COMBINATION OF ASPERGILLUS NIGER FERMENTATION AND BROKEN RICE AS COAGULANT FOR TREATMENT OF PALM OIL MILL EFFLUENTS

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by

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LIST OF ABBREVIATIONS

POME	Palm Oil Mill Effluent
COD	Chemical Oxygen Demand
BOD	Biochemical Oxygen Demand
TSS	Total Suspended Solid
OPF	Oil Palm Fronds
EFB	Empty Fruit Bunches
OPT	Oil Palm Trunks
FFB	Fresh Fruit Bunches
СРО	Crude Palm Oil
OFAT	One-Factor-At-A-Time
PKS	Palm Kernel Shell
MF	Mesocarp Fibers
DOE	Department of Environment
EQA	Environmental Quality Act
GRAS	Generally Recognized as Safe
FTIR	Fourier-Transform Infrared Spectrometry
TKN	Total Kjedahl Nitrogen

LIST OF SYMBOLS

°C	Degree Celsius
%	Percentage
mg	Milligram
g	Gram
kg	Kilogram
vol	Volume
mL	Milliliter
L	Liter
U/mL	Unit Per Milliliter
рН	Potential Of Hydrogen
μm	Micromole
min	Minutes
h	Hours

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KOMBINASI PENAPAIAN ASPERGILLUS NIGER DAN BERAS HANCUR SEBAGAI PENGGUMPAL UNTUK RAWATAN

AIR SISA KILANG KELAPA SAWIT.

ABSTRAK

Salah satu isu terbesar yang telah banyak disumbangkan oleh industri kelapa sawit ialah air sisa kilang kelapa sawit. Sebelum dilepaskan ke alam sekitar, air sisa (efluen) kilang kelapa sawit kerap dirawat pada skala industri menggunakan sistem kolam. Matlamat utama kajian ini adalah untuk merawat efluen kilang kelapa sawit agar pelepasan akhir mematuhi had standard yang ditetapkan. Apabila Aspergillus niger digunakan untuk merawat efluen yang dikumpul, parameter kritikalnya, termasuk permintaan oksigen kimia, jumlah pepejal terampai, kandungan fosforus, kandungan nitrogen ammonia, dan aktiviti amilasenya, dipantau sepanjang penapaian. Sebelum menjalankan penapaian efluen dalam bioreaktor tangki kacau 2.5 L, POME telah diprarawat dengan pelbagai frekuensi sonifikasi. Penapaian POME dalam kelalang goncang pada mulanya dijalankan dengan pelbagai nilai pH. Nilai pH terbaik dengan peratusan penyingkiran permintaan oksigen kimia (COD) tertinggi kemudiannya dipilih. Rawatan sekunder, yang melibatkan rawatan pembekuan menggunakan beras hancur sebagai agen pengumpalan semulajadi, juga diteruskan pada efluen yang dirawat. Pengaruh suhu, pH, masa pengawetan, dan dos beras pecah dinilai untuk pengoptimuman pengumpalan rawatan ini oleh proses saringan OFAT. Pelepasan akhir efluen yang dirawat dalam kajian ini berbanding pelepasan akhir had piawai efluen jelas digambarkan oleh neraca jisim. pH terbaik yang dilaporkan dengan peratusan tertinggi penyingkiran permintaan oksigen kimia (COD) ditentukan ialah pH 9 pada hari ketiga

penapaian dengan 49%. Juga, didapati bahawa pada hari ke-3 penapaian, POME prarawatan dengan frekuensi sonikasi 30 kHz mempunyai peratusan penghapusan COD tertinggi (74%). Apabila beras hancur digunakan sebagai rawatan sekunder, didapati bahawa pH 3, pada suhu bilik, dengan masa rawatan selama 15 minit, dan 12g/L beras hancur menghasilkan keputusan dengan peratusan penghapusan COD tertinggi (48%). Imbangan jisim akhirnya disatukan dengan berkesan. Penyingkiran nutrien dari peringkat terawal hingga peringkat akhir projek ini, iaitu rawatan POME, ditunjukkan dengan jelas oleh keseimbangan jisim, yang juga digunakan untuk membandingkan keputusan (efluen akhir yang dilepaskan) dari projek ini dengan efluen yang telah ditetapkan had. Oleh itu, *Aspergillus niger* yang digunakan dalam kajian ini boleh menjadi sumber berpotensi untuk rawatan biologi efluen selain sistem kolam konvensional dan beras hancur juga menunjukkan hasil yang baik untuk digunakan sebagai agen pengumpalan semula jadi dalam rawatan fizikokimia.

