CONCLUSIONS. The agents that can directly target modification of GAPDH have potential in inhibiting the development and in arresting the progression of diabetic retinopathy. The objective of this study was to understand the mechanism by which high glucose inactivates GAPDH and how the overexpression of GAPDH affects glucose-mediated metabolic abnormalities. Further, our recent studies have shown that reinstitution of good control in diabetic rats does not protect inhibition of retinal GAPDH. We also investigated the effect of reversal of a high glucose exposure to a normal glucose exposure on GAPDH activity and its covalent modification.

RESULTS. High glucose decreased GAPDH activity, expression, and nuclear translocation. Overexpression of GAPDH prevented glucose-induced inhibition of its activity, nuclear translocation, apoptosis, and activation of protein kinase C and hexosamine pathways. Inhibitors of nitration and ribosylation ameliorated glucose-induced inhibition of GAPDH, and their addition during the normal glucose exposure that followed high glucose levels had a beneficial effect on GAPDH activity and the degree of nitration and ribosylation.

METHODS. Bovine retinal endothelial cells (BRECs), transfected with GAPDH, were incubated in 20 mM glucose. The effect of the overexpression of GAPDH on its activity, nuclear translocation and reversal of glucose insult was also evaluated. BRECs were transfected with GAPDH and nuclear translocation and reversal of glucose insult was also evaluated. Overexpression of GAPDH on its activity, nuclear translocation and reversal of glucose insult was also evaluated.

PURPOSE. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) has been hypothesized as a mediator in the activation of multiple pathways implicated in the pathogenesis of diabetic retinopathy. The objective of this study was to understand the mechanism by which high glucose inactivates GAPDH in retinal microvascular cells.

RESULTS. High glucose decreased GAPDH activity, expression, and nuclear translocation. Overexpression of GAPDH prevented glucose-induced inhibition of its activity, nuclear translocation, apoptosis, and activation of protein kinase C and hexosamine pathways. Inhibitors of nitration and ribosylation ameliorated glucose-induced inhibition of GAPDH, and their addition during the normal glucose exposure that followed high glucose levels had a beneficial effect on GAPDH activity and the degree of nitration and ribosylation.

CONCLUSIONS. In hyperglycemia, GAPDH in retinal microvascular cells is inhibited by its covalent modifications, and this activates multiple pathways implicated in the pathogenesis of diabetic retinopathy. The agents that can directly target modification of GAPDH have potential in inhibiting the development and in arresting the progression of diabetic retinopathy. The overall objective of this study was to conclusively establish the role of GAPDH and its signaling pathway in the development and progression of diabetic retinopathy. The retina is a complex tissue with multiple cell types, and microvascular and other nonvascular cells could contribute to the inhibition of GAPDH seen in the rat retina in diabetes. The overall objective of this study was to conclusively establish the role of GAPDH and its signaling pathway in the development and progression of diabetic retinopathy. The use of isolated retinal microvascular cells (endothelial cells and pericytes), we have investigated the mechanism by which high glucose inactivates GAPDH and how the overexpression of GAPDH affects glucose-mediated metabolic abnormalities. Further, our recent studies have shown that reinstitution of good control in diabetic rats does not protect inhibition of retinal GAPDH. We also investigated the effect of reversal of a high glucose exposure to a normal glucose exposure on GAPDH activity and its covalent modification.

METHODS. Retinal Endothelial Cells and Pericytes. Endothelial cells (BRECs) and pericytes were isolated from bovine retina and cultured on dishes coated with 0.1% gelatin. BRECs were grown in Dulbecco’s modified Eagle’s medium (DMEM) containing 15% fetal bovine serum (heat inactivated), 5% growth medium supplement (NuSerum; BD Biosciences, Franklin Lakes, NJ), 50 μg/mL heparin, 50 μg/mL endothelial cell growth supplement, and 1% antibiotic/antimycotic. Pericytes were grown in DMEM containing 15% fetal bovine serum and 1% antibiotic/antimycotic. Cells were incubated in normal (5 mM) or high (20 mM) glucose media with or without 1 μM PJ34 (poly(ADP-ribose) polymerase; PARP inhibitor VIII; Calbiochem/EMD Chemicals, Inc., Gibbstown, NJ), 5 μM FeTPPS (5,10,15,20-Tetakis(4-sulfonatophenyl)porphyrinato Iron (III)), Chlorotetrahydrobiopterin (TH4), and 14.0 mM lactate. All cells received fresh media every 48 hours. Nuclear and cytosol fractions were prepared by differential centrifugation, as previously described by us.

