



# Intraperitoneal Insulin Tolerance Test

Version: 1

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## Summary:

An intraperitoneal insulin tolerance test or ipITT is designed to determine the sensitivity of insulin-responsive tissues in the rodent. This is determined by measurement of glucose remaining in the circulation over time after a bolus ip insulin injection.

## Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Humalin® R	Eli Lilly	R-100
Insulin Syringes	Fisher Scientific	14-826-79
Saline Solution	Fisher Scientific	L97753
Easy Check Glucose test strips	JRS Medical	00-101
Easy Check Glucose monitor	JRS Medical	Y4209

## Reagent Preparation:

Dilute the stock solution (100 U/ml) with saline to 0.5 U/mL (1/200 dilution) by adding 5µl stock (100 U/mL) to 995 µl 0.9% (w/v) sterile saline

## Protocol:

1. Fast mice for 4 h only by taking away food early in the morning (7:00am).
2. Calibrate the glucose meter according to the manufacturer's instructions.
3. Deprive mice from water then remove approximately 5µl of blood (one drop) from the tail via a tail tip cut and transfer directly onto a glucose indicator strip.
4. Measure blood glucose immediately in a glucometer.

5. 4. Give the mouse an intraperitoneal injection of insulin (0.5 U/kg) with a 27 G needle.
6. 5. Continue to take blood samples from the initial tail cut before the insulin injection and at 15, 30, 45, 60 and 120 min.
7. 6. Between each of these time points, return the mouse to its cage and monitor it every minute.

## **NOTE:**

1-The mouse is given an intraperitoneal injection with a 27G needle of insulin. Before performing the experiment, we will have to determine if the mouse strain is insulin resistant or glucose tolerant, so as to avoid giving the wrong dose of insulin.

2-For insulin resistant mice, a higher dose of insulin could be used as they do not respond as well as insulin sensitive mice to insulin. However, we will make sure that the dose of insulin used will not cause hypoglycemia.

3-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.