# abcam

### Product datasheet

## Anti-XLF antibody [EPR15882-36] - C-terminal ab189917





#### 1 References 7 Images

#### Overview

**Product name** Anti-XLF antibody [EPR15882-36] - C-terminal

Rabbit monoclonal [EPR15882-36] to XLF - C-terminal **Description** 

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), ICC/IF, WB, IHC-P

Species reactivity Reacts with: Human

Does not react with: Mouse, Rat

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control Ramos, Jurkat and HepG2 whole cell lysate (ab7900); Human endometrial adenocarcinoma;

HepG2 and NCCIT cells; Ramos cells.

**General notes** 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  $\mathsf{RabMAb}^{\mathsf{®}}$  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 +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR15882-36

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab189917 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
Flow Cyt (Intra)		1/60.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/250 - 1/500.
WB		1/10000 - 1/50000. Detects a band of approximately 39 kDa (predicted molecular weight: 33 kDa).
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

#### **Target**

#### **Function**

DNA repair protein involved in DNA nonhomologous end joining (NHEJ) required for double-strand break (DSB) repair and V(D)J recombination. May serve as a bridge between XRCC4 and the other NHEJ factors located at DNA ends, or may participate in reconfiguration of the end bound NHEJ factors to allow XRCC4 access to the DNA termini. It may act in concert with XRCC6/XRCC5 (Ku) to stimulate XRCC4-mediated joining of blunt ends and several types of mismatched ends that are noncomplementary or partially complementary.

## Tissue specificity

#### Involvement in disease

Ubiquitously expressed.

Defects in NHEJ1 are the cause of severe combined immunodeficiency due to NHEJ1 deficiency (NHEJ1-SCID) [MIM:611291]; also known as autosomal recessive T cell-negative, B cell-negative, NK cell-positive, severe combined immunodeficiency with microcephaly, growth retardation and sensitivity to ionizing radiation or NHEJ1 syndrome. SCID refers to a genetically and clinically heterogeneous group of rare congenital disorders characterized by impairment of both humoral and cell-mediated immunity, leukopenia and low or absent antibody levels. Patients with SCID present in infancy with recurrent, persistent infections by opportunistic organisms. The common characteristic of all types of SCID is absence of T-cell-mediated cellular immunity due to a defect in T-cell development. NHEJ1-SCID is characterized by a profound T- and B-lymphocytopenia associated with increased cellular sensitivity to ionizing radiation, microcephaly and growth retardation. Some patients may manifest SCID with sensitivity to ionizing radiation without microcephaly and mild growth retardation, probably due to hypomorphic NHEJ1 mutations.

Note=A chromosomal aberration involving NHEJ1 is found in a patient with polymicrogyria. Translocation t(2;7)(q35;p22).

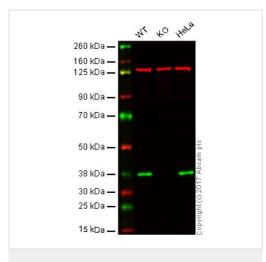
Sequence similarities

Belongs to the XLF family.

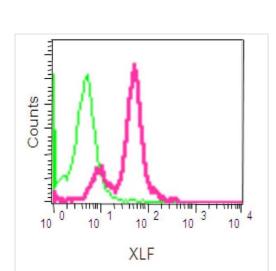
**Cellular localization** 

Nucleus.

#### **Images**



Western blot - Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917)



Flow Cytometry (Intracellular) - Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917)

Lane 1: Wild type HAP1 whole cell lysate (20 µg)

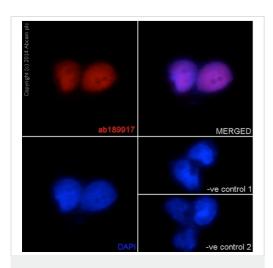
Lane 2: XLF knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

**Lanes 1 - 3:** Merged signal (red and green). Green - ab189917 observed at 38 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

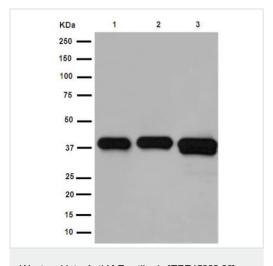
ab189917 was shown to specifically react with XLF in wild type cells as signal was lost in XLF knockout cells. Wild-type and XLF knockout samples were subjected to SDS-PAGE. ab189917 and <a href="mailto:ab18058">ab18058</a> (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/10000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216773">ab216773</a> and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216776">ab216776</a> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

Intracellular flow cytometrical analysis of Ramos cells labeling XLF with ab189917 at 1/60 compared to a negative control cell. FITC-conjugated goat-anti-rabbit secondary antibodies were used for the analysis.



Immunocytochemistry/ Immunofluorescence - Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917)

Immunofluorescent analysis of paraformal dehyde-fixed NCCIT cells labeling XLF with ab189917 at 1/250, Goat anti rabbit  $\lg G$  (Alexa Fluor® 555) at 1/200 and DAPI staining (blue).



Western blot - Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917)

**All lanes :** Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917) at 1/10000 dilution

Lane 1 : Ramos cell lysate
Lane 2 : Jurkat cell lysate
Lane 3 : HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

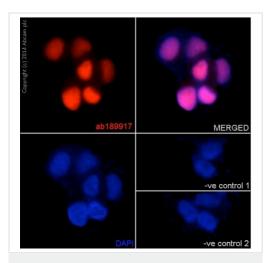
#### Secondary

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

Predicted band size: 33 kDa

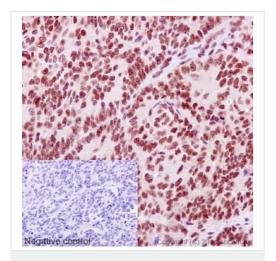
Additional bands at: 39 kDa. We are unsure as to the identity of

these extra bands.



Immunocytochemistry/ Immunofluorescence - Anti-XLF antibody [EPR15882-36] - C-terminal (ab189917)

Immunofluorescent analysis of paraformaldehyde-fixed HepG2 cells labeling XLF with ab189917 at 1/250, Goat anti rabbit  $\lg G$  (Alexa Fluor® 555) at 1/200 and DAPI staining (blue).

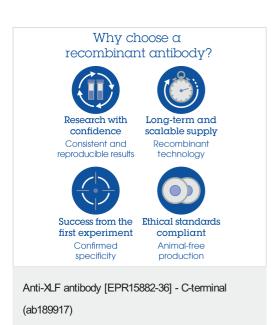


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-XLF antibody

[EPR15882-36] - C-terminal (ab189917)

Immunohistochemical analysis of paraffin-embedded Human endometrial adenocarcinoma tissue labeling XLF with ab189917 at 1/250 with prediluted ImmunoHistoprobe(Ready to use) HRP Polymer for Rabbit IgG as secondary antibody.

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



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