IITB-Monash Research Academy

Indian Institute of Technology Bombay | MONASH University

An Indian-Australian research partnership

Project title

Generation and characterization of cell cycle regulation of induced pluripotent cells (iPSCs)
Project number: IMURA0099

Monash University supervisors: Dr. Paul Verma,
Email: Paul.Verma@med.monash.edu.au

IITB supervisor: Prof. Dulal Panda
Email: panda@iitb.ac.in

Research Academy theme/s

List only the research academy theme/s that is relevant to the project

1. **Biotechnology and stem cell research**

The problem

Cell Therapy has the potential to proved cures for a number of currently incurable diseases. Embryonic Stem Cells (ESCs) offer a promising resource to generate a wide range of tissue types for the treatment of a variety of degenerative and auto-immune diseases, such as Parkinson's Disease, Multiple Sclerosis, spinal cord injuries, diabetes and myocardial infarction. A key issue in the clinical use and commercialization of SC-based therapies is the potential for these cells and their derivatives to elicit immune rejection by receiving patient[1-3]. Generally, compatibility is less than absolute and even tissue matched organ transplant patients need to take immuno-suppressant drugs for the rest of their lives to prevent rejection of the transplanted organ. This is both expensive and has multiple undesirable side effects for the recipient.

While a number of routes have been proposed to overcome or circumvent immune rejection of transplanted cells, the most attractive option for cell therapy is to transplant tissue derived from SC lines genetically identical (autologous) to individual patients, thereby overcoming immune rejection issues.

Recently numerous research papers have reported the generation of human ES-like cells (called iPS cells) by over-expressing 4 key genes in somatic cells.[4] We believe that while this research is incredibly exciting and promising, several issues need to be addressed for iPS technology to be clinically relevant- 1) The approach requires genetic modification of the adult cells. 2) One of the genes used for generation of the human iPSCs is a cancer gene and it was originally reported in mouse studies to result in tumours in mice generated from these cells, and 3) The only method of gene transfer that has worked in both mice and human uses retroviral vectors, that require high levels of infection. Careful evaluation of iPSCs, including comparisons with ESCs is essential before the cells can be considered for clinical use. As cell cycle regulation of ESCs are markedly different from other somatic cells, understanding the cell cycle and cell cycle checkpoints in both stem cells and somatic cells would provide an insight into generation of pluripotent cells efficiently. This would provide the ability to regulate cell proliferation and pluripotency. Several adult tissues contain non-dividing differentiated cells, which are continuously replaced by differentiation of stem progenitor cells. Cancer develops in these tissues due to miss regulation of the stem cells. Therefore, stem cells are attractive targets for cancer chemotherapy and to understanding control of cell proliferation.

Project aims

1. Generation and characterization of iPSCs
2. Comparison of cell cycle check points of iPSC lines with ESCs and investigate approaches to regulate cell cycle in somatic cells aimed at efficient generation of iPSC lines
3. Screen anticancer drugs using stem cells as targets.
4. To look for differences or similarities in the cell cycle and cell cycle checkpoints in stem cells and somatic cells.

Expected outcomes

Understanding the process of reprogramming of iPSCs at the cellular level by comparison with ESCs, which is essential for translation to future clinical outcomes.
The ability to generate reprogrammed somatic cell with therapeutic potential will significantly impact on the application of cell therapy.
The project may provide a significant understanding of cell cycle regulation and differentiation.
Aims to discover novel anticancer drugs.

Project plan

The student will need training with stem cell culture and viral vector generation and transfection for maintenance and generation of iPSCs before generating the initial data. This would be provided at Monash University under the supervision of Drs Verma and Sumer. The training will also include characterization of stem cells by *in vitro* and *in vivo* methods including-

- Vector design and generation
- Viral generation and transduction of target cells
- Isolation and maintenance of pluripotent cells
- In vitro differentiation as embryoid bodies (EBs)
- Characterization by protein localization, gene expression
- In vivo differentiation by teratoma formation in Severe Combined Immune Deficient (SCID) mice

Following this period the student will return to IITB and analyse the cell lines generated. She will also use the acquired skills to transfer stem cell expertise to the laboratory of Prof. Panda and generate additional iPSC lines for analysis. The investigations at IITB will include-
I) Comparison of cell cycle check points of iPSC lines with ESCs and investigate approaches to regulate cell cycle in somatic cells aimed at efficient generation of iPSC lines. II) Screening anticancer drugs using stem cells as targets. III) Understanding differences or similarities in the cell cycle and cell cycle checkpoints in stem cells and somatic cells.