of high O_2 concentration

UNLOADING



Oxyhaemoglobin

LOADING

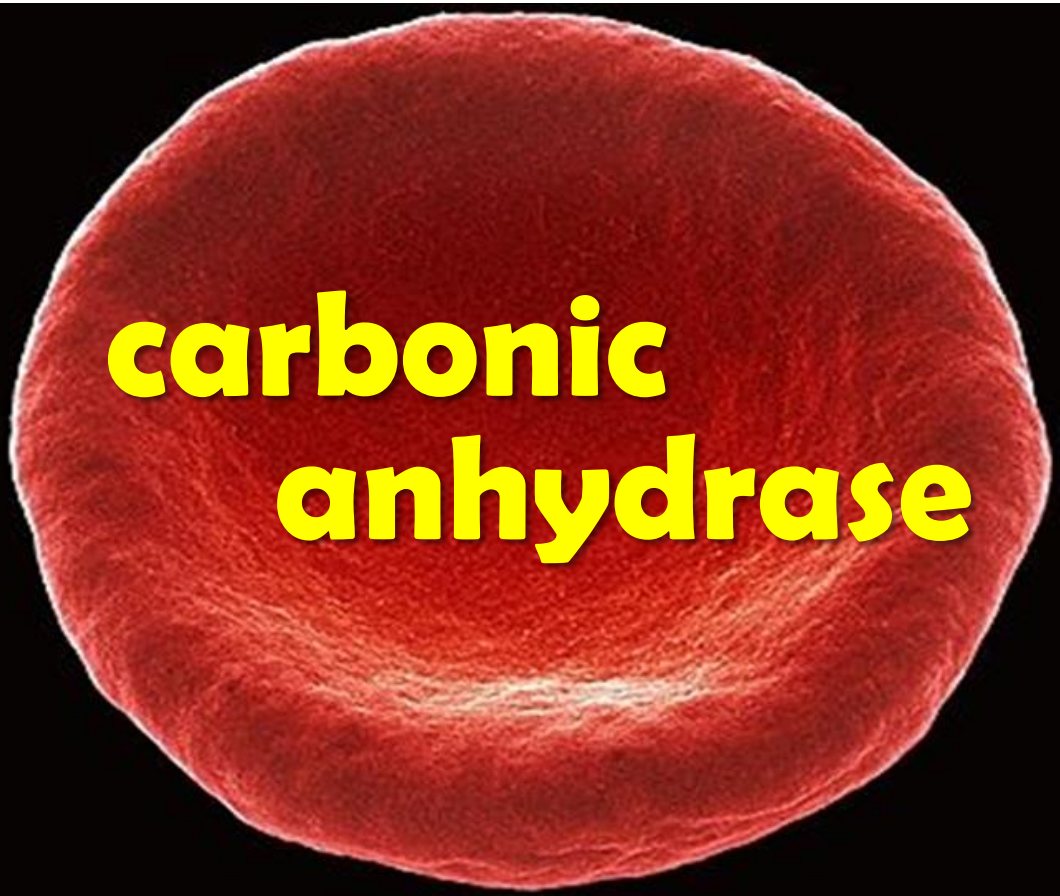
Hb releases the O_2 :

- in regions of low O_2 concentration

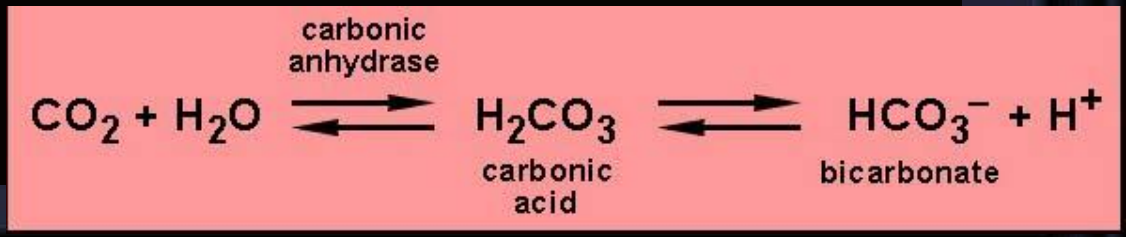
RBC lack mitochondria.
Give *two* advantages of this.

1. More room for carrying hemoglobin
2. Respire anaerobically : do not use up any of the O_2 they carry

RBC contain the enzyme:



plays a role in CO_2 transport



Physiological Characteristics & Functions of RBC

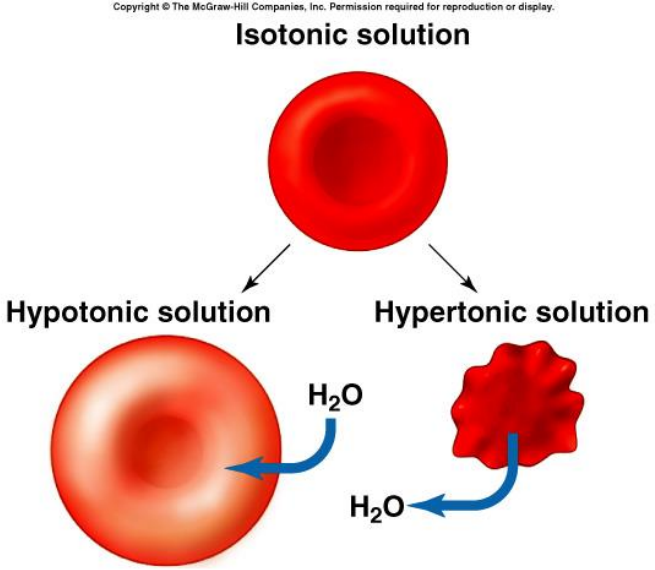
①Permeability: Semipermeable membrane, gas and urea freely passing through.

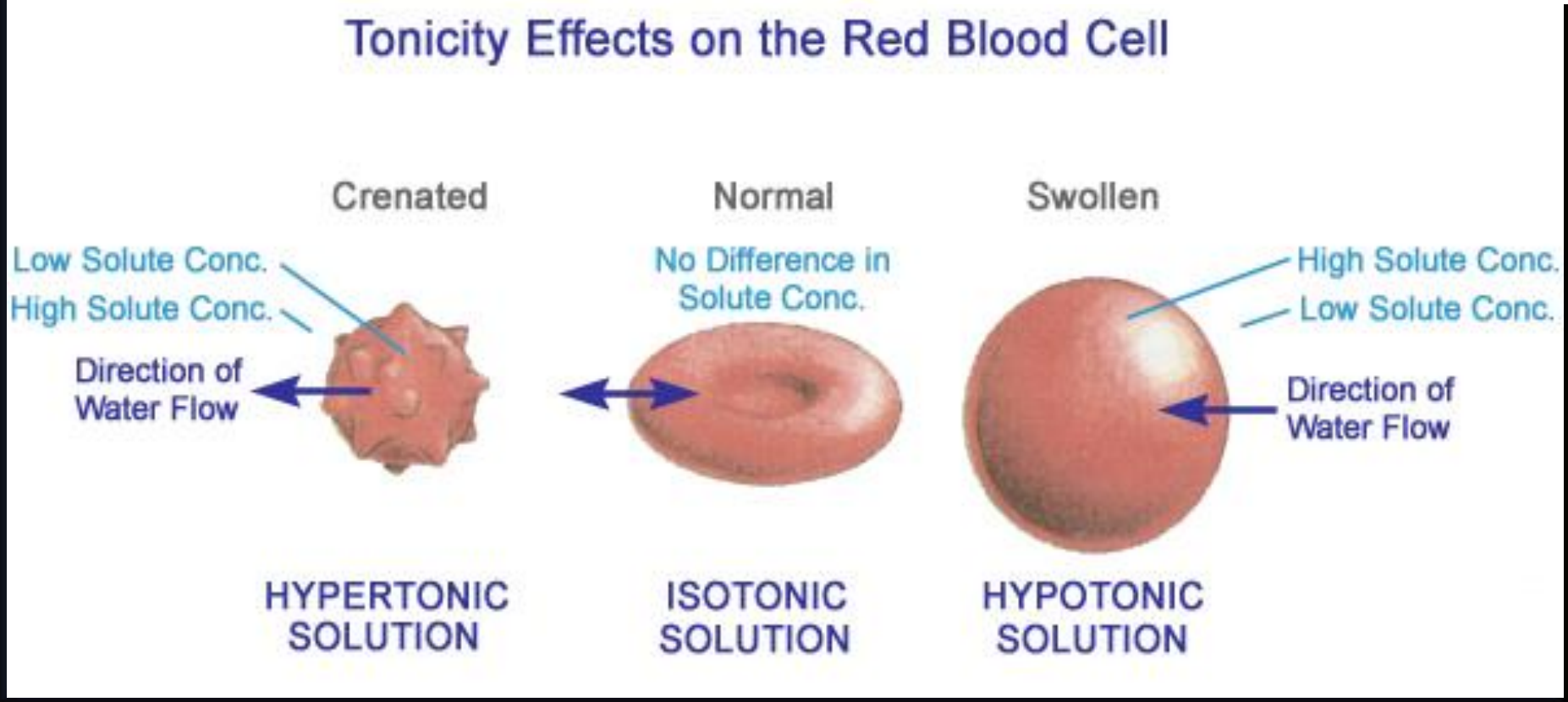
②Plasticity: depends on: 1) surface area-cubage ratio, 2) viscosity of Hb, 3) membrane elasticity and viscosity.

③Osmotic fragility: Changes in RBC put into lower osmotic salty solution. Osmotic fragility of aged RBC is large and easily results in rupture (hemolysis and ghost cell). Isosmotic solution, e.g. 0.85% NaCl & 1.4%$NaHCO_3$, 5% glucose, etc. Isotonic solution, e.g. 0.85% NaCl

④Suspension stability: it can be described by erythrocyte sedimentation rate (ESR) which is RBC descending distance per hour and suspension stability is inverse proportion to ESR. Normal value : male, 0~15 mm/h; female, 0~20 mm/h.

