Glucose Transport Activity in Insulin-Resistant Rat Muscle

Effects of Angiotensin-Converting Enzyme Inhibitors and Bradykinin Antagonism

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Insulin resistance of skeletal muscle glucose disposal underlies the pathogenesis of NIDDM and is associated with hypertension, obesity, and dyslipidemia. Angiotensin-converting enzyme (ACE) inhibitors are used primarily in antihypertensive therapy but also are known to improve whole-body insulin-mediated glucose disposal. However, the exact site of action is not well characterized. We have used the isolated epitrochlearis muscle from a well-established animal model of skeletal muscle insulin resistance, the obese Zucker rat, to test the effect of oral administration of ACE inhibitors on insulin-sensitive muscle glucose transport activity. Both acute and chronic administration of a sulfhydryl-containing ACE inhibitor (captopril) or a non–sulfhydryl-containing ACE inhibitor (tran-dolapril) significantly enhanced in vitro insulin-mediated muscle glucose transport activity. In addition, the acute effect of oral captopril administration was completely abolished by pretreatment of the animal with a bradykinin B_2 receptor antagonist (HOE 140). These findings indicate that ACE inhibitors may improve whole-body glucose metabolism by acting on the insulin-sensitive skeletal muscle glucose transport system. In addition, bradykinin or one of its metabolites may be involved in the action of the ACE inhibitor captopril on insulin-resistant muscle. *Diabetes* 45 (Suppl. 1):S125–S128, 1996

Syndrome X, or the metabolic syndrome (1), describes the clustering of atherogenic risk factors in the hypertensive patient, including insulin resistance of whole-body glucose disposal, hyperinsulinemia, obesity, and dyslipidemia. In addition, ~40% of individuals with non-insulin-dependent diabetes mellitus (NIDDM) are hypertensive and have an increased risk of cardiovascular disease (2,3). A frequent and successful intervention in hypertension is the use of angiotensin-converting enzyme (ACE) inhibitors, which lower blood pressure and improve other abnormal cardiovascular variables in this population (4–6). Whereas some of the other commonly used antihypertensive agents, such as β-adrenergic blockers and thiazides, cause a further decrease in insulin sensitivity (7–10), ACE inhibitors improve insulin sensitivity in both acute (11–14) and chronic studies (7,15–18).

Despite these several studies in this area, it is still not firmly established whether the improved action of insulin on whole-body glucose disposal after ACE inhibitor treatment arises from alterations in the skeletal muscle glucose transport system via nonspecific effects such as improved capillary blood flow and glucose delivery or a combination of the two effects. In this context, this article will review our recent experimental findings on the effects of acute and chronic ACE inhibition on the skeletal muscle glucose transport system in an animal model of severe insulin resistance. In addition, data that shed some light on the possible biochemical mechanism underlying the effect of acute ACE inhibition on this system will be reviewed.

MODE OF ACTION OF ACE INHIBITORS

ACE inhibitors appear to elicit their effects by the inhibition of two enzymatic pathways (Fig. 1). The best-described mode of action of ACE inhibitors involves the inhibition of the conversion of angiotensin I to angiotensin II, both systemically and locally (6). This leads to smooth muscle relaxation and to a reduction of vascular resistance and mean arterial blood pressure. A second, less emphasized mode of action is the inhibition of kininase II, an enzyme identical to ACE (19), which leads to a decreased degradation of bradykinin (6). Therefore a potentially important mechanism of action of ACE inhibitors, both hemodynamically and metabolically, is the increased role of bradykinin and its metabolic products, including prostaglandins (PGs) (20).

THE OBESE ZUCKER RAT: AN ANIMAL MODEL OF SKELETAL MUSCLE INSULIN RESISTANCE

The obese Zucker rat was first described in 1961 by Zucker and Zucker (21); it displays a number of pathophysiological variables associated with human NIDDM (22), as detailed in Table 1. Central among these is the severe resistance to insulin for activation of the glucose transport process, a primary defect in NIDDM (23,24). We have used the isolated epitrochlearis muscle to study skeletal glucose transport activity and have with this approach circumvented the

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ACE, angiotensin-converting enzyme; NIDDM, non-insulin-dependent diabetes mellitus; PG, prostaglandin.

