



**UNIVERSITI PUTRA MALAYSIA**

***BIOSUGARS PRODUCTION FROM NAPIER GRASS USED IN  
PHYTOREMEDIATION OF PALM OIL MILL EFFLUENT FINAL  
DISCHARGE***

**NURUL ATIQAH OSMAN**

**FBSB 2020 20**



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By

**NURUL ATIQAH OSMAN**

**Thesis Submitted to the School of Graduate Studies, Universiti Putra  
Malaysia, in Fulfilment of the Requirements for the Degree of  
Master of Science**

**July 2020**

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

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**July 2020**

**Supervisor : Ahmad Muhaimin Roslan, PhD**  
**Faculty : Biotechnology and Biomolecular Sciences**

Napier grass is well known as a substrate for biosugars production, due to its considerable cellulose content. It also has suitable features as phytoremediator in treating wastewater, for example, high growth rate and high tolerance to contaminants. A study has proven that phytoremediation of palm oil mill effluent final discharge (POME FD) using Napier grass can be applied in constructed wetland treatment system. However, the potential of utilising Napier grass after phytoremediation of POME FD has not been fully studied. Therefore, it is crucial to study the properties of the Napier grass, which include the physical analysis, lignocellulosic composition, and elements and heavy metals concentration deposited in the plant after phytoremediation. Apart from that, the compounds in POME FD may influence cellulose content in plants, and can inhibit saccharification process. Therefore, it is important to determine the effect of POME FD on the yield of biosugars obtained after saccharification.

In this study, the effect of POME FD on the Napier grass cultivars (Common and Red cultivar) in a constructed wetland system was evaluated. The two months old Napier grass cultivars were used in this study. The treatment systems were supplied with 5 litre of POME FD per day, whereas the control systems were supplied with rainwater at 5 litre every day, for 8 weeks. The height and the number of tiller and leaves, were measured for every weeks. The plants were harvested at week 8. The stems were pressed and the juice obtained were analysed for sugar and elements content determination. The dried stems and leaves were used for the lignocellulosic composition analysis, while only stem sample were used in the alkaline pretreatment and

saccharification for biosugars determination. A 20 FPU/g<sub>substrate</sub> of *Acromonium* cellulase was used in the saccharification.

The results revealed that the percentage of height increments of the plant from week 0 to week 8 in treatment system for Common and Red cultivars were  $61.72 \pm 2.36\%$  and  $109.98 \pm 2.05\%$ , respectively, as compared to those in control, which were  $14.42 \pm 2.11\%$  and  $30.30 \pm 1.75\%$ , respectively. These results indicated the ability of Napier grass to absorb the necessary nutrients from POME FD for plant growth. Besides, Napier grass cultivar grown in treatment system have higher cellulose percentage ( $38.77 \pm 0.29\%$  and  $42.29 \pm 0.95\%$ , for Common and Red cultivar, respectively) compared with control ( $34.16 \pm 1.01\%$  and  $36.14 \pm 0.45\%$ , respectively). Thus, it is more suitable as substrate for biosugars production. In addition, the result revealed that heavy metals concentration absorbed and accumulated in Napier grass stem, leaves and juice were below the standard limit of World Health Organization (WHO). As for glucose conversion yield percentage, there is no significant difference between the treatment and control biomass. This suggested that concentration of elements deposited in stem of treatment system biomass does not inhibit saccharification process. The findings of this study can be used as a reference for further utilisation of Napier grass after being used in the phytoremediation of POME FD.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia  
sebagai memenuhi keperluan untuk ijazah Master Sains

**PENGHASILAN BIOGULA DARIPADA RUMPUT NAPIER YANG  
DIGUNAKAN DI DALAM FITOREMEDIASI PELEPASAN AKHIR EFLUEN  
KILANG SAWIT**

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Rumput Napier terkenal sebagai substrat untuk penghasilan biogula, kerana kandungan selulosa yang cukup banyak. Ia juga mempunyai ciri-ciri yang sesuai sebagai organisma fitoremediasi untuk merawat air sisa, contohnya, kadar pertumbuhan yang tinggi dan toleransi yang tinggi terhadap bahan cemar. Kajian telah membuktikan bahawa fitoremediasi pelepasan akhir efluen kilang sawit (PA-EKS) menggunakan rumput Napier dapat diterapkan di dalam sistem rawatan tanah bench. Walau bagaimanapun, potensi penggunaan rumput Napier setelah proses fitoremediasi PA-EKS masih belum dikaji sepenuhnya. Oleh itu, sangat penting untuk mengkaji sifat-sifat rumput Napier, yang merangkumi analisis fizikal, komposisi lignoselulosa, dan unsur-unsur dan kepekatan logam berat yang disimpan di dalam tumbuhan setelah proses fitoremediasi. Selain itu, sebatian dalam PA-EKS dapat mempengaruhi kandungan selulosa pada tanaman, dan dapat menghalang proses sakarifikasi. Oleh itu, adalah penting untuk menentukan kesan PA-EKS terhadap hasil biogula yang diperoleh selepas sakarifikasi.

