TROGLITAZONE, A NOVEL ANTIDIABETIC DRUG
-NEW AVENUE FOR TREATING INSULIN RESISTANCE-

Hiroyoshi Horikoshi
Research Institute and R&D Planning and Management Department
Sankyo Co., Ltd.

Impaired insulin action in Type 2 diabetes is thought to lead to hyperglycemia, with both environmental and complex genetic factors playing key roles. Although the primary lesion in Type 2 diabetes is unknown, a number of studies suggest that metabolic defects in the liver, skeletal muscle and fat, and pancreatic β-cells contribute to the disease. These metabolic abnormalities are characterized by the overproduction of hepatic glucose, impaired insulin secretion, and peripheral insulin resistance.

In current pharmacological treatment of Type 2 diabetes, sulfonylurea (SU) drugs have mainly been used as oral hypoglycemic drugs to stimulate endogenous insulin secretion from β cells. SU drugs, however, sometimes aggravate the disease by causing fatigue of the pancreatic β cells, which leads to reduced drug efficacy after long-term treatment. This class of drugs also leads to enhanced obesity arising from the stimulation of endogenous insulin secretion in obese Type 2 diabetic patients, plus an increased incidence of SU-induced hypoglycemia.

Since 1980, a major challenge has been made by us to develop a potential pharmacological therapy for the treatment of insulin resistance in peripheral tissues and/or suppression of abnormal hepatic glucose production in Type 2 diabetic patients. Such a drug would be expected to have fewer side effects and retain long-term efficacy.

PHARMACOLOGICAL PROFILE OF TROGLITAZONE

To explore the hypoglycemic and hypoinsulinemic effects of our compounds in vitro, we established both an insulin receptor binding assay and glucose uptake studies to further characterize insulin action in adipocytes from drug-treated animals in 1983. We tested many compounds and found, at the end of 1983, that troglitazone was the first sample consisting of a thiazolidinedione ring and a chroman ring of a structure of vitamin E that could improve hyperglycemia, hyperinsulinemia, and hypertriglyceridemia in diabetic KK mice. Troglitazone also increased glucose uptake in drug-treated adipocytes.

These findings suggested that troglitazone increased not only insulin sensitivity but also insulin responsiveness. Finally, troglitazone (CS-045,Noscal) was selected as a potential antidiabetic agent after intensive testing.

Hypoglycemic Actions in Diabetic Rats and Mice

The pharmacological profile of troglitazone was assessed by means of several genetic and acquired animal models of Type 2 diabtes with insulin resistance, including obesity, hyperglycemia, hyperinsulinemia, and hypertriglyceridemia. Troglitazone significantly decreased both plasma glucose and insulin levels in several species of genetically insulin resistant animal models of Type 2 diabetes ,such as KK, ob/ob, and db/db mice, and ZDF rats. Troglitazone also decreased plasma lactate, triglycerides, free fatty acids, and ketone.

bodies.

Similar results were obtained in other genetically obese Zucker fatty rats and in non-genetic models of insulin resistance, high fructose-fed rats. Plasma glucose, insulin, and lipids were observed in these animals. Impaired glucose tolerance was essentially normalized in Zucker fatty rats treated with troglitazone, decreasing remarkedly both postprandial glucose and insulin levels after oral glucose load.

In contrast, troglitazone did not significantly improve hyperglycemia in streptozotocin-treated rats, a model of Type 1 diabetes. Insulin tolerance tests, however, showed that treatment with troglitazone significantly improved insulin sensitivity. Moreover, combined treatment with trolgitazone potentiated the hypoglycemic action by exogenous insulin and lowered the insulin dose to achieve an appropriate plasma glucose level.

Islet Studies

Impaired islet cell function is a characteristic feature of Type 2 diabetes. Pancreatic functions were examined in db/db and KK diabetic mice at early and late stages of diabetes. In both models, chronic treatment with troglitazone increased regranulation of islet β cells and insulin content in pancreas. Furthermore, the direct effect of troglitazone on insulin secretion was assessed by a pancreatic perfusion system. However, troglitazone alone did not stimulate insulin secretion.

Hepatic Glucose Production

Abnormal elevation in hepatic glucose production due to increased gluconeogenesis is a major cause of fasting hyperglycemia in Type 2 diabetes. We, therefore, analysed gluconeogenesis in diabetic KK mice treated with troglitazone. In KK mice, gluconeogenesis is markedly elevated compared with nondiabetic control mice. Troglitazone reduced the rates of gluconeogenesis in diabetic mice but had no effect in normal mice.

We further determined the enzymatic step at which troglitazone influenced the glycolytic/gluconeogenic pathway. The levels of the glycolytic intermediates were measured from liver of control and troglitazone-treated db/db and KK mice and subjected to crossover analysis. A crossover point was observed between fructose-6-phosphate and fructose-1,6-bisphosphate (FBP). These data suggested that trogltiazone affected the interconversion of the two intermediates. Drug treatment led to a significant decrease in FBPase activity.

Over all, these data suggest that the reduced gluconeogenesis may result from a decrease in FBPase protein.

