# Overview

**Product name**  
Anti-Paxillin antibody [Y113] ab32084

**Description**  
Rabbit monoclonal [Y113] to Paxillin

**Host species**  
Rabbit

**Specificity**  
ab32084 recognises Paxillin alpha, beta and gamma isoforms.

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

**Species reactivity**  
Reacts with: Mouse, Rat, Cow, Dog, Human

**Immunogen**  
Synthetic peptide within Human Paxillin aa 1-100 (N terminal). The exact sequence is proprietary.

**Positive control**  
HeLa cell lysate, human breast carcinoma.

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

This product is a recombinant rabbit monoclonal antibody.

## 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. Avoid freeze / thaw cycle.

**Dissociation constant ($K_D$)**  
$K_D = 4.17 \times 10^{-10} \text{ M}$

**Storage buffer**  
PBS 49%, Sodium azide 0.01%, Glycerol 50%, BSA 0.05%

**Clonality**  
Monoclonal
**Clone number**  
Y113

**Isotype**  
IgG

### Applications

Our **Abpromise guarantee** covers the use of **ab32084** in the following tested applications.

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

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<td>Flow Cyt</td>
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<td>Use at an assay dependent concentration.</td>
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<tr>
<td>IP</td>
<td>⭐⭐⭐⭐⭐</td>
<td>1/200.</td>
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### Target

**Function**  
Cytoskeletal protein involved in actin-membrane attachment at sites of cell adhesion to the extracellular matrix (focal adhesion).

**Sequence similarities**  
Belongs to the paxillin family.

Contains 4 LIM zinc-binding domains.

**Post-translational modifications**  
Phosphorylated on tyrosine residues during integrin-mediated cell adhesion, embryonic development, fibroblast transformation and following stimulation of cells by mitogens.

**Cellular localization**  
Cytoplasm > cytoskeleton. Cell junction > focal adhesion.
Immunocytochemistry/ Immunofluorescence - Anti-Paxillin antibody [Y113] (ab32084)

This image is courtesy of Dr. Shaohua Li, UMDNJ-Robert Wood Johnson Medical School

Sample: mouse embryonic fibroblasts

Preparation:

Fix in 3% PFA in PBS for 30 min at RT

Primary antibody: Rabbit anti-paxillin Y113 (ab32084), 1:100

Secondary antibody: Goat polyclonal
Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488) pre-adsorbed (ab150081), 1:200

Rhodamine-phalloidin, 1:100

Nuclei were counterstained with DAPI

Flow Cytometry - Anti-Paxillin antibody [Y113] (ab32084)

Flow cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling with purified ab32084 at 1/100 dilution (10ug/ml) (Red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) (ab150077)(1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black)(ab172730) was used as a isotype control.Cell without incubation with primary antibody and secondary antibody (Blue) were used as unlabeled control.
ab32084 staining paxillin in MEF1 cells treated with (S)-(−)-Blebbistatin (ab120491), by ICC/IF. Decreased membrane expression of paxillin correlates with increased concentration of (S)-(−)-Blebbistatin, as described in literature.

The cells were incubated at 37°C for 2h in media containing different concentrations of ab120491 ( (S)-(−)-Blebbistatin ) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab32084 (1/100 dilution) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (ab96899) at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.

Western blot - Anti-Paxillin antibody [Y113] (ab32084) at 1/10000 dilution + HeLa cell lysate.

**Predicted band size:** 68 kDa

**Observed band size:** 60 kDa
Ab32084 showing positive staining in Normal ovary tissue.

Anti-Paxillin antibody [Y113] (ab32084) at 1/5000 dilution + Mouse RAW264.7 whole cell lysate at 20 µg

Secondary
An HRP Donkey anti-rabbit IgG polyclonal at 1/10000 dilution

Predicted band size: 68 kDa

Blocking step: 5% Milk for 1 hour at 20°C

Gel: SDS-PAGE

Ab32084 showing positive staining in Ovarian carcinoma tissue.
ab32084 showing positive staining in Transitional cell carcinoma of kidney tissue.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Paxillin antibody [Y113] (ab32084)

Immunofluorescence analysis of bovine kidney cells, staining Paxillin with ab32084.

Cells were fixed with paraformaldehyde, permeabilized with 1% Triton X-100 and blocked with 5% BSA for 1 hour. Samples were incubated with primary antibody (1/2500 in 5% BSA) for 1 hour at 25°C. An undiluted AlexaFluor®488-conjugated goat anti-rabbit polyclonal IgG was used as the secondary antibody.
ab32084 staining paxillin in MEF1 cells treated with (+/-)-blebbistatin (ab120425), by ICC/IF. Decreased membrane expression of paxillin correlates with increased concentration of (+/-)-blebbistatin, as described in literature.

The cells were incubated at 37°C for 1h in media containing different concentrations of ab120425 ((+/-)-blebbistatin) in DMSO, fixed with 4% formaldehyde for 10 minutes at room temperature and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab32084 (1/100 dilution) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (ab96899) at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D

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