Journal of Diabetes Science and Technology
Volume 4, Issue 3, May 2010

High-Sensitivity C-Reactive Protein Predicts Cardiovascular Risk in Diabetic and Nondiabetic Patients: Effects of Insulin-Sensitizing Treatment with Pioglitazone

Andreas Pfützner, M.D., Ph.D.,[1] Thomas Schöndorf, Ph.D.,[1] Markolf Hanefeld, M.D.,[2] and Thomas Forst, M.D.[1]

Abstract

Systemic inflammatory activity has turned out to play a key pathogenic role in vascular atherosclerosis, insulin resistance, and type 2 diabetes mellitus. Inflammatory biomarkers may therefore be a valuable tool for risk evaluation. Among them, the best evidence to date supports the use of high-sensitivity C-reactive protein (hs-CRP) to monitor insulin resistance and cardiovascular risk in diabetic and nondiabetic individuals. Data suggest that hs-CRP may also participate directly in the process of atherogenesis. A growing number of clinical trials tested the hypothesis that antidiabetic drugs specifically targeting insulin resistance could benefit individuals by reducing inflammation, atherogenesis, and thus cardiovascular risk. One such class are the thiazolidinediones (pioglitazone and rosiglitazone). These agents act as selective ligands of the nuclear transcription factor peroxisome proliferator-activated receptor-γ (PPARγ). This article reviewed published data on hs-CRP changes with the thiazolidinedione agent pioglitazone. Here we found pronounced insulin-sensitizing and anti-inflammatory properties in different clinical settings, including diabetic and nondiabetic individuals. Coadministration of pioglitazone to antilipidemic statin therapy resulted in additional effects on low-grade inflammation, and hs-CRP reduction has been demonstrated to occur independently of glucose lowering. The anti-inflammatory effect appeared to be a rapid physiologic reaction on PPARγ activation and could be observed within a short-term interval after starting pioglitazone therapy. In summary, clinical study results underline the benefit of an early insulin resistance treatment to oppose systemic vascular inflammation and cardiometabolic syndrome in patients with elevated levels of high-sensitivity C-reactive protein.

J Diabetes Sci Technol 2010;4(3):706-716

Introduction

Over the last decades, evidence has accumulated that systemic inflammatory activity plays a key pathogenic role in atherosclerosis and cardiovascular disease (CVD).[1,2] This rationale has led to a search for clinically useful inflammatory biomarkers to improve CVD risk prediction. Prominent among possible candidates is C-reactive

Author Affiliations: [1]IKFE, Institute for Clinical Research and Development, Mainz, Germany; and [2]GWT, Dresden, Germany

Abbreviations: (BMI) body mass index, (CVD) cardiovascular disease, (HbA1c) hemoglobin A1c, (HOMA) homeostasis model assessment, (hs-CRP) high-sensitivity C-reactive protein, (LDL) low-density lipoprotein, (PPARγ) peroxisome proliferator-activated receptor-γ

Keywords: atherosclerosis, diabetes, hs-CRP, inflammation, insulin resistance, pioglitazone

Corresponding Author: Andreas Pfützner, M.D., Ph.D., IKFE–Institute for Clinical Research and Development, Parcusstr. 8, D-55116 Mainz, Germany; email address *AndreasP@ikfe.de*

protein (CRP) as measured by a highly sensitive assay.[3] C-reactive protein represents the classical acute-phase protein produced in the liver in response to inflammatory stimuli, and plasma levels of high-sensitive C-reactive protein (hs-CRP) provide a sensitive marker of increased inflammatory activity in the arterial wall.[4]

Chronic, systemic subclinical inflammation has also been identified as a driving force for insulin resistance, metabolic syndrome, and type 2 diabetes mellitus. Some related metabolic disorders include abdominal adiposity, hypertension, endothelial dysfunction, and glucose intolerance, which often occur in a cluster. Insulin resistance correlates closely with the risk of CVD, explaining some of the excess mortality in type 2 diabetes patients. There appears to be a more-or-less linear relationship of cardiovascular risk to insulin resistance across the spectrum of normoglycemic patients with insulin resistance up to patients presenting with overt type 2 diabetes.[5]

Among inflammatory biomarkers, the best evidence to date supports the use of hs-CRP as an independent predictor of increased CVD risk in diabetic and nondiabetic patients.[6–8] In addition, numerous clinical trials have established elevated CRP levels to be predictive of the development of insulin resistance, metabolic syndrome, and type 2 diabetes.[9–19] Chronic systemic low-grade inflammation may induce insulin resistance, and this association has prompted studies to evaluate the hs-CRP-reducing effects of antidiabetic drugs that work by targeting insulin resistance directly. One such class are the thiazolidinediones (pioglitazone and rosiglitazone). These agents act as selective ligands of the nuclear transcription factor peroxisome proliferator-activated receptor-γ (PPARγ). Our institute for clinical research and development performed and conducted multiple studies in various clinical settings and patient populations, which evaluated the role of pioglitazone treatment for the management of insulin resistance, type 2 diabetes, and chronic systemic inflammation. The purpose of this review was to provide an overview about published data that specifically illuminate the impact of pioglitazone on hs-CRP concentrations in diabetic and nondiabetic patients.

Role of hs-CRP in Atherogenesis

Recent scientific research has revealed that increased focal inflammatory activity in the arterial wall is one of the main characteristics of atherosclerosis. From a pathological point of view, all stages of the atherosclerotic process, from its initiation to plaque rupture, might be considered an inflammatory response to injury and endothelial dysfunction. Damage to the endothelial wall triggers a cascade of events that modulates the inflammatory response, leading to the recruitment of white blood cells into the blood vessel wall, where they give rise to abnormal foam cells and initiate the development of atherosclerotic lesions.[1,20,21]

Among emerging biomarkers that reflect this inflammatory process, one of the largest databases exists for hs-CRP: a chronic low-level increase of the acute-phase protein turned out to be predictive of the risk of future cardiovascular events.[4] Data suggest that C-reactive protein, beyond its role as a cardiovascular risk marker, may also participate directly in atherogenesis,[22,23] which includes but is not limited to the following effects (**Figure 1**).[24–27]

- C-reactive protein binds the phosphocholine of oxidized low-density lipoprotein (LDL).
- C-reactive protein increases LDL uptake into macrophages and enhances the ability of macrophages to form foam cells.
- C-reactive protein inhibits endothelial nitric oxide synthase expression in endothelial cells. Nitric oxide has important antiatherogenic effects, including decreased platelet aggregration, vasoconstriction, and smooth muscle cell proliferation.
- C-reactive protein increases plasminogen activator inhibitor-1 expression and activity.
- C-reactive protein activates macrophages to secrete tissue factor, a powerful procoagulant.
- C-reactive protein upregulates the expression of adhesion molecules in endothelial cells that will attract monocytes to the site of injury.

High concentrations of CRP mRNA have been demonstrated to be present in atherosclerotic plaques.[28] Two research groups revealed independently that CRP is produced by human artery smooth muscle cells of atherosclerotic lesions in response to inflammatory cytokines.[29,30] Taking all this evidence into account, locally produced CRP may actually participate directly in aspects of atherogenesis, promoting the development of cardiovascular complications.

