abcam

Product datasheet

Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free ab193556

Recombinant RabMAb

16 Images

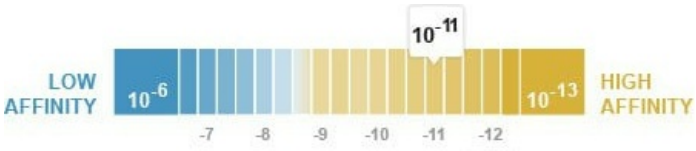
Overview

Product name	Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free
Description	Rabbit monoclonal [EPR3730(2)] to PAX5 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, IP, WB, IHC-P, ChIP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Ramos and Daudi cell lysates and human tonsil and mouse spleen tissue lysates. IHC-P: Human tonsil, hodgkin's lymphoma, spleen and diffuse B-cell lymphoma tissues. ICC/IF: Ramos cells. Flow Cyt (intra): Ramos cells. IP: Mouse spleen tissue lysate and Ramos cells. ChIP: Ramos cells
General notes	ab193556 is the carrier-free version of **ab109443**. 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.
Dissociation constant (K_D)	$K_D = 4.45 \times 10^{-11}$ M



Learn more about K_D

Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3730(2)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab193556 in the following tested applications.

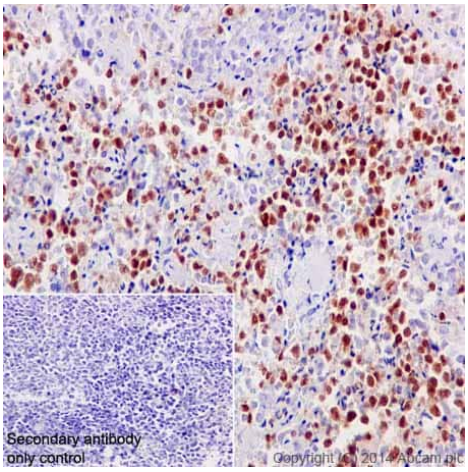
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 42 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
ChIP		Use at an assay dependent concentration.

Target

Function	May play an important role in B-cell differentiation as well as neural development and spermatogenesis. Involved in the regulation of the CD19 gene, a B-lymphoid-specific target gene.
Involvement in disease	A chromosomal aberration involving PAX5 is a cause of acute lymphoblastic leukemia. Translocation t(9;18)(p13;q11.2) with ZNF521. Translocation t(9;3)(p13;p14.1) with FOXP1. Translocation t(9;12)(p13;p13) with ETV6. Leukemia, acute lymphoblastic, 3
Sequence similarities	Contains 1 paired domain.
Developmental stage	Expressed at early B-cell differentiation, in the developing CNS and in adult testis.
Post-translational modifications	O-glycosylated.
Cellular localization	Nucleus.

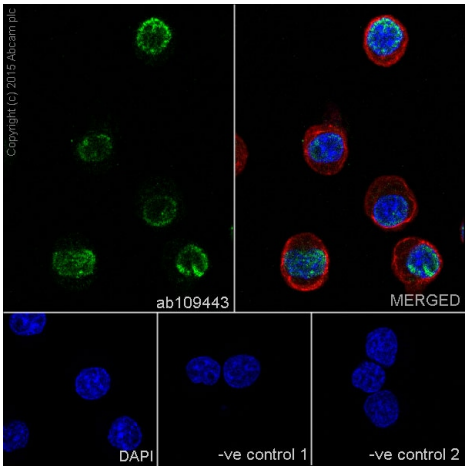
Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat spleen tissue labelling PAX5 with purified **ab109443** at 1/1000. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



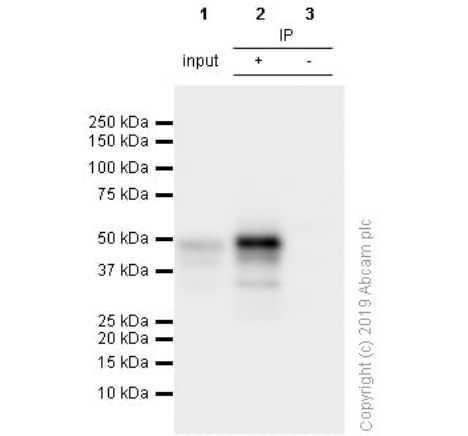
Immunocytochemistry/ Immunofluorescence - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunocytochemistry/Immunofluorescence analysis of Ramos cells labelling PAX5 with purified **ab109443** at 1/250. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. **ab7291**, a mouse anti-tubulin (1/1000) and **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used.

Control 1: primary antibody (1/250) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



Immunoprecipitation - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

ab109443 (purified) at 1/20 dilution immunoprecipitating PAX5 in mouse spleen lysate 10 µg.
Lane 1 (input): mouse spleen lysate 10 µg
Lane 2 (+): **ab109443** & mouse spleen lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab109443** in mouse spleen lysate

For western blotting, **ab109443** at 1/500 dilution (0.268 µg/mL) and veriBlot for IP secondary antibody (HRP) (**ab131366**) at 1/1000 dilution was used.

