abcam

ab75927

Anti-SSB antibody [mAbcam75927]

KO Validated

Key facts

Isotype	IgG2a
Host species	Mouse
Storage buffer	pH: 7.4 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	mAbcam75927
Purification technique	Affinity purification
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	5 µg/mL
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	0.5 µg for 10^6 Cells
Notes	ab170191 - Mouse monoclonal IgG2a, is suitable for use as an isotype control with this antibody.

WB

Tested

Species	Human
Dilution info	1-5 µg/mL
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	5 µg/mL
Notes	-

IHC-P

Tested

Species	Human
Dilution info	5 µg/mL
Notes	-

Target data

See full target information Lupus La protein

Function	Binds to the 3' poly(U) terminus of nascent RNA polymerase III transcripts, protecting them from exonuclease digestion and facilitating their folding and maturation (PubMed:2470590, PubMed:3192525). In case of Coxsackievirus B3

infection, binds to the viral internal ribosome entry site (IRES) and stimulates the IRES-mediated translation (PubMed:12384597).

Storage

Shipped at conditions	Blue Ice
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

Notes

Want a custom formulation?
This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	The single-stranded DNA-binding protein (SSB) also known as La protein in humans has a mechanical role in stabilizing single-stranded DNA (ssDNA) during replication recombination and repair processes. In E. coli the SSB protein weighs approximately 19 kDa and is essential for maintaining genome integrity. SSB expression occurs in various organisms with bacterial SSBs being key to prokaryotic cells while homologs like La/SSB in humans are more involved in post-transcriptional processes. Anti-SSB antibodies target this protein often in research and diagnostic applications.
Biological function summary	The single-stranded DNA-binding protein plays an important role in the orchestration of DNA metabolism. It binds to ssDNA substrates acting as a scaffold for recruiting other proteins and it frequently involves itself in complex formation with other cellular factors such as replication proteins. This function proves important in stabilizing unwound DNA and regulating access for proteins involved in DNA synthesis and repair. Anti-La SSB antibodies are typically used to study aberrant RNA processing events.
Pathways	The SSB protein influences the DNA replication and repair pathways. SSB's interaction with proteins like DNA polymerase and exonuclease suggests its

	cooperation with these enzymes to ensure precise DNA synthesis and to facilitate proper repair of DNA. Anti-SSB Ab can be used to investigate specific pathway dysregulations providing insights into pathway alterations that impact genome stability. Notably SSB's role in coordinating with other repair proteins emphasizes its centrality in maintaining steady state of DNA integrity.
Associated diseases and disorders	The single-stranded DNA-binding protein has vital connections to autoimmune diseases. Sjögren's syndrome and systemic lupus erythematosus often see the presence of anti-SSB antibodies indicating an autoimmune response against this protein. These disorders associate with the human La protein an autoimmune target in these disease states. Understanding SSB's interaction with other proteins such as Ro60 in these conditions gives insights into disease pathology and potential areas for therapeutic intervention.

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

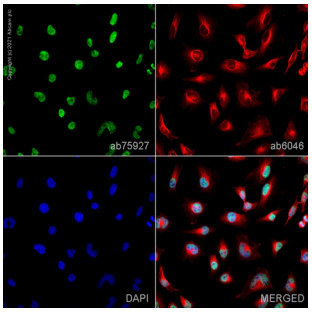
We do not recommend this combination. It is not covered by our product promise.

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

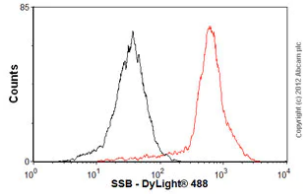


Immunocytochemistry/ Immunofluorescence - Anti-SSB antibody [mAbcam75927] (ab75927)

SSB Immunocytochemistry/ Immunofluorescence staining using mouse Anti-SSB antibody

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).

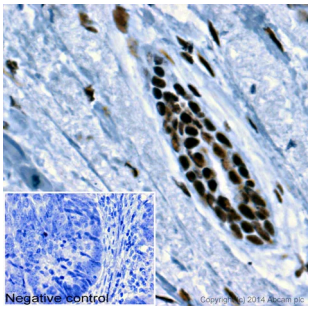
Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



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

SSB Flow Cytometry (Intracellular) staining using mouse Anti-SSB antibody

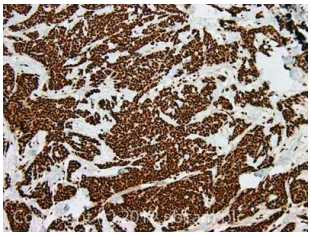
Overlay histogram showing Ramos cells stained with ab75927 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions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

SSB Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-SSB antibody

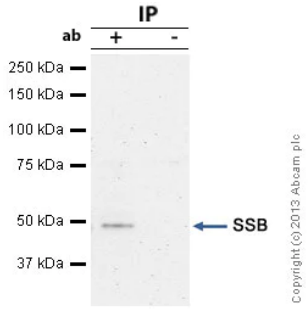
IHC image of SSB staining in Human breast adenocarcinoma formalin fixed paraffin embedded tissue section*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was incubated with ab75927, 0.5µg/ml overnight at +4°C. 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



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

SSB Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-SSB antibody

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.



