abcam

Product datasheet

Anti-SSB antibody [mAbcam75927] ab75927

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Overview

Product name	Anti-SSB antibody [mAbcam75927]
Description	Mouse monoclonal [mAbcam75927] to SSB
Host species	Mouse
Tested applications	**Suitable for:** ICC/IF, IP, WB, IHC-P, Flow Cyt (Intra)
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide corresponding to Human SSB aa 300 to the C-terminus (C terminal). (Peptide available as **ab106476**)
Positive control	WB: HEK293 Whole Cell; HeLa Nuclear; Raji Nuclear; MCF7 Nuclear, MCF7 Whole Cell; Ramos Whole Cell; Human Fetal Heart Tissue. IHC: Human breast adenocarcinoma. ICC: HeLa cells
General notes	This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact **orders@abcam.com**. 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. 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

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 long term.
Storage buffer	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Purity	Protein G purified
Clonality	Monoclonal
Clone number	mAbcam75927

Myeloma	Sp2/0-Ag14
Isotype	IgG2a

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab75927 in the following tested applications.

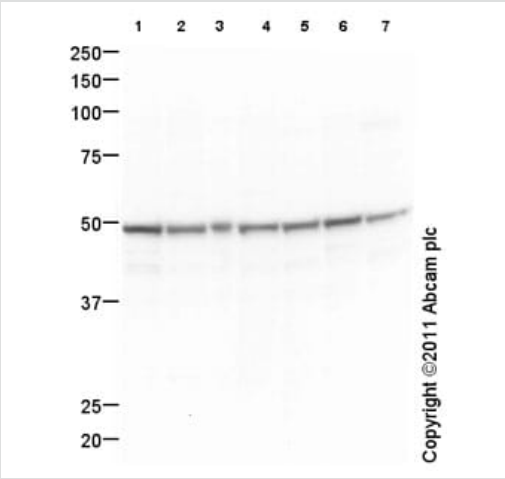
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF	★★★★★ (1)	Use a concentration of 5 µg/ml.
IP		Use a concentration of 5 µg/ml.
WB	★★★★★ (3)	Use a concentration of 1 - 5 µg/ml. Detects a band of approximately 47 kDa (predicted molecular weight: 47 kDa).
IHC-P		Use a concentration of 5 µg/ml.
Flow Cyt (Intra)		Use 0.5µg for 10^6 cells. **ab170191** - Mouse monoclonal IgG2a, is suitable for use as an isotype control with this antibody.

Target

Function	Binds to the 3' poly(U) terminii of nascent RNA polymerase III transcripts, protecting them from exonuclease digestion and facilitating their folding and maturation.
Sequence similarities	Contains 1 HTH La-type RNA-binding domain. Contains 1 RRM (RNA recognition motif) domain.
Post-translational modifications	Phosphorylated. The phosphorylation sites are at the C-terminal part of the protein. The N-terminus is blocked.
Cellular localization	Nucleus.

Images



Western blot - Anti-SSB antibody [mAbcam75927] (ab75927)

All lanes : Anti-SSB antibody [mAbcam75927] (ab75927) at 5 µg/ml

Lane 1 : HEK293 (Human embryonic kidney cell line) Whole Cell Lysate
Lane 2 : HeLa (Human epithelial carcinoma cell line) Nuclear Lysate
Lane 3 : Raji (Human Burkitt's lymphoma cell line) Nuclear Lysate - tumor cell line (**ab30127**)
Lane 4 : MCF7 (Human breast adenocarcinoma cell line) Nuclear Lysate
Lane 5 : MCF7 (Human breast adenocarcinoma cell line) Whole Cell Lysate
Lane 6 : Ramos (Human Burkitt's lymphoma cell line) Whole Cell Lysate
Lane 7 : Heart (Human) Whole Cell Lysate - fetal normal tissue

Lysates/proteins at 20 µg per lane.

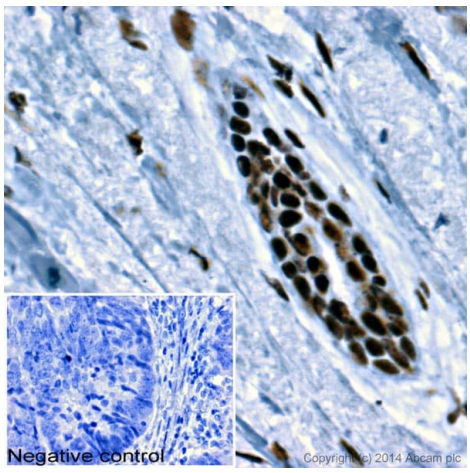
Secondary
All lanes : Goat Anti-Mouse IgG H&L (HRP) preadsorbed (**ab97040**) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 47 kDa
Observed band size: 47 kDa

