


Anti-beta Catenin antibody [E247] - ChIP Grade ab32572

KO 評価済 リコンビナント RabMAb

★★★★★ **75 Abreviews** **944 References** 画像数 30

製品の概要

| | |
|--------------|---|
| 製品名 | Anti-beta Catenin antibody [E247] - ChIP Grade |
| 製品の詳細 | Rabbit monoclonal [E247] to beta Catenin - ChIP Grade |
| 由来種 | Rabbit |
| 特異性 | <p>This antibody is not suitable for ICC testing in mouse and rat species.</p> <p>Our inhouse testing indicated that this antibody does not work in Raw264.7 cell line in western blot. We have an alternative antibody ab68183 detecting weak band in lower expressor Raw264.7.</p> |
| アプリケーション | <p>適用あり: IHC-P, WB, ICC/IF, IP, ChIP</p> <p>適用なし: Flow Cyt</p> |
| 種交差性 | <p>交差種: Mouse, Rat, Human</p> <p>交差が予測される動物種: Sheep, Hamster, Cow, Macaque monkey, African green monkey</p> <p></p> |
| 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: A431, HeLa, wild-type HAP1, HCT 116 and Wild-type MCF7 cell lysates. ICC/IF: A431 and wild-type HAP1 cells. SW480 and SK-N-SH cells. IHC-P: Human lung adenocarcinoma, kidney adenocarcinoma, colon adenocarcinoma, cervical carcinoma, breast carcinoma and papillary carcinoma of thyroid gland tissue, Rat liver and pancreas, Mouse liver and pancreas; IP: A431 whole cell lysate and 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. Avoid freeze / thaw cycle. |
| バッファー | pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 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 | E247 |
| 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ab32572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 |
|----------|------------|--|
| IHC-P | ★★★★★ (25) | 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. |
| WB | ★★★★★ (28) | 1/5000 - 1/10000. Detects a band of approximately 92 kDa (predicted molecular weight: 86 kDa). We recommend Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) . |
| ICC/IF | ★★★★★ (13) | 1/250. We recommend Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody . |
| IP | | 1/30. |
| ChIP | ★☆☆☆☆ (1) | Use at an assay dependent concentration. |

追加情報 Is unsuitable for Flow Cyt.

ターゲット情報

| | |
|-------|---|
| 機能 | Key downstream component of the canonical Wnt signaling pathway. In the absence of Wnt, forms a complex with AXIN1, AXIN2, APC, CSNK1A1 and GSK3B that promotes phosphorylation on N-terminal Ser and Thr residues and ubiquitination of CTNNB1 via BTRC and its subsequent degradation by the proteasome. In the presence of Wnt ligand, CTNNB1 is not ubiquitinated and accumulates in the nucleus, where it acts as a coactivator for transcription factors of the TCF/LEF family, leading to activate Wnt responsive genes. Involved in the regulation of cell adhesion. The majority of beta-catenin is localized to the cell membrane and is part of E-cadherin/catenin adhesion complexes which are proposed to couple cadherins to the actin cytoskeleton. |
| 組織特異性 | Expressed in several hair follicle cell types: basal and peripheral matrix cells, and cells of the outer and inner root sheaths. Expressed in colon. |

関連疾患

Defects in CTNNB1 are associated with colorectal cancer (CRC) [MIM:114500].
Note=Activating mutations in CTNNB1 have oncogenic activity resulting in tumor development. Somatic mutations are found in various tumor types, including colon cancers, ovarian and prostate carcinomas, hepatoblastoma (HB), hepatocellular carcinoma (HCC). HBs are malignant embryonal tumors mainly affecting young children in the first three years of life.
Defects in CTNNB1 are a cause of pilomatixoma (PTR) [MIM:132600]; a common benign skin tumor.
Defects in CTNNB1 are a cause of medulloblastoma (MDB) [MIM:155255]. MDB is a malignant, invasive embryonal tumor of the cerebellum with a preferential manifestation in children.
Defects in CTNNB1 are a cause of susceptibility to ovarian cancer (OC) [MIM:167000]. Ovarian cancer common malignancy originating from ovarian tissue. Although many histologic types of ovarian neoplasms have been described, epithelial ovarian carcinoma is the most common form. Ovarian cancers are often asymptomatic and the recognized signs and symptoms, even of late-stage disease, are vague. Consequently, most patients are diagnosed with advanced disease.
Note=A chromosomal aberration involving CTNNB1 is found in salivary gland pleiomorphic adenomas, the most common benign epithelial tumors of the salivary gland. Translocation t(3;8) (p21;q12) with PLAG1.

配列類似性

Belongs to the beta-catenin family.
Contains 12 ARM repeats.

