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Apparent Digestibility Co - efficient (ADC) of *Clarias gariepinus* (Burchell, 1822) Fingerlings Fed Different Levels of Maca (*Lepidium meyenii,* Walp.) Root Powder as Feed Additive

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Abstract

A study was conducted at the Lay - Joy fish farm, Billiri, Gombe State Nigeria to determine the apparent digestibility co - efficient (ADC) of Clarias gariepinus fingerlings fed different levels of Maca root powder as feed additive. Five (5) isonitrogenous (40% crude protein) and isocaloric (1,732kcal kg^{-1} gross energy) diets were formulated, *where Maca root powder in form of feed additive* was incorporated at *0.0g/100g (TM_0), 0.25g/100g (TM_1), 0.5g/100g (TM_2), 0.75g/100g (TM_3) and 1.0g/100g (TM_4) inclusion levels,* diet without *Maca root powder (TM_0)*, served as the control diet. The formulated diets were fed to *Clarias gariepinus fingerlings (n = 300, 10.0±0.00g)* in *fifteen (15) rectangular white plastic tanks (n = 20) at a fixed feeding rate of 3% body weight twice daily between the hours of 8:00 - 9:00am and 4:00 - 5:00pm at regular interval and adjusted after every two (2) weeks of sampling* for a period *of twelve (12) weeks. Ten (10) hours after feeding, three (3) fish from each treatment and the control were sacrificed, the faecal samples were collected and the ADC values of the feeds were computed. Data obtained were analysed using one - way analysis of variance (ANOVA) at P = 0.05. ADC value of crude protein was highest in fish fed diet TM_3 (90.43±1.4%) and least (84.75±1.6%) in the fish fed the control diet (TM_0), ADC value of crude lipid was highest in fish fed diet TM_3 (84.46±1.8%) and least (81.30±1.0%) in the fish fed control diet (TM_0), ADC value of dry - matter was highest in fish fed diet TM_3 (68.07±1.2%) and least (64.51±1.3%) in the fish fed the control diet (TM_0), while the ADC value of carbohydrate was highest in fish fed control diet (TM_0) (50.38±1.3%) and least (49.14±1.0%) in fish fed diet TM_2, respectively. However, there was a significant difference (p<0.05) in the ADC values of crude protein, crude lipid and dry - matter at 0.5g/100g and above inclusion level of the Maca root powder, while there was no significant difference (p>0.05) in the ADC values of carbohydrate. Inclusion of Maca root powder as feed additive at 0.75g/100g indicated a high significant difference (p<0.05) on the ADC values of crude protein, crude lipid and dry - matter. Therefore, it is recommended that Clarias gariepinus fingerlings be fed with diet incorporated with Maca root powder as feed additive in order to improve the ADC values of some essential nutrients. However, further research should be carried out on other Phyto - additives on their efficacy in determining the ADC values in Clarias gariepinus.*

Keywords: Apparent Digestibility, *Clarias gariepinus*, Fingerlings, *Lepidium meyenii***,** Additive.

