MEDICINE

432 Team

29 Anemia

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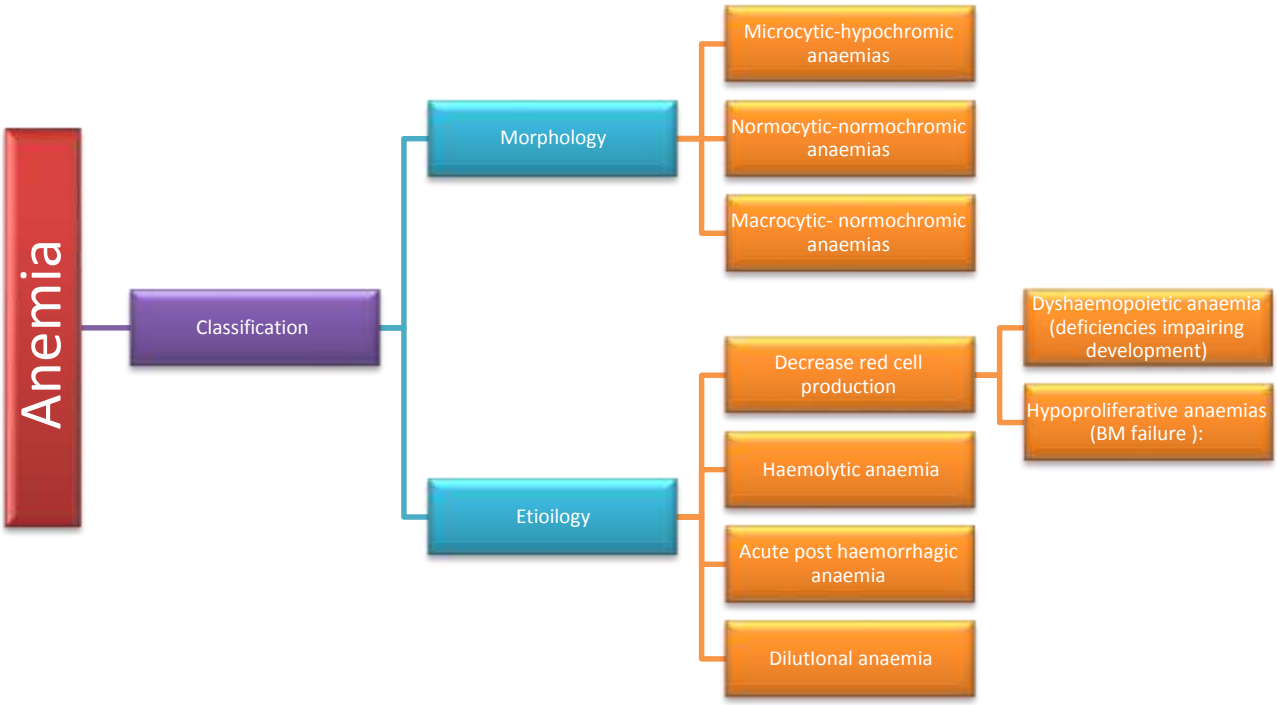
COLOR GUIDE: • Females' Notes • Males' Notes • Important • Additional

Objectives: (not given)

General Outline



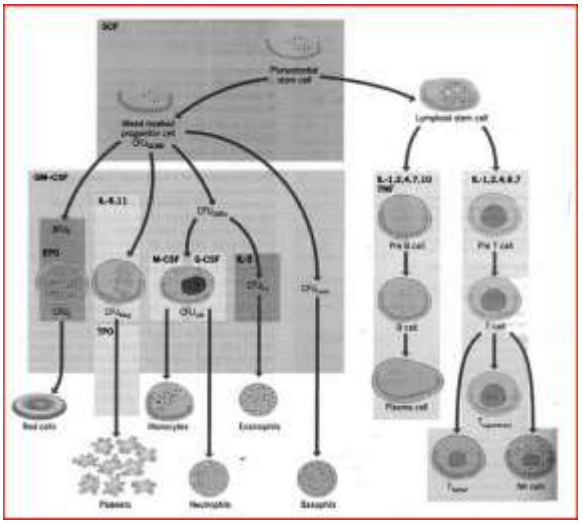
Introduction:

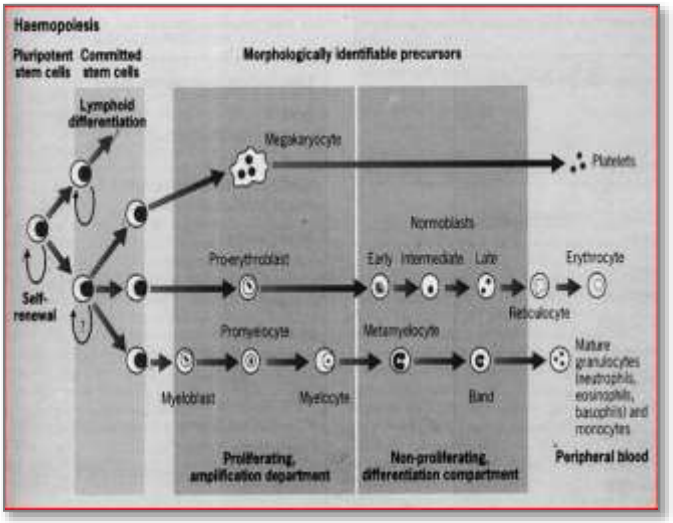
Blood formation:

1. Intrauterine life:
- Blood cells are formed in the liver and spleen up to the fifth months.
- After 5th month: bone marrow shares in the formation of these cells.
2. After birth:
    - Formation of these cells will be restricted to the bone morrow.
3. Adulthood

   The active bone morrow will be restricted to axial skeleton: flat bones, vertebrae, ribs, sternum and ilia.
    - Some extension to the proximal ends of long bone mainly femur.

- Extramedullary haemopoiesis: When demand for blood formation is increased the active red BM extends into the shafts of the long bones.
- The spleen and liver will regain their ability to produce blood elements when bone morrow is affected by some diseases.





Blood Constituents:

- Organelles (formed elements):
    - RBCs
    - WBCs
    - Thrombocytes
- Fluid components: plasma in which the above elements are suspended and contain fibrinogen.

Erythrocytopoiesis

Normal erythrocytopoiesis depends on:

1. Healthy BM: normal stem cells and architecture.
2. Regulatory hormones: GPO, Androgen thyroxin, cortisol and ACT.
3. Nutritional elements: Protein (high biological value),
4. Minerals (Iron, copper, zinc, selenium)
5. Vitamins (B, Folic acid, vit. C)

Anaemia: Reduction of O2 carrying capacity of the blood with inadequate O2 supply to tissues.

- Diagnosed when there is a reduction of (RBCs number) below the reference range for the age and sex of an individual.
- More accurately is defined as reduction in the red cell mass

Classification:

A. By etiology:

1. Decreased red cell production
   - Dyshaemopoietic anaemia
     a) Mineral deficiency: iron, zinc, selenium, cupper
     b) Vitamin deficiency: B12, folic acid, Vit C & pyridoxine
     c) Hormonal deficiency: anemia of renal diseases, pituitary, thyroid or suprarenal deficiency.
     d) Protein deficiency: high class
   - Hypoproliferative anaemia
     a) Aplastic anaemia.
     b) Myelophthisic anaemia (bone-marrow infiltration replaces heamopoietic cells leading to anemia).
     c) Anemia of chronic diseases
2. Haemolytic anaemia:
   - Short life-span of RBCs
3. Acute post haemorrhagic anaemia:
   - Loss of RBCs
4. Mixed anaemia.
   - eg. Megalobastosis associated with haemolysis

5. Dilutional anaemia: (relative anemia: volume is high but cell mass remains the same)
   - Pregnancy
   - Oliguric RF
   - Volume-overload

B. By Morphology

1. Microcytic-hypochromic anaemias:
   - Iron deficiency anaemia.
   - Thalassaemia.
   - Anaemia of chronic disease.
   - Sideroblastic anaemia:
     a) Hereditary
     b) Chronic lead poisoning.
2. Normocytic-normochromic anaemias:
   - Anaemia of chronic diseases.
   - Acute post –haemorrhagic anaemia.
   - Hemolytic anaemia.
   - Aplastic anaemia.
   - Myelophthisic anaemia.
3. Macrocytic- normochromic anaemias:
   - Megaloblastic anaemia.
   - Marked reticulocytosis.
   - Myelodysplastic syndromes.
   - Myxoedema.
   - Acquired sideroblastic anaemia.

