Gliclazide Decreases Cell-Mediated Low-Density Lipoprotein (LDL) Oxidation and Reduces Monocyte Adhesion to Endothelial Cells Induced by Oxidatively Modified LDL

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Low-density lipoprotein (LDL) oxidation has been suggested to play a key role in the pathogenesis of atherosclerosis, a major complication of diabetes mellitus. Gliclazide, a second-generation sulfonylurea, is widely used in the treatment of type II diabetes mellitus. Recently, a free-radical–scavenging activity of gliclazide has been reported. In the present study, we examined the effects of gliclazide on cell-mediated LDL oxidation and monocyte adhesion to endothelial cells induced by oxidatively modified LDL. Incubation of human monocytes and bovine aortic endothelial cells (BAE cells) with increasing concentrations of gliclazide (0 to 10 μg/mL) and native LDL (100 μg/mL) resulted in a dose-dependent diminution of cell-mediated LDL oxidation as assayed by measurement of thiobarbituric acid (TBA)-reactive substances (TBARS). In addition, exposure of BAE cells to gliclazide (0 to 10 μg/mL) and native LDL (100 μg/mL) induced a dose-dependent diminution of the oxidized LDL–induced monocyte adhesion to BAE cells as measured by the myeloperoxidase (MPO) assay. The effects of glyburide, another second-generation sulfonylurea, were also tested on cell-mediated oxidation of LDL and LDL-induced monocyte adhesion to the endothelium. No significant effect of this drug was observed on these two processes. These results therefore demonstrate that gliclazide is effective in vitro in reducing both cell-mediated LDL oxidation and monocyte adhesion to the endothelium. These findings suggest a potential beneficial effect of gliclazide in the prevention of atherosclerosis in diabetic patients.

ATHEROSCLEROSIS occurs prematurely in diabetic individuals[1] and is considered the major complication of diabetes mellitus.[2] Diabetic patients show increased levels of circulating modified lipoproteins[3] and enhanced oxidation of plasma low-density lipoproteins (LDLs).[4] Increased production of malondialdehyde (MDA), a marker of lipid peroxidation, has also been found in erythrocyte membranes of diabetic patients.[5] Moreover, circulating levels of MDA are higher in the plasma of diabetic patients compared with control subjects.[6] LDL oxidation has been suggested to play a key role in atherogenesis associated with diabetes mellitus.[7] The proatherogenic properties of oxidized LDL are likely to result from its biological functions. LDL, once oxidized, is avidly taken up by macrophages via the scavenger receptor, leading to formation of foam cells in vitro.[8-10] Oxidatively modified LDL is also toxic to endothelial cells[11] and alters both growth factor[12] and cytokine[13] production. Finally, it is chemotactic to human monocytes,[14-15] inducing monocyte recruitment and adhesion to the endothelium in vitro[16] and promoting leukocyte-endothelial interactions in vivo.[17] Enhanced binding of monocytes to the vasculature has been documented in diabetes.[18] Since adherence of monocytes to the endothelium is one of the earliest events in the development of atherosclerosis, such alteration may represent one of the mechanisms leading to accelerated atherosclerosis in diabetic patients.

Gliclazide, a second-generation sulfonylurea, is widely used in the treatment of type II diabetic patients. Beside its metabolic effects,[19] gliclazide possesses some nonmetabolic effects specifically related to vascular disease in diabetes. Among them, a free-radical–scavenging activity of gliclazide has been recently documented.[20] Based on this observation, we anticipated that gliclazide may reduce vascular cell-mediated LDL oxidation and oxidized LDL–induced monocyte adhesion to endothelial cells. Our results demonstrate that gliclazide effectively reduces in vitro both cell-mediated LDL oxidation and monocyte adhesion to endothelium. These data suggest that treatment of diabetic patients with gliclazide may prevent or retard the development of atherosclerosis.

MATERIALS AND METHODS

Reagents

Dulbecco's minimal essential medium (DMEM) and L-glutamine were obtained from ICN Biochemicals (Costa Mesa, CA). RPMI 1640 was purchased from GIBCO (Grand Island, NY). Penicillin-streptomycin and fetal calf serum (FCS) were obtained from Flow Laboratories (McLean, VA) and Hyclone Laboratories (Logan, UT). Thiobarbituric acid (TBA) and tetraethoxypropane (TEP) were purchased from ICN Biochemicals. Dianisidine dihydrochloride and hexadecyltrimethylamine ammonium bromide (HTAB) were from Sigma Chemicals (St Louis, MO). Phosphoric acid and butanol were obtained from Fisher Scientific (Nepean, Montreal, Canada). Gliclazide and glyburide were kindly provided by Les Laboratoires Servier (Neuilly, France) and Hoechst (Canada), respectively.

