

Anti-Fyn antibody [EPR19636] ab184276

KO 評価済 リコンビナント RabMAb

5 References 画像数 11

製品の概要

| | |
|--------------|---|
| 製品名 | Anti-Fyn antibody [EPR19636] |
| 製品の詳細 | Rabbit monoclonal [EPR19636] to Fyn |
| 由来種 | Rabbit |
| アプリケーション | 適用あり: WB, IHC-P, IP, Flow Cyt (Intra) |
| 種交差性 | 交差種: Mouse, Rat, 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: Human brain lysate; Ramos, HEK-293, RAW 264.7, PC-12 and NIH/3T3 whole cell lysates; P0 mouse brain lysate; P0 rat brain lysate; Rat spleen lysate. IHC-P: Human tonsil and cerebral cortex tissues; Mouse spleen tissue; Rat testis tissue. Flow Cyt (intra): RAW 264.7 cells. IP: P0 mouse brain lysate. |
| 特記事項 | <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> |

製品の特性

| | |
|-------|---|
| 製品の状態 | Liquid |
| 保存方法 | 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. |
| バッファー | pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol, 0.05% BSA |
| 精製度 | Protein A purified |
| ポリ/モノ | モノクローナル |
| クローン名 | EPR19636 |

アプリケーション

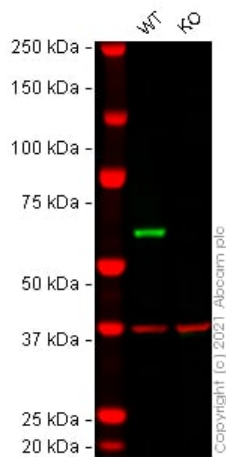
The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab184276の使用に適用されず
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

| アプリケーション | Abreviews | 特記事項 |
|------------------|-----------|---|
| WB | | 1/1000. Detects a band of approximately 61 kDa (predicted molecular weight: 61 kDa). |
| IHC-P | | 1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |
| IP | | 1/40. |
| Flow Cyt (Intra) | | 1/50. |

ターゲット情報

| | |
|--------------|---|
| 機能 | Tyrosine-protein kinase implicated in the control of cell growth. Plays a role in the regulation of intracellular calcium levels, with isoform 2 showing the greater ability to mobilize cytoplasmic calcium in comparison to isoform 1. Required in brain development and mature brain function with important roles in the regulation of axon growth, axon guidance, and neurite extension. Blocks axon outgrowth and attraction induced by NTN1 by phosphorylating its receptor DDC. Phosphorylates RUNX3. |
| 組織特異性 | Isoform 1 is highly expressed in the brain. Isoform 2 is expressed in cells of hemopoietic lineages, especially T lymphocytes. |
| 配列類似性 | Belongs to the protein kinase superfamily. Tyr protein kinase family. SRC subfamily. Contains 1 protein kinase domain. Contains 1 SH2 domain. Contains 1 SH3 domain. |
| 細胞内局在 | Cell membrane. Present and active in lipid rafts. Present in cell body and along the process of mature and developing oligodendrocytes. |
| 製品の状態 | This protein is known to be similar in amino acid sequence to HCK (P08631), LCK (P06239), YES1 (P07947), SRC (P12931), and LYN (P07948). Therefore, cross-reactivity with these homologous proteins may be observed. We would be happy to provide immunogen alignment information upon request. |

画像



Western blot - Anti-Fyn antibody [EPR19636]
(ab184276)

