



STEAMING PROCESS OPTIMIZATION, PHYSICAL TESTING AND ANALYSIS OF GROWTH PERFORMANCE PARAMETERS IN EARTHWORM-BASED PELLETS FOR AFRICAN CATFISH (*CLARIAS GARIEPINUS*)

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ABSTRACT

New formulation of fish pellets was designed by using earthworm powder and other economical ingredients such as fish meal, soybean waste and rice bran which were calculated using Pearson's square method. The optimization of steaming process done by One-Factor-at-One –Time (OFAT) method showed that maximum water stability was achieved within 40 minutes steaming time at 80°C steaming temperature. Physical properties tested for soaking, protein leaching and hardness revealed the positive results of stable and durable pellets to be applied as feeding material. Growth performance was tested by applying earthworm-based fish pellets on African catfish and measuring weight and length increment weekly. Weight increment of 6.58% was achieved to suggest the potential of this formulation in catfish cultivation.

Keywords: Steaming process, Earthworm-based pellets, African catfish

INTRODUCTION

Earthworm powder is one of the alternative protein sources that have been recognized as potential ingredients in providing efficient protein to support animal growth (Gabriel and Dedeke, 2010). Protein in earthworm powder contain the necessary amino acids for catfish (Julendra, 2009). Hence earthworm powder are reported as having high ability to accelerate growth, develops muscles, gain weight, prevent protein and amino acid deficiency and attractive to fish (An Phu Earthworm Farm, 2010). Pearson's square method is a simple method that can be used to formulate animal feed previously. Many researchers used Pearson's square method to generate the amount of ingredients need in feed including the protein content in feed (Bolorunduro, 2002; Wagner, 2012). Moreover, animal protein is necessary to be added to fish feed as compare to plant protein to supply sufficient amino acid (Bolorunduro, 2002). Steaming process and binder agent are recognized among factors

that are suitable to be applied to increase the stability of pellets in water. Upon applying the steam, the binder agent will gelatinize and perform strong bonding among ingredient particles (Yogendra, 2011). Optimization of temperature and time is important to maximize the gelatinization process and hence increase stability of pellets in water or commonly refer as water stability. Generally starch gelatinization can be achieved when subjected to temperature from 70 to 90°C (Tulyathan, 2006). Desired properties of good fish pellets are high water stability, sufficient durability in water, not easily break during storage and minimum leaching of nutrient especially protein when soaking. An efficient feed will increase the weight of catfish in acceptable growth period. The objective of this study is to apply steaming process to manually produce earthworm-based pellets, examine the physical properties of steamed earthworm-based pellets and to apply on African catfish feeding experiment.

MATERIALS AND METHODS

Raw Material Preparations

The earthworm powder, fishmeal, tapioca flour and rice bran were obtained from local market. Rice bran was finely ground to smaller particle. Soybean waste is taken from individual vendors, dry and ground to a fine particle size. Micro ingredients such as the vitamin mix and calcium bis-(dihydrogen phosphate) were purchased from local supplier and Hamburg Chemicals Inc. (Germany) respectively. Protein analysis was performed for all ingredients, excluding micro ingredients, using Kjeldahl's analyzer (BuchiLabortechnik, Switzerland).

Formulation Calculation

Amount of ingredients were determined based on Pearson's square formulation (Hardy, 1980) whereby the protein content was fixed at 32% (Robinson and Li, 1999). Earthworm powder, fishmeal and soybean waste were classified as protein sources whereas rice bran and tapioca flour were grouped as energy sources.

Preparation of Pellets

All ingredients were weighted (Table 1) and mixed at slow pace with distilled water in 2:3 portion (v/w). The mash was pelletized using pelletizer and the steam was applied according to selected temperature and time.

Steaming Process Optimization

Steaming time and temperature were optimized by using the One-Factor-at-One-Time (OFAT) (Ahmad Anas, 2012) technique taking water stability as the response. Steaming times at 10, 20, 30, 40 and 50 minutes were tested at fixed temperature of 90°C (Chapman, 2003; Tulyathan, 2006). Optimum time from OFAT study was chosen and applied for five ranges of temperatures namely 60, 70, 80, 90 and 100°C.

