Adiponectin complex ratios as predictors to clinical response to rosiglitazone treatment in treatment-naïve type 2 diabetic patients

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A. Study Purpose and Rationale

Upon introduction of the thiazolidinedione (TZD) class of antidiabetic medications to the armamentarium of clinicians in the battle for glycemic control in diabetic patients, bold predictions of improved cardiovascular health, prolonged pancreatic β-cell function and vast improvements in the metabolic disarray common to the diabetic population followed. Many of these hopes were initially dashed with the removal of troglitazone (Rezulin™) from the market after scattered reports of hepatotoxicity; with the emergence of other viable TZDs (rosiglitzone, Avandia™; pioglitazone, Actos™), these dreams were renewed. Clinical trials demonstrated efficacy of TZDs in adjunct therapy of type 2 diabetic patients, in conjunction with either a biguanide (metformin) or a sulfonylurea – in fact, combination formulations of both available TZDs with metformin or a variety of sulfonylureas have recently been approved for treatment of diabetic patients and clinical trials demonstrate greater efficacy of these combination medications than either component alone (Rosenstock, 2006). Nevertheless, there has been reluctance by many practioners to fully embrace TZDs in the initial treatment of diabetic patients, preferring to wait for metformin monotherapy failure. This clinical preference appears to be borne from a variety of factors as summarized in David Nathan's treatment algorithm for type 2 diabetes (DM2), recently republished in a NEJM editorial (Nathan, 2006): cost of use, incidence and severity of side effects and availability of a more effective, though relunctantly adopted treatment option (insulin). The accompanying article to this editorial described the ADOPT trial, a double-blind RCT comparing the use of metformin, rosiglitazone (RSG) and glyburide as initial monotherapy for type 2 diabetic patients – briefly, the results of this trial confirm efficacy of rosiglitazone in initial treatment of DM2, and fairly convincingly demonstrates minimally equal if not improved glycemic durability (longer time to treatment failure, defined as addition of second antidiabetic agent) with subtly improved parameters of glycemic control (0.12% improved %HbA1c, improved fasting plasma glucose, improved insulin sensitivity as measured by HOMA). This improved metabolic profile, however, comes at an inconsistent price – 12% of patients in the RSG arm withdrew from the study due to adverse side effects (weight gain most commonly, fractures and CHF most seriously) and another 15% had insufficient glycemic response to the medication (Kahn, 2006). Although both parameters were significantly better than the metformin and glyburide arms of this trial, the nearly 30% of patients (a number which has been demonstrated in other TZD-based trials) that have an inadequate response to TZD treatment preclude, in the eyes of many clincians, the use of Avandia or Actos as initial monotherapy in diabetic patients.

Clearly, what is necessary is a biochemical or clinical screening parameter that will identify and properly triage diabetic patients as responders and non-responders to TZD therapy earlier in the course of treatment – the advantages of such a pre-screen are fairly obvious, including improved glycemic control and durability (if only TZD-responsive patients are selected, the initial choice for monotherapy tilts more favorably towards TZDs as opposed to metformin) and avoidance of preventable side effects. The purpose of the study outlined below is to validate such a pre-screen already proposed by retrospective analysis of prior studies involving a variety of TZDs.

One of the mechanisms proposed for TZD action involves PPARy activation in adipose tissue, leading to changes in the secretory profile of adipocytes and an induction of serum concentrations of an adipocyte-secreted protein (adipokine), adiponectin. Adiponectin has been shown to increase insulin sensitivity, leading to enhanced inhibition of hepatic glucose output in both a variety of animal models as well as in euglycemic-clamp studies performed on human subjects. The human adiponectin gene lies within a novel diabetes susceptibility locus identified on chromosome 3 (3q27) and it has been proposed that genetic variability within the adiponectin gene leads to alteration of serum levels of the protein, and generally predisposes patients to insulin resistance (Kang et al, 2005; Pajvani et al, 2003b). The potential importance of adiponectin as a therapeutic target is underscored by a number of studies which demonstrated that in mice and humans, TZD treatment effects transcriptional upregulation accompanied by increased production and secretion of adiponectin from adipocytes (Yu et al, 2002). However, whether the TZD-mediated induction of adiponectin is causative or simply diagnostic of improved insulin sensitivity remains under debate. A causative role has been challenged by the report that discordance exists between improvements in insulin sensitivity and induction in adiponectin. Although the vast majority of patients induce adiponectin expression and secretion in response to TZD treatment (albeit, to various degrees), only approximately 70% of patients demonstrate clinically improved insulin sensitivity. This suggests that induction of adiponectin in any particular individual is neither predictive nor correlative to quantitative improvements in insulin sensitivity.

