



QUANTITATIVE ASSESSMENT OF FATTY ACID PROFILE AND NUTRITIONAL STATUS OF GENETICALLY MALE TILAPIA (*Oreochromis niloticus* Linnaeus, 1758)

M. U. EFFIONG* and A. W. AKPAN

Department of Zoology, University of Uyo, P.M.B. 1017, Uyo, Akwa Ibom State, Nigeria

*Correspondence Mobile: +234(0)803-625-5488; E-mail: sharonfisheries@yahoo.com

ABSTRACT

The study identified fatty acid profile and nutrient contents of Genetically Male Tilapia with a view to provide nutritional data for dietary planning. Fatty acid methyl esters were determined by gas chromatograph and were identified using retention time in comparison with an external standard (SupelcoTM 37 component FAME Mix). Nutrient contents were determined by atomic absorption spectrophotometer. The major saturated fatty acids (28.20%) were palmitic and myristic acids. Oleic acid was the prominent monounsaturated (16.24%) while dominant polyunsaturated (50.77%) was of ω -6 series and was found chiefly in linoleic fatty acid. The prominent essential fatty acids were C18:3n-3 (27.43%) and C18:2n-6 (23.33%) with a mean free fatty acid component of 0.92%. The mean value of ω -3/ ω -6 ratio of 1:1.13 was within the recommended daily intake of EPA and DHA needed for normal human health. Mean values for nutritional attributes were: moisture 71.13%, protein 16.90%, lipid 6.17%, carbohydrate 0.38% and calorie 6.50kJ/g. High mineral contents were observed. The significance of this study was its revelation that GMT is a good source of essential fatty acids and the fish belongs to high-protein, low-oil category.

Keywords: YY Super-male Tilapia, protein, lipid, essential fatty acids

INTRODUCTION

Aquaculture is an integral component of the overall agricultural production system in Nigeria. The country with hundreds of rivers and ponds is notable for being a fish-loving nation where fish plays an important role in the diets, constituting the main and often irreplaceable animal protein source in both urban and rural households (Otubusin, 2011). The major fish species cultured in Nigeria include catfishes and tilapia. Tilapia is one of the most widely cultured fish in the world. Currently, farmed tilapia represents more than 75% of world tilapia production (FAO, 2013), and this contribution has been exponentially growing in recent years. Several factors have contributed to the rapid global growth of tilapia. Among these are: genetic improvement, ease of culture, highly adaptable to a wide range of environmental conditions (Ponzoni *et al.*, 2008). However, a major problem in tilapia culture is that females grow slower than males. Early sexual maturation diverts energy from growth to reproduction and unwanted breeding results in overcrowding and competition. The most effective solution to this problem is to produce and grow only male fish. Researches have addressed this problem in an innovative way through the application of basic genetics, to develop a unique product in Genetically Male Tilapia (GMT) (Mair *et al.*, 1997). The GMT so developed has proved to be excellent production fish in both extensive and intensive systems using ponds, raceways, cages and tanks (Eknath *et al.*, 2007). They are now in use in more than 20 countries around the world (Gupta and Acosta, 2004) including Nigeria.

Consumers would usually desire to know if there are nutritional differences in various fish species from different sources. This can only be answered through proximate analysis of body composition of various fish species. Karapanogiotidis (2002) reported that some fishes are high in omega-3 (n-3) and omega-6

(n-6) PUFA depending on some external (environment, culture method and climatic effects) and internal (genetic make-up, feeding regime, life cycle stage) factors (Buchtova *et al.*, 2004). The essential fatty acids: eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are of interest in human diets because they reduce the risk of human cardiovascular diseases (Leaf and Kang, 1996). A daily intake of 500mg EPA and DHA has been recommended for the primary prevention of coronary heart disease (ISSFAL, 2004). In addition, fatty acid composition data are needed by food scientists and nutritionists to aid them in dietary formulation, processing and product development. The body can make some of the fats it needs from the foods consumed by human but two essential fatty acids – linoleic and linolenic acids (which are used to build n-3 and n-6 fatty acids) cannot be made in the body. In order to reduce the risk of cardiovascular diseases, emphasis has been placed on the increased consumption of fish and fish products, which are rich in PUFA of the n-6 series (Sargent, 1996). Studies have been carried out on proximate chemical composition and fatty acids profiles of different fish species (Uauy and Valenzuela, 2000). There is, however, dearth of accurate basic chemical composition data for the newly introduced GMT in Nigeria. This however constitutes a barrier to the development on the use of this tilapia species. In view of this, the present study was carried out with the aim of investigating the variation in body composition of GMT, *Oreochromis niloticus* in terms of the fatty acid profile and nutritional status.