Physical Testing

Soaking experiments: The dry weights of 10 pellets of each type of pellets were determined. The pellets were allowed to sink from the surface of water and left to submerge for 2, 5 and 10 minutes. The water was drained out after submerged time and the pellets were dried using absorbent paper. The particles were re-weighted to obtain weight increase after immersion (Paolo, 2006)

Protein leaching experiments: Soaking water from soaking experiments was tested for its protein content using the Kjeldahl's method for all time intervals previously conducted (Oyedapo, 1995).

Hardness tests: 30 pellets were used in the test where the forces (N) applied to break the pellets were recorded (Yogendra, 2011) using Texture Analyzer (Brookfield Model CT3, USA). Length and width of the pellets were measured before the test.

Fish experimental: The steamed earthworm-based pellets were fed to triplicate tanks of eight-week old catfish and commercial feeds were fed to another triplicate tank as control. Each tank fitted with five catfish and feed twice daily with 8% feeding rate. The water in the tanks was maintained with desired oxygen ranging from 4.5 – 5.6mg/L pH, 6.7-7.5 temperature $28 \pm 1^{\circ}\text{C}$ (Agokei, 2011); (Kasi, 2011). Weight and length measurement was recorded weekly for individual fish.

Calculation: The following formula was applied to the data

Specific growth rate (%/day) = $[(\ln W_f - \ln W_i) / T] \times 100$ (Aderolu, 2010)

Voluntary food intake (g/fish/day) = $W_i \times \text{feeding rate (8\%)}$ (Sogbesan, 2007)

Where W_f refers to the mean final weight, W_i is the mean initial weight of fish and T is the feeding trial period in days.

Feed conversion rate = total feed intake (g) / total wet weight gain (g) (Aderolu, 2010)

Protein intake = Percentage protein in feed x total weight of diet applied (Sogbesan, 2007)

Protein efficiency ratio = Mean weight / protein intake (Aderolu, 2010)

RESULTS

Protein Percentage and Weight of Ingredients in Formulation

All ingredients except micro ingredients were analyzed for protein percentage and the percentage of ingredients required to achieve 32% protein contain were calculated using Pearson's square and listed as in Table 1.

Table-1. Percentage of protein for each ingredients and weight of each ingredient according to Pearson's square formulation

Ingredients	Protein (%)	Weight (%)
Earthworm powder	77.1232	12.1633
Fish meal	51.1621	24.3265
Soybean waste	24.2634	36.4898
Rice bran	16.285	16.0137
Tapioca flour	0.1000	8.0067
Vitamin mix	-	2
Calcium bis-(dihydrogen phosphate)	-	1

Steaming Process Optimization Using OFAT

One-Factor-At-One-Time (OFAT) was applied at fixed temperature, 90°C with steaming time of 10, 20, 30, 40 and 50 minutes. The effect on pellets stability in water was recorded as percentage and shown in Table 2.

Table-2. Water stability percentage on 10, 20, 30, 40 and 50 minutes steaming time at fixed steaming temperature, 90°C.

Time (min)	Water stability (%)
10	74.09
20	77.11
30	76.69
40	79.11
50	73.65

According to Table 2, the highest water stability percentage was obtained at 40 minutes steaming time. The optimum time was used for the subsequent study on the effect of steaming temperature at 60, 70, 80, 90 and 100°C. Water stability was determined again and recorded in Table 3. Optimum steaming time and temperature were at 40 minutes and 80°C.

Table-3. Water stability percentage on 60, 70, 80, 90 and 100°C steaming temperature at fixed steaming time 40 minutes.

Temperature (°C)	Water stability (%)
60	78.19
70	76.49
80	82.14
90	71.10
100	73.36

Physical Properties Tests

Table 4 shows the weight increment of pellet in soaking experiment. Steamed pellets have a higher percentage of soaking weight compared to non steam pellets. Steamed pellets and non steamed pellets have increased soaking weight of 18.88% and 10.73% respectively from 2 minutes to 10 minutes soaking time.

