AE005

Leader in Biomolecular Solutions for Life Science

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Mouse anti DDDDK-Tag mAb

Catalog No.: AE005 **281 Publications**

Basic Information

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

Calculated MW

Category
Monoclonal Antibody

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

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 **Swiss Prot**

Immunogen
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℃. Avoid freeze / thaw cycles.
Buffer: PBS with 0.09% Sodium azide,50% glycerol,pH7.3.

Contact

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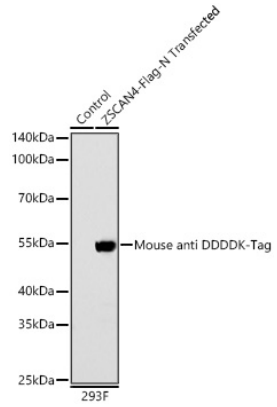
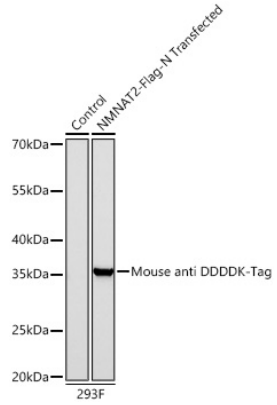
Validation Data



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℃.
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.



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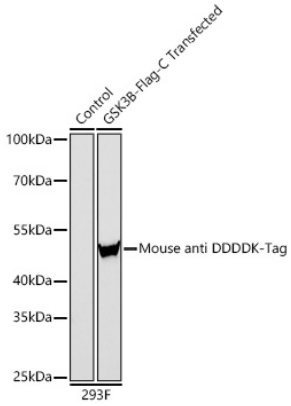

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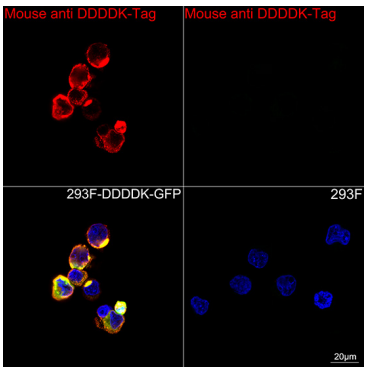
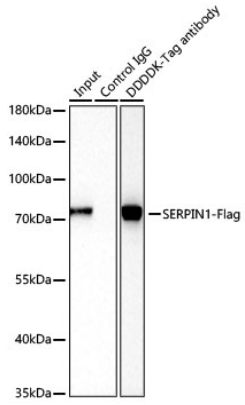
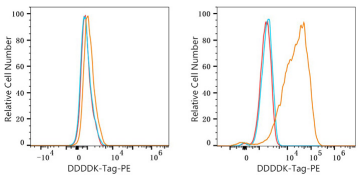


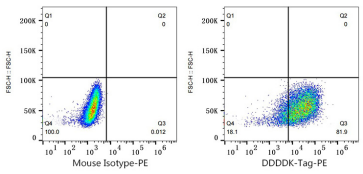
Confocal imaging of 293F cells transfected with DDDDK-Tag using Mouse anti DDDDK-Tag mAb (AE005, dilution 1:200) followed by a further incubation with Cy3 Goat Anti-Mouse IgG (H+L) (AS008, dilution 1:500) (Red). DAPI was used for nuclear staining (Blue). Objective: 100x.



Immunoprecipitation of SERPINB1-Flag from 150 µg extracts of 293T cells transfected with a SERPINB1 expression vector containing a single N-terminal DDDDK-Tag was performed using 0.5 µg of Mouse anti DDDDK-Tag mAb (AE005). Mouse IgG isotype control (AC011) was used to precipitate the Control IgG sample. IP samples were eluted with 1X Laemmli Buffer. The Input lane represents 10 % of the total input. Western blot analysis of immunoprecipitates was conducted using Mouse anti DDDDK-Tag mAb (AE005) at a dilution of 1:5000.



Flow cytometry: 1X10^6 CHO cells (negative control,left) and CHO-Claudin18.2-Flag (Transfection,right) cells were intracellularly-stained 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. Non-



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.

fluorescently stained cells were used as blank control (red line).