

Certificate of Analysis

Product: Anti-Apaf1 Protein A Purified Mouse Monoclonal Antibody

Code: 200-301-A35

Lot # 19595

Size: 100 µg

Physical State: Liquid (sterile filtered)

Clone: 2E10

Isotype: IgG

Antibody Concentration: 1.0 mg/ml (by UV absorbance at 280 nm)

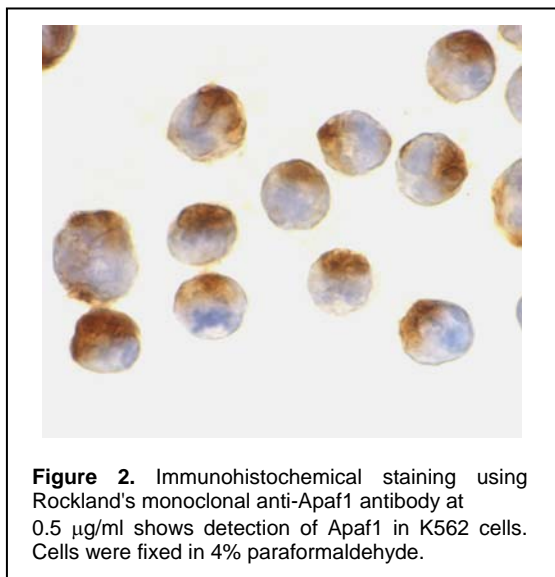
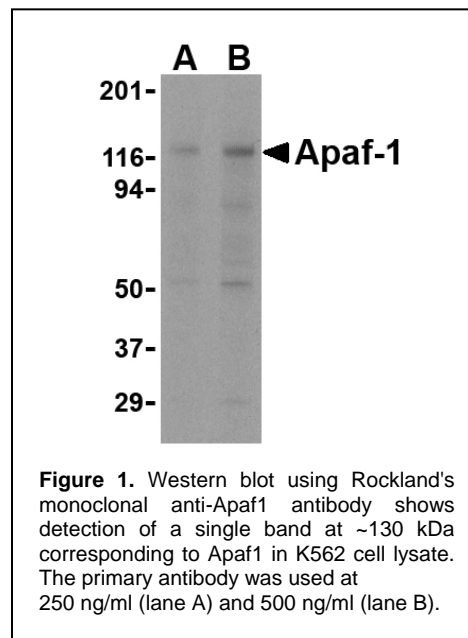
Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

Stabilizer: None

Preservative: 0.02% (w/v) Sodium Azide

Storage Conditions: Store vial at -20° C prior to opening. Dilute only prior to immediate use. For extended storage, aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Expiration date is one (1) year from date of opening.

Background: Apoptosis is related to many diseases and is induced by a family of cell death receptors and their ligands. Cell death signals are transduced by death domain containing adapter molecules and members of the caspase family of proteases. The mammalian homologue of the key cell death gene CED-4 in *C. elegans* was identified recently from human and mouse and designated Apaf1 for apoptosis protease-activating factor 1. Apaf1 binds to cytochrome c (Apaf2) and caspase-9 (Apaf3), which leads to caspase-9 activation. Activated caspase-9 in turn cleaves and activates caspase-3, one of the key proteases responsible for the proteolytic cleavage of many key proteins in apoptosis. Apaf1 can also associate with caspase-4 and caspase-8. Apaf1 transcript is ubiquitously expressed in human tissues.



Application Note(s): This affinity purified antibody has been tested for use in ELISA, immunohistochemistry and western blotting. Specific conditions for reactivity should be optimized by the end user. Paraformaldehyde fixation is recommended for immunohistochemical staining. Expect a band at ~130 kDa in size corresponding to Apaf1 by western blotting in the appropriate cell lysate or extract. K562 cells can be used as a positive control.

Recommended Dilutions:

ELISA	1:10,000 - 1:40,000
WESTERN BLOT	1:200 - 1:2,000
IF MICROSCOPY	1:100 - 1:1,000
OTHER APPLICATIONS	User Optimized

Purity and Specificity: This monoclonal antibody is directed against human Apaf1 protein. The product was purified from Ascites by protein A chromatography. Expect reactivity with human, mouse and rat Apaf1. Cross-reactivity with Apaf1 from other sources has not been determined.

Relevant Link(s): Swiss-Prot [O14727](#) NCBI Link [O14727](#)

Immunogen: This protein A purified monoclonal antibody was prepared using conventional hybridoma technology after repeated immunizations with a synthetic peptide corresponding to a region near the carboxy terminus of human Apaf1 protein.

Hybridoma: Produced by the fusion between mouse splenocytes and mouse myeloma SP2/0 cells using conventional hybridoma technology.

General References:

Zou, H., *et al.* (1997) Apaf-1, a human protein homologous to C. elegans CED-4, participates in cytochrome c-dependent activation of caspase-3. *Cell* **90**:405-13.

Cecconi, F., *et al.* (1998) Apaf1 (CED-4 homolog) regulates programmed cell death in mammalian development. *Cell* **94**:727-37.

Li, P., *et al.* (1997) Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. *Cell* **91**:479-89.

Hu, Y., *et al.* (1998) Bcl-XL interacts with Apaf-1 and inhibits Apaf-1-dependent caspase-9 activation. *Proc. Natl. Acad. Sci. USA* **95**:4386-91.

Related Products:

#[W09-000-364](#) HeLa Whole Cell Lysate in SDS-PAGE Sample Buffer
#[W09-000-365](#) 293 Whole Cell Lysate in SDS-PAGE Sample Buffer
#[W09-001-370](#) Jurkat Whole Cell Lysate in SDS-PAGE Sample Buffer
#[611-703-127](#) Peroxidase Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (DONKEY) MX10
#[611-132-122](#) IRDye800 Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (GOAT) MX10
#[MB-070](#) Blocking Buffer for Fluorescent Western Blotting
#[KIA-003](#) **MaxTag**TM Anti-RABBIT IgG Kit for Immunoblotting

USDA Certification: All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation.

Note: This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information.