Dalam kajian ini, kesan PA-EKS terhadap kultivar rumput Napier (kultivar *Common* dan *Red*) dalam sistem tanah bench telah dikaji. Kultivar rumput Napier yang berusia dua bulan telah digunakan di dalam kajian ini. Sistem rawatan dibekalkan dengan 5 liter PA-EKS setiap hari, manakala sistem kawalan dibekalkan dengan air hujan sebanyak 5 liter setiap hari, selama 8 minggu. Tinggi dan jumlah teler dan daun, diukur pada setiap minggu. Tumbuhan dituai pada minggu 8. Batang ditekan dan jus yang diperolehi dianalisis untuk penentuan kandungan gula dan unsur-unsur. Batang dan dedaun yang telah kering digunakan untuk analisis komposisi lignoselulosa, sementara hanya sampel batang digunakan dalam pra-rawatan alkali dan

sakarifikasi untuk penentuan hasil biogula. Sebanyak 20 FPU/g<sub>substrat</sub> *Acremonium cellulase* digunakan di dalam proses sakarifikasi.

Hasil kajian menunjukkan bahawa peratusan peningkatan ketinggian tanaman dari minggu 0 hingga minggu ke-8 dalam sistem rawatan untuk kultivar *Common* dan *Red* adalah  $61.72 \pm 2.36\%$  dan  $109.98 \pm 2.05\%$ , dibandingkan dengan kawalan, iaitu  $14.42 \pm 2.11\%$  dan  $30.30 \pm 1.75\%$ . Keputusan ini menunjukkan keupayaan rumput Napier untuk menyerap nutrien yang diperlukan dari PA-EKS untuk pembesaran tumbuhan. Selain itu, kultivar rumput Napier yang ditanam dalam sistem rawatan mempunyai peratusan selulosa yang lebih tinggi ( $38.77 \pm 0.29\%$  dan  $42.29 \pm 0.95\%$ , untuk kultivar *Common* dan *Red*) dibandingkan dengan kawalan ( $34.16 \pm 1.01\%$  dan  $36.14 \pm 0.45\%$ ). Oleh itu, ia lebih sesuai sebagai substrat untuk penghasilan biogula. Sebagai tambahan, keputusan menunjukkan bahawa kepekatan logam berat yang diserap dan terkumpul di batang, daun dan jus rumput Napier berada di bawah piawaian Pertubuhan Kesihatan Sedunia (PKS). Bagi peratusan hasil penukaran glukosa, tidak ada perbezaan yang signifikan antara biojisim rawatan dan kawalan. Ini menunjukkan bahawa kepekatan unsur-unsur yang disimpan dalam batang biojisim sistem rawatan tidak menghalang proses sakarifikasi. Penemuan kajian ini dapat dijadikan rujukan untuk penggunaan rumput Napier setelah digunakan dalam fitoremediasi PA-EKS.

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This thesis was submitted to the Senate of the Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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## LIST OF ABBREVIATIONS

ADF	Acid detergent fibre
ADL	Acid detergent lignin
AIL	Acid insoluble lignin
AIR	Acid insoluble residues
Al	Aluminium
APHA	American Public Health Association
As	Arsenic
ASL	Acid soluble lignin
BBL	Biorefinery and Biomass Laboratory
BOD	Biological oxygen demand
Ca	Calcium
Cd	Cadmium
Co	Cobalt
COD	Chemical oxygen demand
CPO	Crude palm oil
Cr	Chromium
C-sink	Carbon sink
Cu	Copper
DMDSe	Dimethyldiselenide
DMSe	Dimethylselenide
DOE	Department of Environmental
EFB	Empty fruit bunches
Fe	Iron
FFB	Fresh fruit bunches
FPU	Filter paper unit
H <sub>2</sub> O <sub>2</sub>	Hydrogen peroxide
H <sub>2</sub> SO <sub>4</sub>	Sulfuric acid
H <sub>3</sub> PO <sub>4</sub>	Phosphoric acid
HCL	Hydrochloric acid
HNO <sub>3</sub>	Nitric acid

HPLC	High-performance liquid chromatography
ICP-MS	Inductive coupled plasma-mass spectrometry (ICP-MS)
ICP-OES	Inductive coupled plasma-optical emission spectrometry
K	Potassium
L	Litre
Mg	Magnesium
Mn	Manganese
n.d	Not detected
NaOH	Sodium hydroxide
NDF	Neutral detergent fibre
Ni	Nickel
OPF	Oil palm fronds
OPT	Oil palm trunks
PAC	Polyaluminium chloride
Pb	Lead
PKC	Palm kernel cake
POME	Palm oil mill effluent
POME FD	Palm oil mill effluent final discharge
POMS	Palm oil mill sludge
ppb	Part per billion
PPF	Palm press fibre
ppm	Part per million
rpm	Rotation per minute
Se	Selenium
SES	Scanning electron microscope
TKN	Total Kjeldahl nitrogen
TN	Total nitrogen
TSS	Total suspended solid
WHO	World Health Organization
Zn	Zinc

# CHAPTER 1

## INTRODUCTION

### 1.1 Research overview

Pollution of soil and water caused by wastewater is one of the major global threats that our environment is facing today. One of the wastewater sources is palm oil mill effluent final discharge (POME FD) that is released to the environment without an effective treatment. Palm oil mill effluent (POME) is one of the by-products generated from the process of palm oil extraction in palm oil production. Since Malaysia is the world's second largest producer and exporter of palm oil, a large amount of POME is generated annually (Kamyab et al., 2018). It is estimated that for every tonne of crude palm oil produced, about 2.5 to 3.5 tonne of POME is generated (Madaki and Seng, 2013a).

