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Impact of Gliclazide Modified Release or Glimepiride as Add-on Therapy to Metformin on Glycemic and Oxidative Stress Parameters in Type 2 Diabetic Patients

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Abstract

Type 2 diabetes mellitus is a condition characterized by an elevation of oxidative stress, which has been implicated in diabetic progression and its vascular complications.

Assessing the impact of gliclazide modified release (MR) versus glimepiride on oxidative stress markers, glycemic indices, lipid profile, and estimated glomerular filtration rate in uncontrolled type 2 diabetic patients on metformin monotherapy.

This was a prospective comparative study conducted in the Thi-Qar specialized diabetic, endocrine, and metabolism center. Sixty-six patients were allocated into two groups based on the addition of the sulfonylureas (SUs). Group 1 (33 patients) was on gliclazide MR, whereas Group 2 (33 patients) was on glimepiride. The measured oxidative stress markers were reduced glutathione (GSH), superoxide dismutase (SOD), malondialdehyde (MDA), and protein carbonyl (PC) evaluated before and after 16 weeks of SUs addition.

There were significant drops in SOD ($P < 0.001$), MDA ($P < 0.001$), and PC ($P = 0.001$) and a significant increase in GSH ($p = 0.029$) levels after gliclazide MR add-on therapy. There were significant drops in SOD ($P = 0.026$) and MDA ($P < 0.001$) levels with non-significant changes in both GSH ($P = 0.214$) and PC ($P = 0.538$) after glimepiride add-on therapy. There was a significant difference in improvement of PC level ($P = 0.048$) in the gliclazide group compared to the glimepiride group, with a non-significant difference in the improvements of GSH, SOD, and MDA between both groups. At the end of the study, there were no significant differences in glycemic control, lipid profile, or eGFR improvement between the two groups.

Glycemic control plays a pivotal role in decreasing oxidative stress. The control of diabetes with the gliclazide-MR-metformin combination reduced oxidative stress more than the glimepiride-metformin combination, indicating its antioxidant property.

Keywords: Oxidative Stress, T2DM, Gliclazide MR, Glimepiride, Metformin.

تأثير جليكلازيد طويل الامد او جليميبرايد كدواء اضافي للميتفورمين على مؤشرات الاجهاد التاكسدي ونسبة السكر في الدم في مرضى السكري من النوع الثاني

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الخلاصة

يتميز داء السكري النوع الثاني بارتفاع الإجهاد التأكسدي الذي يساهم في تقدم المرض ومضاعفته.

تقييم تأثير جليكلازيد طويل الامد بالمقارنة مع جليميبرايد على علامات الإجهاد التأكسدي ومؤشرات نسبة السكر في الدم وملف الدهون ومعدل الترشيح الكبيبي في مرضى السكري النوع الثاني غير المنضبطين على العلاج الأحادي بالميتفورمين.

هذه دراسة مقارنة مستقبلية اجريت في مركز ذي قار التخصصي لأمراض السكري والغدد الصماء والتمثيل الغذائي. تم توزيع ستة وستين مريضا إلى مجموعتين اعتمادا على اضافة أدوية السلفونيل يوريا. المجموعة الاولى (٣٣ مريضا) كانت على جليكلازيد طويل الامد بينما المجموعة الثانية (٣٣ مريضا) كانت على جليميبرايد. تضمنت علامات الإجهاد التأكسدي المقاسة على الجلوتاثيون (GSH), أنزيم سوبر اوكساید ديسموتاز (SOD), ملونديالديهيد (MDA) و بروتين كاربونيل (PC) التي تم تقييمها قبل وبعد ١٦ أسبوعًا من إضافة السلفونيل يوريا .