Pathophysiology

1. **Tissue Hypoxia**
   - Impaired functions of the tissues, the degree of impairment depends on the degree of dependency on O2, so CVS, CNS and skeletal muscles are much affected.
2. **Compensatory mechanisms**
   - Increased COP. **(explaining high output cardiac failure)**
   - Increased O2 delivery from HB to the tissue.
   - Increased erythropoietin production with stimulation of erythropoiesis. **(Explaining the accompanying reticulocytosis)**
   - Increased plasma volume.
   - Redistribution of the blood from less to more vital organs.

3. **Rate of blood loss:**
    - The rapid the rate of blood loss, the more the severe symptoms will occur especially in elderly. While the slowly falling HB allows for hemodynamic compensation with less symptom.
4. **Factors related to a specific cause.**

Symptoms

- Neurological:
    - ✓ Dizziness, fainting, lack of concentration
    - ✓ Blurred or diminished vision
    - ✓ Headache, tinnitus
    - ✓ Paraesthesia in the fingers and toes
    - ✓ Insomnia, irritability.
- CVS:
    - Angina, dyspnea, palpitation and intermittent claudication by exertion
    - Heart Failure in severe cases or presence of other organic cardiac disease, it is high COP failure.
- Musculo skeletal:
    - Easy fatigability.
    - Tiredness and lassitude.
- GIT:
    - Dyspepsia and anorexia
- Genital
    - Loss of libido & impotence
    - Menstrual abnormalities like amenorrhea.
- May be polyuria.

Hematocrit and Symptoms	
Hematocrit	**Expected symptoms**
>30%–35%	None
25%–30%	Dyspnea (worse on exertion), fatigue
20%–25%	Lightheadedness, angina
Under 20%–25%	Syncope, chest pain

Signs

- ✓ Pallor of the skin and mucous membranes
  - The color of the skin is unreliable because it depends upon the degree of skin pigmentation and the amount of fluid in the subcutaneous tissues.
  - Best examined: palmer creases, nail bed, and mucous membranes.
- ✓ Peripheral edema:
  - Slight edema of the legs probably due to increase in the capillary permeability secondary to hypoxia.
- ✓ High COP failure.
- ✓ Fever:
  - Mild fever may occur in severe anemia but other causes should be excluded.
- ✓ Fundal changes:
  - Retinal hemorrhages of the flame shape type, exudates and rarely papilledema.
- ✓ The cardiovascular system:
  - Increased velocity with decreased viscosity of the blood in addition to capillary dilation can cause the following:
    1. Tachycardia, bounding pulse
    2. Cardiac examination: Loud HS, S3 over mitral or tricuspid area
    3. Haemic murmur: ejection systolic, hearted all over the precordium
    4. Raised jugular venous pressure.
- ✓ Proteinuria and impairment of the concentrating power of kidneys due to anoxia of renal tubules.

Now that we established some of the basic principles regarding anemia, let's talk diseases. (Remember that just saying anemia is not establishing a diagnosis)

Microcytic Anemia

How to Answer "What Is the Most Likely Diagnosis?" for Anemia	
Feature in the history	**What is the most likely diagnosis?**
Blood loss (GI bleeding)	Iron deficiency
Menstruation	Iron deficiency
Cancer or chronic infection	Chronic disease
Rheumatoid arthritis	Chronic disease
Alcoholic	Sideroblastic
Asymptomatic	Thalassemia

A.IRON DEFIIENCY ANEMIA

Sources, absorption and metabolism:

- ✓ Red meat and liver, bread, eggs and green vegetables, mainly in ferric form.
- ✓ Daily minimum requirement 10-12mg of which about 1mg is absorbed.
- ✓ In the stomach the iron is released from its complex form and is reduced to ferrous form (action of gastrin and HCl).
- ✓ Iron absorption takes place in the duodenum and proximal jejunum.
- ✓ Iron absorption is under regulatory system (Apoferritin-Transferrin system) present in the intestinal mucosa and regulates absorption of the iron according to body requirement.

➢ Factors enhancing iron absorption
- ✓ Pregnancy
- ✓ Iron deficiency anemia
- ✓ Increased erythropoiesis
- ✓ Vit. C.

➢ Factors decrease iron absorption:
- ✓ Excess phosphate, tannates, phytate in diet
- ✓ Iron overload haemochromatosis
- ✓ Decreased erythropoies eg a plastic anaemia
- ✓ Malabsorption syndrome
- ✓ Decreased HCl atrophic gastritis.

➢ Conditioned deficiency (normal demand and intake but there are defective absorption and utilization) Not common:
- Ferric form cause.

- Decreased HCL
- Iron binder: phosphate, phytate, tannates
- Malobsorption syndrome.

➢ Relative deficiency: (increased requirement) common cause.

- Menstruating females.
- Pregnancy, labor.
- Growing children.
- Convalescence from disease.

✓ Chronic blood loss: the commonest cause

➢ Frank blood loss:

- Menorrhagia
- Repeated GI bleeding, hematemesis, epistaxis, and hematuria.
- Bleeding tendencies
- Repeated blood donation

➢ Occult blood loss via GIT:

- Anckylstoma & schistomiasis
- Oozing OV, PU.
- Neoplasm.
- Inflammatory bowel disease ulcerative colitis
- TB enteritis.

Clinical features

➢ General manifestations of anemia

➢ Angular stomatitis.

➢ Atrophic glossitis (red, glazed, smooth tongue)

➢ Cheilosis (crusting of angle of mouth)

➢ Atrophy of the gastric mucosa

➢ Small splenomegaly

➢ Brittle nails, thinning and ridging and loss of luster.

➢ Koilonychia (spoon-shaped nails) in severe cases

➢ Brittle hair

➢ Features of special types:

a. Plummer-Vinson –syndrome:
   - Common in middle aged female
   - Postcricoid esophageal web causes dysphagia

b. Ankylostoma infestation:
   - Perverted appetite: (pica) eating mud, stones and chalk.
   - Epigastric pain (DD= D. ulcer), altering bowel habits
   - Endemic parasites (trapezoid face)

Note:

Fe KAP:
***Fe** (iron)/ **F**atigue*
***E**xercise intolerance*
***K**oilonychia*
***A**ngular cheilosis*
***P**ica/Pallor*

- The symptoms are mild and variable.
- Investigations:
  - Stool: Ova.
  - CBC: eosinophilia.

Investigations

Unique Features and Diagnoses of Iron Studies	
Unique feature	**Diagnosis**
Low ferritin	Iron deficiency
High iron	Sideroblastic anemia
Normal iron studies	Thalassemia

- CBC:
  - The red cells are microcytic, (MCV<80f1) and hypochromic (MCH < 27pg) anisocytosis and poikilocytosis (variation in size and shape, respectively). **With red cell distribution width RDW>15**
  - Eosinophilia is present in cases of ankylostoma infestation.
- Bone Marrow:
  - Absent iron store
  - Normoplastic hyperplasia (because it is trying to compensate)
  - Decreased Hb in maturely erythroblasts.
- Serum iron study
  - Decreased serum iron (n=70-170 mg %).
  - Increased total iron binding capacity (TIBC). (N=250-450mg %).
  - Decreased serum ferritin levels.
    - Reflect iron stores.
    - The normal values are 30-300 mg/L in males and 15-200 mg/L in females and investigation of gastrointestinal tract are often required.
    - Falsely raised value in cases of acute phase reactant e.g malignancy.
  - Decreased transferrin saturation (n=25-50%).
- Investigations for specific causes
  - ✓ Stool analysis:
    - Occult blood, ankylstoma & Bilharizasis.
  - ✓ GI cause:
    - Imaging: barium swallow, meal or enema.
    - Endoscopic studies: upper and lower.

- Achlorhydria: histamine test.

✓ Hemostatic profile.

