Vitamin B12 and Methylmalonic Acid Testing

Policy Number: AHS–G2014 – Vitamin B12 and Methylmalonic Acid Testing	Prior Policy Name and Number, as applicable:
Initial Presentation Date: 06/01/2021 Revision Date: N/A	

I. Policy Description

Vitamin B12, also known as cobalamin, is a water-soluble vitamin required for proper red blood cell formation, key metabolic processes, neurological function, and DNA regulation and synthesis. Hematologic and neuropsychiatric disorders caused by a deficiency in B12 can often be reversed by early diagnosis and prompt treatment (Oh & Brown, 2003).

Methylmalonic acid (MMA) is produced from excess methylmalonyl-CoA that accumulates when Vitamin B12 is unavailable and is considered an indicator of functional B12 deficiency (SobczynskaMalefora et al., 2014).

Holotranscobalamin (holoTC) is the metabolically active fraction of B12 and is an emerging marker of impaired vitamin B12 status (Langan & Goodbred, 2017).

II. Related Policies

Policy Number	Policy Title
AHS-G2154	Folate Testing
AHS-M2141	Testing of Homocysteine Metabolism-Related Conditions

III. Indications and/or Limitations of Coverage

Application of coverage criteria is dependent upon an individual's benefit coverage at the time of the request. Medical Policy Statements do not ensure an authorization or payment of services. Please refer to the plan contract (often referred to as the Evidence of Coverage) for the service(s) referenced in the Medical Policy Statement. If there is a conflict between the Medical Policy Statement and the plan contract (i.e.,

Evidence of Coverage), then the plan contract (i.e., Evidence of Coverage) will be the controlling document used to make the determination.

Application of coverage criteria is dependent upon an individual's benefit coverage at the time of the request. If there is a conflict between this Policy and any relevant, applicable government policy [e.g. National Coverage Determinations (NCDs) for Medicare] for a particular member, then the government policy will be used to make the determination. For the most up-to-date Medicare policies and coverage, please visit their search website http://www.cms.gov/medicare-coverage-database/overview-and-quick-search.aspx?from2=search1.asp& or the manual website

1. Vitamin B12 testing **MEETS COVERAGE CRITERIA** in individuals being evaluated for clinical manifestations of Vitamin B12 deficiency including:

    a. Cutaneous
        I. Hyperpigmentation
        II. Jaundice
        III. Vitiligo

    b. Gastrointestinal
        i. Glossitis

    c. Hematologic
        i. Anemia (macrocytic, megaloblastic
        ii. Leukopenia
        iii. Pancytopenia
        iv. Thrombocytopenia
        v. Thrombocytosis

    d. Neuropsychiatric

        i. Areflexia

        ii. Cognitive impairment (including dementia-like symptoms and acute psychosis)

        iii. Gait abnormalities

        iv. Irritability

        v. Loss of proprioception and vibratory sense

        vi. Olfactory impairment

        vii. Peripheral neuropathy

2. Vitamin B12 testing **MEETS COVERAGE CRITERIA** when performed no sooner than 3 months after initiation of therapy for individuals undergoing treatment for vitamin B12 deficiency.

3. Screening for Vitamin B12 deficiency **MEETS COVERAGE CRITERIA** for individuals with one or more of the following risk factors:

a. *Decreased ileal absorption*

viii. Crohn disease

ix. Ileal resection

x. Tapeworm infection

xi. Patients that have undergone bariatric procedures such as Roux-en-Y gastric bypass, sleeve gastrectomy, or biliopancreatic diversion/duodenal switch

b. *Decreased intrinsic factor*

i. Atrophic gastritis

ii. Pernicious anemia

iii. Postgastrectomy syndrome

a. *Genetic*

i. Transcobalamin II deficiency

b. *Inadequate intake*

i. Alcohol abuse

ii. Patients older than 75 years or elderly individuals being evaluated for dementia

iii. Vegans or strict vegetarians (including exclusively breastfed infants of vegetarian/vegan mothers)

iv. Eating disorders

c. *Prolonged medication use*

i. Histamine H2 blocker use for more than 12 months

ii. Metformin use for more than four months

iii. Proton pump inhibitor use for more than 12 months

4. Methylmalonic acid testing **MEETS COVERAGE CRITERIA** to confirm vitamin B12 deficiency in asymptomatic high-risk patients with low-normal levels of vitamin B12 or when vitamin B12 deficiency is suspected but the serum vitamin B12 level is normal or low-normal.

5. Methylmalonic acid **MEETS COVERAGE CRITERIA** for the evaluation of inborn errors of metabolism, which is out of scope for this policy.

6. Screening for Vitamin B12 deficiency in healthy, asymptomatic individuals **DOES NOT MEET COVERAGE CRITERIA.**

7. Homocysteine testing **DOES NOT MEET COVERAGE CRITERIA** for the confirmation of vitamin B12 deficiency.

8. Holotranscobalamin testing **DOES NOT MEET COVERAGE CRITERIA** for the screening, testing or confirmation of vitamin B12 deficiency.

