# Effects Of Replacing Soyabeans Meal With Graded Levels Of Boiled Jatropha Kernel Meal In Diets Of *Clarias gariepinus* Fingerlings

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**Abstract:** A feeding trial was conducted in the laboratory for 8 weeks to evaluate the growth response of Clarias gariepinus fingerlings using boiled Jatropha kernel as the protein source in place of soyabean meal. Four experimental diets (40% crude protein) were formulated, three of these diets contained boiled Jatropha kernel meal at varying levels of 0% 10%, 20%, and 30% while the diet without boiled Jatropha kernel meal served as control. Fish fingerlings were fed weekly at 5% body weight. It was discovered that the percentage weight gain was significantly different (p<0.05) among the controlled diet and the experimental diets; specific growth rates were also significantly different (p<0.05) among the controlled diet, diets 3 and 4 but different in diets 1 and 2 with diet 4 (30%) of boiled Jatropha kernel having the highest value. The highest weight gain was recorded in diet 4 containing 30% of boiled Jatropha kernel meal (75.57g) while the least at 10% inclusion (52.47g). The soyabean meal protein increased with increasing boiled Jatropha kernel substitution. Based on the results of this study, it could be concluded that C. gariepinus could tolerate 30% inclusion of boiled Jatropha kernel meal in the diet in place of soyabean meal.

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**Key words:** Growth performance, Nutrient utilization, Clarias gariepinus, boiled Jatropha kernel meal.

## Introduction

Oilseed cake and legume seeds are considered suitable as alternative dietary protein sources for fish feed and are available in sub-Saharan Africa on a large scale (1). Soybean meal (SBM) is one of the most nutritious of all plant protein sources (2). Due to its high protein content, high digestibility and relatively well balanced amino acid profile, it is widely used as feed ingredient for many aquaculture species (3). It is currently the most commonly used plant protein source in fish feed (4). 5 reported that soybean products have been used to replace a significant portion of fish meal in fish feed with nutritional, environmental and economic benefits. However, wider utilization and availability of this conventional source for fish feed is limited by increasing demand for human consumption and by other animal feed industries (6), hence the need to focus on using less expensive and readily available plant protein sources to replace soybean meal without reducing the nutritional quality of the feed becomes imperative (7).

Jatropha kernel meal is a by-product of bio-fuel or bio-diesel processing. Each ton of dry seed produces 200-300 liters of bio-fuel and 700-800 kg of seed meal (8). If not contaminated by seed shell, Jatropha kernel meal contains high crude protein (56.4%) (9), higher than that of soybean meal (48%), so that it is potential to be used as protein source in the diet. Jatropha kernel meal also contains anti-nutritive compounds, such as lectin, trypsin inhibitor (anti-trypsin), saponin, phytate, and phorbol ester (10;,11). Of all the compounds, phorbol ester is considered as the most toxic compound.

To be able to utilize as protein source in the diet, these anti-nutritive compounds need to be removed either physically using heat treatment or chemically by extraction. Physical treatment by heating in autoclave potentially reduces anti-trypsin and lectin contents in seed meal (12). Chemical treatment by extraction can be conducted using hexan and metanol or etanol, or soaking the seed in base solution (13) or potassium bicarbonate (14Martinez-Herrera *et al*. 2006). Phorbol ester in Jatropha kernel could be eliminated by extraction. Combined physical and chemical treatments are able to eliminate lectin, anti-trypsin, and Phorbol ester compounds in Jatropha kernel meal.

Jatropha kernel meal is one of the important annual crops of the world grown for oil. It has nutritional quality comparable to other oilseed proteins including soybean and other conventional legumes (15; 16; 17:19,18, 19 and20) and their potential as dietary protein sources in animal feed is well recognized ( 21).

In fish feeding, Jatropha seed meal had been tested as protein supplement in livestock feed such as goat (22Belewu, *et. al.*, 2010); mice (23) and broiler chicken ( 24Tiurma Pasaribu, *et. al.*, 2010) and with fish species such as *Cyprinus carpio* ( 25; 9 and *Clarias gariepinus* ( 26). Information on aquafish performance fed Jatropha kernel meal is still limited. The aim of this experiment is to evaluate the effects of replacing soyabean meal with graded levels of boiled Jatropha kernel meal in diets of *Clarias gariepinus* fingerlings.