All lanes : Anti-Fyn antibody [EPR19636] (ab184276) at 1/1000 dilution

Lane 1 : Wild-type HEK-293 cell lysate

Lane 2 : FYN knockout HEK-293 cell lysate

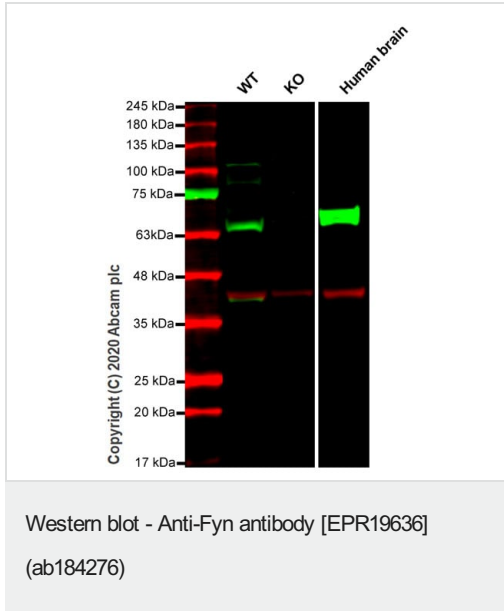
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 61 kDa

Observed band size: 60 kDa

False colour image of Western blot: Anti-Fyn antibody [EPR19636] 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, ab184276 was shown to bind specifically to Fyn. A band was observed at 60 kDa in wild-type HEK-293 cell lysates with no signal observed at this size in FYN knockout cell line [ab269630](#) (knockout cell lysate [ab272440](#)). To generate this image, wild-type and FYN knockout HEK-293 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 (IRDye[®] 800CW) preabsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye[®] 680RD) preabsorbed ([ab216776](#)) at 1/20000 dilution.



All lanes : Anti-Fyn antibody [EPR19636] (ab184276) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T cell lysate

Lane 2 : FYN knockout HEK-293T cell lysate

Lane 3 : Human brain tissue lysate

Lysates/proteins at 20 µg per lane.

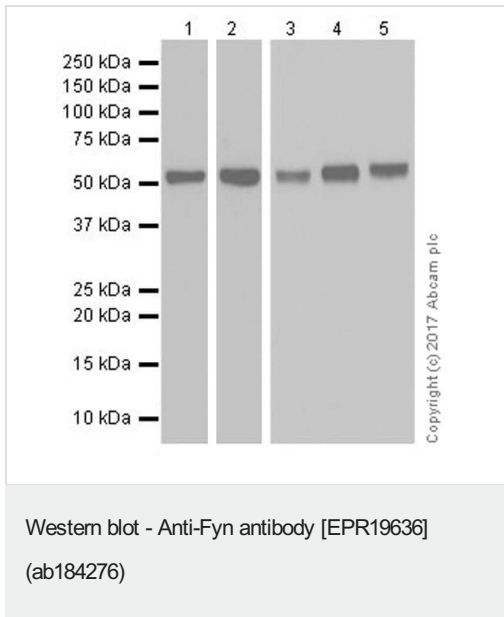
Performed under reducing conditions.

Predicted band size: 61 kDa

Observed band size: 159 kDa

Lanes 1-3: Merged signal (red and green). Green - ab184276 observed at 60 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab184276 Anti-Fyn antibody [EPR19636] was shown to specifically react with Fyn in wild-type HEK293T cells. Loss of signal was observed when knockout cell line **ab266133** (knockout cell lysate **ab257071**) was used. Wild-type and Fyn knockout samples were subjected to SDS-PAGE. ab184276 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.



All lanes : Anti-Fyn antibody [EPR19636] (ab184276) at 1/1000 dilution

Lane 1 : Human brain lysate

Lane 2 : Ramos (human Burkitt's lymphoma cell line) whole cell lysate

Lane 3 : HEK-293 (human epithelial cell line from embryonic kidney) whole cell lysate

Lane 4 : P0 mouse brain lysate

Lane 5 : P0 rat brain lysate

Lysates/proteins at 20 µg per lane.