Inconsistent antidiabetic response and induction of adiponectin led us to hypothesize that increased adiponectin levels alone are insufficient to explain the wide range of clinical responses to TZD treatment. We have recently demonstrated, and other groups have confirmed, that adiponectin exists in multiple oligomeric forms in serum, ranging from small trimeric complexes (LMW) to an intermediate hexamer (MMW) to a range of high molecular weight complexes (HMW) with an apparent molecular composition of 12-18 monomeric subunits of adiponectin (Pajvani et al, 2003a). These oligomeric complexes are stable both *in vitro* and *in vivo* and can readily be resolved by velocity sedimentation, gel filtration chromatography as well as several new commercially-available ELISA/RIA kits. In retrospective analysis of TZD-treated diabetic patients, we identified a statistically significant correlation between induction of HMW adiponectin with improved insulin sensitivity (Pajvani et al, 2004). Regardless of the diabetic cohort examined, total adiponectin levels were induced with TZD treatment, even with significant differences in treatment regimens (troglitazone, rosiglitazone, pioglitazone with treatment periods of between 3-26 weeks). Consistent with other studies (Abbassi et al, 2006), there was no correlation between induction of total adiponectin levels and improvement in various measures of insulin sensitivity, and in fact, there were patients who induced adiponectin who demonstrated no clinical improvement, as well as vice versa - these outliers, however, did not induce HMW adiponectin release. Follow-up studies demonstrated that TZD-mediated changes in serum adiponectin complexes (Bodles et al, 2006), as well as early detectable changes in hepatic glucose output (via clamp studies) can occur as early as 2 weeks into treatment (Tonelli et al, 2004). Collectively, these prior studies conclusively demonstrate that TZD-mediated improvements in insulin sensitivity are always and necessarily accompanied by a relative increase in the %HMW/total adiponectin ratio in serum, and in the absence of change in that ratio, no clinical benefit will be derived.

The purpose of this proposed study is to identify any predictive value of early changes in %HMW/total adiponectin for future TZD-mediated improvements in insulin sensitivity, as a diagnostic assay. It is attractive to speculate that an early induction of HMW adiponectin will be able to portend responsiveness to these medications, allowing earlier notice as to whether, for instance, continued rosiglitazone treatment would be warranted or not worth the clinical side-effects and cost of treatment.

B. Study Design and Statistical Analysis

This prospective, randomized, stratified, controlled and double-blinded study will evaluate the value of increased HMW/total adiponectin ratio in predicting future response in HbA1c (and by proxy, glycemic control) with rosiglitazone treatment. Briefly, as outlined below in (C), 40 treatment-naïve type 2 diabetic patients will be started on an up-titrating dose of rosiglitazone and several metabolic parameters (including total and HMW adiponectin levels) will be measured at baseline and compared to various time points during the course of the study. A detailed list of inclusion and exclusion criteria and means of recruitment are detailed below in sections (G) and (H).

