

Embryo Cryopreservation

Core responsibilities

The core staff will superovulate wild-type females, mate them with the investigator's practiced stud males, isolate 8 cell embryos, freeze embryos in a controlled rate freezer using glycerol as a cryoprotectant, and store the resulting straws in two different locations. The core aims to freeze between 200-400 embryos. It is likely that two freezing sessions are performed to obtain this number. For all initial cryopreservations, the core will thaw a test straw and transplant 20-40 embryos into 2-4 recipients to test viability. ***Due to the variability in fertility and embryo fragility between mouse lines/strains, the core does not guarantee embryo yield and/or viability.*** In instances of low embryo yield/viability, the core will work with investigators to optimize fertility and achieve cryopreservation with minimal expense to the Investigator. The investigator will receive a Cryopreservation Inventory form indicating the number of embryos/straws frozen and their location.

Investigator Responsibilities

Step 1: Complete an Embryo Cryopreservation Service Request form and consult with Andrei Golovko about either doing this in the investigator's room or importing mice into the TIGM quarantine.

Step 2: Provide the core with 6-8 practiced stud males 3-6 months of age.

Step 3: (optional) Females of the following strains are available at TIGM and can be used for timed matings: CD1, FVB, 129/SvEvBrd, C57BL/6J and C57BL/6N. If females of a different background are preferred, please also arrange for importation of 12-15 of those animals, 3-5 weeks of age, to TIGM quarantine.

Timeline

Day 1: Superovulated wild-type females are plugged.

Day 3: Embryos are flushed, cryopreserved and stored in liquid nitrogen.

Day 3-4: 1-2 straw of embryos are thawed and cultured to the blastocyst-stage and transferred to recipient mothers for live birth.

Day 21: Pups born and investigator notified whether the cryopreservation was successful.