PRODUCTION OF BIOSURFACTANT BY MARINE *Pseudomonas* spp. AND ITS CHARACTERISATIOM

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IOOL OF MARINE AND ENVIRONMENTAL SCIENCES
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Lihat Sebelah

PRODUCTION OF BIOSURFACTANT BY MARINE *Pseudomonas* spp. AND ITS CHARACTERISATION

By

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Research Report submitted in partial fulfilment of the requirements for the degree of Bachelor of Science (Marine Science)

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SCHOOL OF MARINE AND ENVIRONMENTAL SCIENCES

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DECLARATION AND VERIFICATION REPORT

FINALYEAR RESEARCH PROJECT

It is hereby declared and verified that this research report entitled Production of Biosurfactant by Marine *Pseudomonas* spp. and its Characterisation by Umahsreerekah a/p Gopala Krishnan , Matric No. UK 28201 have been examined and all errors identified have been corrected. This report is submitted to the School of Marine and Environmental Sciences as partial fulfillment towards obtaining the Degree Bachelor of Science (Marine Science), School of Marine and Environmental Sciences, Universiti Malaysia Terengganu.

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PRODUCTION OF BIOSURFACTANT BY MARINE *Pseudomonas* spp. AND ITS CHARACTERISATION

ABSTRACT

The production of rhamnolipid using renewable resources especially the agro-industrial wastes are more preferred compared to chemically synthesise surfactants. As in the findings from this study, four carbon sources namely glycerol, sweetwater, molasses and glycerin pitch were used for rhamnolipid production by *Pseudomonas aeruginosa* PAO1 and *Pseudomonas putida* KT2440. Conditions of biosynthesis involving carbon to nitrogen ratios (excess of carbon sources and nitrogen source limitation) were conducted in order to enhance the productivity of rhamnolipid. The rhamnolipid concentration was determined using colorimetric orcinol assay. Result showed that *P. aeruginosa* worked best at C/N ratio 40 in glycerol and sweetwater (6.87 g/L and 0.67 g/L respectively), C/N ratio of 30 in molasses (0.36 g/L) whereas *P. putida* worked best in C/N ratio 50 in glycerol (0.51 g/L), C/N ratio 30 in sweetwater (0.29 g/L) and 40 in molasses (0.33 g/L). *P. aeruginosa* produce more rhamnolipid concentration (1.47 g/L) using glycerin pitch compared to *P. putida* (0.46 g/L). From these result, it can be said that the rhamnolipid production using renewable carbon sources will be a good approach in proper waste management and bioconversion of by-products into value added materials.

PENGHASILAN BIOSURFAKTAN MENGGUNAKAN MARIN *Pseudomonas* spp DAN PENCIRIANNYA.

ABSTRAK

Pengeluaran rhamnolipid menggunakan sumber-sumber yang boleh diperbaharui terutamanya sisa-sisa daripada industri pertanian amat digalakkan berbanding surfaktan yang disintesis kimia. Dalam kajian ini, empat sumber karbon iaitu gliserol, sweetwater, molases dan gliserin pitch yang telah digunakan untuk pengeluaran rhamnolipid menggunakan *Pseudomonas aeruginosa* PAO1 dan *Pseudomonas putida* KT2440. Keadaan biosynthesis yang melibatkan ratio carbon dan nitrogen (sumber karbon yang berlebihan dan sumber nitrogen yang terhad) telah dijalankan untuk meningkatkan lagi pengeluaran rhamnolipid. Kepekatan rhamnolipid dikaji menggunakan cara 'colorimetric orcinol assay'. Hasil daripada kajian menunjukkan *P. aeruginosa* hasilkan rhamnolipid paling banyak pada C/N ratio 40 dalam gliserol dan sweetwater (6.87 g/L and 0.67 g/L respectively), C/N ratio 30 dalam molases (0.36 g/L) sementara *P. putida* hasilkan rhamnolipid paling banyak pada C/N ratio 50 dalam gliserol (0.51 g/L), C/N ratio 30 dalam sweetwater (0.29 g/L) dan 50 menggunakan molases (0.32 g/L). Selain itu, *P.aeruginosa* hasilkan rhamnolipid paling banyak (1.47 g/L) menggunakan gliserin pitch berbanding dengan *P. putida* (0.46 g/L). Daripada kajian ini, boleh dibuktikan bahawa pengeluaran rhamnolipid menggunakan sumber karbon ini merupakan pendekatan yang bagus kepada pengurusan pembuangan sisa dan biopenukaran sumber karbon kepada sumber yang berpotensi.

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LIST OF ABBREVIATIONS

Abbreviation	
C/N ratio	Carbon to Nitrogen ratio
NR	Nutrient Rich
MSM	Mineral Salt Medium
°C	Degree Celcius
rpm	Revolution per minute
µL	Microliter
g	Gram
mL	Millilitre
w/v	Weight per volume
%	Percentage
w/w	Weight per weight
mN/m	Millinewton per meter
g/L	Gram per liter
g/mol	Gram per mole
E_{24}	Emulsification Index
HCL	Hydrochloric acid
H_2SO_4	Sulphuric acid
CDW	Cell dry weight

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$$C/N = \frac{\dfrac{\text{No. of Carbon} \times \text{molecular weight of Carbon} \times z\ g/L}{\text{Total molecular weight of Carbon source}}}{\dfrac{\text{No. of Nitrogen} \times \text{molecular weight of Nitrogen} \times 0.5\ g/L}{\text{Total molecular weight of Nitrogen source}}}$$