abcam

Product datasheet

Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free ab271990

KO VALIDATED Recombinant RabMAb

★★★★★ 1 Abreviews 9 Images

Overview

Product name	Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free
Description	Rabbit monoclonal [EPR20374] to Indoleamine 2, 3-dioxygenase - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), IP, ICC/IF, IHC-P, WB
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant full length protein. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Wild-type A549 Treated IFN gamma, Human ovary cancer, placenta and tonsil lysates; SK-OV-3 whole cell lysate; HeLa whole cell lysate treated with 50ng/ml Interferon-gamma (IFN-gamma) for 16 hours. IHC-P: Human spleen, tonsil, placenta and endometrium cancer tissues. ICC/IF: HeLa cells treated with IFN-gamma (50 ng/ml) for 16 hours. Flow Cyt (intra): HeLa cells treated with IFN-gamma (50 ng/ml) for 16 hours. IP: HeLa whole cell lysate treated with 50ng/ml IFN-gamma for 16h.
General notes	ab271990 is the carrier-free version of **ab211017**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production

For more information **see here**.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR20374
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab271990 in the following tested applications.

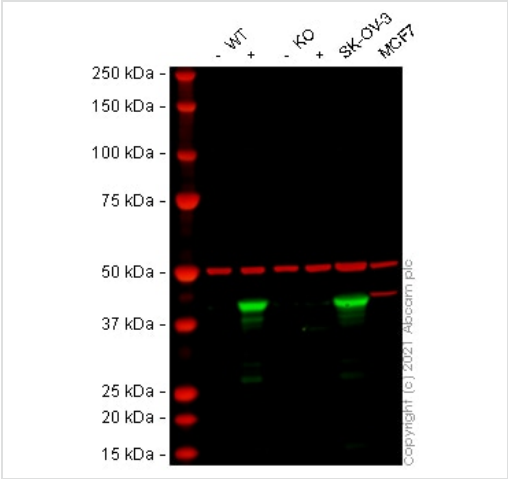
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
IHC-P	★★★★★ (1)	Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 45 kDa.

Target

Function	Catalyzes the cleavage of the pyrrol ring of tryptophan and incorporates both atoms of a molecule of oxygen.
Pathway	Amino-acid degradation; L-tryptophan degradation via kynurenine pathway; L-kynurenine from L-tryptophan: step 1/2.
Sequence similarities	Belongs to the indoleamine 2,3-dioxygenase family.

Images



Western blot - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

All lanes : Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] (**ab211017**) at 1/1000 dilution

Lane 1 : Wild-type A549 Vehicle Control IFN gamma (0 ng/ml, 48 h) cell lysate
Lane 2 : Wild-type A549 Treated IFN gamma (25 ng/ml, 48 h) cell lysate
Lane 3 : IDO1 knockout A549 Vehicle Control IFN gamma (0 ng/ml, 48 h) cell lysate
Lane 4 : IDO1 knockout A549 Treated IFN gamma (25 ng/ml, 48 h) cell lysate
Lane 5 : SK-OV-3 cell lysate
Lane 6 : MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

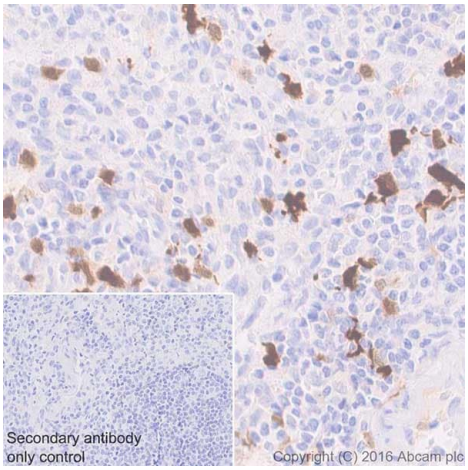
Performed under reducing conditions.

Predicted band size: 45 kDa
Observed band size: 40 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab211017**).

Lanes 1 - 6: Merged signal (red and green). Green - **ab211017** observed at 40 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

ab211017 was shown to react with Indoleamine 2, 3-dioxygenase in treated wild-type A549 cells in Western blot with no signal observed in treated IDO1 knockout cell line **ab266949** (IDO1 knockout cell lysate **ab256948**). Wild-type A549 and IDO1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab211017** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Immunohistochemical analysis of paraffin-embedded human spleen tissue labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

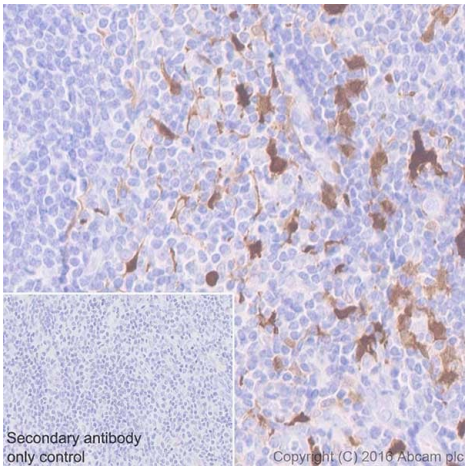
Cytoplasmic and nuclear staining on dendritic cells of human spleen is observed (PMID: 21328335, PMID: 25271151).

