

EVALUATION OF MAYFLIES, *Povilla adusta* Navas, AS
ALTERNATIVE PROTEIN SOURCE IN FEED FOR IMPROVED
GROWTH OF NILE TILAPIA, *Oreochromis niloticus*, REARED IN
EXPERIMENTAL TANKS

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partial fulfilment of the requirements for the award of a Bachelor
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Declaration

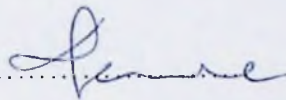
I, Nsega Monic, declare that this report is my original work and has never been submitted to any university or institution for award of a degree or any other qualification.

Signed 

Date 30th June 2006

SUPERVISOR

This work was done under my supervision and has been submitted to the Faculty of Agriculture, Busoga University with my consent

Signed 

Date 30.6.2006

Dr Tamale Andrew (BVM MUK; MPVM (cand) MUK)

Dedication

To my dear parents: Late Safatia Nsega and Miria Namulondo

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List of Abbreviations and Acronyms

g		grams
cm		centimetres
mm		millimetres
L		litres
mg/l		milligrams per litre
µs/cm		microsiemens per centimetre
SD		Standard Deviation
°C		degrees celcius
N		North
E		East
n		number
FIRRI		Fisheries Resources Research Institute
FTI		Fisheries Training Institute
NAADS		National Agricultural Advisory Services
MAAIF		Ministry of Agriculture, Animal Industry and Fisheries

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Definitions of terms

Optimum growth	Highest growth that can be attained with proper feeding using balanced feeds and good management practices
Quality feeds	Formulated balanced diets containing all required nutrient in proper proportions
Stocking	Introducing fish into tanks
Stocking density	Numbers of fish per unit volume of water in tank
Stress	Abnormal physiological condition of fish resulting when its collective adaptive responses to environmental factors are extended to or approaching the fish's limit of tolerance for that factor

Abstract

Supplementary feeds, either single or multi-ingredient, are used to improve fish yields in fish farming systems. Protein-rich ingredients are highly recommended for better performance (growth) of fish. In Uganda, due to high expense of ingredients rich in proteins for example "mukene" (*Rastrineobola argentea*), and the lack of suitable alternatives, farmers tend to use carbohydrate-rich maize bran, a cheap ingredient leading to low farm yield.

The major objective of the study was to evaluate *P. adusta* as alternative protein source in feed for Nile tilapia (*Oreochromis niloticus*). Specific objectives were: to determine and compare nutrient content in "mukene", *P. adusta*, and maize bran; to monitor and compare (weekly) weight and length gains of *O. niloticus* fed on three formulated experimental diets: i) 40% protein diet composed of "mukene" and maize bran; ii) 40% protein diet composed of *P. adusta* and maize bran, iii) maize bran only. Observations were also made on the behaviour of *O. niloticus* fed on the different diets. Experimental glass tanks were used for the study over a 10-weeks period.

Results indicated 64.5%, 46.4% and 8.8% crude protein in "mukene", *P. adusta*, and maize bran respectively. Total carbohydrate values were 33.5% in maize bran, 7.0% in *P. adusta* and 4.8% in "mukene". Lipids constituted 13.4%, 9.5% and 7.9% in "mukene", *P. adusta*, and maize bran respectively. Fish were successfully conditioned to the diets. Fastest conditioning was in fish fed on "mukene" mixed with maize bran, followed by those fed on a mixture of *P. adusta* and maize bran. Results of SPSS One-Way ANOVA at 95% Confidence Interval indicated significant differences in weight gain. Coefficient of Correlation was strongest in a mixture of "mukene" and maize bran (gained 11g) followed by *P. adusta* and maize bran (gained 7g) and lastly maize bran only (gained 2g), with R values of 0.77, 0.52, and 0.48 respectively. Therefore, *P. adusta* can be efficiently utilised as an alternative protein source.

CHAPTER ONE

1.0. INTRODUCTION

1.1. Background

Fish farming is a controlled and managed enterprise that allows the culture and rearing of selected fish species at both subsistence and commercial scales. Provided there is a source of water, fish farming can be practised even in areas where there are no lakes or rivers as is practised in many parts of Israel. Apart from water, other requirements for successful fish farming are appropriate management practices just like applied in agriculture.

Fish farming was introduced in Uganda in 1953 when the Kajjansi Experimental Fish Farm was established to promote aquaculture in the country as a means of supplementing wild stocks in capture fisheries and to increase protein intake for people (Biribonwoha, 1976). Since its introduction, fish farming has been developing slowly at small-scale level. By 1960, there were about 11,000 ponds covering approximately 420 ha with an estimated fish production of 400 – 500 tons, mainly tilapia and carp. Fish yield dropped to less than 100 tons during 1970s to mid-1980s but afterwards increased to 285 tons in 1999, to 2300 tons in 2002, and later 5,200 tons in 2004. By 2004, 95% of about 20,000 fish ponds owned by 12,000 farmers were at subsistence scale, with an average size of 200 m², and only 5% were developing towards commercial scale (Mwanja, 2005).

Major constraints to increased fish farming in Uganda have been attributed to lack of adequate and appropriate fish feeds; inadequate supply of quality seed (fry); inadequate extension services to farmers; poor pond management practices leading to, for example, prolific breeding of Nile tilapia; lack of investment credit schemes; and marketing of cultured fish. Among these constraints, lack of quality feeds has been cited as the most important (Lojjo, 2004). The main issue to consider in quality fish feeds is the protein content and its conversion into fish growth.

There are several factors contributing to lack of quality feeds. Supplementary feeds are costly constituting about 60% of the total expense in a production cycle (Lojjo, 2004), and yet locally available cheap feeds may not be rich in proteins needed for fast growth. Due to high costs of quality (formulated) feeds, many farmers do not routinely feed fish and when fed the quality and quantity of feed is largely inappropriate to enhance proper fish growth. Consequently, stunted fish growth and poor pond yields have discouraged farmers. There is thus need to identify protein sources that should be readily available to fish farmers.

In Uganda, "mukene" meal, a type of fish meal prepared from dried *Rastrineobola argentea* (Pisces: CYPRINIDAE), is the major source of protein in animal feeds for livestock and poultry. "Mukene" is also used as a major protein ingredient in formulated commercial fish feeds. However, the increasing demand for "mukene" for human consumption coupled with rising costs and competition from other livestock feed manufacturers, creates uncertainty about its availability as a major source of protein in fish feeds. Imported fish feeds are more expensive and rarely readily available especially for small scale fish farmers. There is therefore need to explore alternative protein sources for use in fish feeds. Alternative sources of protein for fish feeds in Uganda include Soya, other fishes, *Caridina*, blood meal and lake flies especially mayflies and particularly *Povilla adusta*.

Mayflies, also known as shadflies, belong to the Order Ephemeroptera, with many genera including *Ephemera*, *Hexagenia*, *Ephoron* and *Povilla* (Penak, 1978; Cobert, 1957). *P. adusta* is the most common species observed in the Lake Victoria region (Cobert, 1957). The short-lived adults emerge from Lake Victoria in such large numbers that they are a nuisance, as they invade business and residential places (especially where there is light at night), and accumulate in large swams, as commonly observed in Entebbe, Jinja near lake Victoria, and many fish landings around lakes.

Dried *P. adusta* have an average size of about 1.0 -1.5 cm long; each weighing about 0.005 g. They exhibit seasonal lunar emergence (Hartland- Rowe, 1955; Corbert, 1957), and appear every month shortly after full moon (Hartland- Rowe, 1955), with the main swarm appearing over first three days of full darkness although smaller numbers can emerge during two to three other nights before or just after the main swarm (Corbert, 1957). Pennak (1978) observed variations in seasonal emergence patterns where adults of some species may be collected almost daily for two or three months while other species can emerge only during two or three successive days. Although lake-flies, particularly mayflies, have been known to be rich in proteins and can be eaten by people and processed into animal feeds (Odhiambo, 1978; Kibwika, 2005), this non-conventional resource is still under-utilised and has not been considered as a protein source in fish feeds. In the natural environment, *P. adusta* is a significant dietary item especially for sub-adult Nile tilapia (Baliirwa, 1998). This study investigated the viability of utilisation of *P. adusta* as fish feed and their potential to improve farmed fish production.

