Technical Data Sheet

Purified Mouse Anti-SREBP-1

Product Information

Material Number:	**557036**
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	IgG-2A4
Immunogen:	Human SREBP-1 aa. 301-407
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human Tested in Development: Hamster
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

SREBP-1 and -2 (**s**terol-**r**egulatory **e**lement **b**inding **p**roteins-1 and -2) are transcription factors which participate in the control of cholesterol homeostasis. SREBPs proteins, which are attached to the endoplasmic reticulum and nuclear envelope, are proteolytically cleaved and thus activated in response to conditions of low cellular sterol. Upon activation of SREBP-1 or -2, an ~480-500 amino acid, N-terminal cleavage fragment of these proteins enters the nucleus and activates transcription of enzymes and other proteins required for cholesterol synthesis. Proteases which cleave SREBPs have been identified and include SCA (SREBP-cleavage activity), as well as a key regulator of apoptotic pathways, caspase-3. SREBP proteins containing point mutations at caspase-3 cleavage sites (Asp460 in SREBP-1 and Asp468 in SREBP-2) do not become cleaved following induction of apoptosis, suggesting that SREBPs may play some role in apoptotic processes. However, sterol-regulated vs. apoptosis-associated cleavage of SREBP proteins appears to be independantly regulated. On SDS-PAGE, sterol-regulated cleavage fragments of SREBP proteins migrate more slowly (i.e., higher molecular weight) than do staurosporin-induced fragments. In addition, staurosporin-induced SREBP cleavage products may appear as a doublet, with the upper band representing a phosphorylated form of SREBP. On SDS-PAGE, full length, precursor forms of SREBP-1 and -2 migrate at ~125 kDa, while proteolytic cleavage fragments may be observed as a cluster of bands between 60 - 70 kDa. The IgG-2A4 antibody recognizes human and hamster SREBP-1. The antibody recognizes the N-terminal (basic helix-loop-helix) domain of human SREBP-1. A fusion protein containing N-terminal amino acids 301-407 (the bHLH/leucine zipper domain), was used as immunogen. The antibody recognizes both the 125 kDa precursor and 60-70 kDa mature, cleaved forms of SREBP-1.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

Application Notes

Application

Western blot	Routinely Tested
Immunoprecipitation	Tested During Development

Recommended Assay Procedure:
The IgG-2A4 antibody may be used for western blot analysis (0.5 to 1 µg/ml) and immunoprecipitation (2 µg/ml). U-937 human histiocytic lymphoma cells (ATCC CRL-1593) or HeLa human cervical carcinoma cells (ATCC CCL-2) are recommended as positive controls for these applications.

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

Sakai J, Nohturfft A, Cheng D, Ho YK, Brown MS, Goldstein JL. Identification of complexes between the COOH-terminal domains of sterol regulatory element-binding proteins (SREBPs) and SREBP cleavage-activating protein. *J Biol Chem.* 1997; 272(32):20213-20221.(Clone-specific: Western blot)

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Sato R, Yang J, Wang X, et al. Assignment of the membrane attachment, DNA binding, and transcriptional activation domains of sterol regulatory element-binding protein-1 (SREBP-1). *J Biol Chem.* 1994; 269(25):17267-17273.(Immunogen)
Wang X, Pai JT, Wiedenfeld EA, et al. Purification of an interleukin-1 beta converting enzyme-related cysteine protease that cleaves sterol regulatory element-binding proteins between the leucine zipper and transmembrane domains. *J Biol Chem.* 1995; 270(30):18044-18050.(Biology)
Wang X, Zelenski NG, Yang J, Sakai J, Brown MS, Goldstein JL. Cleavage of sterol regulatory element binding proteins (SREBPs) by CPP32 during apoptosis. *EMBO J.* 1996; 15(5):1012-1020.(Clone-specific: Western blot)