abcam

Product datasheet

Anti-RUNX1 / AML1 antibody [EPR23044-100] ab240639

Recombinant RabMAb

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Overview

Product name	Anti-RUNX1 / AML1 antibody [EPR23044-100]
Description	Rabbit monoclonal [EPR23044-100] to RUNX1 / AML1
Host species	Rabbit
Tested applications	**Suitable for:** ChIC/CUT&RUN-seq, Flow Cyt (Intra), WB, IP, IHC-P, ICC/IF **Unsuitable for:** ChIP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: THP-1, Jurkat and MOLT-4 lysates. IHC-P: Human breast carcinoma and Human breast tissues. ICC/IF: Jurkat cells. Flow Cyt (intra): Jurkat cells. IP: Jurkat cells. ChIC/CUT&RUN-Seq: K-562 cells.

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	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR23044-100
Isotype	IgG

Applications

The Abpromise guarantee	Our **Abpromise guarantee** covers the use of ab240639 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
ChIC/CUT&RUN-seq		Use at an assay dependent concentration. 5µg
Flow Cyt (Intra)		1/500.
WB	★★★★☆ (5)	1/1000. Predicted molecular weight: 48 kDa.
IP		1/30.
IHC-P	★★★★★ (4)	1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★★ (3)	1/100.

Application notes Is unsuitable for ChIP.

Target

Function CBF binds to the core site, 5'-PYGPYGGT-3', of a number of enhancers and promoters, including murine leukemia virus, polyomavirus enhancer, T-cell receptor enhancers, LCK, IL-3 and GM-CSF promoters. The alpha subunit binds DNA and appears to have a role in the development of normal hematopoiesis. Isoform AML-1L interferes with the transactivation activity of RUNX1. Acts synergistically with ELF4 to transactivate the IL-3 promoter and with ELF2 to transactivate the mouse BLK promoter. Inhibits MYST4-dependent transcriptional activation.

Tissue specificity Expressed in all tissues examined except brain and heart. Highest levels in thymus, bone marrow and peripheral blood.

Involvement in disease Note=A chromosomal aberration involving RUNX1/AML1 is a cause of M2 type acute myeloid leukemia (AML-M2). Translocation t(8;21)(q22;q22) with RUNX1T1.
Note=A chromosomal aberration involving RUNX1/AML1 is a cause of therapy-related myelodysplastic syndrome (T-MDS). Translocation t(3;21)(q26;q22) with EAP or MECOM.
Note=A chromosomal aberration involving RUNX1/AML1 is a cause of chronic myelogenous leukemia (CML). Translocation t(3;21)(q26;q22) with EAP or MECOM.
Note=A chromosomal aberration involving RUNX1/AML1 is found in childhood acute lymphoblastic leukemia (ALL). Translocation t(12;21)(p13;q22) with TEL. The translocation fuses the 3'-end of TEL to the alternate 5'-exon of AML-1H.
Note=A chromosomal aberration involving RUNX1 is found in acute leukemia. Translocation t(11,21)(q13;q22) that forms a MACROD1-RUNX1 fusion protein.
Defects in RUNX1 are the cause of familial platelet disorder with associated myeloid malignancy (FPDMM) [MIM:601399]. FPDMM is an autosomal dominant disease characterized by qualitative and quantitative platelet defects, and propensity to develop acute myelogenous leukemia.
Note=A chromosomal aberration involving RUNX1/AML1 is found in therapy-related myeloid malignancies. Translocation t(16;21)(q24;q22) that forms a RUNX1-CBFA2T3 fusion protein.
Note=A chromosomal aberration involving RUNX1/AML1 is a cause of chronic myelomonocytic leukemia. Inversion inv(21)(q21;q22) with USP16.

