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ENZYMATIC AND HEAMATOLOGICAL CHANGES IN RATS (*RATTUS NORVEGECUS*) FED WITH DEFATTED OGIRI (FERMENTED *CITRULLUS VULGARIS)* AND MELON SEEDS

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ABSTRACT
Samples of 'Ogiri' (fermented *Citrullus vulgaris*); fermented with an attested strain of *Bacillus subtilis* (CRBS23) and unfermented melon seeds were defatted using extraction techniques. Diets containing the same quantity (20%) of protein supplement with 4:5 mixture of vitamins and minerals were prepared from the Ogiri and unfermented melon seeds and fed on starved rats (*Rattus norvegecus*) for 4weeks after which they were subjected to enzymatic and heamatological evaluation. The highest weight gained and protein efficiency ratio (PER) was recorded in unfermented *Cucumeropsis manni* (UCM), which was significantly higher (at α=0.05) than others. Generally, the weight of organs of the rats from fermented diet groups were relatively higher than those that are of unfermented diets except in few cases. The highest Acid Phosphate (ACP) (230.2µmol/ml) was obtained in the kidney of rats fed with fermented *Citrullus lanatus* (FCL) Significantly higher (at α=0.05) than all other diet groups. While the ACP activities in the heart and liver were low in rats fed with protein free feed (PFF) (114.38 µmol ml^{-1}) and fermented *Citrullus vulgaris* (FCV) (188.14 µmol ml^{1}). Rats fed with fermented *Citrullus vulgaris* (FCV) showed the highest Alkaline Phosphate (ALP) (230.54 µmol ml^{-1}) and Glutamyl Oxaloacetate Transaminase (GOT) (230.504µmol ml^{-1}) activities in the heart. Highest ALP (2678.30 µmol ml^{-1}) and GOT (80.60µmol $ml^{-}1$) activities were recorded respectively in the kidney and heart of the rats in diet group FCM. The ALP level in the liver varied significantly (at α=0.05) among the diet groups while The GOT level in kidney was generally lower than that of the other organs and blood serum. There was a good indication in the research that the dietary protein was well utilized by rats thus defatted 'Ogiri' may serve as a good food supplements in animal feeds.
Key words: Fermented *Citrullus vulgaris*, ogiri, heamatological, Glutamyl Oxaloacetate Transaminase

1.0 INTRODUCTION

Fermented foods are essential part of diet in all part of the world and these foods have constituted a significant portion of African diets for decades (Achi, 2005a). Among the merit of fermentation is the preservation of substantial amount of food through lactic acid, alcohol, acetic acid and alkaline vitamins; biological enrichment of food substrates with protein, essential amino acids, essential fatty acids and vitamins. Also enrichment of diet through development of diverse flavours, aroma, texture in food substrates and elimination of anti nutrients; couple with decrease in the cooking time and fuel requirements (Osho *et al*., 2009). It is only in recent times that the production of fermented vegetable proteins for use as food condiments is craft-based. By observation and trials, man has selected and developed more conducive methods that give it more desirable changes (Latunde-Dada, 2000). Remarkably in many areas of Nigeria today, they are still made in traditional ways, with success depending upon observance of good manufacturing practices and control of environmental conditions during the manufacturing phase. Some of the most important food condiments are "iru" and "dawadawa" produced from African locust bean (*Parkia biglobosa*) (Osho *et al*., 2009); "Ogiri" which is produced from melon seeds (*Citrullus Vulgaris*) (Ogbonna *et al.,* 2000) and that (Ogiri-Igbo) produced from castor oil seeds (Ibe, 2009).

Melon Seeds are leguminous plants which are annuals with monoecious flowering characteristic; melons are classified as a pepo, a fruit in which the ovary wall is fused with the receptacle tissue to form a hard rind or skin. Fruits are usually spherical or oval, oblong with smooth glabrous, having no hair surfaces some are deeply ridged, while others are covered with a reticulate corky netting (Akpambang, 2008). Flesh colors range form white to green, pink or orange. Melon seed has been well noted for essential compositions associated with its edibility (Oyenuga, 1986).