COMBINATION OF ASPERGILLUS NIGER FERMENTATION AND BROKEN RICE AS COAGULANT FOR TREATMENT OF PALM OIL MILL EFFLUENTS

ABSTRACT

One of the biggest issues faced by the palm oil industry is the released POME in their daily production. Prior to being released into the environment, palm oil mill effluent (POME) is frequently treated at industrial scale using ponding systems. The primary goal of this study was to treat POME prior discharge into environment at established standard limit. When Aspergillus niger was used to treat the collected POME, its critical parameters, including chemical oxygen demand, total suspended solids, phosphorus content, ammoniacal nitrogen content, and its amylase activity, were monitored throughout the fermentation. Before starting the fermentation of the POME in the 2.5 L stirred-tank bioreactor, the POME was pretreated with various sonification frequencies. The POME fermentation in the shake flask was initially carried out by varying the pH value. The pH and the sonification frequency were used as a pretreatment before starting the biological treatment. The best pH value and sonification frequency with the highest chemical oxygen demand (COD) removal percentage was then selected. The secondary treatment, which involves the coagulation treatment utilizing broken rice as the natural coagulant, was also continued on the treated POME. The influence of temperature, pH, curing time, and broken rice dose was assessed for this treatment's coagulation via optimization using one-factor-at-a-time (OFAT). The final discharged of POME treated in this study as compared to the final discharged of POME standard limit was clearly depicted by the

mass balance. The best pH reported with the highest percentage of chemical oxygen demand (COD) elimination was determined to be pH 9 on the third day of fermentation with 49% removal efficiency. Also, it was discovered that on day 3 of fermentation, POME pretreated with 30 kHz sonication frequency has the highest COD elimination percentage (74%). When broken rice was used as a secondary treatment, it was discovered that pH 3, room temperature, 15 minutes of curing time, and 12 g/L of broken rice produced the results with the highest COD elimination percentage (48%). The removal of nutrients from the earliest stages to the end stage of this project, which is the POME treatment, was effectively represented by a mass balance, which was also used to compare the results (finally discharged POME) of this project to the standard POME discharged limit. Hence, the *Aspergillus niger* used in this study could be potential source for POME biological treatment besides the conventional ponding system and broken rice also showed a good result to be used as natural coagulant in physicochemical treatment.

CHAPTER 1 INTRODUCTION

1.1 Research Background

Oil Palm, also known as *Elaeis guineensis*, is one of the most lucrative, well-known, and viable agricultural crops in tropical countries like Malaysia. Today, the Malaysian economy is mostly fueled by the palm oil industry. Despite the worldwide COVID-19 pandemic, this sector still account for around RM73.25 billion of Malaysia's export revenues in 2020 (Parveez et al., 2021).

In the nation, there were around 452 palm oil mills, which processed 98.28 million tonnes of fresh fruit bunches (FFB) (S.K. Loh et al., 2019). Despite Malaysia being one of the world's leading manufacturers of palm oil, this sector produces tonnes of which significant waste each problems year, poses for waste management (N. Abdullah & Sulaim, 2013). The palm oil industry generates a lot of waste, including liquid waste from palm oil mill effluents (POME) as well as solid waste in the form of oil palm fronds (OPF), trunks (OPT), and empty fruit bunches (EFB).