Glucose Utilization

Adipose tissue is one of the major targets that is highly responsive to insulin. We decided to determine whether troglitazone affected adipose cell function. 2-Deoxyglucose uptake by insulin was evaluated in isolated adipocytes in treated and untreated animals. Insulin produced a dose-dependent increase in 2-deoxyglucose uptake in adipocytes from both groups. Troglitazone treatment significantly enhanced basal glucose uptake and shifted the insulin dose response curve to the left. To explore troglitazone's effect in cultured cells, troglitazone was chronically exposed in L6 myocytes. Basal glucose uptake was significantly increased. They also observed increases of Glut 1 transporter content and a small but significant increase in Glut 4 transporter levels.

Insulin promotes the formation of glycogen from glucose. The effect of troglitazone on the activity of glycogen synthase was studied in both HepG2 and BC3H-1 cells. Treatment of both cell types with troglitazone produced a dose-dependent increase in glycogen synthase activity.

In metabolic mode of action, troglitazone appears to directly improve the action of insulin in liver, skeletal muscle, and adipose tissues. Troglitazone increases glucose disposal rates, and decreases hepatic glucose output.

Molecular Mechanism of Action

A number of studies have suggested that thiazolidinediones exert their primary insulin-potenciating effects through the regulation of transcription. This has been examined in cultured adipocyte cells. Addition of troglitazone in 3T3-L1 preadipocytes increased both the differentiation rate as well as the percent of proliferation. However, troglitazone did not significantly influence the stimulation of mitogenesis by insulin or serum in 3T3-L1 fibroblasts.

It has been demonstrated that thiazolidinediones induce adipocyte differentiation by interacting with members of PPAR(peroxisome proliferation activated receptor) family. Thiazolidinediones can serve as ligands for PPARγ, a PPAR family member highly expressed in adipose tissue but also found in other tissues. PPAR activation thus initiates the cascade of transcriptional events which culminate in the expression of C/EBP *a* and adipocyte differentiation. The apparent mechanism of action of the thiazolidinediones like troglitazone involves binding to nuclear receptors that regulate gene expression. Troglitazone may interact with PPARγ. PPAR exists in a heterodimer with another nuclear receptor RXR(retinoid X receptor). The binding of troglitazone to PPARγ can induce the interaction of the complex with specific DNA sequences in thiazolidinedione responsive gene. This DNA binding may involve the displacement of a corepressor molecule on ligand binding.

Data now suggest that the modulation of PPARγ by troglitazone plays a critical role in regulating intermediary metabolism, although the full spectrum of genes that respond to this drug, either directly or indirectly, awaits further characterization.

CLINICAL STUDIES

Clinical trials were conducted in Japan from 1987 through 1993. Troglitazone was first licensed to Warner-Lambert for co-development and co-marketing with Sankyo in North America in 1991. It was also licensed to Glaxo-Wellcome in Europe in 1992. Based on our world-wide development program, clinical trials were conducted in North America from 1991 to 1996 and in Europe from 1992 to 1997.

Based on the results of the preclinical and clinical studies, troglitazone is expected to be a clinically useful hypoglycemic drug in type 2 diabetic patients with insulin resistance who are inadequately responding to diet or SU drug or insulin therapy. In 1997,troglitazone has been marketed for the treatment of Type 2 diabetes in Japan and of Type 2 diabetes requiring insulin in the United States, and will be marketed for the treatment of Type 2 diabetes in the United States and UK by the end of 1997 and the Europian Continent in early 1998.

NEW AVENUES FOR TROGLITAZONE TREATMENT

It has been widely recognized that insulin resistance and associated diseases are closely related by a set of metabolic abnormalities known as insulin-resistance syndrome. This syndrome is manifested by compensatory hyperinsulinemia, including impaired glucose tolerance (IGT), polycystic ovarian syndrome (PCOS), dyslipidemia, vascular diseases, central obesity, and hypertension. Any of these symptoms represent a major risk factor for coronary artery disease. Troglitazone is being studied in several of these human disease states.

Hypertension.

Chronic troglitazone treatment of Zucker fatty rats and high fructose-fed rats, both useful animal models for mild hypertension associated with obesity and insulin resistance, could significantly improve mild hypertension and hyperinsulinemia. If so, troglitazone may provide a new pharmacological approach to the mangement of hypertension in obese and/or Type 2 diabetic patients with insulin resistance.

Impaired glucose tolerance.

An initial study has been conducted in which obese patients with or without IGT were treated with troglitazone for 3 months. The metabolic abnormalities associated with insulin resistance syndrome were strikingly corrected with troglitazone. The National Institute of Health has initiated a large-scale, multicenter study, the Diabetes Prevention Program (DPP), that includes troglitazone as one of the treatment arms in IGT patients with insulin resistance. This study will attempt to determine whether conversion of high-risk individuals to diabetes can be prevented.

Polycystic ovarian syndrome(PCOS).

PCOS is another interesting disease state caused by insulin resistance. This syndrome is characterized by chronic anovulation and hyperandrogenism, which is considered one of the features in insulin resistance states. Hyperinsulinemia is also common in this syndrome. A recent clinical study found that troglitazone improved total body insulin action in PCOS by lowering plasma insulin levels, and that it also reduced concomitantly elevated testosterone and luteinizing hormone levels toward the normal range.