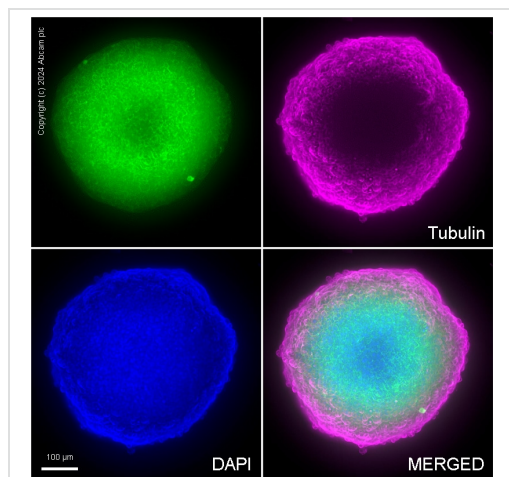
翻訳後修飾

Phosphorylation by GSK3B requires prior phosphorylation of Ser-45 by another kinase. Phosphorylation proceeds then from Thr-41 to Ser-37 and Ser-33.
EGF stimulates tyrosine phosphorylation. Phosphorylation on Tyr-654 decreases CDH1 binding and enhances TBP binding.
Ubiquitinated by the SCF(BTRC) E3 ligase complex when phosphorylated by GSK3B, leading to its degradation. Ubiquitinated by a E3 ubiquitin ligase complex containing UBE2D1, SIAH1, CACYBP/SIP, SKP1, APC and TBL1X, leading to its subsequent proteasomal degradation.

細胞内局在

Cytoplasm. Nucleus. Cytoplasm > cytoskeleton. Cell junction > adherens junction. Cell junction. Cell membrane. Cytoplasmic when it is unstabilized (high level of phosphorylation) or bound to CDH1. Translocates to the nucleus when it is stabilized (low level of phosphorylation). Interaction with GLIS2 and MUC1 promotes nuclear translocation. Interaction with EMD inhibits nuclear localization.

画像

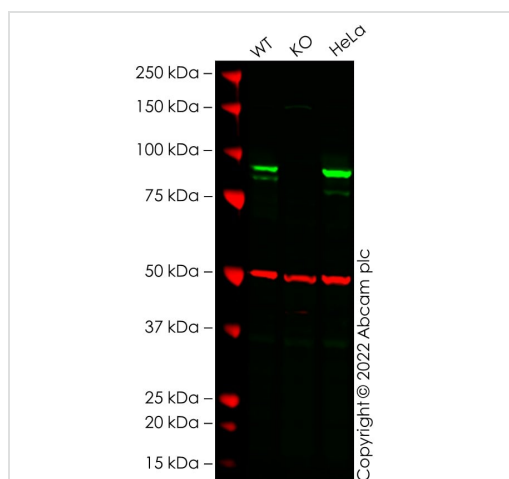


Immunocytochemistry - Anti-beta Catenin antibody
[E247] - ChIP Grade (ab32572)

ab32572 staining of β -catenin in a HCT116 cell spheroid. The cells were fixed with 4% formaldehyde (10 min), permeabilised with 0.5% Triton X-100 for 1h and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween overnight at room temperature. The spheroids were then incubated overnight at room temperature with ab32572 at 2 μ g/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin at 2 μ g/ml. DAPI was used as nuclear counterstain (shown in blue). As secondary antibodies **ab150081** Goat anti-Rabbit (Alexa Fluor[®] 488) (shown in green) and **ab150120** Goat anti-Mouse (Alexa Fluor[®] 594) (shown in magenta) were used, incubated overnight at room temperature. All permeabilization, blocking and antibody incubation steps were performed using a rotary shaker.

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.

The antibody ab32572 also worked using 100% methanol (5 min).



Western blot - Anti-beta Catenin antibody [E247] -
ChIP Grade (ab32572)

All lanes : Anti-beta Catenin antibody [E247] - ChIP Grade
(ab32572) at 1/5000 dilution

Lane 1 : Wild-type MCF7 cell lysate

Lane 2 : CTNNB1 knockout MCF7 cell lysate

Lane 3 : HeLa cell lysate

Lysates/proteins at 20 μ g per lane.

