1. Abstract

Type 2 diabetes is an increasing global challenge to human health. Muscle insulin resistance, defined as impaired insulin-stimulated muscle glucose uptake, is an established key defect in the early development of type 2 diabetes. Muscle insulin resistance involves a blunted insertion of the glucose transporter 4 (GLUT4) in the surface membrane of the muscle fibers, the sarcolemma, following insulin stimulation. Intriguingly, an acute bout of exercise reverses muscle insulin resistance. Whether this is due to an increased ability for insulin to induce insertion of GLUT4 remains uncertain, however, this is indicated by rodent studies.

Despite the great therapeutic potential of understanding the mechanism(s) of how GLUT4 traffics to reach the sarcolemma during insulin resistance and insulin sensitization following acute exercise, our knowledge remains insufficient and primarily based on studies of cultured cells. Thus, this PhD project overall aimed to provide novel insights into GLUT4 trafficking and subcellular distribution in adult muscle.

In Study 1, we tested the hypothesis that the microtubule (MT) network is required for GLUT4 movement and susceptible to insulin resistance. We observed that a high basal rate of GLUT4 trafficking took place on the MTs and was dependent on an intact MT cytoskeleton in mouse muscle. Insulin increased the travelling distance of the MTs while insulin resistance, induced by C2 ceramide treatment, decreased the travelling distance and the polymerization rate of the MTs. Furthermore, MT disruption dispersed GLUT4 localized in large structures to smaller peripheral structures. This demonstrates a novel role of the MT network in GLUT4 trafficking in muscle and implicates MTs as a key step in GLUT4 trafficking in adult muscle, vulnerable to development of muscle insulin resistance.

Since intracellular compartment localization of GLUT4 affects accessibility to the sarcolemma, Study 2 examined the subcellular compartment distribution in insulin-sensitized muscle induced by exercise. We observed that prior exercise redistributed GLUT4 in human muscle as one-legged kicking exercise increased GLUT4 co-localization with the t-tubules and the GLUT4 storage vesicles. This redistribution from prior exercise was accompanied by insulin-induced GLUT4 localization in the endosomes and increased insulin-induced GLUT4 content in the sarcolemma in the prior exercised leg. Together, these data support the hypothesis that increased insulin sensitivity by prior exercise in humans is caused, at least in part, by potentiation of GLUT4 insertion in the sarcolemma due to intracellular redistribution.
Since GLUT4 insertion in the sarcolemma in adult muscle is technically challenging to measure, we established, in Study 3, a model for quantifying exercise-induced GLUT4 insertion in sarcolemma in adult muscle \textit{in vivo}. To do this, we expressed exofacially tagged GLUT4 in mouse muscle. This assay can be used to improve our mechanistic understanding of processes involving GLUT4 trafficking and insertion in the sarcolemma in adult muscle. In study 3, we demonstrated that sarcolemmal insertion of GLUT4 could take place without visual changes in overall GLUT4 localization, in contrast to what has been indicated by previous studies investigating GLUT4 in rodent muscle without directly measuring inserted GLUT4.

In conclusion, the MT network constitutes an important trafficking component for GLUT4 in adult muscle and may contribute to insulin resistance. In addition, exercise-induced insulin sensitization is associated with subcellular redistribution of GLUT4 in unstimulated muscle fibers 4 hours after exercise and augmented insulin-induced GLUT4 insertion in sarcolemma. Finally, GLUT4 insertion in the sarcolemma seemingly occurs without visual changes in global GLUT4 localization. Thus, this PhD project provided novel insights into GLUT4 trafficking and subcellular redistribution in adult muscle which may guide future research efforts attempting to reverse muscle insulin resistance.