

**THE USE OF MICROFILTER RECOVERED PALM OIL
MILL EFFLUENT (POME) SLUDGE AS FISH FEED
INGREDIENT**

By

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LIST OF ABBREVIATIONS

ARA	Arachidonic acid
AOAC	Association of Official Chemist
BOD	Biochemical Oxygen Demand
COD	Chemical Oxygen Demand
DOE	Department of Environment
EFA	Essential Fatty Acid
EAA	Essential Amino Acid
EPA	Eicosapentaenoic Acid
DHA	Docosahexaenoic Acid
FAO	Food and Agricultural Organization
CPO	Crude Palm Oil
CLO	Cod Liver Oil
FO	Fish Oil
FFB	Fresh Fruit Bunch
EFB	Empty Fruit Bunch
FFA	Free Fatty Acid
FCR	Feed Conversion Ratio
FID	Flame Ionization Detection
DEMM	Dead-end Microfiltration Membrane
MDPS	Microfiltered Dried Palm Oil Mill Effluent Sludge
DM	Dry Matter

FAME	Fatty Acid Methyl Esters
O&G	Oil and Grease
TS	Total Solids
TSS	Total Suspended Solids
POME	Palm Oil Mill Effluent
PUFA	Polyunsaturated Fatty Acid
HUFA	Highly unsaturated Fatty Acid
MUFA	Monounsaturated Fatty Acid
MPOB	Malaysia Palm Oil Board
ORFR	Oil Rich Fibrous Residue
PKC	Palm Kernel Cake
LWG	Lean Weight Gain
TKN	Total Kjeldahl Nitrogen
MWG	Mean Weight Gain
MIW	Mean Initial Weight
MFW	Mean Final Weight
PER	Protein Efficiency Ratio
SGR	Specific Growth Rate
PVDF	Polyvinylidene fluoride
GE	Gross Energy
ANOVA	Analysis of Variance.

PENGUNAAN ENAPEEWAR EFFLUENT KELAPA SAWIT YANG TELAH DISARING SECARA MIKRO SEBAGAI RAMUAN MAKANAN IKAN

Abstrak

Kajian ini membincangkan kebolehan menggunakan lipid, protein dan karbohidrat daripada efluen kelapa sawit (POME) yang telah melalui proses mikroturasan bersaiz 0.45µm sebagai ramuan makanan ikan Nile tilapia, *Oreochromis niloticus*. POME mengandungi kandungan bahan organik yang amat tinggi seperti lipid, protein dan karbohidrat yang merupakan kandungan organik dalam bentuk keperluan oksigen kimia (COD), pepejal terampai (SS) dan minyak dan gris (O&G). Jumlah nutrien didalam deposit terdiri daripada 45.63% lipid, 10.49% protein dan 18.82% karbohidrat. Komponen cecair saki baki tersebut telahpun dikeluarkan cecair sambil mengekalkan 96% minyak dan gris, 98.1% pepejal terampai, 81% keperluan oksigen kimia dan 43.75% jumlah nitrophile Kjeldahl (Total Kjeldahl Nitrogen). Kegunaan nutrien yang diperolehi daripada MF POME ini sebagai ramuan makanan ikan dikaji dengan memberi ikan Nile tilapia, *Oreochromis niloticus* makan enapcemar POME yang kering selama 6 minggu. Eksperimen ini dijalankan untuk mengenalpasti kesan menggantikan minyak ikan dengan enapcemar POME yang dimikroturas dari segi kadar pembesaran, faktor pemakanan dan komposisi otot ikan Nile tilapia, *Oreochromis niloticus*. 3 diet praktikal telah disediakan, iaitu minyak ikan (FO), minyak kelapa sawit mentah (CPO) dan enapcemar POME kering yang telah dimikroturas (MDPS) serta formulasi yang menggantikan FO didalam diet CPO dan MDPS. Kadar tumbesaran amat ketara ($P < 0.05$) lebih baik didalam diet DMPS (peningkatan berat ikan 97.40%) berbanding dengan makanan ikan FO (peningkatan berat ikan 64.97%) dan CPO (68.33%). Pemakanan berdasarkan kadar nisbah penukaran makanan (FCR; 1.3) dan nisbah kecekapan protein (PER; 1.97) diperolehi daripada makanan ikan MDPS berbanding dengan diet FO (FCR; 1.82, PER; 1.35) dan CPO (FCR; 1.83, PER; 1.35). Makanan ikan MDPS menunjukkan Kadar Pertumbuhan Spesifik (SGR) sebanyak 1.96 manakala diet FO dan CPO mempunyai nilai SGR 1.20 dan 1.23 masing-masing. Jangkamasa hayat makanan ikan MDPS paling tinggi berbanding makanan ikan yang lain. Keputusan kajian ini juga menunjukkan kualiti bangkai ikan tidak dikompromi dengan penggunaan MDPS. Kandungan lemak aced tepu lipid otot ikan yang diberi MDPS tidak meningkat dan menunjukkan lemak asid politaktepu (PUFA) yang rendah berbanding dengan ikan yang menerima diet FO. Walau bagaimanapun, kepekatan individu PUFA didalam otot lipid dipengaruhi oleh kepekatan PUFA beransurkan dietnya. Daripada kajian, didapati enapcemar POME boleh digunakan sebagai ramuan alternatif didalam makanan ikan Nile tilapia, *Oreochromis niloticus* berdasarkan nilai nutrien dan mudah diperolehi serta kandungan PUFA yang rendah.