Introduction: The apparent digestibility of a certain nutrient is mostly represented by one value, the apparent digestibility co - efficient (ADC). This representation by a single value suggests that an apparent digestibility co - efficient of a nutrient is constant (Ali, 2022). Digestion can be defined as the breakdown of food by catalytic enzymes (Babale, 2016). Digestion of food in fish depends on the kind of food, the extent to which it is susceptible to the effects of the digestive enzymes, the activity of the digestive enzymes and the length of time the food is exposed to the action of the digestive enzymes (Falaye, Elezuo, Ajani and Omoike, 2016). Each of these factors is affected by many secondary factors, some of which are associated with the fish itself, such as species, age, size, physiological condition and environmental condition, such as water temperature, composition of diet, particle size and quantity of diet consumed (Ali, 2022). Digestibility determination is very important in the selection of feed ingredients for feed formulation and determination of nutrient digestibility (Babale, 2016). Digestibility is one of the most important aspects in evaluating the efficiency of animal feedstuffs and a basic requirement for formulating fish diets (Sogbesan, 2023). Findings have shown that the determination of nutrient digestibility is the first step for evaluating the potential of any ingredient for use in the diet of reared species (Ali, 2022). Falaye *et al.* (2016) stated that feed ingredients' digestibility is not only a useful tool for diet formulation but gives the right estimation for fish growth. Nutrient digestibility may vary among fish species due to differences in the digestive system, enzymes and foods consumed (Babale, 2016). Despite these differences and the lack of pepsin in fish without stomach, variation in the digestibility of proteins and lipids among species are very small. Much more pronounced are variations in digestibility of carbohydrates, especially starch (Ali, 2022). Babale (2016) stated that carnivorous fish digest starch to a much lesser degree than omnivorous and herbivorous fish. He also stated that the Differences in digestibility of carbohydrates can also occur among different families of carnivorous fish within the same species. The inclusion of starch to maximise the use of dietary protein for the growth of *Clarias gariepinus* (Burchell, 1822) has been reported (Sogbesan, 2023). The attributes to digest starch, a non - protein energy source in *Clarias gariepinus*, depends on the secretion of α – amylase to the gastrointestinal tract, however, findings have shown that all species of fish possess the characteristics to secrete α - amylase (Onimisi, Balorunduro, Oniye, Balogun, Yunusa and Abdulraheem, 2016). Onimisi *et al.* (2016) also reported that the digestibility of dry matter, lipid and protein was significantly affected by feeding level but not feeding frequency when the feeding level was similar. Also, gastric evacuation rate in *Clarias gariepinus*, increases with increase of dietary material through the digestive tract, resulting in less material digested, and lower apparent nutrient digestibility values. Therefore, it was suggested that *Clarias gariepinus* should be fed two times per day when they are past the fingerling's stages, in order to utilize dietary protein more efficiently. Maca (*Lepidium meyenii*, Walp.) is reported as a vegetable root plant native to Peru in South America which can be used in its dry powder form as Phyto - additive in animal feeds (Mahdy, Abuol - Omran, Desoky, Omnia, Abd El - Salam, Abo - Farw, El - Kholany, Ahmed and Khalifa, 2023). In Hausa language it is known as ''Gadali or Albasar Tamoji' and it possess multiplicity of biological function (Garba, Gaddafi and Yahaya, 2022). Maca is an adaptogen, which helps the body adapt to stress, contains plant sterols which help the body produce the appropriate levels of hormone and also help in the digestion of some vital nutrients (Gaddafi, Yahaya, Garba and Usman, 2023). Maca has been used as a food health - promoting food material, stamina builder, and fertility promoter due its various biological effects, including the regulation of metabolism and hormonal secretion, improvement in memory and digestion, and also antidepressant activity in humans (Zhang, Li, Wang, Yao and Zhu, 2017). Recently studies indicated, Maca has been consumed in juices, soups, extracts, and processed foods enriched with Maca flour (Lee and Chang, 2019). It is stated by the nutrition industry that Maca has the ability to improve, digestion, energy and modulate the response against oxidative stress. Previous studies reported that Maca comprises various classes of bioactive compounds, including saponins, alkaloids, steroid hormones, and polyphenol compounds (Tang, Jin, Xie, Huang, Wang, Chu, Dai, Liu, Wang and Zhang, 2017) and these compounds play a variety of biological role in human and animal body. Maca is also rich in crude protein, crude lipid, some essential acids (Ali, Watafua, Maisheru, Salisu, Remkyes, and Yakubu, 2024a). The objective of this study was to determine the apparent digestibility co - efficient (ADC) of *Clarias gariepinus* fingerlings fed different levels of Maca root powder as feed additive.

Materials and Methods: Study Area: The study was conducted at the Lay - Joy fish farm, Gombe - Yola Road, Billiri local government area (LGA), Gombe State Nigeria. Billiri LGA lies within Lat. 9°50'N; 11°09´E and Long. 9.833°N 11.150°E. It covers an area of 737km² (285 sq. m) and is 50 km away from Gombe the State capital.

