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Protective Effects of Polyphenol-Rich Extracts of Glyphaea brevis Against Oxidative Stress in Streptozotocin-Induced Diabetic Rats

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ABSTRACT

Background and Objective: Diabetes mellitus and its oxidative complications are progressing around the world and plant-based products appear to be promising alternative treatments. The present study aimed to evaluate the antioxidant potential of the leaves of *Glyphaea brevis* (Tiliaceae) in an experimental model of diabetic rats.

Materials and Methods: Diabetes mellitus was induced by intravenous injection of streptozotocin (STZ). Aqueous and hydroethanolic extracts of G. brevis leaves (AE and HEE respectively) were given daily at doses of 250 and 500 mg/kg by gastric intubation for 30 days. Antioxidant status of rats was determined in extracellular as well as in intracellular compartments by assessing ferric-reducing antioxidant power, enzymatic antioxidants (catalase, superoxide dismutase, and glutathione peroxidase) and non-enzymatic antioxidants (protein thiols).

Results: As compared with untreated diabetic rats, administration of extracts led to significantly higher values of ferric-reducing antioxidant power, enzymatic and non-enzymatic antioxidants in all compartments.

Conclusion: These results suggest that *G. brevis* extracts have antioxidant activity that could find applications in the management of oxidative complications of diabetes mellitus.

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INTRODUCTION

Diabetes mellitus has been defined as a metabolic disorder characterized by chronic hyperglycemia of varying degrees coupled to insufficient secretion and (or) action of endogenous insulin.[1] According to statistics of the International Diabetes Federation, it now affects about 463 million people worldwide and this number is likely to rise to 578 million by 2030 and 700 million by 2045 if no adequate resources are deployed.[2] Therefore, diabetes mellitus is the most common serious metabolic disorder and has been classified during the last decade as one of the five leading causes of premature mortality in the world with 4.2 million deaths in the age group 20-79 years in 2019.[3]

Oxidative stress is the result of abnormal increase in the cell oxidation levels resulting from the excessive generation of free radicals as well as reactive oxygen species (ROS) and plays a significant role in the onset and progression of diabetes mellitus.[4,5] Persisting hyperglycemia in diabetes mellitus leads to increased production of free radicals by glucose oxidation and non-enzymatic glycation of proteins. Excessive generation of free radicals and ROS in the cell is known to cause cell membrane proteins breakdown, lipid peroxidation, and possible genomic mutations which may trigger the activation of apoptotic pathways in ß-cells[6,7] a model for high-oxidative stress. The involvement of oxidative stress in the etiology of other pathologies is strongly evidenced as ROS excess has been associated with more than a hundred diseases such as malaria, AIDS-linked opportunistic diseases, coronary heart diseases, diabetes, atherosclerosis, some cancers, skin aging, and neurodegenerative diseases.[8,9]

Diabetes mellitus is nowadays managed by diet, physical exercise, and pharmacotherapy with the last one being most used in the outpatient context. However, pharmacological drugs are either too expensive

KEYWORDS:
Antioxidant, Diabetes mellitus, *Glyphaea brevis*, Oxidative stress, Polyphenols.

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or having side effects – liver dysfunction, kidney tumors, diarrhea, etc. – that may impair the quality of life of patients.[10] Therefore, the search for new biological compounds with antioxidant activity would help develop alternative treatments which in turn could be effective and easily available for the management of diabetes mellitus as well as its oxidative complications. Medicinal plants are the main source of natural compounds and have always served as an important source of medicines throughout the ages.[11] However, despite the widespread use of herbal medicines, many pharmacological aspects of medicinal plants remain unexplored.[12] Previous studies revealed that several plants or their compounds have important antioxidant properties which may explain their health benefits in the relief of pathologies involving oxidative stress.[13,14] In this regard, polyphenols are plant molecules that have been regularly cited as potent antioxidants in the literature.[15]

Glyphaea brevis (Spreng.) Monach., a Tiliaceae, is a medicinal plant with various uses in Africa and South America. The traditional use of the leaves for the treatment of palpitations, hepatitis and poisoning has been reported in Cameroon.[16] In the Ivory Coast, the leaves are used in the treatment of fever and female sterility[17] while they are used to relieve sleepiness, bacterial infections, convulsions, sexual impotency, diabetes, and some age-related brain disorders in Nigeria.[18-21] During the two last decades, a growing number of scientific studies have been unveiling the health beneficial effects of *G. brevis* such as anticonvulsant,[21] α-amylase-inhibitor,[22] anti-inflammatory,[23] antimicrobial,[23,24] and antioxidant. [23,25] *In vitro* antioxidant activity of *G. brevis* leaf extracts has been discovered to be closely linked to their polyphenols which are mostly extracted with aqueous and hydroethanolic solvents.[25] However, no report was available on the effect of *G. brevis* on oxidative stress in vivo, especially during diabetes mellitus.

Streptozotocin (STZ) is a naturally occurring alkylating antineoplastic agent that is particularly toxic to the insulin-producing ß-cells of the pancreas in mammals. STZ stimulates hydrogen peroxide generation in pancreatic ß-cells which triggers DNA fragmentation.[26] It has therefore been widely used as an experimental model of oxidative stress and diabetes mellitus.

Thus, the present study was undertaken to assess the antioxidant activity of polyphenols-rich extracts of *G. brevis* in an animal experimental model of diabetes mellitus.

MATERIALS AND METHODS

Plant material

Fresh leaves of Glyphaea brevis were harvested in March 2018 in Douala, Cameroon, and authenticated at Cameroon National Herbarium (voucher number 10781/SRF/Cam). The leaves were cleaned with water, dried in an oven at 40°C till constant weight, powdered separately, and passed through a sieve (n°40).

