

Original Research Article

Growth performance and nutrient utilization efficiency of *Clarias gariepinus* juvenile fed *Bombyx mori* (Mulberry silkworm) meal as a partial replacement for fish meal

ABSTRACT

A 56-day (Eight weeks) trial was carried out to determine the growth performance and nutrient utilization of *Clarias gariepinus* juveniles fed *Bombyx mori* meal (BMM) as a replacement for fishmeal. Five experimental diets were formulated at various level of inclusion of 25%, 50%, 75% and 100% of BMM designated as diets B, C, D, E respectively. The control diet (diet A) was 100% fishmeal as control (A), 25% BMM inclusion (B), 50% BMM inclusion (C), 75% BMM inclusion (D) and 100% BMM inclusion (E). The experimental fish were collected from.....and stocked in with stocking density of and -with the fish fed x times a day at 5% of their body weight. The effect of the diets on the growth, feed utilization efficiency, carcass and mineral compositions of the fish were evaluated. The results showed that mean weight gain, specific growth rate and protein efficiency ratio of fish fed the experimental diets differed significantly ($P < 0.05$) when compared with the control. In this study, replacement of fishmeal with 25% BMM gave the best result while 100%, 75% and 50% gave better result than the control. Also, carcass yield and mineral depositions differed marginally in the fish fed the various diets. It can be inferred from this study that *B. mori* larvae have the potential to be used as a protein source and can replace and elicit better growth trends in fish more than the conventional fish-meal without any problem with growth.

Keywords: Growth response, Nutrient utilization, *Bombyx mori*, *Clarias gariepinus*

INTRODUCTION

The rapid world-wide expansion of aquaculture to meet up with the global demand of fish is an indication of an impending pressure on the aqua-feed industries in the near future. Since many fisheries are overfished, aquaculture is recognized as the only feasible option to meet the increasing demands for aquatic foods (Allan, 2004; Chebbaki *et al.*, 2010). Feed is one of the major inputs in aquaculture production, and however, fish feed technology is one of the least

32 development sector of aquaculture particularly in Africa and other developing countries of the
33 world (Gabriel *et al.*, 2007). It was reported by Ahmad and Diab (2008) that the feeding cost in
34 aquaculture represents over 50% of the operation cost of fish farming. Fishmeal as a raw material
35 is the choice in aquaculture due to its high quality protein with balanced amino acid profile
36 (Gatlin *et al.*, 2007). According to Goytortu'a, (2006), the production of fishmeal has been
37 relatively stable in the last two decades but the increasing demand ~~could not be matched in the~~
38 ~~present world~~ due to increased aquaculture. Moreover, the cost of fishmeal is increasing which
39 renders the use of alternative protein source which are local, cheap and unsuitable for direct
40 human consumption inevitable to make up for the shortage of fishmeal and secure the stable
41 supply for commercial feeds (Hardy, 2006).

42 Many works have been done on the nutritional value of many alternative feed ingredients to
43 supplement the dietary fishmeal-based diets. Several researches have shown that alternative
44 protein sources derived from grains such as corn, wheat, barley, soy bean, cotton seed, fishmeal,
45 and animal rendering products such as mechanically deboned meal, poultry by-product meal,
46 meat and bone meal, maggot, Bambara groundnut, etc. can be used to formulate nutritious
47 aquaculture feed at various levels of inclusions in different feeds. For instance, Yang (2004)
48 reported that diets with 20% of fishmeal replaced by meat-and-bone meals produced growth
49 equivalent to the control diet in *Gibel carp*. Kasper *et.al.*, (2007) also reported that up to 47.6%
50 of the fishmeal could be replaced with soy-bean meal in the diets of yellow perch (*Perca*
51 *flavescens*) without affecting the feed consumption, weight gain, feed efficiency ~~or and~~ survival
52 of the fish. *Nibeia miichthioides* and *Carassius auratus gibelio* grew well on diets with 50% of
53 the fishmeal replaced by poultry- by- product meal (Wang *et al.* 2006; Yang *et al.*, 2004). Also,
54 Oso *et al.*, (2013) showed that *Clarias gariepinus* fingerlings fed up to 75% inclusion of
55 Bambara groundnut in fish feed was satisfactorily acceptable without affecting growth and feed
56 utilization.

