



Medical College of Georgia Mouse ES Cell & Transgenesis Core

C57BL/6 ES CELL & CHIMERIC MOUSE SERVICES

Investigator: _____ Building/Room #: _____

Contact person: _____ Phone: _____

Address/email: _____

SECTION A

This application represents a request for:

- Generation of chimeras from an existing ES cell clone (go straight to Section C below)
- Generation of homologous recombinant ES cell clones and germline transmission

Name for this job: _____ Date: _____

Besides the information requested in Section B, the targeting construct DNA should be prepared to a final concentration of 1.5 µg/ml (at least 40 µl) as recommended on the Medical College of Georgia Mouse ES Cell & Transgenesis Core Facility website. An agarose gel image should be provided showing both 20 and 200 ng of the purified DNA construct preparation. This gel image **AND** the information requested in Section B below **MUST** be provided before the application will be considered for review.

SECTION B

Advice about targeting vector construction can be obtained from numerous sources including the Mouse ES Cell & Transgenesis Core Facility website. Even the most experienced Investigators are encouraged to submit a preliminary plan **BEFORE** making a targeting construct. Whether this is a preliminary plan or a completed application for service, you **MUST** provide the following information:

- [] 1. Map of the targeting vector with selection cassettes labeled, the genomic locus, and the anticipated targeted locus with positions of endonuclease sites and probes for the screening strategy
- [] 2. Source of genomic DNA (library, mouse strain, etc.) _____
- [] 3. Written description of the strategy for identifying appropriately targeted ES cell clones
- [] 4. Genomic Southern blot using one or more flanking probes, with size markers

In addition, the following information is **required** for evaluation by the Institutional Biosafety Committee:

1. What is the Ensembl gene ID or the MGI symbol ID of the targeted gene? _____
2. What is the anticipated effect of the planned genetic manipulation? _____

3. Did construction of the targeting vector use any viral vector? ____ If yes, provide details on attachment
4. If you have one, provide your Biosafety approval number for the targeting vector _____

(Proceed to Section D)

SECTION C

Source and strain of ES cell clone _____ Passage # _____

Has this clone produced germline transmission before? Yes No

Explain: _____

- [] **Karyotype of ES cell clone** (attach copy of data)
- [] **Mycoplasma test result** (attach test results)

SECTION D

As the Investigator applying for service with job name _____, initially requested on date _____, I, _____ acknowledge and agree that the Core will charge my funding source below a non-refundable set up fee of \$ _____ for electroporation service to generate homologous recombinant clones (if applicable). Upon successful completion of electroporation (as defined below), a further charge of \$ _____ will be due. I acknowledge and agree that any homologous recombinant clones remain the property of the Core, but that they may be obtained if all of the following conditions have been met: **(1)** attempts to generate mice are complete; **(2)** the Core's standard Material Transfer Agreement has been executed, and; **(3)** a fee of \$1,000 for up to 3 clones and all other outstanding charges have been paid.

In order to generate mice as in **Section A** (after the generation of clones, if applicable), I will incur a non-refundable fee of \$ _____. Upon completion of service, the balance owed will be \$ _____.

The balance will be charged upon completion of the service as judged by the Core Director in accordance with the basic service requested (e.g. chimera production or germline transmission based on coat color). Successful completion of an electroporation service is defined as an electroporation that either generates at least 100 clones for screening by the Investigator and results in at least one homologous recombinant clone OR yields at least 3 homologous recombinants if fewer than 100 clones are provided. If the service is not completed successfully as defined above, the Core will repeat the electroporation once more at no charge - however, a minimum number of clones will no longer be guaranteed. Should the Investigator wish to have further attempts performed, this will be only at the discretion of the Core Director and a fresh set-up fee will be charged. The Core will do its best to deliver services within a reasonable timeframe. However, the Core cannot offer time or germline transmission guarantees.

I acknowledge that the Core purchases some stock mice from trusted sources and that despite regular, stringent health status monitoring the possibility of introduction of an infectious agent is real. I understand that germ line-positive founders (or chimeras if requested) will be quarantined for health status monitoring **at my expense** (an **additional** charge of about \$300) before delivery to my vivarium. I agree that the Core is not responsible for any consequences of transfers to my vivarium, and that transfers are subject to Lab Animal Services approval. I agree that I will incur per diem charges for any delay in transfer and/or execution of my responsibilities as detailed on the Mouse ES Cell & Transgenesis Core Facility website.

1. Account # for billing set up fee _____
(*other forms of payment can be arranged for applicants outside of the Medical College of Georgia*)

2. Account # for balance (if different) _____

3. Animal Use Approval #: _____ **Approval Date:** _____

Investigator's Signature _____ **Date** _____

Comments by Core Director: _____

Response by Investigator: _____
