UMMS TRANSGENIC ANIMAL MODELING CORE (TAMC)

ESTABLISHMENT OF NEW MOUSE ES CELL LINES

THE FACILITY WILL PERFORM:

1. Isolation of blastocysts from uterine horns of up to 5 pregnant E3.5 females. Blastocysts (between 10-40) will be rinsed in DMEM, and placed onto an inactivated SNL76 (STO) feeder cell layer in embryo growth media plus LIF. The blastocysts will be grown *ex vivo* in CO_2 incubator for approximately 5 days, at which time the embryos will have hatched and the inner cell mass will have expanded.

2. Clumps of inner cell mass cells will be disaggregated in trypsin, and individual clones of ES cells established in 96-well micro-titer dishes bearing inactivated SNL76 (STO) feeder cells.

3. The Facility will expand select clones for freeze down and DNA preparation, and will provide the DNA to the Investigator for further analysis.

4. Frozen vials of the properly targeted clones will be given to the Investigator.

5. Although the Core will proceed with due care and diligence, no guarantee can be made regarding the viability or pluripotency of each embryo-derived ES cell line.

Charges for establishment of up to 8 new ES cell lines = $1,000.00

P.I. Name _____ *Date Received* _____

Department _____

Account Number _____

Mouse line _____

ES cell line nomenclature _____ *TOTAL CHARGES $* _____

The Facility requires that the plugging and harvesting of the embryos be coordinated, and that the pregnant females (or flushed blasts) be brought to the Core or Rivera lab for further manipulation and plating.

X _____ ***X*** _____

UMMS INVESTIGATOR / date *UMMS TAMC* / date

revised: 7/1/2017