Tonicity effects

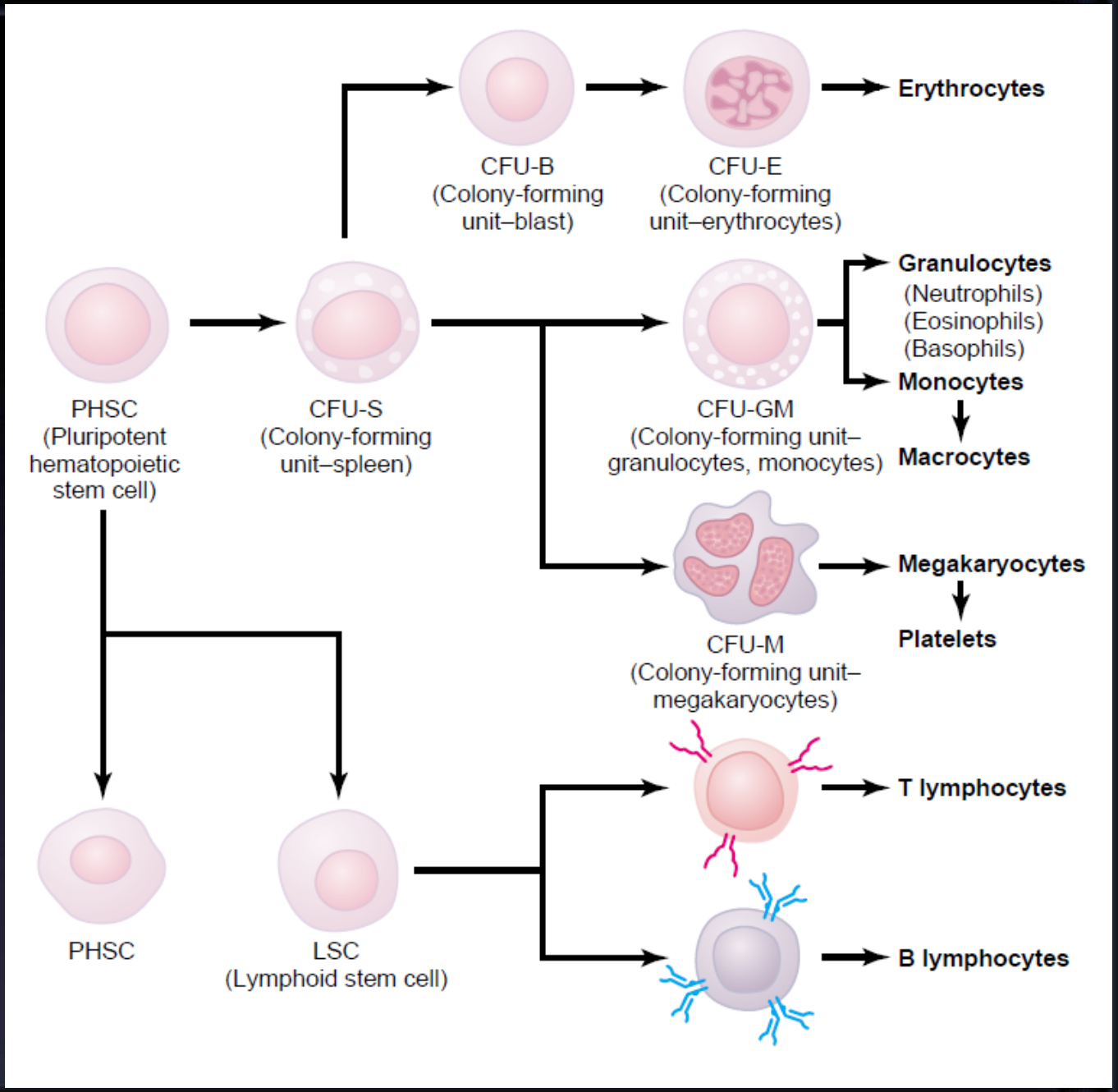


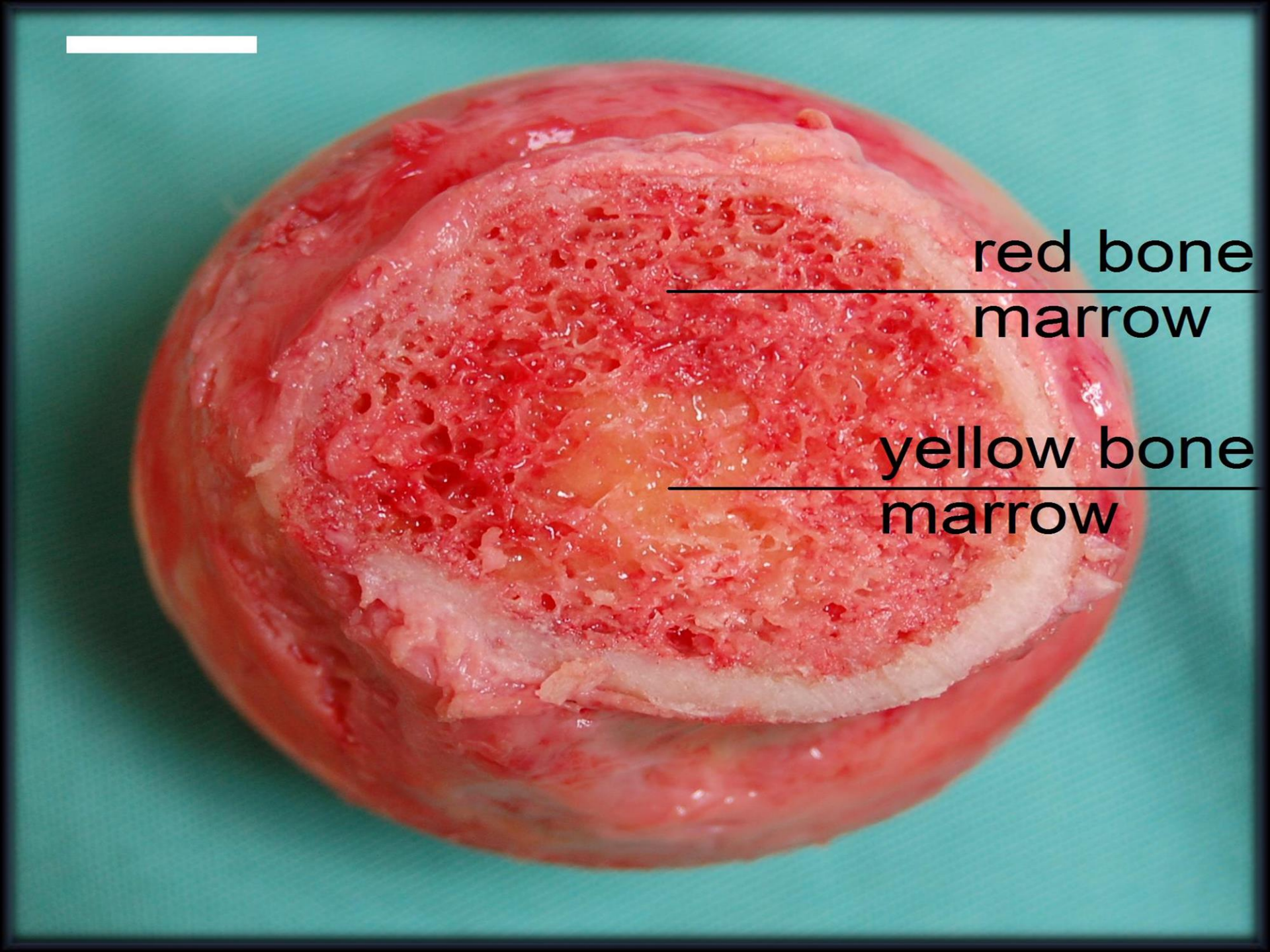


Blood Cells Formation

- Formation of erythrocytes (RBC)
  ► Erythropoiesis
- Formation of leucocytes (WBC)
  ► Leucopoiesis
- Formation of thrombocytes (platelets)
  ► Thrombopiesis
- Formation of blood
  ► Haemopoiesis.

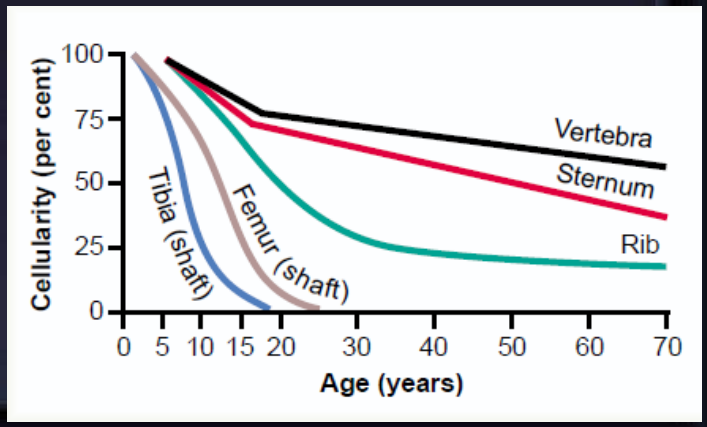
Formation of the multiple different blood cells from the original pluripotent hematopoietic stem cell (PHSC) in the bone marrow



