

**ISOLATION AND CULTIVATION OF FRESHWATER
MICROALGAE FOR BIOPHARMACEUTICAL
APPLICATIONS**

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ABSTRACT

Microalgae are currently being study widely. This research is conducted to isolate microalgae from freshwater and explore their bioactive compounds that have potential for biopharmaceutical application. Twenty-two indigenous microalgae samples from Malaysia freshwater microalgae originated from ponds of Silo Agropark, Temoh, Perak (4°14'0"N, 101°10'0"E) and Universiti Teknologi Mara, UiTM Puncak Alam (3°12'12"N, 101°27'10"E) were collected. The samples have been subjected to four different regimes to isolate the microalgae from the fresh water samples. The regimes were conducted to study the culturing in broth and centrifugation steps prior to culture onto TAP agar. Regimes 3 and 4 yield the highest percentage microalgae recovery of 40% from KU samples and 71% from PA pond. To isolate more microalgae, all the regimes must be conducted even though Regime 1 and 2 were not applicable to KU pond, it had yield about 29% to 57% microalgae recovery from PA pond. Under prolonged and repeated subculturing *in vitro* on Tris Acetate Phosphate (TAP) media, severe contamination remained. Therefore, wide spectrum of antibiotic such as ampicillin was selected to resolve this problem. The microalgae have been exposed with 0.5 g/L of ampicillin in TAP broth at different time of exposure (12, 24, 36 and 48 hours) and have been cultured onto TAP agar containing different concentration of ampicillin (0, 0.5, 0.75 and 1.0 g/L). Microscopic observation shows the diversity of morphology of the microalgae with most of the microalgae have sphere shape and other like oblong shape with diameter in range of 50 to 280 µm. Extraction of microalgae was done to extract the bioactive compound and was analyzed using high performance liquid chromatography (HPLC). However, no UV active compounds have been detected using HPLC method and further study need to be conducted to explore the bioactive compound in the microalgae.

CHAPTER 1.0: INTRODUCTION

1.1 Background

Algae belong to kingdom Protista and are divided into microalgae and macroalgae. Microalgae is a type of unicellular that is generally between 2 - 200 μm in size, and are typically found in freshwater and marine environments. On the other hand, macroalgae is multicellular form of algae which are generally more than 200 μm in size (Graham et al., 2012). Diatoms species is the most diverse group of microalgae. Diatoms are eukaryotic microalgae that are ubiquitously found in rivers, ponds, lakes, wetlands and oceans. They can also be found in household aquariums, usually forming the brownish scum on the walls of the aquarium.

One of the most unique characteristics of diatoms is that their cell walls are made of silica, composed of hydrated silicone dioxide, called frustules. The silica is the same materials that are being used in glass production (Mimouni et al., 2012). The shell or frustules is like a box, form of two overlapping glassy portion as in Petri dish. The side view of the cell is the girdle view and the top and bottom cell is the valve view (Figure 1.1) (Spolaore et al., 2006).