

共五大題，請勿遺漏

Question 1 (10 points): Describe *5 essential know-hows* (or *whatever you know about primer and vector designs*) for successfully cloning a gene into a plasmid for transient expression in a mammalian cell line.

Question 2 (30 points) : HIV infects CD4 T, Macrophage, and Dendritic cells. Patients chronically infected with HIV usually have low CD4 T cell counts and high viral loads. A scientist has collected 100 serum samples from patients who were likely infected with HIV. He wants to know (1) *whether they are recently or chronic infected by HIV*, (2) *whether their IFN- α specific T cell responses (CD4 and CD8) are severely affected in these patients*. Please describe the possible approaches the scientist may perform. You should include (a) the experimental designs and (b) the rationales for the techniques used for studying these two questions. (Hints: MHC I & MHCII, Ag specific peptides, and T cell receptors; Note that you have plenty of resources. 10 points for the first question and 20 points for the second question)

Question 3 (20 points) : Tet-Off and Tet-On is a system using tetracycline (Tet) or its derivative (eg. Doxycycline) to control tightly regulated gene expression. What are the features of Tet-Off? What are the features of Tet-On? Please describe in details the underlying principle of these two systems.

Question 4 (10 points) : Assuming that you are studying a cell cycle-related protein expression. You intend to find how the protein expression will affect the fractions of cell during the various phases of cell cycle, ie, G1/G0, S, G2, M phase. Assuming that you have all the chemicals (ie, pharmacological inhibitors) and apparatuses (eg, flow cytometry) available. Please briefly describe two different chemicals and /or procedures that would help you to demonstrate that the cells you are harvesting are in a specific phase of cell cycle. Note that we are not asking you to state what proteins (eg, which cyclin, or which cyclin-dependent kinase) is expressed at specific phase.

(背面仍有題目,請繼續作答)

Question 5 (30 points) : "A dramatic scientific breakthrough in stem cell research was revealed by studies from Shinya Yamanaka's group at Kyoto University, Japan, showing that somatic cells can be reprogrammed into embryonic stem-like cells through overexpression of 4 genes-- Oct4, Sox2, Klf4 and c-Myc, both in mouse and human cells. These induced pluripotent stem (iPS) cells can differentiate into cells of all three germ layers, thus holding great potentials for many applications such as customized patient-specific cell clones. Based on this finding and your discipline, please propose a project to extend this exciting work. Your proposal should include at least *title* (3 points) and outlines of *background/significance* (8 points), *hypothesis* (3 points), *specific aims* (8 points), and *experimental designs/expected results/alternative approaches* (8 points), all in half a page. Your score will be judged mainly by the **rationale and novelty.**" (30 points in total)