ERNICIOUS anemia is the most common cause of vitamin B₁₂ deficiency. Vitamin B₁₂ deficiency has many causes; the term “pernicious anemia” applies only to the condition associated with chronic atrophic gastritis. A recent population survey revealed that 1.9 percent of persons more than 60 years old have undiagnosed pernicious anemia. Earlier studies suggested that pernicious anemia is restricted to Northern Europeans. However, subsequent studies have reported the disease in black and Latin-American subjects, with an earlier age of onset in black women. Although the disease is silent until the end stage, the underlying gastric lesion can be predicted many years before anemia develops.

Pernicious anemia was first described by Thomas Addison in 1849. The anemia was linked to the stomach by Austin Flint in 1860 and named pernicious anemia soon thereafter. Successful treatment of the anemia with cooked liver suggested that it was caused by the lack of an extrinsic factor that was found in liver (later identified as vitamin B₁₂) and an intrinsic factor in gastric juice. Although pernicious when first discovered, the disease is now controlled by treatment with vitamin B₁₂. The discovery of a serum inhibitor of intrinsic factor (later found to be an autoantibody to intrinsic factor) and of autoantibodies to parietal cells laid the foundation for the immunologic explanation of the underlying gastritis that causes pernicious anemia.

GASTRIC PATHOLOGICAL FINDINGS

There are three regions of the stomach: the fundus and the body, both of which contain acid-secreting gastric parietal cells and pepsinogen-secreting zymogenic cells, and the antrum, which contains gastrin-producing cells. Chronic atrophic gastritis is recognized macroscopically by the loss of gastric mucosal folds and thinning of the gastric mucosa. It can be classified into two types according to whether or not the lesion affects the gastric antrum (Table 1). Type A (autoimmune) gastritis involves the fundus and body of the stomach and spares the antrum, whereas type B (nonautoimmune) gastritis involves the antrum as well as the fundus and body. Type A gastritis is associated with pernicious anemia, autoantibodies to gastric parietal cells and to intrinsic factor, achlorhydria, low serum pepsinogen I concentrations, and high serum gastrin concentrations, the latter resulting from hyperplasia of gastrin-producing cells. Type B gastritis is usually associated with Helicobacter pylori infection and low serum gastrin concentrations, because of destruction of the gastrin-producing cells associated with antral gastritis.

Histopathological Findings

Gastric biopsy specimens from patients with pernicious anemia show a mononuclear cellular infiltrate in the submucosa extending into the lamina propria between the gastric glands (Fig. 1). The cellular infiltrate includes plasma cells, T cells, and a large non-T-cell population (probably B cells). The infiltrating plasma cells contain autoantibodies to the parietal-cell antigen and to intrinsic factor. Extension of the cellular infiltrate into the mucosa is accompanied by degenerative changes in parietal
cells and zymogenic cells. In the fully established lesion, there is marked reduction in the number of gastric glands, and the parietal cells and zymogenic cells disappear and are replaced by mucus-containing cells (intestinal metaplasia).

**NATURAL HISTORY**

The progression of type A chronic atrophic gastritis to gastric atrophy and clinical anemia is likely to span 20 to 30 years. The presence of gastric parietal-cell antibodies in the serum is predictive of the presence of autoimmune gastritis. The pathologic lesion and the anemia can be reversed by treatment with corticosteroids or azathioprine. These observations suggest that precursor cells present in the stomach can differentiate into parietal and zymogenic cells if further autoimmune destruction is controlled. This suggestion is supported by studies of mice with autoimmune gastritis.

**IMMUNOPATHOGENESIS OF GASTRITIS**

**Gastric Parietal-Cell H⁺/K⁺-ATPase**

The pathologic process associated with type A gastritis appears to be directed toward the gastric parietal cells. The pathologic lesion is restricted to the parietal-cell–containing fundus and body regions of the stomach. Parietal cells are lost from the gastric mucosa, and autoantibodies to parietal cells and to their secretory product, intrinsic factor, are present in the serum and in gastric juice.