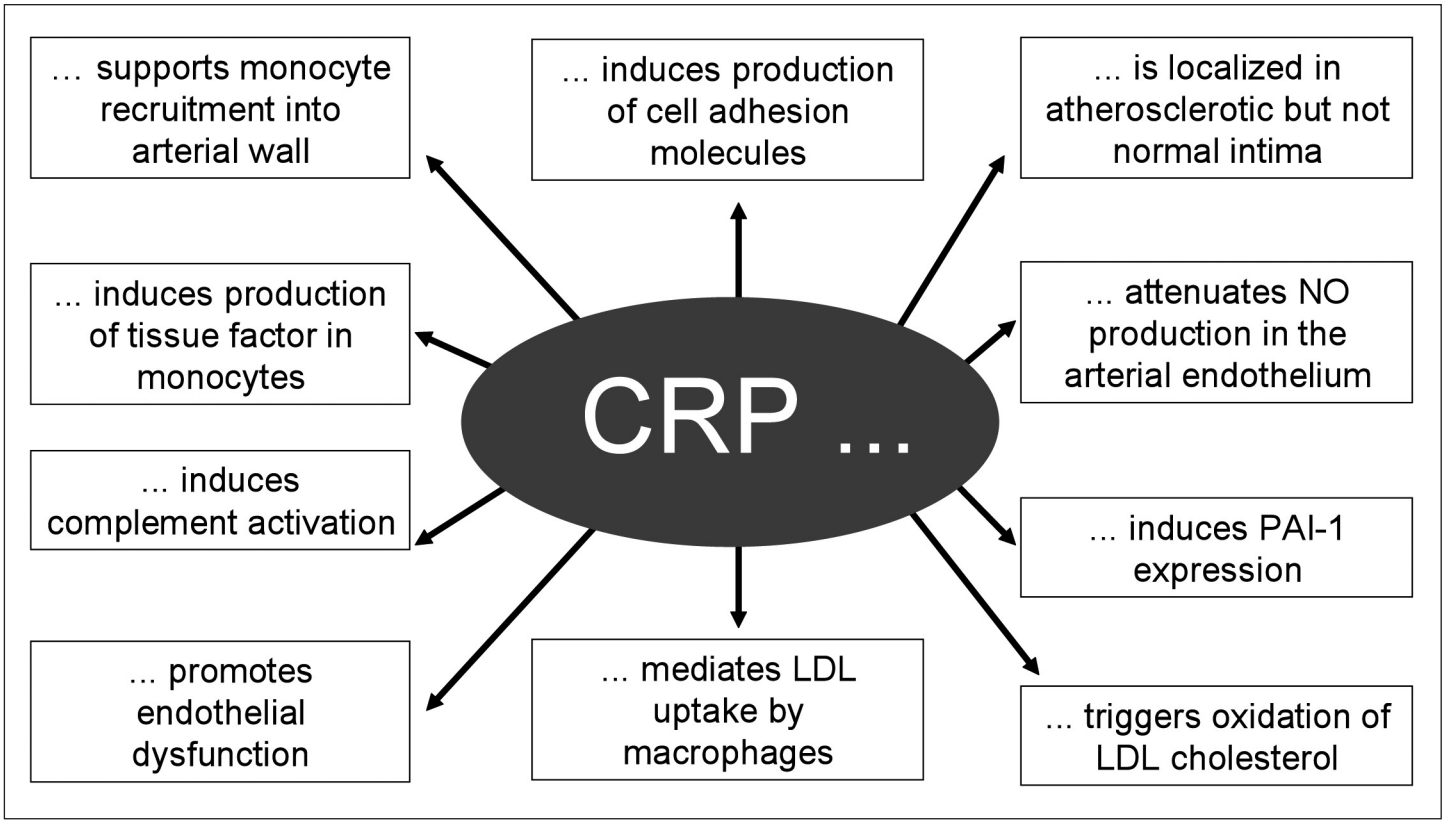

Figure 1. Pathophysiological role of C-reactive protein in atherosclerosis. NO, nitric oxide; PAI-1, plasminogen activator inhibitor-1. Adapted from Ridker *et al.*[23] and *www.crphealth.com*.

Role of hs-CRP in Cardiovascular Risk Prediction

A number of large, prospective epidemiologic studies have examined inflammatory markers as predictors of CVD events in different clinical settings. At present, the best characterized biomarker is hs-CRP. Low-level increases in C-reactive protein appear to be a strong independent predictor of future cardiovascular events, including myocardial infarction, ischemic stroke, peripheral vascular disease, and sudden cardiac death among individuals with and without prior evidence of cardiovascular disease (**Figure 2**).[16,31–36]

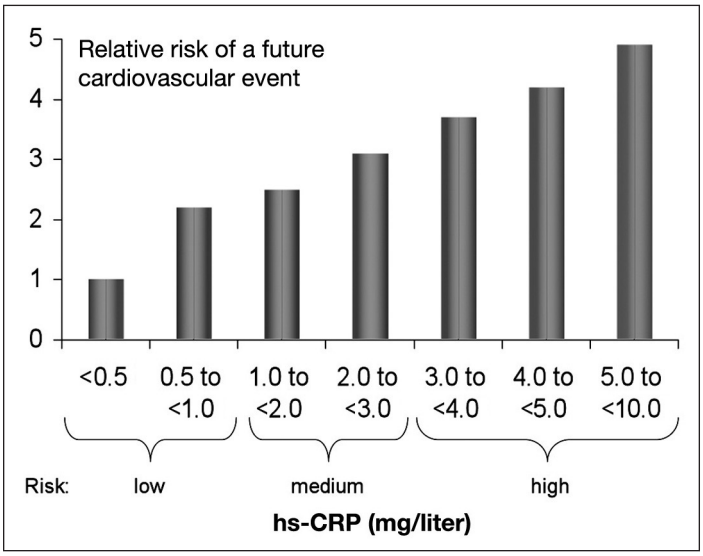

Figure 2. Association between CRP levels and cardiovascular risk. Adapted from Ridker *et al.*[32]

Extensive analysis has demonstrated that hs-CRP adds prognostic information to traditional risk assessment and predicts cardiovascular risk independently of traditional risk factors, including low-density lipoprotein cholesterol.[33,37,38] Measurement of hs-CRP in 27,632 initially healthy U.S. women from the Women's Health Study revealed a predictive value to all LDL cholesterol cutoff points.[2] A further analysis in 15,632 participants of a prospective cohort study yielded, after adjustment for age, blood pressure, smoking, diabetes, and obesity, that hs-CRP added prognostic information beyond that conveyed by all lipid measures.[39]

High-Sensitivity C-Reactive Protein Correlates with Insulin Resistance and Metabolic Syndrome

