


## Product datasheet

# Anti-Vinculin (phospho Y100) antibody ab200812

[1 Abreviews](#) [3 Images](#)

### Overview

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<b>Product name</b>	Anti-Vinculin (phospho Y100) antibody
<b>Description</b>	Rabbit polyclonal to Vinculin (phospho Y100)
<b>Host species</b>	Rabbit
<b>Tested applications</b>	<b>Suitable for:</b> ICC/IF, WB
<b>Species reactivity</b>	<b>Reacts with:</b> Human, Recombinant fragment <b>Predicted to work with:</b> Chicken 
<b>Immunogen</b>	Synthetic peptide corresponding to Human Vinculin (phospho Y100). derived from a region that contains tyrosine 100. Database link: <a href="#">P18206-1</a>
<b>Positive control</b>	WB: Hela untreated and treated with 100 ng/mL of LPS for 20 mins whole cell lysate; COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were treated with vanadate for 24 hr. ICC/IF: 70% confluent log phase HeLa cells.
<b>General notes</b>	<p>The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.</p> <p>If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&amp;As</p>

### Properties

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<b>Form</b>	Liquid
<b>Storage instructions</b>	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.
<b>Storage buffer</b>	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 99% PBS
<b>Purity</b>	Immunogen affinity purified
<b>Clonality</b>	Polyclonal
<b>Isotype</b>	IgG

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab200812 in the following tested applications.

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

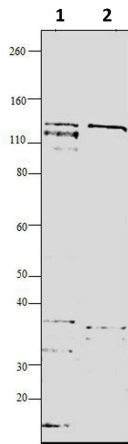
Application	Abreviews	Notes
ICC/IF		1/250.
WB		1/1000. Predicted molecular weight: 124 kDa.

## Target

<b>Function</b>	Actin filament (F-actin)-binding protein involved in cell-matrix adhesion and cell-cell adhesion. Regulates cell-surface E-cadherin expression and potentiates mechanosensing by the E-cadherin complex. May also play important roles in cell morphology and locomotion.
<b>Tissue specificity</b>	Metavinculin is muscle-specific.
<b>Involvement in disease</b>	<p>Defects in VCL are the cause of cardiomyopathy dilated type 1W (CMD1W) [MIM:611407]. Dilated cardiomyopathy is a disorder characterized by ventricular dilation and impaired systolic function, resulting in congestive heart failure and arrhythmia. Patients are at risk of premature death.</p> <p>Defects in VCL are the cause of cardiomyopathy familial hypertrophic type 15 (CMH15) [MIM:613255]. It is a hereditary heart disorder characterized by ventricular hypertrophy, which is usually asymmetric and often involves the interventricular septum. The symptoms include dyspnea, syncope, collapse, palpitations, and chest pain. They can be readily provoked by exercise. The disorder has inter- and intrafamilial variability ranging from benign to malignant forms with high risk of cardiac failure and sudden cardiac death.</p>
<b>Sequence similarities</b>	Belongs to the vinculin/alpha-catenin family.
<b>Domain</b>	<p>Exists in at least two conformations. When in the closed, 'inactive' conformation, extensive interactions between the head and tail domains prevent detectable binding to most of its ligands. It takes on an 'active' conformation after cooperative and simultaneous binding of two different ligands. This activation involves displacement of the head-tail interactions and leads to a significant accumulation of ternary complexes. The active form then binds a number of proteins that have both signaling and structural roles that are essential for cell adhesion.</p> <p>The N-terminal globular head (Vh) comprises of subdomains D1-D4. The C-terminal tail (Vt) binds F-actin and cross-links actin filaments into bundles. An intramolecular interaction between Vh and Vt masks the F-actin-binding domain located in Vt. The binding of talin and alpha-actinin to the D1 subdomain of vinculin induces a helical bundle conversion of this subdomain, leading to the disruption of the intramolecular interaction and the exposure of the cryptic F-actin-binding domain of Vt. Vt inhibits actin filament barbed end elongation without affecting the critical concentration of actin assembly.</p>
<b>Post-translational modifications</b>	<p>Phosphorylated; on serines, threonines and tyrosines. Phosphorylation on Tyr-1133 in activated platelets affects head-tail interactions and cell spreading but has no effect on actin binding nor on localization to focal adhesion plaques.</p> <p>Aceylated; mainly by myristic acid but also small amount of palmitic acid.</p>
<b>Cellular localization</b>	Cytoplasm > cytoskeleton. Cell junction > adherens junction. Cell membrane. Cytoplasmic face of

adhesion plaques. Recruitment to cell-cell junctions occurs in a myosin II-dependent manner. Interaction with CTNNB1 is necessary for its localization to the cell-cell junctions.

