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Full Length Research Paper

Effects of metformin and metformin in combination with omega-3 on newly diagnosed type 2 diabetic Iraqi patients

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ABSTRACT

Type 2 diabetes is defined as a syndrome characterized by insulin deficiency, insulin resistance and increased hepatic glucose output. To compare the effects of an insulin sensitizer, metformin and metformin in combination with omega-3 on blood glucose level, serum insulin and oxidative stress state in newly diagnosed type 2 diabetes. This is an open-label, randomized study carried out on 24 newly diagnosed type 2 diabetic patients. Patients were randomly divided into two groups and assigned for treatment with either metformin (n=12) or metformin in combination with omega-3 (n=12) for 2 months. The level of fasting blood glucose (FBG), post-prandial blood glucose (PPG), glycated hemoglobin (HbA1c), serum insulin and serummalondialdehyde (MDA) were calculated before and after one month and two months of treatment. FBG, post prandial blood glucose and HbA1c significantly decreased in both treated groups after one month and two months of treatment. Serum insulin level decreased non-significantly with metformin and metformin in combination with omega-3 after one and two months of treatment. The level of serum MDA decreased significantly in group treated with metformin in combination with omega-3 after one month and two months of treatment and in metformin treated group after two months, while insignificant decrease observed after one month in metformin treated group. Omega-3 has no significant effect on glycemic control and insulin secretion but has beneficial effect on oxidative stress state.

Keywords: Type 2 DM, MDA, metformin, omega-3.

INTRODUCTION

Diabetes is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The chronic hyperglycemia of diabetes is associated with long-term damage, dysfunction, and failure of different organs, especially the eyes, kidneys, nerves, heart, and blood vessels (The Committee on The Diagnosis and Classification of Diabetes Mellitus, 2003). Several pathogenic processes are involved in the development of diabetes. These range from autoimmune destruction of

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the β-cells of the pancreas with consequent insulin deficiency to abnormalities that result in resistance to insulin action (Alberti et al., 1998).

Type 2 diabetes mellitus is associated with multiple metabolic derangements that result in the excessive production of reactive oxygen species and oxidative stress. These reactive oxygen species set in motion a host of redox reactions which can result in unstable nitrogen and thiol species that contribute to additional oxidative stress. Multiple cellular studies have shown that under oxidative stress conditions, insulin signaling is impaired, resulting in insulin resistance of the cell (Eriksson, 2007).

Metformin is insulin-sensitizing and antihyperglycemic agent used in the treatment of type 2 diabetes mellitus. The main mechanisms include anorexiogenesis, reduction of intestinal carbohydrate absorption, inhibition of hepatic gluconeogenesis, as well as increased glucose uptake by peripheral tissues (Edgerton et al., 2009).Omega-3 fatty acids are essential because they are not synthesized by the body and must be obtained through diet or supplementation, omega-3 improves several metabolic abnormalities underlying the development of diabetes mellitus (DM), such effects include insulin-sensitizing effects via increased production and secretion of adipocytokines such as adiponectin and leptin (Banga et al., 2009); and potential prevention of insulin resistance via anti-inflammatory effects mediated directly (Oh et al., 2010) or through conversion to specialized pro-resolution mediators such as resolvins and protectins(Gonzalez-Periz et al., 2009).

In the present study, we sought to investigate the potential effects of omega-3 with metformin on glycemic parameter and lipid peroxidation in newly diagnosed type 2 Iraqi diabetic patients.

MATERIALS AND METHODS

Patient Selection

This study was carried out on twenty four patients (males and females) with type 2 diabetes mellitus (DM); their ages range 40-55 years with newly diagnosed disease. They are randomly selected and assigned either to metformin, or metformin in combination with omega-3. They are maintained on dietary control program under the supervision of clinical nutrition specialist at National Diabetes Center for Treatment and Research/Al-Mustansiriyah University. All subjects were diagnosed with T2DM in accordance with the WHO diabetes diagnostic criteria of 1999 and had never been treated before.

METHOD

After 12 hours overnight fasting, blood samples were analyzed for FBG, HbA1c, fasting insulin, MDA, and then a solid meal was given and the blood glucose level was measured after 2 hours to calculate post-prandial blood glucose. All subjects were orally administered with either metformin 500 mg given three times daily in a tablet dosage form or metformin 500 mg given three times daily and 1000 mg omega-3 given two times daily in a soft gel form. After one month and 2 months of the treatment, we observed the changes in these parameters. FBG measured by using ready-made kit based on enzymatic colorimetric method, post prandial blood glucose measured by using blood glucose monitoring system (Accu-chek active roche). HbA1c determined by high- performance liquid chromatography (HPLC)(Bio-Rad Variant, U.S.A). Serum insulin was measured by using ready-made kit. The insulin kit is a solid phase enzyme-linked immunosorbent assay (ELISA) based on the sandwich principle. Serum MDA was measured by using ready-made kit(ELISA). This assay employs the competitive inhibition enzyme immunoassay technique.

