Vitamin B₁₂ Deficiency
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A 57-year-old woman reports increasing symptoms of painful paresthesias in both legs for the past 18 months. Physical examination reveals impaired position sense and vibration sense. The serum vitamin B₁₂ level is 205 pg per milliliter (151.2 pmol per liter), which is above the lower end of the laboratory reference range. The hematocrit is 42%, with a mean corpuscular volume of 96 fl. The serum methylmalonic acid level is 3600 nmol per liter (normal level, <400), and the serum homocysteine level 49.1 μmol per liter (normal level, <14). How should this patient be further evaluated and treated?

THE CLINICAL PROBLEM

The recognition and treatment of vitamin B₁₂ deficiency is critical since it is a reversible cause of bone marrow failure and demyelinating nervous system disease. Vitamin B₁₂ (cobalamin) is synthesized by microorganisms and detected in trace amounts mostly in foods of animal origin.¹ Uptake in the gastrointestinal tract depends on intrinsic factor, which is synthesized by the gastric parietal cells, and on the “cubam receptor” in the distal ileum.² The most frequent cause of severe vitamin B₁₂ deficiency is a loss of intrinsic factor due to autoimmune atrophic gastritis,³ historically called “pernicious anemia,” even though many patients present with mainly neurologic manifestations.⁴,⁵

PATHOPHYSIOLOGY OF VITAMIN B₁₂ DEFICIENCY

Vitamin B₁₂ is a cofactor for only two enzymes: methionine synthase and l-methylmalonyl–coenzyme A mutase⁶,⁷ (see Fig. 1 in the Supplementary Appendix, available with the full text of this article at NEJM.org). The interaction between folate and B₁₂ is responsible for the megaloblastic anemia seen in both vitamin deficiencies. Dyssynchrony between the maturation of cytoplasm and that of nuclei leads to macrocytosis, immature nuclei, and hypersegmentation in granulocytes⁶ in the peripheral blood (Fig. 1A). The hypercellular and dysplastic bone marrow can be mistaken for signs of acute leukemia (Fig. 1B).¹⁰ The ineffective erythropoiesis results in intramedullary hemolysis and release of lactate dehydrogenase, features that are similar to those of microangiopathic hemolytic anemia.⁸ Clinical and laboratory findings of megaloblastic anemia in the peripheral blood and bone marrow are shown in Figure 2.

Vitamin B₁₂ is necessary for the development and initial myelination of the central nervous system as well as for the maintenance of its normal function. Demyelination of the cervical and thoracic dorsal and lateral columns of the spinal cord, occasional demyelination of cranial and peripheral nerves, and demyelination of white matter in the brain⁵ (i.e., “combined-systems disease” or “subacute combined degeneration”) can occur with vitamin B₁₂ deficiency (Fig. 2). Pathologi-
KEY CLINICAL POINTS

VITAMIN B₁₂ DEFICIENCY

- Vitamin B₁₂ deficiency causes reversible megaloblastic anemia, demyelinating neurologic disease, or both.
- Autoimmune gastritis (pernicious anemia) is the most common cause of severe deficiency.
- Methodologic problems may compromise the sensitivity and specificity of current vitamin B₁₂ assays.
- Measurement of methylmalonic acid, homocysteine, or both is used to confirm vitamin B₁₂ deficiency in untreated patients; an elevated level of methylmalonic acid is more sensitive and specific for the diagnosis.
- For patients with pernicious anemia or malabsorption, lifelong vitamin B₁₂ therapy is indicated.
- High-dose oral vitamin B₁₂ tablets (1000 to 2000 µg) taken daily are as effective as intramuscular monthly injections in correcting blood and neurologic abnormalities.

C A U S E S O F V I T A M I N B₁₂ D E F I C I E N C Y

Table 1 and Figure 3 list causes of vitamin B₁₂ deficiency and recommended management. Pernicious anemia is discussed below, since this is the most common cause of severe vitamin B₁₂ deficiency worldwide.

Dietary vitamin B₁₂ deficiency in infants and children is also discussed because of the increasing recognition of severe abnormalities in exclusively breast-fed infants of mothers with vitamin B₁₂ deficiency.

