

Protein L (ProL) Probes

Catalog No. 160007

OVERVIEW

Gator™ Protein L (ProL) Probes are useful in measuring the concentration and kinetics of antibodies. Specifically, Protein L binds specifically to certain subtypes of kappa light chain, including human VkI, VkIII and VkIV subtypes, and mouse VkI subtype. As there is no heavy chain involved in binding interaction, Protein L can bind to all antibody classes (IgG, IgM, IgA, IgE and IgD), as well as single chain variable fragments (scFv) and Fab fragments. These probes can be regenerated and reused for multiple experiments.

MATERIALS REQUIRED

Protein L Probes	Catalog No. 160007
Max Plate	Catalog No. 130062
Black Plates	Greiner 655209
Quantitation (Q) Buffer	Catalog No. 120010
Kinetics (K) Buffer	Catalog No. 120011
Regeneration Buffer	10mM Glycine pH 1.5

STORAGE

Store at room temperature in the foil pouch, ensuring zipper is fully sealed to avoid humidity/moisture contamination. In high-humidity environments, storage inside a dry cabinet is recommended.

GENERAL APPLICATIONS

1. Quantitation of crude or purified samples of immunoglobulins
2. Kinetics of interactions of an antigen with an antibody
3. Determination of concentration of an antibody and interaction with antigen in one run (QKR)

GENERAL METHODS

Sample Volume

Black Plate: 200 μ L (180 μ L minimum)
 Max Plate: 250 μ L (280 μ L maximum)

Pre-wet Conditions

250 μ L assay buffer (Q or K) in Max Plate,
 5 min at 1000 rpm

Speed

Q	Standard Protocol: 400 rpm, 120 seconds; 1 μ g/mL – 2000 μ g/mL
	High-Sensitivity Protocol: 1000 rpm, 300 seconds; 25 ng/mL – 500 μ g/mL
K	1000 rpm
Q K R	Use 400 or 1000 rpm for the quantitation step (based on concentration) and 1000 rpm for the kinetics steps

Rapid Quantitation of Unknown Samples

Quantitation of crude and purified unknown samples can rapidly be performed using the Q assay preset on the Gator™ software. For accurate results, make a standard curve of known concentrations in the same buffer as the unknowns. The linear range of ProL probes is 25 ng/mL to 2 mg/mL. Probes can be regenerated in between samples to analyze an entire plate of samples in one run.

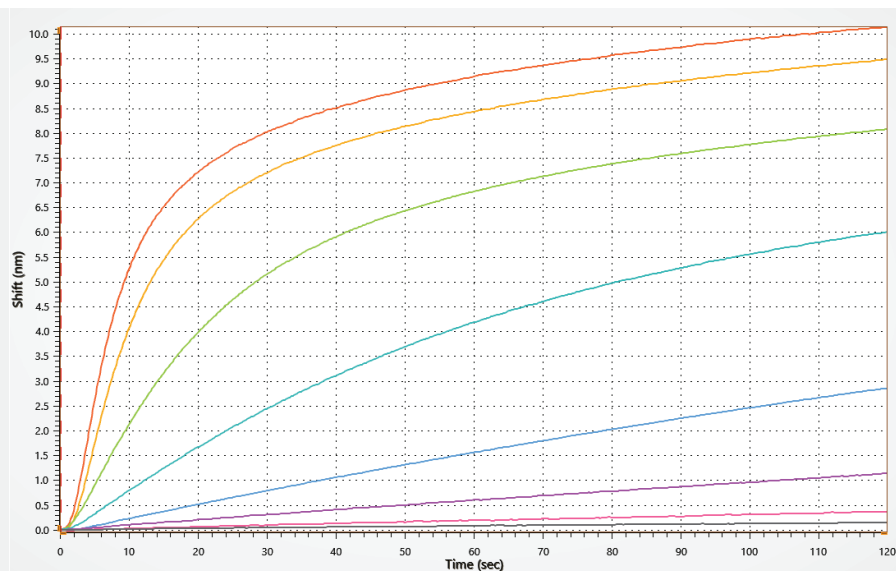


Figure 1: Binding curve of human IgG to ProL probes (1 μ g/mL to 700 μ g/mL in Q Buffer). Assay performed using standard protocol (400 rpm for 120 seconds).

Regeneration to Save on Consumables

ProL probes can be regenerated using the Gator™ software. (Settings are in Assay Setup.) Regeneration buffer and neutralization buffer (Q or K buffer) should be placed in adjacent wells in either the Black Plate or the Max Plate. For ProL probes, 3 cycles of 5 seconds for regeneration is recommended. Regeneration before assay is recommended to ensure run-to-run consistency. After regeneration, probes can be stored in assay buffer and kept at 4°C for >2 weeks.

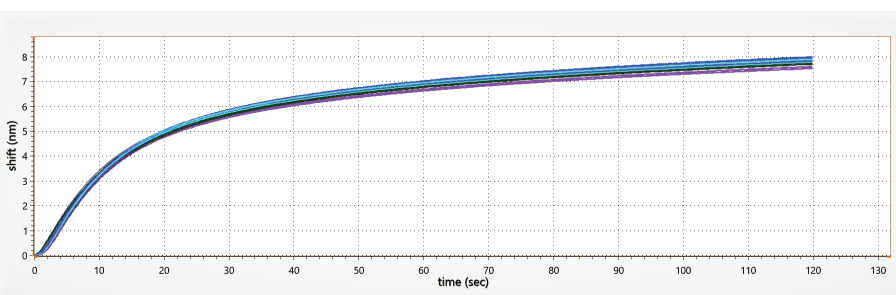


Figure 2: 40 consecutive measurements of human IgG (300 μ g/mL in Q Buffer) on the same ProL probe with regeneration. Assay performed using standard protocol (400 rpm for 120 sec) using the buffer listed under Materials Required.