Experimental Fish: Three hundred (300) *Clarias gariepinus* fingerlings with mean initial weight (10.0±0.00g) were stocked at twenty (20) fingerlings per tank in triplicates per treatment after one (1) week of acclimatization, the study lasted for a period of twelve (12) weeks.

Experimental Feed: The experimental diets contained fish meal (FM), soybean meal (SBM), yellow maize meal (YMM), groundnut cake meal (GNCM) and Maca root powder. All the ingredients were grounded into a fine powder using a hammer mill and sieved by a 0.25 mm sieve. Fish meal, soybean meal, groundnut cake meal and yellow maize meal were obtained from commercial suppliers in Gombe, the vitamin/mineral premix, fish oil and chromic oxide (Cr_2O_3) were purchased from TTS Integrated Farms Nigeria Limited, Agege - Lagos, while the Maca root powder naturally derived from dry Maca (*Lepidium meyenii*) root was obtained from BonAmour Pharmacy Limited Lagos, Nigeria, imported from Piping Rock Health Products, Ronkonkoma, New - York, USA. Prior to the feed formulation, proximate composition of these ingredients was determined (Table 1).

Five (5) isonitrogenous and isocaloric diets were prepared, each diet with crude protein (CP) content at 40% CP as calculated according to Pearson's square method and gross energy content at 1,732kcal kg^{-1} respectively for the feeding the experimental *Clarias gariepinus* as recommended by Ali (2022) was prepared, where the Maca root powder in form of feed additive *was incorporated into the diets at* 0.0g/100g (TM_0), 0.25g/100g (TM_1), 0.5g/100g (TM_2), 0.75g/100g (TM_3) and 1.0g/100g (TM_4) inclusion levels, *diet without* Maca root powder (TM_0), *served as the control diet. (Table 2). Diets were subjected to proximate analysis as described by the AOAC (2005) as presented in Table 3.*

Experimental Design: The fish were cultured in fifteen (15) rectangular white plastic tanks (flow - through system) with a water holding capacity of one thousand litres (1,000L) each in a complete randomized design (CRD). Each tank was washed thoroughly with salt, filled to just a little over 1/3 (350 litre) capacity and stocked with twenty (20) fingerlings of *Clarias gariepinus* with mean initial weight (10.0±0.00g). The *Clarias gariepinus* fingerlings were fed the experimental diet at 3% body weight two (2) times daily between the hours of 8:00 - 9:00am and 4:00 - 5:00pm for a period of twelve (12) weeks. The quantity of feed was adjusted accordingly after every two (2) weeks of sampling for growth performance and survival rate (mean body weight and mortality). Water temperature, pH, dissolved oxygen, and ammonia were measured at the beginning of the experiment after which they were measured weekly throughout the period of the experiment. Water temperature, dissolved oxygen and pH were measured using Horiba U-22 XD multi - parameter water quality checker while ammonia was measured using freshwater aquaculture test kit (Model AQ-2, Code 3633-03, Lamotte U. S. A). Digestibility of diets was determined using the indirect method which relies on use of an inert marker as described by Elezuo (2016). In this study chromium (III) oxide (Cr_2O_3) was used as the marker at 0.5% level of inclusion. Ten (10) hours after feeding, three (3) fish from each treatment and the control were sacrificed through partial decapitation by cutting through the vertebrate column. The gastrointestinal tract was divided into four (4) parts, these were; the stomach, the fore - gut, the mid - gut and the hind - gut as described by Babale (2016). A longitudinal incision was made along the length of the abdomen from the anus to the base of the mouth. The gastrointestinal tract was then carefully removed in other to avoid squeezing the contents from one section of the gastrointestinal tract to another. The hind - gut was severed from the mid - gut. A longitudinal incision was then made in the mid - gut to allowed easy removal of the contents. The faecal sample was then stored in a freezer prior to proximate analysis and the determination of Cr_2O_3. The Cr_2O_3 was quantified as described by Ali (2022).The weight and percentage of the Cr_2O_3 was calculated using the following formulae: $\text{Weight of Cr2O3} = \text{Absorbance} - 0.0032/0.2089$Digestibility co - efficient of diets were estimated to determine bioavailability of nutrients. Apparent digestibility co - efficient (ADC) of the feeds were computed using the formula described by Hossain *et al.* (2003).