Briefly, the cells harvested by trypsinization were pooled from three to four culture dishes and activation of some of the major pathways associated with the pathogenesis of diabetic complications. In addition to serving as a critical checkpoint in glycolysis, inhibition of GAPDH contributes to the diversion of upstream glycolytic intermediates to alternative pathways that could lead to the formation of AGEs, activation of PKC, and induction of the hexosamine and polyol pathways. Our recent studies have demonstrated that GAPDH is reduced in the retina in diabetes and remains compromised even after good glycemic control is reinstated; the enzyme is covalently modified and translocated to the nuclear fraction. Nuclear translocation of GAPDH is shown to be closely associated with the induction of apoptosis, and apoptosis of retinal microvascular cells precedes the histopathology characteristic of diabetic retinopathy. However, how diabetes affects GAPDH in retinal microvascular cells, the target of histopathology, remains unclear. The study was conducted with the approval of the institutional review board and with the consent of all participants. The study was supported by National Eye Institute Grant EY017313 and by the Juvenile Diabetes Research Foundation, the Thomas Foundation, and Research to Prevent Blindness.

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MgCl₂ and 2 U DNA polymerase (GoTaq; Promega, Madison, WI) using
with the nontransfected cells. The values obtained from the cells
significantly increased in BRECs overexpressing GAPDH compared
for all subsequent experiments.

GAPDH Expression Plasmids and Transfection of
Retinal Endothelial Cells

Total RNA isolated from BRECs with reagent (TRIzol; Invitrogen, Carls-
bad, CA) was converted to cDNA using a reverse transcription kit
(High-Capacity cDNA Reverse Transcription; Applied Biosystems, Fos-
ter City, CA) and served as the template for amplification of bovine
GAPDH. PCR primers, designed using Primer-BLAST with GenBank
accession number NM_001034034 were as follows: forward, 5'-CAT
CAG TCT CGA AGA TGG TGA AGG TCG-3', which included the
Kozak translation initiation site and a HindIII restriction site; reverse,
5'-GCT CGT CCT GCT GGT GGG GCT GGG-3', which included a XhoI restriction site. Bovine GAPDH-specific sequences are in bold. GAPDH was amplified in a reaction mixture containing 2.5 μM
each primer, 0.2 mM dNTP, 1 × colorless buffer containing 1.5 mM MgCl₂ and 2 U DNA polymerase (GoTaq; Promega, Madison, WI) using
the following thermal cycling conditions: initial denaturation at 95°C
for 3 minutes, 30 cycles of 95°C for 1 minute; 65°C for 1 minute, 72°C
for 2 minutes; and a final extension of 72°C for 8 minutes. Agarose gel
electrophoresis confirmed amplification of a single band of approxi-
mately 1040 bp. PCR products were purified (Wizard SV Gel and PCR
Reagent; Promega) and served as the template for amplification of bovine
GAPDH using XhoI and HindIII, and digested PCR products were ligated into pcDNA3.1 plasmid with T4 DNA ligase (Promega) before transformation
(One Shot TOP10F' Chemically Competent E. coli; Invitrogen).
Ampicillin-resistant bacterial colonies were screened by restriction
digest with XhoI and HindIII for presence of the 1040-bp insert and
were sequenced to confirm cloning of GAPDH. GAPDH containing
plasmid was purified from cultures (PureYield Plasmid Midiprep sys-
tem; Promega) and was quantified before transfection.

For transfection, BRECs (fourth to fifth passages) were incubated
with 0.5 to 5.0 μg GAPDH plasmid, as previously described.11 Trans-
fec tion complexes were formed with transfection reagent (Effectene;
Promega) and was quantified before transfection.

