

SUPPLEMENTARY DATA

Supplementary Figure 1 Endogenous reggie-1 form tetramers in N2a cells after different cross-linking treatments

N2a cells were subjected to chemical cross-linking for 1 min using DSS (A), DMS (B) or MBS (C) at the concentrations indicated above each lane. Reggie-1 specific mAb was used for detection and reveal ~47 kDa reggie-1 monomers as well as ~200 kDa bands (*arrowheads*) corresponding to reggie tetramers. The decrease of the bands indicative of reggie tetramers upon reduction of the cross-linker concentration is also apparent. No intermediate species were observed even at very low concentrations of cross-linker.

Supplementary Figure 2 Homo-tetramerization of EGFP and BiFC reggie-1 and -2 fusion proteins in N2a cells

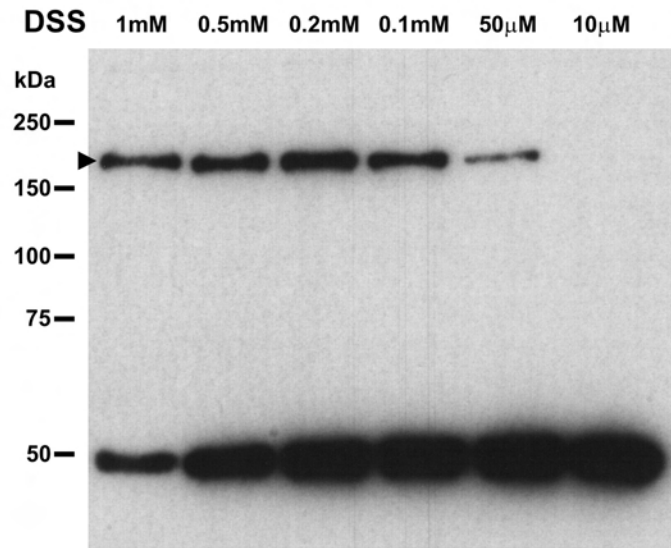
Transiently transfected N2a cells expressing the EGFP- or BiFC-fusion proteins indicated above each lane were cross-linked using 1 mM of DSS for 5 min after 20 h post transfection. GFP specific mAb used for detection reveal the ~75 and ~60 kDa monomers of reggie-EGFP and reggie-BiFC fusion proteins, respectively (*closed and open arrowheads, respectively*). Homo-tetramers of reggie-EGFP and reggie-BiFC migrate as ~300 and ~240 kDa bands, respectively (*light and dark grey arrowheads, respectively*).

Supplementary Figure 3 Homo- and hetero-tetramerization of HA-tagged and EGFP-fused reggie-1 and -2 in N2a cells

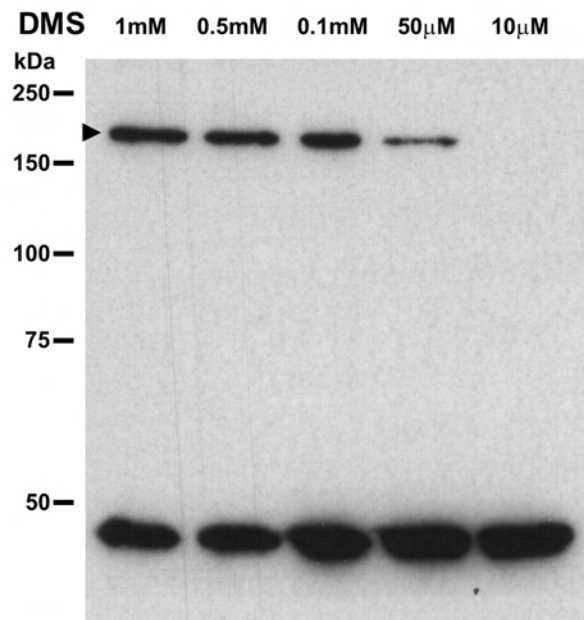
N2a cells were transiently co-transfected with the constructs indicated above each lane. Cells were cross-linked 20 h post transfection using 1 mM of DSS for 5 min. A, GFP specific mAb revealed monomers of reggie-EGFP of ~75 kDa (*closed arrowheads*) as well as homo-tetramers of ~300 kDa (*open arrowheads*). Hetero-tetramers containing EGFP-reggie species migrate as ~250 kDa bands (*grey arrowheads*). B, HA specific mAb revealed bands of ~50 and ~200 kDa corresponding to monomers and homo-tetramers of HA-tagged reggies, respectively (*closed and grey arrowheads, respectively*). Hetero-tetramers containing HA-reggies migrate as ~250 kDa bands (*open arrowheads*).

