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NB100-616 Protocol

Protocol specific for KAT3B / p300 Antibody (NB100-616)

KAT3B/p300 Antibody (RW105): https://www.novusbio.com/products/kat3b-p300-antibody-rw105_nb100-616
Western Blot Protocol

1. Perform SDS-PAGE (4-7% gel) on samples to be analyzed, loading 50ug of total protein per lane.

2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the transfer apparatus.

3. Stain the blot using ponceau S for 1-2 minutes to access the transfer of proteins onto the membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.

4. Rinse the blot in TBS for approximately 5 minutes.

5. Block the membrane using 5% non-fat dry milk in TBS +0.1% Tween-20 for 1 hour.

6. Dilute the mouse anti-KAT3B primary antibody (NB 100-616) in blocking buffer and incubate overnight at 4 degrees Celsius.

7. Wash the membrane 3x 5 min in TBST and apply the diluted mouse-IgG HRP-conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) and incubate 1 hour at room temperature.

8. Wash the blot 3x 5 min in TBST.

9. Apply the detection reagent of choice in accordance with the manufacturer's instructions.