# abcam

# Product datasheet

# Anti-Rb (phospho T356) antibody [EPR2153AY] ab76298



重组 RabMAb

**5 References** 10 图像

概述

产品名称 Anti-Rb (phospho T356)抗体[EPR2153AY]

描述 兔单克隆抗体[EPR2153AY] to Rb (phospho T356)

宿主 Rabbit

适用于: WB, IP, IHC-P, Dot blot 经测试应用

不适用于: Flow Cyt or ICC/IF

与反应: Human 种属反应性

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 Jurkat cell lysate treated with FBS Human breast carcinoma tissue HeLa cells IP: Jurkat whole

cell lysate.

常规说明 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**.

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

these species. Please contact us for more information.

性能

形式 Liquid

存放说明 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

存储溶液 pH: 7.20

Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

纯度 Protein A purified

**克隆** 单克隆

**克隆编号** EPR2153AY

**同种型** IgG

#### 应用

# The Abpromise guarantee Abpromise™承诺保证使用ab76298于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应 <b>用</b>	Ab评论	说明
WB		1/2000. Detects a band of approximately 110 kDa (predicted molecular weight: 106 kDa).
IP		1/20.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Dot blot		Use at an assay dependent concentration.

应用说明 Is unsuitable for Flow Cyt or ICC/IF.

#### 靶标

功能

Key regulator of entry into cell division that acts as a tumor suppressor. Promotes G0-G1 transition when phosphorylated by CDK3/cyclin-C. Acts as a transcription repressor of E2F1 target genes. The underphosphorylated, active form of RB1 interacts with E2F1 and represses its transcription activity, leading to cell cycle arrest. Directly involved in heterochromatin formation by maintaining overall chromatin structure and, in particular, that of constitutive heterochromatin by stabilizing histone methylation. Recruits and targets histone methyltransferases SUV39H1, KMT5B and KMT5C, leading to epigenetic transcriptional repression. Controls histone H4 'Lys-20' trimethylation. Inhibits the intrinsic kinase activity of TAF1. Mediates transcriptional repression by SMARCA4/BRG1 by recruiting a histone deacetylase (HDAC) complex to the c-FOS promoter. In resting neurons, transcription of the c-FOS promoter is inhibited by BRG1-dependent recruitment of a phospho-RB1-HDAC1 repressor complex. Upon calcium influx, RB1 is dephosphorylated by calcineurin, which leads to release of the repressor complex (By similarity). In case of viral infections, interactions with SV40 large T antigen, HPV E7 protein or adenovirus E1A protein induce the disassembly of RB1-E2F1 complex thereby disrupting RB1's activity.

组织**特异性** Expressed in the retina.

疾病相关 Childhood cancer retinoblastoma

Bladder cancer Osteogenic sarcoma

序列相似性 Belongs to the retinoblastoma protein (RB) family.

结构域 The Pocket domain binds to the threonine-phosphorylated domain C, thereby preventing

interaction with heterodimeric E2F/DP transcription factor complexes.

翻译后修饰 Phosphorylated by CDK6 and CDK4, and subsequently by CDK2 at Ser-567 in G1, thereby

releasing E2F1 which is then able to activate cell growth. Dephosphorylated at the late M phase.

SV40 large T antigen, HPV E7 and adenovirus E1A bind to the underphosphorylated, active form of pRb. Phosphorylation at Thr-821 and Thr-826 promotes interaction between the C-terminal domain C and the Pocket domain, and thereby inhibits interactions with heterodimeric E2F/DP transcription factor complexes. Dephosphorylated at Ser-795 by calcineruin upon calcium stimulation. CDK3/cyclin-C-mediated phosphorylation at Ser-807 and Ser-811 is required for G0-G1 transition. Phosphorylated by CDK1 and CDK2 upon TGFB1-mediated apoptosis.

N-terminus is methylated by METTL11A/NTM1 (By similarity). Monomethylation at Lys-810 by SMYD2 enhances phosphorylation at Ser-807 and Ser-811, and promotes cell cycle progression. Monomethylation at Lys-860 by SMYD2 promotes interaction with L3MBTL1.

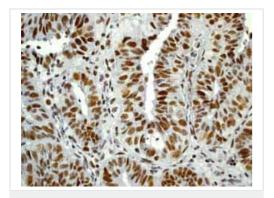
Acetylation at Lys-873 and Lys-874 regulates subcellular localization, at least during keratinocytes

细胞定位

Nucleus.

differentiation.

