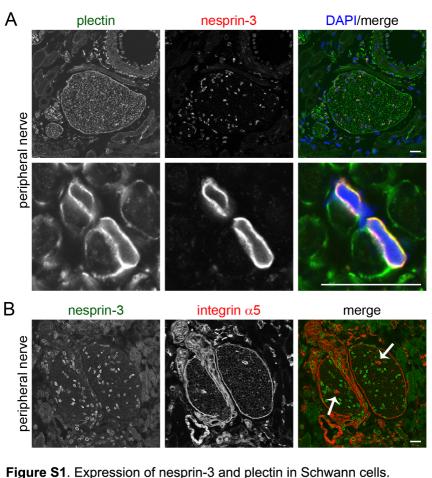
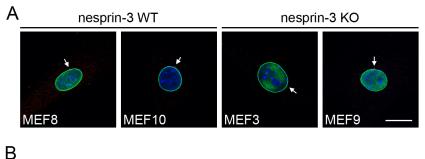
Supplemental Materials Molecular Biology of the Cell

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(A) Frozen sections of the peripheral nerves of 2-mo-old (upper panels) and 4-mo-old (lower panels) wild-type mice were stained for plectin and nesprin-3. Nuclei were counterstained with DAPI. Scale bars, 20 μ m. (B) Frozen section of the peripheral nerve of a 2-mo-old wild-type mouse was double-labeled for nesprin-3 and the integrin α 5 subunit. Arrows show sites of high integrin α 5 expression that indicate the positions of blood vessels. Scale bar, 20 μ m.



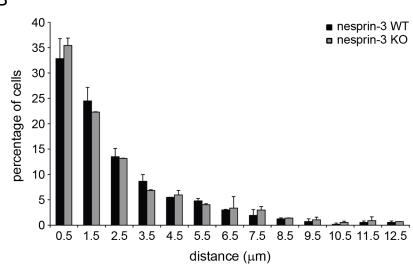


Figure S2. Loss of nesprin-3 does not affect the distance between the nucleus and the MTOC in MEFs.

(A) Primary MEFs were fixed in methanol and stained for lamin B (green) and γ -tubulin (red). Nuclei were counterstained with DAPI (blue) and cells were analyzed by confocal microscopy. Arrows indicate the position of the MTOC. Scale bar, 20 μm . (B) Histogram displaying the distribution of the nucleus-MTOC distance. Distances were determined for 200 cells per individual MEF cell line. Error bars indicate the standard deviation over two wild-type (WT) and two knockout (KO) primary cell lines.

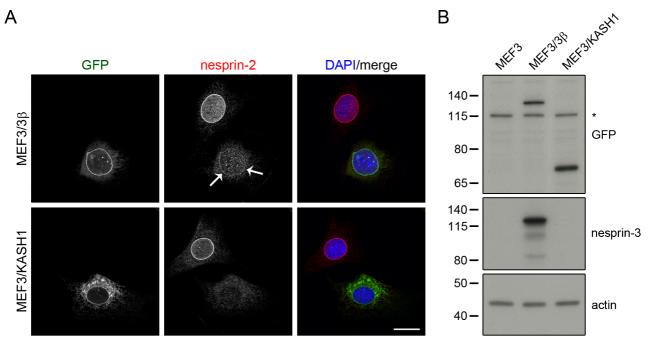


Figure S3. Dominant negative effect of GFP-tagged KASH proteins on endogenous nesprin-2. (A) Immortalized nesprin-3 $^{-/-}$ cells were transfected with GFP-nesprin-3β (MEF3/3β) or GFP-dominant-negative nesprin-1 (MEF3/KASH1) and stained for endogenous nesprin-2. Nuclei were counterstained with DAPI. Arrows indicate localization of nesprin-2 at the NE. Scale bar, 20 μm. (B) Stably transfected nesprin-3 $^{-/-}$ (MEF3) cells were lysed in RIPA lysis buffer and analyzed by Western blot for expression of the indicated proteins. Actin levels served as a loading control. The asterisk indicates a nonspecific band.