Pernicious Anemia

Pernicious anemia³ is an autoimmune gastritis resulting from the destruction of gastric parietal cells and the associated lack of intrinsic factor to bind ingested vitamin B₁₂. The immune response is directed against the gastric H/K-ATPase, which accounts for associated achlorhydria.³,⁴ Other autoimmune disorders, especially thyroid disease, type 1 diabetes mellitus, and vitiligo, are also commonly associated with pernicious anemia. Whether the stomach pathogen Helicobacter pylori plays a causative role in pernicious anemia is unclear.⁵ Autoimmune gastritis may cause malabsorption of iron, with clinical iron deficiency developing early in life and eventually progressing to malabsorption of vitamin B₁₂.⁶ The prevalence of pernicious anemia ranges from 50 to 4000 cases per 100,000 persons, depending on the diagnostic criteria.¹ All age groups are affected, but the median age range in large series is 70 to 80 years.²¹,²² Pernicious anemia is more common in persons of African or European ancestry (4.3% and 4.0% prevalence among older adults, respectively) than in those of Asian ancestry.¹,²¹ Milder forms of atrophic gastritis with hypochlorhydria and an inability to release dietary protein-bound vitamin B₁₂ affect up to 20% of older adults.¹⁰,²³,²⁴

Dietary Deficiency in Infancy and Childhood

The infant of a mother with vitamin B₁₂ deficiency may be born with the deficiency or it may occur if he or she is exclusively breast-fed,¹⁵,¹⁶ usually between 4 and 6 months of age. Typical manifestations of vitamin B₁₂ deficiency in children include failure of brain development and overall growth and development, developmental regression, hypotonia, feeding difficulties, lethargy, tremors, hyperirritability, and coma (Fig. 2).¹⁵,¹⁶ Brain imaging may reveal atrophy and delayed myelination. Anemia may be present. Vitamin B₁₂ replacement results in rapid improvement in responsiveness, and many infants recover fully. However, the longer the period of deficiency, the more likely that there will be permanent disabilities. Mothers of infants with
vitamin B₁₂ deficiency often have unrecognized pernicious anemia, but alternatively, they may have a history of gastric bypass surgery, the short-gut syndrome, or a long-term vegetarian or vegan diet. Tandem mass spectrometry, used in neonatal screening programs in all 50 states, may detect nutritional B₁₂ deficiency owing to an increase in propionyl carnitine, but direct measurement of methylmalonic acid has higher sensitivity. Other causes of B₁₂ deficiency in children, such as ileal resections, the Imerslund–Gräsbeck syndrome, inflammatory bowel disease, and pernicious anemia, are listed in Table 1.

**EVALUATION**

Both the clinical recognition of vitamin B₁₂ deficiency and confirmation of the diagnosis by means of testing can be difficult. An approach to testing is shown in Table 2.

The patient’s history may include symptoms of anemia, underlying disorders causing malabsorption, and neurologic symptoms. The most common neurologic symptoms are symmetric paresthesias or numbness and gait problems. The physical examination may reveal pallor, edema, pigmentary changes in the skin, jaundice, or neurologic defects such as impaired vibration sense, impaired position and cutaneous sensation, ataxia, and weakness (Fig. 2).

Bone marrow biopsy and aspiration are not necessary for the diagnosis of megaloblastic anemia and may be misleading in cases of severe pancytopenia with hypercellularity, increased erythroblasts, and even cytogenetic abnormalities, confusing the diagnosis with acute leukemia. Imaging of the spinal cord is not indicated in patients with recognized vitamin B₁₂ deficiency, but in cases of severe myelopathy that are not initially recognized as the result of vitamin B₁₂ deficiency, there is characteristic hyperintensity on T₂-weighted imaging, described as an inverted V-shaped pattern in the cervical and thoracic spinal cord.

**Vitamin B₁₂ Assay**

The first test performed to confirm the diagnosis of vitamin B₁₂ deficiency is generally measurement of the serum vitamin B₁₂ level. Although an extremely low level (<100 pg per milliliter (<73.8 pmol per liter)) is usually associated with clinical deficiency, such low levels are infrequently observed. Both false negative and false positive values are common (occurring in up to 50% of tests) with the use of the laboratory-reported lower limit of the normal range as a cutoff point for deficiency. The high rate of false negative and false positive results may be
Brain
Altered mental status
Cognitive defects
“Megaloblastic madness”: depression, mania, irritability, paranoia, delusions, lability

Spinal cord
Myelopathy
Spongy degeneration
Paresthesias
Loss of proprioception: vibration, position, ataxic gait, limb weakness; spasticity (hyperreflexia); positive Romberg sign; Lhermitte’s sign; segmental cutaneous sensory level

Optic atrophy, anosmia, loss of taste, glossitis

Infertility

Abnormalities in infants and children
Developmental delay or regression, permanent disability
Does not smile
Feeding difficulties
Hypotonia, lethargy, coma
Hyperirritability, convulsions, tremors, myoclonus
Microcephaly
Choreoathetoid movements

Peristomal

Abnormalities in infants and children
Developmental delay or regression, permanent disability
Does not smile
Feeding difficulties
Hypotonia, lethargy, coma
Hyperirritability, convulsions, tremors, myoclonus
Microcephaly
Choreoathetoid movements

Infertility

Abnormalities in infants and children
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Feeding difficulties
Hypotonia, lethargy, coma
Hyperirritability, convulsions, tremors, myoclonus
Microcephaly
Choreoathetoid movements

Peripheral blood
Macrocytic red cells, macroovalocytes
Anisocytosis, fragmented forms
Hypersegmented neutrophils, 1% with six lobes or 5% with 5 lobes
Leukopenia, possible immature white cells
Thrombocytopenia
Pancytopenia
Elevated lactate dehydrogenase level (extremes possible)
Elevated indirect bilirubin and aspartate aminotransferase levels
Decreased haptoglobin level
Elevated levels of methylmalonic acid, homocysteine, or both

Bone marrow
Hypercellular, increased erythroid precursors
Open, immature nuclear chromatin
Dysynchrony between maturation of cytoplasm and nuclei
Giant bands, metamyelocytes
Karyorrhexis, dysplasia
Abnormal results on flow cytometry and cytogenetic analysis

Peripheral nervous system
Cutaneous sensory loss
Hyporeflexia
Symmetric weakness
Paresthesias

Autonomic nervous system
Postural hypotension
Incontinence
Impotence

Peripheral nervous system
Cutaneous sensory loss
Hyporeflexia
Symmetric weakness
Paresthesias

Bone marrow
Hypercellular, increased erythroid precursors
Open, immature nuclear chromatin
Dysynchrony between maturation of cytoplasm and nuclei
Giant bands, metamyelocytes
Karyorrhexis, dysplasia
Abnormal results on flow cytometry and cytogenetic analysis
due to the fact that only 20% of the total measured vitamin B₁₂ is on the cellular delivery protein, transcobalamin; the remainder is bound to haptocorrin, a protein of unknown function.²⁷ Most laboratories now perform automated assays of vitamin B₁₂ on platforms used for many other analytes. There is often poor agreement when samples are assayed by different laboratories or with the use of different methods.²⁸-³¹ Because intrinsic factor is used as the assay-binding protein, anti–intrinsic factor antibodies (which are common in pernicious anemia) must be removed chemically from the sample, which has proved to be problematic in the automated assays.³²-³⁴ Recent studies show normal values³⁴ or falsely high values³³ of vitamin B₁₂ in many patients with pernicious anemia. New assays of holo transcobalamin (to measure the vitamin B₁₂ saturation of transcobalamin) provide a modest improvement in specificity over that provided by assays of total serum vitamin B₁₂, but they have not been clinically validated²⁷-²⁹ and are not yet available commercially in the United States.

Given the limitations of available assays, clinicians should not use a laboratory’s reported lower limit of the normal range to rule out the diagnosis of vitamin B₁₂ deficiency in patients with compatible clinical abnormalities. Clinicians should also recognize that vitamin B₁₂ values are frequently low in patients without other metabolic or clinical evidence of vitamin B₁₂ deficiency (i.e., megaloblastic anemia or myelopathy).

**Measurement of Serum Methylmalonic Acid and Total Homocysteine**

Measurement of methylmalonic acid, total homocysteine, or both is useful in making the diagnosis of vitamin B₁₂ deficiency in patients who have not received treatment.⁴,²²,²⁴,²⁶,³³,³⁵,³⁶ The levels of both methylmalonic acid and total homocysteine are markedly elevated in the vast majority (>98%) of patients with clinical B₁₂ deficiency (Fig. 4),²² including those who have only neurologic manifestations of deficiency (i.e., no anemia).⁴,²²

Elevated levels of methylmalonic acid and total homocysteine decrease immediately after treatment, and the levels can be remeasured to document adequate vitamin B₁₂ replacement. Levels of these metabolites are normal in up to 50% of patients with low vitamin B₁₂ levels who have no hematologic or neurologic response to replacement therapy, indicating that the low values are false positive results.²⁶ Given the limitations of vitamin B₁₂ assays in confirming the diagnosis of B₁₂ deficiency,²¹,³¹ it may be prudent to measure methylmalonic acid, total homocysteine, or both in patients with compatible clinical findings or provide empirical treatment with the use of defined end points to document a clinical response.

