

Rev05 DATASHEET

Update: Nov,14,2022

THE™ DYKDDDDK Tag Antibody, mAb, Mouse

Cat. No.: A00187

Overview

Specificity	THE™ DYKDDDDK Tag Antibody, mAb, Mouse recognizes DYKDDDDK tags localized at the C-terminal, N-terminal and internal region of DYKDDDDK tagged fusion proteins.	
Host Species	Mouse	
Immunogen	A synthetic peptide (DYKDDDDK) coupled to KLH	
Conjugate	Unconjugated	

Applications

Working concentrations for specific applications should be determined by the investigator. The appropriate concentrations may be affected by secondary antibody affinity, antigen concentration, the sensitivity of the method of detection, temperature, the length of the incubations, and other factors. The suitability of this antibody for applications other than those listed below has not been determined. The following concentration ranges are recommended starting points for this product.

Application	Recommended Usage
ELISA	0.05-0.2 μg/ml
Western Blot	0.1-1.0 μg/ml
Immunoprecipitation (IP)	1 μg/ml
Immunocytochemistry/Immunofluorescence (ICC/IF)	1 μg/ml
Flow Cytometry	1 μg/ml

Properties

Form	Lyophilized
Storage Buffer	Lyophilized with PBS, pH 7.4, containing 0.02% sodium azide
Reconstitution	Reconstitute the lyophilized powder with deionized water (or equivalent) to a final concentration of 0.5 mg/mL.

GenScript USA, Inc.



The lyophilized product remains stable for up to 1 year at -20°C from the date of receipt The reconstituted antibody can be stored for 2-3 weeks at 2-8°C. For long term storage, aliquot and store at -20°C or below. Avoid repeated freezing and thawing cycles.	
Protein A affinity column	
Mouse IgG2b,κ	
Monoclonal	
5A8E5	
GenScript can offer this product per customer's request including product size, buffer components, etc.	

Examples

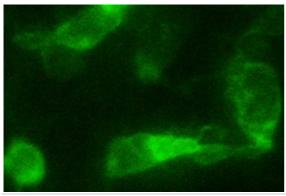


Figure 1. The affinity of THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) for a C-terminal DYKDDDDK tag was analyzed in an immunocytochemistry/immunofluorescence analysis of C-terminal DYKDDDDK tag protein-transfected HEK293 cells. The primary antibody was THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) used at 1 μg/mL concentration. The secondary antibody was Fluorescein Conjugated Anti-mouse IgG used at 2 μg/mL concentration.

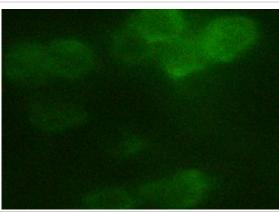


Figure 2. The affinity of THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) for an N-terminal DYKDDDDK tag was analyzed in an

immunocytochemistry/immunofluorescence analysis of N-terminal DYKDDDDK tag protein-transfected HEK293 cells. The primary antibody was THETM DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) used at 1 μ g/mL concentration. The secondary antibody was Fluorescein Conjugated Anti-mouse IgG used at 2 μ g/mL concentration.

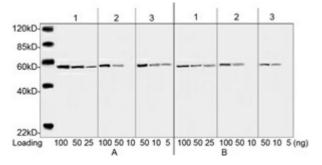


Figure 3. Comparison of DYKDDDDK-tag detection by THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187; panel A) vs. Mouse Anti- DYKDDDDK Tag mAb (Company S, clone M2; panel B) by Western blot.

In both panels A and B:

Lane 1 100 ng, 50 ng, 25 ng N-terminal DYKDDDDK-tagged fusion protein (Predicted Size: 52 kDa)

Lane 2 100 ng, 50 ng, 25 ng Internal DYKDDDDK-tagged fusion protein (Predicted Size: 55 kDa)

Lane 3 50 ng, 10 ng, 5 ng C-terminal DYKDDDDK-tagged fusion



protein (Predicted Size: 55 kDa)

The primary antibody in panel A was THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) used at 1 µg/mL concentration. The primary antibody in panel B was Mouse Anti- DYKDDDDK Tag mAb (Company S, clone M2) used at 1 µg/mL concentration. In both cases, the secondary antibody was IRDye800 Conjugated Goat Anti-Mouse IgG.

