Technical Data Sheet

Purified Mouse Anti-SREBP-2

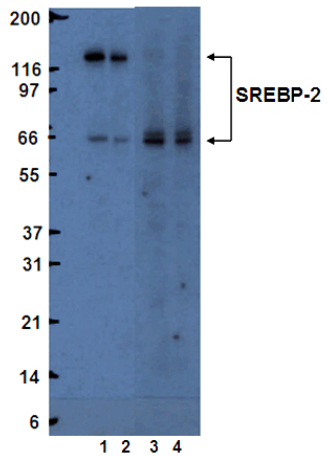
Product Information

Material Number:	**557037**
Alternate Name:	SREBF2; sterol regulatory element-binding protein 2
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	IgG-1C6
Immunogen:	Human SREBP-2 aa. 833-1141
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

SREBP-1 and -2 (sterol-regulatory element binding proteins-1 and -2) are transcription factors which participate in the control of cholesterol homeostasis. SREBP 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, a ~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 caspase-3, a key regulator of apoptotic pathways. 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 independently 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 kD, while proteolytic cleavage fragments may be observed as a cluster of bands between 60 - 70 kDa.

The IgG-1C6 antibody recognizes human SREBP-2. The antibody recognizes the C-terminal of human SREBP-2. A fusion protein containing C-terminal amino acids 833-1141 of human SREBP-2, was used as immunogen. The antibody recognizes both the 125 kDa precursor and the 60-70 kDa COOH-terminal cleaved forms of SREBP-2.



Western blot analysis of SREBP-2. *Jurkat (ATCC TIB-152) cells were left untreated (lanes 1 & 2) or treated with 1 µM Staurosporin for 4 hours (lanes 3 & 4) to induce apoptosis. Cell lysates were probed with Purified Mouse Anti-SREBP-2 (Cat. No. 557037) and visualized with HRP Goat Anti-Mouse Ig (Cat. No.554002) . As apoptosis is induced, the SREBP-2 125 kDa precursor is cleaved to its ~70 kDa size.*

Preparation and Storage

Store undiluted at 4°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

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Application Notes

Application

Western blot	Routinely Tested
Immunoprecipitation	Tested During Development

Recommended Assay Procedure:

The IgG-1C6 antibody may be used for western blot analysis (2 µg/ml) and immunoprecipitation (2 µg antibody/300-500 µg of total protein lysate). U-937 human histiocytic lymphoma cells (ATCC CRL-1593) or HeLa human cervical carcinoma cells (ATCC CCL-2) are recommended as additional positive controls.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1 mL	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. 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.
3. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Hua X, Sakai J, Ho YK, Goldstein JL, Brown MS. Hairpin orientation of sterol regulatory element-binding protein-2 in cell membranes as determined by protease protection. *J Biol Chem.* 1995; 270(49):29422-29427. (Immunogen)

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)

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)