abcam

Product datasheet

Anti-TLS/FUS antibody [EPR5812] ab124923

KO VALIDATED Recombinant RabMAb

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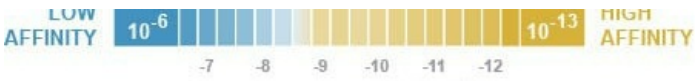
Overview

Product name	Anti-TLS/FUS antibody [EPR5812]
Description	Rabbit monoclonal [EPR5812] to TLS/FUS
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF, Flow Cyt (Intra)
Species reactivity	**Reacts with:** Human **Does not react with:** Mouse, Rat
Immunogen	Synthetic peptide within Human TLS/FUS. The exact sequence is proprietary.
Positive control	WB: Hap1, Hek-293T, K562, Human fetal brain, Caco 2, HeLa and HepG2 lysates. Flow Cyt (intra): HepG2 IHC-P: Human kidney, human breast tissue tissue. ICC/IF: K562, HepG2 cells.
General notes	We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
Dissociation constant (K_D)	K_D = 1.55 x 10^{-10} M



Learn more about K_D

Storage buffer	pH: 7.2 Preservative: 0.05% Sodium azide Constituents: 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR5812
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab124923 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

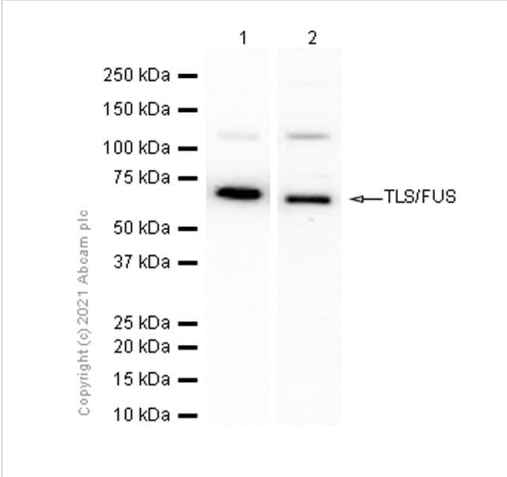
Application	Abreviews	Notes
WB	★★★★☆ (2)	1/1000. Detects a band of approximately 73 kDa (predicted molecular weight: 53 kDa).
IHC-P		1/800. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ICC/IF		1/100.
Flow Cyt (Intra)		1/100.

Target

Function	Binds both single-stranded and double-stranded DNA and promotes ATP-independent annealing of complementary single-stranded DNAs and D-loop formation in superhelical double-stranded DNA. May play a role in maintenance of genomic integrity.
Tissue specificity	Ubiquitous.
Involvement in disease	Note=A chromosomal aberration involving FUS is found in a patient with malignant myxoid liposarcoma. Translocation t(12;16)(q13;p11) with DDIT3. Note=A chromosomal aberration involving FUS is a cause of acute myeloid leukemia (AML). Translocation t(16;21)(p11;q22) with ERG. Defects in FUS may be a cause of angiomatoid fibrous histiocytoma (AFH) [MIM:612160]. A distinct variant of malignant fibrous histiocytoma that typically occurs in children and adolescents and is manifest by nodular subcutaneous growth. Characteristic microscopic features include lobulated sheets of histiocyte-like cells intimately associated with areas of hemorrhage and cystic pseudovascular spaces, as well as a striking cuffing of inflammatory cells, mimicking a lymph node metastasis. Note=A chromosomal aberration involving FUS is found in a patient with angiomatoid fibrous histiocytoma. Translocation t(12;16)(q13;p11.2) with ATF1 generates a chimeric FUS/ATF1 protein.

	Defects in FUS are the cause of amyotrophic lateral sclerosis type 6 (ALS6) [MIM:608030]. ALS6 is a familial form of amyotrophic lateral sclerosis. ALS is a neurodegenerative disorder affecting upper motor neurons in the brain and lower motor neurons in the brain stem and spinal cord, resulting in fatal paralysis. Sensory abnormalities are absent. Death usually occurs within 2 to 5 years. The etiology of amyotrophic lateral sclerosis is likely to be multifactorial, involving both genetic and environmental factors. The disease is inherited in 5-10%.
Sequence similarities	Belongs to the RRM TET family. Contains 1 RanBP2-type zinc finger. Contains 1 RRM (RNA recognition motif) domain.
Post-translational modifications	Arg-216 and Arg-218 are dimethylated, probably to asymmetric dimethylarginine.
Cellular localization	Nucleus.

