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# Replacement of Soyabean Meal with Toasted *Leucaena leucocephala* Seed Meal in the Diet of Catfish Hybrid (*Clarias gariepinus* ♀ X *Heterobranchus longifilis* ♂) Post Fingerlings

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***Authors' contributions***

*This work was carried out in collaboration between both authors. Author BSA designed the study and wrote the draft manuscript. Author DEO performed the laboratory experiments and statistical analysis under the supervision of author BSA. Both authors read and approved the final manuscript.*

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## ABSTRACT

Ground toasted *Leucaena leucocephala* seed (TLLS) was incorporated as a non-convectional feedstuff at 0, 10, 20, 30, 40, 50% replacement level for Soyabean cake; and fed to clariid catfish hybrid "*Heteroclarias"* (*Clarias gariepinus* ♀ x *Heterobranchus longifilis* ♂) post fingerlings in plastic tanks and fed to satiation twice daily. All feeds contained 40% crude protein (CP). The mean weight gain of fish, feed intake and specific growth rate showed no significant difference (ANOVA: P > 0.05) between treatment I and II, but were significantly different from treatments III, IV and V. The percentage weight gain and protein efficiency ratio were not significantly difference (ANOVA: P > 0.05) among all the treatments, feed conversion ratio was significantly difference (ANOVA: P> 0.05). The mean weight-gain of fish, percentage weight gain, feed intake and feed conversion ratio was significantly superior in treatment I (control), while treatment II was the next in terms of superior ranking. Treatment IV, V and VI did not favor fish growth and feed utilization. Survival among treatments slightly varied but not as a result of the feed treatments. At the end of the experiment, the recommended inclusion level of toasted *Leucaena leucocephala* seed meal was 10% as a possible replacement for soybean seed meal.

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## 1. INTRODUCTION

*Heteroclarias* is an intergeneric hybrid of two African Clariid catfishes; *Heterobranchus longifilis*♂ and *Clarias gariepinus* ♀ [1]. *Clarias gariepinus* occupies a unique and prominent position in commercial fisheries in Nigeria because of its tastiness, hardiness and as well as tolerant to poor water conditions, it has an efficient feed conversion ratio in the males and as such attracts high market price, reaches maturity early and has higher fecundity than *Heterobranchus longifilis* [2]. It is also capable of reproducing in captivity and growing to a size of about 7.0 kg; [3,4]. In addition, *Heterobranchus longifilis* can tolerate low dissolved oxygen, high turbidity and importantly grows faster to a size of 14.0 kg, has higher feed conversion but not as hardy and does not have the same high survival rate as *Clarias gariepinus* [5]. Several studies have demonstrated that *Heteroclarias* (*Clarias gariepinus* ♀ X *Heterobranchus longifilis* ♂) exhibited superior growth, improved survival and general hardiness than true breed of either *Clarias gariepinus* or *Heterobranchus longifilis* [6-8]. In aquaculture, feeds play a vital role as it constitutes 60-70% of total investment. The high cost of fish feed has been recognized as a major factor militating against rapid development of aquaculture due to most of the conventional feedstuffs being used in human foods and farm animal feeds hence bringing about high cost and scarcity of these feedstuffs [9]. In a bid to produce feeds at reduced cost and lessen the pressure on conventional feedstuff, studies have been carried out on the use of unconventional feedstuffs as alternative source of protein without compromising fish growth and health.

*Leucaena leucocephala,* a non-convectional plant protein, is generally known as "multipurpose tree" [10] due to its diverse use. It has a crude protein (CP) value of 28 to 45% [11] [12]. It is known to be high in α-carotene, [13] with a rich amino acid profile.

This aim of this study was to determine the utilization and best replacement level of Soyabean meal by dietary toasted *Leuceana leucocephala* seed meals by (*Clarias gariepinus* ♀ X *Heterobranchus longifilis* ♂) hybrid post fingerlings.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Diets

Fishmeal, soybean, corn meal, wheat offal and bone meal were purchased from a Lish private company in Benin City. The multivitamin capsule (vitamin premix) and vitamin E-gel (composed of Natural Tocopherol, soya oil, Gelatin, Glycerin, water and preservatives) were purchased from a pharmaceutical shop and the palm oil was obtained from Ekiosa market in Benin City.

*Leucaena* seeds where harvested from the *L. leucocephala* shrubs around the Faculty of Agriculture, University of Benin. The seeds were sundried for two days to ensure all seeds were dried uniformly. A pot was placed on fire and heated up with low heat for 2 minutes before the seeds were introduced into the pot. They were stirred continuously till the seed coat became reddish brown in colour and emitting a sweet aroma. They are then removed and allowed to cool before milling to flour and there-after sieved. Fresh soybean seed where poured into a preheated pot, toasted, milled and sieved in a similar manner as was carried out with the *Leucaena* seeds. Six isonitrogenous and isocaloric diets with a crude protein level of 40% were formulated. Diets 1 (control), 2, 3, 4, 5, 6 had soybean meal protein substituted with TLLS meal at 0%, 10%, 20%, 30%, 40%, 50% respectively. The composition of the experimental diets is shown in Table 1.