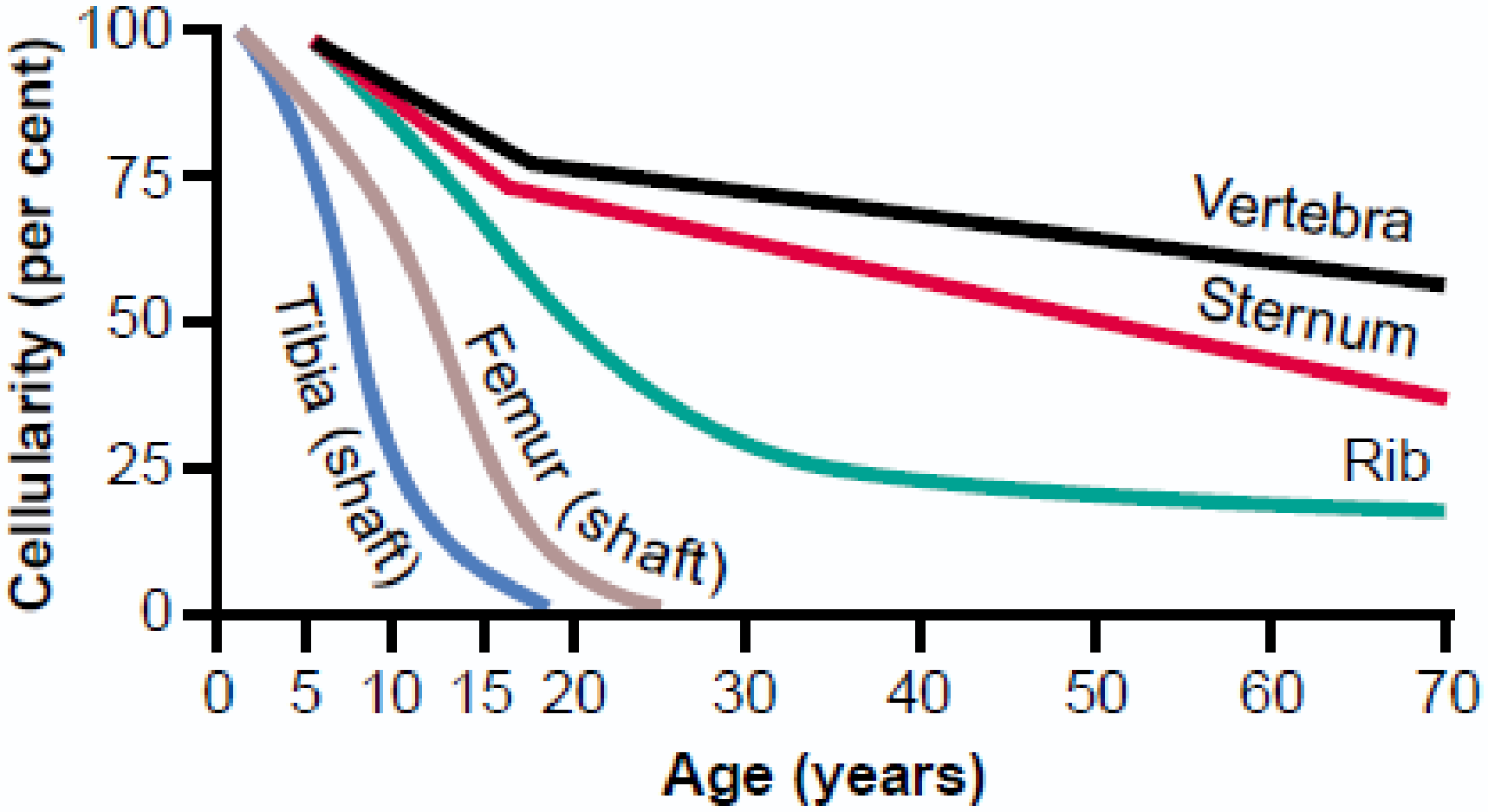

red bone marrow
yellow bone marrow

Sites of blood formation

- Adults ►Bone Marrow
- Children ► Bone Marrow
- Before Birth (In Fetus)
  - 1st 4 months ► Yalk Sac
  - Sec Trim ► Liver,spleen, lymph nodes
  - Third Trim ► Bone Marrow



Sites of blood formation



Erythropoiesis, (Formation of RBC)

Stages of RBC development (7 days)

Pluripotential haemopoietic STEM CELL

↓

Committed Stem cell

↓

Proerthroblast

↓

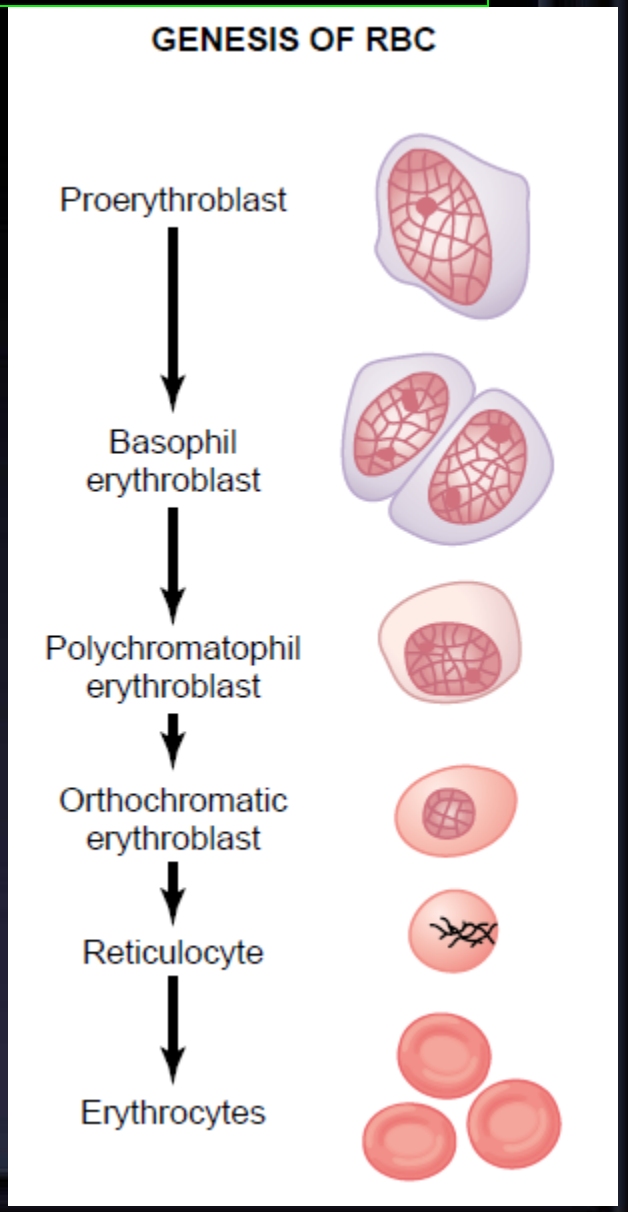
early, intermediate and late normoblast

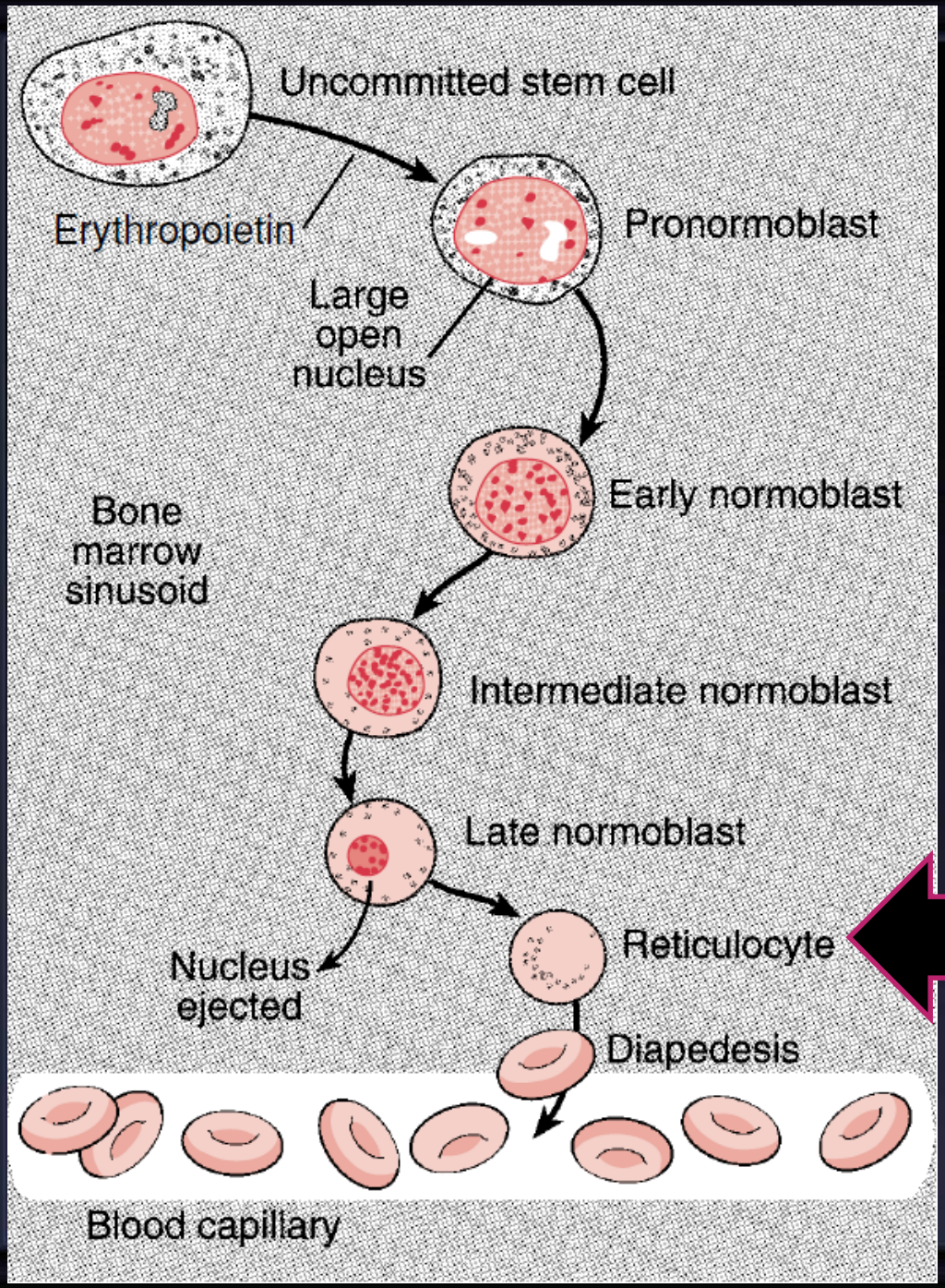
↓

Reticulocytes (Appears in Circulation)

