

NB100-428 Protocol

Western Blot protocol specific for Factor Inhibiting HIF-1 Antibody (NB100-428)

[[URL: https://www.novusbio.com/products/fih-1-hif-1an-antibody_nb100-428]][[Caption:FIH-1/HIF-1AN Antibody]]

1. (30-40ug) of protein were separated via gel electrophoresis (10-20%) under denaturing conditions and transferred to nitrocellulose membrane.
2. Membrane was blocked overnight (4C) in TBS/Tween-20 (TBST) + 5% milk (blocking buffer).
3. Membrane was incubated with primary anti-FIH [cat# NB 100-428] (1:500) in blocking buffer for 2 hours at room temp.
4. Membrane was washed 3 times with TBST. 10 minutes each wash.
5. Membrane was incubated with HRP conjugated Donkey anti-rabbit secondary in blocking buffer for 2 hours at room temp.
6. Membrane was washed 3 times with TBST. 10 minutes each wash.
7. Membranes were developed for visualization using Super Signal(R) West Pico Chemiluminescent Substrate and captured on Hyperfilm ECL (Amersham Biosciences).