ADC (%) = 100 x (% nutrient in faeces/% nutrient in feed) x (% marker in feed/marker in faeces).

Data Analysis: The data obtained were subjected to one - way analysis of variance (ANOVA) using the GraphPad Instant package for windows 2016 of statistical analysis system (SAS, 2016). Mean separation was done (at P = 0.05) using Fisher's least significance difference (LSD) to separate the means in cases of significant difference.

Table 1: Proximate and Energy Composition of Ingredients used for the Experimental Diets

IngredientsCP%	CL%	CF%	Ash%	NFE%	Moisture%		GE	
Fish meal 71.23	7.83	ND	9.22	3.81	7.91		1195	
Soybean meal	43.90	3.10	7.30	5.10	33.90	6.70		1003
GNC meal 43.50	5.17	6.01	5.97	31.00	8.35		2432	
Yellow maize	8.95	3.35	10.38	4.07	64.86	8,39		807

Source: Ali, Danba, Sani and Danzaria, (2024b).

Keys: CP - Crude protein, CL - Crude lipid, CF - Crude fibre, NFE - Nitrogen free extracts, GE - Gross energy, GNC - Groundnut cake, ND - Not detected.

Table 2: Ingredient Percentage Composition (g/100g) of Experimental Diets with Different Levels of Maca Root Powder as Feed Additive

Ingredients (%)	TM_0	TM_1	TM_2	TM_3	TM_4
Fish meal	20.00	20.00	20.00	20.00	20.00
Soybean meal	21.50	21.25	21.00	21.00	21.00
GNC meal	23.00	23.00	23.00	23.00	23.00
Yellow maize	30.00	30.00	30.00	29.75	29.50
Maca root powder	**0.00**	**0.25**	**0.50**	**0.75**	**1.00**
Fish Oil	1.00	1.00	1.00	1.00	1.00
Vegetable oil	1.00	1.00	1.00	1.00	1.00
Starch	1.00	1.00	1.00	1.00	1.00
Lysine	0.25	0.25	0.25	0.25	0.25
Methionine	0.25	0.25	0.25	0.25	0.25
***Vitamin/premix**	1.00	1.00	1.00	1.00	1.00
Salt	0.50	0.50	0.50	0.50	0.50
Cr_2O_3	0.50	0.50	0.50	0.50	0.50
Total	100.00	100.00	100.00	100.00	100.00

Source: Ali *et al.* (2024b).

Keys: TM_0 - Maca root powder (0.0g/100g), TM_1 - Maca root powder (0.25g/100g), TM_2 - Maca root powder (0.5g/100g), TM_3 - Maca root powder (0.75g/100g), TM_4 - Maca root powder (1.0g/100g).

*Vitamin and mineral mixture (product of HEPOMIX): 12.000.000 IU Vitamin A; 2.000.000 IU Vitamin D3; 10g Vitamin E; 2g Vitamin K3; 1g Vitamin B1; 5g Vitamin B2; 1.5 g Vitamin B6; 10g Vitamin B12; 30g Nicotinic acid; 10g Pantothenic acid; 1g Folic acid; 50g Biotin; 250g Choline chloride 50%; 30g Iron; 10g copper; 50g Zinc; 60g Manganese; 1g Iodine; 0.1g Selenium and Cobalt 0.1g.