Endothelial Cell Culture

Subcultured (17th passage) bovine aortic endothelial (BAE) cells were kindly provided by Dr R. Sauvé (Montreal University, Montreal, Quebec, Canada). Cells were plated in tissue culture flasks at 37°C in 95% CO_2/95% air atmosphere and grown in DMEM supplemented with 10% heat-inactivated FCS, 100 U/mL penicillin, 100 μg/mL streptomycin, and 2 mmol/L glutamine. After 6 days, cells were trypsinized and cultured for an additional 48 hours in 96 multiwells (Costar), at which time cell confluence was reached. In all experiments, cells were used between passages 18 and 23.

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Human Monocyte Isolation

Fresh heparinized blood (100 mL) was obtained from healthy, nonsmoking male and female donors. Peripheral blood mononuclear cells were isolated by density centrifugation using Ficoll (Pharmacia, Uppsala, Sweden),[21] allowed to aggregate in the presence of FCS, and then further purified by the rosetting technique. After density centrifugation, recovery of highly purified monocytes (85% to 90%) as assessed by FACS analysis was obtained. Monocytes were resuspended in serum-free RPMI 1640 medium with 2 mmol/L glutamine supplemented with 100 U/mL penicillin and 100 μg/mL streptomycin and used immediately.

Human LDL Isolation

LDL was isolated from plasma obtained from healthy normolipidemic human subjects. Venous blood was drawn into tubes containing EDTA, and LDL was isolated at a density of 1.019 to 1.063 after sequential centrifugation in KBr according to the method of Hatch.[22] LDL was extensively dialyzed for 24 hours at 4°C against 5 mmol/L Tris/50 mmol/L NaCl to remove EDTA, stored at 4°C, and used within 2 days at a final concentration of 100 μg LDL protein/mL. Protein content in LDL preparations was measured according to the Bradford method.[23]

Oxidative Modification of LDL

Cell-free system. Oxidation of LDL was performed by incubating native LDL (100 μg protein/mL) at 37°C for 20 hours in serum-free RPMI 1640 containing 5 μmol/L $CuSO_4$. Incubation was performed in the presence of increasing concentrations of gliclazide (0 to 10 μg/mL) or glyburide (0 to 5 μg/mL). These concentrations were chosen because they are in the range of plasma levels in diabetic patients treated with these drugs.

Cell system. Cellular modification of LDL was obtained by incubating 100 μg LDL protein for 20 hours at 37°C in the presence of human monocytes or BAE cells in serum-free RPMI 1640 containing 3 μmol/L $CuSO_4$. To assess the effect of gliclazide or glyburide on cell-mediated LDL oxidation, cells were pretreated for 1 hour with increasing concentrations of gliclazide (0 to 10 μg/mL) or glyburide (0 to 5 μg/mL) before addition of LDL. At the end of the incubation period, supernatants were removed and EDTA (0.04% final concentration) was added to stop LDL oxidation.

Determination of LDL Oxidation

TBA-reactive substances assay. The lipid peroxide content of oxidized LDL was determined by measuring, in the supernatant, TBA-reactive substances (TBARS) expressed as MDA equivalents (nmol/500 μL medium).[24-25] Samples (500 μL) were mixed with 3 mL phosphoric acid (1%) and 1 mL TBA (0.6%). The mixture was heated to 95°C for 45 minutes in a hot water bath. After cooling, the MDA-TBA complex was extracted by adding butanol, and the optical density was read at 532 nm. Freshly diluted TEP, which produces MDA after hydrolysis, was used as a standard.

Electrophoretic mobility. To assess in the cell-free system the electrophoretic mobility of copper ion–treated LDL, agarose gel electrophoresis of this lipoprotein was performed using the commercial paragon system (Beckman, Brea, CA).