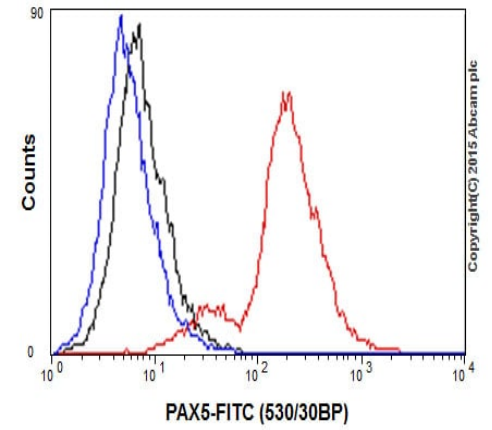
Blocking and diluting buffer: 5% NFDM /TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**)

ChIP - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Chromatin was prepared from Ramos cells according to the Abcam Dual-X-ChIP protocol*. Cells were fixed with 1.5 mM EGS for 30mins and then formaldehyde for 10min.
The ChIP was performed with 25 µg of chromatin, 5 µg of abzz (red), or 5 µg of rabbit normal IgG **ab172730** (gray) and 25 µl of Protein A/G Dynabeads. The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).
Primers and probes are from paper PMID:19806612
*http://www.abcam.com/resources?keywords=X%20ChIP%20protocol

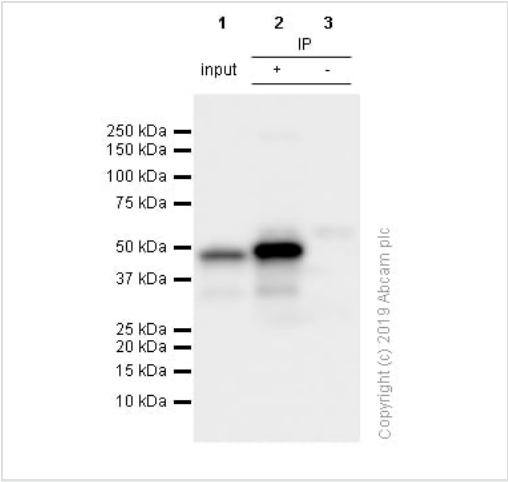
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



Flow Cytometry (Intracellular) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Intracellular Flow Cytometry analysis of Ramos cells labelling PAX5 with purified **ab109443** at 1/30 (red). Cells were fixed with 80% methanol. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



Immunoprecipitation - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

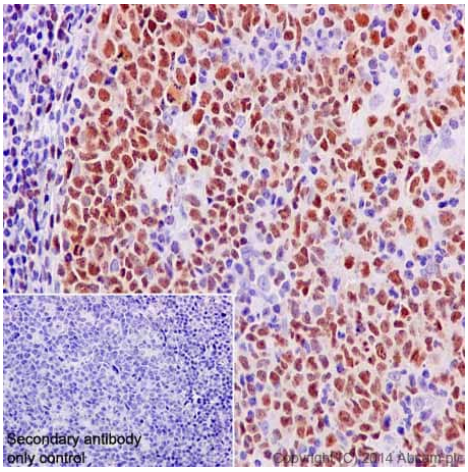
ab109443 (purified) at 1/20 dilution immunoprecipitating PAX5 in Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate 10 µg.

Lane 1 (input): Ramos (Human Burkitt's lymphoma B lymphocyte) whole cell lysate 10 µg
Lane 2 (+): **ab109443** & Ramos whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab109443** in Ramos whole cell lysate

For western blotting, **ab109443** at 1/500 dilution (0.268 µg/mL) and veriBlot for IP secondary antibody (HRP) (**ab131366**) at 1/1000 dilution was used.

Blocking and diluting buffer: 5% NFDM /TBST.

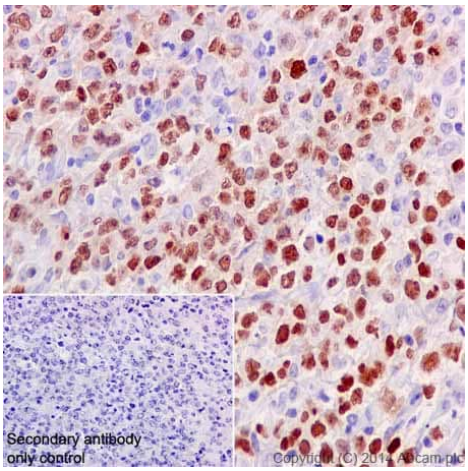
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling PAX5 with purified **ab109443** at 1/1000. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