A major breakthrough in our understanding of the pathogenesis of type A gastritis was the demonstration that gastric H⁺/K⁺-ATPase is the antigen recognized by parietal-cell autoantibodies. This ATPase belongs to a family of electroneutral P-type ATPases that includes Na⁺/K⁺-ATPase and Ca²⁺-ATPase. These enzymes have a highly conserved catalytic (α) subunit that is phosphorylated during...
reaction cycles. Gastric H^+/K^+–ATPase is responsible for secretion of hydrogen ions by parietal cells in exchange for potassium ions (Fig. 2). This enzyme is the major protein of the membrane lining the secretory canaliculi of parietal cells.\(^{20,22}\) Autoantibodies to parietal cells bind to both the 100-kd catalytic (\(\alpha\)) subunit and the 60-to-90-kd glycoprotein (\(\beta\)) subunit of gastric H^+/K^+–ATPase.\(^{17}\)

Although parietal-cell autoantibodies can fix complement\(^{23}\) and lyse parietal cells in vitro,\(^{24}\) it is unlikely that these autoantibodies are pathogenic in vivo, because gastric H^+/K^+–ATPase is not accessible to circulating antibodies. The importance, if any, of an early observation that passive transfer of parietal-cell autoantibodies to rats resulted in reduction in parietal-cell mass without an inflammatory response is therefore uncertain.\(^{25}\) A report describing autoantibodies that bind to the gastrin receptor\(^{26}\) was not confirmed.\(^{27}\) The results of studies showing reactivity of parietal-cell autoantibodies with the surface membranes of parietal cells in vitro\(^{28,29}\) may be explained by the loss of cell polarity after cellular dissociation. Gastric H^+/K^+–ATPase appears to be the only parietal-cell antigen recognized by parietal-cell autoantibodies, because immunoblotting and immunoprecipitation experiments show reactivity only with the two subunits of this ATPase.\(^{14-18}\)

**Murine Models of Autoimmune Gastritis**

The identification of gastric H^+/K^+–ATPase as the target of parietal-cell autoantibodies raises the question of the role of the ATPase in the immunopathogenesis of the gastric lesion. Although this question has not been answered for pernicious anemia, studies in mice suggest that the lesion of autoimmune gastritis is initiated by CD4 T cells that recognize the \(\beta\) subunit of gastric H^+/K^+–ATPase.

Organ-specific autoimmune disease, including gastritis, develops in susceptible strains of mice after neonatal thymectomy (Fig. 3, upper panel).\(^{30-32}\) Gastritis also develops in neonatal mice treated with cyclosporine\(^{33}\) and in adult mice after thymectomy combined with irradiation,\(^{34}\) cyclophosphamide treat-
Lesions are predominantly macrophages and CD4+ zymogenic cells. The mononuclear cells in early gastritis are characterized by submucosal infiltration of lymphocytes, or immunization with murine gastric H+/K+–ATPase. Either thymectomy combined with cyclophosphamide treatment, or immunization of adult mice results in gastritis and serum autoantibodies to gastric H+/K+–ATPase.

Transgenic expression of the β subunit of gastric H+/K+–ATPase in the thymus prevents gastritis induced by neonatal thymectomy, by adult thymectomy combined with cyclophosphamide treatment, or by immunization with murine gastric H+/K+–ATPase (Fig. 3, lower panel). These observations suggest that the pathogenic T cells have been rendered tolerant after encountering the β subunit in the thymus. The α subunit, which is present in the normal thymus, does not appear to have a role in the initiation of gastritis but may have a role in its perpetuation. A single injection of neutralizing anti–interferon-γ antibody prevents the development of gastritis, implicating this Th1-type cytokine in its genesis.

Taken together, these observations suggest that interferon-γ–secreting Th1-type CD4 T cells are important in the pathogenesis of murine autoimmune gastritis (Fig. 4). Whether this is also the case for the gastritis of pernicious anemia in humans is not known. The gastric environment appears to be important for the genesis of the lesion, because transgenic expression of the β subunit of H+/K+–ATPase in pancreatic islets does not induce a destructive insulitis after neonatal thymectomy.

