



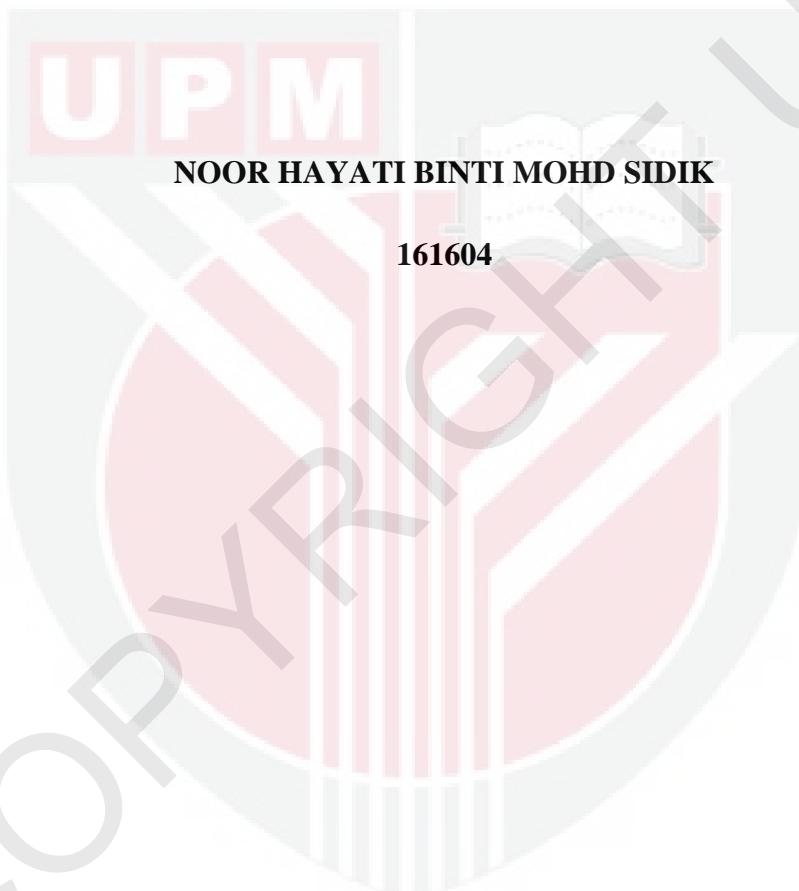
**UNIVERSITI PUTRA MALAYSIA**

**ISOLATION AND ANALYSIS OF *RHLC* and *RHLR* FROM RHAMNOLIPID PRODUCING BACTERIUM, *PSEUDOMONAS AERUGINOSA* strain S5**

**NOOR HAYATI MOHD SIDIK**

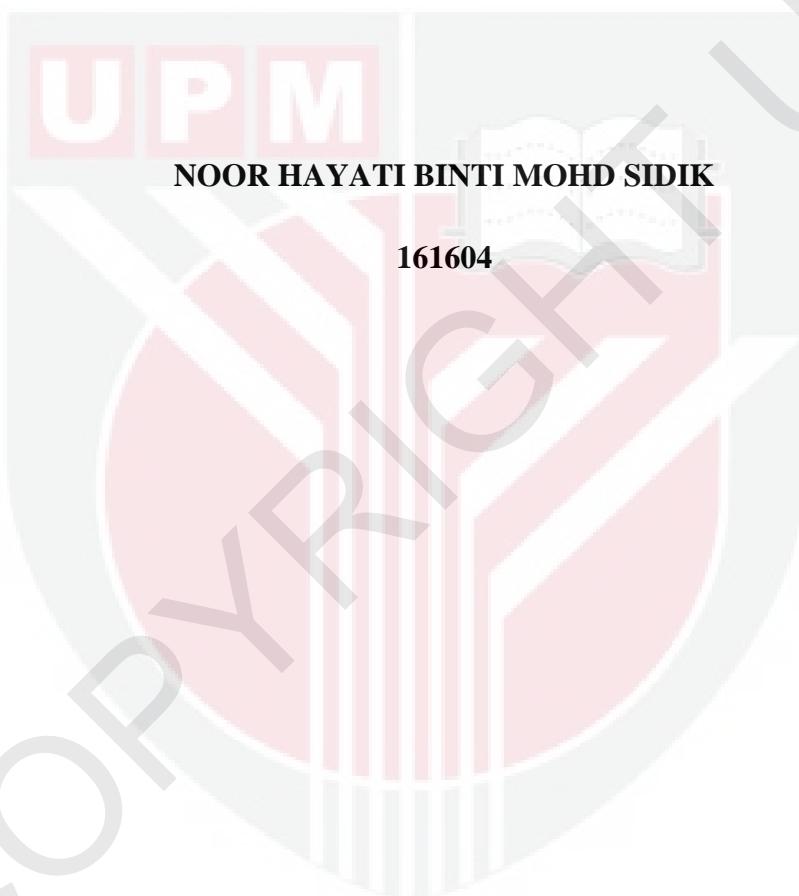
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PRODUCING BACTERIUM, *PSEUDOMONAS AERUGINOSA* STRAIN S5**



