



## Supplementary Figure S2. Arrdc4 interacts with several Nedd4 family members and is ubiquitinated by Nedd4-2.

- **a.** Pull-down using anti-GFP antibody shows an interaction between Arrdc4 and Nedd4, Nedd4-2, WWP1, WWP2 and Itch. There is no interaction with Smurf1 or Smurf2. The dotted line indicates non-adjacent lanes on the same blot.
- **b.** *In vivo* ubiquitination assay shows that Arrdc4 is ubiquitinated by Nedd4-2 but not its catalytically inactive mutant (CM) and that this ubiquitination is dependent on its PY motif. Pull-down was carried out using anti-GFP antibody and ubiquitination was detected using an anti-HA antibody.