## Materials and Methods

### Collection and preparation of the Jatropha kernel for the meal

Two kilograms of mature seeds of *Jatropha curcas* were source from neighbouring village within New Bussa and its environment. The seed were stored with polythene bags at room temperature (approximately 20ºC) until use. Before the commencement of the experiment, the Jatropha seed were deshelled to obtain the kernels. The kernels were washed with tap water to remove dirt and other debris drained properly and sun dried to a constant weight. These were ground into a powder with a domestic hammer mill to pass through a 1mm sieve. The ground kernel was boiled at 100ºC for 30 minutes so as to eliminate the possible detrimental effects of anti-nutritional factors that could be present in the kernel. The Jatropha kernels meal and the experimental diets were analysed for their proximate composition according to the methods of 27.

### Diet formulation and preparation

Experimental diets were formulated with boiled Jatropha kernel replacing soyabean meal on a dry basis to maintain the crude protein and energy levels. Four iso-nitrogenous and iso-energetic diets were formulated to evaluate the nutritive value of catfish, *Clarias gariepinus* fingerlings. The boiled Jatropha kernel meal was included in the diets at levels of 0, 10, 20 and 30%. The diet containing 0% boiled Jatropha kernel meal serves as the control. The ingredients were mixed together using a kitchen mixer before the addition of vitamin premix. Oil was later added to the dry ingredient and mixed thoroughly. Warm water was added to the premixed ingredients and homogenized until a dough-like paste was formed. The dough was passed through an improvised pelleting machine. The moist pellets were sun-dried to a constant weight and kept in air tight containers. The percentage composition of the experimental diets is given in Table 1.

### Experimental design

A complete randomized design (CRD) was adopted with tree replicates per diet. Each replicate had 15 fish and were grouped fed. Fingerlings of *Clarias gariepinus* were purchased from Kofo fish farms, New Bussa, Niger State, Nigeria. The fish were allowed to acclimatize for 5 days, during this period, they were fed on commercial diet. Prior to the commencement of the experiment, all fish were starved for 24 hours. This practice was to eliminate variation in weight due to residue food in the gut and also to prepare the gastro intestinal tract for the experimental diets, while at the same time to increase the appetite of the fish.

### Feeding trials

The feeding trial was conducted in an experimental unit containing a set of 15 rectangular plastic tanks, each with a capacity of 55L of water. 180 catfish, *Clarias gariepinus* fingerlings of average initial weight of 29.52±0.40g were used for the experiment. All fish were fed twice daily at a fixed feeding rate of 5% body weight per day. Feedings were generally done in the mornings at 09:00 and 15:00h except on sampling days when they were fed after weighing. All fish were collectively weighed at the start of the experiment (0 day) and at 7-day intervals (14, 21, 28, 35, 42, 49 and 56 days). Growth performances were evaluated as body weight gain (BWG), Feed conversion ratio (FCR) and specific growth rate (SGR).

### Water Quality measurement

Water quality was monitored every week throughout the feeding trials. Water temperature (°C) and pH were measured daily with a combined digital pen meter, while dissolved oxygen (mg/l) was measured using Winkler's titration method. Conductivity was determined using a digital conductivity meter.

### Statistical analysis of data

One-way analysis of variance (ANOVA) was carried out to determine the effect of diets on growth and nutrient parameters using 16.0 version of 28 statistical package for Windows. Significant differences between individual means were identified using the Duncan's multiple range test (29). Mean differences were considered significant at $P<0.05$.

## Results and Discussion

### Chemical composition of raw and boiled Jatropha kernel

The chemical composition of raw and boiled Jatropha kernel were presented in Table 2. The chemical composition of raw and boiled Jatropha kernel meal showed that the kernel meal contained high crude fat of 27.97 and 30.09% respectively. This resulted in high gross energy of 569.18kcl/g and 596.78kcal/g. The crude protein of boiled Jatropha kernel meal was higher (37.75%) than the raw (31.23%). The raw kernel meal had 31.32% of nitrogen free extract (NFE) while the boiled Jatropha kernel had 24.18% nitrogen free extract (NFE). The digestible energy was also higher in boiled Jatropha kernel meal (436.31kcl/g) than raw Jatropha kernel meal (414.17kcl/g).