> ***Case:***
>
> *A 47 year old man presents with iron deficiency anemia.*
> *Think: colon cancer unless proven otherwise (up to 20%).*
> *Next step: colonoscopy and endoscopy.*

Treatment: The aim of the treatment is to correct the anemia and build up iron stores

I. Correction of underlying cause if possible. (E.g. treat cancer…)

II. Iron replacement

- Oral iron
  - ✓ 200 mg anhydrous ferrous sulphate three times daily.
  - ✓ The response is a rise in Hb concentration of 1 gm/dl per week
  - ✓ Side effects nausea, abdominal pain, diarrhea or constipation.
- Parenteral iron:
  - ✓ Indications:
    - General intolerance of oral preparation even at low dose.
    - Severe malabsorption and those who have gastrointestional diseases.
    - Rapid iron loss.
  - ✓ Preparation
    a. Iron sucrose (ferrosac – venofer)
      - Good safety and efficacy profile.
      - Given IM or slowly IV infusion.
      - Amp: 5ml = 100 mg.

    b. Iron dextran (cosmofer - imferon)
      - Good efficacy but more side effects.
      - Giving IM, or IV infusion.
      - Amp: 2 ml = 100mg.
      - Side effects:
      - Local pain, staining inflammation, abscess formation
      - General: hypersensitivity reaction, fever, rigor, hypotension
  - ✓ N.B: Test dose is required prior to the use of iron therapy.

III. Transfusion therapy:

- Packed RBCs.
- Indications:
  - Severe (Hb<7gm/dl) and symptomatizing anemia)
  - Complicated anemia: HF

B. Anemia of chronic disease

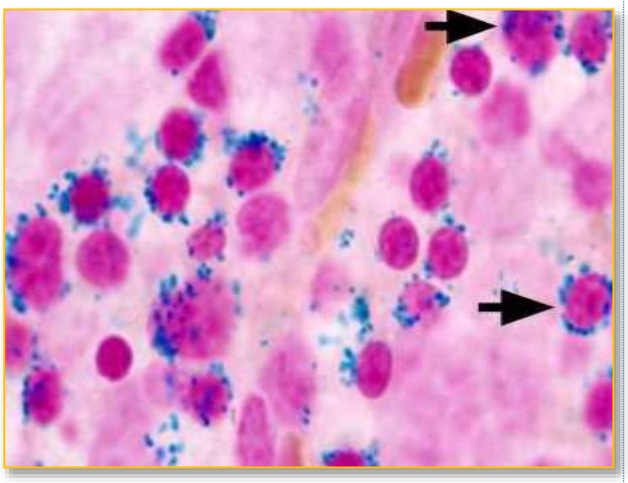
- Type of anemia may be microcytic hypochromic or more common normocytic normochromic.
- Endocrinal disorders: hypothyroidism, hypocorticalism, hypogonadism in male.
- Inflammatory diseases: collagenosis, Chron's disease.
- Rheumatological diseases.
- Chronic infection: as TB, sarcoidosis, oestomylitis.
- Neoplastic diseases.
- Malabsorption.
- Organ failure, liver and kidney

Laboratory:

- Decreased serum iron and TIBC.
- Normal or raised serum ferritin
- Normal BM iron.

C. Sideroblastic anemia

It is a refractory microcytic hypochromic anemia characterized by the presence of sideroblasts in the bone marrow. Sideroblasts are Erythroblasts inside, which iron accumulates into the mitochondria of erythroblasts owing to disordered haem synthesis. A ring of iron granules is formed around the nucleus.



Types:

- Inherited: X-linked disease transmitted by females.
- Acquired:
  1. Primary: one of the myelodysplastic syndromes
  2. Secondary:
     - ✓ Other types of myelodysplasia
     - ✓ Myeloproliferative disorders
     - ✓ Myeloid leukemia
     - ✓ Drugs as isoniazid, alcohol, lead.

	Ferritin	Iron	TIBC	Transferrin saturation	Soluble transferrin receptor
Iron deficiency anaemia	↓	↓	↑	↓	↑
Anaemia of chronic disease	↑/Normal	↓	↓	↓	↓/Normal

(TIBC = total iron binding capacity)

Macrocytic anemia

With megaloblastosis:

The peripheral blood film shows macrocytes with hypersegmented polymorphs with six or more lobes in the nucleus. If severe there may be leucopenia and thrombocytopenia (deficient DNA synthesis)

Causes include:

- Vitamin B12 deficiency.
- Folic acid deficiency
- Congenital enzyme deficiencies in DNA synthesis or drugs interfering with DNA synthesis (hydroxyurea, azathioprine).
- Myelodysplasia due to dyserythropoiesis

1. Vitamin B12 deficiency

- Sources: meat, fish, eggs, and milk.
- Not destroyed by cooking.
- Daily requirement is 1-2 mg / day.
- Absorption and transport: Vit. B12 is liberated in the stomach, bound to intrinsic factor (IF) and absorbed through the terminal ileum, transported by transcobalamin I (and to lesser extent by transcobalamin II and III) to be utilized by the tissues.
- It is essential for:
  - Hematopoiesis
  - GIT mucosa integration
  - Formation of myelin of nervous system.
- Storage: the average adult stores 2-3 mg in the liver, it may take two years or more before B12 deficiency develops as the daily loss are small (1-2mg).

Causes of deficiency:

- Dietary deficiency:
- Malabsorption syndrome. (Celiac) Crohn's disease
- Gastric pathology:
  - Pernicious anaemia.
    - This is an organ-specific autoimmune disorder in which the gastric mucosa is atrophic, with loss of parietal cells causing intrinsic factor deficiency. In the absence of intrinsic factor, less than 1% of dietary vitamin B12 is absorbed.
    - Pernicious anaemia has an incidence of 25/100 000 population over the age of 40 years in developed countries, but an average age of onset of 60 years.

- It is more common in individuals with other auto-immune disease (Hashimoto's thyroiditis, Graves' disease, vitiligo, hypoparathyroidism or Addison's disease) or a family history of these or pernicious anaemia.
- The finding of anti-intrinsic factor antibodies in the context of B12 deficiency is diagnostic of pernicious anaemia without further investigation.
- Antiparietal cell antibodies are present in over 90% of cases but are also present in 20% of normal females over the age of 60 years; a negative result makes pernicious anemia less likely but a positive result is not diagnostic.
- The Schilling test, not favorable anymore.

- Rare congenital enzyme deficiency.
- Increased demand
  - Infancy, pregnancy, lactation
  - Hemolysis and active hematopoiesis
  - Malignancy.
  - Thyrotoxicosis
- Increased loss: Hemodialysis
- Decrease of stores. Far advanced chronic liver diseases.

Causes of vitamin B12 deficiency:

VITAMIN B:
V*egan*
I*leal resection*
T*apeworm*
A*utoimmune (PA)*
M*egaloblastic anemia*
I*nflammation of terminal ileum*
N*itrous oxide*
B*acterial overgrowth*

Clinical features

1. Hematological manifestations
   - General manifestation of anemia.
   - Bleeding tendencies (Thrombocytopenia)
   - Increased risk of infection (Leucopenia)
   - Hepatosplenomegaly.
   - Color of the skin is lemon yellow owing to combination of pallor and jaundice.
2. GIT manifestations
   - Atrophic glossitis.
   - Angular stomatitis.
   - Gastric atrophy dyspepsia, anorexia nausea, vomiting.
   - Intestinal atrophy diarrhea and malabsorption
   - Stomach cancer on top of atrophic gastritis
3. Neurological manifestations (not seen in folate deficiency).
   - Peripheral neuropathy
     - Glove and stocking paraesthesiae
     - Loss of ankle reflexes
   - Subacute combined degeneration of the cord (SCD)
     - Pyramidal tract affection (UMNL) → spastic paraplegia.
     - Posterior column affection→ loss of deep sensation, vibration, proprioception.
   - Dementia
   - Optic atrophy

- Psychosis (rare)
- Autonomic neuropathy

Investigations (you could probably guess those by now☺)

1. CBC

- RBCs:
  - Macrocytic normochromic anemia.
  - Poikilocytosis and anisocytosis
  - Howell-Jolly bodies may be present
- WBCs:
  - Moderate Leucopenia.
  - Shift to the right (hypersegmented neutrophils).
- Platelets
  - Moderate thrombocytopenia.
  - Giant platelets
- Reticulocytes are decreased but increased by treatment with vit. B12

2. Bone marrow:

- Show megaloblastic erythropoiesis. The most characteristic is dissociation between nuclear & cytoplasmic development in erythroblasts with the nucleus maintaining a primitive appearance despite maturation and haemoglobinization of the cytoplasm.
- Erythroid hyperplasia, maturation defect in erythropoiesis, giant metamyelocytes, atypical megakaryocytes with hypersegmented nuclei.

