International Journal of Fisheries and Aquatic Studies 2024; 12(1): 56-62

E-ISSN: 2347-5129
P-ISSN: 2394-0506
(ICV-Poland) Impact Value: 76.37
(GIF) Impact Factor: 0.549
IJFAS 2024; 12(1): 56-62

www.fisheriesjournal.com
Received: 21-12-2023
Accepted: 26-01-2024

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Nutritional characteristics and costs of diets based on fish, spirulina, maggot and earthworm meals at the larval phase of rearing tilapia *Oreochromis niloticus*

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DOI: https://doi.org/10.22271/fish.2024.v12.i1a.2892

Abstract
This study was conducted to assess the nutritional characteristics and costs of diets formulated with fish, Spirulina, maggot and earthworm meals to feed tilapia *Oreochromis niloticus* at larval stage. The protein contents of these ingredients are 56.00±0.21 (fish), 40.34±0.13 (maggot), 40.7±0.22 (Spirulina) and 41.17±0.16% (earthworm). These different ingredients made it possible to formulate four types of diets. Diets based on maggot (MD), Spirulina (SD), earthworm (ED) and fish (FD) meals. The protein contents in each diet formulated are 40%. The costs per kilogram of formulated diets were evaluated. The highest value is obtained for SD (204116 FCFA) and the lowest for MD (296 FCFA). Biochemical composition, mineral composition, Fatty acid profile and Concentration of aflatoxins of experimental diets are evaluated. In conclusion, the total replacement of fish meal by those of Spirulina, maggot and earthworm did not negatively affect the nutritional quality of diet. The values reported are within the recommended limits for rearing this specie. In addition, the use of maggot and earthworm meal helped to reduce the cost per kilogram of feed produced, unlike fish meal. Spirulina meal increased the cost per kilogram of the formulated diet.

Keywords: Nutritional value, invertebrate feeds, spirulina feed, biochemical composition and economic parameters

1. Introduction
Fishmeal-based feed usually used to feed fish is expensive (current prices of fishmeal on the market, 300 to 700 FCFA/kg) and represents more than 50% of the cost of production in farms [1]. The high cost of this feed is often a source of abandonment of fish farming by low-income fish farmers and threatens the survival of aquaculture.
If fishmeal is used so far as the main source of protein in fish feed, it is because of its high protein content (51.1 to 72% dry matter) [2]. Fishmeal contains less than 12% lipids [2] with an essential amino acid profile that covers the needs of fish and the absence of antinutritional factors [3]. It is also a source of minerals and vitamins whose contents are variable [4]. Substitution of fishmeal by other inexpensive protein sources in fish diet is therefore necessary if the cost per kilogram of formulated diet is to be reduced. As a result, increase fish growth, but also decrease the cost per kilogram of fish produced. Nowadays, other protein sources are currently being tested in various research studies and can validly replace fishmeal. These include earthworms [5], fly larvae (maggots) [6, 7] and algae of the genus Arthrospira (Spirulina) [8]. According to Bamba *et al.* (2007) [9], a significant proportion of natural resources (algae and aquatic invertebrates) go into the diet of tilapia, hence the interest in incorporating them into fish feed. Their protein content and essential amino acid profiles are close to those of fishmeal. The crude protein (46 to 70% dry matter) and lipid (7 to 10% dry matter) contents of earthworm meal are comparable to those of fish meal and meat [5]. According to these authors, earthworms are rich in long-chain fatty acids and vitamins like fishmeal and meat. Additionally, earthworm meals have been used as additional protein in the diet of *Heterobranchus longifilis* [10]. As for maggots, their protein content is around 40 to 64% [11].

In addition, maggots are very rich in lipids, essential amino acids and minerals and their metabolizable energy is 3755 kcal [10]. Spirulina, on the other hand, has a protein content of between 34 and 73% [8]. Spirulina also contains high lipid levels (7-16% dry matter) [13]. In view of the many nutritional qualities of these algae and invertebrates which are almost similar to those of fishmeal, their incorporation into fish feed seems to be an opportunity to reduce the cost per kilogram of feed formulated in order to improve the growth of farmed fish and specially to reduce their production costs. The objective of this study is therefore to evaluate the nutritional characteristics and the costs of feed formulated with these protein sources (Spirulina, maggot and earthworm meals) for the rearing of *Oreochromis niloticus* in the larval phase.

