Inhibition of Angiotensin Type 1 Receptor Prevents Decline of Glucose Transporter (GLUT4) in Diabetic Rat Heart

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There is some evidence that inhibition of angiotensin-converting enzyme (ACE) activity can improve the uptake and conversion of glucose by heart and skeletal muscle in diabetes. To study the underlying mechanisms, we treated streptozotocin-induced diabetic rats with the angiotensin type 1 receptor (AT_1) antagonist ICI D8731 and the ACE inhibitor fosinopril for 4 months and determined the expression of the myocardial glucose transporter proteins. In diabetic rats, the expression of the insulin-regulated glucose transporter (GLUT4) was strongly diminished as shown by Western and Northern blots. ICI D8731 prevented the decrease of GLUT4 protein in diabetes but had no influence on the amount of mRNA encoding for GLUT1 and GLUT4. GLUT1 protein was hardly detected in the rat heart and was affected neither by diabetes nor by treatment with the AT_1 antagonist. Additionally, ICI D8731 influenced the translocation of GLUT4 from the intracellular pool to the plasma membrane, because treatment increased the amount of GLUT4 protein in the plasma membranes as well as in intracellular membrane fractions compared with membranes of untreated diabetic control rats. In contrast, inhibition of ACE by fosinopril influenced neither the expression nor the translocation of the glucose transporter proteins. These observations indicate that angiotensin II has a distinct influence on the post-transcriptional regulation of the GLUT4 transporter protein, presumably indirectly as a consequence of hemodynamic effects and structural alterations of the vessel wall.
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A reduced ability of the myocardium to take up and metabolize glucose has been demonstrated in clinical and experimental studies and seems to be characteristic for insulin-dependent and -independent types of diabetes (1–3). This diminished uptake of glucose has been suggested to contribute largely to the defective myocardial energy provision in diabetes and the reduced ability of the diabetic heart to adapt energy provision to metabolic and functional need (4,5). Furthermore, experimental studies using cardiomyocytes and the isolated perfused heart have shown that the reduced uptake of glucose by myocardium is largely caused by impaired translocation and expression of the insulin-regulated glucose transporter GLUT4 (6–8). Thus alterations in the translocation and expression of GLUT4 might be of specific relevance for the insulin resistance described for heart with respect to glucose. The mechanisms underlying the defective expression of GLUT4 in diabetes have not been fully explored. Changes in the coupling of the insulin receptor to the glucose transporter in specific translocation processes or in the gene expression have been discussed (2,3,8).

On the other hand, it has been observed that angiotensin-converting enzyme (ACE) inhibition can improve glucose uptake in skeletal muscle of patients (9–11) and that the activity of ACE is enhanced in diabetes (12). Furthermore, we were able to show that bradykinin amplifies the effects of insulin on the uptake and conversion of glucose (13). These observations suggest that ACE activity plays an important role in the regulation of glucose uptake and insulin sensitivity. However, the underlying mechanisms are not clear yet. The accelerated uptake and conversion of glucose as well as the increased insulin sensitivity observed in heart and muscle might be due to a direct effect of ACE inhibition on the activity and the expression of the insulin-regulated glucose transporter (GLUT4). The question then arises of whether the improvement is due to increased generation of bradykinin, which has been shown to improve glucose uptake (13). Alternatively, ACE inhibition might inhibit the generation of angiotensin II, which is known to change the gene expression in heart (14,15).

To reveal the mechanisms by which ACE inhibition leads to an improvement of glucose metabolism in the diabetic heart, we treated diabetic rats with the ACE inhibitor fosinopril and the antagonist to the angiotensin type 1 receptor (AT_1) antagonist ICI D8731 and compared the effects of both compounds on the expression of GLUT1 and GLUT4.

