# abcam

## Product datasheet

# Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free ab248508

Recombinant RabMAb

# 15 Images

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**Product name** Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho

Y15) antibody [EPR7875] - BSA and Azide free

Rabbit monoclonal [EPR7875] to CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 **Description** 

(phospho Y15) + CDK5 (phospho Y15) - BSA and Azide free

**Host species** Rabbit

Specificity The antibody will cross-react with phosphorylated CDK1 (pY15), CDK2 (pY15), CDK3 (pY15) and

CDK5 (pY15) but not with non-phosphorylated CDK1, CDK2, CDK3 and CDK5. Please see our

ELISA results on the images section.

**Tested applications** Suitable for: ELISA, IHC-P, WB, Dot blot

Species reactivity Reacts with: Mouse, Rat, Human, Recombinant fragment

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

General notes ab248508 is the carrier-free version of ab133463.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

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.

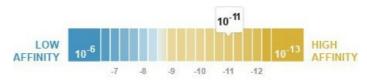
Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**<sup>®</sup> **patents**.

## **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

**Dissociation constant (K<sub>D</sub>)**  $K_D = 4.10 \times 10^{-11} M$ 



Learn more about K<sub>D</sub>

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR7875

**Isotype** IgG

#### **Applications**

## The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab248508 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
ELISA		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Detects a band of approximately 34 kDa (predicted molecular weight: 34 kDa).
Dot blot		Use at an assay dependent concentration.

## **Target**

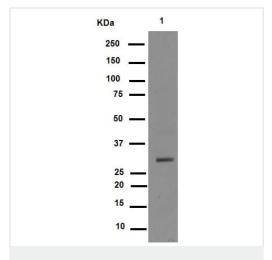
#### **Cellular localization**

CDK1: Nucleus. CDK5: Cytoplasm. Cell projection > lamellipodium. Cell projection > growth cone. In axonal growth cone with extension to the peripheral lamellipodia.

#### **Form**

CDK1: CDK1 can be located to the Nucleus, cytoplasm and Mithocondria. It's cytoplasmic during interphase and reversibly translocated from cytoplasm to the nucleus when phosphorilated before G2-M transition when associated with cyclin-B1. Accumulates in mitochondria in G2-arrested cells upon DNA-damage.

#### **Images**



Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (ab133463) at 1/1000 dilution (purified) + HeLa cell lysate treated with UV at 10  $\mu g$ 

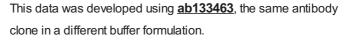
#### **Secondary**

HRP goat anti-rabbit (H+L) at 1/1000 dilution

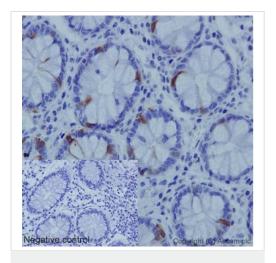
**Predicted band size:** 34 kDa **Observed band size:** 34 kDa

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

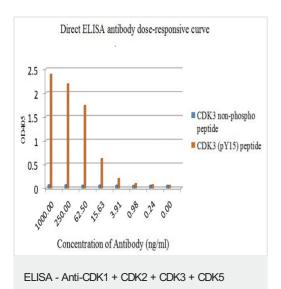
Blocking and dilution buffer: 5% NFDM/TBST.



Immunohistochemical staining of paraffin embedded human colon with purified <u>ab133463</u> at a working dilution of 1 in 75. The secondary antibody used is a HRP polymer for rabbit lgG. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

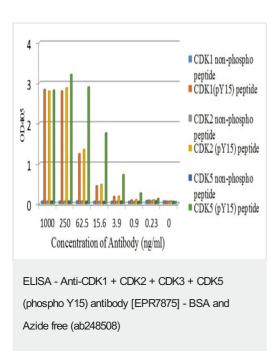


(phospho Y15) antibody [EPR7875] - BSA and

Azide free (ab248508)

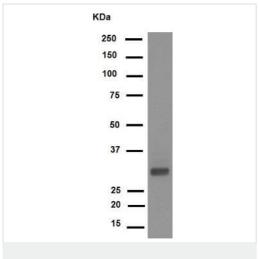
This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Direct ELISA antibody dose-response curve using purified <a href="mailto:ab133463">ab133463</a> at 0-1000 ng/ml. Antigen concentration of 1000 ng/mL. An alkaline phosphatase-conjugated goat anti-rabbit lgG (H+L) (1/2500) was used as the secondary antibody.

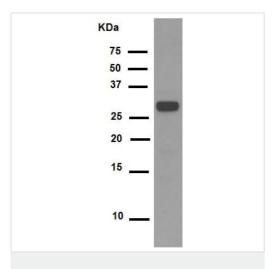


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Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)



Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (**ab133463**) at 1/2000 dilution (purified) + C6 cell lysate at 10 µg

#### Secondary

HRP goat anti-rabbit (H+L) at 1/1000 dilution

**Predicted band size:** 34 kDa **Observed band size:** 34 kDa

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (ab133463) at 1/2000 dilution (purified) + NIH/3T3 cell lysate at 10 µg

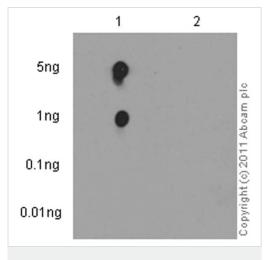
#### Secondary

HRP goat anti-rabbit (H+L) at 1/1000 dilution

**Predicted band size:** 34 kDa **Observed band size:** 34 kDa

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

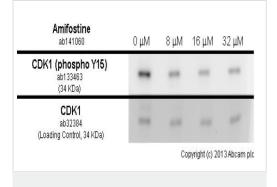
Blocking and dilution buffer: 5% NFDM/TBST.