كان هناك انخفاض معنوي في مستويات علامات الاجهاد التأكسدي لكل من SOD (P <0.001)، MDA (P <0.001)، و PC (P = 0.001) وزيادة معنوية في مستويات GSH (p = 0.029) بعد جليكلازيد إم آر .
كانت هناك انخفاضات معنوية في مستويات SOD (P = 0.026) وMDA (P <0.001) مع تغيرات غيرمعنوية في كل من GSH (P = 0.214) و PC (P = 0.538) بعد اضافة جليميبرايد. كان هناك فرق كبير في تحسين مستوى PC (P = 0.048) في مجموعة جليكلازيد إم آر مقارنة بمجموعة الجليميبرايد، مع اختلاف غير معنوي في تحسينات الـ GSH و SOD و MDA بين المجموعتين. في نهاية الدراسة، لم تكن هناك فروق ذات دلالة إحصائية في ضبط نسبة السكر في الدم ، الدهون ، أو تحسين معدل الترشيح الكبيبي بين المجموعتين.
يلعب التحكم في نسبة السكر في الدم دورًا محوريًا في تقليل الإجهاد التأكسدي. خفضت السيطرة على مرض السكري مع مزيج جليكلا زيد – طويل الامد - ميتفورمين الإجهاد التأكسدي أكثر من مزيج جليميبيريد - ميتفورمين، مما يشير إلى خصائصه المضادة للأكسدة.
الكلمات المفتاحية: الإجهاد التأكسدي ، داء السكري النوع الثاني ، جليكلازيد إم آر ، جليميبرايد ، ميتفورمين.

Introduction

Type 2 diabetes mellitus (T2DM) is a globally growing metabolic disease and is defined as persistent hyperglycemia caused by a progressive decline in insulin secretion and/or resistance [1,2]. Long-term hyperglycemia is associated with both macro- and micro-vascular complications that lead to cardiovascular diseases, diabetic retinopathy, neuropathy, and nephropathy [3,4]. These complications increase the risk of health problems and expose T2DM patients to vulnerable morbidity and mortality [5,6]. The cause of T2DM is related to both various genetic and environmental factors that impact inflammation, autoimmunity, and metabolic stress [7].

High glucose blood levels enhance the buildup of free radicals due to mitochondrial dysfunction and endoplasmic reticulum stress[8]. The phenomenon of oxidative stress (OS) refers to an imbalance between the productions of reactive oxygen species (ROS) and the antioxidant defense system [9]. The overexpressed ROS alters the structure and function of proteins, lipids, and nucleic acids [10]. Beta cell dysfunction and insulin resistance have been related to hyperglycemia-induced ROS overproduction, which is implicated in vascular endothelial dysfunction, by different mechanisms, including auto-oxidation of glucose, polyol pathway flux, hyper-activation of the hexosamine pathway, enhanced activation of protein kinase C (PKC) associated with activation of the nuclear factor kappa B (NF-kB) pathway, increased production of advanced glycation end products (AGEs), and reduced antioxidant capacity [11–13]. Oxidative stress is not only a potential pathophysiologic mechanism for the development of T2DM and insulin resistance, but it is also a critical upstream event for diabetic vascular complications [14].

Many clinical reports have observed the disruption of redox homeostasis during T2DM [15]. This includes an increase in serum total superoxide dismutase (SOD) enzyme expression [16,17], a drop in reduced glutathione (GSH) serum levels [18], an increase in malondialdehyde (MDA) serum levels [19], which is a byproduct of lipid peroxidation, and an increase in protein carbonyl (PC) serum levels [20],a marker of protein oxidation.

The necessity of anti-diabetic treatments for maintaining good glycemic control outweighs the negative patient concerns about the potential adverse effects of medications [21]. Even though there are many new available anti-hyperglycemic agents for treating T2DM that work on different mechanisms, sulfonylureas (SUs), especially modern SUs, in particular both gliclazide modified release (MR) and glimepiride, are still often used in clinical practice [22] because both demonstrate efficacy and safety [23], have a lower incidence of hypoglycemia [24,25], and are affordably priced. In the Action in Diabetes and Vascular Disease: Preterax and Diamicron Modified Release Controlled Evaluation (ADVANCE) [26] and Cardiovascular Outcome Study of Linagliptin Versus Glimepiride in Patients With Type 2 Diabetes (CAROLINA) [27] studies, the cardiovascular safety of second-generation SUs was demonstrated with gliclazide MR and glimepiride, respectively. Many international guidelines, including those from the World Health Organization (WHO) [28,29], recommend the use of modern SUs in the management of T2DM as a first-line add-on treatment to metformin monotherapy when it is not enough to achieve glycemic control.