RESEARCH DESIGN AND METHODS

Materials. The protease inhibitors phenylmethylsulfonyl fluoride, leupeptide, pepstatin, and benzamidine were purchased from Sigma (Munich, Germany), Immobilon protein-transfer membrane from Millipore (Eschborne, Germany), the bicinchoninic acid protein determination kit from Pierce (Rockford, IL), and aprotinin (Trasylol) from Bayer (Leverkusen, Germany). ^{125}I-protein A was from Amersham-Buchler (Braunschweig, Germany), and the polyclonal antisera to the synthetic 12-amino acid peptides identical to the COOH-terminal ends of rat GLUT1 and GLUT4 glucose transporter proteins were from Calbiochem (La Jolla, CA). The AT_1 antagonist ICI D8731 was a gift from ICI (Macclesfield, England), and the ACE inhibitor fosinopril was a gift from Squibb (Munich, Germany). Intensifying screens and Hyperfilm MP were pur-

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ACE, angiotensin-converting enzyme; CM, crude membrane; PBS, phosphate-buffered saline; PM, plasma membrane; SDS, sodium dodecyl sulfate; SSPE, saline–sodium phosphate–EDTA buffer; STZ, streptozotocin.

chased from Amersham-Buchler. All other reagents were of analytic grade.

Animals and induction of diabetes. Diabetes was induced in male Wistar rats (250–300 g body wt) by an intraperitoneal injection of streptozotocin (STZ) (60 mg/kg body wt) as previously described (13,16). After verification of the diabetic state by determination of blood glucose (hexokinase method) and glucosuria, treatment of animals was started. ICI D8731 and fosinopril were administered in the drinking water at doses of 8 and 10 mg · kg body wt^{-1} · day^{-1}, respectively. The concentrations of both compounds in the drinking water were adjusted according to the daily consumption (12–15 and 90–130 ml/day for control and diabetic rats, respectively). ACE activity could not be detected in the serum of fosinopril-treated animals. The blood pressure was only slightly reduced by both compounds (data not shown). Untreated diabetic animals were used as controls.

After treatment for 4 months, the rats were anesthetized with ether, and the hearts were quickly removed after thoracotomy. Heart tissue was washed in ice-cold phosphate-buffered saline (PBS), frozen in liquid nitrogen, and stored at −70°C. Age-matched healthy rats and untreated diabetic rats were used as nondiabetic or diabetic controls.

Membrane preparation. Heart membranes were isolated by a modification of the method described by Dimitrakoudis et al. (17). In brief, hearts were powdered in liquid nitrogen and homogenized (Ultraturrax; Ika-Werk, Staufen, Germany) (three times for 5 s) in the homogenization buffer (25 mmol/l HEPES, 4 mmol/l EDTA, 250 mmol/l sucrose, 0.2 mmol/l phenylmethylsulfonyl fluoride, 1 mmol/l leupeptin and pepstatin, 25 mmol/l benzamidine, and 1 U/ml aprotinin). After further homogenization (glass-Teflon Potter-Elvehjem), the homogenates were centrifuged at 5,000*g* (6,500 rpm) for 10 min at 4°C (SS34; Sorvall-Dupont, Bad Nauheim, Germany). The supernatants were centrifuged at 208,000*g* for 2 h (4°C, Beckman L5–65, Ti70) to obtain a crude membrane (CM) fraction. The CM fractions were resuspended in the homogenization buffer and fractionated by centrifugation on a discontinuous sucrose gradient (consisting of four layers of sucrose with densities of 48, 35, 30, and 25% wt/wt sucrose; 165,000*g* for 2 h [4°C, Beckman L5–65, SW60]). The various membrane fractions were collected at the interphases, diluted, spun down (208,000*g* for 2 h [4°C, Beckman L5–65, Ti70]), resuspended in the homogenization buffer, aliquoted, frozen in liquid nitrogen, and stored at −70°C until further use.

