

***A  
N  
I  
M  
A  
L***

***R  
E  
S  
E  
A  
R  
C  
H***

***I  
N  
T  
E  
R  
N  
A  
T  
I  
O  
N  
A  
L***



Volume 3 Number 1 (2006)



**An International Journal Publishing Original Research Involving  
the Use of Animals and Animal Products**

**ISSN: 159-3115**

**Website: [zoo-unn.org](http://zoo-unn.org)**

## Animal Research International®

Animal Research International is an online Journal inaugurated in University of Nigeria to meet the growing need for an indigenous and authoritative organ for the dissemination of the results of scientific research into the fauna of Africa and the world at large. Concise contributions on investigations on faunistics, zoogeography, wildlife management, genetics, animal breeding, entomology, parasitology, pest control, ecology, malacology, phytonematology, physiology, histopathology, biochemistry, bioinformatics, microbiology, pharmacy, veterinary medicine, aquaculture, hydrobiology, fisheries biology, nutrition, immunology, pathology, anatomy, morphometrics, biometrics and any other research involving the use of animals are invited for publication. While the main objective is to provide a forum for papers describing the results of original research, review articles are also welcomed. Articles submitted to the journal is peer reviewed to ensure a high standard.

### Editor:

Professor F. C. Okafor

### Associate Editor/Secretary

Dr. Joseph E. Eyo

### Editorial Advisory Committee

Prof. M. C. Eluwa	Prof. A. O. Anya
Dr. N. M. Inyang	Prof. E. I. Braide
Dr. B. O. Mgbenka	Dr. G. T. Ndifon
Prof. Bato Okolo	Dr. (Mrs.) R. S. Konya
Prof. I B Igbinosa	Prof. N. Umechue
Prof. A. A. Olatunde	Prof. B. E. B. Nwoke
Prof. O. A. Fabenro	Prof. F. J. Udeh
Prof. R. P. king	Prof. A. A. Adebisi
Prof. E. Obiekezie	Prof. W. S. Richards
Prof. J. A. Adegoke	Dr. W. A. Muse
Prof. D. N. Onah	Prof. O. U. Njoku

### Subscription Information

Animal Research International is published in April, August and December. One volume is issued each year. Subscription cost is US \$200.00 a year (₦1,400.00) including postage, packing and handling. Each issue of the journal is sent by surface delivery to all countries. Airmail rates are available upon request. Subscription orders are entered by calendar year only (January - December) and should be sent to The Editor, Animal Research International, Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka. All questions especially those relating to proofs, publication and reprints should be directed to The Editor, Animal Research International,

Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka

### Change of address

Subscribers should notify The Editor of any change in address, 90 days in advance.

### Advertisements

Apply to Animal Research International, Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka.

## Animal Research International®

### Notice to Contributors

Original research papers, short communications and review articles are published. Original papers should not normally exceed 15 double spaced typewritten pages including tables and figures. Short communications should not normally exceed six double spaced typewritten pages including tables and figures. Manuscript in English should be submitted in triplicate including all illustrations to The Editor/Associate Editor Animal Research International, Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka. Submission of research manuscript to Animal Research International is understood to imply that it is not considered for publication elsewhere. Animal Research International as a policy will immediately acknowledge receipt and process the manuscript for peer review. The act of submitting a manuscript to Animal Research International carries with it the right to publish the paper. A handling charge of US \$ 20.00 or ₦500.00 per manuscript should be sent along with the manuscript to the Editor, Animal Research International. Publication will be facilitated if the following suggestions are carefully observed:

1. Manuscript should be typewritten in double spacing on A4 paper using Microsoft Word. An electronic copy [1.44 MB floppy] should be enclosed, or submit online at [divinelovejoe@yahoo.com](mailto:divinelovejoe@yahoo.com).
2. The title page should include title of the paper, the name(s) of the author(s) and correspondence address (es).
3. Key words of not more than 8 words should be supplied.
4. An abstract of not more than 5% of the length of the article should be provided.
5. Tables and figures should be kept to a minimum. Tables should be comprehensible without reference to the text and numbered serially in Arabic numerals.
6. Figures (graphs in Microsoft excel format, map in corel draw 10 format and pictures in photo shop format) should be comprehensible without reference to the text and numbered serially in Arabic numerals.

7. Symbols and common abbreviations should be used freely and should conform to the Style Manual for Biological Journals; others should be kept to a minimum and be limited to the tables where they can be explained in footnotes. The inventing of abbreviations is not encouraged- if they are thought essential, their meaning should be spelt out at first use.

8. References: Text references should give the author's name with the year of publication in parentheses. If there are two authors, within the text use 'and'. Do not use the ampersand '&'. When references are made to a work by three or more authors, the first name followed by *et al.* should always be used. If several papers by the same author and from the same year are cited, a, b, c, etc., should be inserted after the year publication. Within parentheses, groups of references should be cited in chronological order. Name/Title of all Journal and Proceeding should be written in full. Reference should be listed in alphabetical order at the end of the paper in the following form:

EYO, J. E. (1997). Effects of *in vivo* Crude Human Chorionic Gonadotropin (cHCG) on Ovulation and Spawning of the African Catfish, *Clarias gariepinus* Burchell, 1822. *Journal of Applied Ichthyology*, 13: 45-46.

EYO, J. E. and MGBENKA, B. O. (1997). Methods of Fish Preservation in Rural Communities and Beyond. Pages 16-62. In: Ezenwaji, H.M.G., Inyang, N.M. and Mgbenka B. O. (Eds.). *Women in Fish Handling, Processing, Preservation, Storage and Marketing*. Inoma from January 13 -17, 1997.

WILLIAM, W. D. (1983) *Life inland waters*. Blackwell Science, Melbourne

Manuscripts are copy edited for clarity, conciseness, and for conformity to journal style.

#### Proof

A marked copy of the proof will be sent to the author who must return the corrected proof to the Editor with minimum delay. Major alterations to the text cannot be accepted.

#### Page charges

A subvention of US \$600.00 (₦ 5,000.00) is requested per published article. The corresponding author will receive five off-prints and a copy of the journal upon payment of the page charges.

#### Copy right

Manuscript(s) sent to ARI is believed to have not been send elsewhere for publication. The author upon acceptance of his/her manuscript give ARI the full

mandate to electronically distribute the article globally through African Journal Online (AJOL) and any other abstracting body as approved by the editorial board.

#### Address

Animal Research International, Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka

**Phone:** 042-308030, 08043123344, 08054563188

**Website:** www. zoo-unn.org

**Email:** divinelovejoe@yahoo.com

#### ANNUAL SUBSCRIPTION RATE THREE NUMBERS PER VOLUME

CATEGORY	DEVELOP- ING COUNTRY	DEVELOP- ED COUNTRY	NIGERIA
STUDENT	\$ 200.00	\$ 300.00	N 1,400.00
INDIVIDUALS	\$ 300.00	\$ 350.00	N 2,000.00
INSTITUTION/ LIBRARY	\$ 500.00	\$ 600.00	N 5,000.00
COMPANIES	\$ 600.00	\$ 750.00	N 10,000.00

Pay with bank draft from any of the following banks **only**. (a) Afribank (b) Citizens Bank (c) Intercontinental Bank (d) Standard Trust Bank (e) United Bank for Africa (f) Union Bank (g) Zenith Bank (h) First Bank Nig. PLC (i) Western Union Money Transfer.

Addressed to **The Editor/Associate Editor**, Animal Research International, Department of Zoology, P. O. Box 3146, University of Nigeria, Nsukka.

Alternatively, you may wish to send the bank draft or pay cash directly to **The Editor/Associate Editor** at Animal Research International Editorial Suite, 326 Jimbaz Building, University of Nigeria, Nsukka.

For more details contact, The Secretary, Animal Research International, Department of Zoology, Editorial Suite Room 326, Faculty of Biological Sciences Building (Jimbaz), University of Nigeria, Nsukka. Enugu State, Nigeria.

## PERFORMANCE OF SHEEP GRAZING *Brachiaria decumbens*, *Panicum maximum* and *Pennisetum purpureum* IN COMBINATION WITH *Gliricidia sepium*

OSAKWE, Isaac Ikechukwu and NWAKPU, Petrus Emeka

Department of Animal Production and Fisheries Management, Ebonyi State University, PMB 053, Abakaliki, Nigeria

**Corresponding Author:** Osakwe, Isaac Ikechukwu. Department of Animal Production and Fisheries Management, Ebonyi State University, PMB 053, Abakaliki, Nigeria. Email: [osakwe@yahoo.com](mailto:osakwe@yahoo.com). Phone: 234-43-300-448; 234-8034910687.

### ABSTRACT

*The introduction of forage legumes into grass pastures has generally improved grazing animal production by increasing total edible biomass and nutrient profiles. An experiment was designed to study the performance of sheep grazing Brachiaria decumbens, Panicum maximum and Pennisetum purpureum in combination with Gliricidia sepium. Eighteen paddocks of approximately 0.03 ha were used in the trial. Nine of the paddocks had Gliricidia sepium alley planted in rows 4 m apart and interplanted with 4 rows of either Brachiaria decumbens, Panicum maximum, or Pennisetum purpureum. The other nine paddocks had only the grass species without the Gliricidia sepium. The paddocks were each grazed by 3 sheep. The pure grass stands without the Gliricidia sepium served as controls for the grass species in combination with Gliricidia sepium. The three grasses and their combinations within the alley plots were replicated three times. The animals were grazed continuously for 28 days in the sub plots. Sheep grazing the Gliricidia/Panicum plot had a higher ( $P < 0.01$ ) growth rate (38 g d<sup>-1</sup>) than those animals grazing both the Gliricidia/Bracharia (23 g d<sup>-1</sup>) and Gliricidia/Pennisetum (21 g d<sup>-1</sup>) plots respectively. There was no significant difference ( $P > 0.05$ ) between sheep grazing the Gliricidia/Bracharia and Gliricidia/Pennisetum plots. The total dry matter intake of sheep on the Gliricidia/Panicum plot was higher ( $P < 0.05$ ) (1.33 kg DM d<sup>-1</sup>) than that of sheep on Gliricidia/Bracharia (0.86 kg DM d<sup>-1</sup>) and Gliricidia/Pennisetum (0.43 kg DM d<sup>-1</sup>) plots respectively. The total biomass from the Gliricidia/Bracharia (23 t ha<sup>-1</sup>) and Gliricidia/Panicum (21 t ha<sup>-1</sup>) plots respectively were higher ( $P < 0.01$ ) than the total biomass from the Gliricidia/Pennisetum (13 t ha<sup>-1</sup>) plot. These results demonstrate that grazing West African dwarf sheep in a Gliricidia sepium/Panicum maximum plot improved their growth rate during dry season when feed supplies are limited. It also underscores the poor performance of animals grazing Pennisetum purpureum in Gliricidia sepium alley plot.*

**Keywords:** Grazing, *Bracharia*, *Panicum*, *Pennisetum*, *Gliricidia*, Sheep

### INTRODUCTION

A major problem facing livestock farmers worldwide is how to economically maximize animal production with limited land availability. The situation is even worsened by desertification, leaching and urbanization. The potential to increase ruminant production on these land areas can be realized if innovations in managing rangeland are adopted. Tropical pastures have long been recognized as capable of producing large quantities of forage dry matter; however, individual animal performance is normally less per animal than for similar animals grazing temperate zone forages (Minson and Wilson, 1981; Moore and Mott, 1973). Ellis et al. (1976) reported that grazing behaviour of animals is based on availability and preference for plant species and/or portions of plants. Small ruminant production system in Nigeria is based on indoor feeding, grazing of natural or sown pasture or a combination of these. Grazing of sown pastures however, is limited to universities, research institutions and a few private farms where animal performance can be better evaluated. There have been few grazing trials in the country to determine productivity of pasture, and

small ruminant performance. Sumberg (1985) reported an improvement in the nutritional quality of natural fallow regrowth in a *Gliricidia sepium* alley plot. The planting of browse species such as *Gliricidia sepium* and *Leucaena leucocephala* as hedgerows of alleys in native or productive permanent grass plots may overcome the constraint to animal production caused by lack of fodder in the dry season. The presence of legume forages and tree forages in pasture have been generally accepted to improve ruminant productivity in both tropical (Milford, 1967) and temperate (Ulyatt, 1980) pastures. In addition, the extensive root systems of leguminous trees bind soil and so control soil erosion. Trees also reduce the direct effects of wind erosion. This study was therefore undertaken to determine the performance of sheep grazing *Brachiaria decumbens*, *Panicum maximum* and *Pennisetum purpureum* in *Gliricidia sepium* alley plots.

### MATERIALS AND METHODS

**Study Area:** The study was carried out at the International Livestock Centre for Africa (ILCA), now International Livestock Research Institute (ILRI),

Ibadan, Nigeria. The station is located between latitudes 6°10'1 and 9° 10'1 North of the equator and longitudes 3° and 6° East of the Greenwich, at an altitude of 200 m above sea level with annual rainfall averaging 1500 mm. The vegetation in this area is made up of derived guinea savanna and humid forest zone (Ezenwa, 1995). Mixed farming had been practised in the area for several decades.

**Experimental Design:** For this study, randomised complete block design was used consisting of 18 paddocks of approximately 0.03 ha. Nine of the paddocks had *Gliricidia sepium* alley planted in rows 4 m apart and interplanted with 4 rows of either *Brachiaria decumbens*, *Panicum maximum*, or *Pennisetum purpureum*. The other nine paddocks had only the grass species without the *Gliricidia sepium*. The pure grass stands without the *Gliricidia sepium* served as controls for the grass species in combination with *Gliricidia sepium*. Each plot was grazed by 3 sheep. The three grass combinations within the alley plots were replicated three times. The animals were grazed continuously for 28 days in the sub plots. Sampling of the grasses and *Gliricidia* was done at the start of grazing (i.e. before grazing), then weekly, and at the end of the grazing period. The weekly sampling was to estimate forage quantity and utilization rates. Grazing was discontinued anytime dry-matter (DM) on offer falls below 2.5 % of total body weight of the grazing animals.

**Sheep:** 9 - 12 months old sheep were assigned to the various subplots on the basis of initial forage on offer allowing 20 kg DM for 15 kg sheep. Animals were weighed weekly. The animals were able to harvest the upper foliage of the *Gliricidia* tree by leaning on the tender stem. The matured stem were bent down and tied so that the upper foliage became available for grazing. The animals were treated with an anthelmintic before grazing began. They were provided with shade and mineral salt block in each paddock. Dry matter yield on offer was estimated by cutting from three random 0.5 x 0.5 m quadrats for the grass and 1m x 1m quadrats for *Gliricidia* in each paddock. The cutting was done at 10 cm above ground level with a hand shear for grasses and 25 cm above ground level for the trees. Data were analysed using Analysis of Variance

## RESULTS

**Growth Rate:** The growth data of sheep grazing *Brachiaria*, *Panicum* and *Pennisetum* in *Gliricidia sepium* alley are shown in Figure 1. Sheep grazing the *Gliricidia/Panicum* plot had a higher ( $P < 0.01$ ) growth rate (38 g d<sup>-1</sup>) than those sheep grazing both *Gliricidia/Brachiaria* (23 g d<sup>-1</sup>) and *Gliricidia/Pennisetum* (21 g d<sup>-1</sup>) plots respectively. The mean body weight of sheep grazing *Gliricidia/Brachiaria* plot was not significantly different ( $P > 0.01$ ) from those grazing the *Gliricidia/Pennisetum* plot.

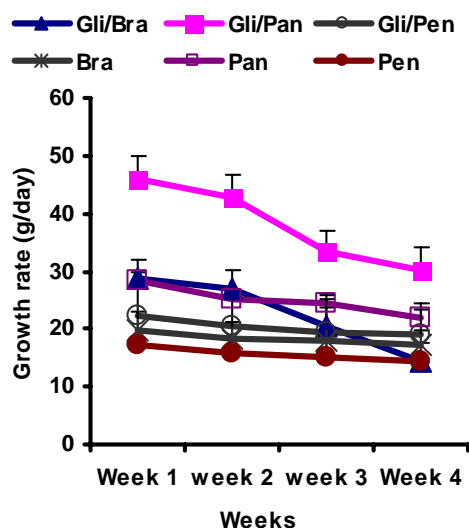


Figure 1: Growth rate of sheep grazing *Brachiaria*, *Panicum* and *Pennisetum* in *Gliricidia* alley.

**Total Dry Matter Intake:** Total dry matter intake (TDMI) of sheep grazing *Brachiaria*, *Panicum* and *Pennisetum* in *Gliricidia sepium* alley are summarised in Figure 2. Sheep grazing *Gliricidia/Panicum* plot had a higher ( $P < 0.05$ ) TDMI (1.33 kg DM d<sup>-1</sup>) than those grazing *Gliricidia/Pennisetum* (0.43 kg DM d<sup>-1</sup>) plot but not significantly different ( $P > 0.05$ ) from sheep grazing *Gliricidia/Brachiaria* (0.86 kg DM d<sup>-1</sup>) plots. The intake of dry matter of sheep on *Pennisetum/Gliricidia* alley plots was not different ( $P > 0.05$ ) from those on *Brachiaria/Gliricidia* plots.

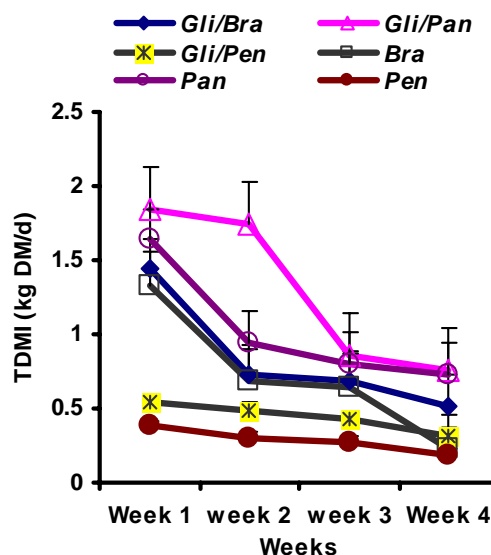


Figure 2: Total dry matter intake of sheep grazing *Brachiaria*, *Panicum* and *Pennisetum* in *Gliricidia* alley



**Total Biomass:** Total biomass yields of the three grasses in *Gliricidia sepium* plots are shown in Figure 3. The total biomass (TBM) yields from the *Gliricidia/Brachiaria* (23 t ha<sup>-1</sup>) and *Gliricidia/Panicum* (22 t ha<sup>-1</sup>) plots were higher ( $P < 0.01$ ) than the TBM (13 t ha<sup>-1</sup>) from the *Gliricidia/Pennisetum* plot. There was however, no significant difference ( $P > 0.01$ ) between the TBM yields of *Gliricidia/Brachiaria* and *Gliricidia/Panicum* plots.

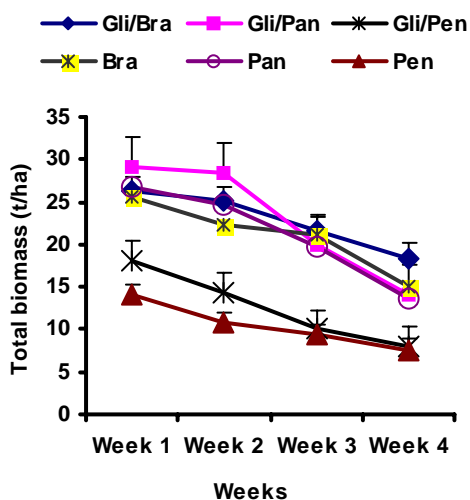


Figure 3: Total biomass yields of *Brachiaria*, *Panicum* and *Pennisetum* in *Gliricidia* alley plots

## DISCUSSION

The effects of supplementing *Gliricidia sepium* to basal grass diets on growth and survival rates of WAD sheep and goats have been reported (ILCA, 1988). In that study, sheep response was twice that of the goats. ILCA (1988) reported a growth rate of 30.3 to 48.9 g/d for male and 25.5 to 37.7 g/d for females sheep fed *Panicum* grass at different levels of *Gliricidia* supplementation. The sheep used in this study were not sexed. The growth rate of 38 g/d observed in *Gliricidia/Panicum* plot is consistent with the report of ILCA (1988).

The introduction of browse trees such as *Gliricidia* and *Leucaena* into planted pasture has contributed substantially to the dietary nutrient profiles of livestock under grazing condition. The higher TDMI in the *Gliricidia/Panicum* plot could be attributed to preference of *Panicum* grass to the other grass species, which resulted in higher growth rate of sheep. It was observed that yields of total biomass declined as the grazing days increased. This is in agreement with the findings of Mears and Humphreys (1974) who reported reductions in green matter of Kikuyu grass (*Pennisetum clandestinum*) as stocking rate and grazing days increased.

Similarly, Watson and Whiteman (1981) reported a drop in green yields of mixed pastures of *Panicum maximum* and *B. decumbens*. The general decline in live-weight gain as grazing progressed was attributed to reduced nutritive value due to advanced

plant maturity (Blunt, 1978), and thus reduction in herbage, especially leaf, on offer (Laredo and Minson, 1973).

**Conclusion:** Data from this study showed that there is a great potential for improvement in growth rate of sheep grazing planted pasture with *Gliricidia sepium* as hedgerows or alleys. The availability of the browse plants during dry season would have improved the quality of the declining nutritive value of the grass species, and hence reduced the characteristic weight loss during this period. In addition, to weight loss reduction, the use of leguminous browse plant improves soil fertility and structure, provides firewood to the household and acts as a windbreak when planted in the farm. The nutrient recycling from the grazing animals would also improve the nutrient status of the rangeland.

## ACKNOWLEDGEMENT

The author wishes to acknowledge with appreciation the contributions of Mr. Sam Ofodile of International Institute of Tropical Agriculture (IITA) Ibadan, Nigeria and the assistance of the technical staff and livestock attendants of ILCA (ILRI) for data collection. I am also grateful to Mr. Usen Sampson and Emmanuel Iweha for their interest and special assistance during the study.

## REFERENCES

- BLUNT, C. G. (1978). The production from steers grazing nitrogen fertilized irrigated pangola grass in the Ord Valley. *Tropical Grasslands*, 12: 90 – 96.
- ELLIS, J. E., WEINS, J. A., RODELL, C. F. and ANWAY, J. A. (1976). A conceptual model of diet selection as an ecosystem process. *Journal of Theoretical Biology*, 60: 93 – 108.
- EZENWA, I. V. (1995). *Forage production potentials of herbaceous and tree legumes in mixtures with grasses in Oyo-State, Nigeria*. Ph.D. Thesis, Agronomy Department, University of Ibadan.
- ILCA (1988). *Effects of polyphenolic compounds in forage from multipurpose fodder trees on growth, intake and digestion in sheep and goats*. ILCA (International livestock Centre for Africa) Annual report 1987. Addis Ababa, Ethiopia. pp 17 – 34.
- LAREDO, M. A. and MINSON, D. J. (1973). The voluntary intake, digestibility and retention time by sheep of leaf and stem fractions of five grasses. *Australian Journal of Agricultural Research*, 24: 875 – 888.
- MEARS, P. T. and HUMPHREYS, L. R. (1974). Nitrogen response and stocking rate of *Pennisetum clandestinum* pastures. 1. Pasture nitrogen requirement and concentration, distribution of dry matter and botanical composition. *Journal of Agricultural Science*, 83: 451 – 467.
- MILFORD, R. (1967). Nutritive values and chemical composition of seven tropical legumes and

- lucerne grown in subtropical south-eastern Queensland. *Australian Journal of Experimental Agriculture and Animal Husbandry*, 7: 540 – 545.
- MINSON, D. J. and WILSON, J. R. (1981). *Comparative digestibility of tropical and temperate grasses and legumes*. Annual Report, Tropical Crops and Pastures, Division of Commonwealth Scientific and Industrial Research Organization (CSIRO).
- MOORE, J. E. AND MOTT, G. O. (1973). Structural inhibitors of quality in tropical grasses. Pages 53 – 98. *In*: MATCHES, A. G. (Ed.). *Anti-quality components of forages*. American Society of Agronomy, Crop Science Society of America (CSSA) Special Publication 4.
- SUMBERG, J. E. (1985). Small ruminant feed production in a farming systems context. Pages 41 – 46. *In*: SUMBERG, J. E. and CASSADAY, K. (Eds.). *Sheep and goat production in humid West Africa*. Proceedings of the workshop on small ruminant production systems in the humid zone of West Africa, held in Ibadan, Nigeria. 23 – 26 January 1984.
- ULYATT, M. J. (1990). The feeding value of temperate pastures. Pages 125 - 141. *In*: MONLEY, F. P. W. (Ed.). *Grazing Animals*. Elsevier, Amsterdam.
- WATSON, S. E. and WHITEMAN, P. C. (1981). Grazing studies on the Guadalcanal Plains, Solomon Islands. 2. Effects of pasture mixtures and stocking rates on animal production and pasture components. *Journal of Agricultural Science*, 97: 353 – 364.