MATERIALS AND METHODS

Collection of Fish Sample: The GMT fingerlings were procured from Durante Fisheries Industries, Ibadan, Oyo State, Nigeria. Fish were transported in oxygen bags to Department of Zoology, University of Uyo, where the experiment was conducted. The fish were kept and nurtured for 12 weeks using commercial fish feed before analysis.

Determination of Nutritional Status of GMT

Estimation of Minerals by Atomic Absorption Spectrophotometer: The estimation of the trace elements (Calcium, Phosphorus, Iron and Zinc) of GMT was performed with flame atomic absorption spectrophotometer as described by (Stansby, 1963).

Proximate Analysis of Fish Carcass: Proximate analysis of fish carcass was done according to standard AOAC method (AOAC, 2004). Moisture content was done by oven-drying to a constant weight; Total ash by muffle furnace combustion; Crude fibre by trichloroacetic acid method; Lipid content by soxhlet extraction method; Protein by micro – kjeldahl method, Carbohydrate was calculated as difference obtained after subtracting moisture, total organic nitrogen (protein), ether extract, ash and fibre from 100%. Caloric value was estimated based on physiological fuel values (0.2364 KJ/g for protein; 0.3954 KJ/g for lipid and 0.1715 KJ/g for carbohydrate) as described by (Henken *et al.*, 1986).

Lipid Extraction and Fatty Acid Analysis: The extraction of total lipids and preparation of fatty acid methyl esters were performed according to Musa (2009). Fatty acid analysis was carried out on a gas chromatograph (Hewlett Parkard: 6890) fitted with an automatic sampler (Model: AS 2000B) and flame ionization detector. The conditions used were: Omega wax fused silica capillary column BPX-70 (60 m x 0.32 mm i.d., 0.25 µm film thickness) (SGE; Melbourn, Australia). The starting temperature was 108°C; this was later raised to 115°C at a rate of 8°C/min and held for 10 minutes. This temperature was finally raised to 240°C at a rate of 8°C/min and held for another 10 minutes. The sample size was 1µl and flashed through helium as the carrier gas at a rate of 1.6ml/min with inlet pressure of 12 psi. Fatty acids methyl esters were identified in comparison to an external standard (Supelco™ 37 component FAME Mix).

Statistical Analysis: Statistical analysis was done using computer software SPSS version 20.0 (2012).

RESULTS AND DISCUSSION

Nutritional Status of GMT: The results of nutritional analysis of the fillet of GMT are presented in Table 1. The data revealed that moisture contents of GMT ranged from 69.74 to 71.27% with a mean value of 71.13%. The mean lipid content of 6.17% was significantly ($P < 0.05$) lower than 14.4%

reported by (Okonji and Daniel, 2013) for Nile tilapia. The percentage dry matter contents of fish were 56.99% for protein and 8.05% for lipid while mean carbohydrate and caloric values were 0.38% and 6.50KJ/g respectively. This may mean that GMT belongs to a high-protein-low-lipid fish category. Proteins are essential to all life. In animals, they help form supporting and protective structures such as cartilage, skin, nails, hair and muscles. They are major constituents of enzymes, antibodies, many hormones, and body fluids such as blood, milk and egg white (Potter and Hotchkiss, 1996). Calcium is the most abundant mineral in the human body and it is essential for growth, bone formation, blood coagulation, milk formation and vitamin D absorption. The deficiency of calcium leads to rickets, osteomalacia and osteoporosis (Islam *et al.*, 2013). The mean amount of calcium observed in GMT was 199.76mg/kg. Phosphorous is a major constituent of all animal cells. Though primary dietary deficiency of phosphorous is not known to occur in man, insufficient amount in food may lead to secondary phosphate depletion resulting into muscle weakness, and other diseases, notably those affecting the kidneys and bones (Rosenquist, 1996). In this study, GMT contained almost the same phosphorus level as that reported in *Clarias batrachus* by (Islam *et al.*, 2013). Iron is an essential life supporting macronutrient in animals and human. Iron plays an important role in cellular metabolism as an active component of various enzymes, especially those associate with the respiration chain of mitochondria. Iron deficiency – anemia is widely prevalent especially in children (Gehring *et al.*, 2011). In this study, mean iron content of GMT was 6.83mg/kg indicating that this fish is a good source of iron. Trace amount of zinc was detected in GMT.

