A STUDY ON THE EFFECT OF ADD-ON ROSIGLITAZONE ON THE ANKLE-BRACHIAL PRESSURE INDEX OF PATIENTS WITH TYPE 2 DIABETES MELLITUS IN HOSPITAL UNIVERSITI SAINS MALAYSIA

by

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LIST OF ABBREVIATIONS

ACE-I	Angiotensin Converting Enzyme Inhibitor
ABPI	Ankle-Brachial Pressure Index
APC	Activated Protein C
ARB	Angiotensin-II Receptor Blocker
AT III	Antithrombin III
CRP	C-Reactive Protein
DP	Dorsalis Pedis artery
FBS	Fasting Blood Sugar
HbA_{1c}	Glycosylated Haemoglobin
HDL-C	High Density Lipoprotein Cholesterol
hsCRP	High Sensitivity C-Reactive Protein
IFG	Impaired Fasting Glucose
IGT	Impaired Glucose Tolerance
IVUS	Intravascular Ultrasound
LDL-C	Low Density Lipoprotein Cholesterol
MDCT	Multi-Detector Computed Tomography
mg	milligram
ml	millilitre
mmHg	millimetres of Mercury
mmol/L	millimoles per Litre
MRI	Magnetic Resonance Imaging

OHA	Oral Hypoglycaemic Agent
PAD	Peripheral Artery Disease
PAI-1	Plasminogen Activator Inhibitor-1
PC	Protein C
PPAR-γ	Peroxisome Proliferator-activated Receptor-Gamma
PS	Protein S
PT	Posterior Tibial artery
RBS	Random Blood Sugar
SD	Standard Deviation
t-PA	tissue-type Plasminogen Activator
TNFα	Tumour Necrosis Factor-Alpha
u-PA	urokinase-type Plasminogen Activator
vWF	von Willebrand Factor

ABSTRAK

KAJIAN MENGENAI KESAN UBAT ROSIGLITAZONE KE ATAS INDEKS TEKANAN DARAH BUKU LALI-BRAKIAL DI KALANGAN PESAKIT DIABETIS MELLITUS JENIS 2 YANG MENJALANI RAWATAN DI HOSPITAL UNIVERSITI SAINS MALAYSIA

Latarbelakang Rosiglitazone adalah salah satu jenis agen pengurangan gula dari kumpulan thiazolidinedione yang digunakan dalam rawatan diabetis mellitus jenis 2. Kajian telah menunjukkan ubat ini, yang merupakan agonis penambah peroksisom reseptor-gamma mempunyai kesan selain dari pengawalan paras gula. Selain dari memperbaiki rintangan terhadap insulin, contoh-contoh kesan rosiglitazone yang tidak berkaitan dengan pengurangan gula adalah pengurangan penanda aras keradangan, memperbaiki fungsi endotelial, memperbaiki aktiviti fibrinolitik dan perubahan dalam profil kolesterol. Kajian ini ingin memperlihatkan secara klinikal kesan pengurangan aterosklerosis serta penambahbaikan sistem fibrinolitik selepas penggunaan rosiglitazone. Objektif kajian ini adalah untuk menilai perubahan secara tidak langsung beban aterosklerosis dengan mengukur indeks tekanan darah buku lali-brakial, bilangan pesakit di dalam kajian ini yang mempunyai penyakit arteri yang signifikan, perubahan dalam sifat kecenderungan untuk salur darah tersumbat di kalangan pesakit kencing manis dengan mengukur tahap aktiviti plasminogen dalam serum, perubahan kawalan paras gula dengan mengukur HbA_{1c} dan perubahan dalam profil kolesterol dengan mengukur kolesterol keseluruhan, lipoprotin kolesterol berketumpatan tinggi serta lipoprotin kolesterol berketumpatan rendah setelah rosiglitazone ditambah ke regim rawatan kencing manis jenis ke-2.

Kaedah Sebuah kajian kohort hirisan lintang direka di mana pesakit serta doktor tahu akan jenis ubat yang diberi. 59 pesakit telah diambil untuk kajian. Pesakit yang belum pernah menggunakan rosiglitazone diberikan 4mg untuk dimakan selama 10 minggu. Ubat ini ditambah kepada ubatan pesakit yang sedia ada. Indeks tekanan darah buku lali-brakial, HbA_{1c}, paras aktiviti serum plasminogen serta profil kolesterol (berpuasa) diambil pada permulaan dan akhir tempoh kajian. Pesakit diminta tidak menukar dos serta regimen ubat-ubatan mereka ketika tempoh kajian.

