

## Product datasheet

# Mouse monoclonal [SB62α] Anti-Rabbit IgG light chain (HRP) ab99697

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### Overview

Product name	Mouse monoclonal [SB62α] Anti-Rabbit IgG light chain (HRP)
Host species	Mouse
Target species	Rabbit
Specificity	ab99697 reacts with the light chain of native Rabbit IgG primary antibodies and does not bind to the reduced and denatured Rabbit IgG heavy chain band (50kD).
Tested applications	<b>Suitable for:</b> IHC-P, ELISA, WB
Immunogen	Full length protein corresponding to Rabbit IgG Light chain. Rabbit Light Chain
Conjugation	HRP

### Properties

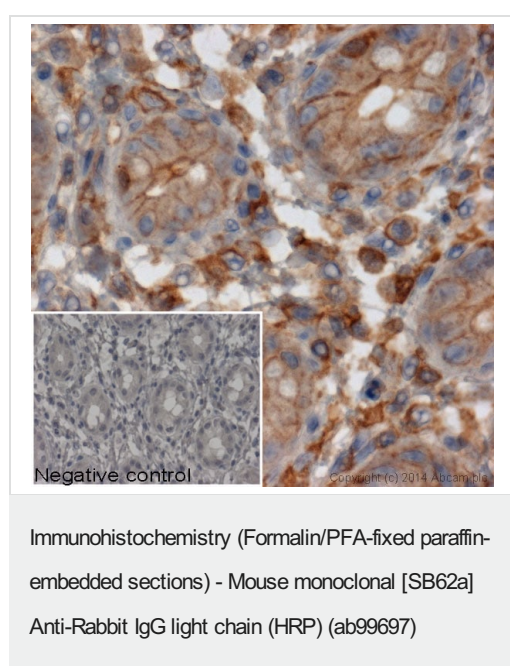
Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C.
Storage buffer	pH: 7.4 Constituents: 50% Glycerol (glycerin, glycerine), 50% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	SB62α
Isotype	IgG2b
Light chain type	lambda

### Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab99697 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
IHC-P		1/8000.
ELISA		1/4000 - 1/8000.
WB		Use at an assay dependent concentration. Note: This antibody is not recommended for sensitive or quantitative detection of the reduced and denatured Rabbit Light Chains on Western Blots.

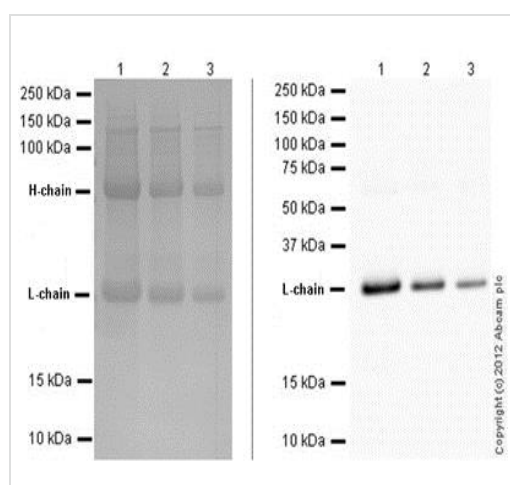
## Images



IHC image of beta actin staining in human colon 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 **ab8227**, 3µg/ml overnight at +4°C. An HRP-conjugated secondary (ab99697, 1/8000 dilution) was used for 1hr at room temperature. The section was counterstained with haematoxylin and mounted with DPX.

The inset negative control image is secondary-only at 1/4000 dilution.

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



Western blot showing that Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) specifically binds to light-chains

**Left blot:** Coomassie stain showing the rabbit heavy (~50kDa) and light-chains (~25 kDa)

Lane 1: 10 µg rabbit monoclonal was loaded

Lane 2: 5 µg rabbit monoclonal was loaded

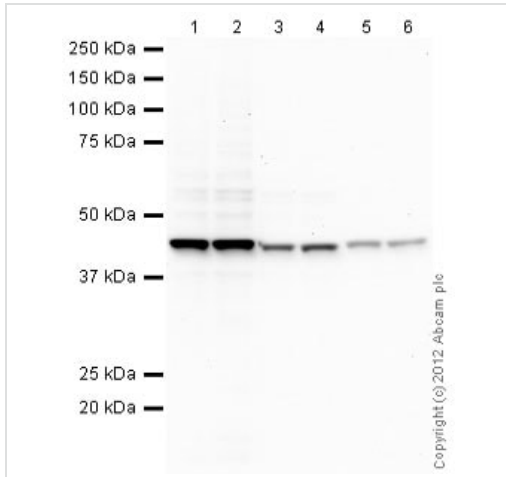
Lane 3: 2 µg rabbit monoclonal was loaded

