Guanghong Jia,[1,2] William Durante,[3] and James R. Sowers[1,2,3,4]

Endothelium-Derived Hyperpolarizing Factors: A Potential Therapeutic Target for Vascular Dysfunction in Obesity and Insulin Resistance

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COMMENTARY

The endothelium consists of a single layer of cells that serves as a barrier between blood and tissues and actively participates in the regulation of vascular tone and function (1). The influence of the endothelium on blood flow in arterioles and capillaries is modulated by the synthesis and release of a number of endothelial-derived relaxing and constricting substances such as nitric oxide (NO), prostacyclin (PGI_2), and endothelium-derived hyperpolarizing factors (EDHF) (2). However, the balance or homeostasis between endothelial relaxing and constricting factors is disrupted in insulin-resistant states including obesity, type 2 diabetes mellitus (T2DM), and hypertension, all of which promote cardiovascular disease (CVD) (1,2). For example, endothelial cell (EC) dysfunction is caused by impairment of endothelial-dependent vasodilation in various vascular beds such as skeletal muscle arterioles and capillaries. Both conduit arteries and microvessels are very important for insulin and glucose metabolism as an increase in capillary surface area prompts both insulin and glucose delivery and subsequent glucose uptake in skeletal muscle tissue (3,4).

In healthy individuals, systemic and local infusion of insulin induces NO production to prompt insulin-mediated capillary recruitment and microvascular blood flow, resulting in an increase in forearm blood volume (2). One study demonstrated that endothelial NO synthase inhibition significantly blunted the insulin-stimulated increase of ultrasound-assessed femoral blood flow without any changes in blood pressure and heart rate in rats (5). In parallel, insulin-stimulated capillary recruitment and glucose uptake were completely abolished (5), suggesting that local capillary NO bioavailability plays a key role in the regulation of insulin-stimulated skeletal muscle capillary recruitment and glucose uptake.

The three main endothelial-derived relaxing substances that regulate arteriole and capillary tone are NO, PGI_2, and EDHF (Fig. 1) (1–3). NO is the principal regulator of flow-mediated dilation, and this process is impaired in insulin-resistant states (1–3). In addition to NO, ECs also produce and release PGI_2 from cyclooxygenase-derived metabolites in response to shear stress. PGI_2 crosses EC membranes and activates vascular smooth muscle cell (VSMC) adenylyl cyclase and protein kinase A, resulting in VSMC relaxation (1–3). In contrast to the well-defined NO and PGI_2 pathways, the molecular constituents and mechanism of EDHF-mediated relaxation remain controversial. Classically, endothelium-derived NO largely mediates large conduit artery relaxation, whereas EDHF plays an important role in modulating vascular tone in small resistance arteries (6). NO-mediated relaxation is easily impaired, whereas EDHF-mediated responses are generally preserved or even enhanced to maintain vascular homeostasis in insulin-resistant states (7). Thus, EDHF is regarded as a backup system for NO-mediated responses in the maintenance of tissue perfusion. It is widely accepted that EDHFs include epoxyeicosatrienoic acids (EETs) derived from a metabolite of cytochrome P450 (CYP) epoxygenase, electrical communication through gap junctions, endothelium-derived potassium ions (K^+), hydrogen sulfide, and endothelium-derived hydrogen peroxide (8). The EET pathway, comprising four EET isomers (5,6-EET, 8,9-EET, 11,12-EET, and 14,15-EET) (9) is well established as an important EDHF. EETs are rapidly metabolized by soluble epoxide hydrolase to form the generally less active dihydroxyeicosatrienoic acids (9). After ECs are stimulated with various agonists, an increase in EC calcium (Ca^{2+}) influx occurs that can potentially increase Ca^{2+}-sensitive small- and intermediate-conductance potassium channels (K_{Ca}) (10). Following

[1]Diabetes and Cardiovascular Research Center, Columbia, MO
[2]Harry S. Truman Memorial Veterans Hospital, Columbia, MO
[3]Department of Medical Pharmacology and Physiology, University of Missouri School of Medicine, Columbia, MO
[4]Dalton Cardiovascular Research Center, Columbia, MO
Corresponding author: James R. Sowers, sowersj@health.missouri.edu.

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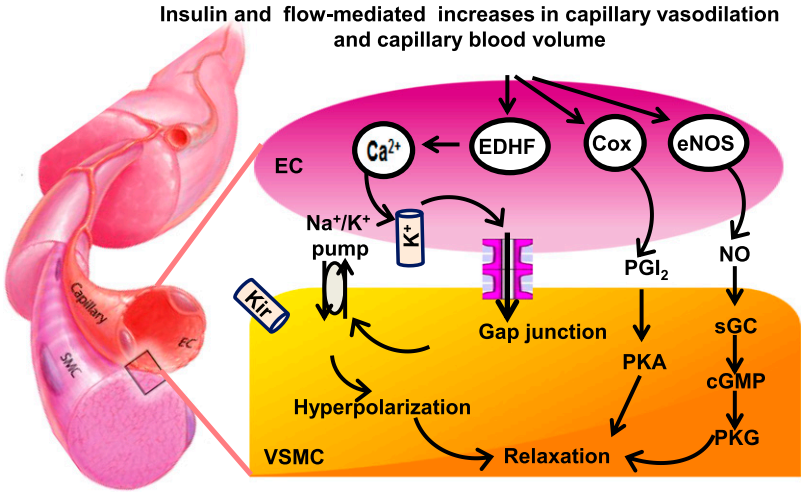

