# **Product Datasheet**

# Polyethylene Glycol Antibody Pair AP0002

Unit Size: 1 Set

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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# AP0002

Polyethylene Glycol Antibody Pair

<b>Product Information</b>	
Unit Size	1 Set
Concentration	Concentration of individual antibodies may be found on the vial label. If unlisted please contact technical services.
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Buffer	50 mM Tris-HCI, 10 mM reduced Glutathione, pH 8.0.

<b>Product Description</b>	
Description	Quality control test: Standard curve using LipoDox as an analyte. This matched antibody pair set binds to the repeating subunits of the polyethylene glycol polymer and can be employed to detect and quantify PEGylated compounds.  Capture antibody: mouse monoclonal anti-PEG IgM (100 ug). Stored in PBS buffer with 0.02% Sodium Azide.  Detection antibody: biotinylated mouse monoclonal anti-PEG IgG1 (100 ug). Stored in PBS buffer with 0.02% Sodium Azide and 50% Glycerol.
Notes	This product is produced by and distributed for Abnova, a company based in Taiwan.

Product Application Details		
Applications	Sandwich ELISA	
Recommended Dilutions	Sandwich ELISA 0.1 ng/ml - 30 ng/ml	

#### **Publications**

Moyers JS, Volk CB, Cao JXC et al. Internalization and localization of basal insulin peglispro in cells. Mol Cell Endocrinol 2017-05-30 [PMID: 28576743]

Chuang KH, Tzou SC, Cheng TC et al. Measurement of poly(ethylene glycol) by cell-based anti-poly(ethylene glycol) ELISA. Anal Chem. 2010-03-15 [PMID: 20178318]

Cheng TL, Cheng CM, Chen BM et al. Monoclonal antibody-based quantitation of poly(ethylene glycol)-derivatized proteins, liposomes, and nanoparticles. Bioconjug Chem. 2005-09-01 [PMID: 16173802]

Chuang KH, Wang HE, Cheng TC et al. Development of a Universal Anti-Polyethylene Glycol Reporter Gene for Noninvasive Imaging of PEGylated Probes. J Nucl Med. 2010-05-19 [PMID: 20484433]

Chuang KH, Wang HE, Chen FM et al. Endocytosis of PEGylated Agents Enhances Cancer Imaging and Anticancer Efficacy. Mol Cancer Ther. 2010-05-25 [PMID: 20501805]

Su YC, Chen BM, Chuang KH et al. Sensitive Quantification of PEGylated Compounds by Second-Generation Anti-Poly(ethylene glycol) Monoclonal Antibodies. Bioconjug Chem. 2010-06-10 [PMID: 20536171]

Sherman MR, Williams LD, Sobczyk MA et al. Role of the Methoxy Group in Immune Responses to mPEG-Protein Conjugates. Bioconjug Chem. 2012-03-07 [PMID: 22332808]



**Procedures** 



# Sandwich ELISA Assay Protocol for Anti-PEG Antibody Pair (AP0002)

Sandwich ELISA Assay Protocol for Anti-PEG Antibody Pair (AP0002):

Please centrifuge before opening the vials.

The test protocol is a guideline, user need to determine their optimal experimental condition for best performance.

# SECTION 1 Equipments & Reagents

#### 1.1 Anti-PEG Antibody pair

Polyethylene Glycol Matched Antibody Pair (Catalog #: AP0002). This matched antibody pair set binds to the repeating subunits of the polyethylene glycol polymer and can be employed to detect and quantify PEGylated compounds.

### 1.2 Secondary reagent

Streptavidin-HRP (Jackson ImmunoResearch, Catalog #: 016-030-084)

#### 1.3 Coating buffer (1 Liter)

5.3 g Na2CO3 + 4.2 g NaHCO3, pH=8.0 (adjust pH with 1N NaOH)

#### 1.4 1x PBS

0.14 M NaCl, 2.7 mM KCl, 1.5 mM KH2PO4, 8.1 mM Na2HPO4, pH 7.4

#### 1.5 Blocking solution

5% skim milk in 1x PBS

#### 1.6 Dilution buffer

2% skim milk in 1x PBS

#### 1.7 PBS-T

1x PBS containing 0.2% Tween-20

#### 1.8 ELISA plates

NUNC MaxiSorp(TM) High Protein-Binding Capacity ELISA plates (Catalog #: 44-2404)

#### 1.9 HRP substrate

50 mg/ml ABTS (Sigma #A-1888) in 100 mM phosphate-citrate buffer pH 4.0 (17.4 g K2HPO4, 21 g citric acid in 1 Liter

Q-H2O). Immediately before use, add 3 ul of 30% H2O2 per 10 ml ABTS substrate solution.

#### SECTION 2 - Assay Protocol

- 2.1 Dilute capture antibody to 5 ug/ml in coating buffer.
- 2.2 Add 50 ul diluted capture antibody per well and incubate at 37C for 4 h and then at 4C overnight.
- 2.3 Wash plates 3 times with 1x PBS.
- 2.4 Add 200 ull blocking solution per well for 2 hours at room temperature.
- 2.5 Dilute PEG-compound (analyte) in dilution buffer to suitable concentrations.
- 2.6 Wash wells 3 times with 1x PBS.
- 2.7 Add graded concentrations of PEG-compound (50 ul/well) and incubate 2 h at room temperature.
- 2.8 Wash with PBS-T 3 times and 1x PBS 2 times.
- 2.9 Add 50 ul/well detection antibody (5 ug/ml in dilution buffer) for 1 h at room temperature.
- 2.10 Wash wells with PBS-T 3 times and with 1x PBS 2 times.
- 2.11 Add 50 ul/well streptavidin-HRP (1 ug/ml in dilution buffer), and incubate for 1 h at room temperature.
- 2.12 Wash wells with PBS-T 6 times and with 1x PBS 2 times.
- 2.13 Add 100 ul/well freshly prepared ABTS substrate for 30 min in dark at room temperature.
- 2.14 Read absorbance of the wells at 405 nm.





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#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Antibody Pairs are guaranteed for 6 months from date of receipt.

For more information on our 100% guarantee, please visit www.novusbio.com/guarantee

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