### **Product Datasheet**

# Rat IgG F(ab)2 Isotype Control NBP1-97015-2mg

Unit Size: 2 mg

Store at 4C short term. For extended storage, add an equal volume of glycerol, aliquot and store at -20C or below. Avoid repeated freeze-thaw cycles.

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#### NBP1-97015-2mg

Rat IgG F(ab)2 Isotype Control

Product Information	
Unit Size	2 mg
Concentration	2 mg/ml
Storage	Store at 4C short term. For extended storage, add an equal volume of glycerol, aliquot and store at -20C or below. Avoid repeated freeze-thaw cycles.
Preservative	0.01% Sodium Azide
Isotype	IgG
Purity	Ion exchange chromatography
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Product Description	
Description	Native Rat IgG, Fab2.
	Source: Serum
Host	Rat
Species	Rat
Specificity/Sensitivity	Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-

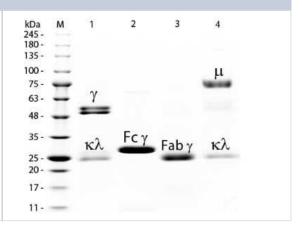
Product Application Details	
Applications	Western Blot, ELISA, Immunohistochemistry, SDS-Page
Recommended Dilutions	Western Blot, ELISA, Immunohistochemistry 1:10 - 1:500, SDS-Page 1:100-1:2000

against anti-Rat IgG F(c) or anti-Pepsin.

Rat IgG, anti-Rat IgG F(ab)2 and anti-Rat Serum. No reaction was observed

#### **Images**

SDS-Page: Rat IgG F(ab)2 Isotype Control [NBP1-97015] - SDS-PAGE of Rat IgG F(ab')2 Fragment (0002). Lane M: 3 uL Opal Prestained Marker. Lane 1: Reduced Rat IgG Whole Molecule. Lane 2: Reduced Rat IgG F(c) Fragment. Lane 3: Reduced Rat IgG F(ab')2 Fragment (0002). Lane 4: Reduced Rat IgM Whole Molecule. Load: 1 ug of IgG, F (c) and F(ab')2; 1.5 ug of IgM. Predicted/Observed size: IgG at 55 and 25 kDa; F(c) at 25 kDa; F(ab')2 at 25 kDa; IgM at 78 and 25 kDa. Observed F(c) Fragment migrates slightly higher.



#### **Procedures**

#### Block/Neutralize Protocol for IgG F(ab)2 Isotype Control (NBP1-97015)

Materials

Assay Buffer: 50 mM Tris, pH 8.0

Recombinant Human Pro TNF alpha Fusion Protein (rhPro-TNF-alpha ) (Catalog # 1012-PS)

Recombinant Human TACE/ADAM17 (rhTACE) (Catalog # 930-ADB)

Positive Control: Recombinant Human TNF-alpha (rhTNF-alpha) (Catalog # 210-TA)

Goat Anti-Human TNF alpha Biotinylated Antigen Affinity-purified Polyclonal Antibody (Catalog # BAF210)

SDS-PAGE followed by Western Blotting

- 1) Dilute rhPro-TNF-alpha to 0.2 mg/mL in Assay Buffer.
- 2) Dilute rhTACE to 0.2 mg/mL in Assay Buffer.
- 3) Prepare the following vials for a final volume of 20 uL.
  - a) 10 uL of rhPro-TNF-alpha at 0.2 mg/mL + 10 uL of rhTACE at 0.2 mg/mL
  - b) 10 uL of rhPro-TNF-alpha at 0.2 mg/mL + 10 uL of Assay Buffer (control-with incubation)
  - c) 10 uL of rhPro-TNF-alpha at 0.2 mg/mL + 10 uL of Assay Buffer (control-without incubation) Note: At this point the concentration of rhPro-TNF-alpha in the reaction tubes is 0.1 mg/mL.
- 4) Incubate vials at 37 degrees C overnight (except for the non-incubated control, store at less than or equal to 20 degrees C).
- 5) Stop the reactions by adding the reducing gel loading buffer for SDS-PAGE to all vials. Heat the samples at 100 degrees C for 3-5 minutes.
  - a) Dilute the reaction tubes (and control without incubation) to 50 ng/15 uL in reducing sample buffer.
- 6) Prepare a sample of Positive Control at 5 ng/15 uL in reducing sample buffer. Heat the sample at 100 degrees C for 3-5 minutes.
- 7) Load the samples on a 15% gel.
  - a) 50 ng/lane (15 uL) of rhPro-TNF-alpha of the incubated reactions (including the control with incubation).
  - b) 50 ng/lane (15 uL) of rhPro-TNF-alpha of the control (without incubation).
  - c) 5 ng/lane (15 uL) of Positive Control (Catalog # 210-TA).
- 8) Follow SDS-PAGE/Western Blotting procedures.
- a) Use the Biotinylated Anti-human TNF-alpha /TNFSF1A antibody at 0.1 ug/mL. (Catalog # AF-210-NA may be used in place of # BAF210.)
- 9) Visually determine processing of rhPro-TNF-alpha to the mature form by rhTACE by comparing the incubated reactions to the Positive Control.<br/>
  <br/>| [URL:https://www.novusbio.com/support/support-by-application.html] [Caption:Support By Application]





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#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Isotype Controls are guaranteed for 1 year from date of receipt.

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