Megaloblastic anemias

Objectives : After completing this lecture, the student will be able to:

1. Describe the criteria that define a macrocytic anemia as megaloblastic.
2. Compare and contrast the morphologic characteristics of megaloblasts and normoblasts in the bone marrow.
3. Differentiate red blood cell and white blood cell changes in the peripheral smear that are seen in the megaloblastic anemias.
4. Describe ineffective hematopoiesis as it relates to the megaloblastic process.
5. Describe the pathway of vitamin B12 and folic acid from ingestion through incorporation into the red blood cell.
6. Describe the clinical symptoms of a patient with megaloblastic anemia.
7. List the causes of vitamin B12 and folic acid deficiency.
8. Define pernicious anemia and its clinical and laboratory findings.
9. Describe the relevant laboratory tests used in the diagnosis of megaloblastic anemia.
10. Describe the treatments for the megaloblastic anemias.
11. Differentiate the anemias that are macrocytic but are not megaloblastic.

MACROCYTIC ANEMIAS AND THE MEGALOBLASTIC PROCESS

- The macrocytic anemias are a morphologic classification of anemias having mean corpuscular volume (MCV) greater than 100 fL, elevated mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin content (MCHC) within normal range.
- These anemias are termed macrocytic and normochromic.
- Broadly defined, the macrocytic anemias are divided into two categories: megaloblastic and nonmegaloblastic processes.
- If the source of the anemia is vitamin B12 or folic acid deficiency, the anemia is termed megaloblastic.
- If the source of the anemia is unrelated to a nutritional deficiency, the anemia is macrocytic but not megaloblastic.
- Vitamin B12 or folic acid deficiency leads to impaired DNA synthesis, a serious condition, and affects all readily dividing cells, skin cells, hematopoietic cells, and epithelial cells. The effects on the bone marrow, the peripheral smear, and the patient's quality of life are dramatic and substantive

RED BLOOD CELL PRECURSORS IN MEGALOBLASTIC ANEMIA

- Because megaloblastic processes damage DNA synthesis, nucleated cells are affected the most.
- There are multiple changes to white blood cells and red blood cells in the bone marrow structure that should be recognized and appreciated.
- The megaloblastic red blood cell precursors are larger, the nuclear structure is less condensed, and the cytoplasm is extremely basophilic or much bluer.
- There is asynchrony between the age of the nuclear material and the age of the cytoplasm, but this can best be appreciated by making a serious comparison of the nuclear and cytoplasmic material in megaloblastic precursor cells versus normoblastic precursor cells.

Consequences of ineffective erythropoiesis

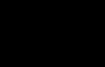
- Bone marrow destruction of erythroid precursors
- Lack of regeneration of bone marrow elements during anemic stress
- Lack of nRBCs in peripheral smear
- Lack of polychromasia in peripheral smear
- Reticulocytopenia
- Intramedullary hemolysis
- Increased bilirubin and LDH