THE USE OF MICROFILTER RECOVERED PALM OIL MILL EFFUENT (POME) SLUDGE AS FISH FEED INGREDIENT

Abstract

This study examined the possible utilization of the retained lipid, protein, and carbohydrate from the palm oil mill effluent microfiltered using membrane pore size 0.45µm as fish feed ingredient for culture of Nile tilapia, *Oreochromis niloticus*. POME contains high concentration of organic matter such as lipids, proteins and carbohydrates, which constitute the high organic load in the form of chemical oxygen demand (COD), suspended solids (SS) and oil and grease (O&G). The amount of the nutrient retained in the residue is 45.63% lipid, 10.49% protein and 18.82% carbohydrate. The residue has the liquid component removed while retaining 96% Oil & Grease, 98.1% Suspended Solids, 81% Chemical Oxygen Demand, and 43.75% Total Kjeldahl Nitrogen. The usefulness of the nutrients obtained from the MF POME as fish feed ingredients was examined by feeding the dried POME sludge to the Nile tilapia for 6 weeks. Feeding experiments were conducted to determine the effect of substituting fish oil with microfiltered POME sludge on growth, feed utilization and muscle composition of the Nile tilapia, *Oreochromis niloticus*. Three practical diets were prepared namely: Fish Oil (FO) diet, Crude Palm Oil (CPO) diet and Microfiltered Dried Palm Oil Mill Effluent Sludge (MDPS) diet, and the formulation contained 100% substitution of FO in the diets of CPO and MDPS. The growth performance was significantly ($P < 0.05$) better in the fish fed MDPS diet (Weight gain; 97.40%) compared to fish fed FO (Weight; 64.97%) and CPO (68.33%) diets. Similarly good Feed Utilization with respect to Feed Conversion Ratio (FCR; 1.3) and Protein Efficiency Ratio (PER; 1.97) was obtained in the fish fed MDPS diet compared to the ones fed with FO (FCR; 1.82, PER; 1.35) and CPO (FCR; 1.83, PER; 1.35) diets. The MDPS fed fish showed Specific Growth Rate (SGR) of 1.97, while the fish fed with FO and CPO diets showed a SGR of 1.20 and 1.23 respectively. There was higher rate of survival in the fish fed with MDPS compared to the other treatments. The result of the study further showed that carcass quality of the experimental fish was not compromised as a result of the use of MDPS diets and also the muscle lipids of fish fed MDPS diet did not increase in saturated fatty acids content but showed significantly lower polyunsaturated fatty acid (PUFA) concentration compared to fish fed FO diet, however, the concentrations of individual PUFA in muscle lipids were influenced by dietary PUFA concentrations. The study revealed that palm oil mill effluent (POME) sludge could be an alternative feed ingredient in the diet of Nile tilapia, *Oreochromis niloticus* based on its nutritional value and abundant availability and, also the advantage of its low PUFA content.

CHAPTER ONE

INTRODUCTION

1.1 Background

Palm oil is produced from oil palm, primarily *Elaeis guineensis*, which originated from West Africa but has adapted well to other tropical lowland regions such as Malaysia. The African oil palm belongs to the family *Palmae* and is classified *Elaeis guineensis*, it is believed to be indigenous to West Africa because the specific name, *guineensis* shows that the first specimen described was collected in Guinea, West Africa. The oil palm (*Elaeis guineensis*- an un-branched monoecious plant) is not a native plant of Malaysia; it was introduced in 1875 as an ornamental plant.

The oil palm industry in Malaysia started 80 years ago in a modest way. The oil palm products started attracting commercial value as cash crop and to some extent as food crop in 1917. The most common type of palm oil species grown in Malaysia is the cross-bred species called the *Tenera* produced from *Dura* (thick shell) and *Pisifera* (shell-less) species. The modern expansion of the agro-based industry can be traced back to the 1960s when the government embarked on a massive programme of agricultural diversification. Today, oil palm is the leading agricultural crop in Malaysia, covering about two million hectares or one third of the total cultivated area. There are over 500 million oil palm trees across various plantations in Malaysia. Today it is the

largest agricultural sector, exceeding rubber plantation more than double in area planted (Malaysia Palm Oil Board-MPOB, 2004).