C-reactive protein has been shown to impair insulin signaling,[40] and emerging laboratory and epidemiological data now link inflammation and hs-CRP to insulin resistance and the metabolic syndrome.[11,18] We conducted a cross-sectional study to assess the association of hs-CRP levels with insulin resistance, β-cell dysfunction, and the prevalence of macrovascular dysfunction in 4270 noninsulin-treated patients with type 2 diabetes mellitus. Results demonstrated that increased hs-CRP levels were linked to homeostatic model assessment, intact proinsulin, insulin, body mass index (BMI), and β-cell dysfunction (all $p < 0.001$)(**Figure 3**), but showed no correlation with disease duration or glucose control.[19] Another cross-sectional trial of participants in the Women's Health Study correlated hs-CRP with insulin resistance in nondiabetic women. The odds ratio for having elevated fasting insulin increased with each tertile of hs-CRP.[17] The Insulin Resistance Atherosclerosis Study explored relationships among cardiovascular risk factors, cardiovascular disease, and insulin resistance as measured directly by a frequently sampled intravenous glucose tolerance test across different ethnic groups and various states of glucose tolerance. Results confirmed that CRP was related independently to insulin sensitivity in healthy, nondiabetic subjects and that chronic subclinical inflammation emerged as part of insulin resistance and other components of the metabolic syndrome.[11]

Some authors hypothesized that decreased insulin sensitivity may lead to enhanced CRP expression by counteracting the physiological effect of insulin on hepatic acute-phase protein synthesis.[41] Clamp studies in normal subjects showed that insulin exerts selective effects on hepatic protein synthesis, reducing the expression of acute-phase response proteins. Resistance to this effect would then in turn lead to increased synthesis of acute-phase proteins, such as CRP.[42] In addition, insulin resistance leads to compensating increased insulin secretion by β cells, supporting visceral lipid tissue growth when sufficient caloric uptake occurs. This process results in an enhanced production of proinflammatory cytokines from the adipose tissue. These proinflammatory cytokines may also contribute to higher levels of C-reactive protein and further impair insulin sensitivity.[5]

Insulin resistance is generally accepted as one of the key components of the metabolic syndrome, a cluster of conditions known to increase the risk of developing

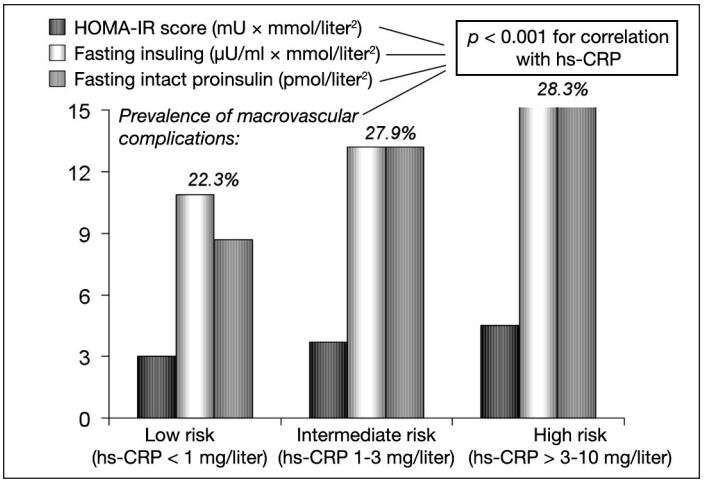

Figure 3. High-sensitivity C-reactive protein is closely linked to insulin resistance. Adapted from Pfützner *et al*.[19]

type 2 diabetes and CVD. General features of the metabolic syndrome include abdominal obesity, hypertriglyceridemia, high-density lipoprotein (HDL) cholesterol, hypertension, and abnormal glucose. All these attributes, however, have been proven to be associated with chronic, low-grade inflammation, as defined by hs-CRP measurement.[8] High hs-CRP levels were also found to be strongly correlated and independently with type 2 diabetes: Elevated levels of hs-CRP remained a significant predictor for future diagnosis of metabolic syndrome and diabetes even after adjusting for BMI, family history of diabetes mellitus, smoking, and other factors.[9–12,14–16,31] Large prospective studies pointed out the involvement of increased hs-CRP on cardiovascular morbidity and mortality in these patients. High levels of hs-CRP have been shown to be an independent predictor of cardiovascular risk for all degrees of severity of metabolic syndrome and type 2 diabetes.[4,16,40]

Effects of Pioglitazone Treatment on hs-CRP Reduction

The recognition that chronic low-grade inflammation plays a fundamental role in atherosclerosis, insulin resistance, and type 2 diabetes has led to clinical studies evaluating whether the treatment of insulin resistance (e.g., with thiazolidinediones) may also reduce hs-CRP as biomarker and conceivable risk factor of cardiovascular disease. Thiazolidinediones (pioglitazone and rosiglitazone) are a class of insulin-sensitizing agents used to treat type 2 diabetes mellitus. For pioglitazone therapy, the PROactive study revealed significant reductions in secondary cardiovascular outcome measures, including macrovascular death, reinfarction, and restroke.[43–45] Because chronic inflammation may be an underlying

cause of both atherosclerosis and insulin resistance, treatments with anti-inflammatory properties may also reduce the incidence of macrovascular complications associated with insulin-resistant states. Indeed, studies have demonstrated that pioglitazone therapy benefits individuals with elevated hs-CRP values.[46–53] **Table 1** provides an overview of the growing number of clinical trials that have examined the impact of pioglitazone on inflammatory processes as measured by hs-CRP levels in various clinical settings.

Table 1.
Effects of Pioglitazone on hs-CRP Levels: Clinical Results in Type 2 Diabetic and Nondiabetic Patients[a]

Reference	Treatment arms	Design	Case numbers	Follow-up	Results with pioglitazone
Type 2 diabetic patients					
Satoh *et al.*[54]	PIO vs control (diet or diet+SU)	Prospective open-label controlled	*N* = 136 Subgroups: HbA1c responders[b] and non-responders	12 weeks	hs-CRP ↓ (PIO: –29% in responders, –27% in nonresponders; $p < 0.01$ vs control)
Pfützner et al.[46] PIONEER	PIO vs GLIM	Prospective open-label controlled, randomized	*N* = 192	24 weeks	hs-CRP ↓ (PIO: –28%, GLIM: –1%; $p < 0.05$ vs GLIM)
Forst *et al.*[48] PIOCARD	PIO vs PBO	Prospective double-blinded controlled randomized	*N* = 92 Patients with CVD	4 weeks	Hs-CRP ↓ (PIO: –18% after 3 days; $p < 0.05$ vs PBO)
Karagiannis *et al.*[50] IRIS V	PIO + previous antidiabetic treatment	Observational trial	*N* = 1.170	20 weeks	Hs-CRP ↓ (PIO: –15%; $p < 0.05$ vs baseline)
Hohberg *et al.*[51] PIOSWITCH	Patients were switched from insulin to PIO+GLIM	Prospective open label	*N* = 98 Patients with residual beta-cell function	24 weeks	hs-CRP ↓ (–21 %; $p < 0.01$ vs baseline)
Musholt *et al.*[52]	PIO+MET after short-term insulin infusion	Prospective open label (pilot study)	*N* = 14	16 weeks	Hs-CRP ↓ (postinsulin value: –15%, end value after PIO intake: –37%; $p < 0.05$ vs baseline)
Pfützner *et al.*[55] PIOGLIM	PIO+GLIM vs GLIM uptitration	Prospective double-blind randomized	*N* = 82	24 weeks	hs-CRP ↓ (PIO+GLIM: –32.6% after 6 months (vs –8.1% with GLIM)
Pfützner *et al.*[56] PIOFIX	PIO+MET (fixed combination) vs GLIM+Met	Prospective double-blind randomized	*N* = 288	24 weeks	hs-CRP ↓ (PIO+MET: –30% after 6 months; $p < 0.05$ vs GLIM+MET: 0%)
Nondiabetic patients					
Szapary *et al.*[57]	PIO vs PBO	Prospective double-blind controlled randomized	*N* = 60 Patients with low HDL-C and MetSyn	12 weeks	hs-CRP ↓ (PIO: –31%; $p < 0.001$ vs placebo)
Hanefeld *et al.*[47] PIOSTAT	PIO+PBO vs SIM+PBO vs PIO+SIM	Prospective double-blind controlled randomized	*N* = 125 Patients with CVD and elevated levels of hs-CRP	12 weeks	hs-CRP ↓ (PIO mono: –32%, SIM mono: –14%, PIO+SIM: –41%; $p < 0.0001$ for PIO+SIM vs baseline)
Forst *et al.*[49] PIOVASC	ATOR+PBO vs ATOR+PIO	Prospective double-blind controlled randomized	*N* = 175 Patients at high cardiovascular risk	24 weeks	hs-CRP ↓ (ATOR+PLB: –32%, ATOR+PIO: –53%; $p < 0.05$ vs ATOR+PLB)
Pfützner *et al.*[53] PIOACE	PIO+PBO vs RAM+PBO vs PIO+RAM	Prospective double-blind controlled randomized	*N* = 149 Patients with arterial hypertension	12 weeks	hs-CRP ↓ (PIO mono: –25%, PIO+RAM: –16%, RAM mono: +20%; $p < 0.05$ vs RAM mono)

