Technical Data Sheet

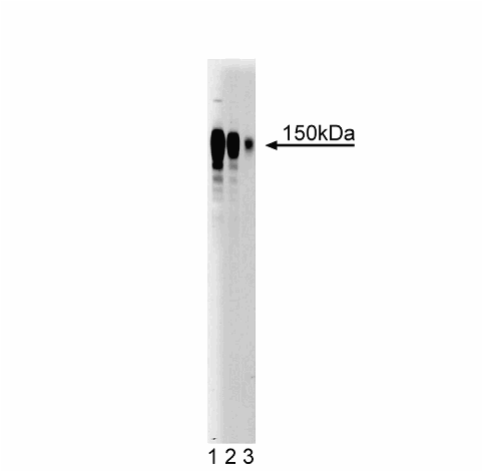
Purified Mouse Anti-Ataxin-2

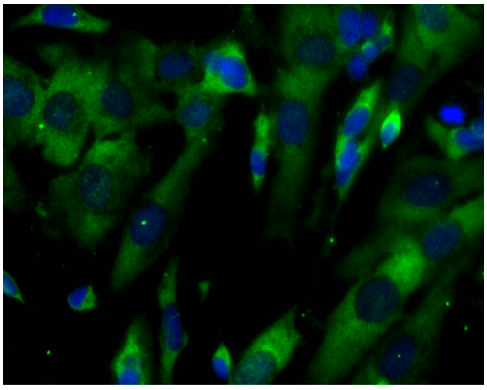
Product Information

Material Number:	**611378**
Size:	50 µg
Concentration:	250 µg/ml
Clone:	22/Ataxin-2
Immunogen:	Human Ataxin-2 aa. 713-904
Isotype:	Mouse IgG1
Reactivity:	QC Testing: Human Tested in Development: Rat
Target MW:	150 kDa
Storage Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09% sodium azide.

Description

The hereditary ataxias are neurodegenerative disorders characterized by abnormalities of balance due to dysfunction of the cerebellum and cerebellar pathways. Spinocerebellar ataxias (SCAs) represent a heterogeneous group of disorders with a prevalence of about 1 in 10^5. Moderate expansion (36 or 37 units) of CAG repeats on the SCA2 gene leads to a gene product, ataxin-2, that has a long polyglutamine tract. Ataxin-2 is a basic protein with two domains (Sm1 and Sm2) that have been implicated in RNA splicing and protein interaction. Human ataxin-2 has significant homology with mouse ataxin-2 and ataxin-2 related protein. Ataxin-2 is expressed as early as day 8 of mouse embryogenesis and is detected in brain, heart, placenta, liver, skeletal muscle, and pancreas, but not in lung or kidney. Expression increases with age and the protein is localized to the cytoplasm in purkinje cells and other neuronal cell types. Thus, ataxin-2 is thought to be important for RNA splicing or protein complex formation in many normal tissues, while polyglutamine containing ataxin-2 leads to neuropathology.





Western blot analysis of Ataxin-2 on Jurkat cell lysate (left). *Lane 1: 1:250, lane 2: 1:500, lane 3: 1:1000 dilution of anti-Ataxin-2.*

Immunofluorescent staining of SH-SY5Y cells (right). *Cells were seeded in a collagen coated 384 well imaging plate (Material # 353962) at ~ 8,000 cells per well. After overnight incubation, cells were stained using the Triton X100 fix/perm protocol (see Recommended Assay Procedure) and the anti-Ataxin antibody. The second step reagent was Alexa Fluor® 488 goat anti mouse Ig (Invitrogen). The image was taken on a Pathway 855 or 435 imager using a 20x objective. This antibody also stained SK-N-SH, C6, U87 and U373 cells using both the Triton X100 and methanol fix/perm protocols (see Recommended Assay Procedure).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at -20°C.

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Application Notes

Application

Western blot	Routinely Tested
Immunofluorescence	Tested During Development

Recommended Assay Procedure:

Methanol Procedure for a 96 well plate:

Remove media from wells. Add 100 µl/well fresh 3.7% Formaldehyde in PBS. Incubate for 10 minutes at room temperature (RT). Flick out and add 100 µl/well 90% methanol. Incubate for 5 minutes at RT. Flick out and wash twice with PBS. Flick out PBS and add 100 µl/well blocking buffer (3% FBS in PBS). Incubate for 30 minutes at RT. Flick out and add diluted antibody (diluted in blocking buffer). Incubate for 1 hour at RT. Wash three times with PBS. Flick out PBS and add second step reagent. Incubate for 1 hour at RT. Wash three times with PBS. Image sample.

Triton-X 100 Procedure for a 96 well plate:

Remove media from wells. Add 100 µl/well fresh 3.7% Formaldehyde in PBS. Incubate for 10 minutes at room temperature (RT). Flick out and add 100 µl/well 0.1% Triton-X 100. Incubate for 5 minutes at RT. Flick out and wash twice with PBS. Flick out PBS and add 100 µl/well blocking buffer (3% FBS in PBS). Incubate for 30 minutes at RT. Flick out and add diluted antibody (diluted in blocking buffer). Incubate for 1 hour at RT. Flick out and wash three times with PBS. Flick out and add second step reagent. Incubate for 1 hour at RT. Flick out and wash three times with PBS. Image sample.

Suggested Companion Products

Catalog Number	Name	Size	Clone
611451	Jurkat Cell Lysate	500 µg	(none)
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)
554001	FITC Goat Anti-Mouse Ig	0.5 mg	Polyclonal

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Huynh DP, Del Bigio MR, Ho DH, Pulst SM. Expression of ataxin-2 in brains from normal individuals and patients with Alzheimer's disease and spinocerebellar ataxia. *Ann Neurol.* 1999; 45(2):232-241.(Biology)

Nechiporuk T, Huynh DP, Figueroa K, Sahba S, Nechiporuk A, Pulst SM. The mouse SCA2 gene: cDNA sequence, alternative splicing and protein expression. *Hum Mol Genet.* 1998; 7(8):1301-1309.(Biology)

Pulst SM, Nechiporuk A, Nechiporuk T, et al. Moderate expansion of a normally biallelic trinucleotide repeat in spinocerebellar ataxia type 2. *Nat Genet.* 1996; 14(3):269-276.(Biology)