# abcam

### Product datasheet

# Anti-APC antibody [EP701Y] - BSA and Azide free ab239828



## 1 References 10 Images

#### Overview

Product name Anti-APC antibody [EP701Y] - BSA and Azide free

**Description** Rabbit monoclonal [EP701Y] to APC - BSA and Azide free

Host species Rabbit

**Specificity** This antibody is predicted to detect isoform 2 (short) of APC based on sequence analysis.

Tested applications Suitable for: ICC/IF, Flow Cyt (Intra), IP, IHC-P, WB

Species reactivity Reacts with: Mouse, Rat, Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HEK-293, C2C12, and C6 whole cell lysates; ICC/IF: HEK-293 cells; Flow Cyt (intra): HEK-

293 cells; IP: HEK-293 whole cell lysate; IHC-P: Human, mouse and rat colon tissues; human

colon carcinoma tissue.

**General notes** ab239828 is the carrier-free version of <u>ab40778</u>.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our <u>conjugation kits</u> for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar<sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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.

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Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

Storage buffer pH: 7.2

Constituent: 59% PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EP701Y

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee Our

Our Abpromise guarantee covers the use of ab239828 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
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.  See IHC antigen retrieval protocols.
WB		Use at an assay dependent concentration. Detects a band of approximately 160 kDa (predicted molecular weight: 312 kDa).

#### **Target**

**Function** 

Tumor suppressor. Promotes rapid degradation of CTNNB1 and participates in Wnt signaling as a negative regulator. APC activity is correlated with its phosphorylation state. Activates the GEF activity of SPATA13 and ARHGEF4. Plays a role in hepatocyte growth factor (HGF)-induced cell migration. Required for MMP9 up-regulation via the JNK signaling pathway in colorectal tumor cells. Acts as a mediator of ERBB2-dependent stabilization of microtubules at the cell cortex. It is required for the localization of MACF1 to the cell membrane and this localization of MACF1 is critical for its function in microtubule stabilization.

Tissue specificity

Expressed in a variety of tissues.

#### Involvement in disease

Defects in APC are a cause of familial adenomatous polyposis (FAP) [MIM:175100]; which includes also Gardner syndrome (GS). FAP and GS contribute to tumor development in patients with uninherited forms of colorectal cancer. FAP is characterized by adenomatous polyps of the colon and rectum, but also of upper gastrointestinal tract (ampullary, duodenal and gastric adenomas). This is a viciously premalignant disease with one or more polyps progressing through dysplasia to malignancy in untreated gene carriers with a median age at diagnosis of 40 years. Defects in APC are a cause of hereditary desmoid disease (HDD) [MIM:135290]; also known as familial infiltrative fibromatosis (FIF). HDD is an autosomal dominant trait with 100% penetrance and possible variable expression among affected relatives. HDD patients show multifocal fibromatosis of the paraspinal muscles, breast, occiput, arms, lower ribs, abdominal wall, and mesentery. Desmoid tumors appears also as a complication of familial adenomatous polyposis. Defects in APC are a cause of medulloblastoma (MDB) [MIM:155255]. MDB is a malignant, invasive embryonal tumor of the cerebellum with a preferential manifestation in children. Although the majority of medulloblastomas occur sporadically, some manifest within familial cancer syndromes such as Turcot syndrome and basal cell nevus syndrome (Gorlin syndrome). Defects in APC are a cause of mismatch repair cancer syndrome (MMRCS) [MIM:276300]; also known as Turcot syndrome or brain tumor-polyposis syndrome 1 (BTPS1). MMRCS is an autosomal dominant disorder characterized by malignant tumors of the brain associated with multiple colorectal adenomas. Skin features include sebaceous cysts, hyperpigmented and cafe au lait spots.

