Mouse anti DDDDK-Tag mAb

Catalog No.: AE005 281 Publications



Basic Information

Observed MW 37kDa/48kDa/57kDa/70kDa

Calculated MW

Category Tag antibody

Applications WB,IF/ICC,IP,FC (intra),ELISA

Cross-Reactivity Species independent

CloneNo number AMC0382

Background

FLAG-tag, or FLAG octapeptide, or FLAG epitope, is a polypeptide protein tag that can be added to a protein using recombinant DNA technology, having the sequence motif DYKDDDDK. It has been used for studying proteins in living cells and for protein purification by affinity chromatography. It has been used to separate recombinant, overexpressed protein from wild-type protein expressed by the host organism. It can also be used in the isolation of protein complexes with multiple subunits, because its mild purification procedure tends not to disrupt such complexes. It has been used to obtain proteins of sufficient purity and quality to carry out 3D structure determination by x-ray crystallography.A FLAG-tag can be used in many different assays that require recognition by an antibody. If there is no antibody against a given protein, adding a FLAG-tag to a protein allows the protein to be studied with an antibody against the FLAG sequence. Examples are cellular localization studies by immunofluorescence or detection by SDS PAGE protein electrophoresis and Western blotting.

Recommended Dilutions

WB	1:10000 - 1:80000
IF/ICC	1:200 - 1:800
IP	0.5µg-4µg antibody for 100µg-200µg extracts of whole cells
FC (intra)	5 μl per 10^6 cells in 100 μl volume
ELISA	Recommended starting concentration is 1 µg/mL. Please optimize the concentration based on your specific assay requirements.

Immunogen Information

Gene ID

Immunogen

Swiss Prot

Synthetic peptide. This information is considered to be commercially sensitive.

Synonyms DDDDK;DDDDK tag;DDDDK-tag

Product Information

Source Mouse **Isotype** IgG1,Kappa **Purification** Affinity purification

Storage

Store at -20°C. Avoid freeze / thaw cycles. Buffer: PBS with 0.09% Sodium azide,50% glycerol,pH7.3.

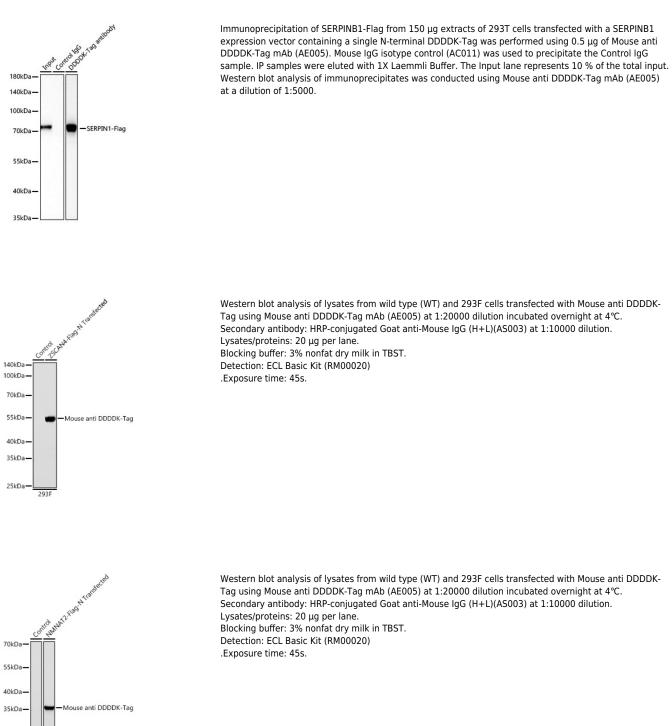
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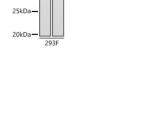
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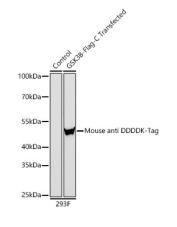
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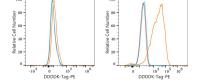
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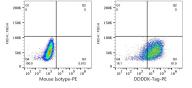


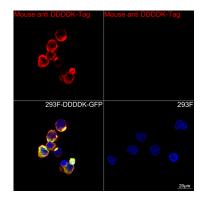




Western blot analysis of lysates from wild type (WT) and 293F cells transfected with Mouse anti DDDDK-Tag using Mouse anti DDDDK-Tag mAb (AE005) at 1:20000 dilution incubated overnight at 4°C. Secondary antibody: HRP-conjugated Goat anti-Mouse IgG (H+L)(AS003) at 1:10000 dilution. Lysates/proteins: 20 µg per lane. Blocking buffer: 3% nonfat dry milk in TBST. Detection: ECL Basic Kit (RM00020) .Exposure time: 45s.







Flow cytometry: 1X10^6 CHO cells (negative control,left) and CHO-Claudin18.2-Flag (Transfection,right) cells were intracellularlystained with Mouse anti DDDDK-Tag mAb (AE005,2 µg/mL,orange line) or Mouse isotype control (2 µg/mL,blue line), followed by PE Goat anti-Mouse pAb staining. Nonfluorescently stained cells were used as blank control (red line). Flow cytometry: 1X10^6 CHO-Claudin18.2-Flag (Transfection,right) cells were intracellularly-stained with Mouse isotype control (2 µg/mL,left) or Mouse anti DDDDK-Tag mAb (AE005,2 µg/mL,right), followed by PE Goat anti-Mouse pAb staining.

Confocal imaging of 293F cells transfected with DDDDK-Tag using Mouse anti DDDDK-Tag mAb (AE005, dilution 1:200) followed by a further incubation with Cy3 Goat Anti-Mouse IgG (H+L) (AS008, dilution 1:500) (Red). DAPI was used for nuclear staining (Blue). Objective: 100x.