Terms of Service for Transgenic Mice

Transgenic Service: TWO transgenic founders are guaranteed for each DNA sample submitted. If you did not obtain two founders, the Transgenic Facility will perform another injection with the same DNA free of charge. If the second injection still does not generate positive mice, it is likely that this is caused by the quality or nature of the DNA samples and the Transgenic Facility will not perform any injection on this construct unless a new request is submitted. Transgenic Facility will guarantee at least 8 pups from second injection. Germline transmission of the transgene is not guaranteed. Guarantee is invalid if transgene causes embryonic lethality or the nature of submitted transgene construct prevents the production of transgenic founder. We produce transgenic mice in B6CBA/F1 background. Cost of C57BL/6 transgenic mice production will be three times the cost of B6CBA/F1.

BAC Transgenic Service: Client purifies the BAC (TMSR's BAC purification protocol can be downloaded HERE). TMSR guarantees 120 injected eggs will be transferred into four fosters. TMSR does not guarantee the number of founders from this injection.

Guaranteed BAC Transgenic Service: Client may pay one additional injection charge and submit bacteria containing the BAC on a plate to TMSR. TMSR will purify and inject the BAC and guarantee TWO BAC transgenic founders.

Terms of Service for Chimeric Mice

ES Cell Injection: A minimum of 16 blastocysts will be injected and implanted (through uterine transfer) into two foster mothers for each ES clone submitted. **There is no guarantee on the outcome of the injection, such as the number of pups, chimerism, and germline transmission.**

Terms of Service for ES CELL ELECTROPORATION

1. Client should provide 100 ug targeting vector at >0.5ug/ul concentration. Transgenic Facility will linearize and electroporate client's gene targeting vector into ES cell line chosen by client (listed in the previous page), carry out drug selection, pick antibiotic-resistant ES clones in two 96-well plates (we guarantee 150 clones that will grow and yield DNA for genotyping after picking), make two duplicated copies of genomic DNA (in 96-well plate format) and one copy of frozen ES cells (in two 96-well plates with MEF feeders, stored at -80C). Client is responsible for screening of targeted ES clones by PCR or Southern hybridization. Client needs to pay additional fee to recover/expand frozen targeted ES cells, extract DNA from targeted ES cells for re-confirmation of targeted ES clones and thaw frozen ES cells for blastocyst injection. Client may then pay the regular fee for blastocyst injection. **BAC targeting vector:** The Facility will pick one 96-well plate and 75 valid DNA samples are guaranteed. BAC targeting vector yields less drug-resistant colonies.

2. Client needs to report back to the Transgenic Facility within **30 days** once the DNA plates arepicked up. This will guarantee the successful recovery of frozen ES cells. The ES cells are frozen in 96-well plates at –80C. Frozen ES cells usually thaw well if thawed within 30 days. Most ofthe frozen ES cells will not survive thawing if stored at –80C for more than 45 days. Client should have the screening strategy (PCR or Southern hybridization) well tested before submitting the targeting vector to Transgenic Facility to ensure timely successful screen of ES cells will survive thawing. **Shorter time is required to expand the targeted ES cells and the ES cells will have higher chance to retain totipotency.** Transgenic Facility will not guarantee the recovery of any ES cells frozen for more than 30 days at -80 C.

3. ES CELL ELECTROPORATION SERVICE doesn't include the recovery/expansion of targeted frozen ES cells and production of chimeric mice. There is **no guarantee** on targeted ES cells in terms of production of chimeric mice and germline transmission.

4. The charge for ES CELL ELECTROPORATION SERVICE is **\$4,400** for Cancer Center member, **\$5,500** for non-member. Please check our fee schedule for other service charges.

Terms of Service for ES CELL EXPANSION

1. Client needs to report back to the Transgenic Mouse Shared Resource (TMSR) within **30 days** after the DNA plates are picked up from TMSR. This will guarantee the successful recovery of frozen ES cells. The ES cells are frozen in 96-well plates at -80C. Frozen ES cells usually thaw well if thawed within 30 days. Most of the frozen ES cells will not survive thawing if stored at -80C for more than **45 days**. Client should have the screening strategy well tested before submitting the targeting vector to TMSR to ensure timely screen of ES cell colonies. The sooner the frozen ES cells are thawed, the higher percentage of the frozen ES cells will survive thawing. Shorter time is required to expand the targeted ES cells and thawed ES cells will retain totipotency better. TMSR will not guarantee the recovery of any ES cells frozen for more than 30 days at -80C. TMSR guarantees the recovery will be greater than 50% if frozen ES clones are thawed within 30 days.

2. TMSR will thaw the frozen ES clones and expand ES clones in wells of 24-well plates. Each ES clone will be frozen in two vials in liquid nitrogen tank and aliquot of the same clone will be expanded further to extract genomic DNA for clients to reconfirm the recovered ES clones. These DNAs serve two purposes: (1) to reconfirm that correct ES clones are recovered, (2) enough genomic DNA will be available for client to re-check these ES clones with multiple probes or PCRs to fully validate these targeted ES clones.