CHAPTER 1

INTRODUCTION

1.1 Background of Study

This research is basically the continuous research on the patchouli growth. Biofertilizer is an alternative product containing variety kinds of microorganisms that are able to convert important nutrient from unavailable form through biological process and is applicable in replacing the used of chemical fertilizer to increase soil fertility, organic matter content and plant production as well (Hegde *et al.*, 1999; Vessey, 2003). Moreover, the uses of biofertilizer are totally out of harm's way as it is a friendly fertilizer to the environment. Based on the previous researches, it shows that the patchouli plant is lacking of nutrients by only using biofertilizer to enhance its growth

Compared to the used of chemical fertilizer, the patchouli grows vigorously. In order to maximize the used of biofertilizer on patchouli growth, N-fixer is added as a nutrient supplier. N-fixer is not only significant for legumes, but also non-legumes (Doebereiner and Pedrosa, 1987). It also has beneficial effect that may derive both from its nitrogen fixation and stimulating effect on root development (Doebereiner and Pedrosa, 1987; Lynch, 1990). The using of these beneficial microorganisms helps in enhancing plant resistant to the unpleasant environment circumstance.

For that reason, a series of experiment will be conducted to obtain the most optimum condition for N-fixer bacteria to grow. The parameters that will be study to obtain the optimum condition of N-fixer are temperature, pH and carbon sources (glucose concentrations). The equipment that will be used in this research is Ultra Violet spectrophotometer (UV-vis) and Design of Experiments (DOE) software. Design of Experiment (DOE) software will be used to design the experimental work to optimize the growth parameter of N-fixer within stated range

1.2 Objective

The aim of this study/research is to determine the optimum growth condition for N-fixer bacteria. Therefore, the objectives of this research are:

- i. To obtain the optimum temperature for the growth of N-fixer bacteria at stated range
- ii. To obtain the optimum pH condition to grow N-fixer bacteria at stated range
- iii. To obtain the requirement of carbon sources needed by N-fixer bacteria to achieve optimum growth at stated range

1.3 Scope of Study

In order to achieve the stated objectives, several scope of study has been identified. A series of experiments will be conducted and the parameters that will be study are temperature, pH and initial carbon concentration. Range of all parameters will be determined based on literature study to obtain combination of growth parameter for N-fixer within stated range.

This research can be divided into two parts which are the growth profile of Nfixer and optimization of the experiments. In the first part, the growth profile of Nfixer which consist the cell concentration and glucose consumption will be studied during its 72 hr fermentation. The purpose of constructing these three curves is to proceed to the second part of the research which is optimizing the experiments.

In order to determine the requirement of carbon sources needed by N-fixer bacteria, DNS method will be used. On the other hand, colony forming unit (CFU) method will be use to determine the cell concentration of the N-fixer bacteria as it is the most accurate technique compared to other technique. Design of the experimental work will be design using DOE software. The purpose of using DOE is to find out the best combination of parameter to maximize the growth of the bacteria.

1.4 Problems Statement

In perfumery industry, patchouli oil is essential ingredients which are use as a base material in producing perfume. The increases of value and demand in the perfumery industry as there are no synthetic substitute for patchouli oil. Based on the world demand, the consumption of patchouli oil is about 2000 tonnes per annum. Beyond it's used as perfume, patchouli oil have been used for centuries in traditional medicine which indicating the skin condition. Other than that, it is also used as rubbing oil and fumigant to prevent fever from spread and to build up strong immune system.

Even though the demand on patchouli oil is very high, the oil extract from one tree is very little. Sometimes, no oil is produced after the fresh leaves being extract. The fresh leaves of patchouli plant producing the best quality of essential oil comparing to other part. In order to keep the patchouli always available in the market, the chemical fertilizer is widely used to enhance its growth. Eventhough, the patchouli plants grow vigorously as the chemical fertilizer is continuing being used, the amount of patchouli oil produce is very little (Ganesha Rao and Munnu Singh, 2008). In addition, the usage of chemical fertilizer gives a side effect to the environment by reducing the soil fertility. Other than that, it also destroyed certain