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

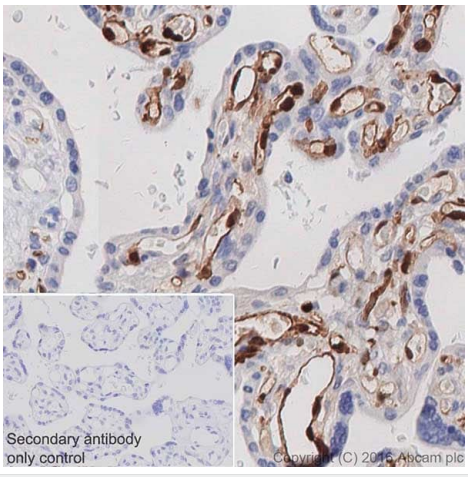
Cytoplasmic and nuclear staining on dendritic cells of human tonsil is observed (PMID: 21328335, PMID: 25271151).

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Immunohistochemical analysis of paraffin-embedded human placenta tissue labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

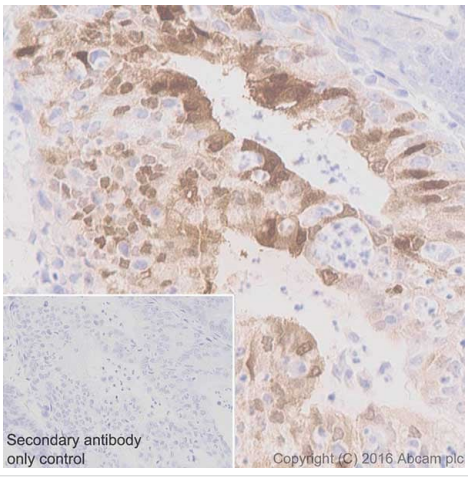
Cytoplasmic and nuclear staining on endothelial cells of human placenta is observed (PMID: 21328335, PMID: 25271151).

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Immunohistochemical analysis of paraffin-embedded human endometrium cancer tissue labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

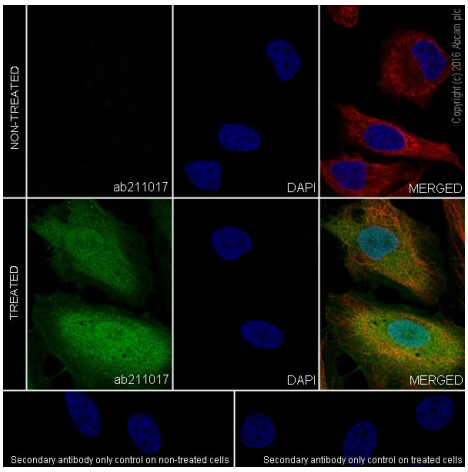
Cytoplasmic and nuclear staining on human endometrium cancer is observed (PMID: 26155395).

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Immunocytochemistry/ Immunofluorescence - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells, treated with 50ng/ml IFN-γ for 16 hours or untreated, labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/2000 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green).

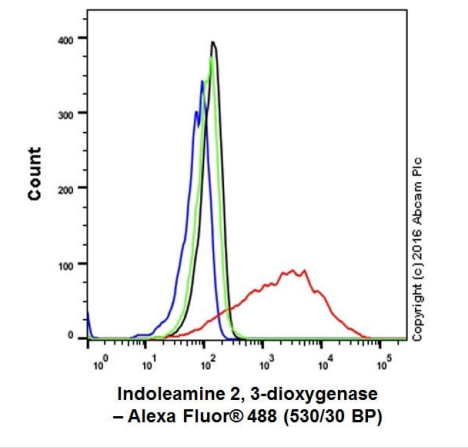
The signal increased after treatment with IFN-γ (50 ng/ml) for 16 hours on HeLa cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with **ab195889** (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) at 1/1000 dilution.

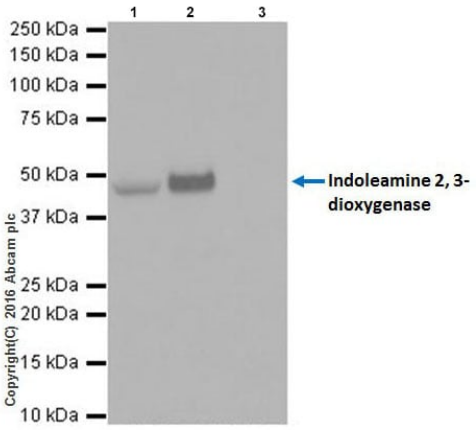
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Flow Cytometry (Intracellular) - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells, treated with 50ng/ml IFN-gamma for 16h (red) or untreated (green), labeling Indoleamine 2, 3-dioxygenase with **ab211017** at 1/500 dilution compared with a rabbit monoclonal IgG isotype control (**ab172730**; black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).



Immunoprecipitation - Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

Indoleamine 2, 3-dioxygenase was immunoprecipitated from 0.35 mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate treated with 50ng/ml IFN-γ for 16h with **ab211017** at 1/40 dilution.

Western blot was performed from the immunoprecipitate using **ab211017** at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

Lane 1: HeLa treated with 50ng/ml IFN-γ for 16h whole cell lysate 10 µg (Input).

Lane 2: **ab211017** IP in HeLa treated with 50ng/ml IFN-γ for 16h whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab211017** in HeLa treated with 50ng/ml IFN-γ for 16h whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab211017**).

Anti-Indoleamine 2, 3-dioxygenase antibody [EPR20374] - BSA and Azide free (ab271990)

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