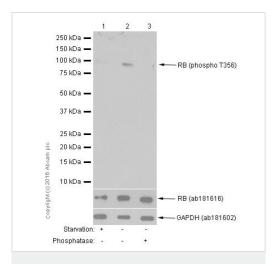
#### 图片



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

ab76298 showing positive staining in Endometrial carcinoma tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

**All lanes :** Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298) at 1/4000 dilution

**Lane 1 :** K562 (Human chronic myelogenous leukemia cell line from bone marrow) 2 day serum starved whole cell lysate.

**Lane 2**: K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate.

**Lane 3 :** K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate. Membrane incubated with phosphatase.

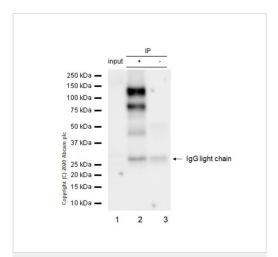
Lysates/proteins at 15 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

**Predicted band size:** 106 kDa **Observed band size:** 106 kDa

Exposure time: 1 minute



Immunoprecipitation - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298) Purified ab76298 at 1/20 dilution (0.5 $\mu$ g) immunoprecipitating Rb in Jurkat whole cell lysate.

Lane 1 (input): Jurkat (Human T cell leukemia T lymphocyte) whole cell lysate 10µg

Lane 2 (+): ab76298 + Jurkat whole cell lysate.

Lane 3 (-): Rabbit monoclonal lgG (<u>ab172730</u>) instead of ab76298 in Jurkat whole cell lysate.

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

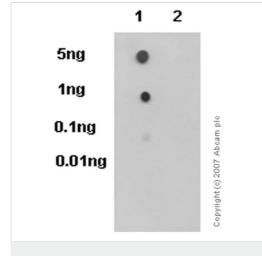
Blocking Buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM/TBST.

Cleavage fragments were observed as described in literature.

(PMID: 11420704)

Observed band size: 110 kDa

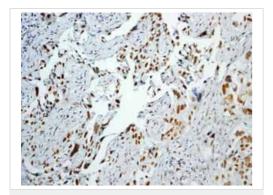


Dot Blot - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

Dot blot analysis of Rb (phospho T356) phospho peptide (Lane 1), Rb Non-phospho peptide (Lane 2), labelling Rb (phospho T356) with ab76298 at a dilution of 1/1000. Peroxidase conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody at a dilution of 1/2500.

Blocking and dilution buffer: 5% NFDM/TBST.

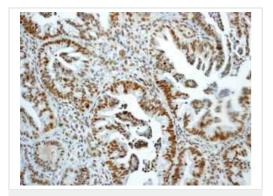
Exposure time: 3 minutes.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

ab76298 showing positive staining in Lung adenocarcinoma tissue.

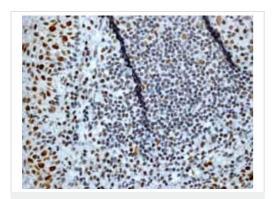
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

ab76298 showing positive staining in Ovarian carcinoma tissue.

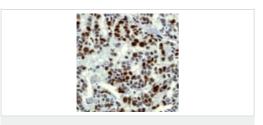
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

ab76298 showing positive staining in Normal tonsil tissue.

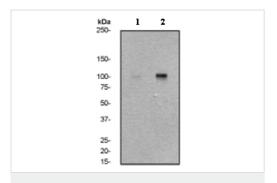
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298)

Immunohistochemical analysis of paraffin-embedded human breast carcinoma using ab76298 at 1/100-1/250 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298) **All lanes :** Anti-Rb (phospho T356) antibody [EPR2153AY] (ab76298) at 1/2000 dilution

Lane 1: Jurkat cell lysate

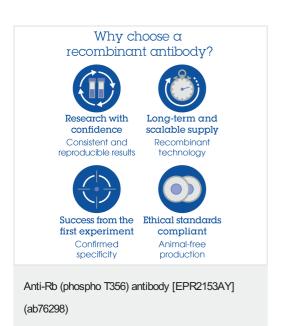
Lane 2: Jurkat cell lysate treated with FBS

Lysates/proteins at 10 µg per lane.

### **Secondary**

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

**Predicted band size:** 106 kDa **Observed band size:** 106 kDa



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