Secondary

All lanes : Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse
IgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 86 kDa

Observed band size: 85/90 kDa

False colour image of Western blot: Anti-beta Catenin antibody [E247] - ChIP Grade 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, ab32572 was shown to bind specifically to beta Catenin. A band was observed at 85/90 kDa in wild-type MCF7 cell lysates with no signal observed at

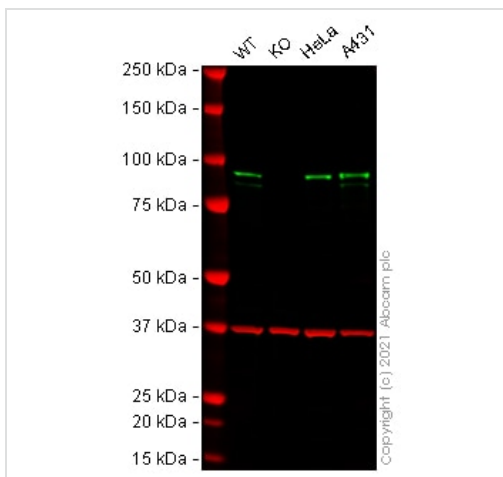
this size in CTNNB1 knockout cell line [ab286762](#). To generate this image, wild-type and CTNNB1 knockout MCF7 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before 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.

| Normal tissue samples | | | | | | Malignant tissue samples | | | | | |
|-----------------------|--------------------------|----------------|---|-----------------------------|---|--------------------------------------|---|----------------------------|---|--------------------------------|---|
| Human cardiac muscle | ✓ (Interrelated discs ✓) | Human placenta | ✓ | Human skeletal muscle | ✓ | Clear cell carcinoma of human kidney | ✓ | Human glioma | ✓ | Human hepatocellular carcinoma | ✓ |
| Human cerebrum | ✓ | Human skin | ✓ | Human breast carcinoma | ✓ | Human bladder cancer | ✓ | Human lung carcinoma | ✓ | Human ovarian carcinoma | ✓ |
| Human colon | ✓ | Human spleen | ✓ | Human cervical carcinoma | ✓ | Human colon carcinoma | ✓ | Human pancreatic carcinoma | ✓ | Human prostatic hyperplasia | ✓ |
| Human endometrium | ✓ | Human stomach | ✓ | Human endometrial carcinoma | ✓ | Human gastric adenocarcinoma | ✓ | Human thyroid carcinoma | ✓ | | |
| Human kidney | ✓ | Human testis | ✓ | | | | | | | | |
| Human liver | ✗ | Human tonsil | ✓ | | | | | | | | |
| Human lung | ✓ | | | | | | | | | | |
| Human mammary gland | ✓ | | | | | | | | | | |
| Human pancreas | ✓ | | | | | | | | | | |

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

Tissue Microarrays stained for Anti-beta Catenin antibody [E247] - ChIP Grade using ab32572 in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negative (cross mark) staining per sample type tested. The section was incubated with ab32572 for 30 mins at room temperature followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Western blot - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

All lanes : Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572) at 1/5000 dilution

Lane 1 : Wild-type HepG2 cell lysate

Lane 2 : CTNNB1 knockout HepG2 cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : A431 cell lysate

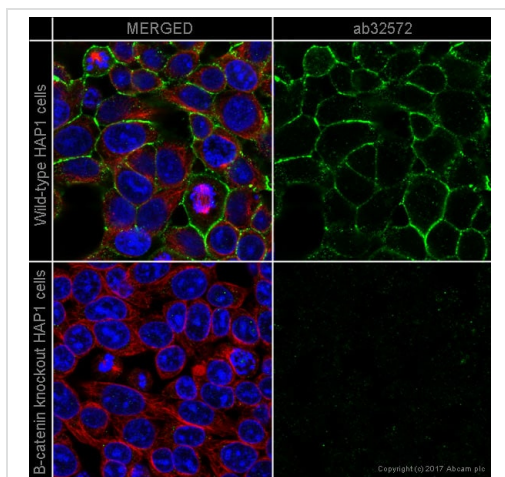
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 86 kDa

Observed band size: 85 kDa

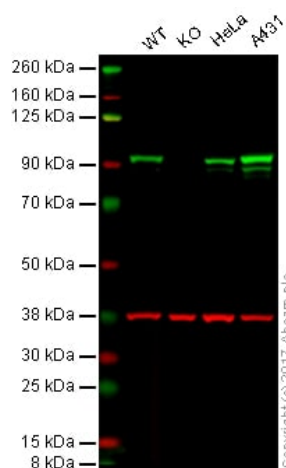
False colour image of Western blot: Anti-beta Catenin antibody [E247] - ChIP Grade staining at 1/5000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] ([ab8245](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32572 was shown to bind specifically to beta Catenin. A band was observed at 85 kDa in wild-type HepG2 cell lysates with no signal observed at this size in CTNNB1 knockout cell line ([ab277911](#)). To generate this image, wild-type and CTNNB1 knockout HepG2 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.



Immunocytochemistry/ Immunofluorescence - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 staining in CTNNB1 (beta Catenin) wild-type HAP1 cells (top panel) and in CTNNB1 (β-catenin) knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab32572 at 1/250 dilution and [ab195889](#) at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with **Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed (ab150081) secondary antibody** at 2 µg/ml (shown in green). Nuclear DNA was labeled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Western blot - Anti-beta Catenin antibody [E247]
(ab32572)

Lane 4 : Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#)) at 1/5000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate

Lane 2 : CTNNB1 (β-catenin) knockout HAP1 whole cell lysate

Lane 3 : HeLa whole cell lysate

Lane 4 : A431 whole cell lysate

Lysates/proteins at 20 µg per lane.