2. Materials and Methods

2.1 Production of *Spirulina*, maggot and earthworm: Spirulina (*Arthrospira platensis*) used was produced in culture media prepared in three basins of capacity 198 liters by-each according to the method of Zarrouk. (1966) [14]. The biomass of the Spirulina harvested (Fig 1) was dried in the sun then crushed using the method of Fox. (1999) [15]. The dried Spirulina was crushed into powdery to obtain Spirulina meal. Housefly maggots used were produced in Ivory Coast from poultry droppings, pig manure and waste from fish evisceration following the description of Mpoame *et al*., (2004) [16]. The collected maggots (Fig 2) were killed in hot water, oven dried at 70 °C for 24 h and ground into powder to obtain maggot meal. The earthworm was produced according to the method of Sogbesan and Madu, (2003) [17]. The earthworm *Eudrilus eugeniae* (Fig 3) was cultured for three months. After these days, they are sorted, washed, killed with hot water and then dried in an oven at 80 °C and crushed into powdery to obtain earthworm meal.

Fig 1: Spirulina

Fig 2: Maggot

Fig 3: Earthworm

2.2 Experimental diets: Proportion (%) of ingredients used in the composition of experimental diets is shown in Table 1. Four isoproteic practical diets (40% crude protein content) were formulated with fishmeal, housefly maggot meal, Spirulina meal and earthworm meal as the main protein sources. Fishmeal being replaced totally with Spirulina (*Arthrospira platensis*), earthworms (*Eudrilus eugeniae*), and housefly maggots. These ingredients were included in diet at the level 18-20%. An industrial commercial diet used as the reference was purchased in local markets of Abidjan. Crude protein content of the commercial diet used at the same larval stage is 34.5%. The four formulated diets were designated as SD (diet containing Spirulina meal), MD (diet containing maggot meal), ED (diet containing earthworm meal) and FD (diet containing fishmeal). The commercial diet was designated as CD. All diets were prepared according to the method of Bamba *et al.* (2014) [18].

Table 1: Proportion (%) of ingredients used in formulated diets (FD, SD, MD and ED)

Ingredients (%)	Diets			
	FD	**SD**	**MD**	**ED**
Corn flour	8	5	5	5
Soybean meal	45	53	54	53
Cotton meal	14	14	14	14
Copra meal	6.5	3.5	4	3.5
Wheat bran	3.5	1.5	3	1.5
Fishmeal	20	--	--	--
Spirulina (Algae)	--	20	--	--
Maggot meal	--	--	18	--
Earthworm meal	--	--	--	20
Palm oil	2	2	1	2
Vitamins	0.25	0.25	0.25	0.25
Salt	0.30	0.30	0.30	0.30
Seashell flour	0.35	0.35	0.35	0.35
Lysine	0.05	0.05	0.05	0.05
Methionine	0.05	0.05	0.05	0.05
Total	100	100	100	100

FD = Fish Diet, SD = Spirulina Diet, MD = Maggot Diet, and ED = Earthworm Diet -- = Absents Ingredients.

2.3 Spirulina, maggot and earthworm meals cost estimate: The costs of the kg of Spirulina produced were calculated on the basis of the prices of the nutrients used for the preparation of the culture medium. As for the costs per kg of maggot and earthworm meal, they were calculated taking into account the cost of loading the gas cylinder used to heat the water that was used to kill the maggots and earthworms. On the one hand, and the price of the kg of sugar used to prepare the sugar water which was used to feed the flies on the other.

2.4 Analytical methods: The feed ingredients, experimental diets and fish samples were analyzed according to AOAC (1990) [19] for dry matter, crude protein, crude lipid, crude fiber, nitrogen free extract (NFE) and ash. The gross energy contents of the diets and fish samples were calculated using factors of 22.22, 38.9 and 17.2 $kJ.g^{-1}$ of protein, lipid and nitrogen free extract respectively [20]. The mineral assays were carried out using an atomic absorption spectrophotometer (VARIAN AA. 20) according to the techniques described by AOAC (2003) [21]. The IUPA (1987) [22] method was used for the separation of methyl esters from free and total fatty acids. Aflatoxins were determined according to AOAC (2005) [23] methods including the steps of extraction, purification on an immunoaffinity column and quantification using calibration after analysis by High Performance Liquid Chromatography.