1.2. Problem Statement

Uganda has a potential to produce an estimated capacity of about 300,000 metric tons of fish annually from capture fisheries but the actual production has been declining and the maximum catches ever realised were 245,000 metric tonnes in 1990 (National Fisheries Policy, 2004). Decreasing catches from capture fisheries (Muhoozi, 2002) in addition to increasing demand for fish for local consumption and for exports have created a deficit in fish supply. The fish supply gap can be filled by fish farming. Uganda has great potential for fish farming that has not fully been exploited due to technological constraints especially availability of feeds.

Lack of appropriate fish feeds at affordable prices has greatly contributed to the slow pace of development of fish farming, especially commercial fish farming, in Uganda. "Mukene", (*Rastrineobola argentea*), a common fish that would provide the necessary feed quality in terms of protein is also used as feed for livestock,

poultry and human consumption, thus making it expensive. Also, increasing exploitation of "mukene" renders doubt to its availability to support the expanding livestock, poultry, and fish farming industry. Protein quality and palatability of some trials of plant sources (e.g. soybean, cotton seed cake) are inferior (lack adequate levels of essential amino acids e.g lysine and methionine) compared to fish meal (Cesar, 2000; Craig & Helfrich, 2002). In addition, soybean and cotton seed cake also contain indigestible carbohydrates and nutritional factors/toxins i.e tpsin and gossypol respectively. Elimination of the inhibitors requires further processing (e.g. heating) which increases costs. Alternative cheap protein sources that could readily be available have not been identified. The potential of the abundant *P. adusta* as alternative protein sources requires investigation of their nutritional content and acceptability by culture species such as Nile tilapia which is a major farmed fish.

1.3. Significance of the Study

Uganda's strategy is to maintain a per capita fish consumption level of about 10 kg per year within 10 years' time, and this will require an estimated 320,000 tons of fish, excluding the additional recommended maximum export of 60,000 tons allowable per year (National Fisheries Policy, 2004). Sustainable fish production may be achieved through promotion of fish farming, an effort which will not only meet the demands for fish and fish products, but also keep in line with the Plan for Modernisation of Agriculture (PMA) which focuses on reduction of poverty and alleviation of living standard especially for rural communities (National Fisheries Policy, 2004).

Tilapia is one of the most important farmed fishes in the world and in freshwater fish ponds (Reuben, 1983); in the tropics (Collart, 1997); and in Uganda (Agaba, 1999; Baseline survey, 2000). Among the tilapia, *Oreochromis niloticus* is one of the widest cultured species (Nalwanga, 2004).

In Uganda, Nile tilapia is a commercial species second to Nile perch on foreign market and one of the most preferred fish locally. Due to tilapia's high demand

for export, its supply on the local market has been unstable with the effect that generally small-sized tilapia and carcass are now common food fish options for local consumers. Strategies to increase Nile tilapia farming at low costs would contribute to meeting local consumption needs, improvements in household income, as well as supplement wild stocks and enhance sustainability of Nile tilapia fishery. The most commonly used fish feeds for Nile tilapia are "mukene" in maize bran, and maize bran.

1.4. Objective

The overall objective of the study was to assess the relative importance of *Povilla adusta*, as an alternative protein source in fish feed for Nile tilapia reared in experimental tanks.

1.4.1. Specific Objectives

- a. To determine and compare nutrient content (Protein, fat, carbohydrates, fibre, ash, calcium, phosphorus) in "mukene", *P. adusta*, and maize bran
- b. To monitor and compare weight gains and total length increase of *O. niloticus* fed on:
 - i. a mixture of "mukene" and maize bran,
 - ii. *P. adusta* and maize bran, and
 - iii. maize bran only.
- c. To make observations on the behaviour of *O. niloticus* fed on:
 - i. a mixture of "mukene" and maize bran,
 - ii. *P. adusta* and maize bran, and
 - iii. maize bran only.

1.5. Scope of the Study

The study was carried out at Fisheries Resources Research Institute (FIRRI) from January 2006 to March 2006 for a period of 10 weeks. Experimental glass tanks, in duplicate, were set under controlled environmental conditions.

1.6. Research Question

Is there any difference in growth (weight gain and increase in total length) of *O. niloticus* fed on:

- (i) a mixture of "mukene" and maize bran,
- (ii) *P. adusta* and maize bran, and
- (iii) maize bran only?

1.7. Hypothesis

H₀: There is no difference in weight gain of *O. niloticus* fed on diets formulated using (i) "mukene" and maize bran, (ii) *P. adusta* and maize bran, and (iii) maize bran only

CHAPTER TWO

2.0. LITERATURE REVIEW

Feeds for farmed fish must be nutritionally complete and balanced but due to their unavailability at low costs, many farmers resort to using cheap ingredients such as cooked or raw maize bran, cassava, or other household food wastes. As proper nutrition is essential to good growth and fish health, Schmittou *et. al.* (1998) suggested the following guidelines for feed quality:

a) Use manufactured floating pellet feeds, b) use only nutritionally complete feeds containing: over 30% protein with complete balanced amino acid composition, addition of vitamin and mineral fish feed supplements fortified with vitamin C and phosphorus, c) Use freshly manufactured feeds and avoid: feeds older than 4 weeks, molded, spoiled or otherwise degraded feed.

It has been difficult for many farmers to adhere to the suggested guidelines due to the high expense for feeds with the above qualities. Although some studies have been done towards formulation and selection of appropriate feed for *O.niloticus*, little has been achieved in developing nutritionally cost effective feeds that cause optimum growth in a short period.

2.1. Feed trials

The performance of fish feed does not only depend on its quality but also on the feeding management (Lojjo, 2004). The quality of the feed is determined by its composition and the extent to which the nutrients are digested and assimilated. Studies by Nalwanga (2004) on feed formulated for Nile tilapia reared in laboratory and fed for 11 weeks on soya-bean, "mukene", a mixture of soya-bean and "mukene", and a mixture of soya-bean, "mukene", maize bran and potato peelings gave average weight gains of 1.32g, 4.41g, 5.87g, and 9.66g respectively. A similar experiment by Agaba, (1999) using Rabbit pellets and cotton seed cake for a period of 8 weeks caused average weight gain of 1.70g and 1.35g respectively. These weight gains could be below optimum.

Fish fed on proteins alone or carbohydrates alone have lower growth rates since the fish spends too much energy digesting the nutrients (Nalwanga, 2004). Proteins are used for fish growth if adequate levels of fats and carbohydrates are present in the diet (Craig & Helfrich, 2002). There is thus need to determine the nutrient composition of candidate feed ingredients prior to formulation of balanced fish feeds.

2.2.0. Nutritional requirements by fish

Artificial feeds are supplied as supplements to provide the required nutrients for improved fish productivity. For optimum growth, fish needs various levels of nutrients (Table 1).

Table 1. Requirements of different nutrients by fish

Nutrient	Percentage	Purpose
Proteins	35 – 42	Body building
Lipids/fats	5 – 8	Energy; Proper functioning of the body
Starch/Carbohydrates	25	Source of energy
Minerals	1 – 3	Body structure; Normal functioning of the body
Vitamins	<1	Protection against diseases

Source: NAADS, 2005

2.2.1. Common feeds used by farmers

Due to the high expense for formulated feeds, many farmers resort to cheaper supplementary feeds such as maize bran, rice bran, household wastes, *Ipomea* (potato leaves), and *Amaranthus* (dodo). Maize bran is the most commonly used. However, maize bran alone is low in protein levels with reported crude protein of 10.9% (Mlay *et al.* 2005) or 6.92% (Aquaculture Extension Manual, 1994) much lower than the recommended protein requirements (Table 2).