Although there is a controversial result regarding the role of oral anti-diabetic agents in oxidative stress reduction, gliclazide showed both in vitro and in vivo its property as a free radical scavenger [30] due to its unique aminoazabicyclo-octyl ring . Despite the fact that several studies have looked into the antioxidant effect of gliclazide [31,32], there are few reports that have looked at glimepiride, with contradictory results. It has been established that the combination of sulfonylurea and metformin improves glycemic control [33]. However, there are limited studies on the potential therapeutic benefits of sulfonylureas added to metformin treatment on oxidative stress, lipid profile, and renal functions. This study aimed to assess the effects of gliclazide modified release (MR) versus glimepiride on oxidative stress markers beyond their glycemic control in patients with T2DM who are inadequately controlled by metformin monotherapy.

Patients and Methods

This non-randomized clinical trial (quasi-experimental) was carried out in Thi-Qar specialized diabetic, endocrine and metabolism center (TDEMC) at Thi-Qar Health Directorate (Thi-Qar – Iraq) after the clearance of Human Research Ethics Committee of the Iraqi Ministry of Health and

approved by research ethics committee of College of Pharmacy/ University of Baghdad under approval name of (REACUBCP4122021A). An informed permission was obtained from all participants after explanations of the study objectives. The study was conducted during the period between February 2022 and November 2022.

Sample size

The need to identify a significant difference between two independent groups by the intent of comparing means using the following equation [34] :

$$n = 2(Z\alpha /2 + Z\beta) 2 / (\mu1 – \mu2 / \sigma) 2$$

Assuming normal distribution and homogenous variance ($\sigma1= \sigma2 = \sigma =1$), equal sample size (n1 = n2 = n = number of participants per group), and 2 tailed $\alpha = 0.05$ so $Z\alpha /2 = 1.96$ and $\beta = 0.02$ (80% power) so $Z\beta = 0.8416$, and ($\mu1 – \mu2 / \sigma$) = effect size = δ which regarded as large in this study = 0.7.

$$n = 2(1.96 + 0.8416) 2 / (0.7) 2 = 32.036$$

We approximate the number to 33 participants in each group or a total of 66.

Inclusion criteria

Uncontrolled T2DM patients of both genders on their conventional therapy (metformin ± statins) were enrolled in this study according to following eligible criteria: adults (35-65 years old) with duration of T2DM ≤ 6 years and using metformin only as anti-diabetic agent in their conventional treatments regardless of the dosage form type or duration of use, and either on the full allowed daily dose of (2 gm) or the maximum tolerated dose of (1 gm) with glycosylated hemoglobin (HbA1c) ≤ 11%.

Exclusion criteria

All patients with chronic kidney disease (CKD), established cardiovascular disease (CVD), severe liver disease, on chemotherapy or radiotherapy, regular users of antioxidants, with systemic comorbidities such as hypothyroidism, on insulin or oral anti-diabetic medication other than metformin as starting monotherapy, and those who changed their doses of antihyperlipidemic agents within 3 months prior to the baseline visit were excluded from the study.

Groups allocation

After enrollment, T2DM patients were allocated into 2 groups based on SUs added to their conventional therapy as follow: the first group of patients was prescribed oral gliclazide MR 60 mg tablet once daily and the second group of patients was prescribed oral glimepiride 2 mg tablet once daily.

Data collection

The socio-demographic data, personal habits, baseline disease characteristics, and medical history were taken from each participant. The weight and height of patients were used to calculate their body mass index (BMI, kg/m2). Both systolic and diastolic blood pressures (SBP & DBP) were measured at the resting phase. The biochemical tests and enzyme-linked immunosorbent assay (ELISA) of the investigated parameters were evaluated at the baseline visit (before adding SUs) and at the follow-up visit after 16 weeks of SU add-on therapy.

