Chemical Analysis and Nutritional Assessment of *Artocarpus heterophyllus* Lam. (Jack Fruit) Defatted Seeds used as Additive in Feed for *Clarias gariepinus* post juveniles

[1] Flora Eyibio Olaifa, [2]Ibironke Adetolu Ajayi and [2]Idris Olawale Raji

[1]Department of Aquaculture and Fisheries Management, University of Ibadan, Ibadan, Nigeria
[2]Department of Chemistry, University of Ibadan, Ibadan, Nigeria
frajayi@yahoo.com

Abstract: A 49-day feeding trial was carried out with feeds supplemented with microgram quantities of the defatted seeds of Artocarpus heterophyllus in the diets of Clarias gariepinus at the post juveinile stage. Five diets at 40% crude protein were formulated containing 0, 15, 30, 45 and $60x10^6$ µg DAH seed as additive. Each dietary treatment was replicated three times with 10 fish per replicate. Proximate composition of the defatted seed showed that it was rich in protein, carbohydrate and minerals. Fish on DAH–supplementd diets had better survival rates than the control. Haematology, plasma biochemistry and gross tissue examination were also carried out. No significant differences ($p<0.05$) were observed between the fish on DAH-containing treatments and the control. There might be need to further process and test defatted A. heterophyllus seeds as either probiotic or prebiotic on young fish for longer periods in order to take advantage of its rich supply of nutrients.
[Olaifa, F.E. Ajayi, I.A, Raji, I.O. **Chemical Analysis and Nutritional Assessment of *Artocarpus heterophyllus* Lam. (Jack Fruit) Defatted Seeds used as Additive in Feed for *Clarias gariepinus* post juveniles.** *J Am Sci* 2013;9(8):128-135]. (ISSN: 1545-1003). http://www.jofamericanscience.org. 19

Keywords: *Artocarpus heterophyllus*, defatted seeds *Clarias gariepinus*, haematology, histology

1. Introduction

Fish require diets with proper balance of nutrients for normal growth and development from conventional and non conventional sources. Available feedstuffs are used by man as food and raw materials for industry. Research is ongoing to source for alternatives with fish feeds generating a lot of interest (El-Dakar et al., 2008; Gabriel et al., 2007; Malik, 2009) since fish is an important and less expensive source of protein and minerals than meat.

Artocarpus heterophyllus (Jackfruit) belongs to the family Moraceae along with Artocarpus altilis (Breadfruit), A. camansi (Breadnut), A. integer (Champedak), A. lakoocha (lakoocha), A. odoratissimus (Maranga), Treculia africana (African breadfruit) and Morus spp. (Mulberries). It is native to Western Ghats of India, Malaysia though also found in many tropical and subtropical countries (Elevitch and Manner, 2006; Rajiv et al., 2009). Jackfruit is a medium- sized, evergreen tree that grows to a height of 8-25m and attains a stem diameter of 30- 80 cm. It is most common in lowland forests up to 250m, decreasing in abundance up to 1,000m above sea level. It thrives best in moist tropical environments below 1000m. Jackfruit grows best in well drained, deep soils of moderate fertility but tolerates a wide range of soils including shallow limestone, sand, and rocky substrates but does not tolerate water stagnation or poor drainage. A distinguishing feature of Jack fruit tree is its ability to produce a higher yield of fruits than any other tree in the Moraceae family producing 70-100kg of fruit per tree depending on variety, cultural practice and environmental factors. The ripe fruit of A. heterophyllus can be eaten raw or processed into several delicacies like jam, jelly and chutney (Elevitch and Manner, 2006).

The fruits of jackfruit are rich in protein (Duke, 1992). The seeds are rich in protein and fat and can be consumed in roasted form (Biswas and Rahmatullah, 2011). Virtually every part of the plant is useful- as a source of timber with anti-termite activity, leaves and fruit wastes as feed for cattle, goats and pigs. Its leaves, bark, roots and fruits have medicinal uses (Odoemelam, 2005; Rajah et al., 2010).

The seeds of A. heterophyllus are light brown, rounded, 2–3 cm in length by 1–1.5 cm in diameter, and enclosed in thin, whitish membranes. Up to 500 seeds can be found in a fruit. Seeds are hardy and can be stored up to a month in cool and humid conditions (Elevitch and Manner, 2006 and Rajiv et al., 2009). The seeds of *A. heterophyllus* are rich in protein (Chowdhury et al., 2012). Soong and Barlow (2004) suggested the use of jack fruit seeds as a source of natural food additives and ingredients while Bhushan et al. (2008) and Zzaman (2012) explored its natural antioxidant properties.

Clarias gariepinus (African sharp tooth catfish) belongs to the family *Clariidae* or air breathing catfishes. It has elongated body, large depressed bony head with small eyes; wide gill openings; air-breathing organs arising from gill arches (FAO, 2011). It is a highly valued fish in Nigeria and can withstand harsh environmental conditions. There is little information available on the use of Jackfruit as an additive or

component of any fish feed. In this study we investigate the chemical composition and nutritional effects of incorporating microgram quantities of defatted seeds of A. heterophyllus as additive to formulated feeds of C. gariepinus post juveniles.

