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Utilization of the larva of the emperor moth, *Cirina forda* (Westwood, 1849) in the diet of *Clarias gariepinus* (Burchell, 1822) fingerlings

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Abstract
This study was done to investigate the utilisation of the Emperor moth larva (*Cirina forda*) as a replacement for fishmeal as a protein source in the diet of *Clarias gariepinus* fingerlings. The proximate composition of the larva of *Cirina* was determined according to the methods of Association of Official Analytical Chemists, AOAC (2003) to be 49% crude protein, 17.55% lipid, 8.57% moisture, 4.83% fibre, 20.05% carbohydrate and 5.54% ash. One hundred and twenty fingerlings of *C. gariepinus* were fed four diets formulated to contain 40% crude protein for 56 days. The diets were prepared by replacing fishmeal with *C. forda* larva meal at 50%, 75% and 100% in triplicate with a control diet without the larva meal. The diets were fed twice a day. The highest final mean weight and mean weight gain were obtained in diet 3 containing 100% *Cirina forda* while the least values were recorded in Control diet of 0% inclusion of *Cirina forda*. *Clarias gariepinus* can tolerate and utilize adequately 100% *Cirina forda* as a protein source without any inclusion of fishmeal.

Keywords: Mperor moth, *Cirina forda*, *Clarias gariepinus*

Introduction
One of the important factors to consider in fish farming is nutrition. The nutritive value of fish diet depends on quality of the protein ingredients used in the diet formulation. Protein is the most expensive component in fish feeds and the fishmeal is the major source of protein in fish diet. Fishmeal is not only expensive but also a scarce commodity due to its large demand. The global production of fishmeal has remained relatively stable over the last decade (1995-2005), and supplies are unlikely to improve (Lunger *et al.*, 2006) [5]. The increasing cost of fishmeal has led many to look for cheaper and available and suitable substitute to fishmeal.

The fishmeal is highly nutritious digestible, and palatable. Any intended substitute must meet the nutritional demand of the fish similar to fishmeal. Plant based protein sources are much cheaper than the animal-based protein sources, however, they are deficient in essential amino acids especially lysine and methionine. Plant proteins also contain endogenous anti-nutritional factors which reduces digestibility and lower fish growth. Edible insect such as the emperor moth, *Cirina forda* larvae is superior to plant proteins and possess the nutritional qualities needed by fish and so can effectively and efficiently replace the fishmeal.

Cirina forda larva is rich in protein, fats, carbohydrate, ash, and low in moisture. It is highly digestible, high in tryptophan and has a relatively balanced amino acid profile which makes it a good protein source for the fish feed industry. Its high ash content is a reflection of the high mineral it contains (Yohanna *et al.*, 2014) [10]. It is rich in potassium, sodium and calcium which are essential minerals lacking in most animal and plant protein sources (Omotoso, 2006) [7]. These qualities of *Cirina forda* larva meets the nutrient requirements that support optimum fish growth.

The African catfish, *Clarias gariepinus* is an omnivorous, hardy fish and tolerant to a wide range of environmental conditions. These qualities make it a good candidate for fish culture in Nigeria. This study provides information on the use of insects such as *Cirina forda* larva as a protein source in the diet of fish. The objectives of this study were to determine the proximate composition of *Cirina forda* larva and the growth performance of *Clarias gariepinus* fed the larva meal as replacement to fishmeal diets.

Materials and Method

Experimental Site

The experiment was carried out at the Department of Fisheries and Aquaculture hatchery Unit, University of Agriculture, Makurdi, Benue State for 56 days.

Collection of larva and fish

The Emperor moth *Cirina forda*, larva was collected from the wild in Apir village, Makurdi, Benue State. This was washed and sundried for three days, ground into powder and stored in air-tight containers. A total of 120 fingerlings of *Clarias gariepinus* were obtained from Aqua haven fish farm, North bank, Makurdi.