The primary areas of production are Southeast Asia followed by the West Coast of Africa and Latin America. Currently, Malaysia is the world's largest producer and exporter of palm oil contributing 49.5% of world's production and 64.5% of world's export (Malaysia Palm Oil Board-MPOB, 2004). The supply gap, high returns of investment and bright external trade opportunities in the oil palm business makes investment in the oil palm sector a safe and profitable venture.

The golden oil as it is called in Malaysia has always had boast in its production, for instance in 1998, a annual production of 7,425,000 tones and monthly production of approximately 618,750 tones was recorded for Worldwide export (Industrial Process and Environmental Handbook No.3, 1999), In May 2001, the amount of crude palm oil production increased, hitting a target as high as 985,063 (Palm oil link, 2001).

Crude Palm oil is produced from palm fresh fruit bunches (PFFB) through major operational processes such as steaming and squeezing. Consequent to the operational processes in the palm oil mills, wastes load in the form of gaseous emissions from boilers and incinerators, solid wastes materials and by-products such as empty fruit bunch (EFB), potash ash, palm kernel, fiber and shells and liquid waste are produced. During the processing of the fresh fruit bunches (FFB), water is the most needed resource, it has been reported that processing/milling of one ton of FFB requires 1.5 m^3

of water, and 50% of the water used during milling is released as liquid waste known as Palm Oil Mill Effluent (POME) while the rest is lost as steam in the boilers blow down, wash waters and leakage (Industrial Processes and the Environmental Handbook No. 3, 1999). The contribution of liquid waste to the total waste load from the palm oil mills has been emphasized in earlier studies, for example Ahmad *et al.*, (2003) reported that during palm oil extraction, about 1.5 tons of POME is produced per ton of FFB processed. It is also reported that 31 million tons of FFB is produced annually and processed in 265 mills, from which 7.7 million tons of oil palm empty fruit (EFB), 6.0 million tons of fiber and 2.4 million tons of fruit shell are generated as solids wastes, and more than 10 million tons POME are generated as wastewater.

1.1.1 Palm Oil Mill Effluent (POME) Wastewater Stream

POME is a thick brownish viscous liquid waste which is non toxic but has unpleasant odor. It represents a complex substrate comprising of un-hydrolyzed materials with a high concentration of complex compounds such as proteins, fats, starch, cellulose, hemi-cellulose, and organic acids which requires hydrolysis prior to their assimilation. Due to its organic composition it is identified as one of the world's most polluting wastewater. The chemical oxygen demand (COD) and biochemical oxygen demand (BOD) of POME are very high with average values of 50,000 and 25,000 mg/l for COD and BOD respectively. However COD values greater than 80,000mg/l⁻¹ are frequently

reported. The incomplete extraction of palm oil from the palm fruits can increase COD values substantially (Oswal *et al.*, 2002). The POME is discharged at a temperature of 80-90°C and has a pH typically between 4 and 5 (Ma and Halim, 1988., Polprasert, 1989; Singh *et al.*, 1999). Wong *et al.*, (2003) reported that in 2001, the value of total crude palm oil produced in May of that year was in the tune of 985,063 tons and a total volume of 1,477,595m³ water was used in the milling process releasing 738,797m³ of POME. The oil palm industry and its processing have always been linked to the environment. According to Ahmad *et al.* (2003), the estimated amount of water required to produce 1 ton of crude palm oil is between 5-7.5 tons and more than 50% of the water will end up as POME which is a source of inland water pollution when released into local rivers or lakes without treatment. POME as one of the major wastes generated by palm oil mills originated from the mixture of sterilizer condensate, separator sludge and hydro-cyclone wastewater. It is rich in organic matter such as protein, carbohydrates, lipids and also nitrogenous compounds such as minerals that can be converted for use as microbial culture (Agamuthu & Tan, 1985; Habib *et al.*, 1997; Wu *et al.*, 2007) and possibly, animal feeds including fish. As a result of this benefit from POME, aquaculturists could reduce current dependence on natural marine resources to farm fish through the use of low-cost, locally available, alternative feed ingredients.

1.1.2 Palm Oil Mill by-product utilization

Palm oil mill effluent, the final liquid discharge after extracting the oil from the fresh fruit bunch, contains soil particles, residual oils and suspended solids but only 5% of dry matter. Palm oil sludge is the material that remains after decanting the palm oil mill effluent (Devendra, 1981). It can be filter-pressed, dried and ground to produce dehydrated palm oil mill effluent, or centrifuged in the wet state, after having undergone anaerobic fermentation. In the latter case, the product is known as fresh centrifuged sludge solids of 15 to 20% dry matter and may be dehydrated to form dry centrifuged sludge solids of between 94% and 97% dry matter.

