THE UNIVERSITY OF HONG KONG
Graduate School

Partnerships in Teaching and Learning in RPg Education

Title:	Genome Editing in Mammalian Cells with CRISPR/Cas9
Pre-requisite(s), if any:	Participants should have biomedical science background (e.g. students from Faculty of Medicine, or School of Biological Science in Faculty of Sciences)

Description

Designed engineered nucleases (ZFNs, TALENs, CRISPR/Cas9 *etc.*) facilitate genome editing in kinds of cell lines, embryos and animals. Among them, CRISPR/Cas9 has generated considerable excitement. It is cost-effective and time-efficient for targeted-genome editing. In this workshop, we will share our experience in engineering the genome of human induced pluripotent cells (hiPSCs). HiPSCs have the potential to differentiate into all kinds of cell types in our body. By combining both technologies, multiple human diseases can be modeled comprehensively, and autologous cell transplantation therapies can also be tested.

The following students may be interested in this workshop: 1) Students who would like to engineer (knock-out/know-in a specific gene or DNA fragment) their own cell types with CRISPR/Cas9. 2) Students who work on stem cells.

Objectives

To help the participants obtain the background of genome editing technology and apply this technology to their own projects.

Learning outcomes

After the workshop, the participants should be aware of：
1. iPSCs and their application in disease modeling;
2. The principles of designing sgRNAs, donor plasmid (to generate reporter cell line) and ssODNs (to correct or introduce mutations);
3. The workflow of genome editing in human iPSCs.

After the workshop, the participants should learn the following skills：
1. Human iPSCs maintenance and passaging;
2. Design of sgRNAs and ssODNs for correcting or introducing mutations in a target site;
3. Single cell cloning.

Teaching and learning activities

Activities	**No. of hours**
1. Lecture / Presentation: 1.1 Reprogramming & application of hiPSCs 1.2 CRISPR/Cas9 mediate knockout 1.3 CRISPR/Cas9 mediate knockin	1.5 hours

2. Group discussion: 2.1 Design of CRISPR/Cas9 2.2 Knockout (NHEJ) strategies 2.3 ssODNs design 2.4 Donor plasmid design	1 hour
3. Hands-on experiments: 3.1 Electroporation with Neon transfection system (Demonstration only) 3.2 Single cell cloning 3.3 Picking colonies manually	1.5 hours
Total:	4 hours

Demonstration of achievement of learning outcomes

This course will help the participants acquire skills of hiPSCs maintenance and genome editing. By the end of the workshop, successful participants will be able to:

- demonstrate awareness of the differentiation potential of hiPSCs;
- demonstrate ability to design sgRNA, ssODNs and donor plasmid;
- demonstrate awareness of the workflow to achieve genome editing in hiPSCs;
- apply knowledge of this workshop to engineer their own cell lines.