## SUSCEPTIBILITY OF *Aedes aegypti* PUPAE TO NEEM SEED KERNEL EXTRACTS

<sup>1</sup>UMAR, Abba., <sup>2</sup>KELA, Santa Larit and <sup>2</sup>OGIDI, Jonathan Asadabe

<sup>1</sup>Department of Biological Sciences, University of Maiduguri, PMB 1069, Maiduguri, Borno State, Nigeria

<sup>2</sup>Biological Sciences programme, Abubakar Tafawa Balewa University, PMB 0248, Bauchi, Bauchi State, Nigeria

**Corresponding Author:** UMAR, Abba, Department of Biological Sciences, University of Maiduguri, PMB 1069, Maiduguri, Borno State, Email: [Aumar66.ng@Hotmail.com](mailto:Aumar66.ng@Hotmail.com)

### ABSTRACT

*Laboratory bioassays were conducted to evaluate the pupicidal activity of neem (Azadirachta indica) seed kernel extracts (NSKE) on Aedes aegypti. The neem seed kernel powder was sequentially extracted with hexane, benzene, ethyl acetate, acetone, DMSO, 2-propanol, ethanol, methanol and distilled water. Ten concentrations (0.0, 0.25, 0.5, 1.0, 2.0, 4.0, 5.0, 10.0, 15.0 and 20.0%) of the neem extracts were used for the bioassays. Each treatment was replicated five times. Twenty-laboratory strains of Aedes aegypti pupae were exposed to each concentration. Pupae were not fed during the exposure periods. Pupal mortality was assessed after 1 and 24 hours of exposure. The results of the effects of 1h exposure indicated decreased pupicidal mortality with decreasing extracts toxicity thus: ethyl acetate ( $LC_{50} = 0.06\%$ ) > acetone > ( $LC_{50} = 0.29\%$ ) > benzene ( $LC_{50} = 0.82\%$ ) > hexane ( $LC_{50} = 3.13\%$ ) and propanol ( $LC_{50} = 7.63\%$ ). No pupal mortality was observed with extracts from Dimethyl sulfoxide (DMSO), ethanol, methanol and distilled water. The results of the effect of extract for 24h exposure indicated pupicidal mortality in 2-propanol ( $LC_{50} = 0.67\%$ ) and ethanol ( $LC_{50} = 1.70\%$ ). No pupicidal mortality was observed with hexane, benzene, ethyl acetate, acetone, Dimethyl sulfoxide (DMSO), methanol and distilled water extracts. The ability of some neem extracts to kill Aedes pupae at relatively low concentrations presents an alternative to the use of synthetic pesticides for control of mosquitoes. This technique is environmental friendly, biodegradable, less expensive, and locally available in mosquito endemic area. Potentials for adoption in mosquito management programmes cannot be overemphasized.*

**Keywords:** *Azadirachta indica*, *Aedes aegypti*, Pupicidal

### INTRODUCTION

*Aedes aegypti* Linn is a prevalent mosquito in the sahel savanna regions of Nigeria (Molineaux and Gramiccia 1980). They are established vector of yellow, dengue and other haemorrhagic viral fevers (Gubler, 1997). The epidemics of these diseases have been reported in Nigeria (Nasidi *et al.*, 1989).

Both private and public health mosquito control in Nigeria is largely based on the conventional synthetic insecticides (Don-Pedro and Adegbite, 1985). These conventional insecticides are associated with high costs (Jackai, 1993), persistent development of resistance in many of the mosquito species (Brown, 2002), adverse effects on non-target organisms (Hennessey *et al.*, 1992), human toxicity reactions (Liu *et al.* 2003) and non-suitability in integrated pest management programmes (Schmutterer, 1990).

The problems associated with synthetic insecticides necessitated investigations in to phytochemicals for mosquito control, since they are environmental friendly, biodegradable, less expensive, and locally available in mosquito endemic area (Novak, 1985). Phytochemicals with diverse mode of actions may be effective against resistant vector species and can be easily integrated with other mosquito control measures in both private and public

mosquito control programmes (Sivagnaname and Kalyanasundaram, 2004).

Neem, *Azadirachta indica* (Family: Meliaceae) have met these requirements (Ipek *et al.*, 2004) and can play a vital role in mosquito control measures (Sukumar *et al.*, 1991). Although intensive work on neem as natural insecticides in Nigeria began in 1981 in the crop protection Unit of the Department of Agricultural Extension Services, University of Ibadan (Ivbijaro, 1987), little attention has been given to the potential of neem in mosquito control in Nigeria (Aleiro, 2003).

This study investigates the effects of neem seed kernel extracts as bio-insecticides against *Aedes aegypti* for possible usage in integrated mosquito control programmes in North Eastern Nigeria and other mosquito endemic areas of the third world.

### MATERIALS AND METHODS

**Insect Culture:** The eggs of *Aedes aegypti* were obtained from the National Vector and Malaria Control Unit, Yaba, Lagos. They were reared inside cages (60 × 60 × 60 cm) in the insectry of the Department of Biological Sciences, University of Maiduguri. The adults of both sexes were fed with 10 % glucose solution (Sneller and Dadd, 1977). In addition, females were fed on blood meal twice a week from restrain chicken with shaved abdominal



feathers (Azmi *et al.*, 1998). A 250 ml glass beaker containing 150 ml of distilled water with a filter paper smoothly adhered to the inner wall serves as oviposition sites. The larvae were held in plastic containers and were daily fed on a pinch of finely powdered liver and brewer's yeast mixed at the ratio of 3: 2 (wt\wt) (Roberts, 1998). When not needed for studies, the eggs were stored in a laboratory shelf at  $37 \pm 5^{\circ}\text{C}$ .

#### Neem Seed Kernel Extraction of Active Ingredient:

Neem seeds were collected from mature disease-free trees in University of Maiduguri in October 2004. The neem seeds were air dried and stored in the laboratory. The dried neem seeds were decorticate to remove the kernels and air dried for 5 days before pulverization with an electric blender and sieved with 40 mesh screen to obtain a fine powder. 500 g of neem seed kernel powder was sequentially extracted either with 2 liters of hexane or benzene or ethyl acetate or acetone or Dimethyl sulfoxide (DMSO) or 2-propanol or ethanol or methanol or distilled water for 48 hours at room temperature ( $37 \pm 5^{\circ}\text{C}$ ). The mixture was stirred and filtered through Whatman number one filter paper and the filtrates were evaporated to dryness in a water bath ( $40^{\circ}\text{C}$ ). The residues were air dried by placing the container near an electric fan (Ascher, 1981). The powder or liquid obtained from each extraction was stored in a refrigerator at  $4^{\circ}\text{C}$  separately in labeled specimen bottles for bioassay.

**Pupal Susceptibility Tests:** The pupicidal effects of the neem seed kernel extracts (NSKE) were determined using standard procedures (WHO, 1970). The bioassay was conducted under varying laboratory conditions ( $37 \pm 5^{\circ}\text{C}$  Temperature,  $80 \pm 5\%$  Relative Humidity and 12:12 Light: Dark Photoperiods). Ten concentrations (0.0, 0.25, 0.5, 1.0, 2.0, 4.0, 5.0, 10.0, 15.0 and 20.0 %) of each organic solvent and distilled water extracted neem seed kernel were used for the bioassay. Each treatment was replicated five times. Twenty-laboratory bred *Aedes aegypti* pupae were used per replicate. The pupae were siphon with eyedropper and immediately exposed in to 100 ml of each concentration of NSKE or distilled water (control). The exposure containers (250 ml glass beakers) were covered with mesh screen to prevent the escape of emerging adults. The pupal mortalities were recorded 1 and 24 hours of exposure. The pupae were probe with needle and moribund pupae were counted as dead (Azmi *et al* 1998). The results were analyzed using StatsDirect Statistical Software Version 4.2 (StatsDirect, 2005) to obtain the probit values.

## RESULTS

The results of the pupicidal effects of 1h exposure of *A. aegypti* pupae to neem seed kernel extracts are presented in Table 1. The result indicated pupicidal effects with decreasing toxicity in ethyl acetate ( $\text{LC}_{50} = 0.06\%$ ) > acetone > ( $\text{LC}_{50} = 0.29\%$ ) > benzene

( $\text{LC}_{50} = 0.82\%$ ) > hexane ( $\text{LC}_{50} = 3.13\%$ ) and propanol ( $\text{LC}_{50} = 7.63\%$ ) extracts.

**Table 1: Effects of 1h exposure of neem seed kernel extracts on laboratory strain *Aedes aegypti* pupae**

Extracts	Lethal concentration values	
	LC <sub>50</sub> (%)	LC <sub>90</sub> (%)
Hexane	3.13 (1.50 -23.28) *	374.03 (35.91 -196.67)
Benzene	0.82 (00.60 -1.94)	6.41 (2.34 -9.05)
Ethyl acetate	0.06 (7.45 - 0.20)	0.42 (0.01 -0.67)
Acetone	0.29 (0.07- 0.74)	1.84 (0.81 -6.28)
Dimethyl sulfoxide (DMSO)	0.00 (0.00-0.00)	0.00 (0.00-0.00)
2-propanol	7.63 (3.18 -847.23)	169.44 (20.57- 328.93)
Ethanol	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Methanol	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Distilled water	0.00 (0.00-0.00)	0.00 (0.00-0.00)

\* Numbers in parentheses are 95% confidence limits

However, no pupal mortalities were observed with extracts from DMSO, ethanol, methanol and distilled water. The results of the pupicidal effects of 24h exposure of *A. aegypti* pupae to neem seed kernel extracts are presented in Table 2.

**Table 2: Effects of 24 h exposure of neem seed kernel extracts on laboratory strain *Aedes aegypti* pupae**

Extracts	Lethal concentration values	
	LC <sub>50</sub> (%)	LC <sub>90</sub> (%)
Hexane	0.00 (0.00-0.00)*	0.00 (0.00-0.00)
Benzene	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Ethyl acetate	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Acetone	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Dimethyl sulfoxide (DMSO)	0.00 (0.00-0.00)	0.00 (0.00-0.00)
2-propanol	0.67 (0.37- 0.96)	1.67 (3.83 -83.08)
Ethanol	1.70 (1.03 -526.91)	19.53 (3.69 -2027.47)
Methanol	0.00 (0.00-0.00)	0.00 (0.00-0.00)
Distilled water	0.00 (0.00-0.00)	0.00 (0.00-0.00)

\* Numbers in parentheses are 95% confidence limits

The result indicated pupal mortalities with decreasing toxicity in extracts from 2-propanol ( $\text{LC}_{50} = 0.67\%$ ) and ethanol ( $\text{LC}_{50} = 1.70\%$ ). However, hexane, benzene, ethyl acetate, acetone, DMSO, methanol and distilled water extracts of NSK had no pupicidal effects.

## DISCUSSION

The toxicities of neem seed kernel extracts to mosquito pupae under laboratory conditions were studied. The findings of this report will serve as base line data for mosquito control in northeastern Nigeria. The present investigation revealed pupal mortalities in ethyl acetate, acetone, benzene, hexane, 2-propanol and ethanol extracts. These results corroborate earlier investigations, that neem seed extracts that are effective against insects were extracted with hexane, ethyl ether, acetone, ethanol, and methanol (Jacobson, 1981). Schmutterer (1990) reported that neem seed extracted with hexane, pentane, ethanol, methanol, esters and dichloromethane as well mixtures of any of these solvents with water possessed insecticidal activities.

The results also showed that the toxicity of neem extracts decreased with polarity of extraction solvent. This contradicts the results of Zebitz (1984) that revealed the toxicity of neem extracts increased with the polarity of the extraction solvents. Schaver and Schmutterer (1981) revealed that nonpolar solvents are more effective than polar ones in extracting substances active against mites from neem kernels.

Although the present findings showed that methanolic extracts had no pupicidal effects on *A. aegypti* pupae, the results of Sivagnaname and Kalyanasundaram (2004) showed that methanolic extracts of the leaves of *A. indica* was effective against *Culex quinquefasciatus* and *A. aegypti* pupae. These authors revealed that the extract was less effective against the larvae of *Anopheles stephensi*, but was more effective against the pupae of *A. stephensi* compared to other species. However, these authors did not use sequential extraction technique and the preceding solvents might have extracted most active components. The potency of the methanolic extract reported by Sivagnaname and Kalyanasundaram (2004) could be due to the ability of methanol to extract a great amount of polar compounds and its effectiveness in eluting salanin, desacetyl-nimbin and nimbin (Feuerhake, 1984).

The result of present Rao *et al.* (1992) experimented under field conditions showed that application of neem cake powder at a dose of 500 kg/ha alone or coated on urea resulted in drastic reduction in the late instars larvae and pupae of culicine mosquitoes. In another studies Rao *et al.* (1995) showed that lipid rich fractions of neem was effective in control of the breeding of culicine vectors of Japanese encephalitis and equally produced significant reduction in populations of anopline pupae.

The pupal mortality observed in the hexane extracts could be due to the effects of the oil fractions on their respiratory system. In earlier studies, Corbet *et al.* (1995) observed susceptibility of mosquito larvae and pupae to surface materials entering their tracheal system and reported that essential oils increased the tendency to tracheal flooding and chemical toxicity. It has been

established that pupal mortality was due to the effects of azadirachtin, the most biologically active substance from neem that modifies the insect's physiology by influencing hormonal systems especially that of ecdysone (Schmutterer, 1990). Mordue and Blackwell (1993) had earlier reported that azadirachtin prevented ecdysis and apolysis and caused pupal death before and during molting. The results showed that the susceptibility of *A. aegypti* pupae to neem seed kernel extracts were lesser than that of 4<sup>th</sup> instar larvae confirming earlier results (Boschitz and Grunewald, 1994) that revealed that sensitivity of *A. aegypti* towards Neemzal decreased with increasing age of the larvae.

Further research on the potentials of these extracts in field conditions become imperative for practical implementations of mosquito control programmes to protect human populations from the scourge of mosquito-borne diseases in the northeastern Nigeria and other mosquito endemics areas of the world.

## REFERENCES

- ALIERO, B. L. (2003). Larvicidal effects of aqueous extracts of *Azadirachtin indica* (neem) on the larvae of *Anopheles* mosquitoes. *African Journal of Biotechnology*, 2(9): 325 – 327.
- ASCHER, K. R. S. (1981). Some physical (solubility) properties and biological sterility for *Epilachna varivestis* females effects of a dried methanolic neem (*Azadirachtin indica*) seed kernel extract. Pages 63 – 74. In: SCHMUTTERER, H. and ASCHER, K. R. S. (Eds). *Natural pesticides from the neem tree (Azadirachtin indica A. Juss) and other tropical plants. Proceedings of 1st International Neem Conference*, Rauschholzhausen, Federal Republic of Germany, 16 – 18 June, 1980.
- AZMI, M. A., NAQVI, S. N. H., AHMAD, I., TABASSUM, M. R. and ANBREEN, B. (1998). Toxicity of neem leaves extract (NLX) compared with Malathion (57 E.C.) against late 3rd instar larvae of *Culex fatigans* (wild strain) by WHO method. *Tropical Journal of Zoology*, 22: 213 – 218.
- BOSCHITZ, C. and GRUNEWALD, J. (1994). The effects of Neemzal on *Aedes aegypti* (Diptera :Culicidae). *Journal of Applied Parasitology*, 35(4): 251 – 256.
- BROWN, A. W. A. (1986). Insecticide resistance in mosquitoes: a pragmatic review. *Journal of the American Mosquito Control Association*, 2: 123 – 139.
- CORBET, S. A., DANAHAH C. W., KING V., CHALMERS, C. L. and TILEY, C. F. (1995). Surfactant-enhanced essential oils as mosquito larvicides. *Journal of Experimental and Applied Entomology*, 75: 229 – 236.
- DON-PEDRO, K. N. and ADEGBITE, T. O. (1985). Nuvar resistance in a strain of *Aedes aegypti* in Lagos, Nigeria. *Environmental Pollution*, 38: 19 – 24.
- FEUERHAKE, K. J. (1984). Effectiveness of technical solvents for the extraction of neem seed components with insecticidal activity. Page 103 – 113. In: Schmutterer, H. and Ascher, K. R. S.

- (Eds.). *Natural pesticides from the neem tree (Azadirachtin indica A. Juss) and other tropical plants. Proceedings of 2nd international Neem Conference*, Rauischholzhausen, Federal Republic of Germany, 25 – 28 May, 1983.
- GUBLER, D. J. (1997). Epidemic Dengue/Dengue Haemorrhagic Fever: A Global Public Health Problem in the 21st Century. *Dengue Bulletin*, 21: 21 – 31.
- HENNESSEY, M. K., NIGG, H. N. and HABECK, D. H. (1992). Mosquito (Diptera: Culicidae) adulticide drift into wildlife refuges of the Florida keys. *Journal of Environmental Entomology*, 21(4): 714 – 721.
- IPEK, G. RALP, P. and MOHAMMED, A. (2004). Ecological risk assessment of neem-based pesticides. *Journal of Environmental Science and Health, Part B- Pesticides, Food Contaminants and Agricultural Wastes*, 39(2): 311 – 321.
- IVBIJARO, M. F. (1987). Prospects for neem in Nigeria agriculture. Pages 525 – 533. In: SCHMUTTERER, H. and ASCHER, K. R. S. (Eds) *Natural pesticides from the neem tree (Azadirachtin indica A. Juss) and other tropical plants. Proceedings of 3<sup>rd</sup> International Neem Conference*, Nairobi, Kenya, 10 - 15 July, 1986.
- JACKAI, L. E. N. (1993). The use of neem in controlling cowpea pests. *International Institute of Tropical Agriculture Research Bulletin*, 7: 5 – 11.
- JACOBSON, M. (1981). Neem Research in the US Department of Agriculture: Chemical, biological and cultural aspects. Pages 33 – 42. In: SCHMUTTERER, H. and ASCHER, K. R. S. (Eds) *Natural pesticides from the neem tree (Azadirachtin indica A. Juss) and other tropical plants. Proceedings of 1st International Neem Conference*, Rauischholzhausen, Federal republic of Germany, 16 – 18 June, 1980.
- LIU, W., ZHANG, L., JAMAL, H. H., JULIANA, J., ZAILINI, H. and BERNANRD, G. D. (2003). Mosquito coils Emissions and health implications. *Environmental Health Perspectives*, 111(12): 1430 – 1437.
- MOLINEAUX, L. and GRAMICCIA, G. (1980). *The Gaarki project research on the epidemiology and control of malaria in Sudan savannah of West Africa*. World Health Organization, Geneva. 406 pp.
- MORDUE, A. J. and BLACKWELL, A. (1993). Azadirachtin: An update. *Journal of Insect Physiology*, 39: 903 – 924.
- NASIDI, A., MONATH, T. P., DEOCK, K., TOMORI, O., CORDELLIER, R., OLALEYE, O. D., HARRY, T. O., ADENIYI, J. A., SORUNGBE, A. O., AJOSE-COKER, A. O., VAN DER LAAN, G. and OYEDIRAN, A. B. O. (1989). Urban yellow fever epidemic in western Nigeria, 1987. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 83(3): 401 – 406.
- NOVAK, D. (1985). Non-chemical approaches to mosquito control in Czechoslovakia. Pages 185 – 196. In: LAIRD, M. and MILES, J. W. (Eds). *Integrated mosquito control methodologies*. Volume 2. Academic Press, Dan Diego.
- RAO, D. R., REUBEN, R., VENUGOPAL, M. S., NAGASAMPAGI, B. A. and SCHMUTTERER, H. (1992). Evaluation of neem, *Azadirachta indica*, with and without water management, for the control of culicine mosquito larvae in rice-fields. *Journal of Medical and Veterinary Entomology*, 6(4): 318 – 324.
- RAO, D. R., REUBEN, R. and NAGASAMPAGI, B. A. (1995). Development of combined use of neem (*Azadirachta indica*) with and without water management, for the control of culicine mosquito larvae in rice-fields. *Journal of Medical and Veterinary Entomology*, 9(1): 25 – 33.
- ROBERTS, D. (1998). Overcrowding of *Culex sitiens* (Diptera: Culicidae) larvae: population regulation by chemical factors or mechanical interference. *Journal of Medical Entomology*, 35(5): 665 – 669.
- SCHAUER, M and SCHMUTTERE, H. (1981). Effects of neem seed kernel extracts on the two-spotted spider mite, *Tetranychus urticae*. Pages 259 – 266. In: SCHMUTTERER, H. and ASCHER, K. R. S. (Eds) *Natural pesticides from the neem tree (Azadirachtin indica A. Juss) and other tropical plants. Proceedings of 1st International Neem Conference*, Rauischholzhausen, Federal republic of Germany, 16 – 18 June, 1980.
- SCHUMTHERER, H. (1990). Properties and potential Natural pesticides from the Neem tree, *Azadirachtin indica*. *Annual Review of Entomology*, 35: 271 – 279.
- SIVAGNANAME, N. and KALYANASUNDARAM, M. (2004). Laboratory Evaluation of methanolic extract of *Atlantia monoplyla* (Family: Rutaceae) against immature stages of mosquitoes and non-target organisms. *Memorias do Instituto Oswaldo Cruz, Rio de-Janeiro*, 99(1): 115 – 118.
- SNELLER, V. P. and DADD, R. H. (1977). Requirement for sugar in a chemically defined diet for *Aedes aegypti* larvae. *Journal of Experimental and Applied Entomology*, 14: 387 – 392.
- STATSDIRECT (2005). StatsDirect Ltd.uk Version 4.2, 11 Gresham Way Cheshire, M33 3UY UK.
- SUKUMAR, K., PERICH, M. J. and BOOBAR, L. R (1991). Botanical derivatives in Mosquito control: A review. *Journal of the American Mosquito control Association*, 7: 210-217.
- WHO (1970) Expert committee on insecticides, 17<sup>th</sup> Reports, *World Health Organization Technical Report Series No. 433 (Annexes 2A and 2B)*, Geneva.
- ZEBTIZ, C. P. W. (1984). Effect of some crude and azadirachtin enriched neem (*Azadirachtin indica*) seed kernel extracts on larvae of *Aedes aegypti*. *Journal of Experimental and Applied Entomology*, 35: 11 – 16.

## SUSCEPTIBILITY OF MOSQUITO LARVAE TO CONVENTIONAL INSECTICIDES IN A TROPICAL ARID ECOSYSTEM

<sup>1</sup>UMAR, Abba, <sup>2</sup>KELA, Santaya Larit and <sup>1</sup>ABDULRAHMAN, Hauwa Tati

<sup>1</sup>Department of Biological Sciences, University of Maiduguri, P. M. B. 1069, Maiduguri, Borno State, Nigeria

<sup>2</sup>Biological Science programme, Abubakar Tafawa Balewa University, PMB 0248, Bauchi, Bauchi State, Nigeria

**Corresponding Author:** UMAR, Abba. Department of Biological Sciences, University of Maiduguri, P. M. B. 1069, Maiduguri, Borno State. [Email-Aumar66.ng@Hotmail.com](mailto:Email-Aumar66.ng@Hotmail.com) Tel: 0806379939, 08026922134

### ABSTRACT

*The susceptibility of 4th instar larvae of Aedes aegypti and Culex quinquefasciatus to dieldrin, dichlovos and cypermethrin were evaluated in laboratory. Larval mortality was assessed 24 hour after exposure. The result showed that the LD<sub>50</sub> values for Aedes aegypti exposed to dieldrin, dichlovos and cypermethrin were 0.48, 37.09 and 0.29 µg per liter respectively. The LD<sub>50</sub> values for Culex quinquefasciatus of exposed to dieldrin, dichlovos and cypermethrin were 0.11, 10.05 and 0.05 µg per liter respectively.*

**Keywords:** *Aedes aegypti*, *Culex quinquefasciatus*, LC<sub>50</sub>, Dieldrin, Dichlovos, Cypermethrin.

### INTRODUCTION

The development of resistance in mosquitoes to a wide variety of conventional insecticides has posed a serious problem for vector control program (Brown, 1986; Boike *et al.*, 1989; WHO, 1992; Deedat, 1994; Chandre *et al.*, 1999). These invariably led to the development of new insecticides for mosquito control besides the use of other control measures and multiple overdosed treatments, thus fostering serious human health concerns (Rozendaal, 1997; Brown, 1983). Pesticide resistant is a major constraint to mosquito control (Busvine, 1978).

To keep tract of these problems, the screening for susceptibility status of mosquitoes in the local environment is imperative. Little work is done to establish the susceptibility status of mosquitoes to commonly used insecticides in arid tropical ecosystem of Maiduguri, Nigeria.

The present study compares the susceptibility status of larvae of two vector mosquito species viz: *Culex quinquefasciatus* and *Aedes aegypti* to three conventional insecticides (Dieldrin, Dichlovos and Cypermethrin) using field strains of mosquitoes.

### MATERIALS AND METHODS

**Study Areas:** The study was conducted in Maiduguri located in the Sahel Savanna region of Northeastern Nigeria at latitude 11°05' North and longitude 13°05' East (BSBS, 2004). Maiduguri has mean annual rainfall of about 625 mm. The mean annual temperature and is about 32°C. The mean annual relative humidity for dry and rainy seasons was 40% and 60% respectively while the mean annual evaporation rate is about 1600 mm (Marte, 1986).

**Dieldrin:** Old stock of dieldrex 20 EC was obtained from a pesticide store. Dieldrex is a brand of dichlovos with a chemical formula: C<sub>12</sub>H<sub>8</sub>Cl<sub>6</sub>O. It is a contact and stomach poison with highly mammalian toxicity. They are Persistent insecticide (Kumar, 1984). The stock

solution was serially diluted to obtain 20, 40, 60, 80 and 10 µg per liter.

**Dichlovos:** Nuvan 100EC was purchased from a pesticide store in Maiduguri. Nuvan is a brand of dichlovos with a chemical formula: C<sub>4</sub>H<sub>7</sub>Cl<sub>2</sub>O<sub>4</sub>P. It is a fumigant, contact and stomach poison with low mammalian toxicity (Kumar, 1984). The stock solution was serially diluted to obtain 20, 40, 60, 80 and 10 µg per liter.

**Cypermethrin:** Cypercot 25 EC was purchased from a pesticide store in Maiduguri. Cypercot 25EC is a brand of cypermethrin with a chemical formula: C<sub>22</sub>H<sub>19</sub>Cl<sub>2</sub>NO<sub>3</sub>. It is a contact poison with low mammalian toxicity (Kumar, 1984). The stock solution was serially diluted to obtain 20, 40, 60, 80 and 10 µg per liter.

**Rearing of Mosquitoes:** The larvae of *A. aegypti* and *C. quinquefasciatus* were collected from their natural breeding habitats in Maiduguri, Borno State. The adults of both sexes were fed with 10 % glucose solution (Sneller and Dadd, 1977). In addition, females were fed on blood meal twice a week from restrain chicken with shaved abdominal feathers (Azmi *et al.*, 1998). A 250 ml glass beaker containing 150 ml of distilled water with a filter paper smoothly adhered to the inner wall serves as oviposition sites for *A. aegypti* and a plastic container with little quantity of water serve as oviposition sites for *C. quinquefasciatus*. The larvae of each species were separately held in plastic containers and were daily fed on a pinch of finely powdered liver and brewer's yeast mixed at the ratio of 3: 2 (wt: wt) (Roberts, 1998).