VITAMIN B12 AND FOLIC ACID: THEIR ROLE IN DNA SYNTHESIS

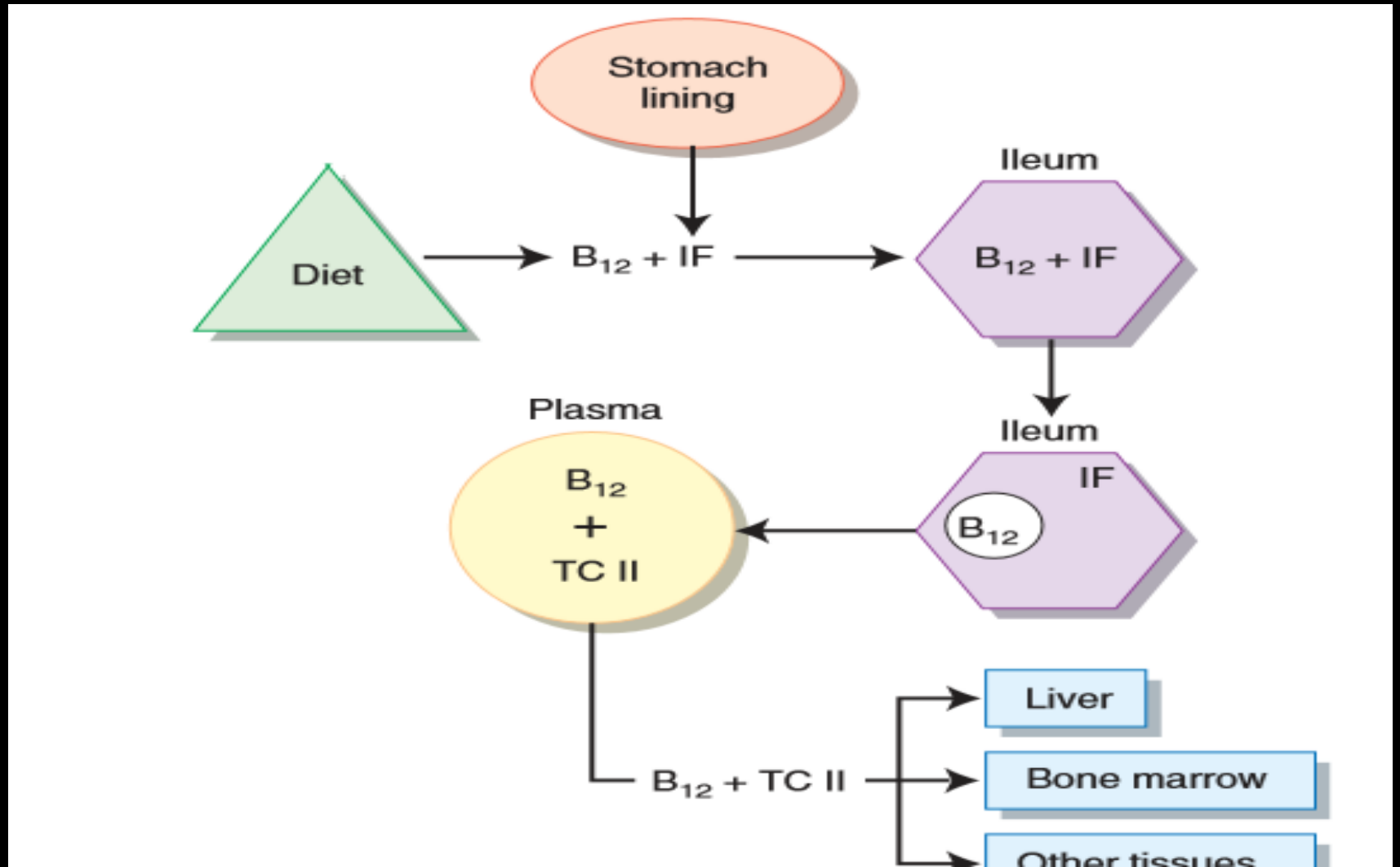
DNA synthesis is dependent on a key structure, thymidine triphosphate (TTP). This structure cannot be formed unless it receives a methyl group from methyl tetrahydrofolate or folic acid. Vitamin B12 is the cofactor responsible for transferring the methyl group to methyl tetrahydrofolate.1 Sufficient quantities of vitamin B12 and folic acid are key to the formation of TTP. If TTP cannot be synthesized, then it is replaced by deoxyuridine triphosphate (DTP). The synthesis of this component leads to nuclear fragmentation and destruction of cells and impaired cell division. For this reason, vitamin B12 and folic acid are essential elements in the DNA pathway.