Citrullus vulgaris is a variety of melon seed which is popularly called "egusi" in West Africa which is a creeping annual plant and intercropping plant which makes use of traditional farming practices, it thrives well on rich light soil in the hot climatic regions of Africa (Akpambang *et al.,* 2008). *Citrullus vulgaris* is a member of the family *Cucurbitacea* and are widely cultivated for their seeds which have high content of fat and protein. The seeds are obtained either in shelled or unshelled forms in West African markets. The seeds can be ground or

milled as a soup condiments named "Ogiri" (Ogbonna *et al.,* 2000). "Ogiri" is a fermented, proteinous Nigerian soup condiment .It is an oily paste produced mainly from oil seeds such as melon seeds, castor seeds, fluted pumpkin seeds. This is mainly fermented by *Bacillus subtilis* (Falegan, 2011). It is one of the condiments consumed in the eastern and western parts of Nigeria especially the Ijebu ethnic groups. "Ogiri" is characterized with very strong pungent odour. "Ogiri" is presumed to be free of mycotoxins.

Enzymes are proteins with catalytic properties due to their powers of specific activation of their substrate (David and Falegan, 2007). Enzymes are retained within their cells of origin by the plasma membrane surrounding the cell. The plasma membrane is metabolically active part of the cell and its integrity depends on the cell's energy production. Any process that impairs energy production, either by depriving the cell of oxidizable substrate or reducing the efficiency of energy production by restricting the access of oxygen, will promote deterioration of the cell membrane. The membrane will become leaky and if cellular injury becomes irreversible, the cell will die. Small molecules are the first leak from the damage or dying cells, followed by larger molecules such as enzymes and ultimately the whole content of the necrotic cells are discharged. Generally speaking, both the haematological and biochemical blood components are influenced by quality and quantity of feed and also the level of anti-nutritional elements or factors present in the feed (Ahamefule *et al.,* 2006). Viscera organs perform different functions in the body. Beside these functions, these organs help in housing a number of marker enzymes that can be used to ascertain the functionality of various organs such as liver, kidney, heart, spleen and brain. The enzymes are predominant in these organs and are released into the blood when there is a damage or biliary obstruction, thereby leading to several fold increase of such enzymes in the serum (Ahamefule *et al*., 2006). Most commonly determined enzyme system include in the alkaline phosphatase (ALP), acid phosphatass (ACP), glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT).

Since the starter culture for the production of ogiri has been isolated and well studied (Falegan, 2012), It is necessary to investigate the possibility of using all cultivars as substitutes in the production of ogiri and the nutritional potentials of fermenting the cultivars of melon. This study will show the extent at which deffated ogiri samples will support the growth of rats. The body and organ weight is compared with rats fed with other feeds; also enzymatic activities of the visceral organs will be quantitatively evaluated.

2.0 MATERIALS AND METHODS

2.1 Experimental animals

Forty (40) weaning albino Wister rats, which were approximately twenty five days old were obtained. The rats (*Rattus norvegecus*) were all males purchased from preclinical Animal House, University of Ilorin. They were randomly distributed into eight treatment groups, and kept in metabolic cages, five animals per cage. All animals were starved for 24 hours before being fed with the composed diets (Table 1). The amount of feed consumed was determined on daily basis. The animals were fed for 28 days.

2.1.1 Fermented "Ogiri" samples

They were produced from attested strain of *Bacillus subtilis* (CRBS23) using different cultivars of melon seeds. Methods of Odunfa were modified and used (Odunfa, 1981).

2.1.2 Defatting of samples

The pulped samples were suspended in petroleum ether (95%) in screw-cap reagent bottles. The suspension was shaken thoroughly and allowed to settle down overnight. And the clear supernatant was later decanted. The residue was re-suspended in the same fresh solvent in a separating funnel. This process was repeated twice to effect complex extraction. The residue was recovered and excess extract solution was removed by squeezing in muslin cloth. The defatted samples (ogiri and unfermented melon seed) were spread on muslin cloth and sun-dried. Total defatting was confirmed by pressing one gram of the defatted sample on a white sheet of paper. Soiling of the paper with oil indicated incomplete defatting of sample.

