1. Wash plate twice with cold PBS.
2. Add 1 mL lysis buffer into P100 plate, sit on ice for 20 min with gentle shaking, centrifuge 14,000rpm/10min/4C, take supernatant.
3. Preclear the lysate with 50 uL protein G slurry, tumble, 45 min/4C, followed by a centrifuge 14,000rpm/15min/4C.
4. Add 1 ug antibody (3 uL anti-ARA9 antibody I used) to 50 uL protein G slurry, add 500 uL cold PBS, tumble, 1 hr/4C. Wash Ab/beads twice by adding PBS. Spin down beads by centrifuge at 1000g/1 min.
5. Add precleared lysate into pre-bond Ab/protein G complex, tumble O/N, 4C Spin down beads by centrifuge at 1000g/1 min.
6. Wash beads five times with lysis buffer.
7. Add SDS sample buffer to beads.