Human/Mouse/Rat β-Catenin Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF1329

DESCRIPTION

Species Reactivity	Human/Mouse/Rat
Specificity	Detects human, mouse and rat β-Catenin in Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human β-Catenin Ala2-Leu781 Accession # P35222
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 μm filtered solution in PBS.

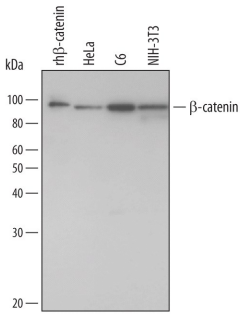
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Chromatin Immunoprecipitation (ChIP)	5 µg/5 x 10^6 cells	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Intracellular Staining by Flow Cytometry	2.5 µg/10^6 cells	See Below
Simple Western	50 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

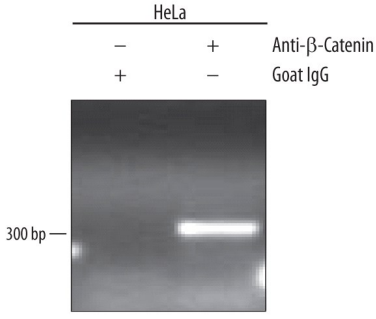
DATA

Western Blot



Detection of Human/Mouse/Rat β-Catenin by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line, C6 rat glioma cell line, and NIH-3T3 mouse embryonic fibroblast cell line. PVDF membrane was probed with 1 µg/mL Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). For additional reference, recombinant human β-catenin (1 ng) was included. A specific band for beta-Catenin was detected at approximately 95 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Chromatin Immunoprecipitation (ChIP)



Detection of β-Catenin-regulated Genes by Chromatin Immunoprecipitation. HeLa human cervical epithelial carcinoma cell line were fixed using formaldehyde, resuspended in lysis buffer, and sonicated to shear chromatin. β-Catenin/DNA complexes were immunoprecipitated using 5 µg Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329) or control antibody (Catalog # AB-108-C) for 15 minutes in an ultrasonic bath, followed by Biotinylated Anti-Goat IgG Secondary Antibody (Catalog # BAF109). Immunocomplexes were captured using 50 µL of MagCellect Streptavidin Ferrofluid (Catalog # MAG999) and DNA was purified using chelating resin solution. The *SU(Z)12* promoter was detected by standard PCR.

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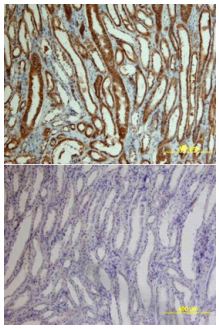
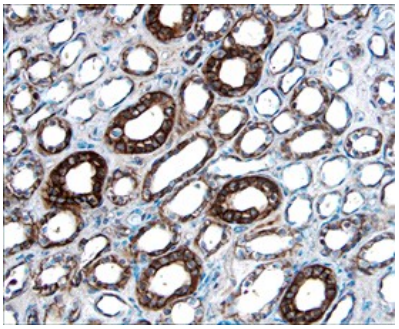
Immunohistochemistry



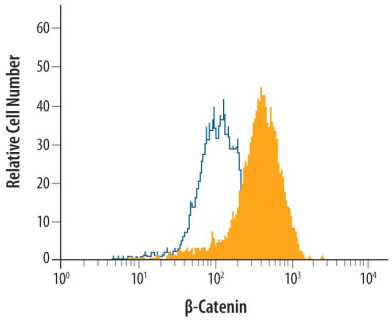
β-Catenin in Human Kidney Cancer Tissue. β-Catenin was detected in immersion fixed paraffin-embedded sections of human kidney cancer tissue using Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Lower panel shows a lack of labeling if primary antibodies are omitted and tissue is stained only with secondary antibody followed by incubation with detection reagents. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Immunohistochemistry



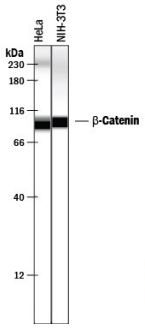
β-Catenin in Human Kidney Cancer Tissue. β-Catenin was detected in immersion fixed paraffin-embedded sections of human kidney cancer tissue using 15 µg/mL Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329) overnight at 4 °C. Tissue was stained with the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific labeling was localized to epithelial cells in collecting tubules in the medulla. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Intracellular Staining by Flow Cytometry



Detection of β-Catenin in HeLa Human Cell Line by Flow Cytometry. HeLa human cervical epithelial carcinoma cell line was stained with Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329, filled histogram) or control antibody (Catalog # AB-108-C, open histogram), followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with saponin.

Simple Western



Detection of Human and Mouse β-Catenin by Simple Western™. Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line and NIH-3T3 mouse embryonic fibroblast cell line, loaded at 0.2 mg/mL. A specific band was detected for β-Catenin at approximately 94-97 kDa (as indicated) using 50 µg/mL of Goat Anti-Human/Mouse/Rat β-Catenin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1329) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system. Non-specific interaction with the 230 kDa Simple Western standard may be seen with this antibody.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

β-Catenin is a cadherin-associated protein that is involved in the regulation of cell adhesion. It also acts as a transcriptional co-activator in the nucleus and is involved the canonical Wnt signal transduction pathways.

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