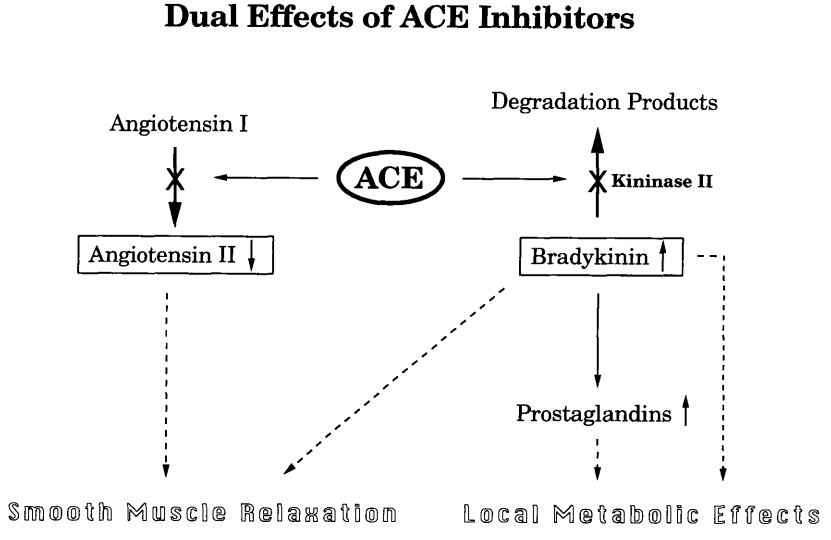

FIG. 1. Effects of ACE inhibitors on enzyme systems and subsequent adaptations in vascular and metabolic parameters.

influence of blood flow on this process. With this preparation, substrate is provided only by diffusion into the extracellular space of the muscle from the incubation medium (25,26). The epitrochlearis muscle, consisting primarily of type IIb fibers (27), is a well-accepted preparation with which to study the muscle glucose transport system, because *1*) there are no diffusion limitations for oxygen or substrates, even in muscles from larger or older rats (28), *2*) high-energy phosphate levels are maintained (28), and *3*) there is an excellent response to stimuli such as insulin or contractions for activation of glucose transport activity (25,28). However, because of its fiber-type composition, results from studies using the epitrochlearis may not necessarily apply to muscle of differing fiber-type compositions.

The ability of a maximally effective dose of insulin to activate 2-deoxyglucose uptake, an accurate index of glucose transport activity (28,29), is <50% ($P < 0.05$) as great in muscle from the obese rat as in weight-matched (~30 mg) muscle from age-matched (10-week-old) lean Zucker rats (30) (Fig. 2). Therefore the isolated epitrochlearis muscle can be used to investigate the underlying mechanisms for and possible interventions against the severe skeletal muscle insulin resistance in the obese Zucker rat.

ACE INHIBITION AND SKELETAL MUSCLE GLUCOSE TRANSPORT

Obese Zucker rats were treated both acutely and chronically with two types of ACE inhibitors. Captopril is a sulfhydryl-containing ACE inhibitor with a relatively short half-life, whereas trandolapril does not contain sulfhydryl groups and has a longer half-life. Neither compound has an effect on glucose transport activity in the absence of insulin (30) (E.J.H., S.J., H.J.A., G.J.D., unpublished data). Therefore the data presented here are restricted to the insulin-mediated increases in glucose transport activity. A single dose of captopril significantly improved insulin-mediated glucose transport activity by 46% ($P < 0.05$) compared with the obese vehicle-treated group (Fig. 2). Likewise, trandolapril acutely enhanced insulin-mediated glucose transport activity by 33%.

Chronic (14 days) treatment with captopril or trandolapril also resulted in a significant improvement of insulin-mediated glucose transport activity in muscle from the obese Zucker rat (Fig. 3). Chronic captopril treatment resulted in a 60% enhancement of insulin-mediated glucose transport activity, whereas this variable was improved by 70% relative to the obese vehicle-treated group after chronic trandolapril treatment. Our observation that both captopril and trandolapril are capable of eliciting an improvement in insulin action on glucose transport indicates that this phenomenon represents a class effect of ACE inhibitors rather than a substrate-specific effect and that the sulfhydryl groups of captopril are not essential for the metabolic effects of ACE inhibitors. This contention is consistent with the study of Paolisso et al. (16), who demonstrated similar improvements in insulin sensitivity of glucose disposal in a placebo-controlled clinical study