Adhesion Assay

The day of the experiment, confluent monolayers of BAE cells were incubated in serum-free RPMI 1640 supplemented with 3 μmol/L $CuSO_4$ and treated with LDL (100 μg protein/mL) for 20 hours at 37°C. At the end of the incubation period, supernatants were removed and fresh serum-free RPMI 1640 medium was added. One hundred microliters of a monocytic cell suspension (2.8×10^6 cells/mL) was then added to each well. After a 30-minute incubation period, nonadherent monocytes were removed by washing twice with phosphate buffered saline without calcium or magnesium (PBS-A). Adherent cells were lysed in 50 μL HTAB (0.5%) in PBS-A at pH 6.0 for 30 minutes. Quantification of adherent monocytes was made by measuring monocyte myeloperoxidase (MPO) activity.[26] Briefly, MPO activity was determined by addition to each well of 250 μL dianisidine dihydrochloride (0.2 mg/mL in PBS-A) warmed at 37°C and mixed with hydrogen peroxide (0.4 mmol/L final concentration). After 2 to 5 minutes of incubation, the optical density of the plate wells was read at 450 nm using a Titertek multiscan spectrophotometer (Flow Laboratories). To test the effect of gliclazide or glyburide on monocyte adhesion to endothelium, BAE cells were pretreated for 1 hour with increasing concentrations of gliclazide (0 to 10 μg/mL) or glyburide (0 to 5 μg/mL).

Determination of Cell Viability

To evaluate the cellular toxicity of gliclazide and glyburide at the maximal concentrations used, cell viability was estimated using trypan blue exclusion and consistently found to be higher than 95%.

Statistical Analysis

Statistical analysis of the results was performed by one-way ANOVA followed by Tukey's test. Results are expressed as the mean ± SEM.

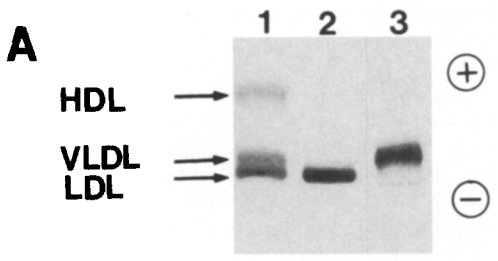
RESULTS

Effect of Gliclazide on the Oxidative Modification of LDL Induced by Incubation With Copper Ions in a Cell-Free System

Incubation of native LDL (100 μg protein/mL) with Cu^{2+} at a concentration of 5 μmol/L for 20 hours at 37°C resulted in oxidative modification of LDL, as reflected by the higher increase in net negative charge of Cu^{2+}-LDL compared with native LDL (Fig 1A) and by the significant increase in TBARS content of the incubation medium (Table 1). Although the TBARS assay is an indirect measure of lipid peroxidation, it is a convenient and widely used method to assess LDL oxidation in both cell-free and cell-mediated systems. Addition of gliclazide in the concentration range of 2.5 to 10 μg/mL decreased the formation of TBARS in a dose-dependent manner. The maximal inhibitory effect was observed with a concentration of 10 μg/mL gliclazide (Fig 1B). At this concentration, TBARS content decreased by 18%. In contrast, addition of increasing concentrations of glyburide (0.05 to 5 μg/mL) under similar experimental conditions did not reduce TBARS formation in the medium (Table 2). Since gliclazide and glyburide were dissolved in dichloromethane, the effects of this vehicle alone were also tested. Incubation of LDL in the presence of 20 μg/mL dichloromethane did not affect the degree of LDL oxidation mediated by copper ions (Fig 1B).

Effect of Gliclazide on the Oxidative Modification of LDL Mediated by Human Monocytes

Incubation of native human LDL (100 μg protein/mL) in medium containing 3 μmol/L Cu^{2+} in the presence of freshly isolated human monocytes for 20 hours at 37°C significantly enhanced oxidative modification of LDL, as measured by lipid peroxidation products in the supernatants (Table 1). Medium TBARS content in the presence of monocytes was 4.9 ± 0.2 MDA equivalents/500 μL medium, whereas TBARS production in the absence of cells was 2.3 ± 0.3 MDA equivalents/500 μL medium ($P < .005$). To evaluate the effects of gliclazide or



