# abcam

#### Product datasheet

# Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] ab76495



Recombinant

RabMAb

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#### Overview

Product name Anti-Thymidine Kinase 1/TK1 antibody [EPR3193]

**Description**Rabbit monoclonal [EPR3193] to Thymidine Kinase 1/TK1

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, IHC-P

Species reactivity Reacts with: Human

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

Positive control WB: HEK-293T, Jurkat, HAP1, SW620, U-2 OS, and MOLT-4 cell lysates. Human tonsil tissue

lysate; IP: MOLT-4 whole cell lysate; IHC-P: Human tonsil tissue; Flow Cyt (intra): HeLa cells.

**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<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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

## **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: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

1

Clonality Monoclonal
Clone number EPR3193

**Isotype** IgG

## **Applications**

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab76495 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)		1/200. For unpurified use at 1/70 <u>ab172730</u> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB		1/1000. Detects a band of approximately 25 kDa (predicted molecular weight: 25 kDa).  For unpurified use at 1/5000 - 1/50000
IP		1/20.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.

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Sequence similarities

Belongs to the thymidine kinase family.

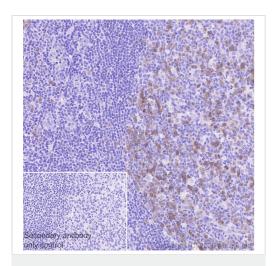
Post-translational modifications

Phosphorylated on Ser-13 in mitosis.

Cellular localization

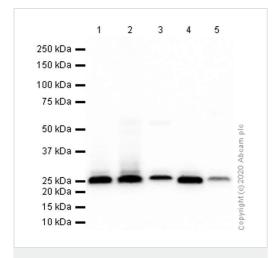
Cytoplasm.

#### **Images**



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human tonsil tissue sections labeling Thymidine Kinase 1/TK1 with purified ab76495 at 1/200 dilution (0.85 µg/mL). Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0). The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Western blot - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

**All lanes :** Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495) at 1/1000 dilution (Purified)

**Lane 1 :** MOLT-4 (Human lymphoblastic leukemia T lymphoblast) whole cell lysate

Lane 2: U-2 OS (Human bone osteosarcoma epithelial cell) whole cell lysate

**Lane 3 :** SW620 (Human colorectal adenocarcinoma epithelial cell) whole cell lysate

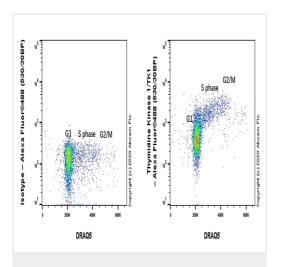
**Lane 4 :** Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate

Lane 5: Human tonsil lysate

#### Secondary

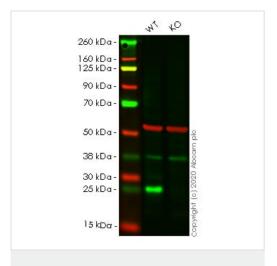
**All lanes :** Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 25 kDa



Flow Cytometry (Intracellular) - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Thymidine Kinase 1/TK1 with Purified ab76495 at 1/200 dilution (0.1  $\mu$ g/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor 488 ,ab150077) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Left). Unlabeled control - /.



Western blot - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

**All lanes :** Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495) at 1/1000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: TK1 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 25 kDa **Observed band size:** 25 kDa

**Lanes 1-2:** Merged signal (red and green). Green - ab76495 observed at 25 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab76495 was shown to react with Thymidine Kinase 1/TK1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line <u>ab266230</u> (knockout cell lysate <u>ab257745</u>)

was used. Wild-type HEK-293T and TK1 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab76495 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

260 kDa — 125 kDa — 25 kDa — 25 kDa — 15 kDa — 15 kDa — 15 kDa — 25 kDa — 2

Western blot - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

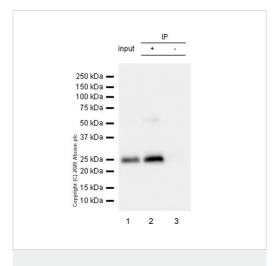
Lane 2: TK1 knockout HAP1 whole cell lysate (20 µg)

Lane 3: Jurkat whole cell lysate (20 µg)

Lane 4: MOLT4 whole cell lysate (20 µg)

**Lanes 1 - 4:** Merged signal (red and green). Green - ab76495 observed at 25 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab76495 was shown to specifically react with TK1 in wildtype HAP1 cells. No band was observed when TK1 knockout samples were examined. Wild-type and TK1 knockout samples were subjected to SDS-PAGE. Ab76495 and <a href="mailto:ab8245">ab8245</a> (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/5000 dilution and 1/10,000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216773">ab216773</a> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216776">ab216776</a> secondary antibodies at 1/20,000 dilution for 1 hour at room temperature before imaging.



Immunoprecipitation - Anti-Thymidine Kinase 1/TK1 antibody [EPR3193] (ab76495)

Purified ab76495 at 1/20 dilution (0.8µg) immunoprecipitating Thymidine Kinase 1/TK1 in MOLT-4 whole cell lysate.

Lane 1 (input): MOLT-4 (Human lymphoblastic leukemia T

Lane 2 (+): ab76495 + MOLT-4 whole cell lysate.

Lane 3 (-): Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab76495 in MOLT-4 whole cell lysate.

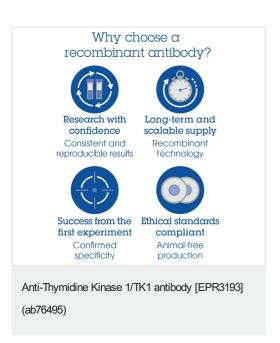
VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) (1/1000 dilution) was used for Western blotting.

Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

Observed band size: 24 kDa

lymphoblast)



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