Datasheet for ABIN2669973

anti-IdU antibody

1 Validation

4 Images

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Overview

Quantity:	100 µL
Target:	IdU
Reactivity:	Chemical
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This IdU antibody is un-conjugated
Application:	Immunohistochemistry (IHC), Immunofluorescence (IF)

Product Details

Immunogen:	Iododeoxyuridine coupled to keyhole limpet hemocyanin.
Clone:	OTI2B10
Isotype:	IgG2b
Purification:	Purified from mouse ascites fluids by affinity chromatography

Target Details

Target:	IdU
Alternative Name:	IdU (IdU Products)

Application Details

Application Notes:	IHC 1:150, IF 1:150,

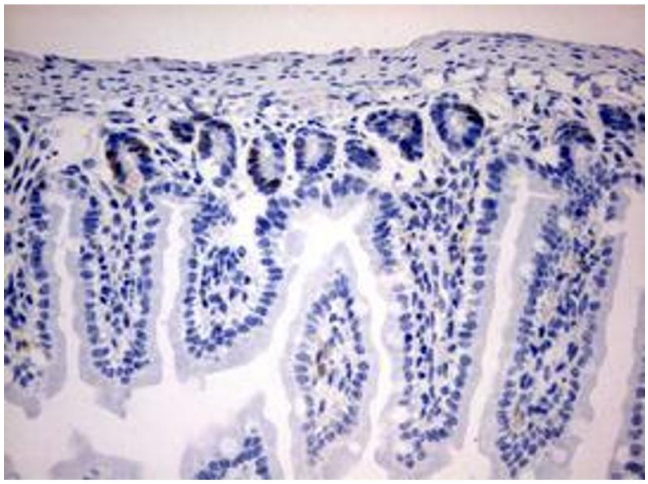
Application Details

Restrictions:	For Research Use only

Handling

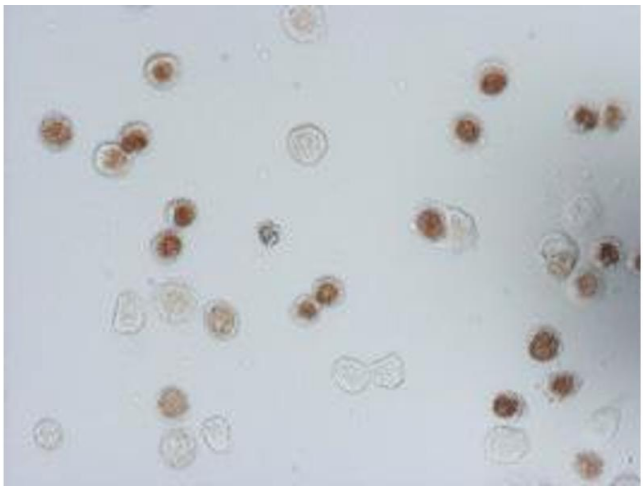
Format:	Liquid
Concentration:	4.54 mg/mL

Images



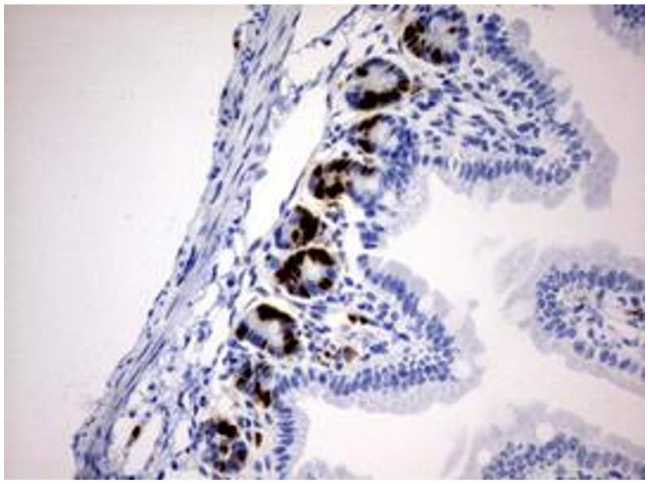
Immunohistochemistry

Image 1.



Immunofluorescence

Image 2.



Immunohistochemistry

Image 3.

Please check the product details page for more images. Overall 4 images are available for ABIN2669973.

