

**BIOSYNTHESIS OF POLY(3-HYDROXYBUTYRATE)  
BY *CUPRIAVIADUS NECATOR* DSM 428 UTILIZING  
WASTE DATE FRUITS AS A CARBON SOURCE**

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Sekian, terima kasih.

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by

**SITI NUR AQILAH BINTI ABDUL WAHAB**

A dissertation submitted in the partial fulfilment of the requirements for the degree of  
Bachelor of Technology (B.Tech) in the field of  
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School of Industrial Technology  
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June 2020

## DECLARATION BY AUTHOR

This dissertation is composed of my original work, and contains no material previously published or written by another person except where due reference has been made in the text. The content of my dissertation is the result of work I have carried out since the commencement of my research project and does not include a substantial part of work that has been submitted to qualify for the award of any other degree or diploma in any university or other tertiary institution.

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## TABLE OF CONTENT

	Page
<b>Declaration by author</b>	ii
<b>Acknowledgements</b>	iii
<b>Table of Contents</b>	iv
<b>List of Tables</b>	vi
<b>List of Figures</b>	vii
<b>List of Symbols</b>	viii
<b>List of Abbreviations</b>	ix
<b>Abstrak</b>	xii
<b>Abstract</b>	xiii
<b>CHAPTER 1 INTRODUCTION</b>	
1.1 Research Background	1
1.2 Problem Statement	3
1.3 Objectives	4
<b>CHAPTER 2 LITERATURE REVIEW</b>	
2.1 Petrochemical-Based Plastic Affects The Health Of Earth.	5
2.2 Biodegradable Plastics	6
2.3 Bacteria For PHB Production	10
2.4 Date Fruits As Low-Cost Carbon Feedstock	12
<b>CHAPTER 3 MATERIALS AND METHODS</b>	
3.1 Flowchart Of Conducted Experiments.	16
3.2 Preparation Of Waste Date Fruits Solution As Fermentation Medium	17
3.3 Determination Of Sugar Content And Concentration In Waste Date Fruits Using HPLC	18

3.4 Determination Of Total Nitrogen Using Kjeldahl Method	18
3.5 Inoculum And Culture Medium Preparation	19
3.6 Bacterial Cultivation	20
3.7 Cell Growth Measurement	21
3.8 Determination Of PHB Content	22
<b>CHAPTER 4 RESULTS AND DISCUSSION</b>	
4.1 Date Fruits Characterisation	
4.1.1 Determination Of Sugar Content And Composition Using HPLC	24
4.1.2 Determination Of Nitrogen Content Using Kjeldahl Method	27
4.2 Fermentation Process Of <i>C. necator</i> Using Waste Of Dates Fruit	
4.2.1 Growth Profile	30
4.3 Production Of Polyhydroxybutyrate (PHB)	
4.3.1 Cell Dry Weight And PHB Content	33
4.4 Correlation Of Sugar And Nitrogen Content On The PHB Accumulation	36
4.5 Statistical Analysis	37
<b>CHAPTER 5 CONCLUSIONS AND RECOMMENDATION</b>	
5.1 Conclusion And Recommendation	38
<b>REFERENCES</b>	39
<b>APPENDICES</b>	43

## LIST OF TABLES

<b>Caption</b>		<b>Page</b>
Table 2.1	Physical properties of PHB	9
Table 2.2	Comparison of various types of bacteria, carbon sources, PHA type and content (%)	10
Table 2.3	Comparison of PHB content by utilization of waste date fruits as carbon feedstock	14
Table 4.1	The analysis of the sugar content and composition before and after fermentation	25
Table 4.2	The percentage of nitrogen content determined using Kjeldahl method in various dates to water ratios	29
Table 4.3	Average amount of cell dry weight (CDW) and PHB content.	35



## LIST OF FIGURES

<b>Caption</b>		<b>Page</b>
Figure 2.1	Chemical structure of Poly (3-hydroxybutyrate)	9
Figure 3.1	Flowchart of methodology conduct in this study	16
Figure 4.1	Bar graph represents the amount of sugars obtained in dates solution before and after fermentation	27
Figure 4.2	Average of nitrogen obtained in different ratios of dates solution before and after fermentation	29
Figure 4.3	Growth curve of <i>Cupriavidus necator</i> throughout 26 hours fermentation, with 200 rpm agitation at 30°C with various dates to water ratio: (a) 1:3, (b) 1:5, and (c) 1:8.	31

## LIST OF SYMBOLS

Symbol	Caption
$\pm$	Plus-minus
%	Percentage
$^{\circ}\text{C}$	Degree Celcius
$^{\circ}\text{C}/\text{min}$	Degree Celcius per minute
X	Multiply

## LIST OF ABBREVIATIONS

Symbol	Caption
PHB	Polyhydroxybutyrate
P3HB	Poly(3-hydroxybutyrate)
g/L	Gram per litre
PHA	Polyhydroxyalkanoate
K	Thousand
PE	Polyethelene
PP	Polypropelene
Kg	Kilogram
PLA	Poly lactid acid
kg/m <sup>3</sup>	Kilogram per Cubic Meter
mPa	Mega pascal
UV	Ultraviolet
P(3HB-co-3HV)	Poly(3-hydroxybutyrate-co-3-hydroxyvalerate)
Scl	Short chain length
CDW	Cell dry weight
3HV	3-hydroxyvalerate
Mmt	Million metric tonnes
ml	Millilitre
Rpm	Revolutions per minute
HPLC	High Performance Liquid Chromatography
GC	Gas Chromatography
g	Gram
pH	Potention of hydrogen