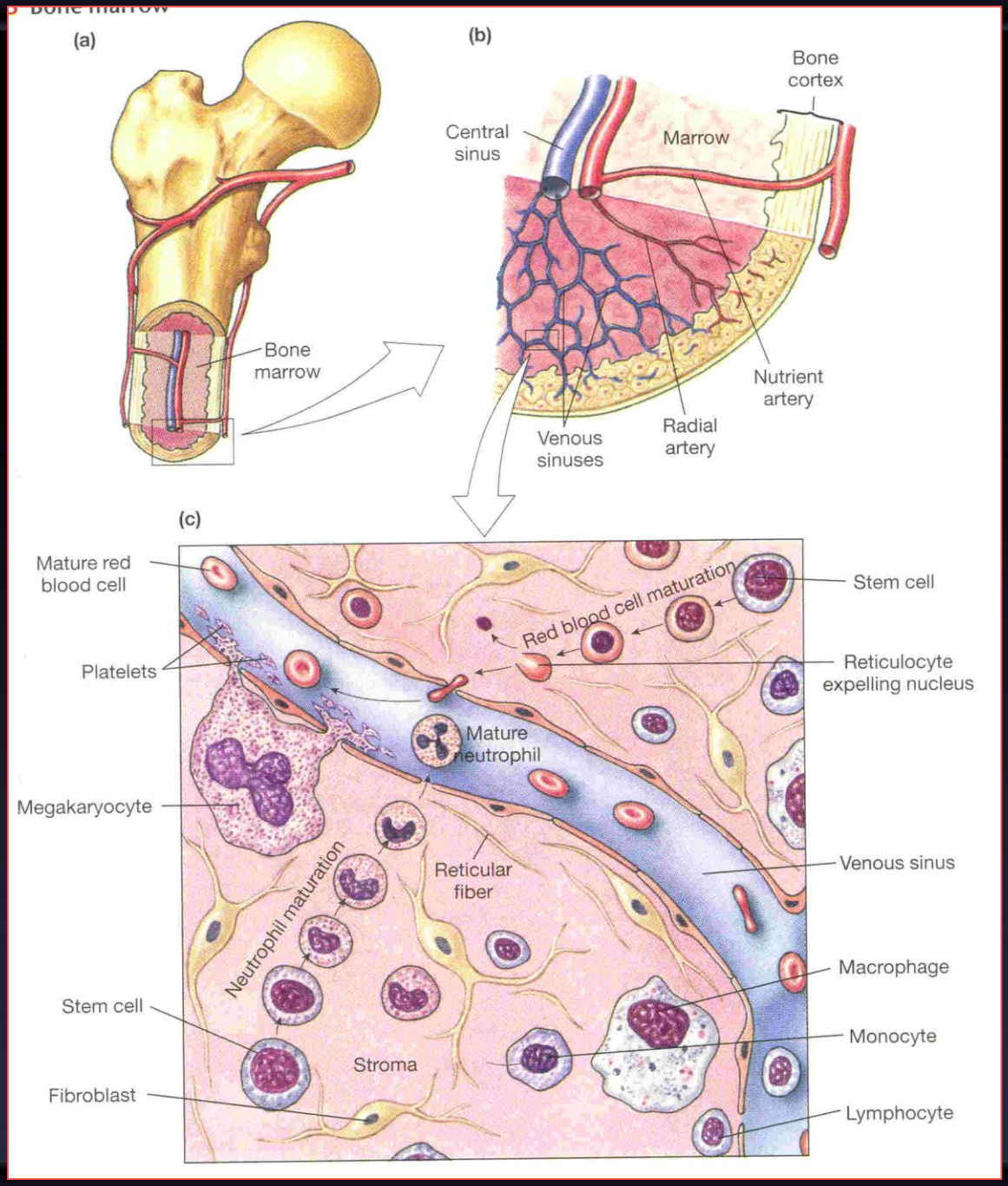
↓

Erythrocytes





Contains remnants of the Golgi apparatus, mitochondria, and a few other cytoplasmic organelles


(a)
Bone marrow
(b)
Bone cortex
Central sinus
Marrow
Nutrient artery
Radial artery
Venous sinuses
(c)
Mature red blood cell
Platelets
Megakaryocyte
Stem cell
Fibroblast
Red blood cell maturation
Stem cell
Reticulocyte expelling nucleus
Mature neutrophil
Reticular fiber
Neutrophil maturation
Venous sinus
Macrophage
Monocyte
Stroma
Lymphocyte

Features of the maturation process of RBC

1. Reduction in size
2. Disappearance of the nucleus
3. Acquisition of haemoglobin

Nutritional requirements for RBC formation

1. **Amino acid**
   - **HemoGlobin**
2. **Iron**
   - **HemoGlobin**
   - **Deficiency → small cells (microcytic anaemia)**

Nutritional requirements for RBC formation *cont.*

3. **Vitamins**
   - **Vit B12 and Folic acid**
     - **Synthesis of nucleoprotein DNA**
     - **Deficiency → macrocytes megaloblastic (large) anemia**
   - **Vit C**
     - **Iron absorption**

Nutritional requirements for RBC formation-*cont.*

- **Vit B6**
- **Riboflavin, nicotinic acid, pantothenic acid, biotin & thiamine (VB)**
  - **Deficiency → normochromic normocytic anaemia**
- **Vit E**
  - **RBC membrane integrity**
  - **Deficiency → hemolytic anaemia**

Nutritional requirements for RBC formation-*cont.*

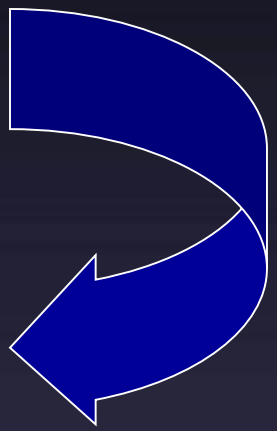
- **Essential elements**
  - Copper, Cobalt, zinc, manganese, nickel
  - Cobalt → ↑Erythropoietin

Vitamin B12 & Folic acid

- Important for cell division and maturation
- Deficiency of Vit. B12 > Red cells are abnormally large (macrocytes)
- Deficiency leads:
  - Macrocytic (megaloblastic) anaemia
- Dietary source: meat, milk, liver, fat, green vegetables

Vitamin B12

- Absorption of VB12 needs <u>intrinsic factor</u> secreted by parietal cells of stomach
- VB12 + intrinsic factor is absorbed in the terminal ileum
- Deficiency arise from
  - Inadequate intake
  - Deficient intrinsic factors
- Pernicious anaemia



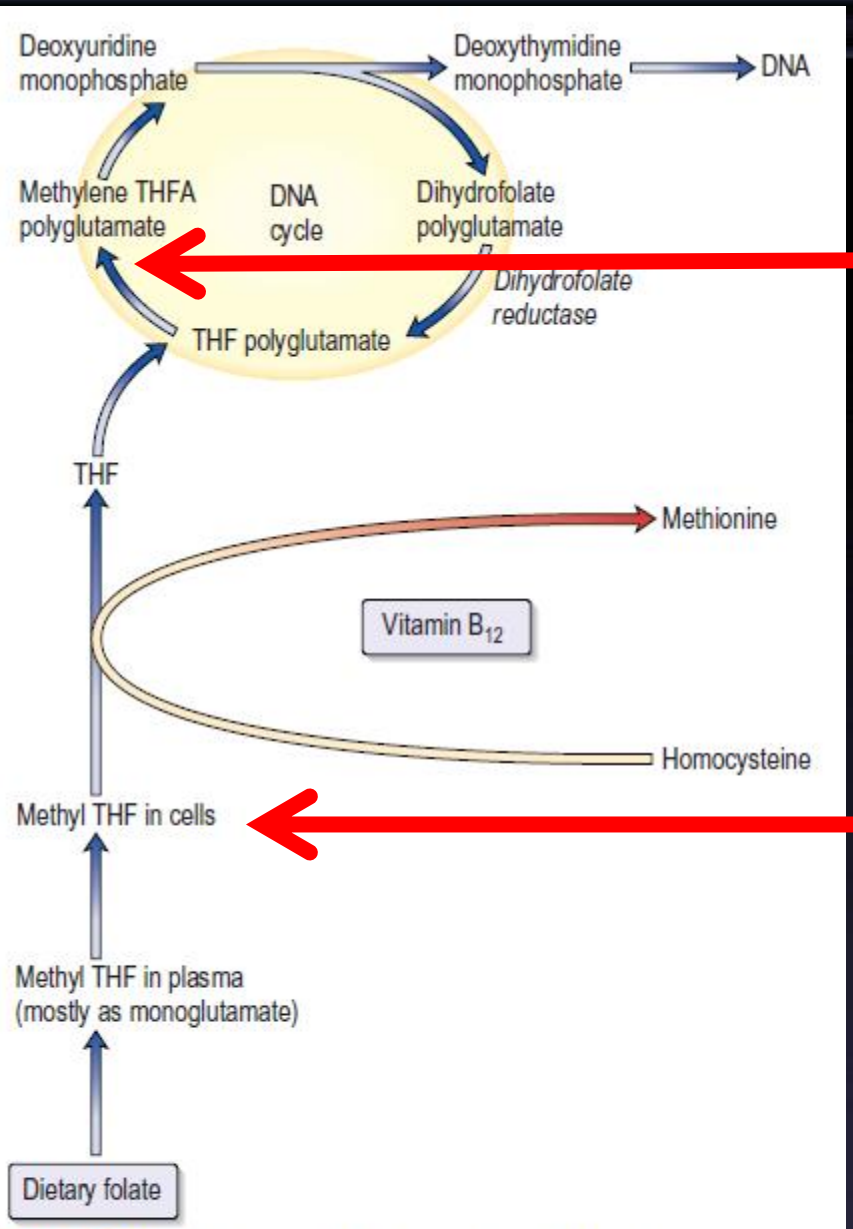


Fig. 8.12 **Biochemical basis of megaloblastic anaemia.** The metabolic relationship between vitamin B_{12} and folate and their role in DNA synthesis. THF, tetrahydrofolate.

Deficiency of folate reduces the supply of the coenzyme M-THFA

Deficiency of vitamin B12 also reduces supply of M-THFA by slowing the demethylation of methyl THF

Control of Erythropoiesis

Hypoxia, (blood loss)

↓

↓ Blood O2 levels

↓

Tissue (kidney) hypoxia

↓

↑ Production of erythropoietin

↓

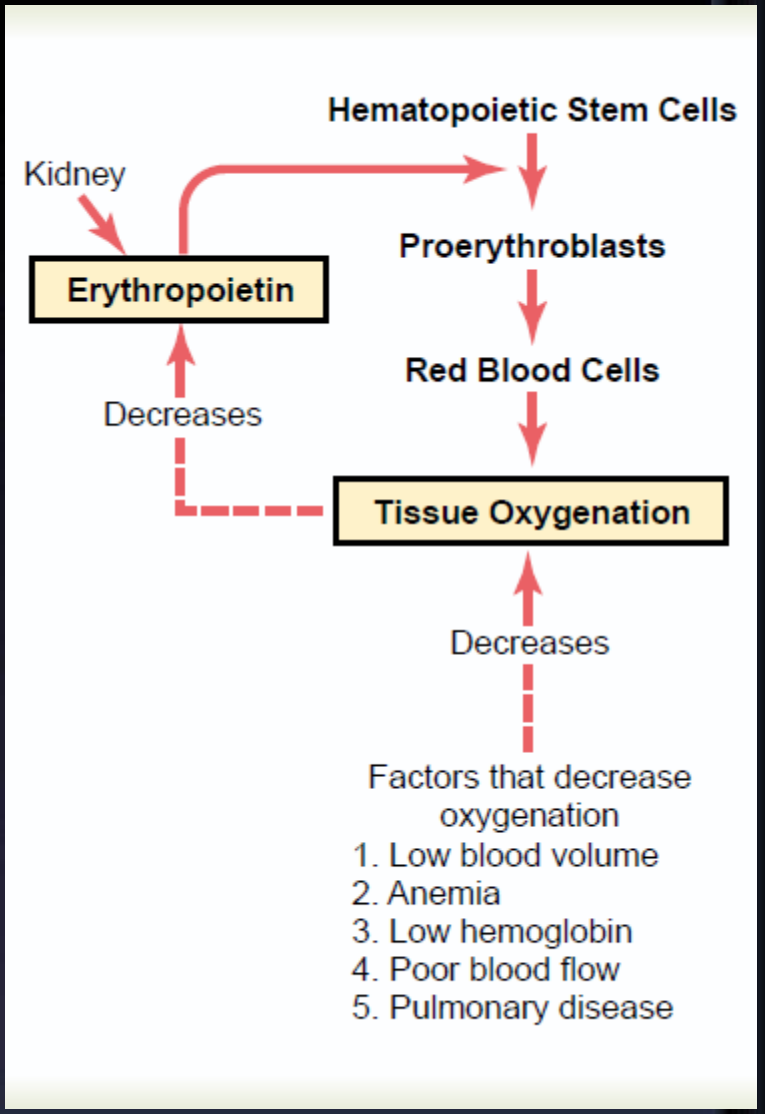
↑ plasma erythropoietin

↓

Stimulation of erythrocytes production

↓

↑ Erythrocyte production



Erythropoietin

- **Chemistry:** Glycoprotein
- **Site of Synthesis:** 90% from renal cortex 10% liver
- **Does not affect maturation process**
- Can be measured in plasma & urine
- High level of erythropoietin are seen in;
  - anemia
  - High altitude
  - Heart failure

- Stimulate the production of proerythroblasts from hematopoietic stem cells in the bone marrow
- once the proerythroblasts are formed, the erythropoietin causes these cells to pass more rapidly through the different erythroblastic stages and can increase it to perhaps 10 or more times normal

Control of erythropoiesis *cont.*

Other hormones

- Androgens, Thyroid, cortisol & growth hormones are essential for red cell formation

- Deficiencies of any one of these hormones results in anaemia

Iron metabolism

Total Iron in the body = 3-5g

1. Haemoglobin: ……. 65-75% (3g)
2. Stored iron…….. 15-30%
3. Muscle Hb (myoglobin) ….. 4%
4. Enzymes (cytochrome) …….. 1%
5. Plasma iron: (transferrin) …. 0.1%

(Serum ferritin → indication of the amount of iron stores)

Iron metabolism cont.

