Supporting Information

Wang et al. 10.1073/pnas.1219203110

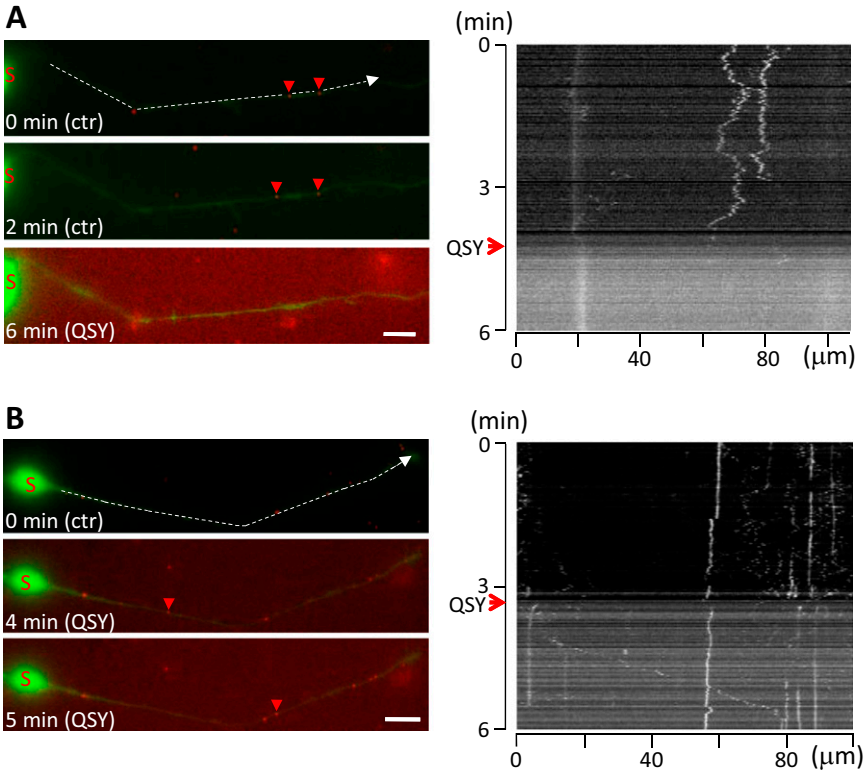

Fig. S1. VAMP2-QDs undergoing random movement on the surface and rapid cytoplasmic transport after internalization in cultured cerebellar granule cells. (*A*) An example of random movement of VAMP2-QDs on the neurite surface before and after QSY-21 incubation. Note the disappearance of all neurite-associated QD fluorescence after QSY-21 incubation (red arrow in kymograph). The kymograph on the right depicts the movements of two surface-bound QDs (red arrowheads on the left). The soma was marked with "S", and the kymograph represents the trajectory of the QD along the neurite at positions marked by the dashed line. The arrow represents the direction of kymograph. (*B*) An example of intracellular transport of VAMP2-QD along the neurite (red arrowhead on the left) after high K^+-induced internalization of VAMP2-OD, visible after quenching of external QDs. The elevated background fluorescence was caused by autofluorescence of the QSY solution. The puncta in the surround are likely internalized QDs in other cells or cell fragments that were inaccessible by extracellular QSY. (Scale bars, 10 μm.)