POME by far the largest waste produced and challenging to handle (Awalludin et al., 2015). POME is made up of various suspended materials. POME when compared to municipal sewage is one hundred times more contaminated. It detected with a high chemical oxygen demand (COD). Additionally, the effluent has larger levels of phosphorus, organic nitrogen, and other supplement substances (Hadiyanto et al., 2013). These wastes are produced daily concomitantly creates a severe problem. Though non-toxic, the impact of POME discharge activities to the environment and human health is unavoidable.

These concerns have kept many people up especially the Department of Environment (DOE) to enforce a strict law on discharge limit of treated POME. Therefore, various treatment strategies have been developed with the goal of maintaining below the discharge limits. Conventionally, prior to being released into the environment, POME from plantations often treated using the open pond system as it believes to be simple and cheap (Chia et al., 2020). Yet, this conventional treatment, which is used by most mills, takes up a lot of space, has a long retention time, and produces gases that are unpleasant and dangerous and there are still several palm oil plantation mills that cannot adhere to the discharge restrictions even after the treatment.

A sophisticated technology for treating POME in a well-managed way must be developed as a novel solution to these persistent POME wastes problem. Prior to getting released into the environment and waterways, POME needs to be treated. Numerous studies have been undertaken as a result to treat POME using different strategies, including biological, physicochemical, thermochemical, and hydrothermal. All in all, biological methods utilizing microorganisms like fungus have drawn considerable interest and are the subject of the current research for the treatment of POME waste.

Besides treating POME, researchers have also looked into the possibility of producing beneficial and value-added materials from treated POME such as enzymes. Razak et al 1999's, who was the first to remove lipases from POME. Ohimain claim that POME is also capable of producing extracellular enzymes such as xylanase, pectinase, and amylase (Ohimain et al., 2012). Since its high organic content, POME is ideal for microbial growth and the production of enzymes., it can be utilized as an inexpensive substrate for various microbes to generate various enzymes (Chia et al., 2020).

Amylase is one of the essential enzymes that is in great demand due to its supposed technological importance and financial benefits (Sindhu et al., 2017). By 2024, the market for amylase is anticipated to reach USD 320.1 million (Grand View Research, 2016). Due to their availability and productivity, microbial source amylases typically satisfy industrial demand. Among these microbial sources, fungal amylases have been employed extensively in industrial production owing to it generally recognized as safe (GRAS) status, cost effectiveness, uniformity, requirement for less time and space during manufacturing, and ease of process adjustment and optimization (Ahmed et al., 2015).

In a nutshell, managing palm oil mill effluent (POME) waste in a sustainable manner is now more difficult than it has ever been. The treatment assessment should be carried out meticulously and correctly to avoid causing lingering problems as the treatment advances. Therefore, researchers today must go a step further by not only improving the way POME are treated, but also by employing POME to produce products with added value that may contribute to the development of our environment and economy.

1.2 Problem Statement

One of Malaysia's greatest industries, the oil palm sector, produces a variety of waste as a byproduct of the oil extraction process. The primary source of waste, according to Cheng, is palm oil mill effluent (POME), which is released at a rate that is 3.12 times higher than the amount of the intended product itself, crude palm oil (CPO) (Cheng et al., 2019). This causes a significant amount of untreated, unused POME waste to be released.

The current treatment process, open ponding system, which has been adopted by most oil palm mills, takes up a longer time and a very large land area needed (Kamin et al., 2020). Keep digging the pond for the storage of POME does not make sense, because it never ends and some of the treated POME still does not meet the standard industrial discharge limit.

Only a few studies have been reported up to this point using POME as a fermentation medium with *A. niger* for biological treatment and producing value added product from it. The literature review also showed that some of the parameters was not well documented such as the removal of nitrogen and phosphorus.

1.3 Research scope and Objectives

The main aim of this study was to treat and utilize the abundant POME wastes generated by oil palm industry. The measurable objectives of this study are:

1.3.1 Objective 1

To characterize the raw POME and sterile POME, study the effect of different pH value and different sonication frequency on chemical oxygen demand (COD), total suspended solid (TSS), nitrogen content, phosphorus content and amylase activity by *Aspergillus niger* fermentation during POME treatment.

