

# Datasheet for ABIN1532205

# anti-c-MYC antibody (AA 31-80)

2 Validations

3 Images



Go to Product page

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Quantity:	100 μL	
Target:	c-MYC (MYC)	
Binding Specificity:	AA 31-80	
Reactivity:	Human, Mouse, Rat	
Host:	Rabbit	
Clonality:	Polyclonal	
Conjugate:	This c-MYC antibody is un-conjugated	
Application:	Western Blotting (WB), Immunohistochemistry (IHC), ELISA, Immunofluorescence (IF)	
Product Details		
Immunogen:	The antiserum was produced against synthesized peptide derived from human MYC.	
Isotype:	IgG	
Specificity:	MYC Antibody detects endogenous levels of total MYC protein.	
Purification:	The antibody was purified from rabbit antiserum by affinity-chromatography using immunogen.	
Purity:	> 95 %	
Target Details		
Target:	c-MYC (MYC)	
Alternative Name:	Myc Proto-Oncogene protein (MYC Products)	
Background:	Synonyms: Myc proto-oncogene protein, Class E basic helix-loop-helix protein 39, bHLHe39,	

### **Target Details**

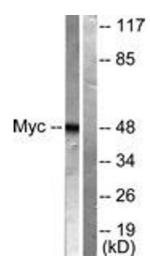
	Proto-oncogene c-Myc, Transcription factor p64 , MYC , BHLHE39  NCBI Gene Symbol: MYC
Molecular Weight:	48 kDa
Gene ID:	4609
OMIM:	113970
UniProt:	P01106
Pathways:	p53 Signaling, Cell Division Cycle, Sensory Perception of Sound, Transition Metal Ion Homeostasis, Mitotic G1-G1/S Phases, Positive Regulation of Endopeptidase Activity, Regulation of Carbohydrate Metabolic Process, Positive Regulation of Response to DNA Damage Stimulus, Warburg Effect

### **Application Details**

Expiry Date:

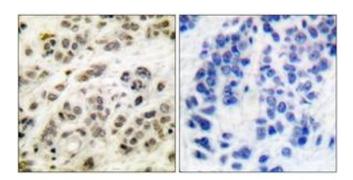
Application Details		
Application Notes:	es: WB: 1:500~1:1000 IHC: 1:50~1:100 IF: 1:100~1:500 ELISA: 1:40000	
Comment:	Unigene-Number: Hs.202453 (NCBI Gene Symbol: MYC)	
Restrictions:	For Research Use only	
Handling		
Format:	Liquid	
Concentration:	1 mg/mL	
Buffer:	phosphate buffered saline (without Mg2+ and Ca2+), pH 7.4, 150 mM NaCl, 0.02 % sodium azide and 50 % glycerol.	
Preservative:	Sodium azide	
Precaution of Use:	This product contains sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which	

12 months



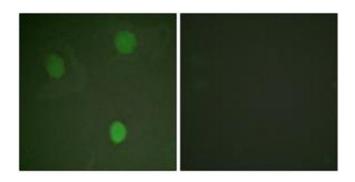
#### **Western Blotting**

**Image 1.** Western blot analysis of extracts from HeLa cells, treated with Forskolin 40nM 30', using MYC (Ab-62) Antibody. The lane on the right is treated with the synthesized peptide.



#### **Immunohistochemistry**

**Image 2.** Immunohistochemistry analysis of paraffinembedded human breast carcinoma tissue, using MYC (Ab-62) Antibody. The picture on the right is treated with the synthesized peptide.



#### Immunofluorescence

**Image 3.** Immunofluorescence analysis of HeLa cells, using MYC (Ab-62) Antibody. The picture on the right is treated with the synthesized peptide.





