

Supplemental Figure 1. FBXL19 reduces Rac1 levels. A. MLE12 cells were transfected with several V5 tagged F-box protein plasmids (*FBXO23, FBXL18, FBXL19, FBXW2,* and *FBXW12*). Cell lysates were analyzed for Rac1, V5 tag, and β -actin by immunoblotting. B. MLE12 cells were transfected with *HACE1-HA, XIAP-flag,* or *FBXL19-V5* plasmids. Cell lysates were analyzed for Rac1, HA tag, Flag tag, V5 tag, and β -actin by immunoblotting. C. Beas2B and MLE12 cell lysates were analyzed for FBXL19 and β -actin by immunoblotting. D. Beas2B cells were transfected with *FBXL19-V5* plasmid. Cell lysates were analyzed by Rac1, V5 tag, and β -actin immunoblotting. Shown are representative blots from three independent experiments.



Supplemental Figure 2. FBXL19 targets Rac1, Rac2 and Rac3. MLE12 cells were transfected with *Rac1-V5*, *Rac2-flag*, or *Rac3-V5* with or without *FBXL19-V5* plasmids. Cell lysates were analyzed for Rac1, flag tag, V5 tag, and β -actin by immunoblotting. Shown are representative blots from three independent experiments.



Supplemental Figure 3. FBXL19 regulates LPA-mediated Rac1 activity. MLE12 cells were transfected with *FBXL19-V5* (A) or *FBXL19* shRNA plasmid (B) followed by LPA treatment (5 μ M, 10 min). Rac1 activity was measured using a Rac1 activation assay kit. Shown are representative blots from three independent experiments.



Supplemental Figure 4. Rac1 regulates MLE12 cell migration and leading edge formation. A. MLE12 cells were transfected with *Rac1* shRNA plasmids (0 -4 μ g). Cell migration was measured by transwell migration assays. **p*<0.01, compared to veh in *shRac1* (0 μ g) transfected cell. ***p*<0.01, ratio between veh and LPA compared to ratio between veh and LPA in *shRac1* (0 μ g) transfected cells. B. The cell lysates from (A) were analyzed for Rac1 and β -actin by immunoblotting. Shown are representative blots from three independent experiments. C. MLE12 cells grown on glass bottom dishes were transfected with *Rac1* shRNA followed by LPA treatment (5 μ M, 1h). Cells were fixed and actin filaments were stained with phalloidin (green) and nuclei were stained with DAPI (blue). Shown are representative images from three independent experiments. D. MLE12 cells were transfected with *FBXL19-GFP* plasmid followed by LPA treatment (5 μ M, 1h). Cells were fixed and localization of FBXL19-GFP (green), Rac1 (red), and nuclei (blue) were examined by immunofluorescence staining. Shown are representative images from three independent system fixed and localization of representative images from three independent system fixed and localization of FBXL19-GFP (green), Rac1 (red), and nuclei (blue) were examined by immunofluorescence staining. Shown are representative images from three independent system fixed and localization of FBXL19-GFP (green).