Diet formulation and preparation

Pearson's square method was used to formulate a 40% crude protein diets with ingredients of maize, groundnut cake, fishmeal, premix, bone meal, palm oil, starch, and salt purchased from the Wadata market, Makurdi, Benue State. The experimental diet was formulated to replace fishmeal with *C. forda* larva at four (4) inclusion levels of 50% *C. forda* larva (Diet 1), 75%, (Diet 2) and 100% (Diet 3). A control diet contained 100% fishmeal and 0% *C. forda* larva (Table 1)

Table 1: Composition of the Experimental diets

Ingredient	Control	1	2	3
Fish meal	67.7	38.85	16.90	0
Cirina forda	0	38.85	50.80	67.7
Maize	14.3	14.3	14.3	14.3
Groundnut cake	10	10	10	10
Bone meal	2	2	2	2
Palm oil	2	2	2	2
Starch	1	1	1	1
Salt	1	1	1	1
Premix	2	2	2	2
Total	100	100	100	100

Experimental set-up

The feeding of the fish was done in twelve (12) plastic 40L tanks with 30 litres of water. The fish were acclimatized for 48hours before the feeding trial commenced. In each tank C. *gariepinus* was stocked at 10mfish per tank and fed their allocated diets twice a day at 09:00 h and 15:00 h replicates. The fish were fed to satiation each feeding time. Faecal samples and uneaten feed was siphoned from each tank daily. Water quality parameters of temperature, dissolved oxygen, pH, electrical conductivity and total dissolved solids were determined weekly. Fish weight and length was also measured weekly.

Proximate analyses of *Cirina forda* larva, Experimental diets and Fish

The proximate composition of the larva, experimental diets and fish were done in the Animal Science laboratory of Federal University of Agriculture using procedures of AOAC (2003) [2].

Growth evaluation parameters

Growth performance characteristics determined were:

Mean Weight Gain (MWG) = Final mean weight (g) – initial mean weight (g)

$$\text{Percentage Weight Gain (PWG)} = \frac{\text{Mean Weight Gain}}{\text{Final Mean Gain}} \times 100$$

$$\text{Growth rate (GR)} = \frac{\text{Mean Weight Gain}}{\text{Number of Days}}$$

$$\text{Specific Growth Rate (SGR \%/day)} = \frac{100(\log eW2 - \log eW1)}{\text{Number of Days}} \times 100$$

$$\text{Feed Conversion Ratio (FCR)} = \frac{\text{Total feed consumed}}{\text{Mean Weight Gain}}$$

$$\text{Feed Conversion Efficiency (FCE)} = \frac{\text{Weight Gain}}{\text{Weight of Feed consumed}} \times 100$$

$$\text{Protein Efficiency Ratio (PER)} = \frac{\text{Weight Gain}}{\text{Protein Consumed}}$$

$$\text{Survival rate (SR)} = \frac{\text{Number of fish stocked}}{\text{Number of fish harvested}} \times 100$$

Statistical analysis

Evaluation of growth performance and nutrient utilization parameters were assessed by one-way analysis of variance (ANOVA) using SPSS 17.0. Significant differences among means were determined using Fishers least significant difference (F-LSD) test.

Results

Proximate composition of *Cirina forda* larva

The proximate composition of *Cirina forda* larva used in this experiment contained 49% crude protein, 17.55% lipid, 8.57% moisture, 4.83% fibre, 20.05% carbohydrate and 5.54% ash. (table 2)

Table 2: Proximate Composition of *Cirina forda* larva in the experiment

Composition	Percentage (%)
Crude protein	49.00
Lipids	17.55
Moisture	8.57
Crude fibre	4.83
Carbohydrate	20.05
Ash	5.54

Analysis of the experimental diets

The mean proximate composition of the experimental diets is shown in table 3. The moisture content was highest in diet 1 (7.70±0.01) and least in the control diet (6.79±0.61) while the ash was highest in the control diet (6.55±0.01) and least in diet 3 (5.46±0.01). The lipid content was highest (16.87±0.01) in diet 3 and least (10.62±0.01) in the control diet. The crude protein was found highest (50.02±0.01) in the Control diet and lowest (43.13±0.01) in diet 3. The fibre content was highest (4.25±0.01) in diet 3 and lowest (1.57±0.01) in the control diet. Carbohydrate content of the diets was highest (26.03±0.01) in the Control diet and lowest (22.38±0.01) in diet 3. (Table 3)

Table 3: Analysed Proximate Compositions of the Experimental Diets

Composition%	Control	50	75	100	P-Value
Protein	$50.02{\pm}0.01^{d}$	$47.48{\pm}0.01^{c}$	$44.26{\pm}0.01^{b}$	$43.14{\pm}0.01^{a}$	0.00
Lipids	$10.62{\pm}0.01^{a}$	$12.68{\pm}0.01^{b}$	$14.08{\pm}0.01^{c}$	$16.87{\pm}0.01^{d}$	0.00
Fibre	$1.57{\pm}0.01^{a}$	$2.92{\pm}0.01^{b}$	$3.58{\pm}0.01^{c}$	$4.25{\pm}0.01^{d}$	0.00
Carbohydrate	$26.03{\pm}0.01^{d}$	$24.08{\pm}0.01^{b}$	$24.95{\pm}0.01^{c}$	$22.38{\pm}0.01^{a}$	0.00
Ash	$6.55{\pm}0.01^{c}$	$5.49{\pm}0.01^{b}$	$5.47{\pm}0.01^{a}$	$5.46{\pm}0.01^{d}$	0.00
Moisture	$6.79{\pm}0.01^{a}$	$7.42{\pm}0.01^{b}$	$7.66{\pm}0.01^{c}$	$7.70{\pm}0.01^{d}$	0.00

means on the same row with different superscripts differ significantly ($P<0.05$)

