Oral Glucose Tolerance Test

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Summary:

The Oral Glucose Tolerance Test (oGTT) measures the clearance of an orally administered glucose bolus from the body. It is used to detect disturbances in glucose metabolism that can be linked to human conditions such as diabetes or metabolic syndrome. Animals are fasted for approximately 16 to 18 hours, fasted blood glucose (baseline), is measured before a solution of glucose is administered by oral gavage. Subsequently, blood glucose is measured at different time points during the following 2 hours.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Ultra2 Glucose meter	ONE Touch	n/a
Ultra test strips	ONE Touch/Any Brands	n/a
Sterile 0.9% saline solution	Any	n/a
50% Dextrose	USP	n/a
Sterile 20 ml syringe w. 16 G needle	Any	n/a
Sterile 1 ml syringe w gavage needle	Any	n/a
Scale	Mettler-Toledo	n/a
Timer	Any	n/a
Stainless steel disposable scalpers	Any	n/a
Gauze & 70% alcohol	Any	n/a
Lab coats/gloves/PPE	Any	n/a
Disinfectant	Nolvasan 10%	n/a
Topical anesthetic cream-optional	Any	n/a

Protocol:

1. SET-UP (fasting & weight of mice; set up of materials & calibration of glucose meters)

a. Fast mice overnight (16-18h) by transferring mice to clean cages with no food. **NOTE**: Ensure access to drinking water.

b. Prepare 20% saline working solution: draw 12 ml of 50% dextrose solution using a 20 ml syringe w. 16G needle into a 30ml sterile via; add 18 ml 0.9% saline using another 20 ml syringe w 16G needle. Mix by inversion. Label and date vial. **NOTE:** working glucose solution shelf life is 30 days.

c. Prepare workspace: lab mat, syringe, gauze, alcohol, glucose strips, glucose meters and freshly made 20% glucose solution, open experimental record sheet (Excel).

d. Calibrate glucose meters: Make sure that glucose meter and strips share the same code. Retrieve Control Solution (NOTE: bottle has a discard date; discard date is set 3 months from first use). Insert one strip in glucose meter. Shake control solution and squeeze one drop of solution on strip, making sure that the solution fills the strip completely. Compare the reading to the ranges written on the control solution bottle. Press the right arrow button to log the reading into the glucometer, L1 will appear on the top of the screen, and then press the power button on the side of the meter. Record reading on spreadsheet.

2. PROCEDURE

a. Weight mice in numerical order (order of oGTT test) so that the mice weighed last have some time to calm down before getting a baseline reading.

b. Place the mouse on the scale and record its weight. Place the mouse in a duplex cage with mouse of same sex. **NOTE**: no food in cage but access to water is critical. Order cages in the order of ipGTT test.

c. Calculate and record the volume of 20% glucose solution required (2g of glucose/kg body mass) for IP injection as follows: volume of IP glucose injection (µl) = 10 x body weight (g).

d. Set the test strip in the glucose meter, part of the way. If left in all the way, the meter tends to turn off before blood is administered. **NOTE: Recap the strip container every time after taking a strip. Do not use strips that have been left open to air for extended periods of time.**

e. Clean tail with gauze soaked with 70% alcohol, then dry tail with dry gauze.

f. Score the tip of the tail using a fresh or sterilized scalpel blade, only a millimeter or two is needed.

g. Push test strip all the way into the glucose meter, drop indicator will show up.

h. Milk the tail and discard the first small drop of blood by dabbing on lab mat.

i. A small drop of blood (<5µl) from the tail is placed on the glucose test strip in the glucose meter. Make sure blood fills up the strip and touches the base before the 5 second count down ends to get an accurate reading.

j. Record this baseline glucose fasting level.

k. Orally gavage the mouse with the appropriate amount of glucose solution determined earlier by weight and note the time-point of injection on the record sheet.

l. The blood glucose levels are measured at 15, 30, 60 and 120 minutes after glucose injection, by placing a small drop of blood on a new test strip and recording the measurements. Start the bleeding again by removing the clot from the first incision; massage the tail if blood flow is inadequate from base to tip. Record results on the record sheet.

m. Ensure that further blood loss from the incision is minimal by briefly applying pressure to the incision after each measurement. At the end of the experiment add food and recovery gel to the cage and make sure water is available to the animals.

n. Monitor the animals carefully to observe any abnormal behavior(s).

NOTE: **change gloves between mice**