Blood sample collection

Ten milliliters of venous blood were taken from each patient during their scheduled visit to the TDEMC after fasting for 12 hours. About 2 ml of the collected blood was immediately transferred to an ethylene tetraacetic acid (ETDA) tube and sent for HbA1c analysis. The remaining 8 ml of blood was placed in a gel tube for 30 minutes to coagulate before being centrifuged at 3000 rpm for 10 minutes to obtain serum. A portion of the serum on the day of collection was tested for fasting blood glucose (FBG), serum creatinine (S.Cr) and lipid profile levels at the chemical laboratory. The residual serum was divided into 4 eppendrof tubes and kept frozen at (-40^{o}C) until the time of the serum MDA, GSH, PC, and SOD assays by ELISA.

Measurement of selected clinical parameters

Serum levels of reduced GSH, SOD, MDA, and PC were measured using commercially available sandwich technology ELISA [35] kits (Shanghai YL Biotech Co., Ltd., China) according to the manufacturer's instructions (HumaReaderHS, Human, Germany). The HbA1c levels were determined using high-performance liquid chromatography [36] with the D-10TM hemoglobin testing system (Bio-Rad, USA). Standard enzymatic methods (Abbott, Architect plus C4000, USA) were used to determine FBG, the lipid profile, and S.Cr. levels [37]. Non-high density lipoprotein cholesterol (Non-HDL), a predictor of atherogenic risk, was calculated as follow: Non-HDL= Total cholesterol (TC) – High density lipoprotein (HDL) [38]. Kidney function was assessed by calculating the estimated glomerular filtration rate (eGFR) depending on S. Cr level according to the Chronic Kidney Disease-Epidemiology Collaboration (CKD-EPI) equation GFR = 141 × min {S.cr/κ, 1} α × max {S.cr/κ, 1}-1.209 × 0.993Age × 1.018 [if female] - 1.159 [if black] [39].

Statistical analysis

The statistical analysis was conducted using (IBM SPSS Statistics, USA version 26) software for Microsoft Windows. The normal distribution of data was evaluated by the Shapiro-Wilk test. Descriptive statistics of continuous variables were expressed as mean ± standard deviation (SD) for parametric data or median and 25-75 interquartile range (IQR) for non-parametric data, while categorical variables were presented as numbers and percentages and tested by the Chi-square test. A paired sample t-test was used to assess difference between two repeated

means within the same group when normality was proven; alternatively, the Wilcoxon signed rank test was used for non-parametric variables repeated measures. The independent t-test was used to compare the means of two independent parametric variables, and the Mann-Whitney test was used for the median comparison of two unrelated non-parametric variables. The significance level of the difference was considered when p value < 0.05 in a two-tailed test. The difference was calculated as (pretreatment value– posttreatment value).

Results

The study profile included 8 weeks screening period for 800 T2DM patients, while those who fulfilled the study requirement and accepted to participate were 80 patients. A total of 33 patients in the first group and 33 patients in the second group completed the study. The socio-demographic and baseline disease characteristics of patients summarized in Table 1 are without significant differences between the studied medication groups.

Table 1. Socio-demographic and baseline disease characteristics of patients

Parameters	First group Add on Gliclazide (n=33)	Second group Add on Glimepiride (n=33)	P-value
Age (year)	49.33 ± 8.96	50.84 ± 8.51	0.484
Duration of T2DM (year)	3.84 ± 1.37	3.75 ± 1.41	0.792
Gender (male/female)	13 (39.4)/20 (60.6)	14 (42.4)/20 (57.6)	0.802
Smoking (Yes/No)	6 (18.2)/27 (81.8)	5 (15.2)/28 (84.8)	0.741
Family history of T2DM (Yes/No)	27 (81.8)/6 (18.2)	28 (84.8)/5 (15.2)	0.741
Metformin type(Extend/Ordinary)	19 (57.6)/14 (42.4)	19 (57.6)/14 (42.4)	1
Metformin daily dose (1gm/2gm)	6 (18.2)/27 (81.8)	4 (12.1)/29 (87.9)	0.492
Statins (Yes/No)	19 (57.6)/14 (42.4)	19 (57.6)/14 (42.4)	1
Hypertension (Yes/No)	9 (27.3)/24 (72.7)	10 (30.3)/23 (69.7)	0.786
Diabetic neuropathy (Yes/No)	14 (42.4)/19 (57.6)	13 (39.39)/20 (60.6)	0.802

Data presented as [n (%), and mean ± (SD)]. n: number of subjects, SD: Standard deviation, *Significant when p < 0.05.