Table 1: Nutritional Analysis of the Fillet (% wet weight) of Genetically Male Tilapia

Nutritional					
Index	Range	*Mean ± SE	Nutritional Index	Range	*Mean ± SE
Moisture	69.74-71.27	71.13±0.76	Calorie (kJ/g)	6.35 – 6.64	6.50±0.08
Protein	16.2.75-17.42	16.90±0.34	Protein (DM)	56.73 – 57.24	56.99±0.15
Lipid	6.05-6.28	6.17±0.07	Lipid (DM)	7.83 – 8.32	8.05±0.04
Fibre	0.03-0.72	0.32±0.21	Calcium (mg/kg)	192.3 – 208.5	199.76±2.5
Ash	4.21-4.82	4.44±0.19	Phosphorus (mg/kg)	68.3 – 70.00	69.27±1.2
Carbohydrate	0.16-0.77	0.38±0.19	Iron (mg/kg)	6.53 – 7.12	6.83±2.1

*Values are given as mean of triplicate experiments.

Fatty Acid Compositions of Lipid of GMT: The sequence of fatty acids was ordered according to their chromatographic retention times and values were given as mole percentages of the total fatty acid methyl esters. A total of eleven fatty acids were identified in lipid of GMT (Table 2). Among the fatty acids found, the prominent was C18 group (specifically; stearic, oleic, linoleic and linolenic acids) comprising 74.83% of total fatty acids. Palmitic and Myristic acids were the main saturated fatty acids with mean concentrations of 11.73% and 6.06% respectively. The availability of high levels of palmitic acid has been described as characteristics of fresh water fish Ackman (1980). This high proportion of palmitic acid among saturated fatty acids was also reported in Nile tilapia and African catfish fillets by (Aggelousis and Lazos, 1991). The principal acids in the PUFA group were linolenic and linoleic acids. This result agreed with those obtained in other studies conducted on freshwater fish species (Pourshamsian *et al.*, 2012). A higher ($P < 0.05$) proportion of linoleic acid (27.33%) was observed in GMT in this study than between 11.1 and 19.64% reported in Nile tilapia (Yones *et al.*, 2013). Das (2006) reported that direct intake of various PUFA alters the cell membrane fatty acid composition which in turn, modulates cell/tissue response to infection, injury and inflammatory actions. The branched chain fatty acids identified were C15:0, C16:0, C17:0, C18:2 and C20:0. These fatty acids were the highest fatty acids noticed. This high level of branched chain fatty acids in this species has important advantages. Uguala *et al.* (2008) reported that branched chain fatty acids influence lower melting point, lower cholesterol levels, provide energy, and form an integral part of biomembranes. The authors further opined that esterification of branched

chain fatty acids to cholesterol causes the fatty acids to stimulate protein synthesis and influence some ribosomal functions which are necessary for peptide elongation.

Fatty acid profile analysis also provides information about the essential fatty acid requirements of fish which would aid the compounding of adequate protein-to-fat ratio feed that would balance energy requirements with caloric intake. The principal essential fatty acids: EPA and DHA concentrations were similar to other studies on tropical (Clement and Lovell, 1994) and temperate (Ahlgen *et al.*, 1994) freshwater fishes. This indicated the dominance of these fatty acids in the tissue lipids of GMT. Sargent (1996) noted that DHA has a role in maintaining the structure and functional integrity of cell membranes; thus desirable for human nutrition and health. The free fatty acids above 1.5% had been reported as indication of unsuitability of lipids for edible purposes (Molla *et al.*, 2007). The mean free fatty acid of the lipid derived from GMT was found to be 0.92%, indicating its suitability for human consumption. The values of ω -3 to ω -6 ratio obtained in muscle fatty acids of GMT was within the recommended daily intake (1:1 to 1:1.5) of EPA and DHA for normal human health (Osman *et al.*, 2000). The study has revealed that GMT is a good source of high-protein, low-lipid with balanced EPA and DHA levels and thus a healthy source of animal protein for human nutrition

Table 2: Fatty Acid Compositions of Genetically Male Tilapia Derived from Methyl Ester Mixture by Gas Chromatograph Analysis.