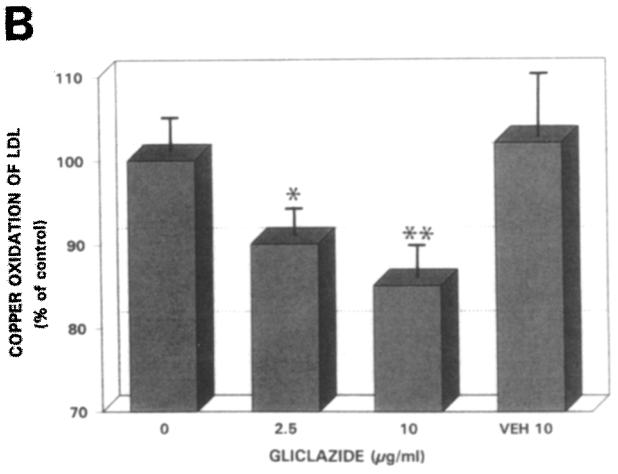


Fig 1. (A) Electrophoretic mobility of human normolipidemic plasma (lane 1), native LDL (lane 2), and copper ion–treated LDL (lane 3). Lipoproteins were separated in a 0.5% agarose gel and stained with Sudan Black. (B) Gliclazide dose-dependently decreases LDL oxidation in a cell-free system. Native LDLs (100 μg/mL) were incubated for 20 hours in RPMI containing 5 μmol/L $CuSO_4$ with increasing concentrations of gliclazide (0 to 10 μg/mL). At the end of the incubation period, TBARS content was measured in the medium. Results are expressed as % TBARS content observed in the absence of gliclazide. Data represent the mean ± SEM of 6 different experiments. *$P < .05$ and **$P < .02$ *v* control.

glyburide on the degree of LDL oxidation mediated by human monocytes, these cells were incubated for 1 hour in the presence of increasing concentrations (1 to 10 μg/mL) of gliclazide or glyburide (0.05 to 5 μg/mL) before addition of native LDL. Addition of gliclazide resulted in a dose-dependent inhibition of the degree of monocyte-induced LDL oxidation (Fig 2). The maximal decrease was about 20% and was observed at a concentration of 10 μg/mL of this drug. In contrast, glyburide had no significant effect on monocyte-induced LDL oxidation (Table 2). Dichloromethane (10 μg/mL) did not significantly affect monocyte-mediated LDL oxidation (data not shown).

Table 2. Effect of Glyburide on the Generation of TBARS by Copper Ions, Monocytes, and BAE Cells

Glyburide (μg/mL)	5 μmol/L $CuSO_4$ (without cells)	Monocytes + 3 μmol/L $CuSO_4$	BAE Cells + 3 μmol/L $CuSO_4$
0	100 ± 4.9	100 ± 2.5	100 ± 4.4
0.05	94 ± 0.6	101 ± 4.2	115 ± 5.8
0.13	97 ± 2.9	104 ± 1.8	105 ± 3.8
0.25	95 ± 1.7	96 ± 8.6	103 ± 3.5
0.5	93 ± 1.5	104 ± 2.6	103 ± 1.4
5	96 ± 0.9	96 ± 4.3	112 ± 6.9

NOTE. Data represent the mean ± SEM of 3 experiments.

Effect of Gliclazide on the Oxidative Modification of LDL Mediated by BAE Cells

Endothelial cells have the capacity to oxidize LDL in a way similar to that of human monocytes. Incubation of BAE cells with LDL (100 μg protein/mL) for 20 hours led to a significant increase in the medium TBARS content (3.9 ± 0.2 MDA equivalents/500 μL medium) as compared with that observed in the absence of cells (1.8 ± 0.3 MDA equivalents/500 μL medium, $P < .005$; Table 1). Preincubation of endothelial cells with gliclazide (1 to 10 μg/mL) for 1 hour before addition of native LDL resulted in a significant decrease of BAE cell–mediated oxidation of LDL (Fig 3). Inhibition of LDL oxidation by gliclazide was dose-dependent, and the maximal effect (18% decrease) was observed at 10 μg/mL of this drug. Preincubation of BAE cells with glyburide (0.05 to 5 μg/mL) was ineffective in reducing LDL oxidation in the presence of BAE cells (Table 2). As already reported, no effect of dichloromethane on BAE cell–mediated LDL oxidation was observed (data not shown).