**Proximate composition of the experimental diets**

The proximate composition of the experimental diets containing 0, 10, 20 and 30% boiled Jatropha kernel meal were presented in Table 3. The results showed that diet 4 has the highest value of crude protein (40.82%), followed by diet 2 and 3 (40.57%) and (40.41%) while diet 1 has the lowest value (40.31%). Diet 1 has the highest value (3.89%) of crude fibre, followed by diet 2 and 3 (3.64% and 3.59%) while diet 4 has the lowest value (3.36%) of crude fibre. Crude lipid was highest in diet 1 (9.94%), followed by diet 2 and 3 (9.52%) and (9.39%) while diet 4 has the lowest value (9.94%) of crude lipid. The nitrogen free extract (NFE) was highest in diet 4 (40.37%), followed by diet 3 and 2 (40.22%) and (39.70%) while diet 1 has the lowest value (39.23%) of nitrogen free extract (NFE). The moisture content of the diets ranged from 7.08% (diet 4) and 9.92% (diet 2). All the diets contained a uniform value of gross energy (Kcl/g) and digestible energy (Kcl/g) of approximately 482.30 and 318.49.

Table 1: Percentage composition of the experimental diets

| Ingredients | Diet 1 | Diet 2 | Diet 3 | Diet 4 |
|---|---|---|---|---|
| Boiled Jatropha seed meal | 0.00 | 7.52 | 15.47 | 23.86 |
| Soyabean meal | 36.55 | 30.06 | 23.20 | 15.92 |
| Fish meal | 21.93 | 22.55 | 23.20 | 23.8 9 |
| Groundnut cake | 14.62 | 15.03 | 15.67 | 15.92 |
| Yellow maize | 16.90 | 14.84 | 12.66 | 1 0.38 |
| Vitamin premix | 3.00 | 3.00 | 3.00 | 3.00 |
| Vegetable oil | 2.00 | 2.00 | 2.00 | 2.00 |
| Methionine | 1.00 | 1.00 | 1.00 | 1.00 |
| Lysine | 1.00 | 1.00 | 1.00 | 1.00 |
| Bone meal | 2.00 | 2.00 | 2.00 | 2.00 |
| Salt | 1.00 | 1.00 | 1.00 | 1.00 |
| Total | 100.00 | 100.00 | 100.00 | 100.00 |

Hi-Nutrients Vitamins/Minerals premix supplies 100g Diet. Palmat A: 1000IU; Cholecalceferol (D): 1000IU; G-Tocopherolacetate (E): 1.1mg; Menadilione (K): 0.02mg; Thiamine B1: 0.63mg; Riboflavin (B12): 0.001mg; Nicotinic Acid: 3.0mg; Folic Acid: 0.1mg; Choline: 31.3mg; Ascorbic Acid ©: 0.1mg; Iron (Fe): 0.05mg; Cu: 0.25mg; Mn:6.00mg; Co: 0.5mg; Zn: 5.0mg; Sn:0.02mg.

**Growth performance and feed utilization of *Clarias gariepinus* fingerlings fed boiled Jatropha kernel meal based diet**

The growth performance and feed utilization of *Clarias gariepinus* fingerlings fed boiled Jatropha kernel meal based diets are presented in Table 4. The results showed that diet 4 has the highest value of mean weight gain (75.57g), followed by diet 3 and 2 (61.08g) and (52.47%) while diet 1 has the lowest value (45.039g) mean weight gain. Also, the daily growth rate per fish was highest in diet 4 (1.35g/day), followed by diet 3 and 1(1.09g/day) and (0.81g/day) while diet 2 has the lowest value (0.64g/day) daily growth rate per fish. Similarly, the specific growth rate was highest in diet 4 (2.25%/day), followed by diet 3 and 2 (2.00%/day) and (1.83%/day) while diet 2 has the lowest value (1.67%/day) specific growth rate. At the end of the feeding trial, the fish body weight gains were different from each other. The best growth response was shown by 30% boiled Jatropha kernel treatment (diet 4) and the worst was by fish received diet containing 10% boiled Jatropha kernel treatment (diet 2). Inclusion of 30% boiled Jatropha kernel treatment in the diet caused increase in growth performance and feed utilization.

The food conversion ratio (FCR) ranged between 2.34 (diet 4) and 2.88 (diet 1) while the protein efficiency ratio (PER) also ranged between 1.13 (diet 1) and 1.85 (diet 4). Percentage survival rate was highest in fish fed diet 4 (96.67%) but significantly different ($p>0.05$) from numerically lowest value (83.33%) in diet 2. Mean weight of *Clarias gariepinus* fingerlings fed graded levels boiled Jatropha kernel meal based diets for 56 days is presented in Figure 1.

**Water quality parameters**

28.50°C, dissolved oxygen 5.10 - 5.80mg/l, pH 7.50 – 8.00 and conductivity 320 - 350µhom/cm$^3$. This water parameter ranges fall within acceptable limit in aquaculture (30).