2. Materials and methods

Mature fruits of A. heterophyllus were collected from the Botanical Garden, University of Ibadan and cut open and to remove the seeds. The outer layers of the seeds were removed manually (Ocloo et al., 2010) and sun-dried for 24 hours before milling with kitchen-sized mortar and pestle. The milled seeds were defatted using a soxhlet extractor with n-hexane (bpt. 60°C) as described by Ajayi et al. (2007). The residue was air-dried to lose the solvent by evaporating for one week, weighed and stored in an air-tight plastic container until needed.

Predetermined quantities of feed ingredients-fishmeal (72% crude protein), soybean (44% crude protein), maize, wheat offal (18% crude protein), vitamin-mineral premix, starch, dicalcium phosphate (DCP), salt, and vegetable oil, were purchased, milled into powder and used to formulate a 40% crude protein diet. Five diets, with four containing microgram quantities of defatted seeds of A. heterophyllus were formulated and turned into dough by adding small quantities of warm water. Treatment1(control) diet did not contain defatted A. heterophyllus seed while treatments 2 - 5 contained 15.0 x 10^6 µg, 30.0x 10^6 µg, 45.0x 10^6 µg and 60.0x 10^6 µg of the DAHS respectively (Table 1). Each diet mixture was treated separately, extruded through a 1/4mm die mincer of Hobart A-200T pelleting machine (Hobart GmbH, Rben-Bosch, Offenburg, Germany). The diets were sun-dried, broken mechanically into suitable sizes for the fish, packaged in labelled polythene bags and stored before use.

Table 1: Gross Compositions of Experimental Diets (%).

Ingredient	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Fishmeal	10.18	10.18	10.18	10.18	10.18
Soybean	20.35	20.35	20.35	20.35	20.35
Maize	30.99	30.99	30.99	30.99	30.99
Wheat offal	30.99	30.99	30.99	30.99	30.99
Vitamin/mineral premx	2.00	2.00	2.00	2.00	2.00
Starch	1.00	1.00	1.00	1.00	1.00
DCP	2.00	2.00	2.00	2.00	2.00
Salt	0.50	0.50	0.50	0.50	0.50
Vegetable oil	2.00	2.00	2.00	2.00	2.00
Total	100	100	100	100	100
DAH (x10^{6}µg)	Nil	15.00	30.00	45.00	60.00

DCP= Dicalcium phosphate, DAHS = Defatted *Artocarpus heterophyllus* seed.

Post juvenile *Clarias gariepinus* numbering one hundred and fifty (mean body weight: 150.08±0.03g) were obtained from a fish farm and transported to the laboratory in two 100-litre bowls. They were held in fifteen 40-litre circular plastic bowls and acclimatized for five days. De-chlorinated water (tap water exposed to air for 24 hours) was used for the study. The fish were randomly divided into five experimental groups with 10 fish per replicate and each treatment had three replicates. Every bowl was supplied with 38 litres of water and covered with nets. Water was changed every three days. Water quality parameters measured during the study were temperature, pH and dissolved oxygen. Fish were fed twice daily at 3% body weight, weighed weekly and feeding adjusted to the new body weights. Lengths of fish were also measured using a 30-cm ruler and recorded for each treatment. The experiment lasted for 49 days.

Changes in weight, length, relative growth rate, specific growth rate, condition factor, survival rate, protein efficiency ratio, feed conversion ratio (FCR) and nitrogen metabolism were monitored and calculations made using the following formulae:

Weight Gain (g) = ($W_1 - W_0$)

W_0 = Initial mean weight

W_1 = Final mean weight

Specific Growth Rate (SGR) = (Ln W_1 – LnW_0) / T × 100 (Brown, 1957)

Ln = Natural log

W_1 = final mean weight

W_0 = Initial mean weight

T = time interval

Relative Growth Rate (RGR) = Weight gained by fish (g)/Initial body Weight (g) x100

Condition Factor (K) =100 x W/L^3

W= final weight

L = Final standard length

Survival Rate, (S) % = $N_1 \times 100/N_0$

N_1 = Final number of fish at the end of experiment.

N_0 = Initial number of fish at the beginning of experiment.

Feed Conversion Ratio (FCR) = Dry weight of feed (g) /weight gain (g)

Nitrogen Metabolism (NM) = $\frac{(0.549)(a+b)h}{2}$

a= initial mean weight of fish
b= final mean weight of fish
h= experimental period in days (Nwanna, 2003)

Protein Efficiency Ratio (PER) = body weight gain (g)/weight of protein fed (g)

The proximate compositions of the defatted seeds, prepared feeds and fish carcass were carried out according to AOAC (2000). Nitrogen free extract was determined by difference [100 − (protein + crude fat + ash + crude fiber)]. The mineral compositions of the defatted A. heterophyllus seeds and fish samples were determined using the method described by Idouraine et al. (1996). One gram of each sample was dry–ashed in a muffle furnace at 550°C for 5 hours until a white ash was obtained. The minerals were extracted from ash by adding 3 ml of concentrated HNO_3 (63%). The digest was carefully filtered into 100 ml standard bottle and made up to mark with deionised water. Minerals were estimated with the use of an atomic absorption spectrophotometer (Perkin Elmer model 703, USA) as described by Onyeike and Acheru (2002). Histology and haematology of fish were carried out (MAFF, 1984; Schalm et al, 1975). Data obtained were subjected to one-way analysis of variance (ANOVA) and Duncan's multiple range test was used as follow up test.

