

Product datasheet

Anti-c-Myc (phospho T58) antibody [EPR17923] - BSA and Azide free ab236021

Recombinant RabMAb

4 Images

Overview		
Product name	Anti-c-Myc (phospho T58) antibody [EPR17923] - BSA and Azide free	
Description	Rabbit monoclonal [EPR17923] to c-Myc (phospho T58) - BSA and Azide free	
Host species	Rabbit	
Tested applications	Suitable for: ICC/IF, WB, Flow Cyt (Intra), Dot blot	
Species reactivity	Reacts with: Human	
	Predicted to work with: Mouse, Rat	
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.	
Positive control	WB: HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate, HeLa cells treated with 200nM Calyculin A and 1uM Okadaic Acid for 60 minutes whole cell lysate. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa cells.	
General notes	ab236021 is the carrier-free version of <u>ab185655</u> .	
	Our <u>carrier-free</u> 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 cell-based 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 [®] 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[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb[®] patents</u>.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR17923
lsotype	lgG

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab236021 in the following tested applications.

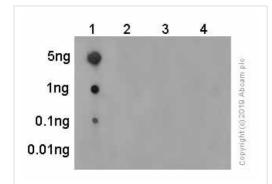
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 57 kDa (predicted molecular weight: 49 kDa).
Flow Cyt (Intra)		Use at an assay dependent concentration.
Dot blot		Use at an assay dependent concentration.

Target	
Function	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.
Involvement in disease	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.
Sequence similarities	Contains 1 basic helix-loop-helix (bHLH) domain.

Post-translational modifications	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.
Cellular localization	Nucleus > nucleoplasm. Nucleus > nucleolus.
Form	c-Myc is also expressed in the cytoplasm.

Images



Dot Blot - Anti-c-Myc (phospho T58) antibody [EPR17923] - BSA and Azide free (ab236021) Lane 1: c-Myc (phospho T58).

Lane 2: c-Myc (pT58) non-phospho peptide.

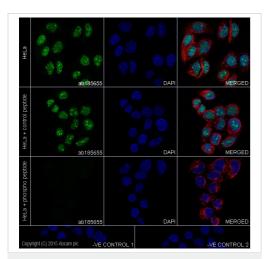
Lane 3: c-Myc (pS62) phospho peptide.

Lane 4: c-Myc (pS62) non-phospho peptide.

Dot blot analysis using <u>**ab185655**</u> at a dilution of 1/1000. Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (<u>**ab97051**</u>) was used as the secondary antibody at a dilution of 1/100000.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



Immunocytochemistry/ Immunofluorescence - Antic-Myc (phospho T58) antibody [EPR17923] - BSA and Azide free (ab236021) Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa cells (Human epithelial cells from cervix adenocarcinoma) labeling c-Myc (phospho T58) with <u>ab185655</u> at 1/250, followed by Goat anti-rabbit IgG (Alexa Fluor[®] 488) (<u>ab150077</u>) secondary antibody at 1/500 (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 counterstain is DAPI (blue).

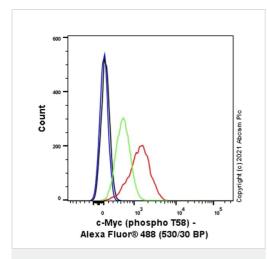
Tubulin is detected with Anti-alpha Tubulin antibody -Loading Control (<u>ab7291</u>) at 1/1000 dilution Goat Anti-Mouse IgG (AlexaFluor[®]594) preadsorbed (<u>ab150120</u>) at 1/500 (red).

The negative controls are as follows:-

-ve control 1 - <u>ab185655</u> at 1/500 followed by <u>ab150120</u> at 1/500.
-ve control 2 -<u>ab7291</u> at 1/1000 followed by <u>ab150077</u> at 1/500.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and

sodium azide (ab185655).



Flow Cytometry (Intracellular) - Anti-c-Myc (phospho T58) antibody [EPR17923] - BSA and Azide free (ab236021)



BSA and Azide free (ab236021)

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This data was developed using <u>ab185655</u>, the same antibody clone in a different buffer formulation.

Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) treated with 200nM Calyculin A and 1uM Okadaic Acid for 60 min cells labeling c-Myc with purified **ab185655** at 1/700 dilution (1 µg/mL) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit IgG (Alexa Fluor[®] 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue). If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <u>https://www.abcam.com/abpromise</u> or contact our technical team.

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