IV. Scientific Background

Vitamin B12 cannot be synthesized by human cells (Means Jr & Fairfield, 2019); rather, it is obtained from animal-derived dietary sources, such as meat, eggs, and dairy products (Hunt, Harrington, & Robinson, 2014), as well as fortified cereals and supplements (Zeuschner et al., 2013). Vitamin B12 deficiency is classically caused by pernicious anemia; however, with modern fortification of western diets, this condition now accounts for only a minority of cases and currently occurs most often due to malabsorption (Means Jr & Fairfield, 2019). The prevalence of vitamin B12 deficiency in the United States and United Kingdom is approximately 6% in persons younger than 60 years, reaching 20% in those older than 60 years. On the contrary, the prevalence is approximately 40% in Latin America, 70% in Kenyan school children, 80% in East Indian preschool-aged children, and 70% in East Indian adults (Hunt et al., 2014). Risk factors for deficiency include (Langan & Goodbred, 2017): decreased ileal absorption (Crohn disease, ileal resection, tapeworm infection), decreased intrinsic factor (atrophic gastritis, pernicious anemia, post-gastrectomy syndrome), genetic defects (transcobalamin II deficiency), inadequate intake (alcohol abuse, patients older than 75 years, vegans, or strict vegetarians), prolonged medication use (histamine H2 blocker use for more than 12 months, metformin use for more than four months, proton pump inhibitor use for more than 12 months).

Vitamin B12 plays an essential role in nucleic acid synthesis. Deficiency can result in cell cycle arrest in the S phase or cause apoptosis (Green, 2017) and ultimately bone marrow failure and demyelinating nervous system disease (Stabler, 2013). Vitamin B12 is also critical in the remethylation of homocysteine (Hcy), and deficiency in Vitamin B12 can lead to hyperhomocysteinemia, a condition that has been associated with various cancers, such as breast and ovarian cancers, as well as Parkinson disease (Fan et al., 2020; Hama et al., 2020).

Clinical manifestations of Vitamin B12 deficiency vary in their presence and severity from mild fatigue to severe neurologic impairment (Langan & Goodbred, 2017). Mild deficiency can present as fatigue and anemia with an absence of neurological features. Moderate deficiency may include obvious macrocytic anemia with some mild or subtle neurological features. Severe deficiency shows evidence of bone marrow suppression, clear evidence of neurological features, and risk of cardiomyopathy. Vitamin B12 deficiency can also cause glossitis and other gastrointestinal symptoms that vary with underlying diseases, such as inflammatory bowel disease or celiac disease (Means Jr & Fairfield, 2019). Early detection and correction of vitamin B12 deficiency with supplementation prevents progression to macrocytic anemia, elevated homocysteine (Hcy), potentially irreversible peripheral neuropathy, memory loss, and other cognitive deficits (Sobczynska-Malefora et al., 2014).

Analytical Validity

Both the clinical recognition of vitamin B12 deficiency and confirmation of the diagnosis by means of testing can be difficult. Several laboratory measures reflecting physiological, static, and functional B12 status have been developed (Hunt et al., 2014); however, there is no universally agreed upon gold standard assay for determining cobalamin levels in humans. The current convention is to estimate the abundance of vitamin B12 using total serum vitamin B12, despite the low sensitivity of this test (Sobczynska-Malefora et al., 2014). Two reportedly highly sensitive vitamin B12 deficiency markers are elevated levels of serum homocysteine and methylmalonic acid, but testing is expensive, and many other conditions may cause an elevation in these markers, including familial hyperhomocysteinemia, folate deficiency, levodopa therapy, and renal insufficiency (Langan & Zawistoski, 2011). Serum methylmalonic acid levels tend to be just as sensitive but more specific than serum homocysteine levels in regards to vitamin B12 deficiency testing, highlighting the former as the preferred testing method by many (Langan & Zawistoski, 2011).

An in-depth meta-analysis by Willis et al. (2011) of serum cobalamin testing included data from 54 different studies. The variability for sensitivity and specificity across the different studies ranged from 13% to 75% for sensitivity and 45% to 100% for specificity, depending on the reference standard used. Researchers conclude that "from the available evidence, diagnosis of conditions amenable to cbl [vitamin B12] supplementation on the basis of cbl [vitamin B12] level alone cannot be considered a reliable approach to investigating suspected vitamin deficiency" (Willis et al., 2011). The test measures total serum cobalamin including both serum holohaptocorrin and serum holotranscobalamin, which may mask true deficiency or falsely imply a deficient state (Hunt et al., 2014).

Vitamin B12 deficiency is present in both infant and pregnant female populations, and monitoring vitamin B12 levels is important in determining maternal and fetal health and growth. Low vitamin B12 levels during pregnancy are associated with a greater risk of preterm birth (Rogne et al., 2017). It seems that current pregnancy-specific cutoffs for vitamin B12 biomarkers are inadequate in the medical field (Schroder et al., 2019). Recently, a new study has identified a novel cutoff value in the vitamin B12

serum of newborns; the B12-related metabolite known as homocysteine (Hcy) is now recommended to have a cutoff value at "4.77 µmol/L (68.4% sensitivity, 58.3% specificity, p = .012) for the detection of vit-B12 deficiency" (Yetim et al., 2019). Other pregnancy-specific B12 biomarkers have been published. According to another study, "The central 95% reference interval limits indicated that serum total B-12 <89.9 and <84.0 pmol/L, holoTC <29.5 and <26.0 pmol/L and MMA >371 and >374 nmol/L, in the first and second trimesters, respectively, may indicate B-12 deficiency in pregnant women. The lower limits of total B-12 and holoTC and the upper limits of MMA significantly differed by ethnicity in both trimesters. According to the change point analysis, total B-12 <186 and <180 pmol/L and holoTC <62.2 and <67.5 pmol/L in the first and second trimesters, respectively, suggested an increased probability of impaired intracellular B-12 status, with no difference between ethnicities (Schroder et al., 2019)."