3. Results

The results obtained during the study to determine the usefulness of A. heterophyllus defatted seeds as an additive in the diet of C. gariepinus post juveniles are presented on tables 2 -12.

The proximate composition of DAHS (Table 2) shows that the moisture content, crude protein, crude fibre, ash, fat and nitrogen-free extract were 12.34, 15.10, 6.09, 3.79, 1.20 and 61.48% respectively. The defatted seed was dominated by carbohydrate as depicted by high nitrogen free extract (60.80± 0.04%) though it also contained an appreciable amount of crude protein (15. 10 ±0.79%).

Table 2: Proximate composition of defatted *A. heterophyllus* seeds (%)

Parameter	%
Crude Protein	15.10 ±0.79
Crude Fibre	6.09 ±0.04
Ash	3.60 ±0.32
Fat	1.21 ±0.15
Moisture	12.34 ±0.78
Nitrogen Free extract	60.80± 0.04

Table 3: Mineral element composition of defatted *A. heterophyllus* seeds

Mineral	(ppm)
Calcium	200
Magnesium	130
Potassium	620
Phosphorus	200
Nitrogen	186
Sodium	7.99
Manganese	14.34
Zinc	32.87
Iron	75.60
Copper	8.46

The mineral profile of DAHS (Table 3) showed the presence of many essential elements with the dominant element as potassium (620ppm) while copper was least with 8.46 ppm.

Table 4: Proximate composition of diets containing defatted *Arthocarpus heterophyllus* seeds as additive (%)

Parameter	Control diet	15x10 ^{6} µg	30x10 ^{6} µg	45x10^6 µg	60 x10^6 µg
Crude Protein	40.64±0.32^a	40.50±0.22^a	40.75±0.22^a	40.41±0.38^a	40.31±0.35^a
Crude Fibre	4.84± 0.15^b	4.50± 0.02^b	3.25± 0.04^a	4.74± 0.05^b	5.07± 0.04^c
Ash	8.42± 0.27^a	8.62± 0.06^a	8.12± 0.03^a	8.21±0.03^a	8.20±0.04^a
Fat	7.70± 0.02^a	18.18± 0.18^c	7.42± 0.03^a	16.84±0.01^b	16.98±0.02^b
Moisture	6.61± 0.06^a	6.91± 0.03^a	6.76± 0.09^a	7.26± 0.02^b	6.71± 0.02^a
Nitrogen Free Extract	31.79± 0.02^a	21.29± 0.18^b	33.71± 0.04^a	22.54± 0.01^b	22.73± 0.03^b

Figures with different superscripts in the same row are significantly (P<0.05) different. DCP= dicalcium phosphate, DAHS = defatted *A. heterophyllus* seed

Table 4 shows the proximate composition of the experimental diets and the control. All the diets were similar in protein, ash and moisture contents but differed in crude fibre, fat and nitrogen free extract. Diet 5(60 x10^6 µg) had the highest crude fibre content (5.07%). Crude fibre and fat differed significantly (p<0.05) among diets.

Table 5: Proximate composition of fish on diets containing defatted *A. heterophyllus* as additive (%)

Composition (%)	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Crude Protein	59.97±1.45^a	63.40±0.20^{bc}	63.07±0.12^b	64.47±0.15^c	59.37±0.25^a
Crude Fibre	0.50±0.00^a	3.30±0.26^c	2.50±0.20^b	2.67±0.21^b	0.43±0.06^a
Ash	12.13±0.56^b	14.00±0.58^c	15.42±1.30^c	8.17±0.54^a	12.91±0.90^b
Fat	11.47±0.06^c	12.03±0.06^a	17.27±0.05^e	12.63±0.12^a	2.53±0.25^d
Moisture	13.35±0.37^a	10.20±1.35^a	23.85±0.10^b	11.22±0.55^a	22.2±6.90^b

Figures with different superscripts in the same row are significantly (P<0.05) different. DCP= Dicalcium phosphate, DAHS = Defatted *Artocarpus heterophyllus* seed.

Table 5 depicts the proximate composition of the fish after the feeding trial. All the fish on the experimental diets had higher crude protein content in their tissues than the control except diet 5 (60 x10 ^{6} µg inclusion of DAHS). Crude fibre and ash followed similar trends. Fat content of fish was lowest in fish on diet 5 (60 x10 ^{6} µg inclusion of DAHS).

