

Anti-Myc tag Antibody

R1208-1



Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Species independent
Applications:	WB, IP, IF-Cell, FC

Description: Myc gene encodes for a transcription factor that is believed to regulate expression of 15% of all genes through binding on Enhancer Box sequences (E-boxes) and recruiting histone acetyltransferases (HATs). c-Myc is commonly activated in a variety of tumor cells and plays an important role in cellular proliferation, differentiation, apoptosis and cell cycle progression. This Myc-Tag antibody detects Myc-tagged fusion proteins.

Immunogen: Synthetic peptide within human Myc aa 410-420.

Positive control: PG-CM cell lysates, C-terminal Myc-tagged recombinant protein, N-terminal Myc-tagged recombinant protein.

Database links: SwissProt: P01106 Human

Recommended Dilutions:

WB	1:1,000-1:2,000
IP	2-5 µg/ml.
IF-Cell	1:200
FC	1:1,000

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4°C after thawing. Aliquot store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.

Purity: Immunogen affinity purified.

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Images

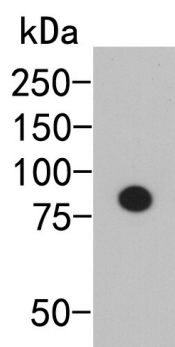


Fig1: Western blot analysis of Myc tag on PG-CM lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (R1208-1, 1/1,000) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.

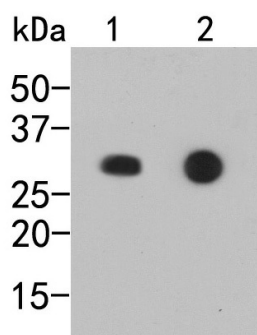


Fig2: Western blot analysis of Myc tag on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (R1208-1, 1/1,000) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.

Positive control:

Lane 1: C-terminal Myc-tagged recombinant protein

Lane 2: N-terminal Myc-tagged recombinant protein

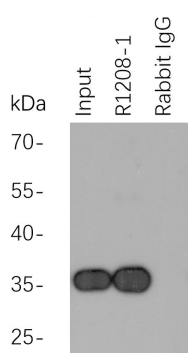


Fig3: Myc tag was immunoprecipitated in 2 μ g C terminal Myc Tag fusion protein lysate with R1208-1 at 2 μ g/20 μ l agarose. Western blot was performed from the immunoprecipitate using EM31105 at 1/1000 dilution. Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:20,000 dilution was used for 60 mins at room temperature.

Lane 1: Myc Tag fusion protein lysate (input).

Lane 2: R1208-1 IP in Myc Tag fusion protein lysate.

Lane 3: Rabbit IgG instead of R1208-1 in Myc Tag fusion protein lysate.

Blocking/Dilution buffer: 5% NFDN/TBST

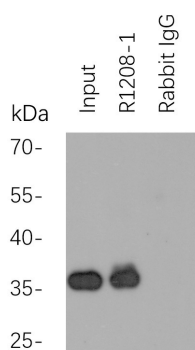


Fig4: Myc tag was immunoprecipitated in 2 μ g N terminal Myc Tag fusion protein lysate with R1208-1 at 2 μ g/20 μ l agarose. Western blot was performed from the immunoprecipitate using EM31105 at 1/1000 dilution. Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:20,000 dilution was used for 60 mins at room temperature.

Lane 1: Myc Tag fusion protein lysate (input).

Lane 2: R1208-1 IP in Myc Tag fusion protein lysate.

Lane 3: Rabbit IgG instead of R1208-1 in Myc Tag fusion protein lysate.

Blocking/Dilution buffer: 5% NFDN/TBST

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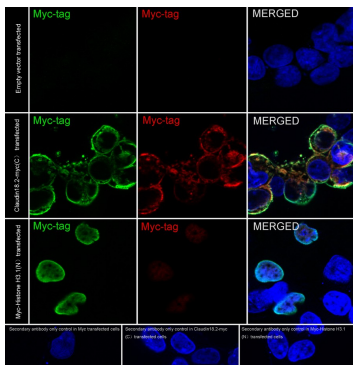
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Fig5: Immunocytochemistry analysis of 293T cells labeling Myc tag with Rabbit anti-Myc tag antibody (R1208-1) at 1/200 dilution.



293T cells, transfected with Myc-tagged empty control, Claudin18.2 (C-terminal) or Histone H3.1 (N-terminal) expression vector, respectively, were fixed in 4% paraformaldehyde for 10 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Myc tag antibody (R1208-1) at 1/200 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Myc Tag (HA601081, green) was stained at 1/1,000 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution.

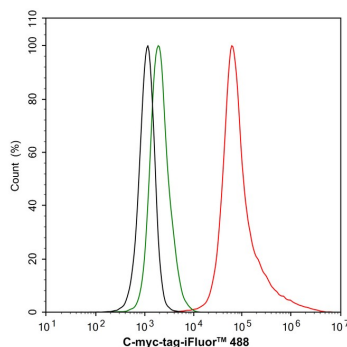


Fig6: Flow cytometric analysis of 293T cells transfected C-myc-tag labeling Myc tag.

Cells were fixed and permeabilized. Then stained with the primary antibody (R1208-1, 1µg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. "A quantitative atlas of mitotic phosphorylation." Dephoure N., Zhou C., Villen J., Beausoleil S.A., Bakalarski C.E., Elledge S.J., Gygi S.P. Proc. Natl. Acad. Sci. U.S.A. 105:10762-10767(2008)
2. "Transactivation of gene expression by Myc is inhibited by mutation at the phosphorylation sites Thr-58 and Ser-62." Gupta S., Seth A., Davis R.J. Proc. Natl. Acad. Sci. U.S.A. 90:3216-3220(1993)

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