### abcam

### Product datasheet

### Anti-c-Myc (phospho S62) antibody [EPR17924] ab185656



重组 RabMAb

36 References 11 图像

概述

产品名称 Anti-c-Myc (phospho S62)抗体[EPR17924]

描述 兔单克隆抗体[EPR17924] to c-Myc (phospho S62)

宿主 Rabbit

经测试应用 适用于: Dot blot, IHC-P, ICC/IF, IP, WB, Flow Cyt (Intra)

种属反应性 与反应: Mouse, Rat, Human

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

阳性对照 WB: HeLa, NIH/3T3 and C6 whole cell lysates. IHC-P: Human endometrium cancer, mouse

spleen and rat testis tissues. ICC/IF: HeLa cells. IP: HeLa whole cell lysate treated with 200nM

TPA for 10 minutes. Flow Cyt (intra): HeLa cells.

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**.

性能

形式 Liquid

存放说明 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.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

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

纯度 Protein A purified

克隆 单克隆

克隆编号 EPR17924

**同种型** IgG

### 应用

### The Abpromise guarantee

### Abpromise™承诺保证使用ab185656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。

应用	Ab评论	说明
Dot blot		1/1000.
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/1000.
IP		1/50.
WB		1/2000. Detects a band of approximately 57 kDa (predicted molecular weight: 48 kDa).
Flow Cyt (Intra)		1/150.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.

#### 靶标

### 功能

## Participates in the regulation of gene transcription. Binds DNA in a non-specific manner, yet also specifically recognizes the core sequence 5'-CAC[GA]TG-3'. Seems to activate the transcription of growth-related genes.

### 疾病相关

# Note=Overexpression of MYC is implicated in the etiology of a variety of hematopoietic tumors. Note=A chromosomal aberration involving MYC may be a cause of a form of B-cell chronic lymphocytic leukemia. Translocation t(8;12)(q24;q22) with BTG1.

Defects in MYC are a cause of Burkitt lymphoma (BL) [MIM:113970]. A form of undifferentiated malignant lymphoma commonly manifested as a large osteolytic lesion in the jaw or as an abdominal mass. Note=Chromosomal aberrations involving MYC are usually found in Burkitt lymphoma. Translocations t(8;14), t(8;22) or t(2;8) which juxtapose MYC to one of the heavy or light chain immunoglobulin gene loci.

### 序列相似性

#### 翻译后修饰

Contains 1 basic helix-loop-helix (bHLH) domain.

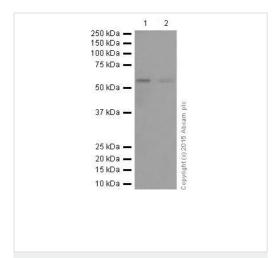
Phosphorylated by PRKDC. Phosphorylation at Thr-58 and Ser-62 by GSK3 is required for ubiquitination and degradation by the proteasome.

Ubiquitinated by the SCF(FBXW7) complex when phosphorylated at Thr-58 and Ser-62, leading to its degradation by the proteasome. In the nucleoplasm, ubiquitination is counteracted by USP28, which interacts with isoform 1 of FBXW7 (FBW7alpha), leading to its deubiquitination and preventing degradation. In the nucleolus, however, ubiquitination is not counteracted by USP28, due to the lack of interaction between isoform 4 of FBXW7 (FBW7gamma) and USP28, explaining the selective MYC degradation in the nucleolus. Also polyubiquitinated by the DCX(TRUSS) complex.

形式

c-Myc is also expressed in the cytoplasm.

### 图片



Western blot - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656) **All lanes :** Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656) at 1/2000 dilution

Lane 1: NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysate

Lane 2: C6 (Rat glial tumor cells) whole cell lysate

Lysates/proteins at 10 µg per lane.

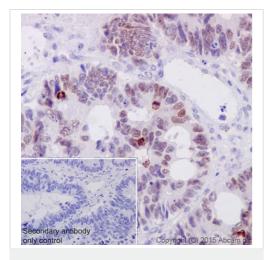
### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG, (H+L),Peroxidase conjugated at 1/1000 dilution

Predicted band size: 48 kDa
Observed band size: 57 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

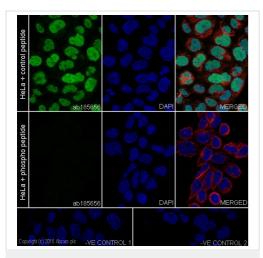
Immunohistochemical analysis of paraffin-embedded Human endometrium cancer tissue labeling c-Myc (phospho S62) with ab185656 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Nuclear staining on cancer cells of Human endometrial cancer is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Antic-Myc (phospho S62) antibody [EPR17924] (ab185656)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling c-Myc (phospho S62) with ab185656 at 1/1000 dilution, followed by Goat anti-rabbit lgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear staining on HeLa cells. The staining decreased after blocking with phospho peptide  $(100\mu g/ml)$  overnight.

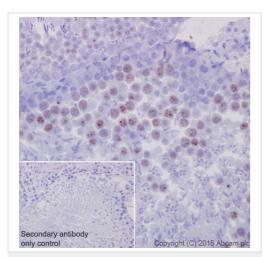
The control peptide is a non-phospho peptide.