POME is considered as the main source of water pollution in Malaysia due to the high biochemical oxygen demand (BOD) and chemical oxygen demand (COD) that cause a reduction of the biodiversity of aquatic ecosystems (Soleimaninanadegani and Manshad, 2014). Furthermore, the damage to the river cannot be undone easily. Since POME is generated in a large volume at a time, it is difficult to be managed, while the treatment of this wastewater is expensive. Consequently, the treated POME is discharged to the nearby river as it is the cheapest and easiest way for disposal. However, Madaki and Seng (2013b) noted that, treated POME (POME FD) still has an effect on the environment; this observation was also supported by the findings of Ibrahim et al. (2017). This is because POME FD still contains a significant amount of organic matter. For that reason, Ujang et al. (2018) has decided to further treat the POME FD using Napier grass constructed wetland and thus, achieved 71.57% of COD and 83.59% of total suspended solid (TSS) reduction.

Napier grass (*Pennisetum purpureum*), also known as “elephant grass”, was first introduced in Malaysia in the 1920's from East Africa and it is traditionally used as fodder grass for livestock (Halim et al., 2013). However, there is another more beneficial use of Napier grass that is less popular, which is as a phytoremediation plant to remediate industrial and agricultural wastewater. Napier grass have been proven to be used as a phytoremediator plant. For instance, Klomjek (2016), in Thailand used two different varieties of Napier grass for the treatment of swine wastewater and both varieties performed more than 70% of removal efficiency of BOD and total Kjeldahl nitrogen (TKN). Besides, from a study conducted by Tayade et al. (2005), 85% of BOD and 83% of TSS removal from municipal wastewater were achieved by planted Napier grass with broadleaf cattail in a horizontal subsurface flow system. This method is made feasible as Napier grass has suitable features in treating wastewater.

Besides removing pollutants in the wastewater, the Napier grass grown on POME FD can also support the growth of the plant, thus leading to the increase in lignocellulosic materials. This can be taken as an advantage since lignocellulosic materials is a promising feedstock for biofuel production, as it can be hydrolysed into biosugars (fermentable sugars) (Kucharska et al., 2018). Since the plant used for the phytoremediation is commonly disposed as waste, this will make biofuel production from this plant more sustainable. Thus, it is adding more value to the use of Napier grass to treat wastewater in a wetland system. This integrated approach can lead to a more eco-friendly, and economical alternative to conventional energy resources (Kumar et al., 2017). This is also one of the zero-waste concept management since all the biomass is being fully utilized.

## 1.2 Problem statement

From the previous study by Ujang et al. (2018), it was proven that the phytoremediation of POME FD using Napier grass can be applied in the constructed wetland treatment system. However, the potential of utilising Napier grass after phytoremediation of POME FD has not yet been fully studied. Mostly, the phytoremediator plant that are contaminated with heavy metal was disposed through ashing, direct disposal, or incineration process (Sas-Nowosielska et al., 2004). Therefore, it is crucial to study the properties of the Napier grass, which include the physical analysis, lignocellulosic composition, and the elements and heavy metals concentration deposited in the plant after phytoremediation. Apart from determining the impact of its disposal to the environment, learning these properties is also important in order to establish a benchmark for further utilisation of Napier grass after being used in the phytoremediation of POME FD.

Apart from phytoremediation, several studies have reported that Napier grass can serve as a source for biosugars production; obtained through pressing or hydrolysis of the fibres (Shakil et al., 2013). However, the compounds in POME FD, for instance, potassium, may influence cellulose content in the plants (Wang et al., 2013). The increase of cellulose content is proportional to the increase of biosugars (glucose) concentration. This is because glucose is the monomer sugar of cellulose. In contrast, iron (Fe) and copper (Cu) can inhibit enzymatic hydrolysis process *via* redox mechanism (Tejirian and Xu, 2010). Therefore, the scope of this study is to used Napier grass in constructed wetland system for the phytoremediation of the POME FD, thus evaluate the feasibility of Napier grass both as a phytoremediator and as a substrate for biosugars production towards zero waste management of POME FD.

### **1.3 Hypothesis**

It is hypothesized that:

1. the Napier grass can absorb nutrients from palm oil mill effluent final discharge, thus can have improved physical and chemical properties compared with control.
2. the elements in the palm oil mill effluent final discharge to Napier grass will enhance the yield of biosugars conversion.

### **1.4 Objectives**

The main objective of this study are:

1. To evaluate the characteristics of Napier grass after the phytoremediation of palm oil mill effluent final discharge in a constructed wetland system.
2. To determine the yield of biosugars that can be obtained from the saccharification of Napier grass stem after being used in the phytoremediation of palm oil mill effluent final discharge.