2.5 Statistical analysis: Data on the biochemical composition of the experimental foods were compared by one-way analysis of variance (ANOVA, $\alpha = 0.05$). Differences were considered significant at $p<0.05$. The Tukey and Duncan tests were used for multiple comparisons of means when the effects were significant. All statistical treatments were carried out with STATISTICA 7.1 software at the significance level of $\alpha = 0.05$.

3. Results

3.1 Biochemical composition (% dry matter) and estimated costs of Spirulina, maggot and earthworm meals: The biochemical composition and the costs of Spirulina, maggot and earthworm used in the manufacture of the experimental foods are recorded in Table 2. The protein contents are 40.34±0.13, 40.7±0.22 and 41.17±0.16% respectively for the maggot, Spirulina and earthworm meal. The estimated costs per kg of these ingredients are 75, 115 and 1019167 FCFA.kg-1 for maggot, earthworm and Spirulina meal respectively. The highest cost is observed with Spirulina meal and the lowest with maggot meal.

Table 2: Biochemical composition (% dry matter) and estimated costs of Spirulina, maggot and earthworm meals used for the manufacture of experimental diets

Components	Spirulina, maggot and earthworm meals		
	SM	MM	EM
Moisture (%)	11.00±0.12^b	8.99±0.17^a	9.01±0.15^a
Crude Protein (%)	40.70±0.22^{ab}	40;34±0.13^a	41.17±0.16^b
Crude Lipid (%)	6.77±0.13^a	25.00±0.14^c	23.68±0.17^b
Ash (%)	15.14±0.10^b	10.12±0.14^a	11.28±0.09^a
Crude Fiber (%)	2.00±0.13^b	2.00±0.15^b	1.00±0.16^a
NFE	24.39±0.15^b	13.55±0.16^a	13.86±0.17^a
EC ($FCFA.kg^{-1}$)	1019167	75	115

Values are expressed as mean ± standard deviation. The mean values bearing the different alphabetical letters on the same line are significantly different at the threshold of σ=0.05. Duncan's test. SM = Spirulina meal, MM = Maggot meal, EM = Earthworm meal.
EC = Estimated cost of 1 kg of Spirulina, maggot and earthworm meals
Nitrogen free extract (NFE) = 100 – (% Moisture + % Protein+ % Ash +% Lipid + % Fiber).
Price in CFA pound: 100 CFA= 0.18 $ based on 2017 exchange prices in Ivory Coast

3.2 Biochemical composition and price of purchased *ingredients* (raw materials): The biochemical composition and the price of the raw materials used for the manufacture of the experimental diets are recorded in Table 3. The protein content is high with fish meal and low with corn meal. The observed difference is statistically significant ($p<0.05$) from one input to another. As for the lipid content, it is high for copra cake (6.95±0.17%) and low for cottonseed cake (2.04±0.15%). Statistical analysis of these different values shows a significant difference ($p<0.05$) from one input to another. As for the prices of raw materials, they are 700, 400 and 240 $FCFA.kg^{-1}$ respectively for fishmeal, soybean and cotton cake. The prices of other raw materials are 140, 110 and 100 FCFA. kg^{-1} respectively for maize flour, wheat bran and copra cake. The price per kg of fishmeal is high and the lowest price is obtained with copra cake.

Table 3: Biochemical composition (% of dry matter) and price of the *ingredient* (raw materials) used for the manufacture of experimental diets

C (%)	*Ingredients* (Raw Materials)					
	FM	CF	SM	CM	COM	WB
M	7.80±0.22^b	10.05±0.21^c	11;88±0.33^d	6.99±0.25^a	8.24±0.17^b	10.79±0.26^c
CP	56.00±0.21^f	11.80±0.19^a	45.00±0.13^e	41.56±0.15^d	21.00±0.14^c	15.30±0.12^b
CL	5.76±0.15^c	3.62±0.44^b	5.11±0.16^c	2.04±0.15^a	6.95±0.17^d	2.88±0.16^{ab}
Ash	18.00±0;24^d	1.57±0.33^a	6.10±0.22^c	5.60±0.24^{bc}	6.05±0.23^c	4.60±0.19^b
Fiber	0.00±0.00^a	1.00±0.13^b	3.00±0.15^c	11.00±0.16^e	16.00±0.13^f	9.00±0.14^d
NFE	12.44±0.11^a	71.96±0.14^f	28.91±0.12^b	32.81±0.13^c	41.76±0.15^d	57.43±0.14^e
Cost	700	140	400	240	100	110