57
58 Moreover, insects are important source of food to many animal species including man (Adedire
59 and Aiyesanmi, 1999). Their larvae may constitute a cheaper source of essential nutrients that are
60 easily available and affordable to the natives within the localities where the insect larvae are
61 found and could form a base for new food products of considerable nutritive value (Ekpo and
62 Onigbinde, 2007). *Bombyx mori* can be used for the production of silk and are ~~also~~ proteinaceous

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63 | in nature. The larvae undergo spinning, vomiting the silk fibre into liquid substance to form
64 cocoon so that they will not emerge out and develop into adult stage called moth. The dead
65 larvae in the cocoon are removed by riling machine and the silk is separated while the dead
66 larvae are used for production of fish feed. The objective of this study is therefore to determine
67 the optimum level of fishmeal replacement with *Bombyx mori* as protein source in the diet of
68 *Clarias gariepinus*. The aim is to evaluate the growth performance of *Clarias gariepinus*
69 juvenile fed *Bombyx mori* as a protein source at different levels of inclusion so as to make
70 cheaper and qualitative supplementary feeds available to culture fish species with a view to
71 bridging the gap between protein supply and consumption through reduction in cost of
72 production in the aquaculture sector.

73

74 MATERIALS AND METHODS

75

76 **Experimental Site-** The experiment was carried out at the postgraduate research laboratory of
77 Zoology Department, Ekiti State University, Ado- Ekiti for a period of 56 days (eight weeks)
78 from June 1 to July 26, 2012. Sixteen plastic bowls of 20 litres water holding capacity were used
79 for the experiment. Water was maintained at 17 litres level throughout the experiment and a
80 mosquito mesh was used to cover the bowls to preclude predators from entering into the bowls
81 and fish jumping out as well.

82

83 Feed Preparation:

84 The feed ingredients used for this study were purchased from Metro Vet Consultant Limited in
85 Ekiti State while the ~~except~~ *Bombyx mori* ~~which was~~ purchased from Ekiti State Sericulture
86 Project. The larvae of *Bombyx mori* were killed during the process of cooking so that they were
87 ~~ill not emerge out of stay in~~ the cocoon ~~to and thus, the silk was not avoid~~ damaged ~~to the silk~~.
88 Inside the cocoon, the larvae were oven dried at 65⁰C and then removed from the cocoon. Later
89 it was pulverized and analyzed for its proximate composition according using to the procedure
90 used by ~~of~~ APHA (2006) as shown in Table 1. Feed ingredients were finely ground and mixed
91 together in appropriate proportions after which the mixture was pelleted (1.8mm diameter.) and
92 then sun dried.

Comment [KBW2]: Why sixteen, as you have five treatments (5x3=15)

Comment [KBW3]: It was better to dry at room temperature since sun drying affect it nutritional content

Table 1: Proximate Composition of *Bombyx mori* larvae

Composition Components	% in larvae
% dry matter	87.50
% crude protein	69.52
% crude fibre	5.01
% ether extracts	11.12
% ash	7.88

Experimental Fish- *Clarias gariepinus* juvenile were used for the experiment. One hundred and fifty mixed sex juvenile with average weight of 7.20g were obtained from a hatchery in Ado-Ekiti. The fish were acclimatized for two days in a plastic bowl during which they were not fed in order to empty their gut and prepare their appetite for the new feed. The fish were randomly stocked at the rate of ten (10) juveniles per bowl. The initial mean weight of fish in each bowl was taken and recorded. Fish were fed at 5% of their body weight twice ~~a day~~ on daily basis, that is, morning ~~from~~ (08:00 – 09:00 am) and evening ~~from~~ (17:00 – 18:00 pm). Subsequent weight and standard length measurements were taken every week and the rations of the feed were adjusted accordingly to fish weight gain. Mortality of the fish was recorded daily. In addition, water in the experimental bowls was renewed totally on daily basis to remove unconsumed food particles and faecal materials and thus good water quality parameters were maintained.