2.1.3 Composition of diets

The composition of test diets is shown in Table 1.The diets were composed to contain the same quantity (20%) of protein supplement. The diets were prepared using the defatted melon seeds and ogiri as the protein sources (Group I-VI). Two control feeding diet groups were prepared. The first control diet (Group VII) was prepared using a soybean meal as the protein source, while second control diet (Group VIII) had no protein source (i.e corn starch was used).Vitamins and mineral mixture was done in ratio 4:5.

Table 1: Composition of diets for experimental rats

COMPONENTS (%)	DIET GROUPS							
	I	II	III	IV	V	VI	VII	VIII
Corn Starch	18.50	25.29	17.50	12.30	0.03	18.50	53.55	81.00
Sucrose	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Oil (corn oil)	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Vitamins/Minerals	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Cellulose (Rice bran)	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
UCM	62.50							
FCM		55.71						
UCL			63.49					
FCL				68.97				
UCV					80.97			
FCV						62.50		
SM							27.55	
PFF (corn starch)								-

Keys:

FCM = fermented *Cucumeropsis manii,*
FCL= fermented *Citrullus lanatus*,
FCV= fermented *Citrullus vulgari,*
SM= soy meal
UCM= unfermented *Cucumeropsis manii,*
UCL= unfermented *Citrullus lanatus,*
UCV= unfermented *Citrullus vulgaris,*
PFF= protein free feed

2.2 Determination of Nutritional Parameters

The following parameters were determined to assess the nutritional quality of the defatted samples as sources of protein.

(I) **Weight gained**: The weight of rats was determined on 48h basis. The mean weight gained and standard deviations of each group were also determined.

(II) **Organ: body weight ratio**: The ratio of the weight of organ to the body weight was determined thus:

$$\text{Ratio of organ to body} = \frac{\text{Weight of organ}}{\text{Weight of the whole rat}}$$

(III) **Protein Efficiency Ratio (PER):** This parameter is based solely on weight gained and does not consider either carcass composition or the need for tissue maintenance

$$\text{PER} = \frac{\text{Weight gained (g)}}{\text{Weight of protein consumed (g)}}$$

(IV) **Assay of enzyme:** Extracts were prepared for enzymes assay. After the organs (heart, kidney and liver) were removed, they were homogenized separately with mortar and pestle in 2.0% (w/v) sucrose solution in ratio 1:4.The serum was obtained from the blood after spinning and kept in 0.25mI sucrose buffer solution. The serum was collected into an anti-coagulant bottle. The extracts were kept immediately in the refrigerator 4^{o}C. The extracts were later used in the determination of enzyme activities in the organs.

2.3 Alkaline (ALP) and acid (ACP) phosphatases

The method of Bergemayer and Brent (1974) was used. The reagents used for the ALP test were prepared thus: Alkaline buffer, 6.36g of Na_2CO_3 and 3.35g of $NaHCO_3$ dissolved in 10ml of distilled water other items include substrate, disodium phenyl phosphate (1.05g/ml); 0.5N NaOH**;** sodium bicarbonate, $NaHCO_3$ (4.2w/v) potassium ferricyanide (2.4g in 100ml distilled H_2O). Reagents used for determination of ACP were similar to ALP but differed only in the buffer. The buffer used for determination of ACP was prepared by adding 21.0g of citric acid to 188ml of NaOH. Distilled water was added to make up to 500ml and the adjusted to 4.8 by adding 1N NaOH.