Patients will be separated into quartiles by improvement in HMW/adiponectin ratio from baseline to 4 weeks post-initiation of rosiglitazone treatment – i.e. the twelve patients with the largest induction of HMW/adiponectin from baseline will constitute group A, the next twelve group B and so on until group D. Previous studies (Pajvani et al, 2004) have demonstrated that groups A and D will be well-separated (change in HMW/adiponectin ratio with TZD treatment ranges from -20% to 400%). Improvements in glycemic control with rosiglitazone treatment (in terms of change in %HbA1c) will be measured for all groups, but only groups A and D will be compared for efficacy. The primary endpoint of this study will be to determine if early rosiglitazone-induced changes in the HMW/total adiponectin ratio at 4 weeks of treatment will predict a TZD-responsive subset of patients that will show the greatest improvement in HbA1c at the 6 month endpoint of the study – theoretically, group A will demonstrate a statistically significant and clinically important improvement in HbA1c when compared to group D. A clinically important change in HbA1c is obviously a debatable subject, but for the purposes of this study, in order for an adiponectin complex assay to add any value to the clinical decision-making process in choosing an antidiabetic regimen, a value of 3-fold improved HbA1c with rosiglitazone in group A versus group D was empirically used in the following power calculations (i.e. average %HbA1c improvement of 1.5% in group A vs. 0.5% in group D). Given an approximate standard deviation and %change of %HbA1c as 0.25% and 1% respectively, (estimated from meta-analysis using clinical trials with TZDs; Phatak et al, 2006), using an unpaired t-test for parallel groups and setting $\alpha=0.05$ and $\beta=0.80$, each quartile will require 2 patients, for a total study size of 8. If, however, the standard deviation in such a small study reaches 0.5%, each quartile will require 5 patients for a total study size of 20. Finally, if a smaller effect size is used in the calculations (2.5-fold, i.e. 1.25% in group A vs. 0.5% in group D) with the larger standard deviation of 0.5%, the number of patients per quartile increases to 8 for a total study size of 32. Due to the variability in clinical response to rosiglitazone across different groups, we will enroll 40 patients in order to avoid a type II error and to account for possible study drop-outs.

Secondary outcomes will include whether (1) change in HMW/adiponectin at 6 months correlates with improved HbA1c; change in total adiponectin at 4 weeks (2) or 6 months (3) correlates with improved HbA1c; change in weight correlates with either change in HMW/adiponectin at 6 months (4) or total adiponectin at 6 months (5).

C. Study Procedure

Diabetic patients who meet eligibility criteria will be invited to participate and will be pre-screened for exclusion criteria. After obtaining informed consent, including advisement as to alternative treatment regimens available for type 2 diabetes, patients will have baseline values of HbA1c, hepatic function panel (LFTs), total adiponectin and HMW/total adiponectin drawn by routine phlebotomy (<5ml total serum necessary). Baseline physical exam will assess pre-treatment edema and thorough ophthalmology exam to monitor for macular edema. Patients will be started on rosiglitazone 4mg dose once daily, provided with glucometer and other necessary implements to check fasting plasma glucose at home and given a follow-up appointment for 4 weeks. Patients will also be given a phone number to call twice weekly to document fasting glucose measurements from the previous days; if patients have a fasting plasma glucose of >180mg/dl on any day after week 2 of the study, or fasting plasma glucose values of >140mg/dl on any consecutive three days after week 2 of the study, their dose of rosiglitazone will be increased to 4mg twice daily. If at the highest dose of rosiglitazone (total 8mg daily), any patient has a fasting plasma glucose measurement of >180mg/dl at any point after week 4 of the study, the patient will be disqualified from the study and referred back to their primary care physician for further diabetes management. Alternatively, patients will be clinically monitored monthly for glycemic control and development of side effects (weight gain, edema), as well as by phlebotomy for changes in LFTs. Any patient with an increase in AST/ALT to 2x ULN will be disenrolled from the study, rosiglitazone discontinued and referred back to their primary care physician for further management. After six months, one final set of blood levels of HbA1c, LFTs, total and HMW adiponectin values will be measured, and final weight and other clinical parameters pre-specified (edema) will be determined.