Also, there were no significant differences between the two groups in terms of anthropometric measurements and blood pressure parameters pre- and post-treatments as shown in Table 2.

Table 2. The impact of studied treatment on the anthropometric measurements of patients

Parameters	First group Add on Gliclazide (n=33)	Second group Add on Glimepiride (n=33)	P-value
Pre weight (Kg)	80.12 ± 14.59	79.39 ± 14.26	0.838
Post weight (Kg)	81 ± 15.11	80.48 ± 15.24	0.891
P-value within group	0.202	0.137	
Difference	-0.87 ± 3.87	-1.09 ± 4.1	0.830
Height (cm)	161.03 ± 9.25	161.84 ± 8.94	0.716
Pre BMl (Kg/m^2)	30.73 ± 3.94	30.27 ± 4.62	0.668
Post BMl (Kg/m^2)	31.12 ± 4.2	30.68 ± 4.93	0.694
P-value within group	0.152	0.151	
Difference	-0.39 ± 1.53	-0.40 ± 1.56	0.979
Pre W.C. (cm)	104.9 ± 9.15	103.3 ± 10.39	0.508
Post W.C. (cm)	105.27 ± 8.93	104.06 ± 10.98	0.625
P-value within group	0.402	0.207	
Difference	0.0 (-1.5 – 0.0)	0.0 (-2.5 – 0.5)	0.444
Pre SBP (mmHg)	130 (120 – 140)	130 (120 – 140)	0.880
Post SBP (mmHg)	120 (120 – 130)	130 (110 – 140)	0.431
P-value within group	0.235	0.936	
Difference	0.0 (-10 – 10)	0.0 (0.0 – 0.0)	0.323
Pre DBP (mmHg)	80 (80 – 90)	80 (70 – 90)	0.325
Post DBP (mmHg)	80 (80 – 80)	80 (70 – 85)	0.539
P-value within group	0.180	0.405	
Difference	0.0 (0.0 – 10)	0.0 (0.0 – 0.0)	0.532

Data presented as [mean ± (SD), and median (Q1-Q3)]. W.C.: waist circumference. Kg: kilogram. m^2: square meter. cm: centimeter. mmHg: millimeters of mercury.

After 16 weeks of SUs add-on therapy, both groups showed significant ($p < 0.05$) reductions in FBS and HbA1c compared to baseline values with no significant difference between the groups despite that there was numerically higher improvement in the gliclazide add-on group than in the glimepiride add-on group. Also, both groups significantly decreased lipid profile levels except non-significant change in triglycerides (TG) levels compared to baseline levels with no significant difference between both groups despite that the improvement was numerically higher in the second group than the first group. First group increased eGFR significantly in contrary to non-significant increase in the second group. Despite that the difference between both groups was non-significant ($p = 0.578$) as shown in Table 3.

Table 3. The impacts of studied treatments on glycemic indices, lipid profile and eGFR of patients

