

Supplemental material legends

Supplemental figure 1: (A) Measurement of the expression levels of Radil and V5-Rap1A(V12) in HUVECs treated with control siRNA (siC) or depleted of Radil (siRadil) and transduced with control lentivirus or Rap1A(V12) containing lentivirus, used in the experiments depicted in Fig. 7. Upper panel: RNA expression levels of Radil assessed by Q-PCR. Lower graph: V5-Rap1A(V12) protein expression levels as assessed by Western blot. (B) Measurement of the expression levels of Rasip1 and V5-Rap1A(V12) in HUVECs treated with control siRNA (siC) or depleted of Rasip1 (siRasip1) and transduced with control lentivirus or Rap1A(V12) containing lentivirus, used in the experiments depicted in Fig. 7. Upper panel: RNA expression levels of Rasip1 assessed by Q-PCR. Lower graph: V5-Rap1A(V12) protein expression levels as assessed by Western blot. (C) Measurement of the expression levels of Radil and Rasip1 in HUVECs treated with control siRNA (siC) or depleted of Radil (siRadil or Rasip1 (siRasip1)), used in the experiments depicted in Fig. 1B. Error bars indicate standard deviation between PCR triplicates. (D) Quantification of the signal correlation between F-actin and β -catenin (0=random, 1=perfect correlation) as a measure of radial stress fiber content throughout the cells. HUVECs transfected with control siRNA (siC), siRNA targeting Radil (siRadil), siRNA targeting Rasip1 (siRasip1) or both (siRadil + siRasip1) were grown to confluency and either not stimulated or stimulated with 007-AM 15 minutes prior to fixation. Cells were stained for β -catenin (red) or F-actin (phalloidin, green). A minimum of three pictures per experiment were analyzed. Graph shows the average correlation of three independent experiments. Error bars depict SD between experiments. *P<0.05, n.s., not significant.

Movie 1: Live imaging of YFP-ArhGAP29 in HEK293T cells transiently transfected with HA-Epac1, HA-Rap1A, V5-Radil, HA-Rasip1 and YFP-ArhGAP29. The movie comprises 11 minutes of filming, with frames being captured every 30 seconds. 007-AM was added when indicated.

Movie 2: Live imaging of mCherry-Radil in HEK293T cells transiently transfected with HA-Epac1, HA-Rap1A, mCherry-Radil, HA-Rasip1 and HA-ArhGAP29. The movie comprises 11 minutes of filming, with frames being captured every 30 seconds. 007-AM was added when indicated.

Movie 3: Live imaging of YFP-Rasip1 in HEK293T cells transiently transfected with HA-Epac1, HA-Rap1A, V5-Radil, YFP-Rasip1 and HA-ArhGAP29. The movie comprises 11 minutes of filming, with frames being captured every 30 seconds. 007-AM was added when indicated.