Intraperitoneal glucose tolerance test (IPGTT) BCMLA_IPG_001

Purpose

The glucose tolerance test measures the clearance of an intraperitoneally injected glucose load 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 hours, fasted blood glucose levels are determined before a solution of glucose is administered by intra-peritoneal (IP) injection. Subsequently, the blood glucose level is measured at different time points during the following 2 hours.

Ontological description: MP:0005559 - increased circulating glucose level, MP:0005560 - decreased circulating glucose level, MP:0005293 - impaired glucose tolerance, MP:0005292 - improved glucose tolerance, MP:0005291 abnormal glucose tolerance, MP:0000188 - abnormal circulating glucose level.

Experimental Design

- **Minimum number of animals :** 7M + 7F
- **Age at test:** Week 56
- **Sex:** We would expect the results of this test to show sexual dimorphism

Equipment

1. Mouse restraining device (optional)
2. Glucose meter
3. Scalpel blade
4. Balance
5. Timer
6. Clean cages

Procedure

1. Weigh mice and fast mice overnight for approximately 16 hours by transferring mice to clean cages with no food or faeces in hopper or bottom of cage. Ensure that they have access to drinking water at all times.
2. Prepare an experiment record sheet, syringe and sticks for glucose measurement and glucose solution.
3. Weight the mouse.

4. 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 (l) = 10 x body weight (g).
5. Optional application of topical anesthetic cream:

a. Apply a small amount of topical anesthetic cream to the tail of the mouse, spreading over the tail evenly
b. Gently massage it in for ~10 seconds to enhance the effect of the anesthetic cream ensuring that the proposed incision site is fully covered
c. Allow an appropriate length of time for the local anaesthetic to take effect.

6. Optional: Restrain the mouse in the restraining device with the tail exposed.
7. Score the tip of the tail using a fresh or sterilized scalpel blade
8. Discard first small drop of blood. A small drop of blood (<5l) is placed on the test strip of the blood glucose meter. This is the baseline glucose level (t = 0) and is recorded in the experiment record sheet.
9. Remove the mouse from the restraining device.
10. Inject the mouse intraperitoneally with the appropriate amount of glucose solution, as previously determined (point 3) and note the time-point of injection.
11. The blood glucose levels are measured at 15, 30, 60 and 120 minutes (t = 15, t = 30, t = 60 and t = 120) 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. Results are recorded in the record sheet.
12. 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 to the cage and make sure that a plentiful supply of water is available to the animals.
13. At the end of the experimental session, place the mouse in a clean cage with water and food available ad libitum.
14. Monitor the animals carefully to observe any abnormal behavior(s).

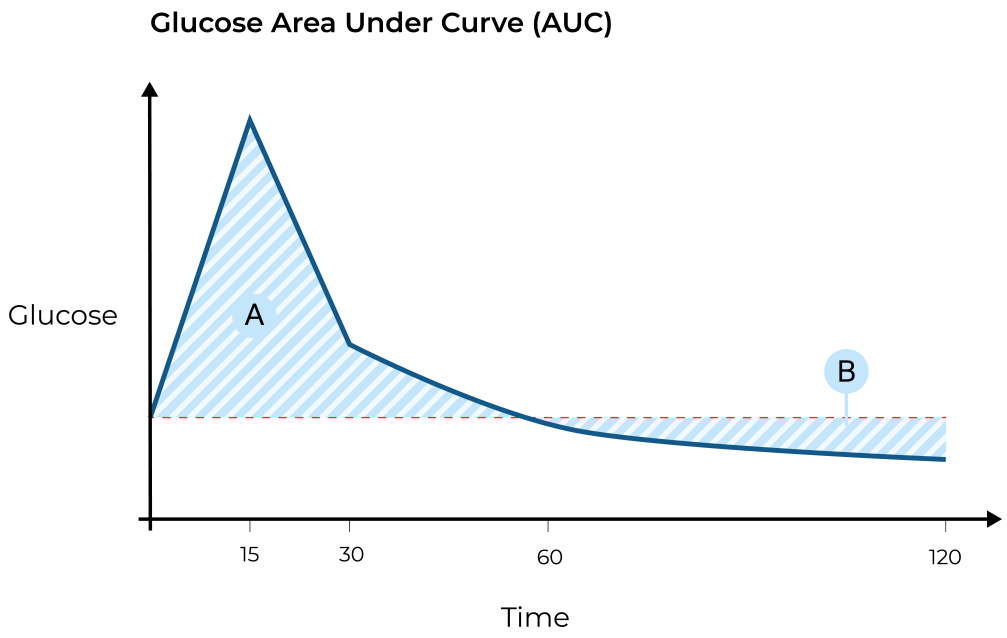
Notes

The information about the date of the experiment, that is the date when the measurement is performed, is an important parameter which is to be submitted in the Experiment xml file (dateOfExperiment="2013-02-28").

If the meter shows a 'high value', a parameter status code IMPC_PARAMSC_008 (âAbove upper limit of quantitationâ) should be submitted with the upper limit of the meter under metadata parameter IMPC_IPG_013_001 (âUpper limit of the meterâ).

