

Protein A Biosensors

For determination of antibody concentration

Key features

- Direct measurement of immunoglobulins (IgG)
- Assay samples without centrifugation
- Broad dynamic range
- Fast turnaround of results
- Correlates to HPLC

Quick facts

- Dynamic range: 1–500 µg/mL for most proteins
- Throughput: 8 samples in ~2 minutes, 96 samples in ~32 minutes
- Precision/accuracy: < 10% CVs
- Limit of detection: typically 1 µg/mL

Bioprocessing applications

Accurate antibody quantitation is critical to selecting cell lines for developing and optimizing antibody production. Traditional methods for measuring antibody concentration include HPLC, ELISA and densitometry—all of which have long analysis times, lack of specificity, and precision.

Using Dip and Read™ Protein A (ProA) biosensors with the Octet® system streamlines a variety of bioprocessing applications by providing precise results which require minimal sample handling and give rapid turnaround of results.

- Cell culture screening
- Process development
- Manufacturing
- Protein purification

Dynamic range

A dynamic range of 2–3 logs is typical. The actual range will be protein-dependent. Protein A biosensors have been shown to quantitate in the range of 1–500 µg/mL for polyclonal human IgGs.

Sample types

Protein A biosensors have been tested on the Octet system with human antibodies and Fc fusion proteins.

Correlation to HPLC

A series of bioreactor samples were assayed both on the Octet system using Protein A biosensors and HPLC.

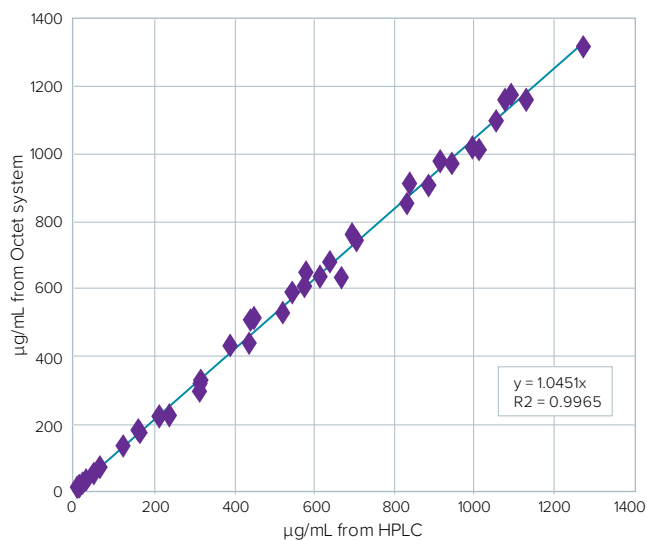


Figure 1: Correlation of sample concentrations determined using the Octet system and HPLC.

Protein A assay principle

The Protein A assay for determining protein concentration is based on the rate of binding of a protein of interest to the bio-sensor surface. Different protein concentrations result in different binding rates. Octet software calculates the binding rates from standards with known values to generate a standard curve — the binding rate of each standard is proportional to its concentration. Concentrations of experimental samples are calculated based on their binding rate compared to that of the known concentrations that make up the standard curve.

Recognition of human IgG Isotypes

ForteBio's Protein A biosensors have been shown to recognize IgG1, IgG2 and IgG4. Little or no binding occurs with IgG3.

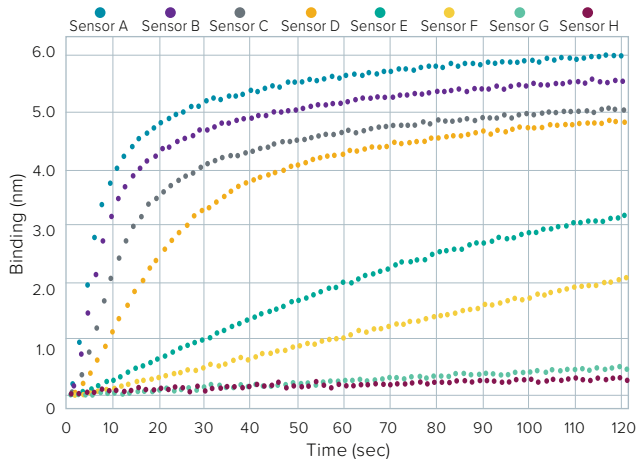


Figure 2: Real-time binding chart of Protein A standards.

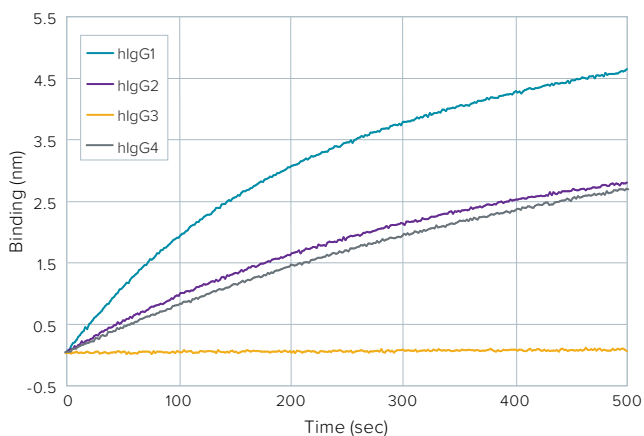


Figure 3: Human antibody isotypes binding to the Protein A biosensors.

Assay parameters

- Sample volume: 200 μ L (after dilution)
- Hydration solution volume: 200 μ L
- Data acquisition: 120 seconds/8 biosensors
- Flow rate: 200 mm/seconds
- Precision/accuracy: <10% CVs
- Biosensor hydration and sample plate equilibration: 10 minutes
- Standard curve fit: linear point-to-point

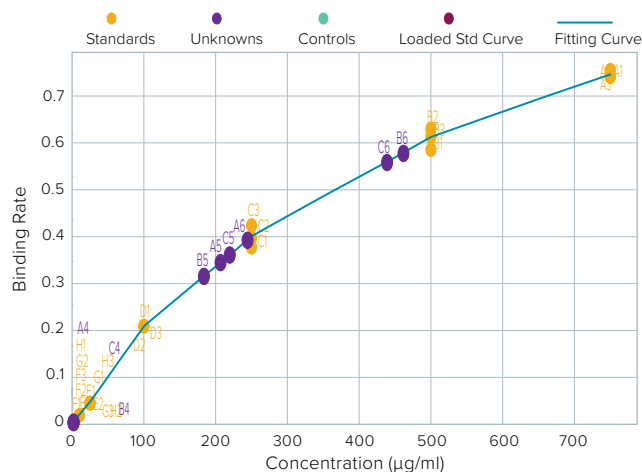


Figure 4: Standard curve with unknowns plotted on the curve.

Ordering information

Part no.	UOM	Description
18-5010	Tray	Tray of 96 biosensors coated with Protein A (recommended Sample Diluent sold separately)
18-5012	Pack	Five trays of 96 biosensors coated with Protein A (recommended Sample Diluent sold separately)
18-5013	Case	Twenty trays of 96 biosensors coated with Protein A (recommended Sample Diluent sold separately)

Note: Additional materials are required to run these assays.

Standards: A purified standard that is identical to the experimental samples is required.

Media for sensor hydration: The biosensor hydration solution must match the standard and experimental sample matrix (e.g., blank media or sample diluent).