## Images



Western blot - Anti-Vinculin (phospho Y100) antibody (ab200812)

**All lanes :** Anti-Vinculin (phospho Y100) antibody (ab200812) at 1/1000 dilution

**Lane 1 :** HeLa whole cell lysate

**Lane 2 :** HeLa, treated with 100 ng/mL of LPS for 20 mins, whole cell lysate

Lysates/proteins at 30 µg per lane.

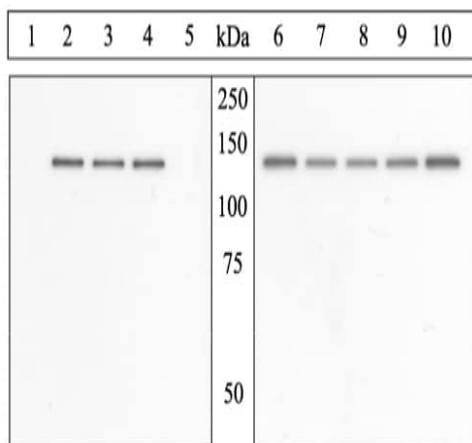
### Secondary

**All lanes :** Goat Anti-Rabbit IgG (H+L) Secondary Antibody, HRP conjugate at 1/5000 dilution

**Predicted band size:** 124 kDa

**Additional bands at:** 116 kDa (possible isoform), 124 kDa (possible isoform)

**Detection:** Chemiluminescence.



Western blot - Anti-Vinculin (phospho Y100) antibody (ab200812)

**Lanes 1-5 :** Anti-Vinculin (phospho Y100) antibody (ab200812) at 1/1000 dilution

**Lanes 6-10 :** Anti-Vinculin pan antibody

**Lane 1 :** COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA untreated

**Lanes 2 & 7-10 :** COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were treated with vanadate for 24 hr

**Lane 3 :** COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were treated with vanadate for 24 hr. with non-phosphopeptide corresponding to the immunogen

**Lane 4 :** COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were treated with vanadate for 24 hr with generic phosphotyrosine-containing peptide

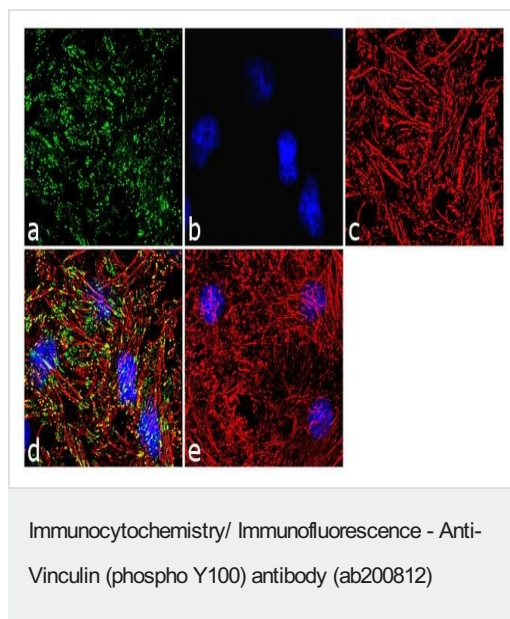
**Lane 5 :** COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were treated with vanadate for 24 hr with phosphopeptide immunogen

**Lane 6** : COS cells co-transfected with activated Src and His-tagged chicken Vinculin cDNA were untreated

Developed using the ECL technique.

**Predicted band size:** 124 kDa

Following immunoprecipitation of Vinculin with an anti-His monoclonal antibody, proteins were resolved by SDS-PAGE on an 8% polyacrylamide gel and transferred to PVDF.



Immunofluorescence analysis of Phospho-Vinculin pTyr100 was performed using 70% confluent log phase HeLa cells. The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, and blocked with 2% BSA for 1 hour at room temperature. The cells were labeled with Phospho-Vinculin pTyr100 Rabbit Polyclonal Antibody (ab200812) at 1/250 dilution in 0.1% BSA and incubated for 3 hours at room temperature and then labeled with Goat anti-Rabbit IgG (H+L) Superclonal™ Secondary Antibody, Alexa Fluor® 488 conjugate at a dilution of 1/2000 for 45 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with SlowFade® Gold Antifade Mountant with DAPI. F-actin (Panel c: red) was stained with Rhodamine Phalloidin. Panel d represents the merged image showing punctate membranous localization. Panel e shows the no primary antibody control. The images were captured at 60X magnification.

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

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