

EFFECT OF COMBINATION OF *NIGELLA SATIVA* AND *TRIGONELLA FOENUM-GRAECUM* WITH GLIBENCLAMIDE ON SERUM TRIGLYCERIDE, HDL AND CREATININE LEVELS IN TYPE-2 DIABETES MELLITUS PATIENTS

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ABSTRACT:

To evaluate the effect of combination of *Nigella sativa* and *Trigonella foenum-graecum* seeds with Glibenclamide on serum triglyceride, HDL, and creatinine levels in type-2 diabetes mellitus patients. Diabetic outpatient clinics of Isra University Hyderabad and Liaquat University of Medical and Health Sciences Jamshoro, from March to August 2008. For this study Type-2 diabetic patients on Glibenclamide, who gave written consent to volunteer in the study, were randomly divided into two groups. Fifty patients in group A (Control Group) were maintained on routine dose of Glibenclamide, while fifty patients in group "B" (Intervention group) were given capsules containing equal amount of combined powder of *N-sativa* and *T. foenum-graecum* seeds, in addition to their routine dose of Glibenclamide. Serum triglyceride, HDL, and creatinine level were measured for every patient at the start and after 3 months of therapy. Comparison of the mean values of parameters measured in both the groups showed that serum HDL levels were significantly ($P < 0.05$) raised in intervention group as against the control, whereas the levels for serum triglycerides was comparable in both the groups. Results also showed that no significant change occurred in serum creatinine level in both the groups. This study provides evidence that the combination therapy of *N. sativa* and *T. foenum-graecum* with Glibenclamide is beneficial for type-2 diabetic patients as it increases serum HDL level in these patients.

Keywords: *Nigella-sativa*, *Trigonella foenum-graecum*, Glibenclamide, HDL, Triglyceride, Type-2 diabetes mellitus.

INTRODUCTION

Diabetes Mellitus Type-2 is a common disorder which develops in man in the middle or later part of the life. It is recorded that diabetes involved about 190 million peoples globally and this figure may increase up to 342 million by the year 2025 (Zimmet, 2005).

Uncontrolled Diabetes Mellitus is associated with marked increased risk of atherosclerotic vascular disorders (macro-

vascular complications), including coronary, cerebrovascular and peripheral artery diseases. These are responsible for about 70% of deaths in patients with type-2 diabetes. (Yamagishi SI., 2007). Beside hyperglycaemia, dyslipidaemia is also an important risk factor for atherosclerotic vascular disorders in patients with type-2 diabetes mellitus. Dyslipidaemia in diabetic patients is characterized by moderate hypertrigly-ceridaemia and decreased level of high density lipoprotein (HDL) cholesterol (Chahil TJ., 2006).

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The beneficial effects of oral Anti-diabetic drugs (allopathic drugs) on glycemic levels are well documented but these drugs can not prevent progressive nature of complications of diabetes associated with dyslipidaemia in all the cases and these have their own side effects (KIM JD *et al.*, 2006).

Now a days, the attractiveness of complementary medicine has been increased (KIM JD *et al.*, 2006) and herbs / plants are used to cure different ailments as these have relatively fewer side effects (Gilani AH., 2005). *Nigella sativa* and *Trigonella foenum-graecum* (Abdel-barry JA., 2000) are two important plants used traditionally in the treatment of diabetes. *N. sativa* has shown its beneficial effects in correcting dyslipidaemia in many animal studies (Le PM., 2004; Badary OA., 2009). Similarly, the anti-hyperlipidemic properties of *T. foenum-graecum* seed powder have been documented in various animal and human trials (Badary OA., 2009).

The aim of the present study was to investigate the effect of combination of *N. sativa* and *T. foenum-graecum* seeds powder with Glibenclamide on serum triglyceride, HDL, and creatinine levels in type-2 diabetes mellitus patients.

SUBJECTS AND METHODS

Study setting: This clinical trial was conducted in the Diabetic outpatient clinics of Isra University Hyderabad and Liaquat University of Medical and Health Sciences Jamshoro, from March to August 2008.

Inclusion Criteria: All type-2 diabetes mellitus patients on Glibenclamide with Fasting blood sugar level more than 131mg/dl and HbA1c more than 7% were included in the study.

Exclusion Criteria: Followings were excluded from study:

- Patients with any other endocrine dysfunction
- Pregnant or lactating women

- Hypertensive patients
- Patients on steroid therapy.

Study population: One hundred type-2 diabetes mellitus patients who fulfilled the inclusion and exclusion criteria were included in the study. All the patients were advised to take their usual meal and maintain their daily routine. Ethics review committee / institutional review board approval was obtained for the study. Written informed consent was obtained from all patients before start of trial. Selected patients were randomly divided into two groups.