Protein was determined by a modification of the Lowry method using the bicinchoninic acid protein assay according to the manufacturer's instructions (Pierce). The activity of the Na^+-K^+-ATPase as a marker for plasma membranes was determined by its ability to hydrolyze *p*-nitrophenylphosphate after stimulation by potassium as described by Bers and Langer (18). All samples were run in triplicate. *p*-Nitrophenol (50 μmol/l) was used as standard. We consistently observed a 4- to 16-fold enrichment of specific activity of Na^+-K^+-ATPase in sarcolemmal fractions compared with homogenates. The specific activities and the mean recovery were similar in each subgroup (47–65%). Thus membrane fractionation did not appear to be affected by the diabetic state, although the activity of Na^+-K^+-ATPase in the plasma membrane was lower in diabetic than in control animals.

The following membrane fractions were used for further studies: CM, the total membrane fraction obtained by high-speed centrifugation of heart homogenates as described; plasma membrane (PM), membrane fractions with the highest Na^+-K^+-ATPase activity collected at the interphases between sucrose densities of 8–25% and 25–30% (there was no significant difference in the Na^+-K^+-ATPase activities, so these fractions were pooled); F2, membrane fraction collected at the interphase between sucrose densities of 30–35%; and F3, membrane fraction collected at sucrose densities of 35–48%. Both F2 and F3 contain membrane fractions of the sarcoplasmic reticulum but with different densities.

Western blots. Identical amounts of protein of each membrane fraction were separated on 10% sodium dodecyl sulfate (SDS)-polyacrylamide gels. After transfer onto Immobilon Teflon membranes (semidry blotting device, Bio-Rad, Munich, Germany) and blocking of unspecific binding sites by a solution containing 10% (wt/vol) nonfat dry milk powder in PBS and 0.05% (wt/vol) Tween 20, the proteins were incubated overnight with the antiserum to rat GLUT1 and GLUT4 glucose transporters (1:500 in a solution of 10% milk powder). After washing (five times with PBS/0.05% Tween 20), the membranes were incubated with ^{125}I-protein A (0.3 μCi/mmol) in a 10% solution of milk powder for 4 h. Thereafter, the filters were extensively washed, air dried, and autoradiographed using intensifying screens and Hyperfilm MP (3–7 days at −70°C).

For quantification, sections of the blot corresponding to GLUT4 or GLUT1 bands on autoradiograms were excised, and bound radioactivity was determined with a γ-counter with correction for background radioactivity on the blot. Only lanes loaded with the same amount of protein and from the same blot were used for direct comparison. As a standard for GLUT1 protein, a preparation of rat brain membranes, which are known to contain GLUT1 protein, (19) was used.

Northern blots. Total RNA was isolated from frozen hearts (−80°C) following the cesium chloride method as described by Chirgwin et al. (20). RNA (20 μg/sample) was submitted to formamide-formaldehyde agarose gel electrophoresis (40 mmol/l 3-[*N*-morpholino]propanesulfonic acid, 10 mmol/l Na acetate, and 1 mmol/l EDTA, pH 7.0, overnight) and transferred to Hybond N^+ nylon membrane (Amersham-Buchler) by means of capillary blotting (Northern transfer). After ultraviolet fixation (5 min) and baking (2 h, 80°C), the membranes were prehybridized in 5× saline–sodium phosphate–EDTA buffer (SSPE) (3.6 mol/l saline 20×, 200 mmol/l sodium phosphate, and 20 mmol/l EDTA, pH 7.7), 5× Denhardt's solution, 0.5% SDS, 50% (wt/vol) formamide, and 50 μl freshly denatured sheared salmon sperm DNA for at least 2 h at 42°C (21).

About 50 ng of the 1.5-kilobase fragment of rat GLUT4 cDNA cloned in pBluescript and transfected in *Escherichia coli* strain JM 109 (22) was random primed labeled with [α-^{32}P]dATP (3,000 Ci/mmol, random primed labeling kit, Boehringer Mannheim, Mannheim, Germany). The labeled fragment was separated from unincorporated nucleotides by gel filtration on a Bio-Gel P-30 column (Bio-Rad). The cDNA for GLUT4 was a gift from Dr. M.J. Birnbaum (Boston, MA) (22) and Dr. H.G. Joost (Aachen, Germany).