Preparation of)olyphenol-rich Extracts

Aqueous extract (AE): AE was obtained by mixing 150 g of leaf powder with 2 l of distilled water. Then, the resulting mixture was boiled for 15 min, cooled at room temperature and filtered to obtain a filtrate that was vacuum-evaporated at 50°C. The yield of the extraction process was 28.2 g of AE that was kept in a sealed flask at 2°C.

Hydroethanolic extract (HEE): HEE was obtained by mixing 150 g of leaf powder with 2 l of a hydroethanolic solvent (1:1 v:v). The resulting mixture was hermetically covered and allowed to macerate at room temperature for 48 h. After maceration, the mixture was filtered at room temperature and the resulting filtrate was collected and kept at 2°C. The process was repeated once and filtrates were vacuum-evaporated at 50°C. The process led to 31.5 g of hydroethanolic extract (HEE) that was kept in the same conditions as for AE.

Polyphenolic Content Assessment

The polyphenolic content of each extract was measured by the Folin-Ciocalteu colorimetric method.[27] Thirty microliters of the extract solution (test), catechin (standard) or distilled water (control) and 1 mL of ten-times diluted Folin-Ciocalteu reagent were introduced in turn into a test tube. The resulting mixture was incubated at room temperature for 30 minutes and the absorbance was read at λ= 750 nm. Polyphenol contents were expressed in milligrams catechin equivalents per gram (mg cat eq/g).K

Animals

Healthy male Wistar rats, aged 3-4 months and weighing 200–250 g, were procured from the animal house of the Laboratory of Nutrition and Nutritional Biochemistry (LNNB), University of Yaounde 1. They were housed at a room temperature of 25 ± 2◦C, with a relative humidity of 75 ± 5%, and a 12 h dark-light cycle. They were kept in polypropylene cages with husk renewed every 24 h. Animals were free to access water and food. Experimental protocols had been approved by the Institutional Review Board of the Faculty of Science, University of Yaoundé I.

Induction of Diabetes Mellitus

After an overnight fast, rats were rendered diabetic using a single intravenous injection (50 mg/kg body weight) of streptozotocin (Sigma-Aldrich) diluted in a freshly prepared citrate buffer (0.1 M; pH 4.5). Diabetic status was assessed five days later by measuring fasting blood glucose concentrations, urinary volume, and by qualitative detection of sugar in the urine. Animals presenting a fasting glycaemia above 235 mg/dl were retained for the study.[28]

Experimental Design

Thirty-five male rats were divided into seven groups of five animals each in metabolic cages (1 animal per cage) according to the following:

- *Group I (Normal Control):* Negative control group (Normal + distilled water)
- *Group II (Diabetic Control):* Diabetic control group (Diabetic + distilled water)
- *Group III (TOLB 80):* Tolbutamide (TOLB) treated group (Diabetic + TOLB 80 mg/kg)
- *Group IV (AE 250):* Aqueous extract (AE) treated group (Diabetic + AE 250 mg/kg)

- *Group V (AE500):* Aqueous extract (AE) treated group (Diabetic + AE 500 mg/kg)
- *Group VI (HEE250):* Hydroethanolic extract (HEE) treated group (Diabetic + HEE 250 mg/kg)
- *Group VII (HEE500):* Hydroethanolic extract (HEE) treated group (Diabetic + HEE 500 mg/kg)

Each group received their assigned product daily through gastro-esophageal intubation over 4 weeks. The rats were provided with food and water ad libitum. At the end of the study period, all the rats were sacrificed under anesthesia (α-ketoglutarate-ketamine) after a 12-hour fast. Blood from the heart was collected in EDTA tubes, centrifuged at 2000 ×g for 10 minutes and the resulting plasma was kept at –20°C in Eppendorf tubes for biochemical analysis. A 0.1mL of the remaining pellet was washed in 2 ml of a 0.9% NaCl solution, centrifuged at 2000 × g for 5 minutes and this process was repeated. Hemolysis was obtained by adding 2 ml of distilled water to the washed pellet which was centrifuged at 2000 × g for 30 minutes. The supernatant (hemolysate) was collected and stored at –20°C for biochemical analysis. Heart, liver, and kidney of each animal were taken after dissection, rinsed in a 0.9% NaCl solution, dried on filter paper and mashed in a buffer (0.15 M KCl – 10 mM potassium phosphate; pH 7.4) with a ratio of 1:10 (w:v) on ice. The resulting mixture was centrifuged at 2000 × g for 30 minutes. The supernatant (homogenate) was collected and stored in Eppendorf tubes at –20°C for the assessment of biochemical parameters.

Estimation of Superoxide Dismutase

Superoxide dismutase (SOD) was assayed at λ= 480 nm using the technique of Misra and Fridovich.[29] The reaction mixture (3 mL) contained 2.5 ml of 0.05 M pH 10.2 carbonate buffer, 0.4 ml of the biological sample, and 0.3 mL of a freshly prepared 0.3 mM adrenalin solution. The increase in absorbance was recorded every 30 seconds for 120 seconds. One unit of enzyme activity was expressed as 50% inhibition of adrenalin reduction/min/mg protein.

Estimation of Catalase

Catalase (CAT) was assayed as described by Beers and Sizer. [30] Briefly, the reaction mixture (3 ml) contained 0.5 ml of the biological sample and 2.5 ml of 30 mM hydrogen peroxide. The decrease of absorbance at λ= 240 nm was recorded every 30 seconds for 90 seconds. One unit of enzyme activity was expressed as 1 µmol hydrogen peroxide reduced/min/mg protein with a molar extinction coefficient of 40 M/cm.[31]

Estimation of Protein Thiols

Protein thiols (-SH) were assessed by the method described by Ellman.[32] The reaction mixture (1 ml) contained 0.1 ml of the biological sample and 0.9 ml of Ellman's reagent (4.96 mg of 5, 5'-dithiobisnitrobenzoic acid (DTNB) in 250 ml of 0.1 M pH 7.4 Tris-HCl buffer). The absorbance was read at λ= 412 nm after 30-min incubation at room temperature.