There have been numerous attempts to convert palm oil mill effluent into a viable animal feed resource; however, most methods have been discontinued due to the large initial capital investment required, and particularly to the cost of fuel for dehydration (Davis & Reilly, 1980).

In Malaysia, one method used to convert fresh palm oil mill effluent into a potential feedstuff involved concentration by centrifugation or decantation, followed by absorption on other dry feeds like tapioca chips, grass meal or palm kernel meal (Devendra, 1992). Perhaps, one idea would be to promote the use of the fresh centrifuged sludge solids (15-20 % dry matter) for finishing pigs which, compared to younger animals, have a greater capacity to effectively use larger amounts of more liquid feeds. Apparently, this material has only been used in dry, concentrated rations

(Ong, 1982). This approach however, might require supplementation to increase the crude protein content to that of a cereal, as well as some molasses to improve palatability.

It would have to be fed immediately, preferably near the factory in order to avoid transportation of a product that contains 80% of water. Interestingly, this approach was indicated by Devendra, (1981) for feeding sheep and cattle (Devendra, 1992); he referred to the use of this residual product alone, or combined with oil-rich fibrous residue. Perhaps, this same recommendation might be applied to feeding pigs. To date, derivatives of the African oil palm have shown potential feeding value for pigs in conventional, cereal-based rations. However, interest has focused on the use of primary products and by-products of the African oil palm as a partial or complete energy source replacement in swine rations, particularly where the protein is offered separately, in the form of a restricted amount of a high-quality supplement. It has been shown that the oil-rich fibrous residue (ORFR), normally used as a source of energy to run the factory, can also furnish the necessary energy for the pig (Ocampo *et al.*, 1990a, 1990b). The successful experimental use of the crude palm oil (Ocampo, 1994b), combinations of the crude palm oil and sugar cane juice (Ngoan and Sarria, 1994) and even the whole fresh palm fruit (Ocampo, 1994a, b) further emphasized the fact that other oil palm byproducts can serve to completely replace cereals in rations for swine. Further study by Habib *et al.* (1998) showed that POME contain organic and essential nutrient which may be used to grow micro-algae as natural food for aquatic organisms and it could

also be used as fertilizer and animal feeds (Wu *et al.*, 2007). POME can be used as components in compound feeds, combining Palm kernel Cake (PKC) and POME can partly provide complete-based diets for low cost feeding systems, particularly fattening rations for beef cattle, field trials with cattle and pigs in estates have shown improved lean weight gain (LWG) with PKC-POME feeding (Wan Zakari & Alimon, 2004), Similarly, combining POME and Sago meal (40%:45%) has successfully been used for feeding local sheep.

1.1.3 Tilapia, Aquaculture and Fishmeal

World tilapia production (fishing and aquaculture) has been booming during the last decade, with an output doubling from 830.000 metric tons in 1990 to 1.68 million metric tons in 1999. Aquaculture was the main factor responsible for this increase, while landings of tilapia (600.000 metric tons) stayed more or less stable over the years (Josupeit, 2000). The global tilapia aquaculture production has surpassed one million metric tons with more than 80% of the production coming from Asian countries (food and agricultural organization-FAO, 1999). The intensive farming of tilapia is rapidly expanding at an annual rate of about 12% and there is currently a great demand for formulated feeds. However, high quality fishmeal and fish oil are the major dietary ingredients from industrial fisheries which are used to service aqua feed industry. It is envisaged that world annual fishmeal production will remain static at 6.5 million metric

tons over the next decade and world annual fish oil production will remain around 1.24 million metric tons during next decade, although this is expected to fluctuate due to El Niño, an event that affects fish meal supplies by changing ocean water temperature in rich fishing grounds (Tacon, 2004).

To keep pace with global aquaculture production, a marked increase in use and production of formulated feed is foreseen for the next 25 years (Barlow, 2000). High quality fishmeal and fish oil are the major dietary ingredients for the production of formulated feed. Against this background, it is predicted that the requirement will increase from 2.115 to 3.262 million metric tons for fishmeal and from 0.708 to 1.308 million tons for fish oil between 2000 and 2025, in-order to be able to support today's intensive aquaculture industry (Barlow, 2000).

While the demand for fishmeal for the aquaculture industry will be in the increase, it is projected that there will be a drastic reduction in the use of fishmeal for the poultry industry (Barlow, 2000) and, as a result, aquaculture will have sufficient fishmeal to 2020 and beyond. It is also predicted that sufficient fish oil will be available to year 2010, although fluctuations caused during El Niño may create temporary shortages. However, beyond this period, there will be a shortfall of marine oil for aquaculture feed which are rich in polyunsaturated fatty acid (PUFA) and invariably the major source of unsaturated fatty acids in compound aqua feeds (NRC, 1993). In view of this notable expectation, the feed industry should look for possible substitutes. The demand for fish

oil could be reduced by using vegetable oil or a blend of fish oil and vegetable oil as a source of unsaturated fatty acids (*n-3* and *n-6*).

