HIF1-alpha Antibody

Rabbit Polyclonal

Catalog No.

Antigen Affinity Purified Protein ID 016665

Lot No. A300-286A-7

APPLICATIONS WB, IP, IHC, ICC-IF

A300-286A

Human, Mouse SPECIES REACTIVITY

PRESUMED REACTIVITY Based on 100% sequence identity, this antibody is predicted to react with Rat, Chicken and

3091

Bovine

100 µl **AMOUNT**

CONCENTRATION $200 \mu g/ml$

2 - 8° C / 1 year from date of receipt STORAGE/SHELF LIFE

PHYSICAL STATE Liquid

BUFFER Tris-buffered Saline containing 0.1% BSA and 0.09% Sodium Azide

GeneID

ISOTYPE IgG **ORIGIN USA**

PRODUCTION

Antibody was affinity purified using an epitope specific to HIF1-alpha immobilized on solid

PROCEDURES support.

> The epitope recognized by A300-286A maps to a region between residues 775 and the Cterminus (residue 826) of human hypoxia-inducible factor 1, alpha subunit using the numbering

given in Swiss-Prot entry Q16665 (GeneID 3091).

Immunoglobulin concentration was determined by extinction coefficient: absorbance at 280 nm

of 1.4 equals 1.0 mg of IgG.

APPLICATIONS Centrifuge tube to remove product from lid. Optimal working dilutions should be determined

experimentally by the investigator. Prepare working dilution immediately before use.

Western Blot 1:2,000 - 1:10,000

Immunoprecipitation 2 – 10 μg/mg lysate

Immunohistochemistry 1:100 to 1:500. Epitope retrieval with citrate buffer pH6.0 for 20

minutes using a pressure cooker is recommended for FFPE tissue

sections.

Immunofluorescence

1:100 - 1:500. Epitope retrieval with citrate buffer pH 6.0 for 20 (ICC)

minutes using a pressure cooker is recommended for FFPE tissue

APPLICATION NOTES Western blot of immunoprecipitates performed using Normal Pig Serum (Cat. No. \$100-020),

Goat anti-Rabbit Light Chain HRP Conjugate (Cat. No. A120-113P) and 4-8% SDS-PAGE

(link to IP-western blot protocol in Additional Info section below).

Western blot of lysates performed using standard western blot reagents and 4-8% SDS-PAGE.

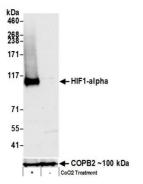
IHC HUMAN CONTROLS Renal Cell Carcinoma, HeLa Cells, HeLa Cells Hypoxic

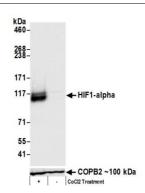
ADDITIONAL INFO https://www.bethyl.com/product/A300-286A

Use the link above to view SDS, a current list of citations, and other product specific information.

IP-western blot protocol https://www.bethyl.com/content/protocol_IP_WB

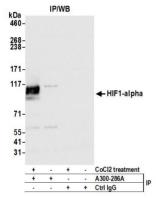
This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc. Eric McIntush, PhD | Chief Scientific Officer Date: January 21, 2020



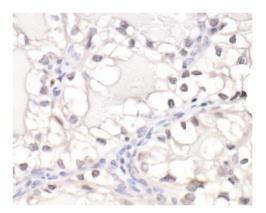


Detection of human HIF1 alpha by western blot. Samples: Whole cell lysate ($50 \mu g$) from HepG2 cells that were either treated with $200 \mu M$ cobalt chloride (+) or mock treated (-). Antibodies: Affinity purified rabbit anti-HIF1 alpha antibody A300–286A (lot number A300–286A–7) used for WB at $0.04 \mu g/ml$. Detection: Chemiluminescence with exposure times of 10 seconds. Lower Panel: Rabbit anti-COPB2 antibody (A304–523A).

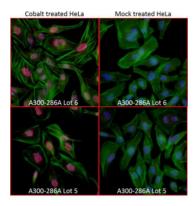
Detection of mouse HIF1 alpha by western blot. Samples: Whole cell lysate ($50 \mu g$) from NIH3T33ells that were either treated with $200 \mu M$ cobalt chloride (+) or mock treated (-). Antibodies: Affinity purified rabbit anti-HIF1 alpha antibody A300–286A (lot number A300–286A–7) used for WB at $0.04 \mu g/ml$. Detection: Chemiluminescence with exposure times of 3 seconds. Lower Panel: Rabbit anti-COPB2 antibody (A304–523A).



Detection of human HIF1 alpha by western blot of immunoprecipitates. Samples: Whole cell lysate (1 mg for IP, 20% of IP loaded) from HepG2 cells that were treated with 200 μM cobalt chloride or untreated. Antibodies: Affinity purified rabbit anti-HIF1 alpha antibody A300-286A (lot number A300-286A-7) used for WB at 0.04 μg/ml and for IP at 6 μg/mg lysate. Detection: Chemiluminescence with exposure times of 3 seconds.



Detection of human HIF1-alpha by immunohistochemistry. *Sample:* FFPE section of renal cell carcinoma. *Antibody:* Affinity purified rabbit anti-HIF1-alpha antibody (Cat. No. A300-286A Lot6) used at a dilution of 1:100 (2µg/ml). *Detection:* DAB



Detection of human HIF1-alpha by immunocytochemistry.

Sample: Formaldehyde-fixed asynchronous HeLa cells treated with 200 μM cobalt chloride (left) or mock treatment (right). Antibody: Affinity purified rabbit anti-HIF1-alpha antibody Cat. No. A300-286A Lot6 (upper images) and A300-286A Lot5 (lower images) used at a dilution of 1:100 (2μg/ml). Detection: Red-fluorescent goat anti-rabbit IgG-heavy and light chain cross-adsorbed Antibody DyLight® 594 Conjugated (A120-201D4). Counterstain: DAPI (blue) and fluorescent phalloidin (green).