1.3.2 Objective 2

To investigate the coagulation effect using broken rice on treated POME.

1.4 Description of the Study

In addition to treating palm oil mill effluents (POME), the goal of this research is to simultaneously develop valuable by-products from *A. niger* fermentation. Currently, POME treatment takes a long time, requires a lot of land, expensive, and lays a heavy load on the government.

To minimize data inconsistency, the POME sample was only once collected. The raw POME and the sterile POME were firstly characterized. Along with the physical characteristics, this study also looked into the chemical components of the POME during the treatment process, including proximate analysis, chemical oxygen demand (COD), total suspended solid (TSS), crude amylase, phosphorus (P), and ammoniacal nitrogen (NH₃-N).

In order to choose an appropriate condition for POME treatment, the study was further investigated on the effect of the different pH on the POME treatment using a shake flask. Next, the best pH with high COD removal were selected for second phase which to study the effect of different sonication frequency on the POME treatment using bioreactor. Subsequently, the sonification frequency with the highest COD removal was selected to final phase.

Final phase of this study was on the coagulation and flocculation treatment of POME using the broken rice. Lastly, for the broken rice treatment, process optimization of different culture condition also was carried out using "one-factor-at-a-time" (OFAT) method. The parameters tested were pH, temperature, curing time, and the dosage of the broken rice.

In essence, this study's findings provide a better understanding on effectiveness of POME biological treatment and expand the range of potential applications for environmentally friendly and sustainable activities.

1.5 Significant of this Study

By planting oil palm seeds at Tennamaram Estate in Batang Berjuntai, Selangor, in 1917, Henri Fauconnier laid the foundation for Malaysia's palm oil business, and this year, Malaysian palm oil celebrates 105 years of cultivation. Along the road, several key milestones have been reached, and with each milestone, this industry has encountered and addressed a sizable number of challenges. However, some of the problems are still open to this day.

The researcher's current key goal is to properly address the palm oil industry's waste problem because the demand for palm oil is expected to rise dramatically and reach 240 Mt by the year 2050 (Mardiharini et al., 2021). A worse situation will arise if a better solution is not established. Table 1.1 shows the comparison between conventional treatment methods with current study.

The palm oil industry as well as the environment may be significantly impacted by the bioremediation of palm oil mill effluents (POME) utilizing *A. niger*. As for industry benefits, it offers a sustainable and eco-friendly approach to treating POME and mitigating its environmental impact. Byproducts like enzymes, organic acids, and microbial biomass can be produced as a result of Aspergillus niger-based bioremediation. These byproducts can be used in a variety of industrial processes, which might result in new sources of income for the palm oil sector. Adopting sustainable practices such as bioremediation aligns with the growing demand for sustainable palm oil production. This can enhance the industry's reputation and marketability, especially in regions with a focus on sustainable sourcing. While the positive impacts on the environment are reduced pollution, lower greenhouse gas emissions, enhanced nutrient recovery, decreased ecological impact and promote of circular economy.

There are seventeen sustainable development goals (SDGs), which were developed by the United Nations with the intention of being accomplished by the year 2030, provide a road map for building a socially, economically, and environmentally sustainable future. This study fits three of the goals, which are: (6) clean water and sanitation, (13) climate action, and (14) life below water. Table 1.1: Comparison between Conventional and Current Treatment Method.

Element	Conventional Treatment	In This Study
Type of Treatment	Biological	Biological + Physicochemical
Substrate	Raw POME	Raw + Sterile POME
Inoculum	Mixture of bacteria, fungus, and microalgae (Co-culture)	Aspergillus niger
		Batch Cultivation
Condition	Ponding treatment	(Shake flask & bioreactor)
Duration	$\approx 115 \text{ days}$	$\approx 8 \text{ days}$
Environment	Lack of control	Easily control

