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Effect of feed energy on testes biochemical characteristics of *Clarias jaensis* (Boulenger, 1909) endogenous catfish in Cameroon

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Abstract
This study aims to investigate the effects of four diets with different energy level; 3000, 3100, 3200 and 3300 Kcal/kg, on testes biochemical characteristics of *Clarias jaensis*. 360 male and female (97 ± 9 g) were divided into four groups, and fed with diets during 5 months. 12 males were sacrificed per group at the end to determine the biochemical characteristics of their testes. The high value of body weight (263.00 ± 29.70 g) and total length (32.63 ± 0.85 cm) were recorded at 3100 Kcal/kg ($P<0.05$). Testes weight were significantly higher at 3000 Kcal/kg. Except the total cholesterol (8.91 ± 1.11 mg/dl) and LDL (2.31 ± 0.01 mg/dl), other biochemical characteristics of testes were significantly higher at 3000 Kcal/kg and the low values were recorded with other energy level. Therefore; 3000 Kcal/kg was found to be the optimal energy level for a good quality of *Clarias jaensis* testes.

Keywords: *Clarias jaensis*, energy, testes, biochemical characteristics

1. Introduction
The fish farming in Cameroon is mainly based on the production of exogenous species like *Clarias gariepinus*, *Oreochromis niloticus*, etc. However, there are endogenous species that have the ability to better adapt to rearing conditions and can be produced at different levels of intensifications (Cacot and Lazard, 2009) [1]. This is the case of *Clarias jaensis* which is currently suffering from uncontrolled exploitation by peasants for profit and food purposes (Zango, 2009) [2]. However, its domestication could help to preserve it and support sustainable aquaculture development like any other local species. To enhance this specie, it is important to have the knowledge of its nutrition and reproduction technics, which are essential for breeding. In fact, nutrition play an important role on fish reproduction. It have a profound effect on gonadal growth and fecundity (Gamage, 2001) [3]. The relationship between nutrition and adult maturation is still little know. Most of the research work conducted on broodstock emphasized factors which effect gonad maturation and spawning behaviour, such as induced spawning by hormone injection, selective breeding, hybridization and environmental factors. However, there is very little information on the effect of nutrition on the biochemical characteristics of the gonads, yet the cholesterol who comes from the food is at the basis of the production of steroids hormones like testosterone and estradiol (Martin, 1985 and Gower, 1988) [4, 5]. The choice of the study of the level of energy in this work is explained by the fact that, it is important in the phenomenon of reproduction. Thus, according to Barnabé (1991) [6], gonad maturation does not occur in emaciated fish that do not have sufficient mobilizable reserves. The work of çek and Yilmaz (2009) [7] and Reidel *et al*. (2010) [8] showed that the energy level in the feed affects the reproductive performance of *Clarias gariepinus* and *Rhamdia quelen*; In *Clarias jaensis*, the main work carried out focused on the evaluation of its reproductive and growth characteristics in wild and captivity (Zango, 2009; Efole *et al*., 2016; Zango *et al*., 2016 and Zango *et al*., 2017) [2, 9, 10, 11]. There is no information on the effect of nutrition (feed energy) on gonad quality especially biochemical characteristics of this specie.
The general objective of this work, is to contribute to a better knowledge of endogenous fish by determining their nutritional needs for their preservation and enhancement. More specifically, evaluate the effect of energy level on the biochemical characteristics of *Clarias jaensis* testes.

2. Materials and methods
2.1 Period and zone of the study
The study took place between May and July 2016 in the west region of Cameroon and at the Application and Research Farm of the University of Dschang (LN 5 ° 44'-5 ° 36 'and 5 ° 44'-5 ° 37'; LE 10 ° 06'-9 ° 94 'and 10 ° 06'-9 ° 85' at an altitude of 1392 -1396 m.

2.2 Animal material
A number of 360 post fingerlings of male and female *Clarias jaensis* with an average weight of 97 ± 9g were fished in the natural environment in the Santchou Area Rivers, located in the western Cameroon region and transported in nurseries to the FAR of the University of Dschang. The fish were immediately acclimatized for two weeks in the 1 m^3 (1 x 1 x 1) concrete tanks. During the acclimation period, a 3A feed containing 40% of protein and consisting of wheat bran,

soybean meal and fishmeal (Lacroix 2004) was distributed to them twice a day.

2.3 Livestock structures
Twelve polystyrene happas of 1 m^3 (1 x 1 x 1), attached to four sides chinese bamboo were built in a pond of 100 m^2 area and depth 0.90 m. This pond was supplied with water from a dam pond located at 150 m. The height of water in each happa was 0.80 m.