Estimation of Glutathione Peroxidase

Glutathione peroxidase (GSH-Px) activity was measured as described by Hegde et al.[33] Briefly, reaction mixture in test tubes contained 0.2 ml of 0.4 M phosphate buffer pH 7.0, 0.1 ml of 10 mM sodium azide, 0.2 ml of tissue homogenate (test) or distilled water (control), 0.2 ml of 2 mM glutathione, 0.1 ml of 0.2 mM hydrogen peroxide. The contents were incubated at 37°C for 10 min. The reaction was stopped by adding 0.4 ml of 10% TCA, and the test tubes were centrifuged. The supernatant (1 ml) was assayed for glutathione content by using Ellman's reagent.

Estimation of Hydroperoxides

Hydroperoxides were quantified by the method of Jiang et al. [34] using ferrous oxidation in orange xylenol (FOX) reagent. The reaction mixture contained 0.9 ml of FOX reagent and 0.1 ml of the biological sample or distilled water (blank). The contents were incubated at 37°C for 30 min. Optical density was measured against the blank at λ= 560 nm.

Estimation of Malondialdehyde (MDA)

Malondialdehyde (MDA) was assayed colorimetrically as described and modified by Gutteridge and Wilkins.[33] A 0.4mL of the biological sample (test) or distilled water (blank) was treated with 2 mL of glacial acetic acid + 2 mL of 1% thiobarbituric acid, placed in a water bath for 15 min, cooled, and centrifuged at room temperature for 10 min at 500×g. The absorbance of clear supernatant was measured against reference blank at λ= 535 nm.

Estimation of Total Proteins

Total proteins concentrations were assessed spectrophotometrically using commercially-available total protein kits as per instructions of manufacturer (Chronolab, Zug, Switzerland).

Statistical Analysis

Results were expressed as means ± SD and were statistically analyzed using one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT). The SPSS for Windows software version 23.0 (SPSS Inc., Chicago, IL, USA) was used for the analysis. P-values less than 0.05 were considered significant.

RESULTS

Polyphenol Content of *G. brevis* Extracts

Polyphenol contents of extracts were as follows: 42.68 ± 1.07 mg cat eq/g for the aqueous extract (AE) and 84.67 ± 1.22 mg cat eq/g for the hydroethanolic extract (HEE).

Effects of Polyphenol-rich Extracts on Plasma Redox Status in STZ-diabetic Rats

The effect of G. brevis extracts on indicators of plasma redox status in diabetic rats is shown in Table 1. Plasma ferric reducing antioxidant power (FRAP) of the tolbutamide group was significantly higher than that of the other groups. Administration of G. brevis extracts resulted in a significant ($p < 0.05$) decrease in plasma hydroperoxide levels in all treated groups compared to the untreated control group. Only animals treated with the aqueous extract at the doses of the study showed significantly lower plasma MDA levels the diabetic control group (EA 250 mg / kg: -41.27%, $p < 0.01$; EA 500 mg

/ kg: -36.91%, p <0.01). There was, in general, no significant difference between the plasma levels of protein thiols in the different groups, with the exception of those in the group treated with the hydroalcoholic extract at a dose of 250 mg / kg which were higher.

Effects of Polyphenol-rich Extracts on Intracellular redox Status in STZ-diabetic Rats

FRAP

Diabetes caused a significant decrease in FRAP (1.5 to 4.5 times) in all organs as well as in erythrocytes (Figure 1). The most severe depletions in FRAP were noticed in the liver (Figure 1A), the kidney (Figure 1B) and the erythrocytes (Figure 1D). Treatment with either AE or HEE led to higher values of FRAP, although these values were comparable to those from the normal control group only in liver (Figure 1A) and erythrocytes (Figure 1D).

Hydroperoxides

Diabetes caused a significant increase in hydroperoxides concentrations which were 2 to 2.5 times higher than those of the normal control (Figure 2). This effect was noticed in plasma as well as in erythrocyte hemolysates and organ homogenates (liver, kidney, and heart). Treatment with AE and HEE resulted in lower concentrations of hydroperoxides. The effects were comparable to those of tolbutamide.

Malondialdehyde (MDA)

Malondialdehyde concentrations were significantly increased in streptozotocin-induced diabetic rats in comparison with the normal control group (Figure 3). This effect was noticed in all organs as well as in erythrocytes. Diabetic rats treated with extracts had lower MDA values with the most important reduction brought about by HEE at dose of 500 mg/kg in all organs. Globally, the MDA concentrations achieved with extract treatments were comparable to those with tolbutamide.

Thiols

Diabetes drastically reduced thiols concentrations with reductions ranging between 60% and 75% depending of the sample analyzed (organs or erythrocytes) as seen in Figure 4. The most important decrease was noticed in the kidneys and the erythrocytes. Treatment with extracts or tolbutamide moderately increased thiols concentrations but they remained lower than those of the normal control group.

Enzymatic antioxidants

Diabetes caused a decrease in antioxidant enzyme activities of 37.80% (SOD), 49.29% (CAT) and 45.01% (GPx) at the erythrocytic level (Figures 5-7). The administration of the aqueous extracts 500mg / kg PC resulted in an increase in the activities of CAT (66.36%), SOD (42.70%), GPx (23.46%) while the administration of hydroethanolic extract at 500mg/kg BW/ kg BW led to an increase in the activities of CAT (52%), SOD (43.96%) and GPx (23.91%) as presented in Figures 5-7.