Group A (Control): This group comprised of fifty type-2 diabetic patients maintained on routine dose of Glibenclamide only.

Group B (Intervention Group): This group comprised of fifty type-2 diabetic patients kept on capsules, containing equal amount of combined powder of *Nigella sativa* and *Trigonella foenum-graecum* seeds, in addition to their routine dose of Glibenclamide.

Period of study: Each patient from both the groups was treated for a period of 3 months.

Preparation of Drugs: One tablet of Glibenclamide (Daonil) composed of 5 mg of Glibenclamide.

One herbal capsule used in intervention group contained powder of combination of crushed *N. sativa* seeds (250 mg) and *T. foenum-graecum* seeds (250 mg). Authentic seeds of *N. sativa* and *T. foenum-graecum* were purchased from local market, identified and confirmed by department of Botany University of Sindh Jamshoro. Seeds, after cleaning and shade drying were grounded in a mechanical grinder and passed through 80 mesh sieve. Powder obtained in each case was kept separately in a glass jar so that it dried completely. The dried powders were then filled in capsules in equal amounts i.e. 250 mg for each of *N. sativa* and *T. foenum-graecum*.

According to fasting blood sugar (FBS) levels, doses prescribed to patients in Group A and B are depicted in Table 1.

Serum Triglycerides level determination:

Serum triglyceride levels were determined spectrophotometrically by Peroxides method, using commercial kit (Greiner Diagnostic GmbH, Bahlingen-Germany). For this purpose, overnight fasting blood samples were collected from every patient before the start of treatment, and after the three months of treatment.

Serum HDL level determination:

HDL levels were measured in serum samples obtained from overnight fasting blood samples of patients, on day one and after three months of treatment. HDL level was estimated spectrophotometrically by Direct Polymer / Detergent method, using commercial kit (Bio-systems S.A. Costa Brava 30, Barcelona Spain).

Serum Creatinine level determination:

Serum creatinine levels were measured in samples collected before and after the three months of treatment by Jaffy's method, using inclusion and exclusion criteria, using commercial kit (DiaSys Diagnostic System GmbH, Holzheim Germany).

Statistical Analyses:

In this study, Serum triglyceride, HDL and Creatinine levels have been statistically compared between groups and within groups for differences in mean values at different intervals, so statistical test "Repeated Measure ANOVA (Analysis of Variance)" using General Linear Model in SPSS. Since Serum triglyceride, HDL, and Creatinine levels were analyzed at two different time intervals, so according to rules of applied statistical test p-

Table-1

Doses prescribed to group A & B patients, according to their fasting blood sugar levels (FBS)

FBS level (mg/dl)	Group A (Control)	Group B (Intervention)
	● Glibenclamide dose	■ Combined dose of <i>N. sativa</i> & <i>Trig foenum-graecum</i> with Glibenclamide
131-150	5 mg (1 tablet)	5 mg + 1000 mg (2 capsules)
151—180	10 mg (2 tablet)	10 mg + 1000 mg (2 capsules)
181-200	15 mg (3 tablet)	15 mg + 1500 mg (3 capsules)
200-230	20 mg (4 tablet)	20 mg + 2000 mg (4 capsules)
231 or more	20 mg (4 tablet)	20 mg + 2000 mg (4 capsules)

- 1. One tablet of Glibenclamide contains 5 mg of Glibenclamide
- 2. One capsule of combined powder of *N. sativa* and *Trig foenum-graecum* contains 500 mg, 250 mg of each

Table-2

Demographic characteristics of patients by group

Characteristics	Group		p-value
	A	B	
<u>Age Group:</u>			0.160
<30			
30 – 39	7	1	
40 & above	43	11	
Mean Age (S.E.M)	51±1.36	38 48±1.50	
<u>Sex:</u>			0.687
Male	21	19	
Female	29	31	

value i.e. <0.05 was divided by 2 (no of time intervals) to obtain level of significance, which came to be <0.025 .

RESULTS

In Table-2, demographic characteristics of patients of both the groups are depicted. Majority of the patients in both the groups were above 39 years of age and were female. No significant differences were noticed between groups for gender and mean age.

In Table-3, Serum triglyceride, HDL and Creatinine levels for control and intervention group patients are depicted. After 3 months of treatment Serum triglyceride levels between groups (p-value=0.075) was comparable. But the table reveals non-significant beneficial effect of combined therapy on serum triglyceride levels. However between control and intervention groups significant difference occurred in HDL levels after 3 months of treatment (p-value=0.013). The table also reveals non-significant effect of combined therapy for the period of 3 months on Serum creatinine level (p-value=0.381).

