Vitamin-Related Anemias

An international symposium on vitamin-related anemias, organized by R. E. Olson and sponsored by Hoffman-La-Roche, Inc., was held in honor of W. B. Castle on 27 and 28 May 1968, at Skytop, Pa. Castle was the first to demonstrate [(W. B. Castle, *Amer. J. Med. Sci.* **178**, 748 (1929)] that a thermolabile substance in normal gastric juice, together with a substance extrinsic to the body, is necessary for hematologic remission in patients with pernicious anemia. He designated the substance of the gastric juice intrinsic factor, in contrast to the extrinsic factor which is derived from the food.

P. A. Marks reported that work in mice, which paralleled studies in man, indicated that in early fetal development the blood islands of the yolk sac produce very primitive hemoglobins that are represented by Gower (ε_4) and Gower II ($\alpha_2\varepsilon_2$) in man and E_1 (X_2Y_2), E_2 (α_2Y_2), and E_3 (α_2Z_2) in mice. The liver, spleen, and bone marrow begin to function later in gestation and produce fetal hemoglobin F ($\alpha_2\gamma_2$) and adult hemoglobin A ($\alpha_2\beta_2$). These hemoglobins thus reflect the cell type responsible for their biosynthesis. Regulation at the level of DNA synthesis and messenger RNA synthesis appears to be a major factor in determining the amounts of different hemoglobins formed by the same and different cell lines.

The function of pyridoxal-5′-phosphate in early steps of heme synthesis was discussed by D. Shemin. δ-Aminolevulinic acid (ALA) synthetase, which appears to be under both genetic and feedback control, is a regulatory enzyme for heme synthesis in both bacteria and mammals. The ALA dehydrase, which links two δ-aminolevulinic acid molecules together, was discussed, and an attractive reaction mechanism was presented. The mitochondrial membrane may limit synthesis of porphyrin under some conditions since the formation of δ-aminolevulinic acid occurs in mitochondria but the dehydrase reaction occurs in the cell sap.

The role of folic acid coenzymes in the biosynthesis of purines and pyrimidines was extensively reported by F. M. Huennekens (Scripps Clinic and Research Foundation, La Jolla). H. Weissbach and R. T. Taylor (National Heart Institute) reviewed the metabolic role of vitamin B_{12}. Vitamin B_{12} coenzyme catalyzes rearrangements in molecules in which both alkyl groups and hydrogen may move. The methylation of homocysteine to methionine by N^5-methyl tetrahydrofolate is an example. The methylation of deoxyuridylic acid, on the other hand, involves a transfer of the hydroxymethyl group from folic acid to acceptor with a simultaneous reduction involving the oxidation of tetrahydrofolate to dihydrofolate. The dihydrofolate is then reduced to tetrahydrofolate by a separate enzyme, dihydrofolate reductase, which has been characterized by Huennekens. This enzyme is markedly inhibited by antifolate compounds and is greatly increased in leukemic white cells. Whether the ribonucleotide reductase of human bone marrow is dependent on vitamin B_{12} is still an unanswered question according to Beck (Harvard Medical School), who discussed deoxyribonucleotide synthesis (in bacteria and animal cells) and the role of vitamin B_{12} in erythropoiesis. If enzymological studies can demonstrate a clear-cut dependency of that reductase on vitamin B_{12} in animals (Beck has already some indications for it), a direct influence of vitamin B_{12} on the synthesis of deoxyribonucleic acid would become evident. If not, the previously mentioned postulation of an indirect influence of vitamin B_{12} on ribonucleic synthesis by means of folic acid (Weissbach) would receive further support.

The function of vitamin E in hemopoiesis and hemolysis was discussed by M. K. Horwitt and C. D. Fitch (St. Louis University School of Medicine). A deficiency of vitamin E causes anemia in the rhesus monkey many months after the appearance of creatinuria and other signs of this deficiency. Abnormal precursors of red cells accumulate in the bone marrow, and incorporation of thymidine into the nuclei of these precursors is increased, which suggests that "marrow arrest" is the cause of the failure to manufacture erythrocytes normally. Studies with chromium-54 indicate that survival of erythrocytes is also decreased. This anemia responds promptly to vitamin E, with a marked reticulocytosis, disappearance of morphological abnormalities in the bone marrow, and normalization of hemoglobin concentration.

C. A. Finch (University of Washington School of Medicine) reported that, as determined by use of Fe^{59}, about 20 mg of Fe pass through the plasma compartment in 1 day, whereas 1 mg of dietary iron is absorbed. In aplastic anemia there is no erythroid marrow to utilize labeled iron, which results in a direct uptake of the radioactivity from the plasma transferrin by the liver.

