

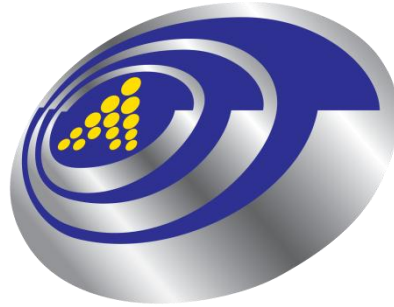
**PHYSICAL TESTING ON FORMULATED  
EARTHWORM-BASED PELLETS FOR GROWTH  
PERFORMANCE EVALUATION IN AFRICAN  
CATFISH (*CLARIAS GARIEPINUS*)**

KULAAB A/P LIAM

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2013



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**Physical Testing on Formulated Earthworm-based  
Pellets for Growth Performance Evaluation in African  
Catfish (*Clarias gariepinus*)**

by

**Kulaab A/P Liam**

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A thesis submitted in fulfillment of the requirements for the degree of  
Master of Science (Bioprocess Engineering)

**School of Bioprocess Engineering  
UNIVERSITI MALAYSIA PERLIS**

2013

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## LIST OF ABBREVIATIONS

ppt	Part per thousand
kg	kilogram
N	Nitrogen
P <sub>2</sub> O <sub>5</sub>	Phosphorus pentoxide
K <sub>2</sub> O	Potassium oxide
NG	Not given
psig	Pounds per square inches gauges
ME	Metabolize energy
µm	Micrometer
m <sup>2</sup> /kg	Meter square per kilogram
DP	Degree of polymerization
d.s.	Dry substances
RH	Relative humidity
OFAT	One-Factor-At-One-Time
JMP Software	JMP software from SAS Institute, USA (Version 10)
Na <sub>2</sub> CO <sub>3</sub>	Sodium carbonate
NaOH	Sodium hydroxide
CuSO <sub>4</sub> .5H <sub>2</sub> O	Copper sulphate solution
BSA	Bovine Serum Albumin
UV-Vis	Ultraviolet Visible Spectroscopy
CCD	Central Composite Design
RSM	Response Surface Methodology
N	Newton
ANOVA	Analysis of Variance
HCl	Hydrochloric acid
GE	Gross energy
ADC	Apparent digestibility coefficients
AOAC	Association of Analytical Communities

HNO <sub>3</sub>	Nitric acid
K <sub>2</sub> CrO <sub>4</sub>	Potassium chromate
ppm	Part per million
Cr <sub>2</sub> O <sub>3</sub>	Chromium (III) oxide
Cr	Chromium
n	One part weight (% wt)
m	Fraction of ratio
DF	Degree of freedom
AAS	Atomic absorption spectroscopy

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## LIST OF SYMBOLS

Symbol		Unit
a	Heating time	minutes
b	Heating temperature	°C
$\alpha$	Significance level	
H <sub>0</sub>	Hypothesis null	
$\mu_s$	Means of heated earthworm-based pellets	
$\mu_u$	Means of unheated earthworm-based pellets	
H <sub>A</sub>	Alternative hypothesis	
U	Unheated earthworm-based pellets	
S	Heated earthworm-based pellets	
C	Commercial pellets	
FE	Feces of African catfish fed by earthworm-based pellets	
FC	Feces of African catfish fed by commercial pellets	
EP	Modified earthworm-based pellets	
CP	Modified commercial pellets	



## **Kajian Fizikal Makanan Ikan yang Diformulasi Berasaskan Tepung Cacing Tanah untuk Penilaian Kadar Pertumbuhan pada Ikan Keli Afrika (*Clarias gariepinus*)**