Four test tubes were arranged and labeled as standard, standard blank, test and test blank respectively. A 0.5ml of the acid buffer solution (citric acid) was added to four test tubes. A 0.5ml of distilled water was added to the standard blank only; 0.5ml of 0.5N NaOH was added to the test tubes with exception of the test. The solution were mixed together and incubated at 37^{o}C for 3 minutes. After incubation, 50µl of the organ extract was pipetted into the test and test blank tubes, mixed and incubated at 37^{o}C for 3 minutes. After incubation,50µl of the organ extract was pipetted into the test and test blank tubes, mixed and incubated at 37^{o}Cfor 15 minutes (for ALP determination) and an hour (for ACP determination). After incubation, 0.5µl of 0.5N NaOH was

pipetted into the test tube alone. Lastly, 0.5ml of aminophenazone (4.2% $NaHCO_3$ and 2.4% potassium ferricyanide) was added to the four test tubes and the optical density (OD) was read at 510nm.

Glutamic oxaloacetic transaminase (GOT) and Glutamic pyruvate Transaminase (GPT): Randox enzyme kits were used for the determination of both GOT and GPT. Phosphate buffer (11.876g/l of Na_2HPO_4) was used for the determination of both GPT and GOT. The substrate for GOT differed From GPT, GOT substrate contained L-aspartic acid (200mmol/l), α-ketoglutarate (30mmol/l) and NaOH (0.4mmol/l). GPT substrate contained L-alanine (200mmol/l), α-oxoglutarate (2.0mmol/l) and NaOH (0.4 mmol/l).

The test tubes were set up, one labeled blank and the other as the test. A 0.5ml of the substrate was pipetted into the two test tubes and 0.5ml of the sample was pipetted only to the test tube labeled "test". The solution were mixed and incubated at 37^0C for 30 minutes. After incubation, 0.5ml of 2.4 dinitrophenyl hydrazine (DNPH) was added to the test tubes and the mixture incubated at room temperature (28 ± 2)^{0}C for 20 minutes. A 0.5ml of the sample was later added to the blank only and 5ml of NaOH solution was pipetted into the test. The solutions were mixed and the OD reading was taken at 546nm. All data obtained were analyzed using Anova multiple range test of SPSS 15.0.1.

3.0 RESULT AND DISCUSSION

Rat fed with diet composed with fermented *C. manni* (FCM) recorded the highest feed intake at the third week of feeding (Table 2). In 50% of the diet groups the feed intake reduced after the second week of feeding; but later increased and reached the peak level at the fourth week. Rats in the positive and negative diet groups i.e. SM and PFF respectively showed different patterns; the feed consumption rate was highest by the second week but reduced subsequently as the number of weeks increased in diet group PFF. In the case of SM reduction of intake started from the first week of consumption to the fourth week. Table 3 shows the weight gained by rats in the different diet groups. The highest weight gained was recorded in unfermented *Cucumeropsis manni* (UCM), which was significantly higher (at α=0.05) than others.

The least weight gain was recorded in unfermented *Citrullus lanatus* (UCI), which was not significantly different from the fermented and unfermented *Citrullus vulgaris* (FCV and UCV) respectively. The rats fed with PFF lost weight, and were not looking healthy.

Table 4 shows that rats fed with diet group of unfermented *Cucumeropsis manni* (UCM) had the highest protein efficiency ratio (PER) which was significantly higher than others (at α=0.05).There were no significant differences in the PER of diet groups UCL,FCV,UCV and SM. A negative PER was recorded for PFF because of the reduction in the weights of the rats.

Table 2: Feed intake (g) per week by experimental rats in diets groups

	TIME (Week)			
DIET GROUPS	**1**	**2**	**3**	**4**
FCM	27.54	32.85	34.11	26.64
FCL	27.25	26.05	20.78	25.90
UCV	24.39	22.36	24.26	26.97
UCL	22.88	24.37	23.07	28.33
FCV	23.88	20.40	22.11	25.22
UCM	25.86	28.70	24.96	29.92
SM	34.56	33.61	32.87	31.61
PFF	24.84	28.45	22.56	19.09

Keys:

FCM= fermented *Cucumeropsis manii* UCM= unfermented *Cucumeropsis manii*
FCL= fermented *Citrullus lanatus* UCL= unfermented *Citrullus lanatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

Table 3: Weight gained (g) by experimental rats fed on diets composed with different protein sources