Keputusan Seramai 48 daripada 59 pesakit dinilai pada akhir tempoh kajian. Purata indeks tekanan darah buku lali-brakial adalah 1.06 ± 0.12 sebelum dan 1.07 ± 0.13 selepas rosiglitazone ditambah (p=0.439). Seramai 4 pesakit (8.3%) mempunyai nilai indeks tekanan darah buku lali-brakial kurang <0.90. Purata aktiviti serum plasminogen(%) adalah 96.00 ± 14.77 sebelum rosiglitazone, dan 111.98 ± 15.83 selepas (p=0.006). HbA_{1c} (%) pada permulaan kajian adalah 9.76 ± 2.06, dan purata selepas kajian 9.25 ± 2.03 (p<0.001). Purata kolesterol keseluruhan (mmol/L) adalah 4.95 ± 1.02 sebelum rosiglitazone dan 5.32 ± 0.94 selepas (p=0.003). Purata lipoprotin kolesterol berketumpatan tinggi (mmol/L) pada permulaan kajian adalah 1.32 ± 0.37 dan 1.47 ± 0.41 di penghujung, p<0.001. Selain dari itu, purata lipoprotin kolesterol berketumpatan rendah (mmol/L) adalah 2.89 ± 0.85 pada awal kajian dan 3.08 ± 0.96 di akhir kajian (p=0.098).

Kesimpulan Kajian ini menunjukkan rosiglitazone 4mg yang dimakan sekali sehari memperbaiki tahap aktiviti serum plasminogen. Ini mungkin menandakan aktiviti

fibrinolitik makin berkesan. Kajian ini walaubagaimanapun tidak dapat menentukan kesannya secara klinikal. HbA_{1c} juga berkurangan secara signifikan. Selain dari ini, terdapat pertambahan jumlah kolesterol keseluruhan serta tahap lipoprotin kolesterol berketumpatan tinggi sama seperti kajian lain sebelum ini. Pertambahan indeks tekanan darah buku lali-brakial serta tahap lipoprotin kolesterol berketumpatan rendah tidak signifikan dalam kajian ini. Bilangan pesakit yang mempunyai penyakit arteri yang signifikan tidak ramai dalam kajian ini berbanding kajian lain yang dijalankan sebelum ini. Kajian yang lebih lanjut diperlukan untuk menunjukkan perkaitan antara penggunaan rosiglitazone dan pengurangan ateroma.

ABSTRACT

A STUDY ON THE EFFECT OF ADD-ON ROSIGLITAZONE ON THE ANKLE-BRACHIAL PRESSURE INDEX OF PATIENTS WITH TYPE 2 DIABETES MELLITUS IN HOSPITAL UNIVERSITI SAINS MALAYSIA

Background Rosiglitazone, an oral hypoglycaemic agent of the thiazolidinedione group is used for Type 2 Diabetes treatment. Research has shown that this medication, being a peroxisome proliferator activator receptor-gamma agonist has effects beyond glycaemic control alone. Apart from improving insulin resistance, the non-hypoglycaemic effects of rosiglitazone include reduction of inflammatory markers, improvement of endothelial function, improvement in fibrinolytic activity as well as changes in cholesterol profile. This study intends to see if the non-hypoglycaemic effects of rosiglitazone translate to a clinically noticable reduction in atherosclerosis as well as improvement of fibrinolytic activity. The objectives of this study were to assess changes in surrogate markers of atherosclerotic burden via ankle-brachial pressure index measurements, the number of patients who have significant peripheral artery disease in the study, changes in the diabetic prothrombotic state via serum plasminogen activity, changes in glycaemic control via HbA_{1c} and changes in cholesterol profile by measuring total-, high density lipoprotein- and low density lipoprotein-cholesterol after rosiglitazone is added to a pre-existing Type 2 Diabetes treatment regime.

Methods A non-blinded cross-sectional cohort study was designed. 59 patients were enrolled. Patients who were rosiglitazone naïve were prescribed 4mg of oral rosiglitazone added-on to their current medication for a period of 10 weeks. Ankle-brachial pressure index, HbA_{1c}, serum plasminogen activity levels and fasting cholesterol profile were

taken at the start and end of the study period. Patients were requested not to change their medication dose nor regime throughout the study.

