abcam

Product datasheet

Anti-CYP27B1 antibody [EPR20271] ab206655

Recombinant RabMAb

10 References 9 Images

Overview

Product name Anti-CYP27B1 antibody [EPR20271]

Description Rabbit monoclonal [EPR20271] to CYP27B1

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, IP

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HaCaT, MeWo and HeLa whole cell lysates; Human fetal heart, fetal brain, fetal kidney and

fetal liver lysates; Mouse heart and kidney lysates; Rat heart, kidney and brain lysates;

Recombinant CYP27B1 protein. IHC-P: Human, mouse and rat kidney tissues. IP: MeWo 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: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number EPR20271

Isotype IgG

Applications

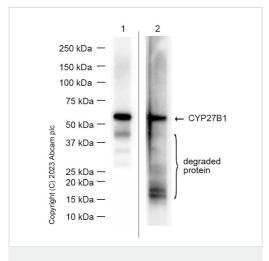
The Abpromise guarantee Our Abpromise guarantee covers the use of ab206655 in the following tested applications.

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

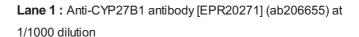
Application	Abreviews	Notes
WB		1/1000. Detects a band of approximately 45, 70 kDa (predicted molecular weight: 57 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		1/40.

rarget		
Function	Catalyzes the conversion of 25-hydroxyvitamin D3 (25(OH)D) to 1-alpha,25-dihydroxyvitamin D3 (1,25(OH)2D) plays an important role in normal bone growth, calcium metabolism, and tissue differentiation.	
Tissue specificity	Kidney.	
Pathway	Hormone biosynthesis; cholecalciferol biosynthesis.	
Involvement in disease	Defects in CYP27B1 are the cause of rickets vitamin D-dependent type 1A (VDDR1A) [MIM:264700]; also known as pseudovitamin D deficiency rickets (PDDR). A disorder caused by a selective deficiency of the active form of vitamin D (1,25-dihydroxyvitamin D3) and resulting in defective bone mineralization and clinical features of rickets.	
Sequence similarities	Belongs to the cytochrome P450 family.	
Cellular localization	Mitochondrion membrane.	

Images



Western blot - Anti-CYP27B1 antibody [EPR20271] (ab206655)



Lane 2: Anti-His-tag antibody at 1/200 dilution

All lanes: Recombinant CYP27B1 protein

Secondary

All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000

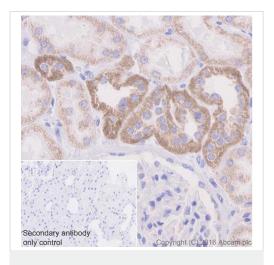
dilution

Predicted band size: 57 kDa **Observed band size:** 56 kDa

Exposure time: 3 seconds

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.



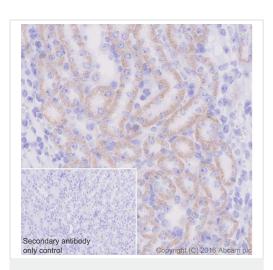
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CYP27B1 antibody
[EPR20271] (ab206655)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labeling CYP27B1 with ab206655 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Strong cytoplasmic staining on distal convoluted tubules of human kidney. Weaker cytoplasmic staining on proximal convoluted tubules [PMID: 23416105] [PMID: 12050281].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



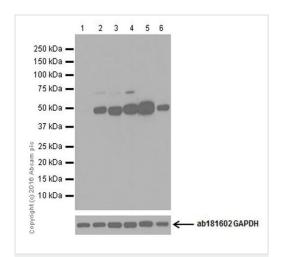
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CYP27B1 antibody
[EPR20271] (ab206655)

Immunohistochemical analysis of paraffin-embedded mouse kidney tissue labeling CYP27B1 with ab206655 at 1/2000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasmic staining on mouse kidney tubules [PMID: 23416105] [PMID: 12050281].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-CYP27B1 antibody [EPR20271] (ab206655)

All lanes : Anti-CYP27B1 antibody [EPR20271] (ab206655) at 1/2000 dilution

Lane 1 : THP-1 (Human monocytic leukemia cell line) whole cell lysate

Lane 2: HaCaT (Human keratinocyte cell line) whole cell lysate

Lane 3 : MeWo (Human malignant melanoma cell line) whole cell lysate

Lane 4 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 5: Human fetal heart lysate

Lane 6: Human fetal brain lysate

Lysates/proteins at 20 µg per lane.