An elevated level of methylmalonic acid is reasonably specific for vitamin B₁₂ deficiency, and the level always decreases with vitamin B₁₂ therapy.²⁴,³⁶ Modest increases (to 300 to 700 nmol per liter) occur with renal failure.³⁶,³⁷ However, nearly all patients with megaloblastic anemia or myelopathy have levels of methylmalonic acid that are higher than 500 nmol per liter, and 86% have levels that are higher than 1000 nmol per liter (Fig. 3). The level of serum total homocysteine is less specific, since it is also elevated in folate deficiency,²²,³⁵ classic homocystinuria, and renal failure.

**Tests to Determine the Cause of Vitamin B₁₂ Deficiency**

If the patient consumes sufficient amounts of vitamin B₁₂ and has clinically confirmed B₁₂ deficiency, then malabsorption must be present. Testing for pernicious anemia is described in Table 2. A positive test for anti–intrinsic factor or anti–parietal-cell antibodies is indicative of pernicious anemia; surveillance for autoimmune thyroid disease is reasonable in patients with positive antibody tests. Chronic atrophic gastritis can be diagnosed on the basis of an elevated fasting serum gastrin level and a low level of serum pepsinogen I.³,¹⁰ Some experts recommend endoscopy to confirm gastritis and rule out gastric carcinoid and other gastric cancers, since patients with pernicious anemia are at increased risk for such cancers.³

The Schilling test of radioactive vitamin B₁₂...
### Table 1. Causes and Treatment of Vitamin B<sub>12</sub> Deficiency.

<table>
<thead>
<tr>
<th>Cause</th>
<th>Treatment</th>
<th>Follow-up</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severe malabsorption</td>
<td>Intramuscular cyanocobalamin at a dose of 100 µg administered intramuscularly daily or every other day for 1 wk, then weekly for 4 to 8 wk, and then monthly for life, or oral cyanocobalamin at a daily dose of 1000 µg for life*</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Pernicious anemia (autoimmune gastritis)</td>
<td>Administer iron and folic acid replacement as needed for full hemoglobin response, especially in patients with dietary deficiencies, perform surveillance for other autoimmune conditions, perform upper endoscopy in patients with symptoms of gastric cancer or malabsorption.</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Total or partial gastrectomy</td>
<td>Same as for pernicious anemia</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Gastric bypass or other bariatric surgery</td>
<td>Same as for pernicious anemia</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Inflammatory bowel disease, tropical sprue</td>
<td>Same as for pernicious anemia</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Imerslund–Gräsbeck and other syndromes‡</td>
<td>Same as for pernicious anemia</td>
<td>Same as for pernicious anemia</td>
</tr>
<tr>
<td>Mild malabsorption</td>
<td>Oral cyanocobalamin at a dose of 500 to 1000 µg daily or intramuscularly daily or every other day for 1 wk, then weekly for 4 to 8 wk, and then monthly for life</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
</tr>
<tr>
<td>Mild atrophic gastritis</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
</tr>
<tr>
<td>Dietary deficiency</td>
<td>Use of metformin†</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
</tr>
<tr>
<td>Adults</td>
<td>Oral cyanocobalamin at a dose of 250 µg daily for 6 wk, then weekly until patient recovers; treatment with 500 µg daily for 24 mo; oral supplementation with 110 µg of vitamin B&lt;sub&gt;12&lt;/sub&gt; daily or vitamin B&lt;sub&gt;12&lt;/sub&gt;-enriched formula or food</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
</tr>
<tr>
<td>Breastfeeding in infants with vitamin B&lt;sub&gt;12&lt;/sub&gt; deficiency</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
<td>Same as for protein-bound vitamin B&lt;sub&gt;12&lt;/sub&gt; malabsorption</td>
</tr>
</tbody>
</table>
absorption is no longer available. A potential replacement absorption test is under development wherein the increase in vitamin B₁₂ saturation of holotranscobalamin is measured after several days of oral B₁₂ loading,¹⁹ but this requires further study.