**Laboratory Bioassay:** A batch of 20; 4th instars larvae of *A. aegypti* or *C. quinquefasciatus* were separately exposed to 20, 40, 60, 80 and 10 µg of dieldrin, dichlovos and cypermethrin per liter o distilled water respectively. Larval mortality was assessed after 24 hours after exposure. The experiment was conducted at 37 ± 5 °C and 80 – 90 % relative humidity. The data obtained was subjected to probit

analysis using Statsdirect Statistical Software Version 4.2. (Statsdirect, 2005).

## RESULTS

The result of the study is presented in table 1. The result showed that the LD<sub>50</sub> values for *A. aegypti* exposed to dieldrin, dichlovos and cypermethrin were 0.48 37.09 and 0.29 µg per liter respectively. The LD<sub>50</sub> values *C. quinquefasciatus* exposed to dieldrin, dichlovos and cypermethrin were 0.11, 10.05 and 0.05 µg per liter respectively.

**Table 1: Comparative toxicity of synthetic insecticides to *A. aegypti* and *C. quinquefasciatus***

Insecticide	Mosquito species	
	<i>A. aegypti</i>	<i>C. quinquefasciatus</i>
Dieldrin	0.481 (0.60-0.05)2	0.111 (0.16-0.08) 2
Dichlovos	37.09 (42.13-32.63)	10.05 (13.62-6.89)
Cypermethrin	0.29 (0.40-0.23)	0.05 (0.59-0.03)

*L* LD<sub>50</sub> values in µg per liter, 295 % Confidence interval

## DISCUSSIONS

Pesticide resistant is a major constraint of insect vector control (Busvine, 1978). Resistance in pest or vector population is expected to develop quickly whenever all individuals in the population are intensively selected with insecticides for several generations (Malcom, 1988). Results obtained in this study reveal that both *A. aegypti* and *C. quinquefasciatus* are less susceptible to dichlovos but more susceptible to cypermethrin and dieldrin. This confirms the findings of Molta and Ali (1998) who indicates that permethrin is potent against *Anopheles* species in northeastern Nigeria. The high mortality recorded with cypermethrin could be due to intoxication effects at different levels of pharmacokinetic interaction, thus; penetration of barrier tissue, distribution, storage, metabolism in internal tissue, and molecular interaction with the target site (Narahashi, 1976; WHO, 1980; Shamaan *et al.*, 1993; Curtis *et al.*, 1996).

Several other studies have revealed organophosphorus resistance in various species of mosquitoes (Don-Pedro and Adegbite, 1985; Amin and Peiris, 1990). The present study has shown that *A. aegypti* and *C. quinquefasciatus* were less susceptible to dichlovos. Thus their use in mosquito control may not be effective in the local environment. However, studies by Georghiou, (1980) and WHO (1992) showed that dichlovos could be use in mosquito control with effective resistance management techniques. These authors used higher concentrations than the ones used in this study. Although the present study did not indicate selection to dieldrin and cypermethrin in the local environment, other studies have revealed the resistance of mosquitoes to dieldrin (WHO, 1986; Amin and Hemingway, 1989) and cypermethrin (Chandre *et al.*, 1999). The studies of Kristan *et al.* (2003) revealed that resistance to pyrethroid insecticides was caused by the *kdr* gene in the malaria vector *Anopheles gambiae* Giles s.s. (Diptera: Culicidae).

Although the present study indicates that both mosquito species were susceptible to dieldrin and cypermethrin, the later is recommended for mosquito control because dieldrin has undesirable effects (Metcalf, 1980) and has been banned in many countries including Nigeria. As suggested by Dorta *et al.* (1993) synthetic pyrethroids could be effectively employed in integrated vector control operations. However, several reports have shown resistance to pyrethroids in several species of mosquitoes (WHO, 1992; Vulule *et al.*, 1994). Their results further revealed that *A. aegypti* is less susceptible to all the three insecticides than *C. quinquefasciatus*. This could be due to interplay of several factors Viz: biochemical (Hill, 1985), genital (Hemingway, 1983), behavioral (Miller and Gibson 1994) and physiological (Lockwood *et al.*, 1984).

It is concluded from the present study that, of the insecticides tested cypermethrin can be effectively used for controlling mosquito vectors and shall play a vital role in reducing the morbidity and mortality of mosquito borne-diseases in northeastern Nigeria and other mosquito endemic countries. However, pyrethroids insecticides could be used rationally, otherwise resistance problem to these insecticides will appear in the local environment in the future.

## REFERENCE

- AMIN, A. M. and HEMINGWAY, H. (1989). Preliminary investigation of the mechanisms of DDT and Pyrethroid resistant in *Culex quinquefasciatus* Say (Diptera: Culicidae) from Saudi Arabia. *Bulletin of Entomological Research*, 79: 361 – 366.
- AMIN, A. M. and PEIRIS, H. T. R. (1990). Detection and selection of organophosphorus and Carbamate resistance *Culex quinquefasciatus* Say (Diptera: Culicidae) from Saudi Arabia. *Medical and Veterinary Entomology*, 4: 265 – 269.
- AZMI, M. A., NAQVI, S. N. H., AHMAD, I., TABASSUM, M. R. and ANBREEN B. (1998). Toxicity of Neem Leaves Extract (NLX) Compared With Malathion (57 EC.) Against Late 3rd Instar Larvae of *Culex fatigans* (Wild Strain) by WHO Method. *Tropical Journal of Zoology*, 22: 213 – 218.
- BOIKE, A. H. JR., RATHBURN, C. B. JR., FLOORE, T. G., RODRIGUEZ, H. M and COUGHLIN, J. S. (1989). Insecticide tolerance of *Culex nigripalpus* in Florida. *Journal of American Mosquito Control Association* 5: 522 – 528.
- BSBLS (2004). *Map of Borno State, Nigeria*. Borno State Bureau of Land and Survey, Maiduguri, Borno State, Nigeria. 1 pp.
- BROWN, A .W .A. (1983). Insecticide resistance as a factor in the integrated control of Culicidae. Pages 161 – 235. *In: Laird, M. and Miles, J. W. (Eds). Integrated mosquito control methodologies*. Academic Press, New York.
- BROWN, A. W. A. (1986). Insecticide resistance in mosquitoes: A pragmatic review. *Journal of the American Mosquito Control Association*, 2: 123 – 140.



- BUSVINE, J. R. (1978). Current problems in the control of mosquitoes. *Nature*, 273: 604 – 607.
- CHANDRE, F., DARRIET, F., MANGA, L., AKOBELLO, M., FAYE, O., MOUCHET, J. and GUILLET, P. (1999). Status of pyrethroid resistance in *Anopheles gambiae* sensu lato. *Bulletin of World Health Organization*, 77: 230 – 234.
- CURTIS, C. F., MYAMBA, J. and WILKES, T. J. (1996). The comparison of different insecticides and fabrics for anti-mosquito bed nets and curtains. *Medical and veterinary Entomology*, 10(1): 1 – 11.
- DEEDAT, Y. D. (1994). Problems associated with the use of pesticides: An overview. *Insect Science and Application*, 15: 247 – 251.
- DON-PEDRO K. N. and ADEGBITE, T. O. (1985). Nuva resistance in a strain of *Aedes aegypti* in Lagos, Nigeria. *Environmental Pollution, (Series A)* 38: 19 – 24.
- DORTA, D., MVASUKI, V. and RAJAVEL, A. (1993). Evaluation of organophosphorus and synthetic pyrethroid insecticides against six vector mosquito species. *Reviews of Saúde Pública*, 27: 391 – 397.
- GEORGHIOU, G. P. (1980). Insecticide resistance and prospects for its management. *Residue Reviews*, 76: 131 – 145.
- HEMINGWAY, J. (1983). The genetics of Malathion resistance in *Anopheles stephensi* from Pakistan. *Transaction of Royal Society of Tropical Medicine and Hygiene*, 7: 106 – 108.
- HILL, N. (1985). The biochemistry of insecticide resistance in *Anopheles sacharovi*. Comparative studies with a range of insecticide susceptible and resistance *Anopheles* and *Culex* species. *Pesticide Biochemistry and Physiology*, 24: 68 – 76.
- KUMAR, R. (1984). *Insect pest control with special reference to African agriculture*. Edward Arnold Publishers. 180 pp.
- KRISTAN, M., FLEISCHMANN, H., DELLA TORRE, A., STICH, A. and CURTIS, C. F. (2003). Pyrethroid resistance/susceptibility and differential urban/rural distribution of *Anopheles arabiensis* and *An. gambiae* s.s. malaria vectors in Nigeria and Ghana. *Medical and Veterinary Entomology*, 17(3): 326 – 332.
- LOCKWOOD, J. A., SPARKS, T. C. and STORY, R. N. (1984). Evolution of insect resistance to insecticides: a reevaluation of the roles of physiology and behavior. *Bulletin of Entomological Society of America*, 30: 41 – 50.
- MALCOM, C. A. (1988). Current status of pyrethroids resistance in Anopheline. *Parasitology Today*, 4: 513 – 515.
- MARTE, M. A. (1986). Groundwater Exploitation and Drilling Technology in Arid Areas of Nigeria, Borno as a Case Study. Pages 141 – 173. In: *Proceedings of the First Annual Symposium and Training Workshop on Groundwater Resources in Nigeria*, University of Lagos, Nigeria.
- METCALF, R. L. (1980). Changing role of insecticide resistances in crop protection. *Annual Review of Entomology*, 25: 219 – 256.
- MILLER, J. E. and GIBSON, G. (1994). Behavioural response of host-seeking mosquitoes (Diptera: Culicidae) to insecticide-impregnated bed netting: a new approach to insecticide bioassays. *Journal of Medical Entomology*, 31: 114 – 122.
- MOLTA, N. B. and ALI A. (1998). Susceptibility of *Anopheles* species of northeastern Nigeria to Permethrin. *Entomological Society of Nigeria Occasional Publication*, 31: 101 – 107.
- NARAHASHI, T. (1976). Nerve membrane as a target of pyrethroids. *Pesticide Science*, 7: 267 – 272.
- ROBERTS, D. (1998). Overcrowding of *Culex sitiens* (Diptera: Culicidae) larvae: Population regulation by chemical factors or mechanical interference. *Journal of Medical Entomology*, 35(5): 665 – 669.
- ROZENDAL, J. A. (1997). *Vector control*. World Health Organization, Geneva, Switzerland
- SHAMAAN, N. A., HAMIDAH, R., JEFFRIES, J., HASHIM, A. J. and WAN NGAH, W. Z. (1993). Insecticide toxicity, glutathione transferases and carboxylesterase activities in the larva of the *Aedes* mosquito. *Comparative Biochemistry and Physiology Part C: Comparative Pharmacology*, 102 (1): 107 – 110.
- SNELLER, V. P. and DADD, R. H. (1977). Requirement for sugar in a chemically defined diet for *Aedes aegypti* larvae. *Journal of Experimental and Applied Entomology*, 14: 387 – 392.
- STATSDIRECT (2005). StatsDirect Ltd.Uk Version 4.2, 11 Gresham Way Cheshire, M33 3UY UK.
- VULULE, J. M., BEACH, R. F., ATIELI, F. K., ROBERTS, J. M., MOUNT, D. L. and MWANGI, R. W. (1994). Reduced susceptibility of *Anopheles gambiae* to permethrin associated with the use of permethrin-impregnated bed nets and curtains in Kenya. *Medical and Veterinary Entomology*, 8: 71 – 75.
- WHO (1980). Resistance of vectors of disease to pesticides: 5th report of expert committee on Vector biology and control. *World Health Organization Technical Report Series*, 655: 1 – 63.
- WHO (1986). Resistance in vectors and reservoirs of diseases to pesticides. *Bulletin of World Health Organization Technical Report Series*, 737: 1 – 88.
- WHO (1992). Vector resistance to pesticides. 15th report of the WHO Expert Committee on Vector Biology and Control. *World Health Organization Technical Report Series*, 818: 1 – 71.

## FOOD AND FEEDING HABITS OF *Campylomormyrus tamandua* IN ANAMBRA RIVER, NIGERIA

<sup>1</sup> NWANI, Christopher Didigwu, <sup>2</sup>EYO, Joseph Effiong and <sup>3</sup>UDEH, Emmanuel Fame

<sup>1</sup>Applied Biology Department, Ebonyi State University, Abakaliki. Ebonyi State, Nigeria

<sup>2</sup>Fisheries and Hydrobiology Research Unit, Department of Zoology, University of Nigeria Nsukka

<sup>3</sup>Department of Animal Production and Fisheries Management, Ebonyi State University, PMB 53, Abakaliki, Ebonyi State, Nigeria

**Corresponding Author:** NWANI. C.D., Applied Biology Department, Ebonyi State University, Abakaliki. Email: [nwaniCD@yahoo.com](mailto:nwaniCD@yahoo.com) Phone: 08037509910

### ABSTRACT

*The food and feeding habits of 417 samples of Campylomormyrus tamandua (Osteichthyes: Mormyridae) in Anambra River, Nigeria were studied from October 2002 to March 2004. Fish samples were collected monthly at Otuocha and Ogorugu river ports along the Anambra river using a fleet of gill nets of various mesh sizes, traps and hook and lines. Out of the eight (8) categories of food consumed, the most dominant group was benthic invertebrates (IFS = 44.92) followed by allochthonous invertebrates (IFS = 33.40) while the least was mud/sand (IFS = 10.02). Variation in the stomach fullness condition showed that 82 (19.66%) of the stomachs studied were empty, 40 (9.59%) were full while 295 (70.74%) were partially filled. Food richness and diet breadth showed no significant difference between the seasons and sex respectively ( $P > 0.05$ ). The trophic variations and flexibility in C. tamandua are discussed.*

**Keywords:** Anambra river, *Campylomormyrus tamandua*, Food, Feeding habits

### INTRODUCTION

*Campylomormyrus tamandua* is among the mormyrid species inhabiting fresh waters of tropical Africa including Anambra River (Lowe –McConnell, 1972). Popularly known as “Onu-Enyi” in the Anambra area, the fish is covered with small scales with a head, which is smooth and fleshy. Roberts (1975) attributes its success primarily to two adaptations, namely their electric organs important in nocturnal movement and communication and diversification of feeding habits. It is also a good specimen for neurological studies (Gosse, 1984). The fish is mostly favoured by the inhabitants of the study area probably because the flesh, though oily is quite tasty and of high flavour. The high oil content makes the fish difficult to dry but when patiently dried and stock-piled are transported to neighbouring towns and markets where they are sold. They are of high food value with price index ranging from two hundred naira (₦ 200:00) to three hundred naira (₦300.00) per kilogram (Olaose-bikan and Raji 1998). Only limited information exists on the biology of the fish especially the food and feeding habits its importance and potentials notwithstanding.

Imevbore and Bakere (1970) noted that *C. tamandua* feed almost exclusively on the larvae of bottom dwelling insect families such as larvae of chironomidae, ephemeropterae, ceratopogonidae, chaoboridae and trichoptera. Imevbore and Okpo (1972) also reported that the mormyrids of Kainji area feed on algae, zooplankton and mud/sand. Olatunde and Moneke (1985) reported that the diet of the mormyrid species in Zaria consist mainly of immature insects and some items of plant origin. Other reports on food and feeding habits of some

mormyrid species in Nigeria include Blake (1977), Hyslop (1986), King (1989), Tugels *et al* (1992), Ikomi (1996), Kouamelau *et al* 1999, 2000 and Nwani 1998, 2004.

Mormyrids especially *C. tamandua* are increasingly becoming important in the world aquarium business and aquaculture, thus, the need arises for better knowledge about the food and feeding habits. Knowledge from such studies would help in proper fish management and feed formulation.

### THE STUDY AREA

The Anambra River has its source from Ankpa highlands of Kogi State of Nigeria. It lies between latitude 6°10' and 7°20' and longitude 7°40' East of River Niger. There is a rainy season (April–September/October) and a dry season (October/November – March). From December to January, the basin is influenced by the harmatan but its effect is not well marked. The vegetation in the basin is guinea savanna but the lentic water bodies are often fringed with macrophytes like *Pterocarpus spp*, *Dalbergia spp*, *Jussiaea spp*, *Vossia cuspidate*, *Pennisetum spp*, *Cyodon spp* and in some areas *Raphia hookeri*. The people of the area are part time fishermen, traders and crop farmers. The farm produce include yam, cassava, rice, potato, vegetables, groundnuts, banana etc. Crop farming activities in the River basin go hand in hand with fisheries activities, which in turn are closely related to the flood regime. During the flood period when the water level becomes increasingly high, active farming becomes increasingly intensified. However, towards

the end of the flood regime, the above cycle alternates with the resumption of fishing activities, which get to the peak during the dry season.

## MATERIALS AND METHODS

Fish samples were collected monthly around Otuocha and Ogurugu river ports along the Anambra river from October 2002 to March 2004 using a fleet of gill nets (38.1 mm, 63.5 mm, 76.2 mm, 88.9 mm, 101.6 mm, 127.0 mm, and 177.8 mm), 20 traps and 200 hook and lines. Fish collected were preserved in ice and transported to the project laboratory of the Department of Zoology University of Nigeria Nsukka where the analysis was done. Fish collected were identified using the keys of Holden and Reed (1972), Lowe-McConnell (1972), Teugels *et al* (1992) and Olaosebikan and Raji (1988). The stomach of each fish was dissected out and slit open and its degree of fullness estimated by arbitrary 0–20 points scale thus 0, 5, 10, 15 and 20 points were representing empty, 1/4 full, 1/2 full and fully extended stomachs respectively. The percentage of partially filled stomachs (PS) i.e. (1/4 – 3/4 full) were used to evaluate patterns of feeding activity.

Stomach contents were sorted out into categories and analysed using relative frequency (RF) and percentage point (PP) methods (Hynes 1950, Hyslop 1980, King 1988). Thus

$\%RF = (a_i/n) \sum A_i = 1$ ; where  $a_i$  = frequency of item  $a$ ,  $A$  = frequency of the  $n$ th item (i.e. sum of all  $a_i$  values).

For the point scheme, each stomach was allotted 20 points regardless of the fish size and these were shared amongst the various categories of food taking into account their relative proportion by volume. The points gained by each food item in all stomachs examined were computed and expressed as a percentage of the total points of all food items. The %RF and %PP were then used to compute the index of food significance as follows:

$IFS = \%RF \times \%PP / \sum \%RF \times \%PP \times 100$ ; Where RF = relative frequency, PP = percentage point.  $IFS \geq 10$  was regarded as primary,  $IFS \geq 3$  but  $< 10$  as secondary whereas food with  $IFS < 3$  was incidental. The IFS data were used to compute diet breadth based on Shannon–Weiner function (H) as follows:  $(H) IFS = - \sum (n_i/N) \log_e (n_i/N)$ ; Where  $n_i$  = IFS of each food item,  $N$  = total IFS of all food items.

Food richness was defined as the number of food items in the diet (King, 1988). Food composition was analysed by students't-test. Differences were considered significant at 5% level of probability.

## RESULTS

**Variations in Stomach Fullness Condition:** The overall stomach fullness condition showed that out of the 417 samples of *C. tamandua* stomachs examined, 82 (19.66%) were empty, 40 (9.59) were full while 295 (70.74 %) were partially filled. Among the partially filled stomachs, 91 (21.82 %) were 1/4 full, 114 (27.34 %) were 1/2 full and 90 (21.58 %) were 3/4 full.

The monthly changes in stomach fullness condition (Figure 1) indicated that the peak of empty stomachs (ES) was in January. This month coincided with the lowest full stomach (FS) suggesting low feeding activity at this period. The peak of the partially filled stomachs (PS) was recorded in September. With respect to seasonal variation in stomach fullness condition (Figure 2), empty and 1/4 full stomachs were dominant during the dry season while 1/2 full and 3/4 full stomachs were dominant during the wet season. There was a significant dry season increase in empty stomach ( $d = 4.06$ ,  $P < 0.05$ ) and 1/4 full stomach ( $d = 3.86$ ,  $P < 0.05$ ) while the 1/2 full and 3/4 full stomachs were significantly higher in the rainy than in the dry season ( $P < 0.05$ ).

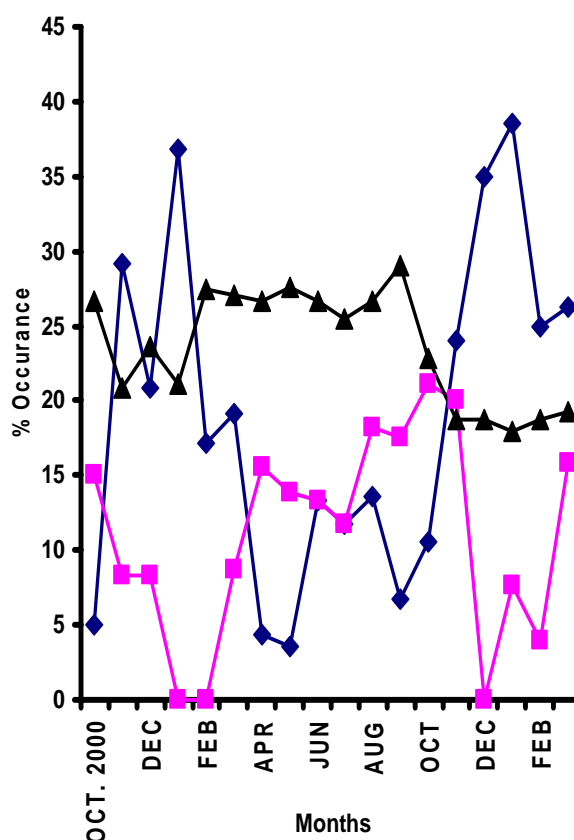
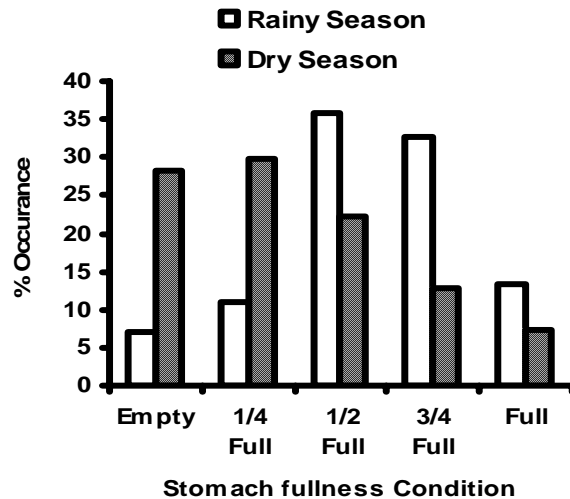


Figure 1: Monthly variations in feeding intensity among *Campylomormyrus* species of Anambra river, Nigeria

**Diet Composition:** Twenty-five food items were recorded in the diet of *C. tamandua* (Table 1). Lepidopteran larvae contributed the highest value (10.00 %) in terms of percentage relative frequency (%RF). This was followed by fine particulate organic matter (9.02 % RF) and mud/sand (8.80 % RF). The lowest value (0.01 % RF) was recorded in the ephemeropteran larvae. Considering the percentage point (PP), lepidopteran larvae contributed the highest value (13.30 % RF) followed by formicidae



**Figure 2: Variation in indices of feeding activities in *Campylomormyrus tamandua* of Anambra river, Nigeria**

**Table 1: Trophic spectrum of the diet of all sizes of *Campylomormyrus tamandua* in Anambra River**

Dietaries	%RF	%PP	IFS
<b>Algae:</b> Filamentous Algae	8.60	5.60	6.95
Colonial Algae	4.02	2.00	1.16
Unicellular Algae: Diatoms	2.02	2.04	0.59
Desmids	2.70	1.50	0.58
Euglenids	0.02	0.04	0.29
<b>Benthic invertebrates</b>			
Diptera: Chironomid larvae	6.05	8.50	7.42
Chironomid pupae	4.04	3.03	1.77
Unid diptera larvae	7.70	10.86	12.07
Odonata Anisoptera nymph	2.60	2.40	0.97
Ephemeropteran larvae	0.01	0.02	0.40
Trichoptera larvae	4.26	6.04	3.31
Crustacean: Ostracoda	0.02	0.03	0.97
Arachnida: Hydracarina	0.03	0.80	0.40
<b>Allochthonous invertebrates</b>			
Hymenoptera:		12.02	13.00
Formicidae imagines	8.03	13.30	19.19
Lepidopteran larvae	10.00	1.50	0.28
Diplopoda polydesmida	1.31	2.60	0.76
<b>Miscellaneous invertebrates</b>	4.70		
<b>Zooplankton</b>			
Crustacea cyclop copepods	1.40	0.60	0.12
Cladocera –Bosmina	0.07	0.04	0.16
Rotifera –Keratella	2.00	0.30	0.10
<b>Macrophyte material:</b>			
Leaf fragments	2.80	2.00	0.41
Seeds	3.00	1.32	0.57
<b>Detritus</b>			
Coarse particulate organic matter	6.53	6.25	5.60
Fine particulate organic matter	9.02	10.11	13.00
<b>Mud/sand</b>	8.80	7.10	10.02

imagines (12.02 % PP) and unidentified dipteran larvae (10.86 % PP). The lowest value (0.02 % PP) was recorded in the ephemeropteran larvae. Considering the value for the index of food significance (IFS) of each food group, the benthic invertebrates were the most dominant food group (IFS = 44.92 %) followed by allochthonous

invertebrates (IFS = 33.40 %) and detritus (IFS = 18.60 %). The lowest value (IFS = 10.02 %) was recorded in the mud/sand. Food of primary importance (IFS >10) was fine particulate organic matter, unidentified dipteran larvae, formicidae imagines, lepidopteran larvae and mud/sand. Other foods of secondary importance (IFS < 10 but ≥ 3) were filamentous algae, coarse particulate organic matter, chironomid larvae and trichopteran larvae. Other food items were of minor importance (IFS < 3).