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## APPENNDICES

### APPENDIX A

#### Appendix A1. Chemical Oxygen Demand (COD)

The COD analysis was performed according to Standard Method by APHA (1999), Method 5220 D. Two millilitres of an aliquot sample was added into the COD Digestion Reagent vial (Low range or High range were used). However, if High range plus COD Digestion Reagent vial was used, only 0.2 mL of the aliquot diluted sample was added. The vials containing sample were heated and refluxed for 2 hours. The vials were allowed to cool. DR 2800 spectrophotometer with a wavelength of 620 nm was used to measure the absorbance of the sample

#### Appendix A2. pH

The pH analysis was performed according to Standard Method by APHA (1999), Method 4500-H<sup>+</sup> B. The pH meter was calibrated first before use by immersing the electrode in buffer with pH approaching the sample. The bulb was cleaned with distilled water and dried with a tissue. Then, the electrode of the pH meter was dipped into the sample and read for the pH of the sample. At the end, the electrode was rinsed and dried with a tissue before it was placed back into the buffer.

#### Appendix A3. Colour

The colour analysis was performed according to Platinum-Cobalt Standard Method (2014), Method 8025. Colour measurements were carried out with a spectrophotometer DR 2800. In this method, the colour was being expressed as “apparent” colour where the sample was analysed without being filtered. The distilled water which acts as blank was poured into sample cell and was measured by using a spectrophotometer at wavelengths 455 nm. Then, the step was followed by measuring the samples. The result of the absorbance was taken as colour contained in the sample.

#### Appendix A4. Total Suspended Solid (TSS)

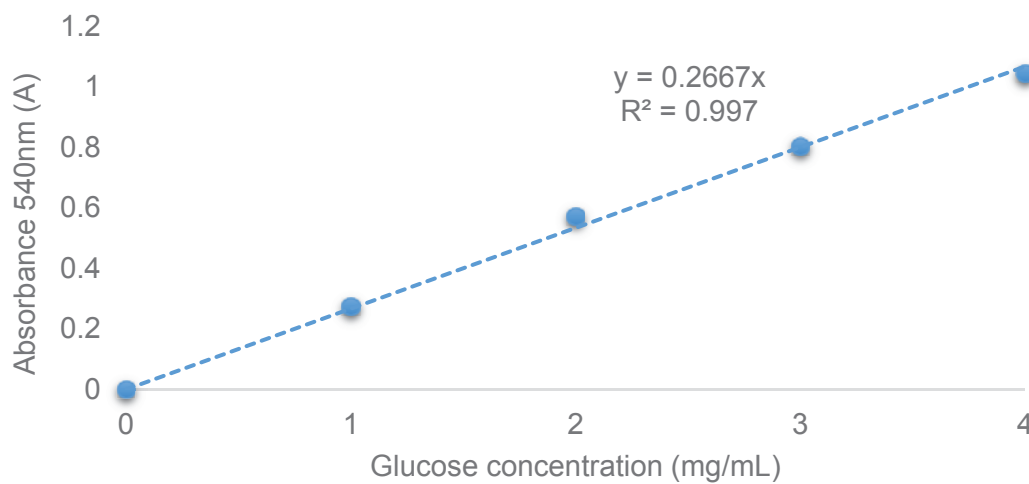
The TSS analysis was performed according to Standard Method by APHA (1999), Method 2540 B. The total suspended solid measurements were carried out by using a spectrophotometer DR 2800. The distilled water which acts as blank was poured into sample cell and was measured at wavelengths of 810 nm. Then, the step was followed by measuring the samples.

## APPENDIX B

### Filter Paper Unit assay for *Acremonium* cellulose

Glucose standard curve using DNS at 540 nm wavelength for FPU assay

Glucose concentration (mg/mL)	Absorbance (A)
0.0	0
1.0	0.274
2.0	0.571
3.0	0.803667
4.0	1.043333



From the graph, value of absorbance for 2.0 mg/ml glucose can be calculated using equation from trendline given

Value of absorbance:

$$y = mx + c$$

Where  $y$  = absorbance (A)

$m$  = gradient obtained from the graph

$x$  = glucose concentration (mg/ml)

