Datasheet for ABIN2913314

anti-SOD1 antibody (AA 2-154)

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Overview

Quantity:	100 µL
Target:	SOD1
Binding Specificity:	AA 2-154
Reactivity:	Human, Horse
Host:	Rabbit
Clonality:	Polyclonal
Conjugate:	This SOD1 antibody is un-conjugated
Application:	Western Blotting (WB), Immunohistochemistry (IHC), Immunoprecipitation (IP), Immunofluorescence (fixed cells) (IF/ICC)

Product Details

Immunogen:	SOD1 (Ala2-Pro154)
Isotype:	IgG
Purification:	Antigen-specific affinity chromatography followed by Protein A affinity chromatography

Target Details

Target:	SOD1
Alternative Name:	Superoxide Dismutase 1 (SOD1 Products)
Pathways:	Sensory Perception of Sound, Transition Metal Ion Homeostasis

Application Details

Application Notes:	Optimal working dilution should be determined by the investigator.
Restrictions:	For Research Use only

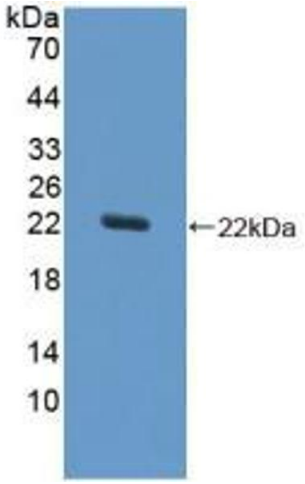
Handling

Format:	Liquid
Concentration:	500 µg/mL
Buffer:	0.01M PBS, pH 7.4, containing 0.05 % Proclin-300, 50 % glycerol.
Preservative:	ProClin
Precaution of Use:	This product contains ProClin: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	4 °C,-20 °C
Storage Comment:	Store at 4 °C for frequent use. Aliquot and store at -20 °C for one year. Avoid repeated freeze/thaw cycles.

Publications

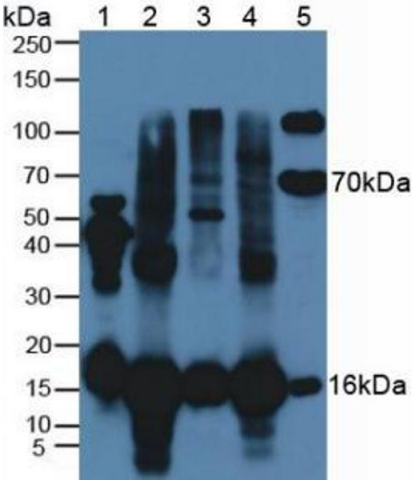
Product cited in:	Kronstein-Wiedemann, Stadtmüller, Traikov, Georgi, Teichert, Yosef, Wallenborn, Karl, Schütze, Wagner, El-Armouche, Tonn: "SARS-CoV-2 Infects Red Blood Cell Progenitors and Dysregulates Hemoglobin and Iron Metabolism." in: **Stem cell reviews and reports**, (2022) (PubMed).

Images



Western Blotting

Image 1. Western blot analysis of recombinant Horse SOD1.



Western Blotting

Image 2. Western blot analysis of (1) Rat Serum, (2) Bovine Liver Tissue, (3) Bovine Heart Tissue, (4) Bovine Brain Tissue and (5) Bovine Serum.

Validation report #100074 for Western Blotting (WB)

Successfully validated (Western Blotting (WB))

