



## ORIGINAL ARTICLE

# Comparison of the Effect of Glitazones and DPP-IV Inhibitors in Type 2 Diabetics as Add on Therapy on Insulin Sensitivity and Serum hs-CRP Levels

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## Abstract

To study & compare the effect of glitazones and DPP-IV inhibitors as add on therapy on Insulin sensitivity and serum hs-CRP levels in type 2 diabetes mellitus patients. Thirty patients (previously known cases of type 2 DM), aged above 18 years, were randomly included in the study. Group A, Glimepiride + Metformin + Pioglitazone (G + M + P) and Group B, Glimepiride + Metformin + Sitagliptin (G + M + S). The study drugs were given on the basis of physician's discretion and the doses of study drugs were fixed according to their clinical presentation & followed up for a period of 3 months. The glycosylated haemoglobin (HbA1c) at the start of study (day 0) of group A was  $10.39 \pm 0.67$  and in group B was  $10.10 \pm 0.62$ . The mean value of HbA1c at the end of the study period (day 90) in group A was  $9.46 \pm 0.61$  and in group B was  $9.04 \pm 0.58$ . Insulin resistance at the start of the study (day 0) in group A, was  $5.549 \pm 0.59$  and in group B was  $6.66 \pm 0.76$ , The mean values of IR at the day 90, in group A was  $3.42 \pm 0.37$  and in group B was  $3.82 \pm 0.61$ . The mean value of hs-CRP at the start of study (day 0) in group A was  $4.34 \pm 0.77$  and in group B was  $6.78 \pm 1.18$ . The mean value of hs-CRP at the end of the study period (day 90) in group A was  $2.8 \pm 0.63$  and in group B was  $4.51 \pm 0.73$ . There was no significant intra group or intergroup difference found in the above mentioned study parameters. Both the study drug groups reduced HbA1c, insulin resistance, though there was no any significant difference. There was decrease in hs-CRP levels in both the groups. Moreover these combination therapies were safe and no serious adverse effects were reported

## Key Words

Type 2 Diabetes mellitus, Pioglitazone, Sitagliptin

## Introduction

The epidemic of type 2 diabetes mellitus and impaired glucose tolerance is one of the major causes of morbidity and mortality worldwide. DM is known not only for raised blood sugar level but more so for its debilitating and life threatening complications (1). Insulin resistance is the prominent feature of type 2 diabetes mellitus and results from combination of inflammation, genetic susceptibility and obesity. Hyperglycaemias, oxidative stress and inflammation contribute to the increased risk of diabetic vasculopathies. Diabetes is a proinflammatory state as evidenced by increased high sensitivity C-reactive protein (hs-CRP), fibrinogen, plasminogen activator inhibitor-1, soluble cell adhesion molecules, and pro-inflammatory cytokines (2,3). Elevated levels of CRP have been reported in patients with diabetes and insulin resistance. The most important current use of serum hs-CRP level

is in primary prevention that is, in the detection of high risk individuals not yet known to have a problem (4). It has been seen that oral hypoglycemic drugs apart from providing optimal glycemic control, also lowers the levels of inflammatory mediators, thus slows down the further disease progression and decrease the incidence of long term complication related to disease. High sensitivity CRP (hs-CRP) is an acute-phase reactant produced primarily in the liver under the stimulation of adipocyte-derived IL-6 and TNF-alpha. hs-CRP is invariably correlated with various parameters relevant to diabetes (5). Glitazones bind to peroxisome proliferator-activated receptors, initiating transcriptional activity that leads to improve insulin action. DPP-4 inhibitors have the potential both for assisting in achieving glycemic goals and for modifying underlying disease course, because DPP-IV is expressed

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as CD26 on cell membranes and CD26 mediates proinflammatory signals, so the drugs of this class also an anti inflammatory effect (6). While all these drugs not necessarily acting primarily to reduce inflammation, it is likely that these drugs, by correction of glucose and insulin levels impinges on the innate immune system that attenuates inflammation and further improve insulin action. All these drugs apart from correcting the hyperglycemia also have comparable anti-inflammatory activity (7). So with this background in this study we used Dipeptidyl peptidase-4 (DPP-4) inhibitors and Thiazolidinediones (TZDs) as add on therapy to see their effect on insulin sensitivity and also on serum hs-CRP level.