Clinical variables	First group Add on Gliclazide (n=33)	Second group Add on Glimepiride (n=33)	P-value
Pre FBG (mg/dl)	169.95 ± 38.38	169.52 ± 38.45	0.964
Post FBG (mg/dl)	137.06 ± 36.18	143.27 ± 37.89	0.498
P value within group	< 0.001*	0.002*	
Difference	32.89 ± 44.42	26.25 ± 43.61	0.542
Pre HbA1c (%)	8.9 ± 1.31	8.92 ± 1.3	0.955
Post HbA1c (%)	7.61 ± 1.42	7.76 ± 1.57	0.689
P value within group	0.001*	0.001*	
Difference	1.28 ± 1.96	1.15 ± 1.81	0.781
Pre Cholesterol (mg/dl)	178.89 ± 29.36	186.67 ± 39.24	0.366
Post Cholesterol (mg/dl)	156.54 ± 37.47	159.47 ± 34.21	0.741
P value within group	< 0.001*	< 0.001*	
Difference	22.35 ± 31.82	27.2 ± 30.68	0.531
Pre Triglycerides (mg/dl)	128 (100.5 – 185)	149 (117.75 – 190)	0.412
Post Triglycerides (mg/dl)	143 (110.5 – 214.5)	145 (106.5 – 199.5)	0.758
P value within group	0.083	0.823	
Difference	-24.5 (-93 – 23.5)	-20 (-59 – 60.45)	0.308
Pre LDL (mg/dl)	117.59 ± 28.08	121.12 ± 35.11	0.654
Post LDL (mg/dl)	93.33 ± 32.04	97.69 ± 32.62	0.586
P value within group	< 0.001*	< 0.001*	
Difference	24.26 ± 30.13	23.43 ± 32.99	0.915
Pre HDL (mg/dl)	42.8 ± 9.24	42.24 ± 8.89	0.805
Post HDL (mg/dl)	45.49 ± 10.04	45.64 ± 9.16	0.949
P value within group	0.004*	0.002*	
Difference	-2.69 ± 4.95	-3.39 ± 5.85	0.599
Pre Non-HDL (mg/dl)	136.09 ± 30.35	144.42 ± 37.13	0.322
Post Non-HDL (mg/dl)	111.05 ± 35.29	113.83 ± 35.47	0.751
P value within group	< 0.001*	< 0.001*	
Difference	25.04 ± 32.98	30.59 ± 29.07	0.471
Pre eGFR (mL/min/1.73m2)	102.81 ± 14.38	101.72 ± 13.98	0.756
Post eGFR (mL/min/1.73m2)	107.93 ± 13.22	105.21 ± 11.29	0.371
P value within group	0.025*	0.082	
Difference	-5.12 ± 12.54	-3.48 ± 11.16	0.578

Data presented as [mean ± (SD), and median (Q1-Q3)]. LDL: low density lipoprotein.
* $P < 0.05$: Comparison pre and post treatment in each group.

The gliclazide add-on group showed significant reductions in SOD, MDA, and PC with significant increases in GSH levels compared to baseline levels, whereas the glimepiride add-on group showed significant reductions in SOD and MDA only. There were no significant differences between both groups regarding GSH (p =0.837), SOD (p = 0.053), and MDA (p = 0.775) after 16 weeks of treatment, except for PC level (p = 0.019). In addition, there was a non-significant numerically higher improvement in GSH (p = 0.534), SOD (p = 0.085), and MDA (p = 0.525) markers with a significant higher improvement in PC levels (p = 0.048) in the gliclazide group than the glimepiride group, as shown in Table 4.

Table 4. The impacts of studied treatments on oxidative stress markers of patients

Clinical variables	First group Add on Gliclazide (n=33)	Second group Add on Glimepiride (n=33)	P -value
Pre GSH (ng/ml)	1.33 (0.87 – 1.74)	1.24 (0.81 – 1.71)	0.734
Post GSH (ng/ml)	1.47 (1.3 – 1.78)	1.51 (1.32 – 1.65)	0.837
P value within group	0.029*	0.214	
Difference	-0.23(-0.64 – 0.13)	-0.2 (-0.54 – 0.28)	0.534
Pre SOD (U/L)	98.4 (62.92 –131.76)	77.44 (59.94–149.26)	0.346
Post SOD (U/L)	57.63(46.09 – 66.57)	61.67 (56.57 – 69.94)	0.053
P value within group	< 0.001*	0.026*	
Difference	46.73 (11.15 – 79.9)	15.38 (-8.72 – 84.13)	0.085
Pre MDA(nmol/ml)	8.06 ± 1.87	7.84 ± 1.93	0.638
Post MDA(nmol/ml)	5.64 ± 1.03	5.72 ± 1.09	0.775
P value within group	< 0.001*	< 0.001*	
Difference	2.42 ± 1.68	2.12 ± 2.06	0.525
Pre PC (ng/ml)	72.97 (67.75 – 81.44)	71.08 (63.04 – 84.54)	0.581
Post PC (ng/ml)	65.17 (57.10 – 72.82)	73.55 (65.65 – 77.69)	0.019**
P value within group	0.001*	0.538	
Difference	12.69 ± 23.1	2.17 ± 19.04	0.048***

Data presented as [mean ± (SD), and median (Q1-Q3)].
* P < 0.05: Comparison pre and post treatment in each group.
** P < 0.05: Comparison between groups.
*** P < 0.05: Comparison between groups' differences.

