### **CHAPTER I**

### INTRODUCTION

# 1.1 Background

Approximately 70% of the captured fisheries are utilized for food processing and a significant amount of the processed fisheries resulted as wastes (Kim and Mendis, 2006). The waste discarded annualy exceed 20 million tons, which is equivalent to 25% of the total production (FAOSTAT, 2001). In crustaceans such as crab and shrimp the waste makes up approximately 45% of the crustaceans. These chitinuous crustacean wastes are considered to be hazardous to the environment due to the high perishability and high polluting effect if discarded offshore. In the sea the crustacean wastes rapidly lead to high biochemical oxygen demand (BOD), while on land the wastes quickly become colonized by pathogens and spoilage organisms resulted in environmental issue and public health concerns (Kim, 2011).

Chitin or poly ( $\beta$ -(1-4)-N-acetyl-D-glucosamine) is an important natural polysaccharide that was first identified in 1884. Generally, the main sources of chitin are crab and shrimp shells (Younes and Rinaudo, 2015). The shell of the shrimp contained 15-20% of chitin, made it a good source of chitin. According to Rinaudo (2006), chitin and its derivatives have antibacterial properties, biodegradability, nontoxicity, physiological inertness, gel-forming property and affinity for proteins. In food industry, it is usually used to immobilize enzyme and

cells, industrial pollutant treatments and films and fibers forming. However, the main development of chitin film is used for medical and pharmaceutical application, such as wound dressing material and controlled drug release. Nacetylglucosamine is a derivative from chitin that is commercially used, it is produced by hydrolyzing glycosydic bond in chitin to set free the glucosamine units. According to Arbia *et al.* (2013), producing N-acetylglucosamine with chemical synthesis can damage the environment, therefore the safe way to produce N-acetylglucosamine without damaging the environment is by using enzyme. However, using enzyme is high cost and alternative method is needed to surpress the cost of using enzyme.

In producing N-acetyl glucosamine using enzyme, fermentation method can be used as during the fermentation chitinolytic microorganisms can produce chitinase (Brzezinska *et al.*, 2014). Unlike chemical extraction that has many drawbacks, microbial fermentation offers high reproducibility in shorter time, simpler manipulation, smaller solvent consumption, lower energy input and environmental-friendly (Younes and Rinaudo, 2015). According to Josephine (2018), *Providencia stuarti* has strong chitinolytic properties. Utilizing these *Providencia stuarti* to produce chitinase shows a good potential. Immobilization of *Providencia stuarti* can be used as an alternative way to suppress the cost of using enzyme to produce N-acetylglucosamine. By immobilizing the *Providencia stuarti*, the bacteria cells can be used repeatedly. It means that there is no additional cost neededed to produce the chitinase. Optimum fermentation cycles of the immobilized

cells have to be tested to know the number of times the cells can be used with the same efficiency.

One of the methods to do the cell immobilization is by entrapment. Entrapment can be done using polymeric gels, however they are limited by the mechanical strength and lack of open spaces to accommodate active cell growth, resulting in the rupture and cell release. The use of natural structural materials such as papaya trunk wood for cell entrapment has added another view of the immobilization methods. The advantages of using natural structural materials are reusability, freedom from toxicity, mechanical strength and open spaces for growing cells. These have suggested to find other types of natural structural materials from other plant sources that may be used for cell entrapment (Iqbal and Saeed, 2005).

Trunk wood of plants, such as papaya can be used for the cell entrapment. According to Iqbal and Saeed (2005), papaya trunk wood is a structural fibrous network (SFN). SFN of papaya trunk wood as an immobilization matrix shows promising potential as to be the material for the cell entrapment. Additionally, the SFN of papaya trunk wood is very cheap and abundantly available. Currently, the papaya trunk wood has no commercial value. Thus, it is expected that SFN of papaya trunk wood can perform well as a matrix to entrap *Providencia stuartii* cells and can be used in N-acetylglucosamine production through submerged fermentation.

#### 1.2 Research Problem

Chitin or poly  $(\beta-(1-4)-N-acetyl-D-glucosamine is a useful compound for$ industry found in the crustacean wastes, such as shrimp shells. The crustacean wastes are considered to be hazardous to the environment and people. One of the ways to minimize the environment and public health issues is by extracting chitin from the crustacean waste. Chitin can be utilized to produce N-acetylglucosamine which is commercially used for industry. The safe way to produce Nacetylglucosamine without damaging the environment is by using chitinase. Using chitinolytic microorganisms, such as Providencia stuarti that is immobilized to produce chitinase suggested to have the potential to surpress the high cost of using enzyme. One of the ways to immobilize cell is by using entrapment. SFN of papaya trunk wood is used as it is very cheap and abundantly available. SFN of papaya trunk wood also already proven by Iqbal and Saeed (2005) as a good immobilization matrix. Providencia stuarti immobilization using SFN of papaya trunk wood research has not been researched, thereby the optimum size, the optimum ratio of SFN of papaya trunk wood and growth medium and the optimum fermentation cycles should be thoroughly researched.

## 1.3 Objectives

## 1.3.1 General Objective

The general objective of this research was to apply the immobilization technique on *Providencia stuarti* cells using SFN (Structural Fibrous Network) of

papaya trunk wood entrapment method and apply it in N-acetylglucosamine production from *Penaeus vannamei* shells powder submerged fermentation.

## 1.3.2 Specific Objectives

The specific objectives of this research were:

- 1. To determine the optimum size of SFN of papaya trunk wood as a matrix for *Providencia stuarti* immobilization in the production of N-acetylglucosamine through submerged fermentation of *Penaeus vannamei* shells powder.
- 2. To determine the optimum ratio of SFN of papaya trunk wood and growth medium in the production of N-acetylglucosamine through submerged fermentation of *Paneus vannamei* shells powder.
- 3. To determine the optimum fermentation cycles of immobilized *Providencia stuarti* cells used in the production of N-acetylglucosamine through submerged fermentation of *Paneus vannamei* shells powder.