VASCULAR CELL TRANSFORMATION

The Redox Molecular Signaling Core utilizes a two-step immortalization technique involving lentiviral expression of a tetracycline-responsive transactivator (rtTA3) and lentiviral expression of a construct encoding both large and small T antigen under the control of a tet-on promoter. When co-transfected in target cells, these constructs induce robust cell proliferation in the presence of doxycycline (a tetracycline analog) due to T antigen expression. However, these cells rapidly lose T antigen expression upon doxycycline removal to better enable study of cellular processes. To initiate Vascular Cell Transformation, contact the Redox Molecular Signaling core facility at RedoxMolSignalCore@lsuhsc.edu and schedule a meeting with Core Leaders to discuss the project timeline and deliverables.

To be provided by investigator:

- A completed Work Order Form brought to the meeting with Core Leaders.
- One 10 cm dish of primary vascular cells to be transformed. Cells should be provided at approximately 40% confluence. Alternatively, investigators can choose to transform vascular cells isolated from their desired mouse models using the core endothelial and smooth muscle isolation services.
- Should specialty media be required for vascular cell culture, 200 mLs of specialty media should be provided.

To be generated by the core:

- Transformed vascular cells showing small and large T antigen expression in response to doxycycline. Transduction efficiency of >50% and sensitivity to doxycycline will be verified prior to cells being returned to the investigator. The percent of transformed cells in the population will increase with further passage.

Timeline: 2-3 weeks