Hybridization was carried out overnight at 42°C. Membranes were washed twice with 2× SSPE/0.1% (wt/vol) SDS at room temperature and twice with 1× SSPE/0.1% SDS at 50°C, air dried, and subjected to autoradiography. Quantification was done by use of the filmless radioactivity monitoring system FUJIX BAS 1000 (Raytest, Straubenhardt, Germany). Results are given as disintegrations per minute per signal after comparison with a known standard exposed simultaneously.

Statistical analysis. Data are given as means ± SE of five or six hearts per group. Statistical analysis was performed by Student's *t* test.

RESULTS

Injection of STZ induced a severe hyperglycemic state. Only animals with a blood glucose concentration >15 mmol/l were considered diabetic. The diabetic state was further characterized by glucosuria, loss of body weight, and polydipsia. The diabetic animals excreted ketone bodies but were not ketoacidotic. A more extensive characterization of the diabetic state induced by STZ is given by Roesen et al. (16).

As expected, GLUT1 and GLUT4 protein could be detected in the CM fractions of hearts of nondiabetic and diabetic control rats. The molecular masses for GLUT1 and GLUT4 identified by the two specific polyclonal antisera were both in the range of 43–45 kDa. The concentration of GLUT4, however, was much higher than that of GLUT1. STZ-induced diabetes (4 months) did not change the expression of GLUT1. There was no significant difference in the amount of GLUT1 protein between hearts of diabetic and nondiabetic control rats. Treatment of the diabetic animals with the ACE inhibitor fosinopril did not cause any significant alteration in the amount of GLUT1 protein as detected by Western blots (data not shown).

In contrast with GLUT1, the amount of GLUT4 was reduced by ~30% in diabetic rats (Fig. 1 and Table 1). This difference between nondiabetic control and diabetic control animals was more pronounced in the PM fraction. A similar difference can be seen in the amount of GLUT4 in the high-density membranes of the sarcoplasmic reticulum. Thus GLUT4 protein is generally reduced in the heart of diabetic rats, in plasma membranes, and in intracellular membranes. Treatment of the diabetic animals with fosinopril did not prevent the diabetes-induced decrease in GLUT4 protein in PM or intracellular membrane fractions (Fig. 1 and Table 1).

Treatment of diabetic animals with the AT_1 antagonist ICI

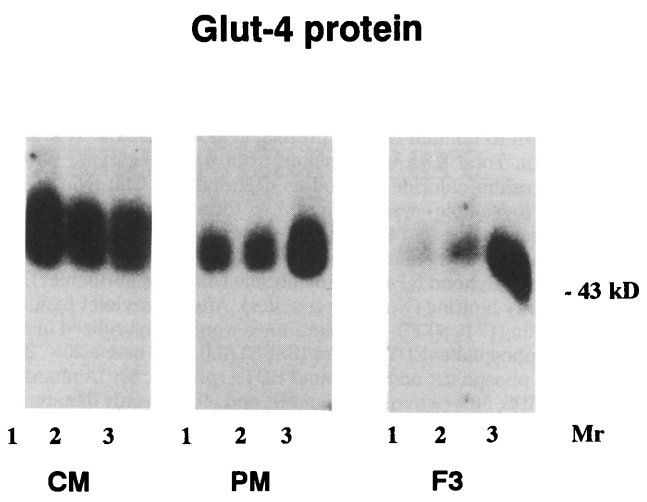

FIG. 1. Influence of STZ-induced diabetes and the ACE inhibitor fosinopril on the expression of GLUT4 protein in myocardial membranes. Equal amounts of protein (50 μg/lane) were subjected to immunoblot analysis using polyclonal antisera against GLUT1 and GLUT4 as described in METHODS. CM: *lane 1*, nondiabetic controls; *lane 2*, diabetic controls; *lane 3*, diabetic rats treated with fosinopril. PM: *lane 1*, diabetic rats treated with fosinopril; *lane 2*, diabetic controls; *lane 3*, nondiabetic controls. F3: *lane 1*, diabetic rats treated with fosinopril; *lane 2*, diabetic controls; *lane 3*, nondiabetic controls.