Statistical Analysis

Data are expressed as means ± SD. Statistics were performed using SPSS (version 19). Differences from baseline were assessed by the paired Student's *t* test. A P-value of <0.05 was considered significant.

RESULTS

Patients

Of 24 patients randomized to treatment, 12 in the metformin group, 12 in the metformin in combination with omega-3 group. All patients received the required doses of the study drug. There were no apparent differences between the two groups with respect to demographic and baseline characteristics (Table 1).

Efficacy

Changes from baseline to the end of the study are summarized in Table 2. FBG, PPG, HbA1c were significantly ($p<0.05$) decreased in both groups after one month and two months of treatment. Treatment with metformin in combination with omega-3 showed no significant difference when compared with metformin treated group ($p>0.05$) whether after one month or two

Table 1. Patient characteristic at baseline

Characteristic	Metformin	Metformin+Omega-3
n=24	12	12
Age; years	40-55	40-55
FSG (mg/dl)	214.58±13.41	220.50±12.85
PPG (mg/dl)	279.92±12.75	272.17±10.73
HbA1c %	9.14±1.43	9.02±1.74
Insulin (µIU/ml)	14.30±2.82	15.21±1.90
MDA (µmol/l)	4.41±2.45	3.94±1.62

Table 2. Changes from baseline and after 1month and 2 months in glycemia and serum MDA

Variable/time point	Metformin only	Metformin in combination with omega-3
FPG (mg/dl)		
Baseline	214.58±13.41	220.50±12.85
After 1 month	177.83±13.76*	174.33±15.40*
Change from baseline	(17.12%)	(20.93%)
After 2 months	140.17±10.25*	138.92±10.22*
Change from baseline	(34.67%)	(36.99%)
PPG (mg/dl)		
Baseline	279.92±12.75	272.17±10.73
After 1 month	231.83±14.54*	227.00±11.24*
Change from baseline	(17.17%)	(16.59%)
After 2 months	196.08±13.31*	189.67±15.70*
Change from baseline	(29.95%)	(30.31%)
HbA1c (%)		
Baseline	9.14±1.43	9.02±1.74
After 1 month	7.83±1.12*	7.65±1.90*
Change from baseline	(14.33%)	(15.18%)
After 2 months	6.70±0.80*	6.55±1.21*
Change from baseline	(26.69%)	(27.38%)
Insulin (µIU/ml)		
Baseline	14.30±2.82	15.21±1.90
After 1 month	14.15±1.16	14.93±1.26
Change from baseline	(1.04%)	(1.84%)
After 2 months	13.98±1.24	14.74±1.37
Change from baseline	(2.23%)	(3.09%)
MDA(*M*mol/L)		
Baseline	4.41±2.45	3.94±1.62
After 1 month	3.64±1.78[a]	2.32±0.95*[b]
Change from baseline	(17.46%)	(41.11%)
After 2 months	2.88±1.64*[a]	1.33±0.65*
Change from baseline	(34.69%)	(66.24%)

Data are given as mean ± SE for baseline and end of study values for change from baseline; *significantly different compared to baseline level (P<0.05); values with non-identical superscripts (a,b) among different groups are considered significantly different (P<0.05).

