Effect of STZ dose and dosing regimen on the diabetes phenotype of C57BL/6J mice fed a high fat diet

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INTRODUCTION

The combination of a high fat diet (HFD) and low dose streptozotocin (STZ) has been developed as a non-genetic model of type 2 diabetes1. Briefly, animals are fed a high fat diet to induce insulin resistance. Subsequently animals are dosed with a low dose of STZ such that frank hyperglycaemia is evident in the presence of, or slightly elevated, plasma insulin concentrations. The HFD/STZ model is believed to offer advantages over genetic models of diabetes (e.g. ob/ob and db/db mice; ZDF rat). For example, whilst such genetic models exhibit a diabetic phenotype, patients with type 2 diabetes do not exhibit the extreme hyperinsulinaemia seen in these models2, and, in addition, the underlying genotype is of limited relevance to the aetiology of diabetes in human patients (i.e. the models have limited face validity and poor construct validity). In the present study we have further evaluated the HFD/STZ model in the mouse using a number of STZ dosing regimens in order to investigate how STZ dose affects the diabetic phenotype of the animals.

MATERIALS AND METHODS

C57BL/6J mice (n=9) were given ad libitum access to a high fat diet (D12492: 60% kcal as fat; Research Diets, USA) or control diet (D12450B: 10% kcal as fat; Research Diets, USA) for 3 weeks. Animals on the high fat diet were randomised on the basis of body weight and dosed by the intraperitoneal route as detailed below with either vehicle or STZ (Sigma):

- Vehicle (0.05M citric acid)
- 50 mg/kg daily for 3 days
- 50 mg/kg daily for 5 days
- 75 mg/kg daily for 3 days

Animals on the control diet (lean) were dosed with vehicle. The first day of STZ treatment was regarded as Day 1. Animals were weighed daily. Glycaemic control was assessed by an oral glucose (2 g/kg) tolerance test (OGTT) after an overnight fast on Days 11 and 33. Animals were terminated on Day 39 and unfasted blood samples collected for determination of HbA1c, ketone bodies and triglycerides using standard kits and reagents. At termination a urine sample was collected from the bladder and assayed for glucose content. The pancreas was dissected and prepared for determination of insulin content (n=5) or β cell mass (n=4; data not shown).

RESULTS

- Control animals fed a HFD exhibited significantly increased body weight (Fig. 1) and impaired glucose tolerance (Fig. 2) compared to the control animals fed a 10% fat diet. This was characterised by a large increase in fasted insulin and insulin AUC consistent with the development of marked insulin resistance though not frank diabetes.
- The combination of HFD and STZ produced varying degrees of weight loss (Fig. 1) and a diabetic phenotype of differing severity (Figs. 2 to 4). All animals treated with STZ exhibited a degree of insulin resistance (i.e. fasting plasma insulin levels were in excess of the lean controls on Day 11 (Fig. 2) and Day 33 (data not shown but similar).
- 50 mg/kg ip for 3 days: non diabetic phenotype (Day 33 fasting glucose: 12.1 mM); glucose intolerance; small elevations in HbA1c and ketone bodies; glycosuria.
- 50 mg/kg ip for 5 days: intermediate diabetic phenotype (Day 33 fasting glucose: 21.7 mM); elevated HbA1c, triglycerides, ketone bodies and reduced pancreatic insulin.
- 75 mg/kg ip for 3 days: marked diabetes (Day 33 fasting glucose: 22.5 mM); markedly elevated HbA1c, triglycerides, ketone bodies and reduced pancreatic insulin.

SUMMARY

- Access to a HFD led to impaired glucose tolerance.
- Small changes in the dosing paradigm for STZ can produce marked changes in the degree of diabetes produced in this model. Accordingly, when using this model to evaluate the efficacy of test compounds to improve glucose control and HbA1c, the selection of an appropriate STZ dose is critical.
- Studies evaluating diabetic nephropathy and/or neuropathy in this HFD/STZ model are ongoing.

REFERENCE


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