

Datasheet for ABIN1112984
anti-DYKDDDDK Tag antibody



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1 Validation

Overview

Quantity:	100 µg
Target:	DYKDDDDK Tag
Host:	Mouse
Conjugate:	This DYKDDDDK Tag antibody is un-conjugated
Application:	Western Blotting (WB), Immunoprecipitation (IP), Immunofluorescence (IF)

Product Details

Immunogen:	A synthetic peptide DYKDDDDK conjugated to KLH.
Sequence:	DYKDDDDK
Isotype:	IgG1

Target Details

Target:	DYKDDDDK Tag
Alternative Name:	Flag Tag (DYKDDDDK Tag Products)
Target Type:	Tag

Background: DYKDDDDK-Tag, or DYKDDDDK octapeptide, is a polypeptide protein tag that can be added to a protein using recombinant DNA technology. It can be used for affinity chromatography, then used to separate recombinant, overexpressed protein from wild-type protein expressed by the host organism. The peptide sequence of the DYKDDDDK-Tag is: N-DYKDDDDK-C (1012 Da). It can be used in conjunction with other affinity tags, for example a polyhistidine tag (His-tag), HA-tag or myc-tag. It can be fused to the C-terminus or the N-terminus of a protein.

Application Details

Application Notes: WB (1:1000-10000), IP (1:100-500), IF (1:200-1000). Other applications have not been tested.
The optimal dilutions should be determined by end user.

Restrictions: For Research Use only

Handling

Format: Liquid

Buffer: PBS (pH7.4), 50% Glycerol and 0.02% Sodium Azide.

Preservative: Sodium azide, Thimerosal (Merthiolate)

Storage: 4 °C

Storage Comment: Store at 2-8 °C for 1 month. Or aliquot and store at -20 °C or below for long term. Avoid repeated freeze and thaw cycles.



Successfully validated (Immunofluorescence (IF))

by [Reveal Biosciences](#)

Report Number: 028889

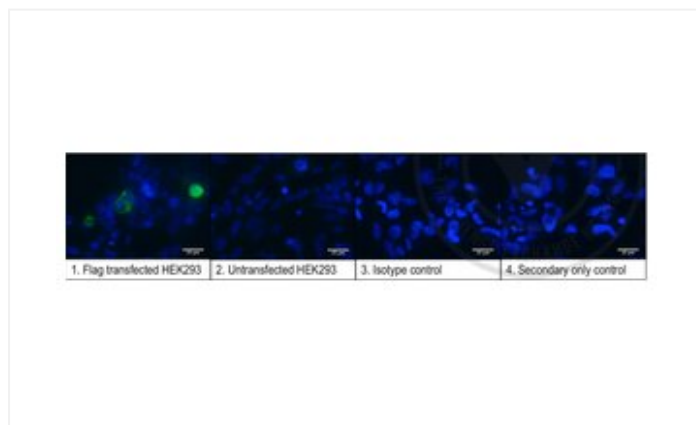
Date: Nov 07 2013

Lot Number:	L21/2013G
Method validated:	Immunofluorescence (IF)
Positive Control:	HEK293 cells transfected with DYKDDDDK Tag
Negative Control:	Untransfected HEK293 cells
Notes:	Signal was detected in positive control samples but not in negative control samples.
Primary Antibody:	- Antibody: DYKDDDDK Tag antibody - Catalog number: ABIN1112984 - Supplier: Biospes - Supplier catalog number: BTL1005 - Lot number: L21/2013G
Secondary Antibody:	- Antibody: Goat anti-mouse IgG (H+L) IF488 conjugated antibody - Supplier: Biospes - Supplier catalog number: BSA1041 - Lot number: S6/2013S
Controls:	<ul style="list-style-type: none">• Positive control: HEK293 cells from ATCC transfected with a DYKDDDDK tagged protein (specimen known to contain the target protein).• Negative control: Untransfected HEK293 cells from ATCC (specimen known to not contain the target protein).• Isotype control: HEK293 cells from ATCC transfected with a DYKDDDDK tagged protein treated with isotype mouse IgG control instead of the primary antibody. Any staining observed is due to non-specific binding of secondary antibody.• Secondary antibody only control: HEK293 cells from ATCC transfected with a DYKDDDDK tagged protein treated with Goat anti-Mouse IF488 secondary antibody only. Any staining observed is due to non-specific binding of secondary antibody.
Protocol:	<ul style="list-style-type: none">• HEK293 cells were grown directly on coverslips and transfected with a DYKDDDDK tagged protein.• Cells were fixed with 4% paraformaldehyde in PBS for 15 min at room temperature (RT).• Fixed cells were rinsed three times in PBS for 5 min each at RT.

- Cells were blocked in 1 X PBS / 5% normal goat serum / 0.3% Triton X-100 for 20 min at RT.
- Cells were incubated with primary antibody diluted 1:100 in Universal Antibody Dilution Buffer (Electron
- Microscopy Sciences, 25885-05) overnight at 4°C.
- Cells were rinsed three times in PBS for 5 min each at RT.
- Cells were incubated with secondary antibody diluted 1:150 in Universal Antibody Dilution Buffer
- (Electron Microscopy Sciences, 25885-05) for 60 min at RT in dark.
- Cells were rinsed three times in PBS for 5 min each at RT.
- Coverslips were mounted on slides with Fluoro-Gel II with DAPI (Electron Microscopy Sciences Cat. No: 17985-50).
- Stained cells were imaged with an EIDAQ 100 fluorescent microscope.

Experimental Notes: None

Image for Validation report #028889



Validation image no. 1 for anti-DYKDDDDK Tag antibody (ABIN1112984)

Figure 1: : Immunofluorescence analysis of cultured cells using DYKDDDDK Tag antibody (Catalog number ABIN1112984, Lot number L21/2013G). Flag is present in the positive control (HEK293 cells transfected with DYKDDDDK Tag panel 1, transfected cells are indicated by green fluorescent staining) and absent from the negative control (untransfected HEK293 cells panel 2). No staining was observed in the isotype (panel 3) and secondary only (panel 4) controls. DAPI was used as a nuclear counterstain (indicated by blue fluorescent staining). Scale bar = 25 µm.