**TREATMENT OF VITAMIN B₁₂ DEFICIENCY**

The daily requirement of vitamin B₁₂ has been set at 2.4 μg,⁴⁰,⁴¹ but higher amounts — 4 to 7 μg per day — which are common in persons who eat meat or take a daily multivitamin, are associated with lower methylmalonic acid values.⁴² Healthy older adults should consider taking supplemental crystalline vitamin B₁₂ as recommended by the Food and Nutrition Board.⁴¹ However, most patients with clinical vitamin B₁₂ deficiency have malabsorption and will require parenteral or high-dose oral replacement. Adequate supplementation results in resolution of megaloblastic anemia and resolution of or improvement in myelopathy.

**Injected Vitamin B₁₂**

There are many recommended schedules for injections of vitamin B₁₂ (called cyanocobalamin in the United States and hydroxocobalamin in Europe).⁶,²³ About 10% of the injected dose (100 of 1000 μg) is retained. Patients with severe abnormalities should receive injections of 1000 μg at least several times per week for 1 to 2 weeks, then weekly until clear improvement is shown, followed by monthly injections. Hematologic response is rapid, with an increase in the reticulocyte count in 1 week and correction of megaloblastic anemia in 6 to 8 weeks. Patients with severe anemia and cardiac symptoms should be treated with transfusion and diuretic agents, and electrolytes should be monitored. Neurologic symptoms may worsen transiently and then subside over weeks to months.⁵ The severity and duration of the neurologic abnormalities before treatment influence the eventual degree of recovery.⁴,⁵ Treatment of pernicious anemia is lifelong. In patients in whom vitamin B₁₂ supplementation is discontinued after clinical recovery, neurologic symptoms recur within as short a period as 6 months, and megaloblastic anemia recurs in several years.⁶

**High-Dose Oral Treatment**

High-dose oral treatment is effective and is increasingly popular. A study performed 45 years ago...
Figure 3. The Normal Mechanisms and Defects of Absorption of Vitamin B$_{12}$.

The vitamin B$_{12}$ (Cbl) released from food protein by peptic action is bound to haptocorrin (HC) in the stomach and travels to the duodenum, where pancreatic proteases digest the HC, releasing Cbl to bind to intrinsic factor (IF). The IF-Cbl complex binds to a specific receptor in the distal ileum (the cubam receptor) and is internalized, eventually released from lysosomes, and transported into the blood. Both HC and transcobalamin (TC) bind Cbl in the circulation, although the latter is the cellular delivery protein. Adapted from Stabler. 6
**Table 2. Laboratory Testing in Vitamin B\textsubscript{12} Deficiency.**

<table>
<thead>
<tr>
<th>Test to detect deficiency</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Measurement to detect deficiency</td>
<td>65–95% for proven clinical deficiency; 50% for detecting elevated level of methylmalonic acid</td>
<td>50–60% for clinical response; 80% for detecting elevated level of methylmalonic acid</td>
<td>Current vitamin B\textsubscript{12} assays are especially problematic in patients with anti–intrinsic factor antibodies</td>
</tr>
<tr>
<td>Serum vitamin B\textsubscript{12} &lt;200 pg/ml or laboratory cutoff level</td>
<td>90%</td>
<td>25% for detecting elevated level of methylmalonic acid</td>
<td></td>
</tr>
<tr>
<td>Holotranscobalamin &lt;20 to 45 pmol/liter</td>
<td>Insufficient data on sensitivity for clinical deficiency; 46–89% for detecting elevated level of methylmalonic acid</td>
<td>Insufficient data on specificity for clinical deficiency; 28–96% for detecting elevated level of methylmalonic acid</td>
<td>Levels of holotranscobalamin increase in renal failure; superior to measurement of total vitamin B\textsubscript{12} in pregnancy, when the total level decreases</td>
</tr>
<tr>
<td>Serum methylmalonic acid &gt;400 nmol/liter</td>
<td>98% for clinical deficiency</td>
<td>Poor specificity for clinical response in patients with modest elevation of level of methylmalonic acid (300–1000 nmol/liter)</td>
<td>Renal failure and volume depletion may increase level of serum methylmalonic acid, but rarely to &gt;1000 nmol/liter</td>
</tr>
<tr>
<td>Serum or plasma total homocysteine &gt;21 µmol/liter</td>
<td>96% for clinical deficiency</td>
<td>Homocysteine level also increased in clinical folate deficiency and renal insufficiency</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Test to determine cause of deficiency</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pernicious anemia</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anti–intrinsic factor antibodies</td>
<td>50%</td>
<td>100%</td>
<td>Must be tested &gt;7 days after vitamin B\textsubscript{12} injection to prevent false positive result</td>
</tr>
<tr>
<td>Anti–parietal-cell antibodies</td>
<td>80%</td>
<td>50–100%</td>
<td></td>
</tr>
<tr>
<td>Atrophic body gastritis (antral sparing)**</td>
<td>85%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting high serum gastrin level (&gt;100 pmol/liter)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Low level of serum pepsinogen I (&lt;30 µg/liter)</td>
<td>90%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Endoscopy with pentagastrin-fast hypochlorhydria</td>
<td>100%</td>
<td>Rarely performed</td>
<td></td>
</tr>
<tr>
<td>Malabsorption of vitamin B\textsubscript{12}††</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vitamin B\textsubscript{12} absorption test</td>
<td>Schilling test no longer available</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Increase in serum holotranscobalamin level after oral loading</td>
<td>Unknown</td>
<td>Unknown</td>
<td>Promising preclinical data, but still experimental</td>
</tr>
</tbody>
</table>