D8731 had no influence on the amount of GLUT1 protein in control or diabetic animals. However, it prevented the diabetes-induced reduction of GLUT4 almost completely (Fig. 2 and Table 1). Treatment with ICI D8731 increased the amount of GLUT4 not only in the CM fraction but also in the PM and the intracellular membrane fractions (F2 and F3) (Fig. 2*B*). These findings indicate that the total amount of myocardial GLUT4 is increased by treatment of diabetic rats with ICI D8731. Additionally, treatment with ICI D8731 influenced the distribution of GLUT4 protein in hearts of diabetic rats (Fig. 3). There is a significant shift of GLUT4 from the intracellular pool (F2 and F3) into the PM fraction. After treatment with ICI D8731, ~50% of the total GLUT4 protein was detected in the PM fraction compared with ~33% in diabetic controls.

Northern blot analysis (Fig. 4) revealed that the steady-state level of GLUT4 mRNA is reduced in diabetes by ~30–40%. Treatment with the AT$_1$ antagonist did not prevent the diabetes-induced reduction of GLUT4 mRNA. The amount of GLUT1 mRNA was not affected by the induction of diabetes or by treatment of the animals with the AT$_1$ antagonist (data not shown).

DISCUSSION

The present study was designed to study the mechanisms underlying the improvement of glucose uptake and metabolism by inhibition of ACE in the diabetic heart. We assumed that the activity of the glucose transporter is affected by ACE inhibition either by increased generation of bradykinin, which had previously been shown to stimulate the uptake and the conversion of glucose in the diabetic rat heart (13), or by an inhibition of angiotensin II formation. To test this hypothesis, diabetic rats were treated with either the ACE inhibitor fosinopril or the AT$_1$ antagonist ICI D8731. Inhibition of ACE has two effects, reduced formation of angiotensin II and an increase in bradykinin, whereas the AT$_1$ antagonist specifically inhibits the actions of angiotensin II on heart tissue.

Induction of diabetes caused a reduction in the total amount of myocardial GLUT4 protein by ~30%. A similar diminution of GLUT4 had already been observed in STZ-induced diabetic rats by others (23,24). The data of Garvey et al. (23) suggested that the reduction of GLUT4 expression and the impairment of glucose uptake are strongly related in hearts of STZ-induced diabetic rats. Thus, in myocardium, a

TABLE 1
Influence of diabetes and treatment with fosinopril or the AT1 antagonist ICI D8731 on the expression of GLUT4 in membranes of control and STZ-induced diabetic rats

	GLUT4 (%)	*P* value
Nondiabetic control	100	
Diabetic control	72 ± 8	<0.05*
Diabetic treated with fosinopril	74 ± 6	<0.05*
Diabetic treated with AT1 antagonist	88 ± 6	<0.05†

*Compared with nondiabetic controls. †Compared with diabetic controls.