**Dissertation submitted in partial fulfillment of the requirement for the course  
BMY 4999 Project in the Department of Microbiology  
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PRODUCING BACTERIUM, *PSEUDOMONAS AERUGINOSA* STRAIN S5**



**DEPARTMENT OF MICROBIOLOGY  
FACULTY BIOTECHNOLOGY AND BIOMOLECULAR SCIENCES  
UNIVERSITI PUTRA MALAYSIA  
2015**

## PENGESAHAN

Dengan ini adalah disahkan bahawa projek yang bertajuk *Isolation and analysis of rhlC and rhlR from rhamnolipid producing bacterium, Pseudomonas aeruginosa strain S5* telah disiapkan serta dikemukakan kepada Jabatan Mikrobiologi oleh Noor Hayati binti Mohd Sidik (161604) sebagai syarat untuk kursus BMY 4999 projek.

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## ABSTRACT

Rhamnolipids are biomolecules with a huge potential in leading global surfactant market, which find applications in various kind of industry in demand for emulsifying and solubilizing agent including food industry, pharmaceutical industry, and petroleum industry. Rhamnolipids are being majorly produced by microorganism, particularly, *Pseudomonas aeruginosa*. This study aims to test rhamnolipid production in *Pseudomonas aeruginosa* strain S5 and to analyze the genes involve for the rhamnolipids biosynthesis which are *rhlC* and *rhlR* that code for rhamnosyltransferase 2 and transcription regulator respectively. Qualitative test on cetyltrimethyl ammonium bromide agar and quantitative tests by emulsification index valued 37.44 %, display capabilities of *Pseudomonas aeruginosa* strain S5 in producing rhamnolipid. In molecular detection of rhamnolipid production, BLAST result shows high similarity between both *rhlC* and *rhlR* sequences, and sequence available in database. The information from the gene analysis could facilitate the understanding on the rhamnolipid biosynthesis in *Pseudomonas aeruginosa* strain S5, hence can be further used in genetic modulation in order to meet future prospects.

## ABSTRAK

Rhamnolipid merupakan biomolekul yang berpotensi tinggi dalam menerajui pasaran surfaktan dunia, seiring dengan kemampuannya untuk diaplikasikan dalam pelbagai industri yang mempunyai permintaan tinggi terhadap bahan emulsifikasi dan bahan pelarut, termasuk industri makanan, industri farmasi, dan juga industri petroleum. Rhamnolipid secara utamanya dihasilkan oleh mikroorganisma, khususnya *Pseudomonas aeruginosa*. Kajian ini bertujuan untuk menguji penghasilan rhamnolipid oleh *Pseudomonas aeruginosa* strain S5 dan untuk menganalisis gen-gen yang terlibat dalam penghasilan rhamnolipid iaitu gen *rhlC* dan *rhlR*, masing-masing mengekod *rhamnosyltransferase 2* dan pengawalatur transkripsi. Ujian kualitatif terhadap agar cetiltrimetilammonium bromida dan ujian kuantitatif oleh indek emulsifikasi yang bernilai 37.44 % menunjukkan keupayaan *Pseudomonas aeruginosa* strain S5 menghasilkan rhamnolipid. Dalam pengesanan molekular, keputusan BLAST merekodkan persamaan yang tinggi di antara kedua-dua gen *rhlC* dan *rhlR* dengan jujukan yang terdapat dalam pengkalan data. Informasi analisis gen yang diperolehi dapat membantu kefahaman tentang penghasilan rhamnolipid oleh *Pseudomonas aeruginosa* strain S5, sekaligus dapat diguna pakai dalam modulasi genetik bagi menepati prospek masa hadapan.

