Megaloblastic anemia:

Anemia characterized by ***macrocytic blood picture*** and ***megaloblastic maturation of haemopoeitic cells***. So in this type of anemia, the erythroblasts in the bone marrow show a characteristic abnormality-maturation of the nucleus being delayed relative to that of the cytoplasm.
Here the ***defect*** is in the ***DNA synthesis*** so the ***maturation of nucleus*** being ***delayed relative to that of cytoplasm***, the nuclear ***chromatin*** maintains an ***open***, ***stippled***, ***lacy*** appearance despite ***normal hemoglobin formation in the cytoplasm*** of erythroblast as they mature.

Causes:

1. Vitamin B12 deficiency.
2. Folate deficiency.
3. Abnormalities of vitamin B12 or folate metabolism e.g. trascobalamin II deficiency.
4. Other defect of DNA synthesis:
    A. Congenital enzyme deficiencies e.g. aortic aciduria.
    B. Acquired enzyme deficiencies e.g. alcohol, drug.

Vitamin B12 (Cobalamin)Absorption:

Average daily diet contains 5-30 µg of vitamin B12 mainly in foods of animal origin such as liver , meat, fish ,eggs and milk – well in excess of 1 µg daily requirements. but does not occur in fruit, cereals or vegetables.
The vitamin consists of a small group of compounds, the Cobalamin, which have the same basic structure.
A normal diet contains a large excess of B12 compared with daily needs.
In the stomach ,gastric enzymes release vitamin B. 12 from food and at gastric PH its binds to carrier protein called R protein .
B12 is combined with glycoprotein intrinsic factor (IF) which is synthesized by parietal cells.
The IF-B12 complex can then bind to specific surface receptors, cubilin, for IF in the distal ileum where B12 is absorbed.

Transport: the trasncobalamines

Vitamin B12 is absorbed into portal blood where it becomes attached to the plasma-binding protein trasncobalamines II (TC II) which delivers B12 to bone marrow and other tissues.
Although TCII is the essential plasma protein for transferring B12 into the cells of the body, TCII deficiency cause megaloblastics anemia because of failure of B12 to enter the marrow (and other cells) from the plasma but the serum B12 level in TCII deficiency is normal. This is because most of B12 in plasma is bound to another transport protein, TCI which is a glycoprotein largely synthesized by granulocytes and macrophages.

In myeloproliferative disorders where granulocytes production is greatly increased, the TCI and B12 levels in the serum both rise considerably.
Note: B12 bound to TCI does not transfer readily to marrow; it appears to be functionally dead.

Folate:

Folates are produced by plants and bacteria, hence dietary leafy vegetable, fruits, and animal protein (liver and kidney)are rich source.
An average western diet contains more than minimum daily intake of 50 µg but excess cooking destroys folates.

Absorption, transport:

Most dietary folates is present as polyglutamates, these are converted to monoglutamatee in the upper small bowel and actively transported into plasma.
Total body stores of folate are small and deficiency can occur in a matter of weeks. .

Biochemical basis for megaloblastics anemia:

Folate is an important substrate of and vitamin B12 a co-factor for generation of essential amino acid methionine from homocysteine .
This reaction produces tetrahydro-folate which is converted to thymidine monophosphate for incorporation into DNA. So deficiency of either vitamin B12 or folate will therefore produce high plasma levels of homocysteine and impaired DNA synthesis.

Causes of vitamin B12 deficiency:

1.Nutritional: especially in vegans.
2.Malabsorption:
- Gastric causes:- pernicious anemia.
  - congenital lack or abnormality of intrinsic factor.
  -Total or partial gastrectomy.
- Intestinal causes: - Intestinal stagnant loop syndrome-Jejunal diverticulosis., blind loop , stricture ,etc

- Chronic tropical sprue.
-ileal resection and Crohn s disease.
- Congenital selective mal absorption with proteinuria.
- Fish tapeworm.

Causes of folate deficiency:

1.Nutritional: Especially old age, institution, poverty, special diet.
2.Malabsorption:Tropical sprue, gluten-induced enteropathy, Crohn s disease., extensive Jejunal resection.
3. Excessive utilization:
- Physiological: pregnancy, lactation, prematurity.
-Pathological: -Hematological diseases e.g hemolytic anemia.
- Malignant disease e.g carcinoma, lymphoma, myeloma.
- Inflammatory diseases: Crohn s disease., tuberculosis, rheumatoid arthritis, etc…

4.Excess urinary folate loss: Active liver diseases, congestive heart failure.
5. Drugs: Anti-convulsants, sulfasalazine.
6. Mixed: Liver disease, alcoholism, intensive care.

Clinical features of megaloblastics anemia:

1. The onset is usually insidious with gradually progressive symptoms and signs of anemia.
2. Mild jaundice (lemon yellow tint).
3. Glossitis (beefy-red, sore tongue).
4. Angular stomatitis.

5. Mild symptoms of malabsorption with weight loss may be present caused by epithelial abnormalities.
6. Purpura as a result of thrombocytopenia.
7. In severe cases, Vitamin B12 neuropathy (sub-***acute combined degeneration of spinal cord***) developed. Neuropathy usually affecting the peripheral sensory nerves and posterior and lateral columns.