Keys: Control: 100% fishmeal, Diet 1: 50% *Cirina forda* larva, Diet 2: 75% *Cirina forda* larva, Diet 3: 100% *Cirina forda* larva

Growth performance and nutrient utilization of the fish

The growth performance and nutrient utilization of *Clarias gariepinus* fingerlings fed control diet, diet 1, diet 2, and diet 3 at P-level (0.05) is shown in table 4. Fish fed control diet had the highest (0.45±0.00) initial mean weight while those fed diet 2 were the least (0.37±0.03). For the final mean weight, fish fed diet 3 had the highest (7.66±0.32) while fish fed the control diet had the least (3.62±0.00). Fish fed diet 3 had the highest mean weight gain of 7.04±0.31 while the fish fed the control diet had the lowest mean weight gain of 3.17±0.00. Fish fed diet 3 had the highest percentage mean weight gain (94.44±0.23) while those fed the control diet had the least (87.57±0.00). Fish fed diet 3 had the highest growth rate (0.13±0.01) while those fed the control diet had the least (0.06±0.00). Fish fed diet 2 and 3 had the highest specific growth rate (0.05±0.00) while fish fed the control diet and diet 1 had the least (0.04±0.00). Fish fed the control diet had the best feed conversion ratio (1.68±0.00) while those of diet 3 were poorest (1.98±0.14). Fish fed the control diet had the best feed conversion efficiency (46.69±0.00) while those of diet 3 had the least (22.69±3.99). Fish fed diet 3 had the best protein efficiency ratio (2.26±0.09) while those of the control diet had the least (1.36±0.00). Fish fed diet 3 had the best survival rate (90±5.00) while those of the control diet had the least (80±0.00).

Table 4: Mean Growth Performance of *Clarias gariepinus* Fingerlings Fed Diets containing different inclusion levels of *Cirina forda* larva

Parameter	Control	Treatment 1	Treatment 2	Treatment 3	P-Value
MIW	$0.45{\pm}0.00^{c}$	$0.43{\pm}0.01^{b}$	$0.37{\pm}0.03^{a}$	$0.41{\pm}0.01^{ab}$	0.05
MFW	$3.62{\pm}0.00^{a}$	$4.32{\pm}0.33^{b}$	$4.94{\pm}0.04^{c}$	$7.46{\pm}0.32^{d}$	0.00
MWG	$3.17{\pm}0.00^{a}$	$3.89{\pm}0.33^{b}$	$4.57{\pm}0.05^{c}$	$7.04{\pm}0.31^{d}$	0.00
PMWG	$87.57{\pm}0.00^{a}$	$89.97{\pm}0.99^{b}$	$92.44{\pm}0.61^{c}$	$94.44{\pm}0.23^{d}$	0.00
GR	$0.06{\pm}0.00^{a}$	$0.07{\pm}0.01^{ab}$	$0.08{\pm}0.00^{b}$	$0.13{\pm}0.01^{c}$	0.00
SGR	$0.04{\pm}0.00^{a}$	$0.04{\pm}0.00^{a}$	$0.05{\pm}0.00^{b}$	$0.05{\pm}0.00^{b}$	0.00
FCR	$1.68{\pm}0.00^{a}$	$1.73{\pm}0.10^{ab}$	$1.69{\pm}0.11^{ab}$	$1.88{\pm}0.14^{b}$	0.00
FCE	$69.58{\pm}0.00^{c}$	$57.89{\pm}0.05^{b}$	$61.67{\pm}2.32^{bc}$	$54.15{\pm}0.08^{a}$	0.00
PER	$1.36{\pm}0.00^{a}$	$1.54{\pm}0.02^{b}$	$1.57{\pm}0.03^{b}$	$2.26{\pm}0.09^{c}$	0.00
Survival	$80.00{\pm}.00^{a}$	$83.33{\pm}3.33^{b}$	$83.33{\pm}3.33^{b}$	$90.00{\pm}5.00^{b}$	0.00

