CHAPTER 1

INTRODUCTION

1.1 Background of Study

Animal cell culture is a widely used technique that involves the isolation of cells, tissues and organs from animals. Cells extracted from animals were then grown *in vitro* or in artificial environment (Oyeleye, Ogundeji, Ola, & Omitogun, 2016). The term 'culture' in animal cell culture generally refers to keeping the cells alive and allowing them to replicate in optimum conditions ie temperature, pH, nutrient, and gas concentration. Animal cell culture has been used as a tool to study the mechanisms, functions and operations that occur within cells. The technique is also important in cancer research, vaccine manufacturing, drug and recombinant protein productions (Yao & Asayama, 2017). Animal cell culture is broadly divided into two types; suspension and adherent cell cultures. Suspension cell culture refers to culture of cells that free-float in the culture medium while adherent cell culture refers to culture of cells that require surface for attachment and growth.

In this research, Vero cells were adapted form serum supplemented medium to serum free medium. Serum provides nutrient for cell growth, proliferation and differentiation (Bottenstein & Sato, 1979). Fetal bovine serum (FBS) is a complex mixture of different factors and contain large amount of growth factors, protein, vitamins, trace elements, and hormones (Valk et al., 2010).

Serum-free medium is the medium without serum. The serum was replaced with selected hormones, promoting growth factor and stimulate differentiation of specific cells (Hayashi & Sato, 1976). There are many advantages of using serum-free medium. Firstly, it is chemically defined and allow controlled culture conditions in vitro. Besides, there is less variability in qualitative and quantitative culture medium

composition. Lastly, using serum-free medium is able to eliminate the potential source of microbiological contamination (Valk et al., 2004).

The conventional way to expand the number of cells was using roller bottles, stack plate propagators, multi-trays, and Povitsky bottles. However, these manual systems have drawbacks. First, it requires many operators. Besides, it is a system which is generally characterized by potential biohazard. Lastly, higher chances of contamination (Merten, 2015). These disadvantages can be alleviated by introduction of microcarrier technology (Wezel, 1967).

The microcarrier technology is used in stirred-tank reactors for large scale culture of adherent cells. There are two types of stirred-tank reactor which are stirred magnetic bar and ball-shaped eccentrically rotating agitator shown in Figure 1.1. When using the stirred-tank reactor, the culture medium will be homogenous where anywhere in the culture vessel the culture environment is the same. Using microcarriers allow alleviation of the limiting space in the flask because microcarriers have high surface to volume ratio. Besides, microcarrier technology uses homogenous stirring suspension allowing monitoring and controlling of environmental parameters such as pH, pO_2 and the concentration of medium components. This allows more optimal and reproducible cull culture process (Merten, 2015).

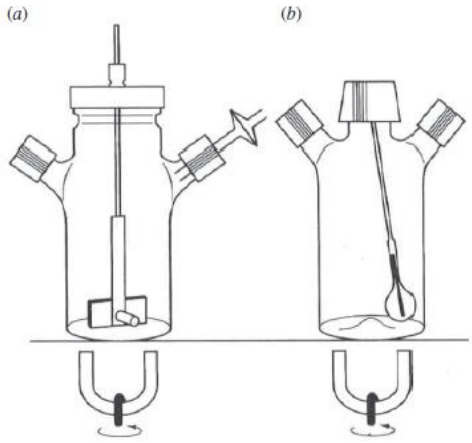

Figure 1.1: Two different types of spinner vessel: (a) Spinner with a magnetic bar stirrer and (b) spinner with a ball-shaped eccentrically rotating agitator (Merten, 2015).

Microcarrier based cell culture offer many advantages in animal cell culture. Microcarriers can produce high-yield culture of anchorage dependent cells by growing the cells in glass, stainless steel or disposable bioreactor. This is because it provides high surface are to volume ratio for the attachment and proliferation of the cell. Besides, using microcarrier can reduce the requirement of labour and culture medium. It has better process monitoring and control as well as simpler purification of biological products compared to conventional cell culture system (Arifin et al., 2016). Figure 1.2 below provides an overview of the experiment.

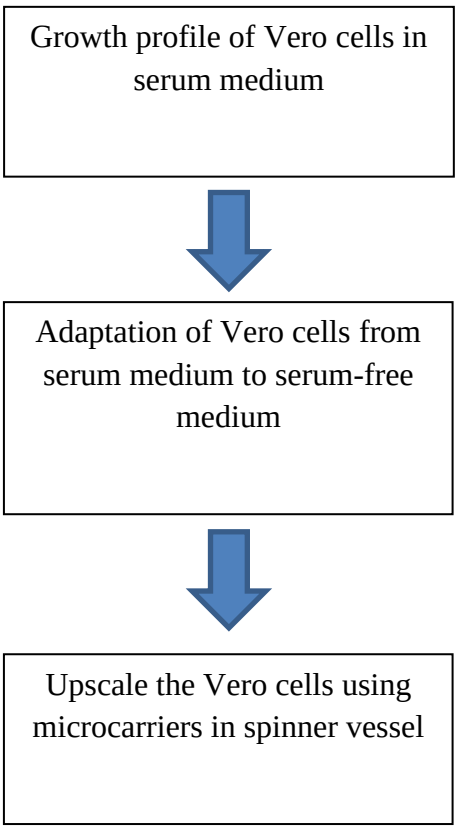

Figure 1.2: Overview of the experiment.