As aquaculture technology evolved, the push toward bigger yield and faster growth has involved the enhancement or replacement of natural foods with prepared diets. Aquaculture production techniques can be categorized based on management intensity. The extensive culture technique occurs where natural productivity meets all the nutrient requirements of fish. Semi-intensive culture occurs when ponds are fertilized with either organic or inorganic fertilizers to stimulate natural production or when supplemental feeds are used to increase fish yields. Intensive culture occurs when all the nutritional requirements are met externally through formulated diets. The semi-intensive system is most widespread in Africa as feed requires over 50% of the operating costs of more intensive production systems (Pillay, 1992). Formulated fish diets are expensive and not readily available to small-scale farmers, in contrast to commercial livestock feeds. As a result, in most developing countries, agricultural by-products used as organic inputs to ponds and single ingredient diets offer opportunity to improve fish farming, and these materials may be effectively combined to compliment their various nutritional properties.

Fish yields are determined by several factors that include the quantity and quality of diets. The primary goal in fish farming is to transform dietary protein into fish protein (Jauncey, 1982). Protein sources in fish diets are mainly of two types, animal protein and plant protein. Inclusion of animal protein into fish diets significantly increases

production costs. As proteins are generally too expensive for use in fish feed, except as feed supplements, the focus of attention becomes maximizing the efficiency of low cost plant proteins, and farm and industrial wastes.

Single ingredients are often deficient in one or more of the nutrients required for growth. To overcome the deficiency, ingredients are mixed together to form a compound feed. Considering the high cost of proteins in fish feed formulation, as evident by the rising cost of fish meal, and also the unsustainable utilization of fish oil for fish feed production due to dwindling fisheries harvests, the possibility of using POME could be a case in point hence it has been used as poultry and cattle feed (Vadiveloo, 1987). The appreciable nutrient contained in POME makes its use potentially interesting in the culture of live fish foods and the preparation of fish feed (Habib *et al.*, 1998) and above all its use as low-cost ingredient for fish feed becomes more than a lucrative consideration. The main ultimate goal of this research is therefore, to evaluate the possibility of using micro-filtered dried POME sludge as a feed ingredient for fish feed formulation.

1.2 Problem statement

Aquaculture has traditionally relied on products from industrial fisheries, namely fishmeal and oil to serve the aqua feed industry and the demand on these traditional fish ingredients which are finite global resources are increasing. Therefore, in order to

balance the ever expanding aquaculture production with high energy fish feed, a suitable alternative feed ingredient from POME should be utilized to provide the essential nutrient and energy needed to fuel the growth of aquaculture production.

1.3 The objectives

The followings are the set objectives for the study:

- (i) To evaluated the capability of the dead-end configured hydrophilic micro-filtration membrane as nutrient recovery system.
- (ii) To evaluate the nutritive value of POME and its substitution as ingredient in Nile Tilapia (*Oreochromis niloticus*) diet.
- (iii) To evaluate food conversion ratios (utilization efficiency) and protein efficiency ratios as parameters used to determine the value of feeds for providing the necessary energy for growth.
- (iv) Evaluate the fatty acids profile in muscle of fish fed the POME sludge diet.

1.4 Scope of investigation

The study was confined to the following scopes:

- (i) The study focused on pre-filtration of the raw POME by sieving with ordinary sieve average mesh size 125mm to remove coarse solids found in the suspension
- (ii) The study evaluated and screened the capability of the dead-end configured hydrophilic micro-filtration membrane with respect to resource recovery and filtrate quality.
- (iii) This study concentrated on conducting feeding trial on Nile Tilapia fish (*Oreochromis niloticus*) trial using the formulated and prepared POME diet as feed ingredient source.

1.5 Hypothesis

The use of microfiltered POME sludge in fish feed gives a good knowledge about its nutritional value and the subsequent utilization of the sludge in diet enhance Nile tilapia growth performance.

CHAPTER TWO

LITERATURE REVIEW

2.1 Palm Oil Mill Effluent (POME)

2.1.1 Introduction

Over the last 30 years, Malaysian palm oil industry has grown and at present it is one of the largest agro-based industries (Wong *et al.*, 2002). The country's palm oil accounted for about 52% of the world palm oil production and the industry has contributed to the export earning of the country in the tune of RM 13 billion. The increase in cultivation of oil palm and production of palm oil is contributing to the growth of the nation's economy, however, the practices has negative impact on the environment. The palm oil mills are large generators of organic wastes, often mismanaged; the disposal of the wastes has posed a major problem which therefore calls for appropriate measures. The palm oil extraction process involves use of water, and a large quantity is required to steam and sterilize the palm fresh fruit bunches (FFB) and as well clarify the extracted oil. It is estimated that for 1 ton crude palm oil (CPO) produced, 5-7.5 tons of water are required, and more than 50% of the water will end up as palm oil mill effluent (POME) (Ahmad *et al.*, 2003). Sterilization and clarification are two main stages in palm oil mill which produces condensate and clarification sludge respectively basically form wastewater otherwise known as palm oil mill effluent.