CHAPTER 2 LITERATURE REVIEW

2.1 Palm Oil (*Elaeis guineensis*)

2.1.1 Palm Oil & Cultivation

Palm Oil or scientifically known as *Elaeis guineensis* is one of the most adaptable plants grown in tropical regions. With its tropical climate and abundant rainfall, Malaysia offers ideal living conditions for oil palm trees (Onoja et al., 2019). British colonists first planted the trees in Malaysia in 1871 as an ornamental plant. The goal for producing crude palm oil (CPO) comes later in 1917. Palm oil was originally from West Africa (Awalludin et al., 2015). The taxonomy of palm oil shown in table 2.1 below. The African palm oil or Macaw Fat, which is a member of the family *Arecaceae*, is thought to be native to West Africa because the epithet *guineensis* indicates that the first specimen reported was found in Guinea, West Africa.

Kingdom	: Plantae
Phylum	: Magnoliophyta

Class : Liliopsida

Order : Arecales

Family : Arecaceae

Species : Elaeis guineensis Jacq.

: Elaeis oleifera

Genus : Elaeis

Table 2.1: Taxonomy of Palm Oil (S. S. S. Abdullah et al., 2015).

As a monoecious plant with no branches, palm oil can produce both male and female flowers on the same tree. The flowers are produced in tight clusters, and each one has three sepals and petals in addition to being petite. Young and mature plants' trunks are covered in fronds, presenting it in a rough manner. Aside from the wounds left by the fronds that have wilted and fallen off, the elder trees' trunks are smoother. It took a palm oil tree about three months to get through the germination stage. It starts to produce fruit in bunches after 2.5 to 3 years of field planting and will continue to do so throughout the whole year. A mature palm oil tree has a single stem that can reach a height of 20 meters and pinnate leaves that can extend up to three to five meters in length (Corley & Tinker, 2015). The usual bunch of an oil palm weighs 10 and 25 kilos, weighing an average of 1,000 to 3,000 fruitlets. The oil palm's fruitlet, also known as the fruit, has a form that resembles a sphere or an elongated cylinder. Typically, the fruitlet has a deep purple color that almost looks black and when it is mature, the color changes to orange red. A hard kernel (seed) and a shell (endocarp) with a soft mesocarp around it

make up each fruitlet. The size of a palm fruit is comparable to a huge olive. The oil palm has a lifespan of at least 100 years and can reach heights of up to sixty feet. Every 25 to 30 years, oil palm trees are typically replaced with newer varieties for economic reasons.

About 30 to 35% of the oil in each palm fruit is present. As mentioned by, Yaap, it is more advantageous to grow oil palm trees in a region with a low-lying altitude, a moist tropical rainforest, and high rainfall (Yaap et al., 2010). Tenera, a hybrid of the *dura* and *pisifera* species, is the oil palm species that is most frequently planted in Malaysia. Tenera was picked because it provides good yields for both palm oil and palm kernel oil. The tenera variety yields approximately 1 tonne of palm kernels and 4 to 5 tonnes of crude palm oil (CPO) per hectare annually (Dian et al., 2017).

2.1.2 Palm Oil Industry in Malaysia

The nation's palm-growing industry currently focuses on growing trees for oil. However, the growth of the oil palm in Malaysia began first as a decorative plant in 1870. In 1917, Tennamaram Estate, Batang Berjuntai in Selangor became the first industrial oil palm estate in Malaysia (Naidu & Moorthy, 2021). Malaysia being one of the world's largest producer and exporter of palm oil. Malaysia's palm oil plantations have grown dramatically over time and in 2017, Malaysia celebrated Malaysian Palm Oil's 100th anniversary. Today, Malaysia's economy, so-cioeconomic progress, political stability, and capacity for innovation have all been supported by the palm oil industries.

The Malaysian government is firmly committed to the development of the sector and supports increased palm oil output on a worldwide scale. The globe now accepts palm oil widely, and Malaysia has sold palm oil to more than 140 countries. The overall export revenue increased by 48.0% to RM108.52 billion from RM73.33 billion in 2020 due to increasing pricing in international trade. When compared to RM45.65 billion in 2020, the revenue from palm oil exports alone dramatically increased in 2021 by 41.6% to RM64.62 billion (Parveez et al., 2021; MPOB, 2021).

