

## Source

Anti-Rituximab Antibodies (recommended for ADA assay) antibody is produced from a hybridoma resulting from fusion of SP2/0 myeloma and B-lymphocytes obtained from a mouse immunized with Rituximab.

## Isotype

Mouse IgG1/kappa

## Specificity

Recognizes Rituximab specifically, no cross reactivity with other humanized antibodies.

## Purity

>95% as determined by SDS-PAGE.

## Formulation

Lyophilized from 0.22  $\mu$ m filtered solution in Tris with Glycine, Arginine and NaCl, pH7.5 with trehalose as protectant.

Contact us for customized product form or formulation.

## Reconstitution

Please see Certificate of Analysis for specific instructions.

*For best performance, we strongly recommend you to follow the reconstitution protocol provided in the CoA.*

## Storage

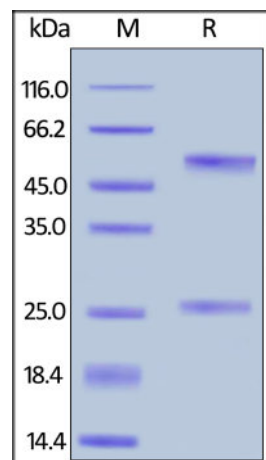
For long term storage, the product should be stored at lyophilized state at  $-20^{\circ}\text{C}$  or lower.

*Please avoid repeated freeze-thaw cycles.*

This product is stable after storage at:

- $4-8^{\circ}\text{C}$  for 12 months in lyophilized state;
- $-70^{\circ}\text{C}$  for 3 years under sterile conditions after reconstitution.

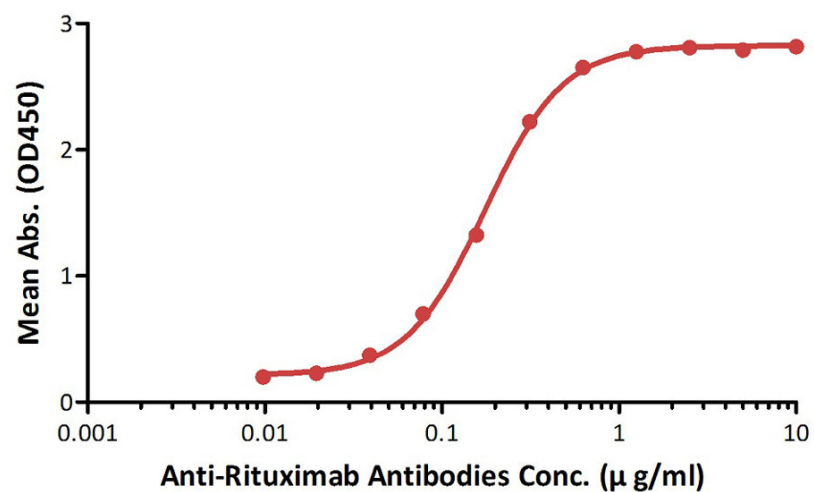
## SDS-PAGE



Anti-Rituximab Antibody (AY36) on SDS-PAGE under reducing (R) condition. The gel was stained overnight with Coomassie Blue. The purity of the protein is greater than 95%.

