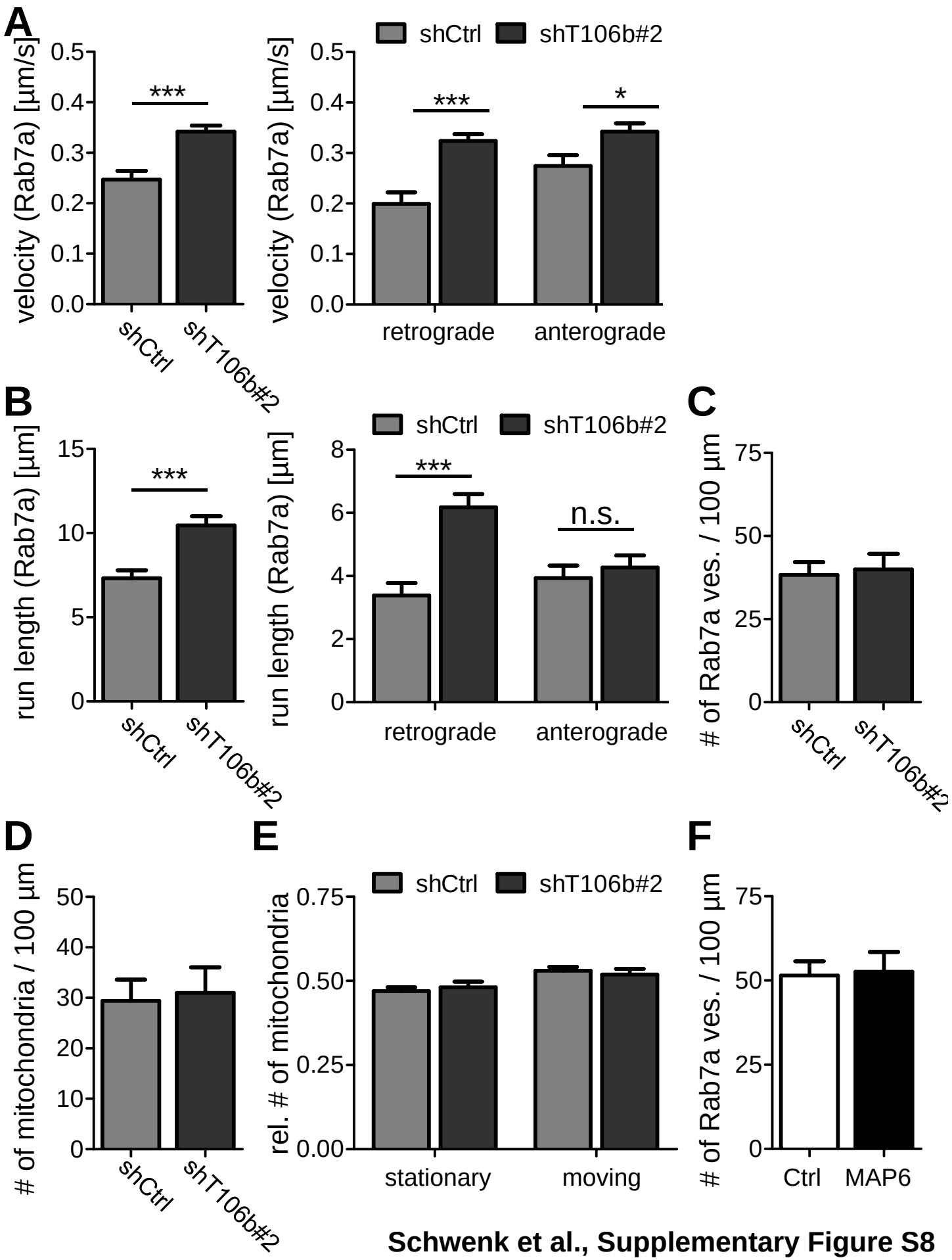

Schwenk et al., Supplementary Figure S8

Supplementary Figure S8: TMEM106B knockdown enhances retrograde movement of late endosomes / lysosomes but not of mitochondria

(A-C) Primary hippocampal neurons (DIV6+3) were transfected with either TMEM106B shRNA #2 or a Ctrl shRNA and Rab7a-GFP. Live cell imaging of Rab7a-GFP labeled vesicles was used to visualize late endosomal / lysosomal trafficking in dendrites. Velocity (A) and total run length (B) of individual moving vesicles was analyzed in kymographs (compare Figure 6). At least 80 moving vesicles per condition were analyzed. The speed of lysosomal movement is in the expected range for organelle transport in dendrites (Bannai et al, 2004; Kwinter et al, 2009; van Spronsen et al, 2013). The number of vesicles per 100 µm of dendrite length did not change (C). (D-E) Live imaging experiments in hippocampal neurons (DIV6+3) transfected with either TMEM106B shRNA #2 or a Ctrl shRNA and mito-dsRed to visualize mitochondrial trafficking in dendrites. TMEM106B had no effect on mitochondrial density (D) and movement (E) in dendrites. (F) MAP6 overexpression had no effect on the density of Rab7a-GFP labeled vesicles in dendrites. Mean +/- SEM, unpaired t-test: * denotes $p<0.05$ *** denotes $p<0.001$.

Supplemental References

Bannai H, Inoue T, Nakayama T, Hattori M, Mikoshiba K (2004) Kinesin dependent, rapid, bi-directional transport of ER sub-compartment in dendrites of hippocampal neurons. *J Cell Sci* **117:** 163-175

Kwinter DM, Lo K, Mafi P, Silverman MA (2009) Dynactin regulates bidirectional transport of dense-core vesicles in the axon and dendrites of cultured hippocampal neurons. *Neuroscience* **162:** 1001-1010

van Spronsen M, Mikhaylova M, Lipka J, Schlager MA, van den Heuvel DJ, Kuijpers M, Wulf PS, Keijzer N, Demmers J, Kapitein LC, Jaarsma D, Gerritsen HC, Akhmanova A, Hoogenraad CC (2013) TRAK/Milton motor-adaptor proteins steer mitochondrial trafficking to axons and dendrites. *Neuron* **77:** 485-502