The clarification sludge shows higher level of solid residues compared to the sterilizer condensate. However, both contain some level of un-recovered oils and fats. POME is a colloidal suspension of 95-96% water, 0.6-0.7% oil, and 4-5% total solid (TS) including 2-4% suspended solid (SS) originating from the mixing of sterilizer condensate, separator sludge and hydro-cyclone wastewater (Table 2.1). For a well-controlled conventional mills about 0.9, 0.1 and 1.5 m³ of sterilizer condensate, separator sludge and hydro-cyclone waste respectively are generated for each ton of crude palm oil produced. In most mills, these three wastewater streams are combined together resulting in viscous brown liquid containing fine suspended solids. POME is acidic (pH 4-5), has discharge temperature of 80-90°C /50-60°C and non toxic (since no chemicals are added during extraction), (Ahmad *et al.*, 2003). The raw POME has an extremely high content of degradable organic matter, which is due in part to the presence of un-recovered palm oil. Apart from the organic composition, POME is also rich in mineral content, particularly phosphorus, potassium, magnesium and calcium (Okwute & Isu, 2007, Ahmad *et al.*, 2003). Thus most of the dewatered POME (sludge) can be recycled or returned to the plantation as fertilizer, however, the application as fertilizer has to be carried out carefully, as overdose will result in nutrient imbalance and lead to undesirable chemical reaction in the soil, a prolonged inadequate utilization of POME may cause the accumulation of magnesium and inhibit the availability of

potassium in the soil. It was indicated that the utilization of POME is limited by the potassium (or magnesium) value (Kittikun *et al.*, 2000).

Table 2.1 Characteristic Properties of Palm Oil Mill Effluent

Parameters	Sterilizer effluent (g/L)	Hydrocyclone effluent (g/L)	Centrifuge effluent (g/L)	Mixed effluent (g/L)
BOD	10-25	-	17-35	11-30
COD	30-60	-	40-75	30-70
TS	40-50	5-15	35-70	30-65
SS	3-5	5-12	12-18	9-25
Oil	2-3	1-5	5-15	5-13
A-N	0.02-0.08	-	0.02-0.08	0.02-0.08
TN	0.35-0.60	0.07-0.15	0.5-0.9	0.5-0.9
pH	4.5-5.5	-	3.5-4.5	3.5-4.5

Source: Borja *et al.*, 1995

The POME is an important source of inland water pollution (due to oxygen depletion and other related effects) when released into local rivers or lakes without treatment. POME contains high chemical oxygen demand (COD) and biochemical oxygen demand (BOD), with COD values greater than 80,000mg l⁻¹ frequently reported. A study showed that incomplete extraction of palm oil from the palm fruits can increase COD values substantially (Oswal *et al.*, 2002). The contaminants present in POME wastewaters are mostly organic in nature. Detailed analysis for each component is either useful or

practically possible; as such most of the analyses give an overall measure of the degree of contamination present

The choice of POME wastewater treatment systems is largely influenced by the cost of operation and maintenance, availability of land, and the location of the mill. The evaluation of any effluent treatment system depend both on the actual load of the effluent discharge (in addition to the concentration) and the capacity and nature of the ultimate receiving body. In Malaysia, the final discharge of the treated POME must follow the standard set by the Department of Environment (DOE) of Malaysia. Every proponent therefore requires a license with which to operate and to ensure high quality effluent discharge in accordance to the DOE regulation (Table 2.2)

Table 2.2 Environmental Regulations for Palm Oil Mill Effluent (POME) Discharge Limits

Parameters	Concentration (mg/L) except for pH value
Biochemical Oxygen Demand (BOD)	100
Chemical Oxygen Demand (COD)	-
Oil and grease (O&G)	50
Suspended Solids (SS)	400
Total nitrogen (TN)	150
Ammoniacal nitrogen (AN)	150
pH	5-9

Source: Abdul Latif *et al*, (2003)

2.1.2 Nutritional Value

POME is a resource that can produce other valuable products. It's sludge contains 11.1% crude protein, 12% crude lipids (ether extract), 17% crude fibre, 9% ash and 50.5% carbohydrates (N-free extract) on a dry matter basis, making it a useful substrate for the production of single-cell protein for feeding farm animals(Wan Zakari & Alimon, 2004).

