



Supplementary information, Figure S6. Myo1D promotes the coalescence of p62 bodies, related to Figure 4

(a) Myo1D knockout NRK cells were transfected with GFP, GFP-Myo1D, GFP-Myo1D K108R GFP-Myo1D Motor-IQ, GFP-Myo1D IQ, GFP-Myo1D IQ-TH1 or GFP-Myo1D TH1 then stained with antibodies against GFP and p62. Scale bar, 10 μm .

(b) Fluorescence intensity recovery of reconstituted p62 bodies after photo bleaching.

(c) Quantification of fluorescence intensity recovery of photobleached p62 bodies (n = 3). Scale bar, 5 μm .

(d) SEM image showing the structure of a reconstituted p62 body on the Arp2/3-derived actin network. Scale bar, 1 μm .

(e) Cells were stained with phalloidin and Myo1D or antibodies against Arpc2 and Myo1D. Scale bar, 10 μm .

(f) Myo1D were labeled with Alexa-488. Total internal reflection fluorescence spectroscopy (TIRF) images showing that p62 body formation. PWCA (300 nM), Actin (1 μM , alexa-647 phalloidin labeled), Arp2/3 complex (100 nM), and CapZ (600 nM) were pre-loaded into the chamber. Then Myo1D (100 nM Alexa-488 labeled), p62 (0.5 μM), and Ub8 (3 μM) were added into the reconstitution system. Scale bar, 5 μm .

(g) Condensate formation during in vitro assays in the presence of WT and K108R Myo1D. Scale bar, 5 μm .

(h) Condensate formation during in vitro assays in the presence or absence of ATP. Scale bar, 5 μm .

(e) A phase diagram of p62, Ub8 and Myo1D in the indicated concentration ranges.