Experimental Design and Diets – The experiment consisted of five treatments with three (3) replicates. Each of the five experimental diets contained *Bombyx mori* as fishmeal substitute except the control. All the experimental diets were formulated to contain common basal diets and different proportions of *Bombyx mori* as shown in Table 2. Each of the diets was analyzed to

determine the proximate composition. The various diets were formulated to contain 0%, 25%, 50%, 75% and 100% *Bombyx mori* designated as Diets A (control), B, C, D and E respectively.

Table 2: Gross composition of experimental diets (Kg)

INGREDIENTS	DIET A Control 0%	DIET B 25% <i>B. mori</i>	DIET C 50% <i>B.mori</i>	DIET D 75% <i>B. mori</i>	DIET E 100% <i>B. mori</i>
Fish meal	10	7.5	5	2.5	–
<i>B. mori</i>	–	2.5	5	7.5	10
Maize	11	11	11	11	11
Rice bran	7	7	7	7	7
Blood meal	16	16	16	16	16
Soya bean	15	15	15	15	15
Groundnut	38	38	38	38	38
Bone meal	1	1	1	1	1
Salt	1.33	1.33	1.33	1.33	1.33
Lysine	0.37	0.37	0.37	0.37	0.37
Methionine	0.1	0.1	0.1	0.1	0.1
Vitamin C	0.1	0.1	0.1	0.1	0.1
Vitamin premix	0.1	0.1	0.1	0.1	0.1
Total	100	100	100	100	100

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Data collection and analysis

Sampling- All the fish were sampled every week to measure their body weight using manual weighing balance. Body length was measured using a measuring board to the nearest 0.1cm. Mortality of the fish was also recorded daily. In addition, physico-chemical parameters including temperature, pH and dissolved oxygen were monitored weekly during the experiment. Based on the data collected, growth performance, feed utilization efficiency and survival rate of fish were calculated as:

Weight gain (WG) = Final weight of fish – Initial weight of fish

Percentage weight gain (%WG) = [(Final weight – Initial weight)/ Initial weight] x 100

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163 Percentage specific growth rate (%SGR) = $[(\text{Log } W_2 - \text{Log } W_1) / (T_2 - T_1)] \times 100$

164 Feed conversion ratio (FCR) = Dry weight of feed/ Total weight gained by fish

165 Protein efficiency ratio (PER) = Total weight gained by fish/ Protein intake

166 Where:

167 W_2 = Weight of fish at time T_2 (final)

168 W_1 = Weight of fish at time T_1 (initial)

169 T = Period of experiment in days

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171 ~~Feed conversion ratio (FCR) = Dry weight of feed/ Total weight gained by fish~~

172 ~~Protein efficiency ratio (PER) = Total weight gained by fish/ Protein intake~~

173 Statistical Analysis

174 The growth performance and feed utilization parameters were analyzed by univariate analysis
175 using SPSS version 17. Univariate analysis is able to analyze two parameters simultaneously to
176 see if there are strong and meaningful links between them. The significant difference used was at
177 the 0.05 level of probability.

178

179 RESULTS

180 The mean values of growth rate, feed conversion ratio and protein efficiency ratio of *Clarias*
181 *gariepinus* juveniles fed the five experimental diets (A, B, C, D and E) for eight weeks were
182 calculated and summarized in Table 3. The result shows that weight gain, % weight gain, the
183 specific growth rate (SGR), protein efficiency ratio and food conversion ratio were significantly
184 affected by the type of feed provided for the fish. The highest weight gain was recorded in fish
185 fed with diet B (3.05) followed by diet E (2.85) while the lowest value was recorded in diet C
186 (2.33). Fish fed with diet B had the highest increase in percentage body weight gain (45.05)
187 followed by diet E (38.41). The lowest value of 31.14 was recorded in diet A. A similar trend
188 was observed for the specific growth rate and protein efficiency ratio in which the highest mean
189 of 2.00 and 0.086 was recorded with diet B followed by diet E (1.76 and 0.080). Fish fed with
190 diet A revealed the least S.G.R. and protein efficiency ratio (1.47 and 0.064) respectively. The

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191 | highest value of feed conversion ratio (4.35) was achieved with the fish fed diet D followed by
192 | diet C (3.84) in feed conversion ratio. The lowest value (2.79) was recorded with diet E.