**Variation of Diet with Season:** The seasonal changes in the index of food significance (IFS) Table 2 indicated that the IFS of anisopteran nymph and fine particulate organic matter were significantly higher in the dry than wet season ( $P < 0.05$ ). The IFS of chironomid larvae, ephemeropteran larvae, leaf fragments and seeds were significantly higher in the rains than in the dry season ( $P < 0.05$ ). No significant seasonality difference was observed in the IFS for other food items. Food of primary importance in the dry season was formicidae imagines while unidentified dipteran larvae were important in the rainy season.

**Table 2: Seasonal variation in the IFS of *Campylomormyrus tamandua* in the Anambra River system**

Dietaries	DRY	RAINY	P*
<b>Algae:</b>			
Filamentous Algae	0.965	1.538	NS
Colonial Algae	1.351	-	
Unicellular Algae: Diatoms	0.998	1.349	NS
Desmids	0.487	1.288	NS
Euglenids	1.527	1.710	NS
<b>Benthic invertebrates</b>			
Diptera: Chironomid larvae	6.902	9.265	<0.05
Chironomid pupae	5.776	5.333	NS
Unid diptera larvae	9.620	13.499	<0.05
Odonata Anisoptera nymph	4.721	2.425	<0.05
Ephemeropteran larvae	0.381	0.658	NS
Trichoptera larvae	6.089	6.666	NS
Crustacean: Ostracoda	0.089	-	
Arachnida: Hydracarina	1.545	1.893	NS
<b>Allochthonous invertebrates</b>			
Hymenoptera:			
Formicidae imagines	10.059	9.756	NS
Lepidopteran larvae	7.850	6.651	NS
Diplopoda polydesmida	1.475	1.561	NS
<b>Miscellaneous invertebrates</b>	6.357	4.230	NS
<b>Zooplankton:</b>			
Crustacea cyclop copepods	2.017	1.114	NS
Cladocera –Bosmina	0.918	0.866	NS
Rotifera –Keratella	0.922	0.477	NS
<b>Macrophyte materials:</b>			
Leaf fragments	3.630	9.389	<0.05
Seeds	4.484	7.922	<0.05
<b>Detritus</b>			
Coarse particulate organic matter	6.487	4.614	NS
Fine particulate organic matter	6.861	3.386	<0.05
<b>Mud/sand</b>	8.579	4.401	
<b>Food richness</b>	25	23	
<b>Diet breadth</b>	2.88	2.81	

\* = Probability @ 0.05 NS = No significant difference.

**Variation of Diet with Sex:** The IFS of female was higher in five (5) food items (filamentous algae, colonial algae, chironomid larvae, formicidae imagines and coarse organic matter) and lower in three (Dipteran larvae, Lepidopteran) larvae and fine particulate organic matter) than males ( $P < 0.05$ ) (Table 3). Other food items were not significantly different between the sexes. Food richness and diet breadth were slightly higher in females than males. Three food items (fine particulate organic matter, unidentified dipteran larvae and formicidae imagines) were of primary importance in both males and females.

**Table 3: Sex dependent variation in IFS of *C. tamandua* in Anambra**

Dietaries	Male	Female	P*
<b>Algae:</b> Filamentous Algae	2.91	6.49	<0.05
Colonial Algae	1.42	1.44	<0.05
<b>Unicellular Algae:</b>			
Diatoms	0.37	0.79	NS
Desmids	0.58	0.90	NS
Euglenids	0.04	-	
<b>Benthic invertebrates</b>			
Diptera: Chironomid larvae	6.01	9.35	<0.05
Chironomid pupae	1.64	2.20	NS
Unid diptera larvae	14.00	10.13	<0.05
Odonata Anisoptera nymph	0.93	1.01	NS
Ephemeropteran	-	0.08	
larvae	3.50	4.16	NS
Trichoptera larvae	0.08	0.32	NS
Crustacean: Ostracoda	-	0.46	
Arachnida: Hydracarina			
<b>Allochthonous invertebrates</b>	10.48	14.01	<0.05
Hymenoptera: Formicidae	20.76	8.92	<0.05
imagines	0.76	1.37	NS
Lepidopteran larvae	3.80	4.30	NS
Diplopoda polydesmida			
<b>Miscellaneous invertebrates</b>			
<b>Zooplankton</b>			
Crustacea cyclopod copepods	-	0.35	
Cladocera	2.76	-	
Bosmina	0.38	0.37	NS
Rotifera -Keratella			
<b>Macrophyte material:</b>			
Leaf fragments	0.76	1.04	NS
Seeds	0.73	0.86	NS
<b>Detritus:</b>			
Coarse particulate organic matter	5.29	8.74	<0.05
Fine particulate organic matter	17.6	12.90	<0.05
<b>Mud/sand</b>	5.64	9.81	10.02
Food richness	22	24	
Diet breadth	2.43	2.60	
No examined	235	182	
No with food	190	145	

\* Probability @ 0.05, NS = No significant difference

## DISCUSSION

The result of this finding shows that Benthic invertebrates (IFS = 44.92%) were the most dominant food group in the diet of *C. tamandua*. Ezenwaji and Inyang (1998) and Ezenwaji (1999) had in agreement with this finding reported that autochthonous and allochthonous insects constituted important proportion of food of many fish species

inhabiting the Anambra river system. Olaosebikan and Raji (1998) also reported similar results on *Mormyrus rume*, *Hyperopisus bebe* and *Gnathonemus petersii* in lower Niger basin. In a similar report, King (1989) noted the high preponderance of chironomid larvae in the diet of *Brienomyrus brachyistius* in a Nigerian rainforest stream. Teugels *et al* (1992) also reported high occurrence of benthic invertebrates and such common inclusions as Zooplankton, terrestrial invertebrates, plant materials, mud and sand in the diet of *B. brachyistius*, *G. petersii*, *Isichthys henri* and *Petrocephalus ansorgi*.

The discovery of the allochthonous invertebrates such as Formicidae imagines, Lepidopteran larvae and polydesmids in the stomach of *C. tamandua* indicates some degree of surface feeding. Mud and sand were also picked from the bottom. It thus appears that *C. tamandua* is able to exploit all food niches (bottom, mid-water and water surface) in its habitats. It thus exhibits wide plasticity (i.e. high trophic flexibility) in its feeding behaviours. This report agrees with the findings of Ezenwaji and Offiah (2003) that reported high trophic flexibility in *Pellonula leonensis* in Anambra River. Nwani (2004) also obtained similar results for *Hyperopisus bebe* in Anambra River. The seasonality in the dietaries of *C. tamandua* indicated that food richness and feeding intensity were higher in the rainy season than dry season. This finding deviated from the optimal foraging theory (King, 1989), which stated that diet breadth expands during the time of scarcity and contracts during the period of plenty. This reports however is in consonance with the reports of Lowe – McConnell (1972) and Welcomme (1979, 1985) that many tropical fresh water fishes have a broader trophic spectrum during the rainy (flood season). This result is also in agreement with the report of Ezenwaji (1999) who attributed a higher food richness and diet breadth of *Clarias albopunctatus* during the rainy season to increased availability of food resource.

## REFERENCES

- BLAKE, B. F. (1977). Food and feeding of the mormyrid fishes of Lake Kainji, Nigeria, with special reference to seasonal variation and interspecific differences. *Journal of Fish Biology*, 11: 315 – 328.
- EZENWAJI, H. M. G. (1999). The abundance and trophic biology of *Clarias albopunctatus* Nichols and LaMonte, 1953 Osteichthyes: Clariidae) in a tropical flood river basin. *Hydrobiologia*, 392: 159 –168.
- EZENWAJI, H. M. G. and INYANG, N. M. (1998). Observation on the biology of *Clarias agboyiensis* Syndaham, 1980. (Osteichthyes: Clariidae) in the Anambra flood river system, Nigeria. *Fisheries Research*, 36: 47 –60.
- EZENWAJI, H. M. G. and OFFIAH, F. N. (2003). The biology of *Pellonula leonensis* Boulenger, 1916 (Osteichthyes: Clariidae) in Anambra river Nigeria. *Journal of Biological Research and Biotechnology* 1(2): 33 – 50.



- GOSSE, J. P. (1984). Mormyridae. Pages 63 – 122. In: DAGET, J., GOSSE, J. P. and THYS VAN DEN AUDENAERDE, D. F. E. (Eds). *Checklist of freshwater fishes of Africa (CLOFFA)*. ORSTOM, Paris and MRAC Tervuren.
- HOLDEN, M. J. and REED, W. (1972). *West African fresh water fish*. Longman, London, 63 pp.
- HYNES, H. B. N. (1950). The food of fresh water sticklebacks (*Gasterostes aculeatus* and *Pygostes pungitius*) with a review of methods used in studies of the food of fishes. *Journal of Animal Ecology*, 19: 36 – 58.
- HYSLOP, E. J. (1980). Stomach contents analysis: a review of methods and their application. *Journal of Fish Biology*, 17: 411 – 429.
- HYSLOP, E. J. (1986). The food habits of four small sized species of Mormyridae from the flood plain pools of Sokoto river basin. *Journal of Fish Biology*, 28: 147 – 151.
- IKOMI, R. B. (1996). Studies on the growth pattern, feeding habits and reproductive characteristics of the Mormyrid *Brienomyrus longianalis* (Boulenger 1901) in the upper Warri River, Nigeria. *Fisheries Research*, 26: 187 – 198.
- IMEVBORE, A. M. A. and BAKERE, O. (1970). The food and feeding habits of non-cichlid fishes of the River Niger in the Kainji reservoir area. *Kainji Lake Studies 1. (Ecology)*. 47 (S. A. VISSEV Ed.) Niger: Ibadan University Press.
- IMEVBORE, A. M. A. and OKPO, W. S. (1972). Aspects of the biology of Kainji Lake fishes. *The ecology of Lake Kainji, Limnology*, 1: 163 – 178.
- KING, R. P. (1988). Observation on *Liza falcipinnis* (Valenciennes) in Bonny River Nigeria. *Review of Hydrobiology in the Tropics*, 2(1): 63 – 70.
- KING, R. P. (1989). Distribution, abundance, size and feeding habits of *Brienomyrus brachyistius* (Gil 1862). Teleostei: Mormyridae in a Nigerian rainforest stream. *Cybiurn*, 13(1): 25 – 36.
- KOUAMELAN, P. E., TEUGEL, G. G., OLLEVIER, F. and THYS VAN DE AUDENAERDE, D. F. E. (1999). The effect of a man-made lake on the diet of the African electric fish *Mormyrus rume* (Valenciennes, 1846, Osteoglossiformes: Mormyridae). *Hydrobiologia*, 380: 141 – 151.
- KOUAMELAN, P. E., TEUGEL, G. G., OLLEVIER, F. and THYS VAN DE AUDENAERDE, D. F. E. (2000). Feeding habits of *Mormyrops anguilloides* (Mormyridae) in Lacustrine and riverine habit of a West African Basin. *Cybiurn*, 2(1): 69 – 79.
- LOWE-MCCONNELL, R. H. (1972). *Fresh water fishes of the Volta and Kainji Lakes*. Ghana University Press, Accra, 284 pp.
- NWANI, C. D. (1998). *Aspects of the biology of Distichodus species in Anambra River, Nigeria*. M.Sc. Project Report, University of Nigeria, Nsukka, 133 pp.
- NWANI, C. D. (2004). *Aspects of the biology of Mormyrids (Osteichythes: Mormyridae) in Anambra river, Nigeria*. PhD Thesis, University of Nigeria, Nsukka, 194 pp.
- OLAOSBIKAN, B. D. and RAJI, A. (1998). *Field guide to Nigerian fresh water fishes*. Federal College of Fresh Water Fisheries Technology, New Bussa, 106 pp.
- OLATUNDE, A. A. and MONEKE, C. C. (1985). The food habits of four Mormyrid species in Zaria, Nigeria. *Arch hydrobiologia*, 102: 502 – 517.
- ROBERTS, R. J. (1975). *Fish pathology*. Bailliere Tiddall, London, 318 pp.
- TEUGELS, G. G., MCGREID, G. and KING, R. P. (1992). *Fishes of the Cross River Basin (Cameroon –Nigeria): Taxonomy, Zoogeography, Ecology and conservation*. Muse, Royale de l' Afrique Centrale, Tervuren Belgium. *Annals of Science (Zoology)*, 182 pp.
- WELCOME, R. L. (1979). *Fisheries ecology of flood plain rivers*. Longmans, London. 167 pp.
- WELCOME, R. L. (1985). River fisheries. *FAO Fishery Technical Paper*, 262: 1 – 330.

## NEURAL TISSUE AND COMPLETE REGENERATION OF THE TAIL OF THE GEKKONID LIZARD, *Hemidactylus flaviviridis*

NDUKUBA, Patrick Ifeanyiichukwu

Division of Endocrine and Environmental Physiology, Department of Animal and Environmental Biology, Abia State University, PMB 2000, Uturu, Nigeria. E-mail: [ndukuba\\_pi@yahoo.com](mailto:ndukuba_pi@yahoo.com) Phone: 08053569774

### ABSTRACT

*Tails of three groups of the Gekkonid lizard, Hemidactylus flaviviridis, were amputated (group I) or autotomized (groups II and III). The animals were exposed to 12 hours of light and 12 hours of darkness. In group I experiment, previously regenerated tails were amputated (repeated autotomy RA) with a pair of sharp scissors, after anesthetizing the animals with ice cubes, at point equivalent to three autotomy segments. The original planes of autotomy have been replaced by ependymal tubes and there were no blood exudates. In group II, the spinal cord at the local site of autotomy was carefully removed (spinal cord removed, SCR), with dissecting instruments, for a length equal to one autotomy segment. Lizards in group III served as controls (Normal Lizard NL). The results show that the initiation of regeneration, the growth rate, the total length of new growth (regenerate) produced, and the total percentage replacement of the lost (amputated/autotomized) tails 30 days after excision were all significantly less in lizards of group II, ( $p < 0.01$ ) and insignificantly less in group I lizards, when compared with the controls (group III). The results show that for complete regeneration of the lizard tail neural tissue must be present.*

**Keywords:** Neural tissue, Regeneration, Tail, Gekkonid lizard, *Hemidactylus flaviviridis*

### INTRODUCTION

With the exception of most tissues, many lower vertebrates are capable of replacing amputated limbs, a process that invariably involves, and is usually dependent upon, the regeneration of the severed spinal cord (Goss, 1964). However, the completeness of cord regeneration varies from one form to another. In the larval lamprey, and in both larval and adult urodele amphibians, the regenerating tail or limb is complete, including the differentiation of new neurons and the production of paired spinal ganglia in each segment (Goss, 1969). In contrast, the spinal cord of larval amphibians and lizards regenerate little more than their ependymal tubes accompanied by elongating nerve fibres; new neurons do not differentiate (Goss, 1969).

Cavanaugh (1951) demonstrated that if some of the cells in a spinal ganglion of a rat are destroyed, as a result of cutting their peripheral fibres, the remaining ones which must grow back into the entire peripheral field undergo enlargement. Considerable evidence suggests that a comparable situation prevails in the regenerating lizard tail (Panness, 1962). This author reported that no new spinal ganglia are differentiated in the process of lacertilian tail regenerating, the regenerate itself becomes innervated by fibres derived from the three pairs of sensory ganglia immediately anterior to the level of amputation or autotomy.

The role of ependyma in the initiation of regeneration and cartilage differentiation has been well documented (Simpson, 1964). The present study attempts to elucidate the effects of repeated amputation and the removal of the spinal cord from the local site of autotomy on tail regeneration in *Hemidactylus flaviviridis*.

### MATERIALS AND METHODS

Adult *H. flaviviridis* of both sexes measuring  $6 \pm 2$  cm ( $\pm$  S. D. snout - vent length) weighing  $10 \pm 5$  gm ( $\pm$  S. D. body mass) were obtained from a local supplier and maintained *ad libitum* on a diet of cockroaches and grasshoppers for a period of 7 days for acclimation to laboratory conditions. 90 lizards were used for the investigation. They were divided into three groups of 10 lizards each and exposed to the normal photoperiod (12 hours of light and 12 hours of darkness).

All the lizards were immobilized with ice cubes at  $4^\circ\text{C}$  before autotomy or amputation was performed. Ten *H. flaviviridis* with their tails fully regenerated previously, thus lacking planes of autotomy, were amputated with a pair of sharp scissors at points equivalent to three autotomy segments from the vent. This forms the group I lizards. In group II, ten *H. flaviviridis*, with normal planes of autotomy were autotomized at the third segment from the vent. The spinal cord at the local site of autotomy was carefully removed with dissecting instruments in order to study the possible influence of the distal spinal cord on tail regeneration in *H. flaviviridis*. In group III, ten *H. flaviviridis* were autotomized at the third segment from the vent. This group of lizards served as the control and was exposed to the same light and ambient temperature conditions. All the lizards were fed *ad libitum* inside their respective cages. The experimental set up was replicated thrice.

The length of new growth (regenerate) was measured in mm, with a graduated meter rule at fixed time intervals of 10, 15, 20, 25, and 30 days post tail amputation or autotomy. This investigation was conducted during the harmattan months (December – March). The recorded average monthly

ambient and cage temperatures are given in Table 1. The data on the length of tail regenerated and the percentage replacement were subjected to an analysis of variance and mean separation using New Duncan's multiple range test at 0.05 and 0.01 probabilities (Duncan, 1955)

**Table 1: Average ambient temperature and cage temperatures during the Harmattan season**

Months	Ambient Temperature	Cage Temperature
January	30 – 31 °C	31- 32 °C
February	31 - 32 °C	32 –33 °C
March	32 - 33 °C	33 - 34 °C
April	33 - 34 °C	34 –35 °C
Average	32 – 32.5 °C	32.50 – 33.5 °C

## RESULTS

**Growth Rate and Total Length Regenerated and Total Percentage Replacement:** A measurable growth occurred in normal (NL) group (Group III) of animals by day 3 while in repeated amputation (RA) group (Group I) and spinal cord removed (SCR) group (Group II) lizards, measurable growth occurred by day 9 and 10, respectively (Table 2). There was also a delay in the appearance of the regeneration blastema in RA when compared to normal *H. flaviviridis* (Table 3).

**Effect of Repeated Amputation (RA):** The total lengths of tails regenerated as well as the total percentage replacement of the RA lizards were less when compared with the controls (Figure 1 and 2). RA *H. flaviviridis* produced an average tail length of 19.18 mm out of a total amputated length of 40.0 mm, accounting for a replacement rate of 49.5% (Table 3)

**Effect of Removal Spinal Cord (SCR) from the Local Site of Autotomy:** The results showed that the initiation of regeneration, the total length of tail regenerated and the total percentage replacement of the lost (autotomized) tails on day 30 post autotomy were all significantly different in SCR group of *H. flaviviridis*, when compared with those of the controls ( $P < 0.01$ ). SCR animals produced an average tail replacement of 9.0 mm out of a total autotomized length of 40.0 mm, accounting for a replacement rate of 22.5% (Table 3).

**Normal Lizards (NL):** On day 30 after tail autotomy, the normal lizards produced an average tail replacement of 2.2 mm out of the total autotomized length of 41.0 mm, accounting a replacement rate of 51.7% (Table 3).

Comparison of the total length of tail regenerated and the total percentage replacement between the three groups of *H. flaviviridis* (ANOVA and Duncan's multiple range test) revealed no statistically significant difference between the RA and the controls (Figures 1 and 2). However, all other

comparisons between the controls and SCR group of lizards on one hand, and between RA and SCR on the other hand, were significantly different at 5% level (Duncan, 1955).

## DISCUSSION

Several lines of evidence suggest that the central nervous system (CNS) and its associated endocrine organs play pivotal roles in vertebrate appendage regeneration (Singer and Salpeter, 1961; Tassava *et al.*, 1987; Goldharmer, 1988). As reviewed by Wallace (1981), the importance of the wound epithelium to amphibian limb regeneration was demonstrated by several successful experiments in which the formation or function of the wound epithelium was inhibited. Pannese (1962) has shown that in the regenerating lizard tail, no new spinal ganglia were differentiated; the regenerate itself became innervated by fibers derived from the three pairs of sensory ganglia immediately anterior to the level of amputation/autotomy.

In a similar series of investigations, Ndukuba and Ramachandran (1988, 1989) and Ramachandran and Ndukuba (1989 a,b) had earlier demonstrated the influence of both intrinsic and extrinsic factors on tail regeneration in *H. flaviviridis*. These authors had shown that there was a positive influence of increasing photoperiodism as well as intensity on lacertilian tail regeneration and a negative influence of decreasing lengths of light from the intermediate photoperiod regimen of 12 hours of light and 12 hours of darkness. Lizards under continuous illumination produced the best regenerative performance while those in continuous (total) darkness produced the worst performance.

When *H. flaviviridis* were blinded by surgical removal of the two lateral eyes, the regenerative process was unaffected as blinded lizards regenerated their lost tails like their sighted counterparts exposed to the same experimental photoperiodic conditions (Ndukuba and Ramachandran, 1988). However, pinealectomy produced a 50% retardation effect in lizards exposed to continuous light, suggesting the involvement of the pineal organ of *H. flaviviridis* in photoperiodic photoreception (Ramachandran and Ndukuba, 1989).

Furthermore, the administration of exogenous prolactin (PRL) enhanced both the length of new tail and the percentage replacement in unoperated lizards exposed to continuous darkness, but did not affect their pinealectomized counterparts, indicating a more intriguing, interdependent interaction among photoperiodism, pineal, and PRL (Ndukuba and Ramachandran 1989). The influence of temperature and seasonal variations on lacertilian tail regeneration showed that the best regenerative performance was obtained during the summer months (temperature: 30 °C) and the worst performance in the winter months (temperature: 17 °C) with the regenerative performance during the monsoon season (temperature: 26 °C) falling in between. (Ramachandran and Ndukuba, 1989).

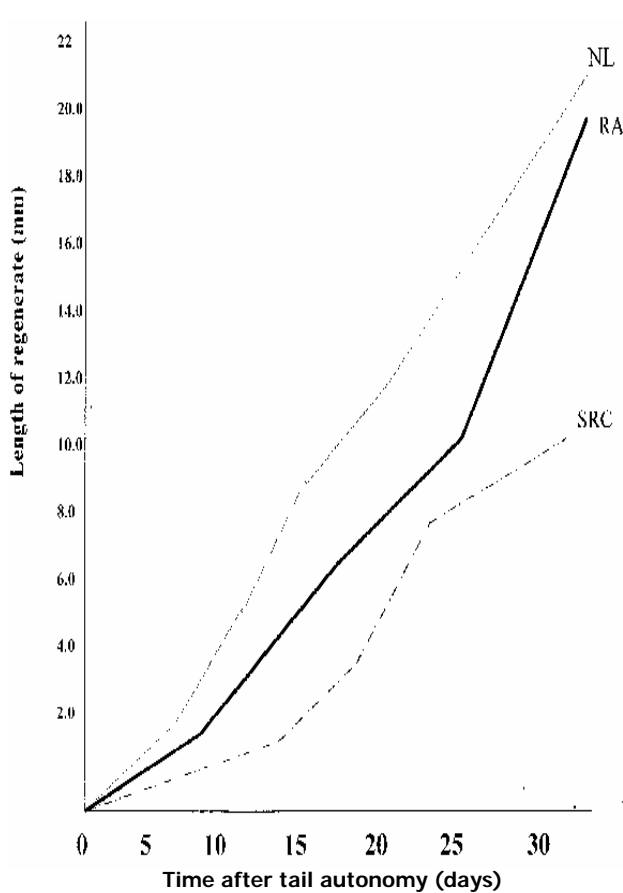
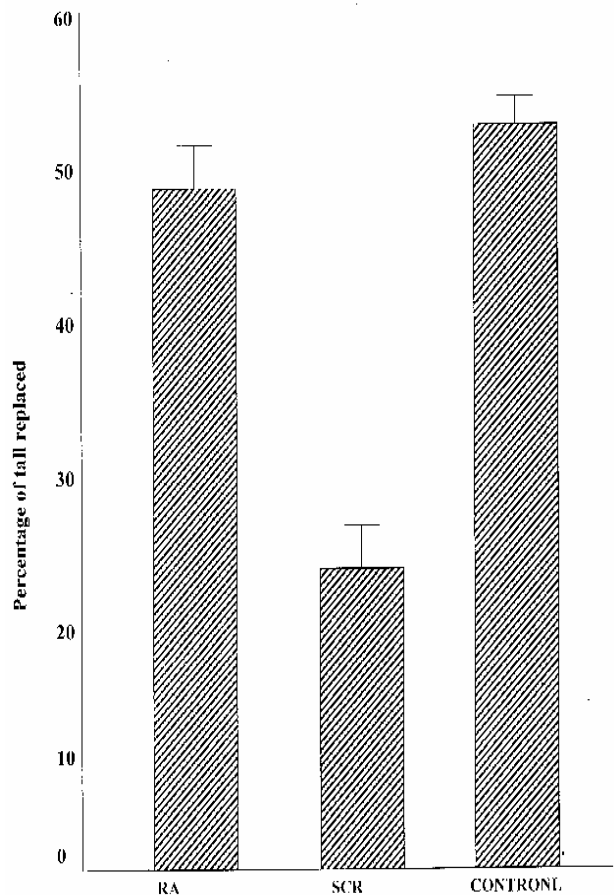
**Table 2: Approximate number of days taken to reach various arbitrary stages of tail regeneration in the house lizard, *Hemidactylus flaviviridis***

Experiment Animals	Wound Healing	Blastema	Early Differentiation	Mid Differentiation	Late Differentiation	Growth <sup>a</sup>
Experiment I (Repeated amputation)RA	1	3	5	6	8	30 <sup>b</sup>
Experiment II (Spinal cord removed) SCR	3	5	8	10	12	30
Experiment III (Normal Lizards (NL)	1	3	5	6	8	30

*a* = Stages of tail regeneration, *b* = Total number of days after tail autonomy, RA = Repeated amputation, SCR = Spinal cord removed, NL = Normal lizards, Average daily room and cage temperature =  $32 \pm 1^\circ\text{C}$ .