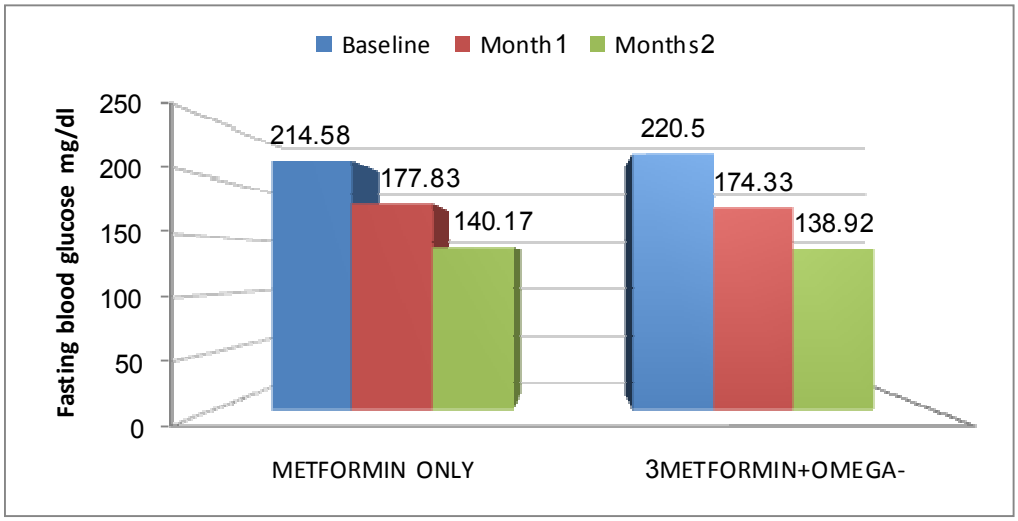

Figure1. Effects of metformin and metformin in combination with omega-3 on fasting blood glucose in type 2 diabetic patients.

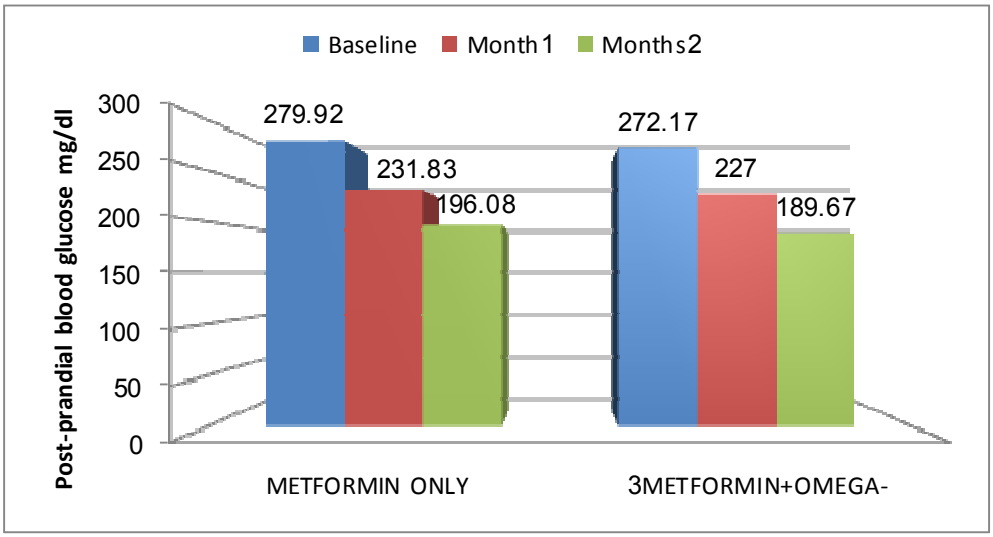

Figure 2. Effects of metformin and metformin in combination with omega-3 on post-prandial blood glucose level in type 2 diabetic patients.

months of treatment, with respect to the change in FBG, PPG, or HbA1c.Treatment with metformin alone or in combination with omega-3 produced slightly but no significant decrease in serum insulin level ($p>0.05$), with no significant difference between these two groups after one month and two months of treatment ($p>0.05$). Treatment with metformin in combination with omega-3 produced significant decrease in serum MDA level compared to baseline values ($p<0.05$) after one month and two months of treatment, and significant reduction observed in metformin treated group after two months of treatment ($p<0.05$), meanwhile insignificant decrease observed after one month of treatment with metformin ($p>0.05$). Treatment with metformin in combination with omega-3 showed significant difference when compared with metformin treated groups after one month and two months of treatment ($p<0.05$).