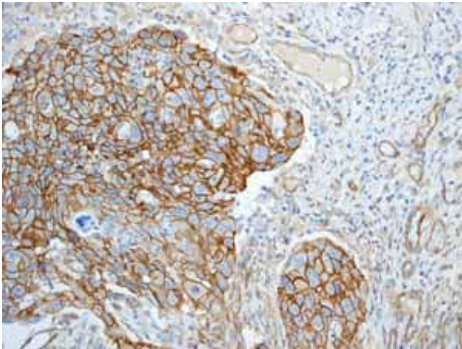
Predicted band size: 86 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab32572 observed at 90 kDa. Red - loading control, [ab8245](#), observed at 37 kDa.

ab32572 was shown to specifically react with CTNNB1 (β-catenin) in wild type HAP1 cells. No band was observed when CTNNB1 (β-catenin) knockout samples were used. Wild-type and CTNNB1 (β-catenin) knockout samples were subjected to SDS-PAGE.

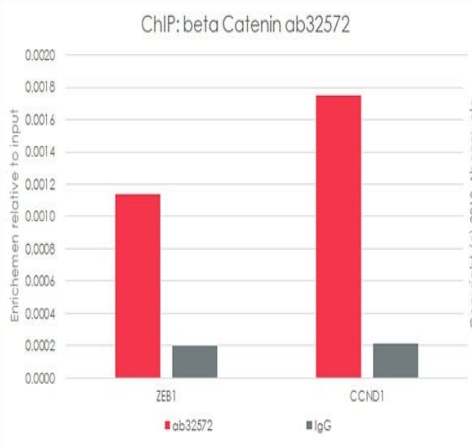
ab32572 and [ab8245](#) (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at a 1/5000 dilution and 1/10000 dilution respectively. Blots were developed with **Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ([ab216773](#))** and **Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ([ab216776](#))**

secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

Immunohistochemical analysis of human cervical carcinoma tissue staining beta Catenin with ab32572 at 1/500 dilution. Heat mediated antigen retrieval was performed with citrate buffer (pH 6)



ChIP - Anti-beta Catenin antibody [E247] (ab32572)

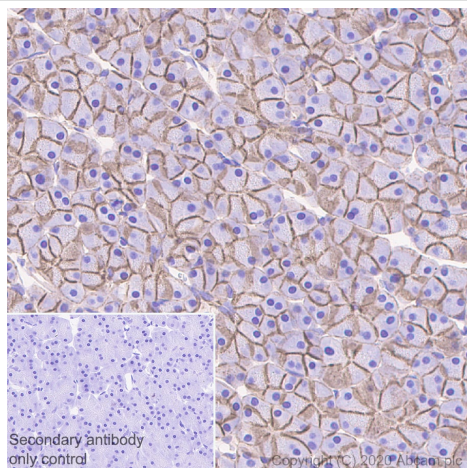
Chromatin was prepared from HCT 116 cells according to the Abcam Dual-X-ChIP protocol*. Cells were fixed with 1.5 mM EGS for 30 mins and then formaldehyde for 10 min.

The ChIP was performed with 25 µg of chromatin, 5 µg of ab32572 (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 from paper PMID: 28625518

*<http://www.abcam.com/resources?>

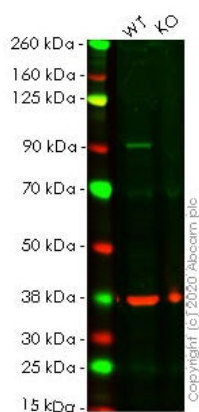
keywords=X%20ChIP%20protocol



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

Immunohistochemical analysis of paraffin-embedded rat liver pancreas labeling beta Catenin with ab32572 at 1/1000 dilution, followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Membranous staining on rat pancreas. The section was incubated with ab32572 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)).