**Table 3: Length of tail regenerated and percentage replacement in three groups of the Gekkonid lizard, *H. flaviviridis***

Group of Lizards	Length of tail removed	Length of tail regenerated	Percentage replacement *
1. Repeated autotomy (RA)	40 mm	19.8 mm	49.5 %
2. Spinal cord removed (SCR)	40 mm	9.0 mm	22.5 %
3. Normal Lizard (NL)	40 mm	21.2 mm	51.7 %

**Figure 1: Length of tail regenerated at the end of 30 days in Normal Lizards (NL), repeated amputation (RA) and spinal cord removed (SCR) *H. flaviviridis*****Figure 2: Repeated amputation (RA) , Spinal Cord removed (SCR) and normal lizards (NL) *H. flaviviridis*. Vertical lines are  $\pm$  S. D. (N = 30)**

Results of the present study showed that tail regeneration in Gekkonid lizard, *H. flaviviridis*, was inhibited by the removal of the spinal cord from the local site of autotomy, but insignificantly retarded when previously regenerated tails were amputated for a second regrowth as compared with the controls. Since according to Goss (1969) lacertilian tails do not regenerate new spinal cords; in their place are regenerated ependymal tubes accompanied by elongating nerve fibers, it could be concussively surmised that the excellent regenerative performance by previously regenerated tails may be due to a possible influence by the ependymal tubes, and their accompanying elongated nerve fibers. Simpson (1964) demonstrated the role of ependyma in the initiation of regeneration and cartilage differentiation in the lizard, *Lygosoma laterale*.

The active participation of the distal spinal cord on tail regeneration in *H. flaviviridis* was evident by a 60 % retardation effect on lizards deprived of their spinal cords at the local sites of autotomy. This finding suggests that for complete tail regeneration in *H. flaviviridis* neural tissue must be present. It further strengthens our earlier report (Ndukuba and Ramachandran, 1989; Ramachandran and Ndukuba, 1989) that 50 % tail replacement is an innate ability which is independent of photoperiodism and associated neuroendocrine mechanism and apparently, occurs under basal level of prolactin secretion.

#### ACKNOWLEDGEMENT

The Author wishes to acknowledge the facilities provided by the Abia State University Sponsored Departmental Research in Animal and Environmental Biology

#### REFERENCES

- CAVANAUGH, M. W. (1951). Quantitative effects of the peripheral innervation area on nerves and spinal ganglion cells *Journal of Comparative Neurology*, 94: 181
- DUNCAN, D. B. (1955). Multiple range and multiple F test. *Biometrics*, 11: 1 – 42.
- GOLDHAMER, D. J. (1988). Analysis of cell proliferation and the roles of nerves and wound epithelium during forelimb regeneration in the adult newt, *Notophthalmus viridescens*. Ph.D. Dissertation, The Ohio State University Ohio, USA.
- GOSS, R. J. (1964). *Adaptive Growth*. Academic Press, New York.
- GOSS, R. J. (1969). *Principles of Regeneration*. Academic Press, New York.
- NDUKUBA, P. I. and RAMACHANDRAN, A. V. (1988). Extratestinal photocereception in lacertilian tail regeneration: the lateral eyes are not involved in photoperiodic photoreception in the gekkonid lizard, *Hemidactylus flaviviridis*. *Journal of Experimental Zoology*. 248: 73 –80.
- NDUKUBA, P. I. and RAMACHANDRAN, V. V. (1989). Is the pineal involved in the stimulatory influence of prolactin on tail regeneration in lizards? Studies with exogenous prolactin in lizards exposed to continuous darkness. *General and Comparative Endocrinology*, 76: 192-199.
- PANNESE, E. (1962). Detection of neurofilaments in the perikaryon of hypertrophic nerve cells. *Journal of Cell Biology*. 13: 457-461.
- RAMACHANDRAN, A. V. and NDUKUBA, P. I. (1989a). Preliminary evidence for pineal mediated extraretinal photoreception in relation to tail regeneration in gekkonid lizard, *Hemidactylus flaviviridis*. *Journal Pineal Research* 6: 121-134.
- RAMACHANDRAN, A. V. and NDUKUBA P. I. (1989b). Tail regeneration in normal, blinded and pinealectomized gekkonid lizards, *Hemidactylus flaviviridis*, exposed to four different light conditions during three seasons (temperatures). *Acta Zoologica*, 70: 205-210.
- SIMPSON, S. B. (1964). Analysis of tail regeneration in the lizard, *Lygosoma laterale*. I. Initiation of regeneration and cartilage differentiation: the role of ependyma *Journal Morphology*, 114: 425-436.
- SINGER, M. and SALPETER, M. M. (1961). Regeneration in vertebrates: the role of the wound epithelium. In "Growth in living systems", M. X. Zarrow. ed., Basic Books Inc., New York. 315 pp
- TASSAVA, R. A., COLDHAMER, D. J. and TOMLINSON, B. L. (1987). Cell cycle controls and the role of nerves and the regenerate epithelium in urodele forelimb regeneration: possible modification of basic concepts. *Biochemical Cell Biol.*, 65: 739-749.
- WALLAGE, H. (1981). "Vertebrate Limb Regeneration" Willey, New York. 200 pp



## PREVALENCE OF CAPRINE STRONGYLE INFECTION AND THE DIAGNOSTIC EFFICACY OF SOME MEDIA FOR FAECAL CULTURE AND NEMATODE LARVAL RECOVERY FROM GOAT FAECES

NWOSU, Chukwunyere Okwudiri<sup>1</sup>, IWUOHA, Chimaobi Lawrence<sup>2</sup>, TORRU, Churchill<sup>2</sup> and MOHAMMED, Ali<sup>1</sup>

<sup>1</sup>Department of Veterinary Microbiology and Parasitology, University of Maiduguri, P.M.B. 1069 Maiduguri, Borno State, Nigeria

<sup>2</sup>Department of Biological Sciences, University of Maiduguri, P.M.B. 1069 Maiduguri, Borno State, Nigeria.

**Corresponding Author:** NWOSU, C. O., Department of Veterinary Microbiology and Parasitology, University of Maiduguri, P. M. B. 1069 Maiduguri, Borno State, Nigeria. Phone: (+234) 0803-4535510. E-mail: [abiamaiduguri@yahoo.com](mailto:abiamaiduguri@yahoo.com)

### ABSTRACT

*The prevalence of caprine strongyle infections and the diagnostic efficacy of some culture media in supporting the recovery of strongyle larvae were evaluated using 840 faecal samples collected from goats during slaughter at Maiduguri Metropolitan abattoir. Faecal examination conducted by the modified McMaster technique revealed that out of 840 goats examined, 708 (83.8 %) shedded strongyle eggs in their faeces. The prevalence of infection was significantly ( $P < 0.05$ ) higher among female, young and diarrhoeic goats than their corresponding male, adult and non-diarrheic counterparts. Faecal culture and larval recovery using the test tube filter paper technique revealed that the direct culture of faecal samples without any additional culture medium supported the recovery of the largest number of nematode infective larvae from the faeces. When this was used as a standard (100% egg hatch or 0% reduction in egg hatch), larval recovery was highest ( $P < 0.05$ ) from goat faeces (98.4 %) followed respectively by sheep faeces (57.7 %), cow faeces (52.4 %), horse faeces (42.3 %) and soil (18.6 %) as culture media. The results therefore indicate the superior diagnostic quality of goat faeces as a culture medium for the recovery of infective nematode stages in goat faeces.*

**Keywords:** Strongyle infection, Faecal culture media, Culture media Efficacy, Goat.

### INTRODUCTION

Among the domesticated small ruminants in Nigeria, goats are much more important in meat production and research purposes than sheep due to their larger population and the wider acceptability of goat meat by the people (ILCA, 1979; Omeke, 1988). However, gastrointestinal helminthiasis, especially parasitic gastroenteritis (PGE) is a major health problem and a serious constraint on the production of small ruminants in Nigeria as a result of the associated morbidity, mortality and cost of treatment and control (Schillhorn van Veen, 1973; Akerejola *et al.*, 1979). In Nigeria, about 20 % of the national goat population die or are slaughtered *in extremis* annually due to helminthiasis (Kuil, 1969). PGE is a complex of diseases contributed to by several nematodes in which *Haemonchus*, *Trichostrongylus*, *Oesophagostomum* and *Gaigeria* species usually predominate in field outbreaks among cattle and small ruminants in Nigeria (Schillhorn van Veen, 1973; Anosa, 1977; Chiejina, 1986, 1987; Nwosu *et al.*, 1996 a,b).

Effective control of PGE depends on efficient diagnosis and establishment of the causative parasites among others. Several methods are available for the diagnosis of PGE but the traditional coprologic examination for nematode ova is the oldest, simplest and most widely used technique for

PGE diagnosis in cattle and small ruminants (Soulsby, 1982; Chiejina, 1987; Blood *et al.*, 1995). However, as a diagnostic technique, coprologic examination lacks specificity since the eggs of most of the nematodes (Trichostrongylids) responsible for PGE are similar in morphology and thus difficult to distinguish from one another. Consequently, the only means of reaching specific diagnosis is to conduct faecal culture, larval recovery and larval identification (Soulsby, 1982; Chiejina, 1987).

Although faecal materials from small ruminants may be cultured directly, in most cases they are mixed with other sterile media to enhance bulk, egg hatchability and larval emergence. Larval recovery from such media has been variable (Nwosu, 1995). In this study, some frequently used culture media were evaluated to determine their efficacy in promoting egg hatchability and larval recovery from the faeces of goats naturally infected with trichostrongylids.

### MATERIALS AND METHODS

**Collection and Preparation of Culture Media:** Faecal culture media were collected from cattle, sheep and goats maintained at the University of Maiduguri Teaching and Research Farm and horses belonging to the Mounted Troops of the Borno State Command of the Nigeria Police Force. In each case,

faecal samples were collected directly from the rectum after confirming that the animals had not been treated with any anthelmintic agent for at least four weeks prior to the study. Soil samples were collected from the University of Maiduguri compound.

About 500 grams of each culture medium was sterilized by autoclaving at 120 °C for 30 minutes. They were oven-dried overnight at 60 °C and individually ground into powder using a Phillips Twist HR 1707 blender. The samples were stored in sealed polythene bags until used.

#### Collection and Processing of Test Faecal Samples:

Test faecal samples were collected directly from the rectum of 840 goats during slaughter and evisceration at the Maiduguri Metropolitan abattoir. The age, sex and health status of the goats were noted. Goats aged six months or below were regarded as young while those above that age range were recorded as adult. Animals that had obvious signs of diarrhoea such as pasting of the hindquarters with watery faeces were regarded as diarrheic. Faecal samples were collected into polythene bags and transported to the laboratory for processing.

Faecal examination and egg counts followed the modified McMaster technique using saturated sodium chloride solution as the floating medium (MAFF, 1977). Faecal culture and larval recovery were conducted using the test tube filter paper method (Harada and Mori, 1955). Faecal cultures were made in triplicates per media type and the average larval recovery taken for each culture medium. In all cases, the identification of nematode ova and infective larvae were based on standard criteria (MAFF, 1977; Sloss and Kemp, 1978; Soulsby, 1982).

**Data Analysis:** Data obtained during the study were summarized as Means  $\pm$  S.D. or in percentages. Statistical differences in the means were determined at the 5% level of significance using the analysis of variance (ANOVA) and Fisher's Exact Test (GraphPad, 1998).

## RESULTS

The prevalence of strongyle eggs in the goats examined during the study is presented in Table 1. Out of the 840 faecal samples examined, 704 (83.8%) contained strongyle eggs. The prevalence of infection was significantly higher among female, young and diarrhoeic goats than their corresponding male, adult and non-diarrheic counterparts ( $P < 0.05$ ).

Egg hatch and larval recovery from the various culture media are shown in Table 2. Larval recovery from direct culture of the faecal samples without any culture medium (Control sample) revealed a range of 42 - 1,915 with a mean of  $688 \pm 507$  larvae. When this was used as a standard (100% egg hatch or 0% reduction in egg hatch), larval recovery was significantly highest ( $P < 0.05$ ) from goat faeces than any other culture media evaluated during the study.

**Table 1: Prevalence of strongyle nematode eggs in goat faeces examined at Maiduguri, Nigeria**

	<i>Number examined</i>	<i>Number (%) infected</i>
<b>All goats</b>	840	708 (83.8)
<b>Age</b>		
Young	580	560 (96.5) <sup>a</sup>
Adult	260	144 (55.4) <sup>b</sup>
<b>Sex</b>		
Male	608	497 (81.7) <sup>b</sup>
Female	232	207 (89.2) <sup>a</sup>
<b>Health status</b>		
Diarrheic	320	314 (98.1) <sup>a</sup>
Non-diarrheic	520	390 (75.0) <sup>b</sup>

<sup>ab</sup>Figures with different superscripts in the same column for sex, age and health status are significantly different ( $P < 0.05$ ).

## DISCUSSION

The results of this study revealed that strongylid nematode infections are highly prevalent in goats in the study area. The prevalence of 83.8% recorded in the study is similar to the range of 77 - 100% reported from other geographical zones of Nigeria (Fagbemi and Dipeolu, 1982; Chiejina, 1986; Nwosu *et al.*, 1996 a,b).

The results also revealed that the type of media used for culturing strongyle nematode eggs significantly ( $P < 0.05$ ) influenced the number of eggs that hatch and thus the number of infective larvae that may be recovered. In the present study where various culture media were used and the samples subjected to similar environmental conditions, most number of infective larvae were recovered from eggs cultured in goat faeces. Preparasitic nematode stages in the environment are known to require optimal conditions of moisture, oxygen tension and warmth for their development, growth and survival (Soulsby, 1982). Since the eggs in the various culture media were subjected to similar environmental conditions, it means that the variations in the number of infective larvae harvested reflected the actual capacity of the various culture media to support the development and hatching of nematode eggs as well as the eventual survival of the hatched larvae to the infective stage.

Generally, only moist and crumbly but not really wet faecal materials are ideal for culture to recover infective larvae (Kaufman, 1996). Consequently, faecal samples that are either very dry or very wet are usually mixed with water or other culture materials respectively to bring them to the ideal consistency for culturing. In this regard, charcoal, peat moss and vermiculite have been used as culture media (Kaufman, 1996). Presently, in Nigeria, these culture media are either scarce or expensive and thus not readily available for faecal culture and larval recovery for routine diagnostic or research purposes. Consequently, faecal materials from other domestic animals, especially horses and cattle are commonly used for this purpose in Nigeria. The results of this study therefore highlight the superior quality of goat faeces for the culture of faecal samples from the goat.

**Table 2: Larval recovery from ovine strongyle eggs cultured in various culture media**

Culture medium	Larval hatch Mean $\pm$ S.D.	Range	% larval hatch	% reduction in larval hatch
Direct (control)	688 $\pm$ 507	42 - 1915	100	0*
Soil	128 $\pm$ 186 <sup>e</sup>	7 - 580	18.6	81.4
Cow faeces	360 $\pm$ 300 <sup>d</sup>	3 - 854	52.4	47.6
Goat faeces	677 $\pm$ 652 <sup>a</sup>	6 - 1855	98.4	1.6
Sheep faeces	397 $\pm$ 407 <sup>c</sup>	1 - 1067	57.7	42.3
Horse faeces	487 $\pm$ 470 <sup>b</sup>	3 - 1382	42.3	29.2

\*Direct culture was used as control and standard (0% reduction in larval hatch) <sup>abcde</sup>Figures with different superscripts in the same column are significantly different (P<0.05).

## REFERENCES

- AKEREJOLA, O. O., SCHILLHORN VAN VEEN, T. W. and NJOKU, C. O. (1979). Ovine and caprine diseases in Nigeria: a review of economic losses. *Bulletin of Animal Health and Production in Africa*, 27: 65 - 70.
- ANOSA, V. O. (1977). Observations on the helminthiasis caused by *Haemonchus contortus* in Nigerian Dwarf Sheep. *Tropical Animal Health and Production*, 9: 11 - 17.
- BLOOD, D. C., HENDERSON, J. A., RADOSTITS, A. M. AND ARUNDEL, J. A. AND GAY, C. C. (1995). *Veterinary Medicine*. 5<sup>th</sup> edn. Bailliere Tindall, London pp. 750 - 798.
- CHIEJINA, S. N. (1986). The epizootiology and control of parasitic gastro-enteritis of domesticated ruminants in Nigeria. *Helminthological Abstracts* (Series A), 55: 413 - 429.
- CHIEJINA, S.N. (1987) Parasitic gastroenteritis in cattle and small ruminants: pathogenesis, diagnosis and treatment. *Zariya Veterinarian*, 2: 45 - 64.
- FAGBEMI, B. O. AND DIPEOLU, O. O. (1982). Strongyle infection of small ruminants in Nigeria. *Veterinary Parasitology*, 110: 347 - 353.
- GRAPHPAD, (1998). GraphPad Software, InStat guide to choosing and interpreting statistical tests, 1998, GraphPad Software, Inc., San Diego California USA, www.graphpad.com.
- HARADA, Y. AND MORI, O. (1955). A new method for culturing Hookworm. *Yonago Acta Medica*, 1: 177 - 179.
- ILCA, 1979). *Small ruminant production in the humid tropics*. International Livestock Centre for Africa, Addis Ababa, Ethiopia, 122 pp.
- KAUFMAN, J. (1996). *Parasitic infections of domestic animals: a diagnostic manual*. Birkhauser Verlag, Basel, Switzerland. 451 pp.
- KUIL, H. (1969) Livestock development and parasites. *Proceedings of the Conference on Livestock Development in the dry and intermediate savanna zone*, Zaria, 1969.
- MAFF, (1977) *Manual of Veterinary Parasitological Laboratory Techniques*. Technical Bulletin No. 18, Ministry of Agriculture, Fisheries and Food, London. 129 pp.
- NWOSU, C. O. (1995) The clinical parasitology and immunology of natural and experimental haemonchosis in Red Sokoto (Maradi) goats Ph.D. thesis, University of Ibadan, Ibadan, Nigeria.
- NWOSU, C. O., OGUNRINADE, A. F. AND FAGBEMI, B. O. (1996a). The seasonal prevalence of *Haemonchus* species in Red Sokoto (maradi) goats in Nigeria. *Veterinary Research Communications*, 20: 367 - 370.
- NWOSU, C. O., OGUNRINADE, A. F. AND FAGBEMI, B. O. (1996b). Prevalence and seasonal changes in the gastro-intestinal helminths of Nigerian goats. *Journal of Helminthology*, 70: 329 - 333.
- OMEKE, B. C. O. (1988). Improving goat productivity in the humid zone of the tropics. *Bulletin of Animal Health and Production in Africa*, 36: 126 - 130.
- SCHILLHORN VAN VEEN, T.W. (1973) Small ruminant health problems in Northern Nigeria with emphasis on the helminthiasis. *Nigerian Veterinary Journal*, 2: 26 - 31.
- SLOSS, W. M. AND KEMP, L. R. (1978). *Veterinary Clinical Parasitology*. 5<sup>th</sup> edition, Iowa State University Press, Ames, Iowa, pp 148 - 219.
- SOULSBY, E.J.L. (1982). *Helminths, Arthropods and Protozoa of domesticated animals*. 7<sup>th</sup> edn., Bailliere Tindall and Cassel, London 809 pp.

## OCTANOL/WATER PARTITION COEFFICIENT AND BIOACCUMULATION INDEX OF BONNY LIGHT CRUDE OIL IN CAT FISH *Clarias agboyiensis* IN LABORATORY-DOSED SEDIMENTS

UBANI, Chibuike Samuel, ONWURAH, Ikechukwu Noel Emmanuel and ALUMANAH, Eddy  
Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria

**Corresponding Author:** UBANI, C. S., Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria. Email: [chkubani@yahoo.com](mailto:chkubani@yahoo.com), Phone: 234-8059389711

### ABSTRACT

*Octanol/water partition coefficient and bioaccumulation index of Bonny light crude oil, having a fractional percentage composition of 81.11 saturate, 7.20 aromatics, 2.48 asphaltene and 9.21 residues, were studied in microcosm aquaria using a fresh water catfish *Clarias agboyiensis*. The partition coefficient ( $K_{ow}$ ) of the crude oil was evaluated to be 0.74. The mean bioaccumulation values of the petroleum hydrocarbons (PHCs) in the homogenates of the whole fish, liver and kidney at intervals of 24, 72 and 120 hours were respectively  $0.845 \pm 0.118$ ,  $11.0 \pm 0.058$  and  $15.0 \pm 0.064$  after exposing the whole fish to sediment and water, respectively containing  $31 \mu\text{g/g}$  and  $190 \mu\text{g/l}$  of the crude oil in the aquarium. The mean bioaccumulation values of petroleum hydrocarbons (PHCs) in the tissue homogenates of the exposed fish were higher than in the control ( $p < 0.05$ ), thus suggesting that Bonny light crude oil with  $K_{ow}$  of 0.74 could be lipophilic.*

**Keywords:** Crude Oil, Bioaccumulation, Partition Coefficient, *Clarias agboyiensis*

### INTRODUCTION

Unwholesome activities by man have created a severe imbalance in our ecosystem. In recent years, public interest in environmental pollution issues has grown that there is extensive coverage in the media. Emphasis on environmental sciences has shifted from direct toxic threat to man towards more general concern regarding pollutants impact on animals and plants, ecosystem and indeed in the whole biosphere (Peakall and Mohtadi, 1980). The society worldwide is increasingly becoming jittery over the safety and quality of the environment in which they live. There is considerable concern on the impact of oil pollution in both the terrestrial and aquatic ecosystems as a result of corrosion/rupture of oil-pipelines sabotage, accidental spills, seepage from storage tanks and tanker wash-off (Inyang, 1996). The fate and effect of crude oil and other petroleum products in the natural ecosystem have been the subject of many investigations. Obviously, the impact of crude oil spillage and discharge on the ecosystem as a result of oil exploration and exploitation activities is a problem of environmental concern (Coon and Dieter, 1981), particularly with regards to the associated heavy metals present in the crude oil. In Nigeria, particularly in the Niger-Delta area, the discharge of crude oil into aquatic environment and its consequent pollution hazard is increasingly becoming a phenomenon of concern (Antai and Mgborno, 1993). Such episodes have created devastating socio-economic problems and health hazard to communities affected, and have been the subject of various litigation between the host community and the oil prospecting companies.

Within the framework of many Environmental Risk Assessment (ERA) procedures for

chemicals, the measuring of bioaccumulation is required under certain circumstances. One main criterion for bioaccumulation potential is the n-octanol/water partition coefficient ( $K_{ow}$ ), which is often used to express hydrophobicity. Chemicals that have  $K_{ow}$  values higher than 2 are usually considered liable to bioaccumulate in biota (Oliver and Charlton, 1984). Bioaccumulation studies are used to assess the rate and extent of contaminant accumulation by an organism from various media including air, water, food, soil and sediment. The rate at which a pollutant bioaccumulates in a given lower trophic level is important for assessing the hazard it poses for a higher trophic level of a food chain (USEPA, 2000). Models used to predict bioaccumulation in food webs have been reported (Sample *et al.*, 1998; 1999)

### MATERIALS AND METHODS

**Test Sample:** The test sample for the study was Bonny light crude oil obtained from the Department of Petroleum Resources (DPR), Nigeria National Petroleum Cooperation (NNPC), Port Harcourt.

**Experimental Design:** *Clarias agboyiensis* caught alive in nets during the rainy season were purchased from fishers at the bank of Anambra River, at Otuocho, Anambra East Local Government Area, Anambra State in the month of May 2002. One hundred (100) healthy *Clarias agboyiensis* of average body weight of  $19.33 \pm 4.40\text{g}$  and length  $15.25 \pm 1.5\text{ cm}$  were selected and distributed into five groups with 10 catfish per group. They were allowed to acclimatize in uncontaminated aquaria in the Zoological garden, University of Nigeria, Nsukka for fourteen days prior to their exposure to crude oil-contaminated aquaria. Water in the aquarium was

changed twice weekly during the acclimatization phase. The catfish were also fed twice weekly with feed composed of 30.41% crude protein obtained from Fishery's Unit, Zoology Department, University of Nigeria, Nsukka.

The acclimatized *Clarias agboyiensis* were divided into two categories A and B. Category A numbering 50 were of average body weight  $16.45 \pm 3.38$ g and length  $14.3 \pm 1.6$  cm while category B also numbering 50 were of average body weight  $22.21 \pm 5.41$ g and length  $6.2 \pm 1.4$  cm. *Clarias agboyiensis* under the two categories A and B were each separated into five treatment groups. All the groups were introduced into the various contaminated aquaria with the exception of the control groups (uncontaminated aquaria).

**Preparation of Oil-Contaminated Sediment:** The method of Landrum *et al.* (2000) was used in the sediment preparation and contamination. In this method 10.0 g portions of characterized soil were spiked with 0.2, 0.4, 0.6 or 5.0g of Bonny light crude oil. The oil-spiked soil samples were mixed thoroughly after which equal volume of tap water (200 ml) was added to each portion and stirred with rod stirrer. The mixture was further shaken vigorously for 10 minutes before allowed to stand for 15 minutes. At the end of the interval, the water was decanted leaving only the sediment at the bottom of the beaker. A similar treatment was further performed on the above samples but with a standing interval of 30 minutes. The same treatment was repeated, but the duration of standing was 24 hours. The prepared sediment was transferred into plastic bowls and appropriate volume of tap water used to wash off the sediment into the plastic aquaria. More water was added to bring the final volume to 4 litres. All the aquaria were allowed to equilibrate for 4 days before introducing the acclimatized *Clarias agboyiensis*. The aquaria containing the fish were all covered with nets fastened with rubber.