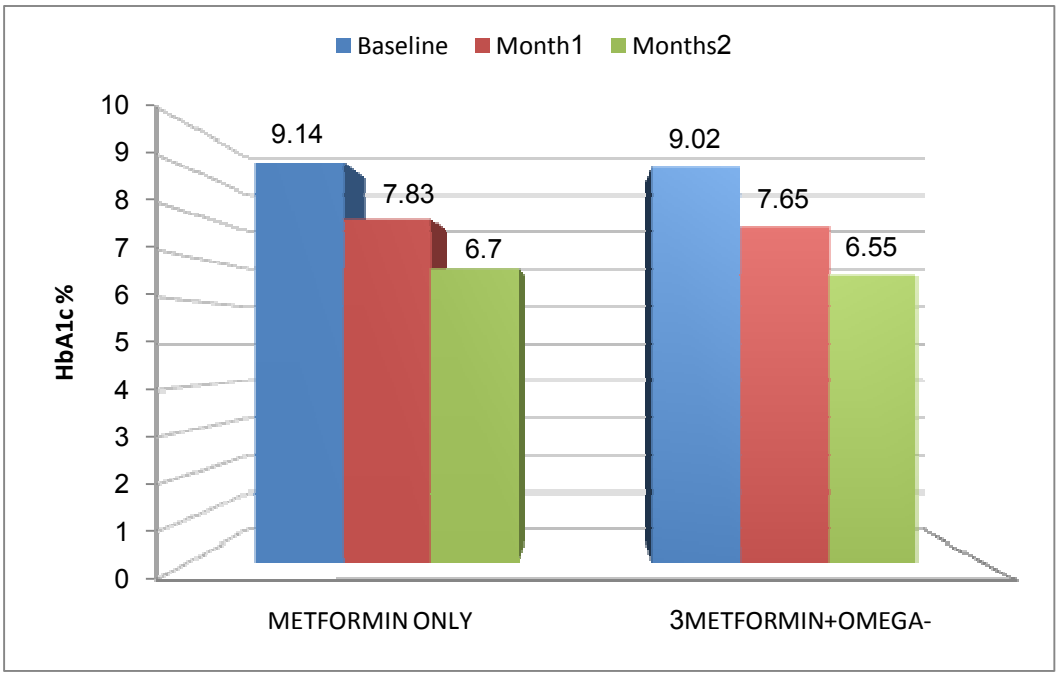

Figure 3. Effects of metformin and metformin in combination with omega-3 onglycated hemoglobin in type 2 diabetic patients.

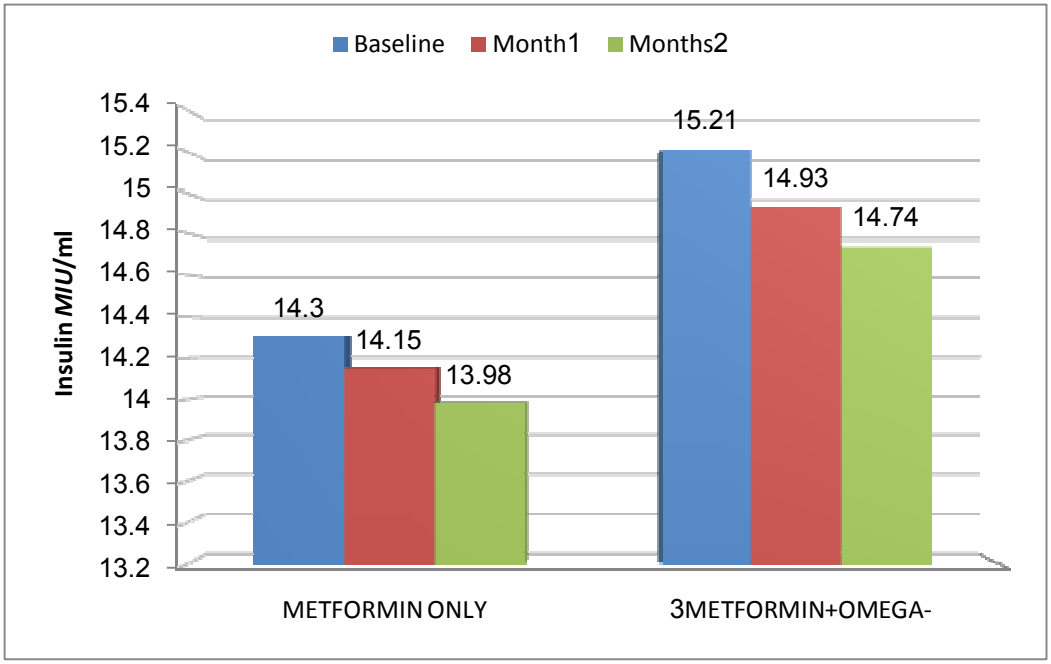

Figure 4. Effects of metformin and metformin in combination with omega-3 on serum insulin level in type 2 diabetic patients.