The nuclear counter stain is DAPI (blue).

Tubulin is detected with <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution and <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

-ve control 1: ab185656 at 1/1000 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

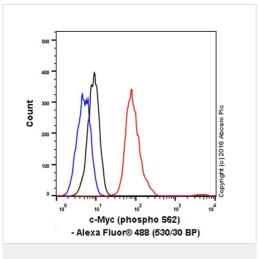
Immunohistochemical analysis of paraffin-embedded Rat testis tissue labeling c-Myc (phospho S62) with ab185656 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Nuclear staining on rat testis is observed.

Counter stained with Hematoxylin.

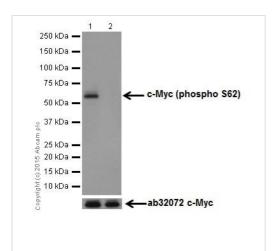
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling c-Myc (phospho S62) with purified ab185656 at 1/150 dilution (red). The secondary antibody was Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 488) at 1/2000 dilution. A Rabbit monoclonal lgG (Black) was used as the isotype control and cells without incubation with primary antibody and secondary antibody (Blue) were used as unlabeled control.



Western blot - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

**All lanes :** Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656) at 1/5000 dilution

**Lane 1 :** HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate

Lane 2: HeLa (Human epithelial cells from cervix adenocarcinoma) treated with Lambda Phosphatase whole cell lysate

Lysates/proteins at 10 µg per lane.

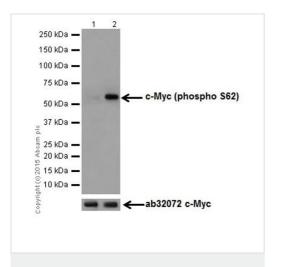
### **Secondary**

**All lanes :** Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

**Predicted band size:** 48 kDa **Observed band size:** 57 kDa

Exposure time: 1 minute

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

**All lanes :** Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656) at 1/5000 dilution

Lane 1: Untreated HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate

Lane 2: HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate treated with 200nM Calyculin A and 1uM Okadaic Acid for 60 minutes.

Lysates/proteins at 10 µg per lane.

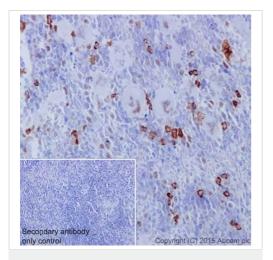
### **Secondary**

**All lanes :** Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

**Predicted band size:** 48 kDa **Observed band size:** 57 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

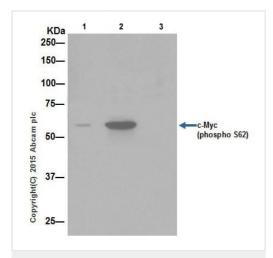
Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling c-Myc (phospho S62) with ab185656 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution.

Nuclear and cytoplasmic staining on mouse spleen is observed.

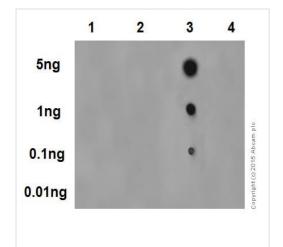
Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)



Dot Blot - Anti-c-Myc (phospho S62) antibody [EPR17924] (ab185656)

c-Myc (phospho S62) was immunoprecipitated from 1mg of HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate treated with 200nM TPA for 10 minutes with ab185656 at 1/50 dilution.

Western blot was performed from the immunoprecipitate using ab185656 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (<u>ab131366</u>) was used for detection at 1/1500 dilution.

Lane 1: HeLa whole cell lysate treated with 200nM TPA for 10 minutes,10  $\mu g$  (Input).

Lane 2: ab185656 IP in HeLa whole cell lysate treated with 200nM TPA for 10 minutes.

Lane 3: Rabbit monoclonal  $\lg G (\underline{ab172730})$  instead of ab185656 in HeLa whole cell lysate treated with 200nM TPA for 10 minutes.

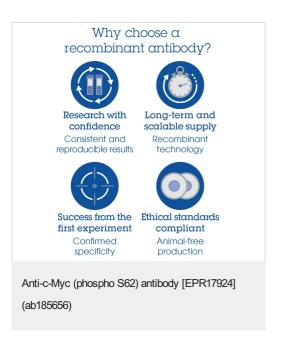
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Dot blot analysis of c -Myc (phospho T58) peptide (Lane 1), c-Myc non-phospho peptide (a control peptide for c-Myc phospho T58) (Lane 2), c-Myc (phospho S62) peptide (Lane 3), and c-Myc non-phospho peptide (a control peptide for c-Myc phospho S62) (Lane 4), labeled using ab185656 at 1/1000 dilution, followed by Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated secondary antibody at 1/1000 dilution.

Blocking/Dilution buffer: 5% NFDM/TBST.

Lanes 1, 2 and 4 are control peptides, lane 3 contains the immunogen peptide.

Exposure time=3 minutes.



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