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# Datasheet for ABIN6261511 anti-ELF1 antibody (C-Term)

1 Validation

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## Overview

Quantity:	100 µL
Target:	ELF1
Binding Specificity:	C-Term
Reactivity:	Human
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This ELF1 antibody is un-conjugated
Application:	Western Blotting (WB), ELISA, Immunofluorescence (IF), Immunocytochemistry (ICC)

# Product Details

Immunogen:	A synthesized peptide derived from human ELF1, corresponding to a region within C-terminal amino acids.
Isotype:	lgG
Specificity:	ELF1 Antibody detects endogenous levels of total ELF1.
Predicted Reactivity:	Horse
Purification:	The antiserum was purified by peptide affinity chromatography using SulfoLink <sup>TM</sup> Coupling Resin (Thermo Fisher Scientific).
Target Details	

Target:

ELF1

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# Target Details

Alternative Name:	ELF1 (ELF1 Products)
Background:	Description: Transcription factor that activates the LYN and BLK promoters. Appears to be required for the T-cell-receptor-mediated trans activation of HIV-2 gene expression. Binds specifically to two purine-rich motifs in the HIV-2 enhancer. Gene: ELF1
Molecular Weight:	67kDa
Gene ID:	1997
UniProt:	P32519

# Application Details

Application Notes:	WB 1:500-1:2000, IF/ICC 1:100-1:500, ELISA(peptide) 1:20000-1:40000
Restrictions:	For Research Use only

# Handling

Format:	Liquid
Concentration:	1 mg/mL
Buffer:	Rabbit IgG in phosphate buffered saline , 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 should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Store at -20 °C. Stable for 12 months from date of receipt.
Expiry Date:	12 months

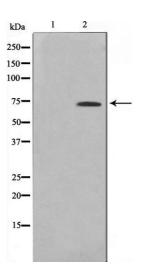
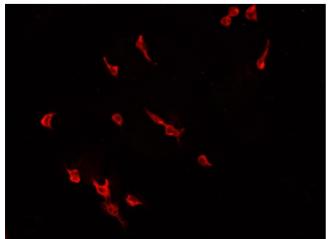
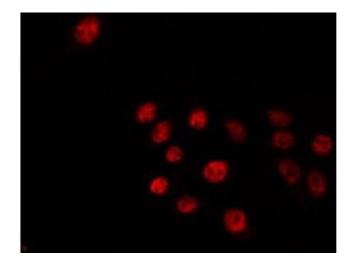




Image 1. Western blot analysis on 293 cell lysate using ELF1 Antibody





#### Immunofluorescence (fixed cells)

**Image 2.** ABIN6266812 staining CACO-2 cells by IF/ICC. The sample were fixed with PFA and permeabilized in 0.1% Triton X-100,then blocked in 10% serum for 45 minutes at 25jãC. The primary antibody was diluted at 1/200 and incubated with the sample for 1 hour at 37jãC. An Alexa Fluor 594 conjugated goat anti-rabbit IgG (H+L) antibody(Cat.# S0006), diluted at 1/600, was used as secondary antibod

#### Immunofluorescence (fixed cells)

**Image 3.** ABIN6266812 staining 293 by IF/ICC. The sample were fixed with PFA and permeabilized in 0.1% Triton X-100,then blocked in 10% serum for 45 minutes at 25<sub>j</sub>ãC. The primary antibody was diluted at 1/200 and incubated with the sample for 1 hour at 37<sub>j</sub>ãC. An Alexa Fluor 594 conjugated goat anti-rabbit IgG (H+L) Ab, diluted at 1/600, was used as the secondary antibod

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

by Dittmann Lab, Microbiology Department, NYU Langone Health, NYU School of Medicine Report Number: 104221 Date: Oct 28 2019

Target:	ELF1
Lot Number:	4318850
Method validated:	Western Blotting (WB)
Positive Control:	A549 (endogenous ELF1), ELF1 recombinant protein
Notes:	Passed. ABIN6261511 recognizes recombinant and endogenous human ELF1. The antibody
	does also reveal an additional larger MW protein band in A549 cell lysates.
Primary Antibody:	ABIN6261511
Secondary Antibody:	goat anti-rabbit HRP-conjugated antibody (Invitrogen, G21234)
Protocol:	<ul> <li>Grow A549 cells (ATCC, CCL-185) in DMEM medium (Corning, 10-013-CV) supplemented with 10% FBS (Atlanta biologicals, S11150) and Penicillin-Streptomycin (Corning, 30-002-CI) at 37°C and 5% CO<sub>2</sub> to 3x10<sup>4</sup> cells/cm<sup>2</sup> in 2ml on a 6 well plate (Costar, 3516).</li> <li>Wash cells once with 1x PBS treat with trypsin (Corning, 25-053-CI).</li> <li>Take cells up in growth medium.</li> <li>Wash cells once with 1x PBS.</li> <li>Resuspend approximately 2.05x10<sup>5</sup>cells/well in 50-100µl 1x LDS sample buffer (Life Technologies, B0007). As positive control, take 20µg recombinant ELF1 (Abnova, H00001997-P01) in 1x LDS sample buffer.</li> <li>Denature samples for 3min at 95°C and subsequently keep them on ice.</li> <li>Separate samples on a Bolt 4-12% Bis-Tris Plus Gel (Invitrogen, NW04122) in an electrophoresis chamber (Mini Gel Tank, Invitrogen, A25977) for 30min at 80V and then for 60min at 120V.</li> <li>Transfer proteins onto nitrocellulose membrane (Invitrogen, IB23002) using an iBlot 2 Gel Transfer Device (Invitrogen).</li> <li>Block the membrane with TBS containing 0.05% Tween (TBST) containing 5% skim milk for 1h at RT.</li> <li>Incubation with primary</li> <li>rabit anti-ELF1 antibody (antibodies-online, ABIN6261511, 4318850) diluted 1:1000 in TBST containing 0.5% skim milk ON at 4°C.</li> <li>loading control mouse anti-beta actin antibody (Invitrogen, MA5-15739) diluted 1:5000 in TBST containing 0.5% skim milk ON at 4°C.</li> </ul>

	Wash membrane 3x for 10min with TBST.
•	Incubation with secondary
	<ul> <li>goat anti-rabbit HRP-conjugated antibody (Invitrogen, G21234) diluted 1:10000 in TBST containing 0.5% skim milk for 1h at RT.</li> </ul>
	<ul> <li>goat anti-mouse HRP-conjugated antibody (Invitrogen, G21040) diluted 1:10000 in TBST containing 0.5% skim milk for 1h at RT.</li> </ul>
	Wash membrane 3x for 10min with TBST.
•	Reveal protein bands using SuperSignal West Dura Extended Duration Substrate
	(ThermoFisher Scientific, 34075) and a ChemiDoc MP Imaging System (Bio-Rad).
Experimental Notes:	ABIN6261511 reveals a protein with an apparent molecular weight of approximately 62kDa. The expected molecular weight for endogenous human ELF1 is approximately 67kDa. The antibody also reveals a recombinant GST-tagged protein at the expected molecular weight (theoretical MW 93.9kDa).
•	Other ELF1-antibody dilutions were tested, but a dilution of 1:2500 was found optimal in
	terms of minimal background and strength of signal.

## Image for Validation report #104221