Images



Western blot - Anti-TLS/FUS antibody [EPR5812] (ab124923)

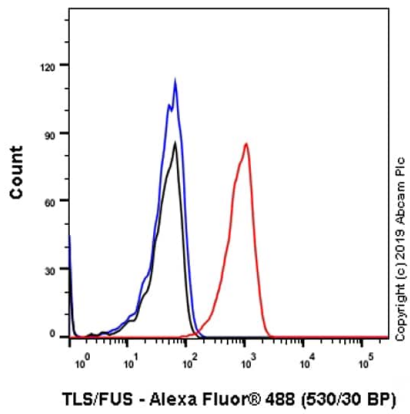
All lanes : Anti-TLS/FUS antibody [EPR5812] (ab124923) at 1/5000 dilution (Purified)

Lane 1 : K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysate at 20 µg
Lane 2 : Caco-2 (Human colorectal adenocarcinoma epithelial cell) whole cell lysate at 15 µg

Secondary
All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

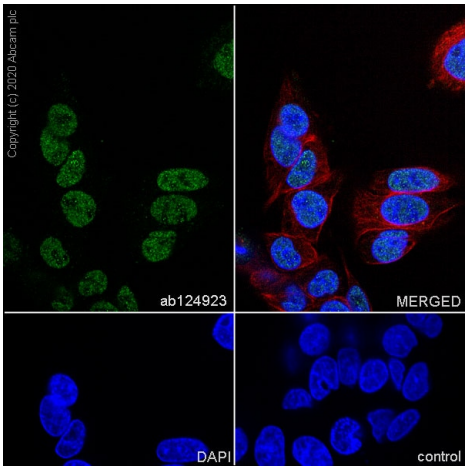
Predicted band size: 53 kDa
Observed band size: 73 kDa

We are unsure about the nature of the extra bands.



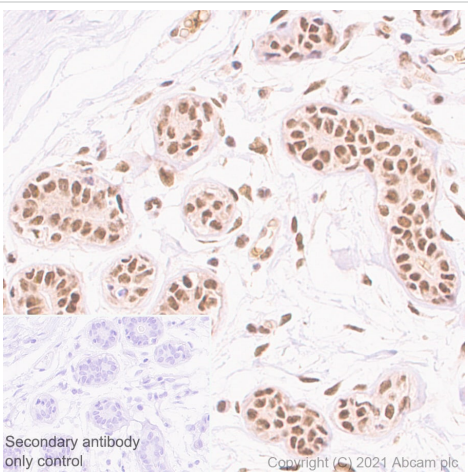
Flow Cytometry (Intracellular) - Anti-TLS/FUS antibody [EPR5812] (ab124923)

Flow Cytometry analysis of HepG2 (Human hepatocellular carcinoma epithelial cell) cells labelling TLS/FUS with Purified ab124923 at 1:100 dilution (10 µg/ml) (Red). Cells were fixed with 5% Paraformaldehyde and permeabilised with 91% Methanol. A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1:2000. Isotype control - Rabbit monoclonal IgG (Black). Unlabelled control - Cell without incubation with primary antibody and secondary antibody (Blue).