Western blot - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

All lanes : Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572) at 1/500 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : CTNNB1 knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

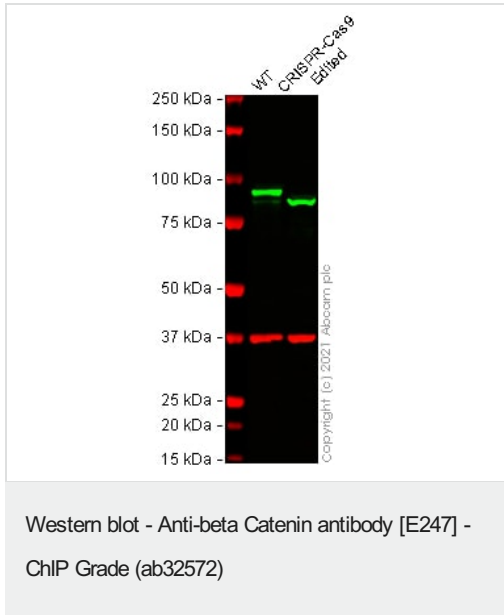
Predicted band size: 86 kDa

Observed band size: 86 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab32572 observed at 86 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) observed at 37 kDa.

ab32572 was shown to react with beta Catenin in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line [ab255352](#) (knockout cell lysate [ab263756](#)) was used. Wild-type HeLa and CTNNB1 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab32572 and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) overnight at 4°C at a 1 in 500 dilution and a 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 20000 dilution for 1 hour at room temperature before imaging.



All lanes : Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572) at 1/5000 dilution

Lane 1 : Wild-type HCT 116 cell lysate

Lane 2 : CTNNB1 CRISPR-Cas9 edited HCT 116 cell lysate

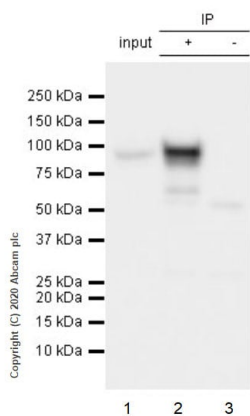
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 86 kDa

Observed band size: 95 kDa

False colour image of Western blot: Anti-beta Catenin antibody [E247] - ChIP Grade staining at 1/5000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] ([ab8245](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab32572 was shown to bind specifically to beta Catenin. A band was observed at 95 kDa in wild-type HCT 116 cell lysates with no signal observed at this size in CTNNB1 CRISPR-Cas9 edited cell line [ab273712](#) (CRISPR-Cas9 edited cell lysate [ab275247](#)). The band observed in the CRISPR-Cas9 edited lysate lane below 95 kDa is likely to represent a truncated form of beta Catenin. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and CTNNB1 CRISPR-Cas9 edited HCT 116 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5 % 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.



Immunoprecipitation - Anti-beta Catenin antibody
[E247] - ChIP Grade (ab32572)

beta Catenin was immunoprecipitated from 0.35 mg mouse brain lysate with ab32572 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab32572 1/1000 dilution (1.962 µg/ml). VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) was used as the secondary antibody at 1/1000 dilution.

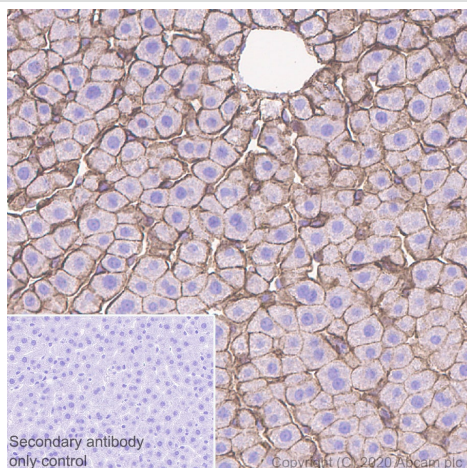
Lane 1: Mouse brain tissue lysate 10 µg

Lane 2: Mouse brain tissue lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab32572 in mouse brain lysate.

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

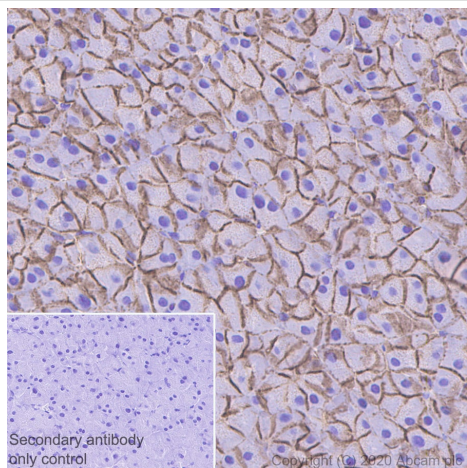
Exposure time: 1 seconds



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody
[E247] - ChIP Grade (ab32572)

Immunohistochemical analysis of paraffin-embedded rat liver tissue labeling beta Catenin with ab32572 at 1/1000 dilution, followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Membranous staining on rat liver. The section was incubated with ab32572 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

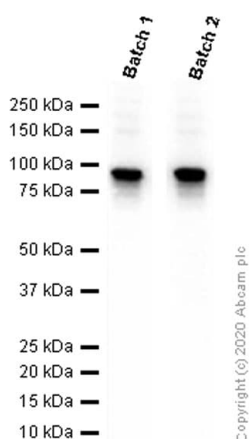
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

