abcam

Product datasheet

Anti-GATA3 antibody [EPR16651] - ChIP Grade ab199428



יוכציטו RabMAb

★★★★★ 13 Abreviews 46 References 画像数 9

製品の概要

製品名 Anti-GATA3 antibody [EPR16651] - ChIP Grade

製品の詳細 Rabbit monoclonal [EPR16651] to GATA3 - ChIP Grade

由来種 Rabbit

特異性 IHC-P is suitable in Human, and may not be suitable for use in Mouse samples.

アプリケーション 適用あり: Flow Cyt (Intra), ChIP, WB, IHC-P, ICC/IF

種交差性 交差種: Mouse. Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: RAW 264.7 whole cell lysate (ab7187), SH-SY5Y cell extracts. EL4, HAP1 and Jurkat whole

> cell lysates. Mouse brain lysate. IHC-P: Human neuroblastoma and Human breast carcinoma tissue. ICC/IF: SH-SY5Y cells. Flow Cyt (intra): Jurkat cells. ChIP: Chromatin prepared from MCF7

特記事項 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 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

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab199428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)		1/500.
ChIP		Use 2 µg for 25 µg of chromatin.
WB		1/1000. Detects a band of approximately 48 kDa (predicted molecular weight: 48 kDa).
IHC-P	★★★★★ (8)	1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★★ (2)	1/250.

ターゲット情報

機能 Transcriptional activator which binds to the enhancer of the T-cell receptor alpha and delta genes.

Binds to the consensus sequence 5'-AGATAG-3'.

組織特異性 T-cells and endothelial cells.

関連疾患 Defects in GATA3 are the cause of hypoparathyroidism with sensorineural deafness and renal

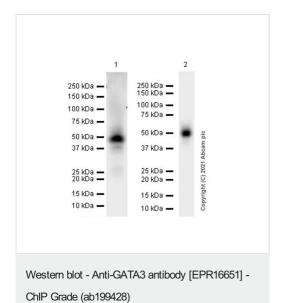
dysplasia (HDR) [MIM:146255]; also known as Barakat syndrome.

配列類似性 Contains 2 GATA-type zinc fingers.

細胞内局在 Nucleus.

製品の状態 There are 2 isoforms produced by alternative splicing.

画像



All lanes : Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428) at 1/1000 dilution

Lane 1 : Mouse brain lysate

Lane 2: EL4 (mouse lymphoma T lymphocyte) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

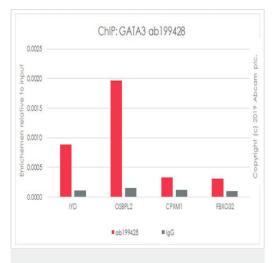
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 48 kDa **Observed band size:** 48 kDa

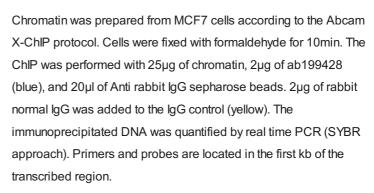
Exposure

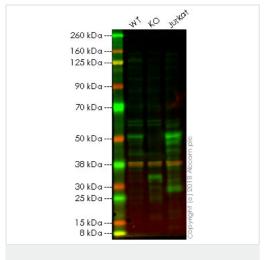
Lane1: 26 seconds

Lane 2: 15 seconds



ChIP - Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428)





Western blot - Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428)

All lanes : Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: GATA3 knockout HAP1 whole cell lysate

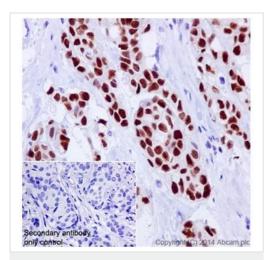
Lane 3: Jurkat whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 48 kDa

Lanes 1 - 3: Merged signal (red and green). Green - ab199428 observed at 48 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

ab199428 was shown to recognize GATA3 in wild-type HAP1 cells as signal was lost at the expected MW in GATA3 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and GATA3 knockout samples were subjected to SDS-PAGE. Ab199428 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



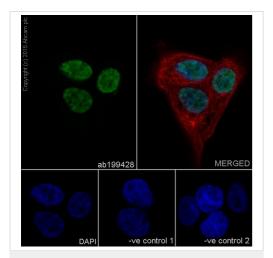
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GATA3 antibody

[EPR16651] - ChIP Grade (ab199428)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling GATA3 with ab199428 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on Human breast carcinoma tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary ab, secondary ab is Goat Anti-Rabbit lgG H&L (HRP) (ab97051).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

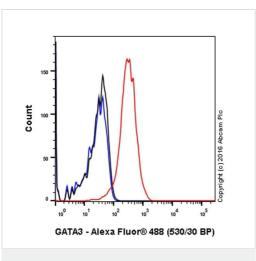


Immunocytochemistry/ Immunofluorescence - Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized SH-SY5Y cells (Human neuroblastoma from bone marrow cells) labeling GATA3 with ab199428 at 1/250 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/500 dilution (green). Nuclear staining on SH-SY5Y cell line is observed. The nuclear counterstain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (Alexa Fluor[®] 594 Goat anti-Mouse secondary) at 1/500 dilution (red).

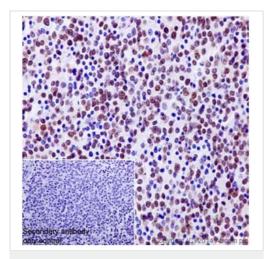
The negative controls are as follows:-

-ve control 1 - ab199428 at 1/250 dilution followed by <u>ab150120</u> (Alexa Fluor[®] 594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2. - <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor[®] 488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Flow Cytometry (Intracellular) - Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428)

Intracellular Flow Cytometry analysis of Jurkat cells labelling GATA3 with ab199428 at 1/500 (red). Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. An Alexa Fluorr® 488-conjugated goat anti-rabbit lgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal lgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.



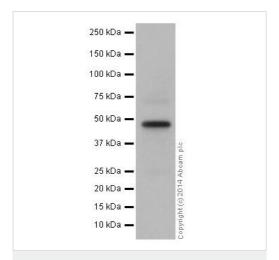
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GATA3 antibody

[EPR16651] - ChIP Grade (ab199428)

Immunohistochemical analysis of paraffin-embedded Human neuroblastoma tissue labeling GATA3 with ab199428 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on Human neuroblastoma tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary ab, secondary ab is Goat Anti-Rabbit IgG H&L (HRP) (ab97051).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428)

Anti-GATA3 antibody [EPR16651] - ChIP Grade (ab199428) at 1/1000 dilution + SH-SY5Y (Human neuroblastoma from bone marrow cells) cell extract at 10 μg

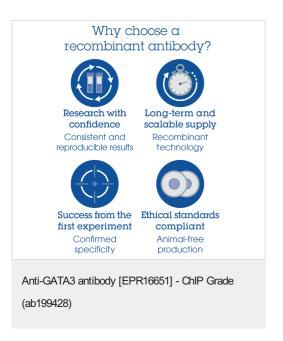
Secondary

Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 48 kDa Observed band size: 48 kDa

Exposure time: 1 minute

Blocking/dilution buffer: 5% NFDM/TBST.



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