

Figure 1-figure supplement 1

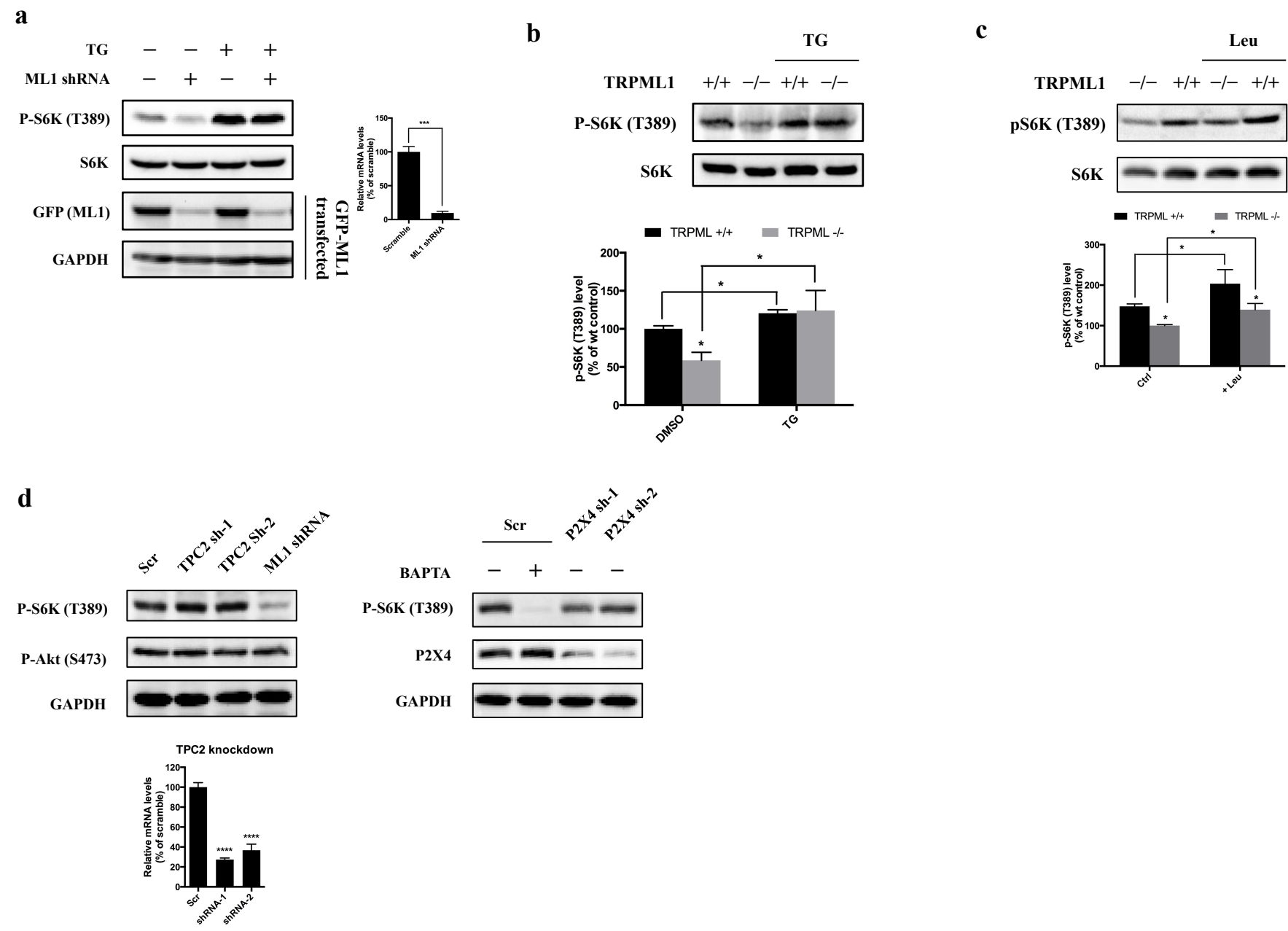


Figure 1-figure supplement 1. Effects of TRPML1 on mTORC1 activation. **(a)** Scrambled shRNA or TRPML1 shRNA-transduced HEK293T cells were treated with vehicle control or thapsigargin (5 μ M) for 2 h , respectively. Cells were lysed and subjected to immunoblotting. RT-qPCR (right panel) and EGFP-TRPML1 transfected cells were used to evaluate the knockdown efficiency. **(b)** WT and mucopolidosis IV patient's fibroblasts (TRPML^{-/-}) were treated with vehicle control or leucine for 10 min. Cells were lysed and subjected to immunoblotting. The plot shows the percentage of p-S6K level compared with vehicle control treated TRPML^{-/-} human fibroblasts normalized by total S6K control. (mean \pm s.d. for n = 2 independent experiments). **(c)** Healthy human fibroblast or mucopolidosis IV patient's fibroblasts (TRPML^{-/-}) were treated with vehicle control or thapsigargin (5 μ M) for 2 h , respectively. Cells were lysed and subjected to immunoblotting. The plot shows the percentage of p-S6K level compared with vehicle control treated wild type human fibroblasts normalized by total S6K control. (mean \pm s.d. for n = 3 independent experiments). **(d)** Lentivirus carrying scrambled shRNA, TRPML1 shRNA, TPC2 shRNA or P2X4 shRNA transduced HEK293T cells were lysed and subjected to immunoblotting. RT-qPCR (bottom panel) was used to evaluate the knockdown efficiency of TPC2.