### Materials & Methods

The present Interventional study was carried out in the department of Pharmacology and General Medicine, Himalayan institute of medical sciences, Swami Ram Nagar, Dehradun, over period of twelve months. Subjects were recruited from Medicine OPD after taking written informed consent

**Sample size-** 30 patients were included in the study & were divided into two groups, each group having 15 patients. **Inclusion criteria:** Patients of both sex and age > 30 years, HbA1c > 8%, Established diagnosis of uncontrolled Type 2 DM, without complications, No concurrent illness, "Body mass index of 25-45 kg/m<sup>2</sup>" **Exclusion criteria:** Patients of Type 1 DM, Age > 65 years, Presence of any acute or long term clinically detectable complication, History of acute or chronic kidney disease, History of cardiovascular disease, congestive heart failure or oedema, Pregnant or lactating women.

**Study Groups-** Patients included in the study were divided into two groups, 15 patients in each group.

Group A-(G+M+P) - Glimepiride + Metformin + Pioglitazone (TZDs). Group B- (G+M+S) - Glimepiride + Metformin + Sitagliptin (DPP-4inhibitor). The drugs were given to patients on the basis of physician's discretion. The dose ranges of drugs were Glimepiride- (1-3mg/day), Metformin- (1-2gm/day), Pioglitazone (30mg/day), and Sitagliptin (100mg/day).

**Follow up:** Study subjects were followed up for a period of 3 months. Glycosylated haemoglobin and the specific investigations (insulin resistance and hs-CRP levels) were done at the time of subject recruitment i.e. at day 0 and at the end of study period i.e. at day 90. Insulin resistance: It was measured by using HOMA-IR method which is represented as: (8, 9).  $HOMA-IR = \frac{\text{Fasting blood sugar} \times \text{Fasting serum insulin}}{405}$  (Where glucose is given in mg/dl and insulin is given in  $\mu\text{U/ml}$ ). Patients were asked to come fasting on the day of test.

The blood sample for both fasting blood sugar and fasting serum insulin estimation was withdrawn. The fasting serum insulin levels were estimated with the help of mercodia insulin ELISA kit, available in the Microbiology department, HIHT. It helped in the quantitative determination of human insulin in serum or plasma. hs-CRP (by ELISA) (10,11). The hs-CRP is based on the principle of a solid phase enzyme-linked immunosorbent assay (ELISA). The assay system utilizes a unique monoclonal antibody directed against a distinct antigenic determinant on the CRP molecule. This mouse monoclonal anti-CRP antibody is used for solid phase immobilization (on the micro titer wells). Antibody-enzyme (horseradish peroxidase) conjugate solution contains goat anti-CRP antibody. The test sample is allowed to react simultaneously with the two antibodies, resulting in the CRP molecule being sandwiched between the solid phase and enzyme-linked antibodies. After 45-minute incubation at room temperature, the wells are washed with water to remove unbound labeled antibodies. A tetramethylbenzidine (TMB) reagent is added and incubated for 20 minutes, resulting in the development of blue color. The color development is stopped with the addition of 1N HCl indicated by change in color to yellow. The concentration of CRP is directly proportional the color intensity of the test sample. Absorbance is measured spectrophotometrically at 450 nm.

**Statistical Analysis** - Interpretation and analysis of obtained results from compared groups was carried out by using unpaired t' test.