Effect of Gliclazide on Monocyte Adhesion to BAE Cells

Preincubation of BAE cells with native LDL (100 μg protein/mL) in the presence of 3 μmol/L Cu^{2+} for 20 hours at 37°C resulted in a 2.7-fold increase in the number of monocytes adhering to the endothelium as assessed by the MPO assay (Table 3). In the presence of LPS (10 ng/mL), used as a positive control, monocyte adhesion to BAE cells was enhanced 2.3-fold (Table 3). Pretreatment of BAE cells with gliclazide (1 to 10 μg/mL) for 1 hour dramatically decreased the ability of oxidized LDL to stimulate BAE cell adhesiveness (Fig 4). Gliclazide-induced inhibition of monocyte adhesion to the endothelium was dose-dependent, and at the highest concentration of gliclazide (10 μg/mL) monocyte adherence to endothelial cells was decreased 43% (Fig 4). Pretreatment of BAE cells with glyburide (0.5 μg/mL) did not affect the LDL-induced enhancement of monocyte adhesion to the endothelium (data not shown). Finally, no effect of dichloromethane on monocyte adhesion to endothelial cells was observed (data not shown).

Table 1. Generation of TBARS by Copper Ions, Monocytes, and BAE Cells

	Copper Ions		Monocytes		BAE Cells	
	−	+	−	+	−	+
TBARS (nmol/500 μL)	0.5 ± 0.1	2.4 ± 0.1*	2.3 ± 0.3	4.9 ± 0.2*	1.8 ± 0.3	3.9 ± 0.2*

NOTE. Data represent the mean ± SEM of 7 different experiments.
*$P < .005$.

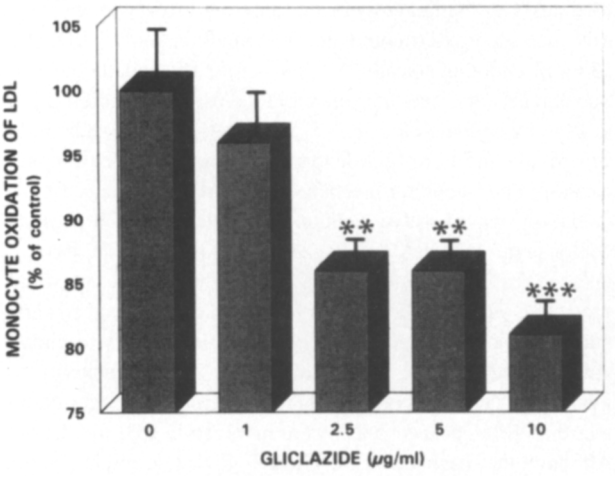


Fig 2. Gliclazide dose-dependently decreases LDL oxidation mediated by human monocytes. Native LDLs (100 μg/mL) were incubated for 20 hours with human monocytes in RPMI containing 3 μmol/L $CuSO_4$ in the presence of increasing concentrations of gliclazide (0 to 10 μg/mL). At the end of the incubation period, TBARS content was measured in the medium. Results are expressed as % TBARS content observed in the absence of gliclazide. Data represent the mean ± SEM of 7 different experiments. **$P < .02$ and ***$P < .005$ *v* control.

DISCUSSION

Our study indicates that gliclazide is clearly an antioxidant and may also have other effects unrelated to its hypoglycemic effects. Indeed, gliclazide inhibits LDL oxidation induced by incubation with copper ions, monocytes, or endothelial cells. Our observation that gliclazide inhibits LDL oxidation by copper in an in vitro system is in accordance with the finding of Luo and O'Brien,[27] who recently reported that in vitro resistance of LDL to copper oxidation is increased in the presence of gliclazide. These results suggest that the antioxidant activity of

Table 3. Monocyte Adhesion to BAE Cells

Treatment	Adhesion (% over basal values)
Control (without LDL)	100 ± 10
LDL + 3 μmol/L $CuSO_4$	280 ± 20*
LPS (10 ng)	230 ± 20*

NOTE. Data represent the mean ± SEM of 6 experiments.
*$P < .01$ *v* control.

gliclazide may also be manifest in the plasma compartment and/or in atherosclerotic plaques where catalytically active copper ions are present. In a cell-free system, Cu^{2+} promotes free radical–mediated LDL oxidation by generating hydroxy radicals such as peroxyl or alkolyl radicals.[28] The reduced Cu^{2+}-induced oxidation of LDL in the presence of gliclazide may suggest that this drug exerts its protective effect by metal chelation or, alternatively, by scavenging free radicals.