Elevated levels of downstream metabolites, MMA and Hcy, are commonly used as adjuvant diagnostics to confirm a suspected diagnosis of cobalamin deficiency (Berg & Shaw, 2013). The sensitivity of elevated serum MMA measurements in detecting patients with overt cobalamin deficiency is reported to be >95%; however, the specificity of this test has not been determined (Hunt et al., 2014). In a study by Rozmarič et al. (2020) the cutoff for MMA as an indicator of B12 deficiency was 0.423 µM with a specificity of 0.90 and sensitivity of 0.91 in newborns; "applying a screening algorithm including only tHCy [total homocysteine] as a second-tier test that may be feasible for many newborn screening labs, newborns with low VitB12, low HoloTC, or elevated MMA can be identified with a positive predictive value between 59% and 87%."

Serum holoTC may be a better indicator of B12-deficiency than serum cobalamin because it represents the biologically active fraction of cobalamin in humans and may be depleted first in subclinical cobalamin deficiency. HoloTC measurements appear to have slighter better sensitivity; however, the specificity of this assay remains to be determined (Oberley & Yang, 2013). It also is not yet clinically validated or available for widespread use (Langan & Goodbred, 2017).

Criteria	Sensitivity	Specificity	Pitfalls
Serum total cobalamin (<200 pg/mL)	95–97%	Uncertain, possibly <80%	*Elevated levels seen with*: Assay technical failure Occult malignancy Alcoholic liver disease Renal disease
Criteria	**Sensitivity**	**Specificity**	**Pitfalls**

			Decreased levels also seen with: Haptocorrin deficiency Folate deficiency Plasma cell myeloma HIV Pregnancy
Elevated serum methylmalonic acid	>95%	Uncertain	*Elevated levels seen with*: Renal insufficiency Hypovolemia Congenital metabolic defects Amyotrophic lateral sclerosis
Elevated serum homocysteine	>95%	Uncertain, less specific than methylmalonic acid	*Elevated levels seen with*: Folate or pyridoxine deficiency Renal insufficiency Hypovolemia Hypothyroidism Psoriasis Congenital metabolic defects Neurodegenerative disease Malignancy Medications
Decreased serum holotranscobalamin	Similar to total cobalamin	Uncertain	*Levels may be affected by*: Liver disease Macrophage activation Autoantibodies

Clinical Utility and Validity

Health Quality Ontario (HQO) performed an extensive meta-analysis of the clinical utility of B12 testing in patients with suspected dementia or cognitive decline because more than 2.9 million serum B12 tests were performed in Ontario alone in 2010 (HQO, 2013). HQO included data from eighteen different studies to address three questions:

1. "Is there an association between vitamin B12 deficiency and the onset of dementia or cognitive decline?
2. Does treatment with vitamin B12 supplementation improve cognitive function in patients with dementia or cognitive decline and vitamin B12 deficiency?

3. What is the effectiveness of oral versus parenteral vitamin B12 supplementation in those with confirmed vitamin B12 deficiency?"

They concluded that "This evidence-based analysis assessed the usefulness of serum vitamin B12 testing as it relates to brain function. This review found very low quality evidence that suggests a connection between high plasma homocysteine levels (a by-product of B vitamin metabolism in the body) and the onset of dementia. Moderate quality of evidence indicates treatment with vitamin B12 does not improve brain function. Moderate quality of evidence also indicates treatment using oral vitamin B12 supplements is as effective as injections of vitamin B12" (HQO, 2013).

Another meta-analysis, completed in 2015, utilized data from 12 studies and a total of 34,481 patients to determine if vitamin B12, vitamin B6, and folic acid supplementation affected homocysteine levels and/or reduced the risk of cardiovascular disease (Li, Li, Qi, & Shen, 2015). A combination of vitamin B12, vitamin B6, and folic acid was found to significantly reduce plasma homocysteine levels, but it did not seem to impact cardiovascular disease risk (Li et al., 2015). Therefore, it was concluded that vitamin B12 should not be utilized as a cardiovascular disease prevention method. Additional research has also concluded that the "Use of vitamin B12 in patients with elevated serum homocysteine levels and cardiovascular disease does not reduce the risk of myocardial infarction or stroke, or alter cognitive decline" (Langan & Goodbred, 2017).
In other indications, vitamin B12 has recently been utilized as a biomarker for patients undergoing therapeutic treatment for tuberculosis (TB); vitamin B12 serum concentrations were observed to have significant differences in TB patients between baseline and 6 months after anti-TB treatment (ATT), attributing the decrements in vitamin B12 to the body "reclaiming normal physiological function of the affected organs and immune function improv[ing] by cleaning or a rapid drop in bacterial load" (Gebremicael et al., 2019). Gebremicael et al. (2019) also found that HIV and HAART therapy status of TB patients at baseline had "no effect on the concentration levels of vitamin B12 and vitamin A," and HAART treatment did not affect vitamin B12 serum concentration in ATT treated HIV+/TB+ patients.