[a] Abbreviations: ATOR, atorvastatin; GLIM, glimepiride; HDL-C, high-density lipoprotein cholesterol; MET, metformin; MetSyn, metabolic syndrome; mono, monotherapy; PBO, placebo; PIO, pioglitazone; RAM, ramipril; SIM, simvastatin; SU, sulfonylureas.
[b] Greater than 1% of reduction in HbA1c.

In 2003, Satoh *et al.*[54] published data of 136 Japanese type 2 diabetes patients showing that pioglitazone treatment (30 mg daily for 3 months) significantly reduced hyperglycemia and hs-CRP relative to an untreated control group. In fact, a stratification into glycemic control responders [>1% of reduction in hemoglobin A1c (HbA1c)] and nonresponders revealed that CRP reduction with pioglitazone occurred independently of changes in parameters related to glucose metabolism.[54] Similar results are now emerging from numerous clinical trials, reinforcing the anti-inflammatory potential of this thiazolidinedione.

The Pioneer study enrolled 192 type 2 diabetes patients into a 6-month, prospective, open-label trial. The patients were randomized to receive either pioglitazone (45 mg) or glimepiride (1 to 6 mg, with the intent to optimize therapy). Significant reductions in hs-CRP were only observed in the pioglitazone group ($p < 0.05$ vs glimepiride). Both groups achieved a comparable reduction in HbA1c ($p < 0.001$), suggesting an independent effect of pioglitazone to improve hs-CRP as a measure of low-grade inflammation.[46] The multicenter, double-blinded, and placebo-controlled PIOcard study investigated inflammatory markers during pioglitazone treatment in 92 type 2 diabetes patients with angiographically proven coronary heart disease. An 18% reduction in hs-CRP levels ($p < 0.05$ vs placebo) was observed after only 3 days of treatment with pioglitazone. We concluded that even before effects on glucose metabolism could be obtained, pioglitazone exerted immediate effects on plasma markers of inflammation.[48] The PioSwitch study converted 98 type 2 diabetes patients with residual β-cell function from insulin therapy to treatment with pioglitazone and glimepiride for 6 months. For the majority of patients, the switch from insulin therapy was possible without deterioration of blood glucose control and resulted in a significant decrease of hs-CRP levels ($p < 0.01$ vs baseline).[51] A combined pioglitazone and metformin treatment study with 14 type 2 diabetes patients examined the efficacy of short-term intravenous insulin intervention followed by oral pioglitazone and metformin therapy to prevent patients from continuous insulin applications. Initially, an in-patient, 34-hour continuous intravenous insulin infusion was performed and metformin was given (2× 850 mg/day). Insulin was stopped and 30 mg/day pioglitazone was added at the second in-patient day. Participants were followed for another 4 months. Results demonstrated that a beneficial effect of a short-term intravenous insulin application on glycemic control could be maintained effectively by pioglitazone/metformin treatment. Improvements in hs-CRP values were demonstrated at end point only after continued glitazone intake ($p < 0.05$ vs baseline).[52] Analyses of data from the IRIS V study, a large population-based observational trial in a routine clinical setting, substantiated the favorable effects of the insulin-sensitizing agent on systemic chronic inflammation: 1170 subjects received pioglitazone alone or in combination with their previous treatment (acarbose, sulfonylurea drugs, and/or metformin). After 20 weeks, hs-CRP levels had decreased significantly with pioglitazone therapy (baseline 3.3 ± 1.0 mg/liter vs end point 2.8 ± 2.3 mg/liter, $p < 0.01$).[50] The PIOglim study was a double-blind, parallel prospective study with 82 patients with type 2 diabetes mellitus treated previously with sulfonylurea monotherapy with insufficient glycemic control (HbA1c >7.0%). They were randomized either to uptitration of glimepiride (4 to 6 mg; $n = 37$; 22 males, 15 females, age: 60 ± 9 years, HbA1c: 7.5 ± 0.6%, BMI: 31.6 ± 5.7 kg/m²) or to receive a glimeperide + pioglitazone combination (uptitration possible with 2 mg + 30 mg, 4 mg + 30 mg, and 4 mg +45 mg; 30/45 mg, $n = 45$; 25 males, 20 females, age: 62 ± 8 years, HbA1c: 7.2 ± 0.6%, BMI: 33.5 ± 6.2 kg/m²) for the next 6 months. A reduction in HbA1c was observed in both groups, which was less pronounced in the glimeperide uptitration group (absolute percent: –0.30%, $p < 0.05$ vs baseline) than in the combination group (–0.84%, $p < 0.001$). The combination group also showed a pronounced relative reduction in hs-CRP from –32.6%, which was more extensive than in the comparator group (–8.1%).[55] The effect of a fixed combination of pioglitazone (15 mg) with metformin (850 mg) given twice daily in comparison to a glimepiride + metformin combination therapy on diabetic dyslipidemia and chronic systemic inflammation was assessed in the double-blind, randomized parallel PIOfix study with 288 patients (187 males, 101 females, age: 59 ± 10 years, BMI: 32.6 ± 5.1 kg/m², HbA1c: 7.3 ± 0.8%). Although similar improvements in HbA1c were seen with both treatments within 6 months, the pioglitazone + metformin combination had a significantly better effect on triglycerides and HDL cholesterol, and a pronounced hs-CRP improvement was seen in this arm (–0.9 ± 1.9 mg/dl), whereas no effect on hs-CRP could be observed with the glimepiride + metformin combination (0.0 ± 1.8 mg/liter, $p < 0.001$).[56]

Insulin resistance may also be present in normoglycemic individuals, and some of these patients can be diagnosed as having (cardio)metabolic syndrome. Research in these study populations confirmed the efficacy of pioglitazone treatment for reducing chronic systemic inflammation and hs-CRP levels independently from glucose-lowering

mechanisms.[57] Our own investigations examined insulin-sensitizing effects on CRP levels compared to the impact of lipid-lowering and antihypertensive therapy in patients at increased macrovascular risk.[47,49,53]

