

Product datasheet

Anti-Spastin antibody [Sp 3G11/1] ab31850

KO VALIDATED

[3 References](#) [5 Images](#)

Overview

Product name	Anti-Spastin antibody [Sp 3G11/1]
Description	Mouse monoclonal [Sp 3G11/1] to Spastin
Host species	Mouse
Tested applications	Suitable for: WB, IHC-P, IP
Species reactivity	Reacts with: Rat, Human
Immunogen	Recombinant full length protein (Human).
Positive control	WB: Total HeLa extract or rat brain synaptosome This antibody gave a positive result in IHC in the following FFPE tissue: Human lung adenocarcinoma. IP: HAP1 cells
General notes	<p>The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing.</p> <p>If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.02% Sodium azide Constituent: 99.98% PBS
Purity	Protein A/G purified
Clonality	Monoclonal
Clone number	Sp 3G11/1
Myeloma	Sp2/0-Ag14
Isotype	IgG2a

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab31850 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
WB		1/500. Detects a band of approximately 52 kDa. There are two splice isoforms of spastin, one without exon4 and two alternative ATG start sites, which may determine the localisation of the translate protein. 52kDa is the major band.
IHC-P		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.

Target

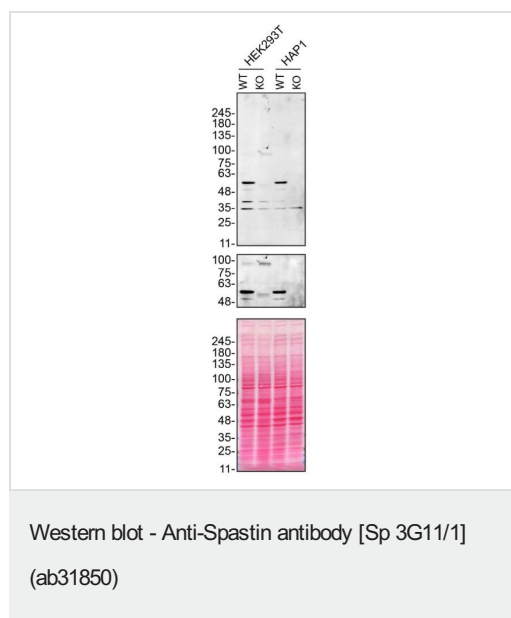
Relevance

Spastin is thought have a role in microtubule dynamics through its function as a microtubule severing protein. It is localised to the centrosome of neuronal cells but is not found in glial cells. Mutation in the ATPase binding domain of spastin causes hereditary spastic paraplegias (HSP), a large group of clinically similar disorders. Mutant forms of spastin are generally found throughout the cytoplasm rather than within the nucleus.

Cellular localization

Cytoplasm, Cytoskeleton, Endoplasmic reticulum, Endosome, Membrane, Microtubule, Nucleus

Images



All lanes : Anti-Spastin antibody [Sp 3G11/1] (ab31850) at 1/500 dilution

Lane 1 : Wild-type HEK-293T cell lysate

Lane 2 : SPAST knockout HEK293T cell lysate

Lane 3 : Wild-type HAP1 cell lysate

Lane 4 : SPAST knockout HAP1 cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

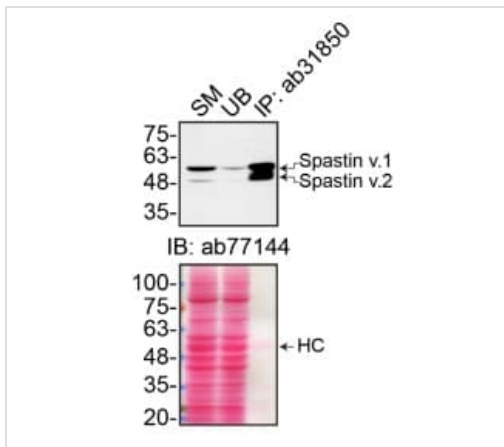
All lanes : goat anti-rabbit HRP at 0.2 µg/ml

Performed under reducing conditions.

ab31850 was shown to react with SPAST in wild-type HEK293T cells in Western blot with loss of signal observed in SPAST knockout cell line **ab267238** (SPAST knockout cell lysate

ab258698). Wild-type HEK293T and SPAST knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5% milk in TBST for 1 hr before incubation with ab31850 overnight at 4 °C at a 1/500 dilution. Blots were incubated with goat anti-rabbit HRP secondary antibodies at 0.2µg/mL before imaging.