Name of Fatty Acid	Retention Time	% of Total Fatty Acid	Name of Fatty Acid	Retention Time	% of Total Fatty Acid
Capric acid	7.46	0.16±0.03	γ -Linolenic Acid	18.48	0.54±0.15
Lauric acid	11.02	1.49±0.70	Arachidic acid	19.59	0.22±0.02
Myristic acid	13.77	5.61±0.30	Gadoleic acid	20.06	0.59±0.01
Myristoleic acid	13.95	0.45±0.05	Eicosadienoic acid	20.09	0.03±0.00
Pentadecanoic	15.91	0.25±0.10	Eicosatrienoic acid	20.18	0.06±0.01
Palmitic acid	16.12	5.39±0.04	Eicosatrienoic acid	20.26	0.51±0.10
Palmitoleic acid	16.64	6.32±0.32	Arachidonic Acid	20.34	0.30±0.00
Heptadecanoic	17.71	0.98±0.14	Arachidonic Acid	20.46	0.60±0.00
Heptadecenoic	17.84	0.26±0.04	Eicosapentaenoic acid	20.52	0.17±0.09
Stearic acid	17.91	7.13±0.12	Behenic acid	21.81	0.04±0.00
Oleic acid	17.97	16.24±0.15	Docosahexaenoic acid	22.07	0.78±0.03
Linoleic Acid	18.01	27.33±0.01	Tricosanoic acid	22.87	0.54±0.06
α -Linolenic Acid	18.37	23.43±0.25	Lignoceric acid	24.82	0.37±0.05

*Values are given as mean of triplicate experiments.

CONCLUSION

The results obtained from this study revealed that lipids of Genetically Male Tilapia contained high amount of EPA and DHA, balanced ω -3 to ω -6 ratio, as well as high amount of macro and micro nutrients. Therefore, GMT is suitable for human consumption. The intensive propagation of this fish is hereby encouraged to boost tilapia production in Nigeria.

REFERENCES

- Ackman, R. G. (1980). Fish Lipids. *In: Connell, J. J. (Ed.). Advances in Fish Sciences and Technology*, pp. 86-103.
- Aggelousis, G. and Lazos, S. E. (1991). Fatty Acid Composition of the Lipids from Eight Freshwater Fish Species from Greece. *J. Food Compos. Anal.*, 4: 68-76.
- Ahlgren, G., Blomquist, P., Boberg, M. and Gustafsson, I. B. (1994). Fatty Acid Content of the Dorsal Muscle – An Indicator of Fat Quality in Fresh Water Fish. *Journal of Fish Biology*, 45: 131-157

- AOAC (Association of Official Analytical Chemists) (2004). Official Methods of Analysis. 18th ed., Gaithersburg, MD, p. 240.
- Buchtova, H., Smutna, M., Vorlova, L., Svobodova, Z. and Flajsans, M. (2004). Fatty Acid Composition of Diploid and Triploid populations of Trench (*Tinca tinca* L.). *Acta. Vet. Brno.*, 73: 235-245.
- Clement, S. and Lovell, R. T. 1994. Comparison of Processing Yield and Nutrient Composition of Nile tilapia (*Oreochromis niloticus*) and catfish (*Ictalurus punctatus*). *Aquaculture* 119: 299-310.
- Das, U. N. (2006). Essential Fatty Acids: Biochemistry, Physiology and Pathology. *Biotechnol. Journal*, 1: 420-439.
- Eknath, A. E., Bentsen, H. B., Ponzoni, R. W., Rye, M., Nguyen, N. H. Thodesen, J. and Gjerde, B. (2007). Genetic Improvement of Farmed Tilapias: Composition and Genetic Parameters of a Synthetic Base Population of *Oreochromis niloticus* for Selective Breeding. *Aquaculture*, 273: 1-14.
- FAO (2013). On-Farm Feeding and Feed Management in Aquaculture. *FAO Fisheries and Aquaculture Technical Paper*, 583, p. 68.
- Gehring, C. K., Gigliotti, J. C., Moritz, J. S., Tou, J. C. and Jaczynski, J. (2011). Functional and Nutritional Characteristics of Proteins and Lipids Recovered by Isoelectric Processing of Fish By-Products and Low Value Fish. *J. Food Chem.* 124 (2): 422-431.
- Gupta, M. V. and Acosta, B. O. (2004). From Drawing Board to Dining Table: The Success Story of the GIFT Project. *NAGA World Fish Center Quarterly*, 27 (3 & 4): 4-14.
- Henken, A. M., Machiels, M. A. M., Dekker, W. and Hogendoorn, H. (1986). The Effect of Dietary Protein and Energy Content on Growth Rate and Feed Utilization of the African Catfish, *Clarias gariepinus* (Burchell, 1822). *Aquaculture*, 58 (1-2): 55-74.
- Islam, R., Mondol, L. K., Sheikh, L., Rahman, S. S., Islam, M. and Rahman, A. (2013). Identification of Fatty Acid Profile, Lipid Characterization and Nutritional Status of *Clarias batrachus*. *Nutr Sci Food Technol.*, 1: 1-6.
- ISSFAL (International Society for the Study of Fatty Acid and Lipids) (2004). Recommendations for Intake of Polyunsaturated Fatty Acids in Healthy Adults. <http://www.issfal.org.uk/>
- Karapanogiotidis, T. (2002). The Effects of Fish Oil-enriched Diets on Growth, Feed Conversion and Fatty Acid Content of Red Hybrid Tilapia, *Oreochromis* sp. *Bulletin of the Japanese Society of Scientific Fisheries*, 46: 1353-1356.
- Leaf, A. and Kang, J. X. (1996). Prevention of Cardiac Sudden Death by N-3 Fatty Acids: A Review of the Evidence. *J. Internal Med.*, 240: 5-12.
- Mair, G. C., Dahilig, L. R., Morales, E. J., Beardmore, J. A. and Skibinski, D. O. F. (1997) Application of Genetic Techniques for The Production of Monosex Male Tilapia in Aquaculture: Early Experiences from The Philippines. Proceedings of the Fourth Central America Symposium on Aquaculture, Tegucigalpa, Honduras, April 22-24, pp. 225-227.
- Molla, A. H., Camellia, S., Ahsan, M. S., Talukder, S. M. and Alam, M. T. (2007). Physicochemical and Microbiological Investigation of the Lipid from Bangladeshi Fresh Water Fish *Mystus vittatus*. *University Journal of Zoology, Rajshahi, University*, 26: 21-25.
- Musa, A. S. M. (2009). National Quality Components of Indigenous Freshwater Fish Species, *Puntius stigma* in Bangladesh. *Bangladesh J. Sci. Ind. Res.*, 44: 367-370.
- Okonji, V. A. and Daniel, E. (2013). Fatty Acids Variation in *Oreochromis niloticus* and *Clarias gariepinus* from Semi-Intensive Fish Tank. *World Journal of Fish and Marine Sciences* 5 (2): 169-175.
- Osman, H., Zurria, A. R. and Law, E. C. (2000). Fatty Acid Composition and Cholesterol Content of Selected Marine Fish in Malaysian Waters. *Chem.*, 73: 55-60.
- Otubusin, S. O. (2011). *Water, Water, Water, Everywhere-----An Enigma!* The University of Agriculture Abeokuta, Nigeria, Inaugural Lecture Series. 32, p. 106.