Figure 1—Proposed mechanisms of endothelium-dependent vasorelaxation by NO, PGI_2, and EDHF. cGMP, cyclic guanosine monophosphate; Cox, cyclooxygenase; eNOS, endothelial NO synthase; PKA, protein kinase A; PKG, protein kinase G; sGC, soluble guanylyl cyclase.

activation of K_{Ca} on ECs, VSMC hyperpolarization is preferentially evoked through myoendothelial gap junctions by passage of a current or diffusion of a factor such as K^+ through the junctions. This, in turn, activates Kir2.1 inward-rectifier K^+ channels and/or Na^+/K^+-ATPase on VSMCs to close voltage-dependent Ca^{2+} channels resulting in VSMC relaxation (10,11) (Fig. 1). To this point, myoendothelial gap junctions provide the means by which hyperpolarization of ECs is transferred to VSMCs (10) (Fig. 1). Myoendothelial gap junctions also facilitate EDHF diffusion from ECs to VSMCs and directly activate VSMC ion channels, resulting in VSMC hyperpolarization and relaxation (11).

Diet-induced obesity and associated insulin resistance are leading causes of vascular dysfunction and impaired tissue perfusion (1–3). In this issue of *Diabetes*, Chadderdon et al. (12) investigate the temporal changes in skeletal muscle capillary responses and endothelial-derived vasodilators in a clinically relevant model of high-fat, high-fructose diet–induced obesity and insulin resistance in primates. This translational diet induced complex temporal responses in skeletal muscle capillary blood volume (CBV) that paralleled dissociation between NO and eicosanoid endothelial-derived vasoregulatory pathways. Indeed, there was a compensatory increase in basal and glucose-stimulated CBV that was associated with an upregulation of lipoxygenase and CYP epoxygenase–derived eicosanoids for up to 18 months, in contrast to an inexorable decline in NO bioavailability, suggesting that a compensatory EDHF response occurs in the early stage of obesity and insulin resistance. That a diet-induced deficiency in NO can upregulate the EDHF response is consistent with the observation that NO inhibits EDHF responses through inhibition of CYP epoxygenase activity and myoendothelial gap junctions (6,7). After 2 years of the high-fat, high-fructose diet, an abrupt loss of the compensatory increase in the lipoxygenase-derived eicosanoid vasodilators coincided with a drop in skeletal muscle microvascular blood flow and peak CBV along with progressive worsening of insulin resistance. Thus, the increase in basal and glucose-mediated CBV in early stages of insulin resistance may represent a compensatory response through EDHF that is lost over time.

This study (12) provides new insights regarding changes of NO and EDHF in the regulation of skeletal muscle blood flow and insulin sensitivity. This is translationally relevant as therapeutic targeting of NO and EDHF also has a potential application for prevention of vascular dysfunction such as vascular stiffness, hypertension, and coronary heart disease that are common causes of morbidity and mortality in T2DM patients. These data highlight a compensatory role of EDHF in the regulation of insulin-mediated capillary recruitment and CBV, consistent with previous observations in other vascular beds. For example, a decrease in EDHF-mediated vasodilatory responses has been reported in mesenteric and carotid arteries as well as in the renal circulation of streptozotocin-induced diabetic rats (13–15). Further, acetylcholine-induced relaxation in sciatic nerve epineurial arterioles is impaired by a reduced EDHF pathway in Zucker diabetic fatty rats (15–17). Therefore, alterations in the EDHF pathway can contribute to both endothelial dysfunction or conversely compensate for the loss in NO bioavailability, depending on the various underlying pathophysiological abnormalities and particular vascular bed being studied. One limitation of the authors interpretation of their results is that they should have acknowledged that NO has been found to inhibit EDHF responses through inhibition of CYP epoxygenase activity (6,7). Thus, measurement of CYP epoxygenase activity and electrophysiology in concert with bioavailable NO in future studies should help us to understand the interactive mechanisms involved in regulating insulin-stimulated skeletal muscle capillary recruitment.

In conclusion, data in the current study (12) defines the skeletal muscle microvascular responses during the development and progression of insulin resistance in diet-induced obesity and describes the relationship between changes in EDHF and NO in the progression of impaired capillary recruitment and increases in insulin and glucose delivery to skeletal muscle tissue. These findings indicate that increasing EDHF may be a potential novel therapeutic strategy in patients with CVD in T2DM. Further studies are warranted to more definitively understand the relative role of NO and EDHF in diet-induced insulin resistance and T2DM.

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Duality of Interest. No potential conflicts of interest relevant to this article were reported.

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