*mean on the same row with different superscripts differ significantly

Keys: Control: 100% fishmeal inclusion, Treatment 1: 50% *Cirina forda* larva inclusion level, Treatment 2: 75% *Cirina forda* larva inclusion level, Treatment 3: 100% *Cirina forda* larva inclusion level, MIW: Mean Initial Weight; MFW: Mean Final Weight, MWG: Mean weight Gain, PMWG: Percentage Mean weight Gain, GR: Growth Rate, SGR: Specific growth rate FCR: Feed Conversion Ratio, FCE: Feed Conversion Efficiency, PER: Protein Efficiency Ratio

The weekly growth rate of the fish is depicted in figure 1. Diet 3 had the highest growth while control diet had the least growth rate.

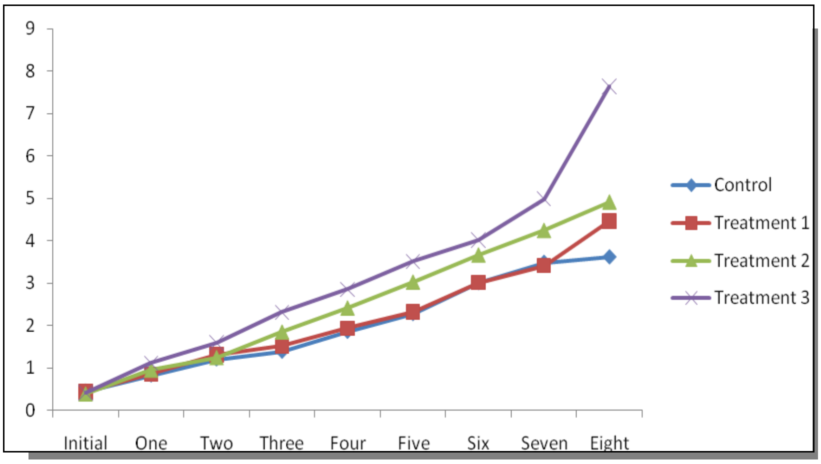

Fig 1: Weekly Weight of *Clarias gariepinus* fed different inclusion levels of *Cirina forda* larva Meal.

Water quality

There was no significant differences ($P>0.05$) in the water quality parameters considered during this study. They however differ in numerical values. Electrical Conductivity was found highest in Control treatment (750.50±50.50) and least in treatment 2 (722.00±53.68), Total Dissolved Solids was highest in Control (375.00±25.00) and least in treatment 2 (360.00±24.79), Temperature was highest in treatment 3 (24.50±1.97) and lowest in Control (22.60±2.50), pH was highest in treatment 1 (10.03±0.15) and least in treatment 3 (9.91±0.16), and Dissolved Oxygen was highest in treatment 2 (3.95±0.05) and least in Control (3.88±0.17)

Table 5: Mean Water Quality Parameters

Parameters	Control	Treatment 1	Treatment 2	Treatment 3	P-Value
Electrical Conductivity	750.50±50.50^a	722.33±52.68^a	722.00±53.68^a	743.00±75.20^a	0.98
Total Dissolved Solids	375.00±25.00^a	360.00±24.79^a	362.33±27.11^a	371.00±37.75^a	0.98
Temperature	22.60±2.50^a	24.37±2.26^a	24.33±2.06^a	24.50±1.97^a	0.94
pH	9.99±0.01^a	10.03±0.15^a	9.97±0.11^a	9.91±0.16^a	0.93
Dissolved oxygen	3.88±0.17^a	3.95±0.05^a	3.95±0.04^a	3.91±0.02^a	0.84

means on the same row with different superscripts differ significantly ($P<0.05$)

Proximate Composition of the Experimental Diets

All the proximate parameters differed significantly ($p<0.05$) across the treatments. The moisture value (10.22±0.01) was highest in initial and lowest (5.22±0.01) in treatment 3. NFE was highest (44.42±0.01) in initial and lowest in control (19.01±0.01). Crude protein and ash values were highest (61.69±0.01 and 8.36±0.01) in control and lowest (38.41±0.01 and 1.53±0.01) in initial, while lipid and fibre values were highest (7.13±0.39 and 2.32±0.01) in treatment 3 and lowest in initial (4.03±0.01 and 1.44±0.01).

Table 6: Proximate composition of the Experimental fish fed different levels of *C. forda* larva meal.