Table 6: Growth performance and nutrient utilization of *Clarias gariepinus* post juveniles on diets containing defatted seeds of *A. heterophyllus*

Parameters	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Initial Mean Weight (g)	51.08±.03^a	51.08±0.03^a	51.08±0.03^a	51.11±0.09^a	51.08±0.03^a
Final Mean Weight (g)	64.17±4.17^a	62.05±4.17^a	60.58±2.37^a	60.50±2.72^a	59.76±5.88^a
Mean Weight Gain (g)	13.10±4.16^a	10.97±4.15^a	9.51±2.39^a	9.39±2.64^a	8.75±5.78^a
Mean Length Gain (cm)	5.97±0.76^a	5.30±0.50^a	5.63±0.58^a	5.47±0.29^a	4.97±0.29^a
Percent Weight Gain (PWG)	25.64±8.13^a	21.48± 8.12^a	18.61± 4.69^a	18.36± 5.13^a	16.81± 11.00^a
Feed Conversion Ratio (FCR)	1.15± 0.44^a	1.46± 0.69^a	1.58±0.37^a	1.35± 0.31^a	2.92± 3.10^a
Specific Growth Rate (SGR)	0.46±0.13^a	0.39± 0.14^a	0.35± 0.08^a	0.34± 0.09^a	0.32±0.20^a
Protein Efficiency Ratio (PER)	0.40±0.13^a	0.41±0.15^a	0.30±0.07^a	0.28±0.08^a	0.26±0.17^a
Condition Factor (K)	0.59±0.05^a	0.62±0.02^a	0.58±0.07^a	0.28±0.08^a	0.26±0.17^a
Nitrogen Metabolism (x10^2)	15.50±56.76^a	15.22±5.76^a	15.02±31.63^a	14.99±39.17^a	14.89±76.63^a
Survival Rate (%)	83.33	86.70	93.30	96.70	90.00

Figures with different superscripts in the same row are significantly (P<0.05) different.

Table 6 shows the performance of fish *C. gariepinus* on diets containing different quantities of A. heterophyllus defatted seeds. The fish on the control diet had the highest final mean weightand feed efficiency ratio while those on 60 x10 ^{6} µg inclusion of DAHS were lowest. Mean weight gain, length gain and nitrogen metabolism showed similar trends. The highest condition factor (0.62) was recorded on diet 2 (15x10^6 µg) while the lowest (0.58) was recorded in diet 3 (30x10^6 µg). The survival rate of fish was highest in diet 4(45 x10 ^{6} µg inclusion of DAHS) but lowest in control fish. Also, the highest and lowest feed conversion ratios, 2.92±3.10 and 1.15±0.44, were recorded in treatments 5 and 1 respectively. No significant differences (p>0.05) were observed in all the parameters.

Table 7: Mineral composition of fish on diets containing defatted *A. heterophyllus* as additive (ppm)

Minerals (ppm)	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Calcium	310.0	158.0	569.50	112.00	456.00
Potassium	329.50	78.70	281.00	166.00	298.00
Magnesium	46.30	16.25	57.25	15.61	56.20
Sodium	162.50	40.10	177.00	54.70	206.00
Zinc	1.61	0.49	1.94	0.47	1.78
Manganese	0.40	0.15	0.55	0.15	0.53
Iron	4.04	1.51	4.03	2.13	5.58
Copper	0.10	0.03	0.11	0.04	0.17

Table 7 shows the mineral composition of fish after the feeding trial. Calcium was the dominant element in the fish with the highest quantity recorded in fish on diet 3(30 x10 ^{6} µg inclusion of DAHS). Potassium was lower in all fish than initially present in the defatted seed of A. heterophyllus alone. Zinc, iron and copper were present in trace amounts in the fish. Aside from this spike of calcium in diet 3, fish in other treatment had potassium followed by sodium as dominant elements with copper being the least.

Table 8: Haematology of fish before and after treatment

Parameters	Before Trial	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
PCV (%)	26.00	46±2.83^c	30±2.83^a	32±2.83^{ab}	38±2.83^b	34±2.83^{ab}
Hb (mg/dl)	8.00	14.8±0.57^d	9.7±0.28^a	10.2±0.28^a	2.4±0.28^c	11.3±0.28^b
RBC (10^6/µl)	1.59	4.12±0.03^d	3.24±0.06^a	3.58±0.03^{bc}	3.66±0.06^c	3.52±0.03^b
WBC (10^3/µl)	19.43	14.85±71^d	15.80±283^a	21.0±141^{bc}	20.1± 141^c	16.1± 141^b
Platelets(10^3/ml)	107.50	150± 7071^b	140±7071^{ab}	242±2828^d	211± 1414^c	129±1414^a
Lymphocyte(%)	63.50	69±0.00^b	70±0.00^b	63±2.83^a	67± 0.00^b	63± 0.00^a
Heterophils(%)	34.00	24±0.00^a	22±0.00^a	33±2.83^b	26± 2.83^a	31± 0.00^b
Monocyte (%)	2.00	3±0.0	3.00±0.0	1.00±0.0	3.0± 0.0	2.0± 0.0
Eosinophils (%)	1.00	3±0.0^a	5±0.0^b	3±0.0^a	4± 0.0^{ab}	4± 0.0^{ab}
Basophils (%)	1.00	1±0.0	0.0±0.0	0.0±0.0	0.0±0.0	0.0± 0.0
MCHC (%)	30.84	32.98±1.9^a	32.53±4.0^a	32.63±2.9^a	32.75± 3.2^a	33.39±3.6^a
MCH (%)	50.29	35.93±1.6^d	29.94±0.4^{ab}	28.48±0.6^a	33.89±1.3^{cd}	32.10±0.6^{bc}
MCV (fl)	163.28	109.3±11^a	92.7±10^a	89.4±9^a	103.8±6^a	96.6±9^a