- Ponzoni, R. W., Nguyen, N. H., Khaw, H. L., Kamaruzzaman, N., Hamzah, A., Bakar, K. R. B. and YEE, H. Y. (2008). Genetic Improvement of Nile Tilapia (*Oreochromis niloticus*) – Present and Future. 8th International Symposium on Tilapia in Aquaculture, World Fish Center, Malaysia, p. 33
- Potter, N. N and Hotchkiss, J. H. (1996). *Food Science*, (5th ed.), CBS Publishers & Distributors, New Delhi, India. 30-32.
- Pourshamsian, K., Ghomi, M. R. and Nikoo, M. (2012). Fatty Acid and Proximate Composition of Farmed. Great Sturgeon (*Huso huso*) Affected by Thawing Methods, Frying and Chill Storage. *Adv. Stud, Biol.*, 4: 67-76.
- Rosenquist, A. and Holmer, G. (1996). Nutritional value of micro-encapsulated fish oils in rats. *Z Ernährungswiss.* 35: 178-84.
- Sargent, J. R. (1996). *Origins and Functions of Egg Lipid: Broodstock Management and Egg and Larval Quality*. In: Bromage, N. R. and Roberts, R. J. (Eds.), Oxford: Blackwell Publishers, pp. 353-372
- Stansby, M. E. (1963). Analytical Methods on Industrial Fishery Technology. New York: Rhenhold Publishing Corporation, pp. 369-370.
- Uauy, R., and Valenzuela, A. (2000). Marine Oils: The Health Benefits of N-3 Fatty Acids. *Nutrition* 6 (7/8): 680-684.
- Ugoala, C., Ndukwe, G. I. and Audu, T.O. (2008). Comparison of Fatty Acids Profile of Some Freshwater and Marine Fishes. *Internet Journal of Food Safety*, 10: 9-17.
- Yones, A. M., El-Saidy, D. M. and Abdel-Hakim, N. F. (2013). Effects of Fish Oil Substitution With Vegetable Oils in Diets of Juvenile Nile Tilapia, *Oreochromis Niloticus (L.)* On Growth Performance, Nutrients Utilization and Muscle Fatty Acids Contents. *Merit Research Journal of Food Science and Technology*, 1 (1): 009-018.