Iron intake:

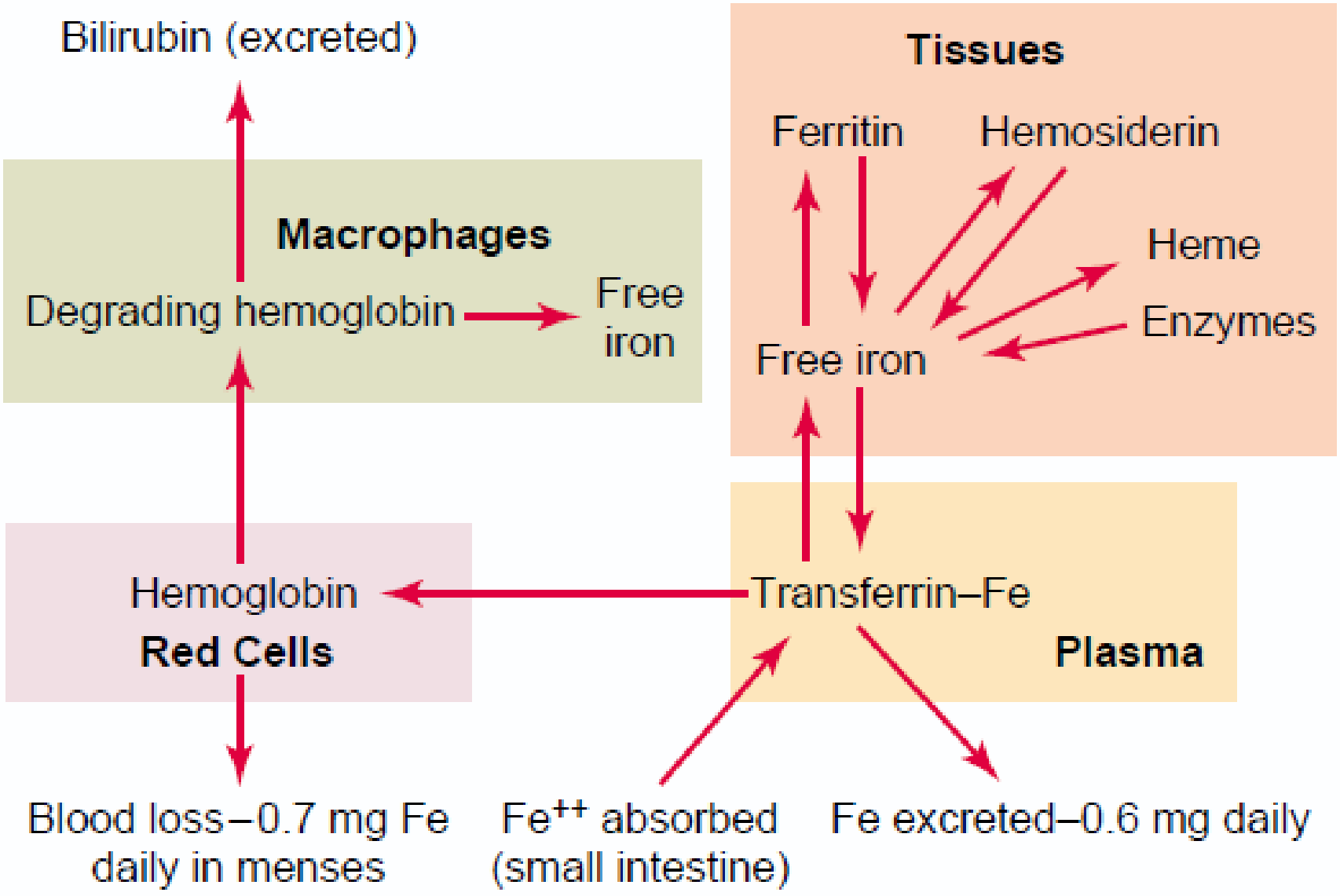
- **Diet provides 10-20 mg iron**
  - **Liver, beef, mutton, fish**
  - **Cereals, beans, lentils and**
  - **Green leafy vegetable**

Iron metabolism cont.

Iron absorption

- **Iron in food mostly in the form of Ferric (F^{+++}, oxidized)**
- **Better absorbed in reduced form Ferrous (F^{++})**
- **Iron in stomach is reduced by gastric acid, Vit. C.**
- **Maximum iron absorption occurs in the duodenum**

Iron transport and metabolism



Iron absorption *cont.*

- **Rate of iron absorption depend on:**
  - **Amount of iron stored (When all the apoferritin is saturated the rate of absorption of iron from intestine is markedly reduced)**
  - **Rate of erythropoiesis**

Iron in plasma:

- **Normally 30-40 saturated with Fe (plasma iron 100-130ug/100ml)**
- **When transferrin 100% saturated >> plasma iron: 300ug/100ml (Total Iron Binding Capacity is low)**

Iron absorption *cont.*

Iron in plasma:

- Transporting protein: TRANSFERRIN
- Normally 30-40 saturated with Fe

  (plasma iron 100-130ug/100ml)
- When transferrin 100% saturated >> plasma iron: 300ug/100ml

  (Total Iron Binding Capacity)

Iron stores

- **Sites: liver, spleen & bone marrow**
- **Storage forms: Ferritin (loose bond) and haemosiderin (firm bond)**

Apoferretin + iron = Ferritin

Ferritin + Ferritin = Haemosiderin

Hemosiderosis

Iron excretion and daily requirement

- **Iron losses**
  - **feces: unabsorbed, dead epithelial cells**
  - **bile and saliva.**
  - **Skin: cell, hair, nail, in sweat.**
  - **Urine**
  - **Menstruation, pregnancy and child birth**

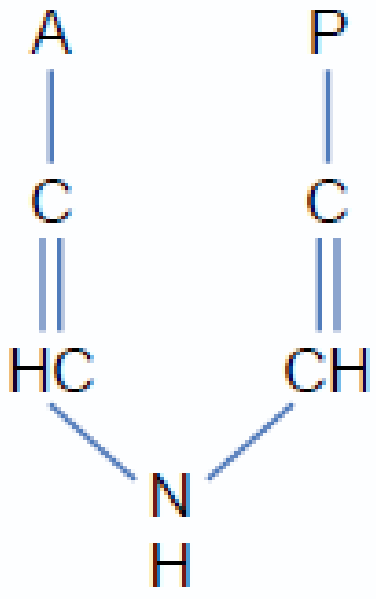
Destruction of Erythrocytes

- **At the end of RBC life span is 120 days:**
- **Cell membrane ruptures during passage in capillaries of the spleen, bone marrow & liver.**
- **Haemoglobin**
  - **Polypeptide → amino acids → amino acid pool**
  - **Heme:**
    - **Iron → recycled (reused)→ iron storage**
    - **porphryn → biliverdin → bilirubin (bile)**

HAEMOGLOBIN

- 14g/dl---18g/dl
- Protein (Globin) + Heme
- Each heme consist of:
  porpharin ring + iron
- The protein (Globin) consist of:
  4 polypeptide chains:
  2 α and 2 β chains

