SARTORIUS

Product Datasheet

Octet® Amine Reactive 2nd-Generation Biosensors

EDC/Sulfo-NHS Based Immobilization for Kinetic Characterization of Primary Amine Containing Biomolecules

Key Features

- Rapid immobilization of proteins and peptides
- Kinetic analysis of ligand-analyte pairs
- Rapid assay development
- Second-generation amine reactive detection surface (AR2G)

Overview

The Dip and Read Amine Reactive 2nd-Generation (AR2G) Biosensors enable kinetic characterization of macro-molecular interactions between purified proteins and target analytes. Immobilization of proteins is achieved through standard EDC-catalyzed amide bond formation to create a covalent bond between a reactive amine on the protein and the carboxy-terminated biosensor surface. Covalent immobilization fastens the protein to the biosensor surface for analysis of binding events and kinetic characterization. The AR2G Biosensor surface is amenable to a wide range of pH and salt conditions, providing robustness and flexibility during the development of regeneration conditions for higher throughput applications.

Second-Generation Performance

The AR2G Biosensor is a newly engineered biocompatible surface matrix that delivers enhanced performance for kinetic assays compared to the original Amine Reactive (AR) Biosensor for most proteins (Figure 1). AR2G routinely achieves greater signal intensity than the AR Biosensor during both the loading (immobilization of ligand) and association (detection of analyte) steps of an assay while delivering the same reliable kinetic values (Figure 2). Evaluation of four antibody proteins demonstrated an increase in the intensity of the loading signal by an average of 4-fold with one example extending to 7.5-fold (Figure 1). A screen of several buffer systems determined that the AR2G chemistry is highly compatible with acetate buffer, which is both commonly available and cost-effective. A decrease in non-specific binding events to the AR2G surface compared to AR has also been demonstrated.

When compared to the original AR Biosensors using a set of model system proteins, the AR2G Biosensor showed:

- Increased robustness towards pH during loading
- Increased robustness towards buffer type during loading
- Decreased non-specific binding
- Increased loading density
- Increased signal detection during analyte detection

Due to the vastly heterogeneous properties of proteins, performance of the AR2G Biosensor will vary for each binding pair. For more information on the use of the AR2G Biosensor, download Technical Note 28, Biotinylation of Protein for Immobilization onto Streptavidin Biosensors.