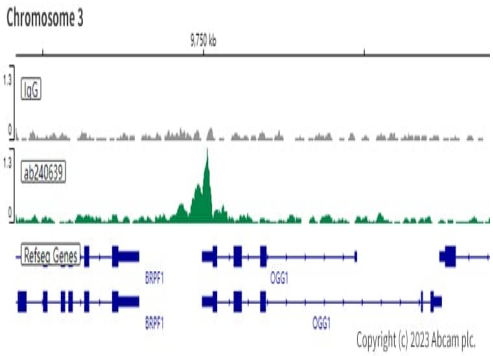
Sequence similarities Contains 1 Runt domain.

Domain A proline/serine/threonine rich region at the C-terminus is necessary for transcriptional activation of target genes.

Post-translational Phosphorylated in its C-terminus upon IL-6 treatment. Phosphorylation enhances interaction with

modifications	MYST3. Methylated.
Cellular localization	Nucleus.

Images

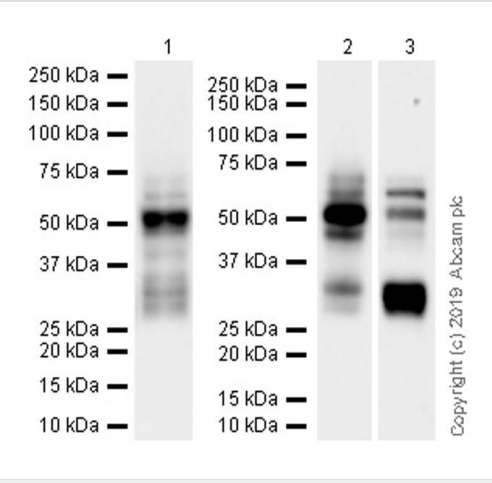


ChIC/CUT&RUN sequencing - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/mL, 2.5 x 10^5 K-562 (Human chronic myelogenous leukemia lymphoblast) cells and 5µg of ab240639 [EPR23044-100]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown.

Additional screenshots of mapped reads can be downloaded **here**.

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Western blot - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

All lanes : Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639) at 1/1000 dilution

Lane 1 : THP-1 (human monocytic leukemia monocyte), whole cell lysate
Lane 2 : Jurkat (human T cell leukemia T lymphocyte), whole cell lysate
Lane 3 : MOLT-4 (human lymphoblastic leukemia T lymphoblast), whole cell lysate

Lysates/proteins at 10 µg per lane.

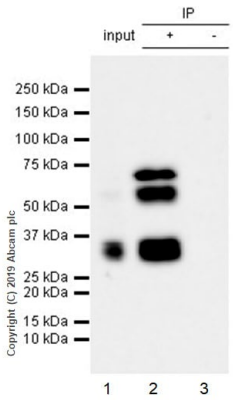
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 48 kDa
Observed band size: 27-55 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST
The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 23352661, 29296779). The RUNX1 gene has several isoforms, 3 major

isotypes. RUNX1b is broadly expressed, and RUNX1a overexpression has been reported in AML.

Exposure time: Lane 1: 48 seconds Lanes 2-3: 26 seconds



Immunoprecipitation - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

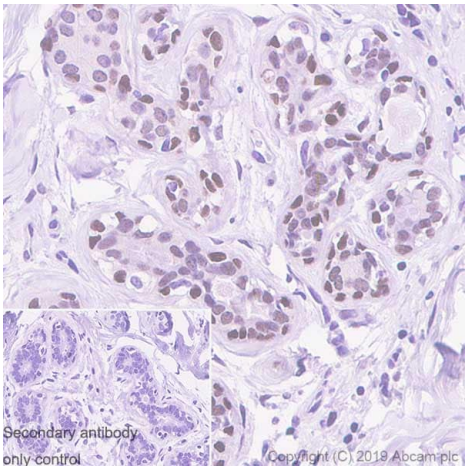
RUNX1 / AML1 was immunoprecipitated from 0.35 mg Jurkat (human T cell leukemia T lymphocyte) whole cell lysate 10ug with ab240639 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab240639 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.
Lane 1: Jurkat (human T cell leukemia T lymphocyte) whole cell lysate 10ug

Lane 2: ab240639 IP in Jurkat whole cell lysate

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab240639 in Jurkat whole cell lysate