3. Estimation of serum B12 level

- Low using radioisotope dilution or immunological assays.

4. Biochemistry

- Serum iron is high, more than 175-mcg/100 ml.
- Increased serum indirect bilirubin reflects mild hemolysis and ineffective erythropoiesis.
- Increased serum lactic dehydrogenase (LDH) reflecting ineffective megaloblastic erythropoiesis.

5. Immunology. (Imp. For diagnosis of pernicious anemia).

- Parietal cells antibodies and gastrin receptor antibodies, present in 70% of patients. (Not specific)
- Intrinsic factor antibodies type I blocking (more specific) and type II precipitating antibodies.

6. Other investigations

- Gastric biopsy: proximal 2/3 of stomach is atrophic.
- Augmented histamine test: achlorhydria.
- Schilling test for pernicious anemia.
- **High homocystine (also seen in folate deficiency)**
- **High methylmalonate in urine.**

Treatment

- Treatment of the underlying cause.
- Replacement therapy.
  - Initial treatment is often as the 1000 µg B12 IM/day for 2 weeks then in a dosage of 1000 µg twice/week till correction of anemia. Maintenance therapy 1000 mg IM every 3 months.
- Neurological damage can be precipitated by treating incorrectly with folic acid.
- Transfusion therapy by packed RBCs in the flowing conditions
  - Severe anemia (Hb < 7 gm/dl)
  - Heart failure
  - Marked symptoms.

Note(s):

The reticulocyte count will peak by the 5th–10th day after starting replacement therapy. The haemoglobin will rise by 10 g/L every week until normalised. The response of the marrow is associated with a fall in plasma potassium levels and rapid depletion of iron stores. If an initial response is not maintained and the blood film is dimorphic (i.e. shows a mixture of microcytic and macrocytic cells), the patient may need additional iron therapy. A sensory neuropathy may take 6–12 months to correct; long-standing neurological damage may not improve.

2. Folate deficiency

- Folates are produced by plants and bacteria, hence dietary leafy vegetables (spinach, broccoli, and lettuce), nuts, yeast, fruits (bananas, melons) and animal protein (liver, kidney) are a rich source.
- Minimum daily requirements 100-200 ug.
- An average Western diet contains more than the minimum daily requirement but excess cooking destroys folates.
- Most dietary folate is present as polyglutamates; these are converted to monoglutamate in the upper small bowel (mostly in the jejunem) and actively transported into plasma.
- Plasma folate is loosely bound to plasma proteins such as albumin and there is an entero-hepatic circulation.
- Total body stores of folate are small and deficiency can occur in a matter of weeks.
- It is essential for DNA synthesis and cell maturation.
- Its deficiency leads to nuclear maturation defect of blood cells and GIT mucosal cells.

Causes of deficiency:

1. Decreased intake
   - Particularly in infancy, old age, poor social conditions, starvation, alcohol excess and in psychiatric patients.
2. Increased demand
   - Physiological: pregnancy, lactation, prematurity
   - Pathological:
     - Hematological disease with excess red cells production e.g hemolysis.
     - Malignant disease with increased cell turnover
     - Inflammatory diseases e.g. rheumatoid arthritis.
     - Metabolic disease e.g. homocystinuria (rare congenital defect in the conversion of homocysteine to cystathion folate).
3. Decreased absorption
   - Small bowel disease especially celiac disease.
4. Drugs affecting folate metabolism.
   - Anticonvulsants **(e.g. phenytoin may inhibit intestinal conjugase inhibiting conversion of polyglutamates into monoglutamates)**, methotrexate **(cytotoxic)**, pyrimethamine and trimethoprim **(inhibit the enzyme responsible for the conversion of dihydrofolate into tetrahydrofolate)** and oral contraceptives.
5. Increased loss
   - Plasma folate is loosely bound to plasma proteins such as albumin; thus, Hemodialysis or peritoneal dialysis can cause loss.

Note:

Pregnant MAN!!
Pregnancy *and lactation*
M*alabsorption (celiac and crohn's)*
A*lcoholism*
N*utritional (tea and toast elderly diet*

Clinical features

- May occur at any age, degrees of folate deficiency are very common and mild deficiency states are frequently unrecognized.
- The clinical picture varies according to possible underlying causes.
- The onset may be insidious or rapid as when negative folate balance is precipitated by e.g infection
- Anemia and sometimes-slight jaundice.
- Glossitis.
- No defined neurological changes.

Investigations

- Essentially as in vit B12 deficiency.

- Howell Jolly bodies and target cells in the blood film would suggest splenic atrophy (coeliac disease)
- Bone marrow shows megaloblastic changes.
- Reduced folate levels: (N= 2.5-25mg/ml) serum levels are labile **(a single folate rich meal could normalize the lvls even in a truly deficient person)** and red cell levels are better reflection of tissue folate by radioisotope dilution or immunological methods.

Treatment

- Folic acid therapy is contraindicated if there is any suspicion of B12 deficiency (Neurological changes may be precipitated).
- Folic acid: 5mg daily is more than adequate.
- Prophylactic folic acid is also given in chronic hematological disorders where there is rapid cell turnover 5mg each week. **As in sickle cell anemia.**
- Prophylactic folic acid (400ug daily) is recommended in pregnancy to reduce the risk of megaloblastosis and fetal neural tube defects. **Pregnancy-induced folate deficiency is the most common cause of megalo-blastosis worldwide and is more likely in the context of twin pregnancies, multiparity and hyperemesis gravidarum.**

Without megaloblastosis:

- A raised MCV with macrocytosis on the peripheral blood film can occur with a normablastic BM. In all these conditions, the level of Vit B12 and folate are normal.
- The exact mechanisms are uncertain, but it seems there is increased lipid deposition in the red cell membrane.
- Physiological: pregnancy and newborn.
- Pathological
  - Alcohol excess
  - Liver disease
  - Reticulocytosis
  - Hypothyroidism
  - Some hematological disorders (e.g aplastic anaemia, sideroblastic anemia)
  - Drugs (e.g. cytotoxic as azathioprine). **Also, don't forget anti-retroviral treatment (ART)**
  - Agglutinated red cell measured on red counters.
  - Cold agglutinins due to autoagglutination of red cells, MCV decreases to normal with warming of the sample to 37C.

Causes of macrocytosis in general:

Feed Them:
F*olate and vitamin B12 deficiency*
E*thanol and liver disease*
D*rugs (ART, methotrexate)*
T*hyroid (hypo)*
H*emolysis (reticulocytes are large cells)*
M*yelodysplastic syndrome*

Case:

A 38 year old with HIV on HAART presents with macrocytosis.
Think: ART

Hemolytic Anemia

- The hemolytic anemias are a group of diseases in which red cell life span is shortened.
- Pathophysiology:
- Hemolysis of RBC can occur either.
  - Intravascular i.e. within the circulation.
  - Extravascular i.e. by phagocytes in RES in the liver, bone, spleen.
- Bone marrow compensatory reactions:
  - Erythriod hyperplasia in BM, can increase erythropoiesis several times, so that anemia may not develop till RBCs life span is less than 20 days
  - Reticuylocytosis is a hallmark.
  - Slight macroytosis in the peripheral blood
- Consequences:
  - Low Hb.
  - Excess hemolysis increases bilirubin (unconjugated) since the liver can increase its capacity several times; the jaundice is mild if there's any at all.
  - The intravascular liberated Hb is bound to haptoglobin. Hb-haptoglobin is rapidly cleared from the circulation by RES, and plasma level of haptoglobin is reduced. In cases of excessive IV haemolysis, haptoglobin is depleted.
  - Haemoglobinuria and dark urine.
  - Extramedullary erythropoiesis will cause specific bone changes.