Values are average from duplicate groups of samples.
FM= Fishmeal, C= Components, CF = Corn flour, SM= Soybean Meal, CM = Cotton meal, COM = Copra Meal, WB = Wheat Bran, M = Moisture, CP = Crude protein, CL = Crude lipid, NFE = Nitrogen free extract. Nitrogen Free Extract (NFE) = 100 – (% Moisture + % Protein + % Ash + % Lipid + % Fiber), Cost (FCFA) = Cost of 1 kg of ingredient (raw material) Price in CFA pound: 100 CFA= 0.18 $ based on 2017 exchange prices in Ivory Coast

3.3. Biochemical composition of experimental diets formulated: The biochemical composition of the different experimental diets is summarized in Table 4. The protein contents of the FD, SD, MD, ED foods vary from 39.37±0.22 (SD) to 39.81±0.23% (FD). Statistical analyzes of these different values show no significant difference ($p>0.05$) from one food to another. Crude protein content of the commercial diet (CD) used at the same larval stage is 34.50±0.18%. The lipid contents of FD, MD, ED and CD foods show no significant difference ($p>0.05$) from one food to another. On the other hand, the lipid content of the MD diet (6.17±0.10%) is higher ($p<0.05$) than those of the other diets.

Table 4: Biochemical composition (% Dry matter) of experimental diets

Components	Diets				
	FD	SD	MD	ED	CD
Moisture (%)	8.88±0.45[a]	11.14±0.48[b]	10.32±0.34[b]	9.14±0.37[a]	12.08±0.32[c]
Crude protein (%)	39.81±0.23[b]	39.37±0.22[b]	39.62±0.19[b]	39.46±0.17[b]	34.50±0.18[a]
Crude Lipid (%)	4.76±0.11[a]	4.14±0.09[a]	6.17±0.10[b]	4.77±0.14[a]	4.38±0.08[a]
Ash (%)	10.51±0.13[c]	9.14±0.16[b]	7.6±0.15[a]	11.04±0.17[c]	12.42±0.14[d]
Crud Fiber (%)	5.37±0.11[a]	5.68±0.15[a]	5.45±0.14[a]	5.56±0.10[a]	6.20±0.09[a]
NFE (%)	30.67±0.21[a]	30.53±0.19[a]	30.83±0.17[a]	30.03±0.15[a]	30.42±0.18[a]
Gross energy (kJ.g^{-1})	15.96±0.12[b]	15.60±0.15[b]	16.49±0.11[b]	15.78±0.10[b]	14.59±0.11[a]

FD = Fish Diet, SD = Spirulina Diet, MD = Maggot Diet, ED = Earthworm Diet and CD = Commercial Diet. NFE = Nitrogen free extract = 100 – (% Moisture + % Protein+ % Ash +% Lipid + % Fiber), Gross Energy = 22.2 × % Protein + 38.9 × % Lipid + 17. 2 × % Nitrogen free extract

3.4. Mineral composition of experimental diets formulated

The mineral compositions of the experimental diets are presented in Table 5. The calcium concentrations are between 17.75±0.12 (CD) and 23.85±0.12 g.kg^{-1} (FD). The highest value is obtained for FD and the lowest for CD. The difference observed is statistically significant ($p<0.05$) from one food to another. However, comparing the means two by two shows no significant difference between MD, ED and CD (Duncan's test, $p>0.05$). Phosphorus concentrations vary from 31.78±0.14 (CD) to 50.34±0.14 g.kg-1 (FD). The highest value is obtained for the FD and the lowest for the CD. The statistical analysis of these different values shows a significant difference ($p<0.05$) from one food to another. However, comparing the means two by two shows no significant difference between the AS and AV foods (Duncan's Test, $p>0.05$).