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## CHAPTER 1

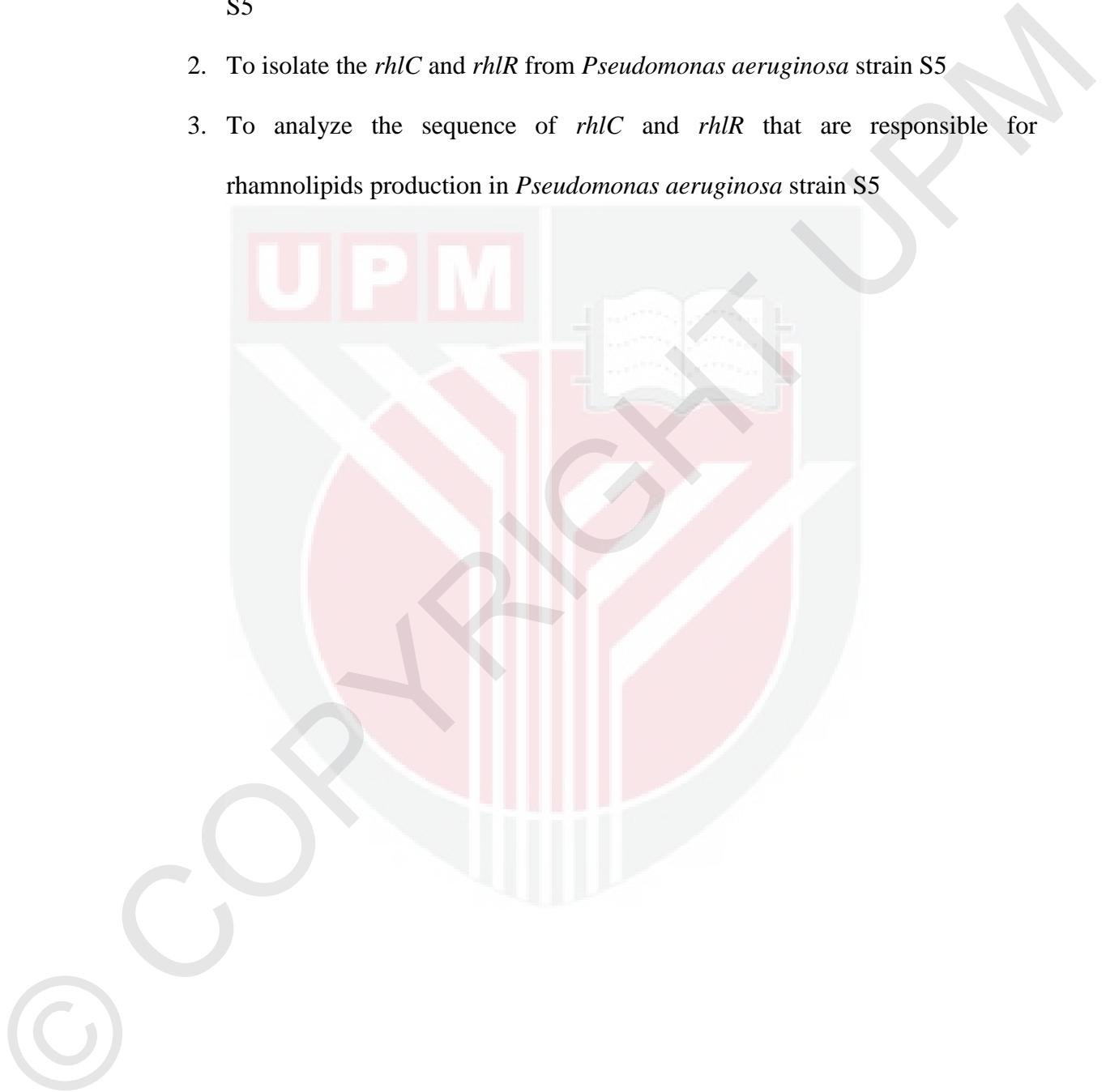
### INTRODUCTION

Rhamnolipid is a biosurfactant that serves an excellence surface and interfacial activities, hence shaping its way to be commercially exploited by many industries in demand for emulsifying, detergency, and surface activity agents. Besides, biosurfactants are well-known to dissolve hydrocarbons in water and vice versa. Throughout the time, intensive studies have been conducted in order to replace the available synthetic surfactant with this biomolecules due to its sustainability sources, less toxicity, and high environmental compatibility when compared to the synthetic surfactant.

It is long known that most *Pseudomonas aeruginosa* is a rhamnolipid producer and vast research have been administered in elucidating the genetic mechanism of the rhamnolipid biosynthesis in order to enhance its production in bigger scale. As for this study, *Pseudomonas aeruginosa* strain S5 was to be identified whether or not this strain is producing rhamnolipid. *Pseudomonas aeruginosa* strain S5 as discussed by Rahman et al. (2006), was originally isolated in the capabilities to degrade benzene, toluene, ethyl-benzene, and xylene (BTEX) and to produce an organic solvent-tolerant lipase. Therefore, it becomes an interesting subject to figure out whether this organism also produces a surface active agent, particularly rhamnolipids.

The objectives of this study are;

1. To determine the rhamnolipids production of *Pseudomonas aeruginosa* strain S5
2. To isolate the *rhlC* and *rhlR* from *Pseudomonas aeruginosa* strain S5
3. To analyze the sequence of *rhlC* and *rhlR* that are responsible for rhamnolipids production in *Pseudomonas aeruginosa* strain S5



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