

# HIF1- $\alpha$ Antibody

Rabbit Polyclonal

Antigen Affinity Purified Protein ID Q16665

Catalog No. A300-286A GeneID 3091

Lot No. A300-286A-5



<b>APPLICATIONS</b>	WB, IP, ICC, PLA
<b>SPECIES REACTIVITY</b>	Human, Mouse
<b>PRESUMED REACTIVITY</b>	Based on 100% sequence identity, this antibody is predicted to react with Rat, Chicken and Bovine
<b>ISOTYPE</b>	IgG
<b>AMOUNT</b>	100 $\mu$ l at 0.2 mg/ml
<b>STORAGE/SHELF LIFE</b>	2 - 8 $^{\circ}$ C / 1 year from date of receipt
<b>PHYSICAL STATE</b>	Liquid
<b>BUFFER</b>	Tris-buffered Saline containing 0.1% BSA and 0.09% Sodium Azide
<b>ORIGIN</b>	USA
<b>PRODUCTION PROCEDURES</b>	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 to 5  $\mu$ g/mg lysate

Immunocytochemistry 1:50 to 1:200. Formaldehyde fixation is recommended. Permeabilization with Triton-X 100 is recommended for formaldehyde-fixed cells.

Proximity Ligation Assay 1:200 - 1:4,000

**APPLICATION NOTES** Western blot of immunoprecipitates performed using ReliaBLOT<sup>®</sup> Reagents (Cat. No. WB120) and 4-8% SDS-PAGE.

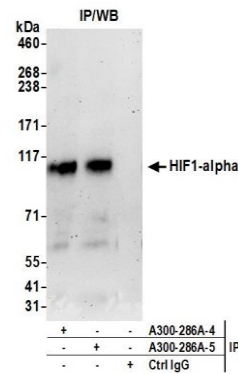
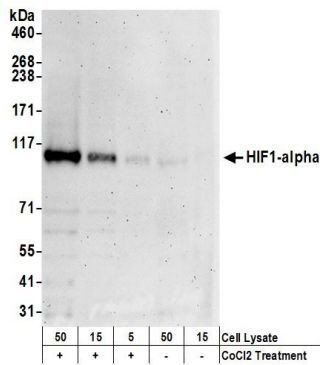
Western blot of lysates performed using standard western blot reagents and 4-8% SDS-PAGE.

**IHC HUMAN CONTROLS** HeLa Cells, Hypoxic HeLa Cells

**ADDITIONAL INFO** <http://www.bethyl.com/product/A300-286A>

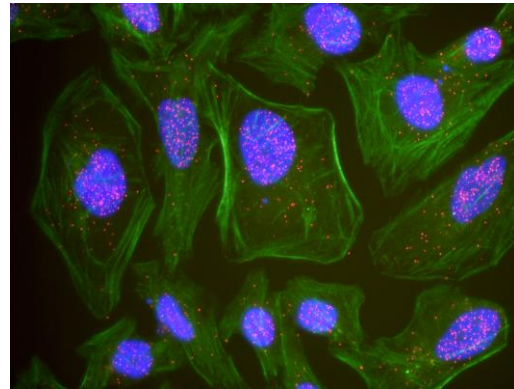
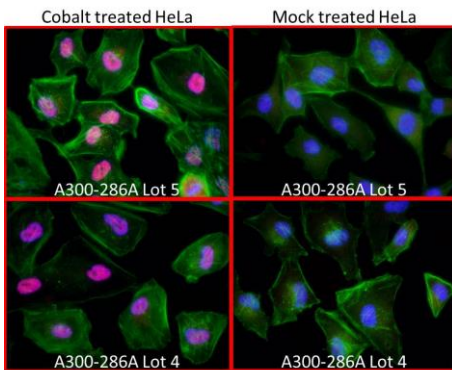
Use the link above to view SDS, a current list of citations, and other product specific information.

# HIF1-alpha Antibo A300-286A



**Detection of Human HIF1 alpha by Western Blot.** *Samples:* Whole cell lysate (5, 15 and 50 µg) from HeLa cells that were treated with cobalt chloride (+; 200 mcM) or mock treated (-). *Antibodies:* Affinity purified rabbit anti-HIF1 alpha antibody A300-286A (lot number A300-286A-5) used for WB at 0.1 µg/ml. *Detection:* Chemiluminescence with exposure times of 30 seconds.

**Detection of Human HIF1 alpha by Western Blot of Immunoprecipitates.** *Samples:* Whole cell lysate (1 mg for IP, 20% of IP loaded) from HeLa cells that were treated with cobalt chloride (200 mcM). *Antibodies:* Affinity purified rabbit anti-HIF1 alpha antibody A300-286A (lot number A300-286A-5) used for WB at 1 µg/ml and for IP at 6 µg/mg lysate. HIF1 alpha was also immunoprecipitated by a previous lot (lot A300-286A-4) of this antibody. *Detection:* Chemiluminescence with exposure times of 10 seconds.



**Detection of Human HIF1 alpha by Immunocytochemistry.** *Sample:* Formaldehyde-fixed asynchronous HeLa cells treated with cobalt chloride-200 mcM (left) or mock treatment (right). *Antibody:* Affinity purified rabbit anti-HIF1 alpha Cat. No. A300-286A Lot5 (upper images) and A300-286A Lot4 (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. *Counterstain:* DAPI (blue) and fluorescent phalloidin (green).

**This antibody is qualified for the Proximity Ligation Assay (PLA).** PLA uses paired antibodies, so please refer to the PLA page of our website for specific imaging obtained by pairing this antibody with other qualified antibodies. The above image shows representative results for PLA using three color fluorescence, including DAPI stained nuclei (blue), phalloidin stained cytoplasmic F-actin (green), and PLA positive signal (red).