Table 5: Mineral composition of experimental diets (g.kg^{-1} diets)

Minerals	Diets				
	FD	SD	MD	ED	CD
Calcium	23.85±0.12[c]	20;33±0.09[b]	18.73±0.08[a]	18.53±0.11[a]	17.75±0.12[a]
Phosphorus	50.34±0.14[d]	40.38±0.13[b]	46.45±0.16[c]	41.87±0.12[b]	31.78±0.14[a]
Potasium	17.74±0.12[b]	25.46±0.11[c]	18.35±0.17[b]	16.24±0.08[a]	17.06±0.09[b]
Sodium	18.74±0.14[d]	26.78±0.13[e]	14.67±0.08[b]	12.33±0.09[a]	16.78±0.10[c]
Magnésium	19.43±0.10[d]	16.04±0.11[c]	14.43±0.11[b]	16.36±0.12[c]	8.56±0.13[a]
iron	0.77±0.06[b]	0.75±0.07[ab]	0.53±0.08[a]	0.84±0.10[b]	0.93±0.09[b]
Manganèse	0.10±0.04[a]	0.54±0.06[b]	0.44±0.08[b]	0.64±0.05[b]	0.76±0.06[b]
Zinc	0.69±0.06[a]	0.55±0.09[a]	0.73±0.08[a]	0.52±0.04[a]	0.63±0.06[a]
Copper	0.38±0.04[a]	0.37±0.05[a]	0.52±0.07[a]	0.34±0.07[a]	0.36±0.06[a]

3.5. Fatty acid profile of experimental diets formulated

The fatty acid compositions of the experimental foods are presented in Table 6 With regard to total saturated fatty acids, their proportions are high with ED (61.23±0.03%), SD (60.04±0.05%) and MD (59.13±0.05) and low for FD (56.42±0.04%) and CD (57.64±0.02%). The proportions of total unsaturated fatty acids are high ($p<0.05$) for FD (43.58±0.04%) and CD (42.36±0.06%) and low for ED (38.77±0.02%), SD (39.96±0.02%) and MD (40.87±0.05%).

Table 6: Fatty acid profile of experimental diets formulated

Fatty acids	Diets				
	FD	SD	MD	ED	CD
C12: 0	8.54±0.01[b]	6.56±0.02[a]	15.09±0.04[d]	10.90±0.01[c]	14.78±0.02[d]
C18:2 (n-6)	11.33±0.02[b]	12.49±0.02[c]	11.83±0.01[b]	10.03±0.03[a]	10.34±0.04[a]
C14:1 (n-5)	15.72±0.04[d]	11.6±0.03[b]	14.69±0.01[c]	8.63±0.03[a]	16.72±0.01[d]
C16:0	34.65±0.07[b]	34.38±0.06[b]	33.61±0.05[b]	32.3±0.01[a]	34.53±0.03[b]
C18: 1 (n-9)	16.53±0.02[b]	15.87±0.04[b]	14.35±0.04[a]	20.11±0.02[c]	15.31±0.03[b]
C18:0	13.23±0.02[b]	19.10±0.04[c]	10.43±0.05[a]	18.03±0.04[c]	11.33±0.03[a]
ST	56.42±0.04[a]	60.04±0.05[b]	59.13±0.05[b]	61.23±0.03[b]	57.64±0.02[a]
TU	43.58±0.04[b]	39.96±0.02[a]	40.87±0.05[a]	38.77±0.02[a]	42.36±0.06[b]

Values are expressed as mean ± standard deviation. The mean values bearing the different alphabetical letters on the same line are significantly different at the threshold of σ=0.05. Duncan's test. C12:0 =Lauric Acid, C18:2 (n-6) = Linoleic Acid; C14:1 (n-5) = Myristoleic Acid, C16:0 = Palmitic Acid, C18: 1 (n-9) = Oleic Acid, C18:0= Stearic Acid, TS = Total saturates; TU = Total unsaturates.

3.6. Concentration, limit and quantification of aflatoxins in the experimental diets formulated for the rearing of *O. niloticus:* The results of the determination of aflatoxins B1, B2, G1 and G2 by HPLC in the différents experimental diets are presented in Table 7. In the diets formulated with fishmeal, the concentrations of aflatoxin B1 are 0.0062 µg.kg^{-1} of diet with the total amount of aflatoxin which is 0.0062 µg.kg- 1 of diet. The concentrations of aflatoxins B2, G1 and G2 are below the quantification limits of these aflatoxins. With regard to the commercial diet Ivograin, the concentrations of aflatoxins B1 and G2 are below the quantification limits for these aflatoxins. These values are 0.00501 and 0.01882 µg.kg^{-1} respectively for aflatoxins B2 and G1 with a total concentration of aflatoxins in this food (CD) which is 0.02383 µg.kg-1 of diet. With regard to diets based on Spirulina, maggot and earthworm flours, no presence of aflatoxins B1, B2, G1 and G2 is observed.