**Tissue Collection:** Two *Clarias agboyiensis* were randomly collected from each aquarium using a plastic sieve. One of the fish was used for whole tissue assay and the other for liver and kidney assay. The sampled *Clarias agboyiensis* were sacrificed by piercing their heads with knife. The skin was wiped with tissue paper after washing in tap water. This helped to remove extra mucus secretion where oil particles could be loosely attached. The *Clarias agboyiensis* were then cut into small pieces (for whole body tissue assay) using sterile stainless steel scissors that was rinsed three successive times in hot distilled water. The second fish was dissected to expose the viscera, liver and kidney. All the tissues were homogenized and suspended in normal saline.

**Determination of Petroleum Hydrocarbons (PHC) in Sediment and Water:** The concentrations of petroleum hydrocarbons (PHCs) in the crude oil adsorbed to the spiked sediment were determined, using 1g portion of the sediment. The PHC was extracted with 5 ml of 1:1 mixture of

chloroform/ethanol by vigorous shaking for two hours, and allowed to stand for four hours. Extraction was done twice and the optical density (OD) of the extract read at 520 nm against the extraction mixture as blank. The same approach was used to determine the concentrations of PHC that partitioned into the water phase.

**Determination of Petroleum Hydrocarbons (PHC) in the Homogenates:** In assaying for the total PHCs that bioaccumulated in the liver, kidney and body of the exposed fish, 1g portions of their homogenates, were mixed with 5 ml of the extraction solvent. The mixture was shaken vigorously and allowed to stand for the supernatant to be separated completely. This was repeated twice and the O.D of the supernatant read at 520 nm against the extraction mixture as the blank.

**Determination of Octanol/Water Partition Coefficient ( $K_{ow}$ ) of Bonny Light Crude Oil:** The octanol/water partition coefficient ( $K_{ow}$ ) of Bonny light crude oil was determined as described by Gobals *et al.* (2002) at 28.5°C. In this method equal volume (5 ml) of water (W) and octanol (O) were equilibrated with each other for 4 hours before adding 2 ml of the crude oil (Bonny light). The mixture was shaken vigorously to effect the distribution of the crude oil to the two phases. The set-up was allowed to stand for 24 hours. When an equilibrium condition was achieved, the net volume of the two phases was determined for estimation of the partition coefficient.

Nernst distribution law defines partition coefficient of a substance ( $X_1$ ) between water and n-octanol phases ( $K_{ow}$ ) as the ratio of equilibrium concentration in the two phases. Most frequently it is given as the logarithm to the base 10 ( $\log K_{ow}$ ) as shown in Equation (1).

$$K_{ow} = \frac{X_i^0 / V^0}{X_i^w / V^w} = \frac{C_i^0}{C_i^w} \quad (1)$$

Where  $X_i^0$  is molar mass of octanol,  $V^0$  the final volume of octanol,  $X_i^w$  the molar mass of water,  $V^w$  the final volume of water,  $C_i^0$  the molar concentration of octanol and  $C_i^w$  the molar concentration of water.

## RESULTS AND DISCUSSION

The distribution of 2 ml of crude oil in the aqueous phase of octanol/water is presented in Table 1. Also the estimated concentrations of PHC in the crude oil spiked sediment and water in each aquarium are shown in Table 2. The partition coefficient ( $K_{ow}$ ) of Bonny light crude oil calculated from Equation (1) was 0.743 and this tends to suggest that the crude oil will most likely not adsorb to particulate organic matter. The octanol/water partition coefficient has been shown to be the measure of a chemical's affinity for lipid portion of an organism's tissue. Since



chemicals with  $K_{ow}$  of 2-6 are said to be lipophilic for some water body (Oliver and Charlton, 1984), it shows that Bonny light crude oil is far less lipophilic in the test microcosm aquarium. The speculation that chemicals with  $K_{ow}$  values  $\geq 2$  are lipophilic also suggests that Bonny light crude oil is not lipophilic. But this seems not the case as shown by the results of bioaccumulation of PHCs in the liver (Tables 3) and the kidney (Table 4) of the exposed fish. The data in Tables 3, 4 and that for bioaccumulation in the body tissue (Table 5) reveal that at 72 hours the bioaccumulation values were highest in the kidney, followed by the liver whereas the whole fish body has the least PHC bioaccumulation. The reason adduced for this increase in bioaccumulation levels in the kidney and liver could be due to their involvement in the "clearance" of toxicants in the body of animals which culminates in their elimination via the kidney. Considering the role of the liver in biotransformation of xenobiotics, the reason for high PHC concentration in the former becomes more glaring. The high PHC bioaccumulation in these organs makes them to be prone to hepatic lesion or organ dysfunction (French et al, 1996). Hence both the liver and the kidney work in a concerted manner as regards detoxification and elimination of pollutants.

**Table 1: Distribution of 2 ml crude oil in aqueous phase of octanol and water**

Compound	Initial volume (ml)	Final volume (ml)	Molar mass of compound
N-octanol	5.0	6.8	130.2
Water	5.0	5.2	18.0
Bonny light crude oil	2.0	-	-
Total volume	12.0	12.0	-

**Table 2: Concentration of PHC in each of the spiked sediment and water**

Aquarium	Initial crude oil dose (g)	Final values of PHC in spiked sediment $\mu\text{g/g}^*$	Final values of dissolved PHC in water $\mu\text{g/l}^*$
Control	No oil	-	-
A	0.2	24	180
B	0.4	31	190
C	0.6	33	220
D	5.0	221	900

\*These values were considered as the concentration of crude oil directly affecting the fish in the aquarium

It should be stated that "purging" of the fish after exposure to the crude oil was not carried out against the U.S. EPA protocol (U.S. EPA, 2000). This is because of the concern that tissue-bound PHC will depurate from the fish during the holding in clean water, thus under-representing the steady state in the organism (U.S. EPA, 2000). The reason for carrying out "purging" is to prevent sediment bound chemicals (PHC) in the gut of the fish from being measured as part of the body tissue burden or concentrations. Hence, Bonny light crude oil having a low  $K_{ow}$  value of 0.74 should have a low tendency to bioaccumulate in the catfish, but the result of this

study suggests the contrary, in that, the bioaccumulation of PHC in the tissue homogenates of the exposed fish was high relative to the control. It has been observed that uptake rates and bioaccumulation levels of a substance within the body tissues are of particular interest because they can be related to toxicity and body burden, and these endpoint parameters can be used to predict impact (Chapman, 1997).

Fish concentrate (bioaccumulate) lipophilic contaminants mainly by exchange across the gills (depending on the gill ventilation rate of the animal). However, Table 4 illustrates the extent to which *Clarias agboyiensis* could bioaccumulate PHC in the liver. Although dietary uptake of the same contaminant is negligible in fresh water fish because of their natural low feeding rates, poor adsorption efficiency and rapid elimination rates of the contaminants (Niimi and Dookhram, 1989; Randall *et al.*, 1998), the bioaccumulation levels of Bonny light crude oil in the various tissue homogenates of the *Clarias agboyiensis* exposed to the crude oil were significantly high ( $P \leq 0.05$ ) relative to the parallel controls. This suggests that the exposed fish are prone to the toxic injury of the test crude oil sample.

The fact that PHC could bioaccumulate in the various tissues of the fish as shown in the study in spite of the "poor" lipophilicity of the crude oil sends a warning signal to the higher trophic levels that may depend on fish for food, especially in areas prone to crude oil spillage, considering the inherent toxicity of crude oil and the fact that its body burden builds up with time. Consequently, incessant consumption of "sea foods" exposed to crude oil pollution could lead to disease conditions caused by the carcinogenic, mutagenic or even teratogenic properties of crude oil and its derivatives. These disease conditions may lead to death as terminal results.

## REFERENCES

- ANTAI, S. P. and MGBORNO, E. (1993). Pattern of Degradation of Bonny light crude oil by *Bacillus sp* and *Pseudomonas sp* isolated from oil spilled site. *West African Journal of Biological and Applied Chemistry*, 39: 1 – 4.
- CHAPMAN, P. M. (1997). Is bioaccumulation useful for predicting impacts? *Marine Pollution Bulletin*, 34: 282 – 283.
- COON, N. C. and DIETER, M. C. (1981). Responses of adult Mallard Ducks to ingested south Louisiana crude oil. *Environmental Research*, 24: 309 – 314.
- GOBAS, F. A. P. C., LAHILTAT, J. M., GAROTALO, G., SHIU, W. Y. and MACKAY, D. (2002). A novel method for measuring membrane water partition coefficient of hydrophobic organic chemicals: Comparison with n-octanol-water partitioning. *Journal of Pharmacological Science*, 77: 265 – 272.
- INYANG, L. E. D. (1996). Biomarker fingerprints pattern of Biodegradation and Characterization of Qua-Ibo light crude oil as

**Table 3: Bioaccumulation of petroleum hydrocarbon in fish liver homogenate after 24 hourly interval of exposure in crude oil- contaminated aquaria**

Treatment group	Crude oil concentration		Concentration of PHC in homogenates (mg/g)		
	Sediment (µg/g)	Water (µg/l)	T <sub>1</sub> = 24 hrs	T <sub>2</sub> = 72 hrs	T <sub>3</sub> = 120 hrs
Control	No oil	No oil	0.00	0.00	0.00
A	24	180	9.0 ± 0.119**	10.0 ± 0.190**	9.0 ± 0.099**
B	31	190	9.0 ± 0.091**	11.0 ± 0.058**	8.0 ± 0.058**
C	33	220	9.0 ± 0.110**	14.0 ± 0.100**	7.0 ± 0.101**
D	221	900	8.0 ± 120**	11.0 ± 0.020**	ND*

\*\* Results are significantly different ( $P \leq 0.05$ ) from each other; thus showing effect of concentration and time of exposure. \* Not determined because the fish died before this time

**Table 4: Bioaccumulation of petroleum hydrocarbon in fish kidney homogenate after 24 hourly interval of exposure in crude oil- contaminated aquaria**

Treatment group	Crude oil concentration		Concentration of PHC in homogenates (mg/g)		
	Sediment µg/g	Water µg/L	T <sub>1</sub> = 24 hrs	T <sub>2</sub> = 72 hrs	T <sub>3</sub> = 120 hrs
Control	No oil	No oil	0.00	0.00	0.00
A	24	180	10.0 ± 0.068**	20.0 ± 0.071	14.0 ± 0.081**
B	31	190	7.0 ± 0.040**	15.0 ± 0.064**	19.0 ± 0.046**
C	33	220	9.0 ± 0.085**	39.0 ± 0.122	17.0 ± 0.091**
D	221	900	10.0 ± 0.113**	29.0 ± 0.122**	ND*

\*\* Results are significantly different ( $P \leq 0.05$ ) from each other; thus showing effect of concentration and time of exposure. \* Not determined because the fish died before this time

**Table 5: Bioaccumulation of petroleum hydrocarbon in whole fish homogenate after 24 hourly interval of exposure in crude oil – contaminated aquaria**

Treatment group	Crude oil concentrations		Concentration of PHC in homogenates (mg/g)		
	Sediment µg/g	Water (µg/l)	T <sub>1</sub> = 24 hr	T <sub>2</sub> = 72 hr	T <sub>3</sub> = 120 hr
Control	No-oil	No-oil	0.00	0.00	0.00
A	24	180	0.2160 ± 0.106**	0.9193 ± 0.023**	0.8173 ± 0.050**
B	31	190	0.3630 ± 0.046**	0.8453 ± 0.118 <sup>xx</sup>	0.9897 ± 0.100**
C	33	220	0.8949 ± 0.076**	1.0670 ± 0.113**	1.0900 ± 0.100**
D	221	900	1.0787 ± 0.110**	1.1250 ± 0.044**	ND*

\*\* Results are significantly different ( $P \leq 0.05$ ) from each other; thus showing effect of concentration and time of exposure. \*Not determined because the fish died before this time

- correlation tools for identification of mystery oil spill. *Paper presented at the International Environment Seminar on the Petroleum Industry and the Nigerian Environment*, Port Harcourt, River State.
- LANDRUM, P. E., KUKKONEN, J. K., JUSSI, V. K., HOOFF, V. and PATRICK, L. (2000). Impact of sediment manipulation on the bioaccumulation of polycyclic aromatic hydrocarbons from Field-contaminated and Laboratory-Dosed Sediments by an Oligochaete. *Environmental Toxicology and Chemistry*, 20: 1752 – 1761.
- NIIMI, A. J and DOOKHRAM, G. P. (1989). Dietary absorption efficiencies and elimination rates of polycyclic aromatic hydrocarbons (PAHs) in rainbow trout *Salmo gairdneri*. *Environmental Toxicology and Chemistry*, 8: 719 – 722.
- OLIVER, G. and CHARLTON, M. N. (1984). Chlorinated organic contaminated on settling particulates in Niagara River Vicinity of lake Ontario. *Environmental Science and Technology*, 18: 903 – 905.
- PEAKALL, D. E. and MOHTADI, M. F. (1980). Oil spill on land as potential sources of ground water contamination. *Environment International*, 3: 107 – 120.
- RANDALL, D. J., CONNELL, D. N., YANG, R. and WA, S. S. (1998). Concentration of persistent lipophilic compounds in fish determined by exchange across the gills not through the food chain. *Chemosphere*, 37: 1263 – 1270.
- SAMPLE, B. E., BEAUCHAMP J. J., EFROYMSON R. A. and SUTER, G. W. (1998). *Development and validation of bioaccumulation models for small mammals*. ES/ER/TM-219. U.S. Development of Energy, Oak Ridge National Laboratory. Oak Ridge, TN.
- SAMPLE, B. E., BEAUCHAMP J. J., EFROYMSON R. A. and SUTER, G. W. (1999). Literature-derived bioaccumulation models for earthworms: Development and validation. *Environmental Toxicology and Chemistry*, 18: 2110 – 2120.
- U.S. EPA (2000). *Methods for measuring the toxicity and bioaccumulation of sediment associated contaminants with fresh water invertebrates*. 2<sup>nd</sup> ed. EPA/600/R-99/064, Office of Record and Development, Washington DC
- WALKER, C. H. and JOHNSON, G. O. (1989). Interactive effects of pollutants at the toxicokinetic level-implication for the marine environment. *Marine Environment Research*, 28: 521 – 525.

## LENGTH-WEIGHT RELATIONSHIP AND CONDITION FACTOR OF FOUR MORMYRID SPECIES OF ANAMBRA RIVER

<sup>1</sup>NWANI, Christopher Didigwu; <sup>2</sup>EZENWAJI, Henry Maduka Godfrey; <sup>2</sup>EYO, Joseph Effiong and <sup>3</sup>UDE Emmanuel Fame

<sup>1</sup>Department of Applied Biology, Ebonyi State University P.M. B. 53, Abakaliki, Ebonyi State, Nigeria

<sup>2</sup>Fisheries and Hydrobiology Research Unit, Department of Zoology, University of Nigeria Nsukka

<sup>3</sup>Department of Animal Production and Fisheries Management, Ebonyi State University P.M.B. 53, Abakaliki, Ebonyi State, Nigeria

**Corresponding Author:** Nwani, C. D., Applied Biology Department, Ebonyi State University, Abakaliki. Email: [cdnwani@yahoo.com](mailto:cdnwani@yahoo.com) Phone 08037509910

### ABSTRACT

*The length-weight relationship and condition factor of four mormyrid species namely Mormyrus rume, Hyperopisus bebe, Campylomormyrus tamandua and Gnathonemus petersii from Anambra river were investigated from October 2002 to March 2004. In all, 400 species M. rume, 384 H. bebe, 417 C. tamandua and 335 G. petersii were sampled for the study. Length-weight relationship showed that the exponent "b," were 3.067, 2.459, 3.201 and 3.114 for M. rume, H. bebe, C. tamandua and G. petersii respectively. The mormyrid species studied with exception of H. bebe exhibited isometric growth and the correlation coefficients were positive and highly significant ( $P < 0.05$ ). The condition factor ( $k$ ) varied from  $0.69 \pm 0.22$  in G. petersii to  $1.17 \pm 0.59$  in M. rume. There were no significant difference in the mean condition factor between the males and females in all the mormyrid species. The importance of condition factor in the breeding activities of the mormyrid species revealed that not much energy is diverted into gonad synthesis and maturation during the breeding cycle season.*

**Keywords:** Mormyrids, Length-weight relationship, Condition factor

### INTRODUCTION

Mormyrid species are widespread in Afro-tropical river ecosystems (Lowe-McConnel, 1972). Commonly known as 'Elephant snout' or 'Elephant nose' fishes, mormyrids are well represented in Nigeria water with about thirty-one (31) different species belonging to eleven (11) genera (Olasoebikan and Raji, 1998). However, four species namely *Mormyrus rume*, *Hyperopisus bebe*, *Campylomormyrus tamandua* and *Gnathonemus petersii* inhabitants of Anambra river were studied in this report.

Mormyrid fishes are preferred by the inhabitants of Anambra area because they are readily available, tasty and relatively cheap. They account for a significant proportion of the total fish landing in most fresh fish landing sites in Nigeria (Reed *et al.*, 1967; Ita, 1978; Victor and Tetteh, 1988). Mormyrids are increasingly becoming important in the world aquarium business, aquaculture and neurological studies (Gosse, 1984). Works available on Mormyrids include those of King 1989, King 1996 a and b, Ikomi 1996 and Ezenwaji 2004. The present study is intended to add unto the existing information on the biology of Mormyrids with emphasis on the length-weight relationship and condition factor.

**Description of Study Site:** The Anambra River has its source from Ankpa highlands of Kogi State, Nigeria. It lies between latitude  $6^{\circ}10'$  and  $7^{\circ}20'$  and longitude  $7^{\circ}40'$  East of river Niger. There is a rainy season (April – September / October) and a dry

season (October / November – March). From December to January / February, the basin is influenced by the harmattan, but its effect is not well marked (Ezenwaji 1989). The ranges of some key physico-chemical parameters of the water are: water temperature  $21.19 - 28.40^{\circ}\text{C}$ , transparency 0.4-1.4m, water level, 4.70-6.76m, air temperature  $19.89^{\circ}\text{C} - 27.02^{\circ}\text{C}$  and pH 6.0 – 7.4. The mean rainfall data was  $109.58 \pm 104.5$  mm with the highest record of 280 mm in July. There was no rainfall from December to March.

The vegetation in the basin is guinea Savanna but the lentic water bodies are often fringed with macrophytes like *Pterocaropus spp*, *Jussiaea spp*, *Eupatorium odoratum*, *Pennisetum spp*, *Cynodon spp* and in some areas *Raphia hookeri*. Relevant human activities in the area are fishing sand mining, bathing, domestic washing, trading (mainly on food items) and rice processing.

### MATERIAL AND METHODS

Fish samples were collected monthly at Otuocha and Ogurugu water ports of Anambra river from October 2002 to March 2004 using gill, drag, drift and cast nets of mesh sizes between 70 to 120 mm. Baskets, traps and hook and line were also used. Fish collected were preserved in ice and transported to the laboratory for measurements. Each fish was weighed to the nearest 0.1 g, total and standard length were determined to the nearest 1 mm.

The length weight relationship of the fish was determined by the equation:  $W = aL^b$ , where W is weight in grams, L is standard length in centimeters, and a & b are regression constants. The logarithm transformed data gave the straight line relationship:  $\log W = \log a + b \log L$ .

The condition factor for each specimen was calculated using the method of Bagenal and Tecsh (1978) thus:  $K = W/L^3 \times 100/1$ ; where K = condition factor, W = weight of fish in grams and L = length of fish in centimeters. The coefficient of variation (CV) was determined as:  $CV = (S/\bar{x} \times 100/1) \%$  (King and Udo, 1996); where S = standard deviation and  $\bar{x}$  = population mean.

**Data Analysis:** The average value of "b" for each species was tested to verify whether it was significantly different from '3' using t – test at 0.05 significance difference. The sexual and seasonal variations in condition factor were also determined using t – test while the coefficient of variations (CV) were tested using F – test.

## RESULTS

**Length-weight Relationship:** The length-weight relationship parameters of mormyrid species in Anambra River are presented in Table 1. The "b" values for male, female and combined sex for *M. rume* were 3.013, 3.114 and 3.076 respectively. For *C. tamandua*, the "b" values for males, females and combined sex were 3.014, 3.215 and 3.201 respectively. Similarly, the "b" value for *G. petersii* males, females and combined sex were 3.001, 3.235 and 3.114 respectively. *M. rume*, *C. tamandua* and *G. petersii* thus exhibited isometric growth pattern for both sexes. The situation was however different in *H. bebe* where the "b" values for males, females and combined sexes were 2.355, 2.062 and 2.459 respectively. *H. bebe* therefore exhibited allometric growth pattern. In all the mormyrid species studied, the 'b' values for the males were not significantly different from the females ( $t = 2.04$   $df_{34}$ ,  $p > 0.05$ ).

**Condition Factor (K):** The monthly variations in the condition factors for the four mormyrid species are presented in Table 2. The annual mean value of the condition factor for *M. rume* (combined sex) was  $1.117 \pm 0.59$ . The mean condition factor for the male was  $1.20 \pm 0.09$  while that of the female was  $1.17 \pm 0.08$ . There was no significant difference in the annual average condition factor for the males and females *M. rume* ( $t = 1.15$   $df_{34}$ ,  $P > 0.05$ ).

The dry season value of  $1.39 \pm 0.56$  for *M. rume* (combined sex) was significantly different from that of the wet season value of  $0.81 \pm 0.44$  ( $t = 3.23$   $df_{16}$ ,  $P > 0.05$ ). However, the average condition of the male *M. rume* in the wet season (April – October)  $0.93 \pm 0.70$  was not significantly different from the dry season (November – March) value of  $1.38 \pm 0.84$  ( $t = 2.06$ ,  $df_{16}$ ,  $P > 0.05$ ). The interseasonal coefficient of variation (CV) in condition factor among the males for the dry (cv = 53.53%) and wet (cv = 49.68%)

seasons were not significantly different ( $F = 1.86$ ,  $P > 0.05$ ). However, the interseasonal variability in female condition factor for the dry (cv = 27.60%) and wet (cv=20.22%) seasons were not significantly different ( $F = 3.01$ ,  $P > 0.05$ ).

Female average condition factor for wet season ( $0.81 \pm 0.46$ ) significantly differed from that of the dry season value of  $1.40 \pm 0.63$  ( $t = 3.06$   $df_{16}$ ,  $P < 0.05$ ).

Considering *H. bebe* (combined sex), the mean condition factor was  $0.81 \pm 0.41$ . There was no significant difference in the mean condition factor between the males ( $0.83 \pm 0.61$ ) and females ( $0.79 \pm 0.11$ ) during the period studied ( $t = 0.86$   $df_{34}$ ,  $P > 0.05$ ). The dry season value of  $0.98 \pm 0.40$  for the combined sex was significantly different from the wet season value of  $0.55 \pm 0.27$  ( $t = 3.83$   $df_{16}$ ,  $P < 0.05$ ). Among the male *H. bebe*, the average condition factor for the dry season ( $1.01 \pm 0.73$ ) was not significantly different from the wet season value of  $0.55 \pm 0.54$  ( $t = 1.16$ ,  $df_{16}$ ,  $P > 0.05$ ). There was no significant difference in the interseasonal coefficient of variation in condition factor for the dry (cv = 51.89 %) and wet (cv = 53.75 %) seasons, ( $F = 2.98$ ,  $P > 0.05$ ). Among the female *H. bebe*, the mean condition for the dry season ( $0.95 \pm 0.46$ ) significantly exceeded the wet season value ( $0.35 \pm 0.46$ ) ( $t = 3.44$   $df_{16}$ ,  $P < 0.05$ ). The interseasonal coefficient of variation between the dry (cv = 22.2 %) and the wet (cv = 38.83 %) seasons were significantly different ( $F = 5.16$ ,  $P < 0.05$ ).

Among combined sex *C. tamandua*, the annual mean condition was  $0.73 \pm 0.29$ . There was no significant difference in the average condition between the males ( $0.67 \pm 0.14$ ) and females ( $0.79 \pm 0.29$ ) ( $t = 2.86$ ,  $df_{34}$ ,  $P > 0.05$ ). The mean condition factor for the combined sex in the dry season ( $0.88 \pm 0.26$ ) was significantly different from that of the wet season value of  $0.49 \pm 0.16$  ( $t = 5.15$ ,  $df_{16}$ ,  $P < 0.05$ ). Among the male *C. tamandua* the average condition factor for the dry season ( $0.82 \pm 0.49$ ) was statistically similar to the wet season values of  $0.44 \pm 0.40$  ( $t = 2.06$ ,  $df_{16}$ ,  $P > 0.05$ ). Similarly the interseasonal Coefficient of variation for the dry season (cv = 29.28%) was statistically similar to the wet (cv = 35.09%) season value, ( $F = 2.02$ ,  $P > 0.05$ ).

Among the female *C. tamandua* the mean condition factor for the dry season ( $0.95 \pm 0.51$ ) was not significantly different from the wet season value of  $0.5 \pm 0.39$  ( $t = 2.16$   $df_{16}$ ,  $P > 0.05$ ). There was also no significant difference in interseasonal coefficient of variation between the dry (cv = 27.48%) and wet (cv = 28.27) seasons ( $F = 1.54$ ,  $P > 0.05$ ).

