Vitamin D, 25 Hydroxy by LC-MS/MS

**CPT Code:** 82306  
**Order Code:** C339  
**ABN Requirement:** No  
**Synonyms:** Vitamin D (Total); 25-Hydroxycalciferol; 25-OH-D; Cholecalciferol Metabolite; Vitamin D; Vit D; Total Vitamin D  
**Specimen:** EDTA Plasma or Serum  
**Volume:** 0.5 mL  
**Minimum Volume:** 0.2 mL  
**Container:** EDTA (Lavender Top tube) or Gel-barrier tube (SST, Tiger Top)

**Collection:**

EDTA Plasma:

1. Draw and gently invert 8 to 10 times.  
2. Centrifuge for 10 minutes.  
3. Pre-squeeze transfer pipet bulb and draw off approximately 2/3 of the upper plasma layer.  
   **Note:** This ensures that the buffy coat and red cells remain undisturbed.  
4. Aliquot plasma into labeled transport tube and cap tightly. Discard original tube.  
5. Store transport tube refrigerated at 2-8°C until ready to ship.

Serum:

1. Collect and label sample according to standard protocols.  
2. Gently invert tube 5 times immediately after draw. DO NOT SHAKE.  
3. Allow blood to clot 30 minutes.  
4. Centrifuge for 10 minutes.

**Special Instructions:** Minimize sample exposure to direct sunlight.

**Transport:** Store serum at 2°C to 8°C after collection and ship the same day per packaging instructions provided with the Cleveland HeartLab shipping box.
Stability:

- **Ambient (15-25°C):** 21 days
- **Refrigerated (2-8°C):** 21 days
- **Frozen (-20°C):** 185 days
- **Deep frozen (-70°C):** 185 days

**Causes of Rejection:** Specimens other than EDTA plasma or serum; improper labeling; samples not stored properly; samples older than stability limits; or grossly hemolyzed samples.

**Methodology:** High Performance Liquid Chromatography/Tandem Mass Spectrometry (LC-MS/MS)

**Turn Around Time:** 3 to 5 days

**Relative Risk - Status of Vitamin D Sufficiency:**

**Comment:** Therapy is based on measurement of Total 25-OHD, with levels <20 ng/mL indicative of Vitamin D deficiency, while levels between 20 ng/mL and 30 ng/mL suggest insufficiency. Optimal levels are ≥30 ng/mL. Vitamin-D is fat-soluble and therefore inadvertent or intentional ingestion of excessively high amounts could be toxic. Studies in children and adults suggest blood levels would need to exceed 150 ng/ml before there is any concern. Holick MF, Binkley NC, Bischoff-ferrari HA, et al. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. J Clin Endocrinol Metab. 2011;96(7):1911-30.

**Use:** Monitoring vitamin D status

**Clinical Significance:** Vitamin D is a fat-soluble vitamin and plays a major role in calcium homeostasis. Deficiency of vitamin D and subsequent hypocalcemia have been linked to many skeletal disorders including osteoporosis. Recent studies have shown evidence that vitamin D is linked to protecting the body from a wide range of diseases. Diseases linked to vitamin D deficiency include: stroke, cardiovascular disease, osteoporosis, and several forms of cancer, multiple sclerosis, rheumatoid arthritis, and type 1 and type 2 diabetes.

Although 25-hydroxy vitamin D is further hydroxylated in the kidneys to its biologically active form, 1, 25-dihydroxy vitamin D, the half-life of 1, 25 dihydroxy
vitamin D is very short and its serum levels do not correlate well with overall vitamin D levels. The measurement of serum 25-hydroxy vitamin D is the optimal indicator of biological reservoir of vitamin D.

The CPT codes provided are based on AMA guidelines and are for informational purposes only. CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.