

UC Davis MMPC-Live Protocol Glucose-stimulated Insulin Secretion (in vivo)

Version: 1.0

Revision Date: 9/17/2012 Replaces version: None

Edited by: Fawaz G. Haj, - UC Davis MMPC-Live

Summary

Reagents and Materials

Protocol

Reagent Preparation

Summary:

The *in vivo* glucose-stimulated insulin secretion test is designated to determine alterations in insulin secretion by the pancreas upon a bolus IP glucose injection.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
45% Glucose solution	Fisher Scientific	NC0025179
Insulin Syringes	Fisher Scientific	14-826-79
Saline Solution	Fisher Scientific	L97753
Ultra Sensitive Mouse	Crystal Chem	90080
Insulin ELISA kit		
Heparin Sodium	Abraxis	401586B
Easy Check Glucose	JRS Medical	00-101
test strips		
Easy Check Glucose	JRS Medical	Y4209
monitor		

Protocol:

- 1. Fast mice for 16hours by taking away food the day before (3:00pm)
- 2. The following day, Calibrate the glucose meter according to the manufacturer's instructions.
- 3. Deprive mice from water then measure blood glucose level using a glucometer and remove immediately approximately $50 \mu l$ of blood from the tail via a tail tip cut and transfer directly onto a sterile 0.5 ml microcentrifuge tube containing 2 ul Heparin.
- 4. Centrifuge at 8000rpm for 5min then transfer plasma (supernatant) to a new 0.5ml microcentrifuge tube and freeze at -80°C.
- 5. Give the mouse an intraperitoneal injection of Glucose (2g/kg) with a 27 G needle
- 6. Continue to take blood samples from the initial tail cut at 2, 5, 15 and 30 min flowing injection and repeat step3 and 4.

NOTE: At the end of the experiment, wipe tail with 70% alcohol and allow drying. Ensure that blood loss from the tail stopped before placing the animal back to its cage.

Reagent Preparation:

• Dilute the glucose stock solution (45%) with saline to 20% by adding 20ml stock to 25ml 0.9% (w/v) sterile saline.