The PIOSTAT study demonstrated that nondiabetic patients with vascular insulin resistance and dyslipidemia treated with pioglitazone therapy experienced an overall reduction in chronic systemic inflammation, which was significantly more pronounced than with alternative simvastatin therapy. Giving both drugs in combination resulted in a synergistic effect on the inflammatory biomarker hs-CRP.[47] Insulin resistance as measured by homeostasis model assessment (HOMA) decreased in those receiving pioglitazone, and the correlation between changes in HOMA and hs-CRP was significant (r = 0.43; $p < 0.05$). The PIOVASC study investigated the anti-inflammatory effects of atorvastatin monotherapy and combined treatment with atorvastatin and pioglitazone in 148 patients (BMI 29.2 ± 4.1 kg/m^2) with increased cardiovascular risk. Again, the addition of pioglitazone to atorvastatin resulted in a significant further reduction of high-sensitivity C-reactive protein ($p < 0.05$).[49] The primary objective of the PIOace study was to investigate the effects of pioglitazone, ramipril, or their combination on low-grade inflammation in nondiabetic hypertensive individuals. Patients were treated with 30/45 mg pioglitazone (dose titration), 2.5/5 mg ramipril, or their combination for 12 weeks. A reduction in hs-CRP was observed with pioglitazone (–0.89 ± 1.98 mg/liter) and combination therapy (–0.49 ± 2.11 mg/liter), whereas an increase occurred with ramipril therapy alone (0.58 ± 2.13 mg/liter). Improvements in insulin sensitivity were seen in the pioglitazone and the combination arms only, supporting the link between insulin resistance and chronic low-grade inflammation.[53] A summary of hs-CRP changes observed in our interventional controlled trial, adjusted for the activity of comparator treatments, is provided in **Figure 4**. Combined analysis revealed an overall reduction of hs-CRP by 26.8% in diabetic patients, by 28.4% in nondiabetic patients with metabolic syndrome, and an overall reduction by 27%.

Available study data suggest that rosiglitazone, the other currently available thiazolidinedione, may have similar effects on hs-CRP reduction in diabetic and nondiabetic patients.[57–64] Analyses from a study entitled A Diabetes Outcome Progression Trial examined long-term effects of rosiglitazone, glyburide, and metformin on hs-CRP in 904 diabetic subjects over 4 years: CRP decreased more markedly in the rosiglitazone group by –47.6% relative to glyburide and –30.5% to metformin at 48 months.[64]

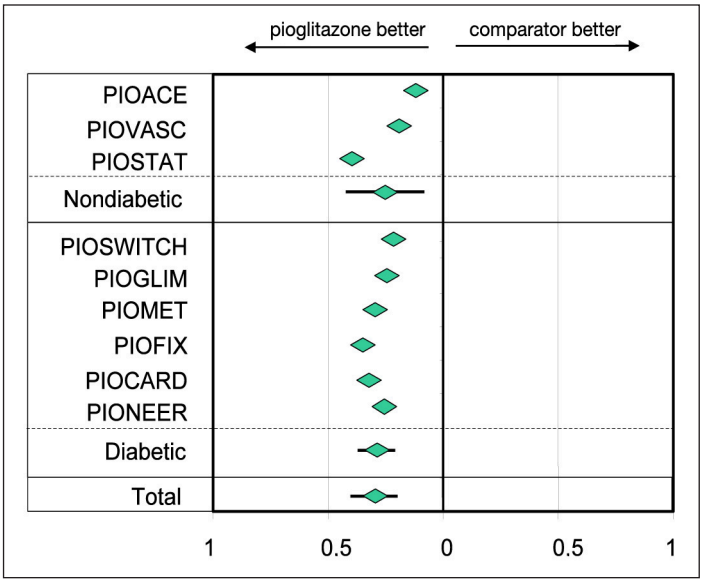

Figure 4. Meta-analysis of hs-CRP changes by pioglitazone in interventional clinical trials performed by IKFE adjusted for the effects of comparator treatments.

Does Antidiabetic Therapy *per se* Influence hs-CRP Levels?

As described earlier, hs-CRP is found to be an independent predictor of risk for metabolic disorders, and increased CRP levels have been described in type 2 diabetes patients. Evidence to date argues for the role of insulin resistance that may be an underlying cause for the association among type 2 diabetes, CVD risk, and low-grade inflammation. The antidiabetic agent metformin provides a weak insulin-sensitizing effect, and it has been found to reduce hs-CRP levels, although to a lesser extent than with glitazone treatment.[65] In contrast, antidiabetic agents without insulin-sensitizing properties appear to have little or no influence on systemic inflammatory processes. This applies to insulin secretagogues, i.e., sulfonylureas, which did not reduce CRP levels in type 2 diabetes patients.[46,66] Anti-inflammatory effects of incretin-based therapies (glucagon-like peptide-1 analogs and dipeptidyl peptidase-4 inhibitors) have not been established yet and need further investigation.[67]

Evidence for anti-inflammatory activity of insulin itself remains controversial. Data appeared that newly initiated insulin therapy in patients with poorly controlled type 2 diabetes decreased serum hs-CRP.[68] Another trial pointed out that the decrease in CRP was significantly more pronounced in intensive insulin-treated patients compared to conventional insulin therapy.[69] In contrast to this, improvement in glycemic control with insulin monotherapy was not associated with a

parallel improvement in markers of vascular inflammation in young, obese, type 2 diabetes patients.[70]

Lifestyle Modification

A higher body mass index was proved to be linked to insulin resistance and elevated CRP concentrations, suggesting a state of low-grade systemic inflammation in overweight and obese persons.[9] Nondrug strategies used to reduce insulin resistance include diet and physical activity. Accordingly, effective weight loss has been shown to reverse elevated hs-CRP levels in diabetic and nondiabetic populations.[9-11,71,72] Favorable effects on the inflammatory response, including a decrease in hs-CRP levels, have also been seen with physical activity interventions.[72,73]

High-Sensitivity C-Reactive Protein in Daily Clinical Practice

It is important to distinguish between standard and high sensitivity assays for the measurement of CRP. The traditional CRP assay, using a polyclonal antibody, assists in the diagnosis and assessment of acute inflammation, i.e., associated with infections and neoplastic diseases. A more sensitive CRP test, the highly sensitive C-reactive protein assay, allows detection of CRP levels down to and below 3 mg/liter. These lower levels toward the upper end of normal reflect low-grade inflammation and have a predictive value of future risk for CVD events.[72]

Measurement of hs-CRP should be done twice, optimally 2 weeks apart, to provide a more stable estimate of level of this marker. The relative cardiovascular risk categories for average hs-CRP levels are[74]:

- low risk <1.0 mg/liter
- average risk 1.0–3.0 mg/liter
- high risk 3.0–10.0 mg/liter
- unspecific elevation >10 mg/liter (should be reevaluated for acute inflammatory conditions)

Conclusions and Summary

The excess cardiovascular risk in insulin-resistant patients is substantial. Research of recent decades revealed that low-grade inflammation is linked significantly to insulin resistance, type 2 diabetes, and increased risk of cardiovascular disease. Elevated levels of high-sensitivity C-reactive protein emerged as a reliable biomarker for the subclinical inflammatory state. Current evidence supports the usefulness of hs-CRP measurement for vascular risk and treatment efficacy assessment in insulin-resistant diabetic and nondiabetic individuals.