### **ABSTRAK**

Formulasi baru pellet ikan telah direka dengan menggunakan tepung cacing tanah dan bahan-bahan yang menjimatkan seperti dedak padi dan kanji ubi kayu sebagai bahan alternatif untuk menggantikan tepung ikan yang berkurangan. Formulasi ini dikira menggunakan kaedah Pearson's square dengan membahagikan bahan-bahan kepada kumpulan protein dan kumpulan tenaga dengan meletakkan bahan yang paling murah sebagai bahan yang mempunyai nisbah tertinggi. Pengoptimuman proses pemanasan dijalankan menggunakan kaedah One-Factor-At-One-Time (OFAT) untuk mengoptimumkan masa dan suhu pemanasan untuk mendapatkan kestabilan air yang tertinggi. Ujikaji kestabilan pellets dalam air (82.14%) telah dicapai pada suhu 80°C dalam masa 40 minit. Ciri-ciri fizikal pelet seperti ujian rendaman, ujian larutan protein dan ujian kekerasan untuk memastikan kebolehan pelet untuk diaplikasikan pada eksperimen ikan. Keupayaan pellet dinilai dengan menentukan prestasi pertumbuhan ikan keli (16%) yang diberi makan dengan pellet berasaskan cacing tanah yang dipanaskan. Ujikaji ikan dijalankan selama 8 minggu dengan menyukat kenaikan berat dan panjang setiap minggu dan dibandingkan dengan pelet komersial. Kadar pertumbuhan ikan keli Afrika yang diberi makan pelet berasaskan cacing tanah yang dipanaskan menunjukkan pencapaian berat dan panjang yang lebih tinggi berbanding pelet komersial. Ujian penghadaman jelas telah dijalankan keatas protein, jumlah lemak, tenaga kasar dan bahan kering. Pelet berdasarkan tepung cacing tanah yang dipanaskan dan pelet komersial diubahsuai dengan menambahkan kromium oksida sebagai penanda lengai untuk menentukan pekali bagi penghadaman jelas. Najis daripada eksperimen ikan telah dikumpul pada minggu yang keempat untuk dianalisis. Kebolehadaman protein, jumlah lemak, tenaga and bahan kering untuk pellet ikan berasaskan tepung cacing adalah 94.92%, 98.09%, 78.53% and 77.78% sementara pellet komersial ialah 66.98%, 98.18%, 74.14% dan 71.43% . Keseluruhan kajian ini menunjukkan kadar kestabilan air adalah lebih tinggi dalam pelet berdasarkan tepung cacing tanah yang dipanaskan dan keupayaan untuk menaikkan kadar pertumbuhan yang lebih tinggi berbanding pelet komersial. Pengenalan kepada formulasi baru dan proses pemanasan telah menambahbaik tumbesaran ikan sebanyak 12.48% dari pellet komersial.

## Physical Testing on Formulated Earthworm-based Pellets for Growth Performance Evaluation in African Catfish (*Clarias gariepinus*)

### ABSTRACT

New formulation of fish pellets was designed by using earthworm powder and other economical ingredients such as soybean waste, rice bran and tapioca starch as an alternative protein source for substitution of fishmeal due to the worldwide shortage. The formulation was calculated using Pearson's square method by dividing ingredients into protein sources ingredients and energy sources ingredients with higher ration on low cost ingredients. The optimization of heating process carried out by using One-Factor-at-One-Time (OFAT) based on the highest water stability. The high water stability (82.14%) was achieved when heated at 80°C within 40 minutes. Physical properties of producing pellets such as soaking test, protein leaching test and hardness test were conducted to confirm the applicability of pellets to fish experimental. Pellets potential was evaluated by determining the growth performance of African catfish (*Clarias gariepinus*) fed with heated earthworm-based fish pellets. Fish experimental was conducted for 8 weeks by comparing weekly weight of the fish fed with heated earthworm-based pellets and commercial pellets. The growth performance of heated earthworm-based pellets showed higher weight and length increment compared to existing commercial pellets. Apparent digestibility tests were also performed for protein, fat, energy, and dry matter. Heated earthworm-based pellets and commercial pellets were modified by adding chromic oxide as an inert marker to determine the apparent digestibility coefficient. The feces of experimented catfish were collected at fourth weeks of fish experimental for the analysis. Digestibility of protein, total fat, gross energy and dry matter for earthworm-based pellets are 94.92%, 98.09%, 78.53% and 77.78% while commercial pellets are 66.98%, 98.18%, 74.14% and 71.43% respectively. Introduction of new formulation and heating process had resulted in growth improvement of the fish by 12.48% from commercial pellets.