Secondary

Lane 1 : VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) at 1/4000 dilution

Lanes 2-5 : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

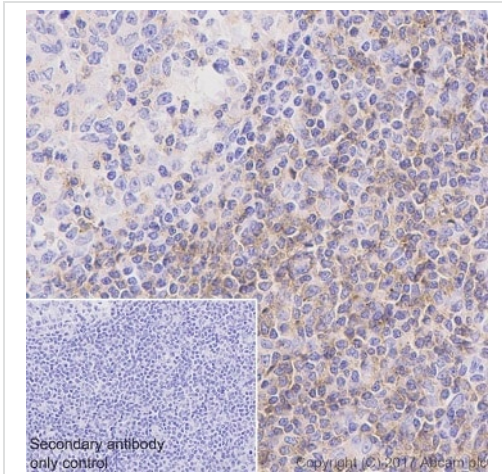
Developed using the ECL technique.

Predicted band size: 61 kDa

Observed band size: 60 kDa

Blocking/Dilution buffer: 5% NFDm/TBST.

Exposure time: Lanes 1 and 2: 3 minutes; Lanes 3,4 and 5: 8 seconds.

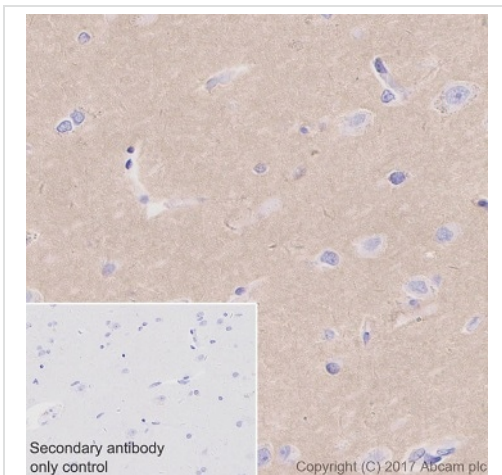


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fyn antibody [EPR19636] (ab184276)

Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling Fyn with ab184276 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining on T cells of human tonsil (PMID: 10523617). 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.

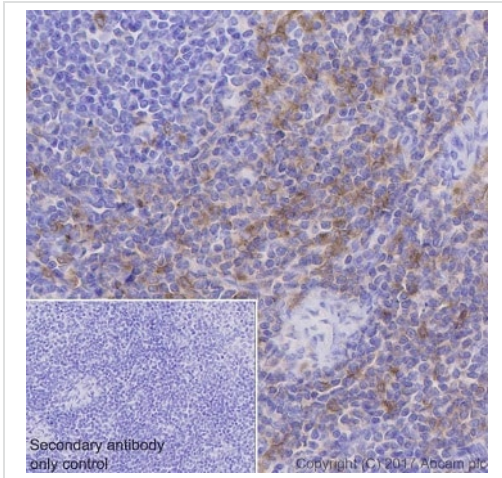


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fyn antibody [EPR19636] (ab184276)

Immunohistochemical analysis of paraffin-embedded human cerebral cortex tissue labeling Fyn with ab184276 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining on human cerebral cortex (PMID: 7544314). 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.

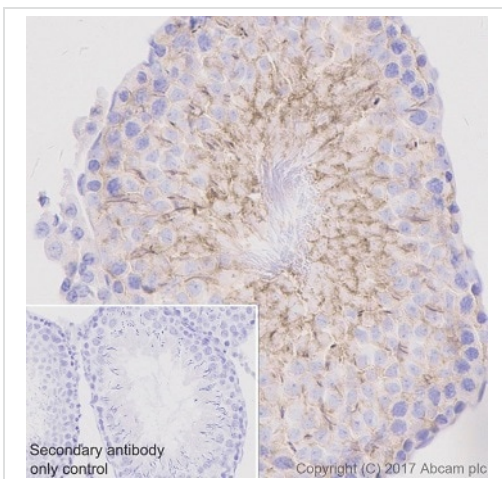


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fyn antibody [EPR19636] (ab184276)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling Fyn with ab184276 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining on T cells of mouse spleen (PMID: 7544314, PMID:10523617). 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.