For transfection, BRECs (fourth to fifth passages) were incubated
with 0.5 to 5.0 μg GAPDH plasmid, as previously described.11 Trans-
fec tion complexes were formed with transfection reagent (Effectene;
Qiagen, Valencia, CA) and were incubated with BRECs for 8 hours
before incubation in 5 mM or 20 mM glucose media for 4 days. Parallel
incubations were carried out by incubating the cells in the transfection
reagent alone for 8 hours followed by incubation in 5 mM or 20 mM
glucose media. (Our previous studies have shown that control plasmid
transfection did not induce a significant increase in glycolytic activity.
) GAPDH abundance was assessed by Western blot analysis, and its glycolytic activity was assessed by measuring the production of NADH at 340 nm. Each parameter was
measured in duplicate using four different cell preparations. Results are
presented as mean ± SD of the values obtained from three different
cell preparations, with each measurement made in duplicate. 5 mM, 5
mM glucose; 20 mM, 20 mM glucose; 5 + GAP and 20 + GAP, GAPDH-
transfected BRECs incubated in 5 mM or 20 mM glucose, respectively,
5 + R and 20 + R, BRECs treated with the transfection reagent alone,
followed by incubation in 5 mM or 20 mM glucose, respectively. *P <
0.05 relative to 5 mM glucose.

GAPDH Enzyme Activity

Glycolytic activity of GAPDH was assayed in 2 to 5 μg protein accord-
ing to a method recently used by us by quantifying difference in
absorbance before and after the addition of GAPDH. Activity was
adjusted for the amount of protein.4

Western Blot Analysis

Proteins (20–30 μg) were subjected to SDS-PAGE and transferred to a
nitrocellulose membrane. Immunodetection was performed using an-
tibodies against GAPDH, caspase-3, and PARP (Santa Cruz Biotechnol-
ogy, Santa Cruz, CA) and β-D-linked N-acetylglucosamine (O/GlcNAc;
Covance, Emeryville, CA). Membranes were stripped and reprobed
with β-actin to evaluate the lane-loading control (for nuclear fractions,
histone 2B was used as the loading control). Band intensities were
quantified using gel digitizing software (Un-Scan-It; Silk Scientific Inc.,
Orem, UT).

Apoptosis

Apoptosis was determined by ELISA (Cell Death Detection ELISA™
kit; Roche Diagnostics, Indianapolis, IN) and was confirmed by mea-
suring the enzyme activity of caspase-3 and the cleavage of PARP.
For ELISA, mononucleosomes and oligonucleosomes generated from the apoptotic cells were quantified using monoclonal antibodies directed against DNA and histones, respectively, as described by us previously. Absorbance generated by incubation with 2,2'-Azino-di-[3-ethylbenzthiazolone sulfonate] diaminonium salt (Roche Diagnostics) was measured at 405 nm.

Enzyme activity of caspase-3 was assayed by measuring the formation of p-nitroanilide by the cleavage of Ac-DEVD-pNA (Biomol Research Laboratory, Plymouth Meeting, PA) at 405 nm. Each sample was analyzed in duplicate.

Cleavage of PARP into a 85-kDa subunit was determined by Western blot technique, as routinely used in our laboratory.

Quantification of GAPDH Ribosylation and Nitration
Total cell protein (80–100 µg) was immunoprecipitated with 1 µg polyclonal anti-GAPDH (Santa Cruz Biotechnology) overnight at 4°C. The antibody-protein complexes were collected (Protein A/G Plus-Agarose; Santa Cruz Biotechnology) and subjected to SDS-PAGE. Covalent modification was analyzed using monoclonal antibodies against nitrotyrosine (Upstate Biotechnology, Lake Placid, NY) or poly (ADP-ribose) (PAR; Alexis Biochemicals, San Diego, CA). The membranes were stripped and reprobed with GAPDH to evaluate the lane-loading control.

Reversal of High-Glucose Exposure
Confluent BRECs from the fourth to fifth passages were incubated in either 5 mM glucose or 20 mM glucose media. At day 4, a group of cells incubated in 20 mM glucose were rinsed with DMEM and incubated for 4 additional days in 5 mM glucose medium in the presence or absence of 1 µM PJ34 or 2.5 mM FeTPPS (reversal group). Cells incubated in continuous 5 mM glucose or 20 mM glucose for the entire duration of 8 days served as controls.

Statistical Analysis
Data are presented as the mean ± SEM. Differences attributed to treatment were statistically analyzed using the Kruskal-Wallis test followed by the Mann-Whitney U rank sum test of paired comparisons. P < 0.05 was considered statistically significant.