2.4 Experimental diet
Four isoproteic experimental diet (Table 1) were formulated with different energy levels 3000, 3100, 3200 and 3300 kcal/kg of food. The chosen energy levels refer to those of *Clarias gariepinus.* Except the metabolize energy who were calculated by the formulation table, the analyzed chemical composition of different diet were obtained by AOAC (2000) [12] method.

Table 1: Composition of expérimental diets

Ingredients (Kg)	R1 (3000 kcal/kg)	R2 (3100 kcal/kg)	R3 (3200 kcal/kg)	R4 (3300 kcal/kg)
Maize	28.00	24.50	22.50	20.30
Wheat bran	6.00	5.10	3.50	3.80
Cottonseed cake	1.50	1.30	1.00	1.50
Soybean meal	3.00	4.40	6.00	4.50
Fish meal	56.70	57.00	56.50	57.00
Shell meal	0.10	0.10	0.10	0.10
Bone meal	0.10	0.10	0.10	0.10
Palm oil	2.60	5.50	8.30	10.70
Premix 2%	2.00	2.00	2.00	2.00
Total	100.00	100.00	100.00	100.00
chemical composition (% /MS)				
CP (%)	39.76 ± 0.49	40.06 ± 0.75	40.66 ± 0.37	40.56 ± 0.50
ME (kcal/kg) (kcal/kg)	3000.21	3102.69	3200.62	3303.65
CE (kcal/kg) (kcal/kg)	3284.67 ± 25.40	3354.67 ± 39.50	3780.00 ± 22.09	3848.00 ± 28,35
Lipid (%)	7.00 ± 0.00	11.33 ± 0.28	12.00 ± 0.00	15.33 ± 0.57
Ash (%)	2,93 ± 0.05	1.96 ± 0.05	2.70 ± 0.17	2.06 ± 0.05

CP= Crude protein, ME= Metabolize energy, CE= Crude energy, P/E= Protein/energy ratio, Ca/P= Calcium/phosphorus ratio

2.5 Experimental set up
Post fingerlings of *Clarias jaensis* were randomly divided into four comparable groups (45 ♂ and 45♀/group) of size and weight with three replication. At each group was randomly assigned one of the experimental diet R1 (3000 kcal/kg), R2 (3100 kcal/kg), R3 (3200 kcal/kg) and R4 (3300 kcal/kg of energy) previously formulated.

2.6 Conduct of the test and data collected
The food was distributed twice a day (8 am and 18 pm) at 3% of the ichtyobiomasse. Each month, a check fishery was conducted during which 20% of the fish in each group were individually weighed using a 0.1 g precision electronic scale (OHAUS) and measured with an icthyometer. The physico-chemical parameters of the water (temperature, dissolved oxygen, pH and conductivity) were measured weekly just to have the state of water. At the end of the experiment, 12 males from each group were sacrificed in the laboratory and the testes, seminal vesicles and fat were collected and weighed each. After this, one of the testes were crushed in a mortar placed on an ice block and containing 0.9% NaCl solution, so as to obtain homogenates 15%. The crushed material obtained was centrifuged at 3000 rpm for 30 min. The supernatant was removed and stored in labeled tubes at -20 ° C and were used for the determination of total protein and cholesterol, HDL (High density lipoprotein), LDL (Low density lipoprotein and TGS (Triglycerids) (Ngoula, 2008) [13]. These assays were performed using kits sold and ready for use, at the Laboratory of Physiology and Animal Health of the University of Dschang.

2.7 Studied characteristics
Condition factor K2 = 100 x (Live weight – Gonads weigh)/ Total length3 Fat index FI = Fat weight / Live weight

2.8 Statistical analysis
The one-way ANOVA was used to test the effect of energy level on K2 factor, testes and seminal vesicles weight, fat index and biochemical characteristics of testes. When there were significant differences between the means, they were separated by the Duncan test at 5% significance level. SPSS 20.0 statistical software was used for the analyzes.

3. Results
3.1 Effect of feed energy on weight and total length of male *Clarias jaensis*
Table 2 show some growth characteristics of male of *Clarias jaensis*. It appears that, feed energy has significantly affect body weight and total length. These characteristics were highest at 3100 Kcal/kg and lowest with ration at 3300 Kcal/kg except total length ($P<0.05$).