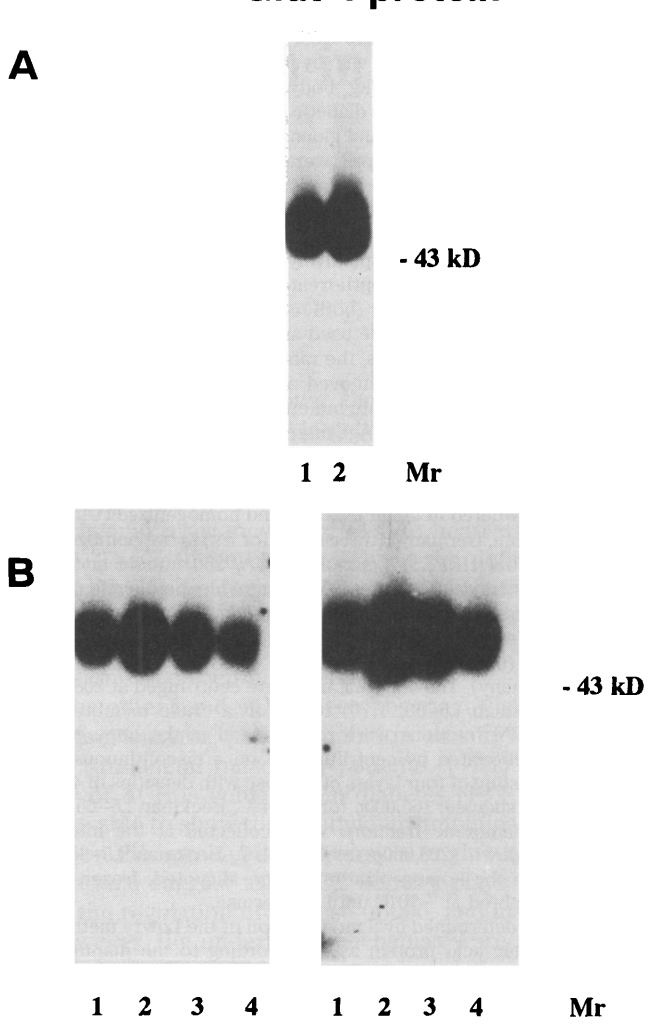

FIG. 2. Influence of the AT$_1$ antagonist ICI D8731 on the expression of GLUT4 protein in myocardial membranes from diabetic rats. Equal amounts of protein (30 μg/lane) were subjected to immunoblot analysis using polyclonal antisera against GLUT1 and GLUT4 as described in METHODS. *A: lane 1*, CM of diabetic controls; *lane 2*, CM of hearts of diabetic rats treated with ICI D8731. *B: lane 1*, CM; *lane 2*, PM; *lane 3*, high-density membrane fraction (F2); lane 4, high-density membrane fraction (F3). Left, diabetic controls; right, diabetic rats treated with ICI D8731.

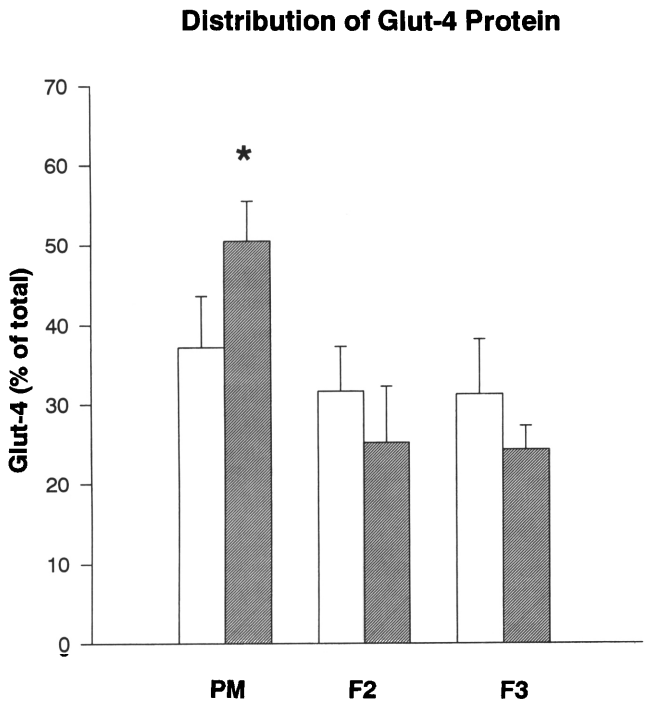

FIG. 3. Influence of the AT_1 antagonist ICI D8731 on the distribution of GLUT4 protein in cardiac membranes of diabetic rats. Cardiac membranes were isolated from hearts of treated (▨) and untreated (□) diabetic rats as described in METHODS. The highest activity of Na^+-K^+-ATPase was found in the PM fraction. The fractionation procedure was not influenced by diabetes. GLUT4 protein was identified using a polyclonal antibody specific for GLUT4 protein and ^{125}I-protein A after polyacrylamide gel electrophoresis and Western blotting. The percentage of GLUT4 is given as the mean ± SE of four different membrane preparations. The total amount of GLUT4 in the isolated membrane fractions was taken as 100%. *$P < 0.05$.