Secondary

Lanes 1-4: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Lanes 5-6: VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) at 1/4000 dilution

Predicted band size: 57 kDa **Observed band size:** 45,70 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

THP-1 is a negative control (PMID: 16930540).

The molecular weight observed is consistent with what has been

described in the literature (PMID: 16061850).

1 2
250 kDa —
150 kDa —
100 kDa —
75 kDa —
50 kDa —
25 kDa —
20 kDa —
15 kDa —
15 kDa —
10 kDa —

Western blot - Anti-CYP27B1 antibody [EPR20271] (ab206655)

All lanes : Anti-CYP27B1 antibody [EPR20271] (ab206655) at 1/1000 dilution

Lane 1 : Human fetal kidney lysate

Lane 2 : Human fetal liver lysate

Lysates/proteins at 20 µg per lane.

Secondary

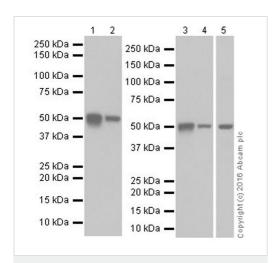
All lanes : VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) at 1/4000 dilution

Predicted band size: 57 kDa **Observed band size:** 45 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 16061850).



Western blot - Anti-CYP27B1 antibody [EPR20271] (ab206655)

All lanes : Anti-CYP27B1 antibody [EPR20271] (ab206655) at 1/1000 dilution

Lane 1 : Mouse heart lysate
Lane 2 : Mouse kidney lysate
Lane 3 : Rat heart lysate
Lane 4 : Rat kidney lysate
Lane 5 : Rat brain lysate

Lysates/proteins at 10 µg per lane.

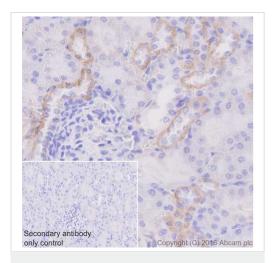
Secondary

All lanes : Goat Anti-Rabbit $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$ at 1/100000 dilution

Predicted band size: 57 kDa **Observed band size:** 57 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1, 2, 3 and 4: 1 second; Lane 5: 3 seconds.



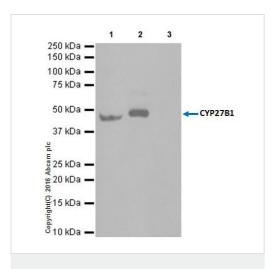
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CYP27B1 antibody
[EPR20271] (ab206655)

Immunohistochemical analysis of paraffin-embedded rat kidney tissue labeling CYP27B1 with ab206655 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasmic staining on rat kidney tubules [PMID: 23416105] [PMID: 12050281].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-CYP27B1 antibody [EPR20271] (ab206655)

CYP27B1 was immunoprecipitated from 0.35 mg of MeWo (Human malignant melanoma cell line) whole cell lysate with ab206655 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab206655 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

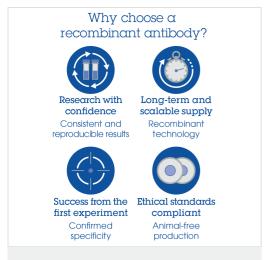
Lane 1: MeWo whole cell lysate, 10 µg (Input).

Lane 2: ab206655 IP in MeWo whole cell lysate.

Lane 3: Rabbit monoclonal $\lg G$ ($\underline{ab172730}$) instead of ab206655 in MeWo whole cell lysate.

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

Exposure time: 30 seconds.



Anti-CYP27B1 antibody [EPR20271] (ab206655)

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