Table 2. Protein requirements by fish

Percentage range	Type of fish	Source
35 – 42	Tropical fishes	NAADS, 2005
32 – 38	Tilapia	Craig & Helfrich, 2002
30 – 50	Many fish species	Cesar, 2000
30 – 36	Many warm water fishes	Lovel, 1979 (as reported by Rondolph, 1986)
30 – 50	Tilapia	Tamir, 2005

2.2.2. Importance of proteins

Proteins constitute about 70% dry weight of the organic material in fish tissue and they are important nutrients for fast fish growth (Schmittou, *et al.*, 1998; Lojjo, 2004;) hence a basis in feed formulation (Nalwanga, 2004). Proteins/amino acid deficiency causes a reduction or cessation of growth, and continued elimination of indispensable amino acids eventually causes mortality (George, 1983). Protein requirement by fish varies with species - Carnivorous > omnivores > herbivores (Craig & Helfrich, 2002) and stage of fish growth. As fish grows its protein requirement decreases (Cesar, 2000; Craig & Helfrich, 2002; NAADS, 2005). "Mukene" meal has been the major protein source for most farmed fish species in Uganda because of its high protein content of 59% (Aquaculture Extension Manual, 1994). Fish meal is highly palatable, allows high protein digestibility and has an amino acid profile that closely matches fish dietary requirements (Cesar, 2000).

2.3. Importance of mayflies and other lake flies

Mayflies are a chief food for fish and dried mayflies can be used as fish baits (Meyer, 2005). It has been shown that Nile tilapia in Lake Victoria feeds on aquatic insects including mayflies (Balirwa, 1998). However, its contribution to fish performance especially growth is not clearly known.

Flour made of dried lake flies - Chironomidae, *Chaoborus*, and *Povilla* had protein content of 67g/100g as well as excellent nutritive properties (Bergeron *et.*

al, 1988), giving the lake flies substantial potential for use as an inexpensive high quality dietary constituent. Embury & Gordon, (1924) reported that the proximate chemical composition of natural insects fed on by wild trout was 49% protein, 15-16% lipid, 8% fiber and 10% ash but these potential sources of protein have not been investigated in dried *P. adusta*.

2.4. Feeding habits of *O. niloticus*

O. niloticus is flexible in its trophic characterisation. It is known to practice ontogenetic shift in diet, the young being omnivorous while sub-adults feed on phytoplankton (Nalunkuma, 1998). It has been reported as omnivorous and opportunistic feeder (Bairwa, 1998) while Keenleyside (1991) described it as omnivore with high feeding plasticity. Tilapia can accept a varied natural and artificial feeding (Collart, 1997; Miller 1976) and can be successfully conditioned to diverse feeds under experimental conditions, for example, feeding on rabbit pellets (Agaba, 1999).

2.5. Feed formulation

No single ingredient contains all nutrients needed by fish in their correct proportions, thus the need to mix different ingredients to produce a diet with the required quantities of essential nutrients, even though it is impractical to formulate rations which will exactly meet the requirements of all species. (Lall, 1991). This is because different fish species differ in their essential amino acid requirements which act as the base for dietary composition (Rondolph, 1986; Nalwanga 2004).

2.6. Conclusion

Lack of cost effective feed ingredients that provide protein for fast growth of Nile tilapia has greatly contributed to reduction in farmed fish production. *P. adusta* may be rich in protein but has not been investigated for utilisation in fish feeds. An experiment to evaluate the potential of *P. adusta* as alternative source of protein was considered vital.

CHAPTER THREE

3.0. MATERIALS AND METHODS

3.1. Study Area/station

The experiment was set up in an aquarium positioned at 00° 43.699' N, 038° 06. 216' E at FIRRI located on plot 39/45 Nile Crescent, Jinja, Uganda.

3.2. Study materials and their sources

The principle study materials were three weeks' old Nile tilapia (*O. niloticus*) fingerlings, and three diets formulated from mayflies (*P. adusta*), "mukene" (*R. argentea*) and maize bran obtained after grinding of maize grain (plate 1).

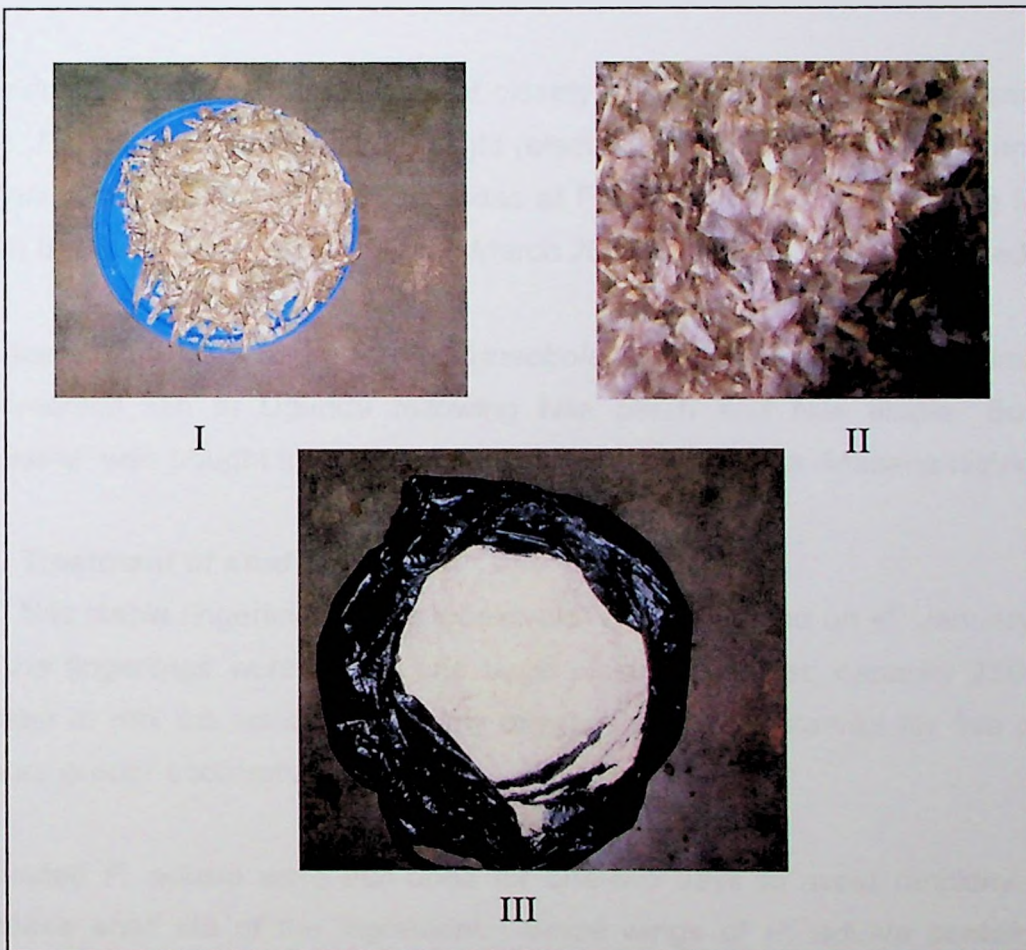


Plate 1: Feed ingredients used (I). "Mukene" (II). *P. adusta* (III) Maize bran in black polythene bag.

O. niloticus belongs to the family CICHLIDAE and genera *Oreochromis*. The fingerlings used in the study were collected from Mr Musomero's fish farm located in Bukanga sub-county, Busalaamu parish, Lukuhnu A village.

Maize bran is a by-product of maize grain milling. There are usually three layers of maize bran: the outermost layer composed of mainly the testa; the middle layer that contains a combination of small particles of maize grain, little maize flour and testa; and a third layer mostly maize flour and a few particles of small maize grain. Farmers consider the third layer most appropriate for fish consumption (Musomero, 2005) probably because of its fine texture, and so it was chosen for this study. The maize bran was bought from Jinja maize mill operators.

P. adusta is a type of lakeflies that closely resemble white ants, but smaller in size. *P. adusta* were attracted by light (electricity) from overhead bulbs and were collected (by sweeping) from verandas at FIRRI and Fisheries Training Institute (FTI) between December 2005 and March 2006 on the days they emerged.

"Mukene" is a local name for *Rastrineobola argentea*, the third most important commercial fish in Uganda following Nile perch and Nile tilapia. Sun-dried "mukene" was bought from Lambu landing on Lake Victoria (Masaka district).

3.3. Treatment of study materials

250 Nile tilapia fingerlings (three weeks old) were collected on 4th January 2006. All the fingerlings were put in one large plastic container, capacity 250L, with aerator to mix the water and supply oxygen, and were starved for five days to ensure proper acclimatisation.

Collected *P. adusta* were sun-dried for one-two days to avoid rancidity and to increase shelf life of the ingredient. Since wings of *P. adusta* contain chitin (Entomology Fact sheet, 1997) whose digestibility by fish is not clearly known, most of the wings were blown off (Plate 2).