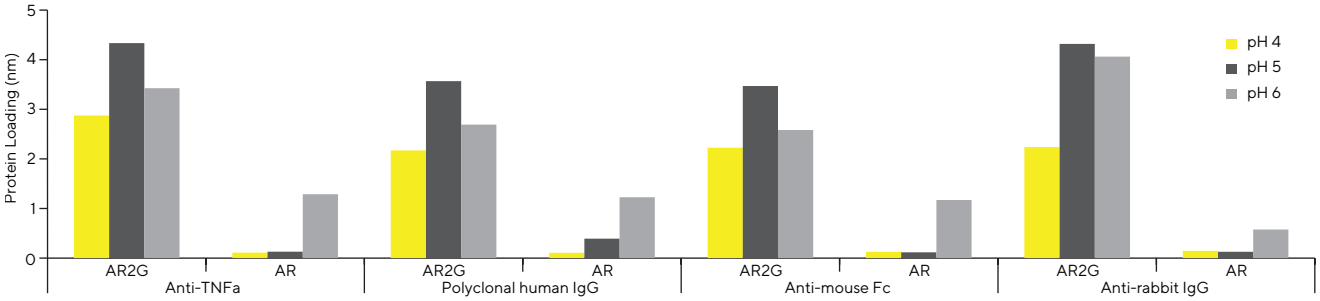
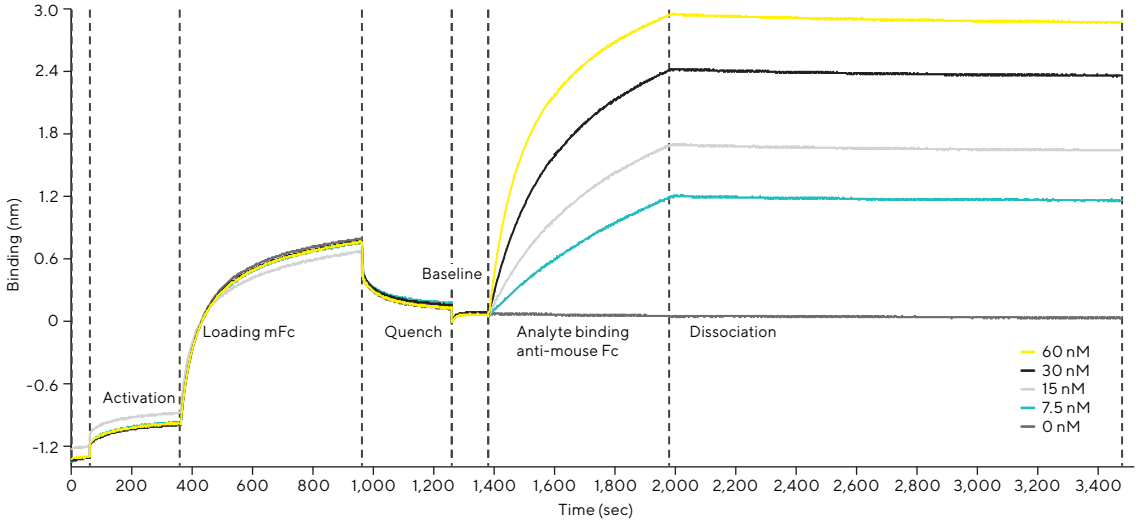
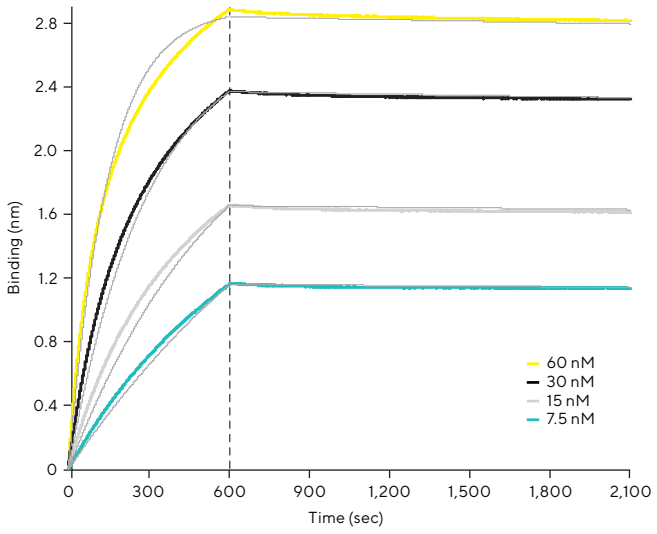


Figure 1: Performance comparison of AR2G and AR Biosensors. Four proteins (20 mg/mL)were immobilized on AR2G Biosensors using EDC/S-NHS (acetate buffer at pH 4, 5 and 6) and on AR Biosensors using EDC-NHS (MES buffer at pH 4, 5 and 6). The signal intensity achieved with the AR2G Biosensors during loading was significantly greater than that with the AR Biosensors for all proteins. The average increase was approximately four-fold with a maximum increase of 7.5-fold (anti-rabbit IgG). The enhanced performance of the AR2G Biosensor was robust towards pH with increased loading density overserved at all pH values.

A



B



C

K_D (M)	k_{on} (1/Ms)	k_{on} Eror	k_{dis} (1/s)	k_{dis} Error
9.19E-11	1.13E+05	2.74E+02	1.04E-05	4.58E-07

Figure 2: (A) Kinetic characterization between a mouse Fc domain and an anti-mouse antibody on the AR2G Biosensor. Mouse Fc was immobilized at 20 µg/mL in 10 mM acetate at pH 6 (optimal conditions determined based on data in Figure 1. Baseline, dissociation and analyte were run in 1X kinetics buffer. The association and dissociation kinetics of the analyte, anti-mouse Fc antibody, were performed at 7.5, 15, 30 and 60 nm.
(B) The kinetic dataset was globally fit using a 1:1 binding model.

Ordering Information

Part No.	UOM	Description
18-5092	Tray	One tray of 96 Octet® Amine Reactive Second-Generation (AR2G) Biosensors
18-5093	Pack	Five trays of 96 Octet® Amine Reactive Second-Generation (AR2G) Biosensors
18-5094	Case	Twenty trays of 96 Octet® Amine Reactive Second-Generation (AR2G) Biosensors

Note: Amine Reactive Second-Generation Reagent Kit (Part no. 18-5095) is also required.

Germany
Sartorius Lab Instruments GmbH & Co. KG
Otto-Brenner-Strasse 20
37079 Goettingen
Phone +49 551 308 0

USA
Sartorius Corporation
565 Johnson Avenue
Bohemia, NY 11716
Phone +1 888 OCTET 75
Or +1 650 322 1360

For further contacts, visit
www.sartorius.com/octet-support

DS-2012 Rev E