# CHAPTER 1

## INTRODUCTION

### 1.1 Background

The term aquaculture refers to the cultivation of both marine and freshwater species which can be ranged from land-based to open-ocean production in farming of aquatic organism such as fish, shellfish and plants (Department of Marine Resources, 2006).

The performance of Malaysian aquaculture industry has increased over the years and contributed to the growth of our economy. In the year 2010, the Malaysian aquaculture has recorded an increase of 7.93% as compared to year 2009 (DOF, 2012). Moreover Malaysian agriculture industry is a source of supplier to big market worldwide such as Asia, Australia and United State (Tan, 2011) and it is envisioned that Malaysia will continue to do if greater effort can be made to improve the industry further.

Besides being a source of protein to the population, aquaculture is a potential sector that can improve the national economic performance through global exportation and new job creators (BERNAMA, 2011). Hitherto Malaysian aquaculture industry has carried out many expansion programs to improve export values of local aquaculture through assistance from foreign experts (Sipalan, 2012).

Freshwater aquaculture is also one of the potential sub-sectors of aquaculture industry. One of the famous freshwater species is catfish. Many of the local catfish farms are made up of the small and large scale industries developed for local consumption and export market. The Malaysian freshwater aquaculture sector recorded a production of 14,568 metric tons of catfish, leading other freshwater species (DOF, 2004).

## **1.2 Problem statements**

### **1.2.1 Production cost**

Existing fish pellets normally were produced using extrusion or pelletizing method. Extruded fish pellets have higher durability compared to pelletized pellets. However, extrusion machine were expensive and will contribute a higher cost in pellets production. On the other hand, pelletizing machine is cheaper however the pellets quality can be lower than extruded pellets in term of physical properties of pellets. The used of pelletizing method are more economical compared to extrusion. Hence a study of the pelletizing method to improve the physical properties of the pelletized pellets is required.

In Malaysia, there are only a few company that produce fish pellets for local supply. Most of the pellets were imported from several countries such as Thailand and China. High market demand and insufficient production of pellets cause the increasing price of both local and exported pellets. The high cost pellets effected the overall production of catfish especially small scale sector. An affordable production method is necessary to cover the needs of fish pellets.

### **1.2.2 Existing fish feed issues**

Catfish farmers always encountered problems in finding new protein sources for catfish feeding. The main protein source used in the current formulated fish pellets is fishmeal which is not only cheap but contains high protein. Most of the commercial fish pellets used fishmeal as the main ingredient. However, production of fishmeal may vary between countries. The increasing usage of fishmeal has led to the shortage of fishmeal

supply in the longer term. Hereby, new protein source is necessary to overcome fishmeal shortages. New formulation of fish feed using new protein sources is required to overcome the problem. The new source of protein can be used as replacement or in combination to fishmeal in fish pellets production. One of the protein sources is earthworm powder which can easily provided. Moreover, the used of earthworm powder in the formulation will increase the utilization of earthworm powder that was produced locally. Earthworm is a proven good source of protein for fish as many studies were conducted using earthworm as an ingredients for fish feed production. High feed conversion ratio (FCR) in earthworm powder and have been used in several fish diet production.