* To convert the values for vitamin B\textsubscript{12} to picomoles per liter, multiply by 0.7378.

† Available assays are largely chemiluminescent microparticle immunoassays performed with the use of automated analyzers that in general show higher values than the radiodilution and microbiologic assays used in past studies of clinically confirmed deficiency.4,22,24,26 Thus, these tests are likely to have lower sensitivities and specificities than the older assays.

‡ The holotranscobalamin assay has been studied widely in Europe27-30 but is not yet commercially available in the United States. The appropriate lower end of the reference range is still under debate.33 The values for sensitivity and specificity are reviewed in Heil et al.29

§ Urinary methylmalonic acid has not been extensively studied, but values greater than 2.5 µmol per millimole of creatinine suggest deficiency.

¶ Elevated levels of methylmalonic acid fall with vitamin B\textsubscript{12} therapy, but an associated clinical response is highly variable, depending largely on the presence of vitamin B\textsubscript{12}–related disease.

‖ Evidence of a causal pathologic process does not confirm coexisting B\textsubscript{12} deficiency, since underlying gastrointestinal disease may predate the deficiency by many years.

** The relationship between atrophic body gastritis (autoimmune gastritis) and infection with Helicobacter pylor is variable. Antral sparing is a type of atrophic body gastritis in which the cells in the antrum can produce high levels of gastrin.

†† There is malabsorption if clinically proven vitamin B\textsubscript{12} deficiency is present in a patient who eats meat, receives multivitamin therapy, or both.
showed that 0.5 to 4% of radioactively labeled oral vitamin B$_{12}$ can be absorbed by passive diffusion in both normal controls and patients with pernicious anemia.$^{43}$ Thus, oral doses of 1000 µg deliver 5 to 40 µg, even if taken with food.

A randomized trial that compared an oral dose of 2000 µg daily with parenteral therapy (seven injections of 1000 µg of cyanocobalamin over a period of 1 month, followed by monthly injections) in patients with pernicious anemia, atrophic gastritis, or a history of ileal resection showed similar reductions in the mean corpuscular volume and increases in the hematocrit at 4 months in both groups.$^{38}$ All participants (four in each group) with paresthesias, ataxia, or memory loss had resolution or improved with treatment. However, levels of methylmalonic acid after treatment were significantly lower with daily oral treatment (169 nmol per liter, vs. 265 nmol per liter with parenteral treatment) and vitamin B$_{12}$ levels were significantly higher (1005 pg per milliliter vs. 325 pg per milliliter [741.5 vs. 239.8 pmol per liter]). A more recent trial with a similar design involving a proprietary oral vitamin B$_{12}$ preparation also revealed significantly lower levels of methylmalonic acid in the oral-treatment group at the 3-month follow-up.$^{30}$ In a randomized trial comparing oral with intramuscular vitamin B$_{12}$ (1000-µg doses, daily for 10 days, then weekly for 4 weeks, and monthly thereafter), the two groups had similar improvements in hematologic abnormalities and vitamin B$_{12}$ levels at 90 days.$^{44}$ Case series of patients treated with oral vitamin B$_{12}$ have yielded variable results; elevated levels of methylmalonic acid, homocysteine, or both were reported in about half of patients with malabsorption who were treated with twice-weekly oral doses of 1000 µg,$^{45}$ whereas normal homocysteine levels were reported in patients treated with 1500 µg daily after gastrectomy.$^{46}$ Data are lacking from long-term studies to assess whether oral treatment is effective when doses are administered less frequently than daily. Studies involving older adults, many of whom had chronic atrophic gastritis, showed that 60% required large oral doses (>500 µg daily) to correct elevated levels of methylmalonic acid.$^{47,48}$

