Effect of combined gliclazide/metformin treatment on oxidative stress, lipid profile, and hepatorenal functions in type 2 diabetic patients

Mansour Alsharidah a,b, Metab. Algeffar c, Abdel-Moneim Hafez Abdel-Moneim a,d,*, Mohamed Faisal Lutfi a, Haila Alshelowi e

a Physiology Department, College of Medicine, Qassim University, Saudi Arabia
b College of Pharmacy, Qassim University, Saudi Arabia
c Department of Family Medicine, College of Medicine, Qassim University, Saudi Arabia
d Physiology Department, Faculty of Medicine, Mansoura University, Egypt
e Department of Pediatric Endocrinology, Albassam Diabetes and Endocrine Center, Saudi Arabia

1. Introduction

Diabetes mellitus (DM) compromises cardiovascular (Chawla et al., 2016), kidneys (Narres et al., 2016), liver (Morling et al., 2015), functions and affects the antioxidant capacity of the body (Grindel et al., 2016). Recent studies have reported that most of diabetic complications result from disturbed oxidative stress in different body organs (Asmat et al., 2016) as well as dyslipidemia (de Souza Bastos et al., 2016). Understanding the mechanisms of diabetic complications constitutes the cornerstone of the evaluation of drug regimens used for the treatment of DM (Zhang et al., 2013).
In clinical practice, metformin and gliclazide are used either separately or in combination for the treatment of type 2 DM (T2DM). Metformin monotherapy was proved to improve metabolic disturbances (Chakraborty et al., 2011; Esteghamati et al., 2013) and attenuate oxidative stress (Kim et al., 2013) in diabetic patients. More recently, it has been proved that adding gliclazide to metformin-treated type 2 diabetic patients synergistically improved the fasting blood glucose (FBG) and lower glycosylated hemoglobin (HbA1c) level (Al-Gareeb et al., 2016). However, there is a lack of sufficient reports on the therapeutic benefits of combined metformin/gliclazide treatment on oxidative stress, lipid profile, and hepatorenal functions (Hong et al., 2013). Moreover, there is a scarcity of research dealing with the combination therapy of metformin/gliclazide for T2DM management. The present study aims at comparing the major therapeutic potentials of combined metformin/gliclazide treatment and metformin monotherapy when used for T2DM patients.

2. Subjects and methods

2.1. Subjects

The study recruited 80 T2DM patients from Al Basam Diabetes Center – Unaiza – Qassim – KSA during the period from March 2015 to March 2016. The control group consisted of 40 T2DM patients on metformin treatment (500 mg BD). The metformin monotherapy group was matched for age and duration of DM with a test group of 40 patients on combined metformin/gliclazide therapy (combined therapy group) (500 mg BD + 80 mg OD, respectively). The patients with other systemic diseases, major diabetic complications, treatment with beta blocker, steroids, thiazides, and/or insulin were excluded from the study. The study was approved by the Institutional Review Board (IRB) of College of Medicine, Qassim University.

2.2. Methods

Five ml of venous blood were taken from each patient during their regular follow-up in the general diabetes clinic. To determine HbA1c level, 1.5 ml of the blood sample was added to EDTA tube. The blood serum was separated from the remaining 3.5 ml to determine oxidative stress status, FBG level, hepatorenal function, and lipid profile.

HbA1c level was determined according to the standard technique (Bunn et al., 1976) using a specific glycated hemoglobin kit (Sigma–Aldrich, St. Louis, MO, USA). FBG level was determined in mmol/L with the standard oxidase methods (Barham and Trinder, 1972). The extent of lipid peroxidation was determined colorimetrically (Ohkawa et al., 1979) using lipid peroxide (LPD) kit (GenWay Biotech, Inc, USA). Total antioxidant capacity (TAC) was measured colorimetrically (Koracevic et al., 2001) using TAC kit (GenWay Biotech, Inc, USA). Total cholesterol was measured using enzymatic, colorimetric test – CHOD/PAP method with MIDI diagnostics kits (CAT.NO EL24-1200, KSA) (Fossati and Prencipe, 1982). Serum triglycerides were measured through enzymatic colorimetric GPO method with MIDI diagnostics kits (CAT.NO EL59L-1000, KSA) (Warnick and Albers, 1978). LDL was indirectly measured using the Friedewald equation (Nauck et al., 2002) based on the presence of total cholesterol, HDL, and triglyceride levels [LDL = total cholesterol – HDL – (triglycerides/5)]. Serum creatinine was determined according to the standard colorimetric method (Fabiny and Ertingshausen, 1971). Aspartate (AST) and alanine (ALT) transaminase (IU) were assessed colorimetrically (Reitman and Frankel, 1957).

2.3. Statistical analysis

Data were analyzed using SPSS for Windows (version 16.0, Chicago, SPSS Inc. USA). Normal distribution of variables was examined using Shapiro-Wilk test. The studied variables were described with median and 25–75 interquartile (Q1–Q3) and Box-plot charts. Significant statistical differences of studied variables were assessed among diabetic groups using Mann-Whitney U test. A P value of <.05 was considered significant.