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197 | Table 3: Mean growth performance and feed utilization Efficiency of *Clarias gariepinus*
198 | juvenile.

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Parameters	Diet A	Diet B	Diet C	Diet D	Diet E
Initial weight	7.29 ^a	6.77 ^b	7.17 ^a	7.33 ^a	7.42 ^a
Final weight	9.56 ^b	9.82 ^{ab}	9.50 ^b	9.77 ^{ab}	10.27 ^a
Weight gain	2.27 ^{de}	3.05 ^a	2.33 ^d	2.44 ^c	2.85 ^b
% Weight gain	31.14 ^d	45.05 ^a	32.50 ^{cd}	33.29 ^c	38.41 ^b
Specific growth rate	1.47 ^d	2.00 ^a	1.53 ^{bc}	1.56 ^{bc}	1.76 ^b
Protein efficiency					
Ratio	0.064 ^c	0.086 ^a	0.065 ^c	0.068 ^c	0.080 ^{ab}
Feed conversion ratio	3.55 ^c	3.12 ^d	3.84 ^b	4.35 ^a	2.79 ^e

211 | Mean values with the same alphabets across rows are not significantly different at $P > 0.05$

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214 | The Pproximate composition of all the experimental feed is summered shown in tTable 4. The
215 | ash content was very high in diet A and low in diet D with 11.95 and 9.84 respectively, while the
216 | percentage fat was high in diet E (3.70) and low in diet A (3.563). The highest and lowest crude
217 | protein was recorded in diet A (37.70) while 35.46 was recorded for both diets D and E. Diets C
218 | and B gave the highest (7.37 and 6.74) and lowest (6.40 and 6.42) for moisture content and
219 | percentage fibere respectively. The percentage soluble carbohydrate was high in diet B (37.73)
220 | and low in diet A (35.21).

221

222 | Table 4: Pproximate composition of experimental feed

Parameters	Diet A	Diet B	Diet C	Diet D	Diet E
Ash	11.95 ^a	10.24 ^b	11.35 ^{ab}	9.84 ^c	10.23 ^b
Moisture	6.54 ^d	6.40 ^e	7.37 ^a	6.86 ^c	7.22 ^b
Crude protein	35.70 ^a	35.60 ^b	35.50 ^c	35.46 ^d	35.46 ^d
Fat	3.53 ^c	3.63 ^b	3.58 ^b	3.60 ^b	3.70 ^a
Fibere	6.54 ^b	6.42 ^c	6.74 ^a	6.64 ^{ab}	6.57 ^b

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231	CHO	35.21 ^d	37.73 ^a	35.76 ^c	36.25 ^b	37.72 ^a
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233 | Mean values with the same alphabets across rows are not significantly different at P> 0.05

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237 | The proximate composition of *Clarias gariepinus* juvenile flesh is ~~summarized~~shown in Ttable

238 | 5. The highest ash content was recorded in diet D (16.44) and the lowest in diet E (15.49), while

239 | the highest moisture content was recorded in diet A (11.41) and lowest in diet D (10.26). Diet E

240 | had the highest crude protein (67.54), while the lowest was recorded in diet D (66.34). The

241 | highest and lowest percentage fat was recorded in diet E (5.76) and C ~~with 5.76 and (5.39)~~

242 | respectively. The percentage soluble carbohydrate was very high in diet C (1.80) and low in diet

243 | D (0.42). Low mortality was also observed in all the treatments throughout the period of the

244 | experiment. Diet D had the highest mortality of 13.33%, while Diet B recorded the lowest rate of

245 | 6.67%.