$c$  = y-intercept

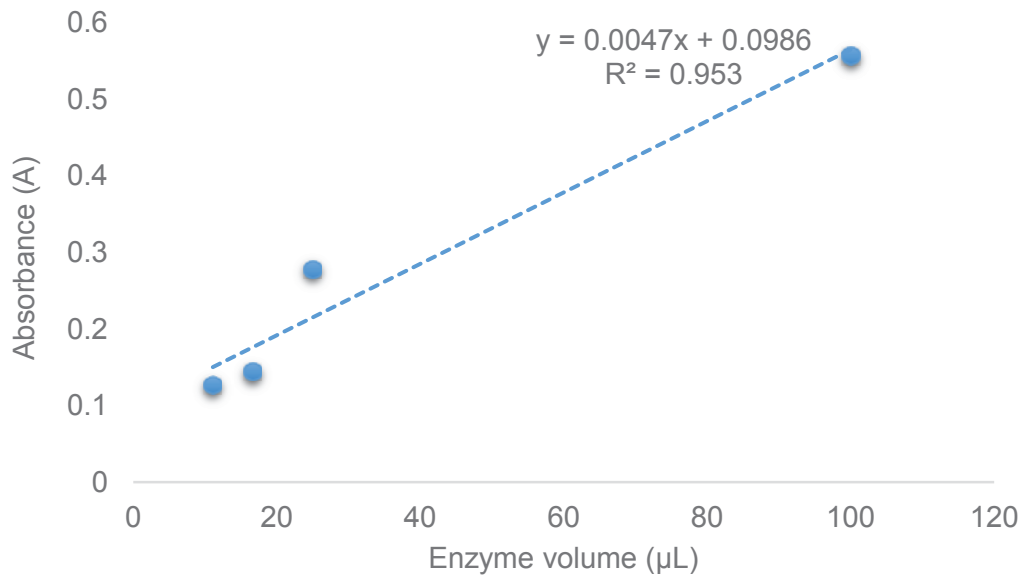
Absorbance value for 2.0 mg/ml glucose

$$= 0.2667 \times 2.0$$

$$= 0.5334$$

### Preparation for enzyme curve

Enzyme dilution	Enzyme volume ( $\mu\text{L}$ )	Absorbance (A)
100	100	0.557
400	25	0.278
600	16.65	0.1445
900	11.1	0.127



From the graph, value of enzyme that producing absorbance similar to 2.0 mg/ml glucose can be calculated using equation from trend line given

Value of absorbance:

$$y = mx + c$$

Where  $y$  = absorbance (A)

$m$  = gradient obtained from the graph

$x$  = glucose concentration (mg/ml)

$c$  = y-intercept

Absorbance value for 2.0 mg/ml glucose from glucose standard curve was 0.5334

Therefore, enzyme volume

$$\begin{aligned}y &= 0.0047x + 0.0986 \\0.5334 &= 0.0047x + 0.0986 \\x &= 92.5106 \mu\text{L}\end{aligned}$$

For FPU calculation, enzyme concentration that would have released exactly 2 mg of glucose, need to be calculated first:

Enzyme concentration =

$$\frac{\text{volume of enzyme in dilution (uL)}}{\text{total volume of enzyme diluted in 50mM sodium citrate buffer (uL)}}$$

$$\begin{aligned}\text{Enzyme concentration} &= \frac{92.5106 \mu\text{L}}{10000 \mu\text{L}} \\&= 0.00925\end{aligned}$$

The FPU is based on the released of exactly 2.0 mg of glucose equivalent, that is 2/0.18  $\mu\text{mol}$  from 0.5 mL of diluted enzyme in 60 minutes

$$2 \text{ mg glucose} = \frac{2}{0.18 \mu\text{mol} \times 0.5 \text{ mL} \times 60 \text{ min}}$$

$$2 \text{ mg glucose} = 0.37 \mu\text{mol.mL.min}^{-1}$$

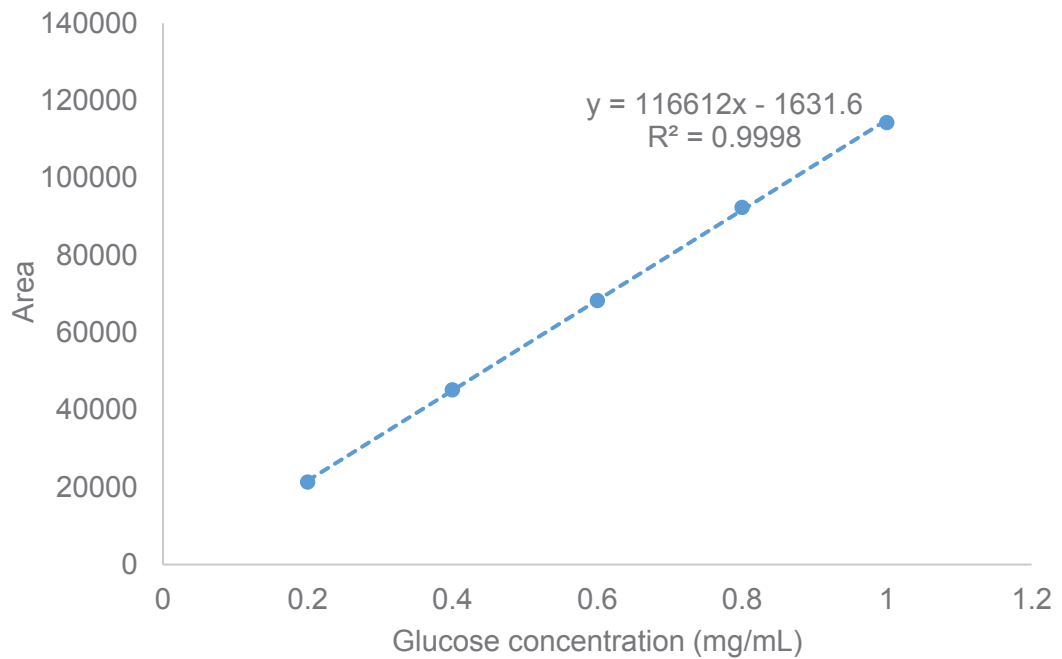
Since initially, enzyme was diluted on 1 g of enzyme into 10 ml sodium citrate which allow the enzyme to transform into solution form, therefore, FPU need to be multiply with 10 mL

$$\begin{aligned}\text{FPU} &= (0.37 / 0.00925) \times 10 \text{ mL} \\&= 400\end{aligned}$$

