

## DESCRIPTION

<b>Species Reactivity</b>	Human/Mouse
<b>Specificity</b>	Detects human FIH-1/HIF-1AN in ELISAs.
<b>Source</b>	Monoclonal Mouse IgG <sub>2B</sub> Clone # 745122
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant human FIH-1/HIF-1AN Met1-Asn349 Accession # Q9NWT6
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

## APPLICATIONS

**Please Note:** Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
<b>Western Blot</b>	0.25 µg/mL	See Below
<b>Simple Western</b>	12.5 µg/mL	See Below

## DATA

**Western Blot**

**Detection of Human and Mouse FIH-1/HIF-1AN by Western Blot.** Western blot shows lysates of A172 human glioblastoma cell line and C2C12 mouse myoblast cell line. PVDF membrane was probed with 0.25 µg/mL of Mouse Anti-Human/Mouse FIH-1/HIF-1AN Monoclonal Antibody (Catalog # MAB7874) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for FIH-1/HIF-1AN at approximately 42 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

**Simple Western**

**Detection of Mouse FIH-1/HIF-1AN by Simple Western™.** Simple Western lane view shows lysates of C2C12 mouse myoblast cell line, loaded at 0.5 mg/mL. A specific band was detected for FIH-1/HIF-1AN at approximately 40 kDa (as indicated) using 12.5 µg/mL of Mouse Anti-Human/Mouse FIH-1/HIF-1AN Monoclonal Antibody (Catalog # MAB7874). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

Non-specific interaction with the 230 kDa Simple Western standard may be seen with this antibody.

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Sterile PBS to a final concentration of 0.5 mg/mL.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

## BACKGROUND

Factor-inhibiting hypoxia-inducible factor 1 (FIH-1), also known as HIF-1AN, is a 40-45 kDa oxygenase that hydroxylates asparagine, aspartate, and histidine residues in target proteins. Its homodimerization is required for substrate recognition. Under normoxic conditions, FIH-mediated hydroxylation inhibits the function of HIF-1 alpha and HIF-2 alpha by preventing their association with the transcriptional coactivator p300. FIH activity is inhibited under hypoxic conditions which allows HIF to mediate hypoxia-induced gene transcription. FIH also hydroxylates amino acids within the ankyrin repeat domains (ARD) of various other proteins including IκB-alpha, NFκB1, AnkyrinR, AnkyrinB, Tankyrase 2, MYPT1 smooth muscle myosin phosphatase, and Notch. Human FIH-1 shares 97% aa sequence identity with mouse and rat FIH-1.