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

Exposure time: 10 seconds

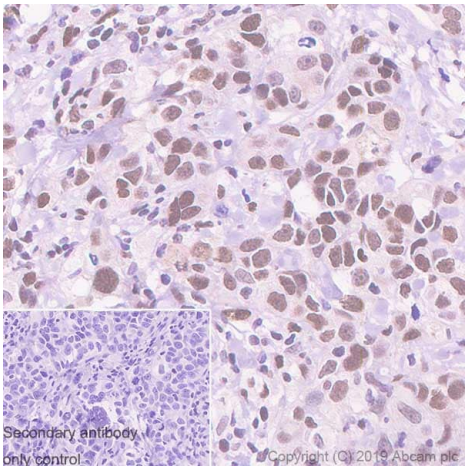


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

Immunohistochemical analysis of paraffin-embedded Human breast tissue labeling RUNX1 / AML1 with ab240639 at 1/2000 dilution (0.25 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on human breast tissue is observed. The section was incubated with ab240639 for 15 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

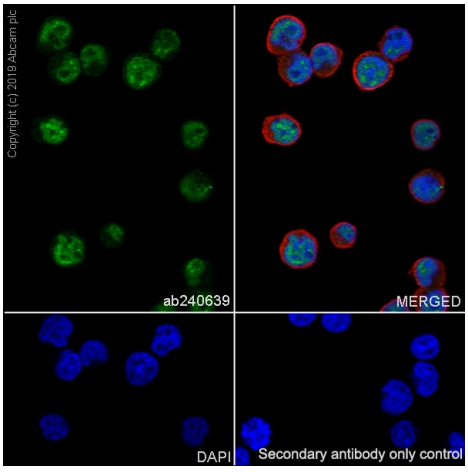


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling RUNX1 / AML1 with ab240639 at 1/2000 dilution (0.25 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on human breast carcinoma (PMID: 24967588) tissue is observed. The section was incubated with ab240639 for 15 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).

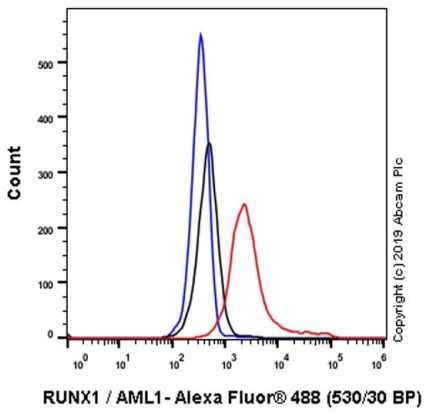
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



Immunocytochemistry/ Immunofluorescence - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

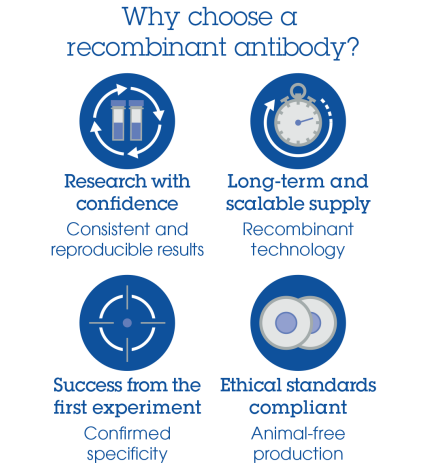
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Jurkat (Human T cell leukemia T lymphocyte) cells labelling RUNX1 / AML1 with ab240639 at 1/100 dilution, followed by ab240639 anti-RUNX1/AML1 **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal image showing strong nuclear and weak cytoplasmic staining in Jurkat cell line is observed. Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab240639 anti-RUNX1/AML1 **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Jurkat (Human T cell leukemia T lymphocyte) cells labelling RUNX1 / AML1 with ab240639 at 1/500 (Red) compared with a Rabbit monoclonal IgG (**ab172730**, Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



Anti-RUNX1 / AML1 antibody [EPR23044-100] (ab240639)

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