Previous in vitro studies have demonstrated that gliclazide is a general free radical scavenger.[29] It has been suggested that the azabicyclo-octyl ring present in this sulfonylurea may be responsible for this property. This hypothesis is supported by our observation that glyburide which does not present this particular chemical structure is ineffective to reduce LDL oxidation. Free radical scavengers have been shown to inhibit the propagation of lipid peroxidation by breaking the chain of oxidative reactions. Such a mechanism may account for the antioxidant action of vitamin E, which inactivates peroxyl radicals, and of probucol, which seems to prevent LDL oxidation by trapping hydroxyl radicals.[30,31] A protective effect of LDL against oxidative modification has also been reported in the presence of several other drugs such as paracetamol[32] and dipyridamole.[33] Our results demonstrate that gliclazide inhibits Cu^{2+}-induced oxidation of LDL at 20 hours with the same order of potency as these drugs, suggesting that similar mechanisms

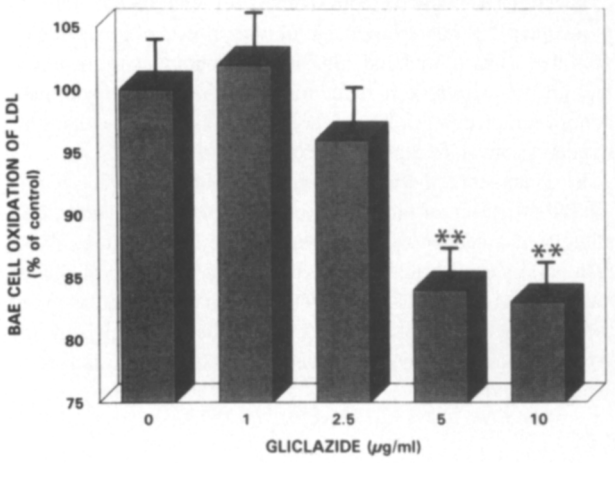


Fig 3. Gliclazide dose-dependently decreases LDL oxidation mediated by BAE cells. Native LDLs (100 μg/mL) were incubated for 20 hours with BAE cells in RPMI containing 3 μmol/L $CuSO_4$ in the presence of increasing concentrations of gliclazide (0 to 10 μg/mL). At the end of the incubation period, TBARS content was measured in the medium. Results are expressed as % TBARS content observed in the absence of gliclazide. Data represent the mean ± SEM of 7 different experiments. **$P < .02$ *v* control.

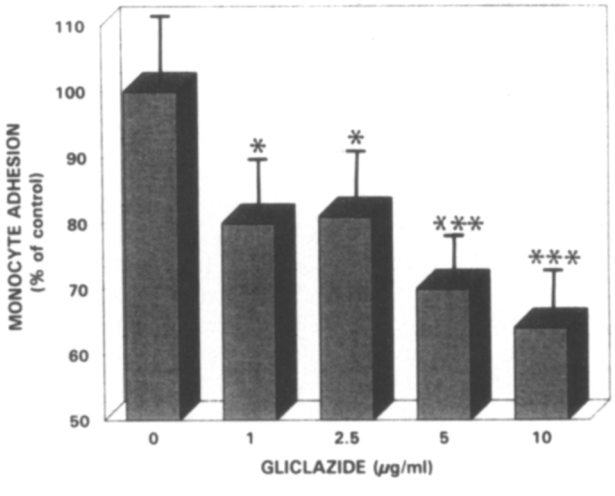


Fig 4. Gliclazide dose-dependently decreases monocyte adhesion to BAE cells induced by oxidized LDL. Endothelial cells were treated for 20 hours with native LDL (100 μg/mL) in the presence of increasing concentrations of gliclazide (0 to 10 μg/mL) Monocyte adhesion to endothelial cells was measured by the MPO assay. Results are expressed as a % of control. Data represent the mean ± SEM of 7 different experiments. *$P < .05$, **$P < .01$, and ***$P < .005$ *v* control.

are used by these drugs to reduce metal ion–catalyzed free radical reactions.

