**Insulin and Glucose Tolerance Test Protocol**

**Razani lab**

**REAGENTS & SUPPLIES**

* Stock Insulin (100 units/mL) = 0.1 units/ uL Humulin R
* 29.5G 500 uL syringes (Exel #26028)
* Glucose test strips (Arkray # 762050)
* Glucometer (Arkray #760001)

Insulin Tolerance Test

* Weigh mice in morning (8-9 am) and fast for 5 hours. Can mark tails with sharpie to make mice easier to distinguish during ITT.
* To make **working** solution dilute Humulin stock 1:1000 in PBS **under hood in a sterile manner.**
* Volume to inject **of working solution (uL)** is weight of mouse in g **times 7.5**
* Prepare and label syringes with appropriate volumes for each individual mouse
* Inject mice staggered every 2 minutes (or 1 minute if you are confident you can keep up)
* Measure tail vein glucose via glucometer at 0, 30, 60, 90, 120 mins. (Measure at 0 **before** injecting). Always wipe residual blood from tail with a paper towel before using glucometer. Can do 15 minute time point if there are fewer mice or if you are fast.

Glucose Tolerance Test

* Use 50% D-Glucose (Medical grade)
* Dilute under hood in sterile PBS 1:5 to make 10% D-Glucose
* Weigh mice in morning (8-9 am) and fast for 5 hours. Can mark tails with sharpie to make mice easier to distinguish during GTT.
* Multiply weight (in Grams) by 10 to get volume in ul of 10% D-glucose to inject (final dosing = 1g gluc /kg body mass)
* Aliquot appropriate volume of 10% D-glucose per mouse in individual syringes
* Inject mice staggered every 2 minutes (or 1 minute if you are confident you can keep up)
* Measure tail vein glucose via glucometer at 0, 30, 60, 90, 120 mins. (Measure at 0 **before** injecting). Always wipe residual blood from tail with a paper towel before using glucometer. Can do 15 minute time point if there are fewer mice or if you are fast.