Wolffenbuttel, Heiner-Fokkema, Green, and Gans (2020) recently conducted a study obtaining data from the general population of National Health and Nutrition Examination Survey (NHANES). A total of 24462 patients were included. The authors found a positive association between low serum B12 concentration and all-cause mortality (hazard ratio [HR] = 1.39), as well as between low serum B12 concentration and cardiovascular mortality (HR = 1.64). The authors also found a positive association of high serum B12 concentration and cardiovascular mortality (HR = 1.45), although the authors noted that participants with diagnoses such as hyperlipidemia and CVD tended to use vitamin B12containing supplements more often than those without such diagnoses. However, the authors did not find an association between vitamin B12 supplement intake and mortality. This demonstrates the importance of testing for B12 in the long run to adjust dietary intake and reduce mortality.

V. Guidelines and Recommendations

American Academy of Family Physicians (AAFP) (Langan & Goodbred, 2017)

The AAFP does not recommend screening persons at average risk of vitamin B12 deficiency. Screening should be considered in patients with risk factors, and diagnostic testing should be considered in those with suspected clinical manifestations. These manifestations are listed below:

- "Cutaneous ○
  Hyperpigmentation ○
  Jaundice ○ Vitiligo
- Gastrointestinal ○ Glossitis
- Hematologic ○ Anemia (macrocytic, megaloblastic) ○ Leukopenia ○ Pancytopenia ○ Thrombocytopenia ○
  Thrombocytosis
- Neuropsychiatric ○ Areflexia ○ Cognitive impairment
  (including dementia-like symptoms and acute psychosis) ○
  Gait abnormalities ○ Irritability ○ Loss of proprioception
  and vibratory sense ○ Olfactory impairment ○
  Peripheral neuropathy"

"The recommended laboratory evaluation for patients with suspected vitamin B12 deficiency includes a complete blood count and serum vitamin B12 level. In patients with a normal or low-normal serum vitamin B12 level, complete blood count results demonstrating macrocytosis, or suspected clinical manifestations, a serum methylmalonic acid level is an appropriate next step and is a more direct measure of vitamin B12's physiologic activity; although not clinically validated or available for widespread use, measurement of holotranscobalamin, the metabolically active form of vitamin B12, is an emerging method of detecting deficiency."

AAFP also notes that different causes of vitamin B12 deficiency have corresponding "time to improvement" after initiation of treatment. For abnormalities related to "Homocysteine or methylmalonic acid level, or reticulocyte count", AAFP lists an "expected time until improvement" of one week; for neurologic symptoms; six weeks to three months; for anemia, leukopenia, mean corpuscular volume, or thrombocytopenia; eight weeks.

Finally, AAFP lists risk factors for vitamin B12 deficiency, which are listed below:

- "Decreased ileal absorption ○ Crohn disease ○ Ileal resection ○ Tapeworm infection
- Decreased intrinsic factor ○ Atrophic gastritis ○ Pernicious anemia ○ Postgastrectomy syndrome (includes Roux-en-Y gastric bypass)
- Genetic ○ Transcobalamin II deficiency
- Inadequate intake ○ Alcohol abuse ○ Patients older than 75 years ○ Vegans or strict vegetarians (including exclusively breastfed infants of vegetarian/vegan mothers)
- Prolonged medication use ○ Histamine H2 blocker use for more than 12 months ○ Metformin use for more than four months ○ Proton pump inhibitor use for more than 12 months" (Langan & Goodbred, 2017).

AAFP also comments on pernicious anemia, stating that "Patients diagnosed with vitamin B_{12} deficiency whose history and physical examination do not suggest an obvious dietary or malabsorptive etiology should be tested for pernicious anemia with anti-intrinsic factor antibodies (positive predictive value = 95%), particularly if other autoimmune disorders are present." AAFP also notes that "Patients with pernicious anemia may have hematologic findings consistent with normocytic anemia" (Langan & Goodbred, 2017).

American College of Gastroenterology (ACG) (Rubio-Tapia, Hill, Kelly, Calderwood, & Murray, 2013)

According to the ACG, "people with newly diagnosed celiac disease should undergo testing and treatment for micronutrient deficiencies. Deficiencies to be considered for testing should include, but not be limited to, iron, folic acid, vitamin D, and vitamin B12 (conditional recommendation, low level of evidence)" (Rubio-Tapia et al., 2013).

American Academy of Neurology (AAN) (Knopman et al., 2001)

"The American Academy of Neurology recommends serum vitamin B12 testing as part of the assessment of elderly patients with dementia" (Knopman et al., 2001). Currently, this guideline is being updated as of 10/26/2020.

British Committee for Standards in Haematology (Devalia, Hamilton, & Molloy, 2014)

"Serum cobalamin currently remains the first-line test, with additional second-line plasma methylmalonic acid to help clarify uncertainties of underlying biochemical/functional deficiencies. Serum holotranscobalamin has the potential as a first-line test, but an indeterminate 'grey area' may still exist. Plasma homocysteine may be helpful as a second-line test but is less specific than methylmalonic acid. The availability of these second-line tests is currently limited" (Devalia et al., 2014).

The Doctors of BC (formerly the British Columbia Medical Association) (BCMA, 2013)

The Doctors of BC recommend vitamin B12 testing for individuals with "unexplained neurologic symptoms such as paresthesias, numbness, poor motor coordination, memory lapses, or cognitive and personality changes," and anemia. They also recommend consideration of testing of elderly individuals (>75 years old), those with inflammatory bowel disease (of small intestine), gastric or small intestine resection, prolonged vegan diet, and long-term use of H2 receptor antagonists or proton pump inhibitors (at least 12 months), or metformin (at least 4 months).