RESULTS
Endothelial Cells: Effect of High Glucose on GAPDH
Protein abundance of GAPDH was decreased in BRECs incubated in high glucose (Fig. 1b) with significant attenuation of its total glycolytic activity (Fig. 1c) compared with the cells incubated in normal glucose. High glucose also altered the subcellular distribution of GAPDH. As shown in Figure 2, nuclear accumulation of GAPDH was increased by more than 55% compared with the cells incubated in normal glucose. In addition, the glycolytic activity of GAPDH was decreased by 25% in the cytosol compared with >50% in the nuclear fraction in the cells incubated in high glucose compared with the cells incubated in normal glucose (Fig. 3).

Given that nuclear translocation of GAPDH is associated with proapoptotic activity, apoptosis was determined. As expected, cell apoptosis (quantified by ELISA) was increased by approximately 50% in the endothelial cells incubated in 20 mM glucose medium (Fig. 4a). In the same cells, the activity of apoptosis execution enzyme caspase-3 and the cleavage of PARP were also increased by approximately 30% to 40% compared with the cells incubated in 5 mM glucose (Figs. 4b, 4c). The signaling pathways that are postulated to be under the control of GAPDH (PKC and hexosamine pathways) were activated, as indicated by increased PKC IIβ expression (Fig. 5a) and O-GlcNAc modification of several proteins (Fig. 5b).

Reversal of Glucose Exposure
Four days of normal glucose levels that followed 4 days of high glucose levels had no beneficial effect on the activity of GAPDH in both cytosolic and nuclear fractions of BRECs; the activity remained subnormal (Fig. 3), and the enzyme continued to be covalently modified, as evidenced by its increased nitration and ribosylation (Fig. 6). Values obtained in the reversal group were not different from those obtained from the experimental group.
To determine the effect of direct inhibition of peroxynitrite or ribosylation on the metabolic memory phenomenon, FeTPPS or PJ34 was included during the normal glucose incubation period that followed 4 days of high glucose exposure. As shown in Figure 3, addition of FeTPPS during 4 days of normal glucose exposure had a beneficial effect on GAPDH activity in cytosol and nuclear fractions. The degree of nitration and ribosylation were also decreased, as determined by immunoprecipitation (Fig. 6). Thus, our results show in principle that the direct inhibition of covalent modification of GAPDH during the reversal phase has a more beneficial effect than the reversal of high glucose alone.

GAPDH in Retinal Pericytes

Given that the loss of pericytes is considered one of the earliest morphologic changes seen during the development of diabetic retinopathy and that the possible mechanism of their loss is apoptosis, some key experiments were also performed in pericytes isolated from bovine retina. As observed with isolated retinal endothelial cells, high glucose inactivated GAPDH in both cytosol and nuclear fractions of retinal pericytes (Fig. 7), and PJ34 or FeTPPS were able to ameliorate glucose-induced inactivation of GAPDH and inhibited their apoptosis (Fig. 8). Incubation of pericytes in 5 mM glucose medium with PJ34 or FeTPPS did not produce any effect on either GAPDH activity or pericyte apoptosis.

In the same pericyte preparations, the inactivation of GAPDH was not reversed by 4 days of incubation with normal glucose that followed 4 days of high glucose, suggesting its role in the development of diabetic retinopathy.

**FIGURE 3.** GAPDH activity in retinal endothelial cells. (a) Cytosol and (b) nuclear fractions of BRECes were prepared by differential centrifugation, and GAPDH activity was assessed with the use of 1 to 2 μg protein by measuring the production of NADH. Each measurement was made in duplicate using three or more cell preparations, and the values are represented as mean ± SD. 5 mM, 5 mM glucose; 20 mM, 20 mM glucose; 20+Fe, 20 mM glucose and 2.5 μM FeTPPS; 20+PJ, 20 mM glucose and 1.0 μM PJ34; Rev, BRECes incubated in 20 mM glucose for 4 days followed by 5 mM glucose for another 4 days without any addition; Rev+Fe or Rev+PJ, 2.5 μM FeTPPS or 1.0 μM PJ34 addition, respectively, during only the 5 mM glucose exposure that followed 20 mM glucose exposure. *P < 0.05 compared with 5 mM glucose. #P < 0.05 compared with 20 mM glucose alone.