Figures with different superscripts in the same row are significantly (P<0.05) different.
PCV=packed cell volume, Hb= haemoglobin concentration, RBC= red blood cell counts, WBC =white blood cell counts, Lymp= lymphocytes, mono= monocytes, MCHC= mean corpuscular haemoglobin concentration, MCH = mean corpuscular haemoglobin, MCV = mean corpuscular volume.

The haematology (Table 8) of fish showed marked differences (p<0.05) in the packed cell volume, haemoglobin, red blood cell counts and white blood cell counts of fish between initial and final observations. There were also significant differences (p<0.05) among the diets in PCV, RBC, platelets and MCHC (mean corpuscular haemoglobin concentration). While the PCV and RBC increased in all treatments above initial values, drops from initial values were observed for mean corpuscular haemoglobin (MCH) and mean corpuscular volume (MCV). The RBC was highest in control fish but lowest before the commencement of the study. WBC was highest in diet 3 (30x10 ^{6} µg). Monocytes, eosinophils basophils and MCHC showed no significant differences among diets.

Table 9: Blood biochemistry of fish before and after the feeding trial

Parameters	Before study	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Total Protein	3.30	3.5±0.14^a	3.7±0.28^a	4.2±0.00^a	3.5±0.71^a	4.0±0.00^a
Albumin	1.30	1.2±0.28^a	1.3±0.00^a	1.5±0.14^a	1.2±0.28^a	1.5±0.00^a
Globulin	2.00	2.3±0.00^a	2.4±0.28^a	2.7±0.00^a	2.3±0.42^a	2.5±0.14^a
AG Ratio	0.65	0.5±0.00^a	0.5±0.14^a	0.6±0.00^a	0.5±0.0^a	0.60±0.0^a
AST(IU/L)	35.50	38±1.41^b	30±0.00^a	32±2.83^{ab}	35±1.4^{ab}	33±4.24^{ab}
ALT(IU/L)	20.50	29±0.00^b	21±1.41^a	23±4.24^{ab}	27±0.0^{ab}	25±4.24^{ab}

Superscripts with same letters in the same row are not significantly (P>0.05) different. AG ratio=albumin-globulin ratio, AST=Aspartate amino transferase, ALT= Alanine amino transferase.

Table 9 shows the blood biochemistry of fish during the study. Except for AST and ALT, no significant differences were observed among the diets with regards to blood biochemistry.

Table 10: The histology of fish performed at the end of the study

Organs	Control	15x10 ^{6} µg	30x10 ^{6} µg	45x10 ^{6} µg	60x10 ^{6} µg
Liver	Diffuse vacuolation of hepatocytes	Diffuse vacuolation of hepatocytes	Diffuse vacuolation of hepatocytes	Diffuse vacuolation of hepatocytes	Diffuse vacuolation of hepatocytes
Kidney	Marked congestion of the interstitium	Marked congestion of the interstitium	No visible lesions seen	No visible lesions seen	No visible lesions seen
Heart	No visible lesions seen	No visible lesions seen	No visible lesions seen	No visible lesions seen	No visible lesions seen
Gills	No visible lesions seen	No visible lesions seen	No visible lesions seen	No visible lesions seen	Severe sub-mucosal congestion

Table 10 depicts the histology of liver, kidney, heart and gill tissues at the end of the experiment. While vacuolations were observed in liver cells of all groups, marked congestion of the interstitium was observed in the control group and fish on diets containing 15x10 ^{6} µg. No visible lesions were observed in the kidneys of fish on greater inclusion levels of *A. heterophyllus* (30-60 10 ^{6} µg). No lesions were also were observed in the heart and gills of fish

except for severe sub mucosal congestion of gills of fish on diet 5 ($60x10^6$ µg).

Discussion

The growth performance of *C. gariepinus* post juveniles on feeds with defatted seeds of *Arthocarpus heterophyllus* included as additive was investigated. Diets with similar gross composition (Table 1) were formulated with the addition of varying quantities of defatted seeds of A. heterophyllus as 0, 15, 30, 45 and 60 $x10^6$ µg representing diets 1-5 respectively. Feed formulation was based on 40% crude protein which was more than adequate for C.gariepinus at the post juvenile stage.