HAEMOGLOBIN SYNTHESIS

I. 2 succinyl-CoA + 2 glycine → 

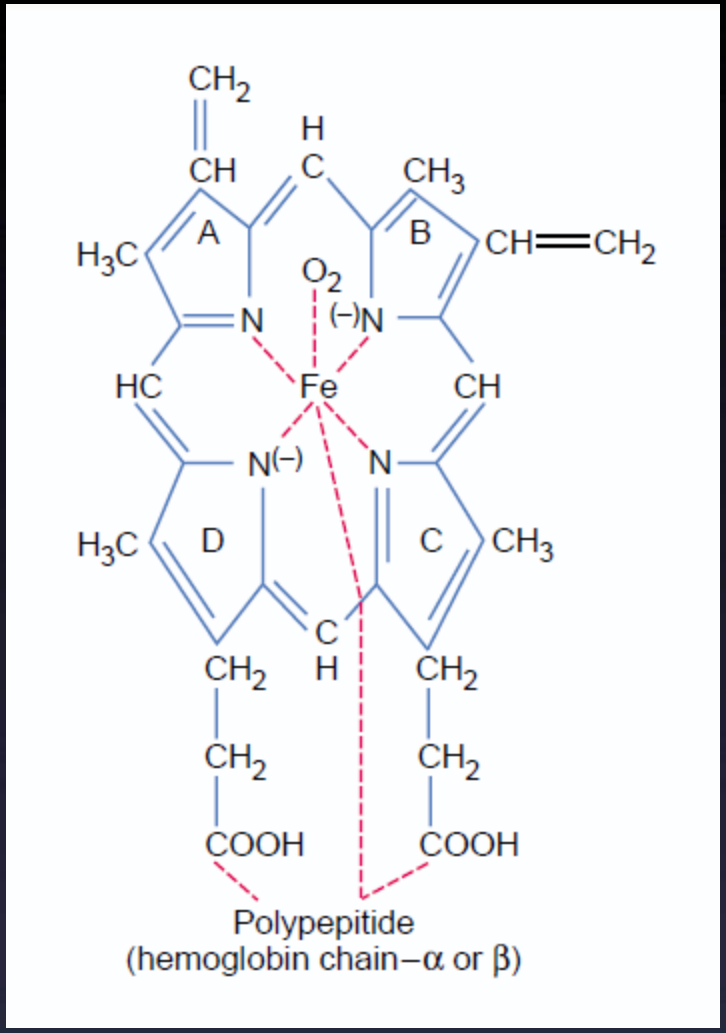
II. 4 pyrrole → protoporphyrin IX

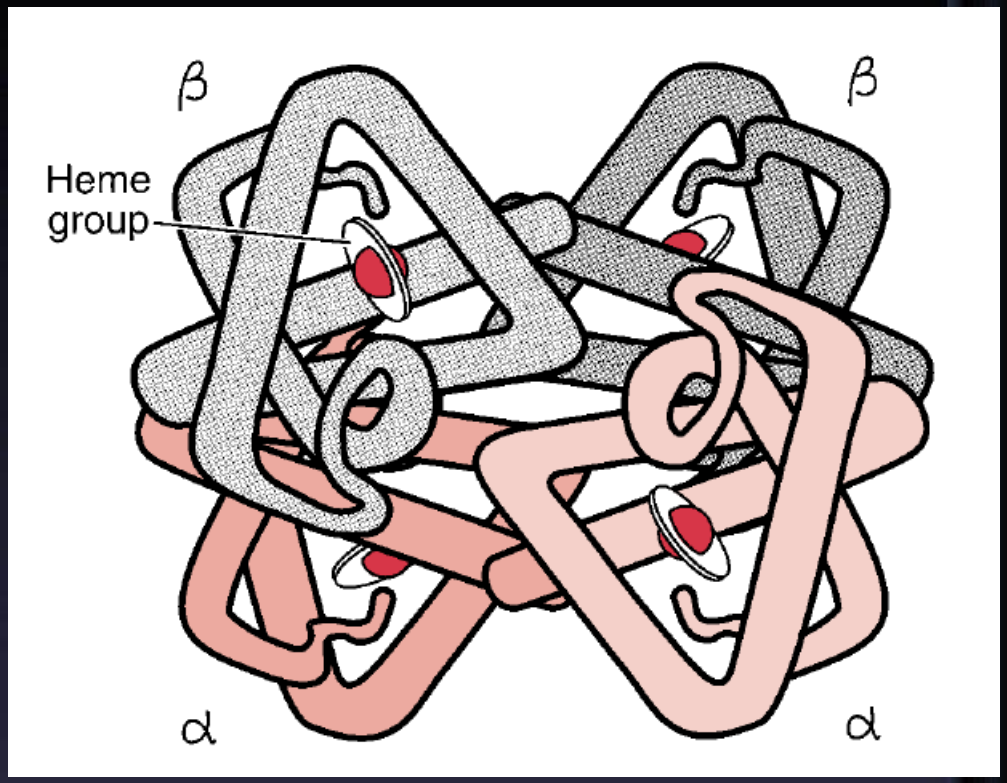
III. protoporphyrin IX + Fe^{++} → heme

IV. heme + polypeptide → hemoglobin chain (α or β)

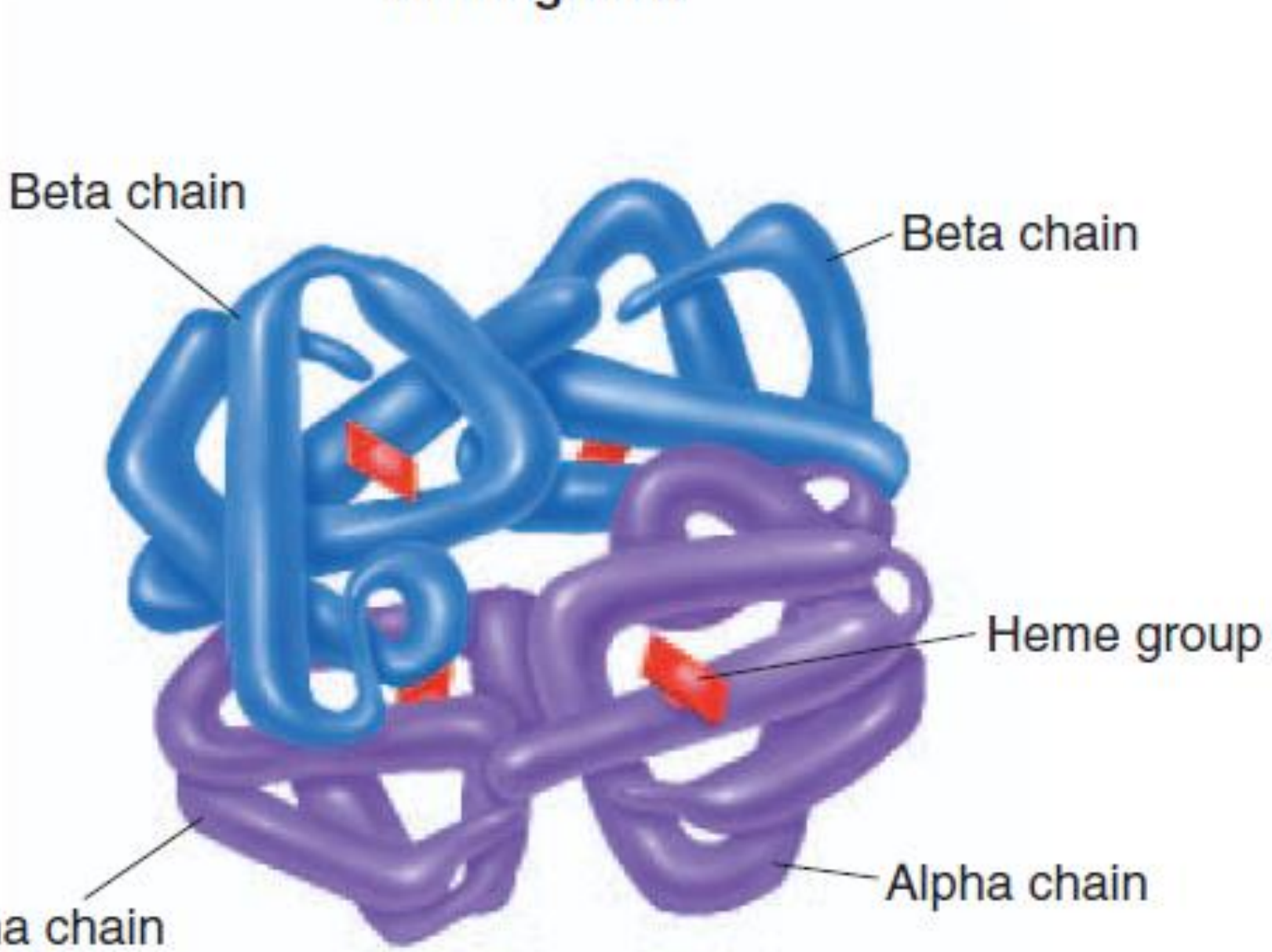
V. 2 α chains + 2 β chains → hemoglobin A

Basic structure of hemoglobin molecule, showing one of the 4 heme chains that bind together to form the hemoglobin molecule.





Hemoglobin



a)

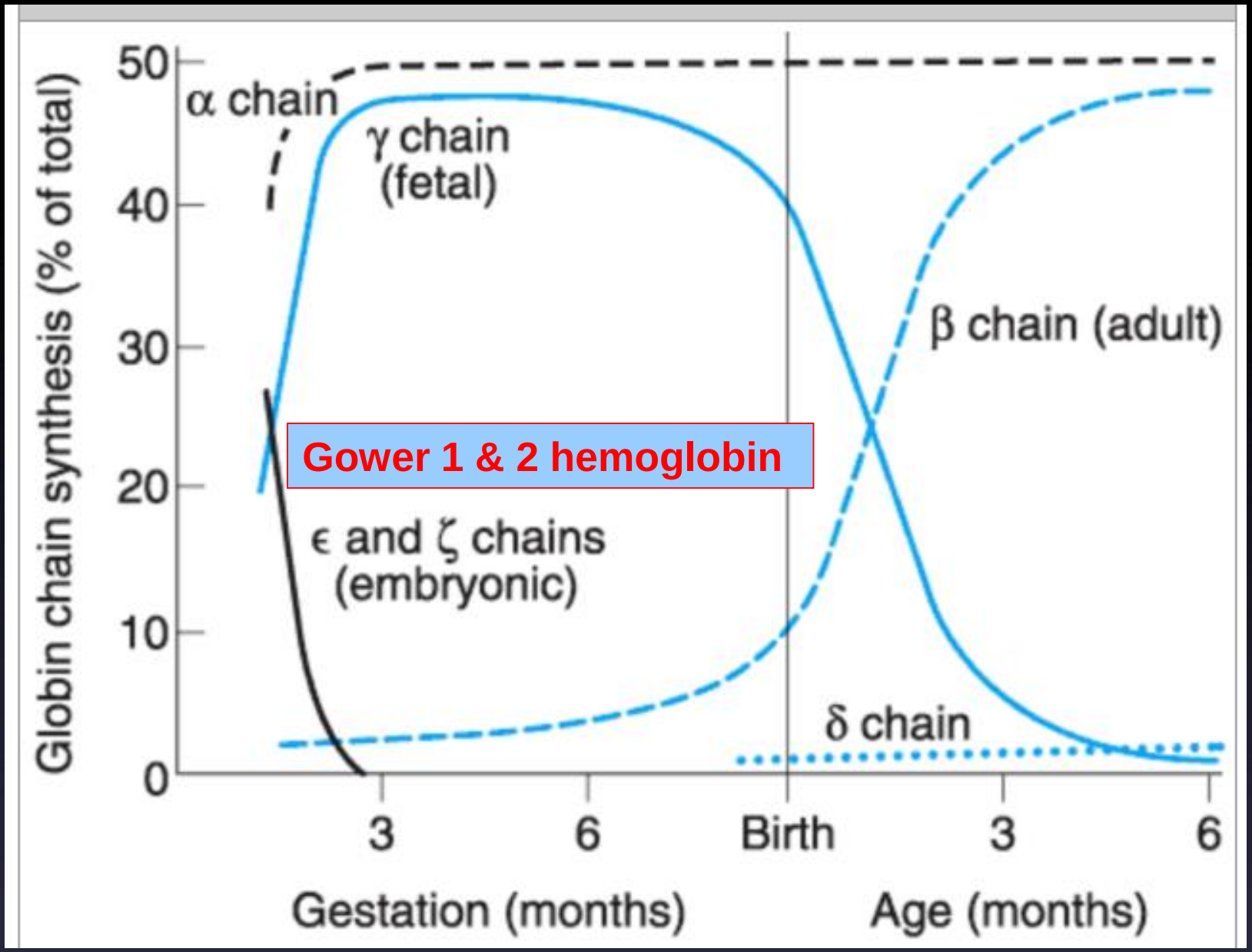
TYPES OF NORMAL HEMAGLOBIN

- HbA: 98% of adult Hb its polypeptide chains (2α & 2β)
- HbA2: 2.5% of adult Hb (2α & 2δ)
- HbF: 80-90% of fetal Hb at birth (2α & 2γ)

Abnormality in the polypeptide chain α & β results in abnormal Hb (hemoglobinopathies) e.g thalassemias, sickle cell

Functions of Hb

- Carriage of O2 and CO2
- Buffer
- (Bind CO Smokers)


Globin chain synthesis (% of total)
50
40
30
20
10
0
α chain
γ chain
(fetal)
β chain (adult)
Gower 1 & 2 hemoglobin
ϵ and ζ chains
(embryonic)
δ chain
3
6
Birth
3
6
Gestation (months)
Age (months)

Jaundice

Yellow coloration of skin, sclera

- **Deposition of bilrubin in tissues**
- **If Bilrubin level in blood > 2 mg/ ml > jaundice**

- **Causes of Jaundice**
  - **Excess breakdown of RBC (hemolysis)**
  - **Liver damage**
  - **Bile obstruction: stone, tumor**

Clinical Correlate

What is Anaemia?