3. Results

The duration of T2DM and age at the time of diagnosis of the diabetic patients undergoing metformin treatment (Median (Q1–Q3) = 50.00 (45.00–57.75) and 2.00 (1.00–5.00) years, respectively) were not significantly different as compared to those in the combined therapy group [Median (Q1–Q3) = 53.00 (45.25–60.00) and 4.00 (2.00–6.75) years, respectively; P > .05]. Patients on met-

Table 1
Comparison of glycemic control, lipid profile, hepatorenal functions and oxidative stress among diabetic patients on mono and combined anti-diabetic therapy.

| Glycemic control | Diabetic patients on combined therapy N = 40 Median (Q1–Q3) | Diabetic patients on Metformin monotherapy N = 40 Median (Q1–Q3) | P |
|------------------|----------------------------------------------------------|--------------------------------------------------------------|---|
| FBG (mmol/L)     | 9.00 (7.30–10.68)                                        | 7.61 (6.70–8.89)                                             | .022 |
| HBA1c (%)        | 8.20 (7.20–9.75)                                         | 7.00 (6.40–7.65)                                             | <.001 |
| Oxidative stress|                                            |                                                             |    |
| LPD (nmol/dl)    | 22.00 (20.00–23.00)                                      | 21.00 (20.00–24.00)                                          | .888 |
| TAC (nmol/ml)    | 70.94 (67.37–73.02)                                      | 70.64 (67.37–74.41)                                          | .751 |
| Lipid profile    |                                            |                                                             |    |
| Triglycerides (mmol/L) | 1.70 (1.13–2.58)                                         | 1.50 (1.30–2.16)                                            | .582 |
| Cholesterol (mmol/L)  | 5.00 (4.50–5.60)                                        | 5.00 (4.20–5.68)                                            | .599 |
| HDL (mmol/L)     | 2.65 (2.01–3.60)                                         | 2.65 (2.00–3.40)                                            | .630 |
| LDL (mmol/L)     | 1.05 (0.94–1.38)                                         | 1.20 (0.94–1.50)                                            | .498 |
| Hepatorenal functions |                                             |                                                             |    |
| ALT (IU)         | 22.50 (17.70–34)                                         | 24.00 (15.00–34.00)                                          | .841 |
| AST (IU)         | 18.00 (14.93–21.75)                                      | 16.00 (14.00–23.50)                                          | .898 |
| Creatinine (mmol/L)| 58.50 (47.25–75.00)                                    | 58.00 (43.00–71.00)                                          | .563 |

FBG: fasting blood glucose; HBA1c: haemoglobin A1c; LPD: lipid peroxidase; TAC: total antioxidant capacity; HDL: high density lipoproteins; LDL: low density lipoproteins; ALT: alanine transaminase; AST: aspartate transaminase.

*Statistically significant.
formin treatment exhibited significantly lower levels of FBG [7.61 (6.70–8.89) mmol/L vs. 9.00 (7.30–10.68) mmol/L, P = .022] and HBA1c [7.00 (6.40–7.65)% vs. 8.20 (7.20–9.75)%, P < .001] compared to those on combined therapy (Table 1).

Glycemic control (Fig. 1), oxidative stress (Fig. 2), lipid profile (Fig. 3), and hepatorenal functions (Fig. 4) were comparable in patients in mono and combined therapy groups (P > .05) (Table 1).

As shown in Fig. 1, the FBG (7.61 mmol/L) and HBA1c (7%) levels of metformin monotherapy group were lower compared to the levels of those on combined therapy.

Fig. 2 showed improved results for LPD (21 nmol/dL) levels in patients undergoing metformin monotherapy. However, there was no significant difference in the TAC of the studied groups.
The triglycerides level in combined therapy group and LDL levels in monotherapy group were found to have significantly higher values (Fig. 3). However, there was no difference in the levels of HDL and cholesterol in both the groups.

AST level in the monotherapy group was lower compared to that of the combined therapy group (Fig. 4). In contrast, ALT level was higher in monotherapy group while creatinine level showed no improvements in both studied groups.

4. Discussion

The combined metformin/gliclazide therapy is commonly used for treating T2DM; however, the therapeutic benefits of this combination on oxidative stress, lipid profile, and hepatorenal functions have not been thoroughly studied before. In the present study, it is evident that the above-mentioned parameters were comparable in both studied groups. Glycemic control was poor in
the diabetic patients undergoing combined therapy compared to those on metformin monotherapy.

Nowadays, oxidative stress in diabetic patients is the cornerstone for the pathogenesis of all complications in diabetes (Asmat et al., 2016). Increased free radical production greatly affects glycemic control, lipid profile, and hepato-renal function in diabetic patients. The observed insignificant differences between monotherapy and combined therapy groups in this study is consistent with the results of previous studies (Meis-goli and Tuerkeli, 2008), which demonstrated that separate (not combined) administration of metformin or gliclazide greatly improved oxidative stress status in diabetic patients. Previous reports proved that administration of gliclazide or metformin decreased oxidative stress as evidenced by decreased catalase (CAT), glutathione S-transferase (GST), erythrocytes glutathione peroxidase (Gpx) levels, and malondialdehyde (MDA) levels with no significant differences between gliclazide or metformin-treated groups (Meis-goli and Tuerkeli, 2008). Moreover, Chen and his colleagues also recommended the use of combined gliclazide/metformin therapy over metformin monotherapy for better oxidative stress status in T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010).

In conclusion, the combined metformin/gliclazide treatment for better oxidative stress status in T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010). Contrary to the results of T2DM patients as evidenced by improved MDA and superoxide dismutase (SOD) levels (Chen et al., 2010).
Warnick, G.R., Albers, J.J., 1978. A comprehensive evaluation of the heparin-manganese precipitation procedure for estimating high density lipoprotein cholesterol. J. Lipid Res. 19, 65–76.

Zhang, F., Xiang, H., Fan, Y., Ganchuluun, T., Kong, W., Ouyang, Q., Sun, J., Cao, B., Jiang, H., Nie, S., 2013. The effects of sulfonylureas plus metformin on lipids, blood pressure, and adverse events in type 2 diabetes: a meta-analysis of randomized controlled trials. Endocrine 44, 648-658. https://doi.org/10.1007/s12020-013-9970-6.