American Association of Clinical Endocrinologists (AACE) and the American College of Endocrinology (ACE) and the Obesity Society (TOS) (Gonzalez-Campoy et al., 2013)

"Vitamin B12 levels should be checked periodically in older adults and patients on metformin therapy (Grade A, BEL 1). With the exception of early treatment of patients with neurologic symptoms, pernicious anemia, or malabsorptive bariatric surgery requiring parenteral (intramuscular or subcutaneous) vitamin B12 replacement, patients with vitamin B12 deficiency can generally be treated with oral vitamin B12 (1,000 µg per day of oral crystalline cobalamin) and may benefit from increasing the intake of vitamin B12 in food (Grade A, BEL 1)" (Gonzalez-Campoy et al., 2013).

American Association of Clinical Endocrinologists (AACE), the Obesity Society, and American Society for Metabolic & Bariatric Surgery (ASMBS) (Mechanick et al., 2013)

"Baseline and postoperative evaluation for vitamin B12 deficiency is recommended in all bariatric surgery and annually in those with procedures that exclude the lower part of the stomach (e.g., LSG, RYGB) (Grade B; BEL 2)" (Mechanick et al., 2013).

American Society for Metabolic and Bariatric Surgery (ASMBS) Integrated Health Nutritional Guidelines (2016 Update) (Parrott et al., 2017)

Concerning vitamin B12 screening and weight loss surgical (WLS) practices, the ASMBS states that "routine pre-WLS screening of B12 is recommended for all patients (Grade B, BEL 2)." Further, serum MMA [methylmalonic acid] testing is recommended to evaluate a possible B12 deficiency for both asymptomatic and symptomatic patients as well as in "those with history of B12 deficiency or preexisting neuropathy (Grade B, BEL 2)"

The ASMBS also makes the following recommendations for post-WLS nutrient screening:

- "Routine post-WLS screening of vitamin B12 status is recommended for patients who have undergone RYGB [Roux-en-Y gastric bypass], SG [sleeve gastrectomy], or BPD/DS [biliopancreatic diversion/duodenal switch]."
- "More frequent screening (e.g., every 3 mo) is recommended in the first post-WLS year, and then at least annually or as clinically indicated for patients who chronically use medications that exacerbate risk of B12 deficiency: nitrous oxide, neomycin, metformin, colchicine, proton pump inhibitors, and seizure medications."
- "Serum B12 may not be adequate to identify B12 deficiency. It is recommended to include serum MMA with or without homocysteine to identify metabolic deficiency of B12 in symptomatic and asymptomatic patients and in patients with history of B12 deficiency or preexisting neuropathy." (Parrott et al., 2017).

American Association of Clinical Endocrinologists/American Collee of Endocrinology (AACE/ACE), The Obesity Society (TOS), American Society for Metabolic & Bariatric Surgery (ASMBS), Obesity Medicine Association (OMA), and American Society of Anesthesiologists (ASA) (2019 Update) (Mechanick et al., 2019)

The AACE/ACE, TOS, ASMBS, OMA, and ASA published clinical practice guidelines for perioperative nutrition, metabolic, and nonsurgical support of patients undergoing bariatric procedures in 2019. In the preprocedure [sic] checklist, the recommendation includes "nutrient screening with iron studies, B12, and folic acid (RBC folate, homocysteine, methylmalonic acid optional), and 25-vitamin D (vitamins A and E optional); consider more extensive testing in patients undergoing malabsorptive procedures based on symptoms and risks. In postprocedure [sic], for early postoperative care, vitamin B12 should be assessed "as needed for normal range levels," and in follow-up "annually; MMA and HCy optional; then q 3-6 mo if supplemented)" (Mechanick et al., 2019). In addition, the societies state:

- Vitamin B12 screening is "recommended for patients who have undergone RYGB [Roux-en-Y gastric bypass], SG [sleeve gastrectomy], or BPD/DS (biliopancreatic diversion/duodenal switch]"
- "Patients who become pregnant following bariatric procedure should have nutritional surveillance and laboratory screening for nutrient deficiencies every trimester, including iron, folate, vitamin B12, vitamin D, and calcium, and if after a malabsorptive procedure, fat-soluble vitamins, zinc, and copper (Grade D)
- Baseline and annual post-bariatric procedure evaluation for vitamin B12 deficiency should be performed in all patients (Grade B; BEL 2)

- More frequent aggressive case finding (e.g., every 3 mo) should be performed in the first postoperative year and then at least annually or as clinically indicated for patients who chronically use medications that exacerbate risk of B12 deficiency, such as nitrous oxide, neomycin, metformin, colchicine, proton-pump inhibitors, and seizure medications (Grade B, BEL 2)
- Because serum B12 may not be adequate to identify B12 deficiency, consider measuring serum methylmalonic acid, with or without homocysteine, to identify a metabolic deficiency of B12 in symptomatic and asymptomatic patients and in patients with a history of B12 deficiency or preexisting neuropathy (Grade B, BEL 2)
- B12 status should be assessed in patients on higher-dose folic acid supplementation (>1000 µg/d) to detect a masked B12- deficiency state (Grade D)" (Mechanick et al., 2019).