The proximate composition of DAHS (Table 2) showed higher moisture and protein contents obtained than those reported for the whole seed (Chowdhury, 2012 and Ocloo *et al*., 2010). These differences could be due to the defatting process which removed oil from the seeds, different varieties of the jackfruit used, maturation of the seeds and environmental conditions (Rahman *et al*., 1999). The protein content in DAHS was higher than what obtains in many cereals (8-9%). The ash content of the defatted seed of A. heterophyllus was higher compared to Garcinia mangostana (Ajayi *et al*., 2007) but lower than 4.5% for Caesalpinia pulcherima (Yusuf *et al*., 2007). The fat content of DAHS was lower than for whole seed (Ajayi *et al*., 2007). Carbohydrate dominated the proximate composition of the defatted seed (60.8%) implying that it is a good energy source. The crude fibre content was within range reported for legume seeds (Prakash *et al*., 2001).

The mineral assay of defatted seeds of *A. heterophyllus* (Table 3) showed potassium as the dominant mineral (620 ppm) which was similar to previous reports for mature whole seeds (SCUC, 2006; Ajayi, 2008). The defatted seed flour was also a good source of other minerals such as calcium, iron, magnesium and sodium.

The proximate analysis of the formulated diet (Table 4) indicated that the inclusion of DAHS in microgram quantities did not have significant effect on the protein contents of the diets. Mixing of DAHS with other ingredients tended to increase the ash content of the formulated diet. However crude fibre and nitrogen free extract contents of the diets were lower on mixing with other ingredients.

The proximate composition of fish (Table 5) showed significant differences ($p<0.05$) in protein, crude fibre, ash fat content and moisture contents of fish. The fish responded differently to the presence of DAHS in their diets. The growth performance and nutrient utilization of the fish during this study are presented in Table 6. No significant differences were observed in all growth indices among the treatments. Though the control fish performed best in all growth parameters, it was lowest on survival rate when compared with diets containing DAHS. This implied that the addition of DAHS as an additive to the diet supported better survival of the fish during the study. The high survival rates recorded in the DAHS-containing diets may also be attributed to the boosting of the immune system of fish by the addition of the minerals and amino acid–rich DAHS to the test diets.

The condition factor indicated that fish in all the treatments were in good condition throughout the study but less than 1 on all diets. Edward *et al.* (2010) stated that a condition factor above 1 indicates better utilization of feeds by the fish for growth and development. This could be due to the brevity of the experimental period (49 days) which may not have allowed adequate adjustment of fish to the test diets. Feed conversion ratio is inversely proportional to effective nutrient utilization. This implies that the control diet with lower feed conversion ratio was better utilized for growth than diets with DAHS. The absence of significant differences ($p>0.05$) among diets in the feed conversion ratios in all the treatments implied almost equal levels of nutrient utilization in all the treatments with the best for fish on control diet.

The mineral composition of fish (Table 7) at the end of the feeding trials showed potassium as the dominant element in the control group. However there was a very high concentration of calcium in fish on diet 3 ($30x10^6$µg). This was a departure from the dominance of potassium in the defatted seeds of *A. heterophyllus*. The prevalence of calcium in the tissue of fish over the other minerals in diet 3 differed from previous observations by Fawole *et al.* (2007) and Onyia *et al.* (2010) who obtained sodium and potassium respectively to be the dominant mineral in their studies.

Haematology (Table 8) is an important tool in the monitoring of physiological and pathological changes in fishes. Normal haematology in fishes has been investigated, (Xiaoyun *et al*., 2009). Haemoglobin concentration reflects the amount of oxygen available to an organism. A reduction or increase in haemoglobin concentration reflects a decrease or increase in the amount of oxygen available to the organism. Since oxygen is required for normal body metabolism and proper functioning of the brain, the fish with a higher haemoglobin concentration is expected to perform better than those with lower concentration (Adeyemo, 2007).

Plasma biochemistry (Table 9) showed no significant changes ($p>0.05$) in protein, globulin and albumin–globulin ratio. Alanine amino transferase (ALT) increased while and aspartate amino transferase AST) decreased in all groups from the initial values and differed significantly ($p<0.05$) among treatments. These observed changes were similar to the

observations of Shalaby *et al.* (2006). The changes in these liver enzymes were regarded as a reflection of the activities occurring in the liver due to the presence of DAHS as an additive in the feed. Blood biochemistry can also be used as health indicators in fish (De Pedro *et al*., 2005 and Satheeshkumar *et al*., 2011).

A gross examination of the tissues (Table 10) showed alterations mainly in the liver and kidney of the fish. All groups of fish including the control fish showed some degree of vacuolation of the hepatocytes. The liver is the organ involved in the metabolism, detoxification, excretion of chemicals and xenobiotics in the body (Pathan *et al*., 2010). Vacuolation has been observed to be a common response to the presence of chemicals in fish (Shaw and Handy, 2006). In the kidneys, congestion of the interstitium was only observed in the control (0 µg DAHS) and the diet with $15x10^6$µg DAHS. No visible lesions were observed in diets containing $30\text{-}60x10^6$µg DAHS. The higher quantities of DAHS in the diets must have produced some positive effects on the kidney as shown by the absence of lesions. The kidney is responsible for osmoregulation in the fish and excretion of wastes. No lesions were observed in all the heart and gill tissues of fish during this study.