Plate 2. Processing (blowing off wings) of *P. adusta*

Both “mukene” and *P. adusta* were processed into powder form using mortar and pestle (Plate 3a). A 1 mm mesh was used to sieve the three ingredients to reduce particle size that can be eaten by the small fish and to increase digestibility (Schmittou *et al.*, 1998) (Plate 3b). The feeds were stored in sealed containers to avoid contamination with moisture.



(a)



(b)

Plate 3. Processing of feed (a) pounding (b) sieving

3.4. Experimental Design

Two sets of glass tanks: set one and set two of different sizes were available. Each set comprised of three glass tanks A1, B1, C1; and A2, B2, C2 in set one and set two respectively (Fig. 3.1).

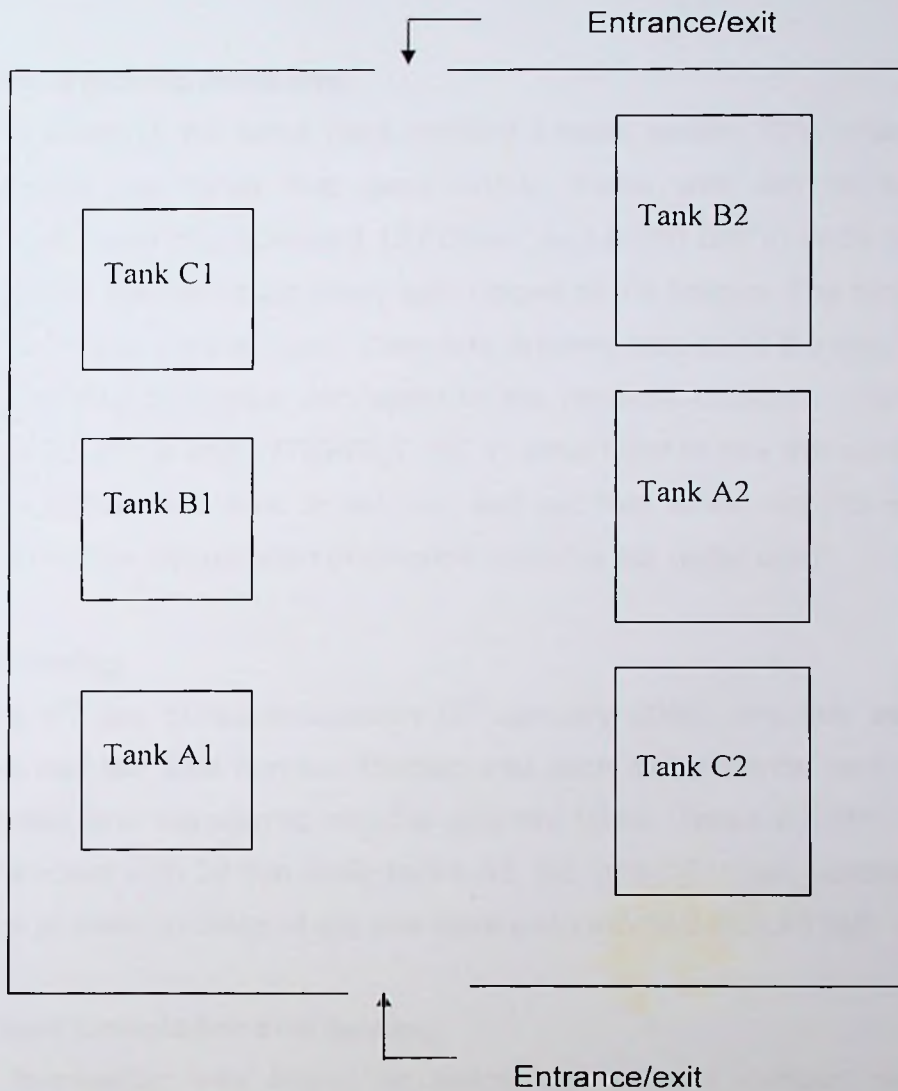


Fig. 3.1. A sketch showing the general lay out of the experimental set up in the aquarium room

Tanks in set one (A1, B1, and C1) measured (60 cm x 29 cm x 38 cm) each and contained approximately 50.4L of water. Tanks in set two (A2, B2 and C2) measured (90 cm x 35 cm x 45 cm) and contained twice the volume of water in set one i.e. about 100.8L of water in each tank. These were arranged as shown in Figure 3.1.

3.5. Pre- stocking measures

Prior to stocking, the tanks were cleaned a week earlier. 70% ethanol was used to disinfect the tanks that were finally rinsed with tap water. Previously disinfected sand that occupied 12,600cm³ and 8,700 cm³ in each of the tanks in set two and set one respectively was placed at the bottom. The tanks were filled to the brim and left overnight. Complete draining was done the following morning before refilling the tanks with water to the required capacity. Aerators, (make INTERPET IPF 2 and INTERPET IPF 3) were fixed to mix the water and supply oxygen within each tank in set one and set two tanks respectively. This also enabled further evaporation of chlorine from the tap water used.

3.6. Stocking

On the 6th day of acclimatisation (9th January 2006), the fish were randomly picked, half the total number stocked into each experimental tank weighed and measured, and transferred into the different tanks. Tanks A1, B1, and C1 were each stocked with 20 fish while tanks A2, B2, and C2 which contained twice the volume of water in tanks of set one were each stocked with 40 fish.

3.7. Feed formulation and feeding

Feed formulation was based on percentage nutrient content results of feed ingredients analysed at Faculty of Veterinary Medicine, Nutrition laboratory, Makerere University. Three feed ingredients – “mukene” (R); *P. adusta* (E); and maize bran (Z) were used to formulate three types of experimental diets: (i) “mukene” + maize bran (named RZ); (ii) *P. adusta* + maize bran (named EZ), each balanced at 40% protein level, and (iii) maize bran only (named Z).

Feeding of fish commenced the following day after stocking. Fish in tanks A1 and A2 were fed on diet RZ ("mukene" + maize bran); fish in tanks B1 and B2 on diet EZ (*P. adusta* + maize bran); while those in tanks C1 and C2 on diet Z (maize bran only). Thus, diets RZ and EZ were test feeds while diet Z was the control.



Plate 4: Feeding fish in tank B1 on diet RZ (*P. adusta* + maize bran)

Fish were fed daily at 5% mean body weight with hourly feeding interval between 10.00 am – 6.00 pm (except on days when sampling was done; feeding started at 12.00 p.m and feed consumed was approximately 3.5% body weight). The 5% mean body weight feed was maintained for the first seven weeks. As the quantity of feed increased with increase in fish body weight, it was observed that after seven weeks fish could not consume to completion more than 5% (of the attained weight within seven weeks) quantity of feed. This was probably because as fish grows it requires feed of a smaller percentage of their body weight (Jobling, 1985). To avoid waste of feed and deterioration in water quality which could affect fish growth (Nwana, 2003), the attained respective quantity of feed per tank fed at 7th week was maintained till the end of the experiment, i.e. 10 weeks.

Although cooking is known to increase digestibility of feeds, especially carbohydrates, by fish (Schmittou *et al.*, 1998), cooked food administered on the first day was observed to sink to the bottom and much of it had not been eaten by the following day. Hence, the feeds were given in powder form, a similar method used by Nalwanga (2004). The total feed for each tank was divided into nine equal portions and one portion was given at a time.

3.8. Fish behaviour

Swimming activity, fish distribution within the water column in tanks, fish response to feeds (intensity of feeding and feed location), "courtship" and "nest building" as well as fish body coloration were observed at different times of the day i.e. at 8.00 a.m before changing water, between 8.05 -9.30 a.m during draining and refilling of water, at 9.35 a.m after refilling water, at 9.55 a.m just before feeding, and every after an hour starting 10.00 a.m – 6.00 p.m during feeding. The observations were made between 8.00 am - 6.00 pm daily for 70 days.

