abcam

ab200837

Anti-CstF-64 antibody [EPR15698]

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR15698
Purification technique	Affinity purification Protein A
Concentration	0.147 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Human
Dilution info	1/500
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/20
Notes	-

IP

Tested

Species	Human
Dilution info	1/30
Notes	-

WB

Tested

Species	Human
Dilution info	1/5000
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/50
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target data

See full target information Cleavage stimulation factor subunit 2

Function	One of the multiple factors required for polyadenylation and 3'-end cleavage of mammalian pre-mRNAs. This subunit is directly involved in the binding to pre-mRNAs.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

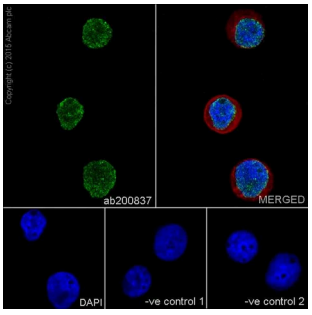
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

---

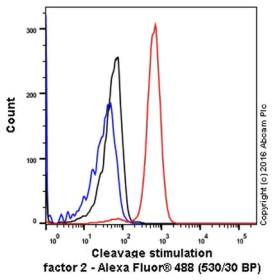
10 product images



Immunocytochemistry/ Immunofluorescence - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CstF-64 antibody

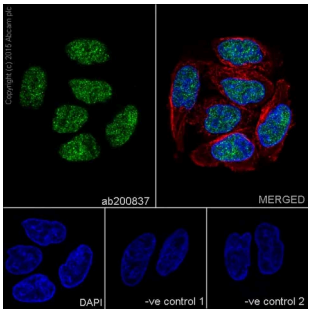
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling CstF-64 with ab200837 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green). Nuclear staining on Jurkat cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows;
-ve control 1: ab200837 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



Flow Cytometry (Intracellular) - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Flow Cytometry (Intracellular) staining using rabbit Anti-CstF-64 antibody

Intracellular Flow Cytometry analysis of Jurkat (human acute T cell leukemia) labelling CstF-64with purified ab200837 at 1/25000 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Alexa Fluor® 488 goat anti-rabbit IgG (1/2000) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

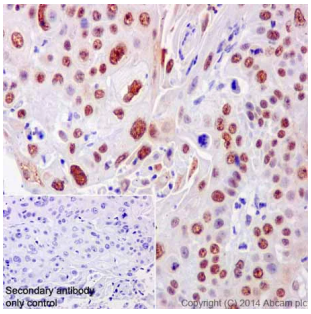


Immunocytochemistry/ Immunofluorescence - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CstF-64 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling CstF-64 with ab200837 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at

1/500 dilution (green). Nuclear staining on HeLa cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows;
-ve control 1: ab200837 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2: ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.

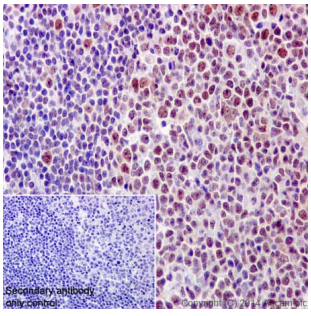


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CstF-64 antibody

Immunohistochemical analysis of paraffin-embedded Human cervix carcinoma tissue labeling CstF-64 with ab200837 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear staining on Human cervix carcinoma tissue is observed. Counter stained with Hematoxylin.
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

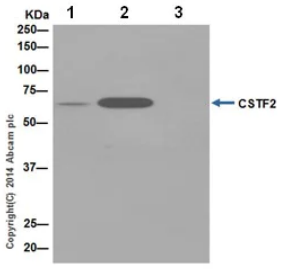


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CstF-64 antibody

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling CstF-64 with ab200837 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear staining on Human tonsil tissue is observed. Counter stained with Hematoxylin.
Secondary antibody only: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



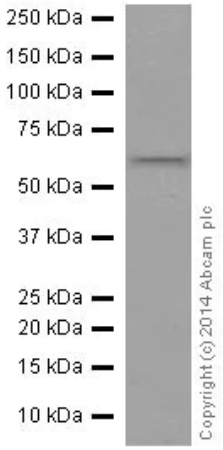
Immunoprecipitation - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 was immunoprecipitated from 1mg of K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate with ab200837 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab200837 at 1/1000 dilution. Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG, was used as secondary antibody at 1/1500 dilution.
Lane 1: K562 whole cell lysate10 µg (Input). Lane 2: ab200837 IP in K562 whole cell lysate. Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab200837 in K562 whole cell lysate.
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 5 seconds

All lanes:
Immunoprecipitation - Anti-CstF-64 antibody [EPR15698] (ab200837)

Predicted band size: 61 kDa



Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837) at 1/200 dilution

All lanes:
Human testis tissue lysate

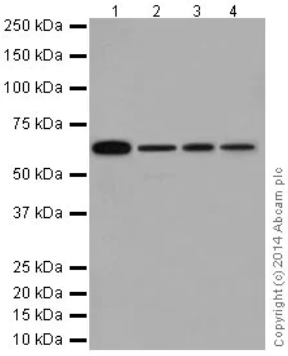
Secondary

All lanes:
Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 61 kDa

Observed band size: 61 kDa

Exposure time: 3min



Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Western blot staining using rabbit Anti-CstF-64 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837) at 1/5000 dilution

Lane 1:
K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate at 10 µg

Lane 2:
Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate at 10 µg

Lane 3:
THP-1 (Human monocytic leukemia cells) whole cell lysate at 10 µg

Lane 4:
HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate at 10 µg

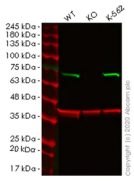
Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 61 kDa

Observed band size: 61 kDa

Exposure time: 3min



Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Western blot staining using rabbit Anti-CstF-64 antibody

Lanes 1-3: Merged signal (red and green). Green - ab200837 observed at 65 kDa. Red - loading control ab8245 observed at 36 kDa.
ab200837 Anti-CstF-64 antibody [EPR15698] was shown to specifically react with CstF-64 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265207 (knockout cell lysate ab257402) was used. Wild-type and CstF-64 knockout samples were subjected to SDS-PAGE. ab200837 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 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.

All lanes:
Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837) at 1/1000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

Lane 2:
CSTF2 knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human CSTF2 (CstF-64) knockout HeLa cell line (ab265207)

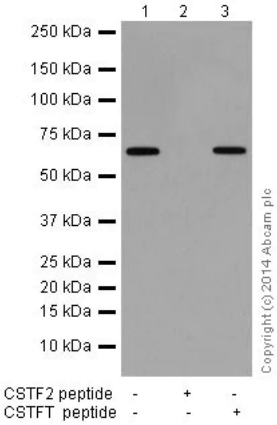
Lane 3:
K-562 cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 61 kDa

Observed band size: 65 kDa



Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837)

CstF-64 Western blot staining of K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate using rabbit Anti-CstF-64 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
Based on sequence analysis, ab200837 shares 78% homology with family member CSTFT. The levels of XR were tested in the accompanying peptide blocking experiment.

All lanes:
Western blot - Anti-CstF-64 antibody [EPR15698] (ab200837) at 1/5000 dilution

Lane 1:
K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate

Lane 2:
K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate with CSTF2 peptide

Lane 3:

K562 (Human chronic myelogenous leukemia cells from bone marrow) whole cell lysate with CSTFT peptide

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 61 kDa

Observed band size: 61 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.