Causes:

1. CORPUSCULAR CAUSES:

- ***Congenital Abnormalities***
  - I. Membrane defects
    - Hereditary spherocytosis
    - Hereditary elliptocytosis
    - Hereditary stomatocytosis
  - II. Haemoglobinopathies
    - Sickle cell anemia
    - Thalassemia
  - III. Enzymopathies
    - Abnormal aerobic glycolysis e.g. G6PD deficiency
    - Abnormal anaerobic glycolysis e.g pyruvate kinase deficiency
    - Non glycolytic enzymopathies
- ***Acquired Abnormalities***
  - Paroxysmal nocturnal haemoglobinuria (abnormal membrane)
  - Vitamin E deficiency.

2. EXTRACORPUSCULAR CAUSES:

i. Immune mechanisms

- ❖ Alloantibodies:
  - Incompatible blood transfusion
  - Hemolytic disease of the newborn
  - After allogeneic BM or organ transplantation
- ❖ Autoantibodies:

1. Warm antibodies (react at 37° C and do not cause agglutination. Type of AB= IgG).

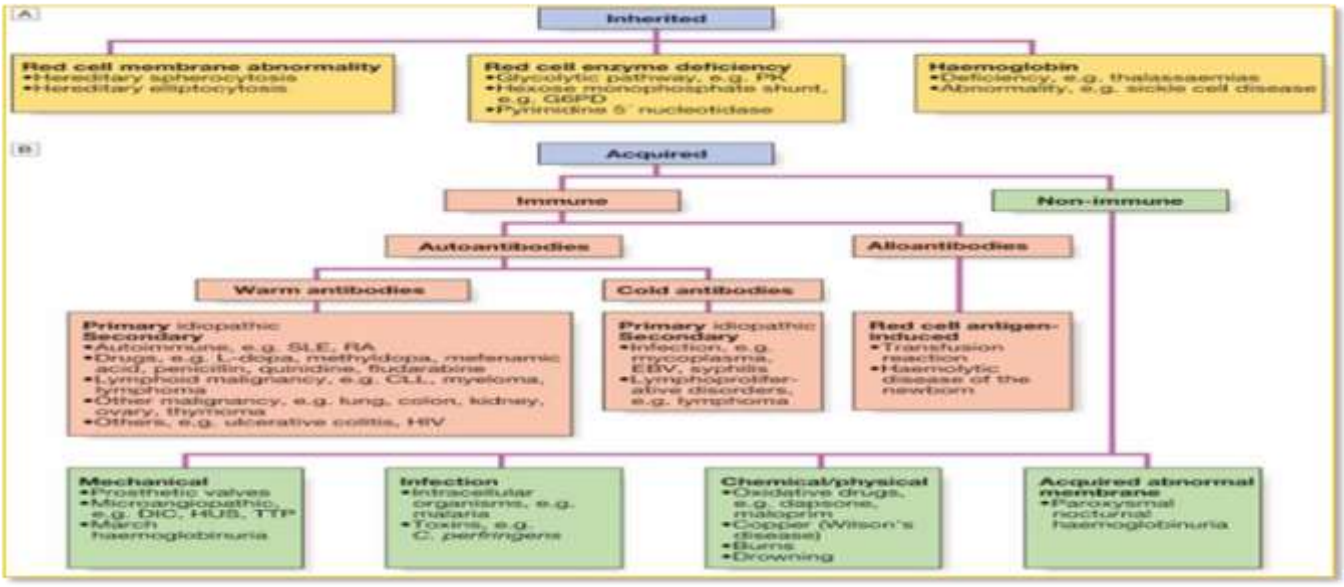
- Primary: Idiopathic
- Secondary:
  - CLL, Lymphoma, SLE and rheumatoid disease
  - Drugs e.g methyldopa.

2. Cold antibodies (react at 32°C (best at4°C) and usually agglutinates and hemolyse red cells. Type of AB = IgM).

- Cold haemoagglutinin disease
  - This affects elderly patients and may be associated with an underlying low-grade B cell lymphoma. It causes a low-grade intravascular haemolysis with cold, painful and often blue fingers, toes, ears or nose (so-called acrocyanosis). The latter is due to red cell agglutination in the small vessels in these colder exposed areas.
- Primary: Idiopathic
- Secondary (mycopolasma pneumonia infection, infectious mononucleosis and lymphomas.
- Paroxysmal cold haemoglobinuria
  - Idiopathic
  - Secondary (some viral infections congenital & tertiary syphilis).

- ❖ Immunochemical mechanisms: drug induced hemolytic anemia.

ii. Non-immune mechanism



Clinical features

- General features of anemia
- General features of haemolytic anaemia
  - Mild jaundice
  - Hepatosplenomegaly:
    - Common in chronic hemolysis except in cases of sickle cell anemia where there's self-splenectomy (repeated infarctions)
- Biliary obstruction and /or gall bladder stones.
  - Pigment stones: extra hepatic obstruction.
  - Viscid bile: intrahepatic obstruction.
  - NB. In this situation jaundice becomes, mixed (hemolytic + obstructive) and abdominal pain may be present.
- Leg ulcer (common with sickle cell)
  - Chronic leg ulcer surrounded by pigmentation cause by extra vascular iron deposition. Ulcers develop on the malleoli.
- Crises:

A. Heamolytic crisis

- Fever, rigors: pyrogens release from RBCs death and subsequent inflammatory responce.
- Generalized bone pain
- Acute abdominal pain, backache, may be vomiting.
- Aggravation of anaemia (pallor), increased jaundice, and dark urine.
  - Investigations:
    - Dark urine: haemoglobinuria.
    - Increased serum bilirubin
    - Reticulocytosis
    - BM erythroid hyperplasia.
    - Cause may be infection

B. Aplastic crisis

- Inability of BM to replace the destructed RBCs
- Severe anaemia without jaundice.
- Reticulocytopenia.
- BM erythroid hypoplasia.

- Cause: viral infection especially parvovirus B19 and HBV.

C. Sequestration crisis

- Trapping and pooling of RBCs in the spleen (mostly in children) and liver (in the adult).
- **Can also cause sickle chest syndrome. The most common cause of death in SCA.**
- **Can cause priapism in male SCA patients.**

D. Megaloblastic crisis

- Anaemia severe without jaundice.
- Decreased reticulocytic count
- Macrocytic anaemia.
- BM: megabloblastic
- Cause: folic acid deficiency.

E. Vaso-occlusive crisis (Thrombotic phenomenon)

- Only in cases of sickle cell anemia.
- This is the most common crisis with SCA.
- Plugging of small vessels in the bone produces acute severe bone pain.
- **This affects areas of active marrow: the hands and feet in children (so-called dactylitis) or the femora, humeri, ribs, pelvis and vertebrae in adults.**
- **Patients usually have a systemic response with tachycardia, sweating and a fever.**

  - Complications of hemolytic anemia:
    - Hemolytic crisis:
    - Aplastic crisis:
    - Folate deficiency caused by increased BM requirement
    - Gall stones
    - In sickle cell anaemia: signs and symptoms of thrombotic phenomena.

Investigations

Diagnostic Tests

The best **initial** test is a **peripheral smear.** Sickle cell trait (AS disease) does **not** give sickled cells. The most **accurate** test is the hemoglobin **electrophoresis.**

1. CBC
    - Normocytic normochromic anemia with reticulocytosis
    - Microcytic anaemia: thalassaemia, spherocytosis.
    - Macrocytic + reticulocytosis: megaloblastic crisis
    - Reticulocytopenia: aplastic crisis.
    - Morphological evidence of RBC damage: as red cell fragment, cell containing malarial parasites.
    - Morphology of RBCs: eg spherocytes & sickle cells
    - WBC and thrombocytes are normal but may increase due to bone marrow stimulation.
2. Short life span of RBCs:
    - Measured by radioactive chromium^{51} labeled RBCs
3. Bone marrow
    - Hypercellular normabloastic marrow.
    - Hypocellular in a plastic crisis.
    - Megaloblastic in folate deficiency.
4. Increased serum LDH
5. Incresesd serum biliurbin, fecal stercobilinogen and urinary urobilinogen.
6. In intravascular haemolysis vs. extravascular the following parameters are presents.
    - Haemoglobinuria and haemosiderinuria
    - Reduced plasma haptoglobin and haemopexin

- Presence of methaemalbumin in plasma.