Table-4. Percentage of weight increment in 2, 5, and 10 minutes of soaking for steamed and non steamed pellets

Time (minutes)	Soaking weight increment (%)	
	Steam pellets	Non steam pellets
2	59.86 ± 0.0298	26.00 ± 0.0112
5	67.77 ± 0.0496	26.65 ± 0.0304
10	71.16 ± 0.0022	28.79 ± 0.0406

Table 5 shows leaching protein concentration of water soaked by steamed pellets and non steamed pellets. Higher protein concentration in non steam pellets was detected compared to steam pellets. From 2 minutes to 10 minutes soaking, the protein leaching concentration increased 61.18% for steam pellets and 156.2% for non steam pellets which is higher than the initial weight of pellets tested.

Table-5. Leaching protein concentration in 2, 5, and 10 minutes for steam and non steam pellets

Time (minutes)	Leaching protein concentration (mg/ml)	
	Steam pellets	Non steam pellets
2	0.0255 ± 0.0022	0.0411 ± 0.0029
5	0.0348 ± 0.0031	0.0617 ± 0.0053
10	0.0411 ± 0.0015	0.1053 ± 0.0008

Table 6 showed forces (N) were required to break steam and non steam pellets. Steam pellets required higher breaking force compare to non steam pellets. All tested pellets have same average width and length with a standard deviation represented error of pellet shape.

Table-6. Breaking force for steam and non steam pellets

Pellets type	Breaking Force (N)	Width (mm)	Length (mm)
Steam	12.3799 ± 0.0439	0.50 ± 0.0068	10.00 ± 0.0785
Non steam	1.9035 ± 0.3960	0.50 ± 0.0111	10.00 ± 0.0371

Steam pellets possessed all the positive attributes required by the pellets before proceeds to the next phase which is application of the pellets to the catfish.

Growth Performance

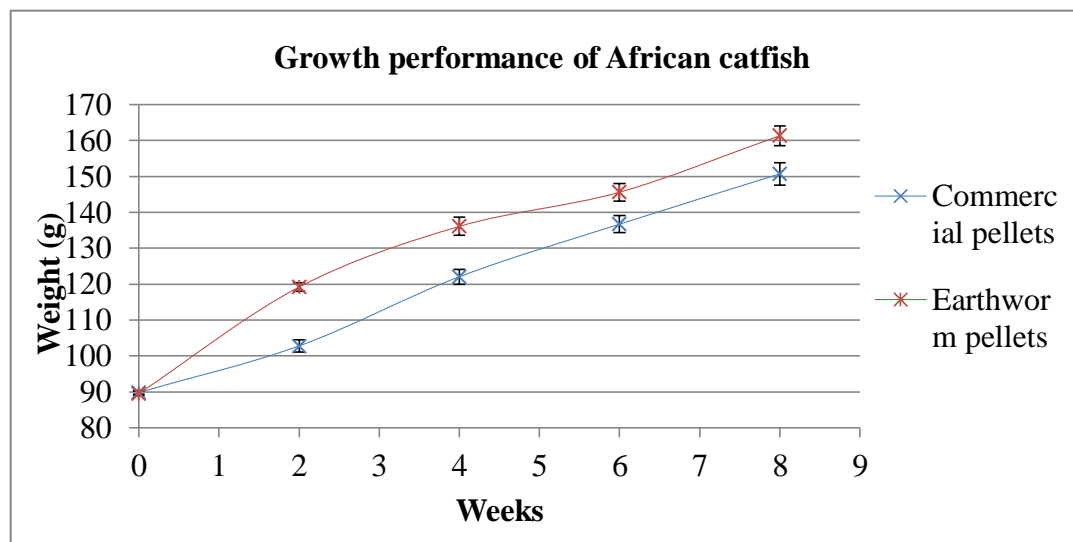
Table 7 presented the growth parameters of catfish for eight weeks. Percentage weight gain and specific growth rate of catfish feed with earthworm-based pellets are higher than commercial pellets. Lower amount of feed from earthworm-based pellets need to increase fish weight compare to commercial pellets. Higher protein efficiency ratio showed in earthworm-based pellets compare to commercial pellets.