At the hepatic level, STZ-induced diabetes decreased the activities of CAT (53.19%), SOD (48.15%) and GPx (54.37%) respectively (Figures 5-7). The administration of the aqueous extracts 500 mg / kg PC resulted in an increase in the activities of CAT (62.71%), SOD (47.47%) and GPx (18.55%) while the administration of the hydroethanolic extracts 500mg / kg PC led to an increase in the activities of CAT (69.23%), SOD (47.47%) and GPx (19.58%).

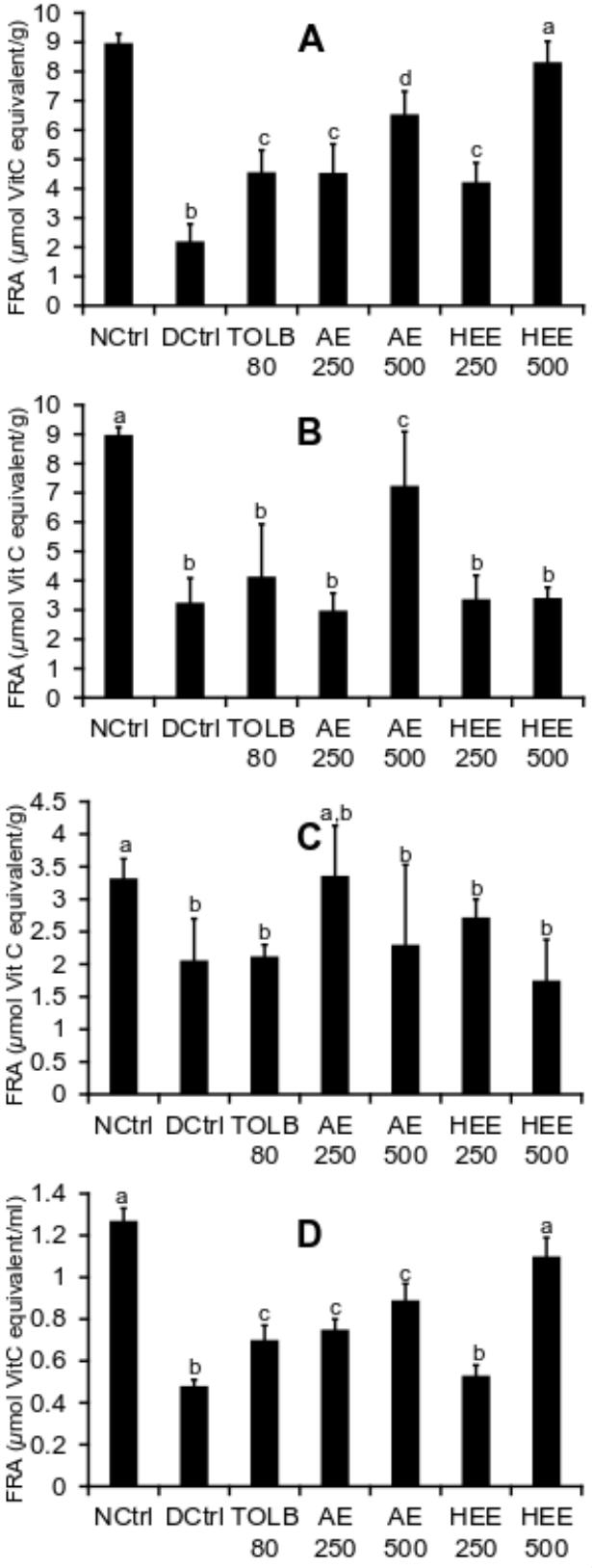

Fig. 1: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the ferric-reducing antioxidant power (FRA) in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

At the renal level, diabetes led to a decrease in SOD (50.64%) and GPx (50.14%) activities. CAT activity did not significantly decrease. The administration of the aqueous extract 250 mg/kg BW resulted in an increase in CAT activity of 53.70% and an increase of 52.83% with the hydroethanolic extract of 250 mg / kg BW. With regard to the activity of renal SOD, the administration of the aqueous extract 500 mg / kg BW resulted in an increase of 37.90% and the hydroethanolic extract 500 mg / kg BW an increase of 46.90%. Regarding the activity of renal GPx, the aqueous extract at 500mg/kg Bw increased the renal GPx activity up to 38.13% while the HEE increased it by 40.7%.