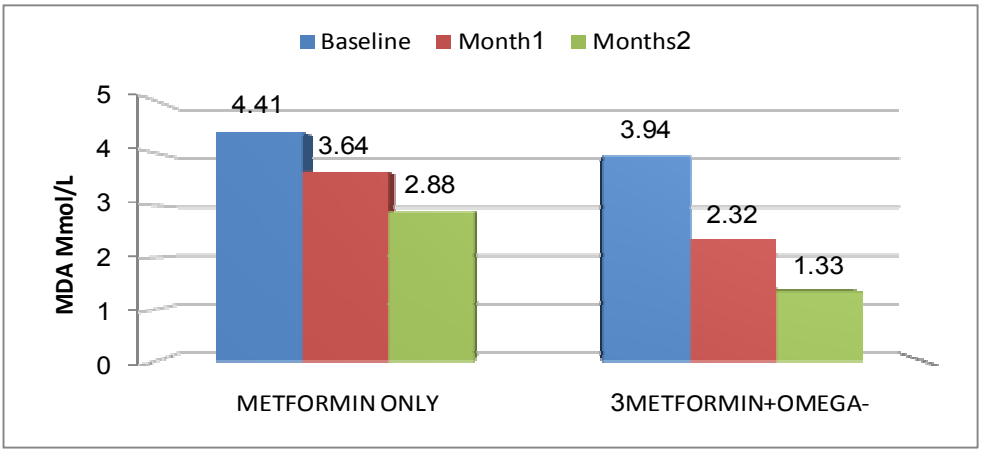

Figure 5. Effects of metformin and metformin in combination with omega-3 on serum MDA level in type 2 diabetic patients.

DISCUSSION

In the present study, our results regarding plasma glucose and glycosylated hemoglobin (HbA1c) indicate that there was a significant improvement in plasma glucose levels and HbA1c after one month and two months of treatment with metformin treated group, our results in agreement with previous studies (Aschner et al., 2010),(Bennett et al., 2010). The improvements in glycemic parameters with metformin alone and in combination with omega-3 were similar. This effect may be related to mechanism of action of metformin treatment which is an insulin-sensitizing drug, it mainly acts by decreasing hepatic glucose production (He et al.,2009),(Beatriz et al.,2013), increase skeletal myocyte glucose uptake (Turban et al., 2012), (Esfahanian et al., 2012), and by increasing insulin sensitivity in muscles and liver (Teranishi et al., 2007). The results in the present study showed that the combination of omega-3 with metformin had no statistically significant effect on glycaemic control, this is supported by reports of a non-significant change in fasting glucose after consumption of omega-3 fatty acids or fish oils (Harding et al., 2004), other study also found that administration of fish oil supplementation at relatively lower doses (1 to 2 g/day) for a period from 2-32 weeks may not have detrimental effect on glycemic status (Holman et al., 2009).In contrast, the other reports found a significant increase in HbA1c and fasting glucose in type 2 diabetes patients after omega-3 fatty acids supplementation (Hu et al., 2003). While, others observed that fasting blood glucose and HbA1c declined after twelve weeks of treatments with 2 g of omega-3 fatty acids in overweight diabetic patients (Mahmoud et al., 2013). Our results supported by other studies, demonstrated that metformin only slightly reduce fasting serum insulin levels compared to baseline values (Derosaet al., 2012), and its glucose lowering effect on increasing peripheral glucose uptake and reducing gluconeogenesis is mediated by the potentiation of insulin action (Shashank, 2005),(Enrique et al., 2009). The results of the present study showed that there was no difference in fasting serum insulin level between metformin alone and in combination with omega-3 treated groups. This agree with other previous studies that approved omega-3 supplementation has no effect on fasting serum insulin level in type 2 diabetic patients (Nettleton et al., 2005). Oxidative stress is proposed to be an early event in the pathology of DM and may influence the onset and progression of late complications (Singh et al., 2008). Furthermore, elevated levels of serum MDA, as a marker of oxidative stress, have been reported in type 2 diabetes mellitus (Mahreen et al., 2010).The present work disclosed a significant decrease in serum MDA levels in newly diagnosed type 2 DM patients after two months treatment with metformin group. These results were in agreement with the study conducted by (Pavlović et al., 2000) who reported a significant reduction in MDA levels in both erythrocytes and plasma in newly diagnosed patients with type 2 DM who had been on metformin treatment for a period of 4 weeks. These results were also in agreement with those reported by (Tessier et al., 1999) who observed that metformin treatment significantly decreased serum MDA levels in adult diabetic patients after a period of treatment for 24 weeks. Our results showed that treatment with metformin in combination with omega-3 resulted in significant decrease in serum level of MDA after one month and two months of treatment. Meanwhile, metformin in

combination with omega-3 showed significant difference when compared with metformin group for the same period of treatment. Omega-3 fatty acid supplementation led to a significantly lower the level of MDA compared to the baseline values in type 2 diabetic patients in consistent with previous studies (Kesavulu et al., 2002).Potential mechanisms for the decrease in MDA level may be related to the assembly of omega-3 fatty acids in membrane lipids and lipoproteins making the double bonds less available for free radical attack, inhibition of the pro-oxidant enzyme phospholipase A2 and stimulation of anti-oxidant enzymes (Mori et al., 2003).

CONCLUSION

- Omega-3 has no significant effect on glycemic control and insulin secretion but has beneficial effect on oxidative stress state.

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