In the case of monocyte-induced modification of LDL, initiation of the oxidative process has been shown to be related, at least in some systems, to secretion of the superoxide anion.[34-36] In contrast, the subsequent propagation and amplification of the oxidative reactions have been reported to involve a superoxide-independent pathway.[34-36] A role for cellular lipoxygenases in LDL modification by monocytes and macrophages has also been postulated.[37,38] In vitro, respiratory burst–dependent generation of superoxide is induced in activated monocytes and macrophages by specific particulate and nonparticulate agents.[39,40] Although incubation of LDL with activated human monocytes has been shown to result in dramatic LDL oxidation, no significant oxidation was reported in the supernatants of unactivated mononuclear cells incubated with LDL.[34-36] These observations are at variance with our results, which demonstrate a twofold increase in the concentration of lipid peroxidation products in culture supernatants of resting human monocytes incubated with LDL as compared with that observed in cell-free medium. Although the reasons for such a discrepancy are unclear, activation of the cells resulting from isolation procedures used in the present study does not seem to account for such an effect. Indeed, basal production of superoxide or tumor necrosis factor-alpha by these cells was consistently found to be undetectable or very low (data not shown). Considering the controversy about the participation of the superoxide anion in human monocyte/macrophage-mediated oxidation of LDL,[41-43] it may be hypothesized that under our experimental conditions LDL oxidation does not require superoxide. Recent reports have proposed that these cells enhance LDL oxidation by enhancing the redox reaction of metal ions present in the medium.[44-46] The similar effectiveness of gliclazide to reduce LDL oxidation mediated by copper ions or monocytes suggests that most of the inhibitory effects of gliclazide we observed on monocyte-mediated LDL oxidation may be related to its ability to decrease metal-catalyzed propagation reactions. Alternatively, since 15-lipoxygenase may be responsible for cell-mediated oxidation of LDL, gliclazide may reduce LDL oxidation by reducing lipoxygenase activity.

Although aortic endothelial cells derived from different species have been found to modify LDL,[47-50] several studies have suggested that BAE cells do not oxidize LDL.[47,49,51,52] However, in a recent study, Morgan et al[53] found that BAE cells are effective to modify LDL, although they seem to require more time for doing so. Our results are in accordance with these data. Indeed, we observed a twofold increase of LDL oxidation after incubation of BAE cells with LDL in the presence of copper ions as compared with that observed in cell-free medium. The contradictory results leave unresolved the hypothesis that superoxide could mediate modification of LDL by cultured endothelial cells,[48,51] but a role for endothelial cell lipoxygenase has been postulated in the oxidative modification of LDL by endothelial cells.[37,49] Since BAE cells produce little superoxide and have high levels of superoxide dismutase and catalase, one important mechanism in BAE cell–induced LDL oxidation may involve cellular lipoxygenases. Our results demonstrate that BAE cells exposed to gliclazide have a reduced capacity to oxidize LDL. Although the maximal protective effect of gliclazide is similar in endothelial cells and human monocytes, BAE cells exhibit less sensitivity to gliclazide than monocytes. Such a difference in the sensitivity to gliclazide of the two systems might be interpreted in view of the fact that different mechanisms are involved in LDL oxidation. Although the mechanism(s) by which gliclazide might protect LDL from BAE cell–promoted oxidation are unknown, the low rate of superoxide production by BAE cells may suggest that gliclazide exerts antioxidative properties by reducing endothelial cell lipoxygenase activity.

Oxidized LDL has been shown to enhance monocyte adhesion to the vascular endothelium.[16] In accordance with these observations, our results demonstrate that BAE cell–induced LDL oxidation is associated with a significant increase in monocyte adhesion to the endothelium. A similar observation was made by Haller et al,[54] who recently reported that exposure of human endothelial cells to native or acetylated LDL led to an increase of monocyte adhesion. The antioxidative properties of gliclazide may be at least partly responsible for the inhibitory effect of this drug on modified LDL-induced human monocyte adhesion. Conversely, the lack of antioxidant action of glyburide may explain its ineffectiveness to reduce the LDL-induced enhancement of monocyte adhesion to the endothelium. Three adhesion molecules, VCAM-1, ELAM-1, and ICAM-1, and especially the former two, may be involved in the oxidized LDL–induced adhesiveness of endothelial cells to monocytes.[54,55] Therefore, the inhibitory effects of gliclazide on modified LDL–stimulated adhesion of monocytes to the endothelium may involve a reduction of the expression of these adhesion molecules at the surface of the endothelial cells. This hypothesis awaits further studies to be tested.

In conclusion, these data provide evidence for a specific inhibitory effect of gliclazide on LDL oxidation and LDL-stimulated adhesion of monocytes to the endothelium. These results suggest that treatment of diabetic patients with gliclazide may attenuate in vivo LDL oxidation and monocyte adhesion to the endothelium. Such effects could be beneficial in the prevention and treatment of accelerated atherosclerosis associated with diabetes.

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