abcam

Product datasheet

SMAD1 (pS463/465) ELISA Kit ab186036

SimpleStep ELISA

1 References 7 Images

Overview

Product name SMAD1 (pS463/465) ELISA Kit

Detection method Colorimetric

Precision Intra-assay

Sample	n	Mean	SD	CV%
C2C12	6			2.3%

Inter-assay

Sample	n	Mean	SD	CV%	
C2C12	3			7.5%	

Sample type Cell Lysate, Tissue Homogenate

Assay type Semi-quantitative

Sensitivity 10 µg/ml

Range 10 μg/ml - 500 μg/ml

Assay time 1h 30m

Assay duration One step assay

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat

Product overview

Abcam's SMAD1 (pS463/S465) *in vitro* SimpleStep ELISA™ (Enzyme-Linked Immunosorbent Assay) kit is designed for the semi-quantitative measurement of SMAD1 (pS463/S465) protein in human and mouse cells.

The SimpleStep ELISA™ employs a labeled capture and detector antibody which immunocaptures the sample analyte in solution. This entire complex (capture antibody/protein/detector antibody) is in turn immobilized in the well by immunoaffinity via the antitag antibody. Samples or standards are added to the wells, followed by the antibody mix. After incubation, the wells are washed to remove unbound material; the TMB substrate is then added. The reaction is stopped by addition of Stop Solution which stops the color development and completes any color change from blue to yellow. Signal is generated proportionally to the amount

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of bound analyte and the intensity is measured at 450 nm. Optionally, instead of the endpoint

reading, development of TMB can be recorded kinetically at 600 nm.

Notes Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of

products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of REACH

Authorisation, and any other relevant authorisations, for their intended uses.

Platform Microplate

Properties

Storage instructions Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Wash Buffer PT	1 x 15ml
50X Cell Extraction Enhancer Solution	1 x 1ml
5X Cell Extraction Buffer PTR	1 x 12ml
Lyophilized SMAD1 (pS463/S465) Control Lysate	1 vial
Plate Seal	1 unit
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
SMAD1 (pS463/S465) Capture Antibody	1 x 3ml
SMAD1 (pS463/S465) Detector Antibody	1 x 3ml
Stop Solution	1 x 12ml
TMB Substrate	1 x 12ml

Function Transcriptional modulator activated by BMP (bone morphogenetic proteins) type 1 receptor

kinase. SMAD1 is a receptor-regulated SMAD (R-SMAD). SMAD1/OAZ1/PSMB4 complex

mediates the degradation of the CREBBP/EP300 repressor SNIP1.

Tissue specificity Ubiquitous. Highest expression seen in the heart and skeletal muscle.

Sequence similarities Belongs to the dwarfin/SMAD family.

Contains 1 MH1 (MAD homology 1) domain. Contains 1 MH2 (MAD homology 2) domain.

Post-translational

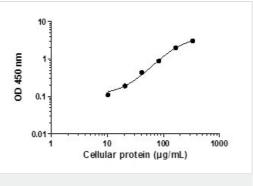
Phosphorylated on serine by BMP type 1 receptor kinase.

modifications Ubiquitin-mediated proteolysis by SMAD-specific E3 ubiquitin ligase SMURF1.

Cellular localization Cytoplasm. Nucleus. Cytoplasmic in the absence of ligand. Migrates to the nucleus when

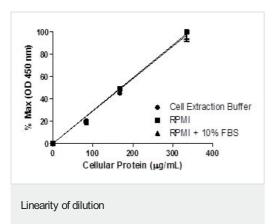
complexed with SMAD4. Co-localizes with LEMD3 at the nucleus inner membrane.

Images

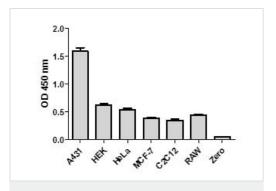


Example of a typical SMAD1 (pS463/S465) cell lysate dilution series. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.

Typical cell lysate dilution series

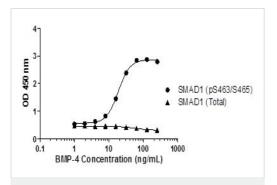


Linearity of dilution in representative sample matrices. Cellular lysates were prepared at 3 concentrations in common media containing 1X Cell Extraction Buffer PTR. Data from duplicate measurements of SMAD1 (pS463/S465) are normalized and plotted.



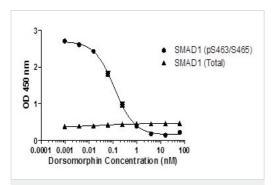
Cell line analysis for Total SMAD1 from 300 μ g/mL preparations of cell extracts. Data from triplicate measurements (mean +/- SD) are plotted and compared to 1X Cell Extraction Buffer PTR (zero).

Comparison of total SMAD1 expression in different cell lines.



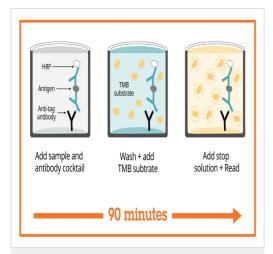
SMAD1 (pS463/S465) phosphorylation in response to BMP-4 treatment.

Induction of SMAD1 (pS463/S465) phosphorylation in HeLa cells in response to BMP-4 treatment. HeLa cells were cultured in 96-well tissue culture plates, serum-starved and treated (30 min) with a dose-range of BMP-4 before cell lysis. Data from quadruplicate measurements of SMAD1 (pS463/S465) are plotted.



SMAD1 (pS463/S465) phosphorylation in response to dorsomorphin treatment.

Inhibition of SMAD1 (pS463/S465) phosphorylation in HeLa cells in response to dorsomorphin treatment. HeLa cells were cultured in 96-well tissue culture plates and treated with a dose-range of dorsomorphin (30 min). Cells were then stimulated with BMP-4 (30 min) and lysed. Data from quadruplicate measurements of SMAD1 (pS463/S465) and SMAD1 (Total) are plotted.



Sandwich ELISA - SMAD1 (pS463/465) ELISA Kit (ab186036)

SimpleStep ELISA technology allows the formation of the antibodyantigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.



(ab186036)

To learn more about the advantages of SimpleStep ELISA[®] kits see <u>here</u>.

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