Force, Dimensionality and Stem Cell Fate

Grant Award Details

Force, Dimensionality and Stem Cell Fate

Grant Type: SEED Grant

Grant Number: RS1-00449

Investigator:

Name:	Valerie Weaver
Institution:	University of California, San Francisco
Type:	PI

Human Stem Cell Use: Embryonic Stem Cell

Award Value: $529,762

Status: Closed

Progress Reports

Reporting Period: Year 2

View Report

Grant Application Details

Application Title: Force, Dimensionality and Stem Cell Fate

Public Abstract:

Human embryonic stem cells (hESCs) are cells derived from human embryos early in development before their fate has been sealed. These cells grow and differentiate in response to a variety of stimuli to eventually give rise to all of the differentiated tissues in the body. By exploiting the remarkable potential of hESCs to differentiate into multiple cell lineages, medicine stands to benefit enormously. To do so requires a comprehensive understanding of the optimal conditions to grow and differentiate these cells. What is known is that the physical environment in which hESCs reside plays an important role in regulating their tissue-specific differentiation.

Recent work has highlighted the importance of the composition and structure of the extracellular matrix (ECM), within which hESCs exist in vivo, in directing hESC differentiation during embryonic development. In an embryo, hESCs differentiate in a dynamic and structurally distinct three-dimensional (3D) ECM, rich in nutrients and exogenous stimuli (force). Mechanical stimulation (via matrix compliance and externally applied force) dramatically influences the formation and development of the embryo. Despite these compelling observations, information regarding the mechanisms whereby matrix compliance and external force regulate hESC differentiation in 3D is extremely limited. Instead, the majority of the research on hESCs has been in two-dimensional (2D) culture on stiff plastic substrates, despite the lack of physiological relevancy.

To address this issue, we will investigate the role of the ECM in 2D and 3D on hESC behavior using biomaterials with well-defined compositional and physical properties. We will assess the role of exogenous force by building a bioreactor designed to impart oscillatory compressive loading on hESCs cultured in 3D ECMs. We will test whether force modulates hESC fate by altering the function of the small RhoGTPase Rac. We will achieve this goal by: determining whether matrix compliance influences hESC differentiation in 2D and 3D and exploring the role of force on Rac activity and function, building a bioreactor capable of imparting controlled cyclic compressive loading to 3D hESC embedded in engineered biomaterial constructs, and by characterizing the effects of dynamic compression on hESC fate by manipulating the loading system. Because our appreciation and understanding of the mechanisms whereby matrix compliance and external force regulate hESC fate is extremely limited, this work would not likely be federally funded. These studies are essential to illustrate the critical role of matrix force in hESC fate and lay the foundation for future studies aimed at clarifying molecular mechanisms. The work will also assist in establishing defined, in vitro systems that more closely recapitulate the in vivo behavior of hESCs to permit their pluripotent propagation, and ensure their correct specification thereby ensuring the safe application of hESCs for human therapy.

Statement of Benefit to California:	The growing worlds of human embryonic stem cell (hESC) science, bioengineering and regenerative medicine offer hope in the treatment of many diseases ranging from breast cancer to diabetes to Parkinson's. Integral to the stem cell therapy treatment of these diseases is a fundamental understanding of the intricate mechanisms that govern stem cell growth, differentiation, maintenance and commitment. Much effort has concentrated on the role of exogenous biochemical supplementation to direct hESC differentiation and commitment. We seek to bridge the gap between the worlds of stem cell biology and bioengineering, adding an innovative approach to direct hESC lineage specification through the three-dimensional (3D) modulation of the extracellular matrix (ECM) microenvironment. We propose the building of a novel bioreactor to elucidate the roles that mechanical stimulation and the structure-function relationship of the ECM environment play in hESC differentiation and commitment in 3D. Through the development of this novel bioreactor, we will clarify and optimize parameters and mechanisms governing the growth, differentiation, maintenance and stability of hESCs that might otherwise go unnoticed with biochemical stimulation alone. These objectives are particularly relevant given the critical importance of establishing defined in vitro conditions in which embryos and ESCs can be derived and propagated with minimal contamination. Our studies also will have significance with regards to assisting investigators to improve their ability to rigorously maintain non-differentiated hESCs under conditions that more accurately recapitulate the in vivo situation, and thereafter aid in the generation of directed lineage specification of hESC differentiation. The latter point is particularly relevant because directed cell lineage specification should greatly reduce the potential for transplanted hESC to spawn terato-carcinomas upon in vivo transplantation. We envision that once our prototype Force Bioreactor has been generated and validated, the Scientific community will have access to our facilities and experimental approaches so that they will be able to apply similar methods to their basic and translational stem cell studies. We will facilitate this information and technology transfer through the active dissemination of our research findings, as well as via the establishment of the {REDACTED} of which the P.I. {REDACTED} has been appointed as Director. The approach in this proposal is both innovative and multidisciplinary, bridging together multiple genres of science and engineering. In today's rapidly evolving world of research, the greatest impacts in stem cell research will be made by those willing to break out of traditional scientific paradigms, merging fields that will ultimately contribute solutions to the diseases that we face.