### Results

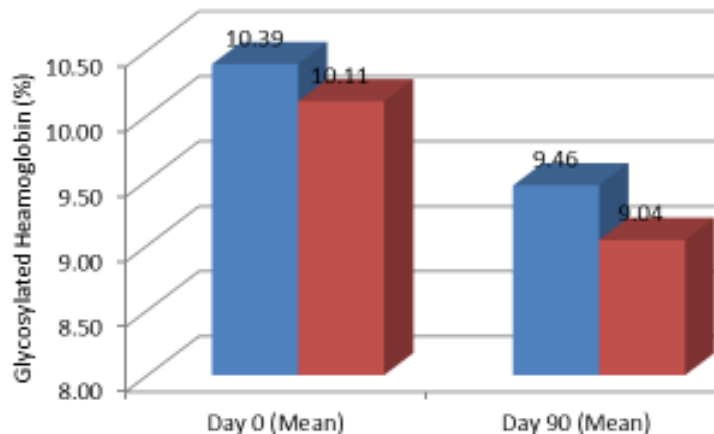
Change in Glycosylated Haemoglobin value at the day 0 and day 90 during study period. Glycosylated haemoglobin (HbA1c) at the start of study (at day 0) in group A was  $10.39 \pm 0.67$  and in group B was  $10.10 \pm 0.62$ . The mean value of HbA1c at the end of the study period (at day 90) in group A was  $9.46 \pm 0.61$  and in group B was  $9.04 \pm 0.58$ . The intergroup change in values of HbA1c as compared to day 0 was not significant. The study drug groups were similar with respect to the overall change in HbA1c (Fig-1).

Change in Insulin Resistance- The mean value of insulin resistance (IR), as calculated by the homeostatic model of assessment for insulin resistance (HOMA-IR) at the start of the study (at day 0) in group A, was  $5.549 \pm 0.59$  and in group B was  $6.66 \pm 0.76$ . The mean values of IR at the day 90, in group A was  $3.42 \pm 0.37$  and in group B was  $3.82 \pm 0.61$  (Fig-2). Change in hs-CRP value at the day 0 and day 90 during study period

The mean value of hs-CRP at the start of study (at day 0) in group A was  $4.34 \pm 0.77$  and in group B was