Dot Blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Dot blot analysis of CDK1+CDK2+CDK3+CDK5 (pY15) phospho peptide (lane 1) and CDK1+CDK2+CDK3+CDK5 non-phospho peptide (lane 2) labelling CDK1+CDK2+CDK3+CDK5 (phospho Y15) with unpurified **ab133463** at a dilution of 1/1000. A peroxidase-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/2500). Blocking and dilution buffer: 5% NFDM/TBST. Exposure time: 10 seconds.

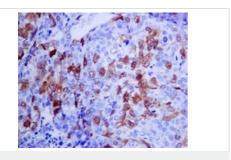


Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Saso2 cells were incubated at 37°C for 24 hours with vehicle control (0  $\mu$ M) and different concentrations of Amifostine (<u>ab141060</u>). Decreased expression of CDK1 (phospho Y15) (unpurified <u>ab133463</u>) in Saso2 cells correlates with an increase in Amifostine concentration, as described in literature.

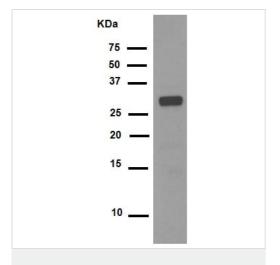
Whole cell lysates were prepared with RIPA buffer (containing protease inhibitors and sodium orthovanadate), 10 $\mu$ g of each were loaded on the gel and the WB was run under reducing conditions. After transfer the membrane was blocked for an hour using 5% BSA before being incubated with unpurified <u>ab133463</u> at 1  $\mu$ g/ml and <u>ab32384</u> at 1  $\mu$ g/ml overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP (<u>ab97051</u>) at 1/10000 and visualised using ECL development solution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin embedded human breast carcinoma tissue labelling CDK1+CDK2+CDK3+CDK5 with unpurified <u>ab133463</u> at 1/50 dilution. Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (ab133463) at 1/1500 dilution (unpurified) + NIH/3T3 cell lysate at 1/1000 dilution

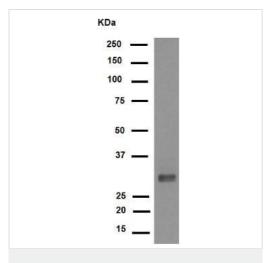
### Secondary

HRP goat anti-rabbit (H+L) at 1/1000 dilution

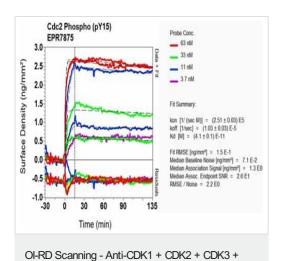
**Predicted band size:** 34 kDa **Observed band size:** 34 kDa

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

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



Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)



CDK5 (phospho Y15) antibody [EPR7875] - BSA

and Azide free (ab248508)

Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (**ab133463**) at 1/1500 dilution (unpurified) + C6 cell lysate at 10 μg

## **Secondary**

HRP goat anti-rabbit (H+L) at 1/1000 dilution

**Predicted band size:** 34 kDa **Observed band size:** 34 kDa

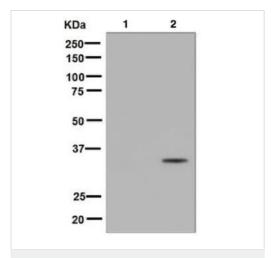
This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

This data was developed using  $\underline{ab133463}$ , the same antibody clone in a different buffer formulation. Equilibrium disassociation constant ( $K_D$ )

Learn more about K<sub>D</sub>

Click here to learn more about K<sub>D</sub>



Western blot - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

**All lanes :** Anti-CDK1 (phospho Y15) + CDK2 (phospho Y15) + CDK3 (phospho Y15) + CDK5 (phospho Y15) antibody [EPR (ab133463) at 1/1000 dilution (unpurified)

Lane 1: HeLa cell lysate

Lane 2: HeLa cell lysate treated with UV

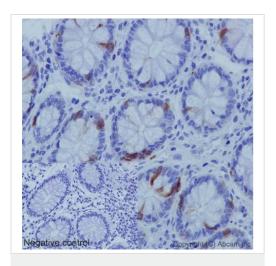
Lysates/proteins at 10 µg per lane.

### Secondary

All lanes: HRP labelled goat anti-rabbit lgG at 1/2000 dilution

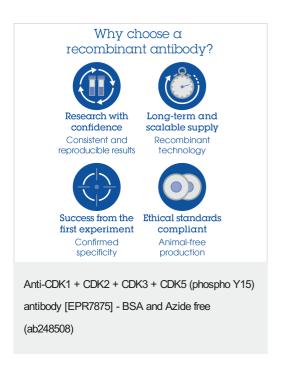
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Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CDK1 + CDK2 + CDK3 + CDK5 (phospho Y15) antibody [EPR7875] - BSA and Azide free (ab248508)

This data was developed using <u>ab133463</u>, the same antibody clone in a different buffer formulation.lmmunohistochemical staining of paraffin embedded human colon with unpurified <u>ab133463</u> at a working dilution of 1 in 50. The secondary antibody used is a HRP polymer for rabbit lgG. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



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