Water quality was monitored every week throughout the feeding trials. Temperature ranged between 26.98 - This study reveals the possibility of utilizing boiled Jatropha kernel in the diet of fish using *Clarias gariepinus* as a case study. From the analysis carried out, proximate composition of the raw and boiled Jatropha kernel agrees with the findings of 25) who obtained 35.3% and 35.6% crude proteins for unheated and heated (30mins) to feed rats and fish (Carp, *Cyprinus carpio*).

It has also been reported that the level of essential amino acids of the Jatropha kernel meal of the non-toxic variety are higher than that of FAO reference protein except for lysine (9) which is necessary for growth and development. A comparison between Jatropha meal and soybean reveals an almost similar pattern for all essential amino acids except lysine and sulphur-amino acids; these are lower and higher respectively in Jatropha meals. Protein requirement is given high priority in any nutritional study because it is the single nutrient that is required in the largest quantity for growth and development and also the most expensive ingredient in diet formulation (32;33).

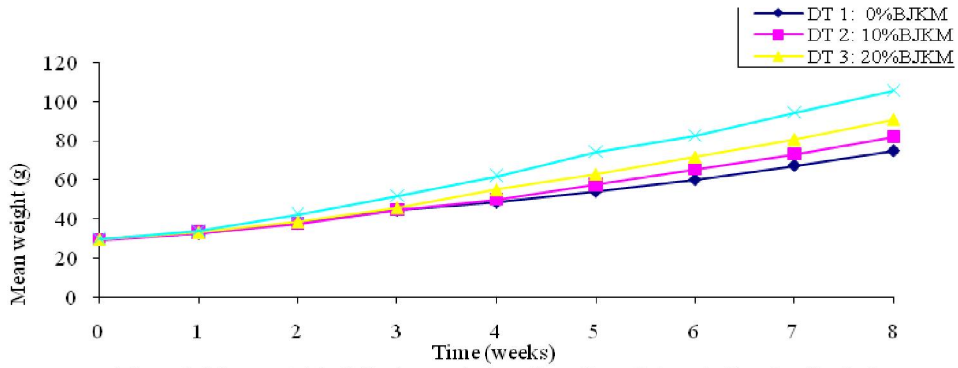


Figure 1: Mean weight of *Clarias gariepinus* fingerlings fed graded levels of boiled Jatropha kernel-based diets

Previous works of 34 showed the need to use plant meal in combined form to produce the cheapest and required nutrient for fish and this formed the basis of this research work. The replacement of fishmeal by alternate sources of protein has met with varied degree of success, depending on the nature and composition of ingredients, inclusion level and method of processing.

This study agrees with the findings of 32 who conducted a study on the nutritional evaluation of sunflower and sesame seed meal in *Clarias gariepinus*: An assessment by growth performance and nutrient utilization. Toxic or anti-nutritive compounds in Jatropha seed or kernel meal may cause irritation of digestive tract which then decreases feed intake and growth. Boiling or cooking is one of the methods of removing anti-nutritional factors in Jatropha kernel meal. This resulted in improvement of quality of feed, ensures regular feed supply even during slump period (Seasonal) and reduces the cost of production.

**Conclusion**

Based on the findings of this study, it is inferred that Jatropha kernel meal is rich in protein and with essential amino acids. It can also be inferred that inclusion of Jatropha kernel meal in the diet of fish will improve growth yield of *Clarias gariepinus*. Though, the Jatropha kernel meal can be included up to 50%, since the fish showed good appetite for all the treatment diets. Growth performance of *Clarias gariepinus* increases with increase in inclusion level of boiled Jatropha kernel meal in the diet. Inclusion of boiled Jatropha kernel meal in the diet does not have detrimental effect on *Clarias gariepinus* as revealed by the survival rate. 30% boiled Jatropha kernel meal replacement with soyabean meal is optimal for *Clarias gariepinus* growth performance.

Table 2: The chemical composition of raw and boiled Jatropha kernel

| **Constituent** | **Raw Jatropha kernel** | **Boiled Jatropha kernel** |
|---|---|---|
| Moisture contents | 5.28 | 7.43 |
| Crude protein | 31.23 | 37.75 |
| Crude fat | 27.97 | 30.09 |
| Crude ash | 5.20 | 4.08 |
| Crude fibre | 4.28 | 3.90 |
| Nitrogen free extract (NFE) | 31.32 | 24.18 |
| Gross energy (kcl/g) | 569.18 | 596.78 |
| Digestible energy (kcl/g) | 414.17 | 436.31 |

NFE (Nitrogen Free Extract) = 100 – (Crude protein + crude lipid + crude fibre + total ash).

