

HIF1 –alpha Antibody

Rabbit Polyclonal

Antigen Affinity Purified Protein ID Q16665

Catalog No. A300–286A GeneID 3091

Lot No. A300–286A–7

APPLICATIONS	WB, IP, IHC, ICC–IF
SPECIES REACTIVITY	Human, Mouse
PRESUMED REACTIVITY	Based on 100% sequence identity, this antibody is predicted to react with Rat, Chicken and Bovine
AMOUNT	100 µl
CONCENTRATION	200 µg/ml
STORAGE/SHELF LIFE	2 – 8°C / 1 year from date of receipt
PHYSICAL STATE	Liquid
BUFFER	Tris–buffered Saline containing 0.1% BSA and 0.09% Sodium Azide
ISOTYPE	IgG
ORIGIN	USA
PRODUCTION PROCEDURES	Antibody was affinity purified using an epitope specific to HIF1 –alpha immobilized on solid support.

The epitope recognized by A300–286A maps to a region between residues 775 and the C–terminus (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.

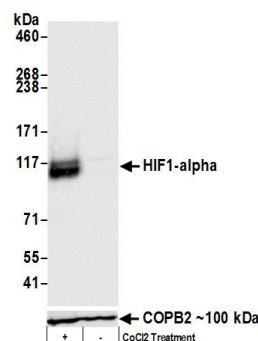
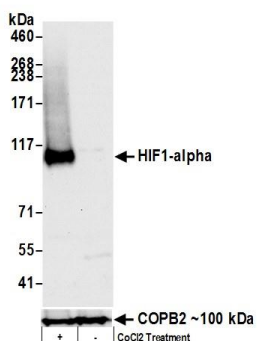
APPLICATIONS Centrifuge tube to remove product from lid. Optimal working dilutions should be determined experimentally by the investigator. Prepare working dilution immediately before use.

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 (ICC)	1:100 – 1:500. Epitope retrieval with citrate buffer pH 6.0 for 20 minutes using a pressure cooker is recommended for FFPE tissue sections.

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

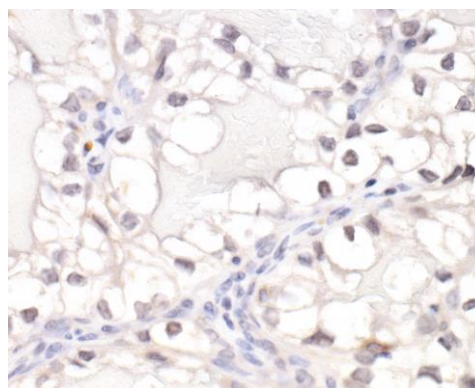
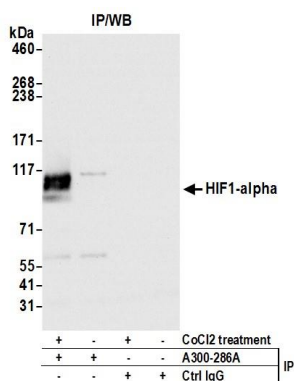
ADDITIONAL INFO <https://www.bethyl.com/product/A300–286A>
Use the link above to view SDS, a current list of citations, and other product specific information.
IP–western blot protocol: https://www.bethyl.com/content/protocol_IP_WB

This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc.
Michael Spencer, PhD Date: April 14, 2022



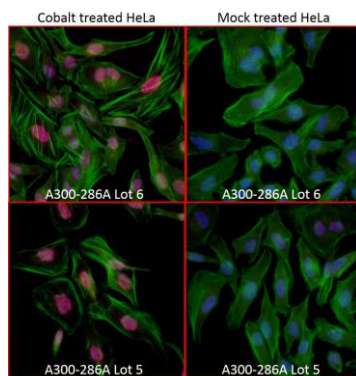
Detection of human HIF1 alpha by western blot. *Samples:* Whole cell lysate (50 µg) from HepG2 cells that were either treated with 200 µM cobalt chloride (+) or mock treated (-). *Antibodies:* Affinity purified rabbit anti-HIF1 alpha antibody A300-286A (lot number A300-286A-7) used for WB at 0.04 µg/ml. *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 µg) from NIH3T3 cells that were either treated with 200 µM 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 µg/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 μ M 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 (2 μ g/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).