Defects in APC are a cause of gastric cancer (GASC) [MIM:613659]; also called gastric cancer intestinal or stomach cancer. Gastric cancer is a malignant disease which starts in the stomach, can spread to the esophagus or the small intestine, and can extend through the stomach wall to nearby lymph nodes and organs. It also can metastasize to other parts of the body. The term gastric cancer or gastric carcinoma refers to adenocarcinoma of the stomach that accounts for most of all gastric malignant tumors. Two main histologic types are recognized, diffuse type and intestinal type carcinomas. Diffuse tumors are poorly differentiated infiltrating lesions, resulting in thickening of the stomach. In contrast, intestinal tumors are usually exophytic, often ulcerating, and associated with intestinal metaplasia of the stomach, most often observed in sporadic disease. Defects in APC are a cause of hepatocellular carcinoma (HCC) [MIM:114550]. This defect includes also the disease entity termed hepatoblastoma.

Sequence similarities

Belongs to the adenomatous polyposis coli (APC) family.

Contains 7 ARM repeats.

Domain

The microtubule tip localization signal (MtLS) motif; mediates interaction with MAPRE1 and targeting to the growing microtubule plus ends.

Post-translational modifications

Phosphorylated by GSK3B.

Ubiquitinated, leading to its degradation by the proteasome. Ubiquitination is facilitated by Axin. Deubiquitinated by ZRANB1/TRABID.

**Cellular localization** 

Cell junction > adherens junction. Cytoplasm > cytoskeleton. Cell projection > lamellipodium. Cell projection > ruffle membrane. Cytoplasm. Cell membrane. Associated with the microtubule network at the growing distal tip of microtubules. Accumulates in the lamellipodium and ruffle membrane in response to hepatocyte growth factor (HGF) treatment. The MEMO1-RHOA-DIAPH1 signaling pathway controls localization of the phosophorylated form to the cell membrane.

#### **Images**



Western blot - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

**All lanes :** Anti-APC antibody [EP701Y] (<u>ab40778</u>) at 1/5000 dilution (Purified)

Lane 1 : HEK-293 (Human embryonic kidney epithelial cell) whole cell lysate

Lane 2: C2C12 (Mouse myoblasts myoblast) whole cell lysate

Lane 3: C6 (Rat glial tumor glial cell) whole cell lysate

Lysates/proteins at 15 µg per lane.

#### Secondary

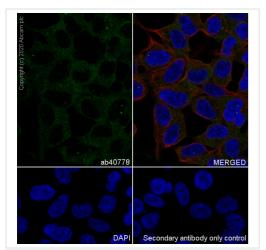
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 312 kDa
Observed band size: 160 kDa

The molecular weight observed represents the truncated APC as described in PMID: 17595655.

Blocking/Diluting buffer: 5% NFDM/TBST

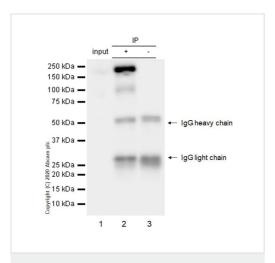
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).



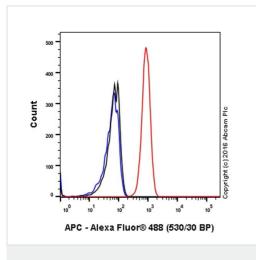
Immunocytochemistry/ Immunofluorescence - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

Immunocytochemistry/ Immunofluorescence analysis of HEK-293 (Human embryonic kidney epithelial cell) cells labeling APC with Purified <a href="mailto:ab40778">ab40778</a> at 1:100 dilution (10 µg/mL). Cells were fixed in 4% Paraformaldehyde. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) 1:200 (2.5 µg/mL). Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488, <a href="mailto:ab50077">ab150077</a>) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab40778</u>).



Immunoprecipitation - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)



Flow Cytometry (Intracellular) - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

Purified <u>ab40778</u> at 1:70 dilution ( $2\mu g$ ) immunoprecipitating APC in HEK-293 whole cell lysate.