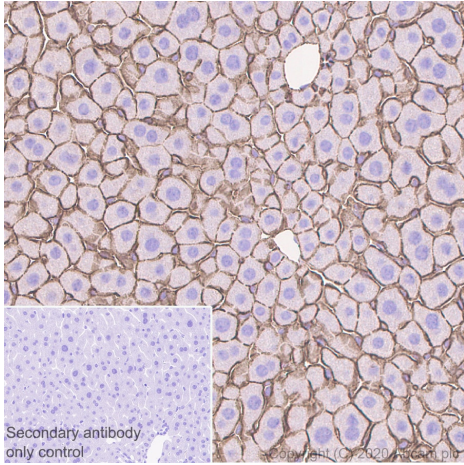
Immunohistochemical analysis of paraffin-embedded mouse pancreas tissue labeling beta Catenin with ab32572 at 1/1000 dilution, followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Membranous staining on mouse pancreas. The section was incubated with ab32572 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)).



Western blot - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

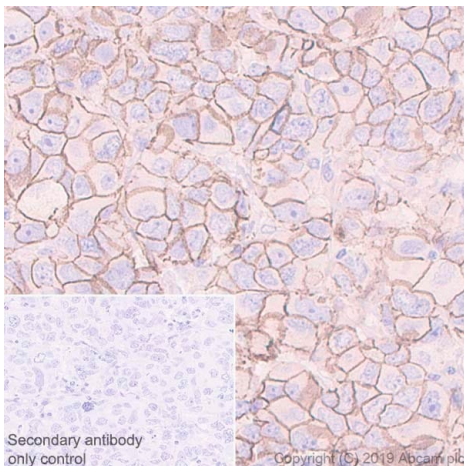
Different batches of ab32572 were tested on A431 (Human epidermoid carcinoma epithelial cell) lysate at 2.0 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 92 kDa.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labeling beta Catenin with ab32572 at 1/1000 dilution, followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)). Membranous staining on mouse liver. The section was incubated with ab32572 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

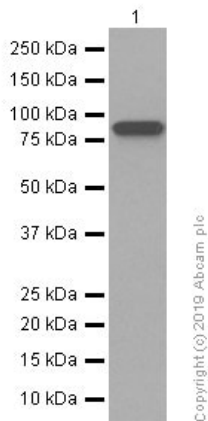
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB ([ab209101](#)).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572)

Immunohistochemical analysis of paraffin-embedded Human breast carcinoma tissue labeling beta Catenin with ab32572, followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP). Membranous staining on human breast carcinoma. The section was incubated with [ab229902](#) for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval using [ab93684](#) (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP).



Western blot - Anti-beta Catenin antibody [E247]
(ab32572)

Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572) at 1/10000 dilution + A431 (Human epidermoid carcinoma epithelial cell) whole cell lysates at 15 µg

Secondary

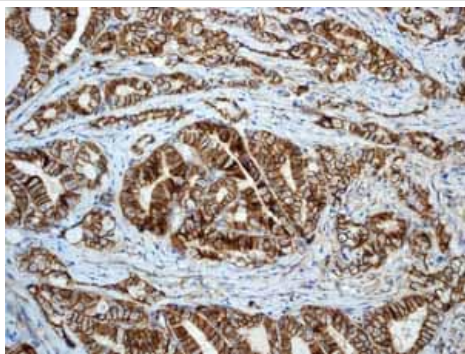
Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/20000 dilution

Predicted band size: 86 kDa

Observed band size: 92 kDa

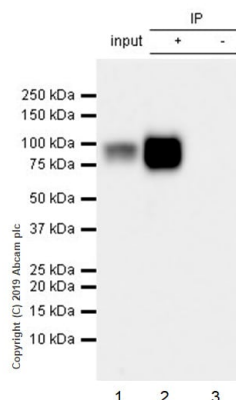
Blocking/Diluting buffer and concentration: 5% NFDM/TBST.

Exposure time: 180 seconds



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

Immunohistochemical analysis of human papillary carcinoma tissue staining beta Catenin with ab32572 at 1/500 dilution. Heat mediated antigen retrieval was performed with citrate buffer (pH 6).



Immunoprecipitation - Anti-beta Catenin antibody [E247] (ab32572)

beta Catenin was immunoprecipitated from 0.35 mg A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate with ab32572 at 1/50 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab32572 1/500 dilution (2 µg/ml). VeriBlot for IP Detection Reagent (HRP) ([ab131366](#)) was used as the secondary antibody at 1/1000 dilution.

Lane 1: A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate 10µg

Lane 2: ab32572 IP in A431 whole cell lysate

Lane 3: Rabbit monoclonal IgG ([ab172730](#)) instead of ab32572 in A431 whole cell lysate.

