

His-probe (H-15): sc-803

BACKGROUND

Plasmid vectors for the expression of coding regions of eukaryotic genes in bacterial, insect and mammalian hosts are in common usage; such expression vectors are frequently used to encode hybrid fusion proteins consisting of a eukaryotic target protein and a specialized region designed to aid in the purification and visualization of the target protein. A system that has proven to be very successful relies on the insertion of a six histidine (His6) sequence in the N-terminus of the encoded protein, allowing for efficient coupling to Ni²⁺-chelating resins and purification by single step affinity chromatography. This polyhistidine sequence can then be removed by specific cleavage at sites recognized by enzymes such as thrombin or enterokinase, permitting the separation of the target protein from the polyhistidine tag. Visualization of such fusion proteins can be achieved by utilizing antibodies generated against specific peptide sequences downstream from the multiple cloning site.

SOURCE

His-probe (H-15) is available as either rabbit (sc-803) or goat (sc-803-G) polyclonal affinity purified antibody raised against a peptide mapping to the polyhistidine domains of pET and Xpress polyhistidine expression vectors.

PRODUCT

Each vial contains 100 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-803 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

Available as agarose conjugate for immunoprecipitation, sc-803 AC, 500 µg/0.25 ml agarose in 1 ml.

Available as fluorescein conjugate for use in immunofluorescence, sc-803 FITC, 200 µg/ml.

APPLICATIONS

His-probe (H-15) is recommended for detection of fusion proteins encoded by prokaryotic pET, pRSET A, B, C and pTrc His A, B, C; baculovirus pBlue Bac His A, B, C; and mammalian pEBV His A, B, C expression vectors by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500), flow cytometry (1 µg per 1 x 10⁶ cells) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

STORAGE

Store at 4° C, ****DO NOT FREEZE****. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

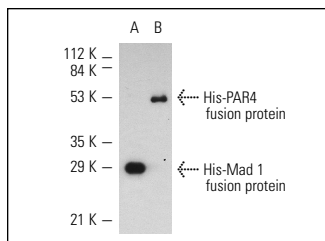
PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

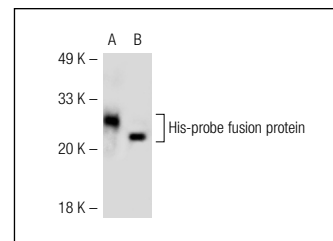
RESEARCH USE

For research use only, not for use in diagnostic procedures.

DATA



His-probe (H-15): sc-803. Western blot analysis of Mad 1 (1-221): sc-4086 WB (A) and PAR4 (1-334): sc-4216 WB (B) polyhistidine tagged fusion proteins using primary antibody at 0.1 µg/ml.



His-probe (H-15)-G: sc-803-G. Western blot analysis of mouse recombinant His-probe tagged GRIP1 (A) and His-probe tagged IKKα (B) fusion protein.

SELECT PRODUCT CITATIONS

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- Shah, Z.H., et al. 2012. A deacetylase-deficient SIRT1 variant opposes full-length SIRT1 in regulating tumor suppressor p53 and governs expression of cancer-related genes. *Mol. Cell. Biol.* 32: 704-716.
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