Many aspects of folic acid deficiency in man were discussed by V. Herbert (Mt. Sinai School of Medicine). These included the increased folate requirement in pregnancy and hyperthyroidism; frequency of folate deficiency (especially in economically disadvantaged people of the world); etiology of folate deficiency (inadequate ingestion, absorption, or utilization, and increased excretion); blood and nerve damage in folate deficiency; laboratory diagnosis, and the treatment of folate deficiency. In his report on pernicious anemia and its variants, F. Thedering (Pius Hospital, Oldenburg, Germany) reviewed mainly the known causes and pathogenesis of pernicious anemia, that is, vitamin B_{12} deficiency caused by lack of intrinsic factor (hereditary lack, antibodies against intrinsic factor, gastric atrophy, and so forth); inadequate intake or malabsorption of vitamin B_{12}; infestation with the fish tapeworm; and the blind loop syndrome. When antibodies against intrinsic factor or the parietal cells of the gastric mucosa are found, a repeated, or even lifelong, therapy with B_{12} becomes necessary.

In their review of the pyridoxine-responsive anemias in man J. W. Harris and D. L. Horrigan (Western Reserve University) reported two cases of patients with pyridoxine-responsive anemia accompanied by sideroblastosis. One patient, observed since 1956, showed a normalization of the blood after prolonged pyridoxine treatment. When the pyridoxine therapy was withdrawn, a progressive decrease of erythrocytes and concentration of serum folate were observed, and morphological abnormalities in the marrow and peripheral blood become evident. In the second patient, under study for 14 years, a previously unrecognized metabolic relation in erythropoiesis between pyridoxine, tryptophan, and indolic substances derived from mammalian liver is now indicated.

F. E. Viteri and J. Alvarado (Institute of Nutrition of Central America and Panama) pointed out that the

anemia of protein-calorie malnutrition in children is often a multiple etiology and may reflect simultaneously occurring deficiencies in hemopoietic substances in addition to deficiencies in protein and calories. Thus, hematologic recovery is complete only if the supply of folic acid, iron, vitamin B_{12}, riboflavin, and pyridoxine is adequate. In the type of protein-calorie malnutrition seen in Central America, it appears that the complicating concurrent multiple deficiency diseases are not as prominent as in other parts of the world. In Guatemalan children, the red cell mass is reduced in proportion to lean body mass and was repaired during therapy in proportion to lean body tissue. Interaction between protein anabolism and stores of other required nutrients such as iron and folic acid was emphasized; there generally is a decrease in the plasma concentrations of these substances during the brisk anabolism of hemoglobin.

Erythropoietin, an endocrine-like substance that is capable of stimulating red cell production in mammals, has not been fully characterized but it appears to be a glycoprotein of relatively low molecular weight (70,000) synthesized by the kidney in response to hypoxia or anemia (C. Gurney, Rutgers). It is believed that erythropoietin stimulates the synthesis of DNA, RNA, and δ-aminolevulinic acid, a precursor of heme in hemopoietic tissues.

E. V. Cox (Royal Berkshire Hospital) reported that normocytic or macrocytic anemia occurs in patients with scurvy. The bone marrow in these patients appeared to be macronormoblastic and hypercellular but not megaloblastic. The controversial question of whether vitamin C is a primary agent in hemopoiesis was discussed. There is conflicting evidence that other vitamin deficiencies may coexist in scurvy and complicate the interpretation of the response of the anemia of scurvy to vitamin C. Nonetheless, there are uncomplicated cases of anemia in scurvy that do respond to vitamin C. The data suggest that anemia of scurvy has more than one cause and that ascorbic acid may potentiate the use of other nutrients required in hemopoiesis.

W. J. Darby (Vanderbilt University) reviewed the tocopherol-responsive anemias in man which have been observed in infants with protein-calorie malnutrition in Jordan, Nigeria, and Thailand. The amount of tocopherol in serums of children with this anemia range from 0.2 to 0.6 mg/100 ml percent, a value considered low but not deficient when compared with animal studies. In some investigations, a full reticulocyte response accompanied by hemoglobin synthesis was observed. In others studies, particularly those in South India, where megaloblastic anemia is common, vitamin E causes a reticulocyte response without net hemoglobin synthesis. In contrast to kwashiorkor, L. Barness (University of Pennsylvania) has observed that vitamin E arrests the hemolytic anemia observed in premature infants, reduces reticulocyte counts, and extends the life of the red cell.

All of the papers presented in this symposium will be edited by R. S. Harris and published in volume 26 (1968) of *Vitamins and Hormones.*

F. WEBER
Department of Vitamin and Nutritional Research, F. Hoffman-La Roche & Co., Ltd., Basel, Switzerland

ROBERT E. OLSON
Department of Biochemistry, St. Louis University School of Medicine, St. Louis, Missouri