Table 7: Concentration, limit and quantification of aflatoxins

Diets	Aflatoxins	Co (µg.kg-1)	AT (µg.kg-1)	DL (µg.kg-1)	QL (µg.kg-1)
FD	G-2	< QL	0.0062	0.00564	0.01878
	G-1	< QL		0.00151	0.0050
	B-2	< QL		0.00136	0.0045
	B-1	0.0062		0.00143	0.0047
SD	G-2	N.D.	ND	0.00564	0.01878
	G-1	N.D.		0.00151	0.0050
	B-2	N.D.		0.00136	0.0045
	B-1	N.D.		0.00143	0.0047
MD	G-2	N.D.	ND	0.00564	0.01878
	G-1	N.D.		0.00151	0.0050
	B-2	N.D.		0.00136	0.0045
	B-1	N.D.		0.00143	0.0047
ED	G-2	N.D.	ND	0.00564	0.01878
	G-1	N.D.		0.00151	0.0050
	B-2	N.D.		0.00136	0.0045
	B-1	N.D.		0.00143	0.0047
CD	G-2	0.01882	0,02383	0.00564	0.01878
	G-1	< QL		0.00151	0.0050
	B-2	< QL		0.00136	0.0045
	B-1	0,00501		0.00143	0.0047

FD = Fish Diet, SD = Spirulina Diet, MD = Maggot Diet, and ED = Earthworm Diet
AT = Aflatoxin totals, Co = concentration, DL = Detection Limit, QL= Quantization Limit, ND= Not detected

3.7 Financial evaluation of experimental diets formulated: The costs of the different experimental diets and the rates of reduction in cost per kilogram of formulated diets are summarized in Table 8. The highest value is obtained for SD and the lowest for MD. The use of maggot and earthworm meal contributed to reduce the cost per kilogram of the formulated diet by 26 (MD) and 22.50% (ED) respectively compared to the FD. In the same order, the cost per kg of the formulated diet was reduced by 4.52 (MD) and 0.00% (ED) compared to the CD.

Table 8: Financial evaluation of experimental diets formulated for the rearing of *O. niloticus*

Paramètres	Diets				
	FD	SD	MD	ED	CD
CF (FCFA.kg-1)	400	204116	296	310	310
RxR CF/FD (%)	--	--	26	22.50	--
RxR CF/CD (%)	--	--	4.52	0.00	--

CF= Cost of 1 kg of diet, RxR CF/FD = Reduction Rate of CF compared to fish diet (FD), RxR CF/CD = Reduction Rate of CF compared to commercial diet (CD).
Price in CFA pound: 100 CFA= 0.18 $ based on 2017 exchange prices in Ivory Coast -- = Absents values.

