Anti-Strep-Tag II Antibody [A5D11]

HA600038

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Species independent
Applications:	WB, IF-Cell, FC
Clone number:	A5D11

Description: The Strep-tag® system is a method which allows the purification and detection of proteins by affinity chromatography. The Strep-tag II is a synthetic peptide consisting of eight amino acids (Trp-Ser-His-Pro-Gln-Phe-Glu-Lys). This peptide sequence exhibits intrinsic affinity towards Strep-Tactin®, a specifically engineered streptavidin, and can be N- or C-terminally fused to recombinant proteins. By exploiting the highly specific interaction, Strep-tagged proteins can be isolated in one step from crude cell lysates. Because the Strep-tag elutes under gentle, physiological conditions it is especially suited for generation of functional proteins.

Immunogen: Synthetic peptide corresponding to Strep-tag II.

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:1,000
FC	1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.2% BSA, 50% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein G affinity purified.

Images

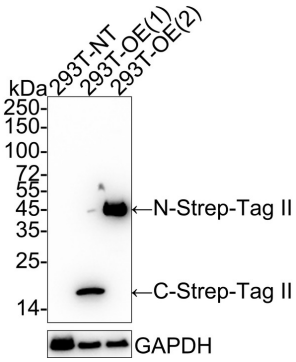

Fig1: Western blot analysis of Strep-Tag II on different lysates with Mouse anti-Strep-Tag II antibody (HA600038) at 1/1,000 dilution.

Lane 1: 293T-NT cell lysate
Lane 2: 293T transfected with Strep-Tag II-tagged Histone H3.1 (C-terminal) cell lysate
Lane 3: 293T transfected with Strep-Tag II-tagged ACAT2 (N-terminal) cell lysate

Lysates/proteins at 20 µg/Lane.

Exposure time: 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA600038) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

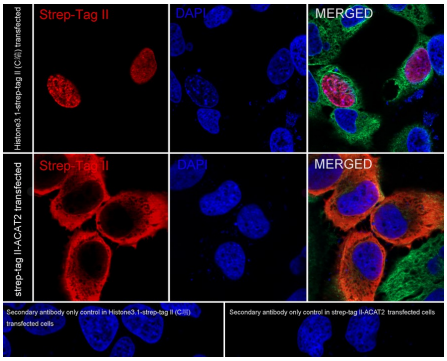

Fig2: Immunocytochemistry analysis of HeLa cells labeling Strep-Tag II with Mouse anti-Strep-Tag II antibody (HA600038) at 1/1,000 dilution.

HeLa cells, transfected with Strep-Tag II-tagged Histone H3.1 (C-terminal) or ACAT2 (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 Mouse anti-Strep-Tag II antibody (HA600038) at 1/1,000 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) 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.

beta Tubulin (ET1602-4, green) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) were used as the secondary antibody at 1/1,000 dilution.

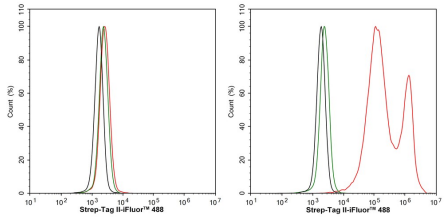

Fig3: Flow cytometric analysis of HeLa cells labeling Strep-Tag II.

HeLa cells, transfected with Strep-Tag II-tagged empty control or ACAT2 (N-terminal) expression vector, respectively, were fixed and permeabilized. Then stained with the primary antibody (HA600038, 1μg/mL) (red) compared with Mouse IgG1 Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Mouse IgG Secondary antibody (HA1125) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Arne Skerra. et. al. The Strep-tag system for one-step purification and high-affinity detection or capturing of proteins. Nat Protoc. 2007;2(6):1528-35.