NUTRITIONAL REQUIREMENTS, TRANSPORT, AND METABOLISM OF VITAMIN B12 AND FOLIC ACID

- Microorganisms and fungi are the main producers of vitamin B12, a group of vitamins known as cobalamins.
- This vitamin may also be embedded in liver, meat, fish, eggs, and dairy products.
- The recommended daily allowance of vitamin B12 is 2.0 µg/day with the daily diet providing approximately 5 to 30 µg/day and storage of 1 to 2 mg, in the liver.
- Dietary requirements will increase during pregnancy and lactation.
- Depletion of vitamin B12 stores takes years to develop.
- Folic acid, on the other hand, is readily available in green leafy vegetables, fruit, broccoli, and dairy products. The minimum daily requirement is 200 µg/day, a much higher requirement than that of vitamin B12, with body stores of 5 to 10 mg. in the liver. Folic acid is quickly depleted in a matter of months because the daily requirement is so much higher. Pregnant women are encourage to increase their folic acid intake because decreased folate may lead to neural tube defects.



Vitamin B12 processing



- Moving folic acid into the circulation and tissues occurs with a little more ease. Once folic acid is ingested and absorbed through the small intestine, it is reduced to methyl tetrahydrofolate through dihydrofolate reductase, an enzyme available in mucosal cells. It is the reduced form that is delivered to the tissues.
- Once inside the tissues, the methyl group is released to combine with homocysteine, an early precursor to DNA synthesis.
- Homocysteine is converted to methionine, an amino acid. If folate or vitamin B12 metabolism is flawed, homocysteine will accumulate and can potentially lead to thrombosis, a potential consequence to the hemostatic system that is just being realized.

CLINICAL FEATURES OF PATIENTS WITH MEGALOBLASTIC ANEMIA

Megaloblastic anemia is usually a disease of middle aged to older age with a high predilection for women. Severe anemia, in which the hemoglobin drops to 7 to 8 g/dL, is accompanied by symptoms of anemias such as shortness of breath, light-headedness, extreme weakness, and pallor. Patients may experience glossitis (sore or enlarged tongue), dyspepsia, or diarrhea. Evidence of neurological involvement may be seen with patients experiencing numbness, vibratory loss (paresthesias), difficulties in balance and walking, and personality changes. Vitamin B12 deficiency causes a demyelinizationof the peripheral nerves, the spinal column, and the brain, which can cause many of the more severe neurological symptoms such as spasticity or paranoia. Jaundice may be seen, because the average red cell life span in megaloblastic anemia is 75 days, a little more than one half of the average red cell life span of 120 days. The bilirubin level is elevated, and the lactate dehydrogenase (LDH) level is high, signifying hemolysis.

HEMATOLOGICAL FEATURES OF MEGALOBLASTIC ANEMIAS

- The CBC shows a pancytopenia (low white count, low red count, and low platelet count), although the platelet count may be only borderline low.
- Pancytopenia in the CBC combined with macrocytosis should raise the index of suspicion toward a megaloblastic process because few other conditions (aplastic anemia, hypersplenism) show this pattern.
- Red cell inclusions such as basophilic stippling and Howell-Jolly bodies may be observed. Howell-Jolly bodies formed from megaloblastic erythropoiesis are larger and more fragmented in appearance than normal Howell-Jolly bodies.
- There is a low reticulocyte count (less than 1%) and the RDW is increased, owing to schistocytes, targets, and teardrop cells. The blood smear in megaloblastic anemia is extremely relevant in the diagnosis and shows macrocytes, macro-ovalocytes, hypersegmented multilobed neutrophils, and little polychromasia with respect to the anemia.

Clinical Features

Many symptomless patients are detected through the finding of a raised mean corpuscular volume (MCV) on a routine blood count. The main clinical features in more severe cases are those of anemia.

Anorexia is usually marked, and there may be weight loss, diarrhea, or constipation. Glossitis, angular cheilosis, a mild fever in more severely anemic patients, jaundice (unconjugated), and reversible melanin skin hyperpigmentation also may occur with a deficiency of either folate or cobalamin.

Thrombocytopenia sometimes leads to bruising, and this may be aggravated by vitamin C deficiency or alcohol in malnourished patients.