**Tolerance of and Autoimmunity to Gastric H+/K+–ATPase**

Murine autoimmune gastritis occurs only when pathogenic T cells are transferred to immunocompromised mice. This observation, together with the induction of gastritis by thymectomy, immunosuppressive drugs, and irradiation, suggests that pathogenic T cells expand only in a lymphopenic host. Expansion of these pathogenic T cells can be prevented by transfer of normal adult CD4 T cells. The pathogenic CD4 T cells are probably “resting” T cells, because they do not express the T-cell–activation marker CD25, whereas disease-preventing CD4 T cells express CD25. Furthermore, gastritis can be induced in mice after the administration of antibodies to CD25. The mechanisms that prevent the activation and expansion of “resting” pathogenic T cells and their homing to the stomach in normal subjects are not known. The simplest explanation is competition for space by CD4 T cells within defined lymphoid compartments, a process that appears to be under as yet undefined homeostatic control.
In both human and murine autoimmune gastritis, zymogenic cells are lost together with parietal cells from the gastric mucosa. There is no evidence of an autoimmune reaction directed toward zymogenic cells. Therefore, the loss of these cells is probably secondary to the primary autoimmune reaction directed toward parietal-cell $H^+/K^-$-ATPase. The induction of autoimmune gastritis by direct immunization with gastric $H^+/K^-$-ATPase in Freund’s adjuvant is also characterized by loss not only of parietal cells but also of zymogenic cells.

**Mechanisms of Vitamin B$_{12}$ Malabsorption**

Intrinsic factor is a 60-kd glycoprotein produced by gastric parietal cells that avidly binds dietary vitamin B$_{12}$. The vitamin B$_{12}$-intrinsic factor complex is carried to the terminal ileum, where it is absorbed after binding to intrinsic-factor receptors on the luminal membranes of ileal cells. Malabsorption of vitamin B$_{12}$ in patients with pernicious anemia is due to intrinsic-factor deficiency. Two mechanisms are responsible. First, the progressive destruction and eventual loss of parietal cells from the gastric mucosa lead to failure of intrinsic-factor production. Indeed, the severity of the gastric lesion correlates with the degree of impaired secretion of intrinsic factor and the reduction in vitamin B$_{12}$ absorption. Second, blocking autoantibodies present in the gastric juice can bind to the vitamin B$_{12}$-binding site of intrinsic factor, thereby preventing the formation of the vitamin B$_{12}$-intrinsic factor complex. Vitamin B$_{12}$ is required for DNA synthesis. Therefore, the major organs affected by vitamin B$_{12}$ deficiency are those in which cell turnover is rapid, such as the bone marrow and the gastrointestinal tract.

**Predisposing Genetic Factors**

A genetic predisposition to pernicious anemia is suggested by the clustering of the disease and of gastric autoantibodies in families, and by the association of the disease and gastric autoantibodies with the autoimmune endocrinopathies. There are reports of a number of white families with a high frequency of pernicious anemia over several generations. About 20 percent of the relatives of patients with pernicious anemia have pernicious anemia. These relatives, especially first-degree female relatives, also have a higher frequency of gastric autoantibodies than normal subjects. Concordance with respect to pernicious anemia has been observed in 12 sets of monozygotic twins, implicating a strong genetic predisposition to development of the disease. In contrast to some other autoimmune diseases, there is little evidence of an association between pernicious anemia and particular molecules of the major histocompatibility complex.

**Association with Other Autoimmune Diseases**

Pernicious anemia may be associated with autoimmune endocrinopathies and antireceptor autoimmune diseases. These diseases include chronic autoimmune thyroiditis (Hashimoto’s thyroiditis), insulin-dependent diabetes mellitus, Addison’s disease, primary ovarian failure, primary hypoparathyroidism, Graves’ disease, vitiligo, myasthenia gravis, and the Lambert–Eaton syndrome.