DIET GROUPS	WEIGTH GAIN (g)
FCM	6.0275
UCM	7.7975
FCL	5.205
UCL	2.1875
FCV	2.8
UCV	2.595
SM	4.67
PFF	- 0.91

Keys:
FCM= fermented *Cucumeropsis manii* UCM= unfermented *Cucumeropsis manii*
FCL= fermented *Citrullus lanatus* UCL= unfermented *Citrullus lanatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

Table 4: Protein efficiency ratio (per) of rats fed on diets composed with different sources of protein

DIET GROUPS	PROTEIN EFFICIENCY RATIO
FCM	0.85
UCM	1.5625
FCL	1.125
UCL	0.485
FCV	0.48
UCV	0.55
SM	0.46
PFF	- 0.3

Keys:
FCM= fermented *Cucumeropsis manii* UCM= unfermented *Cucumeropsis manii*
FCL= fermented *Citrullus lanatus* UCL= unfermented *Citrullus lanatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

The highest heart-body weight ratio (bwr) of 0.71% was recorded in diet group UCL and the least (0.54%) from diet group UCM (Table 5); the difference was not significant. The highest liver-body weight ratio (6.31%) was from diet group UCV: while the lowest (4.42%) was from diet group PFF. The highest kidney-body weight ratio was 1.30% in diet group SM, and the least (1.07%) from diet group UCM. Generally, the weight of organs of the rats from fermented diet groups were relatively higher than those that are of unfermented diets except in a few cases.

Table 6 shows the level of acid phosphatase ACP in the organs of the experimental animals. The ACP was highest in the heart of rats fed with diet UCL; however, there were no significant differences in the amount of ACP among the diet groups except PFF (at α=0.05). Diet group PFF showed the lowest ACP activities in the heart (114.38 µmol ml^{-1}). The ACP in the liver of rats was highest in diet group FCL and lowest in the rats fed with diet FCV (188.14 µmol ml^{1}) .The ACP was highest in the kidney of rats fed with diet group FCL (230.2µmol/ml) which was Significantly higher (at α=0.05) than all other diet groups. The lowest level of ACP 182.9µmol ml^{-1} in the kidney was recorded in diet group SM which served as positive control (Table 6).

The highest ALP activities was in the heart of rats fed with diet FCV (230.54 µmol ml^{-1}); and was significantly (at α =0.05) higher than all other diet groups (Table 7). The lowest ALP (120.21 µmol ml^{-1}) in heart was from the rats fed with diet FCM. In the liver, the highest ALP (823.33µmol/ml) was recorded from rats fed with diet UCL. The ALP level in the liver varied significantly (at α=0.05) among the diet groups. The enzyme activity was very high in the kidney, with the highest value of 2678.30 µmol ml^{-1} from rat in diet group FCM. The lowest ALP was recorded in rats fed with diet PFF.

Glutamyl oxaloacetate transaminase (GOT) had the highest value (230.504µmol ml^{-1}) in the hearts of rats fed with diet FCV (Table 8); while the lowest values were recorded in the hearts of rats fed with diet UCL. The highest GOT recorded (700.00 µmol ml^{-1}) in the blood serum was from the rats fed with diet PFF.

Table 5: weight (g) of organs and organ: body weight ratio of experimental Rats in diet groups composed with different protein sources

	WEIGHT OF ORGAN (g)			ORGAN-BODY WEIGHT RATIO		
DIET GROUPS	Heart	Liver	Kidney	Heart	Liver	Kidney
FCM	0.36^{a}	3.78^{a}	0.79^{a}	0.55	5.75	1.20
UCM	0.39^{a}	4.10^{a}	0.77^{a}	0.54	5.57	1.07
FCL	0.39^{a}	3.61^{a}	0.76^{a}	0.64	6.2	1.25
UCL	0.37^{a}	3.28^{a}	0.57^{bc}	0.71	6.22	1.11
FCV	0.37^{a}	3.07^{a}	0.64^{b}	0.70	5.7	1.21
UCV	0.32^{ab}	3.70^{a}	0.62^{b}	0.62	6.31	1.20
SM	0.34^{ab}	3.34^{a}	0.78^{a}	0.56	5.57	1.30
PFF	0.28^{b}	1.77^{b}	0.49^{c}	0.66	4.42	1.22