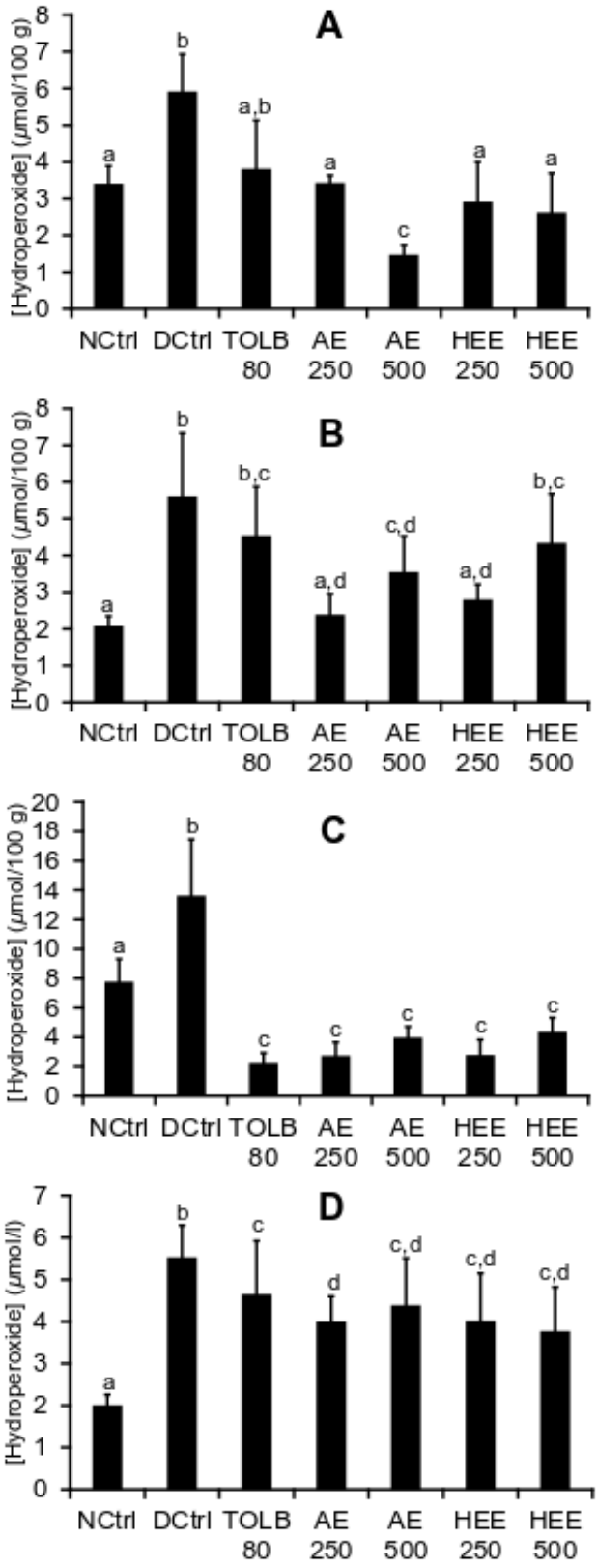

Figure 2: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the hydroperoxides concentration in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

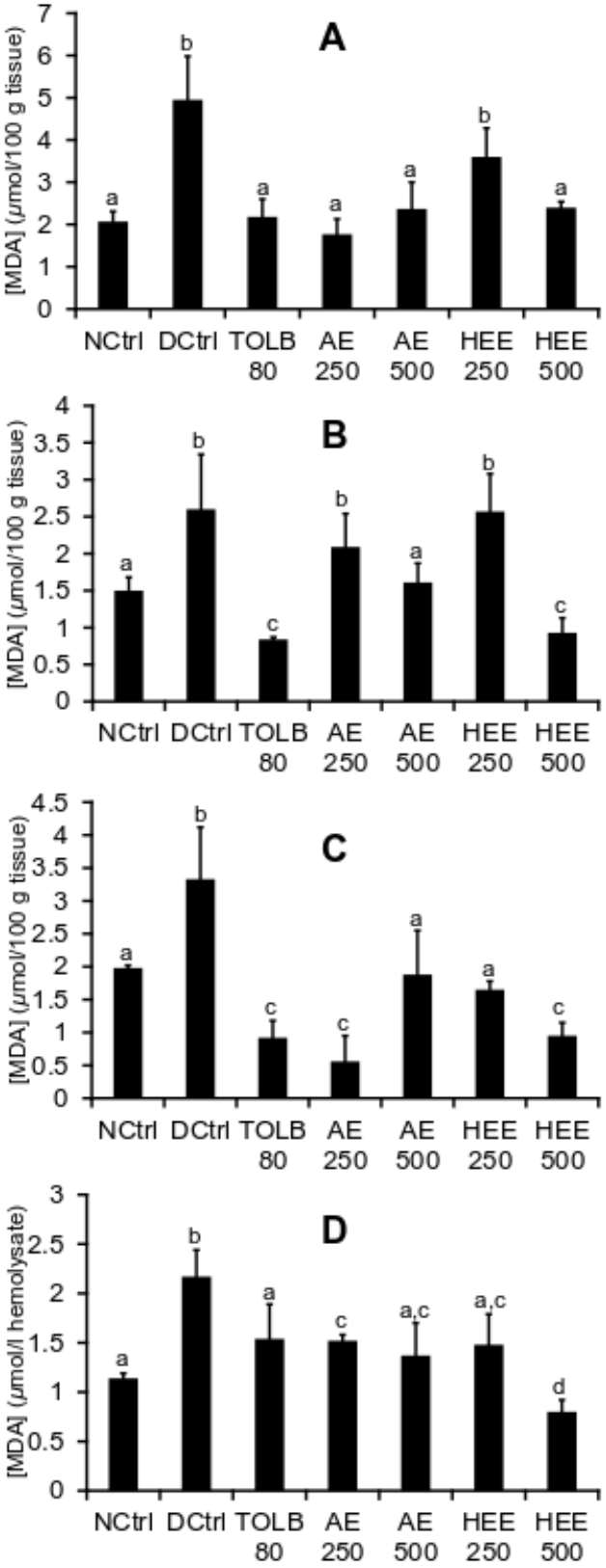

Figure 3: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the malondialdehyde (MDA) concentration in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

At the cardiac level, the induction of diabetes led to a decrease in CAT (70.33%) and GPx (44.44%). SOD activity remains unchanged. The administration of the extracts resulted in an 83.6% increase in CAT activity with the aqueous extract 250 mg / kg BW and 77.5% with the hydroethanolic extract 500 mg / kg BW. Moreover, an increase in SOD activity of 62.5% was observed with the aqueous extract 500 mg / kg BW and of 58.90% with the hydroethanolic extracts 500 mg / kg BW. Regarding GPx activity, the administration of the aqueous extract 500 mg / kg BW resulted in an increase of 23.56% and the hydroethanolic extract at dose of 500 mg / kg BW, an increase of 27.84%.