TABLE 1
Conditions common to the obese Zucker (*fa/fa*) rat and human NIDDM

Hyperinsulinemia
Hyperlipidemia
Adipocyte hypertrophy, especially in central fat depots
Abnormal oral glucose tolerance
Skeletal muscle insulin resistance of glucose transport

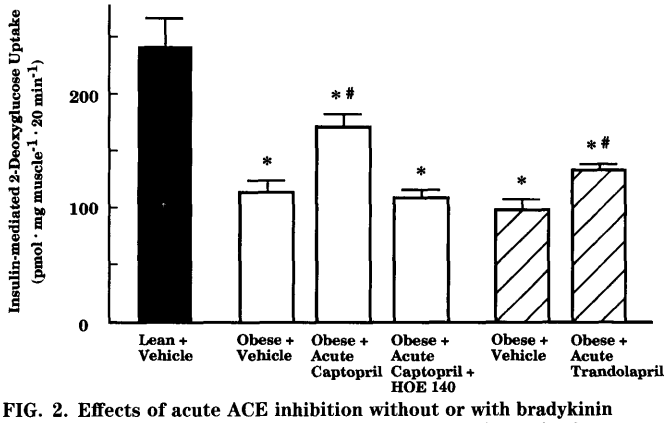

FIG. 2. Effects of acute ACE inhibition without or with bradykinin antagonism via HOE 140 on insulin-mediated skeletal muscle glucose transport activity. Captopril (50 mg/kg body wt) was administered by gavage with or without pretreatment with HOE 140 (100 μg/kg i.p.). After 1 h, insulin-mediated glucose transport activity (2-deoxyglucose uptake in the presence of 13.3 nmol/l insulin minus 2-deoxyglucose uptake in the absence of insulin) was determined (30). Trandolapril (3 mg/kg) was administered by gavage, and 6 h later insulin-mediated glucose transport activity was determined. Values are means ± SE for 5–12 animals per group. *$P < 0.05$ vs. lean + vehicle group; #$P < 0.05$ vs. respective obese + vehicle group by analysis of variance.

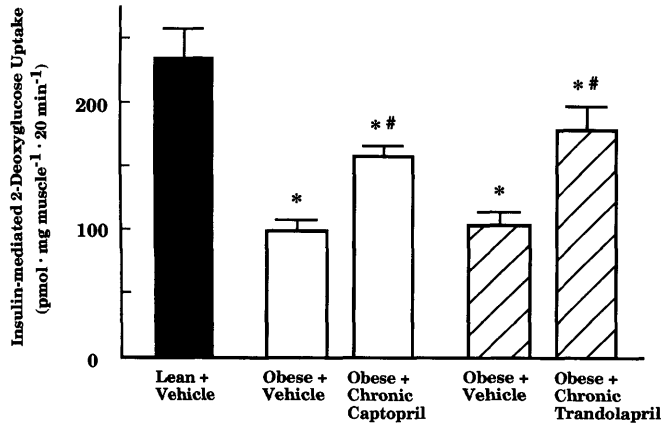

FIG. 3. Effects of chronic ACE inhibition on insulin-mediated skeletal muscle glucose transport activity. Captopril (50 mg · kg body wt^{-1} · day^{-1}) and trandolapril (1 mg · kg^{-1} · day^{-1}) were administered by gavage for 14 consecutive days. At ~20 h after the final treatment, insulin-mediated glucose transport activity was determined as described in the legend to Fig. 2. Captopril data are from Henriksen and Jacob (30). Values are means ± SE for 5–12 animals per group. *P < 0.05 vs. lean + vehicle group; #P < 0.05 vs. respective obese + vehicle group by analysis of variance.

after treatment with five different ACE inhibitors (including captopril).

Interestingly, the improvement in glucose transport activity after chronic ACE inhibitor treatment of obese Zucker rats appears to be restricted to the insulin-dependent pathway for activation of this process, because chronic captopril treatment had no effect on glucose transport activity stimulated by muscle contractions (30). In support of this is our finding that basal glucose transport activity is unaffected by acute or chronic ACE inhibitor treatment (30) (E.J.H., S.J., H.J.A., G.J.D., unpublished data).