Hallmarks of haemolysis	
• ↓Haemoglobin • ↑Unconjugated bilirubin • ↑Lactate dehydrogenase	• ↑Reticulocytes • ↑Urinary urobilinogen
Additional features of intravascular haemolysis	
• ↓Haptoglobin • ↑Methaemalbumin	• Positive urinary haemosiderin • Haemoglobinuria

7. Tests for the cause
   - RBC morphology.
   - Osmotic fragility.
   - Hb electrophoeresis.
   - Estimation of G6PD.
   - Coomb's test.

Now, let's take a closer look at some of those diseases

1. Hereditary spherocytosis

Hereditary Spherocytosis

Etiology

This is a defect in the cytoskeleton of the red cell leading to an abnormal round shape and loss of the normal flexibility characteristic of the biconcave disc that allows red cells to bend in the spleen.

"What Is the Most Likely Diagnosis?"

- Recurrent episodes of hemolysis
- Intermittent jaundice
- Splenomegaly

- Family history of anemia or hemolysis
- Bilirubin gallstones

Diagnostic Tests

- Low MCV
- Increased mean corpuscular hemoglobin concentration (MCHC)
- Negative Coombs test

The most accurate test is osmotic fragility. When cells are placed in a slightly hypotonic solution, the increased swelling of the cells leads to hemolysis.

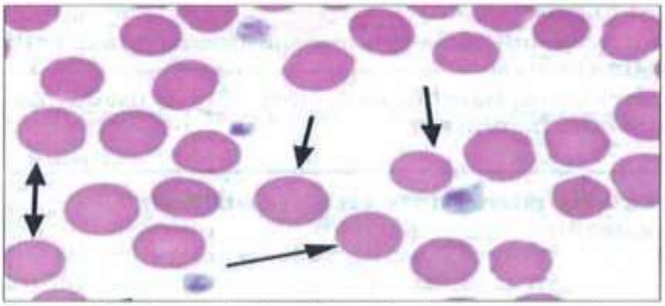


Figure 7.4: Spherocytes lose the central pallor of normal red cells. The MCHC is elevated. *Source: Alireza Eghtedar, MD.*

Treatment

1. Chronic **folic acid** replacement supports red cell production.
2. **Splenectomy** stops the hemolysis but does not eliminate the spherocytes.

Haemoglobinopathies

- Normal hemoglobin consists of:
  - Globin: protein, which is formed from 4 polypeptide chains. Two alpha and two non-alpha globin chains. **Alpha globin chains are produced throughout life, including in the fetus, so severe mutations may cause intrauterine death. Production of non-alpha chains varies with age; fetal haemoglobin (HbF-αα/γγ) has two gamma chains, while the predominant adult haemoglobin (HbA-αα/ββ) has two beta chains. Thus, disorders affecting the beta chains do not present until after 6 months of age. A constant small amount of haemoglobin A2 (HbA2-αα/δδ, usually less than 2%) is made from birth.**
  - Haem: iron-protoporphyrin complex
- Normal electrophoresis pattern in an adult:
  - Hemoglobin A (96%): α2/β2 (normal hemoglobin)
  - Hemoglobin F (3%): α2/γ2 (normal till 6 months of age)
  - Hemoglobin A2: α2/δ2.
- Haemoglobinopathies are clinical syndromes resulting from abnormalities in the structure of globin molecule or with a reduction in the rate of production of one or other of the globin chains.
- There are three main categories:
  - Structural variant of Hb: e.s HbS.
  - Failure to synthesize Ht: e.g thalassemia.
  - Failure to switch from fetal Hb (HbF) to adult Hb (HbA) : Heridetary persistence of Fetal Hb (HPFH).

1. Sickle cell anemia

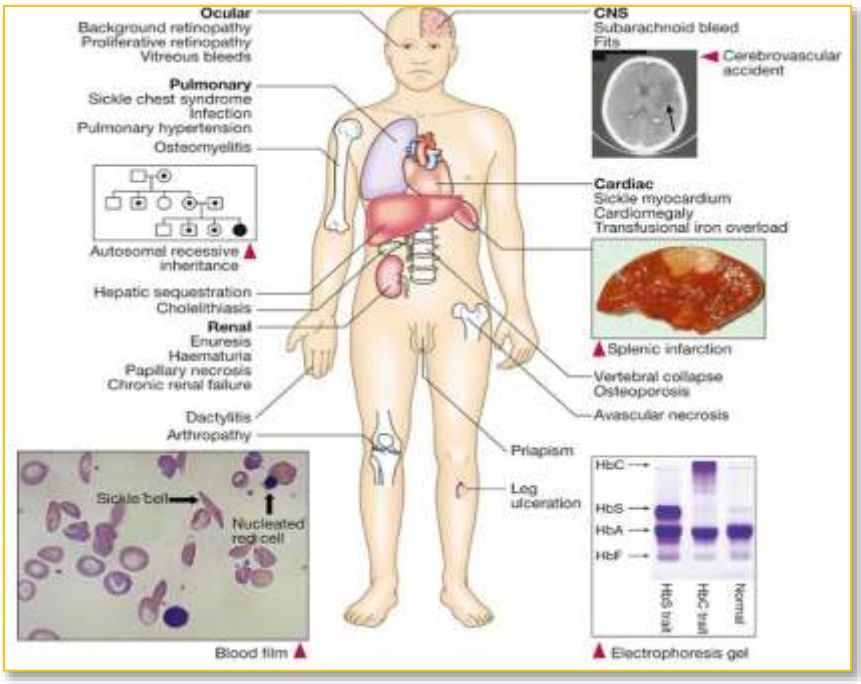
- A hereditary autosomal recessive disorder characterized by the presence of HbS **(formed by substitution of valine for glutamic acid in the sixth position of the β –hemoglobin chain)**, which on exposure to hypoxia, the deoxygenated Hbs forms insoluble aggregates that distort the erythrocytes and increase their rigidity causing sickling deformation and subsequent fragmentation. In addition, the unyielding elongate crescentic erythrocytes form aggregates that block terminal

Chronic disease manifestations:

Skeletal: aseptic necrosis of femoral head and salmonella osteomyelitis.
Biliary disease: pigmented gallstones (high bilirubin)
Renal chronic hematuria (renal papillary necrosis)
Liver disease: viral hepatitis (transfusion) and secondary hemochromatosis.
Pulmonary: local infection and vascular occlusion (acute chest syndrome), pulmonary hypertension.
Heart: enlarged, flow murmur,MI.
Immune: functional splenism increase chance of sepsis with encapsulated organisms (strep. Pneumonia, H. influenza and salmonella osteomyelitis)
Eye: ischemic retinopathy.
Neuro: increased risk for TIA and CVAs.

arterioles, capillaries and veins, resulting in tissue infractions and perivascular oedema in the involved organs (vascular occlusion)

- Symptoms vary from a mild asymptomatic disorder to severe haemolytic anaemia and recurrent severe painful crises.



Signs of SCA:

SICKLE:
S*plenomegaly/****S****ludging*
I*nfection*
C*holelithiasis/acute* ***C****hest syndrome*
K*idney (hematuria, papillary necrosis)*
L*iver congestion/****L****eg ulcer*
E*ye changes*

- People with sickle cell trait (heterozygous sickling disorder) have no symptoms unless extreme circumstances cause anoxia such as flying in a non-pressurized aircraft or problems with anaesthesia.
- Sickle cell trait protects against plasmodium falciparum malaria. (Controversy)
    - 60% Hb A and 40% Hb S
    - The blood count and film are normal
    - The diagnosis is made by a positive sickle test or by Hb electrophoresis.

Investigations:

- Features of hemolytic anemia.
- CBC
    - The level of Hb is in the range 6-8gm/dl with high reticulocytic count 10-20%. (Blood films show features of hyposplenism)
    - Sickling of red cells on a blood film can be induced in the presence of sodium metabisulphite, which cause hypoxia.
- Hb electrophoresis:
    - There is no Hb A, 80-85% Hb S and 2-20% HbF .
    - The parents of the affected child will show features of sickle cell trait.

Treatment

1. Begin with oxygen/hydration/analgesia.
2. If fever or a white cell count higher than usual is present, then antibiotics are given. Use ceftriaxone, levofloxacin, or moxifloxacin.
3. Folic acid replacement is necessary on a chronic basis.
4. Give pneumococcal vaccination because of autosplenectomy.
5. Hydroxyurea prevents recurrences of sickle cell crises by increasing hemoglobin F.