**Right blot:** WB using Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) as secondary antibody at 1/10,000

Lane 1: 1 µg rabbit monoclonal was loaded

Lane 2: 0.5 µg rabbit monoclonal was loaded

Lane 3: 0.25 µg rabbit monoclonal was loaded



Western blot - Mouse monoclonal [SB62a] Anti-Rabbit IgG light chain (HRP) (ab99697)

**All lanes :** Anti-beta Actin antibody - Loading Control ([ab8227](#)) at 1 µg/ml

**Lanes 1-6 :** HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate ([ab27252](#))  
Lysates/proteins at 10 µg per lane.

#### Secondary

**Lanes 1 - 2 :** Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) at 1/2000 dilution

**Lanes 3 - 4 :** Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) at 1/10000 dilution

**Lanes 5 - 6 :** Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) at 1/20000 dilution

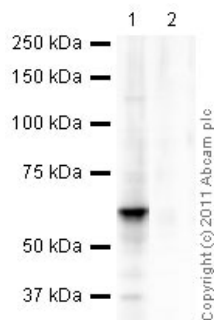
Developed using the ECL technique.

Performed under reducing conditions.



Immunoprecipitation - Mouse monoclonal [SB62a] Anti-Rabbit IgG light chain (HRP) (ab99697)

LAP2 alpha was immunoprecipitated using 0.5mg HeLa whole cell extract, 5µg of Rabbit polyclonal to LAP2 alpha ([ab5162](#)) and 50µl of protein G magnetic beads (lane 1). The antibody was incubated with the Protein G beads for 10min under agitation. No antibody was added to the control (lane 2). HeLa whole cell extract diluted in RIPA buffer was added to each sample and incubated for 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 [ab5162](#). Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).  
Bands: 83kDa: LAP2 alpha.



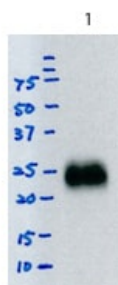
Immunoprecipitation - Mouse monoclonal [SB62a]  
Anti-Rabbit IgG light chain (HRP) (ab99697)

Catalase was immunoprecipitated using 0.5mg Hela whole cell extract, 5µg of Rabbit polyclonal to Catalase ([ab1877](#)) and 50µl of protein G magnetic beads (lane 1). The antibody was incubated with the Protein G beads for 10min under agitation. No antibody was added to the control (lane 2). Hela whole cell extract diluted in RIPA buffer was added to each sample and incubated for 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 [ab1877](#). Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697). Bands: 60kDa: Catalase



Immunoprecipitation - Mouse monoclonal [SB62a]  
Anti-Rabbit IgG light chain (HRP) (ab99697)

Rad21 was immunoprecipitated using 0.5mg Hela whole cell extract, 5µg of Rabbit polyclonal to Rad21 and 50µl of protein G magnetic beads (lane 1). The antibody was incubated with the Protein G beads for 10min under agitation. No antibody was added to the control (lane 2). Hela whole cell extract diluted in RIPA buffer was added to each sample and incubated for 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 [ab42478](#). Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697) .. Band: 100kDa: Rad21; Non specific - 50kDa: We are unsure as to the identity of this extra band. Although the predicted band size is 75kDa based on Swiss-prot data, a band of 120kDa has been previously observed. Mol Cell Biol. 2002 Dec;22(23):8267-77 PMID: 12417729. Th



Western blot - Mouse monoclonal [SB62a] Anti-  
Rabbit IgG light chain (HRP) (ab99697)

Lane 1: Rabbit IgG loaded at 1ug/well  
ab99697 diluted to 1:2000

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