abcam

Product datasheet

Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free ab269457

KO 評価済 リコンビナント RabMAb

画像数 11

製品の概要

製品名	Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free
製品の詳細	Rabbit monoclonal [EPR23436-45] to AICDA - ChIP Grade – BSA and Azide free
由来種	Rabbit
アプリケーション	**適用あり:** Flow Cyt (Intra), WB, IHC-P, ICC/IF, IP, ChIP
種交差性	**交差種:** Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: Ramos,NAMALWA and Raji lysates. IHC-P: Human tonsil, Human Hodgkin lymphoma and Human diffuse large B-cell lymphoma tissues. ICC/IF: NAMALWA cells. Flow Cyt (intra): NAMALWA cells. IP: Ramos and NAMALWA cells.ChIP: Chromatin prepared from Ramos cells.
特記事項	ab269457 is the carrier-free version of **ab269454**. 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**.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C. Do Not Freeze.
バッファー	pH: 7.2 Constituent: PBS
キャリア・フリー	はい
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EPR23436-45
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab269457の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 24 kDa (predicted molecular weight: 23 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
ChIP		Use at an assay dependent concentration.

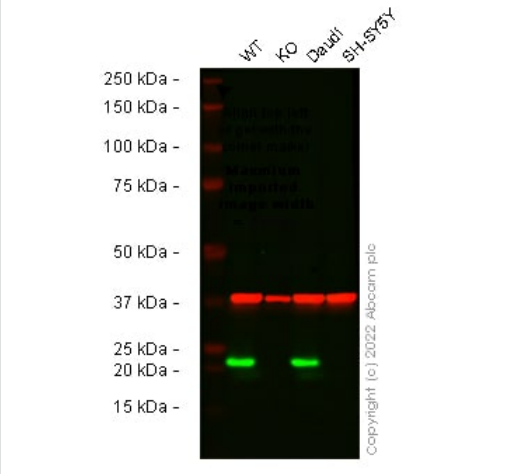
ターゲット情報

機能	RNA-editing deaminase involved in somatic hypermutation, gene conversion, and class-switch recombination. Required for several crucial steps of B-cell terminal differentiation necessary for efficient antibody responses.
組織特異性	Strongly expressed in lymph nodes and tonsils.
関連疾患	Defects in AICDA are the cause of hyper-IgM immunodeficiency syndrome type 2 (HIGM2) [MIM:605258]; also known as hyper-IgM syndrome 2. HIGM2 is an autosomal recessive disorder characterized by normal or elevated serum IgM levels with absence of IgG, IgA, and IgE, resulting in a profound susceptibility to bacterial infections. HIGM2 causes the absence of Ig class switch

recombination (CSR), the lack of Ig somatic hypermutations, and lymph node hyperplasia caused by the presence of giant germinal centers.

配列類似性 Belongs to the cytidine and deoxycytidylate deaminase family.

画像



Western blot - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

All lanes : Anti-AICDA antibody [EPR23436-45] - ChIP Grade (**ab269454**) at 1/1000 dilution

Lane 1 : Wild-type Raji cell lysate
Lane 2 : AICDA knockout Raji cell lysate
Lane 3 : Daudi cell lysate
Lane 4 : SH-SY5Y cell lysate

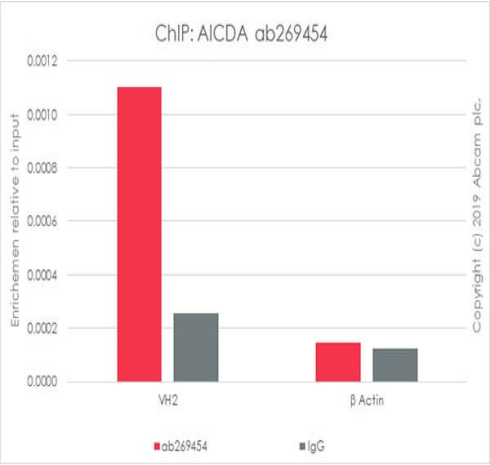
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 23 kDa
Observed band size: 22 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).

False colour image of Western blot: Anti-AICDA antibody [EPR23436-45] - ChIP Grade staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab269454** was shown to bind specifically to AICDA. A band was observed at 22 kDa in wild-type Raji cell lysates with no signal observed at this size in AICDA knockout cell line **ab277185** (knockout cell lysate **ab277227**). To generate this image, wild-type and AICDA knockout Raji cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.