## APPENDIX C

### Appendix C1. Standard curve for glucose using Aminex HPX-87P column (Biorad, USA)

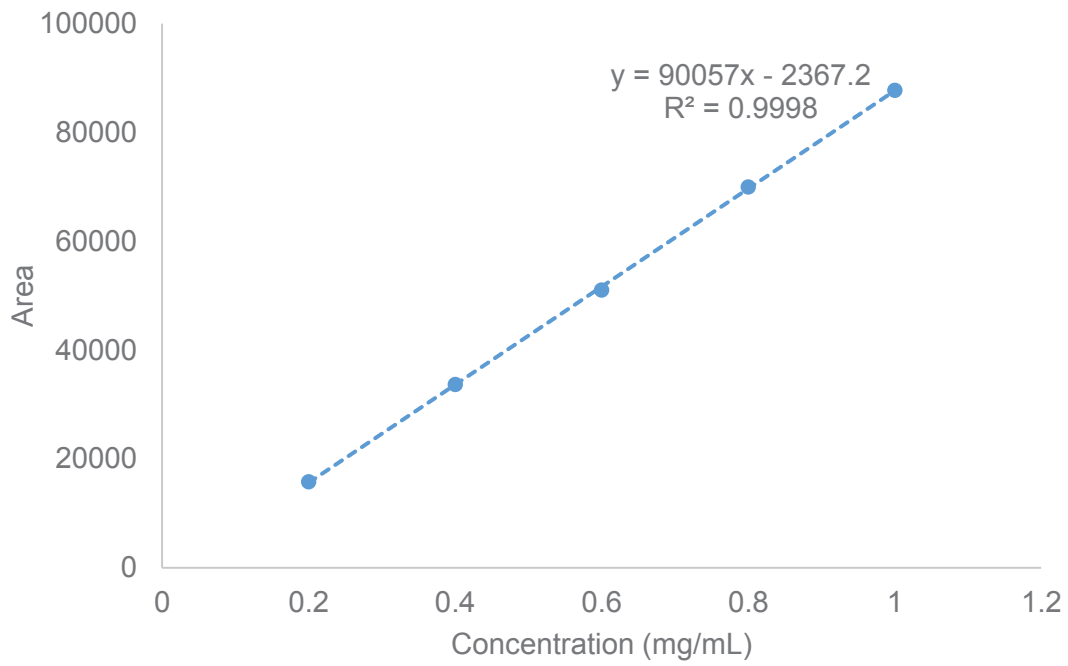
Glucose concentration (mg/mL)	Area
0.2	21364.0
0.4	45217.5
0.6	68292.5
0.8	92439.0
1.0	114365.5





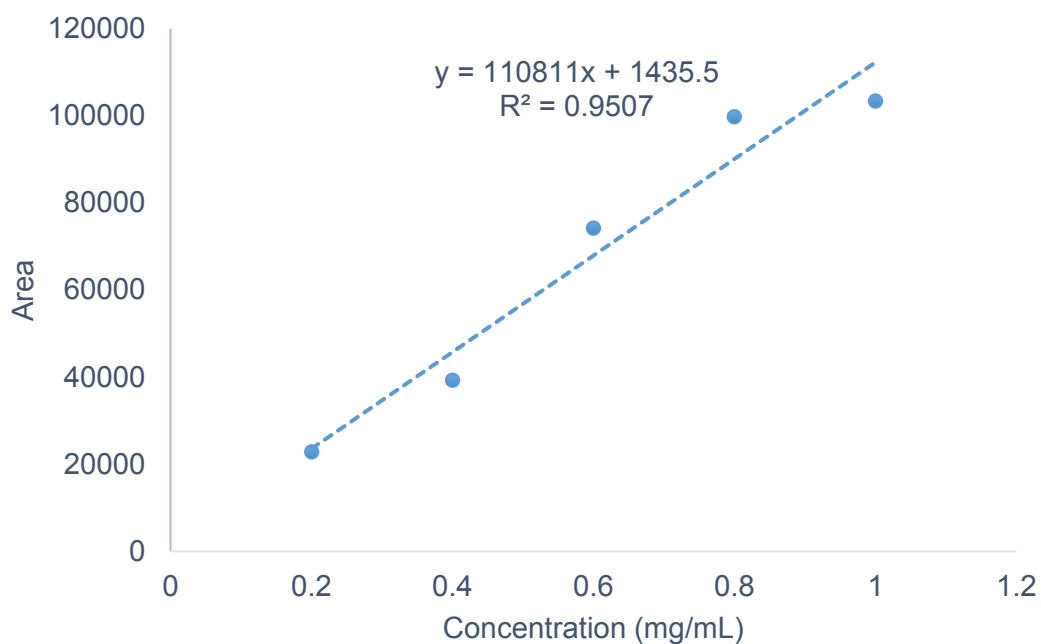
**Appendix C2. Standard curve for xylose using Aminex HPX-87P column (Biorad, USA)**

Glucose concentration (mg/mL)	Area
0.2	15839.0
0.4	33702.0
0.6	51050.5
0.8	69993.0
1.0	87750.5



**Appendix C3. Standard curve for fructose using Aminex HPX-87P column (Biorad, USA)**

Glucose concentration (mg/mL)	Area
0.2	22848
0.4	39307
0.6	74241
0.8	99802
1.0	103411