The anemia and low leukocyte count may predispose to infections, particularly of the respiratory and urinary tracts. Cobalamin deficiency has also been associated with impaired bactericidal function of phagocytes.

Neurological sign & symptoms of vitamin B12 deficiency

Peripheral nerves

- Glove and stocking paraesthesiae
- Loss of ankle reflexes

Spinal cord

- Subacute combined degeneration of the cord
  - Posterior columns – diminished vibration sensation and proprioception
  - Corticospinal tracts – upper motor neuron signs

Cerebrum

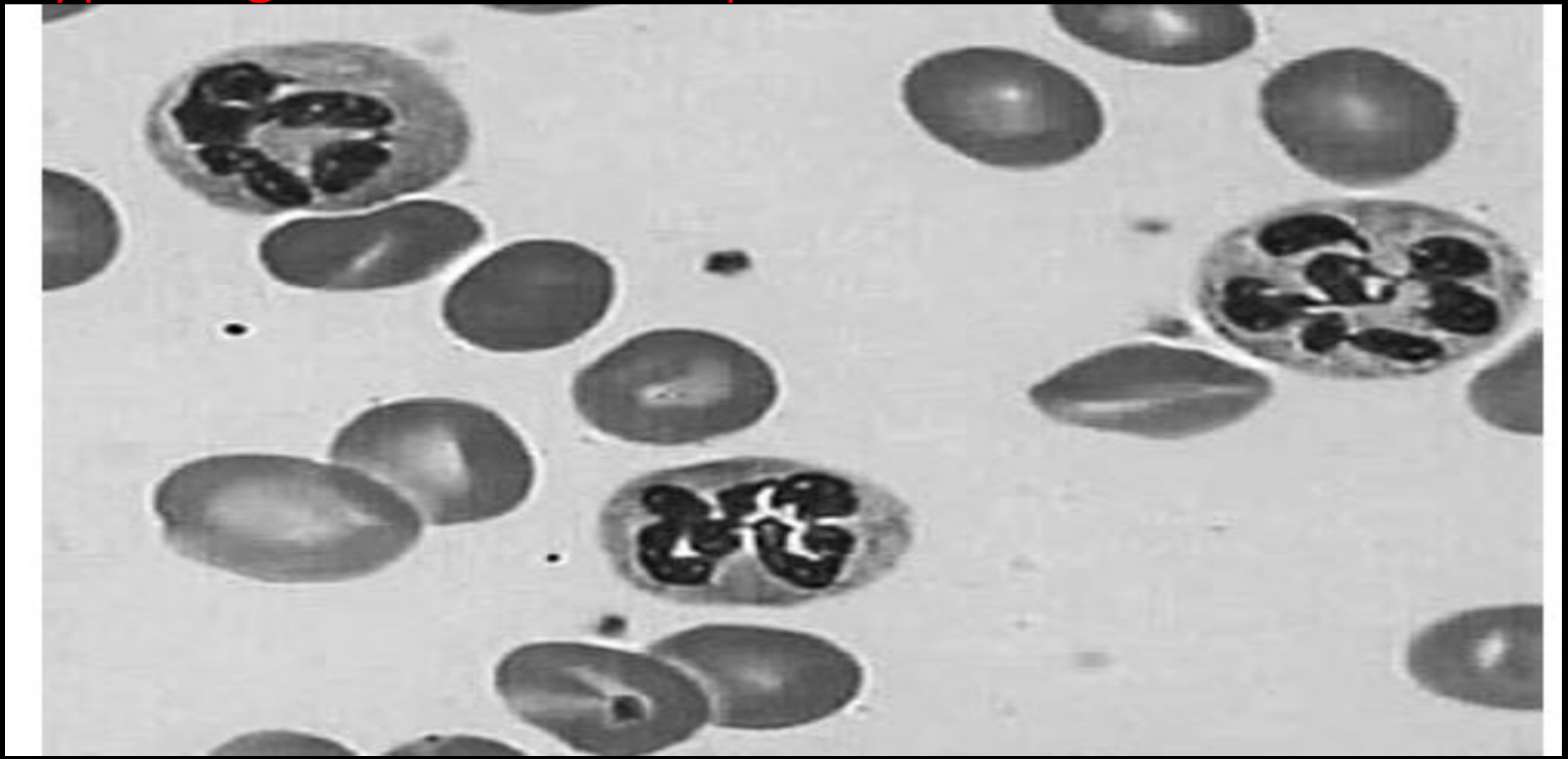
- Dementia
- Optic atrophy

Autonomic neuropathy

Causes of vitamin B12 deficiency

Pernicious anaemia	Commonest, due to autoimmune gastric atrophy resulting in loss of intrinsic factor production required for absorption of B_{12}. Incidence ↑ >40 years and often associated with other autoimmune problems, e.g. hypothyroidism.
Following total gastrectomy	May develop after major partial gastrectomy.
Ileal disease	Resection of ileum, Crohn's disease.
Blind loop syndromes	E.g. diverticulae or localized inflammatory bowel changes allowing bacterial overgrowth which then competes for available B_{12}
Fish tapeworm	*Diphyllobothrium latum*.
Malabsorptive disorders	Tropical sprue, coeliac disease.
Dietary deficiency	E.g. vegans

Hypersegmented neutrophil



Pernicious anaemia

- In the absence of intrinsic factor, less than 1% of dietary vitamin B12 is absorbed.
- This is an organ specific autoimmune disorder in which the gastric mucosa is atrophic, with loss of parietal cells causing intrinsic factor deficiency.
- 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 autoimmune disease (Hashimoto's thyroiditis, Graves' disease, vitiligo, hypoparathyroidism or Addison's disease; or a family history of these or pernicious anaemia.
- The finding of antiintrinsic 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 anaemia less likely but a positive result is not diagnostic.