IMPORTANT NOTES FROM EXTERNAL RESOURCES

Notes

	Management of SCA
Davidson	All patients with sickle-cell disease should receive prophylaxis with daily folic acid, and penicillin V to protect against pneumococcal infection, which may be lethal in the presence of hyposplenism. These patients should be vaccinated against pneumococcus, meningococcus, Haemophilus influenzae B, hepatitis B and seasonal influenza. Vaso-occlusive crises are managed by aggressive rehydration, oxygen therapy, adequate analgesia (which often requires opiates) and antibiotics. Transfusion should be with fully genotyped blood wherever possible. Simple top-up transfusion may be used in a sequestration or aplastic crisis. A regular transfusion programme to suppress HbS production and maintain the HbS level below 30% may be indicated in patients with recurrent severe complications, such as cerebro-vascular accidents in children or chest syndromes in adults. Exchange transfusion, in which a patient is simultaneously venesected and transfused to replace HbS with HbA, may be used in life-threatening crises or to prepare patients for surgery. A high HbF level inhibits polymerisation of HbS and reduces sickling. Patients with sickle-cell disease and high HbF levels have a mild clinical course with few crises. Some agents are able to increase synthesis of HbF and this has been used to reduce the frequency of severe crises. The oral cytotoxic agent hydroxycarbamide has been shown to have clinical benefit with acceptable side effects in children and adults who have recurrent severe crises. Relatively few allogeneic stem cell transplants from HLA-matched siblings have been performed but this procedure appears to be potentially curative

Case:

A young African American man with microcytic hypochromic anemia is found to have a high ferritin and iron with normal RDW.
Think: SCA
Next step: Hb electrophoresis.

Indications for EXCHANGE transfusion in SCA:

Stroke/TIA
Acute chest syndrome
Priapism
Third term pregnancy
Intractable vaso-occlusive crises.

2-The Thalassemias:

The Thalassaemias are anemias originally found in people living on the shores of the Mediterranean, now known to affect people throughout the world. The defective synthesis of globin genes in thalassaemia leads to imbalanced globin chain production leading to precipitation of globin chains within the red cell precursors and resulting in ineffective erythropoiesis. Precipitation of globin chains in mature red cell leads to haemolysis. There are two α genes and one β gene on each chromosome, making a total of four α genes and two β genes. In alpha-thalassaemia, disruption of one or both alleles on chromosome 16 may occur, with production of some or no alpha globin chains. In beta-thalassaemia, defective production usually results from disabling point mutations causing no (β0) or reduced (β–) beta chain production.

i. α-thalassemia:

- In contrast to β -thalassaemia it is caused by gene deletions.
- If all four genes are absent (__/__) there is no α-chain synthesis and Hb Barts (γ4) is present. Hb. Barts cannot carry oxygen and is incompatible with life and infants are either stillborn or die very shortly after birth. (hydrops fetalis)
- If three genes are deleted (__/_α) there is moderate anaemia and splenomegaly (Hb H disease). Hb A, Hb Barts and Hb H (B4) are present, Hb A2 is normal or reduced
- If two genes are deleted (__/αα)(α -thalassaemia trait), there is microcytosis with or without mild anaemia. **(αα/__) thalassemia trait is more common in Asians. (α_/ α_) is more common in Africans)**
- If only one gene is deleted (αα /α _), the patient is usually asymptomatic.

ii. β-thalassemia:

- There is defective production of beta-chain with excess production of Hb F and Hb A2.
- Types
  1. β-thalassaemia major (Cooley's anemia)
     - Homozygous (2 abnormal gene)
     - Hb F > 80-90%
     - The onset is during the first year of life (3-6 mouth)
     - The children fail to thrive, (stunted growth) mongoloid faces, bone deformities.
     - Feature of hemolytic anaemia, jaundice and huge splenomegaly

- Extramedullary haematopoiesis: that soon causes hepatosplenomgaly, bone expansion giving rise to classical thalassaemic facies (mongoloid facies).
- Feature of iron overload: liver cirrhosis, bronze skin, and cardiomegaly.
- The disease is more common among Mediterranians.
- Positive family history.
- Usually recurrent bacterial infection.

2. β-thalassaemia intermedia Heterozygous (only one abnormal gene)
   - Both Hb F (10%) and Hb A2 (10%).
   - Presented in adults by mild Microcytic hypochromicmic anemia and splenomegaly.
   - Commonly symptomatic and require treatment.
3. β-Thalassaemia minor (trait)
   - This common carrier state is asymptomatic, anaemia is mild or absent, the red cells are hypochromic and microcytic with a low MCV and MCH.
   - DD: Iron deficiency anemia. Must be excluded.
   - Rarely requires treatment.

Genotype	HbA	HbA_2	HbF (%)	Other Hemoglobins
Normal β/β	97	2.5 – 3.2	<1	None
Thalassaemia major	0 Present	1.0 – 5.9	>94	Free α-chains Free α-chains None
Thalassaemia intermedia	Present	5.4 – 10.0	30 – 73	None
Thalassaemia minor	>90	3.5 – 8.0	1 – 2	None
Thalassaemia minima β^{silent}/β	97	<3.2	<1	None

Electrophoresis Findings	
Alpha thalassemia	**Beta thalassemia**
One gene deleted: normal	Increased hemoglobin F and A_2
Two genes deleted: mild anemia, normal electrophoresis	N/A
Three genes deleted: moderate anemia with hemoglobin H, which are beta-4 tetrads; increased reticulocytes	Beta thalassemia intermedia • Normal hemoglobin F • No transfusion dependence
Four genes deleted: gamma-4 tetrads or hemoglobin Bart; CHF causes death in utero	N/A

Enzymopathies

1. Glucose-6- phosphate dehydrogenase deficiency:

"What Is the Most Likely Diagnosis?"

Look for African American or Mediterranean **men** with **sudden anemia** and jaundice who have a normal-sized spleen with an infection or are using one of the drugs previously listed.

Diagnostic Tests

The best initial test is for Heinz bodies and bite cells. The G6PD level will be **normal** after a hemolytic event. The most accurate test is the G6PD level after waiting 1 to 2 months after an acute episode of hemolysis.

- The enzyme glucose-6-phosphate dehydrogenase (G6PD) is pivotal in the hexose monophosphate shunt pathway. (**It reduces nicotinamide adenine dinucleotide phosphate (NADPH) while oxidizing glucose-6- phosphate. It is the only source of NADPH in red cells and as NADPH is needed for the production of reduced glutathione, a deficiency renders the red cell susceptible to oxidative stress)**
- In the absence of G6PD, irreversible oxidative denaturation of Hb occurs which becomes attached to red cell membrane to form Heinz bodies. Red cells containing such inclusion bodies become destroyed in the liver and spleen. **Resulting also in bite cells**
- Agents which may cause haemolysis in (G6PD) deficiency
    - Infections and other acute illnesses e.g diabetic ketoacidosis
    - Fava beans.
    - Drugs
        1. Sulfonamides e.g co-trimoxazole
        2. Ant. Bacterial e.g interferons, chloramphenicol
        3. Analgesics e.g aspirin, acetamenophin
        4. Anti-malarial e.g primaquine and quinine
- Rapidly developing intravascular haemolysis with haemoglobinuria.
- Investigations:
    - Between crises the blood count is normal
    - Direct enzyme assay.
    - During crisis, the blood film may show contracted and fragmented cells "bite" cells and blister" cells which have Red Heinz bodies removed by the spleen.
    - Haemoglobinaemia and haemoglobinuria in the acute stage.

ABCs of G6PD deficiency:

***A**ntimalarials*
***B**actrim/Bite cells*
***Ci**profloxacin*
***D**KA*
*Inf**E**ction*
***F**ava beans*

Treatment

Nothing reverses the hemolysis. Avoid oxidant stress.

- Withdrawal of the drug or toxic substance
- Blood transfusion is lifesaving in severe cases.