The annual mean condition factor for the combined sex of *G. petersii* was  $0.69 \pm 0.22$ . The annual mean condition factor for the males ( $0.62 \pm 0.09$ ) was not significantly different from those of the females ( $0.77 \pm 0.06$ ), ( $t = 1.02$   $df_{16}$ ,  $P > 0.05$ ). The dry season mean value for the combined sex was  $0.64 \pm 0.21$  and was not significantly different from the wet season value of  $0.79 \pm 0.22$  ( $t = 2.07$ ,  $df_{16}$ ,  $P > 0.05$ ). The male average condition factor for the dry season ( $0.55 \pm 0.46$ ) and wet season ( $0.72 \pm$

Table 1: Length-weight relationship of Mormyrid species of Anambra river basin

Species	Sex	Number (n)	Length L (min)	Range (TL) L (max)	a	b	r	TL/SL
<i>Mormyrus rume</i>	F	183	19.60	73.40	0.0338	3.013	0.988	1.060
<i>Mormyrus rume</i>	M	217	16.20	60.00	0.0310	3.114	0.977	1.080
<i>Mormyrus rume</i>	M & F	400	17.60	70.34	0.0240	3.076	0.969	1.186
<i>Hyperopisus bebe</i>	F	173	12.40	46.10	0.0035	2.062	0.965	1.043
<i>Hyperopisus bebe</i>	M	211	11.30	41.20	0.0137	2.355	0.954	1.054
<i>Hyperopisus bebe</i>	M & F	384	11.04	40.10	0.0235	2.459	0.976	1.105
<i>Campylomormyrus tamandua</i>	F	182	8.70	42.70	0.0570	3.014	0.997	1.132
<i>Campylomormyrus tamandua</i>	M	235	7.70	42.40	0.0194	3.215	0.947	1.145
<i>Campylomormyrus tamandua</i>	M & F	417	7.50	43.07	0.0381	3.201	0.966	1.144
<i>Gnathonemus Petersii</i>	F	179	13.50	30.06	0.0275	3.001	0.980	1.123
<i>Gnathonemus Petersii</i>	M	156	11.90	27.00	0.0086	3.235	0.999	1.120
<i>Gnathonemus Petersii</i>	M & F	335	11.09	33.40	0.0176	3.114	0.049	1.125

\*TL = Total length, SL = Standard length, a = regression intercept, b = slope and r correlation coefficient

Table 2: Monthly variations in the condition factor (cf = w. 100/L<sup>3</sup>) of Mormyrid species in Anambra river basin

Months	Number	Male	Number	Female	Number	Male	Number	Female
<i>Mormyrus rume</i>				<i>Hyperopisus bebe</i>				
Oct. 2002	8	0.84 ± 0.06	12	0.86± 0.11	11	0.63 ± 0.04	11	0.61 ± 0.06
Nov	10	1.22 ± 0.08	9	1.11 ± 0.06	9	0.90 ± 0.19	10	0.70 ± 0.01
Dec	14	0.70 ± 0.03	11	0.64 ± 0.03	12	0.44 ± 0.83	12	0.64 ± 0.13
Jan	15	1.06 ± 0.04	10	1.93 ± 0.09	10	0.89 ± 0.71	9	1.20 ± 0.18
Feb. 2003	15	1.11 ± 0.05	12	1.70 ± 0.03	13	0.77 ± 0.42	10	0.96 ± 0.09
March	15	2.47 ± 0.06	9	1.55 ± 0.16	13	1.87 ± 0.88	10	0.87 ± 0.05
April	9	0.85 ± 0.08	10	0.91 ± 0.15	9	0.53 ± 0.77	8	0.66 ± 0.020
May	12	0.33 ± 0.11	11	0.45 ± 0.11	13	0.22 ± 0.81	8	0.26 ± 0.13
June	15	0.55 ± 0.13	9	0.84 ± 0.02	13	0.30 ± 0.96	8	0.38 ± 0.04
July	10	0.52 ± 0.17	7	0.93 ± 0.03	11	0.20 ± 0.87	6	0.57 ± 0.19
Aug.	11	1.13 ± 0.18	13	0.71 ± 0.07	9	0.76 ± 0.11	10	0.34 ± 0.13
Sept	11	1.13 ± 0.13	11	0.93 ± 0.14	11	1.01 ± 0.90	11	0.77 ± 0.19
Oct	14	1.84 ± 0.06	11	0.90 ± 0.19	11	0.83 ± 0.45	11	0.88 ± 0.07
Nov	12	0.70 ± 0.03	9	1.21 ± 0.01	10	0.61 ± 0.71	7	1.16 ± 0.18
Dec	9	0.91 ± 0.19	9	1.20 ± 0.08	8	0.83 ± 0.71	10	1.08 ± 0.09
Jan. 2004	12	1.95 ± 0.14	10	1.82 ± 0.04	19	0.99 ± 0.67	11	1.33 ± 0.04
Feb	15	1.52 ± 0.08	11	1.73 ± 0.03	16	1.03 ± 0.53	10	0.89 ± 0.01
March	10	2.68 ± 0.06	9	1.64 ± 0.06	13	2.17 ± 0.35	11	1.00 ± 0.31
Rainy season	90	0.93 ± 0.70	84	0.81 ± 0.40	88	0.55 ± 0.54	73	0.55 ± 0.46
Dry season	127	1.38 ± 0.84	99	1.40 ± 0.63	123	1.01 ± 0.73	100	0.95 ± 0.46
Annual mean	13	1.20 ± 0.09	11	1.17 ± 0.08	12	0.83 ± 0.61	10	0.79 ± 0.11
<i>Campylomormyrus tamandua</i>				<i>Gnathonemus petersii</i>				
Oct. 2002	12	0.66 ± 0.03	8	0.72 ± 0.02	9	0.44 ± 0.06	10	1.00 ± 0.07
Nov	13	0.49 ± 0.15	11	0.52 ± 0.71	16	0.49 ± 0.71	11	0.84 ± 0.14
Dec	14	0.89 ± 0.11	9	0.99 ± 0.11	12	0.37 ± 0.04	12	0.79 ± 0.13
Jan. 2003	12	1.02 ± 0.03	9	1.14 ± 0.02	5	0.88 ± 0.02	9	1.01 ± 0.02
Feb	18	1.09 ± 0.22	10	1.11 ± 0.07	8	0.63 ± 0.02	8	0.81 ± 0.11
March	15	0.45 ± 0.51	8	0.52 ± 0.01	8	0.94 ± 0.24	12	0.61 ± 0.01
April	25	0.32 ± 0.26	20	0.42 ± 0.11	9	0.99 ± 0.53	7	0.01 ± 0.01
May	16	0.19 ± 0.01	13	0.33 ± 0.20	8	0.79 ± 0.01	12	0.89 ± 0.05
June	9	0.49 ± 0.03	6	0.66 ± 0.14	10	1.00 ± 0.06	7	1.03 ± 0.07
July	10	0.59 ± 0.02	7	0.61 ± 0.32	7	0.63 ± 0.02	9	0.88 ± 0.01
Aug	12	0.36 ± 0.08	10	0.51 ± 0.60	11	0.71 ± 0.07	14	0.98 ± 0.09
Sept	8	0.50 ± 0.06	9	0.47 ± 0.20	7	0.58 ± 0.08	6	0.69 ± 0.04
Oct	9	0.62 ± 0.13	10	0.80 ± 0.62	8	0.33 ± 0.01	13	0.57 ± 0.08
Nov	12	1.07 ± 0.02	13	1.14 ± 0.80	6	0.24 ± 0.01	10	0.46 ± 0.05
Dec	11	1.04 ± 0.42	9	1.11 ± 0.91	5	0.60 ± 0.09	10	0.64 ± 0.06
Jan. 2004	16	0.71 ± 0.1	10	1.20 ± 0.09	9	0.53 ± 0.01	8	0.57 ± 0.04
Feb	13	0.94 ± 0.08	11	1.22 ± 0.01	9	0.42 ± 0.3	10	0.53 ± 0.04
March	10	0.63 ± 0.18	9	0.80 ± 0.23	9	0.54 ± 0.20	11	0.63 ± 0.02
Rainy season	101	0.44 ± 0.40	83	0.54 ± 0.39	69	0.72 ± 0.49	78	0.86 ± 0.41
Dry season	134	0.82 ± 0.49	99	0.95 ± 0.51	87	0.55 ± 0.46	101	0.72 ± 0.41
Annual mean	14	0.67 ± 0.14	11	0.79 ± 0.29	9	0.62 ± 0.09	10	0.77 ± 0.06



**Table 3: Seasonal variation in condition factor and coefficient of variation (CV) among four mormyrid species of Anambra river**

Species	Condition factor			Coefficient of Variation		
	Dry season	Wet season	T -value	Dry season	Wet Season	T-value
<i>Mormyrus rume</i>						
M	1.38 ± 0.84 <sup>a</sup>	0.93 ± 0.70 <sup>a</sup>	2.06	53.53 <sup>a</sup>	49.68 <sup>a</sup>	1.86
F	1.40 ± 0.63 <sup>a</sup>	0.81 ± 0.46 <sup>b</sup>	3.06	27.60 <sup>a</sup>	20.22 <sup>a</sup>	3.01
M & F	1.39 ± 0.56 <sup>a</sup>	0.81 ± 0.44 <sup>b</sup>	3.23	29.56 <sup>a</sup>	28.74 <sup>a</sup>	2.02
<i>Hyperopisus bebe</i>						
M	1.01 ± 0.73 <sup>a</sup>	0.55 ± 0.54 <sup>a</sup>	1.16	51.89 <sup>a</sup>	53.75 <sup>a</sup>	2.98
F	0.95 ± 0.46 <sup>a</sup>	0.35 ± 0.46 <sup>b</sup>	3.44	22.20 <sup>a</sup>	38.83 <sup>b</sup>	5.16
M & F	0.98 ± 0.40 <sup>a</sup>	0.55 ± 0.27 <sup>b</sup>	3.83	30.40 <sup>a</sup>	31.40 <sup>a</sup>	3.34
<i>Campylomormyrus tamandua</i>						
M	0.82 ± 0.49 <sup>a</sup>	0.44 ± 0.40 <sup>a</sup>	2.06	29.28 <sup>a</sup>	35.09 <sup>a</sup>	2.02
F	0.95 ± 0.51 <sup>a</sup>	0.50 ± 0.39 <sup>a</sup>	2.16	27.48 <sup>a</sup>	28.27 <sup>a</sup>	1.54
M & F	0.88 ± 0.26 <sup>a</sup>	0.49 ± 0.16 <sup>b</sup>	5.15	26.60 <sup>a</sup>	25.80 <sup>a</sup>	2.02
<i>Gnathonemus petersii</i>						
M	0.55 ± 0.46 <sup>a</sup>	0.72 ± 0.49 <sup>a</sup>	2.60	38.19 <sup>a</sup>	35.12 <sup>a</sup>	2.01
F	0.72 ± 0.41 <sup>a</sup>	0.86 ± 0.41 <sup>a</sup>	2.04	24.41 <sup>a</sup>	18.99 <sup>a</sup>	3.11
M & F	0.64 ± 0.21 <sup>a</sup>	0.79 ± 0.22 <sup>a</sup>	2.07	27.60 <sup>a</sup>	26.40 <sup>a</sup>	2.47

*a* and *b* indicate significant corresponding means at *P* = 0.05

0.49) were not significantly different (*t* = 2.60, *df*<sub>16</sub>, *P* > 0.05). There was no significant difference in interseasonal coefficient of variation between the dry (*cv* = 38.19 %) and the wet (*cv* = 35.12 %) seasons (*F* = 2.01, *P* > 0.05). Among the females, the average condition factor for the dry season (0.72 ± 0.41) and wet (0.86 ± 0.41) seasons were not significantly different (*t* = 2.06 *df*<sub>16</sub>, *P* > 0.05). The interseasonal coefficient of variation for the dry (*cv* = 24.41 %) and wet (*cv* = 18.99 %) seasons were not significantly different (*F* = 3.11, *P* > 0.05).

Comparing the mean condition factor for the combined sexes of the mormyrid species, *M. rume* (1.17 ± 0.59) significantly differed from that of *H. bebe* (0.81 ± 0.41), (*t* = 2.95 *df*<sub>34</sub>, *P* < 0.05), *C. tamandua* (0.73 ± 0.29) (*t* = 3.97, *df*<sub>34</sub>, *P* < 0.05) and *G. petersii* (0.69 ± 0.22) (*t* = 4.52, *df*<sub>34</sub>, *P* < 0.05). The value for *H. bebe* (0.81 ± 0.41) was not significantly different from that of *C. tamandua* (0.73 ± 0.29) (*t* = 0.97 *df*<sub>34</sub>, *P* > 0.05). The annual mean condition factor for *C. tamandua* (0.73 ± 0.29) was not significantly different from that of *G. petersii* (0.69 ± 0.22) (*t* = 0.62, *df*<sub>34</sub>, *P* > 0.05).

## DISCUSSION

Length-weight relationships of fishes are often used to study the indication of fatness, general well-being or gonad development. It is also assumed that heavier fish of a given length are in better condition. Venu and Kurup (2003) noted that for an ideal fish, which maintain dimensional equality, the isometric value of *b* would be 3. The estimates of *b* values (2.062 – 3.235, *x* = 2.905 ± 0.63) obtained in this study fall within the limits reported by Largler *et al.* (1977), 2.998, King (1996a, 1996b) 3.012 & 2.912, Anibeze (2000) 2.153, Stergiou and Moutopoulos (2001) 2.989, Venu and Kurup (2003) 3.021 and Ezenwaji and Inyang (1998) 2.970. The mean value, 2.905 is approximately 3 thus indicating isometric growth pattern.

Information on condition factor (*K*) is relevant for ascertaining the fish optimum environmental requirements, feeding regime and stocking density (Tsadu and Adebisi, 1997). The result of the present study also indicated that among the mormyrid species studied, *M. rume* had the best condition (1.17 ± 0.59) with about 55.6 % of the male and 50% of the female attaining a condition factor above 1.0 while *G. petersii* was in the worst condition (0.69 ± 0.22) with 94.4% of the males and 77.7 % of the females attaining condition factor less than 1.0.

There was no significant difference between the mean condition factor of the males and females of all the mormyrid species. The non-significant difference noted between the mean condition factors of males and females in all the Mormyrid species probably indicates that not much energy was diverted by the females during breeding activities.

This result is similar to the report of Olatunde (1978) on the male and female Schilbeid species of lake Kainji Nigeria and Ikomi (1996) for *Brienomyrus longionalis* in the upper Warri River Nigeria but differed from Anibeze and Inyang (2000) report for male and female *Heterobranchus longifilis* from Idodo River, Nigeria, and Fawole and Adewoye (2002) report for male and female *Clarias gariepinus* in Oba reservoir, Ogbomosho Nigeria.

Except for *G. petersii*, there was seasonality difference in the condition factor among the combined sexes of the mormyrid species. This agrees with the report of Anibeze and Inyang (2000) on *H. longifilis* at Idodo River but contrasted the report of Ezenwaji and Offiah (2003) for *Pellonula leonensis* of Anambra river, Nigeria.

## REFERENCES

- ANIBEZE, C. I. P. and INYANG, N. M. (2000). Oocyte structure, fecundity and sex ratio of *Heterobranchus longifilis* (Valenciennes 1940) in Idodo river basin (Nigeria) with comments on the breeding biology. *Journal of Aquatic Sciences*, 15: 59 – 61.



- BAGENAL, T. B and TESCH, F. W. (1978). Age and growth. Pages 101 – 136. *In*: BAGENAL, T. B. (Editor). *Methods of Assessment of Fish Production in Freshwater*. Blackwell Scientific Publication, Oxford.
- EZENWAJI, H. M. G. (2004). Length – weight relationships of fishes from Anambra river, South-Eastern Nigeria. *Animal Research International*, 1(1): 1 – 6.
- EZENWAJI, H. M. G and INYANG N. M. (1998). Observation of the biology of *Clarias agboyiensis* Sydenham, 1980 (Osteichthyes: Clariidae) in the Anambra flood river system, Nigeria. *Fisheries Research*, 36: 47 – 60.
- EZENWAJI, H. M. G and OFFIAH, F. N. (2003). The biology of *Pellonui leaonensis* Boulenger, 1916. (Osteichthyes: Clariidae) in Anambra river Nigeria. *Bio-Research*, 1(2): 33 – 50.
- FAWOLE, O. O. and ADEWOYE, S. O. (2002). Aspects of the biology of *Clarias gariepinus* (Burchell, 1822) in Oba reservoir, Ogbomosho, Nigeria. *The Zoologist*, 2(1): 144 – 156.
- GOSSE, J. P. (1984). Mormyridae. Pages 63 – 122. *In*: DAGET, J., GOSSE, J. P. and THYS VAN DEN AUDENAERDE, D. F. E. (Eds). *Checklist of freshwater fishes of Africa (CLOFFA)*. ORSTOM, Paris and MRAC Tervuren.
- IKOMI, R. B. (1996). Studies on the growth pattern, feeding habits and reproductive characteristics of the Mormyrid *Brienomyrus longianalis* (Boulenger, 1901) in the Upper Warri River, Nigeria. *Fisheries Research*, 6:187 – 198.
- ITA, E. O. (1978). An analysis of fish distribution in Kanji Lake, Nigeria. *Hydrobiologia*, 58: 233 – 244.
- KING, R. P. (1989). Distribution, abundance, size and feeding habits of *Brienomyrus brachyistius* (Gill 1862). Teleostei Mormyridae in a Nigerian rainforest stream. *Cybiu*, 13(1): 25 – 36.
- KING, R. P. (1996a). Length – weight relationship of Nigeria freshwater fishes. *Naga ICLARM Quarterly*, 19(3): 49-52.
- KING, R. P. (1996b). Length-weight relationship of Nigerian coastal water fishes. *Naga ICLARM Quarterly*, 19(4): 53-58.
- LARGLER, K. F., BARDACK, J. E., MILLER, R. R. and PASSION, D. R. M. (1977). *Ichthyology* (2<sup>nd</sup> ed). John Wiley and Sons, New York. 506 pp.
- LOWE-McCONNEL, R. H. (1972). *Freshwater fishes of the Volta and Kainji lakes*. University Press Accra, Ghana, 284 pp.
- OLAOSEBIKAN, B. D. and RAJI, A. (1998). *Field guide to Nigerian fresh water fishes*. Federal College of Freshwater Fisheries Technology, New Bussa, 106 pp.
- OLATUNDE, A. A. (1978). The food and feeding habits of *Eutropius niloticus* (Ruppell), Family Schilbeidae (Osteichthyes: Siluriformes) in Kainji, Nigeria. *Hydrobiologia*, 57: 197 – 207.
- REED, W., BURCHARD, J., HOPSON, A. J., JENNES, J. and YARO, I. (1967). *Fish and Fisheries of Northern Nigeria*. Ministry of Agriculture, Northern Nigeria, 220 pp.
- STERGIOU, K. I. and MOUTOPOULOUS, D. K. (2001). A review of length-weight relationships of fishes from Greek marine waters. *Naga ICLARM Quarterly*, 24(1-2): 23 – 24.
- TSADU, S. M. and ADEBISI, A. A. (1997). Condition indices of pond raised and wild population of African Catfish *Clarias gariepinus* (Burchell, 1822) (Pisces - Clariidae) in Plateau and Niger States, Nigeria. *Journal of Aquatic Science*, 12: 1 – 7.
- VENU, T. J. and KURUP, B. M. (2003). Length-weight relationship of some deep sea fish inhabiting the continental slope beyond 250 depths along the West Coast of India. *Naga ICLARM Quarterly*, 26(2): 17 – 21.
- VICTOR, R. and TETTEH, J. O. (1988). Fish communities of a perturbed stream in southern Nigeria. *Journal of Tropical Ecology*, 4: 49 – 59.

## MOSQUITO CONTROL STRATEGIES IN ISHIAGU RURAL COMMUNITIES: IMPLICATIONS TO PUBLIC HEALTH

<sup>1</sup>NDUKA, Florence Onyemachi and <sup>2</sup>NWOSU, Eugene Chidebem

<sup>1</sup>College of Biological Science, Abia State University Uturu, Nigeria.

<sup>2</sup>Pest Management Technology Department, Federal College of Agriculture, Ishiagu, Ebonyi, State, Nigeria.

**Corresponding Author:** Nwosu, E. C., Federal College of Agriculture, Ishiagu, Ebonyi State, Nigeria.

### ABSTRACT

*Mosquito control strategies adopted by the residents of two Ishiagu Communities (Okue and Ihie) were investigated. 53 households in Okue and 54 in Ihie were sampled. Simple structured questionnaires were prepared and administered. Percentages and chi square ( $\chi^2$ ) test of significance was employed in analysing the data. The result revealed that 42.9 % of the residents of the two communities used insecticides, 25.2% used mosquito coils, 6.5% used smoke of local herbs and 5.6% were insensitive to mosquito nuisance and careless about mosquito control. Choice of control measures by residents of the two communities were not statistically significant ( $P > 0.05$ ). Occupational related preferences revealed that farmers ranked highest in the use of insecticides and coils (21.5%) each. 13.1% of teachers use insecticides and 3.7% use nettings. Statistical analysis of the occupational related choice patterns was significant ( $P < 0.05$ ).*

**Key Words:** Mosquito, Rural Communities, Control, Implications, Public Health.

### INTRODUCTION

Mosquitoes have through human history constituted a problem to man and animals. About 60 different species of mosquitoes are found World wide (Crans and Farida, 2004). Of these species, members of the *Anopheles*, *Culex*, *Aedes*, *Hemagogus* and *Mansonia* complexes are important pests in Nigeria (Igbinosa, 1990).

Mosquitoes develop in pools of water formed by rainy season storms and permanent swamps of the wetlands. Nwoke *et al* (1993) observed that septic tanks also formed breeding sites for mosquitoes such as *Aedes aegypti*, *Anopheles vitatus*, *Culex horridus*, *Culex cinereus*, *Culex pipiens quinquefasciatus* and *Culex tigripes*. Other breeding sites for mosquitoes are water pots, discarded cans, plastic containers, discarded automobile tyres and notches in forest trees (Tonye, 1978).

Mosquitoes not only inflict biting pains on man but also suck human blood and transmit disease pathogens. Sutherland and Crans (2004) observed that the female mosquito bites humans and animals because they need blood proteins for the development of eggs. The males are short-lived, do not suck blood but nectars and plant juices, and die soon after mating. The haematophagous habit of the female mosquitoes is of public health importance. Various parasitic and viral diseases have been successfully transmitted through the biting mosquitoes. *Wuchereria bancrofti* and *Brugia malayi* which cause lymphatic filariasis in humans (WHO, 1991) are transmitted by members of the *Aedes*, *Culex* and *Mansonia* complexes. Yellow fever and dengue viruses are equally transmitted by these mosquitoes (Rappole *et al*, 2004; WHO, 1994).

The need to control mosquitoes becomes paramount when its threats to public health are

considered. In the developed parts of the World, Organised Mosquito control strategies are carried out by various agencies supported by the Government. Their efforts are channelled towards water management, biological control agents and the use of insecticides in controlling the larvae and adults. Apart from organised approaches, Sutherland and Crans (2004) posited that residents can help significantly by controlling mosquitoes around their homes. The method adopted by each family may differ depending on their economic standing as well as awareness. This work therefore seeks to identify the various mosquito control strategies adopted by residents of Ishiagu communities, and to discover any occupational related preferences among the control strategies identified.

### MATERIALS AND METHODS

**Study Area:** Ishiagu is found in Ivo Local Government Area of Ebonyi State. It is located on a low-lying marshy Riceland of South Eastern Nigeria and usually inundated by flood during the rainy seasons. Raised surfaces that are free from inundation form settlement points with the result that homes are surrounded by marshy flood lands which may account for the explosion of mosquito populations during the rains.

Ishiagu is made up of three autonomous communities (Ihie, Okue and Ishiagu). Two autonomous communities Ihie and Okue were selected for the study. Standard questionnaires indicating various methods of controlling mosquitoes at home were prepared and administered to 54 households in Ihie and 53 in Okue communities. Simple percentages and chi square test of significance were employed in the result analysis.

## RESULTS AND DISCUSSION

The result in Table 1 revealed that spraying of insecticides was the preferred choice (43.0%) followed by the use of mosquito coils (25.2%) and the use of nettings (19.6%). The least preferred control method was the use of smoke from local herbs (5.6%). It was also observed that about 6.5% of the people in the two communities do not care about mosquito nuisance hence they do nothing to control them. The spraying of insecticides was the most preferred mosquito control approach in Ihie (24.3%) and Okue (18.7%) communities. The use of mosquito coils was predominant in Okue (15.0%). More people in Ihie used netting (11.2%) than in Okue (8.4%). More residents of Okue were insensitive to mosquito control activities (3.7%) than Ihie (2.8%). The observed mosquito control strategies in both communities were not statistically significant ( $P>0.05$ ), indicating a high level of knowledge and practise of mosquito control in both communities.

**Table 1: Distribution of respondents according to the control methods adopted in the two communities**

Control method	Ihie	Okue	Total
Use of Netting	12 (11.2)	9 (8.4)	21 (19.6)
Mosquito Coils	11 (10.3)	16 (15.0)	27 (25.2)
Insecticide Spraying	26 (24.3)	20 (18.7)	46 (42.9)
Smoke of herbs	3 (2.8)	4 (3.7)	7 (6.5)
Use nothing	2 (1.9)	4 (3.7)	6 (5.6)
Total	54 (50.5)	53 (49.5)	107 (100)

Number in parenthesis = %

The percentage of those indifferent to the use of any form of insect control in both communities was low. This group poses a potential danger to others. Based on the transfer of malaria parasites from them to those using insect control measures. The high preference to the use of Insecticide (43.0%) in both communities is recommendable. Pesticides are potent in the reduction of mosquito populations, the problem of emergence of resistant mosquitoes (Okon *et al.*, 1992) should be seriously considered.

Table 2 revealed that of the 107 respondents, 15(14.0%) were Traders (TD), 10(9.4%) civil servants (CS), 20(18.7%) were Teachers (TC) and 62(57.9%) were farmers (FM). Choice preference among the farmers tilted towards the use of insecticides and mosquito coils, 23(21.5%) respectively for both. The Use of netting 6 (5.6%) and smoke from local herbs 5 (4.7%) were unpopular among farmers. Although the percentage of farmers using mosquitoes strategies were high, few farmers, 5(4.7%) were insensitive about mosquito nuisance and control. Traders showed equal preference for the use of netting and insecticides with 6(5.6%) for both cases. Teachers preferred mostly the use of insecticides 14(13.1%) followed by netting 4(3.7%). The civil servants preferred mostly netting 5(4.7%) followed by insecticides 3(2.8%). The use of smoke

of local herbs was unpopular among teachers and civil servants with zero preferences. Analysis of the preference pattern among the various occupational groups was significantly different ( $p<0.05$ ).