Results 48 out of 59 patients completed the study. Mean ankle-brachial pressure index was 1.06 ± 0.12 pre-, and 1.07 ± 0.13 post-rosiglitazone (p-value was 0.439). 4 patients (8.3%) had an ABPI ratio of less than 0.90 indicating presence of significant peripheral artery disease. Mean serum plasminogen activity (%) was 96.00 ± 14.77 before rosiglitazone, and 111.98 ± 15.83 after (p-value of 0.006). Initial mean HbA_{1c} (%) was 9.76 ± 2.06, and second mean was 9.25 ± 2.03 (p-value was <0.001). Mean total cholesterol (mmol/L) was 4.95 ± 1.02 before rosiglitazone and 5.32 ± 0.94 after (p=0.003). Mean high density lipoprotein cholesterol (mmol/L) at the beginning was 1.32 ± 0.37 and 1.47 ± 0.41 at the end (p<0.001). Finally, mean low density lipoprotein cholesterol (mmol/L) concentration was 2.89 ± 0.85 at the start and 3.08 ± 0.96 at the end (p= 0.098).

Conclusion This study shows that oral rosiglitazone 4mg daily significantly improves serum plasminogen activity levels, indicating improvement in fibrinolytic activity. There is also a significant reduction in HbA_{1c}, rise in total cholesterol as well as high density lipoprotein-cholesterol levels in line with the findings of previous studies. The rise in ankle-brachial pressure index and low density lipoprotein-cholesterol measurements however, were not significant in this study. Not many patients had significant peripheral artery disease in this study compared to previous ones before this. However, more

research is needed regarding the relation between use of rosiglitazone and atheroma reduction.

1. INTRODUCTION

1.1 Background and Epidemiology

The first ever recorded case of diabetes was by the 3rd Dynasty Egyptian physician Hesy-Ra in 1552 (Shampo et al, 1981). Since then, much has been learnt regarding this disease: its pathophysiology, the types of persons affected, the natural history of the different types of diabetes as well as the complications associated with it, to name a few. According to the World Health Organisation (WHO) Fact Sheet on Diabetes Mellitus, patients can generally be divided into 3 groups: those that have Type 1 Diabetes Mellitus (which occurs due to lack of insulin production), Type 2 Diabetes Mellitus patients (those having impaired insulin secretion and/or peripheral insulin resistance) as well as diabetes which presents in pregnancy, termed Gestational Diabetes Mellitus. The American Diabetes Association also recognises secondary diabetes, ie diabetes mellitus which is due to another underlying pathology eg haemochromatosis or Cushing's syndrome.

The WHO estimates that currently more than 180 million people worldwide have diabetes, and this number is likely to more than double by 2030. An estimated 1.1 million people died from complications related to diabetes in 2005. Initially thought to be a disease afflicting affluent nations, it has been shown that almost 80% of diabetes deaths occur in low and middle-income countries. Having said this however, diabetes deaths are projected to increase by over 80% in upper-middle income countries between 2006 and 2015 (WHO Fact Sheet, 2006). The majority of patients are Type 2 diabetics and thus efforts are targeted towards reducing the incidence of new cases as well as associated morbidity and mortality. In Malaysia, the prevalence of diabetes was estimated to be

0.65% in 1960, 2.1% in 1982, and 6.3% in 1986. This increased to 8.3% in 1996 (Mustaffa, 1990; Second National Health and Morbidity Survey, 1997; Zaini, 2000). A study by Mafauzy et al in 1999 though showed that the prevalence of diabetes in the Peninsular Malaysia North-Eastern state of Kelantan to be as high as 10.5%. Apart from this the subjects studied also had a high prevalence of obesity, hypertension and hypercholesterolaemia (Mafauzy et al, 1999).

1.2 Pathophysiology of Type 2 Diabetes Mellitus

Type 2 Diabetic patients by far comprise the largest group at more than 90% of overall diabetic patients. The progress of this disease is relentless; it advances from an early asymptomatic stage with insulin resistance to mild postprandial hyperglycaemia and finally full blown diabetes requiring pharmacological intervention. Three basic metabolic defects define Type 2 Diabetes Mellitus: insulin resistance, a non-autoimmune pancreatic β-cell insulin secretory defect, as well as increased hepatic gluconeogenesis. The cause of these metabolic defects, and therefore the cause of Type 2 Diabetes Mellitus, is largely unknown. It has been shown though to have a strong genetic component and is found more frequently in certain families and ethnic minority groups. Many acquired factors also play a role in the pathogenesis of the disease. Factors that contribute to insulin resistance include obesity, aging, and a sedentary lifestyle. Other contributing factors to the insulin secretory deficiency include long-term glucotoxicity and elevated free fatty acid levels.