Keys:

FCM= fermented *Cucumeropsis manii* UCM= unfermented *Cucumeropsis manii*
FCL= fermented *Citrullus lanatus* UCL= unfermented *Citrullusl anatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

Table 6: Acid phosphatase (ACP) µmol/ml in the organs of rats in diets groups

	ORGANS			
DIET GROUPS	**Heart**	**Liver**	**Kidney**	**Blood Serum**
FCM	173.10^{a}	459.70^{a}	200.14^{bc}	250.37^{d}
UCM	180.27^{a}	207.14^{ed}	204.85^{b}	204.60^{de}
FCL	175.20^{a}	462.90^{a}	230.24^{a}	450.70^{a}
UCL	181.03^{a}	209.03^{b}	209.03^{b}	234.60^{de}
FCV	178.25^{a}	188.14^{e}	188.59^{cd}	200.10^{f}
UCV	177.60^{a}	366.55^{b}	207.50^{b}	340.40^{c}
SM	175.62^{a}	226.71^{d}	182.90^{d}	220.60^{ef}
PFF	114.38^{b}	218.37^{d}	183.04^{d}	400.20^{b}

Keys:

FCM= fermented *Cucumeropsis manii* UCM= unfermented *Cucumeropsis manii*
FCL= fermented *Citrullus lanatus* UCL= unfermented *Citrullus lanatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

Table 7: Alkaline phosphatase (ALP) µmol/ml in the organs of rats in diet groups

	ORGANS			
DIET GROUPS	**Heart**	**Liver**	**Kidney**	**Blood Serum**
FCM	120.21^{f}	69.20^{fg}	2678.30^{a}	486.40^{b}
UCM	146.63^{c}	160.00^{e}	2207.64^{b}	462.00^{cb}
FCL	137.37^{cde}	363.65^{c}	2620.37^{a}	435.00^{cd}
UCL	105.65^{b}	823.33^{a}	1753.24^{c}	412.10^{d}
FCV	230.54^{a}	325.70^{d}	1339.55^{e}	290.00^{f}
UCV	140.10^{cd}	488.08^{b}	1507.10^{d}	300.00^{f}
SM	124.95^{ef}	93.73^{f}	1211.74^{f}	350.00^{e}
PFF	129.39^{def}	53.62^{g}	968.75^{f}	700.00^{a}

Keys:

FCM=fermented *cucumeropsis manii* UCM=unfermented *cucmeropsis manii*
FCL=fermented *citrullus lanatus* UCL=unfermented *citrullusl anatus*
FCV=fermented *citrullus vulgaris* UCV=unfermented *citrullus vulgaris*
SM=soy meal PFF=protein free feed

Table 8: Glutamyl oxaloacetate transaminase (GOT) µmol/ml in the organs of rats in diet groups

DIET GROUPS	ORGANS			
	Heart	Liver	Kidney	Blood serum
FCM	121.80^{a}	193.54^{a}	80.60^{a}	140.50^{cd}
UCM	100.80^{ab}	87.00^{f}	13.00^{d}	100.40^{f}
FCL	122.20^{a}	163.20^{c}	70.53^{ab}	110.20^{ef}
UCL	76.20^{bc}	191.20^{a}	26.80^{c}	120.30^{def}
FCV	128.80^{a}	128.00^{d}	7.30^{d}	175.10^{b}
UCV	110.40^{a}	175.97^{b}	67.40^{b}	150.00^{c}
SM	103.33^{ab}	122.00^{d}	15.66^{cd}	130.00^{cde}
PFF	68.03^{c}	107.70^{e}	8.00^{d}	250.00^{a}

Keys:

FCM= fermented *Cucumeropsismanii* UCM= unfermented *Cucumeropsismanii*
FCL= fermented *Citrulluslanatus* UCL= unfermented *Citrulluslanatus*
FCV=fermented *Citrullus vulgaris* UCV= unfermented *Citrullus vulgaris*
SM= soy meal PFF=protein free feed

The GOT level in kidney was generally lower than that of the other organs and blood serum. The highest GOT value in the kidney was 80.60µmol ml^{-1}in rat fed with diet group FCM, and this was significantly higher than the other diet groups (at α = 0.05) The diet group PFF and FCV had the least GOT value in kidney, 8.00 and 7.30 µmol/ml respectively.