## APPENDIX D

### Calculation of glucose yield conversion

#### Untreated common cultivar

Percentage cellulose content = 38.77%

Glucose concentration = 11.07 g/L

#### Saccharification condition

Weight biomass = 1.2 g

Working volume = 20 mL

Total weight glucose =  $\frac{\text{Percentage cellulose content} \times \text{Weight biomass (g)}}{100}$

$$= \frac{38.77 \times 1.2 \text{ g}}{100}$$

$$= 0.465 \text{ g}$$

Theoretical glucose concentration =  $\frac{\text{Total weight glucose} \times 1000}{\text{Working volume}}$

$$= \frac{0.465 \text{ g} \times 1000}{20 \text{ mL}}$$

$$= 23.25 \text{ g/L}$$

$$= 23.25 \text{ g/L}$$

% glucose conversion yield =

=  $\frac{\text{Concentration glucose after saccharification}}{\text{Theoretical glucose concentration}} \times 100$

Theoretical glucose concentration

$$= \frac{11.07 \times 100}{23.25 \text{ g/L}}$$

$$= 47.59 \%$$

$$= 47.59 \%$$

## AAPPENDIX E

### Calculation for percentage increment in height for Napier grass cultivars

Common cultivar treatment

Week	0	1	2	3	4	5	6	7	8
Height (cm)	180.0	186.5	190.3	199.1	211.5	231.6	253.0	273.7	291.1
Percentage increment (%)	0	3.61	5.72	10.61	17.50	28.67	40.56	52.06	61.72

Percentage of height increment =  $\frac{\text{Height final (cm)} - \text{Height initial (cm)}}{\text{Height initial (cm)}} \times 100$

Height initial (cm)

$$= \frac{291.1 \text{ cm} - 180.0 \text{ cm}}{180.0 \text{ cm}} \times 100$$

180.0 cm

$$= 61.72 \%$$

## APPENDIX F

### List of elements concentrations in POME FD (treatment) and rain water (control)

Element	POME FD (ppb)	Rain water (ppb)
<b>Macronutrient</b>		
Calcium (Ca)	75,721.30 ± 11,302.60	8,210.92 ± 284.56
Potassium (K)	9,571.31 ± 1,537.03	3,816.69 ± 93.71
Magnesium (Mg)	320,639.44 ± 3,379.20	980.28 ± 32.85
<b>Micronutrient</b>		
Copper (Cu)	6.69 ± 0.32	0.38 ± 0.03
Iron (Fe)	514.76 ± 9.91	60.48 ± 1.75
Manganese (Mn)	10.86 ± 0.70	14.29 ± 0.78
Zinc (Zn)	3.64 ± 0.17	1.34 ± 0.48
<b>Heavy Metal</b>		
Silver (Ag)	0.002 ± 0	0.086 ± 0.01
Aluminium (Al)	100.33 ± 8.91	30.65 ± 2.26
Arsenic (As)	35.73 ± 0.04	0.89 ± 0.01
Beryllium (Be)	0.022 ± 0.01	0.078 ± 0.03
Bismuth (Bi)	0.51 ± 0.01	0.673 ± 0.03
Cadmium (Cd)	0.053 ± 0.02	0.074 ± 0.05
Cobalt (Co)	2.20 ± 0.08	0.14 ± 0.11
Chromium (Cr)	8.27 ± 0.13	2.89 ± 0.24
Gallium (Ga)	0.011 ± 0.02	0.968 ± 0.02
Indium (In)	0.094 ± 0.01	0.006 ± 0.01
Nickel (Ni)	11.65 ± 0.48	1.17 ± 0.17
Lead (Pb)	0.19 ± 0.01	0.08 ± 0.01
Thallium (Tl)	0.169 ± 0	0.001 ± 0.03
Uranium (U)	0.623 ± 0.03	0.081 ± 0.01
Vandium (V)	12.704 ± 0.17	1.639 ± 0.02
<b>Non-essential element</b>		
Barium (Ba)	0.99 ± 0.04	16.446 ± 0.54
Caesium (Cs)	111.509 ± 0.9	0.705 ± 0.02
Sodium (Na)	14383.72 ± 452.14	5377.803 ± 132.16
Rubidium (Rb)	4762.044 ± 31.67	21.106 ± 0.3
Selenium (Se)	3.464 ± 0.06	1.069 ± 0.2
Strontium (Sr)	299.35 ± 7.6	24.908 ± 0.4

## APPENDIX G

### Acceptable conditions for discharge of industrial effluent of standards A and B, Environmental Quality (Industrial Effluents) Regulations 2009