3.9. Water management and disease control

To remove metabolic end products excreted by fish (ammonia and ammonium compounds) which can reduce feed intake and eventually cause death (Aanyu, 2004), glass tanks were cleaned every morning by siphoning wastes (faeces, feeds) with 3 cm diameter plastic tubing. To maintain adequate quality environment, the water was completely changed every morning, and partially replaced every evening depending on observed water turbidity. Dissolved oxygen (mg/l), temperature, °C, pH and conductivity (µs/cm) were monitored in situ once every week (using Hanna Instruments) when there was variation in the quantity of feed given. To avoid pathogens development that could cause disease to fish leading to reduction in feed intake, retardation in growth and eventually death (George, 1983), used equipment and surface of aerators were disinfected every after three days using 70% ethanol. Also, fish that was hurt and incurred wound(s) during sampling was isolated. Treatment of injured fish was not possible since there were no extra tanks to nurse the affected fish.

3.10. Data Collection

Both qualitative and quantitative primary and secondary data were collected through observations and measurements of weight and total length of fish. Experimental data was the main source of primary data while secondary data was gathered mainly from libraries and internet.

3.10.1. Sampling frequency and order

To allow fish to acclimatise to the conditions, the first sampling was done after 14 days, and then once after every week for a period of 10 weeks. Initial sampling and recording of investigated parameters was done prior to stocking. The order of sampling that was maintained was tank A1, B1, C1, C2, A2 and lastly B2.

3.10.2. Total length (mm) and weight (g)

Total length (TL) and weight of randomly picked individual fish, half the number at initial stocking per tank, were recorded at every sampling using a measuring board with 1 mm interval and a Digita weighing scale model No. ACB-300 respectively (Plate 5).



Plate 5 Weighing of fish sample

During samplings, a scoop net was used to collect fish into a basin. Measurements were done as fast as possible to avoid death due to oxygen deficiency.

3.11. Data Analysis

Data that was obtained was analysed using computer programmes Excel and SPSS. Parameters analysed included weight gain and length increase. Bar graphs were used to compare the mean weight gains $\pm$ Standard deviations (SD), and increase in total length $\pm$ SD of fish fed on the different diets over the ten weeks' period. One way ANOVA statistical analysis was used to test for significance in growth of fish with feed type over period.

3.11.1. Growth indicators

Changes in weight and total length were considered. When using weight it was assumed that at the time of weighing the stomachs were empty and food did not influence weight.

3.12. Limitations to the study

These included nature, number and size of tanks used; type and nature of feeds (diets) used; duration and time of the study, as well as water and surrounding environment of the tanks. These could have caused stress that reduced fish growth (Schmittou, *et al.*, 1998). The limitations were:

Replication was not possible due to the limited number of tanks available. Two sets of tanks with variable sizes were used. It would have been desirable if they were replicated and of the same size.

Unlike in outdoor environments such as ponds, the experimental fish were confinement in small transparent glass tanks with each fish limited to a space of about 0.01 m³ and volume of 2.5L of water. Variation of space was impossible unlike in ponds where happens can be adjusted to increase space as fish grows.

The tanks did not have a recirculation system to allow automatic regulation of chemical parameters e.g. ammonia. Therefore draining and refilling was done manually which resulted into agitation of fish which would have led to stress.

Electric Power shedding done every after two days (for about 12 hours a day) affected continuous aeration of tanks to provide dissolved oxygen. Although generators were switched on whenever power would go off, they were not automatic and switching on would take about 5 -10 minutes.

Feed formulation was based on protein requirements only; other important nutrient requirements, for example, Carbohydrates, lipids, etc, were not balanced. This is because only two ingredients were used and yet balancing all the nutrients requires more than two ingredients. Also, the amino acid types and quantities were not investigated. Minerals and vitamin premixes were not available.

Lack of uniformly mixed floating feeds in form of pellets to control waste of feed through possible various ways such as dissolving in water and mixing with sand. There was frequent clogging of aerators by feed which required daily cleaning of the aerators, causing disturbance to fish.

The fish specimen did not remain uniform throughout the experiment. For example, mortalities in some tanks affected the initial stocking density, allowing more space to reduced fish numbers. This could have affected individual weight gain.

Changes in individual fish feeding, growth and feed utilisation could not be monitored in random sampling. Tagging of fish for easy identification of each fish would have facilitated this. Unfortunately, tags were lacking.

Environmental disturbances that included civil works outside and within the aquaria building, occasional sound from generator, regular visits by staff and/or

visitors at a frequency of about two people per day twice daily were sources of stress.

Limited time and timing of observations and feeding of fish carried out for 10 weeks only. This period may not have been enough to observe all the changes in feeding, growth of the fish and feed utilisation/conversion. Also, observations were made during day time only.

CHAPTER FOUR

4.0. RESULTS AND DISCUSSION

4.1. Results

4.1.1. Nutrient content of feed ingredients.

The primary feed ingredients ("mukene", *P. adusta*, and maize bran) varied in nutrient content of eight analysed parameters (crude protein, crude lipid, total carbohydrates, dry matter, crude fibre, ash, calcium and phosphorus (Table 3).

Table 3: Percentage nutrient content of "mukene", *P. adusta*, and maize bran

Nutrient contents	Feed type		
	R ("mukene")	E (<i>P. adusta</i>)	Z (maize bran)
Dry matter	91.03	90.22	87.85
Crude Protein	64.48	46.37	8.79
Crude Lipid	13.37	9.47	7.91
Total Carbohydrates	4.84	6.97	33.50
Crude Fibre	1.11	1.61	2.91
Ash	27.25	20.67	4.74
Calcium	0.01	0.31	0.02
Phosphorus	0.27	0.45	0.03

"Mukene" had the highest crude protein content (64.5%) as well as crude lipid (13.4%) and ash (27.3%) followed by *P. adusta*. Maize bran characterised the highest carbohydrates content (33.5%) while *P. adusta* had a higher content of calcium and phosphorus than the other two feeds (Table 3).

The proximate percentage composition of 40% protein diets formulated using "mukene" and maize bran (RZ); *P. adusta* and maize bran (EZ) constituted 8.31 and 4.99 crude lipid respectively (Appendix 1). Carbohydrates in feed RZ was 14.97 while that in feed EZ was 5.95 (Appendix 1).

4.1.2. Fish growth

Fish growth was monitored using changes in weight and length (weight gain and length increase) as indicators for growth (Appendix 2). Weekly weight gain and length increase were highest in fish fed on a mixture of "mukene" and maize bran (RZ) but lowest in fish fed on maize bran only (Z) (Fig. 4.1).

