

3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

Anti-PCAF

produced in rabbit, affinity isolated antibody

Catalog Number P7493

Product Description

Anti-PCAF is produced in rabbit using a synthetic peptide corresponding to the C-terminal (amino acids 817–832) of human PCAF with N-terminal added cysteine conjugated to KLH as immunogen. The corresponding sequence is identical in mouse and differs by 2 amino acids residues in chicken. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-PCAF recognizes human and dog PCAF. Applications include immunoblotting (~88 kDa) and immunoprecipitation. In some extract preparations, additional band(s) may appear. Staining of PCAF band(s) by immunoblotting is inhibited by the PCAF immunizing peptide.

The basic repeating unit of eukaryotic chromatin is the nucleosome, which consists of 147 bp of DNA wrapped around an octameric protein core consisting of two molecules each of histones H2A, H2B, H3, and H4. These core histones are important for nuclear packaging and DNA organization as well as for regulating various cell processes.

Among the various post-translational histone modifications, acetylation is the most extensively studied. Histone acetyltransferases (HATs) are members of a superfamily of enzymes that transfer the acetyl moiety from acetyl-coenzyme A cofactor onto one or more ϵ -amino groups of lysines contained in the extended N-terminal tail domains of core histone proteins. Type A HATs are located in the nucleus and many of them play a role as transcriptional coactivators. Type B HATs, traditionally thought to locate to the cytoplasm, acetylate nascent cytoplasmic histones prior to chromatin assembly.

PCAF (p300/CBP associated factor, P/CAF), a type A HAT, has been identified in several species. 1,2 Human PCAF is a ubiquitous protein member of the GCN5-related N-acetyltransferase (GNAT) superfamily. It interacts with the p300 and CBP (CREB-binding protein) transcriptional coactivators and may be found in multi-subunit protein complexes formed from more than 20 polypeptides ranging from 10–400 kDa. 5.6 PCAF shows extensive sequence similarity to human and mouse GCN5 HAT. Human PCAF acetylates all four core-histones. It also acetylates nonhistone proteins such as p53, MyoD, TFIIE-β, TFIIE, GATA-1, EKLF, HMGI (Y), and several other cellular and viral proteins.³ PCAF contributes to transcriptional activation by modifying chromatin and transcriptional factors. It is required in many cell growth and development processes such as myogenesis, and in nuclear receptor mediated or growth factor-signaled cell activation.

Reagent

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

Antibody concentration: 1.0-1.5 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 prolonged storage, freeze in working aliquots at –20 °C. 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 minimum working dilution of 1:500 is determined using whole extracts of dog MDCK kidney cells and a chemiluminescent detection reagent.

<u>Immunoprecipitation</u>: 5–10 μ g of antibody immunoprecipitates PCAF from RIPA lysate with $\sim 4 \times 10^7$ human acute T cell leukemia Jurkat cells.

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

References

- 1. Sterner, D.E., and Berger, S.L., *Microbiol. Mol. Biol. Rev.*, **64**, 435-459 (2000).
- 2. Roth, S.Y. et al., *Ann. Rev. Biochem.*, **70**, 81-120 (2001).
- 3. Kouzarides, T., EMBO J., 19, 1176-1179 (2000).
- 4. Grant, P.A., and Berger, S., *Cell Dev. Biol.*, **10**, 169-177 (1999).
- 5. Marmorstein, R., *Cell. Mol. Life Sci.*, **58**, 693-703 (2001).
- 6. Ogryzko, V.V. et al., Cell, 94, 35-44 (1998).
- 7. Yang, X.J. et al., *Nature*, **382**, 319-324 (1996).

DS,KAA,PHC 01/14-1