

Overview**Useful For**

Diagnosis of pernicious anemia

Diagnosis of vitamin B12 deficiency-associated neuropathy

Reflex Tests

Test ID	Reporting Name	Available Separately	Always Performed
IFBPA	Intrinsic Factor Blocking Ab, S	Yes, (order IFBA)	No
MMAPA	Methylmalonic Acid, QN, S	Yes, (order MMAS)	No
GASTR	Gastrin, S	Yes, (order GAST)	No

Testing Algorithm

If vitamin B12 is <150 ng/L, then intrinsic factor blocking antibody (IFBA) is performed. If IFBA is negative or indeterminate, then gastrin is performed.

If vitamin B12 is 150 to 400 ng/L, then methylmalonic acid (MMA) is performed. If MMA is >0.40 nmol/mL, then IFBA is performed.

[See Vitamin B12 Deficiency Evaluation](#) in Special Instructions.

Special Instructions

- [Vitamin B12 Deficiency Evaluation](#)

Method Name

Immunoenzymatic Assay

NY State Available

Yes

Specimen**Specimen Type**

Serum

Specimen Required**Container/Tube:****Preferred:** Red top**Acceptable:** Serum gel**Specimen Volume:** 4 mL

Collection Instructions:

1. Fasting (8 hours)
2. Divide specimen into 3 plastic vials, 1 containing 1 mL, 1 containing 1.5 mL, and 1 containing 1.5 mL
3. Band specimens together.

Additional Information: This test should not be ordered on patients who have received vitamin B12 injection within the last 2 weeks.

Forms

If not ordering electronically, complete, print, and send a [Benign Hematology Test Request Form](#) (T755) with the specimen.

Specimen Minimum Volume

2.3 mL

Reject Due To

Gross hemolysis	Reject
Gross lipemia	OK

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Serum	Frozen	14 days	

Clinical and Interpretive

Clinical Information

[Vitamin B12 deficiency can be caused by many factors, one of which is pernicious anemia, a condition resulting in deficient production of intrinsic factor in the parietal cells of the stomach. Intrinsic factor is a protein that is needed to assist in the absorption of vitamin B12 into the small intestine. Vitamin B12 is converted into adenosylcobalamin, which converts L-methylmalonic acid to succinyl coenzyme A; hence, a decrease in vitamin B12 absorption in the intestine can cause an excess of methylmalonic acid within the body. ¹](#)

Vitamin B12 deficiency may present with any combination of the following: macrocytic anemia, glossitis (painful inflammation of the tongue), peripheral neuropathy, weakness, hyperreflexia, ataxia, loss of proprioception, poor coordination, and affective behavioral changes. These manifestations may occur in any combination; many patients present with neurologic symptoms without macrocytic anemia.

A group of tests is often required to establish the correct diagnosis as determination of vitamin B12 in serum does not detect all cases of vitamin B12 deficiency. Mayo Clinic's Department of Laboratory Medicine and Pathology offers a diagnostic algorithm to expedite the identification of patients with vitamin B12 deficiency. This algorithm takes into account the following facts:

- The most sensitive test for vitamin B12 deficiency at the cellular level is the assay for methylmalonic acid (MMA).

-Nearly half of the cases of pernicious anemia can be unambiguously identified if the serum test for intrinsic factor blocking antibody is positive (this is a simpler and less expensive test than the MMA).

-Serum gastrin is usually markedly increased in pernicious anemia (as a result of gastric atrophy) and this test can be used as a substitute for the more complicated and more expensive Schilling test of intestinal absorption of vitamin B12.

The algorithm is similar to that published by Green,(1) except that the serum gastrin assay is performed in place of the Schilling test. Experience with both Mayo Clinic and Mayo Clinic Laboratories' cases has corroborated that this is a cost-effective alternative to the Schilling test.

In our experience, >90% of laboratory test costs can be saved by using the algorithm rather than ordering all of the services for a patient suspected of having B12 deficiency. Furthermore, the substitution of the serum gastrin assay for the Schilling test offers 3 advantages:

1. It is an in vitro test that does not require administration of radioisotopes to patients
2. It can be performed on mailed-in specimens
3. It is much less expensive

Only those tests that are appropriate, as defined by the algorithm, will be performed.

Reference Values

180-914 ng/L

Interpretation

Vitamin B12 >400 ng/L	Results do not suggest B12 deficiency-no further testing.
Vitamin B12 150 to 400 ng/L	Borderline vitamin B12 level-methylmalonic acid (MMA) is performed. If MMA is >0.40 nmol/mL, then intrinsic factor blocking antibody (IFBA) is performed.
Vitamin B12 <150 ng/L	Vitamin B12 deficiency-IFBA is performed. If IFBA is negative or indeterminate, then gastrin is performed.
MMA < or =0.40 nmol/mL	This value implies that there is no vitamin B12 deficiency at the cellular level.
IFBA positive	Consistent with pernicious anemia, Graves disease, or Hashimoto thyroiditis.
Gastrin >200 pg/mL	Result consistent with pernicious anemia.
Gastrin <200 pg/mL	Result does not suggest pernicious anemia.

[See Vitamin B12 Deficiency Evaluation](#) in Special Instructions.

Cautions

See individual test listings.

Clinical Reference

1. Green R, Kinsella LJ: Current concepts in the diagnosis of cobalamin deficiency. Neurology 1995;45:1435-1440
2. Lahner E, Annibale B: Pernicious anemia: new insights from a gastroenterological point of view. World J Gastroenterol. 2009 Nov 7;15(41):5121-5128

Performance

Method Description

The instrument used is a Beckman Coulter DXI 800. The Access Vitamin B12 assay is a competitive-binding immunoenzymatic assay. A sample is added to a reaction vessel along with alkaline potassium cyanide and dithiothreitol. This treatment denatures B12 binding proteins and converts all forms of vitamin B12 to the cyanocobalamin form. After neutralization, intrinsic factor-alkaline phosphatase conjugate and paramagnetic particles coated with goat antimouse IgG:mouse monoclonal anti-intrinsic factor are added to the sample. Vitamin B12 in the sample binds to the intrinsic factor conjugate, preventing the conjugate from binding to the solid phase anti-intrinsic factor. After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field, while unbound materials are washed away. The chemiluminescent substrate Lumi-Phos 530 is added to the vessel and light generated by the reaction is measured with a luminometer. The photon production is inversely proportional to the concentration of vitamin B12 in the sample. The amount of analyte in the sample is determined by means of a stored, multipoint calibration curve.(Instruction manual: Beckman Coulter Assay 2009, Beckman Coulter Inc, Fullerton, CA)

PDF Report

No

Day(s) and Time(s) Test Performed

Monday through Friday 5 a.m.-12 a.m.

Saturday 6 a.m.-6 p.m.

Analytic Time

Same day/1 day

Maximum Laboratory Time

4 days

Specimen Retention Time

2 weeks/3 months

Performing Laboratory Location

Rochester

Fees and Codes

Fees

- Authorized users can sign in to [Test Prices](#) for detailed fee information.
- Clients without access to Test Prices can contact [Customer Service](#) 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact [Customer Service](#).

CPT Code Information

82607-Vitamin B12 assay

82941-Gastrin (if appropriate)

83921-MMA (if appropriate)

86340-IFBA (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
ACASM	Pernicious Anemia Cascade	2132-9

Result ID	Test Result Name	Result LOINC Value
B12PA	Vitamin B12 Assay, S	2132-9