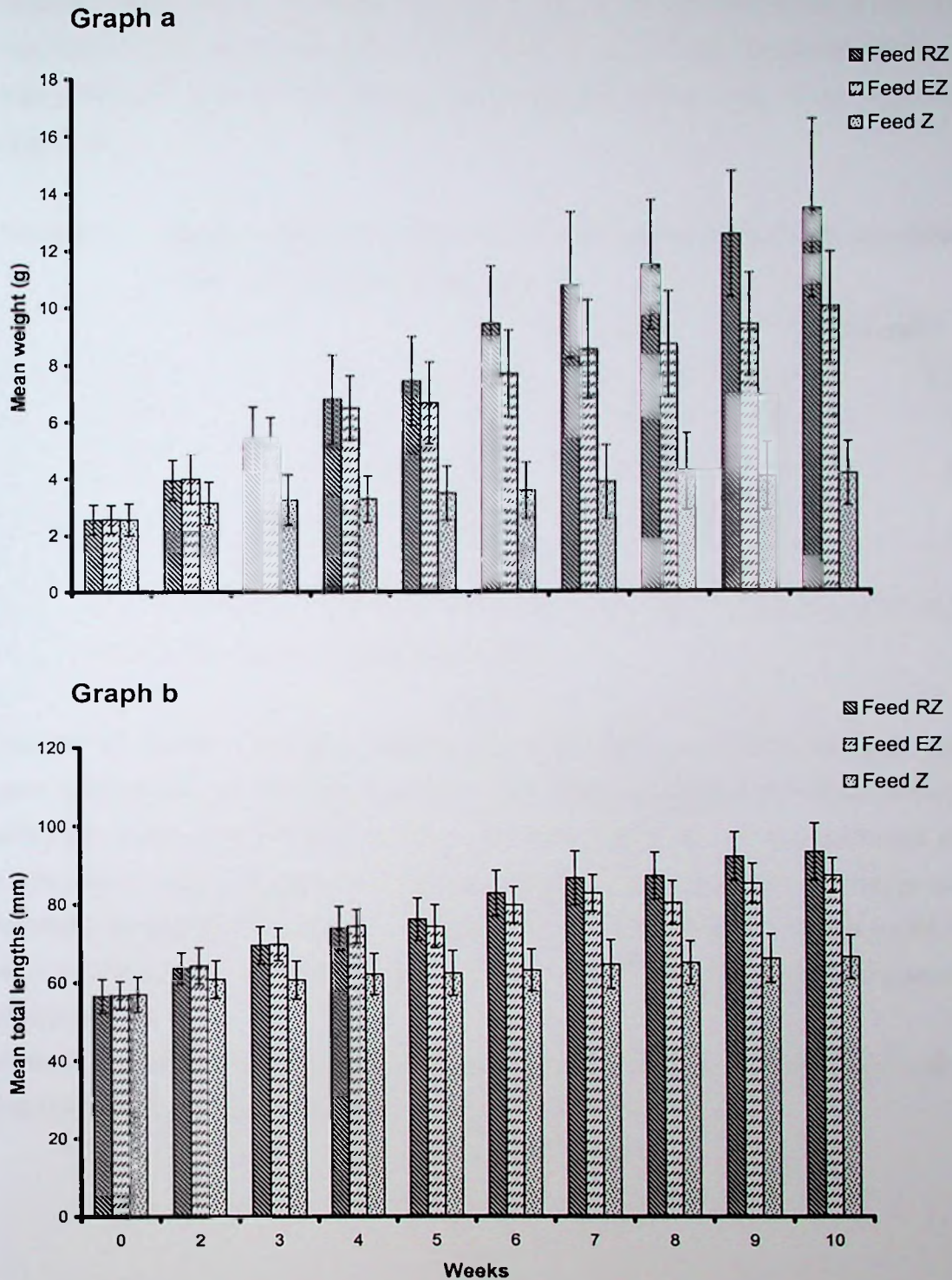


Fig. 4.1 Variations in a) weight (mean $\pm$ SD) and b) TL (mean $\pm$ SD) of fish fed on different diets (RZ = "mukene" + maize bran); EZ = (*P. adusta* + maize bran); Z = maize bran only) with period.

Overall weight gains of 10.9g, 7.4g and 1.6g; and length increase of 36 mm, 30 mm and 9 mm were registered for fish fed on diet RZ ("mukene" and maize bran), EZ (*P. adusta* and maize bran), and Z (maize bran only) respectively (Table 4).

Table 4: Mean weight (g) and length (mm) gains of fish fed on different diets over experimental period

Diet	Initial		Final		Total gain	
	weight (g)	TL (mm)	weight (g)	TL (mm)	weight (g)	TL (mm)
RZ	2.54	56.53	13.42	92.87	10.88	36.34
EZ	2.55	56.70	9.99	86.87	7.44	30.17
Z	2.54	56.97	4.14	66.17	1.60	9.20

Diet: RZ = a mixture of "mukene" and maize bran; EZ = a mixture of *P. adusta* and maize bran; Z = maize bran only

Results of SPSS One-Way ANOVA indicated significant differences in weight gain with period for fish fed on each of the three experimental diets. However, although there was a linear trend in growth of fish fed on the different diets, correlation was strongest in fish fed on diet RZ ("mukene" + maize bran), followed by diet EZ (*P. adusta* + maize bran), and lastly diet Z (maize bran only) with coefficient of correlation (R) values of 0.77, 0.52 and 0.48 respectively (Appendix 3).

Plate 6 shows the relative size of *O. niloticus* fed on the different diets over the experimental period (10 weeks).



Plate 6: Relative size of Nile tilapia fed on: a) “mukene” + maize bran
(b) *P. adusta* + maize bran (c) maize bran only

Note: TL of 114 mm in a; 105 mm in b, and 75 mm in c.

4.1.3. Observations on behaviour of fish in experimental tanks

Observations on behaviour of fish in experimental tanks were made in the morning (8.00 a.m) before changing water, after draining and during refilling (8.05 – 9.30 a.m), after refilling the tanks (9.35 a.m) just before feeding (9.55 a.m), and during feeding (hourly from 10.00 a.m to 6.00 p.m) daily for 70 days. Parameters observed included swimming activity, distribution of fish within tanks, feed location and intensity of feeding, fish colour, aggression, “courtship” and “nest building”. Mortalities per tank were recorded.

Environmental parameters (dissolved oxygen, temperature, pH, conductivity) were regulated by changing water every morning. Mean dissolved oxygen, temperature, pH and conductivity values were (5.63 ± 0.47) mg/l, (25.31 ± 0.72) °C, (6.26 ± 0.61) , and (117.54 ± 3.84) µs/cm respectively over the study period (Appendix 4).

In the morning before changing the water (8.00 am), fish in all tanks were within the water column but not very actively swimming except during the first and second week. After draining and during refilling of water (8.05 – 9.30 am) fish

started to swim more actively crowding at the point of water inlet, an observation noted in all tanks throughout the experimental period. After refilling the tanks (9.35 am), fish in tanks A1, A2 (fed on diet RZ i.e "mukene" + maize bran) and those in tanks B1, B2 (on diet EZ i.e *P. adusta* + maize bran) were seen to move to the feeding corner while those fed on diet Z (maize bran only) were distributed throughout the water column in tanks C1 and C2.

Just before feeding (about 25 minutes after changing water), from the second day of stocking, fish in tanks A1 and A2 (diet RZ = "mukene" + maize bran) tended to crowd at feeding points and moving nearer the tanks seemed to induce fish to begin struggling. From third day after stocking, moving nearer tanks B1 and B2 (diet EZ = *P. adusta* + maize bran) attracted fish to the feeding point and mild struggling compared to fish in tanks A1 and A2 (diet RZ = "mukene" + maize bran) was observed. Fish in tanks C1 and C2 (diet Z = maize bran only) remained distributed throughout the water column in tanks, and moving nearer the tanks did not attract fish to the feeding point. These patterns were noted throughout the experimental period.

During feeding, fish in tanks A1 and A2 (diet RZ = "mukene" + maize bran) were aggressive while feeding mainly from the surface and within top quarter of the water column at the feeding point. No intense struggling was observed within tanks B1 and B2 (diet EZ = *P. adusta* + maize bran) where feeding was mainly at the surface although fish sometimes fed within the water column and at the bottom. On introducing diet Z (maize bran only) in tanks C1 and C2, about half of the total numbers of fish in each tank were observed to move to the feeding point and ingest food. Feeding on diet Z (maize bran only) was at surface, within the water column, and sometimes from the bottom.

From the fourth week, some fish in tank A1 (diet RZ = "mukene" + maize bran) developed bright colours. In the same tank (A1), "Courtship" behaviour, together with "nest building" and "territorial" behaviour were observed from fourth week till the ninth week. In tanks A2 (diet RZ = "mukene" + maize bran), B1 and B2 (diet

EZ = *P. adusta* + maize bran), similar observations (as in tank A1) were noted from the end of fourth week till the tenth week; while in tank C1 (diet Z = maize bran only) such behaviour was observed from the fifth week till the tenth week; and in tank C2 (diet Z = maize bran only) bright colours were observed during the tenth week and a single "nest" seen once during the same week. "Courtship" was also noted in tank C2 but no "territorial" behaviour was observed. In all tanks, during the period of "nest building", response to feed by "nest"-building fish was reduced. Also observed was "territory" guarding (except in tank C2) that involved chasing of some fish by brightly coloured fish, an action that scared them away from feeding. One fish in each of the tanks C1 and C2 (diet Z = maize bran only) developed dark coloration throughout the body during the third and ninth week respectively.

4.1.4. Mortality

About 6% total mortality occurred in all the tanks over the experimental period (Tables 5a & b). Half of the total mortality (3%) was noted during the third week of the experiment. Highest mortality (4%) was recorded in fish fed on diet Z (maize bran only) especially from tank C2 (3%) and 1% from tank C1 as a result of isolation. Mortality of fish fed on diet RZ ("mukene" + maize bran) was about 2% while that fed on diet EZ (*P. adusta* + maize bran) was due to isolation (1%). No visible disease signs were identified.