Several attempts have been made to utilize these effluents, among which are: (a) direct use of the sludge as land fertilizer, and (b) utilization of the effluent as animal feed (Hutagalung *et al.*, 1975). Related report by Habib *et al.*, 1998 showed that POME contains a large amount of nutrients (Table 2.3), the amino and fatty acids, and has gross energy content of about 454Kcal/100g, which is comparable to that of maize (500 Kcal /100g) (Okiy, 1987). The potential of its utilization as a feedstock for bioconversion, as feed ingredient and as well for growth of aquatic plants and organisms is vast hence the substantial amount of organics (Okiy, 1987; Phang, 1990; Yusoff *et al.*, 1996).

Table 2.3 Proximate Composition (g/100g dry sample) of Raw POME.

Proximate Composition of Raw POME	Content (%)
Moisture	6.76
Crude protein	9.07
Crude lipid	13.21
Carbohydrate	32.12
Ash	20.55
NFE	19.47

Source: Habib *et al.*, 1998

Besides the organics, the effluent is a rich source of inorganic as where residual liquor from any bioconversion process may possibly be used as a fertilizer. Partially treated effluent is commonly used for irrigation in the oil palm estate and it has been shown to reduce the use of inorganic fertilizers. Since no chemicals are added in the milling process, the waste produced is biodegradable.

The application of microbial and enzymatic processes for converting the wastes to useful products have been studied most extensively by various workers (Vairappan & Yen, 2008, Alam *et al.*, 2007, Wu *et al.*, 2006).

Besides the production of biogas by Whiting (1983), POME has been subjected to fermentation to upgrade its nutritive value, especially the protein levels. Barker *et al.*, (1981), determined that *Aspergillus oryzae* grown on palm effluent have a high crude protein suitable as a dietary protein for non ruminant animals. The report of Habib *et al.*, (1998), also showed that all of the essential and other seven nonessential amino

acids were present in POME (Table 2.4). The POME was found to contain high amount of essential amino acids such as phenylalanine, methionine, leucine and lysine and low amount of histidine and tyrosine. The non essential amino acids such as aspartic and glutamic acids, serine, glycine and cystine were also found to be high in POME.

Table 2.4 Amino Acids (g/100 protein) content of Raw POME

Amino acids contained in raw POME	Quantity (g)
Aspartic acid	9.66 \pm 0.19
Glutamic acid	10.88 \pm 0.21
Serine	6.86 \pm 0.15
Glycine	9.43 \pm 0.17
Histidine *	1.43 \pm 0.04
Arginine *	4.15 \pm 0.10
Threonine*	2.58 \pm 0.05
Alanine	7.70 \pm 0.16
Proline	4.57 \pm 0.10
Tyrosine *	3.26 \pm 0.06
Phenylalanine *	3.20 \pm 0.07
Valine*	3.56 \pm 0.06
Methionine*	6.88 \pm 0.15
Cystine	3.37 \pm 0.06
Isoleucine*	4.53 \pm 0.11
Leucine*	6.86 \pm 0.15
Lysine*	5.66 \pm 0.14
Tryptophan*	1.26 \pm 0.05

Source: Habib et al., 1998.

Means (\pm SE), n=9 of (* essential) amino acids

It was also found too that most of the unsaturated fatty acids, including oleic acid (18:1*n*-9), linoleic (18:2*n*-6), linolenic acid (18:3*n*-3), arachidonic acid (20:4*n*-6) and

eicosapentaenoic acid (20:5n-3) were contained in POME in high amount (Habib *et al.*, 1998), (Table 2.5). These findings again proved the suitability of raw POME as a good source of micronutrients (Okiy, 1987)

Table 2.5 Fatty acids (g/100g lipid) content of raw POME

Fatty acids contained in raw POME	Quantity(g)
Capric acid(10:0)	2.29 ± 0.03
Lauric acid(20:0)	9.22 ± 0.03
Myristic acid(14:0)	12.66 ± 0.11
Palmitic acid(16:0)	14.45 ± 0.12
Heptadecanoic(17:0)	1.39 ± 0.02
10 Heptadecanoic(17:1)*	1.12 ± 0.02
Stearic acid (18:0)	11.41 ± 0.08
Oleic acid (18:1n-9)*	8.54 ± 0.06
Linoleic acid(18:2n-6)*	9.53 ± 0.05
Linolenic acid(18:3n-3)**	4.72 ± 0.04
Arachidic acid(20:0)	7.56 ± 0.03
Eicosatrienoic acid(20:3n-6)**	1.49 ± 0.02
Arachidonic acid (20:4n-6)**	1.12 ± 0.03
Eicosapentaenoic acid(20:5n-3)**	0.36 ± 0.02
Behenic acid(22:0)	2.620 ± 0.03

Source: Habib et al., 1998.