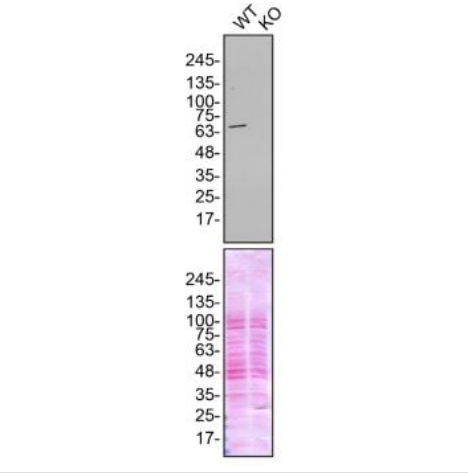
Immunocytochemistry/ Immunofluorescence - Anti-TLS/FUS antibody [EPR5812] (ab124923)

Immunocytochemistry analysis of HepG2 (Human hepatocellular carcinoma epithelial cell) cells labeling TLS/FUS with Purified ab124923 at 1:100 dilution (10 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



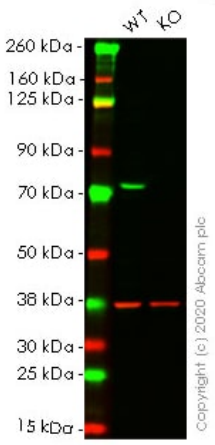
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TLS/FUS antibody [EPR5812] (ab124923)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast tissue sections labeling TLS/FUS with Purified ab124923 at 1:800 dilution (1.429 µg/mL). Heat mediated antigen retrieval was performed using Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used. PBS instead of the primary antibody was used as the negative control.



Western blot - Anti-TLS/FUS antibody [EPR5812] (ab124923)

ab124923 was shown to react with FUS in wild-type HeLa cells in Western blot with loss of signal observed in a FUS knockout cell line. Wild-type HeLa and FUS knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5% milk in TBST for 1 hr before incubation with ab124923 overnight at 4 °C at a 1/5000 dilution. Blots were incubated with secondary antibodies at 1/5000 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.



Western blot - Anti-TLS/FUS antibody [EPR5812] (ab124923)

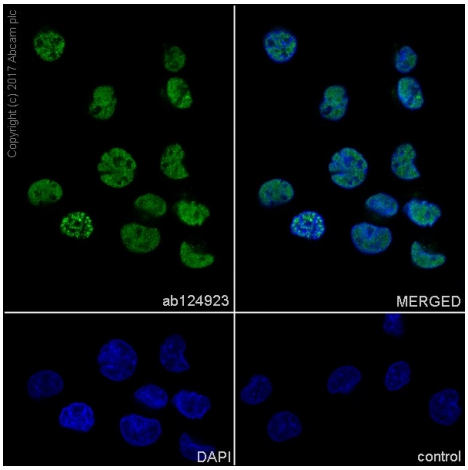
Anti-TLS/FUS antibody [EPR5812] (ab124923) at 1/1000 dilution + FUS knockout HEK-293T cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 53 kDa
Observed band size: 75 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab124923 observed at 75 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

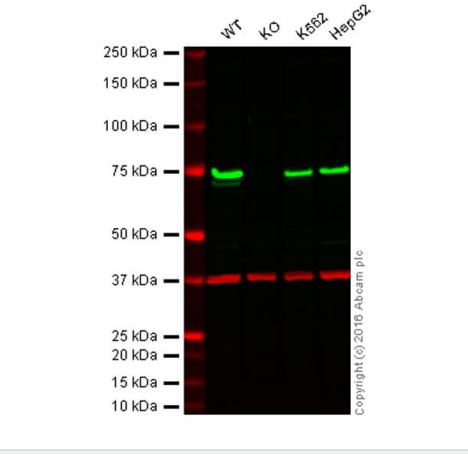
ab124923 was shown to react with TLS/FUS in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line **ab266587** (knockout cell lysate **ab257100**) was used. Wild-type HEK-293T and FUS knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab124923 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-TLS/FUS antibody [EPR5812] (ab124923)

Immunocytochemistry/ Immunofluorescence analysis of K-562 (human chronic myelogenous leukemia lymphoblast) cells labeling TLS/FUS with ab124923 at 1/250 (8.3 µg/mL). Cells were fixed with 4% Paraformaldehyde and permeabilised 0.1% TritonX-100. **ab150077**, AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 (2 µg/mL) was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.

Confocal image showing nuclear staining in K-562 cells.



