# abcam

### Product datasheet

## Anti-CYR61/CCN1 antibody [EPR20681] ab230947





#### ★★★★★ 1 Abreviews 3 References 7 Images

#### Overview

**Product name** Anti-CYR61/CCN1 antibody [EPR20681]

**Description** Rabbit monoclonal [EPR20681] to CYR61/CCN1

**Host species** Rabbit

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

Species reactivity Reacts with: Human

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

Positive control WB: HeLa, PC-3, MDA-MB-231 and Saos-2 whole cell lysate. ICC/IF: MDA-MB-231 cells. Flow

Cyt (intra): MDA-MB-231 cells. IP: Saos-2 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 EPR20681

Isotype lgG

#### **Applications**

#### The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab230947 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
Flow Cyt (Intra)		1/60.
WB	*** <u>*</u>	1/1000. Predicted molecular weight: 42 kDa.
ICC/IF		1/500.
IP		1/30.

#### **Target**

#### **Function**

Promotes cell proliferation, chemotaxis, angiogenesis and cell adhesion. Appears to play a role in wound healing by up-regulating, in skin fibroblasts, the expression of a number of genes involved in angiogenesis, inflammation and matrix remodeling including VEGA-A, VEGA-C, MMP1, MMP3, TIMP1, uPA, PAI-1 and integrins alpha-3 and alpha-5. CYR61-mediated gene regulation is dependent on heparin-binding. Down-regulates the expression of alpha-1 and alpha-2 subunits of collagen type-1. Promotes cell adhesion and adhesive signaling through integrin alpha-6/beta-1, cell migration through integrin alpha-v/beta-3.

#### Sequence similarities

Belongs to the CCN family.

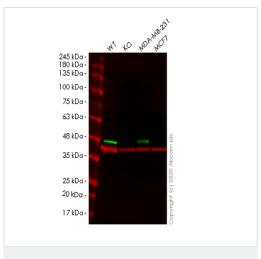
Contains 1 CTCK (C-terminal cystine knot-like) domain.

Contains 1 IGFBP N-terminal domain. Contains 1 TSP type-1 domain.

Contains 1 VWFC domain.

Cellular localization Secreted.

#### **Images**



Western blot - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)

**All lanes :** Anti-CYR61/CCN1 antibody [EPR20681] (ab230947) at 1/500 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: CYR61 knockout HeLa cell lysate

Lane 3: MDA-MB-231 cell lysate

Lane 4: MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

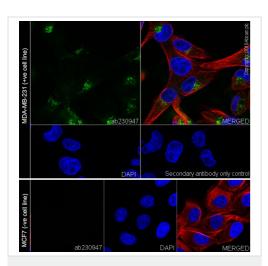
#### **Secondary**

**All lanes :** Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) at 1/10000 dilution

**Predicted band size:** 42 kDa **Observed band size:** 47 kDa

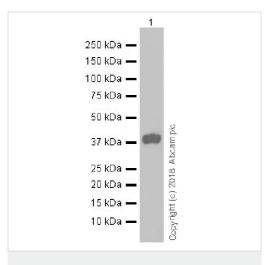
**Lanes 1-4:** Merged signal (red and green). Green - ab230947 observed at 47 kDa. Red - loading control <u>ab8245</u> observed at 36 kDa.

ab230947 Anti-CYR61/CCN1 antibody [EPR20681] was shown to specifically react with CYR61/CCN1 in wild-type HeLa cells. Loss of signal was observed when knockout cell line <a href="mailto:ab265288">ab265288</a> (knockout cell lysate <a href="mailto:ab257406">ab257406</a>) was used. Wild-type and CYR61/CCN1 knockout samples were subjected to SDS-PAGE. ab230947 and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8230947</a> and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8230947</a> and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8230947</a> and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8230947</a> and In 20000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MDA-MB-231 (human breast adenocarcinoma epithelial cell) cells labeling CYR61/CCN1 with ab230947 at 1/500 dilution, followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in the MDA-MB-231 cell line. **Negative control:** MCF7 (PMID: 11059746). The nuclear counter stain is DAPI (blue). Counterstained with ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red). The negative control is the secondary antibody only.