#### Successfully validated (Western Blotting (WB))

by Developmental Biology, Johann-Friedrich-Blumenbach-Institute for Zoology and

Anthropology, Georg-August-University of Göttingen

Report Number: 102753

Date: Jul 03 2018

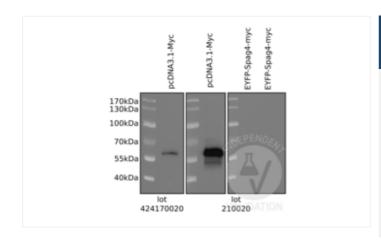
Target:	MYC
Lot Number:	210020 and 424170020
Method validated:	Western Blotting (WB)
Positive Control:	HEK-293 cells transfected with Myc expression plasmid
Negative Control:	HEK-293 cells transfected with HA-Spag4-myc expression plasmid
Notes:	Passed. ABIN1532205 detects Myc protein in a western blot. No cross-reaction was observed
	with the myc-tag sequence positioned at the C-terminus of a recombinant protein.
Primary Antibody:	ABIN1532205
Secondary Antibody:	rabbit anti-mouse IgG, HRP-linked (Sigma A9044, Lot 034M4761)
Protocol:	<ul> <li>Grow HEK-293 cells (ATCC, CRL-1658) in DMEM+GlutaMAXDMEM+GlutaMAX (Gibco, 10567-014, lot 1922818) supplemented with fetal bovine serum (Gibco, 10500-064, lot 08F0477K) and Pen/Strep (Gibco, 15140-122, lot 1924797) at 37°C and 5% CO<sub>2</sub> to 70% confluency.</li> <li>Transfect cells either with a Myc expression plasmid (pcDNA3.1-Myc) or HA- and myctagged SPAG4 expression plasmids using EndofectinMax (GeneCopoeia) following the manufacturer's instructions.</li> <li>Grow cells for 24h.</li> <li>Lyse cells in SDS-sample buffer and denature total cellular lysates for 5min at 95°C. Subsequently separate them on a denaturing 10% SDS-PAGE (Laemmli 1970).</li> <li>Transfer proteins onto 0.2µm Protran membrane (GE Healthcare, 10600004, A10043108) by wet blotting for 1h at 400mA (Towbin et al., 1979).</li> <li>Block the membrane in TBST (50mM Tris-HCl, pH7.4, 150mM NaCl, 0.2% Tween 20) containing 5% milk (blocking solution) for 60min at RT.</li> <li>Incubate membrane with primary rabbit anti-myc antibody (antibodies-online, ABIN1532205 Lot#424170020 and Lot#210020) diluted 1:500 in blocking solution overnight at 4°C.</li> <li>Wash membrane with TBST for 45min at RT.</li> <li>Incubate membrane with secondary goat anti-rabbit IgG (H+L), HRP-linked (Jackson Immuno Research, 111-035-003, lot 123450) diluted 1:1000 in blocking solution for 45min at RT.</li> <li>Wash membrane 6x for 5min with TBST.</li> <li>Reveal protein bands using Clarity Max Western ECL substrate (Bio-Rad, 1705062) and</li> </ul>

- Incubate membrane with primary mouse anti-HA-tag antibody (clone 12CA5) diluted 1:500 in blocking solution overnight at 4°C.
- · Wash membrane with TBST for 45min at RT.
- · Incubate membrane with secondary rabbit anti-mouse IgG, HRP-linked (Sigma A9044, Lot 034M4761) diluted 1:1000 in blocking solution for 45min at RT.
- Reveal protein bands using Clarity Max Western ECL substrate (Bio-Rad, 1705062).

#### **Experimental Notes:**

In lysates of HEK-293 cells ectopically expressing recombinant protein with an N-terminal HAand a C-terminal myc-tag ABIN1532205 did not detect the labeled protein at the expected molecular weight. In contrast, an anti-HA-tag antibody (clone 12CA5) revealed a protein at the expected MW.

#### Image for Validation report #102753



## Validation image no. 1 for anti-Myc Proto-Oncogene protein (MYC) (AA 31-80) antibody (ABIN1532205)

ABIN1532205 lot 424170020 (let panel) and lot 210020 (middle and right panel) detected a protein in the expected molecular mass range exclusively when Myc is expressed (pcDNA3.1-Myc) but did not react with the Myc-tagged SPAG4 (EYFP-Spag4-myc; expected MW approximately 50kDa).





