abcam

Product datasheet

Anti-GRB2 antibody [Y237] ab32037



重组 RabMAb

★★★★★ 3 Abreviews 22 References 9 图像

概述

产品名称 Anti-GRB2抗体[Y237]

描述 兔单克隆抗体[Y237] to GRB2

宿主 Rabbit

经测试应用 适用于: WB, Flow Cyt (Intra), IP, IHC-P, ICC/IF

种属反应性 与反应: Mouse, Rat, Human

免疫原 Synthetic peptide within Human GRB2 aa 200 to the C-terminus (C terminal). The exact sequence

is proprietary.

阳性对照 WB: HeLa, HL-60, Caco-2, HEK-293, PC-12 and NIH/3T3 cell lysates. IHC-P: Human ovarian

carcinoma tissue. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa cells.

常规说明 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**.

性能

形式

存放说明 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA

纯度 Protein A purified

克隆 单克隆 克隆编号 Y237 同种型 ΙgG

The Abpromise guarantee Abpromise™承诺保证使用ab32037于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

Golgi apparatus.

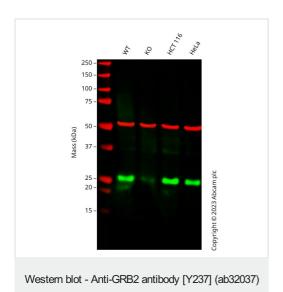
应用	Ab评论	说明
WB	★★★★☆ (2)	1/5000 - 1/10000. Detects a band of approximately 25 kDa (predicted molecular weight: 25 kDa).
Flow Cyt (Intra)		1/20. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IP		Use at an assay dependent concentration.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF	★★★ ☆☆ <u>(1)</u>	1/50 - 1/100.

功能	Adapter protein that provides a critical link between cell surface growth factor receptors and the Ras signaling pathway. Isoform GRB3-3 does not bind to phosphorylated epidermal growth factor receptor (EGFR) but inhibits EGF-induced transactivation of a RAS-responsive element. Isoform GRB3-3 acts as a dominant negative protein over GRB2 and by suppressing proliferative signals, may trigger active programmed cell death.
序列相似性	Belongs to the GRB2/sem-5/DRK family. Contains 1 SH2 domain. Contains 2 SH3 domains.
结 构域	The SH3 domains mediate interaction with RALGPS1 and SHB.

图片

细胞定位

靶标



All lanes: Anti-GRB2 antibody [Y237] (ab32037) at 1/5000 dilution

Lane 1: Wild-type A431 cell lysate

Lane 2: GRB2 knockout A431 cell lysate

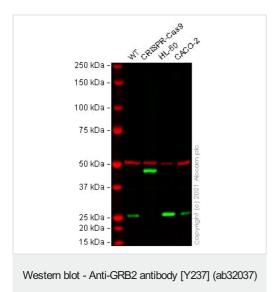
Lane 3 : HCT 116 cell lysate
Lane 4 : HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 25 kDa
Observed band size: 26 kDa

Anti-GRB2 antibody [Y237] (ab32037) staining at 1/5000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (ab7291) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32037 was shown to bind specifically to GRB2. A band was observed at 26 kDa in wild-type A431 cell lysates with a reduction in signal observed at this size in GRB2 heterozygous knockout cell line. To generate this image, wild-type and GRB2 heterozygous knockout A431 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 lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution.



All lanes: Anti-GRB2 antibody [Y237] (ab32037) at 1/5000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 2 : GRB2 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 3 : HL-60 (Human promyelocytic leukemia cell line) whole cell lysate

Lane 4 : Caco-2 (Human colorectal adenocarcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

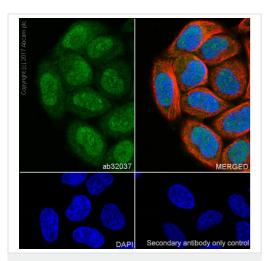
Performed under reducing conditions.

