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COMMENTARY

Exercise and Mitochondrial Function in Adipose Biology: All Roads Lead to NO

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Insulin resistance in rodents and humans is highly correlated with inflammation of white adipose tissue (WAT) and the presence of proinflammatory immune cells. Such chronic immune challenge leads to a variety of metabolic effects in the adipocyte, including endoplasmic reticulum stress, mitochondrial dysfunction, oxidative stress, and altered adipokine and cytokine secretion, which in sum play a major role in affecting whole-body insulin sensitivity (1–3). Of the factors regulating mitochondrial biogenesis, the regulation of the endothelial nitric oxide synthase (eNOS) has garnered considerable attention as a major metabolic control point.

eNOS is a constitutively expressed nitric oxide (NO)–producing enzyme classically linked to smooth muscle contraction and platelet aggregation (4). However, eNOS and downstream NO signaling are now appreciated as a major metabolic determinant of the peroxisome proliferator–activated receptor γ coactivator 1α (PGC1α) (5). Although the molecular mechanism(s) is still not completely understood, NO signaling and activation of the soluble guanylyl cyclase–protein kinase G (PKG) system upregulate PGC1α expression and, moreover, are required for increased expression of SirT1, the major NAD^+-dependent deacetylase controlling PGC1α activity (6–8). NO signaling via PKG stimulates the expression of the entire mitochondrial biogenesis program, including the activation of Nrf1 and Tfam, two major transcription factors regulating expression of mitochondrial enzymes.

A second major theme controlling mitochondrial biogenesis linked to sympathetic drive and eNOS is G-protein–coupled receptor–dependent activation of CREBP. Studies by a number of laboratories have shown that CREBP is a major control point in skeletal and aortic muscle affecting development of insulin resistance and that CREBP is a critical regulator of mitochondrial biogenesis and dynamics (9,10). Adrenergic stimulation and cAMP production activate not only CREBP but also protein kinase A, which in turn phosphorylates and potentiates PGC1α (9), thereby providing an additional mechanism by which catecholamines can upregulate mitochondrial biogenesis.

In 2009, Sutherland et al. (11) published that exercise and adrenergic stimulation upregulated the expression of PGC1α in rat WAT. In their report, exercise training increased PGC1α expression in both epididymal and retroperitoneal fat depots while epinephrine treatment alone increased PGC1α mRNA levels selectively in epididymal WAT, suggesting that catecholaminergic stimulation may be a potential mechanism by which exercise induces mitochondrial biogenesis in fat. Consistent with this, β-blockade attenuated, but did not eliminate, the effects of exercise on PGC1α upregulation. Previous studies by the groups of Nisoli and Vettor (6,12,13) have clearly demonstrated that eNOS mRNA and activity is highly regulated in muscle and WAT and that eNOS is required for the upregulation of SirT1 in response to caloric restriction. Extending these studies, Koh and colleagues (14,15) have shown when using $eNOS^{-/-}$ mice and 3T3-L1 adipocytes that NO signaling plays a major role in controlling adiponectin synthesis. The observation that mitochondrial function and integrity in WAT are linked to adiponectin secretion provides a potential mechanism by which insulin resistance may be regulated in $eNOS^{-/-}$ mice.

In this issue, Trevellin et al. (16) used exercise training of wild-type and whole-body $eNOS^{-/-}$ mice to test the hypothesis that chronic exercise would increase mitochondrial biogenesis in WAT in an eNOS-dependent manner. Using a swim-training model in C57BL/6J mice, the studies confirmed the original observation that chronic exercise increased mitochondrial DNA, mitochondrial enzymes, and expression of mitochondrial biogenesis transcription factors (Nrf1 and Tfam) and coactivators (PGC1α) in subcutaneous adipose tissue (SAT) compared with sedentary controls. In contrast, the beneficial effects

