

Product datasheet

Anti-beta Catenin antibody [E247] ab32572

KO VALIDATED Recombinant RabMAB[®]

★★★★★ 61 Abreviews 199 References 17 Images

Overview

Product name	Anti-beta Catenin antibody [E247]
Description	Rabbit monoclonal [E247] to beta Catenin
Host species	Rabbit
Tested applications	Suitable for: IHC-Fr, IHC-P, WB, ICC/IF, IP, ChIP Unsuitable for: Flow Cyt
Species reactivity	Reacts with: Mouse, Rat, Sheep, Hamster, Cow, Human, Macaque monkey, African green monkey
Immunogen	Synthetic peptide within Human beta Catenin aa 1-100 (N terminal). The exact sequence is proprietary.
Positive control	WB: A431, HeLa and wild-type HAP1 cell lysate. 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.
General notes	A trial size is available to purchase for this antibody.

Our RabMAB[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to [RabMab[®] patents](#)

This product is a recombinant rabbit monoclonal antibody.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: PBS, 50% Glycerol, 0.05% BSA
Clonality	Monoclonal
Clone number	E247

Isotype IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab32572** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-Fr	★★★★★	1/200.
IHC-P	★★★★★	1/500.
WB	★★★★★	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	★★★★★	1/250. We recommend Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody .
IP		1/100.
ChIP		Use at an assay dependent concentration. PubMed: 28923827

Application notes Is unsuitable for Flow Cyt.

Target

Function 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.

Tissue specificity Expressed in several hair follicle cell types: basal and peripheral matrix cells, and cells of the outer and inner root sheaths. Expressed in colon.

Involvement in disease 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.

Sequence similarities

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

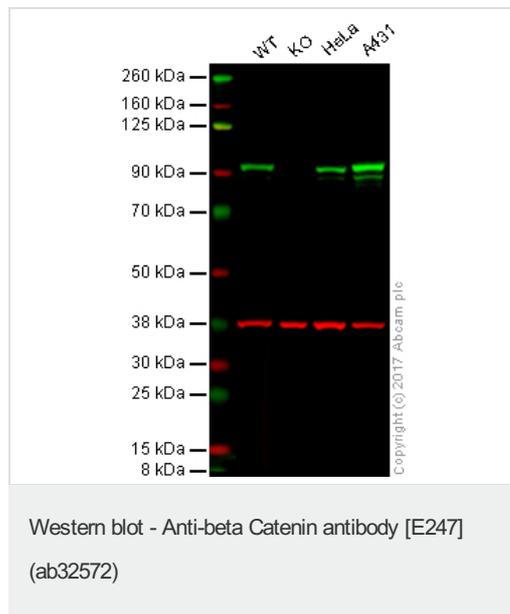
Post-translational modifications

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.

Cellular localization

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.

Images



Lane 1: Wild type HAP1 whole cell lysate (20 µg)

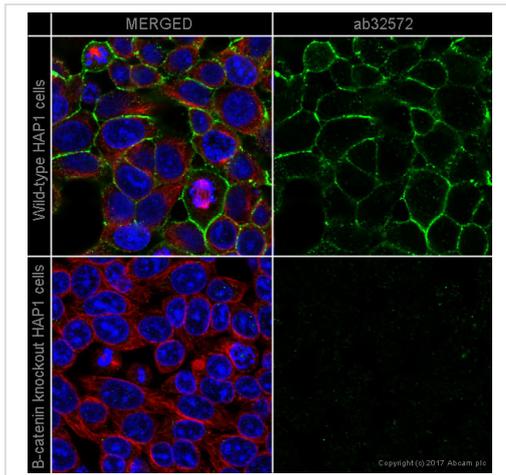
Lane 2: CTNNB1 (β-catenin) knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: A431 whole cell lysate (20 µg)

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

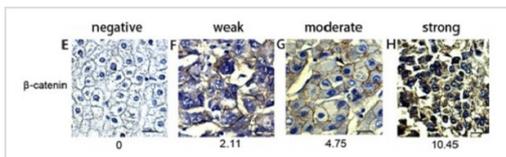
ab32572 was shown to specifically react with CTNNB1 (β-catenin) in wild type HAP1 cells. No band was observed when CTNNB1 (β-catenin) knockout samples were used. Wild-type and CTNNB1 (β-catenin) knockout samples were subjected to SDS-PAGE. ab32572 and ab8245 (Mouse anti-GAPDH loading control) were incubated 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.