Western blot - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)

Anti-CYR61/CCN1 antibody [EPR20681] (ab230947) at 1/5000 dilution + MDA-MB-231 (human breast adenocarcinoma epithelial cell), whole cell lysate at 20  $\mu g$ 

#### Secondary

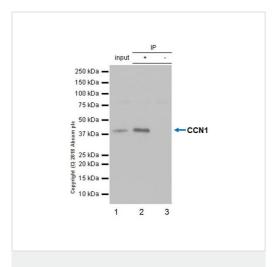
Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

**Predicted band size:** 42 kDa **Observed band size:** 42 kDa

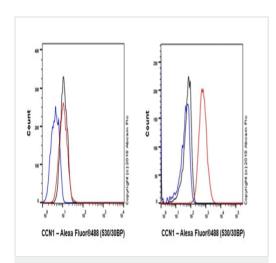
Exposure time: 3 minutes

Blocking/ Dilution buffer concentration: 5% NFDM/TBST.

The molecular mass observed is consistent with that of the full-length protein (42 kDa) (PMID: 23798676).



Immunoprecipitation - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)



Flow Cytometry (Intracellular) - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)

CYR61/CCN1 was immunoprecipitated from 0.35 mg Saos-2 (human osteosarcoma epithelial) whole cell lysate with ab230947 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab230947 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

**Lane 1:** Saos-2 (human osteosarcoma epithelial) whole cell lysate  $10 \mu g$  (lnput).

Lane 2: ab230947 IP in Saos-2 whole cell lysate (+).

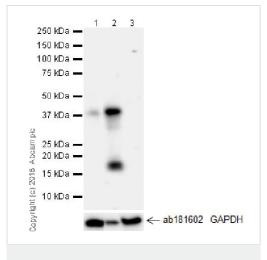
**Lane 3:** Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab230947 in Saos-2 whole cell lysate (-).

Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 8 seconds.

The molecular mass observed is consistent with that of the full-length protein (42 kDa) (PMID: 23798676).

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol permeabilized MCF7 (human breast adenocarcinoma epithelial cell, Left) / MDA-MB-231 (human breast adenocarcinoma epithelial cell, Right) cell line labeling CYR61/CCN1with ab230947 at 1/60 (red) compared with a Rabbit monoclonal lgG (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit lgG (Alexa Fluor \$\tilde{i}\cdot\frac{\gamma}{2}\$ 488, ab150077), at 1/2000 dilution was used as the secondary antibody.

Negative control: MCF7 (PMID: 11059746).



Western blot - Anti-CYR61/CCN1 antibody [EPR20681] (ab230947)

**All lanes :** Anti-CYR61/CCN1 antibody [EPR20681] (ab230947) at 1/1000 dilution

**Lane 1 :** PC-3 (human prostate adenocarcinoma epithelial cell) whole cell lysate

Lane 2 : Saos-2 (human osteosarcoma epithelial) whole cell lysateLane 3 : MCF7 (human breast adenocarcinoma epithelial cell)whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

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

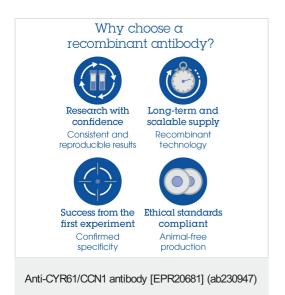
Predicted band size: 42 kDa

Exposure time: 3 minutes

#### Blocking/Dilution buffer concentration: 5% NFDM/TBST.

The expression profile observed is consistent with the literature, with the 42 kDa band being the full-length protein and the 19 kDa band likely a degradation product (PMID: 23798676; PMID: 16266990; PMID: 8657105).

Negative control: MCF7 (PMID 11059746).



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