The functional analysis of a novel myosin Myo13 in Tetrahymena thermophila

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We have recently found that 13 myosin genes are expressed in vegetative growing cells in *T. thermophila.* One of their deduced gene products, Myo13, contains characteristic coiled-coil domains in a tail. Therefore, we would expect that Myo13 forms bipolar filaments like type II myosin and generates mechanical force *in vivo*. First, we prepared antiserum against the first coiled-coil domain of Myo13. Immunofluorescence microscopy revealed that Myo13 localized to the oral apparatus, the deep fiber, cortical dots surrounding nascent and old food vacuoles and the cytoproct a slit for defecation. In particular, Myo13 colocalized with actin in the deep fiber and the food vacuole membrane. By treating cells with Lat-B, an inhibitor for actin polymerization, we found that Myo13 remained in the cytoproct in the absence of F-actin. Surprisingly, the cytoproct was changed dramatically from a linear shape to a ring by Lat-B treatment. SEM revealed that the cytoproct was abnormally opened in these cells, and a protrusion was seen. Live image observation using optical microscopy showed that the protrusion was formed from remnant membranes of egested food vacuoles. This result suggests that F-actin is required for endocytosis to recycle membrane in the cytoproct. It is possible that Myo13 is involved in the process of membrane recycling by interacting with F-actin during egestion of food vacuole.