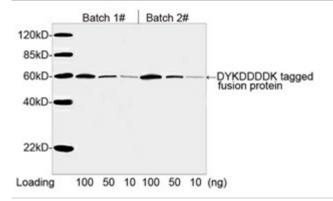


Figure 4. Consistency analysis of Batch 1# and 2# of THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187, 1 μg/mL) by Western blot, showing that signal remains consistent from Lot to Lot. The assay was performed with DYKDDDDK-tagged fusion protein.

The primary antibody were 2 lots of THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) used at 1 µg/mL concentration. The secondary antibody was IRDye™ 800 Conjugated Goat Anti-Mouse IgG.

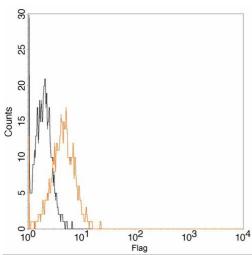


Figure 5. The detection of DYKDDDDK tag by THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) was analyzed by flow cytometric analysis of non-transfected or DYKDDDDK gene-transfected CHO cells (black and orange, respectively) using THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187). The signal was developed with FITC conjugated Goat Anti-Mouse IgG.

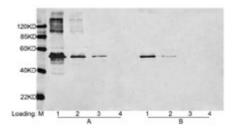


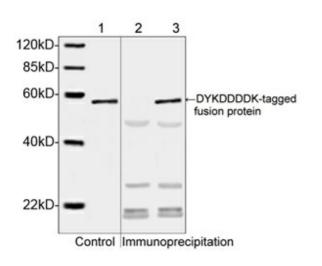
Figure 6. Comparison of DYKDDDDK tag detection of Multiple Tag Protein by THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187; panel A) vs. Mouse Anti- DYKDDDDK Tag mAb (Company S, clone M2; panel B) by Western blot. In both panels A and B:

Lanes 1-3 400 ng, 80 ng,16 ng Multiple Tag Protein (Purified) (GenScript, M0101)

Lane 4. 10 µL HEK293 cell lysate (negative control)

The primary antibody in panel A was THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) used at 1 µg/mL concentration. The primary antibody in panel B was Mouse Anti- DYKDDDDK Tag mAb (Company S, clone M2) used at 1





µg/mL concentration. In both cases, the secondary antibody was IRDye800 Conjugated Goat Anti-Mouse IgG.

Figure 7. The affinity of THE™ DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187) for DYKDDDDK tag in immunoprecipitates from DYKDDDDK-tagged proteintransfected *E.coli* lysates was analyzed using Western blotting. Lane 1: *E.coli* lysate containing DYKDDDDK-tagged fusion protein

Lane 2: Immunoprecipitates of untransformed *E.coli* lysate incubated with THE™ DYKDDDDK Tag Antibody, mAb, Mouse

(GenScript, A00187, 2 μ g/mL) and Protein A. Lane 3: Immunoprecipitates of *E.coli* lysate containing DYKDDDDK-tagged fusion protein incubated with DYKDDDDK Tag Antibody, mAb, Mouse (GenScript, A00187, 2 μ g/mL) and Protein A.

Background

Target Background: The DYKDDDDK peptide is a small epitope tag which is widely used in expression vectors. The tag does not appear to interfere with the bioactivity or bio-distribution of the tagged recombinant proteins. THE™ DYKDDDDK Tag Antibody, mAb, Mouse is a high-affinity monoclonal antibody that can be used to detect DYKDDDDK-tagged proteins

Synonyms: Mouse anti DYKDDDDK-tag mAb; Mouse anti flag-tag mAb

For laboratory research use only. Direct human use, including taking orally and injection and clinical use are forbidden.