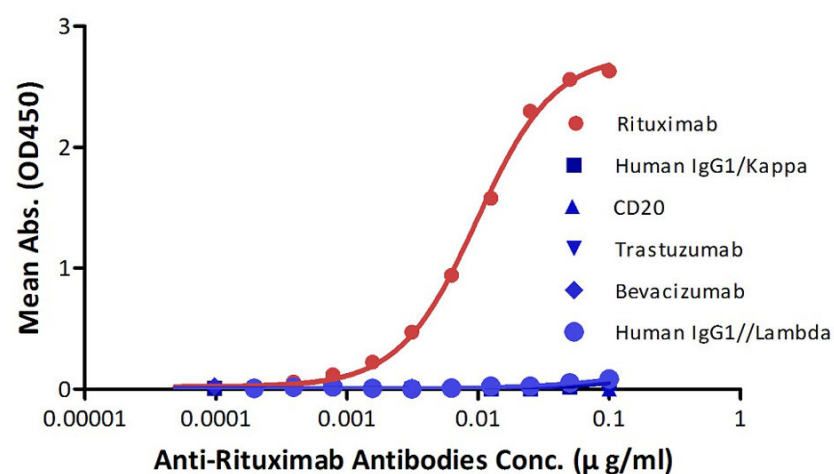
## Bioactivity-Elisa

**Anti-Rituximab Antibodies—Bridging ELISA**



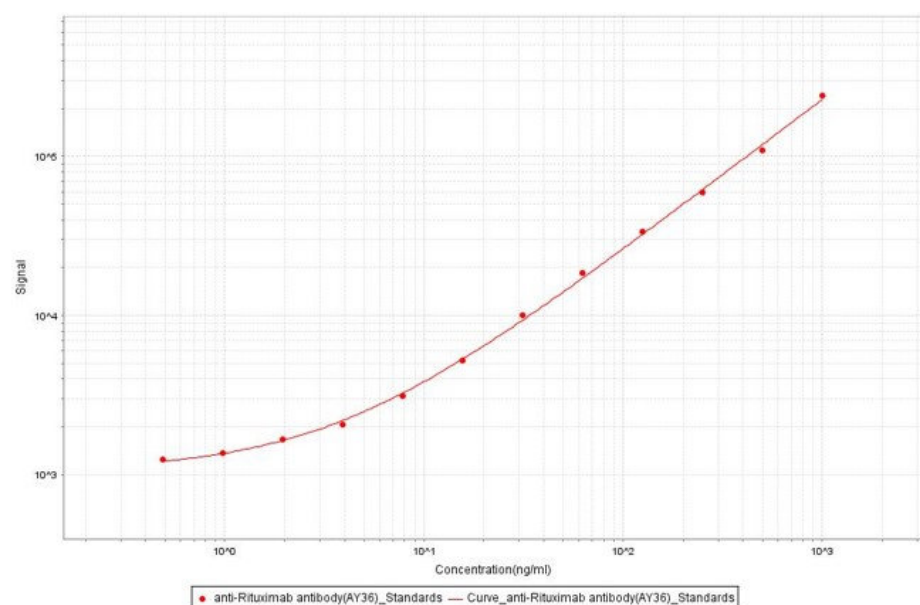
Anti-Rituximab Antibodies bridging ELISA for Anti-Drug Antibody (ADA) assay development. Immobilized rituximab at 1 µg/mL, add increasing concentrations of Anti-Rituximab Antibody (AY36) (Cat. No. RIB-Y36, 10% human serum) and then add biotinylated rituximab at 2 µg/mL. Detection was performed using HRP-conjugated streptavidin with a sensitivity of 9.7 ng/mL.

**Determination of Anti-Rituximab Antibodies Specificity**



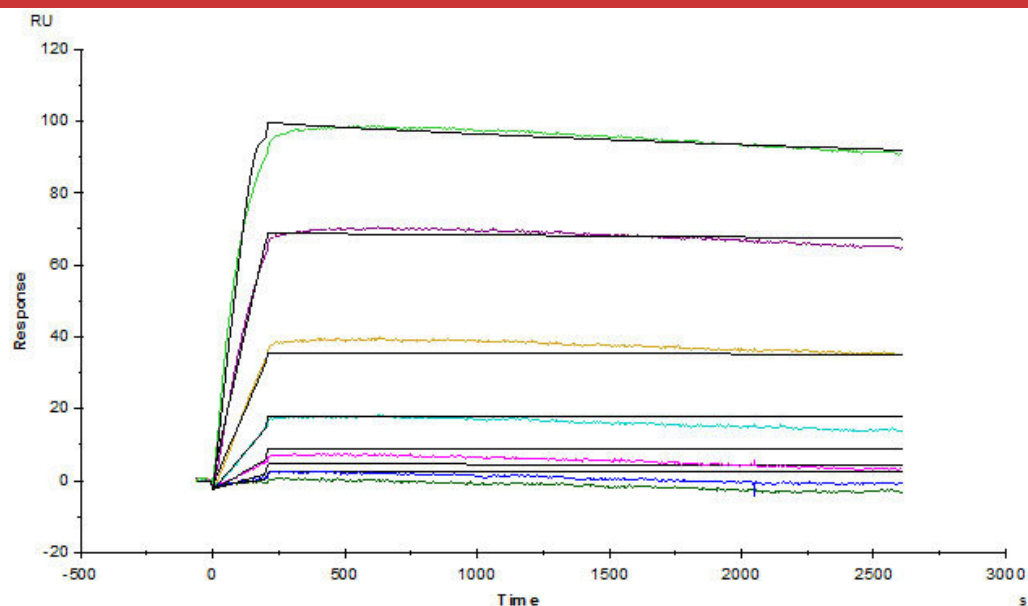
Demonstration of the specificity of Anti-Rituximab Antibody (AY36) (Cat. No. RIB-Y36) to the rituximab.