- The Schilling test, involving measurement of absorption of radiolabelled B12 after oral administration before and after replacement of intrinsic factor, has fallen out of favour with the availability of autoantibody tests, greater caution in the use of radioactive tracers, and limited availability of intrinsic factor.

Causes of folate deficiency

Diet
• Poor intake of vegetables
Malabsorption
• e.g. Coeliac disease
Increased demand
• Cell proliferation, e.g. haemolysis • Pregnancy
Drugs*
• Certain anticonvulsants (e.g. phenytoin) • Contraceptive pill • Certain cytotoxic drugs (e.g. methotrexate)

*Usually only a problem in patients deficient in folate from another cause.

Management of megaloblastic anaemia

- If a patient with a severe megaloblastic anaemia is very ill and treatment must be started before vitamin 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 vitamin B12 deficiency may result in worsening of neurological deficits. Rarely, if severe angina or heart failure is present, transfusion can be used in megaloblastic anaemia.
- The cardiovascular system is adapted to the chronic anaemia present in megaloblastosis, and the volume load imposed by transfusion may result in decompensation and severe cardiac failure. In such circumstances, exchange transfusion or slow administration of 1 U of red cells with diuretic cover may be given cautiously

Vitamin B12 deficiency

- Vitamin B12 deficiency is treated with hydroxycobalamin 1000 µg IM for 6 doses 2 or 3 days apart, followed by maintenance therapy of 1000 µg every 3 months for life.
- 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; longstanding 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 reduces the risk of fetal neural tube defects.
- Prophylactic supplementation is also given in chronic haematological disease associated with reduced red cell lifespan (e.g. haemolytic anaemias).
- There is some evidence that supraphysiological supplementation (400 µg/day) can reduce the risk of coronary and cerebrovascular disease by lowering plasma homocysteine levels.
- This has led the US Food and Drug Administration to introduce fortification of bread, flour and rice with folic acid.