

Quantification of human IL-6 using VICTOR Kira monochromator-based multimode plate reader.

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Introduction

Interleukin-6 (IL-6) is a multifunctional cytokine involved in immune regulation, inflammation, and disease progression. Accurate quantification of IL-6 is essential for immunology research, biomarker studies, and therapeutic development. ELISA remains the gold standard for cytokine measurement due to its sensitivity and reproducibility.

The LEGEND MAX™ Human IL-6 Kit from BioLegend provides a robust solution for detecting IL-6 in serum, plasma, and cell culture supernatants. This application note demonstrates the performance of the VICTOR Kira™ multimode plate reader for human IL-6 ELISA using the LEGEND MAX Human IL-6 ELISA Kit (cat. no.: 430507).

The VICTOR Kira reader offers several advanced features that make it ideal for ELISA and other biochemical assays. Its monochromator-based optics, covering 230–1000 nm, provide flexibility for precise wavelength selection.

The instrument supports a high optical density (OD) range of up to 4.0, ensuring accurate readings even at saturation points. Low detection noise enhances sensitivity, while endpoint and kinetic reading modes allow versatility of the assay. Additional features, such as plate shaking and temperature control, contribute to reproducibility and consistent assay performance.

VICTOR Kira monochromator-based multimode plate reader



Materials and methods

The LEGEND MAX Human IL-6 Kit (cat. no.: 430507) includes a pre-coated 96-well strip microplate, detection antibody, human IL-6 standard, Avidin-HRP solution, assay buffer, wash buffer (20X), substrate solution, stop solution, and plate sealers. The standard range for the assay is 500-7.8 pg/mL. The procedure involves adding standards and samples to pre-coated wells, incubating with detection antibody and Avidin-HRP, followed by substrate addition and stop solution. Absorbance was measured at 450 nm with a correction wavelength of 570 nm using the VICTOR Kira multimode plate reader.



| Figure 1: LEGEND MAX kit, BioLegend

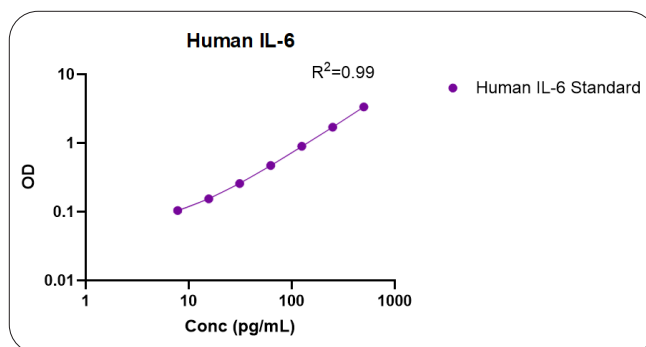
Data analysis

Data analysis was performed using GraphPad Prism 10, which enabled nonlinear regression for curve fitting, calculation of R^2 values, and interpolation of unknown sample concentrations.

The 4PL curve fitting graph can also be plotted with the VICTOR Kira control software.

Results

The standard curve for human IL-6 showed excellent performance with a correlation coefficient (R^2) of 0.99, confirming assay reliability. The curve demonstrates accurate quantification across the dynamic range of the assay.

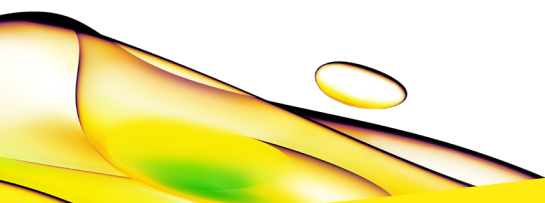


| Figure 2: Human IL-6 ELISA standard curve

Conclusion

The VICTOR Kira reader delivered accurate detection across the full concentration range and demonstrated excellent reproducibility, aided by plate shaking and temperature control. Its monochromator optics allow dual-wavelength correction, which is essential for minimizing background interference.

This study confirms the suitability of the VICTOR Kira multimode plate reader for ELISA assays. The measured dynamic range and assay linearity were in line with the LEGEND MAX Human IL-6 ELISA Kit manual.



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