Table 5a. Mortality per tank over the experimental period

Tank	No. dead	No. isolated	% mortality per tank	No. stocked
A1	2	0	10	20
A2	1	0	2.5	40
B1	0	1	5	20
B2	0	0	0	40
C1	1	1	5	20
C2	4	0	10	40
Total	10		5.5	180

Table 5b: Weekly mortality/isolation of fish in experimental tanks

Period	Tank	No. dead	Comments on likely cause of death/isolation
Week 2	B1	1*	Inactive, developed wound on head
Week 3	A1	1	Unidentified
	A2	1	unidentified
	C2	3	unidentified
Week 5	A1	1	Jumped out of tank and found dead on floor
	C1	1*	Inactive. Had wound on head and lost scales on part of the body
	C2	1	Unidentified
Week 8	C1	1	Unidentified

* Isolated fish; Fish in tanks A1, A2 were fed on diet RZ ("mukene" + maize bran); fish in tanks B1, B2 on diet EZ (*P. adusta* + maize bran); and fish in tanks C1, C2 on diet Z (maize bran only)

4.2. Discussion

4.2.1. Nutrient content of feeds

From the results (Table 3), it is evident that none of the ingredients could independently provide all the required levels of nutrients for optimum growth, even after feed formulation using two ingredients (Appendix 1). However, considering recommended levels of protein requirement (NAADS, 2005; Craig & Helfrich, 2002; Cesar, 2000; Rondolph, 1986; Tamir, 2005), "mukene" and *P. adusta* were within the ranges of protein requirements.

It is noted that although the lipid content in diets RZ ("mukene" + maize bran), EZ (*P. adusta* + maize bran), and Z (maize bran only) were within the recommended ranges, the carbohydrates in both diets RZ and EZ were less (NAADS 2005) except that in feed Z. All the three diets were incomplete to provide optimum fish growth (Rondolph, 1986). The formulated diets would only give an indication of the impact of proteins (in RZ and EZ) on fish growth in comparison to Z with inadequate protein level for proper fish growth. For optimum fish growth, more ingredients would have been used to formulate a balanced diet.

4.2.2. Fish growth

Results of this experiment are in line with Schmittou *et al.* (1998) and Lojjo (2004) who observed that fish growth was directly proportional to protein level in feed. Fish fed on diet RZ ("mukene" + maize bran) and EZ (*P. adusta* + maize bran) with 40% crude protein gained more weight than that fed on feed Z (maize bran only) with low protein content of 8.79%. The least weight gain in fish fed on maize bran only could partly be due to the demand for too much energy to digest carbohydrates (Nalwanga, 2004).

Despite containing almost the same quantity of proteins in diet RZ ("mukene" + maize bran) and EZ (*P. adusta* + maize bran) (Appendix 1), weight gain by fish was higher in fish fed on diet RZ. The performance of a feed is influenced by its composition, the extent to which the nutrients are digested and assimilated and most important the protein quality (Lojjo, 2004; Nalwanga, 2004) which is

determined by its amino acid profile that should closely match the fish requirements (Rondolph, 1986). It is likely that the protein quality of "mukene" was different from that of *P. adusta*. "Mukene", being a fish meal, that is highly palatable with high protein digestibility and an amino acid profile that closely matches fish dietary requirements (Cesar, 2000), could have influenced higher acceptability, digestibility as well as assimilation of diet RZ ("mukene" + maize bran) than EZ (*P. adusta* + maize bran). In addition, the floating nature of ingredient E (*P. adusta*) in diet EZ (*P. adusta* + maize bran) could have been a twofold disadvantage: Firstly, since feeding on diet EZ (*P. adusta* + maize bran) was mainly at surface, it is possible that the fish fed mostly on the floating ingredient E (*P. adusta*) and were not fully utilising ingredient Z (maize bran), which tended to sink, hence missing much of nutritional value (mainly energy) in maize bran. Secondly, there was frequent "sucking" of diet EZ (*P. adusta* + maize bran), particularly ingredient E (*P. adusta*), and clogging into the aerators compared to diets RZ ("mukene" + maize bran) and Z (maize bran only) hence an unequal reduction in the quantity of different diets (RZ, EZ, Z) available for fish consumption, yet fish growth is not only influenced by the quality of feed but also the quantity (Aanyu, 2004).

A balanced diet is important for proper fish growth (NAADS 2005; Rondolph, 1986, Schmittou *et al.*, 1998). Craig & Helfrich (2002) report that proteins are used for fish growth if adequate levels of fats and carbohydrates are present in diet. Inadequate energy source in a feed can result into metabolism of protein to produce energy, rather than for growth (SRAC, 1998). In situations of limited energy source in feed, fish first use the available energy for other body functions (e.g. swimming, digestion, breathing, etc) giving growth the last priority (Swift, 1993). From the formulation (Appendix 1), it is noted that although the lipid percentage composition in diets RZ ("mukene"+ maize bran) and EZ (*P. adusta* + maize bran) could have been adequate, carbohydrate was lower than the recommended 25% (NAADS, 2005) even though fish is known to primarily utilise (and metabolise better) proteins and lipids, but secondarily carbohydrates to get their energy demands (Schmittou, *et al.*, 1998). Therefore, with proper balanced

diets comprising "mukene" and *P. adusta* as sources of proteins, higher weight gains could be attained.

Considering the weight gains of 11g, 7g and 2g for fish fed on different diets i.e. RZ ("mukene"+ maize bran), EZ (*P. adusta* + maize bran), and Z (maize bran only) respectively (Table 4), and from results of One-Way ANOVA (Appendix 3), it can be concluded that there was a significant difference in weight gain of *O. niloticus* fed on the different diets. Therefore, the hypothesis that there would be no weight gain difference as set out is rejected.

4.2.3. Observation of fish behaviour

Inactive behaviour of fish before changing water could have been due to deterioration in water quality (Appendix 3) as a result of accumulated fish wastes and feed left-over. This was overcome by refilling water which appeared to induce increased activity (swimming), probably a response to increased dissolved oxygen availability from the incoming water as a result of bubbling at water inlet. Struggling noticed in fish fed on diet RZ ("mukene" + maize bran) was probably due to higher palatability of "mukene" ingredient which possibly influenced higher competition for diet RZ ("mukene" + maize bran) compared to EZ (*P. adusta* + maize bran) and Z (maize bran). Floating of *P. adusta* in diet EZ (*P. adusta* + maize bran) gave the fish almost equal chance to accessibility to feed hence reduced struggling. The poor response of fish to diet Z (maize bran only) could be attributed to satiation caused by excess energy in feed (Schmitou *et al.*, 1998) hence reducing desire for feed consumption.

Development of bright colours could be a social interaction behavioural mechanism of aggression leading to intimidation of subordinate (not brightly coloured) fish by dominant fish (brightly coloured) so that the dominant fish could successfully compete for food (Aanyu, 2004). Variations in rate of development of bright colours by fish fed on the different diets could be due to difference in feed palatability which probably influenced intra-specific competition. Dark coloration/pigmentation in fish fed on diet Z (maize bran only) could be an

indication of poor assimilation of carbohydrates (Lannan, *et. al.* 1986), but probably not a sign for subordinates (Aanyu, 2004) since no aggressive behaviour was observed. It was not clearly established whether "nest" building and "territory guarding" were associated with hormone development which is influenced by protein availability and quality (De Silva, 1991).

4.2.4. Mortality

The high survival rate (94%) over the study period was probably due to application of standard management practices. For example, there was adequate oxygen and no major fluctuations in environmental conditions. In addition, feeding was administered according to set times. Also, the strict preventive measures that included healthy seed (fingerlings), frequent disinfection of equipment and surface of aerators, as well as regular changing of water provided a favourable environment for fish survival.

Mortality observed during third week was probably a result of stress caused by first sampling especially before the fish got fully acclimatised to the new environment. The high mortality recorded in fish fed on diet Z (maize bran) could be due to a nutritional disease caused by low protein quantity and probably quality (lack of some indispensable amino acids) (George, 1983).

CHAPTER FIVE

5.0. CONCLUSIONS AND RECOMMENDATIONS

5.1. Introduction

This study evaluated the importance of mayflies, *Povilla adusta* Navas as an alternative protein source in fish feed for Nile tilapia. The study recognised that many subsistence fish farmers have difficulty in accessing commercial fish feeds, others cannot afford "mukene" (*Rasrineobola argentea*) the commonly used protein ingredient in feed, while many of the farmers use maize bran alone. This chapter draws out the main issues identified and provides conclusions based on the results of the study. Recommendations and areas for future research are also suggested.

