

Technical Data Sheet

Purified Rabbit Anti-AIF w/control

Product Information

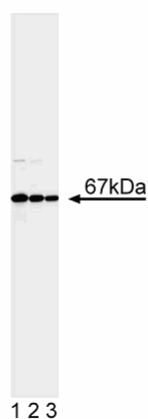
Material Number: 551429
Size: 100 µl
Reactivity: QC Testing: Human
 Reported: Mouse, Rat

Component: 51-8103KC
Description: Purified Rabbit Anti-AIF
Size: 100 µg (1 ea)
Clone Name: Polyclonal
Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Component: 51-16576N
Description: K-562 Control Lysate
Size: 50 µg (1 ea)
Concentration: 1.0 mg/ml
Storage Buffer: SDS-PAGE buffer (62mM Tris pH 6.8, 2% SDS, 0.9% β-mercaptoethanol, 0.003% bromophenol blue, 5% glycerol)

Description

Apoptosis is essential in maintaining normal cellular homeostasis and for normal development. Morphological as well as biochemical changes are a hallmark of apoptosis, and include membrane blebbing, chromosomal condensation and DNA fragmentation. The mitochondria plays an essential role in apoptosis. During apoptosis, the outer mitochondrial membrane (OMM) becomes permeable, releasing cytochrome c into the cytosol, which then binds Apaf-1 and in the presence of dATP/ATP can bind procaspase-9. Procaspase-9 is activated and forms a complex with cytochrome c/Apaf-1 called an apoptosome, which in turn can activate additional caspases, such as caspase-3. In addition to the release of cytochrome c, another protein has been identified, AIF (apoptosis-inducing factor), which is also released through the OMM. AIF has been cloned and has a predicted molecular weight of 57 kDa based upon its cDNA sequence. Characterization of AIF shows it to be a flavoprotein which has some homology to bacterial oxidoreductases. Upon induction of apoptosis, AIF translocates through the OMM to the cytosol and nucleus where it can initiate chromatin condensation. Bcl-2 can block the release of AIF from the mitochondria, but the apoptotic effect cannot be blocked if AIF has been released into the cytosol. Use of the pan caspase inhibitor, Z-VAD-FMK cannot block the phenotypic effects caused by AIF, indicating that AIF may act independently of caspases and Bcl-2 regulation. Furthermore, AIF has been shown to be essential for programmed cell death in murine embryonic morphogenesis and cavitation. Likewise, when AIF is genetically inactivated, embryonic stem cells are resistant to cell death following serum starvation. The antibodies recognize mouse, rat, and human AIF. A peptide corresponding to amino acids 517-531 of human AIF was used as the immunogen.



Western blot analysis of AIF. Lysate from K-562 cells was probed with anti-AIF at dilutions of 1:1000 (lane 1), 1:2000 (lane 2), and 1:4000 (lane 3). AIF is identified as a band of ~67 kDa.

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Preparation and Storage

The polyclonal antibody was purified from antiserum by negative adsorption and affinity chromatography.

Store the antibody at 4°C, store positive control lysate (Cat. No. 51-16576N) at -20°C.

Application Notes

Application

Western blot	Routinely Tested
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Recommended Assay Procedure:

Applications include western blot analysis (1:1000-1:4000). Store the antibody at 4°C. For long-term storage, the antibody may be stored at -20°C. Avoid multiple freeze-thaw cycles. K-562 control lysate [50 µg (1 µg/µl)] is provided as a positive control (Cat. No. 51-16576N; store lysate at -20°C). Additional K-562 control lysate (Cat. No. 611550) is sold separately as a ready-to-use western blot control. The antibodies are routinely tested by western blot analysis on K-562 cells.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554021	HRP Goat Anti-Rabbit Ig	1.0 ml	(none)
611550	K-562 Cell Lysate	500 µg	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

References

Joza N, Susin SA, Daugas E, et al. Essential role of the mitochondrial apoptosis-inducing factor in programmed cell death. *Nature*. 2001; 410(6828):549-554. (Biology)
Martinou JC, Green DR. Breaking the mitochondrial barrier. *Nat Rev Mol Cell Biol*. 2001; 2(1):63-67.(Biology)
Susin SA, Lorenzo HK, Zamzami N, et al. Molecular characterization of mitochondrial apoptosis-inducing factor. *Nature*. 1999; 397(6718):441-446.(Biology)