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

Exposure time: 3 seconds

Lane 1 : Anti-beta Catenin antibody [E247] - ChIP Grade
(ab32572) at 1/500 dilution (2µg/ml)

Lane 1 : A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate at 10 µg

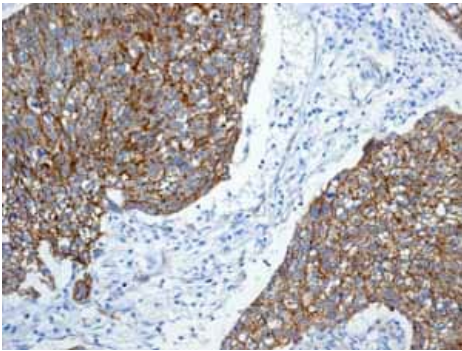
Lane 2 : ab32572 IP in A431 whole cell lysate

Lane 3 : Rabbit monoclonal IgG (**ab172730**) instead of ab32572 in A431 whole cell lysate

Secondary

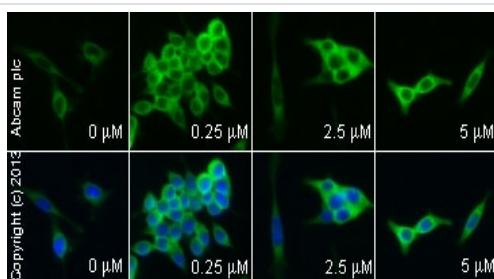
Lane 1 : VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at 1/1000 dilution

Observed band size: 90 kDa



Immunohistochemical analysis of human lung adenocarcinoma tissue staining beta Catenin with ab32572 at 1/500 dilution. Heat mediated antigen retrieval was performed with citrate buffer (pH 6).

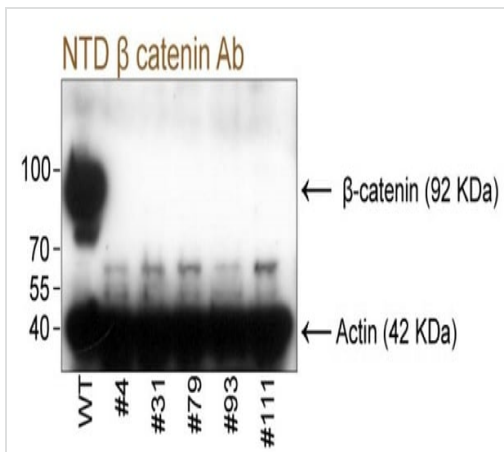
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)



Immunocytochemistry/ Immunofluorescence - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 staining beta Catenin in SW480 (Human colorectal adenocarcinoma cell line) cells treated with BIO (**ab120891**), by ICC/IF. Increase of beta Catenin expression correlates with increased concentration of BIO, as described in literature. The cells were incubated at 37°C for 48h in media containing different concentrations of **ab120891** (BIO) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab32572 (1/200) dilution was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A **Goat Anti-Rabbit IgG H&L (DyLight® 488) preadsorbed (ab96899) secondary antibody** at

1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.



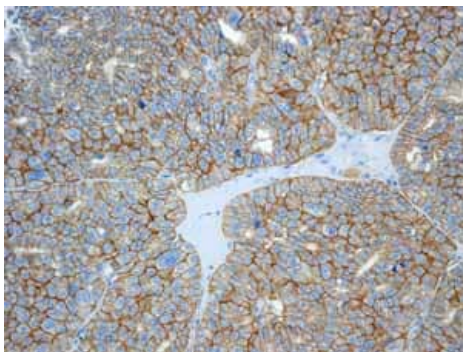
Western blot - Anti-beta Catenin antibody [E247]
(ab32572)

Olsen et al PLoS One. 2014 Dec 23;9(12):e115496. doi: 10.1371/journal.pone.0115496. eCollection 2014. Fig 3. Reproduced under the Creative Commons license <http://creativecommons.org/licenses/by/4.0/>

WB analysis of total cell extracts from WT and gene disrupted cells using ab32572 at a 1/5000 dilution together with anti actin antibody. The position and full length β -catenin, truncated β -catenin and actin bands are indicated. For wild type cells 5 μ g of TP and for the gene disrupted clones 30 μ g of TP was applied for each lane.

Cells were lysed in RIPA buffer containing protease inhibitor and phosphatase inhibitor tablets. Cell lysates were cleared by centrifugation and protein concentration 5–30 μ g of total protein in SDS sample buffer was loaded per lane and separated.