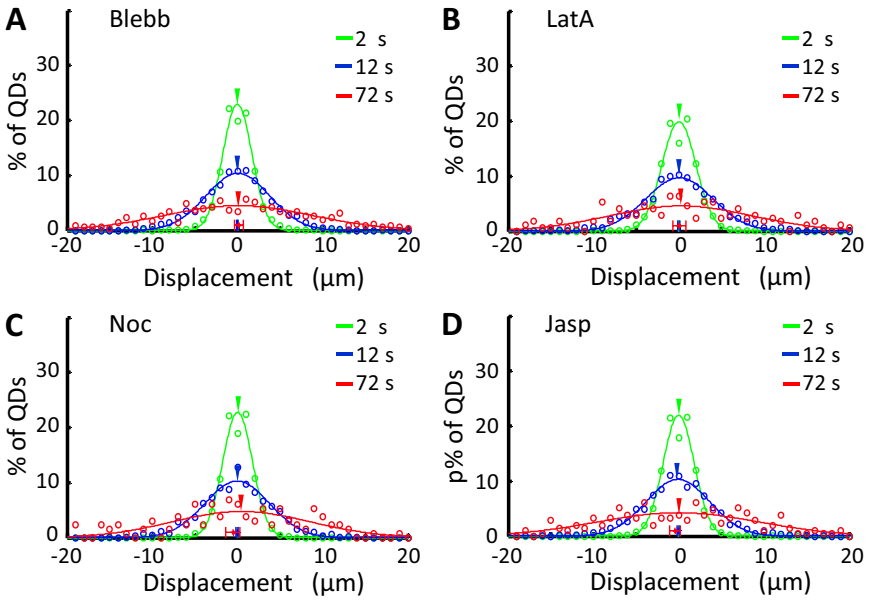

Fig. S2. No directional transport of membrane proteins was seen in the stalling process after treatment of drugs. The distribution of QD movement in the stalling processes in 2-s (green), 12-s (blue), and 72-s (red) intervals after treatment with blebbistatin (Blebb) (*A*), latrinculin A (LatA) (*B*), nocodazole (Noc) (*C*), and jasplakinolide (Jasp) (*D*). Data points with error bars close to the x axis are mean values of the displacement (± SEM). Arrowheads indicate the center for the Gaussian curve.

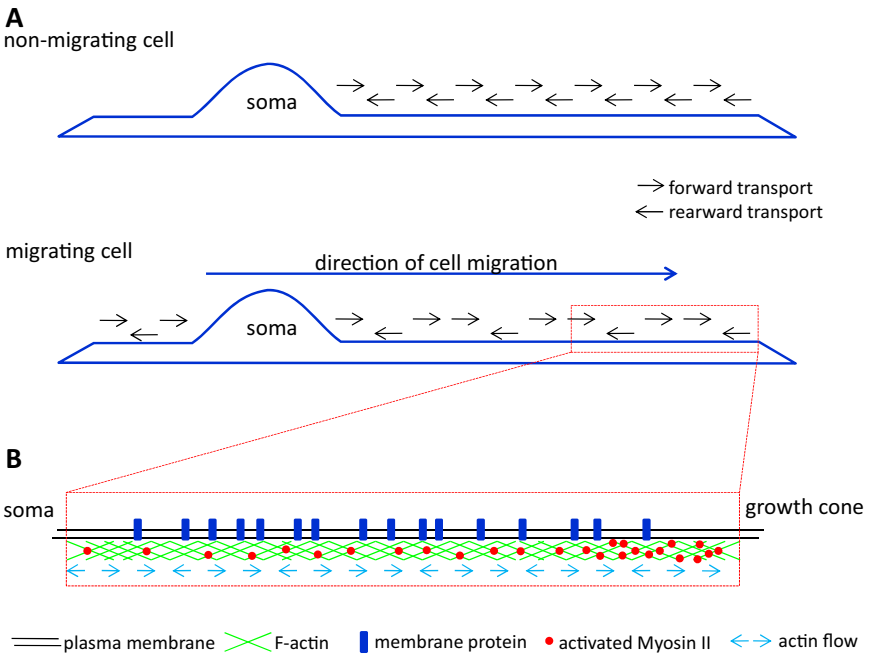

Fig. S3. A schematic diagram showing the potential mechanism of membrane protein movement. (*A*) Membrane proteins exhibit both forward and rearward drift with similar drift speed in migrating and nonmigrating cells, but in migrating cells the frequency of the forward drift is significantly higher than rearward transport, resulting in overall forward transport of membrane proteins. (*B*) The higher frequency of the forward drift of membrane proteins may result from more frequent forward movement of cortical F-actin, driven by the front-high and rear-low distribution of myosin II activity in migrating cells.