abcam

Product datasheet

Anti-Cdk8 antibody [EPR21005] ab229192

KO VALIDATED Recombinant RabMAb

1 Abreviews 2 References 5 Images

Overview

Product name	Anti-Cdk8 antibody [EPR21005]
Description	Rabbit monoclonal [EPR21005] to Cdk8
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Wild-type HeLa, Daudi, HAP1, HCT 116, K562 and SW480 whole cell lysates; Human testis lysate. IP: SW480 whole cell lysate.
General notes	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**.

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 long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR21005
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab229192 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
WB		1/1000. Detects a band of approximately 53 kDa (predicted molecular weight: 53 kDa).
IP		1/30.

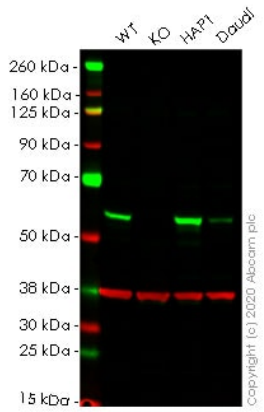
Target

Function Component of the Mediator complex, a coactivator involved in regulated gene transcription of nearly all RNA polymerase II-dependent genes. Mediator functions as a bridge to convey information from gene-specific regulatory proteins to the basal RNA polymerase II transcription machinery. Mediator is recruited to promoters by direct interactions with regulatory proteins and serves as a scaffold for the assembly of a functional preinitiation complex with RNA polymerase II and the general transcription factors. Phosphorylates the CTD (C-terminal domain) of the large subunit of RNA polymerase II (RNAp II), which may inhibit the formation of a transcription initiation complex. Phosphorylates CCNH leading to down-regulation of the TFIIH complex and transcriptional repression. Recruited through interaction with MAML1 to hyperphosphorylate the intracellular domain of NOTCH, leading to its degradation.

Sequence similarities Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. CDC2/CDKX subfamily.
Contains 1 protein kinase domain.

Cellular localization Nucleus.

Images



Western blot - Anti-Cdk8 antibody [EPR21005] (ab229192)

All lanes : Anti-Cdk8 antibody [EPR21005] (ab229192) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CDK8 knockout HeLa cell lysate
Lane 3 : HAP1 cell lysate
Lane 4 : Daudi cell lysate

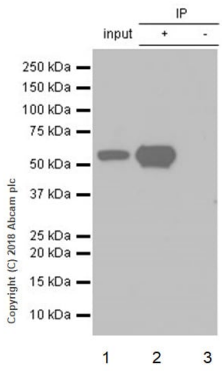
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 53 kDa

Lanes 1-4: Merged signal (red and green). Green - ab229192 observed at 53 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab229192 Anti-Cdk8 antibody [EPR21005] was shown to specifically react with Cdk8 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265087** (knockout cell lysate **ab257885**) was used. Wild-type and Cdk8 knockout samples were subjected to SDS-PAGE. ab229192 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 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.

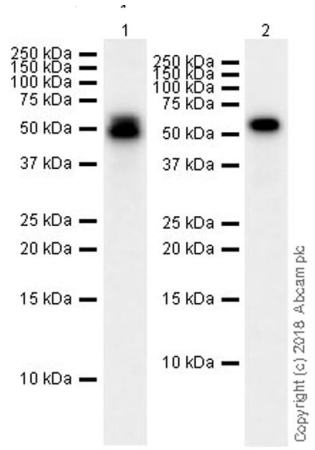


Immunoprecipitation - Anti-Cdk8 antibody [EPR21005] (ab229192)

Cdk8 was immunoprecipitated from 0.35 mg of SW480 (human colorectal adenocarcinoma cell line) whole cell lysate with ab229192 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab229192 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1/10,000 dilution.

Lane 1: SW480 whole cell lysate 10 µg (Input).
Lane 2: ab229192 IP in SW480 whole cell lysate.
Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab229192 in SW480 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.
Exposure time: 3 minutes.



Western blot - Anti-Cdk8 antibody [EPR21005] (ab229192)

All lanes : Anti-Cdk8 antibody [EPR21005] (ab229192) at 1/1000 dilution

Lane 1 : Human testis lysate
Lane 2 : K562 (human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate

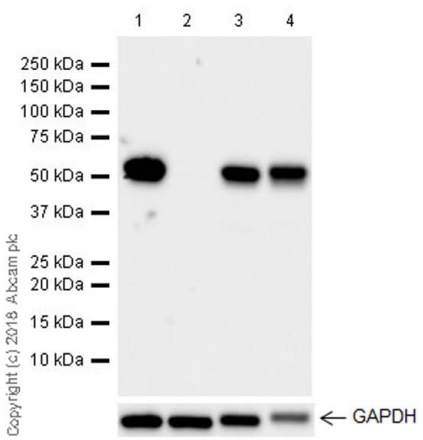
Lysates/proteins at 10 µg per lane.

Secondary
Lane 1 : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution
Lane 2 : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/50000 dilution

Predicted band size: 53 kDa
Observed band size: 53 kDa

Exposure time: 58 seconds

Blocking and dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Cdk8 antibody [EPR21005] (ab229192)

All lanes : Anti-Cdk8 antibody [EPR21005] (ab229192) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate at 20 µg
Lane 2 : Cdk8 knockout HAP1 whole cell lysate at 20 µg
Lane 3 : HCT 116 (human colorectal carcinoma cell line) whole cell lysate at 10 µg
Lane 4 : SW480 (Human colorectal adenocarcinoma cell line) whole cell lysate at 10 µg

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

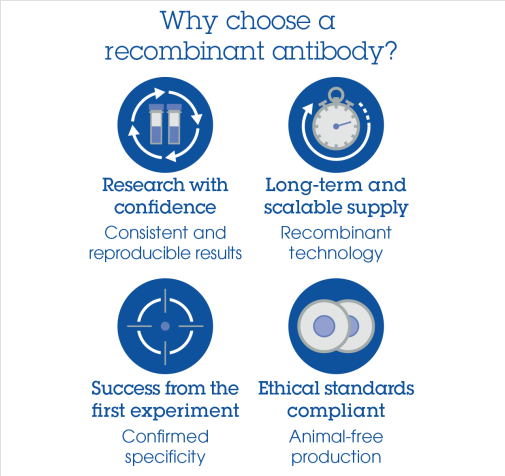
Predicted band size: 53 kDa
Observed band size: 53 kDa

Exposure time: 48 seconds

Blocking and dilution buffer: 5% NFDM/TBST.

ab229192 was shown to specifically react with Cdk8 in wild-type HAP1 cells as signal was lost in Cdk8 knockout cells.

Wild-type and Cdk8 knockout samples were subjected to SDS-PAGE. ab229192 and **ab181602** (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument using the ECL technique.



Anti-Cdk8 antibody [EPR21005] (ab229192)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors