

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

# **ProductInformation**

# Anti-XPA (N-terminal)

produced in rabbit, affinity isolated antibody

Catalog Number X1504

# **Product Description**

Anti-XPA (N-terminal) is produced in rabbit using as immunogen a synthetic peptide corresponding to amino acids 15-33 of human XPA (GeneID: 7507), conjugated to KLH via a C-terminal added cysteine residue. The immunizing peptide differs from the rat sequence in three amino acids. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-XPA (N-terminal) specifically recognizes human XPA by immunoblotting (doublet at 35 kDa), immunoprecipitation, and immunofluorescence. Staining of the XPA 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. The primary structure of DNA is constantly subjected to alteration by cellular metabolites and exogenous DNA-damaging agents, which cause alterations such as base changes of deletions, fusions, translocations or aneuploidy. The four types of response pathways elicited by DNA damage are DNA repair, DNA damage checkpoints, transcriptional response, and apoptosis. Defects in these pathways may cause genomic instability. 1, 2 DNA repair mechanisms include direct repair, base excision repair, nucleotide excision repair, double-strand break repair, and cross-linking repair. <sup>1-3</sup> Nucleotide excision repair (NER) is the major repair system for removing bulky DNA lesions formed by exposure to UV light or environmental chemicals. The damaged bases are removed by a multi-subunit enzyme system that makes dual incisions bracketing the lesion in the damaged strand. 1, 4-6 The basic steps of NER are (a) damage recognition, (b) dual incisions bracketing the lesion to form a 24-32-nt oligomer in eukaryotes, (c) release of the oligomer, (d) repair synthesis to fill in the resulting gap, and (e) ligation. In human, excision repair is carried out by six repair factors (RPA, XPA, XPC, TFIIH, XPG and XPF/ERCC1), composed of 15 polypeptides. Defects in excision repair cause a photosensitivity syndrome called xeroderma pigmentosum (XP), which is characterized by a very high incidence of light-induced skin cancer.

With respect to the mechanism of NER, the order of arrival of each factor at a lesion remains controversial. However, it is widely accepted that the XPC-hHR23B complex recognizes the DNA damage-induced helical distortion, and the transcription factor TFIIH, XPA (possibly in its homodimeric form), and replication protein A (RPA) arrive sequentially at the site of damage. APA encodes a hydrophilic protein of 273 amino acids. It is a metalloprotein that interacts with many other NER subunits, such as replication protein A (RPA), excision repair complementing 1 protein (ERCC1), and TFIIH. It displays a distinct zinc finger motif, indicating that it interacts directly with DNA; the interaction is weak, and RPA is required for its stabilization. 12, 13

## 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  $\mu$ g/mL is recommended using Jurkat cell lysates.

Immunoprecipitation: a working concentration of 5-10  $\mu$ g/mL is recommended using HEK-293T cell lysates.

Indirect immunofluorescence: a working concentration of 5-10  $\mu$ g/mL is recommended using A549 cells fixed with paraformaldehyde/Triton.

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

## References

- 1. Sancar, A., et al., *Ann. Rev. Biochem.*, **73**, 39-85 (2004).
- Ataian, Y., and Krebs, J.E., Biochem. Cell Biol., 84, 490-504 (2006).
- 3. Lindahl, T., Nature, 362, 709-715 (1993).
- 4. Wood, R.D., *J. Biol. Chem.*, **272**, 23465-23468 (1997).

- 5. Wood, R.D., Biochimie, 81, 39-44 (1999).
- 6. You, J.-S., et al., *J. Biol. Chem.*, **278**, 747-7485 (2003).
- 7. Sancar, A., and Reardon, J.T., *Adv. Protein Chem.*, **69**, 43-71 (2004).
- 8. Neumann, A.S., et al., *Mol. Carcinog.*, **42**, 65-92 (2005).
- 9. Park, C.-J., and Choi, B-S., *FEBS*, **273**, 1600-1608 (2006).
- 10. Mer, G., et al., Cell, 103, 449-456 (2000).
- 11. Tanaka, K., et al., Nature, 348, 73-76 (1990).
- 12. Park, C.H., and Sancar, A., *Proc. Natl. Acad. Sci. USA*, **91**, 5017-5021 (1994).
- 13. Liu, Y., et al., Biochemistry, 44, 7361-7368 (2005).

NV,KAA,PHC 03/07-1