

RABBIT ANTI-PARP CLEAVAGE SITE [214/215] POLYCLONAL ANTIBODY

CATALOG NUMBER:	AB3565	QUANTITY:	100 µL
LOT NUMBER:			
		EPITOPE:	Cleavage site 214-215
BACKGROUND:	Poly (ADP-Ribose) Polymerase (PARP) is a 116 kDa nuclear protein, strongly activated by binding to DNA strand breaks. PARP plays a role in DNA repair as well as in other cellular processes, including DNA replication, cell proliferation and differentiation. During apoptosis, ICE family members such as caspase 3 and 7 cleave PARP to yield an 85 kDa and 25 kDa fragment. PARP cleavage is considered to be classically characteristic of apoptosis.		
SPECIFICITY:	The antibody specifically recognizes the 85 kDa fragment of cleaved PARP and can be used as a marker for detecting apoptotic cells.		
IMMUNOGEN:	Synthetic peptide corresponding to the N-terminus of cleavage site (214/215) of human PARP.		

APPLICATIONS: <u>Western blot</u>: 1:1000 Previous lots have been used in immunostaining applications. Optimal working dilutions must be determined by the end user.



Western blot

Extracts from Jurkat cells untreated (1, 2) or stimulated with 0.5 μ M staurosporine for 3 hours (3, 4) to induce apoptosis were transferred to PVDF. The membrane was blocked with 5% BSA-TBST buffer overnight at 4°C, and then incubated with anti-PARP pan (1, 3) or PARP (214/215) cleavage site-specific antibody (CSSA) (2, 4) for 2 hours at room temperature in 3% BSA-TBST buffer. After washing, the membrane was incubated with goat F(ab)₂ anti-rabbit IgG HRP-conjugate and signals detected using standard methods.

Data shows PARP (214/215) CSSA recognizes only the 85 kDa fragment of PARP in apoptotic cells (4) and does not react with full length PARP (2). The anti-PARP pan confirms that non-apoptotic cells express full length PARP of 116 kDa (1), which is then cleaved when apoptosis is induced (3).





Figure 2. Immunohistochemistry

HeLa cells untreated (A) or induced into apoptosis with 0.5 µM staurosporine for 5 hours (B) and fixed in cold acetone for 5 minutes. Cells were incubated with PARP (214/215) CSSA at 10 µg/mL, then incubated with biotinylated goat anti-rabbit IgG and detected by standard DAB staining methods. Data shows the PARP (214/215) CSSA specifically recognizes PARP in apoptotic cells.

- **SPECIES REACTIVITY:** Human. Reactivity with other species has not been confirmed. This sequence is 86% homologous in mouse, rat, and hamster.
- **CONTROL:** Jurkat or HeLa cells treated with staurosporine or etoposide (25 µM for 3 hours).
- **FORMAT:** Affinity purified from rabbit serum using a peptide corresponding to the PARP cleavage site.
- **PRESENTATION:** PBS (without Mg²⁺/Ca²⁺), pH 7.3, 50% glycerol,1 mg/mL BSA, and 0.05% sodium azide.
- **STORAGE/HANDLING:** Store at –20°C in undiluted aliquots for up to one year from date of receipt. For shipment or short-term storage (up to one week), 2-8°C is sufficient.
- **REFERENCES:** Le Page, F., *et al.* (2003) Poly(ADP-ribose) polymerase-1 (PARP-1) is required in murine cell lines for base excision repair of oxidative DNA damage in the absence of DNA polymerase beta. J. Biol. Chem. 278(20):18471-18477.

Turturro, F., *et al.* (2002) Model of inhibition of the NPM-ALK kinase activity by herbimycin A. Clin. Cancer Res. 8(1):240-245 (cites the use of this antibody).

Leemans, J.C., *et al.* (2001) Depletion of alveolar macrophages exerts protective effects in pulmonary tuberculosis in mice. J. Immunol. 166(7):4604-4611 (cites the use of this antibody).

Soldani, C. and A.I. Scovassi (2002) Poly(ADP-ribose) polymerase-1 cleavage during apoptosis: an update. Apoptosis 7(4):321-328.

Germain, M., *et al.* (1999) Cleavage of automodified Poly (ADP-ribose) polymerase during apoptosis. Evidence for involvement of caspase-7. J. Biol. Chem. 274(40):28379-28384.

Kaufmann, S.H., *et al.* (1993) Specific proteolytic cleavage of poly (ADP-ribose) polymerase: an early marker of chemotherapy-induced apoptosis. Cancer Res. 53(17):3976-3985.

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