**FIGURE 4.** Effect of GAPDH overexpression on endothelial cell death: GAPDH-transfected BRECes (using 2.5 μg plasmid) or untransfected BRECes were incubated in 5 mM or 20 mM glucose for 4 days. (a) Apoptosis was determined by cytoplasmic histone-associated DNA fragments using ELISA. The graph shows the mean ± SD adjusted to the total DNA in each sample. (b) Activation of apoptosis execution enzyme caspase-3 was determined in the cells by measuring the cleavage of the substrate Ac-DEVD-pNA. Each experiment was repeated with at least three different BREC preparations, and measurements were made in duplicate. (c) The appearance of a 85-kDa band of PARP was determined by Western blot technique, and β-actin was used as a loading standard. The values, represented as mean ± SD, obtained with 5 mM glucose were considered 100%. *P < 0.05 compared with 5 mM glucose. #P < 0.05 compared with 20 mM glucose.
in the metabolic memory phenomenon (Fig. 7). However, supplementation with PJ34 or FeTPPs during the normal glucose period had a beneficial effect on glucose-induced inactivation of GAPDH.

**DISCUSSION**

GAPDH is considered a potential linking mechanism between hyperglycemia-induced oxidative stress and the major pathways implicated in the pathogenesis of diabetic complications. In diabetes, retinal capillary cells experience increased oxidative stress, and their apoptosis is accelerated. Our results, for the first time, demonstrate that high glucose exposure decreases the activity and abundance of GAPDH in retinal capillary cells. Translocation of the enzyme from cytosol to the nucleus is facilitated, allowing it to increase apoptosis, and the enzyme is covalently modified. However, when GAPDH is overexpressed in retinal endothelial cells, glucose-induced inhibition of GAPDH, its translocation to the nucleus, and the acceleration of apoptosis are ameliorated, and the enzyme is covalently modified. Our results also show that the reversal of high-glucose exposure of retinal microvascular cells by normal glycemic control has a better effect than does normal glycemic control alone. These results could have great clinical implications because the prevention of GAPDH covalent modification could help prevent activation of the major pathways that are important contributors to the development and progression of diabetic retinopathy.

**FIGURE 5.** Activation of the signaling pathways upstream of GAPDH and the effect of GAPDH overexpression. Activation of (a) PKC and (b) hexosamine pathways were quantified in BRECs transfected with 2.5 μg GAPDH plasmid incubated in 5 mM or 20 mM glucose for 4 days by Western blot technique using polyclonal antibodies against PKC βII and O-GlcNAc, respectively. β-Actin was used as a loading standard. Blots are representative of four to five samples in each treatment group. Arrows: some of the bands that were increased in 20 mM glucose compared with 5 mM glucose. Western blot analyses are representative of three or more experiments. 5 mM, 5 mM glucose; 20 mM, 20 mM glucose; 20-GAP, GAPDH-transfected BRECs incubated in 20 mM glucose. *P < 0.05 compared with 5 mM glucose.

**FIGURE 6.** Covalent modification of GAPDH in BRECs. GAPDH was immunoprecipitated using 80 to 100 μg protein and was analyzed by Western blot for (a) nitration and (b) ribosylation. To ensure equal loading, the membranes were reprobed for GAPDH. The blots are representative of three or more experiments.

5 mM, 5 mM glucose; 20 mM, 20 mM glucose; 20-GAP, GAPDH-transfected BRECs incubated in 20 mM glucose. *P < 0.05 compared with 5 mM glucose.
activation of PKC, hexosamine, and polyol pathways. These pathways that could lead to the formation of AGEs and the diversion of upstream glycolytic intermediates to alternative development of diabetic microvascular complications through the its glycolytic activity is postulated to contribute to the devel-

Recent studies have shown that GAPDH is inactivated in the retina in diabetes when histopathology findings characteristic of retinopathy. GAPDH has nonglycolytic functions, and its translo-
cation to the nucleus is considered to initiate apoptosis. Here we show that high-glucose exposure of retinal endothelial cells increases the nuclear localization of GAPDH, and glycolytic activity is significantly decreased. This supports the role of GAPDH in capillary cell apoptosis, the phenomenon that pre-
cedes the histopathology observed in diabetic retinopathy. In addition, the inhibition of glucose-induced apoptosis of retinal capillary cells by the overexpression of GAPDH and the inhib-
itors of nitration and ribosylation further establishes its role in the development of retinal histopathology.