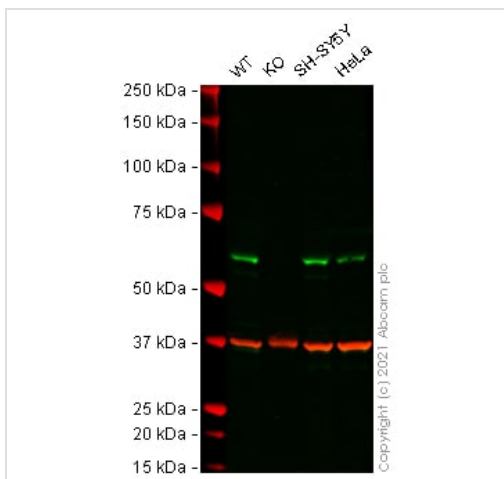
These data were provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.



Immunoprecipitation - Anti-Spastin antibody [Sp 3G11/1] (ab31850)

Immunoprecipitation of SPAST in HAP1 cells. Lysates were prepared and immunoprecipitation was performed using 1µg of ab31850 pre-coupled to Protein A beads. Samples were washed and processed for western blot with **ab77144** at 1/500.

This data was kindly provided by the YCharOS Inc., an open science company with the mission of characterizing every commercially available antibody reagent. Abcam are working with YCharOS to support their mission of antibody characterisation using knock out cell lines.



Western blot - Anti-Spastin antibody [Sp 3G11/1] (ab31850)

All lanes : Anti-Spastin antibody [Sp 3G11/1] (ab31850) at 1/500 dilution

Lane 1 : Wild-type HEK-293T cell lysate

Lane 2 : Spastin knockout HEK-293T cell lysate

Lane 3 : SH-SY5Y cell lysate

Lane 4 : HeLa cell lysate

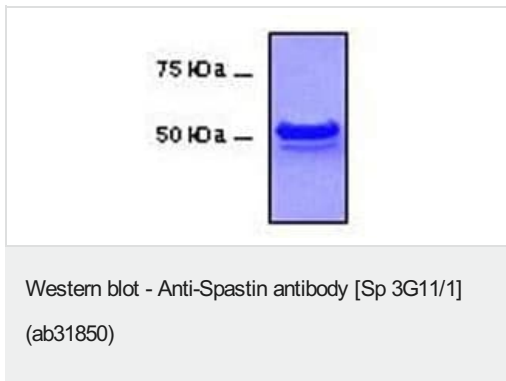
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 55 kDa

False colour image of Western blot: Anti-Spastin antibody [Sp 3G11/1] staining at 1/500 dilution, shown in green; Rabbit Anti-

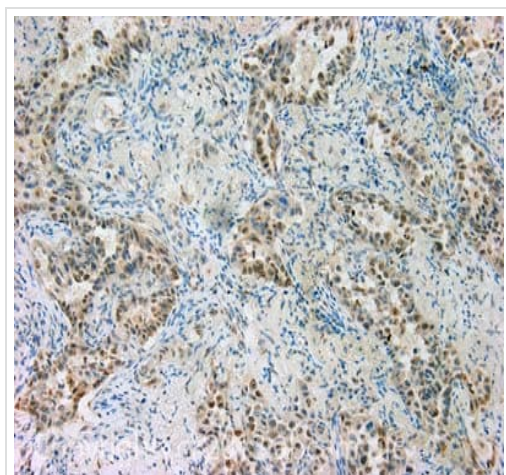
GAPDH antibody [EPR16891] ([ab181602](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab31850 was shown to bind specifically to Spastin. A band was observed at 55 kDa in wild-type HEK-293T cell lysates with no signal observed at this size in SPAST knockout cell line [ab267238](#) (knockout cell lysate [ab258698](#)). To generate this image, wild-type and SPAST knockout HEK-293T cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed ([ab216772](#)) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed ([ab216777](#)) at 1/20000 dilution.



Anti-Spastin antibody [Sp 3G11/1] ([ab31850](#)) at 1/500 dilution + rat brain synaptosome

Observed band size: 52 kDa

There are two splice isoforms of spastin (one without exon4) and two alternative ATG start sites, which may determine the localisation of the translate protein. 52kDa is the major band.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Spastin antibody [Sp 3G11/1] ([ab31850](#))

IHC image of Spastin staining in Human lung adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with [ab31850](#), 10µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

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