Area Under Curve

Area under curve is calculated relative to the fasted blood glucose concentration (concentration at T0), rather than an absolute value relative to the X-axis (0mg/dl). By this method, the response of the animal is measured while taking into account its starting value.



NB In this example Area B is a negative value, as glucose concentrations have dropped to lower than the initial fasted value. If the animal shows a brief/weak spike followed by an extended period where glucose concentration drops far below the fasted value, area A might be smaller than area B i.e. the resultant total area under curve might be negative.

Data QC

Perform a QC check of the glucose meter routinely as outlined in the manual.

Metadata and examples

Metadata	Example
Equipment ID	ID of the machine used when more than 1 is used having same model and manufacturer. E.g. machine 1, machine 2, machine Minnie, machine Mickey Mouse, etc.
Equipment manufacturer	Manufacturer of the equipment. E.g. Roche Diagnostics Ltd.
Equipment model	Model of the equipment. E.g. Accu-Chek Aviva.

Mouse restrained	If the mouse is restrained in order to score its tail for blood analysis. E. g. Yes/No.
Type of strip	The strip used to analyse the blood can be for whole blood or for plasma. E.g. Plasma/Whole blood/Plasma, no maltose interference.
Experimenter ID	An ID of any format to be used coherently both inside the same procedure and for all procedures. E.g. Harw_001, or 1/2/3.
Date equipment last calibrated	Most recent date in which the equipment (or any part of) used in the procedure was subject to a calibration event.

Parameters and Metadata

Body Weight BCMLA_IPG_001_001 | v1.0

simpleParameter

Req. Analysis: false **Req. Upload:** true **Is Annotated:** false

Unit Measured: g

Description: body_weight

Blood glucose concentration BCMLA_IPG_002_001 | v1.0

seriesParameter

Req. Analysis: false **Req. Upload:** true **Is Annotated:** true

Unit Measured: mg/dl

Description: blood_glucose_concentration

Increments: 0, 15, 30, 60, 120,

Equipment ID BCMLA_IPG_003_001 | v1.0

procedureMetadata

Req. Analysis: false **Req. Upload:** true **Is Annotated:** false

Description: equipment_name

Equipment manufacturer BCMLA_IPG_004_001 | v1.0

procedureMetadata

Req. Analysis: true **Req. Upload:** true **Is Annotated:** false

Description: equipment_manufacturer

Options: Abbott, Roche Diagnostic, Lifescan, ARKRAY factory.Inc, Analox, Roche Diabetes Care,

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Equipment model BCMLA_IPG_005_001 | v1.0

procedureMetadata

Req. Analysis: true **Req. Upload:** true **Is Annotated:** false

Description: equipment_model

Options: 32004-02, Accu-Chek Aviva, Accu-Chek Performa, One Touch Ultra 2, Super Glucocard II GT-1640, GLUCOCARD G+meter GT-1820, Alphatrak 2, GM9, Accu-Chek Aviva Connect, Accu-Chek Guide,

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Mouse restrained BCMLA_IPG_006_001 | v1.0

procedureMetadata

Req. Analysis: false **Req. Upload:** true **Is Annotated:** false

Description: mouse_restrained

Options: yes, No,

Type of strip BCMLA_IPG_007_001 | v1.0

procedureMetadata

Req. Analysis: true **Req. Upload:** true **Is Annotated:** false

Description: type_of_strip

Options: Whole blood, Plasma, Plasma, no maltose interference,

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Experimenter ID BCMLA_IPG_008_001 | v1.0

procedureMetadata

Req. Analysis: false **Req. Upload:** true **Is Annotated:** false

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Date equipment last calibrated BCMLA_IPG_009_001 | v1.0

procedureMetadata

Req. Analysis: false **Req. Upload:** false **Is Annotated:** false

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Fasted blood glucose concentration BCMLA_IPG_010_001 | v1.1

simpleParameter

Req. Analysis: false **Req. Upload:** false **Is Annotated:** true

Unit Measured: mg/dl

Derivation: incrementValue('BCMLA_IPG_002_001', 0)

Initial response to glucose challenge BCMLA_IPG_011_001 | v1.1

simpleParameter

Req. Analysis: false **Req. Upload:** false **Is Annotated:** true

Unit Measured: mg/dl

Derivation:
sub(incrementValue('BCMLA_IPG_002_001', 15), incrementValue('BCMLA_IPG_002_001', 0))

Area under glucose response curve BCMLA_IPG_012_001 | v1.1

simpleParameter

Req. Analysis: false **Req. Upload:** false **Is Annotated:** true

Unit Measured: (minutes*mg)/dl

Derivation: areaUnderCurve('BCMLA_IPG_002_001', 5)

Upper limit of the meter BCMLA_IPG_013_001 | v1.3

procedureMetadata

Req. Analysis: false **Req. Upload:** false **Is Annotated:** false

Unit Measured: mg/dl