Parameter	Unit	Standard	
		A	B
Temperature	°C	40	40
pH Value	-	6.0-9.0	5.5-9.0
BOD, at 20°C	mg/L	20	50
Suspended Solids	mg/L	50	100
Mercury	mg/L	0.005	0.05
Cadmium	mg/L	0.01	0.02
Chromium, Hexavalent	mg/L	0.05	0.05
Chromium, Trivalent	mg/L	0.20	1.0
Arsenic	mg/L	0.05	0.10
Cyanide	mg/L	0.05	0.10
Lead	mg/L	0.10	0.5
Copper	mg/L	0.20	1.0
Manganese	mg/L	0.20	1.0
Nickel	mg/L	0.20	1.0
Tin	mg/L	0.20	1.0
Zinc	mg/L	2.0	2.0
Boron	mg/L	1.0	4.0
Iron (Fe)	mg/L	1.0	5.0
Silver	mg/L	0.1	1.0
Aluminium	mg/L	10	15
Selenium	mg/L	0.02	0.5
Barium	mg/L	1.0	2.0
Fluoride	mg/L	2.0	5.0
Formaldehyde	mg/L	1.0	2.0
Phenol	mg/L	0.001	1.0
Free Chlorine	mg/L	1.0	2.0
Sulphide	mg/L	0.50	0.50
Oil and Grease	mg/L	1.0	10
Ammoniacal Nitrogen	mg/L	10	20
Colour	ADMI	100	200

## BIODATA OF STUDENT

Nurul Atiqah Osman was born on January 24, 1994. She attended her preliminary school at Sekolah Kebangsaan Sungai Siput (U), Perak for her Ujian Penilaian Sekolah Rendah (UPSR) on 2006. She continued her secondary education at Sekolah Menengah Kebangsaan Tok Muda Abdul Aziz, Sungai siput (U), Perak (2007-2009) and Sekolah Menengah Sains Raja Tun Azlan Shah, Taiping, Perak (2010-2011). Later, in 2012, she went to Perak Matriculation College, Gopeng, Perak for one-year matriculation program (Module 1: Mathematics, Chemistry, Physics, and Biology) organized by the Malaysia Education Ministry (MOE). After completion of her matriculation program, she was promoted to continue her First Degree in Bachelor of Science (Honour) Biotechnology, a four-year program at Universiti Putra Malaysia, Serdang, Selangor. She attended three-month internship from June to September 2016 at Veterinary Research Institute, Perak and being placed in Virology Avian Department. During her final semester of Bachelor's Degree, she managed to complete a final year project, entitled "Comparison between Open System Wetland and Closed System Wetland in Treating Palm Oil Mill Effluent Final Discharge". She started her Master's Degree in the field of Environmental Biotechnology under supervision of Dr. Ahmad Muhaimin Roslan. The result of her research is as presented in this thesis.

## LIST OF PUBLICATIONS

### Paper publication:

- Farhana Aziz Ujang, Nurul Atiqah Osman, Juferi Idris, Mohd Izuan Effendi Halmi, Mohd Ali Hassan and Ahmad Muhaimin Roslan. (2018, June). Start-up treatment of palm oil mill effluent (POME) final discharge using Napier Grass in wetland system. In IOP Conference Series: Materials Science and Engineering (Vol. 368, No. 1, p. 012008). IOP Publishing.
- Nurul Atiqah Osman, Ahmad Muhaimin Roslan, Mohamad Faizal Ibrahim and Mohd Ali Hassan. (2020). Potential use of *Pennisetum purpureum* for phytoremediation and bioenergy production: a mini review. *Asia Pacific Journal of Molecular Biology and Biotechnology (APJMBB)*
- Nurul Atiqah Osman, Farhana Aziz Ujang, Ahmad Muhaimin Roslan, Mohamad Faizal Ibrahim and Mohd Ali Hassan. (2020). The effect of Palm Oil Mill Effluent Final Discharge on the Characteristics of *Pennisetum purpureum*. *Scientific Report*

### Abstract in conference/symposium:

- Nurul Atiqah Osman, Farhana Aziz Ujang and Ahmad Muhaimin Roslan. (2017) Comparison between Open System Wetland and Closed System Wetland in Treating Palm Oil Mill Effluent Final Discharge. In the Wood & Biofibre International Conference (WOBIC) 2017, Hotel Bangi-Putrajaya, Selangor, Malaysia. (Poster presenter)
- Nurul Atiqah Osman, Farhana Aziz Ujang, Ahmad Muhaimin Roslan, Mohamad Faizal Ibrahim and Mohd Ali Hassan. The Effect of Palm Oil Mill Effluent Final Discharge on the Growth of *Pennisetum Purpureum* Cultivars. In the 6<sup>th</sup> SAES – International Symposium on Applied Engineering and Sciences (SAES2018) UPM – Kyutech, Kyutech Tobata Campus, Fukuoka, Japan. (Poster presenter)
- Nurul Atiqah Osman, Farhana Aziz Ujang, Ahmad Muhaimin Roslan, Mohamad Faizal Ibrahim and Mohd Ali Hassan. Effect of Palm Oil Mill Effluent Final Discharge on the Characteristics of Napier Grass. In the 7<sup>th</sup> SAES – International Symposium on Applied Engineering and Sciences (SAES2019) UPM – Kyutech, UPM, Selangor, Malaysia. (Poster presenter)
- Nurul Atiqah Osman, Farhana Aziz Ujang, Ahmad Muhaimin Roslan, Mohamad Faizal Ibrahim and Mohd Ali Hassan. Effect of Palm Oil Mill Effluent Final Discharge on the Characteristics of Napier Grass. In the AFOB-Malaysia Chapter International Symposium (AFOBMCIS 2019), The Everly Hotel Putrajaya, Malaysia. (Oral presenter)





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