Discussion

Type 2 diabetes mellitus is a condition characterized by elevated oxidative stress, which has been linked to the development of diabetes and its complications. The regulation of glucose, lipids, and renal function are all severely impacted by increased free radical generation. So, measuring oxidative stress biomarkers is seen as a useful tool in addition to the currently used parameters to select proper treatment for type 2 diabetes patients [(40)]. To the best of our knowledge, there has been no previous study evaluating the antioxidant benefits of the glimepiride plus metformin combination versus gliclazide MR plus metformin combination, despite the common use of this combination in our daily clinical practice.

In this study, the SUs add on treatment resulted in non-significant change in anthropometric measurements and blood pressure in both groups with no difference between both groups. It is reported that modern SUs, gliclazide MR and glimepiride, have a neutralizing effect on body weight and cardiovascular risk [(41)].

Furthermore, the addition of gliclazide or glimepiride to metformin therapy in T2DM found no differential effects on arterial distensibility, endothelial function, or vasodilator mechanisms [(42)]. These findings come in accordance with The Zwolle Outpatient Diabetes project Integrating Available Care (ZODIAC-39) study done by Schrijnders *et al.* that neither found a significant change in body weight after treatment with SUs/metformin combination nor between varied SUs combinations [(43)]. A meta-analysis showed the SUs combined metformin therapy did not significantly decrease blood pressure [(44)].

The significant improvement of glycemic indexes in our study was driven by both combination treatment groups, and the difference between the two groups was not significant, with reductions in HbA1c (1.28–1.15%) and FBG (32.89–26.25 mg/dl) for the gliclazide MR and glimepiride groups, respectively. Our findings were compared to those of the Schernthaner *et al.* study, which found that adding gliclazide MR or glimepiride to inadequately

controlled patients on metformin or α-glucosidase inhibitor monotherapy resulted in decreases in HbA1c (1.1–1%) and FBS (1.4–1.3 mmol/L), respectively[23]. The effectiveness of adding gliclazide or glimepiride to metformin combination therapy with similar results to our finding for glycemic control were reported in different studies [45,46]. A systematic review showed the HbA1c reduction of gliclazide alone or in combination was not significantly different from other SUs with significantly less risk of hypoglycemia, but gliclazide reduced HbA1c significantly more than other insulinotropic agents with no significant difference in hypoglycemic risk [47].

Uncontrolled T2DM is associated with lipid disarrangements, and the relationship between dyslipidemia and coronary heart disease has been well established [48]. In our study, non-HDL, a measure of all atherogenic lipoproteins, was studied as a predictor of cardiovascular risk. Both groups showed a significant reduction in non-HDL levels, and this result reflects the significant reduction in total cholesterol levels after treatments. These findings aligned with the Hassan and Abd-Allah study, which reported that both gliclazide and glimepiride, in combination with metformin, significantly improved lipid levels in comparison to the control group [49]. Also, Banik *et al*. found a reduction of 5% in the atherogenic index in patients treated with the gliclazide and metformin combination [50].

Furthermore, the improvement of renal function in our patients after treatment was noticed in both groups, as represented by an increment in eGFR due to reduced creatinine levels. In the ADVANCE trial, intensive glycemic control with gliclazide MR reduced the progression of nephropathy by 21% [26]. Lee *et al.* looked at the safety of gliclazide and glimepiride regarding the risk of end stage renal disease (ESRD), and they found that only gliclazide was associated with a low risk of doubling of creatinine levels in patients with preserved kidney function [51].