Western blot - Anti-TLS/FUS antibody [EPR5812] (ab124923)

All lanes : Anti-TLS/FUS antibody [EPR5812] (ab124923) at 1/1000 dilution

Lane 1 : Wild-type HAP1 cell lysate
Lane 2 : TLS/FUS knockout HAP1 cell lysate
Lane 3 : K562 cell lysate
Lane 4 : HepG2 cell lysate

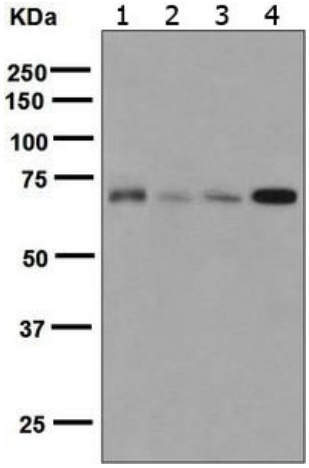
Lysates/proteins at 20 µg per lane.

Predicted band size: 53 kDa

Lanes 1 to 4: Merged signal (red and green). Green - ab124923 observed at 75 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab124923 was shown to specifically react with TLS/FUS when TLS/FUS knockout samples were used. Wild-type and TLS/FUS knockout samples were subjected to SDS-PAGE. ab124923 and **ab28245** (loading control to GAPDH) were both diluted at 1/1000 and 1/10 000 respectively and incubated overnight at 4°C. Blots

were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-TLS/FUS antibody [EPR5812] (ab124923)

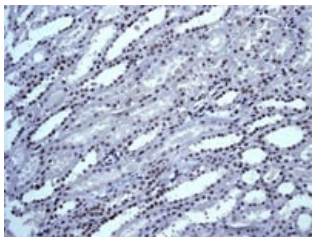
All lanes : Anti-TLS/FUS antibody [EPR5812] (ab124923) at 1/1000 dilution

Lane 1 : K562 cell lysate
Lane 2 : Human fetal brain lysate
Lane 3 : Caco 2 cell lysate
Lane 4 : HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat anti-rabbit HRP at 1/2000 dilution

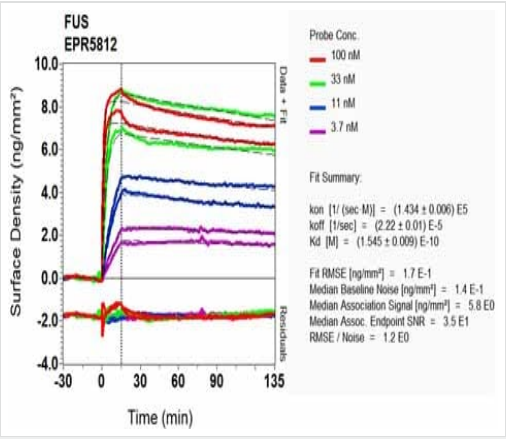
Predicted band size: 53 kDa
Observed band size: 73 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-TLS/FUS antibody [EPR5812] (ab124923)

ab124923, at 1/50 dilution, staining TLS/FUS in formalin-fixed, paraffin-embedded Human kidney tissue, by Immunohistochemistry.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

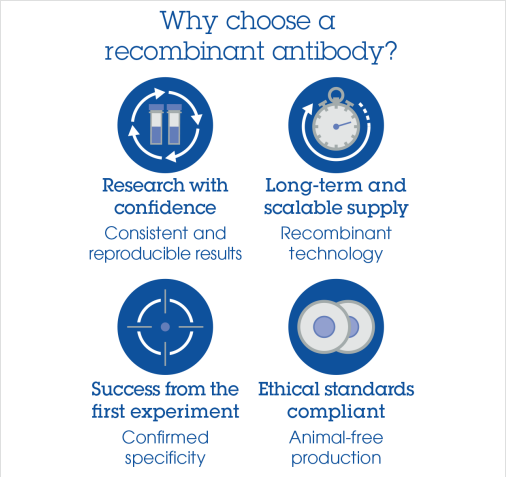


OI-RD Scanning - Anti-TLS/FUS antibody [EPR5812] (ab124923)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Anti-TLS/FUS antibody [EPR5812] (ab124923)

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