Anti-PARP CSSA FITC

Apoptosis Detection Kit

Catalog Number: **AHM2011**

Poly (ADP-Ribose) Polymerase (PARP) is a 116 kDa nuclear protein, which is strongly activated by DNA strand breaks. During apoptosis, ICE family members, such as caspase-3 and -7, cleave PARP to yield an 85 kDa and a 25 kDa fragment. PARP cleavage is considered to be one of the classical characteristics of apoptosis. This FITC-conjugated anti-PARP antibody specifically recognizes the 85 kDa fragment of cleaved PARP and can be used as a marker for detecting apoptotic cells. In this method, apoptosis is detected by flow cytometry using an intracellular staining method. Adherent or suspension cells are induced into apoptosis using the researchers preferred method. The cells are collected by centrifugation and fixed with provided buffer. The fixed cells are permeabilized to allow the entry of the FITC-labeled anti-PARP cleavage site specific antibody into the intracellular environment of the cell. The FITC-conjugated antibody specifically binds to the cleaved PARP, not the full length intact protein. Cells are then analyzed by FACS and positive FITC staining is a measure of the percentage of cells undergoing apoptosis.

Reagents Provided (sufficient for 100 tests):

1. Rabbit anti-PARP cleavage site specific antibody (CSSA) polyclonal antibody (FITC conjugate); 1 mL in buffered saline containing BSA and 0.1% sodium azide.
2. IC Fix™, 100 mL paraformaldehyde in buffered saline.
3. IC Perm™, (5x solution); 250 mL containing buffered saline, normal serum, and saponin. 1x IC Perm™; dilute 100 mL 5x solution with 400 mL of distilled H_2O. 1x solution is stable at 4°C for up to 6 months.
4. PARP cleavage site peptide; 50 µg reconstituted peptide at 0.05 mg/mL.

References:

1. Duriez, P.J. and G.M. Shah (1997) Cleavage of poly (ADP-ribose) polymerase: a sensitive parameter to study cell death. Biochem. Cell. Biol. 75(4):337-349.
2. 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.
3. 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.
4. Tewari, M. et al. (1995) Tama/CPP32 beta, a mammalian homolog of CED-3, is a CrmA-inhibitable protease that cleaves the death substrate poly (ADP-ribose) polymerase. Cell 81(5):801-809.
5. Kumar, A.P. et al. (2001) 2-Methoxyestradiol blocks cell-cycle progression at G(2)/M phase and inhibits growth of human prostate cancer cells. Mol. Carcinogenesis (3):111-124.