4. Discussion

This study evaluates the nutritional value of the ingredients, the experimental diets and the reference diet. The protein contents of earthworm (41.17±0.06%), Spirulina (40.7± 0.22%) and maggot (40.34±0.13%) are relatively high, but low compared to that of fish (56.00±0.18). These results are in agreement with those of Ossey *et al.* (2012) [6] and John (2015) [11]. These authors indicated the protein contents of maggots which are between 40 and 64%. The crude protein content of the Spirulina obtained corroborates the results of Radhakrishnan *et al*. (2017) [8]. These authors obtained protein contents of Spirulina ranging from 34 to 73%. The protein content of earthworms in this study is low (41.17±0.06%) and approaches that (46.57%) obtained by Hasanuzzaman *et al.* (2010) [24]. The highest lipid content (25.00±0.14%) observed in the maggot is in agreement with that obtained by Ossey *et al*. (2012) [6] (28.95%). The low lipid contents (6.77±0.13%) obtained with Spirulina meal are in agreement with the results of Ahmadzadenia *et al*. (2011) [13]. These authors indicated the lipid contents of Spirulina ranging from 7-16%. The high ash contents of Spirulina meal (15.14±0.10%) confirm the results of Abdudraman (2014) [25]. This author indicated that Spirulina is rich in minerals. In this study, the protein (56.00±0.21%) and ash (18.00±0.24%) contents of fishmeal are high compared to those of other ingredients. These results are in agreement with those of Guillaume *et al.* (1999) [26]. These have shown that fish meal contains all the minerals best suited to fish rearing. The protein contents of wheat bran (15.30±0.12%), corn flour (11.80±0.19%) and copra meal (21.00±0.14%) are weak. However, these ingredients can be used as a source of energy because they are rich in starch. These results are in agreement with those of Burel and Médale (2014) [3] who indicated that vegetable products and cereal flour are rich in carbohydrates and therefore in starch, but low in proteins. However, soybean and cottonseed meal obtained high protein contents. These results corroborate those of Guillaume *et al*. (1999) [26] and Médale and Kaushik. (2009) [27] who showed that soybean and cottonseed meals can be used in fish feed because of their high protein contents. The fat contents of purchased raw materials of plant origin are low, varying from 2.04±0.15 to 6.95±0.17%. These results are in agreement with those of Burel and Médale (2014) [3] who indicated that plant products used in aquaculture feed contain little lipids. The formulated diets have the same protein contents at the larval phase (40% of crude protein) in accordance with what is recommended for the rearing of this species. [28, 29, 30, 31]. In this work, the method used for diet formulation is the same as that proposed by Blé *et al*. (2008) [32]. The latter indicated that the formulation of diets takes into account the nutritional value of the raw materials, their availability, their cost but also the needs of the species to be fed, as well as its size and age. However, a slight difference was observed in the lipid contents. The diets based on maggot meal is richer in lipids (6.17±0.10%). This would be explained by the high lipid content of maggot meal. However, these values are in agreement with those of Kestemont *et al*. (1989) [30] and Creswell (2005) [33]. These authors showed that the dietary lipid requirements of tilapia *Oreochromis niloticus* are between 4 and 10%. The similarity in protein contents diet formulated at this larval stage could be explained by the fact that the formulated diets are iso-protein. According to Blé *et al*. (2008) [32], an excess of proteins and lipids in a fish's diet can respectively lead to the excretion of ammonia in the rearing environment and lipid deposition in the carcass. The incorporation of lipids in the formulation makes it possible to spare dietary proteins from catabolism and therefore to cover the growth and energy substrate needs of fish [34] The gross energy of the experimental diets ranges from 14.59±0.11 to 16,49±0,11 $kJ.g^{-1}$. These values are within the recommended limits for farming tilapia *O. niloticus* (10 to 25 $kJ.g^{-1}$) [28, 35, 36]. The use of maggot, earthworm and Spirulina meals did not negatively influence the calcium and phosphorus contents in the formulated diets. However, the phosphorus content of diet formulated with fishmeal was high. These results are in agreement with those of NRC (1993) [2] which showed that fish meals contain a high phosphorus content. In this study, linoleic fatty acid levels were elevated with the Spirulina flour-based diet. Which is in agreement with the results of Diraman *et al.* (2009) [37]. Furthermore, palmitic fatty acid levels were elevated with fishmeal-based food. These results are in the same direction as those of Bahurmiz and Ng (2007) [38] who showed that high concentrations of palmitic acid are characteristic of fish meal.

The results relating to aflatoxins B1, B2, G1 and G2 by HPLC in the different experimental diets showed no presence of these toxins in MD, SD and ED diets. Furthermore, the total quantity of these aflatoxins was below the limits of quantification in FD and CD diets. It appears from these results that the experimental diets were not contaminated with these toxins and could be used as fish feed. The use of maggot and earthworm meal led to a reduction in the cost per kg of the formulated diet while this increased with the diet formulated based on fish meal and Spirulina. This could be explained by the price of the ingredients. Indeed, fish meal is relatively expensive (700 FCFA/kg). Regarding Spirulina, the high cost per kilogram of the formulated diet could be explained by the production method. Indeed, due to a lack of cheaper technical chemicals, we have often resorted to more expensive analytical chemicals, which has overestimated the costs of spirulina culture media.

5. Conclusion

In conclusion, the total replacement of fish meal with Spirulina, maggot and earthworm meal did not negatively affect the nutritional quality of diets. The reported values are within the recommended limits for rearing the species at this stage. Apart from Spirulina, use of maggot and earthworm has contributed to reduce the cost per kilogram of formulated diet compared to that based on fish meal.

6. References

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