Predicted band size: 25 kDa
Observed band size: 25 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab32037 observed at 25 kDa. Red - loading control, <u>ab7291</u> (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

ab32037 was shown to react with GRB2 in western blot. The band observed in the CRISPR-Cas9 edited cell lysate lane above 25 kDa is likely to represent an insertion. This has not been investigated further and the functional properties of the gene product have not been determined. Cell lysates were subjected to SDS-PAGE.

Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab32037 and ab7291 (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 5000 dilution and a 1 in 20000 dilution respectively. Blots were incubated 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 in 20000 dilution for 1 h at room temperature before imaging.

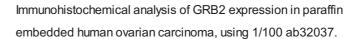


Immunocytochemistry/ Immunofluorescence - Anti-GRB2 antibody [Y237] (ab32037)

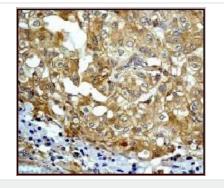
Immunocytochemistry/immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% TritonX-100 permeabilised HeLa (Human cervix adenocarcinoma epithelial cell) cells staining GRB2 with ab32037 at 1/200 dilution (green), followed by AlexaFluor®488 Goat anti-Rabbit secondary antibody (ab150077) at 1/1000 dilution (blue). Confocal image showing nuclear and weak cytoplasmic staining in HeLa cell line. Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) was used as a counterstain at 1/200 dilution (red). DAPI was used as a nuclear counterstain.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is [secondary antibody name] **ab150077** at 1/1000 dilution.

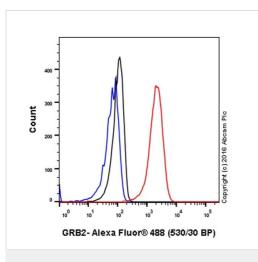
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



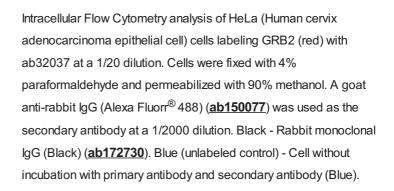
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

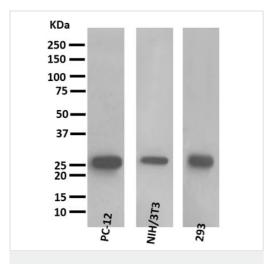


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-GRB2 antibody [Y237] (ab32037)



Flow Cytometry (Intracellular) - Anti-GRB2 antibody [Y237] (ab32037)





Western blot - Anti-GRB2 antibody [Y237] (ab32037)

All lanes : Anti-GRB2 antibody [Y237] (ab32037) at 1/10000 dilution

Lane 1: PC-12 (rat adrenal gland pheochromocytoma) cell lysate

Lane 2: NIH/3T3 (mouse embryonic fibroblast) cell lysate

Lane 3: HEK293 (human epithelial cell line from embryonic kidney)

cell lysate

Lysates/proteins at 10 µg per lane.

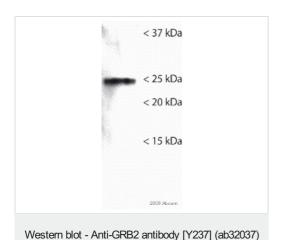
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/2000 dilution

Predicted band size: 25 kDa Observed band size: 25 kDa

Blocking/Dilution buffer: 5% NFDM/TBST

Exposure time: 3 minutes



Anti-GRB2 antibody [Y237] (ab32037) at 1/5000 dilution + Lysate prepared from human Mesothelial cell line MeT-5A at 4 μg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/2000 dilution

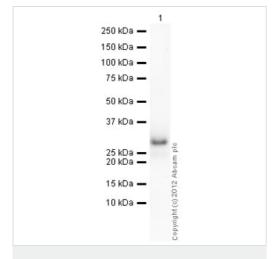
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 25 kDa **Observed band size:** 24 kDa

This image is a courtesy of Anonymous Abreview

Exposure time: 1 minute



Anti-GRB2 antibody [Y237] (ab32037) at 1/5000 dilution + Recombinant Human GRB2 protein (ab85917) at 0.01 μg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 25 kDa

Western blot - Anti-GRB2 antibody [Y237] (ab32037)

Exposure time: 2 minutes



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