2.1.3 Palm Oil Production and Processing Industries

Nearly all of Malaysia's palm oil mills, employ a similar process of extraction and production that comprises of few different stages and multiple working units. Standard schematic of the extraction process for crude palm oil is display in Figure 2.1. Within 24 hours after being harvested, the red fruit bunches are delivered to the mills. Once there, various groups of these fresh fruit bunches (FFBs) are divided to assess their quality. How ripe the oil palm fruit is one of the most important factors influencing the quality of the bunches. Initially, the graded fresh fruit bunch (FFB) is sterilized, then digested, and pressed to make pressed liquid and cake (Foong et al., 2019).

These FFBs are steam sterilized for 75–90 minutes at 140 °C and 3 bars of pressure. The importance of this procedure is to facilitate stripping, reduce further synthesis of free fatty acids owing to enzyme activity, and get the fruit mesocarp ready for processing. One of the main sources of liquid effluent is the steam condensate that is produced by the sterilizer. The fruits are separated from the bunches after the FFBs have been sterilized and put onto a rotating drum-stripper. The fruits that have been removed are fed through the bar screen of the stripper, gathered below by a bucket conveyor, and then dumped into a digester (Vincent et al., 2014).

Fruits are crushed by spinning arms in the digester. At this stage, the mesocarp's oil-bearing cells are broken by the boiling and mashing of the fruits. Under intense pressure, oil is often extracted from the digested fruit mash using twin screw presses. To improve the flow of the oils, hot water is introduced. A clarification system is then fed the crude oil slurry to separate and purify the oil. To separate the fiber and nut (press cake), the material is transported to a depericarper (Vincent et al., 2014).

The crude palm oil (CPO) produced by screw presses is a blend of palm oil (35 to 45%), water (45 to 55%), and fibrous components in various amounts. Then, for oil separation, it is sent to a horizontal or vertical clarity tank. The clarified oil in this device is continuously skimmed off the top of the tank used for clarification. Before being sent to the storage tanks, it is then put through a vacuum dryer and a high-speed centrifuge (Vincent et al., 2014).

The press cake that is discharged from the screw press is made up of moisture, oily fiber, and nuts. The cake is then sent to a depericarper for the separation of the nuts and fibers. A suction fan-induced strong air stream separates the fiber and nuts. Typically, the fiber is sent to the boiler building where it is used as boiler fuel. The nuts are then delivered to a spinning drum where any fiber is removed before being delivered to a nutcracker. A hydrocyclone is routinely used to separate the kernels and shells. The final source of wastewater stream is the outflow from this procedure (Vincent et al., 2014)



Figure 2.1: A typical palm oil mill's unit operations (Tan & Lim, 2019).

2.1.4 Waste Generated from Palm Oil Industries.

As the Palm Oil industries proliferate, the waste produced by these operations also expands and dangerously polluting the environment in its vicinity. Typically, an oil palm tree's biomass makes up 90% of its weight and its palm oil 10% (Onoja et al., 2019). Oil palm biomass is made up of two different forms of lignocellulosic wastes: solid wastes and liquid wastes. This biomass is produced during the plantation's pruning, harvesting, replanting, and milling processes as shown in Figure 2.2. These wastes are oil palm fronds (OPF), oil palm trunks (OPT), empty fruit bunches (EFB), palm kernel shell (PKS), mesocarp fibers (MF) and palm oil mill effluent (POME) (Onoja et al., 2019). The components of FFB produced annually is shown in Table 2.2. (Soh Kheang Loh, 2017). POME was the fresh fruit bunch with the greatest availability or abundance (67% of FFB on a wet basis), followed by 22% empty fruit bunch (EFB), 13.5% mesocarp fiber (MF), and 5.5% palm kernel shell (PKS) (Soh Kheang Loh, 2017).



Figure 2.2: Oil palm biomass source of generation and residue.