defect in the expression of GLUT4 might contribute to insulin resistance and the disturbed uptake and conversion of glucose by myocardium. The reduced expression of the insulin-regulated glucose transport protein could be fully prevented by treatment of the diabetic animals with insulin (23). The finding that both GLUT4 protein and mRNA were reduced to a nearly similar extent suggests that a pretranslational event is responsible for the depletion of the cellular membranes of GLUT4 transporter protein. Similar results were also observed in adipose tissue (1), whereas in skeletal muscle conflicting results have been reported (25), indicating a tissue-specific regulation of GLUT4.

In contrast to GLUT4, the amount of GLUT1 was very low in myocardium. It was not affected by diabetes, ACE inhibition, or AT_1 antagonism. In agreement with other data (23), GLUT1 seems therefore to play only a minor role in the provision of the heart with glucose.

Treatment with the AT_1 antagonist prevented the diminution of GLUT4 protein in diabetic rats but not the reduction of GLUT4 mRNA. If the steady-state levels of GLUT4 mRNA are taken as a parameter for expression of the GLUT4 gene, the AT_1 antagonist did not exert any effect on the activity of this gene. Rather, our data suggest that angiotensin II modulates the stability of the GLUT4 mRNA or the translation of the GLUT4 mRNA, resulting in enhanced formation of this protein, but not the transcription rate. Moreover, treatment of the diabetic rats with ICI D8731 seems to favor the translocation of GLUT4 protein from an intracellular membrane compartment to the PM fraction. Thus our data indicate that activation of the angiotensin II receptor is involved in the regulation of both the expression and translocation of GLUT4 protein.

The mechanisms underlying the effects of angiotensin II and other kinins on the impaired expression of GLUT4 in diabetic hearts are not quite clear. Our data suggest that, at least in the rat heart, increased formation of angiotensin II plays a decisive role in impairment of glucose uptake in diabetes, but presently no data demonstrate that angiotensin II directly inhibits the uptake of glucose by the myocardium. Instead, in vascular smooth cells, angiotensin II has been shown to activate protein kinase C and thereby to cause an increase in GLUT1 (26). Similarly, activation of protein kinase C by angiotensin II results in stimulation of the insulin-dependent glucose uptake in other tissues (27,28). Thus at this time we have no direct explanation for the prevention of the diabetes-induced impairment of GLUT4 expression by AT_1 antagonism.

Because the diabetic animals in this study were treated for 4 months, the changes observed by treatment with the AT_1 antagonist might represent an indirect rather than a direct long-term effect of angiotensin II inhibition. There is evidence that treatment with the AT_1 antagonist protects the vasculature and prevents severe disturbances of the myocardial nerve tissue observed in diabetes (29). Furthermore, several studies have shown that angiotensin II increases the sympathoadrenal activity by enhancing norepinephrine release (30,31) and that increased sympathoadrenal activity can lead to insulin resistance and to inhibition of glucose uptake (32). Therefore it is intriguing to speculate that increased mobilization of sympathetic neurotransmitters by angiotensin II action is responsible for the inhibition of