British Obesity & Metabolic Surgery Society (BOMSS) (2020 Update) (O'Kane et al., 2020)

The BOMSS released 2020 perioperative and postoperative guidelines on biochemical monitoring and micronutrient replacement for patients undergoing bariatric surgery. On measuring vitamin B12 concentrations, the BOMSS has included checking a "full blood count including haemoglobin, ferritin, folate and vitamin B12 levels" in their preoperative nutritional assessment with a grade B and evidence level (EL) of 2. For postoperative care and biochemical monitoring, the BOMSS stated,

- "check vitamin B12 levels at regular intervals following, SG, RYGB and malabsorptive procedures such as BPD/DS (Grade B, EL2)
- Consider the following frequency of vitamin B12 levels: 3, 6 and 12 months in the first year and at least annually thereafter so that changes in status may be detected (GPP – good practice point)"

With relation to folic acid deficiency, O'Kane et al. (2020) mentions, "check and treat for vitamin B12 deficiency, before initiating folic acid treatment to avoid precipitation of subacute combined degeneration of the spinal cord (Grade D, EL4)." For any presence of neurological symptoms/Wernicke's encephalopathy, the guidelines recommend to "check for vitamin B12, copper, and vitamin E deficiencies and treat (GPP)." In pregnant women after undergoing bariatric surgery, checking for vitamin B12 deficiency, among other nutritional deficiencies, has been recommended for each trimester and prior to additional folic acid supplementation in the preconception period (O'Kane et al., 2020).

Guidelines for Diagnosis and Management of the Cobalamin-related Remethylation Disorders cblC, cblD, cblE, cblF, cblG, cblJ and MTHFR Deficiency (Huemer et al., 2017).

This international consortium of scientists from Europe and the U.S. issued guidelines "within the frame of the 'European network and registry for homocystinurias and methylation defects' (E-HOD) project." For Recommendation 5, they state (Quality of the evidence: moderate), "we strongly recommend that in the case of high total homocysteine, plasma and urine samples for determination of MMA, methionine, folate and vitamin B12 are to be obtained before treatment is started" (Huemer et al., 2017).

The American Diabetes Association (ADA) (ADA, 2020a, 2020b)

The ADA states that in patients with type 2 diabetes, the long-term use of metformin may be associated with a vitamin B12 deficiency; therefore, a Grade B recommendation has been made which states that the "periodic measurement of vitamin B12 levels should be considered in metformintreated patients, especially in those with anemia or peripheral neuropathy" (ADA, 2020a). The ADA also recommended that "measurement of vitamin B12 levels should be considered in patients with type 1 diabetes and peripheral neuropathy or unexplained anemia" as well (ADA, 2020b).

American Psychiatric Association (APA) (Yager et al., 2006)

The APA released guidelines which include a table of the physical complications of anorexia nervosa and potential laboratory testing methods. This table contains a few vitamin assays that may be used to monitor endocrine or metabolic processes including vitamin B12 assays "in severe cases" (Yager et al., 2006).

U.S. Preventative Services Task Force (USPSTF) (Langan & Zawistoski, 2011)

Currently, the USPSTF has not published guidelines for vitamin B12 deficiency screenings of asymptomatic or low-risk adults.

VI. State and Federal Regulations, as applicable

A search of the FDA Device database on 10/20/2020 using the term "vitamin B12" yielded 44 results. Additionally, many labs have developed specific tests that they must validate and perform in house. These laboratory-developed tests (LDTs) are regulated by the Centers for Medicare and Medicaid (CMS) as high-complexity tests under the Clinical Laboratory Improvement Amendments of 1988 (CLIA '88). As an LDT, the U. S. Food and Drug Administration has not approved or cleared this test; however, FDA clearance or approval is not currently required for clinical use.

VII. Applicable CPT/HCPCS Procedure Codes

Billing applicable codes is not a guarantee of payment; see Section III for indications and limitations of coverage that may affect payment

Code Number	Code Description
82607	Cyanocobalamin (Vitamin B-12)
83090	Homocysteine

83921	Organic acid, single, quantitative
84999	Unlisted chemistry procedure

VIII. Evidence-based Scientific References

ADA. (2019). Standards of Medical Care in Diabetes-2019 Abridged for Primary Care Providers. *Clin Diabetes, 37*(1), 11-34. doi:10.2337/cd18-0105

ADA. (2020a). 3. Prevention or Delay of Type 2 Diabetes: Standards of Medical Care in Diabetes—2020. *Diabetes Care, 43*(Supplement 1), S32-S36. doi:10.2337/dc20-S003

ADA. (2020b). 4. Comprehensive Medical Evaluation and Assessment of Comorbidities: Standards of Medical Care in Diabetes—2020. *Diabetes Care, 43*(Supplement 1), S37-S47. doi:10.2337/dc20-S004

BCMA. (2013, May 1). Cobalamin (vitamin B12) Deficiency - Investigation & Management - Province of British Columbia. *Clinical Practice Guidelines.* Retrieved from https://www2.gov.bc.ca/gov/content/health/practitioner-professional-resources/bcguidelines/vitamin-b12

Berg, R. L., & Shaw, G. R. (2013). Laboratory evaluation for vitamin B12 deficiency: the case for cascade testing. *Clin Med Res, 11*(1), 7-15. doi:10.3121/cmr.2012.1112