**Fig. 1 Change in Glycosylated Haemoglobin value at the day 0 and day 90**

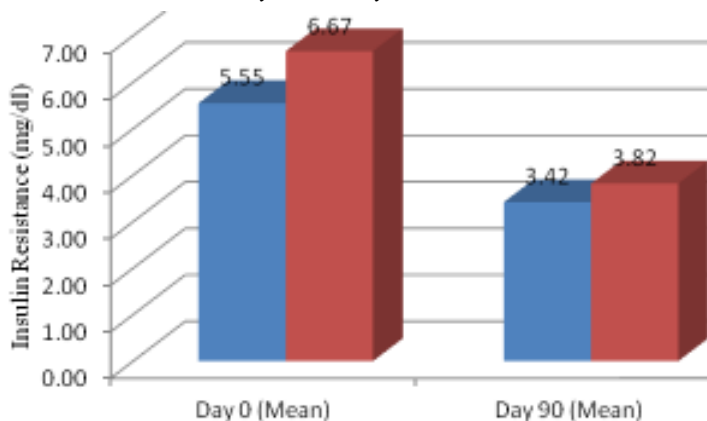


Group A\* = G+M+P, Group B\*\*= G+M+S

G-Glimiperide, M-Metformin, P-Pioglitazone, S- Sitagliptin

■ Group A ■ Group B

**Fig. 2 Change in insulin resistance value at the day 0 and day 90**

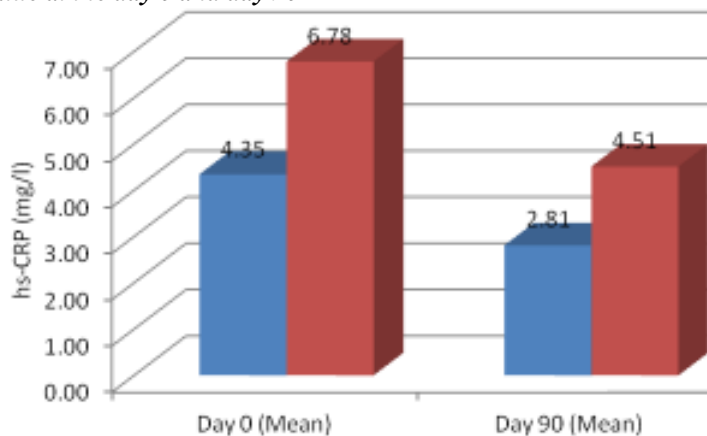


Group A\* = G+M+P, Group B\*\*= G+M+S

G-Glimiperide, M-Metformin, P-Pioglitazone, S- Sitagliptin

■ Group A ■ Group B

**Fig. 3 Change in hs-CRP value at the day 0 and day 90**



Group A\* = G+M+P, Group B\*\*= G+M+S

G-Glimiperide, M-Metformin, P-Pioglitazone, S- Sitagliptin

■ Group A ■ Group B



$6.78 \pm 1.18$ . The mean value of hs-CRP at the end of the study period (at day 90) in group A was  $2.8 \pm 0.63$  and in group B was  $4.51 \pm 0.73$  (Fig-3). There was change in values of hs-CRP in intragroup as compared to day 0, signifies the positive effect of studied drug on serum hs-CRP level, however the change in hs-CRP in intergroup was not significant. The study drug groups were similar with respect to the overall change in hs-CRP (Fig-3).

### Discussion

The relation between diabetes and inflammation explore the therapeutic potential of oral anti-hyperglycaemic agents in the management of type 2 diabetic subjects with increased level of inflammatory markers. There was a decrease in mean baseline glycosylated Hb in both the study groups and in both the groups the difference was comparable with the studies done in same context, but may be because of short duration of study the change in HbA1c as compared to baseline was not significant. The efficacy of Tzds and DPP-IV inhibitors has been proven to be similar in many studies done in past (12, 13). The efficacy of DPP-4 inhibitors and Tzds has also been studied in improving insulin resistance. Both the DPP-4inhibitor and Tzds are individually effective in lowering insulin resistance (14). Several cross-sectional studies showed that insulin resistance and type 2 DM are associated with higher level of CRP, IL-6 and TNF-alpha, markers of subclinical inflammation (15). The difference in insulin resistance at the end of study period (3 months) in group A was 2.2 mg/dl and in group B was 2.8 mg/dl as compared to day 0. Several studies shows the role of OHAs (sulfonylureas, Metformin, DPP-4 inhibitors and Tzds) in reducing the level of hs-CRP in type 2 diabetic subjects, apart from controlling blood glucose level. The anti-inflammatory role of OHAs is helpful in decreasing the disease related long-term complications in type 2 diabetics (16, 17). Keeping this fact in mind the present study was done to see the effect of anti-diabetic drugs on hs -CRP level. With the use of DPP-Iv inhibitors and Tzds as add on therapy with Metformin and Glimepiride combination there is decrease in hs-CRP level at the end of the study period (at day 90) as compared with baseline values (at day 0). The difference in reduction of hs-CRP (normal level <3mg/L) values from day 0 to day 90 in group A (G+M+P) was 1.5 mg/L and in group B (G+M+S) was 2.2 mg/L. However in our study, the change in hs-CRP level in both group was not statistically significant, may be because of short duration of study period. As the change in level of inflammatory markers is the continuous process, so we can say that if treatment is continued for the long duration in both study drug groups, the results

might be significant. Moreover, these combination therapies were found to be safe and no serious adverse events was reported.

### Conclusion

Both the study drug groups reduced HbA1c, insulin resistance, though there was no any significant difference. There was decrease in hs-CRP levels in both the groups but was not significant. Thus, there is a potential of these drugs in not only achieving good glycemic control but also halts the progression of disease related complications.

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