ROLE OF BRADYKININ IN THE ACTION OF ACE INHIBITORS

The role of bradykinin in the improvement of insulin action on glucose transport by acute ACE inhibition was investigated by pretreating obese Zucker rats with an intraperitoneal injection of the bradykinin B_2 receptor antagonist HOE 140 (D-arginyl-L-arginyl-L-prolyl[4R0–4-hydroxyprolyl]-glycyl-L-[3-(2-thienyl)alanyl]-L-seryl-D-[1,2,3,4-tetrahydroisoquinololin-3-ylcarbonyl]-L-[(3aS,7aS)-octahydroindol-2-ylcarbonyl]-L-arginine acetate; Hoechst-Roussel, Somerville, NJ) 1 h before captopril administration. This bradykinin antagonism completely abolished the enhanced insulin effect on glucose transport activity normally observed after captopril treatment (Fig. 2). These data provide clear evidence that bradykinin or one of its metabolites, such as PGs, is involved in the improvement of insulin-mediated skeletal muscle glucose transport activity after administration of an ACE inhibitor.

This finding is in agreement with the recent study of Uehara et al. (18), who demonstrated that bradykinin antagonism prevents significant increases in whole-body insulin-mediated glucose disposal due to acute captopril treatment in insulin-resistant dogs and in human patients with NIDDM. Most previous investigations have attributed the influence of the ACE inhibitors on glucose disposal to improved capillary blood flow and an accompanying increased delivery of insulin and glucose to the muscle (11–13,31,32). Additionally, Hirooka et al. (32) reported an improvement of endothelium-dependent vasodilation after administration of captopril, and Kodama et al. (31) reported an improvement of glycemic control in NIDDM subjects accompanied by an increase in forearm blood flow. Although the potential contribution of hemodynamic influences of ACE inhibitors on glucose disposal cannot be ruled out, the findings presented here, using an isolated muscle preparation, support an additional effect of ACE inhibitors and bradykinin and/or its metabolites on the skeletal muscle glucose transport system.

CONCLUDING REMARKS AND FUTURE DIRECTIONS

The foregoing material has clearly demonstrated that ACE inhibitors can elicit local effects on the insulin-dependent glucose transport system of skeletal muscle and can significantly improve insulin-stimulated glucose transport activity independently of any direct hemodynamic influence. Additionally, we have provided evidence that the bradykinin system plays an important role in mediating the acute effect of ACE inhibition on insulin-stimulated glucose transport activity in insulin-resistant rat muscle. These local adaptive responses in the muscle glucose transport system likely are part of a global effect of ACE inhibition, which would include an increase in substrate and hormone delivery via enhanced muscle blood flow. Together, these two independently regulated alterations likely account for the improvements in whole-body insulin-mediated glucose disposal observed after treatment with ACE inhibitors.

Although in these investigations we have not identified any cellular mechanisms responsible for the effects of the ACE inhibitors on the glucose transport system, there are several potential candidates. First, the translocation of glucose transporters (GLUT4) to the plasma membrane in response to insulin in muscle from obese Zucker rats is defective (33), and the possibility exists that these compounds might enhance this process. Second, because the muscle level of GLUT4 appears to be closely correlated to the maximal ability of insulin to activate glucose transport (34), it is possible that long-term treatment with ACE inhibitors can elevate GLUT4 expression in muscle and thereby increase insulin action on glucose transport.

Finally, it is currently unknown exactly what role bradykinin and/or bradykinin-derived products, such as the PGs, might play in the ACE inhibitor-mediated improvement in insulin-stimulated glucose transport activity. As mentioned above, ACE is identical to kininase II (19), and its inhibition leads to an increase in bradykinin (18) and PGs (6). In this context, it is of importance that an intra-arterial infusion of PGE_1 can increase muscle glucose uptake to a much greater extent than can be accounted for by slightly increased blood flow (20). In addition, administration of PGE_2 in vitro significantly increases insulin-stimulated glucose transport activity in rat skeletal muscle (35,36). The elucidation of the cellular mechanisms responsible for the observed improvements in insulin action after acute and chronic ACE inhibition will be the focus of future investigations.

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