Table-7. Growth parameters of catfish for 56 days (8 weeks)

Parameters	Commercial pellets	Earthworm-based pellets
Initial weight (g)	89.57	89.03
Final weight (g)	149.50	166.45
Mean weight (g/fish)	59.93	77.42
Percentage weight gain (%/fish)	166.91	186.96
Specific growth rate (%/fish)	0.91	1.12
Voluntary food intake (g/fish/day)	7.12	7.12
Feed conversion rate	6.65	5.15
Protein intake	115.63	127.60
Protein efficiency ratio	0.52	0.61

Figure-1. showed weight increment of African catfish for 8 weeks of feeding time with steamed earthworm-based pellets and commercial pellets. The growth of catfish fed with steamed earthworm-based pellets is 6.58% greater than commercial pellets.

Figure-1. Growth performance for 8 weeks feeding time with steamed earthworm-based pellets and commercial pellets.



DISCUSSION

Pearson's square method was used in the calculation of the amount of the ingredients used in formulation process. The ratio of earthworm, fishmeal and soybean used was 1:2:3. This formulation minimized the used of earthworm powder since this material is not considered as waste material compare to other two ingredients and eventually will reduce the pellets cost.

In water stability results, there is a trend when the longer steaming time was applied to the pellets, the higher the water stability observed. The 40 minutes steaming time provided optimized water and heat to the pellet to gelatinize the starch of the tapioca flour. The gelatinized starch binds the ingredients together and therefore pellets exhibited higher stability and durability in water. When exposed to longer than 40 minutes steaming time, the water stability was dropped to the lowest percentage. Exposure to long duration of heat may caused destruction to glycosidic linkages of starch polymer and therefore loosens the binding (Srilakshmi, 2003).

Pellets processed at 80°C steaming temperature and 40 minutes steaming time were then further tested for the physical properties. Steamed pellet were able to absorb water up to 71% of their weight within 10 minutes soaking time without breaking down. This property is important so that the pellets can retain their shape until the fish realized their presence and feed on them. If pellets were to break too fast (less than 10 minutes), there is a likelihood that fish will not feed them at all in their powder form. Minimum protein leaching within 10 minutes soaking time also an important feature to ensure fish obtained sufficient amount of protein. Data shown that optimized gelatinization of the starch had resulted the strong binding within pores in the pellet and hence prevent the leaching of protein. The occurrence of strong binding due to complete gelatinization of binder in the ingredients mixture provides a stable pellets structure. A significantly higher force was required to break the steam pellets. This property is important for storage and transportation of pellet so as the impact of pressure on the pellets will not affect their shape.

The effect of earthworm-based formulation on the catfish growth was directly shown during the 2nd week of data collection. There was 14.4% of weight increment during this period and the increment was about constant throughout the 8th weeks of feeding time. The instant increase of growth upon feeding with earthworm-based formulation right in the 1st week suggested that the pellets consumption was efficient and directly contributed to the mass composition. The most important factor here is the use of earthworm powder in the formulation since it is high in protein which is specifically amino acid lysine (Zarina Zakaria, 2012). Lysine is known for the only important role which is tissue deposition (Miles, 2012). This data emphasis the influenced of earthworm nutritional value to enhance the growth of catfish and therefore the use of this ingredient is highly recommended since voluntary food intake for both treated and commercial pellet are similar, which is 7.12.

CONCLUSION

Steaming process has high potential to increase water stability of fish pellets without the use of expensive extruder. Optimization using OFAT method showed that the steaming temperature and time were 80°C and 40 minutes respectively for maximum water stability achievement. Physical testing of steamed pellets showed positive results indicated the stability and durability of the pellets to be applied as fish feeding material. Steamed earthworm-based pellets were proved to enhance catfish growth.

ACKNOWLEDGEMENT

The authors gratefully acknowledge Malaysia Ministry of Agriculture for the Science Fund (9006-0006) and School of Bioprocess Engineering, University Malaysia Perlis for the facilities support.

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