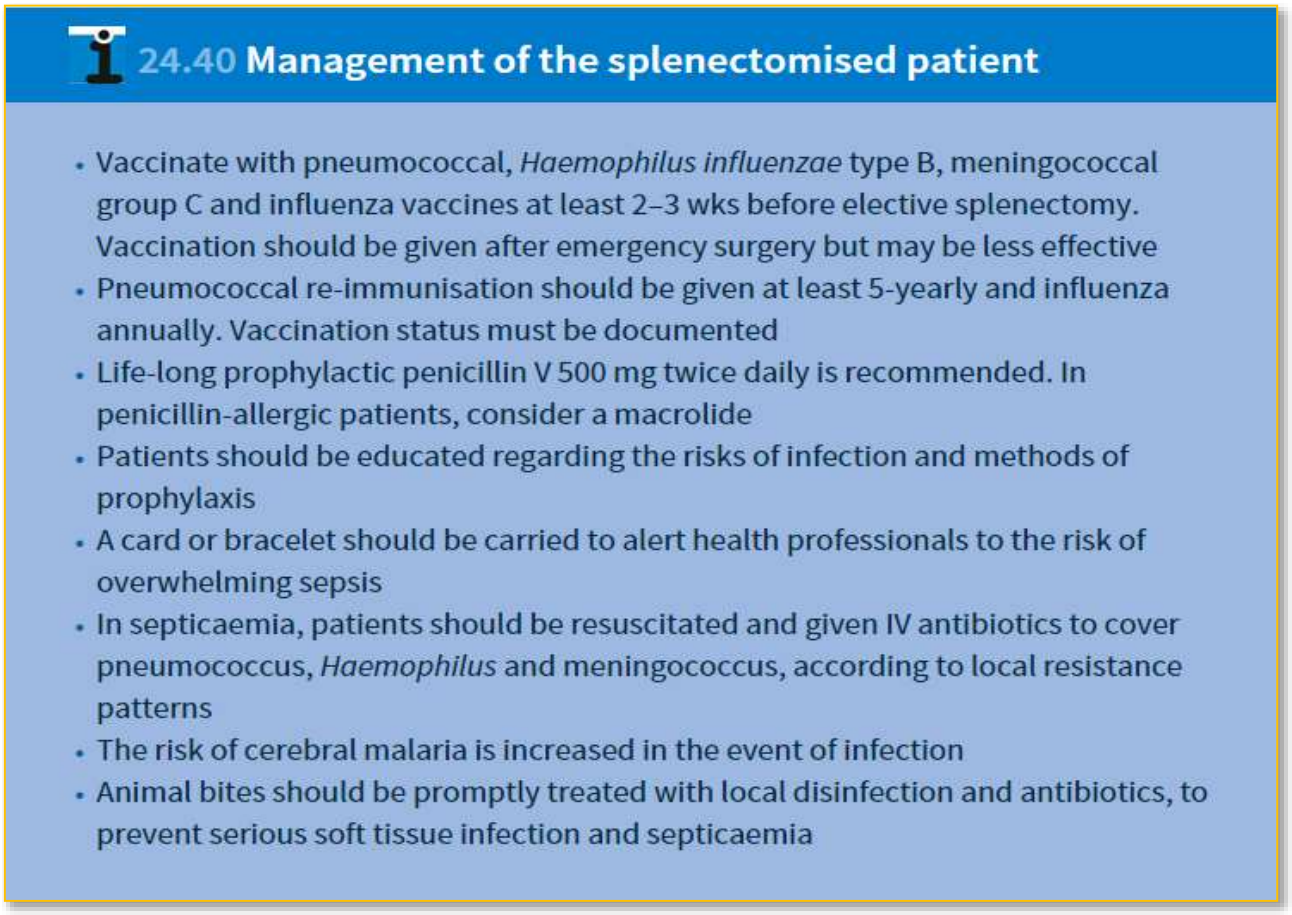
Case:

A 31 year old Italian male with back pain, dark urine, jaundice, and anemia after 2 days of ciprofloxacin.
Think: G6PD deficiency.
Next step: check peripheral smear looking for "bite cells", transfuse if severe anemia and check renal function.

2. Pyruvate kinase deficiency:

- This is inherited as an autosomal recessive, the affected patients being homozygous or less common heterozygous. The red cells become rigid as a result of reduced adenosine triphosphate (ATP) formation.
- Direct enzyme assay is needed to make the diagnosis.

EXTRA



Questions

1) All of the following may cause microcytic anemia, EXCEPT?

a) IRON deficiency.

b) Vitamin B12 deficiency.

c) Chronic disease.

d) Thalassaemia.

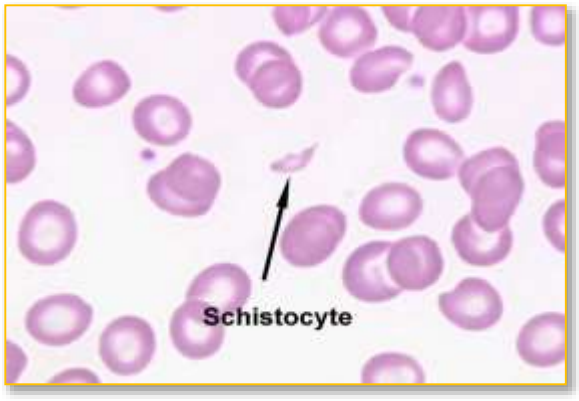
2) Female pt developed anemia. The following is her blood film. schistocytes were shown what is the most likely cause?

a) Microangiopathic hemolytic anemia

b) Sickle cell disease

c) Thalassemia

d) G6PD deficiency.



3) A 73-year-old man comes to the office with fatigue that has become progressively worse over the last several months. He is also short of breath when he walks up one flight of stairs. He drinks 4 vodka martinis a day. He complains of numbness and tingling in his feet. On physical examination he has decreased sensation of his feet. His hematocrit is 28% and his MCV is 114 fl (elevated).

What is the most appropriate next step in management?

a) Vitamin B^{12} level
b) Folate level
c) Peripheral blood smear
d) Schilling test
e) Methylmalonic acid level

4) A 73-year-old woman comes with decreased position and vibratory sensation of the lower extremities, a hematocrit of 28%, MCV of 114 fl, and hypersegmented neutrophils. Her B_{12} level is decreased, but near the borderline of normal.

What is the most appropriate next step in the management of this patient?

a) Methylmalonic acid level
b) Anti-intrinsic factor antibodies
c) Anti-parietal cell antibodies
d) Schillings test
e) Folate level
f) Homocysteine level

5) Which of the following is a complication of B12 or folate replacement?

a. Seizures

b. Hemolysis

c. Hypokalemia

d. Hyperkalemia

e. Diarrhea

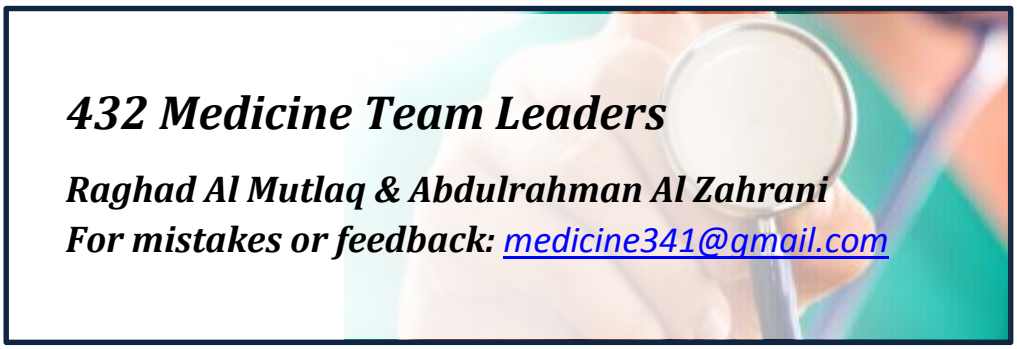
6) Which of the following can be found on smear in sickle cell disease?

a) Basophilic stippling
b) Howell-Jolly bodies
c) Bite cells
d) Schistocytes
e) Morulae

7) A 43-year-old man with sickle cell disease is admitted with an acute pain crisis. His only routine medication is folic acid. His hematocrit on admission is 34%. On the third hospital day, the hematocrit drops to 22%.

What is the best initial test?

a) Reticulocyte count
b) Peripheral smear
c) Folate level
d) Parvovirus B-19 lgM level
e) Bone marrow



Answers:

1: b

2: a

3:c

4:A

5:c

6:b

7:a