**Clinical Presentation**

Anemia

Animal products are the primary dietary source of vitamin B$_{12}$. The recommended daily dietary allowance of the vitamin is 2 μg. The average Western
The onset and progression of pernicious anemia are slow. The median age at diagnosis is 60 years. Slightly more women than men are affected. The usual presentation is with symptoms of anemia; asymptomatic patients can be identified by routine hematologic investigations.

Gastrointestinal Manifestations

Vitamin B₁₂ deficiency results in several abnormalities of the digestive tract. The tongue is usually smooth and beefy red because of atrophic glossitis. Megaloblastosis of the epithelial cells of the small intestine may result in diarrhea and malabsorption.

Neurologic Complications

Vitamin B₁₂ deficiency may cause peripheral neuropathy and lesions in the posterior and lateral columns of the spinal cord (subacute combined degeneration) and in the cerebrum. These lesions progress from demyelination to axonal degeneration and eventual neuronal death. These are serious complications, because they may not be reversed after replacement therapy with vitamin B₁₂. The most frequent manifestations of peripheral neuropathy are paresthesias and numbness. The manifestations of a lesion in the spinal cord are a mixture of signs of a posterior column lesion (loss of vibration and position sense, and sensory ataxia with positive Romberg’s sign) and those of a lateral column lesion (limb weakness, spasticity, and extensor plantar responses). Cerebral manifestations range from mild personality defects and memory loss to frank psychosis (“megaloblastic madness”).

Gastric Complications

Intestinal metaplasia is a risk factor for adenocarcinoma. Achlorhydria and bacterial overgrowth may also lead to the formation of carcinogenic nitrosamines. Population-based studies have revealed an excess risk of gastric carcinoma as well as gastric carcinoid tumors in patients with pernicious anemia. The gastric carcinoid tumors are probably due to hypergastrinemia. The evolution of these endocrine cells from hyperplasia to neoplasia has been attributed to the trophic action of gastrin. In a recent population-based cohort study in Sweden, the risk of gastric carcinoma was increased 3 times and that of gastric carcinoid tumors 13 times in patients with pernicious anemia. The prevalence of gastric carcinoma in patients with pernicious anemia is 1 to 3 percent, and 2 percent of patients with gastric carcinoma have pernicious anemia. One study suggested that regular endoscopic surveillance is warranted in patients with pernicious anemia.

Pernicious Anemia and Immunodeficiency

Pernicious anemia associated with common variable immunodeficiency and low serum immunoglobulin concentrations or with selective IgA deficiency should be distinguished from classic pernicious anemia. It occurs in younger patients and has the features of a type B gastritis.

Childhood Pernicious Anemia

Childhood pernicious anemia is also not associated with chronic atrophic gastritis or achlorhydria but is the result of a genetically determined failure to secrete intrinsic factor or the secretion of a defective intrinsic factor.

LABORATORY DIAGNOSIS

Hematologic Studies

In established megaloblastic anemia, examination of the peripheral blood reveals macrocytosis with hypersegmented polymorphonuclear leukocytes (Fig. 5), anemia, leukopenia, and thrombocytopenia or pancytopenia. Examination of bone marrow reveals megaloblasts and large myeloid precursors (“giant metamyelocytes”). Examination of the marrow is not indicated if the diagnosis is unequivocal. Vitamin B₁₂ deficiency as the cause of megaloblastic anemia is established by a low serum vitamin B₁₂ concentration and normal serum folate concentration.

A Schilling test will confirm that the vitamin B₁₂ deficiency is the result of intestinal malabsorption due to intrinsic-factor deficiency. In patients with pernicious anemia, urinary excretion of orally administered vitamin B₁₂ is low, and it increases if vitamin B₁₂ is administered with intrinsic factor. A simpler test is measurement of serum holotranscobalamin II, the circulating protein that delivers vitamin B₁₂ to cells. In patients with vitamin B₁₂ deficiency, serum concentrations of holotranscobalamin II fall before those of vitamin B₁₂.

Figure 5. Peripheral-Blood Film Showing Macrocytic Red Cells and a Hypersegmented Polymorphonuclear Leukocyte from a Patient with Pernicious Anemia.