The metabolic sequence of events that eventually lead to type 2 diabetes herald the development of hyperglycaemia by years or even decades. Insulin resistance is the initial

metabolic defect and as it worsens, more defects in insulin secretion occur that result in increased hepatic glucose production. At this point, there is a further rise in fasting blood glucose (termed "impaired fasting glucose"- IFG). This worsens later on, leading to impaired glucose tolerance – IGT. The progression from IGT to early type 2 diabetes is marked by a decrease in β-cell function and thus a further decline in insulin secretion. It is the gradual pancreatic β-cell failure to compensate for insulin resistance as well as hyperinsulinaemia that precedes the development of Type 2 Diabetes. Two more pathophysiological changes become apparent during the transition from IGT to type 2 diabetes: 1) insulin resistance becomes more severe due to the factors mentioned above, and 2) there is a concomittant increase in baseline hepatic glucose production. Though early Type 2 diabetes may be as asymptomatic as IGT, the degree of hyperglycaemia is now severe enough for the development of microvascular complications. Apart from this, IGT and insulin resistance are associated with low levels of HDL cholesterol, increases in triglycerides, and hypertension. These metabolic problems, in combination with changes in factors involved in the coagulation cascade, may result in accelerated atherosclerosis and early macrovascular complications (Ramlo-Halsted et al, 2000).

1.3 Treatment of Type 2 Diabetes Mellitus

Thus, in an effort to reduce cardiovascular-associated mortality treatment goals are directed at improvement of classical cardiovascular risk factors eg optimal control of glycaemia, hyperlipidaemia (namely LDL-C), and blood pressure according to guidelines. Treatment essentially can be divided into two groups, ie non-pharmacological interventions which include lifestyle changes eg cessation of smoking, proper diet control and exercise. Pharmacological interventions eg oral hypoglycaemic agents or insulin

injections may also be required; these will be elaborated on in later paragraphs. According to the International Diabetes Federation (IDF) Western Pacific Region guidelines regarding control of diabetes, the optimal fasting plasma glucose level should be between 4.4-6.1mmol/L, optimal non-fasting plasma glucose level of 4.4-8.0mmol/L and optimal HbA_{1c} of less than 6.5% (Asian-Pacific Type 2 Diabetes Policy Group, 2002).

Apart from the lifestyle modifications mentioned above, there are many classes of oral hypoglycaemic agents (OHAs) available for the pharmacological treatment of Type 2 diabetes. There is metformin, a biguanide which suppresses hepatic gluconeogenesis and increases peripheral insulin sensitivity. It is usually used as a first-line agent in obese patients as it promotes peripheral glucose uptake and also has an appetite suppressing effect. Secondly, there is the sulphonylurea group of OHAs. These act as insulin secretagogues; they may be used as first-line agents in non-obese Type 2 diabetics or as add-on therapy. Thirdly are the meglitinides. These are also insulin secretagogues, but have a shorter duration of effect than sulphonylureas. Meglitinides are better suited to address the issue of post-prandial hyperglycaemia. Fourthly are the thiazolidinediones. These drugs are peroxisome proliferator-activated receptor-gamma agonists which amongst other effects, acts as an insulin sensitiser. Then there are the α-glucosidase inhibitors which reduce glucose absorption at the intestinal brush border.

Patients who fail to achieve glycaemic targets using OHAs should be considered for insulin therapy. Insulin itself comes in many preparations; from the ultra-short acting

ones up to insulins with 24-hour duration of action. A combination of OHAs and subcutaneous insulin injections may also be considered depending on the situation. An inhalable insulin preparation is also available commercially (Hite et al, 2006).

A new drug which has entered the market is exenatide, an incretin mimetic which promotes pancreatic insulin secretion. It comes in the form of an subcutaneous injection administered twice a day. It has been approved for adjuvant, as well as monotherapy in Type 2 diabetics.

The International Diabetes Federation has also produced Global Guidelines for the treatment of Type 2 Diabetes Mellitus. Recommendations are divided into comprehensive, standard and minimal care. With standard care, a patient is started on OHAs when lifestyle interventions alone are unable to maintain target glycaemic control. Support for lifestyle measures are maintained throughout the periods of use of oral medications.

The glycaemic response is monitored every 2-6 months via FBS, RBS or HbA_{1c}. If targets are not achieved, it should be considered whether to increase the dose of the oral agent or to initiate another. Patients ought to be started with metformin unless there is evidence or risk of renal impairment, titrating the dose over several weeks to minimise discontinuation due to gastro-intestinal upset. The renal function should also be regularly monitored. The next step is to use sulfonylureas when metformin fails to control glucose concentrations to target levels, or as a first-line option in the non-overweight person. A

low-cost drug should be considered first before initiating more expensive ones, unless the situation warrants differently.