DISCUSSION

Diabetes is usually accompanied by high production of free radicals[5] or impaired endogenous antioxidant defenses.[35,36]

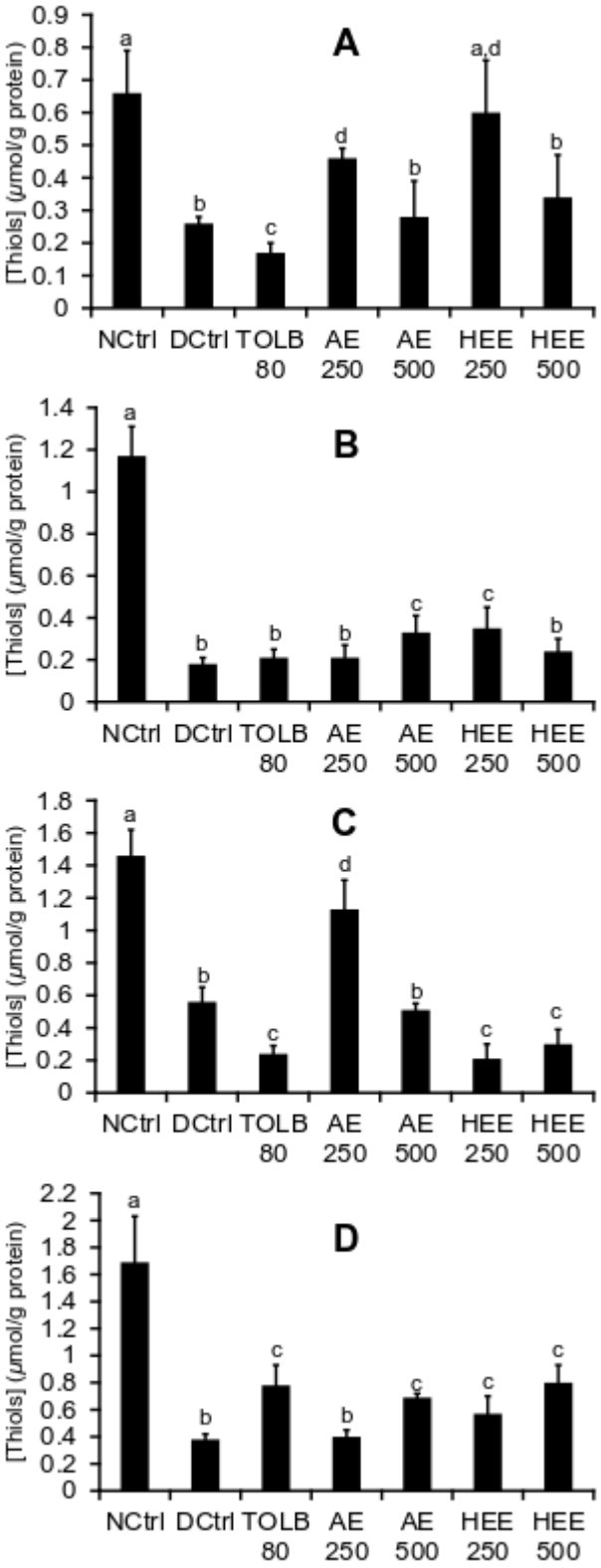

Fig. 4: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the protein thiols concentrations in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

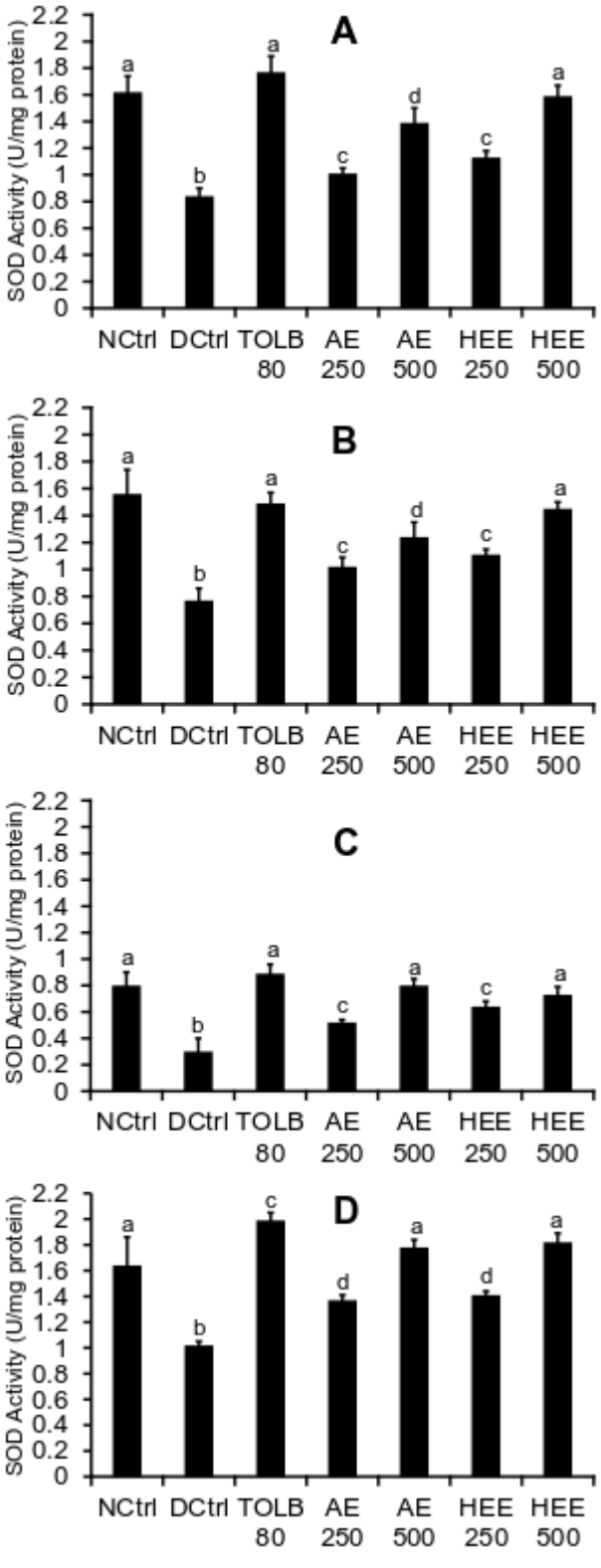

Fig. 5: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the activity of superoxide dismutase (SOD) in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

