Osteocalcin

Method:	Enzyme-linked immunosorbent assay (ELISA)
Kit Manufacturer:	ALPCO Diagnostics, Salem, NH
Description:	The hOST-EASIA is a solid phase Enzyme Amplified Sensitivity Immunoassay performed on microtiterplates. The assay uses monoclonal antibodies (MAbs) directed against distinct epitopes of human osteocalcin. Calibrators and samples react with the capture monoclonal antibody (MAb 1) coated on the microtiter wells and with a monoclonal antibody (MAb 2) labelled with horseradish peroxidase (HRP). After an incubation period, which allows the formation of a sandwich: coated MAb 1 – human osteocalcin – MAb 2 – HRP, the microtiterplate is washed to remove unbound enzyme labeled antibody. Bound enzyme-labeled antibody is measured through a chromogenic reaction. Chromogenic solution (TMB, ready for use) is added and incubated. The reaction is stopped with the addition of Stop Solution and the microtiterplate is then read at the appropriate wavelength. The amount of substrate turnover is determined colorimetrically by measuring the absorbance, which is proportional to the osteocalcin concentration. A calibration curve is plotted and the OST concentration in samples is determined by interpolation from the calibration curve. The use of the EASIA reader (linearity up to 3 OD units) and a sophisticated data reduction method (polychromatic data reduction) result in a high sensitivity in the low range and in an extended calibration range.

Collection and Performance Characteristics

Tube type:	Preferred: SST Alternate: EDTA or Heparin
Minimum Volume:	2 X 0.1 mL (special aliquot tube) Reconstitute 1 aprotinin pellet (protease inhibitor) with 2mL water. Pipette 10 ul of the solution into 100 uL of serum. Store unused cocktail tubes at 4'C for up to 6 months. Aliquot 2 x 0.1 ml serum into 2 ml Micro tubes with skirted base containing Inhibitor Cocktail, cap securely. Store aliquoted samples at -80'C until analysis is performed.
Lowest Reportable Value:	0.4 ng/mL
Dynamic Range:	0.4-75.0 ng/mL
Precision:	Intra assay variation 5.5-5.7% Inter assay variation 4.8-5.5%
Reference Range:	5-25 ng/mL