### **1.3 Research objectives**

The objectives of this research are itemized below:

- i. To develop a new formulation of earthworm-based pellets using earthworm powder and other agriculture waste by Pearson's square method
- ii. To optimize the heating time and temperature in the production of earthworm-based pellets
- iii. To evaluate the potential of heating process by comparing physical properties and proximate analysis of earthworm-based and commercial pellets
- iv. To study the growth performance and digestibility of catfish fed by heated earthworm-based pellets

#### **1.4 Scope of research**

The research is therefore focuses on creating new formulation of earthworm-based pellets by using earthworm powder, soybean wastes, rice bran, fishmeal, and tapioca flour. The formulation also used only a few types of ingredients compared to existing fish pellets which used more than six ingredients. The Pearson's Square Method was performed to calculate the formulation. Since the protein level and ratio of each ingredient can be fixed, there is only one formulation of fish pellets need in this research. The optimum time and temperature of heating process were determined. Physical test of pellets was conducted to confirm applicability of the pellets. The pellets were fed to African catfish to study the growth performance and digestibility compared with commercial pellets.

#### **1.5 Dissertation organization**

This thesis contains of six chapters with chapter one provided brief background about the study, problem statement, scope and objective of research, and methodology outline. Chapter two give details about previous research related to this study, the pros and contras of existing fish pellets and methodology. Chapter three explains about materials and methods adopted in the execution of the research. Results and discussion covered in Chapter four while chapter five contains conclusions and recommendation for further study.

## CHAPTER 2

### LITERATURE REVIEW

#### 2.1 Catfish

Catfish is one of the freshwater species that have high market demand in the country. Catfish is mostly found in freshwater environment with variety of shapes depending on their species. The ray-finned fish differs physically from the others. Among the popular catfish species that have been farmed are *Clarias macrocephalus*, *Clarias batrachus* and *Clarias gariepinus* (Peteri, Nandi & Chowdhury, 1992).

##### 2.1.1 *Clarias gariepinus*

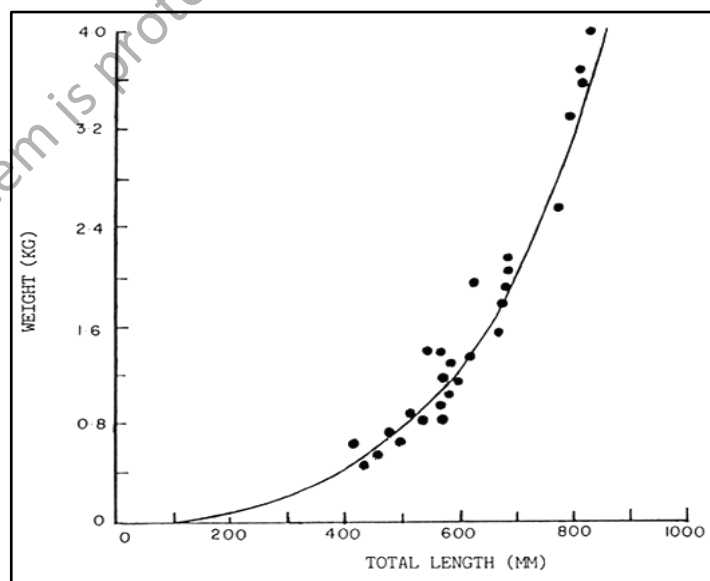
*Clarias gariepinus* (Figure 2.1) or commonly known as African catfish is one of the popular freshwater species that have been commercialized in many countries in the world. The African catfish is also well known on its ability to grow fast and very robust, a trait which is very suitable for tropical country. The high resistance to changing water conditions and the ability to tolerate different kind of feeds are also some of the advantages of African catfish.

African catfish have a scaleless skin with dark dorsal and lateral body surface with grayish-white at ventral side. Fish coloration may be influenced by light (color of water) or water turbidity. Black spots will appear when the African catfish are in stress condition. African catfish will ideally develop at temperature 25°C to 30°C with maximum salinity tolerance of 9.6 ppt for fingerlings and higher for adults. After the development of

accessory respiratory system organ, the fish can survive in extreme environment; even can survive in brackish water (Peteri et al., 1992).



Figure 2.1: A market size of African catfish



Source: Clay (1981)

Figure 2.2: Body length-weight relationship of catfish