Table 3: Proximate and Gross Energy Composition of Experimental Diets with Different Levels of Maca Root Powder as Feed Additive

Proximate composition (%)	TM_0	TM_1	TM_2	TM_3	TM_3
Crude protein (%)	40.01	40.05	40.08	40.21	40.11
Crude lipid (%)	5.80	5.86	5.88	5.96	5.91
Crude fibre (%)	2.81	2.86	2.89	2.92	2.90

Moisture (%)	5.47	5.79	5.30	5.15	5.19
Ash (%)	13.11	12.87	12.94	12.69	13.01
Nitrogen - free extract (%)	32.80	32.57	32.91	33.07	32.88
Gross energy (Kcal/kg)	3,330	3,310	3,201	3,180	3,197

Source: Ali *et al.* (2024b).

Keys: TM_0 - Maca root powder (0.0g/100g), TM_1 - Maca root powder (0.25g/100g), TM_2 - Maca root powder (0.5g/100g), TM_3 - Maca root powder (0.75g/100g), TM_4 - Maca root powder (1.0g/100g).

Results: The results of the apparent digestibility co - efficient (ADC) of the *Clarias gariepinus* fingerlings fed different levels of Maca root powder as feed additive is presented in Table 4. Apparent digestibility co - efficient (ADC) of crude protein values range from 84.75±1.6% – 90.43±1.4%; ADC value of crude protein was highest in fish fed diet TM_3 (90.43±1.4%), while the fish fed the control diet (TM_0) had the least ADC value of crude protein (84.75±1.6%). Apparent digestibility co - efficient (ADC) of crude lipid values range from 81.30±1.0% – 84.46±1.8%; ADC value of crude lipid was highest in fish fed diet TM_3 (84.46±1.8%), while the fish fed the control diet (TM_0) had the least ADC value of crude lipid (81.30±1.0%). Apparent digestibility co - efficient (ADC) of dry - matter values range from 64.51±1.3% – 68.07±1.2%; ADC value of dry - matter was highest in fish fed diet TM_3 (68.07±1.2%), while the fish fed the control diet (TM_0) had the least ADC value of dry - matter (64.51±1.3%). Apparent digestibility co - efficient (ADC) of carbohydrate values range from 49.14±1.0% – 50.38±1.3%; ADC value of carbohydrate was highest in fish fed control diet (TM_0) (50.38±1.3%), while the fish fed diet TM_2 had the least ADC value of carbohydrate (49.14±1.0%). However, there was a significant difference ($p<0.05$) in the ADC values of crude protein, crude lipid and dry - matter at 0.5g/100g inclusion level and above of the Maca root powder, while there was no significant difference ($p>0.05$) in the ADC values of carbohydrate.

Table 4: Apparent Digestibility Co - efficient (ADC) of *Clarias gariepinus* Fed Different Levels of Maca Root Powder as Feed Additive

Digestibility Indices (%)	TM_0	TM_1	TM_2	TM_3	TM_4
ADC of crude protein	84.75 ± 1.6^a	85.29 ± 1.5^a	86.70 ± 1.9^b	$90..43\pm1.4^c$	87.93 ± 1.7^b
ADC of crude lipid	81.30 ± 1.0^a	81.39 ± 1.3^a	82.61 ± 2.0^b	84.46 ± 1.8^c	83.00 ± 1.6^b
ADC of dry – matter	64.51 ± 1.3^a	65.01 ± 1.7^a	66.76 ± 1.5^b	68.07 ± 1.2^c	67.45 ± 1.1^b
ADC of carbohydrate	50.38 ± 1.3^a	50.01 ± 1.1^a	49.14 ± 1.0^a	49.40 ± 1.4^a	49.21 ± 1.7^a

Mean values in each row with similar superscripts are not significantly different ($p>0.05$).

Keys: TM_0 - Maca root powder (0.0g/100g), TM_1 - Maca root powder (0.25g/100g), TM_2 - Maca root powder (0.5g/100g), TM_3 - Maca root powder (0.75g/100g), TM_4 - Maca root powder (1.0g/100g).