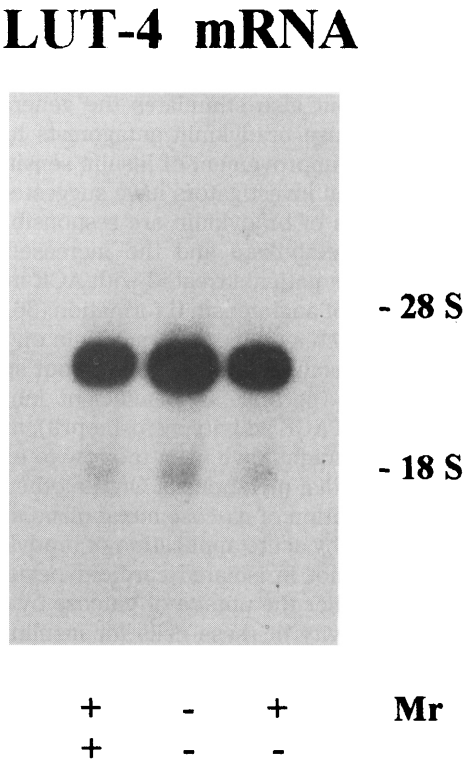

FIG. 4. Northern blot analysis of GLUT4 mRNA. A total of 20 μg of total RNA was subjected to Northern blot analysis and detected with a labeled fragment of rat GLUT4 cDNA. *Lane 1*, diabetic rats treated with AT_1 antagonist ICI D 8731; *lane 2*, nondiabetic controls; *lane 3*, diabetic controls.

glucose uptake and reduction of GLUT4 protein in the diabetic rat heart. Conversely, because AT_1 antagonists are able to reduce the increased sympathetic activity and to prevent the dramatic depletion of catecholamine stores in the diabetic heart (29), it is intriguing to suggest that AT_1 antagonism can counteract the catecholamine-induced disturbances of glucose transport in diabetes and thereby improve myocardial glucose metabolism.

In the rat heart, we recently demonstrated that both local perfusion and diffusion properties are severely impaired in diabetes (33) and that these changes can largely be prevented by treatment of diabetic rats with the AT_1 antagonist ICI D8731 (29). Furthermore, we know that the local perfusion and capillary density are determinants of insulin resistance and glucose uptake (34). Thus the augmented expression of GLUT4 in the diabetic heart by AT_1 antagonism might be a consequence of the improvement in nutritional flow to facilitate the uptake and conversion of glucose.

In contrast with the AT_1 antagonist, we did not observe any effect of ACE inhibition by fosinopril on the expression of GLUT4. Treatment with fosinopril did not consistently prevent the loss of GLUT4 glucose transport protein. How can we explain this unexpected observation? We assume that the systemic ACE activity was inhibited by the application of fosinopril, because the dose of fosinopril was effective enough to prevent myocardial fibrosis and to affect the coronary perfusion (35). Additionally, the ACE activity in the serum was totally inhibited under these conditions. However, these observations do not exclude the possibility that, specifically in myocardium, some angiotensin II is still generated by the local ACE system, the chymase activity recently described (14). It is suggested that the local formation of angiotensin II prevents the effect of systemic ACE inhibition on the expression of GLUT4 observed by AT_1 antagonism.

ACE inhibition not only inhibits the generation of angiotensin II but also stimulates the generation of bradykinin (14). Because bradykinin antagonists have been shown to nullify the improvement of insulin sensitivity by ACE inhibition, several investigators have suggested that elevated concentrations of bradykinin are responsible for the improved glucose metabolism and the increased insulin sensitivity observed in patients treated with ACE inhibitors but not the inhibition of angiotensin II formation (36,37). Our data do not exclude such an effect of bradykinin on myocardial metabolism and insulin sensitivity. Under our specific experimental conditions (because of insufficient inhibition of the local myocardial ACE activity by fosinopril), the rate of bradykinin formation might have been too low to exert a specific effect on myocardial metabolism. On the other hand, we observed an acceleration of glucose metabolism and increased insulin sensitivity by acute application of bradykinin in the perfused heart but not in isolated cardiomyocytes. Bradykinin influenced neither the uptake of glucose by cardiomyocytes nor the sensitivity of these cells for insulin (13). Therefore we conclude that the metabolic effects of bradykinin are mediated by changes in coronary perfusion and in vascular permeability but not by a direct effect on the expression of myocardial glucose transporters studied (13). Recent studies using specific bradykinin antagonists support this conclusion (37).

In summary, our data demonstrate that inhibition of angiotensin II action prevents the decrease in GLUT4 protein expression induced by diabetes. These metabolic effects of the AT_1 antagonist might be of specific importance in pathophysiological states such as diabetes and the ischemic heart, in which myocardial metabolism prefers glucose as a substrate for energy metabolism instead of fatty acids (5) but in which less glucose is available because of the depletion of GLUT4 glucose transporter.

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