Conclusion

All parts of *A. heterophyllus* are useful as food for man and raw materials for industry. The use of its seeds and defatted seeds needs to be further explored in fish diets for possible probiotic or prebiotic effects on younger fish (fingerlings, juveniles). Exposure of the fish to the diets for longer periods would give fish a better opportunity to adapt to the feed. Fry, fingerlings and juveniles also tend to grow faster and respond to dietary treatments better. Other methods of processing the seeds could also be explored such as cooking or toasting which would not require the use of chemical. Further study could be undertaken without defatting the seeds of A. heterophyllus. Since defatted A heterophyllus seeds contain a lot of carbohydrate, research could be targeted at completely substituting a carbohydrate component of fish feed with defatted *A. heterophyllus* seeds.

References

1. El-Dakar, A. Y., Hassanien, G. D., Gad, S. S. and Sakr, S. E. (2008). Use of Dried Basil leaves as a feeding attractant for hybrid tilapia, Oreochromis niloticus X Oreochromis aureus, fingerlings. Med. Aquaculture Journal, 1: 35-44.
2. Gabriel, U. U., Akinrotimi, O. A., Bekibele, D. O., Onunkwo, D. N. and Anyanwu, P. E.(2007). Locally produced fish feed: potentials for aquaculture development in Sub-Saharan Africa. Afr. Journal of Agricultural Research, 2(7), 287-295.
3. Malik, M. E. K. (2009). Utilization of some Medical plants as feed additives for Nile Tilapia, Oreochromis niloticus, feeds. Med. Aquaculture Journal 2, 10-19.
4. Elevitch, C.R., and Manner, H.I. (2006). Artocarpus heterophyllus (jackfruit), ver. 1.1v. In: Elevitch, C.R. (ed.). Species Profiles for Pacific Island Agroforestry. Permanent Agriculture Resources (PAR), Hōlualoa, Hawai'i. (http://www.traditionaltree.org)
5. Rajiv, G., Prakash, O., Rajesh, K. and Anurag, M., (2009). Artocarpus heterophyllus (Jackfruit): An Overview. Pharmacognosy Review 3, 353- 358.
6. Duke, J.A. (1992). Handbook of phytochemical constituents of GRAS herbs and other, economic plants. Boca Raton, FL. CRC Press.
7. Biswas, K.R; Rahmatullah, M. (2011).A survey of non-conventional plants consumed during times of food scarcity in three adjoining villages of Narail and Jessore Districts, Bangladesh. American-Eurasian Journal of Sustainable Agriculture, 5, 1-5.
8. Odoemelam, S. A. (2005). Functional properties of raw and heat processed Jackfruit *(Artocarpus heterophyllus)* flour. Pakistan Journal of Nutrition, 4, 366-370.
9. Rajah, S., Reddy, P., Sushma, Y., Rani, B. V., and Reddy, G. H. (2010). Potentiation of local Anaesthetic activity of *Artocarpus heterophyllus* latex with adrenaline and pH by infiltration method. International Journal of Research in Ayurveda & Pharmacy, 1, 553-558
10. Chowdhury, A.R., Bhattacharyya, A.K.; and Chattpadyyay, P. (2012). Study on functional properties of raw and blended jackfruit seed flour (a non conventional source) for food application. Indian Journal of Natural Products and Resources, 3, 347-353.
11. Soong, Y.Y. and Barlow, P.J. (2004). Antioxidant activity and Phenolic content of selected fruit seeds. Food Chemistry 88, 411-417.
12. Bhushan, S., Kalia, K., Sharma, M., Singh, B., Ahuja, P.S. (2008). Processing of apple pomace for bioactive molecules. Critical Reviews in Biotechnology, 28, 285-296.
13. Zzaman, W. (2012). Optimization of anti oxidant extraction from jackfruit (Artocarpus heterophyllus Lam.) seeds using response surface methodology. M.Sc. Dissertation, Ghent University, 2012.
14. Food and Agriculture Organization of the United Nations (FAO, 2011). Cultured Aquatic Species Information Programme, Clarias gariepinus (Burchell, 1822).
15. Ocloo, F. C. K., Bansa, D., Boatin R., Adom, T. and Agbemavor, W. S. (2010). Physico-chemical, functional and pasting characteristics of flour produced from Jackfruit (Artocarpus heterophyllus)