#### Successfully validated (Immunocytochemistry (ICC))

by Developmental Biology, Johann-Friedrich-Blumenbach-Institute for Zoology and Anthropology, Georg-August-University of Göttingen

Report Number: 103023

Date: Jul 03 2018

Target:	MYC	
Lot Number:	210020 and 424170020	
Method validated:	Immunocytochemistry (ICC)	
Positive Control:	HEK-293 cells transfected with Myc expression plasmid	
Negative Control:	HEK-293 cells transfected with HA-Spag4-myc expression plasmid	
Notes:	Passed. ABIN1532205 (lot 424170020 and lot 210020) detects Myc in immunocytochemistry. No cross-reaction was observed with the myc-tag sequence positioned at the C-terminus of a recombinant protein.	
Primary Antibody:	ABIN1532205	
Secondary Antibody:	goat anti-rabbit IgG MFP590 (Mobitec MFP-A1037, lot 3002088), F(ab')2-Goat anti-Rabbit IgG (H+L) Alexa Fluor 555 (Life Technologies, A21430, lot 1637266)	
Protocol:	<ul> <li>Grow HEK-293 cells (ATCC, CRL-1658) in DMEM+GlutaMAXDMEM+GlutaMAX (Gibco, 10567-014, lot 1922818) supplemented with fetal bovine serum (Gibco, 10500-064, lot 08F0477K) and Pen/Strep (Gibco, 15140-122, lot 1924797) at 37°C and 5% CO2 to 70% confluency.</li> <li>Transfect cells either with a Myc expression plasmid (pcDNA3.1-Myc) or HA- and myctagged SPAG4 expression plasmids using EndofectinMax (GeneCopoeia) following the manufacturer's instructions.</li> <li>Grow cells for 24h.</li> <li>Fix cells in 3.7% paraformaldehyde (in PBS) for 20min at 4°C followed by incubation in 0.3% Triton X-100 for 10min at 4°C.</li> <li>Block cells in PBS containing 1% BSA and 0.5% Tween-20 (PBT) for 1h at RT.</li> <li>Incubate cells with primary antibody</li> <li>rabbit anti-Myc antibody (antibodies-online, ABIN1532205, lot 424170020 or lot 210020) diluted 1:50 in PBS and mouse anti-HA tag antibody (clone 12CA5) ON at 4°C (images G, H).</li> <li>rabbit anti-Myc antibody (antibodies-online, ABIN1532205, lot 210020 or lot 210020) diluted 1:50 in PBS ON at 4°C (images A, D).</li> <li>Wash cells with 50mM Tris-HCl, pH7.4, 150mM NaCl, 0.1% Tween 20 (TBST) for 15min at RT.</li> </ul>	

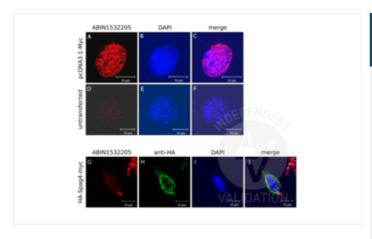
Incubate cells with secondary antibody

- goat anti-rabbit IgG MFP590 (Mobitec MFP-A1037, lot 3002088) diluted 1:300 in PBS and goat anti-mouse IgGDylight488 (Thermo Scientific, 35503) diluted 1:10000 in PBS for 1h at 37°C (images G, H).
- ∘ F(ab')2-Goat anti-Rabbit IgG (H+L) Alexa Fluor 555 (Life Technologies, A21430, lot 1637266) diluted 1:2000 in PBS for 45min at 37°C (images A, D).
- · Counterstain cells with DAPI (Sigma, D-9542).
- Image acquisition on Zeiss LSM 510 confocal microscope and processing using Adobe Photoshop 5.0.

**Experimental Notes:** 

The antibody detects Myc proteins by immunoblotting and by immunocytology. No crossreaction with the C-terminal Myc-tag.

#### Image for Validation report #103023



## Validation image no. 1 for anti-Myc Proto-Oncogene protein (MYC) (AA 31-80) antibody (ABIN1532205)

ABIN1532205 detects ectopically expressed Myc (pcDNA3.1-Myc) (A, red). No cross-reactivity is observed in untransfected cells (D). DAPI was used to visualize the nucleus (B and E, blue). Images on the right show the merged red and blue channels. ABIN1532205 did not detect the myc-tag (G, red) of HA-Spag4-myc whereas an anti HAtag antibody (H, green) detected HA-Spag4-myc at the nuclear membrane as expected. The picture on the right (J) shows the red and green channels merged with DAPI counterstain (I, blue).