DISCUSSION

Diabetes control and complication trial had demonstrated that intensive blood glucose control successfully delay the onset and retard the progression of diabetic retinopathy, nephropathy and cardiovascular diseases (CVD).

CVD and other atherosclerotic vascular disorders are responsible for about 70% of deaths in patients with type-2 diabetes (Yamagishi SI., 2007). Dyslipidaemia in association with hyperglycemia is an important risk factor for CVD in type 2 diabetes mellitus patients. Moderate hypertriglyceridemia and low HDL-cholesterol level are two important indicators of dyslipidaemia in patients with type-2 diabetes mellitus (Chahil, 2006). Other factors such as total serum cholesterol and LDL-cholesterol (not measured in this study) do contribute to this risk which is perhaps twice as high as it is for non diabetic person (Brinton, 2005).

Oral anti diabetic agents used in the treatment of type 2 diabetes such as Sulfonylureas, Biguanides, α -glucosidase inhibitors, Thiazolidinediones and Glinides, which are used as monotherapy or in combination have many side effects (Haberbosch W., 2007), and only one of these agents i.e. Thiazolidinediones, the newer oral antidiabetic drugs provide good glycemic control along with beneficial effects on dyslipidemia (Haberbosch, 2007). However, some recent studies have indicated that some of these agents are associated with risk of myocardial infarction (Nissen, 2007) and heart failure (Granberry, 2007). *N. sativa* has shown its beneficial effects in correcting hyperglycemia and dyslipidemia by enhancing glucose induced insulin release from beta cells in the islets of langerhans (Le, 2004). It exerts an insulin sensitizing action by enhancing the

Table-3
Serum triglyceride, HDL, and creatinine levels (mg/dl) in control and intervention group at base line and after 03 months of treatment

Parameter		Control Group (Mean \pm SEM)	Intervention Group (Mean \pm SEM)	* p-value
Triglyceride	Base line	184.00 \pm 9.16	171.00 \pm 10.75	0.359
	After 03 months	190.00 \pm 8.68	169.00 \pm 8.33	0.075
HDL	Base line	35.86 \pm 0.68	36.70 \pm 0.70	0.393
	After 03 months	35.32 \pm 0.73	38.16 \pm 0.86	0.013
Creatinine	Base line	0.87 \pm 0.02	0.914 \pm 0.03	0.235
	After 03 months	0.82 \pm 0.04	0.86 \pm 0.03	0.381

* p-value <0.025 was considered significant.

activity of two major intracellular signal transduction pathways of the hormone's receptor (Rchid *et al.*, 2004).

Trigonella foenum-graecum has shown hypoglycemic and antihyperlipidemic effects. It delays the digestion and absorption of carbohydrates and enhances insulin action owing to presence of saprogenins which increase biliary cholesterol excretion as well (Basch *et al.*, 2003).

In this study, the beneficial effects of combination of two herbal drugs namely *Nigella sativa* and *Trigonella foenum-graecum* on dyslipidaemia were investigated in type 2 diabetic patients, who were already on Glibenclamide. Results of present study demonstrate that combination therapy given for 3 months did not produce any significant change in serum triglycerides levels, although these were markedly reduced in intervention group. However, serum HDL-cholesterol levels became significantly raised in intervention group compared to control group patients. This suggests that combination therapy in addition to controlling hyperglycemia (Memon *et al.*, 2010) and causing some reduction in BMI (Memon *et al.*, 2010) also control dyslipidemia in diabetic patients. Furthermore, our finding that combination therapy taken upto three months does not produce any significant change in serum creatinine levels supports the findings of other investigators (Ali, 2004; Meral, 2001; Bhatia, 2006 and Kaviarasan, 2007) that both *N.sativa* and *T. foenum-graecum* do not produce any nephrotoxicity.

As this study has not measured the serum total cholesterol and LDL cholesterol levels hence, we can not comment on the effects of this combination therapy on these variables.

Limitations of study: This study had few limitations such as limited sample size, convenient sampling technique (non-probability). Therefore, these findings shall be interpreted carefully.

CONCLUSION

From this study, it may be concluded that combination therapy of *N. Sativa* and *T. foenum-graecum* with Glibenclamide has beneficial effects in controlling dyslipidaemia (moderate decrease in serum triglycerides and significant increase in HDL level) in type-2 diabetes mellitus patients. A larger study with adequate sample size is recommended for better results.

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