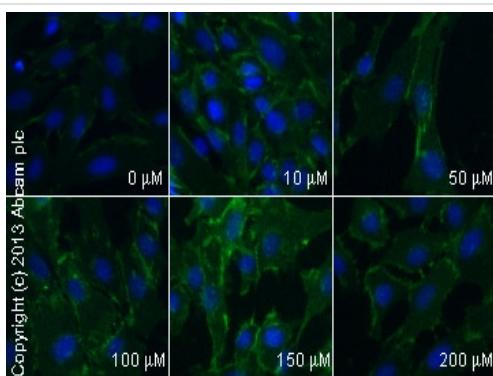
Secondary antibody was a donkey anti-rabbit IgG-HRP used at a 1:5000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 showing positive staining in human kidney carcinoma tissue.

Immunohistochemical analysis of human kidney carcinoma tissue staining beta Catenin with ab32572 at 1/500 dilution. Heat mediated antigen retrieval was performed with citrate buffer (pH 6).



Immunocytochemistry/ Immunofluorescence - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 staining beta Catenin in SK-N-SH (Human neuroblastoma cell line) cells treated with olanzapine (**ab120736**), by ICC/IF.

Increase in expression of beta Catenin correlates with increased concentration of olanzapine, as described in literature.

The cells were incubated at 37°C for 24h in media containing different concentrations of **ab120736** (olanzapine) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab32572 (1/200 dilution) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A **Goat Anti-**

Rabbit IgG H&L (DyLight® 488) preadsorbed (ab96899)

secondary antibody at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.



Western blot - Anti-beta Catenin antibody [E247] (ab32572)

This image is courtesy of an anonymous Abreview

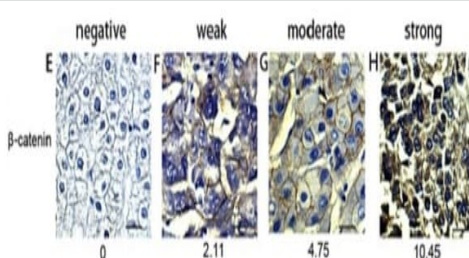
Anti-beta Catenin antibody [E247] - ChIP Grade (ab32572) at 1/5000 dilution + U-2 OS (Human bone osteosarcoma epithelial cell line) whole cell lysate

Performed under reducing conditions.

Predicted band size: 86 kDa

Observed band size: 90 kDa

Western blot image of ab32572 staining whole cell lysate of U-2 OS (Human bone osteosarcoma epithelial cell line) cells. The gel was blocked with 5% milk for 1 hour at 21°C. The primary antibody was diluted 1/5000 and incubated for 12 hours at 4°C. An HRP conjugated swine anti-rabbit antibody was used as the secondary.



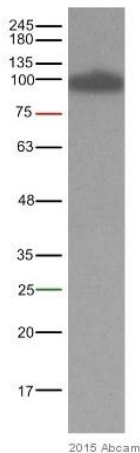
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

Jin et al PLoS One. 2015 Aug 7;10(8):e0133770. doi: 10.1371/journal.pone.0133770. eCollection 2015. Fig 2. Reproduced under the Creative Commons license <http://creativecommons.org/licenses/by/4.0/>

Different expression level of beta Catenin in HCTs (hepatocellular carcinoma tissues) and PLTs (para-cancerous liver tissues).

The HCTs, PLTs were paraffin-embedded and cut into sections with 5 µm-thickness for hematoxylin-eosin and immunohistochemistry (IHC) analysis. ab32572 was used at a dilution of 1:400. The second antibody was a biotinylated IgG to incubate 40 minutes at 37°C. Finally, the tissue slices were visualized by the 3, 3'-diaminobenzidine solution and counterstained with hematoxylin. Substitution of the primary antibody with phosphate-buffered saline was served as a control for IHC.

The beta Catenin with negative, weak, moderate and strong staining activity was respectively detected in HCTs (E-H) and PLTs (M-P). Section E shown above, for full image please see original paper.



Western blot - Anti-beta Catenin antibody [E247] -
ChIP Grade (ab32572)

Brain (mouse) whole tissue lysate at 20 µg

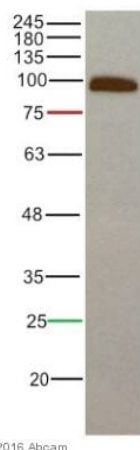
Performed under reducing conditions.

Predicted band size: 86 kDa

Brain (Mouse).

Blocking with 5% milk. The blocking time os 1 hour at 22°C.

Detected by ECL. Exposure time: 5 seconds.



Western blot - Anti-beta Catenin antibody [E247] -
ChIP Grade (ab32572)

Rat Pericytes whole cell lysate

Performed under reducing conditions.

Predicted band size: 86 kDa

Rat Pericyte cells.

Blocking and dilution buffer and concentration: 5% Milk. Blocking
time 1 hour and temperature at 22°C

Exposure time: 10 seconds

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Anti-beta Catenin antibody [E247] - ChIP Grade
(ab32572)

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