The required quantity of ingredient for each of the diet were weighed and mixed into a homogenous mixture with the exception of corn meal. The component of corn meal in each diet was gelatinized (boiled in water to form gel), which served as a binder was poured on the homogenized mixture and mixed properly. They were all made into pellets with a pelleting machine available in the Department of Fisheries experimental farm at the University of Benin. The diets were dried using Altona smoking kiln in the department of fisheries experimental farm and then stored in air-tight containers throughout the experimental period.

**Table 1. Percentage composition of experimental diets for *Heteroclarias* post-fingerlings (%)**

| Ingredients | Diets | | | | | |
|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 |
| Fishmeal | 25.00 | 25.00 | 25.00 | 25.00 | 25.00 | 25.00 |
| Soybean | 54.00 | 44.00 | 34.00 | 24.00 | 14.00 | 4.00 |
| Leucaena seed meal | 00.00 | 10.00 | 20.00 | 30.00 | 40.00 | 50.00 |
| Maize | 3.00 | 3.00 | 3.00 | 3.00 | 3.00 | 3.00 |
| Wheat offal | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 |
| Palm oil | 8.00 | 8.00 | 8.00 | 8.00 | 8.00 | 8.00 |
| Bone meal | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 | 4.00 |
| Vitamin premix | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Vitamin E (gel) | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 | 1.00 |
| Estimated crude protein (%) | 40.00 | 40.00 | 40.00 | 40.00 | 40.00 | 40.00 |
| Analysed crude protein (%)* | 47.75 | 42.5 | 45.50 | 35.50 | 35.00 | 36.75 |

** Based on the A. O. A. C. method (1990). Vitamin E-gel composition: Natural Tocopherol, Soya oil, Gelatin, Glycerin, water and preservatives*

## 2.2 Heteroclarias Post Fingerlings

Heteroclarias post-fingerings (initial mean body weight of 48.66 ± 0.125 g) were obtained from the outdoor fish tanks of the Department of Fisheries Experimental Farm, University of Benin, Benin City.

## 2.3 Feeding Trials

The experimental design consisted of 6 dietary treatments with 3 replicates per treatment. Diets 1 with 0% TLLS inclusion served as control. The study was conducted in the Departmental of Fisheries wet laboratory, University of Benin using 18 plastic aquaria tanks (30 cm×36 cm×52 cm), each tank was filled with water up to 2/3 of its volume with bore-hole water attached to the laboratory. *Heteroclarias* post fingerlings from the same brood stocks were stocked randomly at 5 post fingerlings per aquarium and fed to satiation crumbled pellets of the experimental diets for 8 weeks, split between 2 feedings per day at 8:00 - 9:00 hrs and 15:00 - 16:00 hrs. Aquaria were cleaned daily with replacement of up to 2/3 of the water on each occasion. All fish per aquarium were weighed in bulk and recorded at weekly intervals to monitor growth parameters, weekly feed intake was also recorded. A complete change of water was made during the weekly weighing of fish.

## 2.4 Growth Parameters

Determination of growth parameters such as Weight gain (WG), Feed intake, Specific growth rate (SGR), Relative weight gain (RWG), Food conversion ratio (FCR), Protein efficiency ratio (PER) and Survival rate were computed using the following formular;

1. Weight gain (WG) = $W_2 - W_{1\,(g)}$

   Where; $W_1$ = initial weight
   $W_2$ = final weight

2. Feed intake = initial weight of feed – final weight of feed

3. Specific growth rate (SGR) %/day = $\frac{\text{Loge W2} - \text{loge W1}}{\text{T2} - \text{T1}} \text{ X } 100$

   Where: $T_1$ and $T_2$ are time of experiment in days.
   $W_2$ = final weight at $T_2$
   $W_1$ = initial weight at $T_1$
   Loge = natural logarithm.

4. Relative weight gain (RWG) % = $\frac{\text{Weight Gain}}{\text{Initial Weight}} \text{ X } 100$

5. Food conversion ratio (FCR) $= \frac{\text{Feed Intake(g)}}{\text{Wet Weight Gain(g)}}$

6. Protein efficiency ratio (PER) $= \frac{\text{Weight Gain (g)}}{\text{Protein Intake}}$

7. Survival rate % $= \frac{\text{Initial stocked} - \text{mortality}}{\text{Initial stocked}} \text{X } 100$

The data so generated were analyzed by one-way ANOVA (analysis of variance) using Genstat 12th edition analytical package. Differences in mean were compared by Duncan's multiple range test.

