

ISOLATION OF POLYHYDROXYALKANOATE PRODUCING
BACTERIA FROM BRACKISH WATER SURROUNDING
BAKAU TINGGI, KEMAMAN

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FACULTY OF MARITIME STUDIES AND MARINE SCIENCE
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Isolation of polyhydroxyalkanoate producing bacteria from brackish water surrounding Bakau Tinggi, Kemaman / Iezza Idaffi Tahir.

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**ISOLATION OF POLYHYDROXYALKANOATE PRODUCING BACTERIA
FROM BRACKISH WATER SURROUNDING BAKAU TINGGI, KEMAMAN**

By

Iezza Idaffi binti Tahir

**Research Report Submitted in partial fulfillment of
the requirement for the degree of
Bachelor of Science (Marine Biology)**

**Department of Marine Science
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**DEPARTMENT OF MARINE SCIENCE
FACULTY OF MARITIME STUDIES AND MARINE SCIENCE
UNIVERSITI MALAYSIA TERENGGANU**

**DECLARATION AND VERIFICATION REPORT
FINAL YEAR RESEARCH PROJECT**

It is hereby declared and verified that this research report entitled:
Isolation of polyhydroxyalkanoate producing bacteria from brackish water surrounding
Bakau Tinggi, Kemaman by Iezza Idaffi bt Tahir, Matric No. UK22725 have been
examined and all errors identified have been corrected. This report is submitted to the
Department of Marine Science as partial fulfillment towards obtaining the Degree of
Bachelor of Science (Marine Biology), Faculty of Maritime Studies and Marine Science,
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SYMBOLS AND ABBREVIATIONS

SYMBOLS AND ABBREVIATIONS	FULL NAME
%	Percentage
°C	Degree celcius
mL	Mililiter
L	Liter
mg	Miligram
min	Minute
M	Molar
KOH	Potassium hydroxide
HCl	Hydrochloric acid
NR	Nutrient Rich
MSM	Mineral Salt Medium
CaCl ₂	Calcium Chloride
FeCl ₃	Iron (III) Chloride
CuSO ₄ .5H ₂ O	Copper sulphate pentahydrate
CrCl ₃ .6H ₂ O	Chromium chloride hexahydrate
KH ₂ PO ₄	Potassium dihydrogen phosphate
NH ₄ Cl	Ammonium chloride
MgSO ₄ .7H ₂ O	Hydrated magnesium sulphate
Na ₂ HPO ₄	Disodium hydrogen phosphate

CoCl ₂ ·6H ₂ O	Cobalt (II) chloride hexahydrate
NiCl ₂ ·6H ₂ O	Nickel chloride hexahydrate
PHA	Polyhydroxyalkanoate
SCL-PHA	Short-chain-length polyhydroxyalkanoate
MCL-PHA	Medium-chain-length polyhydroxyalkanoate
P(3HB)	Poly (3-hydroxybutyrate)
C12	3-hydroxydodecanoate
C14	3-hydroxytetradecanoate
PHAc	PHA synthase
GC	Gas Chromatography
CME	Caprylic methyl ester

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ABSTRACT

This study was conducted to evaluate the potential production of polyhydroxyalkanoate (PHA) by bacteria isolated from water sample of mangrove environment in Bakau Tinggi, Kemaman, Terengganu. A total of 134 bacteria colonies isolated and screened using Nile red staining in order to observe the potential PHA producer. A total of 15 colonies when glucose used as a carbon source and 6 colonies when fructose used as a carbon source was observed to emit pink fluorescence when viewed under UV light. This indicates the probable present of PHA granules in those bacteria. There are possibilities where the Nile red staining was only indicates the present of fatty acid in bacteria cell. Shaken-flasks culture with nutrient limitation and addition of excess carbon sources (glucose, fructose, cooking oil, oleic acid) resulting on the growth of bacteria strain in medium supplemented with glucose and fructose. However, no growth observed in medium supplemented with cooking oil and oleic acid. This phenomenon probably occurred due to the different ability of the bacteria to synthesize and utilize the carbon sources supplement for growth or PHA production. The result of gas chromatography indicates the production of MCL-PHA by strain R6(-6)50.1 using glucose as a carbon source. MCL-PHA produced consist a monomer of 3-hydroxydodecanoate (C12) and 3-hydroxytetradecanoate (C14) with a total PHA content of 2.8 wt%. The strain was tested as gram-negative by using Gram staining method. The complete identification of bacteria species by using Polymerase Chain Reaction (PCR) is proposed on the future study.

Pengasingan bakteria penghasil polihidroksialkanoat (PHA) dari air payau sekitar

Bakau Tinggi. Kemaman

ABSTRAK

Kajian ini dijalankan untuk menilai penghasilan polihidroksialkanoat (PHA) oleh bakteria daripada sampel air di kawasan bakau bertempat di Bakau Tinggi, Kemaman, Terengganu. Sejumlah 134 koloni bakteria daripada sampel air diuji menggunakan kaedah palitan Nile red untuk mengesan kehadiran awal PHA di dalam sel bakteria. Hasil kajian menunjukkan bahawa sebanyak 15 koloni dan 6 koloni yang masing-masing dibekalkan glukosa dan fruktosa sebagai sumber karbon memancarkan cahaya pendarfluor merah jambu apabila dilihat di bawah cahaya UV. Pengkulturan bakteria menggunakan goncangan kelalang menggunakan glukosa, fruktosa, minyak masak dan asid oleik menunjukkan pertumbuhan koloni bakteria di dalam media yang menggunakan glukosa dan fruktosa sebagai sumber karbon. Walaubagaimanapun, tiada pertumbuhan dalam media yang menggunakan minyak masak dan asid oleik sebagai sumber karbon. Fenomena ini mungkin berpunca daripada kelainan kebolehan setiap bakteria untuk menggunakan sumber karbon yang dibekalkan sebagai sumber tenaga untuk pertumbuhan ataupun untuk penghasilan PHA. Keputusan analisis gas kromatogram menunjukkan penghasilan MCL-PHA oleh strain R6(-6)50.1 apabila glukosa digunakan sebagai sumber karbon. MCL-PHA yang dihasilkan terdiri daripada monomer 3-hydroxydodecanoate (C12) dan 3-hydroxytetradecanoate (C14) dengan kandungan PHA sebanyak 2.8 wt%. Strain R6(-6)50.1 dikenalpasti sebagai gram-negatif menggunakan kaedah pewamaan Gram. Pengenalpastian spesis bakteria menggunakan kaedah Polymerase Chain Reaction (PCR) adalah diusulkan dalam kajian pada masa depan.