Introduction

25-OH Vitamin D and D2 metabolites have emerged in recent years as molecules of clinical diagnostic importance. Furthermore, LC/MS/MS technology has been demonstrated as the most accurate tool for quantification of these metabolites in serum or plasma. The most pressing need is for a combined, automated sample preparation and analysis system. Herein we report on our development of an integrated and automated solid phase extraction (SPE) method and LC-MS/MS system for quantification of 25-OH Vitamin D3 and D2 metabolites in serum. The key advance is enhanced automation that is integrated into the same platform as the LC-MS/MS system instrument.

Results (Agilent)

Agilent has demonstrated and LOD of 3 pL, linearity between 3 and 150 pL and inter-day imprecision of 8.5%

Example 25-OH Vitamin D calibration

Results (Agilent, preliminary)

The UTAK Vitamin D Plus controls were tested for absence/presence of Vitamin D3. In vivo, Vitamin D3 was chosen as the internal standard for preliminary investigations. An initial assessment of linearity was made using replicate samples of the Tri Level Vitamin D Serum Toxicology Controls. Regression of the ratios of the 25-OH Vitamin D metabolite peak areas to Vitamin D (preliminary IS) against the verified Vitamin D metabolite concentrations (11.1, 27.6 and 65.1 ng/mL for 25-OH Vitamin D3; 13.6, 38.4 and 97.2 ng/mL for 25-OH Vitamin D2) suggested a linear relationship for each of the two metabolites.

References


For Further Information

www.Opsans.com
Contact Ken Gamble at Kim.Gamble@Microliter.com
Microliter Analytical Supplies, PO Box 808, Suwanee, GA 30024
(770) 233-7840

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