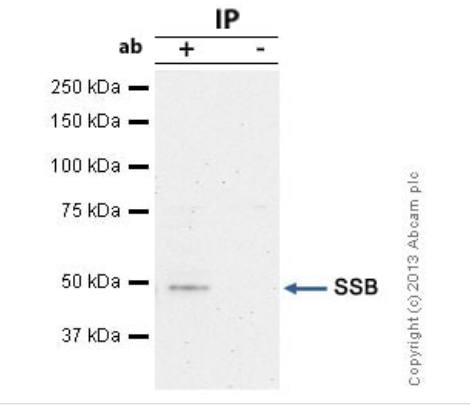
Exposure time: 30 seconds



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SSB antibody [mAbcam75927] (ab75927)

IHC image of SSB staining in Human breast adenocarcinoma formalin fixed paraffin embedded tissue section*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was incubated with ab75927, 0.5µg/ml overnight at +4°C. An HRP-conjugated secondary (Ab97245, 1/200 dilution) was used for 1hr at room temperature. The section was counterstained with haematoxylin and mounted with DPX.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunoprecipitation - Anti-SSB antibody [mAbcam75927] (ab75927)

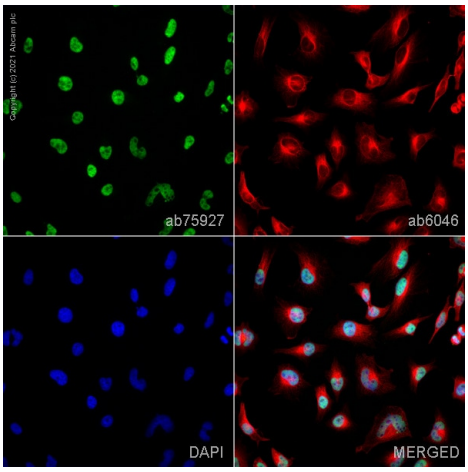
SSB was immunoprecipitated using 0.5mg Hek293 whole cell extract, 5µg of Mouse monoclonal to SSB and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, Hek293 whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab75927.

Secondary: Goat polyclonal to mouse IgG light chain specific (HRP) at 1/20,000 dilution.

Band: 47kDa; SSB

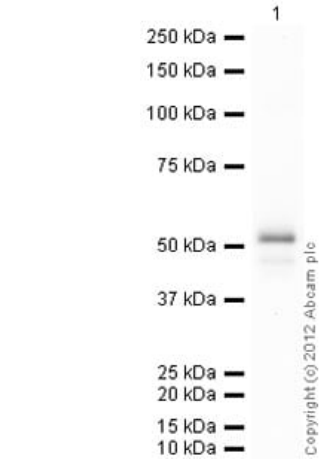


Immunocytochemistry/ Immunofluorescence - Anti-SSB antibody [mAbcam75927] (ab75927)

ab75927 staining SSB in HeLa cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab75927 at 5µg/ml and **ab6046**, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with **ab150117**, Goat polyclonal Secondary Antibody to Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and **ab150080**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 100% methanol (5 min).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Western blot - Anti-SSB antibody [mAbcam75927] (ab75927)

Anti-SSB antibody [mAbcam75927] (ab75927) at 1 µg/ml + Recombinant Human SSB protein (**ab84477**) at 0.1 µg

Secondary

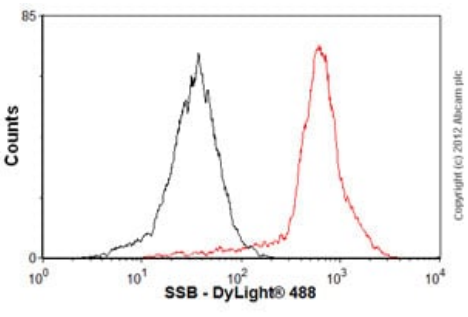
Goat Anti-Mouse IgG H&L (HRP) preadsorbed (**ab97040**) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 47 kDa

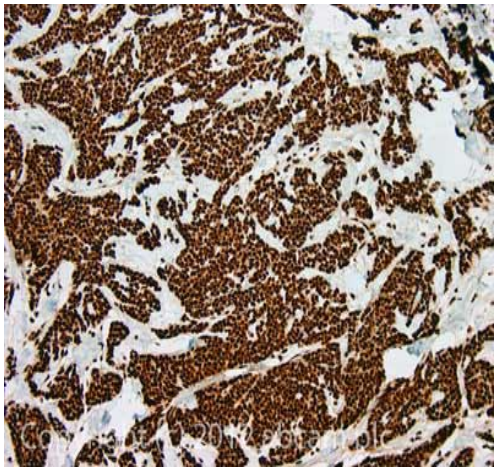
Exposure time: 30 seconds



Flow Cytometry (Intracellular) - Anti-SSB antibody [mAbcam75927] (ab75927)

Overlay histogram showing Ramos cells stained with ab75927 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab75927, 0.5µg/1x10^6 cells) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (**ab96879**) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was mouse IgG2a [ICIGG2A] (**ab91361**, 1µg/1x10^6 cells) used under the same conditions.

Acquisition of >5,000 events was performed. This antibody gave a positive signal in Ramos cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-SSB antibody [mAbcam75927] (ab75927)

IHC image of SSB staining in Human breast adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica BondTM 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 ab75927, 5µg/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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