Lane 1 (input): HEK-293 (Human embryonic kidney epithelial cell) whole cell lysate 10µg

Lane 2 (+): <u>ab40778</u> + HEK-293 whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of <u>ab40778</u> in HEK-293 whole cell lysate.

Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

Observed band size: 160 kDa

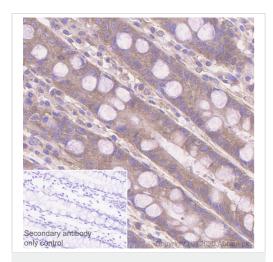
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).

Purified <u>ab40778</u> staining APC in the human cell line 293 (human embryonic kidney) by intracellular flow cytometry. Cells were fixed with 4% paraformaldehyde, permabilised with 90% methanol and the sample was incubated with the primary antibody at a dilution of 1/150. A goat anti rabbit IgG (Alexa Fluor<sup>®</sup> 488) at a dilution of 1/2000 was used as the secondary antibody.

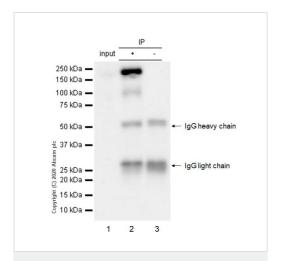
Isoytype control: Rabbit monoclonal IgG (Black)

Unlabelled control: Cell without incubation with primary antibody and secondary antibody (Blue)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)



Immunoprecipitation - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue sections labeling APC with purified <u>ab40778</u> at 1/1000 dilution (1.383 µg/mL). Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

The immunostaining was performed on a Leica Biosystems  $\mathsf{BOND}^{\circledR}$  RX instrument.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).

Unpurified <u>ab40778</u> at 1/70 dilution (2 $\mu$ g) immunoprecipitating APC in HEK-293 whole cell lysate.

Lane 1 (input): HEK-293 (Human embryonic kidney epithelial cell) whole cell lysate 10µg

Lane 2 (+): <u>ab40778</u> + HEK-293 whole cell lysate.

Lane 3 (-): Rabbit monoclonal  $\lg G$  (ab172730) instead of ab40778 in HEK-293 whole cell lysate.

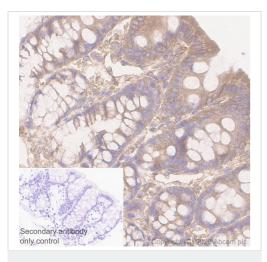
 $\label{eq:continuous} \mbox{VeriBlot for IP Detection Reagent (HRP) } (\mbox{$\underline{ab131366}$}) \mbox{ (1/1000 dilution) was used for Western blotting.}$ 

Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

Observed band size: 160 kDa

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).

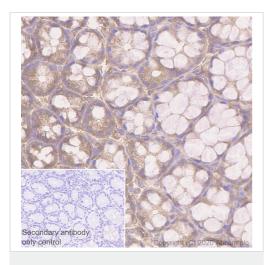


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat colon tissue sections labeling APC with purified <u>ab40778</u> at 1/1000 dilution (1.383 µg/mL). Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).

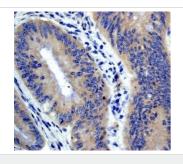


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse colon tissue sections labeling APC with purified <u>ab40778</u> at 1/1000 dilution (1.383 µg/mL). Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

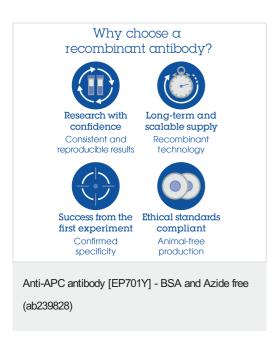
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-APC antibody [EP701Y] - BSA and Azide free (ab239828)

<u>ab40778</u> (1:50), unpurified, staining human colon carcinoma by immunohistochemistry, paraffin-embedded tissue. Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab40778).



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