Discussion:The apparent digestibility co - efficient (ADC) of crude protein values, 84.75% – 90.43% recorded from this study are comparable with the values, 86.97% – 89.82% reported by Ali. (2022) for *Clarias gariepinus* fingerlings fed β - glucan additive diet and the values, 87.06 % – 89.03 % reported by Elezuo (2016) for *Clarias gariepinus* fingerlings fed processed almond (*Terminalia catappa*) kernel meal. The highest ADC value of crude protein recorded from the fish fed diet TM_3 might be as a result of the efficacy of the Maca root powder at 0.75g/100g feed level of inclusion, thereby allowing the crude protein in the feed to be more available to the fish, which was in agreement with the findings of Garba, Yahaya and Gaddafi (2023) which reported that Maca root powder plays an important role in protein metabolism in yankasa rams fed Maca root powder as feed additive. The ADC of crude lipid values, 81.30% – 84.46%; recorded from this study are comparable with the values, 83.50 – 85.35% reported by Ali. (2022) for *Clarias gariepinus* fingerlings fed the co - mixed of betaine/β - glucan additive diet and the values, 74.30 % – 87.38 % reported by Onimisi *et al.* (2016) for *Clarias gariepinus* fingerlings fed *Senna obstusifolia* seed meal. The high ADC values for crude lipid recorded from all the diets suggest that the amount of lipid ingested and digested were the required quantity by *Clarias gariepinus* for the best conversion of feed into flesh and was in support with the findings of Garba *et al*. (2023) which reported that crude lipid retention in the diets with Maca root powder was higher than that of the control in a study with rams**.** Ali. *et al.* (2024a) reported that a dry Maca root powder is rich in crude protein, crude lipid and some essential acids which aid in digestion. The ADC of dry - matter values, 64.51±1.3% – 68.07±1.2%; recorded from this study are comparable with the values, 60.15 % – 70.58 % reported by Onimisi *et al.* (2016) for *Clarias gariepinus* and higher than the values, 56.44 % – 64.43 % reported by Babale (2016) for same species. Falaye *et al.* (2016) stated that the ADC value of dry - matter provides a better estimate of the quantity of digestible materials in the feed rather than individual nutrients. The fairly high ADC values of dry - matter recorded from all the fish fed diets with Maca root powder and the control diet (TM_0) indicated good diets acceptance and utilization by the *Clarias gariepinus* fingerlings. The ADC of carbohydrate values, 49.14±1.0% – 50.38±1.3%; recorded from this study are comparable with the values, 51.24% - 52.58% reported by Ali. (2022) for *Clarias gariepinus* fingerlings fed betaine additive diet and significantly higher than the values, 23.48 % – 41.59 % reported by Onimisi *et al.* (2016) for *Clarias gariepinus*. The fairly low ADC of carbohydrate values recorded from all the *Clarias gariepinus* fingerlings fed diets with Maca root and the control diet (TM_0) indicated that the amount of carbohydrate ingested and digested were the required quantity needed by *Clarias gariepinus* for the best conversion of feed into flesh. Gül, Olgun, Yıldız, Tüzün and Sarmiento - García (2022) reported that the incorporation of Maca root powder as feed additive into animal feeds greatly enhanced the ADC values of some essential nutrients.

Conclusion: The incorporation of Maca root powder in the form of a feed additive at 0.5g/100g inclusion level and above in the diet of *Clarias gariepinus* fingerlings had significant ($p<0.05$) effects on the ADC values of crude protein, crude lipid and dry - matter.

Recommendations: The incorporation of Maca root powder as feed additive into the diet of *Clarias gariepinus* fingerlings is highly recommended, incorporation of Maca root powder at 0.75g/100g inclusion level as feed additive in the diet of *Clarias gariepinus* fingerlings is recommended for improving the ADC values of some essential nutrients in *Clarias gariepinus*. However, further research should be carried out on other Phyto - additives on their efficacy in determining the ADC values in *Clarias gariepinus*.

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