

## NB300-147 Protocol

### Western Blot protocol for Sodium Potassium ATPase Beta 1 Antibody (NB300-147)

[[URL:<https://www.novusbio.com/products/sodium-potassium-atpase-beta-1-antibod...>]][[Caption:Sodium Potassium ATPase Beta 1 Antibody (464.8 (also known as 8A))]]

1. Run ~20 ug of rat kidney homogenate on a 7.5% SDS-PAGE gel.
2. Transfer protein to the membrane using a Tris-Glycine/Methanol buffer.
3. Block membrane with TBST/5% NFDM for 30 min. at room temperature (~23-27 degrees C).
4. Wash membrane twice, for 5 minutes each, with TBST.
5. Incubate membrane with 1:2,500 dilution of NB300-147 (anti-Na,K-ATPase), diluted in TBST, for 1 hour at room temperature.
6. Wash membrane once for 15 minutes, then four times for 5 minutes each, with TBST.
7. Incubate membrane with 1:15,000 dilution of goat anti-mouse IgG-HRP [(Pierce) stocked 1:2 in glycerol], diluted in TBST, for 1 hour at room temperature.
8. Wash membrane once for 15 minutes, then four times for 5 minutes each, with TBST.
9. Detect cross-reacting proteins using SuperSignal West Pico Chemiluminescent substrate (Pierce), diluted according to manufacturer's guidelines.

\*NOTE: Do not boil the protein samples, as boiling causes aggregation of the Na,K-ATPase. The aggregate band will appear at ~150 kDa on Western Blots.