The neuropathy is symmetrical and affects the lower limbs more than the upper limbs.

The patients notice tingling in the feet, difficulty in walking and may fall over in the dark.

8. Neural tube defect (NTD): Folate or B12 deficiency in the mother predispose to NTD in the fetus.

Hematological finding:

-Decrease Hb, RBC, PCV, MCH, ***but*** normal MCHC.
-Increase MCV USUALLY >120fl
-Reticulocytes count low for degree of anemia.
-Serum ferritin elevated.
-Plasma LDH elevated often markedly due to hemolysis in marrow cells..
-Blood film will show:
-RBC: Macrocytic norm chromic, oval cells, teardrop cells.
-WBC: Are reduced in number (leucopenia) with hyper-segmented neutrophils
-Platelets: Are reduced in number.

So there is ***pancytopenia*** because all blood elements (i.e. RBC, WBC and Platelets) are reduce

-***Bone marrow aspiration***: To diagnose megaloblastic anemia we have to do bone marrow aspiration which will show the following:

-Hyper-cellular marrow with megaloblastics changes in Erythroid series..
-Erythroid hyperplasia, increase Erythroid precursors, with delayed in nuclear maturation.
-Giant metamyelocytes.
-Megakaryocytes are hyper polyploidy.

-***Special tests used for diagnosis of vitamin B12 and Folate deficiency***:

- Serum B12 assays.
- Serum and red cell folate.
- The deoxyuridine suppression test.
- Absorption tests using an oral dose of radioactive cobalt can used to distinguished malabsorption from inadequate diet.

When the test is repeated with an active IF preparation gastric lesions such as those associated with pernicious anemia can be distinguish from intestinal lesion.

- Schilling test: Absorption is most frequently measured indirectly by indirectly by the urinary excretion technique.

- Other tests include assign gastric function, testing for antibodies to gastric antigen, endoscopy

Managements of megaloblastics anemia:

-If a patients with severe megaloblastics anemia is very ill , the treatment must be started before vit.B12 and red cell folate results are available., that treatment should always include both folic acid and vitamin B12 .
The use of folic acid alone in the presence of vit.B12 deficiency may results in worsening of neurological deficits.
- Blood transfusion rarely needed except if angina or heart failure present.

Vitamin B12 deficiency:
Vitamin B12 deficiency treated with hydroxycobalamin 1000 µg IM for 6 doses 2 or 3 days apart., followed by maintenance therapy1000 µg every 3 months for life.
Response to treatments:

- The patient feels better after 24-48 hr of correct vitamins therapy with increase appetite and well-being.
- **Reticulocytes** count will peak by the 5^{th}-10^{th} day after starting the replacement therapy.
- **Hemoglobin** level will be rise by 1mg/dl every week until normalized.
-If an initial response is not maintained and the blood film is dimorphic , the patient may need additional iron therapy.
- The white cells and platelets counts become normal in 7-10 days and marrow is normoblastic in about 48 hr. although the giant metamyelocytes persists for up to 12 days.
- A sensory neuropathy may take 6-12 months to correct, long standing neurological damage may not improve.

Folate deficiency:
-Oral folic acid 5 mg daily for 3 weeks will treat acute deficiency and 5 mg once weekly is adequate maintenance therapy.
- Prophylactic folic acid in pregnancy prevents megaloblastosis in women at risk and reduce of fetal neural tube defect.
Prophylactics supplementation is also given in chronic hematological disease associated with reduced red cell lifespan.

Prophylactic therapy:
-Vitamin B12 is given to patients who have gastrectomy or ileal resection.
-Folic acid is given in pregnancy and all women of child-bearing age. Also folic acid give to patient undergoing chronic dialysis and with sever hemolytic anemia .
- Prophylactics supplementation is also given in chronic hematological disease associated with reduced red cell lifespan.

Pernicious anemia:

-Its type of megaloblastics anemia in which there is ***atrophy of stomach mucosa*** which will results in ***lack of intrinsic factors*** which will lead to ***defect in vitamin B12 absorption***.
-Its more common in females than males.
-Peak occurrence at 60 years.
-There may be associated with autoimmune diseases e.g. Vitiligo, Hashimatos diseases and Addison's diseases.
-The diseases most common in northern Europeans.
-Tend to occur in families.
-There is association with ***blood group A***, ***Blue eyes*** and ***early graying of hair*** and there is ***an increased incidence of carcinoma of stomach***.
-***90%*** of patients show ***parietal cell antibody*** in the serum.
-***50%*** have **type I** or **blocking** antibody to intrinsic factor.
-***35%*** have **type II** or **precipitating** antibodies which inhibits its Ileal binding sites.

Non- Megaloblastics anemia:

There are many non-megaloblastics causes of macrocytic anaemia. The exact mechanism creating large red cells in each of these conditions is not clear although increased lipid deposition on the red cell membrane or alternations of erythroblast maturation time in the marrow may be implicated.

Causes of macrocytosis other than megaloblastics anaemia:

1. Alcohol.
2. Liver disease.
3. Myxedema.
4. Myelodysplastic syndromes.
5. Cytotoxic drugs.
6. Aplastic anaemia.
7. Pregnancy.
8. Smoking.
9. Reticulocytosis.
10. Myeloma.
11. Neonatal.