ChIP - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

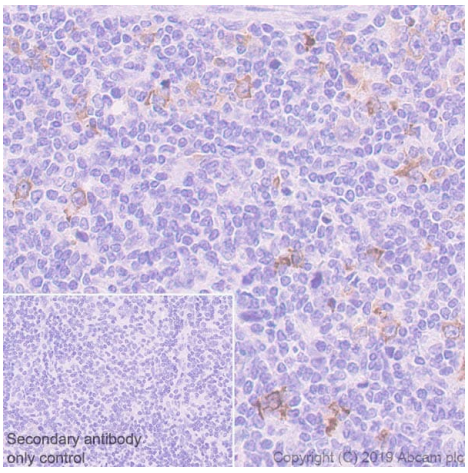
Chromatin was prepared from Ramos cells according to the Abcam Dual-X-ChIP protocol*. Cells were fixed with 1.5 mM EGS for 30mins and then formaldehyde for 10min.

The ChIP was performed with 25 µg of chromatin, 5 µg of **ab269454** (red), or 5 µg of rabbit normal IgG **ab172730** (gray) and 20 µl of Protein A/G sepharose beads. The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci).

Primers and probes are commercial primers from paper: PMC2905439

***https://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 (**ab269454**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

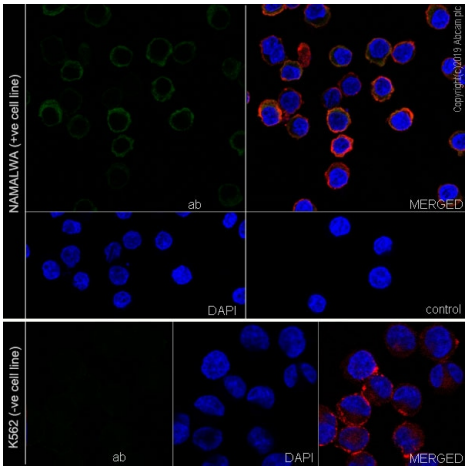
Immunohistochemical analysis of paraffin-embedded Human diffuse large B-cell lymphoma tissue labeling AICDA with **ab269454** at 1/4000 dilution (0.12ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Mainly cytoplasmic staining (weak nuclear staining) in part of tumor cells of human diffuse large B-cell lymphoma (PMID: 29251015).

The section was incubated with **ab255611** for 30 mins at room temperature. The immunostaining 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).

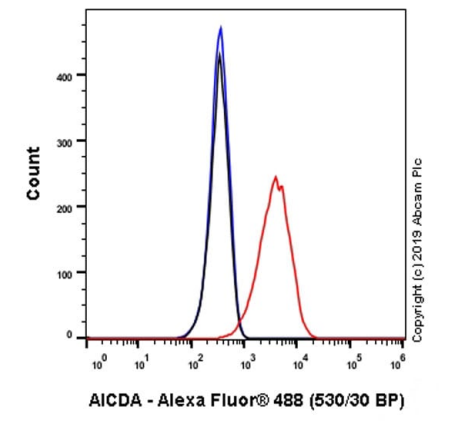


Immunocytochemistry/ Immunofluorescence - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NAMALWA (human Burkitt's lymphoma B lymphocyte) and K-562 (human chronic myelogenous leukemia lymphoblast) cells labelling AICDA with **ab269454** at 1/50 dilution, followed by **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in NAMALWA cell line. **Negative control:** K-562 cell line (PMID: 27217538). **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 Ab269454 anti-AICDA **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution.

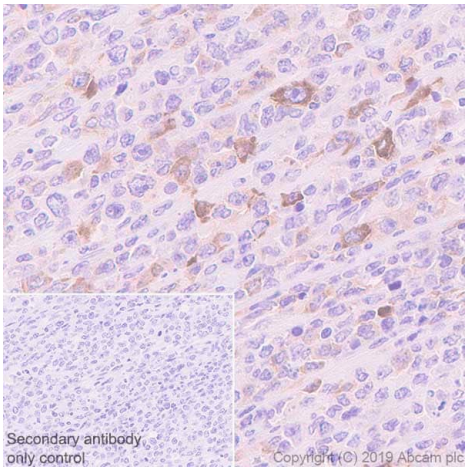
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Flow Cytometry (Intracellular) - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized Ramos (human Burkitt's lymphoma B lymphocyte) cells labelling AICDA with **ab269454** at 1/500 dilution (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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

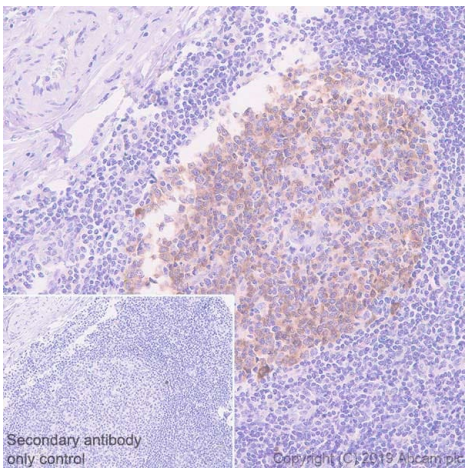
Immunohistochemical analysis of paraffin-embedded Human Hodgkin lymphoma tissue labeling AICDA with **ab269454** at 1/4000 dilution (0.12ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Mainly cytoplasmic staining (weak nuclear staining) in part of tumor cells of human Hodgkin lymphoma (PMID: 15732141).