**Bioactivity-MSD**



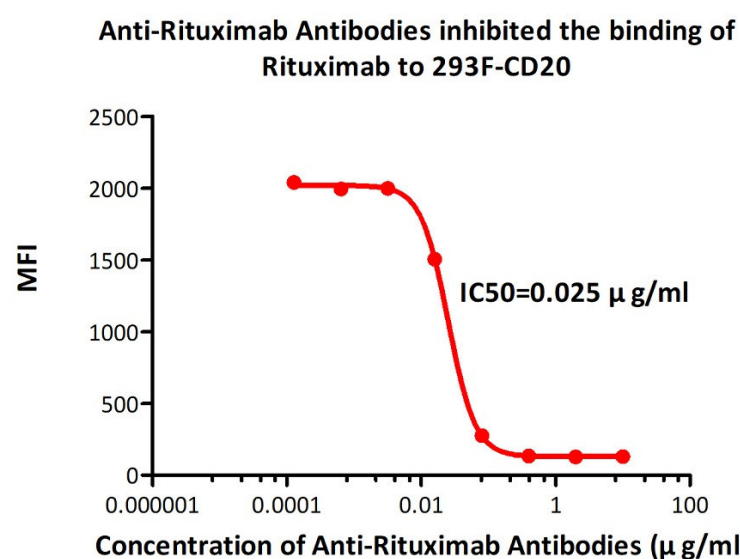
Anti-Rituximab Antibodies bridging MSD for Anti-Drug Antibody (ADA) assay development. Added the mix solution (biotinylated Rituximab at 5 µg/mL, SULFO-Rituximab at 5 µg/mL and increasing concentrations of Anti-Rituximab Antibody (AY36) (Cat. No. RIB-Y36, 100% human serum). Detection was performed using MSD Assay with a sensitivity of 0.97 ng/mL.

**Bioactivity-SPR**



Anti-Rituximab Antibody (AY36) (mouse IgG1, Cat. No. RIB-Y36) captured on CM5 chip via anti-mouse antibodies surface, can bind human rituximab with an affinity constant of 0.01 nM.

### Bioactivity-FACS



FACS analysis shows that the binding of rituximab to 293F overexpressing CD20 was inhibited by increasing concentration of Anti-Rituximab Antibody (AY36) (Cat. No. RIB-Y36). The concentration of rituximab used is 10 ng/ml. The IC50 is 0.025 µg/ml (Routinely tested).

### Background

Rituxan is a genetically engineered chimeric murine/human monoclonal antibody directed against the CD20 antigen found on the surface of normal and malignant B lymphocytes. The antibody is an IgG1 kappa immunoglobulin containing murine light- and heavy-chain variable region sequences and human constant region sequences. Rituximab is composed of two heavy chains of 451 amino acids and two light chains of 213 amino acids

### Clinical and Translational Updates

Please contact us via [TechSupport@acrobiosystems.com](mailto:TechSupport@acrobiosystems.com) if you have any question on this product.