Therapies that address insulin resistance may benefit individuals by reducing inflammation, atherogenesis, and thus macrovascular complications. Treatment aimed at improving the insulin-resistant state, whether non pharmacological, such as exercise and weight reduction, or pharmacological, such as metformin and thiazolidinediones, results in a decrease of CRP levels beyond mere glucose lowering.

For the thiazolidinedione drug pioglitazone, we proved both pronounced insulin-sensitizing and anti-inflammatory properties in different clinical settings. Coadministration of pioglitazone to antilipidemic statin therapy resulted in additional effects on low-grade inflammation, and hs-CRP reduction has been demonstrated to occur independently from glucose lowering. The anti-inflammatory effect appears to be a rapid physiologic reaction on PPARγ activation and can be observed within a short-term interval after starting a pioglitazone therapy. In summary, results underline the benefit of an early insulin resistance treatment to oppose systemic vascular inflammation and the cardiometabolic syndrome in diabetic and nondiabetic individuals with elevated levels of high-sensitivity C-reactive protein.

Acknowledgments:

The authors thank all colleagues and patients who participated in the clinical trials, which contributed data to this analysis. We gratefully acknowledge the support of Anja Lütke, M.D., in preparing, editing, and proofreading this manuscript.

Disclosure:

Many studies included into this analysis have been funded by grants from Takeda Pharma. All authors have received research grants, speaker fees, and travel support from Takeda Pharma over the recent years.

References:

1. Ross R. Atherosclerosis--an inflammatory disease. N Engl J Med. 1999;340(2):115-26.
2. Ridker PM, Rifai N, Rose L, Buring JE, Cook NR. Comparison of C-reactive protein and low-density lipoprotein cholesterol levels in the prediction of first cardiovascular events. N Engl J Med. 2002;347(20):1557-65.
3. Clearfield MB. C-reactive protein: a new risk assessment tool for cardiovascular disease. J Am Osteopath Assoc. 2005;105(9):409-16.
4. Pfützner A, Forst T. High-sensitivity C-reactive protein as cardiovascular risk marker in patients with diabetes mellitus. Diabetes Technol Ther. 2006;8(1):28-36.
5. Ritchie SA, Connell JM. The link between abdominal obesity, metabolic syndrome and cardiovascular disease. Nutr Metab Cardiovasc Dis. 2007;17(4):319-26.
6. Ridker PM, Stampfer MJ, Rifai N. Novel risk factors for systemic atherosclerosis: a comparison of C-reactive protein, fibrinogen, homocysteine, lipoprotein(a), and standard cholesterol screening as predictors of peripheral arterial disease. JAMA. 2001;285(19):2481-5.
7. Albert CM, Ma J, Rifai N, Stampfer MJ, Ridker PM. Prospective study of C-reactive protein, homocysteine, and plasma lipid levels as predictors of sudden cardiac death. Circulation. 2002;105(22):2595-9.
8. Ridker PM. Clinical application of C-reactive protein for cardiovascular disease detection and prevention. Circulation. 2003;107(3):363-9.
9. Visser M, Bouter LM, McQuillan GM, Wener MH, Harris TB. Elevated C-reactive protein levels in overweight and obese adults. JAMA. 1999;282(22):2131-5.
10. Fröhlich M, Imhof A, Berg G, Hutchinson WL, Pepys MB, Boeing H, Muche R, Brenner H, Koenig W. Association between C-reactive protein and features of the metabolic syndrome: a population-based study. Diabetes Care. 2000;23(12):1835-9.
11. Festa A, D'Agostino R Jr, Howard G, Mykkänen L, Tracy RP, Haffner SM. Chronic subclinical inflammation as part of the insulin resistance syndrome: the Insulin Resistance Atherosclerosis Study (IRAS). Circulation. 2000;102(1):42-7.
12. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. JAMA. 2001;286(3):327-34.
13. Festa A, D'Agostino R Jr, Tracy RP, Haffner SM. Elevated levels of acute-phase proteins and plasminogen activator inhibitor-1 predict the development of type 2 diabetes: the insulin resistance atherosclerosis study. Diabetes. 2002;51(4):1131-7.
14. Freeman DJ, Norrie J, Caslake MJ, Gaw A, Ford I, Lowe GD, O'Reilly DS, Packard CJ, Sattar N; West of Scotland Coronary Prevention Study. C-reactive protein is an independent predictor of risk for the development of diabetes in the West of Scotland Coronary Prevention Study. Diabetes. 2002;51(5):1596-600.
15. Han TS, Sattar N, Williams K, Gonzalez-Villalpando C, Lean ME, Haffner SM. Prospective study of C-reactive protein in relation to the development of diabetes and metabolic syndrome in the Mexico City Diabetes Study. Diabetes Care. 2002;25(11):2016-21.
16. Ridker PM, Buring JE, Cook NR, Rifai N. C-reactive protein, the metabolic syndrome, and risk of incident cardiovascular events: an 8-year follow-up of 14,719 initially healthy American women. Circulation. 2003;107(3):391-7.
17. Pradhan AD, Cook NR, Buring JE, Manson JE, Ridker PM. C-reactive protein is independently associated with fasting insulin in nondiabetic women. Arterioscler Thromb Vasc Biol. 2003;23(4):650-5.
18. Ndumele CE, Pradhan AD, Ridker PM. Interrelationships between inflammation, C-reactive protein, and insulin resistance. J Cardiometab Syndr. 2006;1(3):190-6.
19. Pfützner A, Standl E, Strotmann HJ, Schulze J, Hohberg C, Lübben G, Pahler S, Schöndorf T, Forst T. Association of high-sensitive C-reactive protein with advanced stage beta-cell dysfunction and insulin resistance in patients with type 2 diabetes mellitus. Clin Chem Lab Med. 2006;44(5):556-60.
20. Libby P, Ridker PM. Novel inflammatory markers of coronary risk: theory versus practice. Circulation. 1999;100(11):1148-50.
21. Plutzky J. Inflammatory pathways in atherosclerosis and acute coronary syndromes. Am J Cardiol. 2001;88(8A):10K-15K.
22. Blake GJ, Ridker PM. C-reactive protein and other inflammatory risk markers in acute coronary syndromes. J Am Coll Cardiol. 2003;41(4 Suppl S):37S-42S.
23. Ridker PM, Bassuk SS, Toth PP. C-reactive protein and risk of cardiovascular disease: evidence and clinical application. Curr Atheroscler Rep. 2003;5(5):341-9.
24. Pasceri V, Willerson JT, Yeh ET. Direct proinflammatory effect of C-reactive protein on human endothelial cells. Circulation. 2000;102(18):2165-8.
25. Nakagomi A, Freedman SB, Geczy CL. Interferon-gamma and lipopolysaccharide potentiate monocyte tissue factor induction by C-reactive protein: relationship with age, sex and hormone replacement treatment. Circulation. 2000;101(15):1785-91.
26. Verma S, Li SH, Baliwala MV. Endothelium antagonism and interleukin-6 inhibition attenuate the proatherogenic effects of C-reactive protein. Circulation. 2002;105(16):1890-6.
27. Jialal I, Devaraj S, Venugopal SK. C-reactive protein: risk marker or mediator in atherothrombosis? Hypertension. 2004;44(1): 6-11.
28. Yasojima K, Schwab C, McGeer EG. Generation of C-reactive protein and complement components in atherosclerotic plaques. Am J Pathol. 2001;158(3):1039-51.
29. Jabs WJ, Theissing E, Nitschke M, Bechtel JF, Duchrow M, Mohamed S, Jahrbeck B, Sievers HH, Steinhoff J, Bartels C. Local generation of C-reactive protein in diseased coronary artery venous bypass grafts and normal vascular tissue. Circulation. 2003;108(12):1428-31.
30. Calabro P, Willerson JT, Yeh ET. Inflammatory cytokines stimulated C-reactive protein production by human coronary artery smooth muscle cells. Circulation. 2003;108(16):1930-2.
31. Ridker PM, Wilson PW, Grundy SM. Should C-reactive protein be added to metabolic syndrome and to assessment of global cardiovascular risk? Circulation. 2004;109(23):2818-25.
32. Ridker PM, Cook N. Clinical usefulness of very high and very low levels of C-reactive protein across the full range of Framingham Risk Scores. Circulation. 2004;109(16):1955-9.
33. Ridker PM. High-sensitivity C-reactive protein, inflammation, and cardiovascular risk: from concept to clinical practice to clinical benefit. Am Heart J. 2004;148(1 Suppl):S19-26.
34. Pai JK, Pischon T, Ma J, Manson JE, Hankinson SE, Joshipura K, Curhan GC, Rifai N, Cannuscio CC, Stampfer MJ, Rimm EB. Inflammatory markers and the risk of coronary heart disease in men and women. N Engl J Med. 2004;351(25):2599-610.
35. Danesh J, Wheeler JG, Hirschfield GM, Eda S, Eiriksdottir G, Rumley A, Lowe GD, Pepys MB, Gudnason V. C-reactive protein and other circulating markers of inflammation in the prediction of coronary heart disease. N Engl J Med. 2004;350(14):1387-97.
36. Cushman M, Arnold AM, Psaty BM, Manolio TA, Kuller LH, Burke GL, Polak JF, Tracy RP. C-reactive protein and the 10-year incidence of coronary heart disease in older men and women. Circulation. 2005;112(1):25-31.
37. Ridker PM, Glynn RJ, Hennekens CH. C-reactive protein adds to the predictive value of total and HDL cholesterol in determining risk of first myocardial infarction. Circulation. 1998;97(20):2007-11.