D. Study Drugs

Rosiglitazone (Avandia™) is a member of the novel oral antidiabetic class of agents known as thiazolidinediones (TZDs) – the mechanism of action of TZDs is thought to involve activation of the nuclear receptor PPARγ, promoting glucose disposal in muscle and suppressing hepatic gluconeogenesis and thus improving insulin sensitivity. Rosiglitazone is FDA-approved for initial and maintenance therapy of type 2 Diabetes mellitus, beginning with a starting dose of 4mg, maximum daily dose of 8mg independent or in conjunction with other antidiabetic therapies (biguanides or sulfonylureas). Rosiglitazone is generally well-tolerated with a relatively low incidence of adverse effects when taken correctly. The most common adverse effects include edema (4.8%) and weight gain (incidence unknown); serious side effects include congestive heart failure, cholestatic hepatitis and hepatotoxicity (rare), diabetic macular edema (very rare) and pulmonary edema. The serious side effects preclude the use of rosiglitazone in patients with NYHA Class III/IV heart failure and caution should be used in patients with any class of CHF. Rosiglitazone may exacerbate pre-existing edema, hepatic dysfunction or macular edema. Rosiglitazone is not recommended as independent therapy for type 1 Diabetes mellitus as insulin is required for the action of all TZDs. Drug interactions are few (established only for Trimethoprim and Rifampin through actions of these medications on the hepatic P450 CYP2C8 and CYP2C9 enzymes that predominantly metabolize rosiglitazone). Rosiglitazone is pregnancy class C (potentially harmful in animal studies although no studies in humans are available) and should preferably not be used in pregnant patients.

E. Medical Devices

No medical devices will be used in this study.

F. Study Questionnaires

No questionnaires will be used in this study.

G. Study Subjects

Subjects eligible for this study will be treatment-naïve (only past and current therapeutic intervention would be diet/lifestyle management) type 2 diabetic patients between the ages of 30 and 75 years with baseline HbA1c measurements between 7.5% and 9%. Exclusion criteria include pregnancy, clinically significant hepatic disease (as assessed by AST/ALT twice the upper limit of normal, liver disease by history or proven hepatic dysfunction as per biopsy), clinically significant peripheral edema, macular edema or known congestive heart failure (NYHA class I-IV). Type I diabetic patients are not eligible for this study.

H. Recruitment of Subjects

Eligible subjects will be recruited from the individual practices of physicians within the Columbia-Presbyterian Medical Center – electronic and paper notices of this study will be posted to notify CPMC physicians of this study, and an investigator will be available by beeper at all times to help recruit patients. Subjects will be recruited only if their primary care physician (as well as the patient themselves) are in agreement with the study parameters.

I. Confidentiality of Study

All participants will be coded by a random number system in order to ensure confidentiality. Data will be stored in a secure location, only accessible to the study investigators.

J. Location of Study

The study will be performed within the auspices of the Columbia-Presbyterian Medical Center, including but not limited to the Irving Center for Clinical Research, Naomi-Berrie Diabetes Center, Milstein Hospital and the Allen Pavillion.

K. Potential Risks

Risks of enrolling in this study are limited to the potential side effects of the study drug, rosiglitazone (outlined in D). Aside from minimal risk involved in routine phlebotomy involved in this study, there are no other significant risks involved.

L. Potential Benefits

Benefits of this study include improved glycemic control with the resultant beneficial health benefits therein, including but not limited to improved cardiovascular health and limiting morbidity associated with diabetes (renal failure, blindness, vasculopathy, etc.).

Improved glycemic control has been shown to delay morbidity and improve mortality of this diabetes.

M. Alternative Therapies

There are other viable therapeutic options for treatment-naïve type 2 diabetic patients, which will be discussed individually with each research subject. Management of newly diagnosed diabetes has traditionally involved the use of a biguanide (i.e. metformin), with sulfonylureas and thiazolidinediones used as adjunct therapy, although a recent randomized clinical trial evaluating the use of either metformin or rosiglitazone in initial treatment of diabetic patients demonstrated improved glycemic durability with rosiglitazone (Kahn et al, 2006). The natural course of diabetes (progressive pancreatic β-cell dysfunction) usually necessitates insulin replacement therapy in later-stages of its course, but some providers offer this option earlier in disease in hopes of preventing future morbidity.

N. Compensation to Subjects

Subjects will be reimbursed for travel expenses incurred for participating in this study; no other compensation will be offered.

O. Costs to Subjects

Subjects will not incur additional costs from participating in this study.

P. Minors as Research Subjects

All subjects will be required to be over 18 years of age.

Q. Radiation or Radioactive Substances

Not applicable to this study.

R. References

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