Table 3: Proximate composition of the experimental diets

| Parameters | 1 (0%) | 2 (10%) | 3 (20%) | 4 (30%) |
|---|---|---|---|---|
| Moisture content | 8.02 | 9.92 | 7.76 | 7.08 |
| Crude protein | 40.31 | 40.57 | 40.40 | 40.82 |
| Crude lipid | 9.94 | 9.52 | 9.39 | 9.13 |
| Crude ash | 6.63 | 6.50 | 6.40 | 6.32 |
| Crude fibre | 3.89 | 3.64 | 3.59 | 3.36 |
| NFE | 39.23 | 39.70 | 40.22 | 40.37 |
| Gross energy (kcl/g) | 482.52 | 482.24 | 481.90 | 482.43 |
| Digestible energy (kcl/g) | 319.68 | 318.53 | 318.01 | 317.75 |

NFE (Nitrogen Free Extract) = 100 – (Crude protein + crude lipid + crude fibre + total ash).
Gross energy = Caloric value of protein 5.65, NFE 4.1 and lipid 9.45 kcal/g (NRC, 1993).
Digestible energy = Caloric value of protein 3.5, NFE 2.5 and lipid 8.1 kcal/g (Brett, 1973).

Table 4: Growth performance and feed utilization of *Clarias gariepinus* fingerlings fed boiled Jatropha kernel meal based diets

| | Diets | | | |
|---|---|---|---|---|
| Parameters | 1 (0%) | 2 (10%) | 3 (20%) | 4 (30%) |
| Initial body weight (g) | 29.36±0.25$^a$ | 29.30±0.24$^a$ | 29.56±0.46$^a$ | 29.85±0.52$^a$ |
| Final body weight (g) | 74.75±0.75$^d$ | 81.76±3.25$^c$ | 90.64±1.10$^b$ | 125.35±0.91$^a$ |
| Body weight gain (g) | 45.39±0.52$^d$ | 52.47±3.49$^c$ | 61.08±0.75$^b$ | 75.57±0.95$^a$ |
| Daily growth rate (g/day) | 0.81±0.01$^b$ | 0.64±0.47$^b$ | 1.09±0.02$^{ab}$ | 1.35±0.02$^a$ |
| Percentage weight gain (%) | 154.59±0.83$^d$ | 179.17±13.3$^c$ | 206.68±2.6$^b$ | 253.20±5.56$^a$ |
| Specific growth rate (%/day) | 1.67±0.01$^d$ | 1.83±0.09$^c$ | 2.00±0.02$^b$ | 2.25±0.03$^a$ |
| Total feed consumed (g) | 130.72±0.64$^d$ | 136.26±4.27$^c$ | 145.82±1.9$^b$ | 164.66±1.58$^a$ |
| Food conversion ratio (FCR) | 2.88±0.02$^a$ | 2.60±0.09$^b$ | 2.39±0.01$^b$ | 2.34±0.27$^b$ |
| Protein Intake (%) | 40.31±0.10$^c$ | 40.57±0.05$^b$ | 40.40±0.02$^c$ | 40.82±0.13$^a$ |
| Protein efficiency ratio (PER) | 1.13±0.01$^d$ | 1.30±0.09$^c$ | 1.51±0.02$^b$ | 1.85±0.02$^a$ |
| Survival rate (%) | 93.33±5.77$^a$ | 83.33±2.89$^b$ | 95.00±5.00$^a$ | 96.67±2.89$^a$ |
| Culture period (days) | 56 | 56 | 56 | 56 |

Mean values within row with the same superscript are not significantly different (P>0.05)

Table 4.4: Mean water quality of the 55L rectangular plastic tank at weekly intervals during the experimental period

| Weeks | Water temperature (°C) | Dissolved Oxygen (mg/l) | pH | Conductivity (μhom/cm$^3$) |
|---|---|---|---|---|
| 1 | 26.80 | 5.20 | 7.5 | 320 |
| 2 | 26.90 | 5.10 | 7.8 | 335 |
| 3 | 27.00 | 5.10 | 8.0 | 340 |
| 4 | 27.00 | 5.10 | 8.0 | 340 |
| 5 | 27.00 | 6.10 | 8.0 | 340 |
| 6 | 28.50 | 5.10 | 8.0 | 320 |
| 7 | 27.00 | 5.80 | 8.0 | 350 |
| 8 | 27.00 | 5.80 | 8.0 | 350 |
| **Mean** | **27.15** | **5.49** | **7.91** | **337** |

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