However, excessively high levels of free radicals damage cell proteins, membrane lipids, nucleic acids, and may even lead to cell death. Various mechanisms of reactive oxygen species formation have been suggested. Glucose oxidation is believed to be the main source of free radicals. While in its ene-diol form, glucose is oxidized (through a reaction requiring transition metals) into an ene-diol radical anion which is converted to reactive ketoaldehydes and anionic superoxide radicals. These anionic superoxide radicals undergo dismutation processes to yield hydrogen peroxide. The latter, if not degraded by catalase (CAT) or glutathione peroxidase (GPx), and in the presence of transition metals, may pave the way to the production of extremely reactive hydroxyl radicals.[37,38] Anionic superoxide groups can also react with nitric oxide to form reactive peroxynitrite groups.[39,40] It has also been found that hyperglycemia promotes the peroxidation of low-density lipoproteins (LDL) through a superoxide-dependent pathway resulting in the production of free radicals.[41,42]

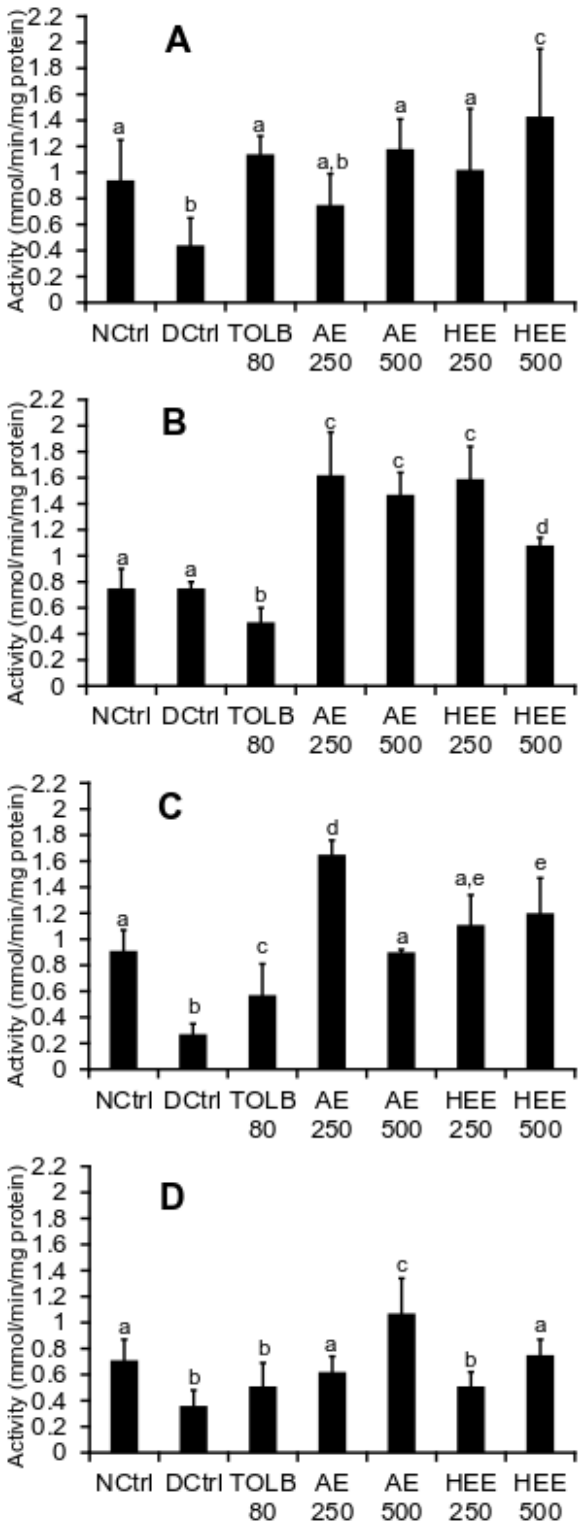

Fig. 6: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the activity of catalase in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

