Validation of the First Automated Chemiluminescence Protein-Binding Assay for the Detection of 25-Hydroxycalciferol

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SUMMARY

Background: Measurement of 25-hydroxyvitamin D, 25(OH)D is the ideal parameter to indicate the access of the organism to vitamin D. Numerous studies have shown that serum levels of 25(OH)D are the best markers of vitamin D deficiency, normal vitamin D supply or vitamin D intoxication. The aim of the study was to validate a beta-site version of the first fully automated chemiluminescence protein-binding assay (CLPBA) for the detection of 25-hydroxycalciferol. Methods: The newly developed CLPBA run on the Nichols Advantage² Specialty Systems was compared to an inhouse radioimmunoassay (RIA), focusing on the major assay features such as imprecision, functional sensitivity, linearity, method comparison and suitability of serum or EDTA-plasma as well as establishing a preliminary reference range. Results: Within-run imprecision is ~ 4.5% and total imprecision ~ 6% respectively (NCCLS protocol), functional sensitivity 6.8 µg/l. With mean recovery values of 96.9% and 98.7% for two diluted serum samples linearity is given over the measuring range. Due to different calibrations used for RIA and CLPBA the CLPBA reads approximately 70% lower (CLPBA = 0.321xRIA + 0.571, n = 469) but correlates well with the RIA (r = 0.9045). Method comparison with HPLC reveals a regression line of CLPBA = 0.8921xHPLC + 0.1358 (n = 54, r = 0.9117). Serum or EDTA-plasma is not equally suitable. Plasma samples read on average 5 µg/l higher than serum samples. The preliminary reference range is 11 µg/l to 84 µg/l (95% of all values). Conclusion: The validated 25(OH)D CLPBA is a very promising alternative to established commercially available 25(OH)D measurements and will, with its use on a fully automated platform, simplify the reliable quantification of 25-hydroxycalciferol significantly. (Clin. Lab. 2001;47:357-365)