

Product datasheet

Anti-PAX5 antibody ab183575

[2 References](#) [5 Images](#)

Overview

Product name	Anti-PAX5 antibody
Description	Rabbit polyclonal to PAX5
Host species	Rabbit
Tested applications	Suitable for: WB, ICC/IF, IP, Flow Cyt
Species reactivity	Reacts with: Mouse, Human
Immunogen	Recombinant fragment corresponding to Human PAX5 (internal sequence). Expressed in E. coli. Database link: Q02548
Positive control	WB: Ramos, Raji, Daudi, A20 IP: Raji ICC: Ramos Flow Cyt: Ramos
General notes	<p>The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.</p> <p>If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.05% Sodium azide Constituents: 0.01% BSA, 30% Glycerol (glycerin, glycerine), 69% PBS
Purity	Protein G purified
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab183575 in the following tested applications.

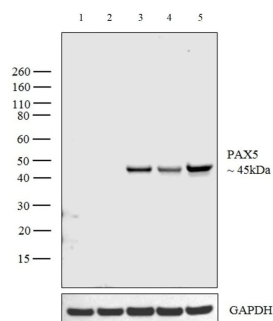
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/500 - 1/2000. Predicted molecular weight: 42 kDa.
ICC/IF		1/50 - 1/200.
IP		Use at 2-5 µg/mg of lysate.
Flow Cyt		1/50. ab171870 - Rabbit polyclonal IgG, is suitable for use as an isotype control with this antibody.

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



Western blot - Anti-PAX5 antibody (ab183575)

All lanes : Anti-PAX5 antibody (ab183575) at 1/1000 dilution

Lane 1 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lane 2 : HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lane 3 : Ramos (Human Burkitt's lymphoma cell line) whole cell lysate

Lane 4 : Raji (Human Burkitt's lymphoma cell line) whole cell lysate

Lane 5 : Daudi (Human Burkitt's lymphoma cell line) whole cell lysate

Lysates/proteins at 30 µg per lane.

Secondary

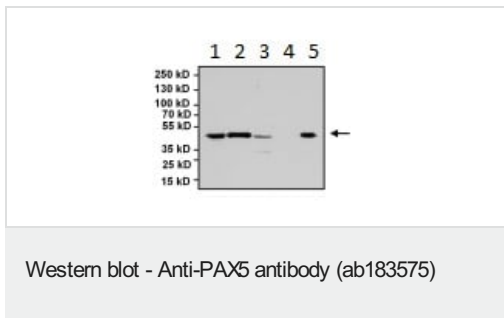
All lanes : Goat anti-Rabbit IgG (H+L) Superclonal™ Secondary Antibody, HRP conjugate at 1/4000 dilution

Predicted band size: 42 kDa

Additional bands at: ~45 kDa. We are unsure as to the identity of these extra bands.

Detection: chemiluminescence.

Western blot demonstrating antibody specificity of the target across different cell lysates. Expression of PAX5 was observed in all cell lysates tested in comparison to Jurkat and Hep G2 using ab183575 in western blot.



All lanes : Anti-PAX5 antibody (ab183575) at 1/1000 dilution

Lane 1 : Ramos whole cell lysate

Lane 2 : Daudi whole cell lysate

Lane 3 : Raji whole cell lysate

Lane 4 : Jurkat whole cell lysate

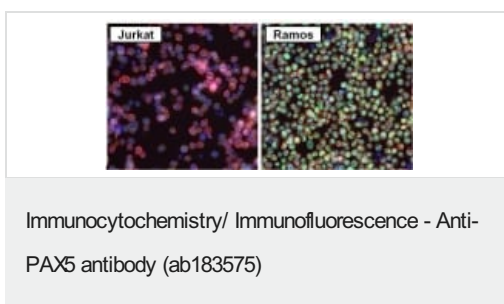
Lane 5 : A20 whole cell lysate

Lysates/proteins at 75 µg per lane.

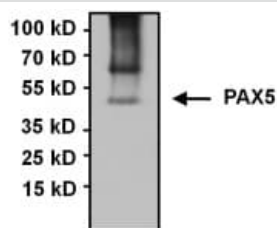
Secondary

All lanes : Goat anti rabbit HRP at 1/500 dilution

Predicted band size: 42 kDa

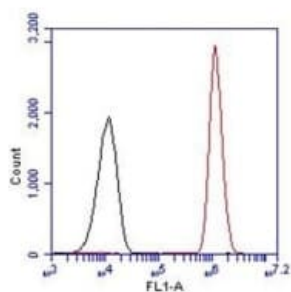


Immunofluorescent analysis of PAX5 (green) in Ramos and negative control Jurkat cells. Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature. Cells were blocked with 2% Blocker BSA for 15 minutes at room temperature. Cells were probed with ab183575 at a dilution of 1/100 for at least 1 hour at room temperature, washed with PBS, and incubated with a DyLight 488-conjugated goat anti-rabbit IgG secondary antibody at a dilution of 1/200 for 30 minutes at room temperature. F-Actin (red) was stained with DyLight-554 Phalloidin and nuclei (blue) were stained with Hoechst 33342 dye.



Immunoprecipitation - Anti-PAX5 antibody
(ab183575)

Immunoprecipitation of PAX5 was performed using Raji whole cell lysates. Antigen-antibody complexes were formed by incubating 75 ug of lysate with 5ug of ab183575 overnight on a rocking platform at 4°C. The immune complexes were captured on 50ul Protein A/G Agarose, washed extensively, and eluted with 5X Lane Marker Reducing Sample Buffer. Samples was resolved on a 4-20% Tris-HCl polyacrylamide gel, transferred to a PVDF membrane, and blocked with 5% BSA/TBS-0.1%Tween-20 for 1 hour. The membrane was probed with ab183575 at a dilution of 1/1000 overnight rotating at 4°C. Membranes were washed in TBST, and probed with an IP Detection Reagent at a dilution of 1/1000 for at least one hour. Chemiluminescent detection was performed.



Flow Cytometry - Anti-PAX5 antibody (ab183575)

Flow cytometry analysis of PAX5 (red histogram) on Ramos cells. Cells were harvested, fixed with 4% formaldehyde, permeabilized, washed with PBS, and incubated with ab183575 at a 1/50 dilution or with PBS alone (black histogram) for 1 hour at room temperature. For flow cytometry analysis, 30-minute incubation with DyLight 488 goat anti-rabbit IgG secondary antibody was performed. 30,000 cells were acquired for analysis.

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