abcam

Product datasheet

Anti-N6, N6-dimethyladenosine (m6,6A) antibody [EPR-19831-44] ab208198

Recombinant RabMAb

3 Images

Overview

Product name	Anti-N6, N6-dimethyladenosine (m6,6A) antibody [EPR- 19831-44]
Description	Rabbit monoclonal [EPR- 19831-44] to N6, N6-dimethyladenosine (m6,6A)
Host species	Rabbit
Specificity	Has been developed to discriminate between the modified base N6, N6-dimethyladenosine (m6,6A) and the unmodified counterpart Adenosine (A).
Tested applications	**Suitable for:** IP, ELISA
Species reactivity	**Reacts with:** Synthetic fragment
Immunogen	Chemical/ Small Molecule. This information is proprietary to Abcam and/or its suppliers.
Positive control	IP: 5' Biotin-mN.mN.mN.mN.mN.[m6,6A]*.mN.mN.mN.mN.mN 3' * - phosphorothioate protection. ELISA: Biotinylated m6,6A-modified oligonucleotides.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information see here. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified

Clonality	Monoclonal
Clone number	EPR- 19831-44
Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab208198 in the following tested applications.

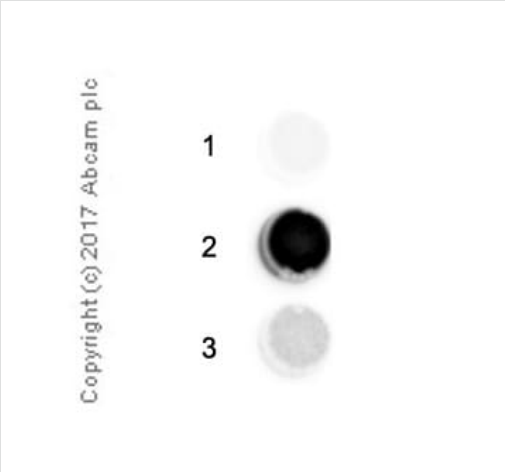
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP		Use at an assay dependent concentration. 0.2 µg of ab208198 was coated onto 50 µL Dynabeads® sheep-anti-rabbit IgG.
ELISA		Use a concentration of 0.005 - 4 µg/ml.

Target

Relevance N6,N6-dimethyladenosine(m6,6A) is an RNA modification found in rRNA in archaea, bacteria and eukaryotes.

Images



Immunoprecipitation - Anti-N6, N6-dimethyladenosine (m6,6A) antibody [EPR- 19831-44] (ab208198)

The IP was performed in a U-bottom non-adsorbing propylene 96-well plate.

ab208198 (0.2 µg) was coated into Dynabeads® sheep-anti-rabbit IgG (50 µl) for 1h at RT.

Unmodified/modified oligonucleotides (5 µM) were added to samples containing the antibody/bead complexes and incubated with agitation for 1 hour at RT.

After washing, Peroxidase-conjugated Streptavidin was incubated at 1/1000 dilution with agitation for 1 hour at RT.

ECL substrate was then added and the results read in a non-transparent 96-well plate with a digital detector and analyzed using ImageJ.

Lane 1: Buffer only.

Lane 2: Modified oligonucleotide (5 µM), 5' Biotin-mN.mN.mN.mN.mN.[m6,6A]*.mN.mN.mN.mN.mN 3'

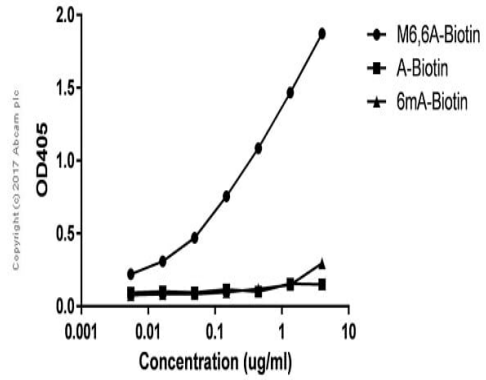
Lane 3: Unmodified oligonucleotide (5 µM), 5' Biotin-mN.mN.mN.mN.mN.[A]*.mN.mN.mN.mN.mN 3'

N - equimolar mixture of (A/U/G/C)

m - 2'O methyl protection
* - phosphorothioate protection

Blocking buffer and concentration: 5% NFDM/TBST

Dilution buffer and concentration: TBST/0.1% Triton X-100/1 mM EDTA



ELISA - Anti-N6, N6-dimethyladenosine (m6,6A) antibody [EPR- 19831-44] (ab208198)

Biotinylated m6,6A (modified), m6A (modified) and A (unmodified) oligonucleotides with the below sequence were coated onto wells of a 96 well plate.

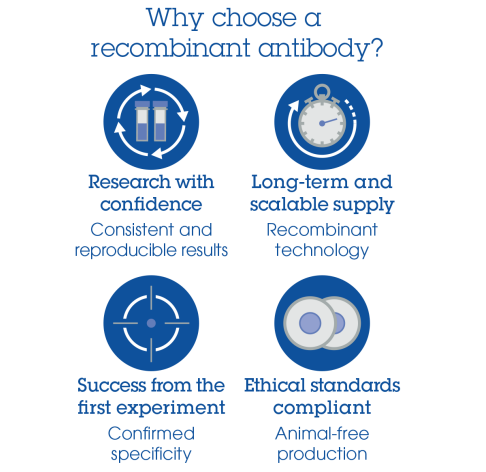
Modified oligonucleotide (5 µM), 5' Biotin-mN.mN.mN.mN.mN. [m6,6A]*.mN.mN.mN.mN.mN 3'

Modified oligonucleotide (5 µM), 5' Biotin-mN.mN.mN.mN.mN. [m6A]*.mN.mN.mN.mN.mN 3'

Unmodified oligonucleotide (5 µM), 5' Biotin-mN.mN.mN.mN.mN. [A]*.mN.mN.mN.mN.mN 3'

N - equimolar mixture of (A/U/G/C)
m - 2'O methyl protection
* - phosphorothioate protection

ELISA was performed on 1.0 µg/ml of antigen using ab208198 at a concentration range of 0.005-4.000 µg/ml, followed by Goat Anti-Rabbit IgG, (H+L), alkaline phosphatase conjugated secondary antibody at 1/2500 dilution.



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