Embryonic Stem Cell/Chimeric Mouse Production Policies of the Mouse Genetics Laboratory (MGL)

Mouse Strains:
The MGL maintains the inbred stock strain C57BL/6 for production of chimeric mice. The MGL will maintain this stock strain as part of the basic embryonic stem cell injection service. If your laboratory requires an alternate strain, a special request must be made to the MGL and any additional costs, including mouse purchase and housing per diems, becomes the responsibility of the Investigator.

Embryonic Stem Cells and MEFs:
The MGL can provide low passage, germline competent ES cells to the Investigator. The MGL provides CJ7 ES cells, derived from the mouse strain 129S1/Sv+P +Tyr-cKitSl-/+J. The appropriate control for this strain is JAX#002448, 129S1/SvImJ. The MGL can also provide primary NEOMEFS for use as the feeder layer for the CJ7 ES line. The MGL is also able to provide DR4 MEFS as a primary feeder resistant to G418, puromycin, hygromycin and 6-thioguanine.

Generation of modified ES clones:
The MGL has established a Gene Targeting Service. The investigator develops the targeting vector and provides this vector to the MGL as a linearized fragment. The MGL Gene Targeting facility will generate G418 resistant colonies using CJ7 ES cells. Please refer to the Gene Targeting Policy page for more specific details of this service. Individual labs may choose to do the targeting and then provide targeted clones to the MGL.

Targeted Clones:
Targeted clones will be accepted for injection based on the following quality assurances. Every clone must be a low passage cell line that exhibits good “ES” morphology and has a normal karyotype. We recommend that the Investigator provide multiple clones for injection. The MGL can receive your clones as cryopreserved vials or growing on plates - this is the choice of the investigator.

Injection Service:
Three days of blastocyst injection is performed with your clone of genetically-modified ES cells. We will inject at least 75 blastocysts and implant into at least 5 pseudopregnant recipients. All offspring are transferred to investigator after weaning. Charges will be made once injections are completed. No guarantee is made about the number of live born mice or the quality of chimerism that will be obtained. High quality ES cell lines (e.g. CJ7) normally generate at least 15 offspring. Prior to injection all clones must be MAP tested (RAR website: http://www.ahc.umn.edu/rar/mapsub.html) and tested for a normal diploid karyotype.
Finally, a photograph, taken at 40X power, of a field of colonies of the ES clone in question growing on MEFs must be emailed to Sandi Horn at hornx001@umn.edu.

At the time of embryo injection and implantation, the recipient females will be moved to the Protocol of the Investigator and all housing per diems for recipient females and resulting offspring will become the responsibility of the Investigator. A successful round of injection is expected to result in 10-40 pups. The number of chimera produced and the degree of chimerism is dependant on the quality of your ES clone. Your chimera will remain in the MGL until they are mated to determine germline transmission, the chimera will be mated to C57BL/6 females. Agouti offspring, N1’s from the chimera X B6 mating, will be ear punched for identification and a 1 cm tail clip will be provided to the Investigator. We ask that you provide genotype results to the MGL in no more than 3 weeks from receipt of your tail clips. The MGL highly recommends confirming all positive gene targeted offspring using a Southern Blot. Once germline transmission has been confirmed, all positive offspring and germline chimera will be transferred by the MGL to the Investigators assigned animal holding facility.