***In vitro* kinase activity**

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**Abstract:** This protocol details methods for the *in vitro* kinase activity testing of purified LRRK2.

Materials: SuperSep Phos-tag Gel (FUJIFILM, 195-1799); Anti-LRRK2 (phospho T1357) antibody (Abcam, ab270606).

**Solutions to prepare:**

10x kinase buffer: 200mM Tris–HCl (pH7.5), 75mM MgCl2 and 1mM EGTA.

**Protocol:**

1, Set up the reaction mixtures with 1x kinase buffer (diluted from 10x kinase buffer), 200 nM LRRK2 or 8 μg Rab8 protein, for the reaction group add 1 mM ATP, and for the control add H2O instead, the total volume we used is 40 μL.

2, Incubate samples for 2 hours at 30°C.

3, Quench reactions through addition of SDS sample loading buffer and heat at 95°C for 10 min.

4, Resolve samples by SDS/PAGE or Phos-tag SDS/PAGE

5, Detect proteins by Coomassie Blue staining or Western blot using antibodies of Rab8 and LRRK2 phospho-specific (pT1357), respectively.