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 μ g/ml (shown in green). Nuclear DNA was labeled in blue with DAPI.

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

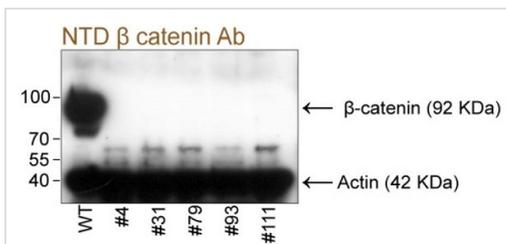


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

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 μ m-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] (ab32572)

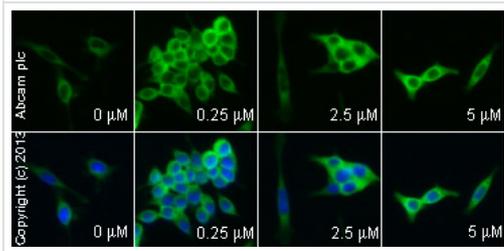
WB analysis of total cell extracts from WT and gene disrupted cells using ab32572 at a 1/5000 dilution together with anti actin antibody. The position and full length β -catenin, truncated β -catenin and actin bands are indicated. For wild type cells 5 μ g of TP and for the gene disrupted clones 30 μ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

Olsen et al PLoS One. 2014 Dec 23;9(12):e115496. doi: 10.1371/journal.pone.0115496. eCollection 2014. Fig 3.

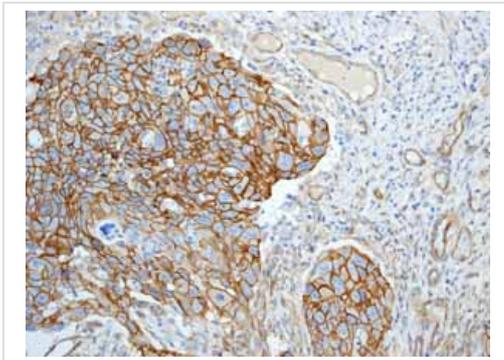
1:5000 dilution.



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

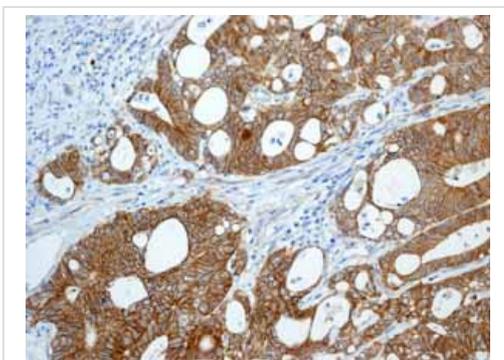
ab32572 staining beta Catenin in SW480 (Human colorectal adenocarcinoma cell line) cells treated with BIO ([ab120891](#)), by ICC/IF. Increase of beta Catenin expression correlates with increased concentration of BIO, as described in literature.

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.



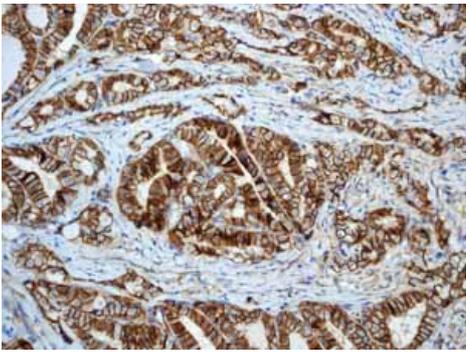
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 showing positive staining in human cervical carcinoma tissue.