Anemia is reduced amount/concentration of Haemoglobin or RBC count in blood less than the amount appropriate for that age, sex, race and physiological status.

Normal ranges of Hb

Men: Hb 13.5 -17.5 g/dL

Women: Hb 11.5-16 g/dL

Infants : Hb 14 – 20 g/dL

Symptoms & Signs: Tiredness, Fatigue, Dyspnea (shortness of breathing), pallor, tachycardia

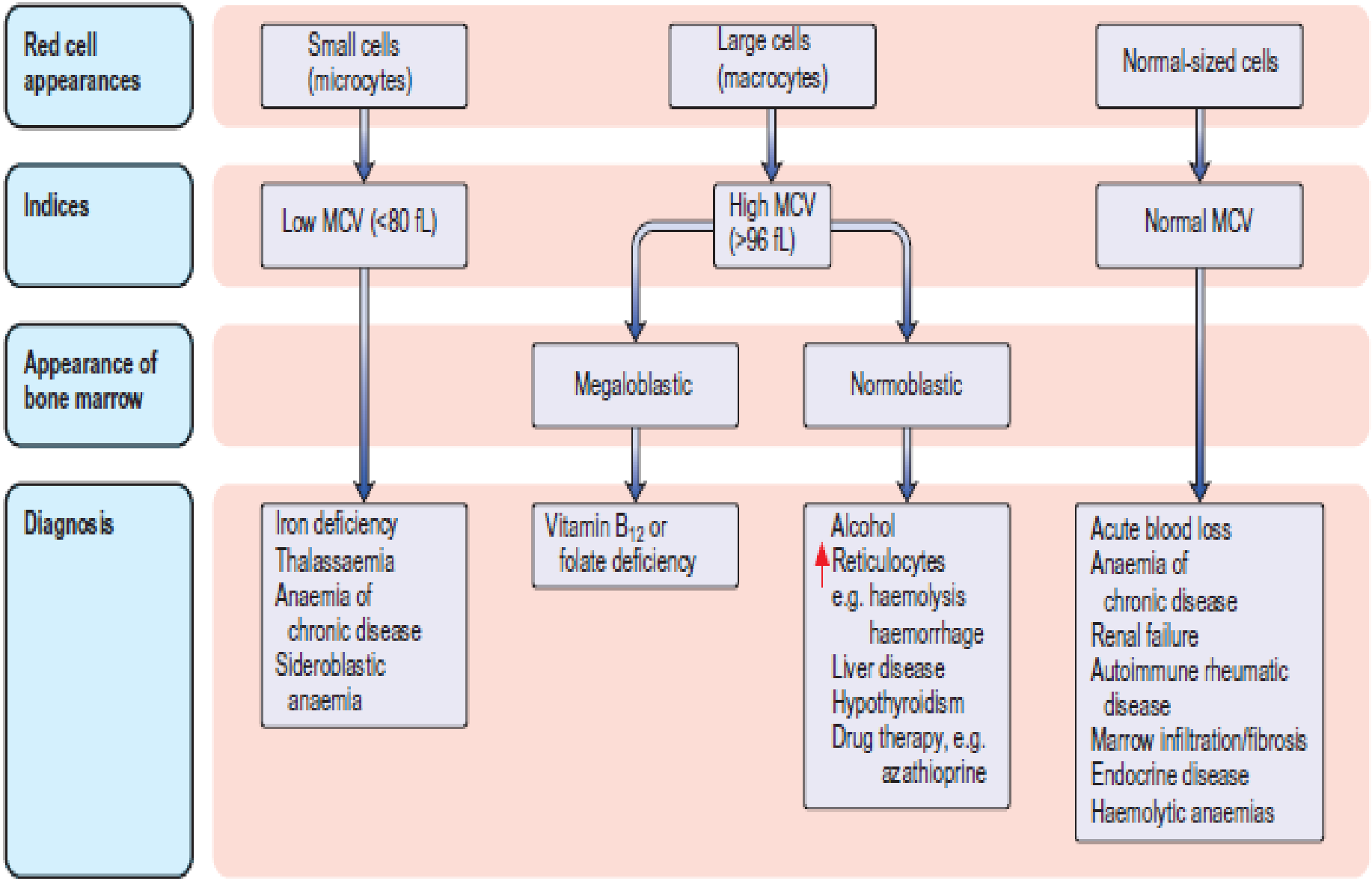
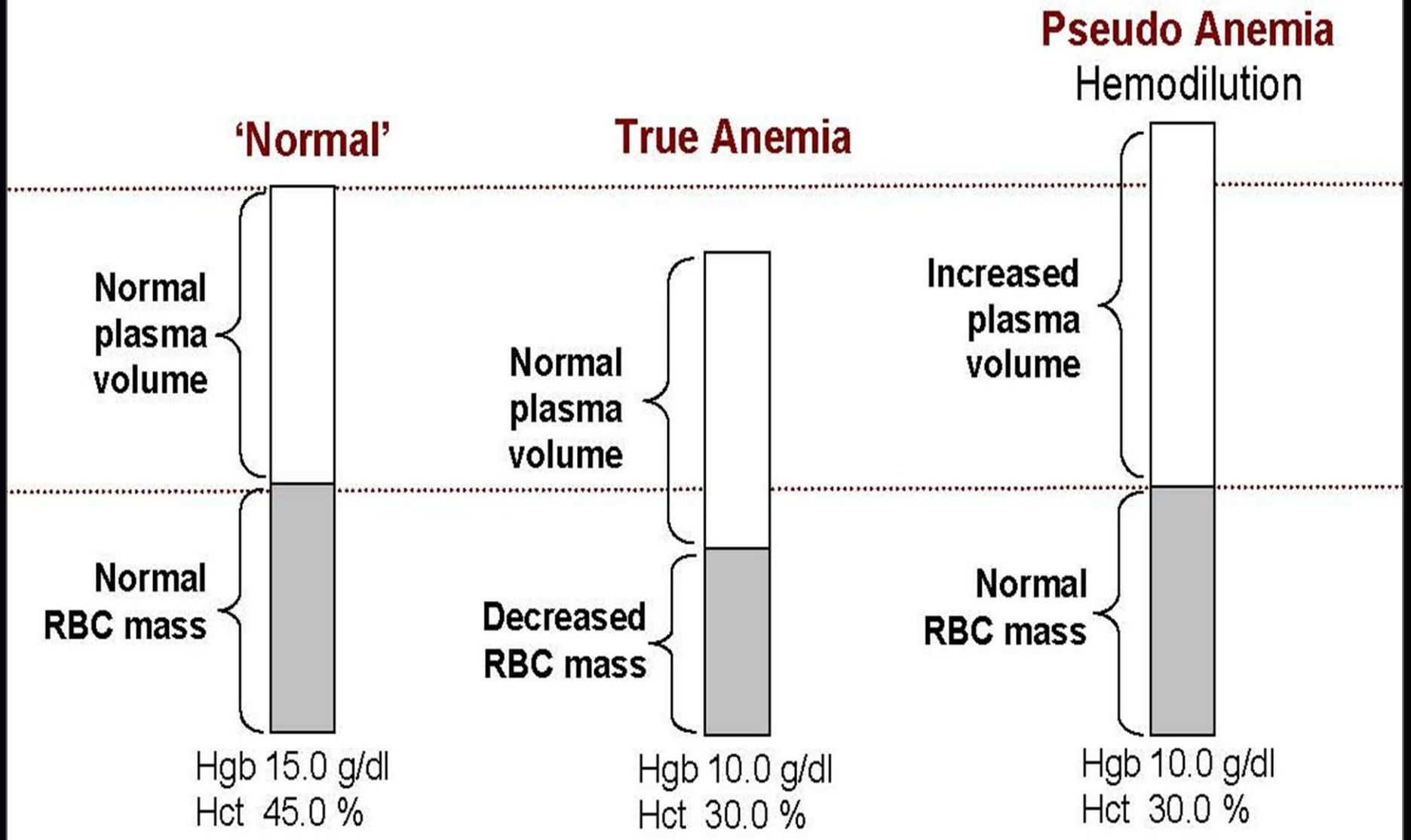
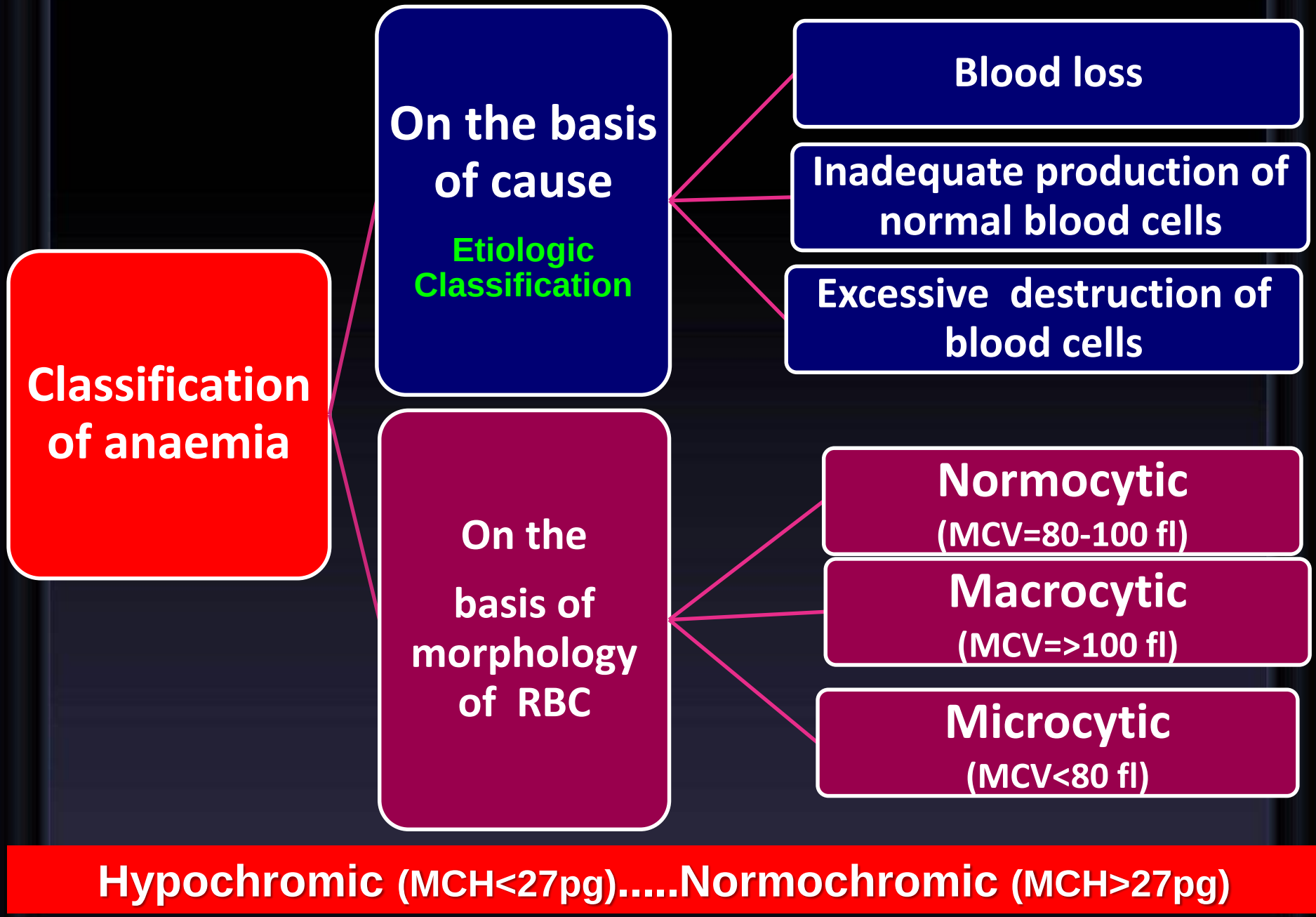


