Recombinant Human R-Spondin 1

Catalog Number: 4645-RS/CF

DESCRIPTION

Source	Chinese Hamster Ovary cell line, CHO-derived Arg31-Ala263 Accession # Q2MKA7
N-terminal Sequence Analysis	Arg31
Predicted Molecular Mass	25.6 kDa

SPECIFICATIONS

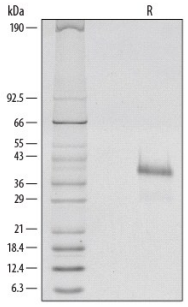
SDS-PAGE	39 kDa, reducing conditions
Activity	Measured by its ability to induce Topflash reporter activity in HEK293T human embryonic kidney cells. The ED_{50} for this effect is typically 1-4 ng/mL in the presence of 5 ng/mL Recombinant Mouse Wnt-3a (Catalog # 1324-WN).
Endotoxin Level	<0.10 EU per 1 µg of the protein by the LAL method.
Purity	>90%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 100 µg/mL in PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 3 months, -20 to -70 °C under sterile conditions after reconstitution.

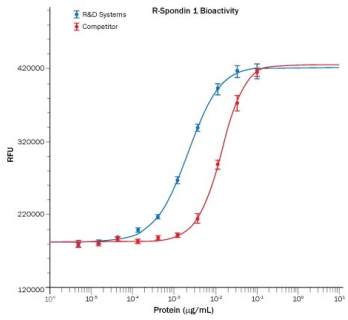
DATA

SDS-PAGE



1 µg/lane of Recombinant Human R-Spondin 1 was resolved with SDS-PAGE and visualized by silver staining, under reducing (R) conditions, showing a single band at 39 kDa.

Bioactivity



Recombinant Human R-Spondin 1 (Catalog # 4645-RS/CF), in the presence of Recombinant Mouse Wnt-3a (Catalog # 1324-WN; 5 ng/mL), induces activation of beta-catenin in HEK293T cells measured using the Topflash assay (blue). The activity is approximately 7-fold greater than the competitor's R-Spondin 1 (red).

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BACKGROUND

R-Spondin 1 (RSPO1, Roof plate-specific Spondin 1), also known as cysteine-rich and single thrombospondin domain containing protein 3 (Cristin 3), is a 27 kDa secreted protein that shares ~40% amino acid (aa) identity with three otherR-Spondin family members (1, 2). All R-Spondins regulate Wnt/β-catenin signaling, but have distinct expression patterns (1-3). Like other R-Spondins, R-Spondin 1 contains two adjacent cysteine-rich furin-like domains (aa 34-135) with one potential N-glycosylation site, followed by a thrombospondin (TSP-1) motif (aa 147-207) and a region rich in basic residues (aa 211-263). Only the furin-like domains are needed for β-catenin stabilization (2, 4). A putative nuclear localization signal at the C-terminus may allow some expression in the nucleus (5). Potential isoforms of 200 and 236 aa have an alternate, shorter N-terminus or are missingaa 146-208, respectively (6). Over aa 21-263, human R-Spondin 1 shares 89%, 87%, 92%, 91%, 91% and 89% aa identity with mouse, rat, equine, canine, caprine and bovine RSPO-1, respectively. R-Spondin 1 is expressed in early development at the roof plate boundary and is thought to contribute to dorsal neural tube development (3, 5). In humans, rare disruptions of the R-Spondin 1 gene are associated with tendencies for XX sex reversal (phenotypic male) or hermaphroditism, indicating a role for R-Spondin 1 in gender-specific differentiation (7, 8). Disruption is also associated with palmoplantar keratosis (7, 8). Postnatally,R-Spondin 1 is expressed by neuroendocrine cells in the intestine, adrenal gland and pancreas, and by epithelia in kidney and prostate (9). Injection of recombinant R-Spondin 1 in mice causes activation of β-catenin and proliferation of intestinal crypt epithelial cells, and ameliorates experimental colitis (9, 10). R-Spondin 1 regulates Wnt/β-catenin by competing with the Wnt antagonist DKK-1 for binding to the Wnt co-receptors, Kremen and LRP-6, reducing their DKK-1-mediated internalization (11). Reports differ on whether R-Spondin 1 binds LRP-6 directly (11-13).

References:

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