PHYSICIAN UPDATE

Diagnosing Vitamin B₁₂ Deficiency with Serum Methylmalonic Acid

BENEFITS
- Provides a more sensitive method to screen for vitamin B₁₂ deficiency by measuring serum methylmalonic acid (MMA) and homocysteine, which are increased in early vitamin B₁₂ deficiency.
- Aids in clarifying the diagnosis when vitamin B₁₂ concentrations are indeterminate (100-400 pg/mL).

BACKGROUND
Effective May 11, methylmalonic acid testing for confirmation of vitamin B₁₂ deficiency will be performed at the Central Laboratory in Springfield, Oregon to improve turnaround time and improve service to you and your patients.

Vitamin B₁₂ (cobalamin) deficiency is a common cause of macrocytic anemia and has been implicated in a spectrum of neuropsychiatric disorders. Diagnosis of vitamin B₁₂ deficiency is typically based on measurement of serum vitamin B₁₂ levels. However, patients with low-normal or even normal serum vitamin B₁₂ levels may be truly vitamin B₁₂ deficient and respond to replacement therapy.

Approximately 50 percent of patients with subclinical disease have normal vitamin B₁₂ levels. In addition, older patients tend to present with neuropsychiatric disease in the absence of hematologic findings.

THE ROLE OF METHYLMALONIC ACID IN VITAMIN B₁₂ DEFICIENCY DIAGNOSIS

Vitamin B₁₂ is involved in two enzymatic reactions (see Figure 1):
- In the first reaction, MMA is converted to succinyl-CoA using vitamin B₁₂ as a cofactor. Vitamin B₁₂ deficiency will cause an elevation of serum MMA.
- In the second reaction, homocysteine is converted to methionine using vitamin B₁₂ and folic acid as cofactors. Deficiency in vitamin B₁₂ and/or folic acid lead to elevated homocysteine levels, while deficiency in Vitamin B₁₂ alone leads to an increase in serum MMA and homocysteine.

Figure 1

continued on next page
INTERPRETATION

Vitamin B\textsubscript{12} and folic acid deficiency often coexist and are not easily differentiated on a clinical basis. When vitamin B\textsubscript{12} deficiency is present but undiagnosed, folate repletion will correct the megaloblastic anemia, but not the potential neuropathic changes. Accordingly, patients should be evaluated for both deficiencies.

MMA is more sensitive for detection of vitamin B\textsubscript{12} deficiency than homocysteine. Specifically, patients with megaloblastic anemia and/or neurologic symptoms should have a serum MMA test when the vitamin B\textsubscript{12} level is between 100-400 pg/mL.\textsuperscript{4} See Figure 2 algorithm for testing indications.

MMA levels can be elevated in patients with renal disease as a result of decreased urinary excretion, thus elevated MMA must be interpreted with caution in renal failure patients.\textsuperscript{4} Serum MMA levels are also elevated in patients with methylmalonic aciduria.

**Figure 2**

![Figure 2](image-url)
Vitamin B₁₂ Replacement and MMA levels
Elevated values of MMA and homocysteine return to normal with vitamin B₁₂ replacement, providing further evidence of vitamin deficiency.⁶

In a series of patients with documented pernicious anemia receiving inadequate treatment, MMA and total homocysteine concentrations increased earlier than did decreases in serum vitamin B₁₂, establishing superior sensitivity of these metabolites in detecting early vitamin B₁₂ deficiency.⁷

FOLLOW-UP
After the diagnosis of vitamin B₁₂ deficiency has been made and a treatment plan has been initiated, follow-up is important to determine the patient’s response to therapy. If vitamin B₁₂ deficiency is associated with severe anemia, correction of the deficiency state should lead to a marked reticulocytosis in one to two weeks.

In mild vitamin B₁₂ deficiency, repeat measurements of serum vitamin B₁₂, homocysteine, and MMA levels two to three months after initiating treatment is recommended.¹

QUESTIONS?
If you have any questions, please contact:

Stephen Erfurth, PhD, DABCC, DABCC/TC
Director of Science and Technology
☎ 541-341-8092
☎ 800-826-3616 ext. 8092
serfurth@peacehealthlabs.org

Grant Beardsley, MS, MT(ASCP), NRCC/TC
Manager, Drug Testing Services
☎ 541-687-2134 ext. 8137
☎ 800-826-3616 ext. 8137
gbeardsley@peacehealthlabs.org

ordering information on back

REFERENCES
Serum Methylmalonic Acid (continued)

ORDERING INFORMATION

36327: Methylmalonic Acid (MMA)
Methodology: High Performance Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)
Performed: Monday–Friday
Released: Within 48 hours of testing
CPT Code: 83921

SPECIMEN REQUIREMENTS

Collect: One 7.5 mL serum separator tube (SST) or one 5 mL red top tube. Also acceptable: One 4 mL green top tube (sodium Heparin)
Handling: Allow to clot, centrifuge and separate serum or plasma from cells and pour into a plastic vial. Freeze if stored beyond 7 days.
Stability: After separation from cells; ambient 7 days, refrigerated 7 days, frozen 1 month*
Standard Volume: 3 mL serum or plasma
Minimum Volume: 1.5 mL serum or plasma
Transport: Ambient, refrigerated or frozen (if frozen, send on dry ice)
Rejection Criteria: Specimen kept at ambient temperature more than 7 days; gross hemolysis or lipemia
Reference Ranges: 0.00 – 0.40 μmol/L