Photomicrograph courtesy of Dr. Alison Street, Haematology Unit, Pathology Services, Alfred Hospital, Melbourne, Australia.
Serologic Studies

Serum antibodies to gastric parietal cells can be detected by indirect immunofluorescence with unfixed, air-dried, frozen sections of mouse stomach in which the antibodies stain parietal cells. Mouse stomachs are preferable to rat stomachs because the latter may give false positive heterophile reactions.58 These autoantibodies are found in about 90 percent of patients with pernicious anemia but also in about 30 percent of nonanemic first-degree relatives of patients with pernicious anemia and in patients with autoimmune endocrinopathies. In normal subjects there is an age-related increase in the prevalence of parietal-cell autoantibodies, from 2.5 percent in the third decade to 9.6 percent in the eighth decade.59 The explanations for the seronegative results in 10 percent of patients with pernicious anemia include faulty diagnosis, complete binding of antibody to antigen so that none is circulating at the time of measurement, disappearance of antibody because of disappearance of the antigen, or failure of production of the antibody.

Two types of autoantibodies to intrinsic factor have been described.60 Type I autoantibodies block the binding of vitamin B\textsubscript{12} to intrinsic factor. They are demonstrable in the serum of about 70 percent of patients with pernicious anemia. Type II autoantibodies bind to a site remote from the vitamin B\textsubscript{12}—binding site, are found in the serum of about 35 to 40 percent of patients, and rarely occur in the absence of the first type of antibody. However, the use of a sensitive enzyme-linked immunosorbent assay for the detection of both autoantibodies has shown that type II autoantibodies appear to be more common than previously reported.61 Both types of autoantibodies can be detected more frequently in gastric juice than in the serum. The demonstration of circulating intrinsic-factor autoantibodies is almost diagnostic of type A gastritis and pernicious anemia.

Gastric Biopsy, Achromia, and Serum Pepsinogen Concentrations

The presence of type A chronic atrophic gastritis can be confirmed by gastric biopsy. Total (pentagastrin-resistant) achlorhydria, the direct result of the loss of gastric parietal cells, is diagnostic of pernicious anemia because it is the only gastric lesion that results in total achlorhydria. Hypergastrinemia is the result of sparing of the antrum and stimulation of the gastrin-producing C cells by achlorhydria. A low serum pepsinogen I concentration is the result of the destruction of the chief cells.

TREATMENT

The standard treatment is regular monthly intramuscular injections of at least 100 \(\mu\)g of vitamin B\textsubscript{12} to correct the vitamin deficiency.52 This treatment corrects the anemia and may correct the neurologic complications if given soon after their onset. It has been suggested that elderly patients with gastric atrophy should take tablets containing 25 \(\mu\)g to 1 mg of vitamin B\textsubscript{12} daily to prevent vitamin B\textsubscript{12} deficiency.52 This recommendation is based on the observation that about 1 percent of vitamin B\textsubscript{12} is absorbed by mass action in the absence of intrinsic factor.

CONCLUSIONS

Pernicious anemia is the end stage of type A chronic atrophic (autoimmune) gastritis. The gastritis results in the loss of parietal cells in the fundus and body of the stomach. The loss of these cells is associated with the failure of intrinsic-factor production and results in vitamin B\textsubscript{12} deficiency and megaloblastic anemia. An autoimmune basis for the gastritis is supported by the presence of mononuclear-cell infiltration into the gastric mucosa with loss of parietal and zymogenic cells, autoantibodies to parietal cells and intrinsic factor, regeneration of parietal and zymogenic cells after therapy with corticosteroids or immunosuppressive drugs, familial predisposition, and association with autoimmune endocrinopathies and antireceptor autoimmune diseases.

The identification of gastric H\textsuperscript{+}/K\textsuperscript{−}–ATPase as the target of parietal-cell autoantibodies was a major breakthrough in our understanding of the molecular and immunologic basis of autoimmune gastritis. The immunologic mechanisms that allow the initiation and progression of the T-cell response to this enzyme, leading to autoimmune gastritis, remain to be established.

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