The section was incubated with **ab255611** for 30 mins at room temperature. The immunostaining 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

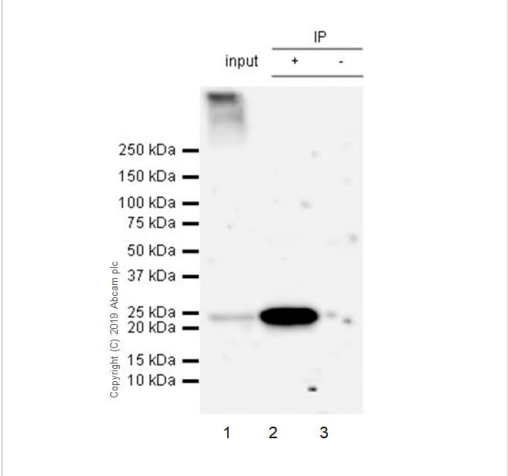
Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling AICDA with **ab269454** at 1/4000 dilution (0.12ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Mainly cytoplasmic staining (weak nuclear staining) in germinal center cells of human tonsil (PMID:23877718, 15732141, PMID: 29251015).

The section was incubated with **ab255611** for 30 mins at room temperature. The immunostaining 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Immunoprecipitation - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

AICDA was immunoprecipitated from 0.35 mg NAMALWA (human Burkitt's lymphoma B lymphocyte) whole cell lysate with **ab269454** at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab269454** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

Lane 1: NAMALWA whole cell lysate 10ug.

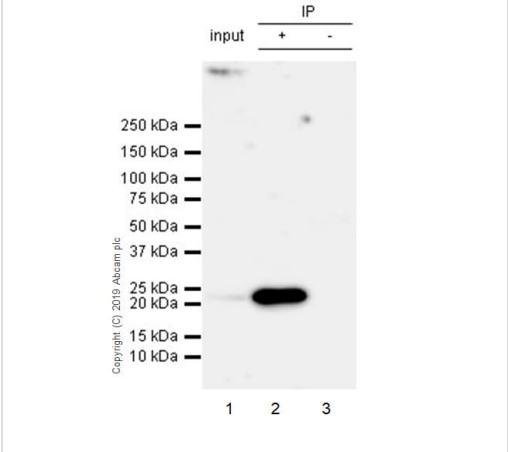
Lane 2: **ab269454** IP in NAMALWA whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab269454** in NAMALWA whole cell lysate.

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

Exposure time: 90 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Immunoprecipitation - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

AICDA was immunoprecipitated from 0.35 mg Ramos (human Burkitt's lymphoma B lymphocyte) whole cell lysate with **ab269454** at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab269454** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

Lane 1: Ramos whole cell lysate 10ug.

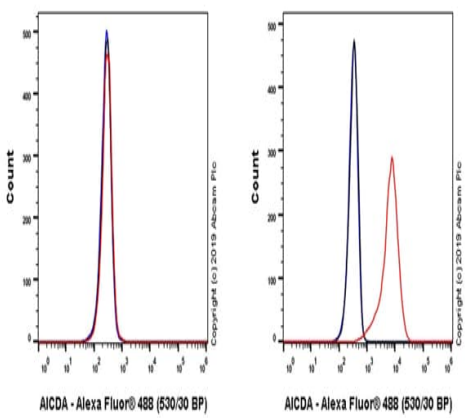
Lane 2: **ab269454** IP in Ramos whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab269454** in Ramos whole cell lysate.

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

Exposure time: 90 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).

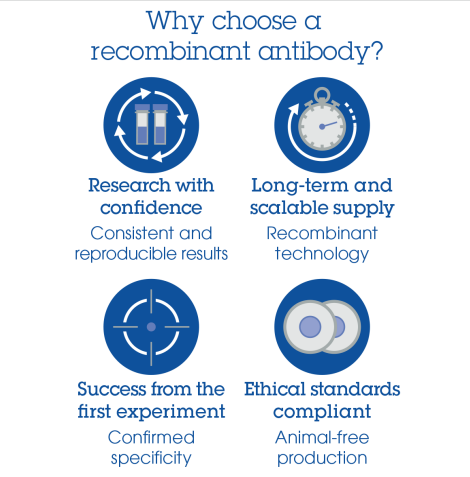


Flow Cytometry (Intracellular) - Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized K-562 (human chronic myelogenous leukemia lymphoblast, Left) / NAMALWA (human Burkitt's lymphoma B lymphocyte, Right) cells labelling AICDA with **ab269454** at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Blac) 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.

Negative control: K-562 cell line (PMID: 27217538).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab269454**).



Anti-AICDA antibody [EPR23436-45] - ChIP Grade - BSA and Azide free (ab269457)

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