Education needs to be provided and, if appropriate, self-monitoring to guard against the consequences of hypoglycaemia. If compliance is an issue, once-daily sulfonylureas should be an available option. In patients who may not have regular meals due to a hectic schedule for example, the rapid-acting insulin secretagogues (eg meglitinide group of OHAs) may be useful compared to sulfonylureas. However, this may only work if the patient can still produce insulin and has good insulin sensitivity. PPAR-γ agonists (thiazolidinediones) are used when glucose concentrations are not at target levels, adding it: a) to metformin as an alternative to a sulfonylurea, or b) to a sulfonylurea where metformin is not tolerated, or c) to the combination of metformin and a sulfonylurea. In such cases the patient should be screened for cardiac failure, as this is a contra-indication for the use of thiazolidinediones, and the patient should be warned of the possibility of developing significant peripheral oedema. Failing these steps, α-glucosidase inhibitors can be used as a further option. These may also have a role in patients who do not tolerate other therapies. Doses are increased stepwise, and other oral glucose-lowering drugs are added one by one until blood glucose control is at target levels. All along, the patient should be monitored for deteriorating glycaemic control despite these measures; the patient may need early insulin therapy if OHAs fail to adequately control blood sugar levels (Clinical Guidelines Task Force, 2005).

1.3.1 Thiazolidinediones

It has been increasingly shown however that a specific pharmacological intervention can also confer benefit beyond its initial scope of use. An example of this would be the use of statins which have been shown to reduce vascular inflammatory markers as well as improve plaque stability, an effect beyond just LDL-c reduction alone (Kinlay et al, 2003). Amongst the plethora of oral hypoglycaemic agents which are available though, more evidence is emerging to show that rosiglitazone (Avandia®, Glaxo-SmithKline) has effects other than reduction of blood glucose levels eg improvement in fibrinolysis, reduction in inflammatory markers as well as reduction in blood pressure and atherosclerotic plaque (Haffner et al, 2002; Mohanty et al, 2001; Law et al 2000; RECORD study preliminary report, due 2008; Raji et al, 2003).

Rosiglitazone is a member of the thiazolidinedione class of antidiabetic agents which improves glycaemic control by improving insulin sensitivity. It is a highly selective and potent agonist for the peroxisome proliferator-activated receptor-gamma (PPAR-γ). In humans, PPAR receptors are found in key target tissues for insulin action such as adipose tissue, skeletal muscle, and liver. Activation of PPAR-γ nuclear receptors regulates the transcription of insulin-responsive genes involved in the control of glucose production, transport, and utilisation. In addition, PPAR-γ responsive genes also participate in the regulation of fatty acid metabolism (Avandia product monograph, 2006). With regards to insulin sensitivity and secretion, the thiazolidinediones have been proven to lower fasting and postprandial glucose concentrations as well as free fatty acid concentrations in clinical studies. Studies also showed that insulin concentrations were also reduced,

indicating that thiazolidinediones act as insulin sensitisers. This has been confirmed by direct measurements in human subjects. Nondiabetic subjects or those with Type 2 Diabetes were treated for three to six months with troglitazone, rosiglitazone, or pioglitazone. This increased their insulin-stimulated glucose uptake in peripheral tissues. Other similar studies showed that thiazolidinediones increased hepatic insulin sensitivity (ie the ability of insulin to reduce endogenous glucose production) and insulin sensitivity in adipose tissue (measured from the ability of insulin to suppress free fatty acid concentrations). This improvement in insulin secretory responses were also shown to be present in subjects with impaired glucose tolerance and Type 2 Diabetes Mellitus, even after being adjusted for an improvement in insulin sensitivity. An unwanted side-effect though is that these improvements are generally accompanied by weight gain and an increase in the subcutaneous adipose-tissue mass.

At the cellular level, PPAR-γ is needed for normal adipocyte differentiation and proliferation as well as fatty acid uptake and storage. What thiazolidinediones do is that they increase the number of small adipocytes and the subcutaneous adipose-tissue mass in studies in animal models. Due to this as well as the high level of PPAR-γ expression in adipose tissues, have led to the hypothesis that thiazolidinediones exert their insulin-sensitising actions either directly (the "fatty acid steal" hypothesis) or indirectly, by means of altered adipokine release, thus modulating insulin sensitivity outside adipose tissue. The "fatty acid steal" hypothesis describes thiazolidinediones as promoting fatty acid uptake and storage in adipose tissue and thus increasing adipose-tissue mass and protects other insulin-sensitive tissues eg skeletal muscle and the liver, and possibly