246

247 | Table 5: Proximate composition of experimental fish

248						
249	Parameters	Diet A	Diet B	Diet C	Diet D	Diet E
250						
251	Ash	15.35 ^c	16.39 ^a	15.35 ^c	16.44 ^a	15.49 ^b
252	Moisture	11.41 ^a	11.38 ^a	10.64 ^{ab}	10.26 ^b	10.69 ^{ab}
253	Crude protein	66.49 ^c	66.79 ^b	66.62 ^b	66.34 ^d	67.54 ^a
254	Fat	5.59 ^b	5.42 ^c	5.39 ^c	5.57 ^b	5.76 ^a
255	CHO	0.86 ^b	0.41 ^d	1.80 ^a	0.42 ^d	0.53 ^c
256						

257 | Mean values with the same alphabets across rows are not significantly different at P> 0.05

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259

260 **DISCUSSION AND CONCLUSION**

261 In the present study, fish fed the various diets showed increase in weight without any external

262 sign of nutritional deficiency even though growth performances of fish fed the *Bombyx mori*

263 diets with up to 100% replacement varied and even better in terms of weight gain, % weight

264 gain, specific growth rate and protein efficiency ratio when compared with the control. This

265 shows the possibility of using *B. mori* as alternative protein source in the diet of *C. gariepinus* as

266 it contains all the necessary nutrients that are required for growth. Orilogbon and Adewole

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(2011) reported that insect meals have a higher level of methionine, lysine and valine compared to soybean meal. The **present** study showed significant differences ($P < 0.05$) in growth performance of *Clarias gariepinus* fed the experimental diet at various inclusion of *B. mori* as diet B, E, D and C were shown to be more significant than diet A (control) in terms of weight gain, percentage weight gain, specific growth rate and protein efficiency ratio. **This result agrees with the work of Olaniyi and Babasanmi (2013) on silkworm meal used as a replacement for fishmeal in the diet of African Catfish up to 100%.** The best performance of fish was **observed in** 25% inclusion of BMM. This supports the work of Sogbesan *et.al.* (2006) that replacement of fishmeal with 25% maggot meal gave the best performance in the fish. Also, more than 25% BMM inclusion gave a better growth performance when compared with control. This contradicts the report made by Sogbesan *et.al.* (2006) that higher replacement levels than 25% **magmeal** gave a lower growth.

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The *Clarias gariepinus* juvenile fed diet D (i.e. 75% inclusion of *Bombyx mori*) showed superior feed conversion ratio. This agreed with the findings of Davies & Gouveia (2010) who worked on the dietary nitrogen utilization in African cat fish fed oil seed meal diets under sub-optimal growth condition consistent with a restricted regime. It was stated that fish fed the experimental diets showed superior feed conversion ratio. The result of the proximate composition of carcass of fish fed experimental diets showed significant difference at various levels of *B. mori* inclusion. This however disagreed with the work done by Nwanna (2003) where he reported no significant difference ($P < 0.05$) in the carcass composition and mineral deposition of *Clarias gariepinus* fed chemically preserved shrimp head waste silage diets. Also, the higher lipid deposits in the carcass of fish fed experimental diets showed better growth in terms of weight which may be due to lipid rather than crude protein gain. This is supported by the report of Adeparusi and Agbede (2004) on growth, nutrient utilization and carcass crude protein deposit in *Oreochromis niloticus* fed diets with leaf protein cake which showed that leaf proteins made from *Gliricidia* and *Leucaena* could be viable means of improving fish feed especially for herbivorous fish like *O. niloticus*.

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~~Also, the~~ The result of this study **also** showed the preference of *Clarias gariepinus* juvenile for small amount of lipids in their diet for growth because the highest **growth performance** was observed in fish fed diet B. This may be attributed to the fact that the lipids of animal origin such

298 as silkworm have shorter chain and lower degree of fatty acids rather than Omega 6-fatty acid in
299 contrast to terrestrial animal, which require long chain of omega 6- fatty acids (Kanazawa,
300 1993).

301
302 In conclusion, although 25% gave the best result in this present study, however 100%, 75% and
303 50% levels of replacement of fish meal protein *Bombyx mori* in the diet of *Clarias gariepinus*
304 juvenile are acceptable to the fish without affecting the growth and feed utilization. This will
305 ultimately reduce the cost of fish meal to be included in the diet and consequently reduce the cost
306 of fish production. It can be said that whatever was responsible for a drop in weight gain and
307 feed utilization of fish fed diet C (50% *B. mori* inclusion) could be taken care of by using a better
308 processing method. Moreover, *B. mori* replacement can be tested on later stage of the fish to
309 make a general conclusion.

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