Devalia, V., Hamilton, M. S., & Molloy, A. M. (2014). Guidelines for the diagnosis and treatment of cobalamin and folate disorders. *Br J Haematol, 166*(4), 496-513. doi:10.1111/bjh.12959

Fan, X., Zhang, L., Li, H., Chen, G., Qi, G., Ma, X., & Jin, Y. (2020). Role of homocysteine in the development and progression of Parkinson's disease. *Ann Clin Transl Neurol*. doi:10.1002/acn3.51227

Gebremicael, G., Alemayehu, M., Sileshi, M., Geto, Z., Gebreegziabxier, A., Tefera, H., . . . Kassa, D. (2019). The serum concentration of vitamin B12 as a biomarker of therapeutic response in tuberculosis patients with and without human immunodeficiency virus (HIV) infection. *Int J Gen Med, 12*, 353-361. doi:10.2147/ijgm.S218799

Gonzalez-Campoy, J. M., St Jeor, S. T., Castorino, K., Ebrahim, A., Hurley, D., Jovanovic, L., . . . Thomas, K. T. (2013). Clinical practice guidelines for healthy eating for the prevention and treatment of metabolic and endocrine diseases in adults: cosponsored by the American Association of Clinical Endocrinologists/the American College of Endocrinology and the Obesity Society. *Endocr Pract, 19 Suppl 3*, 1-82. doi:10.4158/ep13155.gl

Green, R. (2017). Vitamin B12 deficiency from the perspective of a practicing hematologist. *Blood, 129*(19), 2603-2611. doi:10.1182/blood-2016-10-569186

Hama, Y., Hamano, T., Shirafuji, N., Hayashi, K., Ueno, A., Enomoto, S., . . . Nakamoto, Y. (2020). Influences of Folate Supplementation on Homocysteine and Cognition in Patients with Folate Deficiency and Cognitive Impairment. *Nutrients, 12*(10). doi:10.3390/nu12103138

HQO. (2013). Vitamin B12 and cognitive function: an evidence-based analysis. *Ont Health Technol Assess Ser, 13*(23), 1-45. Retrieved from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3874776/

Huemer, M., Diodato, D., Schwahn, B., Schiff, M., Bandeira, A., Benoist, J. F., . . . Dionisi-Vici, C. (2017). Guidelines for diagnosis and management of the cobalamin-related remethylation disorders cblC, cblD, cblE, cblF, cblG, cblJ and MTHFR deficiency. *J Inherit Metab Dis, 40*(1), 21-48. doi:10.1007/s10545-016-9991-4

Hunt, A., Harrington, D., & Robinson, S. (2014). Vitamin B12 deficiency. *Bmj, 349*, g5226. doi:10.1136/bmj.g5226

Knopman, D. S., DeKosky, S. T., Cummings, J. L., Chui, H., Corey–Bloom, J., Relkin, N., . . . Stevens, J. C. (2001). Practice parameter: Diagnosis of dementia (an evidence-based review). *Neruology, 56*(9), 1143-1153. doi:10.1212/WNL.56.9.1143

Langan, R. C., & Goodbred, A. J. (2017). Vitamin B12 Deficiency: Recognition and Management. *Am Fam Physician, 96*(6), 384-389.

Langan, R. C., & Zawistoski, K. J. (2011). Update on vitamin B12 deficiency. *Am Fam Physician, 83*(12), 1425-1430.

Li, J., Li, B., Qi, J., & Shen, B. (2015). [Meta-analysis of clinical trials of folic acid, vitamin B12 and B6 supplementation on plasma homocysteine level and risk of cardiovascular disease]. *Zhonghua Xin Xue Guan Bing Za Zhi, 43*(6), 554-561.

Means Jr, R. T., & Fairfield, K. M. (2019, October 22). Causes and pathophysiology of vitamin B12 and folate deficiencies. *UpToDate.* Retrieved from https://www.uptodate.com/contents/causesandpathophysiology-of-vitamin-b12-and-folatedeficiencies?source=see_link

Mechanick, J. I., Apovian, C., Brethauer, S., Garvey, W. T., Joffe, A. M., Kim, J., . . . Still, C. D. (2019). CLINICAL PRACTICE GUIDELINES FOR THE PERIOPERATIVE NUTRITION, METABOLIC, AND NONSURGICAL SUPPORT OF PATIENTS UNDERGOING BARIATRIC PROCEDURES - 2019 UPDATE: COSPONSORED BY AMERICAN ASSOCIATION OF CLINICAL ENDOCRINOLOGISTS/AMERICAN COLLEGE OF ENDOCRINOLOGY, THE OBESITY SOCIETY, AMERICAN SOCIETY FOR METABOLIC & BARIATRIC SURGERY, OBESITY MEDICINE ASSOCIATION, AND AMERICAN SOCIETY OF ANESTHESIOLOGISTS - EXECUTIVE SUMMARY. *Endocr Pract, 25*(12), 1346-1359. doi:10.4158/gl2019-0406

Mechanick, J. I., Youdim, A., Jones, D. B., Garvey, W. T., Hurley, D. L., McMahon, M. M., . . . Brethauer, S. (2013). Clinical practice guidelines for the perioperative nutritional, metabolic, and nonsurgical support of the bariatric surgery patient--2013 update: cosponsored by American Association of Clinical Endocrinologists, the Obesity Society, and American Society for Metabolic & Bariatric Surgery. *Endocr Pract, 19*(2), 337-372. doi:10.4158/ep12437.gl