by Royal Veterinary College, London

Report Number: 100074

Date: Nov 17 2016

Target:	SOD-1
Method validated:	Western Blotting (WB)
Positive Control:	Horse Fibroblasts, human SH-SY5Y
Notes:	ABIN3058918 successfully labels SOD-1 in lysates of equine fibroblast and human SH-SY5Y cells.
Primary Antibody:	ABIN3058918
Secondary Antibody:	Goat anti-rabbit-IgG HRP-linked whole antibody (Dako, D0487)
Protocol:	• Seed 2x10^5 horse fibroblasts in a T25 flask and grow for 48h at 5% CO_2 in supplemented with Dulbecco's modified Eagle media (Sigma-Aldrich, D5546), with 10% heat-inactivated fetal calf serum (FCS) and 2mM (1%) L-glutamine and 1% penicillin (10IU/ml)/streptomycin (100µg/ml) to form a monolayer of 70-90%. • Human SH-SY5Y are cultured in identical conditions. • Wash cells once ice cold PBS. • Add 500µl chilled RIPA buffer (50mM Tris pH 7.5, 150mM NaCl, 1% NP40, 0.5% Na-deoxychlorate, 0.5% SDS, 1mM EDTA, 1mM EGTA, 1mM PMSF and 1x protease inhibitor) to each flask and incubate for 1min on ice. • Scrape each monolayer and transfer protein aggregates to a separate 1.5ml microcentrifuge tube. • Incubate for 20min on ice. • Centrifuge at 14000rpm for 20min at 4°C. • Remove the supernatant and determine the concentration of soluble proteins against a BSA protein standard using a DC protein assay (Biorad, 5000112). • Adjust protein samples to 0.3mg/ml total protein, using RIPA buffer and loading buffer. • After boiling for 5-10min, 1-10µg of each sample is separated on an Any kD Mini-PROTEAN TGX Stain-Free Precast Gel (Biorad, 4568123) at 100V for 90min. • Transfer proteins onto a PVDF blotting membrane (Hybond P, GE Healthcare Life Sciences, RPN303D) for 90min at 300mA. • Block the membrane with PBST containing 10% dry milk for 1h at RT. • Incubation with primary SOD-1 antibody (antibodies-online, ABIN3058918) diluted 1:1000 and a TUBB loading control antibody (abcam, ab6046) diluted 1:1000 in PBST containing 5% dry milk ON at 4°C.

- Wash 5 times with PBST over 1 hour whilst shaking.
- Incubation with secondary goat anti-rabbit-IgG horseradish peroxidase-linked whole antibody (Dako, D0487) diluted 1:5000 in PBST for 1h at RT.
- Wash 5 times with PBST over 1 hour whilst shaking.
- Blots were developed with Amersham ECL Prime Western Blotting Detection reagent (GE Healthcare Life Sciences, RPN2232) and visualized on a Kodak developer.

Experimental Notes:	The same human cell lysates were also used for a different blot with an anti-human SOD-1 antibody (lab stock). A band of the same molecular weight as in the horse fibroblast lysates was revealed.

Image for Validation report #100074

Validation image no. 1 for anti-Superoxide Dismutase 1, Soluble (SOD1) (AA 2-154) antibody (ABIN3058918)

Total protein lysates from horse fibroblasts and human SH-SY5Y cells were separated were separated on a polyacrylamide gel under denaturing conditions. SOD-1 and TUBB protein bands were revealed as described in the protocol section.

Validation report #100075 for Immunocytochemistry (ICC)

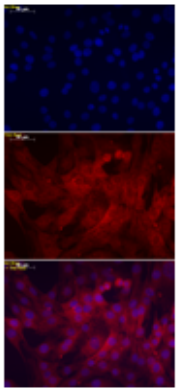
Successfully validated (Immunocytochemistry (ICC))

by Royal Veterinary College, London

Report Number: 100075

Date: Nov 17 2016

Target:	SOD-1
Method validated:	Immunocytochemistry (ICC)
Positive Control:	Horse Fibroblasts
Notes:	ABIN3058918 achieves SOD-1 staining in horse fibroblasts when grown in a monolayer.
Primary Antibody:	ABIN3058918
Secondary Antibody:	anti-rabbit labeled with Alexa Fluor 594
Protocol:	• Horse fibroblasts are seeded onto glass coverslips pre-treated with 1:10 diluted Matrigel (BD Biosciences, 354234) at a density of $1x10^5$ cells per coverslip in a 24 well plate. • Incubate for 48h at 37°C in 5% CO_2. • Wash coverslips with PBS. • Fix coverslips with 4% paraformaldehyde (PFA) and 0.1% triton for 20min at 4°C, followed by 8% PFA for 30min at RT. • Permeabilize cells with 0.5% triton for 30min at RT. • Wash coverslips 3 times with PBS. • Incubate coverslips with primary rabbit anti SOD-1 antibody (antibodies-online, ABIN3058918) diluted 1:40 in a dark, humidified chamber for 1h at RT. A negative control was incubated without primary antibody. • Wash coverslips 3 times with PBS. • Incubate with secondary goat anti rabbit labeled with Alexa Fluor 594 (ThermoFisher Scientific, A10239) diluted 1:500 for 1h at RT. • Wash coverslips 3 times with PBS. • Mount coverslips in a hard setting mount with VECTASHIELD HardSet Antifade Mounting Medium with DAPI (Vector Laboratories, H-1500). • Fluorescence microscopy is then used to image each coverslip.



Validation image no. 1 for anti-Superoxide Dismutase 1, Soluble (SOD1) (AA 2-154) antibody (ABIN3058918)

Horse fibroblasts were grown in a monolayer and stained with ABIN3058918 (red) as desecribed in the protocol. DAPI (blue) was used to visualize nuclei.