128 Megaloblastic Anemias

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The megaloblastic anemias are a group of disorders characterized by the presence of distinctive morphologic appearances of the developing red cells in the bone marrow. The marrow is usually hypercellular and the anemia is based on ineffective erythropoiesis. The cause is usually a deficiency of either cobalamin (vitamin B_{12}) or folate, but megaloblastic anemia may occur because of genetic or acquired abnormalities that affect the metabolism of these vitamins or because of defects in DNA synthesis not related to cobalamin or folate (Table 128-1). Cobalamin and folate absorption and metabolism are described next, followed by the biochemical basis, clinical and laboratory features, causes, and treatment of megaloblastic anemia.

COBALAMIN

Cobalamin (vitamin B_{12}) exists in a number of different chemical forms. All have a cobalt atom at the center of a corrin ring. In nature, the vitamin is mainly in the 2-deoxyadenosyl (ado) form, which is located in mitochondria. It is the cofactor for the enzyme methylmalonyl coenzyme A (CoA) mutase. The other major natural cobalamin is methylcobalamin, the form in human plasma and in cell cytoplasm. It is the cofactor for methionine synthase. There are also minor amounts of hydroxocobalamin to which methyl- and adocobalamin are converted rapidly by exposure to light.

DIETARY SOURCES AND REQUIREMENTS

Cobalamin is synthesized solely by microorganisms. Ruminants obtain cobalamin from the foregut, but the only source for humans is food of animal origin, e.g., meat, fish, and dairy products. Vegetables, fruits, and other foods of nonanimal origin are free from cobalamin unless they are contaminated by bacteria. A normal Western diet contains 5–30 μg of cobalamin daily. Adult daily losses (mainly in the urine and feces) are 1–3 μg (~0.1% of body stores), and because the body does not have the ability to degrade cobalamin, daily requirements are also about 1–3 μg. Body stores are of the order of 2–3 mg, sufficient for 3–4 years if supplies are completely cut off.

ABSORPTION

Two mechanisms exist for cobalamin absorption. One is passive, occurring equally through buccal, duodenal, and ileal mucosa; it is rapid but extremely inefficient, with <1% of an oral dose being absorbed by this process. The normal physiologic mechanism is active; it occurs through the ileum and is efficient for small (a few micrograms) oral doses of cobalamin, and it is mediated by gastric intrinsic factor (IF). Dietary cobalamin is released from protein complexes by enzymes in the stomach, duodenum, and jejunum; it combines rapidly with a salivary glycoprotein that belongs to the family of cobalamin-binding proteins known as haptocorrins (HCs). In the intestine, the haptocorrin is digested by pancreatic trypsin and the cobalamin is transferred to IF.

TABLE 128-1 CAUSES OF MEGALOBLASTIC ANEMIA

Cobalamin deficiency or abnormalities of cobalamin metabolism (see Tables 128-3, 128-4)
Folate deficiency or abnormalities of folate metabolism (see Table 128-5)
Therapy with antifolate drugs (e.g., methotrexate)
Independent of either cobalamin or folate deficiency and refractory to cobalamin and folate therapy:
Some cases of acute myeloid leukemia, myelodysplasia
Therapy with drugs interfering with synthesis of DNA (e.g., cytosine arabinoside, hydroxyurea, 6-mercaptopurine, azidothymidine [AZT])
Orotic aciduria (responds to uridine)
Thiamine-responsive

IF (gene at chromosome 11q13) is produced in the gastric parietal cells of the fundus and body of the stomach, and its secretion parallels that of hydrochloric acid. Normally, there is a vast excess of IF. The IF-cobalamin complex passes to the ileum, where IF attaches to a specific receptor (cubilin) on the microvillus membrane of the enterocytes. Cubilin also is present in yolk sac and renal proximal tubular epithelium. Cubilin appears to traffic by means of amnionless (AMN), an endocytic receptor protein that directs sublocalization and endocytosis of cubilin with its ligand IF-cobalamin complex. The cobalamin-IF complex enters the ileal cell, where IF is destroyed. After a delay of about 6 h, the cobalamin appears in portal blood attached to transcobalamin (TC) II.

Between 0.5 and 5 μg of cobalamin enter the bile each day. This binds to IF, and a major portion of biliary cobalamin normally is reabsorbed together with cobalamin derived from sloughed intestinal cells. Because of the appreciable amount of cobalamin undergoing enterohepatic circulation, cobalamin deficiency develops more rapidly in individuals who malabsorb cobalamin than it does in vegans, in whom reabsorption of biliary cobalamin is intact.

TRANSPORT

Two main cobalamin transport proteins exist in human plasma; they both bind cobalamin—one molecule for one molecule. One HC, also known as TC I, is closely related to other cobalamin-binding HCs in milk, gastric juice, bile, saliva, and other fluids. The gene *TCNL* is at chromosome 11q11-q12.3. These HCs differ from each other only in the carbohydrate moiety of the molecule. TC I is derived primarily from the specific granules in neutrophils. Normally, it is about two-thirds saturated with cobalamin, which it binds tightly. TC I does not enhance cobalamin entry into tissues. Glycoprotein receptors on liver cells are involved in the removal of TC I from plasma, and TC I may play a role in the transport of cobalamin analogues (which it binds more effectively than IF) to the liver for excretion in bile.

The other major cobalamin transport protein in plasma is transcobalamin, also known as TC II. The gene is on chromosome 22q11-q13.1. As for IF and HC, there are nine exons. The three proteins are likely to have a common ancestral origin. TC II is synthesized by liver and by other tissues, including macrophages, ileum, and vascular endothelium. It normally carries only 20–60 ng of cobalamin per liter of plasma and readily gives up cobalamin to marrow, placenta, and other tissues, which it enters by receptor-mediated endocytosis involving the TC II receptor and megalin (encoded by the *LRP-2* gene). The TC II cobalamin is internalized by endocytosis via clathrin-coated pits; the complex is degraded, but the receptor probably is recycled to the cell membrane as is the case for transferrin. Export of "free" cobalamin is via the ATP-binding cassette drug transporter alias multidrug resistance protein 1.

FOLATE

DIETARY FOLATE

Folic (pteroylglutamic) acid is a yellow, crystalline, water-soluble substance. It is the parent compound of a large family of natural folate compounds, which differ from it in three respects: (1) they are partly or completely reduced to di- or tetrahydrofolate (THF) derivatives, (2) they usually contain a single carbon unit (Table 128-2), and (3) 70–90% of natural folates are folate-polyglutamates.

Most foods contain some folate. The highest concentrations are found in liver, yeast, spinach, other greens, and nuts (>100 μg/100 g). The total folate content of an average Western diet is ~250 μg daily, but the amount varies widely according to the type of food eaten and the method of cooking. Folate is easily destroyed by heating, particularly in large volumes of water. Total body folate in the adult is ~10 mg, with the liver containing the largest store. Daily adult requirements are ~100 μg, and so stores are sufficient for only 3–4 months in normal adults and severe folate deficiency may develop rapidly.

ABSORPTION

Folates are absorbed rapidly from the upper small intestine. The absorption of folate polyglutamates is less efficient than that of monoglutamates; on average, ~50% of food folate is absorbed. Polyglutamate