Validation report #103750 for Immunohistochemistry (IHC)

Successfully validated (Immunohistochemistry (IHC))

by Prof. Merighi, Laboratory of Neurobiology, Department of Veterinary Sciences, University of Turin

Report Number: 103750

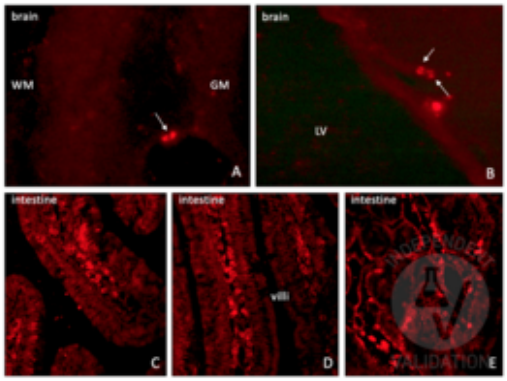
Date: Apr 19 2019

Target:	IdU
Lot Number:	W001
Method validated:	Immunohistochemistry (IHC)
Positive Control:	Adult (24 months) mouse brain and intestine (duodenum)
Negative Control:	We incubated slices overnight with the blocking solution only and then processed them with the secondary antibody.
Notes:	Passed. ABIN2669973 works in IHC-P with a microwave antigen retrieval pretreatment and concentration of 1:50.
Primary Antibody:	ABIN2669973
Secondary Antibody:	goat anti-mouse AF594-conjugated (Invitrogen by Thermo Fisher Scientific, A11032, lot 819561)
Protocol:	• Inject mouse intraperitoneally at the dose of 0.057mg/g body weight with IdU (Sigma, I7125) dissolved in sterile distilled water at final concentration of 100mg/ml, pH7.5. • Perfuse mouse with 4% paraformaldehyde in 0.1M phosphate buffer (PB) pH7.4 and post-fix samples in the same fixative for an additional 2h at RT. • Wash, dehydrate, and embed samples in paraffin wax. • Following several washes in PBS, cut intestines and brain with a microtome (6µm-thick sections) and mount sections on glass slides. • After paraffin removal, process sections by microwave antigen retrieval for 10min (95-100°C) in 10mM sodium citrate buffer pH6.0. • Let sections cool for 20min. • Wash sections 2x 5min with distilled water. • Wash sections 5min with PBS. • Incubate sections in PBS containing 1% albumin from chicken egg white (Sigma, A5378) and 0.3% Triton-X-100 (BioRad, 161-0407, lot 00583) for 1h at RT to block non-specific binding sites. • Incubate sections with primary antibody mouse anti-IdU (antibodies-online, ABIN2669973, lot W001) diluted 1:50 in PBS-BSA-PLL ON at 4°C. • Wash sections 3x 5min with 0.01M PBS.

- Incubate sections with secondary goat anti-mouse AF594-conjugated (Invitrogen by Thermo Fisher Scientific, A11032, lot 819561) diluted 1:500 in 0.1M PBS for 1h at RT.
- Wash sections 3x 5min with 0.01M PBS.
- Mount sections in Fluoroshield (Sigma, F6182, lot MKCB0153V).
- Acquire images with Leica DM 6000B fluorescence microscope equipped with the manufacturer's FITC filter set and digital camera at magnifications of 20x and 40x. Parameters for image acquisition (Exposure 2.2s, Gain 2.3x, Saturation 1.5, Gamma 1.10) were maintained unchanged for all images.

Experimental Notes:	For the incubation with the primary antibody ABIN2669973 and 1:20, 1:70, and 1:100 dilutions were also tested with 0.01M PBS, 5% Normal Goat Serum (NGS Sigma, G9023, lot SLBV1396) and 0.1% Triton-X -100 as the blocking solution, but 1:50 and PBS containing 1% albumin from chicken egg white and 0.3% Triton-X gave the best results.

Image for Validation report #103750



Validation image no. 1 for anti-Iododeoxyuridine (IdU) antibody (ABIN2669973)

Staining of mouse brain and intestine sections using ABIN2669973. A-B: A bright staining in the nuclei of individual cells scattered across the brain is present, especially in the sub-cortical areas at the border between the gray and white matters (A) and ventricular wall (B). Abbreviations: GM = gray matter; LV = lateral ventricle; WM = white matter. C-E: positive control staining in the intestine (duodenum). IdU+ proliferating cells are clearly detected in the germinal layer of the intestinal epithelium, at the basis of the duodenal villi.