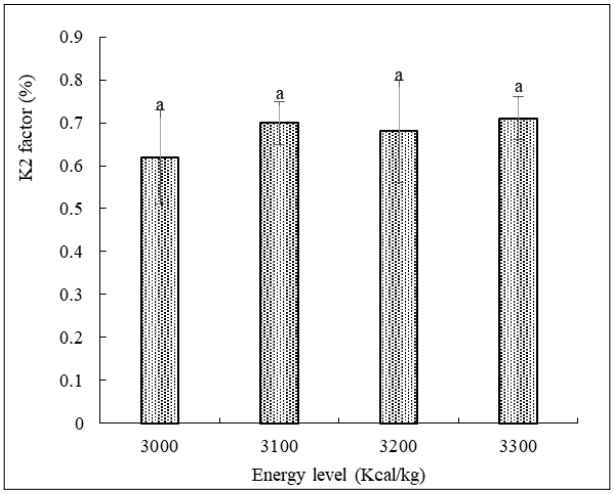
Table 2: Mean weight and total length of male *Clarias jaensis* in function of energy level

	Live weight	Energy level (Kcal/kg)		
and total length	**3000**	**3100**	**3200**	**3300**
LW (g)	197,75 ± 37,87[a]	263,00 ± 29,70[b]	206,61 ± 33,14[a]	194,50 ± 10,00[a]
TL (cm)	28,71 ± 3,69[a]	32,63 ± 0,85[b]	30,55 ± 1,89 [c]	29,43 ± 0,83 [ca]

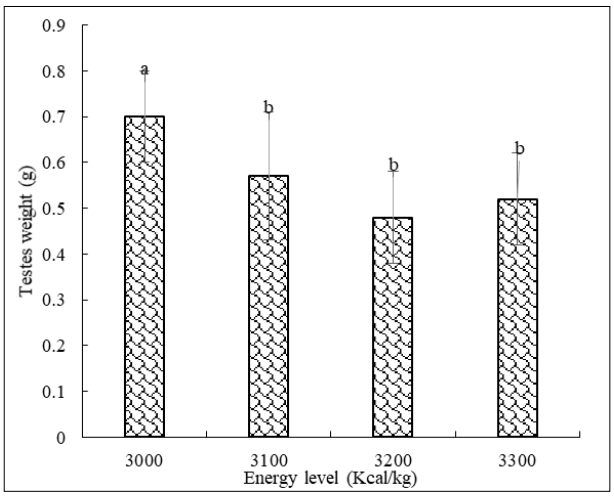
a, b and c : Means with same letter on same line are not significantly different (P>0,05) LW= Live weight, TL= Total length

3.2 Effect of feed energy on K2 factor, testes and seminal vesicles weight and fat index of male *Clarias jaensis*

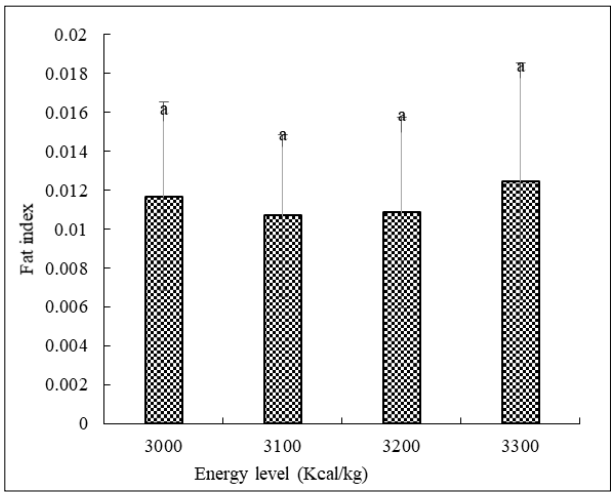
The figure 1 a, b, c and d show that the feed energy had a significant effect only on testes weight (P<0.05). The group fed at 3000 Kcal/kg recorded the higher testes (0.70 ± 0.10 g) and seminal vesicles (1.60 ± 0.28 g) weight and the lower value (0.48 ± 0.10 g) and (1.25 ± 0.96 g) respectively for testes and seminal vesicles weight were obtained at 3200 Kcal/kg. The condition factor K2 (0.71 ± 0.05 %) and fat index (0.012 ± 0.006) were most higher with ration containing 3300 Kcal/kg and the lower values were recorded at 3000 and 3100 Kcal/kg respectively for K2 factor (0.64 ± 0.11 %) fat index (0,010 ± 0,004).



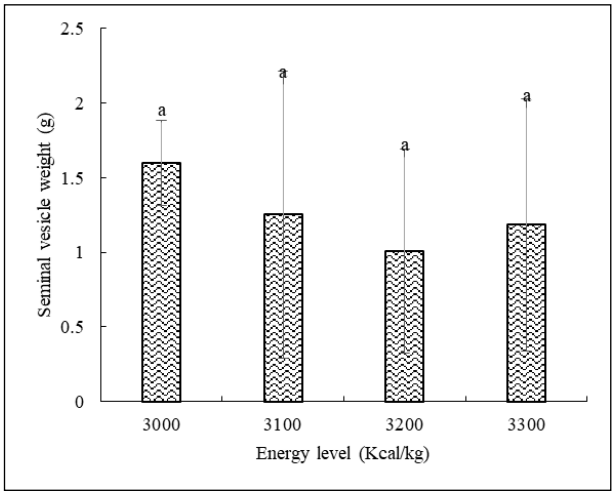
A



B



C



D

Fig 1: K2 factor (a), testes weight (b), fat index (c) and seminal vesicles weight (d) in function of feed energy a and b : Means with same letter are not significantly different (P>0,05)