Table 2.2: Industrial oil palm mill residual (S. S. S. Abdullah et al., 2015; Soh Kheang Loh, 2017)

Component of Fresh Fruit Bunch (FFB)	Residue Production
Oil Palm Fronds (OPF)	46837 ktonnes
Oil Palm Trunks (OPT)	10827 ktonnes
Palm Kernel Shell (PKS)	5.5% of FFB (Wet basis)
	85% of PKS wet basis (dry basis)
Empty Fruit Bunch (EFB)	22% of FFB (wet basis)
	35% of EFB wet basis (dry basis)
Mesocarp Fibre (MF)	13.5% of FFB (wet basis)
	60% of MF wet basis (dry basis)
Palm Oil Mill Effluent (POME)	67% of FFB (wet basis)

2.1.4 (a) Palm Oil Mill Effluent (POME)

Considering the environment, POME is a non-toxic oily wastewater generated from extraction process of oil palm. In general, for each tonne of FFB (fresh fruit bunches) processed, a standard palm oil mill generates *circa* 1 tonne of POME which is a combined waste from sterilizer condensate and cooling water. It should be highlighted that the largest source of POME production occurs during the clarification process, followed by sterilizer condensate and hydro-cyclone effluent during separation, each contributing 60%, 36%, and 4% to POME production, respectively (Tan & Lim, 2019).

Table 2.3 shows the characteristics of POME. It is hot (80-90°C) and has a high chemical oxygen demand (COD) and biochemical oxygen demand (BOD) (Kamyab et al., 2018) as well as acidic with a pH around 4.5 due to the presence of organic acids

that are suitable to be used as carbon sources (Kamyab et al., 2018). However, POME comprises of different suspended materials. POME which is 100 times more polluted than the municipal sewage. Besides the high COD and BOD value, it also contains higher concentration of organic nitrogen, phosphorus, and different supplement substance (Phaik Eong Poh et al., 2010).

Every batch of POME generated may vary significantly due to batches variation, days, and factories, processing procedures, and the age or variety of fruit (M S, 2020). In addition to that, the nature of POME may be affected by the factory's discharge limit, the climate, and the state of palm oil processing (Akhbari et al., 2020). Raw or partially processed POME has an organic substance, which is attributed in part to the presence of unrecovered palm oil. Most POME's components, which are lipids, carbohydrates, minerals, proteins, and nitrogenous substances, making it amenable to biological handling techniques (A Aziz et al., 2020).

Parameter	Average
Temperature (°C)	80-90
pH	4.2
Oil and grease	4000
Biochemical oxygen demand (BOD)	25000
Chemical oxygen demand (COD)	51000
Total solids (TS)	40000
Total suspended solids (TSS)	18000
Ammoniacal nitrogen (NH3-N)	35
Total nitrogen (TN)	750
Phosphorous	180
Potassium (K)	2270
Magnesium (Mg)	615
Calcium (Ca)	439
Iron (Fe)	46.5
Manganese (Mn)	2.0
Copper (Cu)	0.89
Zinc (Zn)	2.3

Table 2.3: Distinctive characteristics of POME (Madaki & Seng, 2013).

*All values are in mg/L except pH & Temperature

2.1.5 Treatment of Palm Oil Mill Effluent (POME)

The numerous potential treatments for POME are shown in Figure 2.3. Since some methods or systems might not be able to satisfy the minimal conditions set forth by the authorities for POME discharge, researchers are looking for ways to improve POME treatments. Every treatment approach has pluses and minuses, so it is critical to investigate all the factors that affect a treatment before deciding which approach is best to use. The removal effectiveness of the important factors such as COD, TSS, P, and N determines whether to utilize a single approach or a combination of methods to treat POME.



Figure 2.3: Difference Treatment of Palm Oil Mill Effluent (POME).

