Product Information

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Anti-XLF (C-terminal)
produced in rabbit, affinity isolated antibody

Catalog Number **X4629**

Product Description

Anti-XLF (C-terminal) is produced in rabbit using as immunogen a synthetic peptide corresponding to amino acids 283-299 of human XLF (GeneID: 79840, also known as Nonhomologous end-joining factor 1), conjugated to KLH via an N-terminal added cysteine residue. The immunizing peptide sequence differs from the rat and mouse sequences by three and four amino acids, respectively. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-XLF (C-terminal) specifically recognizes human XLF. Applications include immunoblotting (~ 33 kDa), immunoprecipitation and immunofluorescence. Staining of the XLF band in immunoblotting is specifically inhibited by the immunizing peptide.

The integrity of genetic information depends on the fidelity of DNA replication and on the efficiency of several different DNA repair processes. DNA repair mechanisms include direct repair, base excision repair, nucleotide excision repair, double-strand break repair, and cross-linking repair.[1-3] DNA damage by double-strand DNA breaks (DSBs) is important for cell integrity. Unrepaired DNA ends can cause cell death, chromosomal instability and neoplastic transformation. DSBs can be generated during normal metabolic processes such as DNA replication, recombination, e.g., lymphoid cells during V[D]J recombination, or exposure to exogenous agents, e.g., ionizing radiation or chemotherapeutic compounds.[4, 5] These lesions can be recognized by several pathways among them the nonhomologous end-joining (NHEJ) pathway. This pathway serves to protect and directly ligate broken ends. Six NHEJ factors were discovered: Ku70, Ku80, the DNA-dependent protein kinase catalytic subunit (DNA-PKcs), Artemis, XRCC4 and DNA ligase IV. XLF (also known as cernunnos) protein is an important factor in the NHEJ process. The gene encoding XLF was cloned from individuals with growth retardation, microcephaly, and immunodeficiency with sensitivity to ionizing radiation and defective in V(D)J recombination, indicating defects in the NHEJ pathway.[6, 7] The gene was cloned by transfection of mutant cells with a library of cDNA expression vectors and screening for restoration of ionizing radiation resistance and V(D)J recombination.[6] XLF gene was also cloned from a yeast two-hybrid screen for proteins that interacted with XRCC4.[7] Individuals with sensitivity to ionizing radiation or immunodeficiency have mutations in the XLF gene.[6, 7] This protein may serve as a bridge between XRCC4-Ligase IV and the other NHEJ factors located at the DNA ends, may facilitate recruitment of other factors to the site of repair, and may regulate XRCC4-Ligase IV activity via modulation of active and inactive multimeric states of XRCC4.[1-4]

Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody concentration: ~1 mg/mL

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing, or storage in “frost-free” freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

Immunoblotting: a working concentration of 0.5-1.0 µg/mL is recommended using HEK-293T cell lysates.

Recommendation: for immunoblotting, we strongly advice diluting the antibody in PBS containing 1-3 % non-fat dry milk and 0.05 % TWEEN® 20.

Immunofluorescence: a working concentration of 0.25-0.5 µg/mL is recommended using HEK-293T cells transfected with XLF and fixed with paraformaldehyde-Triton®.

Immunoprecipitation: 2.5-5 µg of antibody are recommended using HEK-293T cell lysate.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

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5. Tsai, C.J., et al., *Proc. Natl. Acad. Sci. USA*, **104**, 7851-7856 (2007).
6. Buck, D., et al., *Cell*, **124**, 287-299 (2006).
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