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of exercise were not evidenced in eNOS-null animals. The effects of exercise on the upregulation of UCP1 as a biomarker for "browning" of SAT were also confirmed in wild-type mice but markedly reduced in eNOS$^{-/-}$ animals (17,18). To test the hypothesis that adrenergic stimulation may be playing a role in the exercise-induced mitochondrial biogenesis program, the authors challenged wild-type and eNOS-null mice with norepinephrine and demonstrated that the increase in PGC1α and cytochrome c oxidase expression was markedly attenuated despite the presence of a functional PKA-extracellular signal–related kinase 1 and 2 signaling system. Exercise also increased the insulin-stimulated component of glucose transport in isolated subcutaneous adipocytes, and this effect was totally lost in SAT from eNOS-null mice. These results may suggest that some of the effects of exercise on adipocyte mitochondrial biogenesis may be mediated by catecholaminergic signaling, while a second pathway independent of sympathetic drive linked to eNOS in the adipocyte may play a crucial role in affecting mitochondrial number and activity (Fig. 1).

The finding that exercise upregulates PGC1α expression in adipose tissue in an eNOS-dependent manner is consistent with other findings reporting that exercise training upregulates PGC1α in nonmuscle cells (19). Despite these advances, there are still major themes left unresolved. First, as muscle is the major glucose-utilizing tissue, it is not clear how eNOS-dependent upregulation of mitochondrial biogenesis in adipose tissue per se affects whole-body energy metabolism. With the suggestion of an endocrine loop system that connects muscle to fat and back to muscle via the myokine-adipokine axis (17), the whole-body utilization of glucose is likely intimately connected by multiple factors. Second, as exercise training reduces inflammation in adipose tissue (20) and inflammatory cytokines downregulate the mitochondrial biogenesis program in fat cells (3), it is not clear whether the effects are directly mediated on the biology of adipocytes or via attenuation of inflammatory macrophages. Indeed, downregulation of GSTA4 in adipocytes leads to downregulation of eNOS and the entire mitochondrial biogenesis program (3). Lastly, the eNOS-null mouse used in the study by Trevellin et al. (16) was a whole-body knockout and the specific role of adipocyte eNOS has not been established. Clearly, NO donors in isolated adipocytes have the capacity to improve glucose metabolism (16), but the work to date does not demonstrate that exercise training does not affect metabolic processes in nonadipocytes that in turn indirectly influence the expression of eNOS. As such, while the study by Trevellin et al. (16) advances our understanding of the complex regulatory circuits that control mitochondrial biogenesis, more questions are revealed. What is clear is that muscle–fat balance controlling energy metabolism is likely to be mediated by control of eNOS.

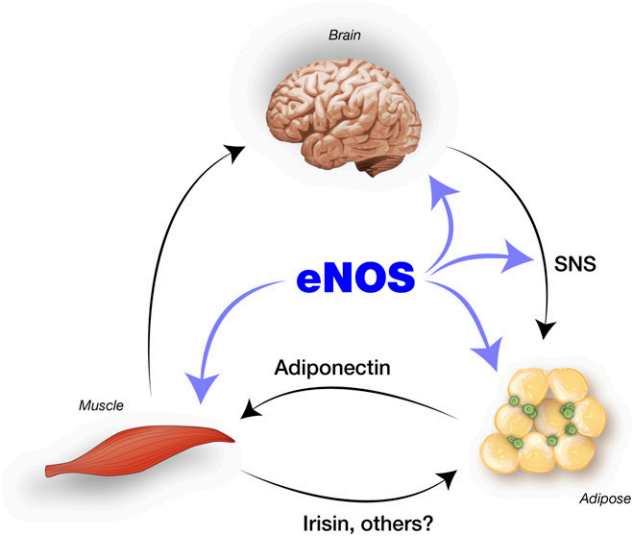

Figure 1—Central role of eNOS in exercise-induced mitochondrial metabolism. Depiction of the role eNOS may play in exercise-induced mitochondrial biogenesis in fat tissue, either indirectly via the sympathetic nervous system (SNS) or directly via a myokine-adipokine axis. Within adipose depots, the role of eNOS may be linked to the biology of the adipocyte or to the immune cells present in the tissue, or both.

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