

NB100-1194 Protocol

Western Blot Protocol for Park7(DJ-1) Antibody (NB100-1194)

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Western blot Protocol:

- 1) Transfer onto PVDF membrane
- 2) Block in 3% (w/v) skimmed milk in TBS-T (0.05% Tween 20) for 1 hr at room temperature with agitation (can be blocked overnight at 4C without agitation).
- 3) Incubate NB100-1194 (2ug/ml) for 1 hr at room temperature with agitation.
- 4) Wash with TBS-T three times
- 5) Incubate with secondary antibody (1:20,000 or higher). We use Jackson or Rockland anti-goat secondary (affinity purified and with minimal cross-reactivity with Human and rodent serum proteins) for 1 hr at room temperature with agitation.
- 6) Wash with TBST three times.
- 7) Apply the detection reagent of choice in accordance with the manufacturers instructions (We use ECL-plus (Amersham) rather than ECL, which is more sensitive for detection.)

Notes:

*All antibodies are incubated in blocking buffer.

*QC testing was done under reducing conditions (with beta mercaptoethanol at 100C).