seeds. Agriculture and Biology Journal of North America, 1, 903-908.
16. Ajayi, I. A., Oderinde, R. A., Ogunkoya, B. O., Egunyomi, A. and Taiwo, V.O. (2007). Chemical analysis and preliminary toxicological evaluation of Garcinia mangostana seeds and seed oil. Food Chem. 101, 999.
17. Brown, M. E. (1957). Experimental Studies on growth. In: Brown, M.E. (Ed.). The Physiology of Fishes, 361-400. Academic Press, NY, USA
18. Nwanna, L. C. (2003). Nutritional value and digestibility of Shrimp head waste meal by African Catfish, *Clarias gariepinus* (Burchell 1822). Ph.D Thesis. University of Ibadan, Nigeria.
19. Association of Official Analytical Chemists International (2000). Official Methods of Analysis.17th ed. AOAC International, Gaithersburg, MD, USA.
20. Idouraine, A., Kohlhepp, E.A. and Weber, C.W. (1996). Nutrient constituents from eight lines of naked seed squash. Journal of Agricultural Food Chemistry, 44, 721– 724.
21. Onyeike, E. N., Acheru, G. N. (2002). Chemical composition of selected Nigerian oil seeds and physicochemical properties of the oil extracts. Food Chemistry, 77, 431-437.
22. MAFF (1984). Ministry of Agriculture and Food Manual of Veterinary investigation. Laboratory Techniques. Volume 2. Reference Book 390. Third Edition.
23. Schalm, O. W., Jain, N. C. and Carrol, E. J. (1975). Veterinary haematology 3rd ed. Lea and Febiger Philadelphia. p. 340; 360-470.
24. Rahman, M.A., Nahar, N., Jabbar, M. A., and Mosihuzzaman, M., (1999). Variation of carbohydrate composition of two forms of fruit from jack tree (Artocarpusheterophyllus L.) with maturity and climatic conditions. Food Chemistry, 65, 91–97.
25. Yusuf, A. A., Mofio, B. M. and Ahmed, A. B. (2007). Nutrient contents of Pride of Barbados (Caesalpinia pulcherrima Linn.) seeds. Pakistan Journal of Nutrition, 6, 117-121.
26. Prakash, D. N., Abhishek, N., Tewari, S. K., and Pushpangadan, P. (2001).Underutilized legumes: Potential sources for low-cost protein. International Journal of Food Science and Nutrition, 52, 337-341
27. SCUC (2006). Jackfruit Artocarpus heterophyllus, Field Manual for Extension Workers and Farmers, SCUC, Southampton, UK.
28. Ajayi, I. A. (2008). Comparative study of the chemical composition and mineral element content of Artocarpus heterophyllus and Treculia africana. Bioresource Technology, 99, 5125–5129.
29. Edward, A., Ladu, B. M. B. and Elihu, A. (2010). Growth, survival and production economics of C. gariepinus fingerlings at different stocking densities in concrete tanks. African Journal of General Agriculture, 6, 59-66.
30. Fawole, O. O., Ogundiran, M. A., Ayandiran, T. A., and Olagunju, O. F. (2007). Proximate and mineral composition in some selected fresh water fishes in Nigeria. Internet Journal of Food Safety, 9, 52-55.
31. Onyia, L. U.; Milam, C.; Manu, J. M. and Allison D. S. (2010). Proximate and mineral composition of some fresh water fishes in upper river Benue, Yola, Nigeria. Continental Journal of Food Science and Technology, 4, 1-6.
32. Xiaoyun, Z, Minyun,L, Khalid, A, Welmin, W. (2009). Comparison of haematology and serum biochemistry of cultured and wild Dojo Loach Misgurnrus anguillicaudatus. Fish Physiology Biochemistry, 35, 435-441.
33. Adeyemo, O. K. (2007). Haematological profile of Clarias gariepinus (Burchell, 1822) exposed to lead. Turkish Journal of Fisheries and Aquatic Sciences, 7(2) 163-169.
34. Shalaby, A. M, Khattab, Y. A and Abdel -Rahman, A. M. (2006). Effects of garlic (*Allium sativum*) and chloramphenicol on growth performance, physiological parameters and survival of Nile tilapia. Journal of Venomous of Animal Toxins include Tropical Diseases, 12, 172 – 201.
35. De Pedro, N. Guijarro, A.I, Lopez-Patiño, M.A. Martinez-Ălvarez, R., Delgado M.J. (2005). Daily and Seasonal variations in haematological and blood biochemical parameters in the tench, *Tinca tinca* Linnaeus, 1758. Aquaculture Research, 36, 1185-1196.
36. Satheeshkumar, P., Ananthan, G. and Senthilkumar, D., Lagadeesam, L. (2011). Haematology and biochemical parameters of different feeding behaviour of teleost fishes from Vellar Estuary, India. Comparative Clinical Pathology, 1-7. Springer Verlag, London, Ltd.
37. Pathan, T. S., Shinde, S. E., Thete, P. B. and Sonawane, D. L. (2010). Histopathology of liver and kidney of Rasbora daniconius exposed to paper mill effluent. Research Journal of Biological Sciences, 5, 389-394.
38. Shaw, B. J., and Handy, R. D. (2006). Dietary copper exposure and recovery in Nile Tilapia. Aquatic Toxicology, 76, 111-121.