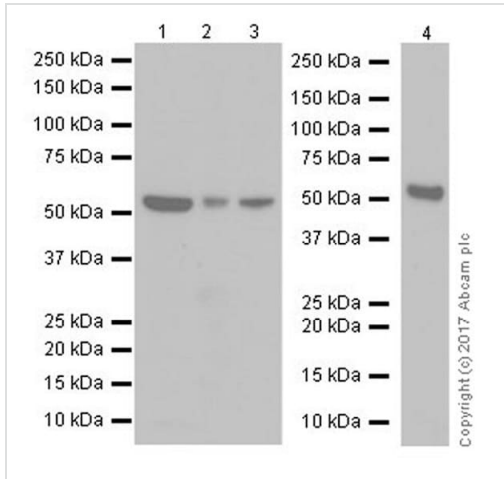


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Fyn antibody [EPR19636] (ab184276)

Immunohistochemical analysis of paraffin-embedded rat testis tissue labeling Fyn with ab184276 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Membranous staining on rat testis (PMID: 7544314). 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.



Western blot - Anti-Fyn antibody [EPR19636]
(ab184276)

All lanes : Anti-Fyn antibody [EPR19636] (ab184276) at 1/1000 dilution

Lane 1 : RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate

Lane 2 : PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysate

Lane 3 : NIH/3T3 (mouse embryo fibroblast cell line) whole cell lysate

Lane 4 : Rat spleen lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

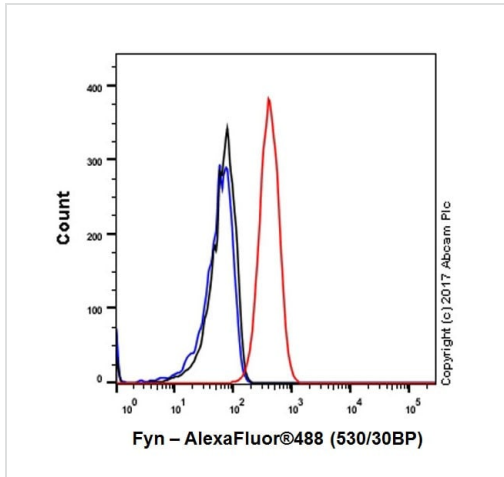
Developed using the ECL technique.

Predicted band size: 61 kDa

Observed band size: 60 kDa

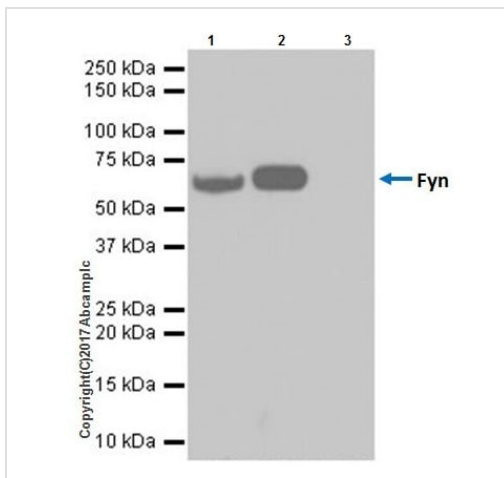
Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Flow Cytometry (Intracellular) - Anti-Fyn antibody [EPR19636] (ab184276)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) cell line labeling Fyn with ab184276 at 1/50 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (**ab172730**) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-Fyn antibody [EPR19636] (ab184276)

Fyn was immunoprecipitated from 0.35 mg of P0 mouse brain lysate with ab184276 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab184276 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

Lane 1: P0 mouse brain lysate 10 µg (Input).





Lane 2: ab184276 IP in P0 mouse brain lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab184276 in P0 mouse brain lysate.

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

Exposure time: 3 minutes.

Why choose a recombinant antibody?

| | |
|--|--|
|  <p>Research with confidence Consistent and reproducible results</p> |  <p>Long-term and scalable supply Recombinant technology</p> |
|  <p>Success from the first experiment Confirmed specificity</p> |  <p>Ethical standards compliant Animal-free production</p> |

Anti-Fyn antibody [EPR19636] (ab184276)

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