Proponents of parenteral therapy state that compliance and monitoring are better in patients who receive this form of therapy because they have frequent contact with health care providers, whereas proponents of oral therapy maintain that compliance will be improved in patients who receive oral therapy because of convenience, comfort, and decreased expense. High-dose vitamin B$_{12}$ tablets (500 to 1500 µg) are available in the United States without a prescription. Self-administered injections are also easily taught, economical, and in my experience, effective. Patients should be informed of the pros and cons of oral versus parenteral therapy, and regardless of the form of treatment, those with pernicious anemia or malabsorption should be reminded of the need for lifelong replacement.

### Areas of Uncertainty

Vitamin B$_{12}$ deficiency is the major cause of hyperhomocysteinemia in countries with folate-fortified food, such as the United States and
Canada. Epidemiologic studies show significant associations between elevated homocysteine levels and vascular disease and thrombosis. However, large randomized trials of combined high-dose vitamin B therapy in patients with vascular disease have shown no reduction in vascular events. Vitamin B₁₂ status should be evaluated in patients with hyperhomocysteinemia before folic acid treatment is initiated.

The potential role of mild vitamin B₁₂ deficiency in cognitive decline with aging remains uncertain. Epidemiologic studies indicate an inverse association between vitamin B₁₂ supplementation and neurodegenerative disease, but results of randomized trials have been largely negative.50 Besides oral tablets, vitamin B is available in sublingual preparations, oral sprays, nasal gels or sprays, and transdermal patches. Data on the absorption and efficacy of these alternative preparations are lacking.

Nutritional guidelines for vitamin B₁₂ intake are published by the Food and Nutrition Board, and nutritional guidelines for vegetarians are published by the American Dietetic Association. There are no recommendations from the American Society of Hematology for the diagnosis and treatment of vitamin B₁₂ deficiency. The American Academy of Neurology recommends measurements of vitamin B₁₂, methylmalonic acid, and homocysteine in patients with symmetric polyneuropathy. The American Society for Gastrointestinal Endoscopy recommends a single endoscopic evaluation at the diagnosis of pernicious anemia.52

CONCLUSIONS AND RECOMMENDATIONS

The patient in the vignette has neurologic abnormalities that are consistent with vitamin B₁₂ deficiency. Since vitamin B₁₂ levels may be above the lower end of the laboratory reference range even in patients with clinical deficiency, methylmalonic acid, total homocysteine, or both should be measured to document vitamin B₁₂ deficiency before treatment is initiated; the elevated levels in this patient confirm the diagnosis. In the absence of dietary restriction or a known cause of malabsorption, further evaluation is warranted — in particular, testing for pernicious anemia (anti-intrinsic factor antibodies). Either parenteral vitamin B₁₂ treatment (8 to 10 loading injections of 1000 μg each, followed by monthly 1000-μg injections), or high-dose oral vitamin B₁₂ treatment (1000 to 2000 μg daily) is an effective therapy. I would review both options (including the possibility of self-injection at home) with the patient. Effective vitamin replacement will correct blood counts in 2 months and correct or improve neurologic signs and symptoms within 6 months.

Dr. Stabler reports holding patents (assigned to the University of Colorado and Competitive Technologies) on the use of homocysteine, methylmalonic acid, and other metabolites in the diagnosis of vitamin B₁₂ and folate deficiency, but no longer receiving royalties for these patents. No other potential conflict of interest relevant to this article was reported.

Disclosure forms provided by the author are available with the full text of this article at NEJM.org.

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