Psi	Pound per square inch
$\mu\text{l}$	Microlit
$\mu\text{m}$	Micrometer
Mm	Millimeter
Min	Minutes
ml/min	Millilitre per minute
NaOH	Sodium Hydroxide
HCl	Hydrochloric Acid
Mg	Milligram
NB	Nutrient Broth
L	Litre
M	Molar
MSM	Mineral salt medium
$(\text{NH}_4)_2\text{SO}_4$	Ammonium sulfate
$\text{KH}^2\text{PO}_4$	Potassium dihydrogen phosphate
$\text{Na}_2\text{HPO}_4$	Sodium phosphate dibasic
$\text{MgSO}_4 \cdot \text{XH}_2\text{O}$	Magnesium sulphate hydrate
$\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$	Ferrous sulfate heptahydrate
$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	Zinc sulphate heptahydrate
$\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$	Copper sulphate pentahydrate
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	Manganese sulphate monohydrate
$\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$	Calcium chloride dihydrate
$\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$	Sodium tetraborate decahydrate
$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$	Ammonium molybdate tetrahydrate
OD	Optical Density

v/v	Volume to volume
kPa	Kilopascal
TCA	Tricarboxylic acid
Nm	Nanometre
H	Hour
ANOVA	Analysis of variance

# **BIOSINTESIS POLI (3-HIDROKSIBUTIRAT) OLEH *CUPRIAVIDUS NECATOR* DSM 428 MENGGUNAKAN BUAH KURMA TERBUANG SEBAGAI SUMBER KARBON.**

## **ABSTRAK**

Banyak penyelidikan telah dilakukan untuk mencari pengganti plastik yang dihasilkan dari petroleum konvensional. Polihidroksibutirat (PHB) adalah polimer biodegradasi yang dihasilkan dari sumber yang boleh diperbaharui telah mendapat perhatian untuk menggantikan plastik yang dihasilkan dari petroleum konvensional kerana ia memiliki sifat kimia dan fizikal yang sangat baik, dengan kelebihan kebolehbidegradasi yang lengkap. Batasan dalam menghasilkan PHB dalam skala besar adalah kerana kos sumber karbonnya yang mahal. Kajian ini memberi tumpuan kepada penghasilan PHB oleh *Cupriavidus necator* menggunakan sumber karbon yang berkos rendah seperti hasil buangan buah kurma. Tiga nisbah kepekatan larutan buah kurma terhadap air disediakan iaitu 1:3, 1:5, 1:8 dengan keadaan penapaian 30 °C, 200 rpm dan penapaian sehingga 29 jam. Analisis kandungan gula dalam larutan buah kurma dilakukan sebelum dan selepas proses penapaian. Kandungan glukosa menunjukkan jumlah tertinggi berbanding fruktosa dan sukrosa iaitu  $48.5 \pm 6.3\%$  (v/v),  $55.7 \pm 3.8\%$  (v/v) dan  $60.6 \pm 5.1\%$  (v/v). Purata berat sel kering terhadap nisbah 1:3, 1:5 dan 1:8 adalah  $0.179 \pm 0.03\text{g/L}$ ,  $0.166 \pm 0.01\text{g/L}$  dan  $0.180 \pm 0.01\text{g/L}$ . Manakala kandungan PHB adalah,  $7.9 \pm 3.0\%$ ,  $4.1 \pm 3.4\%$  dan  $10.4 \pm 9.6\%$ . Nisbah 1:8 mencatatkan berat sel kering dan PHB tertinggi dibandingkan dengan semua nisbah walaupun ia adalah larutan buah kurma yang paling cair. Pemerhatian ini adalah berdasarkan penggunaan sumber karbon yang berkesan kerana mengandungi glukosa tertinggi dibandingkan dengan nisbah lain, dan jumlah nitrogen yang terhad ( $1.45 \pm 1.06\%$ ) yang meningkatkan kesan positif terhadap penghasilan PHB.

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**ABSTRACT**

Many researches have been conducted to find a replacement for conventional petroleum-derived plastics. Poly(3-hydroxybutyrate) (PHB) is biodegradable polymers produced from a renewable source have gained many interest to replace conventional plastic, as it possesses excellent chemical and physical properties, with an advantages of complete biodegradability. The limitation in producing huge amount of PHB is due to its expensive cost of carbon sources. This study focus on the production of PHB by *Cupriavidus necator* using low-cost carbon sources such as waste of date fruits. Three concentration ratios of date fruits to water was prepared which are 1:3, 1:5, and 1:8 with the fermentation condition of 30°C, 200 rpm and up to 29 hours of fermentation. Analysis of sugar content in date fruits solution was performed prior and after the fermentation process. Glucose content shows the highest amount compared to fructose and sucrose which are  $48.5 \pm 6.3\%$ ,  $55.7 \pm 3.8\%$  and  $60.6 \pm 5.1\%$ . The average CDW of ratio 1:3, 1:5 and 1:8 are  $0.179 \pm 0.03\text{g/L}$ ,  $0.166 \pm 0.01\text{g/L}$  and  $0.180 \pm 0.01\text{g/L}$ . While the PHB content are,  $7.9 \pm 3.0\%$ ,  $4.1 \pm 3.4\%$  and  $10.4 \pm 9.6\%$  respectively. Ratio 1:8 state the highest CDW and PHB in comparison to all ratios even it is the most diluted date fruits solution. This observation is due to effective utilization of carbon sources as it contained the highest glucose compared to other ratios, and limited amount of nitrogen ( $1.45 \pm 1.06\%$ ) which enhanced the positive effect on PHB production.