

Figure S9

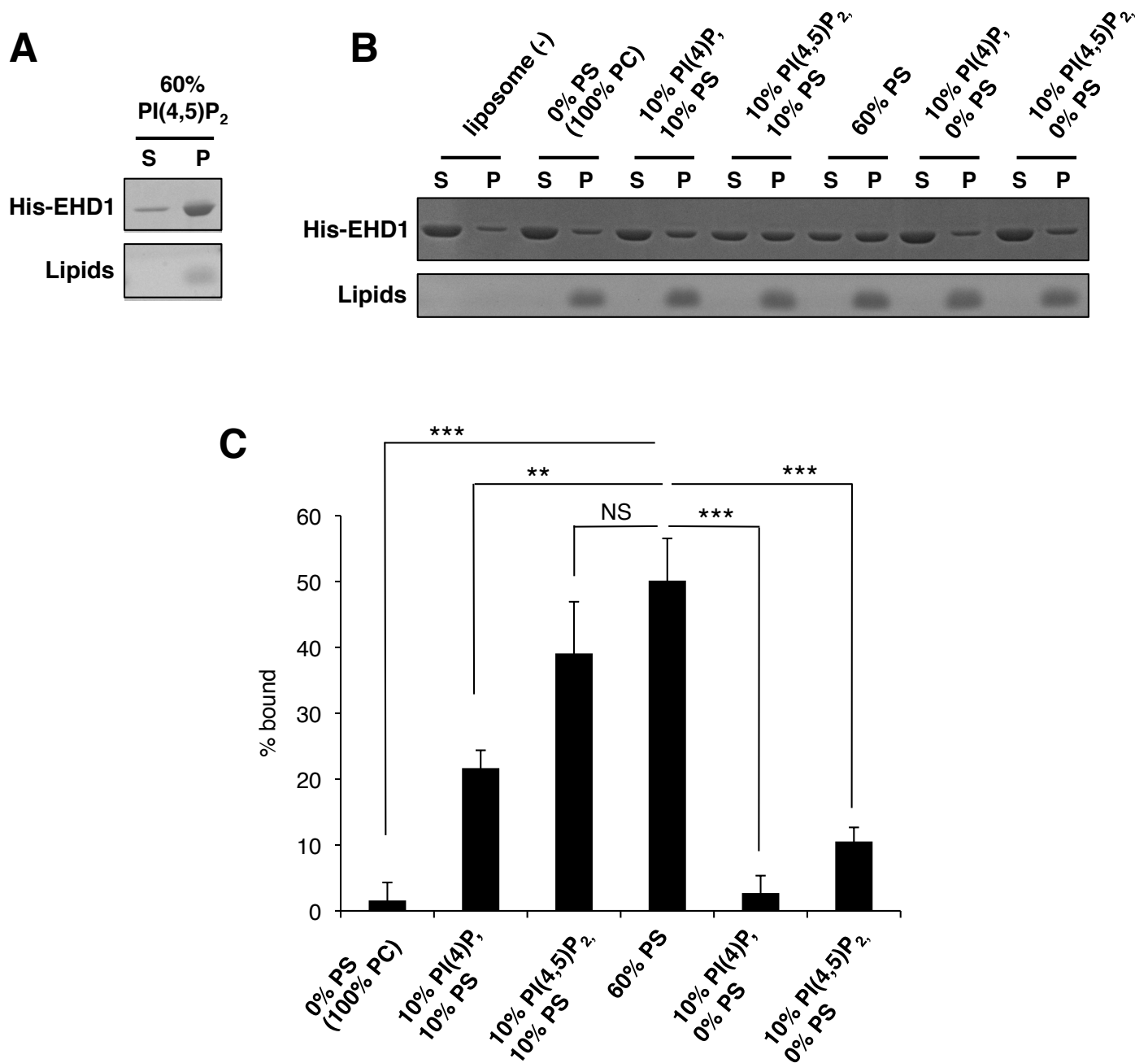


Figure S9. PI(4)P or PI(4,5)₂ affects EHD1 binding to liposomes containing 10% PS.

(A and B) His-tagged EHD1 was mixed with liposomes containing the indicated % (mol/mol) of lipids. After 15 min, the mixture was spun at 100,000 g for 30 min, and the resultant supernatant (S) and pellet (P) were subjected to SDS-PAGE. The proteins and lipids were then stained with Coomassie blue. In (A), “60% PI(4,5)P₂” liposomes consist of 40% PC and 60% PI(4,5)P₂. In (B), “10% PI(4)P or PI(4,5)P₂, 10% PS” liposomes consist of 35% PC, 35% PE, 10% PS, 10% cholesterol, and 10% PI(4)P or PI(4,5)P₂, respectively. “10% PI(4)P or PI(4,5)P₂, 0% PS” liposomes consist of 45% PC, 35% PE, 0% PS, 10% cholesterol, and 10% PI(4)P or PI(4,5)P₂, respectively.

(C) The intensities of individual bands in (B) were quantified with ImageJ, and the percentage of bound proteins was calculated. Data represent the mean ± SD of *n* = 3 independent experiments, and were analyzed with Student’s *t*-test. ***P* < 0.01; ****P* < 0.001; NS, not significant.