

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human F-Spondin/SPON1 in direct ELISAs and Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human F-Spondin/SPON1 Phe29-Cys807 Accession # Q9HCB6
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

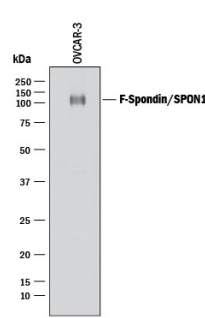
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Simple Western	10 µg/mL	See Below
Neutralization	Measured by its ability to neutralize the the enhancement of neurite outgrowth of dorsal root ganglion neurons from E13 chick embryos induced by F-Spondin. 3 µg/mL is sufficient to block neurite outgrowth induced by 90 ng/3 µL drop/well of F-Spondin immobilized on nitrocellulose coated plate.	

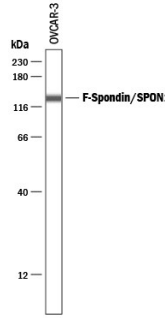
DATA

Western Blot




Detection of Human F-Spondin/SPON1 by Western Blot. Western blot shows lysates of OVCAR-3 human ovarian carcinoma cell line. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human F-Spondin/SPON1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3135) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for F-Spondin/SPON1 at approximately 110 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Simple Western



Detection of Human F-Spondin/SPON1 by Simple Western™. Simple Western lane view shows lysates of OVCAR-3 human ovarian carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for F-Spondin/SPON1 at approximately 134 kDa (as indicated) using 10 µg/mL of Goat Anti-Human F-Spondin/SPON1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3135) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.



PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

F-Spondin ("Floor plate" and "thrombospondin" homology; also Spondin-1 and VSGP) is a member of the F-Spondin family of proteins that collectively belong to a subgroup of TSR (thrombospondin) type I class molecules (1). Class I molecules are either membrane-bound or ECM-associated. F-Spondin is a 110 kDa, secreted, heparin-binding extracellular matrix glycoprotein first characterized in rat for its high expression in embryonic floor plate (2-4). Human F-Spondin is synthesized as an 807 amino acid (aa) precursor that contains a 28 aa signal sequence and a 779 aa mature region (3, 4). The mature region includes an N-terminal reelin-like domain (aa 1-200), a centrally placed F-Spondin (FS) type segment (aa 201-440), and six C-terminal class 2 thrombospondin type I repeats (1, 3, 5). Class 1 and 2 repeats differ in the placement of their cysteine residues. The fifth and sixth TSP repeats (aa 668-806) apparently bind ECM, while TSP repeats 1-4 (aa 442-666), plus the spondin segment, are suggested to mediate either repulsive activity (on motor neurons), or outgrowth promoting activity (on sensory neurons) (1, 6). At least two isoforms of F-Spondin are known. Both are proteolytically-generated, one by plasmin, another by an unidentified protease. Plasmin cleaves the C-terminus at two points, generating a soluble, 95 kDa, 656 aa F-Spondin that contains all but TSP repeats #5 and 6 (7). The unidentified protease appears to cleave F-Spondin between the FS segment and the first TSP repeat, generating 60 kDa and 50 kDa fragments, respectively (6). F-Spondin shows highly unusual glycosylation, exhibiting both C-mannosylation (mannose bound to Trp) and O-fucosylation (fucose bound to Ser/Thr) (4). The significance of these glycosidic modifications is unknown. Mature human F-Spondin is 98%, 97%, 98%, and 97% aa identical to mature canine, rat, bovine and mouse F-Spondin, respectively. Mammalian cells known to express F-Spondin include floor plate epithelium, ventral motor neurons, Schwann cells, fibroblasts, hippocampal pyramidal cells, endothelial cells, vascular smooth muscle cells and some tumor cells (6, 8, 9).

References:

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