4.0 DISCUSSION

The entire rat fed with both fermented and unfermented melon seed gained weight; while the rats fed with protein-free diet (PFF) lost weight. This indicates that all the three melon types, both fermented and unfermented may serve as food supplement for animals. The fermentation of melon seed into 'ogiri' did not have significant effect on the nutritional quality of the protein and the level of ACP in the liver was significantly higher (at α =0.05) than in the kidney and heart. The GOT was highest in the liver, but low in kidney and heart; this result is similar to the result of Ajayi *et al*. (2010) the concentration of GOT, ALP and ACP were relatively lower in the heart than in the kidney and liver; a similar result was also observed by Moss (1999). The activities of the enzymes were generally low in organs of rats fed with diet PFF. This might be due to leakage of enzymes from damaged organs to the blood system. The level of the enzymes in the organs is used as indices of biochemical changes. The feeds were good since there was no wasting or catabolism of muscle tissues. And those animals were not surviving at the expense of body reserve. This was a good indication that dietary protein was well utilized by rats (Ahamefule *et al.,* 2006).

Values obtained for kidney and liver weights in this study showed no significant difference (at α= 0.05) among treatment groups. It is a common practice in feeding trials to use weights of some internal organs like liver and kidney as indicators of toxicity. It is a fact that if there is any toxic element in the feed, abnormalities in weight of liver and kidney would be observed. The abnormalities will arise because of increased metabolic rate of the organs in attempt to reduce this toxic elements or anti-nutritional factors to non-toxic metabolities (Ahamefule *et al.,* 2006).

The proximate analysis of commercial 'ogiri' from different locations revealed different chemical compositions. The strains of *Bacillus subtilis* group isolated from other food condiments were not as effective as those isolated from commercial 'ogiri' samples when used in starter culture fermentations the quality of the 'ogiri' produced by the former was poorer and lower when compared to that produced by the latter (Falegan, 2012: Aderibigbe and Adebayo, 2002). 'Ogiri' compared favorably with soybeans meal as a source of protein in the diet of experimental animals. The protein concentrates in 'ogiri' prepared from fermented *Cucumeropsis manii, Citrullus lanatus* and unfermented *C. manni* were significantly better than soybeans meal with regards to protein efficiency ratio (PER) and weight gain in the rats.

With the use of starter cultures and controlled fermentation processes, the quality of the condiment can be standardized (Steinkraus, 2005). Selection of appropriate starter cultures for the production of 'ogiri' in this study provided very vital database for the optimization of traditional fermented condiment in Nigeria., development of starter cultures that can produce 'ogiri' having desirable sensory qualities, (taste, flavor, colour, texture). Absence of pathogenic and spoilage microorganisms, improved shelf life, highly nutritional, and devoid of anti-nutritional factors is essential in fermentation. There is need for technical improvement on the small scale, low-tech food fermentation (Achi, 2005b). The proper identification of the associated fermentation bacterial strains constituted the most essential requirement for the industrial take off of condiment production (Sanni *et al*., 2002).

5.0 CONCLUSION

The output of this research work showed that defatted 'ogiri' may serve as a good food supplements in animal feeds. Both the defatted fermented and unfermented melon performed well as protein sources in the composed diets, irrespective of the species of melon seed used. Thus, fermentation process did not have a significant effect on the nutritional quality of the protein in melon seed during production of 'ogiri'. Infused molecular typing techniques having high differentiation powers may however be a more realistic method for the identification of the microorganisms associated with condiment production. These techniques are reproducible and provide adequate information about the diversity of the microflora to aid maximization of their bio-technology properties.

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