5.2. Conclusions

"Mukene" had the highest crude protein as well as crude lipid and ash followed by *P. adusta*, but maize bran contained the highest carbohydrates. From these results, it is concluded that "mukene" and *P. adusta* are good sources of proteins, lipid and minerals while maize bran mostly provides carbohydrates for energy.

A mixture of "mukene" and maize bran produced the highest fish growth (weight gain and length increase) followed by a mixture of *P. adusta* + maize bran while maize bran only had the least growth. In order to achieve optimum fish growth and hence yield, it is important for fish farmers to include proteins in feeds.

Just like "mukene" the commonly used source of protein in fish feeds, *P. adusta* can be efficiently used to improve Nile tilapia growth. The insects can be utilised as an alternative protein source in absence of "mukene" or they can form an additional feed ingredient together with "mukene" to increase fish yield. Although it would require large quantities of *P. adusta* to feed fish, insects do not need to be the main composition of a meal; they can be feed of season so that whenever available they can be utilised.

Although tilapia can be conditioned to diverse feeds, the rate of conditioning differs with diet. Fastest conditioning was achieved with a mixture of "mukene" and maize bran followed by that of *P. adusta* and maize bran, and lastly maize bran. Protein content and palatability of feeds are therefore important in conditioning tilapia to artificial feeds.

5.3. Recommendations for action

There is need for better formulation of nutritionally complete feeds with desirable qualities in form of floating pellets of various sizes depending on age of fish.

Similar experiments should be conducted for a longer period of time so as to study possible changes in growth and feed utilisation. Also, longer intervals, such as two weeks, between samplings should be considered in order to minimise stress in fish which reduces growth.

Changes in individual fish feeding, growth and feed utilisation can not be monitored in random sampling. Tagging including labelling of fish is recommended for easy identification of each fish.

Reserve tanks for isolating and treating fish should be available in studies of this type.

Each tank should have a tap for outlet, overflow pipe, and an overhead tap for re-filling water to minimise agitation of fish.

This study was conducted in a confined environment (aquaria laboratory) that was prone to noise and movements of people. Experiments of this type should in future be set up in isolated spaces free from disturbances

Subsistence farmers who can not afford formulated feeds and have been using maize bran only as the major supplement, but have access to *P. adusta* are encouraged to utilise the insects in combination with maize bran. A lighting

system such as pressure lamp near lake shores could attract the insects for ease of trapping them.

5.4. Areas for future research

Protein quality (amino acid types and quantities) of the ingredients -"mukene", *P. adusta*, and maize bran should be established to determine the differences in quality for guidance in formulating feeds with balanced amino acids. Studies using different balanced diets composed of "mukene", and *P. adusta* as protein sources should be carried out.

Similar studies using fresh *P. adusta* (i.e. as soon as they emerge) could be undertaken to give an insight into the quality and performance of *P. adusta* directly trapped onto fish ponds.

The biology and ecology of *P. adusta* is not fully known. Studies in this area could give information that may help in the culture of *P. adusta*. There is also need to determine emergence patterns of *P. adusta*, their distribution, and abundance. This information could be used to guide maximum harvesting and utilisation of *P. adusta* in farming of Nile tilapia.

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APPENDICES

Appendix 1: Proximate percentage composition of formulated diets using

(i) "mukene" and maize bran (RZ), (ii) *P. adusta* and maize bran (EZ)

FEED RZ

Ingredient	Rations (%)	CP	CL	CB	Fibre	Ash	Ca	P
Maize bran	44	3.8676	3.4804	14.74	1.2804	2.0856	0.0088	0.0132
"mukene"	56	36.109	4.8277	0.2337	0.0026	0.0007	7E-08	2E-10
	100	39.976	8.3081	14.974	1.283	2.0863	0.0088	0.0132

FEED EZ

Ingredient	Rations (%)	CP	CL	CB	Fibre	Ash	Ca	P
Maize bran	17	1.4943	1.3447	5.695	0.4947	0.8058	0.0034	0.0051
<i>P. adusta</i>	83	38.487	3.6447	0.254	0.0041	0.0008	3E-06	1E-08
	100	39.981	4.9894	5.949	0.4988	0.8066	0.0034	0.0051

CP = crude protein
P = phosphorus

CL = crude lipid

CB= carbohydrates

Ca = calcium

Appendix 2 : Variations in mean weight (g) and total lengths (mm) of fish fed on different diets with period

Mean wt ± STD										
Week	0	2	3	4	5	6	7	8	9	10
Feed RZ	2.54 ± 0.52	3.93 ± 0.70	5.38 ± 1.14	6.78 ± 1.55	7.41 ± 1.55	9.41 ± 2.01	10.74 ± 2.56	11.45 ± 2.26	12.53 ± 2.20	13.42 ± 3.13
Feed EZ	2.55 ± 0.49	3.98 ± 0.85	5.20 ± 0.94	6.47 ± 1.13	6.64 ± 1.43	7.66 ± 1.52	8.50 ± 1.72	8.68 ± 1.85	9.37 ± 1.80	9.99 ± 1.91
Feed Z	2.54 ± 0.56	3.14 ± 0.74	3.25 ± 0.89	3.27 ± 0.82	3.48 ± 0.94	3.57 ± 0.99	3.86 ± 1.29	4.22 ± 1.35	4.05 ± 1.20	4.14 ± 1.13
Mean TL ± STD										
Feed RZ	56.53 ± 4.32	63.63 ± 3.96	69.13 ± 4.77	73.77 ± 5.56	75.83 ± 5.41	82.47 ± 5.79	86.33 ± 6.86	86.90 ± 5.94	91.77 ± 6.12	92.87 ± 7.20
Feed EZ	56.70 ± 3.59	64.27 ± 4.65	69.43 ± 4.13	74.30 ± 4.28	74.03 ± 5.51	79.47 ± 4.67	82.47 ± 4.67	80.03 ± 5.54	84.93 ± 5.03	86.87 ± 4.42
Feed Z	56.97 ± 4.50	60.50 ± 4.80	60.30 ± 4.84	61.87 ± 5.26	62.13 ± 5.73	62.87 ± 5.31	64.33 ± 6.24	64.73 ± 5.56	65.83 ± 6.20	66.17 ± 5.57
No. of fish	30	30	30	30	30	30	30	30	30	30

RZ = "mukene" + maize bran

EZ = Povilla + maize bran

Z = maize bran

**Appendix 3: Results of SPSS One-Way ANOVA for weight gain in fish fed
on the different diets over time**

Feed	model	Sum of squares	df	Mean square	F	Sig.	R value
RZ	Regression	2668.221	1	2668.221	430.272	.000	.769
	Residual	1847.971	298	6.201			
	Total	4516.192	299				
EZ	Regression	768.496	1	768.496	111.333	.000	.522
	Residual	2057.004	298	6.903			
	Total	2825.500	299				
Z	Regression	831.274	1	831.274	88.436	.000	.478
	Residual	2801.130	298	9.400			
	Total	3632.404	299				

At 95% Confidence Interval

RZ = "mukene" + maize bran; EZ = *P. adusta* + maize bran; Z = maize bran

Appendix 4: Weekly variations in mean dissolved oxygen (mg/l), pH, temperature (°C) and conductivity (µs/cm) in experimental tanks

Week	Temperature	pH	Dissolved oxygen	Conductivity
2	24.37 ± 0.31	6.94 ± 0.06	6.10 ± 0.53	116.18 ± 5.37
3	24.77 ± 0.19	6.40 ± 0.01	5.64 ± 0.64	116.35 ± 5.28
4	24.8 ± 0.22	6.39 ± 0.01	5.64 ± 0.6	118.42 ± 3.83
6	26.08 ± 0.2	5.95 ± 0.75	5.48 ± 0.26	118.17 ± 3.43
7	25.91 ± 0.08	5.94 ± 0.74	5.43 ± 0.15	118.05 ± 2.89
9	24.80 ± 0.08	6.39 ± 0.70	5.64 ± 0.10	118.42 ± 2.37
1 to 9	25.31 ± 0.72	6.26 ± 0.61	5.63 ± 0.47	117.54 ± 3.84