Fig. 8.7 **Classification of anaemia.** MCV, mean corpuscular volume.

RBC Mass and Plasma Volume Relationship




Classification of anaemia
On the basis of cause
Etiologic Classification
Blood loss
Inadequate production of normal blood cells
Excessive destruction of blood cells
On the basis of morphology of RBC
Normocytic (MCV=80-100 fl)
Macrocytic (MCV=>100 fl)
Microcytic (MCV<80 fl)
Hypochromic (MCH<27pg).....Normochromic (MCH>27pg)

Causes of anaemia

1. Blood Loss

- acute→ accident
- Chronic → ulcer, worms (Parasitic Infections)

2. Decrease RBC production

- Nutritional causes
  - Iron → microcytic anaemia
  - VB12 & Folic acid → megaloblastic anaemia
- Bone marrow destruction by cancer, radiation, drugs → Aplastic anaemia.

3. Haemolytic → excessive destruction

- Abnormal Hb (sickle cells)
- Incompatible blood transfusion

Microcytic Hypochromic Anemia

MCV↓, MCH ↓, MCHC↓

1. Fe deficiency anemia : Chronic blood loss, Inadequate diet, Malabsorption, Increased demand, etc.
2. Abnormal globin synthesis : Thalassemia with or without Hemoglobinopathies
3. Abnormal porphyrin and heme synthesis : Pyridoxine responsive anemia, etc.

Macrocytic Anemia

MCV↑, MCH↑, MCHC=May be normal

- Vit. B12 deficiency : Pernicious anemia
- Folic acid deficiency : Nutritional megaloblastic anemias, Sprue, Other malabsorption syndromes

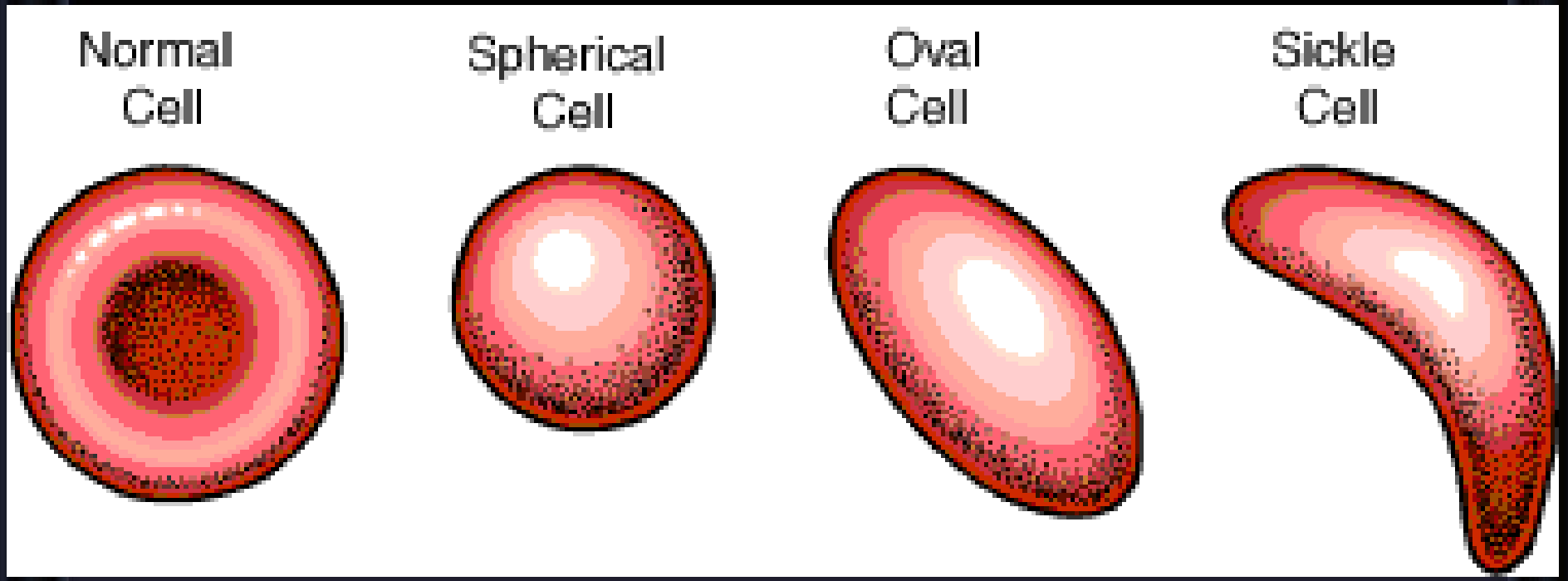
Normocytic Normochromic Anemia

MCV=Normal, MCHC=May be Normal

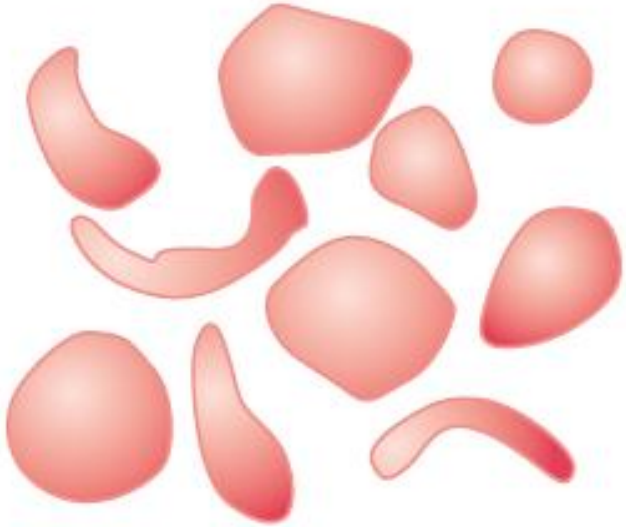
1. Blood loss
2. Hemolytic anemia : depend on each cause
3. Hypoplastic marrow : Aplastic anemia, RBC aplasia
4. Infiltrate BM : Leukemia, Multiple myeloma, Myelofibrosis, etc.
5. Endocrine diseases: Hypothyroidism, Adrenal insufficiency, etc.
6. Kidney disease / Liver disease / Cirrhosis

Polycythemia

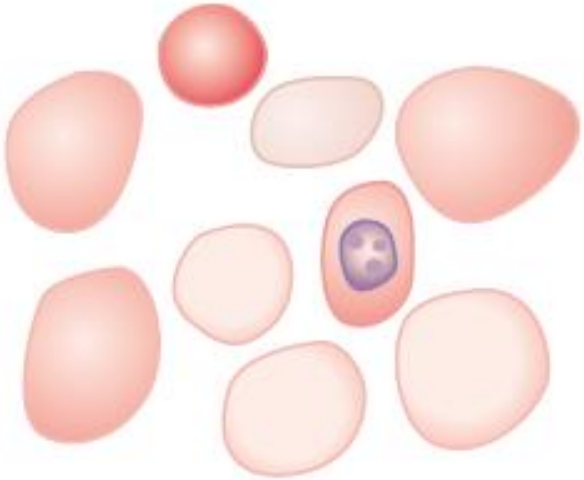
- Increased number of RBC
- Types:
  - True or absolute
    - Primary (polycythemia rubra vera): uncontrolled RBC production
    - Secondary to hypoxia: high altitude, chronic respiratory or cardiac disease
  - Relative
    - Haemoconcentration:
      - » loss of body fluid in vomiting, diarrhea, sweating


Normal Cell
Spherical Cell
Oval Cell
Sickle Cell

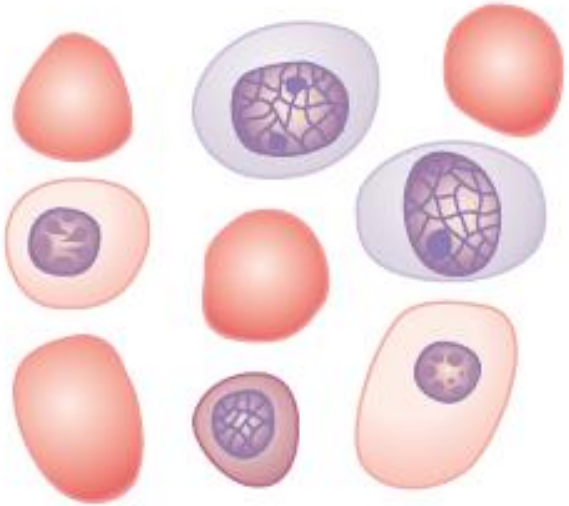
Microcytic,
hypochromic anemia



Sickle cell anemia



Megaloblastic anemia



Erythroblastosis fetalis