Exercise Biochemistry Review

Proceedings of IBEC 2018, Beijing, China, October 23-25
PO-033

Mitochondria-associated Ampk mediates mitochondrial quality control in skeletal muscle

Joshua Drake[1,2],Rebecca Wilson[1,2],Rhianna Laker[1,2],Yuntian Guan[1,4],Tatiana Coverdell[1,4],Maya Dorn[1,2],Poonam Sharma[1,5],George Christ[1,5],Mei Zhang[1,2],Zhen Yan[1,2,6]
1.University of Virginia
2.Center for Skeletal Muscle Research at Robert M. Berne Cardiovascular Research Center
3.Department of Biochemistry and Molecular Genetics
4.Deparment of Pharmacology
5.Department of Biomedical Engineering
6.Department of Medicine

Objective Mitochondria exist as a complex, interconnected reticulum with the degree of complexity at homeostasis varying by cell type. Declines in mitochondrial quality is a hallmark of numerous diseases and is partly maintained through the targeted removal and degradation of damaged/dysfunctional regions of the reticulum known as mitophagy. We have demonstrated that acute exercise causes targeted degradation of a subset (<1%) of the mitochondrial reticulum through mitophagy in an AMPK-dependent manner. However, how such spatial regulation is achieved is unclear. Given that AMPK is a well-known bioenergetics sensor critical for maintenance of energetic homoeostasis, and numerous studies have linked AMPK-signaling to mitochondrial remodeling and functional adaptations under physiological and pathological conditions, we hypothesized that AMPK regulates mitophagy by being physically associated with the mitochondria.
Methods C57BL/6 mice (3 or 20 months of age) were kept in standard conditions (12:12 light, dark cycle) on normal chow. Enriched mitochondrial fractions from skeletal muscle were obtained via differential centrifugation and percoll gradient isolation. Skeletal muscle energetic stress was induced via acute treadmill running, direct electrical stimulation, or hindlimb ischemia-reperfusion. Somatic gene transfer into skeletal muscle was done via electraporation. Confocal microscopy of whole mount skeletal muscle was performed at least 10 days post-electraporation to assess MitoTimer fluorescence Red:Green Ratio and Mitophagy (i.e. presence of pure red puncta).
Results We have discovered that AMPK is enriched in isolated mitochondria from both mouse and human skeletal muscle (mitoAMPK) and that, in mice, mitoAMPK consists exclusively of α1/β2/γ1 isoforms. Furthermore, skeletal muscle mitoAMPK Thr172 phosphorylation, indicative of activation, is increased following electrical stimulation, ischemia-reperfusion, and acute treadmill running. By co-transfecting mouse FDB muscle with pMitoTimer and a mitochondrially-targeted AMPK inhibitor peptide (mitoAIP), we show that mitoAMPK activity is required to maintain mitochondrial quality (as evidenced by increased MitoTimer Red:Green ratio) and exercise-induced mitophagy (MitoTimer pure red puncta). Interestingly, association of mitoAMPK with mitochondria declines by ~50% in skeletal muscle in old mice and coincides with poor mitochondrial quality (increased MitoTimer Red:Green ratio), which may explain the loss of mitochondrial quality and metabolic flexibility with aging.
Conclusions Our current working hypothesis is that activation of mitoAMPK by exercise plays an instrumental role in promoting mitochondrial remodeling, hence contractile and metabolic adaptations, by spatially recognizing damaged regions of the mitochondrial reticulum. Future research on mitoAMPK will significantly improve the mechanistic understanding of exercise training-induced adaptation in skeletal muscle and AMPK biology.