O'Kane, M., Parretti, H. M., Pinkney, J., Welbourn, R., Hughes, C. A., Mok, J., . . . Barth, J. H. (2020). British Obesity and Metabolic Surgery Society Guidelines on perioperative and postoperative biochemical monitoring and micronutrient replacement for patients undergoing bariatric surgery—2020 update. *Obesity Reviews, 21*(11), e13087. doi:10.1111/obr.13087

Oberley, M. J., & Yang, D. T. (2013). Laboratory testing for cobalamin deficiency in megaloblastic anemia. *Am J Hematol, 88*(6), 522-526. doi:10.1002/ajh.23421

Oh, R., & Brown, D. L. (2003). Vitamin B12 deficiency. *Am Fam Physician, 67*(5), 979-986. Retrieved from http://dx.doi.org/

Parrott, J., Frank, L., Rabena, R., Craggs-Dino, L., Isom, K. A., & Greiman, L. (2017). American Society for Metabolic and Bariatric Surgery Integrated Health Nutritional Guidelines for the Surgical Weight Loss Patient 2016 Update: Micronutrients. *Surg Obes Relat Dis, 13*(5), 727-741. doi:10.1016/j.soard.2016.12.018

Rogne, T., Tielemans, M. J., Chong, M. F., Yajnik, C. S., Krishnaveni, G. V., Poston, L., . . . Risnes, K. R. (2017). Associations of Maternal Vitamin B12 Concentration in Pregnancy With the Risks of Preterm Birth and Low Birth Weight: A Systematic Review and Meta-Analysis of Individual Participant Data. *Am J Epidemiol, 185*(3), 212-223. doi:10.1093/aje/kww212

Rozmarič, T., Mitulović, G., Konstantopoulou, V., Goeschl, B., Huemer, M., Plecko, B., . . . Zeyda, M. (2020). Elevated Homocysteine after Elevated Propionylcarnitine or Low Methionine in Newborn Screening Is Highly Predictive for Low Vitamin B12 and Holo-Transcobalamin Levels in Newborns. *Diagnostics (Basel), 10*(9). doi:10.3390/diagnostics10090626

Rubio-Tapia, A., Hill, I. D., Kelly, C. P., Calderwood, A. H., & Murray, J. A. (2013). ACG clinical guidelines: diagnosis and management of celiac disease. *Am J Gastroenterol, 108*(5), 656-676; quiz 677. doi:10.1038/ajg.2013.79

Schroder, T. H., Tan, A., Mattman, A., Sinclair, G., Barr, S. I., Vallance, H. D., & Lamers, Y. (2019). Reference intervals for serum total vitamin B12 and holotranscobalamin concentrations and their change points with methylmalonic acid concentration to assess vitamin B12 status during early and mid-pregnancy. *Clin Chem Lab Med*. doi:10.1515/cclm-2018-1337

Sobczynska-Malefora, A., Gorska, R., Pelisser, M., Ruwona, P., Witchlow, B., & Harrington, D. J. (2014). An audit of holotranscobalamin ("Active" B12) and methylmalonic acid assays for the assessment of vitamin B12 status: application in a mixed patient population. *Clin Biochem, 47*(12), 82-86. doi:10.1016/j.clinbiochem.2013.08.006

Stabler, S. P. (2013). Clinical practice. Vitamin B12 deficiency. *N Engl J Med, 368*(2), 149-160. doi:10.1056/NEJMcp1113996

Willis, C. D., Elshaug, A. G., Milverton, J. L., Watt, A. M., Metz, M. P., & Hiller, J. E. (2011). Diagnostic performance of serum cobalamin tests: a systematic review and meta-analysis. *Pathology, 43*(5), 472-481. doi:10.1097/PAT.0b013e3283486435

Wolffenbuttel, B. H. R., Heiner-Fokkema, M. R., Green, R., & Gans, R. O. B. (2020). Relationship between serum B12 concentrations and mortality: experience in NHANES. *BMC Medicine, 18*(1), 307. doi:10.1186/s12916-020-01771-y

Yager, J., Devlin, M., Halmi, K., Herzog, D., Mitchell, J., Powers, P., & Zerbe, K. (2006). Treatment of patients with eating disorders, third edition. American Psychiatric Association. *Am J Psychiatry, 163*(7 Suppl), 4-54. Retrieved from https://www.ncbi.nlm.nih.gov/pubmed/?term=PRACTICE+GUIDELINE+FOR+THE+Treatment+of+Patients+With+Eating+Disorders+Third+Edition

Yetim, A., Aygun, E., Yetim, C., Ucar, A., Karakas, Z., Gokcay, G., . . . Dagoglu, T. (2019). Measurement of serum vitamin B12-related metabolites in newborns: implications for new cutoff values to detect B12 deficiency. *J Matern Fetal Neonatal Med*, 1-9. doi:10.1080/14767058.2019.1633301

Zeuschner, C. L., Hokin, B. D., Marsh, K. A., Saunders, A. V., Reid, M. A., & Ramsay, M. R. (2013). Vitamin B(1)(2) and vegetarian diets. *Med J Aust, 199*(4 Suppl), S27-32. Retrieved from http://dx.doi.org/

IX. Revision History

Revision Date	Summary of Changes
06/01/2021	Initial presentation