Increased peroxynitrite levels seen in the retina and its microvascular cells are considered to play an important role in the development of diabetic retinopathy. Peroxynitrite induces the nitration of GAPDH, and nitration inhibits its activity. Nitration is also associated with the nuclear accumu-
lution of GAPDH and is a factor in cell death independent of glycolytic impairment. Further, increases in reactive oxygen species can lead to increased DNA damage, which, in turn, can result in the activation of PARP. The inhibition of PARP with PJ34 reverses aortic endothelial dysfunction in dia-
betic animals for several weeks. Activation of PARP, which is shown to contribute to retinal capillary cell death, leukostasis, and the development of retinopathy in diabetic rats, also regulates GAPDH. Here we show that retinal microvascu-
lar (endothelial cells and pericytes) is one of the targets of
such covalent modifications. Both nitration and ribosylation of GAPDH are increased in these cells in high-glucose conditions, and the addition of a nitrotyrosine breaker or a PARP inhibitor prevents glucose-induced inhibition of GAPDH and apoptosis, suggesting that covalent modification is one of the important mechanisms that regulate its activity and subcellular translocation.

GAPDH is shown to regulate multiple upstream pathways that are considered major contributors in the development of diabetic retinopathy. GAPDH increases flux through the hexosamine pathway by fructose-6 phosphate, increases AGEs formation by increasing methylglyoxal, and activates PKC by the formation of diacylglycerol. GAPDH antisense oligonucleotides are shown to activate each of these pathways induced by high glucose in bovine aortic endothelial cells. Here we show that high glucose increases PKC expression and activates the hexosamine pathway in retinal endothelial cells and that the overexpression of GAPDH inhibits such glucose-induced increases. We must acknowledge that our present study did not directly measure PKC activity; however, hyperglycemia activates PKC in the retina and its endothelial cells, and reestablishment of normal glycemic metabolism after a period of poor glycemic control, in addition to failing to provide benefits to retinal GAPDH activity, also fails to normalize PKC expression, suggesting a strong relationship between increased PKC activity and its expression. Thus, our data strongly imply that the inhibition of GAPDH has the potential to contribute to the development of diabetic retinopathy by modulating its upstream signaling pathways.

In the pathogenesis of diabetic retinopathy, along with the breakdown of the blood-retinal barrier, early losses of pericyte and endothelial cells are also detected in the retina, which is followed by the development of acellular capillaries and microaneurysms. Our results show that high glucose also inhibits GAPDH in retinal pericytes, the cells that provide support to the capillaries. The possible mechanism appears to be its covalent modification. This strongly implies that GAPDH could be important in both endothelial cell loss and pericyte ghost formation.

Good glycemic control provides great benefits in the progression of diabetic retinopathy, and the benefits of prior glycemic control continue even after its termination, suggesting a metabolic memory phenomenon. Our laboratory has shown that the retina continues to experience oxidative and nitrosative damage; GAPDH remains compromised and apoptosis-related genes remain upregulated, even when hyperglycemic insult is terminated in rats. Their retinal microvasculature continues to undergo apoptosis, and many apoptosis-encoding genes continue to be upregulated. Here we show that the exposure of retinal endothelial cells and pericytes to normal glucose after a period of high glucose does not provide any benefit to GAPDH activity, its translocation to the nucleus, and covalent modification. However, if the period of normal glucose that has followed high glucose is supplemented with the peroxynitrite scavenger or the inhibitor of ribosylation, inhibition of the enzyme activity and its covalent modification are ameliorated, suggesting that the normal glucose that follows high glucose exposure does not provide any benefit to alterations in GAPDH activity. When this normal glucose period is supplemented with FeTPPs or PJ34, however, alterations in GAPDH are not observed. These results imply that supplementation with the agents that can directly target covalent modification of GAPDH during good glycemic control have greater potential to arrest the progression of diabetic retinopathy than simply good glycemic control itself.

In summary, we show that GAPDH is one of the targets of hyperglycemia-induced changes in the retinal microvasculature and that the reversal from high glucose exposure to normal glucose is not enough to abrogate the effects of high glucose, but supplementation with the inhibitors of covalent modification during the normal glucose period provides protection. These could have immense clinical significance because the mechanisms involved in nitrosylation and ribosylation of GAPDH could serve as potential targets for future therapeutics to inhibit and also to aid glycemic control in regulating cytosolic GAPDH. This could help prevent accelerated apoptosis of retinal microvascular cells and activation of the major pathways that are important in the development and progression of diabetic retinopathy.

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References