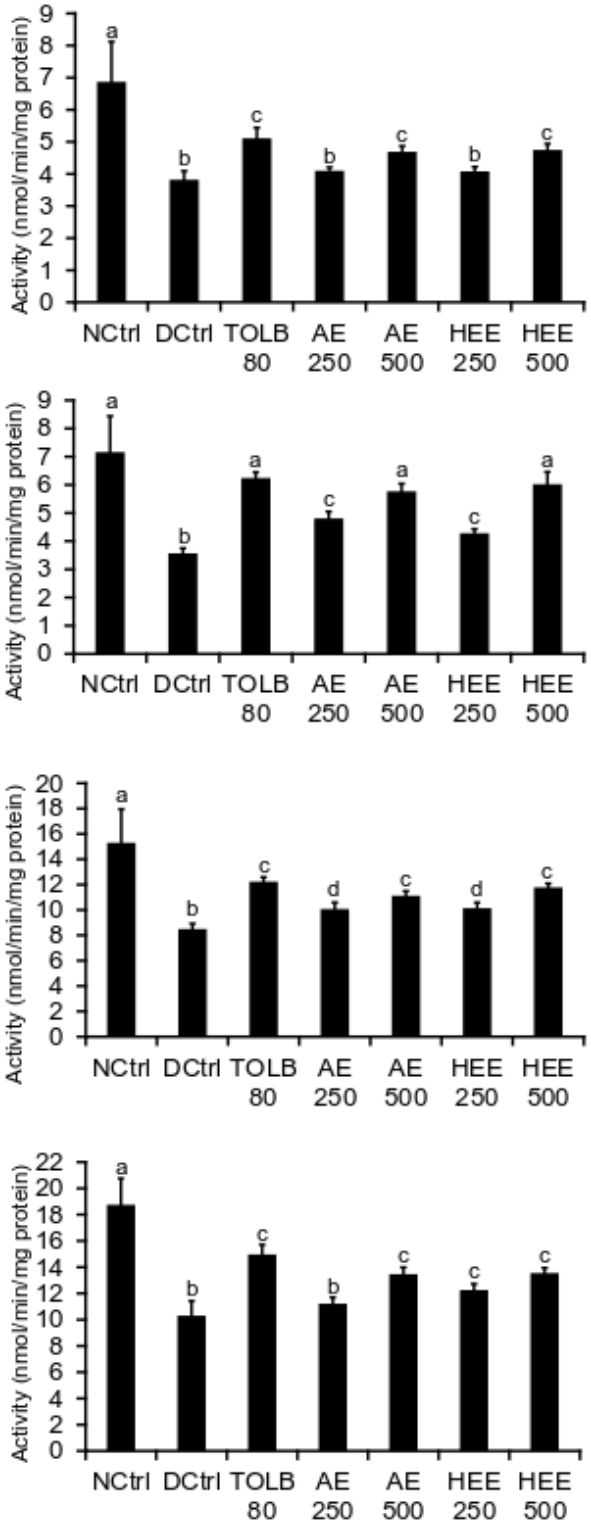

Fig. 7: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the activity of glutathione peroxidse in the liver (A), kidney (B), heart (C) and erythrocytes (D) of diabetic rats.

Table 1: Effects of aqueous (AE) and hydroethanolic (HEE) extracts of G. brevis on the plasma redox status of diabetic rats.

Group	*FRAP (µmol VCE*/l)*	*ROOH (µmol/l)*	*MDA (µmol/l)*	*Thiols (µmol/g protein)*
NCtrl	390.18 ± 53.36[a]	6.28 ± 3.23[a]	1.04 ± 0.07[a]	1.29 ± 0.25[a]
DCtrl	181.60 ± 33.89[a]	19.49 ± 2.01[b]	2.98 ± 0.42[a]	0.22 ± 0.01[a]
TOLB 80	276.07 ± 35.07[b]	17.41 ± 0.85[b]	2.25 ± 0.38[ab]	0.15 ± 0.03[b]
AE 250	218.40 ± 46.28[ab]	9.90 ± 2.43[a,c]	1.75 ± 0.42[b]	0.20 ± 0.03[a]
AE 500	168.10 ± 49.95[a]	10.66 ± 2.05[a,c]	1.88 ± 0.54[b]	0.22 ± 0.03[a]
HEE 250	195.09 ± 34.99[a]	7.44 ± 3.12[a]	2.98 ± 0.40[a]	0.32 ± 0.04[c]
HEE 500	179.55 ± 23.32[a]	12.33 ± 2.48[c]	2.86 ± 0.56[a]	0.18 ± 0.,02[ab]

In addition to this source of free radicals in diabetes, the interaction of excess glucose with proteins leads to the formation of Amadori products and therefore of advanced glycation end-products (AGE).[43] These AGEs, via their receptors, inactivate enzymes by altering their structure,[44] encourage the formation of free radicals[5] and block the antiproliferative effects of nitric oxide.[45,46] AGEs also lead to mutations in DNA at certain points as well as to breakdown of its double-stranded structure, activation of caspases, and apoptosis.[47] They also generate reactive oxygen species (ROS) inducing oxidative damages especially in the heart and the kidney.[48] By increasing the level of intracellular oxidative stress, AGEs activate the transcription factor NF-κB and allow the upregulation of target genes controlled by this factor. The transcription factor NF-κB increases the production of nitric oxide, which is thought to mediate ß-islet cell damage.[49]

Previous studies revealed that injection of streptozotocin to rats triggers an increase in lipid peroxidation characterized by raised levels of malondialdehyde (MDA) and hydroperoxides.[50,51] Therefore, quantification of MDA and hydroperoxides is useful in assessing the extent of tissue damages linked to lipid peroxidation.[51] Administration of *G. brevis* extracts to diabetic rats reduced the levels of MDA and hydroperoxides in erythrocytes and cells from organs. Previous in vitro studies highlighted the free radical scavenging activity of *G. brevis* polyphenols.[23,25] These observations suggest that *G. brevis* would exert an upstream inhibition on lipid peroxidation in diabetic rats by scavenging ROS in the cells.

Enzymatic antioxidants (SOD, CAT, GPx) are the first line of cell defense against oxidative damage initiated by ROS.[52] Previous studies showed that injecting streptozotocin to rats results in a decrease in the levels of both enzymatic and non-enzymatic antioxidants.[50,53,54] Diabetic rats treated with the extracts had higher values of SOD, CAT, and GPx than those from the untreated diabetic group. Once more, the free radical scavenging activity of *G. brevis* may be responsible for this observation.[23,25] On the other hand, the antihyperglycemic effect of *G. brevis* extracts may also be considered as it would limit the oxidative damages especially on functional proteins caused by free radicals and glucose during diabetes.[5,44] The hypothesis of stimulation of the expression of the genes coding these proteins may also explain these increases in activity. The different patterns of antioxidant activity in the organs studied may be the result of differences in the rate of oxidative exposition to which each organ is subject.

CONCLUSION

At the end of this study, it appears that the leaves of Glyphaea brevis have antioxidant effects in streptozotocin-induced diabetic rats. These antioxidants have been extracted in solvents with low toxicity, suggesting the potential application of our findings in the safe management of human diabetes.

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