In this study, we measured patients' serum levels of different OS markers and we observed a drop in GSH levels and increased SOD, MDA and PC levels before SUs add-on therapy with no significant difference between both groups. After 16 weeks of add on treatment, the levels of OS markers in both groups were arranged differently. The Gliclazide add-on group improved all markers significantly by lowering SOD, MDA, and PC levels and raising GSH levels. The Glimepiride add-on group, on the other hand, only significantly lowered SOD and MDA levels. Gliclazide group showed a significant ($p = 0.048$) difference in the improvement (reduction) of follow up PC values compared to glimepiride group. Moreover, the higher improvement of GSH, SOD, and MDA markers was in favor of gliclazide, even if the difference between the two groups was not significant. This indicates the antioxidant impact of gliclazide was greater than that of glimepiride. The partial improvement of OS in the glimepiride group could be attributed to its ability to regulate the antioxidant enzyme expression of catalase and SOD via agonistic activation of the peroxisome proliferator-activated receptor-γ (PPARγ); and reduction of redox-sensitive NF-kB activation [52–54].

Other studies have looked at the beneficial antioxidant effect of gliclazide when added to metformin therapy and showed similar findings to our results. A comparison study by L. Chen *et al.* showed that the combination of gliclazide and metformin therapy was significantly superior to metformin therapy in improving MDA, SOD, and circulating endothelial progenitor cells (EPCs) [55]. Also, Banik *et al.* found a significant improvement in both MDA ($p = 0.001$) and nitric oxide (NO) ($P = 0.015$) levels with gliclazide plus metformin in comparison to metformin alone [50]. Contrary to our findings, AlSharidah *et al.* found a non-significant differences between the gliclazide/metformin combination and metformin monotherapy in improving OS, lipid levels, and hepatorenal function, while glycemic control worsened with combined therapy, indicating the protective effect of combined therapy against OS and preserving the antioxidant activity even after glycemic deterioration [56].

A preliminary report by Nakamura *et al.* raised the possibility of glimepiride's antioxidant property due to a significant decrease in the levels of glyceraldehyde-derived advanced glycation end products (glycer-AGE) after 24 weeks of treatment [57]. Zhao *et al.* recently found a significant difference between combined glimepiride/sitagliptin therapy and glimepiride monotherapy in improving glycemic and OS indexes, even though both groups showed significant improvements compared to pretreatment levels [58]. On other hand, Bibra *et al*. found a non-significant ($p = 0.814$) reduction of MDA levels after 16 weeks of treatment with glimepiride plus metformin therapy in comparison with rosiglitazone plus metformin therapy [59]. Nomoto *et al.* also showed that the antioxidant effect of glimepiride on SOD, and BAP (biological antioxidant potential) was not as significant as sitagliptin in a randomized controlled trail [60].

In the current study, the partial improvement in OS markers in the glimepiride add-on group may be due to indirect action through controlling hyperglycemia, as glucose auto-oxidation is the primary source of free radicals, as well as the improvement in OS markers in the gliclazide add-on group being greater than that in the glimepiride add-on group, indicating its potent antioxidant activity independent of its glycemic control, as both groups are almost showing comparable glycemic control.

Limitation

The study had some limitations, like the participants were recruited from single center and there was no calculated–calorie diet program to be followed by the patients.

Conclusion

To sum up, in T2DM patients who are inadequately controlled with metformin monotherapy, the add-on therapy of gliclazide MR or glimepiride has comparable beneficial impacts on glycemic control, lipid profile, and kidney function, but gliclazide MR may be a preferable first-choice over glimepiride in clinical practice since its additional antioxidant property can probably preserve beta cell function and at least delay the vascular complications related to oxidative stress.

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Conflict of Interest

There are no conflicts of interest.

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Authors' Contributions

Fadhel A. Shihab designed the study, investigation, methodology, data analysis and interpretation, software, preparation of the original manuscript, writing review and editing. Ali L. Jasim supervised and revised the original manuscript, project administration. Adel Gh. Mohammed planned the study conception, participants' enrollment, management and follow-up. All authors reviewing and approving the final version of manuscript.

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