Identification of a distal GLUT4 trafficking event controlled by actin polymerisation

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The insulin-stimulated trafficking of GLUT4 to the plasma membrane in muscle and fat tissue constitutes a central process in blood glucose homeostasis. The tethering, docking and fusion of GLUT4 vesicles with the plasma membrane represent the most distal steps in this pathway and have been recently shown to be key targets of insulin action. However, it remains unclear how insulin influences these processes to promote the insertion of the glucose transporter into the plasma membrane. In this study we have identified a previously uncharacterized role for cortical actin in the distal trafficking of GLUT4. Using high frequency total internal reflection fluorescence microscopy (TIRFM) imaging we show that insulin increases actin polymerisation near the plasma membrane and that disruption of this process inhibited GLUT4 exocytosis. Using TIRFM in combination with probes that could distinguish between vesicle transport and fusion we found that defective actin remodelling was accompanied by normal insulin-regulated accumulation of GLUT4 vesicles close to the PM, but the final exocytotic fusion step was impaired. These data resolve multiple steps of the final stages of GLUT4 trafficking, demonstrating a crucial role for actin in the final stage of this process. We are now using super resolution TIRF microscopy to begin the characterize the carriers that deposit GLUT4 at the plasma membrane and this has revealed multiple classes of carriers thus further adding to our ability to distinguish between discrete steps in this important process.