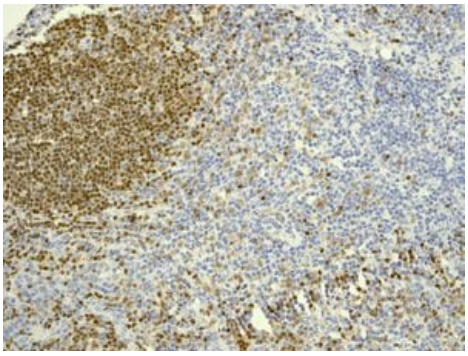
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human B-cell lymphoma tissue labelling PAX5 with purified **ab109443** at 1/1000. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

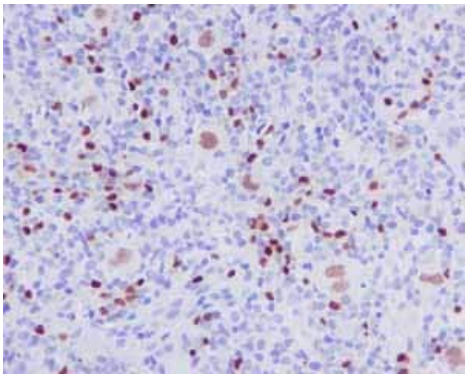


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling PAX5 with unpurified **ab109443**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

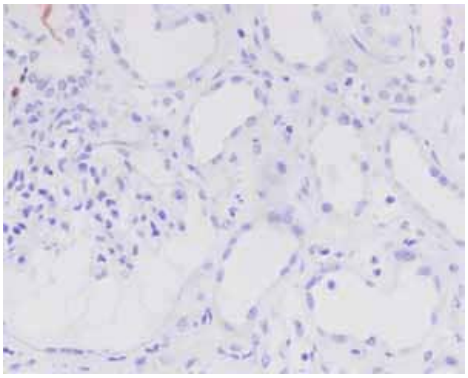


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human Hodgkin's lymphoma tissue labelling PAX5 with unpurified **ab109443**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

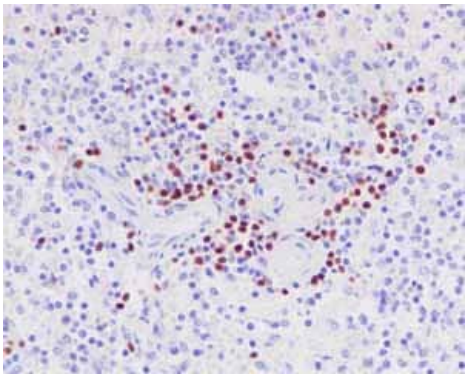


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

ab109443 showing negative staining in Normal kidney tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

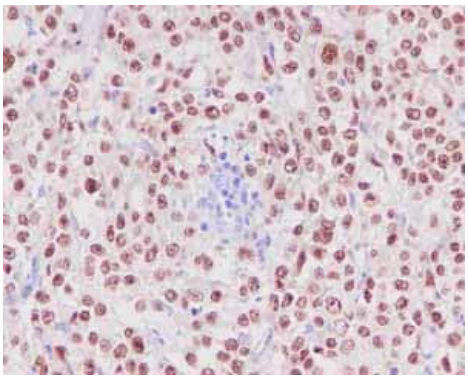


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human normal spleen tissue labelling PAX5 with unpurified **ab109443**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

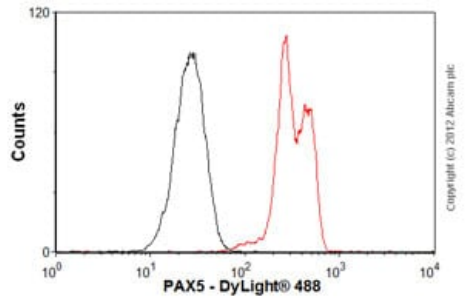


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human diffuse B-cell lymphoma tissue labelling PAX5 with unpurified **ab109443**.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

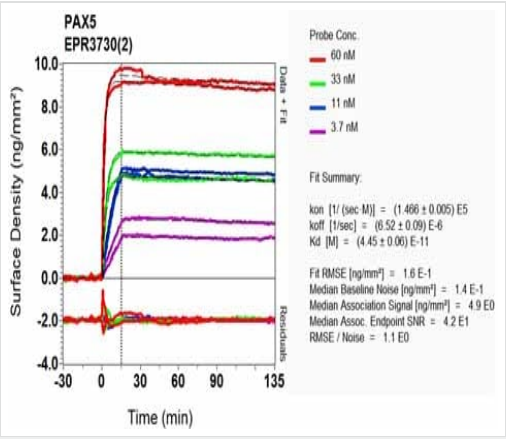
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Overlay histogram showing Ramos cells stained with unpurified **ab109443** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (unpurified **ab109443**, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).



OI-RD Scanning - Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Anti-PAX5 antibody [EPR3730(2)] - BSA and Azide free (ab193556)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab109443**).

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