Discovery of mechanisms that control epigenetic states in human reprogramming and pluripotent cells

Grant Award Details

Discovery of mechanisms that control epigenetic states in human reprogramming and pluripotent cells

Grant Type: Basic Biology III

Grant Number: RB3-05080

Project Objective: The objective of this grant is to gain understanding of human X chromosome inactivation, using naive hiPSC as a model, and to study human X chromosome reactivation during reprogramming toward pluripotency.

Investigator:

Name:	Kathrin Plath
Institution:	University of California, Los Angeles
Type:	PI

Human Stem Cell Use: iPS Cell

Cell Line Generation: iPS Cell

Award Value: $1,364,598

Status: Closed

Progress Reports

Reporting Period: Year 1

View Report

Reporting Period: Year 2

View Report

Reporting Period: Year 3

View Report

Grant Application Details

Application Title: Discovery of mechanisms that control epigenetic states in human reprogramming and pluripotent cells

Public Abstract: The CIRM Basic Biology Award III was developed to support basic research that enables the realization of the full potential of human stem cells and reprogrammed cells for therapies and as tools for biomedical innovation. This is particularly important since many fundamental issues related to the regulation of stem cell fate and reprogramming, especially with regard to human cells, remain to be resolved. X chromosome inactivation (XCI) is one of those fundamental processes of human development related to stem cell biology and reprogramming, that we know surprisingly little about, and we therefore propose to study the regulation of XCI in human cells in this proposal using human induced pluripotent stem cells (iPSCs) as model system.

A normal female has two X chromosomes and no Y chromosome and males have one X and one Y chromosome. To be equal with males, females must shut off one of two X chromosomes during embryonic development by inducing XCI, such that only one X chromosome remains active in every cell of the female body. Females even become genetic mosaics by randomly inactivating either the X chromosome inherited from the father or the mother, which has important consequences for the clinical phenotype of X-linked diseases between the two sexes.

Studies on XCI in the mouse model system have revealed that female embryonic stem cells (ESCs) carry two active X's and that XCI must be initiated when these cells are induced to differentiate. XCI is an epigenetic phenomenon that occurs without alterations in the primary sequence of DNA by formation of a repressive heterochromatin structure. Intriguingly, this heterochromatin structure can be erased when adult murine cells are reprogrammed to the ESC-like state of iPSCs.

Findings in the human system are less clear as typical female human ESCs and iPSC lines have an inactive X chromosome that can change its composition with extended culturing indicating potential epigenetic instability. It is now also thought that these cells don't represent the same developmental state as mouse ESCs and iPSCs. In agreement with this notion, human pluripotent cells with two active X chromosomes have recently been generated that appear to resemble the mouse ESC state. Thus, there are now at least two different human pluripotent states that also differ in their X chromosome status. We believe that our proposed studies of XCI regulation during differentiation and reprogramming in human cells and in these different human ESC states will not only unveil mechanisms underlying this fundamental silencing process and human development, but also be instrumental for the careful characterization of these different human pluripotent states. This is particularly important given that human ESCs and iPSCs carry a tremendous promise for therapeutic applications and for modeling of human development and diseases, and that the XCI status in these cells also will have specific implications for modeling of X-linked diseases.

Statement of Benefit to California: The proposed project will benefit the state of California and its citizens as follows:

1. Better characterization of human induced pluripotent cells (iPSCs) for use in disease studies and cell-based treatments. Human iPSCs hold great potential for regenerative medicine to treat many devastating injuries and diseases such as Alzheimer's disease, Parkinson's disease, diabetes, cancer, rheumatoid arthritis, and spinal cord injuries. Our studies will help to define different human iPSC states and assess their epigenetic stability. This would be beneficial to the people of California as tens of millions of Americans suffer from diseases and injuries that could benefit from a detailed characterization and understanding of the biology of human iPSCs. Such advances would benefit the health as well as the economy of the state of California.

2. Advances in understanding the reprogramming process to the iPSC state. X chromosome inactivation is one of the most dramatic forms of developmentally regulated heterochromatin formation that is reset during the reprogramming process. Understanding how the inactive X chromosome reactivates should reveal epigenetic mechanisms that stabilize the differentiated state and therefore eventually enable the development of safer and more efficient reprogramming approaches. In addition, the X chromosome status may be used as an indicator of complete reprogramming to the naïve pluripotent state.

3. Studies of X-linked diseases using human iPSCs. As X chromosome inactivation can alter the consequences of X-linked mutations, a better understanding of the X chromosome state in human hiPSCs is essential for their use in the modeling of X-linked diseases in vitro.

4. Education and jobs for next-generation California scientists. In addition to creating highly skilled jobs, the proposed research activities will create an interdisciplinary education environment for training the next generation of California citizens at all levels, including high school, undergraduate, graduate students, as well as postdoctoral fellows.