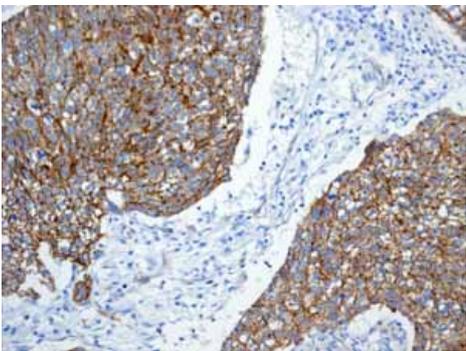
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 showing positive staining in human breast carcinoma tissue.



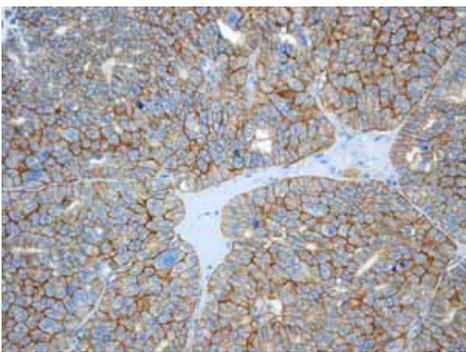
ab32572 showing positive staining in human papillary carcinoma of thyroid gland tissue.

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



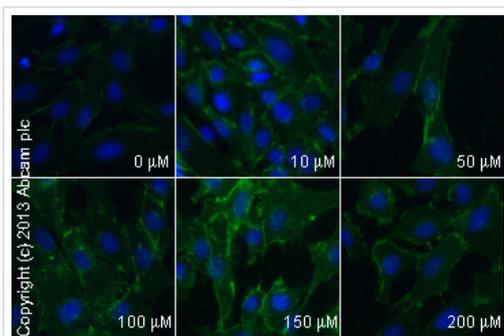
ab32572 showing positive staining in human lung adenocarcinoma tissue.

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



ab32572 showing positive staining in human kidney carcinoma tissue.

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

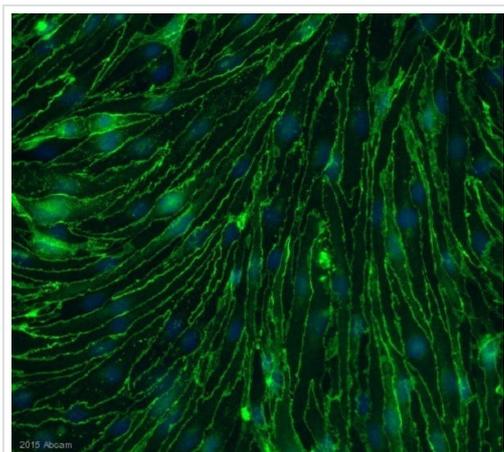


Immunocytochemistry/ Immunofluorescence - Anti-beta Catenin antibody [E247] (ab32572)

ab32572 staining beta Catenin in 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.

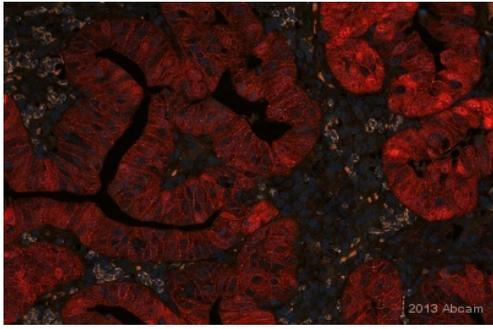


Immunocytochemistry/ Immunofluorescence - Anti-beta Catenin antibody [E247] (ab32572)

This image is courtesy of an anonymous Abreview.

ab32572 staining beta Catenin in the bEnd.5 murine cell line by ICC/IF (Immunocytochemistry/immunofluorescence).

Cells were fixed with paraformaldehyde, permeabilized with 0.1% Triton in PBS and blocked with 10% serum for 30 minutes at 22°C. Samples were incubated with primary antibody (1/300) for 16 hours at 4°C. An Alexa Fluor® 488-conjugated goat anti-rabbit IgG polyclonal (1/500) was used as the secondary antibody.

