

**Supplementary Movie 1. Live imaging of GFP-ArgBP2 $\gamma$  in HeLa and U2OS cells.**

Time-lapse confocal imaging of GFP-ArgBP2 $\gamma$  in HeLa and U2OS cells. ArgBP2 $\gamma$  containing puncta are observed to originate at the cell periphery and undergo retrograde flow towards the cell centre. Frames were taken every 25 secs for 35 mins for HeLa and every 15 secs for 22.5 mins for U2OS.

**Supplementary Movie 2. Wound healing assay of GFP-ArgBP2 $\gamma$  expressing stable cells versus control cells.**

Time-lapse phase contrast imaging of either control cells or cells expressing GFP-ArgBP2 $\gamma$  migrating into a wound area for 25 hours. Frames were taken every 30 mins.

**Supplementary Movie 3. Live imaging of ArgBP2 $\gamma$  expressing U2OS cells**

Time-lapse differential interference contrast of stable U2OS lines expressing GFP-ArgBP2 $\gamma$ , GFP-ArgBP2 $\gamma$ -S259A or GFP-ArgBP2 $\gamma$ - $\Delta$ AnBD as indicated. Cells were allowed to freshly spread on fibronectin for 2 hours before observing random cell migration for 5 hours. Frames were taken every 10 mins for 5 hours. ArgBP2 and ArgBP2-S259A significantly reduce cell migration of these cells compared to controls or the  $\alpha$ -actinin binding mutant.