3.3 Effect of feed energy on biochemical characteristics of *Clarias jaensis* testes

The biochemical characteristics of testes were significantly (P<0.05) affected by feed energy as presented in table 3. The total protein concentration were the most high at 3000 Kcal/kg, while HDL and TGS concentration were higher at 3100 Kcal/kg and lower with other energy level. Total cholesterol and LDL concentration were lower at 3000 Kcal/kg and the higher 3200 Kcal/kg.

Table 3: Biochemical characteristics of *Clarias jaensis* testes in function of feed energy

Biochemical Characteristics	Energy levels (Kcal/kg)				
	3000	3100	3200	3300	P
TP (g/dl)	0,73 ± 0,26[a]	0,31 ± 0,09[b]	0,38 ± 0,11[b]	0,41 ± 0,04[b]	0,04
T Chol (mg/dl)	8,91 ± 1,11[a]	11,48 ± 1,83[b]	11,89 ± 2,08[b]	11,48 ± 0,93[b]	0,01
HDL (mg/dl)	8,60 ± 0,71 [a]	9,24 ± 1,25 [a]	7,25 ± 1,14 [a]	3,71 ± 0,29 [b]	0,02
LDL (mg/dl)	2,31 ± 0,01 [a]	3,00 ± 0,01 [ab]	6,57 ± 0,97 [c]	5,03 ±1,45 [bc]	0,01
TGS (mg/dl)	31,10 ± 10,60 [a]	32,21 ± 9,02 [a]	11,58 ± 4,16 [b]	18,71 ± 5,10 [ab]	0,04

A, b and c : Means with same letter on same line are not significantly different (P>0,05) Totales Proteins, T Chol= Total Cholestérol, HDL = High density lipoprotein, LDL = Low density lipoprotein, TGS = Triglycerids, P = probability

4. Discussion

The higher weight and total length were recorded with the lowest energy level (3100 Kcal/kg). This result is similar to that found by Çek and Yilmaz (2009) [7] in *Clarias gariepinus* fed various diatary energy, who showed that the best growth characteristics are obtained with feed containing a few energy level. The condition factor were not significantly affect by energy level. But, it was lower at 3000 Kcal/kg while testes and seminal vesicle weight were the most high in the same group. This result can be explained by the fact that, the fish would have taken the reserves necessary for the maturation of its gonads in the muscles. Regardless of the energy level, the K2 factor was lower than 1, this showing that the fish were not well (Fulton, 1909) [14]. Çek and Yilmaz (2009) also find that the weight of male gonad of *Clarias gariepinus* is most high with the treatment containing the less energy level. This is also supported by Hernandez *et al*. (2001) [15] and Keembiyetty and Wilson (1998) [16] who showed that feed with a poor energy level allows a good gonad development. The high value of seminal vesicles weigh would be due to the fact that, energy level would have led to a high production of nutrients, useful for the maturation of spermatozoa. In this work, the mean value of seminal vesicles weight were higher than that found in *Hemibagrus nemurus* by Sularto *et al*. (2010) [17] and in *Clarias gariepinus* by Gbemisola and Adebayo (2014) [18]. Total protein were the most high at 3000 Kcal/kg, HDL and TGS were first higher at 3100 and 3000 Kcal/kg where the testes weight were also higher. We did not find information on the effect of energy level on testes biochemical characteristics but, the high level of total protein concentration may be due to the maturation of testes and mobilization of protein in the form of lipoprotein. Indeed, the proteins concentration increase significantly during the sexual maturation process (Yeganeh, 2011) [19]. Total cholesterol and LDL were the lower at 3000 Kcal/kg and the higher 3200 Kcal/kg. This can be explained by the use of total cholesterol and LDL in the production and transport of steroid hormone like testosterone in the group fed with 3000 Kcal/kg (Martin, 1985 and Gower, 1988) [4, 5].

5. Conclusion

In general, the studied characteristics in this work were significantly affected by the energy level in the diet. The high live weight and total length were recorded at 3100 Kcal/kg while the best testes and seminal vesicle weight and biochemical characteristics of testes were observed in fish fed diet 3000 Kcal/kg. Therefore, on the basis of the present results among energy levels tested, 3000 Kcal/kg was found to be optimal for a good quality testes, despite the fact that the best growth characteristics were obtained with diet at 3100 Kcal/kg.

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