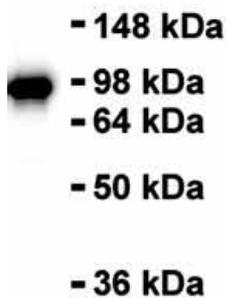


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

This image is courtesy of an anonymous Abreview

ab32572 staining beta Catenin in dog colon tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections).

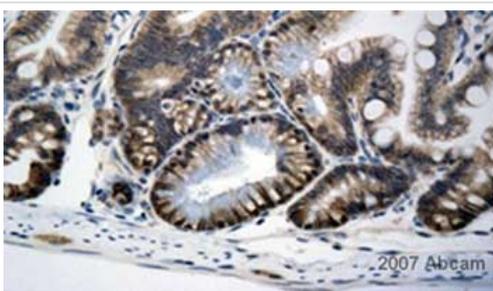
Tissue was fixed with formaldehyde and blocked with 10% serum for 30 minutes at 25°C; antigen retrieval was by heat mediation in 10 mM citrate buffer, pH6. Samples were incubated with primary antibody (1/250 in PBS with 1x casein) for 90 minutes at 25°C. A biotin-conjugated Goat anti-rabbit IgG polyclonal (1/200) was used as the secondary antibody.



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

This image is courtesy of an anonymous Abreview

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.

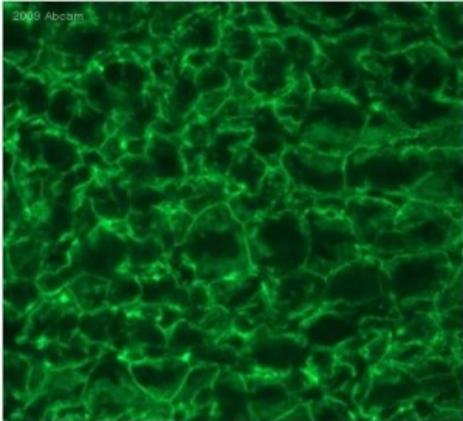


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-beta Catenin antibody [E247] (ab32572)

This image is courtesy of an anonymous Abreview

ab32572 at 1/200 staining mouse small intestine tissue sections by IHC-P.

The tissue was formaldehyde fixed and a heat mediated antigen retrieval step was performed before incubation with the primary antibody. An HRP conjugated goat anti-rabbit antibody was used as the secondary.

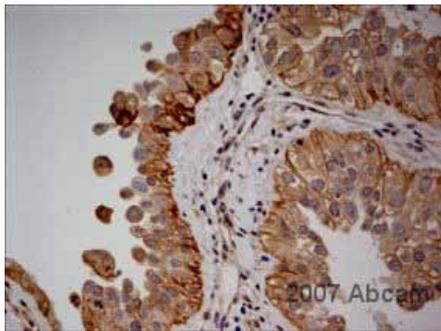


Immunohistochemistry (Frozen sections) - Anti-beta Catenin antibody [E247] (ab32572)

This image is a courtesy of Anonymous Abreview

ab32572 staining beta Catenin in mouse liver tissue section by Immunohistochemistry (Frozen sections).

Tissue samples were fixed with formaldehyde and blocked with 5% serum at 4⁰C for 30 minutes. The sample was incubated with primary antibody (1/200) in dilution buffer containing PBS and 3% goat serum at 4⁰C for 9 hours. An Alexa Fluor[®]488-conjugated Goat polyclonal to rabbit IgG was used as secondary antibody at 1/200 dilution.



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

This image is courtesy of an anonymous Abreview

ab32572 staining human renal carcinoma tissue sections by IHC-P.

Sections were formaldehyde fixed and subjected to heat mediated antigen retrieval in citrate buffer (pH 6) prior to blocking with 1% milk for 45 minutes at 22°C. The primary antibody was diluted 1/200 and incubated with the sample for 1 hour at 22°C. An HRP conjugated goat anti-rabbit antibody, diluted 1/400, was used as the secondary.

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