Supplementary Figure 1

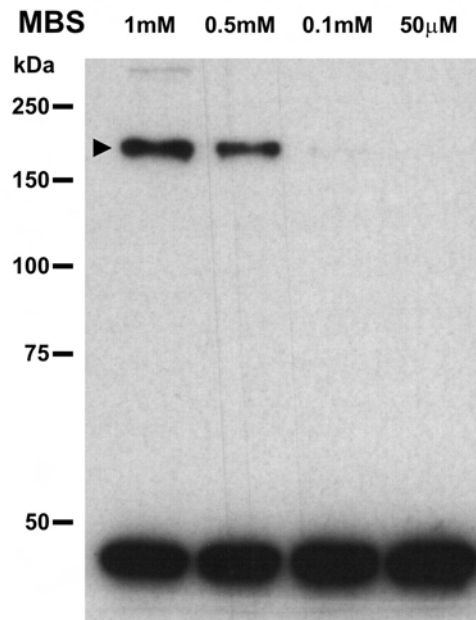
A. WB: Reggie-1



B. WB: Reggie-1

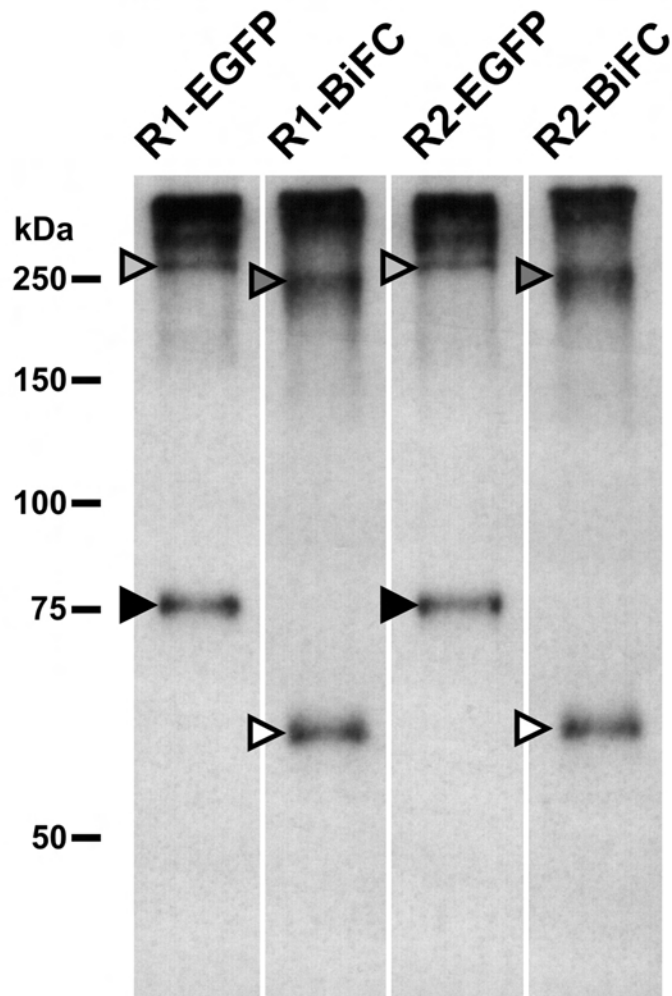


C. WB: Reggie-1



Supplementary Figure 2

WB: GFP



Supplementary Figure 3