38. Razzouk L, Muntner P, Bansilal S, Kini AS, Aneja A, Mozes J, Ivan O, Jakkula M, Sharma S, Farkouh ME. C-reactive protein predicts long-term mortality independently of low-density lipoprotein cholesterol in patients undergoing percutaneous coronary intervention. Am Heart J. 2009;158(2):277-83.

39. Ridker PM, Rifai N, Cook NR, Bradwin G, Buring JE. Non-HDL cholesterol, apolipoproteins A-I and B100, standard lipid measures, lipid ratios, and CRP as risk factors for cardiovascular disease in women. JAMA. 2005;294(3):326-33.

40. Devaraj S, Singh U, Jialal I. Human C-reactive protein and the metabolic syndrome. Curr Opin Lipidol. 2009;20(3):182-9.

41. Campos SP, Baumann H. Insulin is a prominent modulator of the cytokine-stimulated expression of acute-phase plasma protein genes. Mol Cell Biol. 1992;12(4):1789-97.

42. De Feo P, Volpi E, Lucidi P, Cruciani G, Reboldi G, Siepi D, Mannarino E, Santeusanio F, Brunetti P, Bolli GB. Physiological increments in plasma insulin concentrations have selective and different effects on synthesis of hepatic proteins in normal humans. Diabetes. 1993;42(7):995-1002.

43. Dormandy JA, Charbonnel B, Eckland DJ, Erdmann E, Massi-Benedetti M, Moules IK, Skene AM, Tan MH, Lefèbvre PJ, Murray GD, Standl E, Wilcox RG, Wilhelmsen L, Betteridge J, Birkeland K, Golay A, Heine RJ, Korányi L, Laakso M, Mokán M, Norkus A, Pirags V, Podar T, Scheen A, Scherbaum W, Schernthaner G, Schmitz O, Skrha J, Smith U, Taton J; PROactive investigators. Secondary prevention of macrovascular events in patients with type 2 diabetes in the PROactive Study (PROspective pioglitAzone Clinical Trial In macroVascular Events): a randomised controlled trial. Lancet. 2005;366(9493):1279-89.

44. Erdmann E, Dormandy JA, Charbonnel B, Massi-Benedetti M, Moules IK, Skene AM; PROactive Investigators. The effect of pioglitazone on recurrent myocardial infarction in 2,445 patients with type 2 diabetes and previous myocardial infarction: results from the PROactive (PROactive 05) Study. J Am Coll Cardiol. 2007;49(17):1772-80.

45. Wilcox R, Bousser MG, Betteridge DJ, Schernthaner G, Pirags V, Kupfer S, Dormandy J; PROactive Investigators. Effects of pioglitazone in patients with type 2 diabetes with or without previous stroke: results from PROactive (PROspective pioglitAzone Clinical Trial In macroVascular Events 04). Stroke. 2007;38(3):865-73.

46. Pfützner A, Marx N, Lübben G, Langenfeld M, Walcher D, Konrad T, Forst T. Improvement of cardiovascular risk markers by pioglitazone is independent from glycemic control: results from the pioneer study. J Am Coll Cardiol. 2005;45(12):1925-31.

47. Hanefeld M, Marx N, Pfützner A, Baurecht W, Lübben G, Karagiannis E, Stier U, Forst T. Anti-inflammatory effect of pioglitazone and/or simvastatin in high cardiovascular risk patients with elevated high sensitive CRP: the PIOSTAT study. J Am Coll Cardiol. 2007;49(3):291-8.

48. Forst T, Karagiannis E, Lübben G, Hohberg C, Schöndorf T, Dikta G, Drexler M, Morcos M, Dänschel W, Borchert M, Pfützner A. Pleiotrophic and anti-inflammatory effects of pioglitazone precede the metabolic activity in type 2 diabetic patients with coronary artery disease. Atherosclerosis. 2008;197(1):311-7.

49. Forst T, Wilhelm B, Pfützner A, Fuchs W, Lehmann U, Schaper F, Weber M, Müller J, Konrad T, Hanefeld M. Investigation of the vascular and pleiotropic effects of atorvastatin and pioglitazone in a population at high cardiovascular risk. Diab Vasc Dis Res. 2008;5(4):298-303.

50. Karagiannis E, Pfützner A, Forst T, Lübben G, Roth W, Grabellus M, Flannery M, Schöndorf T. The IRIS V study: pioglitazone improves systemic chronic inflammation in patients with type 2 diabetes under daily routine conditions. Diabetes Technol Ther. 2008;10(3):206-12.