Treatments	Moisture	Ash	Lipid	Fibre	Protein	NFE
Initial	10.22±0.01^e	1.53±0.01^a	4.03±0.01^a	1.44±0.01^a	38.41±0.01^a	44.42±0.01^e
Control	6.18±0.42^c	8.36±0.01^e	5.99±0.01^b	1.47±0.01^b	61.69±0.01^e	19.01±0.01^a
1	6.24±0.01^b	6.48±0.01^b	6.79±0.01^c	1.82±0.01^c	56.23±0.00^d	21.99±0.01^b
2	5.65±0.01^b	6.90±0.01^c	7.04±0.01^d	2.03±0.01^d	54.24±0.01^c	24.19±0.01^c
3	5.22±0.01^a	7.14±0.30^d	7.13±0.39^e	2.32±0.01^e	50.80±1.26^b	31.10±1.57^d
P-Value	0.00	0.00	0.00	0.00	0.00	0.00

Means on the same column with different superscript differ significantly ($p<0.05$)

Keys: Treatment 1: 50% *Cirina forda* larva inclusion level, Treatment 2: 75% *Cirina forda* larva inclusion level, Treatment 3: 100% *Cirina forda* larva inclusion level.

Discussion

The crude protein obtained was 49.00%. This was within the protein range of 16-60% for Lepidopteran caterpillars (Ramos-Elorduy *et al.*, 1998) [9] and 55.5% (Omotoso, 2006) [7]. Since the dietary crude protein requirement in the diets of catfish fingerlings is 40%, FAO (2017) [3], this larva could serve as a protein source in the diets of catfish fingerlings and adults.

Low moisture content of *Cirina forda* larva is an indication of a good shelf characteristic. The moisture content of the larva of *C. forda* in this experiment was 8.57%. The low moisture in this larva shows that if incorporated in fish feed, it can be stored for a long time without deterioration (Yohanna *et al.*, 2014) [10].

The lipid of 17.554% was obtained from this work. It has been shown that *C. forda* lipids are richer with the important unsaturates than any animal lipids (Ande, (2003) [1] This is good in fish diet as it will increase the palatability of fish diet and provide energy for fish, thus saving the protein for growth.

The crude fibre content of 4.83% found the larva in this work is low and is good in fish diet. Fish are known to utilize feedstuff with low fibre content. The low fibre content is an indication that the larva can be properly digested by fish.

The carbohydrate content of 20.05% was obtained from emperor moth in this work. Fish utilizes carbohydrates for energy in fish diet.

The ash content in this larva indicates the amount of minerals in it. Ash content of 5.54% obtained in this work shows that the larva can provide the minerals needed in fish diet for the wellbeing of the fish (Yohanna *et al.*, 2014) [10]. All the diets analyzed were within the recommended proportion and composition of fish diets for *C. gariepinus* fingerlings. The content of crude protein, lipid, fibre, carbohydrate, ash, and moisture in the diets, meets the nutritional requirements of the fish (NRC, 2011) [6]. The results obtained from this report shows that *Clarias gariepinus* fingerlings can utilize high inclusion levels of *C. forda* larva in their diets. the fish had better mean weight gain (MWG), growth rate (GR), specific growth rate (SGR), and protein efficiency ratio (PER) with increasing inclusion levels of *C. forda* larva. Diet 3 which had 100% inclusion level of *C. forda* larva had the best growth performance. This result was similar with the report of Oso and Ola-Oladimeji (2016) [8], who reported that *C. gariepinus* grows better with increasing inclusion levels at 25%, 75%, and 100% of *C. forda* larva feed. The high growth performance of *C. gariepinus* at increasing inclusion level of *C. forda* larva, suggests that larva of the emperor moth in the diets contains a protein level which is comparable and even higher in quality to that present in the conventional fishmeal (Oso and Ola-Oladimeji, 2016) [8].

Fish fed control diet (with 100% fishmeal inclusion) had a feed conversion ratio of 1.68±0.00. Which was not differ significantly from the fish fed Diet 1 and 2. The fish fed Diet 3 (100% *C. forda* larva), however, had a feed conversion ratio of 1.98±0.14% which had the best growth performance. It has been shown that *C. gariepinus* fingerlings utilizes *C. forda* larva effectively and efficiently. All the diets in this work had no effect on the quality of water as there were no significant differences in all the water quality parameters measured.

Conclusion

This work has indicated that the larva of *C. forda* larva, is rich in nutrients especially protein (49%) and can serve as a protein source in fish diets. It has also shown that inclusion levels of a 100% *C. forda* are possible. The lipid, ash, fibre, carbohydrate, and moisture content in the larva were within the range recommended for use in fish feed.

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