## Human Myosin VIIa Is a Very Slow Processive Motor Protein on Various Cellular Actin Structures

Osamu Sato, Satoshi Komatsu, Tsuyoshi Sakai, Yoshikazu Tsukasaki, Ryosuke Tanaka, Takeomi Mizutani, Tomonobu M. Watanabe, Reiko Ikebe, and Mitsuo Ikebe

## SUPPLEMENTAL MOVIE LEGENDS

Supplemental Movie 1. Myosin VIIa movement on single actin filaments. The movement of HM7A $\Delta$ Tail/LZ on single actin filaments was observed using IX83-based TIRF microscope system as described in "Experimental Procedures." Rhodamine-phalloidin labeled F-actin was used in this experiment. The movie was captured at the frame rate of 2 fps. The time was shortened X100 using Adobe Premiere software because of extremely slow velocity of myosin VIIa (~11 nm/s). The time line was shown in the movie (~8 min). The length of one side of the movie = 34  $\mu$ m.

Supplemental Movie 2. Myosin VIIa movement on demembranated MEF-3T3 cells. MEF-3T3 cells were demembranated with Triton X-100, stained with Alexa Fluor 568 phalloidin, blocked with casein, and the movement of HM7A $\Delta$ Tail/LZ-Qdot 525 was observed using IX83-based TIRF microscope system as described in "Experimental Procedures." The red signal of Alexa 568 and concomitant green signal of Qdot 525 were captured at the frame rate of 4 fps, and shortened the time X50. The movie shows the movements of HM7A $\Delta$ Tail/LZ-Qdot 525 on stress fibers, lamellipodia and filopodia. Note that myosin VIIa moves more slowly with a shorter run length in stress fibers. The velocities & run length of myosin VIIa on stress fibers, lamellipodia and filopodia were 6.6 nm/s & 0.41 µm, 8.1 nm/s & 0.59 µm and 9.5 nm/s & 0.69 µm, respectively (see text). The length of one side of the movie = 2.6 µm.

Supplemental Movie 3. Myosin VIIa movement at the cell periphery on demembranated MEF-3T3 cells. The signals of Alexa 568 and Qdot 525 were simultaneously captured using single camera. The movie was captured at frame rate of 10 fps, and shortened the time X10. The length of one side of the movie =  $27.6 \mu m$ .