

Mouse Bone Marrow Cell Preparations

(Greg A. Perry, Ph.D.)

Equipment:

Mouse Sterile scissors Petri dish 1cc Syringe 25 gauge Needle 15ml Conical tubes Kim-Wipes Gauze

Reagents:

70% Ethanol Sterile media (HBSS or RF10)

Method:

- 1. Sacrifice mouse by cervical dislocation.
- 2. Wet fur with 70% ethanol. Clip fur at hip area with small scissors.
- 3. "De-pants" the leg by pulling the fur down off the leg toward the foot.
- 4. Remove the leg from mouse by cutting into pelvis. (Be sure not to cut into the femur!)
- 5. Holding the femur in one hand and the tibia in the other, bend the bones backward until they pop out of the kneecap.
- 6. Use scissors to cut foot from tibia and femur from pelvis.
- 7. Remove as much muscle and connective tissue from the bones as possible. For the femurs the muscle will peel back after popping the knee cartilage off with your fingernail.
- 8. Place cleaned bones on sterile gauze soaked in sterile media in a Petri dish.
- 9. Under a sterile hood, flush the marrow from the bones using a 25 gauge needle and a 1cc syringe into a 15ml conical tube with 5-10ml of RF10 media.
 - a. For the Femur:

Clip off the head of the femur with a sterile scissor. Puncture the epiphyseal plate with the needle. Fill the syringe with media, then insert the needle into the epiphyseal end and flush the marrow out the other end. Refill the syringe and repeat the flush from the femoral-head end.

b. For the Tibia:

Clip both ends of the bone. Fill the syringe with media, then insert the needle into one end and flush the marrow out the other end. Refill the syringe and repeat the flush from the other end.