Means (±SE), n=9 of (*unsaturated and **polyunsaturated) fatty acids

2.1.3 Utilization of POME Fish feed.

The palm oil manufacturing processes generate wastes which are either solid matter residuals or waterborne residuals. These residuals are often utilized as a beneficial by-product in the co-production of animal feeds (Wan Zakari & Alimon, 2004). The waterborne residuals (i.e. dissolved and particulate matter) are generally treated in a wastewater treatment process and further processed into valuable products for use in the farm animal production, and in this farming system, food destined for animals such as fish, birds, and other livestock usually consists of raw ingredients or food additives that may include whole, unprocessed food materials, marginally processed foods (e.g. fishmeal, and soy meal), and waste products generated in the production of other food.

An appreciable number of food materials are identified in aquaculture as feed; however, a variety of other alternative food additives may be employed to provide nutrition to animals. Often, the motivation for employing alternative food additives in feed formulations is to reduce the cost of the protein component. Against this background, researchers have examined the use of vegetable products (such as soy) and monocultures (or well characterized mixed communities) of single-cell (i.e., microbial) protein sources as a primary food additive. These microorganisms are grown on substrates including natural gas. Additional organisms that may be incorporated into animal food include algae, yeast, and zooplankton.

An extreme example for an alternative animal food is found in some developing-world aquaculture operations where the faces of pigs, ducks, cows, and other animals have been utilized as a feed in order to recover the nutritional value remaining in these waste products. Fish meal provides a common source of protein for animal food, particularly in the aquaculture, pig, poultry, and pet food industries. However, concerns abound regarding the use of fish meal as a food additive. Overall, worldwide consumption of fish meal was approximately seven million tons in 2001 and the magnitude of this demand for fish meal has led to concerns about depleting wild fish stocks due to over-fishing. These fears are further heightened by seasonal fluctuations and meteorological events (e.g El Nino, La Nina) that influence market prices for fish meal.

In view of this, efforts were made and developed with a desire to coordinate wastewater treatment with aquaculture operations in order to take advantage of the high-quality water achieved as a result of biological treatment and the use of biosolids obtained from treatment as feed ingredients. However, this desire exposed the potential difficulties found in delivering large quantities of high-quality fish food to operations in potentially impoverished regions. Trials have begun for utilization of Palm Oil Mill Effluent (POME) as component in compound cattle, poultry and pig feeds, this research is therefore intended to benefit from the nutritional values in the POME and utilize them as fish feed ingredient.

2.1.4 Aquaculture

Aquaculture could be defined as the art of cultivating the natural produce of water; the raising or fattening of fish in enclosed ponds; and also rearing of aquatic organisms under controlled conditions (De Silva & Anderson, 1995). Researchers such as Landau (1992) and Meade (1989) also defined aquaculture as the large-scale husbandry or rearing of aquatic organisms for commercial purposes and as the practice of rearing, growing or producing products in water or in managed water systems respectively. However, the internationally accepted definition of aquaculture is by the FAO of the United Nations Organization (UNO) which is the farming of aquatic organisms including fish, mollusks, crustaceans and aquatic plants.

Over the last decade, the world has witnessed spectacular growth in the aquaculture industries of many developing countries. It is unequivocally agreed that global aquaculture production will continue to increase, and much of this will occur in the developing countries of Asia and Africa, through the expansion of semi-intensive, small-scale pond aquaculture. Nutrition and feeding play a central and essential role in the sustained development of aquaculture and, therefore, fertilizers and feed resources continue to dominate aquaculture needs. Aquaculture development is also confronted with the choice between using established culture of herbivorous/omnivorous species under extensive or semi-intensive systems or developing more intensive systems to meet increasing production demands. Similarly, issues and conflicts, such as the demand for food verses availability of marine resources, productivity verses

environmental quality, and choice of species verses biodiversity, warrant critical examination.

Growth, health and reproduction of fish and other aquatic animals are primarily dependent upon an adequate supply of nutrient, both in terms of quantity and quality, irrespective of the culture system in which they are grown. Supply of inputs (feeds, fertilizers etc.) has to be ensured so that the nutrients and energy requirements of the species under cultivation are met and the production goals of the system are achieved (De Silva & Anderson, 1995). Complete data on nutrient requirements are only available for a limited number of species. Although dietary protein and lipid requirements and carbohydrate utilization are relatively well investigated for several fish and shrimp species, data on the requirements of micronutrients such as amino acids, fatty acids and minerals are only available for the most commonly cultivated carnivorous and selected omnivorous fish species (Francis-Floyd, 2002). Improvement of feed and nutrition in aquaculture may give us the opportunity to further improve the nutritional quality and benefits of the fish consumed. Nutritional value, colour and appearance, smell and taste, texture and storing capacity may all be affected by the quality of nutrition, however, feed provided during culture production has expanded at a rate of 15% per year and is predicted to continue to grow at this rate for at least the next decade (Barlow, 2000). Tables 2.6 and 2.7;