51. Hohberg C, Pfützner A, Forst T, Lübben G, Karagiannis E, Borchert M, Schöndorf T. Successful switch from insulin therapy to treatment with pioglitazone in type 2 diabetes patients with residual beta-cell function: results from the PioSwitch study. Diabetes Obes Metab. 2009;11(5):464-71.

52. Musholt PB, Schöndorf T, Pfützner A, Hohberg C, Kleine I, Fuchs W, Hehenwarter S, Dikta G, Kerschgens B, Forst T. Combined pioglitazone and metformin treatment maintains the beneficial effect of short-term insulin infusion in patients with type 2 diabetes: results from a pilot study. J. Diabetes Sci. Technol. 2009;3(6):1442-50.

53. Pfützner A, Hanefeld M, Afzal Dekordi L, Müller J, Kleine I, Fuchs W, Forst T. Effect of pioglitazone and ramipril on low grade inflammation and vascular function in patients with increased cardiovascular risk and an activated inflammation: results from the PIOace study. Poster No. 1164 20th IDF Conference, Montreal; 2009.

54. Satoh N, Ogawa Y, Usui T, Tagami T, Kono S, Uesugi H, Sugiyama H, Sugawara A, Yamada K, Shimatsu A, Kuzuya H, Nakao K. Antiatherogenic effect of pioglitazone in type 2 diabetic patients irrespective of the responsiveness to its antidiabetic effect. Diabetes Care. 2003;26(9):2493-9.

55. Pfützner A, Afzal-Dekordi L, Hohberg C, Dissel S, Bluemmer E, Lehmann U, Fuchs W, Forst T. Limitations of the HOMA-B score for assessment of beta-cell functionality in interventional trials-results from the PIOglim study. Diabetes Technol Ther. In press 2010.

56. Pfützner A, Schöndorf T, Tschöpe D, Lobmann R, Merke J, Müller J, Lehmann U, Fuchs W, Forst T. PIOfix-study: effects of pioglitazone/metformin fixed combination in comparison to a combination of metformin with glimepiride on diabetic dyslipidemia. Diabetes. In press 2010.

57. Szapary PO, Bloedon LT, Samaha FF, Duffy D, Wolfe ML, Soffer D, Reilly MP, Chittams J, Rader DJ. Effects of pioglitazone on lipoproteins, inflammatory markers, and adipokines in nondiabetic patients with metabolic syndrome. Arterioscler Thromb Vasc Biol. 2006;26(1):182-8.

58. Haffner SM, Greenberg AS, Weston WM, Chen H, Williams K, Freed MI. Effect of rosiglitazone treatment on nontraditional markers of cardiovascular disease in patients with type 2 diabetes mellitus. Circulation. 2002;106(6):679-84.

59. Sidhu JS, Cowan D, Kaski J. The effects of rosiglitazone, a peroxisome proliferator-activated receptor-gamma agonist, on markers of endothelial cell activation, C-reactive protein, and fibrinogen levels in non-diabetic coronary artery disease patients. J Am Coll Cardiol. 2003;42(10):1757-63.

60. Wang G, Wei J, Guan Y, Jin N, Mao J, Wang X. Peroxisome proliferator-activated receptor-gamma agonist rosiglitazone reduces clinical inflammatory responses in type 2 diabetes with coronary artery disease after coronary angioplasty. Metabolism. 2005;54(5):590-7.

61. Samaha FF, Szapary PO, Iqbal N, Williams MM, Bloedon LT, Kochar A, Wolfe ML, Rader DJ. Effects of rosiglitazone on lipids, adipokines, and inflammatory markers in nondiabetic patients with low high-density lipoprotein cholesterol and metabolic syndrome. Arterioscler Thromb Vasc Biol. 2006;26(3):624-30.

62. Esposito K, Ciotola M, Carleo D, Schisano B, Saccomanno F, Sasso FC, Cozzolino D, Assaloni R, Merante D, Ceriello A, Giugliano D. Effect of rosiglitazone on endothelial function and inflammatory markers in patients with the metabolic syndrome. Diabetes Care. 2006;29(5):1071-6.

63. Pfützner A, Schöndorf T, Seidel D, Winkler K, Matthaei S, Hamann A, Forst T. Impact of rosiglitazone on beta-cell function, insulin resistance, and adiponectin concentrations: results from a double-blind oral combination study with glimepiride. Metabolism. 2006;55(1):20-5.

64. Kahn SE, Haffner SM, Viberti G, Herman WH, Lachin JM, Kravitz BG, Yu D, Paul G, Holman RR, Zinman B; Diabetes Outcome Progression Trial (ADOPT) Study Group. Rosiglitazone decreases C-reactive protein to a greater extent relative to glyburide and metformin over 4 years despite greater weight gain: observations from a Diabetes Outcome Progression Trial (ADOPT). Diabetes Care. 2010;33(1):177-83.

65. Chu NV, Kong AP, Kim DD, Armstrong D, Baxi S, Deutsch R, Caulfield M, Mudaliar SR, Reitz R, Henry RR, Reaven PD. Differential effects of metformin and troglitazone on cardiovascular risk factors in patients with type 2 diabetes. Diabetes Care. 2002;25(3):542-9.

66. Yudkin JS, Panahloo A, Stehouwer C, Emeis JJ, Bulmer K, Mohamed-Ali V, Denver AE. The influence of improved glycaemic control with insulin and sulphonylureas on acute phase and endothelial markers in Type II diabetic subjects. Diabetologia. 2000;43(9):1099-106.

67. Richter B, Bandeira-Echtler E, Bergerhoff K, Lerch C. Dipeptidyl peptidase-4 (DPP-4) inhibitors for type 2 diabetes mellitus. Cochrane Database Syst Rev. 2008;(2):CD006739.

68. Takebayashi K, Aso Y, Inukai T. Initiation of insulin therapy reduces serum concentrations of high-sensitivity C-reactive protein in patients with type 2 diabetes. Metabolism. 2004;53(6):693-9.

69. Hansen TK, Thiel S, Wouters PJ, Christiansen JS, Van den Berghe G. Intensive insulin therapy exerts anti-inflammatory effects in critically ill patients and counteracts the adverse effect of low mannose-binding lectin levels. J Clin Endocrinol Metab. 2003;88(3):1082-8.

70. Avilés-Santa L, Salinas K, Adams-Huet B, Raskin P. Insulin therapy, glycemic control, and cardiovascular risk factors in young Latin Americans with type 2 diabetes mellitus. J Investig Med. 2006;54(1):20-31.

71. Tchernof A, Nolan A, Sites CK, Ades PA, Poehlman ET. Weight loss reduces C-reactive protein levels in obese postmenopausal women. Circulation. 2002;105(5):564-9.

72. Camhi SM, Stefanick ML, Ridker PM, Young DR. Changes in C-reactive protein from low-fat diet and/or physical activity in men and women with and without metabolic syndrome. Metabolism. 2009;59(1): 54-61.

73. Rothenbacher D, Hoffmeister A, Brenner H, Koenig W. Physical activity, coronary heart disease, and inflammatory response. Arch Intern Med. 2003;163(10):1200-5.

74. Pearson TA, Mensah GA, Hong Y, Smith SC Jr; CDC; AHA. CDC/AHA workshop on markers of inflammation and cardiovascular disease: application to clinical and public health practice: overview. Circulation. 2004;110(25):e543-4.