SANTA CRUZ BIOTECHNOLOGY, INC.

His-Tag (H-3): sc-8036

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

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 Ni^{2+}-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.

REFERENCES

1. Maniattis, T., et al. 1982. Molecular Cloning. Cold Spring Laboratory, Cold Spring Harbor, NY.
2. Smith, D.B., et al. 1988. Single-step purification of polypeptides expressed in *Escherichia coli* as fusions with glutathione S-transferase. Gene 67: 31-40.
3. Hochuli, E. 1988. Large-scale chromatography of recombinant proteins. J. Chromatogr. 444: 293-302.

SOURCE

His-Tag (H-3) is a mouse monoclonal antibody raised against a His tagged recombinant protein.

PRODUCT

Each vial contains 200 µg IgG_1 kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

His-Tag (H-3) is available conjugated to agarose (sc-8036 AC), 500 µg/0.25 ml agarose in 1 ml, for IP; to HRP (sc-8036 HRP), 200 µg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-8036 PE), fluorescein (sc-8036 FITC), Alexa Fluor® 488 (sc-8036 AF488), Alexa Fluor® 546 (sc-8036 AF546), Alexa Fluor® 594 (sc-8036 AF594) or Alexa Fluor® 647 (sc-8036 AF647), 200 µg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor® 680 (sc-8036 AF680) or Alexa Fluor® 790 (sc-8036 AF790), 200 µg/ml, for Near-Infrared (NIR) WB, IF and FCM.

In addition, His-Tag (H-3) is available conjugated to biotin (sc-8036 B), 200 µg/ml, for WB, IHC(P) and ELISA.

APPLICATIONS

His-Tag (H-3) is recommended for detection of fusion proteins encoded by polyhistidine expression vectors by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) and immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)].

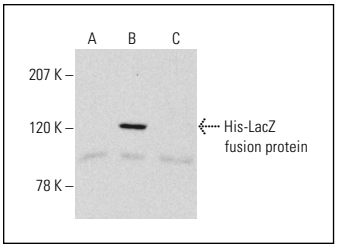
STORAGE

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

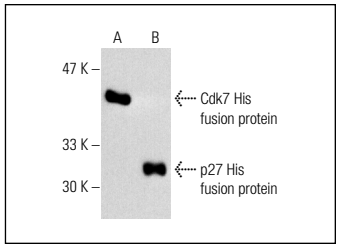
RESEARCH USE

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

DATA



pCruz His: sc-5044. Western blot analysis of His-LacZ fusion protein expression in COS cells transfected with pCruz Myc-Lac Z (**A**), pCruz His-LacZ (**B**) and non-transfected COS cells (**C**). Blot probed with HA-Tag (H-3): sc-8036.



HA-Tag (H-3): sc-8036. Western blot analysis of His-tagged Cdk7 (**A**) and p27 (**B**) fusion proteins.

SELECT PRODUCT CITATIONS

1. Chernukhin, I.V., et al. 2000. Physical and functional interaction between two pluripotent proteins, the Y-box DNA/RNA-binding factor, YB-1, and the multivalent zinc finger factor, CTCF. J. Biol. Chem. 275: 29915-29921.
2. Li, P.S., et al. 2014. The clathrin adaptor Numb regulates intestinal cholesterol absorption through dynamic interaction with NPC1L1. Nat. Med. 20: 80-86.
3. Shibata, N., et al. 2015. Degradation of stop codon read-through mutant proteins via the ubiquitin-proteasome system causes hereditary disorders. J. Biol. Chem. 290: 28428-28437.
4. Fagnocchi, L., et al. 2016. A Myc-driven self-reinforcing regulatory network maintains mouse embryonic stem cell identity. Nat. Commun. 7: 11903.
5. Roelens, B., et al. 2017. Maintenance of heterochromatin by the large subunit of the CAF-1 replication-coupled histone chaperone requires its interaction with HP1a through a conserved motif. Genetics 205: 125-137.
6. Dubois, N., et al. 2018. The C-terminal p6 domain of the HIV-1 Pr55Gag precursor is required for specific binding to the genomic RNA. RNA Biol. 15: 923-936.
7. Seifert, O., et al. 2019. Diabody-Ig: a novel platform for the generation of multivalent and multispecific antibody molecules. MAbs 11: 919-929.
8. Nardi, I.K., et al. 2020. USP22 interacts with PALB2 and promotes chemotherapy resistance via homologous recombination of DNA double-strand breaks. Mol. Cancer Res. 18: 424-435.
9. Cal-Kayitmazbatir, S., et al. 2021. CRY1-CBS binding regulates circadian clock function and metabolism. FEBS J. 288: 614-639.

PROTOCOLS

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