

Figure S1

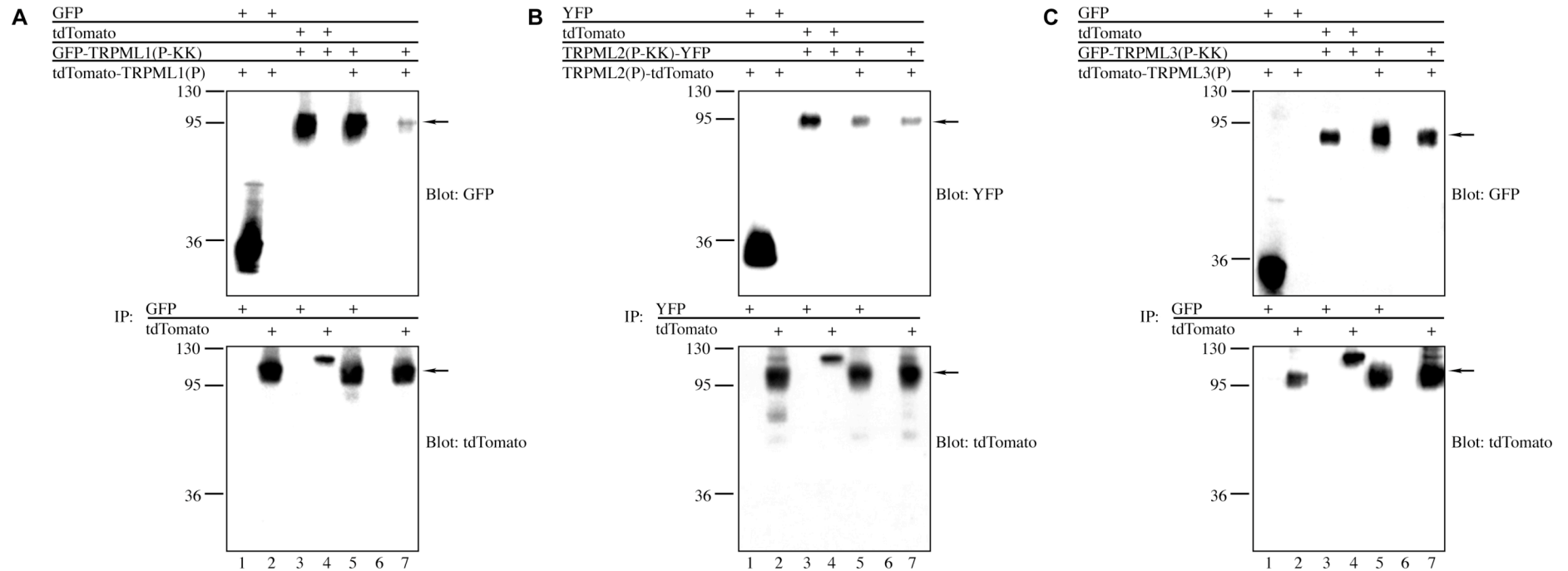


Fig. S1. Pore region mutations do not interfere with TRPML subfamily physical interactions involving proteins of the same TRPML origin. (A-C) HEK cells were co-transfected with the indicated expression constructs (above top panels) and GFP/YFP- or tdTomato-tagged constructs were then immunoprecipitated (as indicated between top and bottom panels). Immunoprecipitates were separated on gels followed by immunoblotting against the indicated targets (to the right of each panel). Size markers (in kDa) are to the left of each panel and bands corresponding with monomeric TRPML isoforms are demarcated with arrows to the right. Lane numbers are indicated below bottom panels. Top panels, lanes 1,3,5: positive controls. Top panels, lanes 2: negative controls—tdTomato-tagged TRPML(P) channels do not co-IP with GFP/YFP. Top panels, lanes 4: negative controls—GFP/YFP-tagged TRPML(P-KK) channels do not co-IP with tdTomato. Top panels, lanes 6: Blank. Bottom panels, lanes 2,4,7: positive controls. Bottom panels, lanes 1: negative controls—tdTomato-tagged TRPML(P) channels do not co-IP with GFP/YFP. Bottom panels, lanes 3: negative controls—GFP/YFP-tagged TRPML(P-KK) channels do not co-IP with tdTomato. Bottom panels, lanes 6: Blank. The tdTomato fluorescent protein is detected as a dimer (~125 kDa) in our running conditions. (A) Lane 7 in top panel and lane 5 in bottom panel: GFP-TRPML1(P-KK) co-IPs with tdTomato-TRPML1(P). (B) Lane 7 in top panel and lane 5 in bottom panel: TRPML2(P-KK)-YFP co-IPs with TRPML2(P)-tdTomato. (C) Lane 7 in top panel and lane 5 in bottom panel: GFP-TRPML3(P-KK) co-IPs with tdTomato-TRPML3(P).