## 3. RESULTS

The growth response and nutrient utilization data evaluated (Table 2) displayed an irregular trend at various substitution levels. The weight gain was significantly different (P>0.05) among the treatments, the highest weight gain was recorded in control Diet with mean weight gain of 7.714 g but was not significantly different (P > 0.05) from diet II which had a mean weight gain of 7.31 g, the least amount of weight gain was however recorded in diet IV which has a weight gain of 1.798 g. Feed intake in diets I and II were not significantly different (P > 0.05) from each other but both were significantly superior to diets III, IV, V and VI. Relative weight gain showed no significant difference (P > 0.05) between all the diets. Specific growth rate in control diet and Diet II showed no significant difference (P > 0.05) from each other but was however significantly differently from diets III, V, VI and IV. There was significant difference (P > 0.05) in terms of feed conversion ratio of all the diets, the FCR was uneven with increasing inclusion levels. Protein efficiency ratio showed no significant difference (P > 0.05) between all the diets. The % survival of the *Leucaena leucocephala* seed meal-fed post fingerlings was comparable to that of the control, thus the values gotten indicate that inclusion levels did not affect the survival of the fish.

## 4. DISCUSSION

This study clearly demonstrated that the growth rate and nutrient utilization varied with different inclusion levels of leucaena seed meal. This variation is in agreement with [14], who

**Table 2. Growth response and nutrient utilization of *Heteroclarias* post fingerling fed *Leucaena leucocephala* seed meal based diets**

| Parameter | Diets | | | | | | SEM |
|---|---|---|---|---|---|---|---|
| | I | II | III | IV | V | VI | |
| | 0% | 10% | 20% | 30% | 40% | 50% | |
| Mean weight gain (g) | $7.71^{a}$ | $7.31^{a}$ | $5.18^{ab}$ | $1.80^{c}$ | $4.11^{bc}$ | $3.75^{bc}$ | 1.49 |
| Feed intake (g) | $11.01^{a}$ | $10.99^{a}$ | $9.03^{b}$ | $8.22^{b}$ | $9.06^{b}$ | $8.27^{b}$ | 0.61 |
| Percentage weight gain (%) | $10.53^{a}$ | $10.08^{a}$ | $9.89^{a}$ | $8.61^{a}$ | $7.70^{a}$ | $6.72^{a}$ | 2.07 |
| Specific growth rate (%) | $1.41^{a}$ | $1.50^{a}$ | $1.13^{ab}$ | $0.27^{b}$ | $1.07^{ab}$ | $1.05^{ab}$ | 0.40 |
| Feed conversion ratio | $1.43^{d}$ | $1.50^{cd}$ | $1.74^{bc}$ | $4.57^{a}$ | $2.20^{abc}$ | $2.21^{ab}$ | 0.51 |
| Protein efficiency ratio (%) | $0.82^{a}$ | $0.82^{a}$ | $0.67^{a}$ | $0.29^{a}$ | $2.27^{a}$ | $0.55^{a}$ | 1.02 |
| Survival rate (%) | 93.33 | 100 | 100 | 73.33 | 93.33 | 100 | - |

*Mean values in each row with the same superscript are not significantly different (P > 0.05)*
*SEM = standard error of mean*

**Table 3. Proximate composition (%) of experimental diets**

| Proximate composition | Treatment | | | | | |
|---|---|---|---|---|---|---|
| | I | II | III | IV | V | VI |
| Moisture content (%) | 10.44 | 14.72 | 10.24 | 10.75 | 12.73 | 13.23 |
| Crude protein (%) | 47.75 | 42.5 | 45.5 | 35.5 | 35.00 | 36.75 |
| Ether extract (%) | 26.12 | 27.22 | 26.52 | 28.12 | 25.42 | 25.32 |
| Crude fibre (%) | 5.30 | 5.43 | 5.43 | 2.13 | 2.13 | 2.22 |
| Ash % | 8.52 | 8.22 | 8.75 | 8.32 | 8.31 | 8.76 |
| N. F. E (%) | 2.07 | 1.91 | 3.56 | 15.18 | 16.41 | 13.72 |

reported that the growth and nutrient utilization of diets by fish decreased as level of toasted *Leucaena leucocephala* seed inclusion increases in the diets. This may have been as a result of non-inclusion of essential amino acid 'methionine' which is known to be limiting in both test ingredients (soybean and *leucaena*) [15]. Heat treatment is known to detoxify anti-nutrients but retarded growth and nutrients utilization recorded in this study may have been as a result of the presence of heat resistance anti-nutrients [16] as this was in line with the findings of [9], who reported that heat treated *Leucaena* seeds gave lower performance than *Leucaena* soaked in water and sundried. Heating of the seeds could have also resulted in the destruction of the amino acid bonds thereby reducing the protein quality of the feed ingredients. In conclusion, the result obtained from this study showed that Diet I with 0% inclusion of toasted *Leucaena leucocephala* seed was the best but this was not significantly different from Diet II with 10% inclusion level which performed best among the other Diet that had toasted *Leucaena leucocephala* seed present in it. Since weight gain of fish is what would translate into income for the fish farmer at the end of the production cycle, 10% inclusion rate of toasted *Leucaena leucocephala* seed in catfish diet would produce better and profitable result at present.

## 5. CONCLUSION

The result obtained from this study showed that control diet had the best response but this was not significantly different from Diet II with 10% inclusion level which performed best among the other Diet that had LSM present in it. Therefore, from the study carried out, the recommended level of LSM is 10% for catfish Hybrid (*Heteroclarias*) since they performed better.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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