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

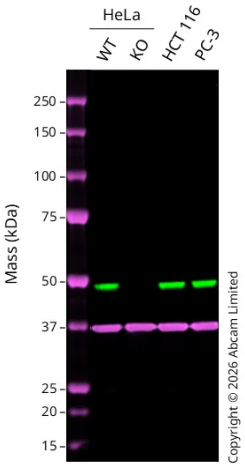
All lanes:
Immunoprecipitation - Anti-SSB antibody [mAbcam75927] (ab75927)

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 46 kDa

Exposure time: 20min



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

SSB Western blot staining using mouse Anti-SSB antibody

Western blot: Anti-SSB antibody [mAbcam75927] ab75927 staining at 5 µg/mL, shown in green; Rabbit anti GAPDH ab181602 loading control staining at 1/20000 dilution, shown in magenta. A band was observed at 49 kDa in Wild-type HeLa cell lysates with no signal observed at this size in SSB knockout HeLa cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 5% 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 800CW and Goat anti-Rabbit 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-SSB antibody [mAbcam75927] (ab75927) at 5 µg/mL

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

Lane 2:
Western blot - Human SSB knockout HeLa cell line (ab274949) at 20 µg

Lane 3:
HCT 116 cell lysates at 20 µg

Lane 4:
PC-3 cell lysates at 20 µg

Secondary

All lanes:
Goat anti-Mouse 800CW & Goat anti-Rabbit 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 47 kDa

Observed band size: 49 kDa



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

SSB Western blot staining using mouse Anti-SSB antibody

All lanes:
Western blot - Anti-SSB antibody [mAbcam75927] (ab75927) at 1 µg/mL

All lanes:
Western blot - Recombinant Human SSB protein (ab84477) at 0.1 µg

Secondary

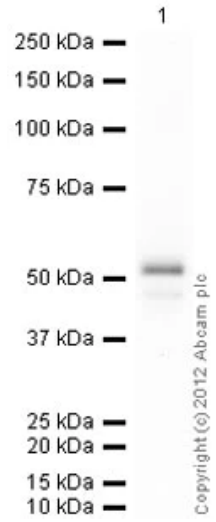
All lanes:
Western blot - Goat Anti-Mouse IgG H&L (HRP) preadsorbed (ab97040) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 46 kDa

Exposure time: 30s



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

SSB Western blot staining using mouse Anti-SSB antibody

All lanes:
Western blot - Anti-SSB antibody [mAbcam75927] (ab75927) at 1 µg/mL

All lanes:
Western blot - Recombinant Human SSB protein (ab84477) at 0.1 µg

Secondary

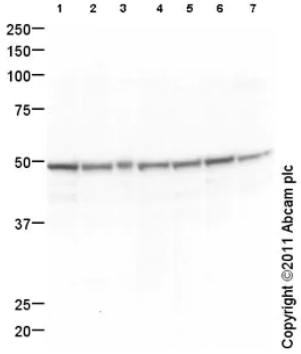
All lanes:
Western blot - Goat Anti-Mouse IgG H&L (HRP) preadsorbed (ab97040) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 46 kDa

Exposure time: 30s



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

SSB Western blot staining using mouse Anti-SSB antibody

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

Lane 1:
HEK293 (Human embryonic kidney cell line) Whole Cell Lysate at 20 µg

Lane 2:
HeLa (Human epithelial carcinoma cell line) Nuclear Lysate at 20 µg

Lane 3:
Raji (Human Burkitt's lymphoma cell line) Nuclear Lysate - tumor cell line (ab30127) at 20 µg

Lane 4:
MCF7 (Human breast adenocarcinoma cell line) Nuclear Lysate at 20 µg

Lane 5:
MCF7 (Human breast adenocarcinoma cell line) Whole Cell Lysate at 20 µg

Lane 6:
Ramos (Human Burkitt's lymphoma cell line) Whole Cell Lysate at 20 µg

Lane 7:
Heart (Human) Whole Cell Lysate - fetal normal tissue at 20 µg

Secondary

All lanes:
Western blot - 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: 46 kDa

Observed band size: 47 kDa

Exposure time: 30s

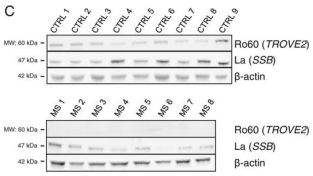


Image collected and cropped by CiteAb under a CC-BY license from the publication

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

SSB Western blot staining using mouse Anti-SSB antibody

SSB western blot using anti-SSB antibody [mAbcam75927] ab75927. Publication image and figure legend from Spurlock, C. F., Tossberg, J. T., et al., 2015, Genome Biol, PubMed 25885816.

ab75927 was used in this publication in western blot. This may not be the same as the application(s) guaranteed by Abcam. For a full list of applications guaranteed by Abcam for ab75927 please see the product overview.

Ro60 and La proteins are depressed in RRMS. (A) Ro60 (TROVE2) and La (SSB) transcript levels in CTRL (N = 24), CIS-MS (N = 16), RRMS (N = 22), RA (N = 18), SLE (N = 24), NMO (N = 22), and PD (N = 19) were determined by quantitative PCR after cDNA synthesis using oligo-dT. Results are normalized to CTRL = 1.0 after normalization to transcript levels of GAPDH, error bars are S.D. (B) As in (A) using whole genome RNA-sequencing data. (C) Western blot to determine Ro60 and La protein levels in PBMC from CTRL (N = 9) and RRMS (N = 8). (D) Quantitative estimates of protein abundance relative to β-actin. *p < 0.05, **p < 0.01.

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