2.1.5 (a) Palm Oil Mill Effluent (POME) Current Treatment (*Ponding System*)

POME disposal before treatment is banned due to its physicochemical features, which not only disrupt the aquatic ecology and pollute water (Tan & Lim, 2019) it may also cause harm to human health. Different POME therapy methods have been investigated over time. Generally, in Malaysia, conventional open ponding systems are employed by at least 85% of the palm oil mill to treat POME and to comply with government regulation (Soh Kheang Loh, 2017). Due to its inexpensive capital outlay, low upfront capital requirements, and minimal technical requirements, this strategy is popular (Ng, 2021) and due to the lack of mechanical mixing, operation management, or monitoring (Choong et al., 2018). Traditional open ponding systems, according to Liew, include facultative ponds, aerobic ponds, settling ponds, cooling ponds, acidification ponds, and others (Liew et al., 2015). In constructing an effective pond, the depth is very crucial (Choong et al., 2018).

The process of POME treatment (Figure 2.4) starts right after the raw POME being discharge from the mill into the oil trapping tank, then the raw POME was directed into acidification pond and held it there for roughly 6 days. Prior to the anaerobic phase, POME temperature was decreased by the cooling pond in range 35 to 38°C from the initial temperature which is *circa* 90-98°C and stabilized the pH. Basically, it establishes favorable circumstances for the breakdown of organic substances. Via cooling tower, the POME which was previously kept for 7 days was pumped into a cooling pond. Highstrength wastewater was found to be best treated in anaerobic treatment ponds because it serves the dual purposes of particle settlement and organic removal (Ahmed et al., 2015). Anaerobic ponds had a depth of around 5 m and a hydraulic retention period of approximately 60 days, respectively. In this stage, during hydrolysis process, complex polymers including proteins, lipids, and carbohydrates are broken down into their corresponding monomers. During acidogenesis, acidogenic bacteria break down the hydrolyzed compounds such as soluble organic monomers of sugars and amino acids to form alcohols, aldehydes, volatile fatty acids (VFAs), and acetate along with H_2 and CO_2 (Sambusiti et al., 2014). Then, hydrogenotrophic methanogens broke down hydrogen and carbon dioxide, while acetolactic methanogens used acetic acid (HAc) and carbon dioxide (CO_2) to produce methane gas. The methane gas is regarded as the final valueadded product for the biogas production. The lignin in anaerobically treated POME partially degraded into phenolics, which caused it to become alkaline and exist as a dark brown substance (M M Bello et al., 2013). Before being released into the facultative ponds, anaerobic POME is further treated in aerobic ponds with floating aerators. After around 20 days in the aerobic pond the POME flows into facultative ponds. The facultative ponds were designed to further reduce the organic content in the effluent, for about 20 days before being discharged into polishing pond. Prior to the wastewater being subsequently discharged into natural water bodies, polishing is the tertiary and last effluent treatment stage where it needed 2 days for sedimentation and elucidation of suspended microbes.

According to several researchers, this traditional ponding system satisfies the disposal standard need, although there are certain restrictions to the minimum disposal standard requirement. The downsides of this method include the extended degrading retention time of organic contaminants (*circa* 6-7 months), large land area is needed (Zainal et al., 2018), the periodic cleaning of created biomass in anaerobic ponds, and

the creation of greenhouse gases like methane (Ng, 2021). In addition, the effluent discharge limit set by the Malaysian Department of Environment is not only violated, but it also necessitates a large environmental imprint (Akhbari et al., 2020), this approach also calls for a lengthy treatment period of up to 115 days (Tan & Lim, 2019).



Figure 2.4: Ponding System for POME Treatment in Malaysia.

2.1.5 (b) Palm Oil Mill Effluent (POME) Biological Treatment

The term "biologically-based processing technique" refers to the employment of microorganisms to break down complex organic substances found in wastewater. Microorganisms or organic material is used as an activator in process. Biological treatment (Bioremediation) is often referred to as a secondary treatment, it is employed due to cost and space concern. POME in known as a high strength pollutant. By biologically degrading pollutant including organic carbon, nutrients, heavy metals, suspended particles, and inorganic salts in the presence of microorganisms, biological processing systems for wastewater treatment aim to eliminate them (Rajasulochana & Preethy, 2016). Recent studies are shown in table 2.4 below.