**Table 2: Occupational related choice preference among the respondents**

Control methods	TD	FM	TC	CS	TOTAL
Use of Netting	6 (5.6)	6 (5.6)	4 (3.7)	5 (4.7)	21 (19.6)
Mosquito Coils	2 (1.9)	23 (21.5)	1 (0.9)	1 (0.9)	27 (25.2)
Insecticide spraying	6 (5.6)	23 (21.5)	14 (13.1)	3 (2.8)	46 (43.0)
Smoke of herbs	1 (0.9)	5 (4.7)	0 (0.0)	1 (0.9)	7 (6.5)
Use Nothing	0 (0.0)	5 (4.7)	1 (0.9)	0 (0.0)	6 (5.6)
Total	15 (14.0)	62 (57.9)	20 (18.7)	10 (9.4)	107 (100)

Number in parenthesis = %, TD = Traders, FM = Farmers, TC = Teachers, CS = Civil Servants

The preference of farmers to the use of insecticide is related to their knowledge and practise of using insecticides to control pest in their farms. This positive attitude influences their choice of mosquito control strategy. The availability of varieties of pesticides in the local market coupled with large number of clients may have helped to reduce the service cost charged by the local pest control agents. With the pesticides becoming competitively cheap, many people became interested in its use in homes for mosquito control.

**Recommendations:** Workshops should be organized for the rural population on the proper use of insecticides and coils, and their associated risks. Research into the characterization of the active ingredient in the identified local herbs is needed. The use of netting which ranked third should be popularised among Ishiagu residents, as it is cheap and last longer. Organised mosquito control programmes should be encouraged. Both the Ministry of Health and Environmental Protection Agency (Ebonyi State) should be courageous enough to fashion out feasible programmes in the area. Private participation in organised mosquito control activities should be encouraged and supported.

## REFERENCES

- CRANS, J. W. and FARIDA, M. (2004). *Controlling mosquitoes around the home*. <http://WWW-rei.rutgers.edu/insects/house.Htm>. Pages 1 – 4.
- IGBINOSA, I. B. (1990). Parasitic Arthropods in *Parasitology for medical students*. Pages 48 – 49. In: EZIGBO J. C. ed. New Frontier Publishers Ltd. Lagos
- NWOKE, B. E. B., NDUKA, F. O., OKEREKE, O. M. and EHIGIBE, O. C. (1993). Sustainable Urban development and human health, septic tank

- as major Breeding South Eastern Nigeria. *Applied Parasitology*, 34:1 – 10.
- RAPPOLE, J. H., DERRICKSONS, S. R. and HUBALEK, U. U. (2000). Migratory birds and spread of west Nile virus in the Western Hemispheres. *Emerging infect Disease*, 6:319 – 328.
- RAILINGS J. A., HENDRICKS, K. A., BRUGES, C. R., CAMPMAN, R. M., CLERK, G. G., TABONY, L. J., and PATTERSON, M. A. (1998). Dengue Surveillance in Texas. *American Journal of Tropical Medicine and Hygiene*, 59: 95 – 99.
- SUTHERLAND, D. J. and CRANS, W. J. (2004). *Mosquitoes in your life*. <http://www.ric.rutgers.edu/inscts/moslife.htm> Pages 1 – 5.
- TONYE, G. O (1978) The Breeding site preference of mosquitoes in Ibadan, Nigeria. *Nigeria Journal of Entomology*, 1(3): 71 – 80.
- OKON, O. E., BRAIDE, E. I., EKONG, R. G. and ITAM, I. H. (1992). Transplacental transmission of malaria in Cross River State. *The Nigerian Journal of Parasitology*, 13: 9 – 12.
- WHO (1991). *Lymphatic filariasis and anchocerciasis*. Tropical Disease progress in research 1989 – 1990. Tenth Program Report. NUDP/ WHO /TDR. 49 - 58 pp.
- WHO (1994) *Parasites of Poverty*. Highlights from a seminar on tropical Diseases, Society and the environment. Geneva, Switzerland.

## A MODULE FOR THERMAL PEST CONTROL IN STORED RAW MATERIALS USED IN FEED MILLS / FOOD MANUFACTURING INDUSTRIES

<sup>1</sup>MGBEMENE, Chigbo Aghaegbusi, <sup>1</sup>AGWUBILO, Ikenna and <sup>2</sup>OKAFOR, Fabian Chukwuemenam

<sup>1</sup>Department of Mechanical Engineering, Faculty of Engineering, University of Nigeria, Nsukka

<sup>2</sup>Department of Zoology, Faculty of Biological Sciences, University of Nigeria, Nsukka

**Corresponding Author:** Mgbemene, C. A., Department of Mechanical Engineering, Faculty of Engineering, University of Nigeria, Nsukka.

### ABSTRACT

*Pests have continued to be problematic in warehouses of most feed mills and food manufacturing industries. Pests are heterogeneous both in space and time, creating gradients and patterns depending on the prevailing environmental variables. Pest control efforts have utilized manipulations of these variables. This project is on a module for thermal control of pests using the hitherto waste steam from the industries. The module is an engineering contraption, which uses steam to raise temperature within it to insecticidal levels (above 45°C). This causes halt in development and protein denaturing (leading to mortality) of the pests (eggs, larvae and adults). This applied heat also toasts the material inside the module thereby improving its quality. The module consists of a rotary drum with steam passing through convoluted tube (without coming in contact with the handled material), with a capacity of 2 tons/hr of commodities. It accepts steam at 130°C and discharges it at 110°C. The steam pressure (permissible) is 2.7 bars. Steam velocity is 8m/s. The volume of the drum is 4 m<sup>3</sup>. A two- screw- 90 degrees-lead –counter screw-rotation type baffle is installed to achieve even distribution of heat on the material within the drum. The module is efficient, effective and useful in any integrated pest management effort.*

**Keywords:** Module, Thermal Pest Control, Stored Raw Materials, Feed Mills, Food Manufacturing Industries

### INTRODUCTION

Stored products in the tropics suffer serious damages due to pest attacks. According to Ogunlana (1976) up to 27.7% losses have resulted from insect damage to maize stored in Nigeria without pest control. Even at 4% level such losses translate to an annual loss of ₦300, 000,000 or 2 million tons. Such Figure for cowpea is 4.5% loss; for fish it is £10 million sterling (Toye, 1976). The most common pests encountered are: *Tribolium castaneum*, *Tribolium confusum*, *Sitophilus oryzae* and *Callosobruchus maculatus* (Osuji, 1985). The study of pest control has grown vastly since the introduction of the first synthetic insecticide (DDT) in 1940 (Kumar, 1986). Most advances in our knowledge in this field have taken place in the last sixty years. The literature on the subject is expanding rapidly.

In order to consider using economically feasible control measures against pests reliable information is needed on losses as a result of pest attacks. Stern et al; (1959) used two terms in discussing control measures against pests namely: economic injury level and economic damage. According to Talpaz and Fristic (1975) another term "economic threshold" is a dynamic measure which may vary with infestation level, cost of control, and time of assessment. In practice, all these boil down to the fact that we need to know the accurate estimates of pests population levels that affect production and yield. Stern (1973) concludes that it is important to know the yield per pest density ratio so as to ascertain when control is inevitable. In most of our warehouses where raw materials are stored the

pest density ratio is so high that instituting control must be instituted one way or the other. Control is often by chemical and by physical means. Physical control is divided into physical methods and environmental manipulation.

Physical control or physical elimination of the pests often involves the alteration of the environment to make it inimical to the pest. Physical barriers are effective in the control of flour beetles that affect most raw materials like Soya bean cakes, palm kernel cake and cassava flours used in feed mills (Dayakar and Ray, 1998). As pests are part of the major production bottlenecks in the feed and food related industries, practitioners always take the easy way out by using insecticides but because of the side effects of these insecticides, physical methods would always serve as suitable control alternatives.

Hand picking of pests is hardly practical in any large-scale control effort but removal of foci of infestations in storehouses is known to greatly affect, reduce and sometimes eliminate the insect population from the stored commodities. This removal of foci of infestations may be possible using various devices. In his studies, Ebeling (1971) listed such devices as jarring, destruction of egg masses and removal by a strong stream of water in considering field pests. For stored products and with a through knowledge of the ecology of the pests, it is possible to develop novel physical control measures. It is against this background that this study was initiated to use thermal control as a means of control in industries. As many industries have steam that is mostly wasted, this would be harnessed as the source heat. In doing this, a module is designed and fabricated that use the

known biological information on raw materials and pests to effect a cost-effective control of pests in stored raw materials.

### The Role of Environmental Manipulation in Pest Control:

Banks (1976) summarized the use of ecological factors against insects, even though the use of this is not very widespread. The use of physical control methods is growing in importance in stored commodities in view of the problems posed by the increasing use of insecticides. Busvine (1968) says that time is ripe for biologists to use engineers to develop modern technology for pest control that will reduce cost, be environmentally friendly and effective. This presentation is our modest effort in this direction.

Most industries that store commodities that are attacked by pests produce steam as waste products of their boilers. The steam can be channeled to two processes both of which will help in pest control. These are: 1. Dehydration. 2. Raising the temperature;

**Dehydration:** Most set ups use dust desiccants which kill insects by destroying the water proofing properties of the insect cuticles. This permits a lethal rate of water loss from the insect body. The use of relative humidity also is very efficient in stored products. The death of insects is largely due to excessive water loss from insect bodies caused by prolonged opening of their spiracles. Howe (1965) noted that many storage pests cannot reproduce at ambient relative humidity of 50% and Banks (1976) noted that the possibility of producing an atmosphere dry enough to be insecticidal but which is only out of equilibrium with the commodity merits further research.

**Temperature:** Temperatures as high as 45°C normally used in artificially drying grains for storage, for feed and food processing facilities are lethal to insects. Efficient portable grain drying equipment has been used for the purpose of elevating temperatures to control pests to stored commodities. This project has designed a module capable of generating high temperature over a short period to effect insecticidal action in stored commodities prior to use in product processing.

## MATERIALS AND METHODS

### Engineering Theory and Design

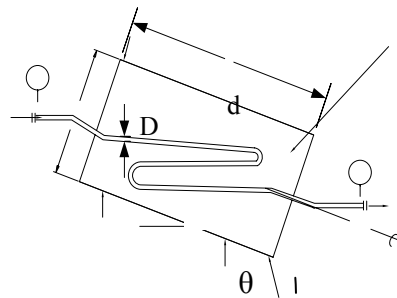
**Type:** Rotary drum steam-through-tube module.

**Working Fluid:** Wet saturated steam.

**Material Handled:** Grains, Cakes, Flours.

**Capacity:** 2 tons of grain/other commodities. The module was designed as a rotary shell and tube heat exchanger with the working fluids as wet saturated steam making the tube pass and air making the shell passes.

From an existing boiler, wet saturated steam at 130°C and 2.7 bars is channeled into the drum. The steam is expected to heat the air in the drum to 100°C before exiting at 110°C. The choice of steam at 130°C is to avoid condensation within the tube. Again, the exit temperature was set at 110°C for the same reason. In event of condensation, the tilt of the drum will force the condensate to drain out (Figure 1). Through the grain inlet; the grains are introduced into the drum. The drum, which has baffles inside, rotates at 4 rpm. The baffles mix the commodities inside the drum and ensure that each commodity comes into contact with the 100°C air. The enclosed raw materials will stay for 30 minutes in the drum before being discharged.



**Figure 1: The schematic diagram of the module**

### Definition of Parameters

$C_p$  = Specific heat of fluid (kJ/kg. K)  
 $\mu$  = Dynamic viscosity (kg/m.s)  
 $k$  = Thermal conductivity (kW/m.K)  
 $p$  = Pressure (bar, Pa)  
 $Q$  = Quantity of heat transferred (W)  
 $\rho$  = Density (kg/m<sup>3</sup>)  
 $Pr$  = Prandtl number  
 $Re$  = Reynolds number  
 $Nu$  = Nusselt number  
 $m$  = Mass flow rate (kg/s)  
 $h$  = Heat transfer coefficient (kW/m<sup>2</sup> K)  
 $\theta$  = Drum tilt angle (°)  
 $L$  = Length of drum (m)  
 $D$  = Diameter of drum (m)  
 $d$  = Pipe diameter (m)  
 $d_i$  = Inside pipe diameter (m)  
 $N$  = Drum rotation (rev/min)  
 $T_1$  = Steam inlet temperature (°C)  
 $T_2$  = Steam outlet temperature (°C)  
 $R_1, R_2$  = Support reactions (N)  
 $T_{air}$  = Temperature of air inside drum (°C)  
 $v$  = Velocity (m/s)  
 $\sigma_c$  = Circumferential stress (MPa)  
 $t$  = Sheet thickness (m)  
 $r$  = Radius (m)

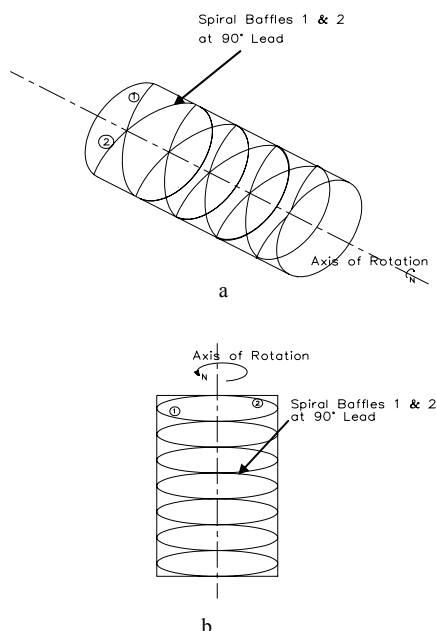
**Design Determinants:**  $D$ ,  $d$ ,  $L$ , steam flow rate, inlet and outlet temperatures of steam, heat transfer from steam, drum rotation speed, duration of grain in the drum, size of motor, entry and discharge of grain, gear parameters, number of pipe turns, baffle design, drum tilt, load borne by the drum, materials to be used, fabrication and cost.



**The Design:** The density of sample commodity e.g. – rice was determined to be  $865 \text{ kg/m}^3$ . From this, the volume to contain 2 tons of raw materials was determined ( $4\text{m}^3$ ) and the length followed suit as 3 metres.  $D$  was then determined.  $d$  was chosen from the table of available pipe diameters. The number of pipe turns was determined to be 9. Therefore, the pipe length used for the design was 27 metres.

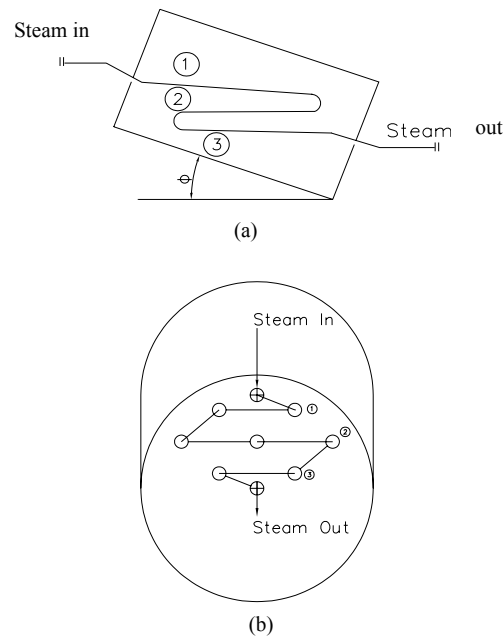
**Particulars:** Temperature of steam at entry =  $130^\circ\text{C}$ ; Temperature of steam at discharge =  $110^\circ\text{C}$ ; Steam pressure = 2.7 bars; Steam velocity = 8 m/s; Final temperature of air in drum =  $100^\circ\text{C}$ ; Length of drum = 3 m; Volume of drum =  $4\text{m}^3$

**Thermal Considerations:** Having selected the following parameters:  $d = 0.0779\text{m}$ ,  $\rho_{130}$  = density of steam at  $130^\circ\text{C} = 0.5542 \text{ kg/m}^3$ ,  $v = 8\text{m/s}$  and  $p = 2.7$  bars, the mass flow rate of the steam was calculated as 0.21 kg/s. The heating load was calculated as  $Q = 3487.6 \text{ W}$  for the 27m length of pipe. This means that 3487.6 Joules of heat energy is given out per second from steam for heating of air to  $100^\circ\text{C}$ . From energy balance, the mass flow rate of air was 0.0495 kg/s. From this, the velocity of air inside the drum was calculated to be 0.012m/s. This translated to drum rotation of approximately 4.0 rpm considering losses and using a generous factor of safety. For effective heating of the raw materials, the drum should actually rotate at a very slow pace. This will effect good heat transfer from steam to grain. Good mixing of grain was achieved by the use of baffles attached inside the drum shell. A 2-screw-n/2-lead-counterscrew-rotation type baffles were used, (Figure 2 a, b).



**Figure 2 (a) and (b): The baffles**

The heating tubes were arranged in a three-layer pattern. This is to achieve even heat distribution within the drum, (Figure 3 a, b).

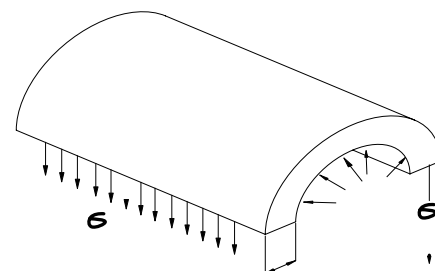


**Figure 3 (a) and (b): The heating tube arrangement**

## RESULTS

**Structural Analysis:** The weight of the whole structure was approximated to be the weight of the drum and its accessories plus the weight of the grain. The weight of the grain is already known i.e. 2000 kg but the weight of the drum is yet to be known. This can be calculated if the thickness of the end plates of the drum and the thickness of the steel sheet are known.

The end plates were made of 12 mm thick steel plates. The thickness of the steel sheet was determined by analyzing the drum as a thin walled pressure vessel see Figure 4.



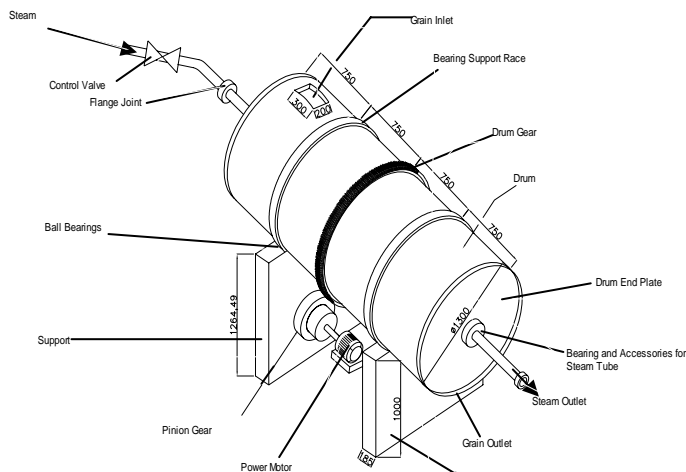
**Figure 4: Thin walled pressure vessel**

In stress analysis,  $\sigma_c$  must not exceed the permissible working stress  $\sigma_{\max}$ . For this steel,  $\sigma_{\max}$  is 241 MPa. Using a factor of safety of 4.0 and  $\sigma_c = \sigma_{\max} / 4$ , thickness of sheet was determined to be 3mm. Finally, the working load was found to be approximately 30 kN. From the analysis, we have the following specifications:

(a) **Prime mover:** 3.5 hp, 715 rpm, 220/400 V induction motor.

- (b) **Gear train:** Worm set/spur set.
  - (i)  $\phi 50$  mm driver motor worm.
  - (ii)  $\phi 130$  mm driven gear on the same shaft with
  - (iii)  $\phi 136$  mm pinion driving
  - (iv)  $\phi 1430$  mm spur (Drum).
- (c) **Support:** Simple support type.
- (d) **Bearings:** (i) Specialized tapered roller bearings on the supports.
  - (ii) Pipe bearing: Single-row 02-series deep-groove angular-contact ball bearings.Bore –  $\phi 90$  mm.

Figure 5 is a diagram of a fully assembled module.



**Figure 5: A fully assembled module**

## The Benefits of Control Using the Module

1. The steam to be used is from boiler operations for other production processes.
2. Highly skilled engineers and scientists are not really needed to operate the module.
3. Investment cost is high on acquiring the equipment but will be used for several years with minimal repairs.
4. The high temperature highly and adversely affects the biology and behaviour of the insects and dehydration reduced relative humidity of the operations.
5. The biological integrity of the commodities is retained and is intact.
6. Yield and profitability of production is increased.

## DISCUSSION

The module presented has succeeded in greatly harnessing the heat energy from the otherwise waste steam while leaving behind some sensible heat, which could be used for other operations. Thus, beyond the role as a veritable pest control contraption it is also an energy saving device.

Temperature is the single most important factor regulating life processes in nature. Other regulatory processes such as predation, disease and competition act on the residual populations, which have survived the effects of temperature. Survival and abundance can be ecologically predicted by

relating known levels of temperature tolerance to the severity of either the very low temperatures of refrigeration or the high temperature such as the one generated by this module or the other such heat generating systems to periods of exposure

Scientists are well aware of the problem of invertebrate survival at sub zero temperatures and this subject has attracted the most attention. From such studies we now know that insects exhibit one of two seemingly immutable strategies to survive such temperature variations but not high temperatures. Insects switch between tolerance and intolerance.

The degree to which they do this, gives the indication of the ecological relevance of the spectrum of the adaptations of which the operations of this module deals a death blow.

Chemical engineers developed theories, which describe and predict how temperature changes cause hygroscopic compounds and lipids with some porous solid in insects, to dry and these theories also are used to explain drying behaviour of insects subjected to the operations of this equipment. It also explains drying found to occur within the raw materials, thus extending their shelf life.

Any interested parties are free to access the designers of this module for its construction for their use. It is cost effective, efficient and easy to operate. It provides a useful tool in an integrated pest management in the industries.

## REFERENCES

- ANON (1953). *Alternation Current Electric Motor*. Brooks Motor Limited, Huddersfield and Bernsley, England.
- BANKS H. J. (1976). Physical Control of Insects: Recent Developments. *Journal of Australian Entomological Society*, 15(1): 89 – 100.
- BUSVINE, J. R. (1968). The Future of Pest Control. Part 2. Alternative to Insecticides. *PANS*, 14(3): 318 – 328.
- DAYAKAR, S. AND RAY, S. N. (1998). Effect of Physical Barriers on Growth and Development of *Tribolium castaneum* (Herbst) on Wheat. *Insect Environment*, 4: 1 – 9.
- EBELING, W. (1971). Sorptive Dusts for Pest Control. *Annual Review of Entomology*, 18: 259 – 280.
- HOLMAN, J. P. (1998). *Heat transfer*. Tata-McGraw-Hill Book Company, New Delhi, 553 – 597 pp.
- HOWE, R. W. (1965). A Summary of Estimates of Optimal and Minimal Conditions for Population Increase of Some Stored Products Insects. *Journal of Stored Product Research*, 1: 177 – 184.

- KUMAR, R. (1986). *Insect Pest Control with Special Reference to African Agriculture*. ELBS Publications. The Camelot Press, Southampton, 298 pp.
- OGUNLANA M. C. (1976). Opening Address. Pages 118 – 123. *In: Proceedings of 9<sup>th</sup> Annual Conference and 10<sup>th</sup> Anniversary Celebration. Entomological Society of Nigeria*.
- OSUJI, F. N. C. (1985). *Outlines of Stored Products Entomology for the Tropics*. Fourth Dimensions Publishing Co. Ltd Enugu, Nigeria.
- POPOV, E. P., NAGARAJAN, S. and LU, Z. A. (1976). *Mechanics of Materials S. I Version*. Prentice-Hall, Englewood, New Jersey.
- ROGERS, G. F. C. and MAYHEW, Y. R. (1980). *Engineering Thermodynamics; Work and Heat Transfer, 3<sup>rd</sup> Edition*. ELBS/Longman, Hong Kong.
- SHIGLEY, E. S. and MISCHKE, C. R. (1989). *Mechanical Engineering Design. 5<sup>th</sup>. Edition* McGraw-Hill Book Company.
- SOLBERG, H. L, CROMER, O. C. and SPALDING, A. R. (1954). *Elementary Heat Power, 2nd. Edition* John Wiley and Sons Incorporated, New York.
- STERN, V. M. (1973). Economic Thresholds. *Annual Review of Entomology, 18*: 259 – 280.
- STERN, V. M., SMITH, R. F., VAN DEN BOSCH, R. and HAGEN, K. J. (1959). The Integration of Chemical and Biological Control of the Spotted Alfalfa Aphid. Part 1: The Integrated Control Concept. *Hilgardia, 29*: 81 – 101.
- TALPAZ, H. and FRIBIE, R. E. (1975). An Advanced Method for Economic Threshold Determination: A Positive Approach. *Southern Journal of Agriculture Economics, 12*: 19 – 25.
- TOYE S. A. (1976). The Role of Entomology in the Economic Development of Nigeria. Pages 3 – 10. *In: Proceedings of 9<sup>th</sup> Annual Conference and 10<sup>th</sup> Anniversary Celebration. Entomological Society of Nigeria*.
- VAN AMERONGEN, C. (Translation 1967). *The Way Things Work. Vol. 2*. Simon and Schuster. New York.
- WALTON, H. (1968). *The How and Why of Mechanical Movements*. Popular Science Publishing Company and E. P. Dutton Company Incorporated, New York.

# ANIMAL RESEARCH INTERNATIONAL

Volume 3 Number 1 April 2006

CONTENTS	PAGES
1. PERFORMANCE OF SHEEP GRAZING <i>Brachiaria decumbens</i> , <i>Panicum maximum</i> and <i>Pennisetum purpureum</i> IN COMBINATION WITH <i>Gliricidia sepium</i> - OSAKWE, Isaac Ikechukwu and NWAKPU, Petrus Emeka	399 – 402
2. SUSCEPTIBILITY Of <i>Aedes aegypti</i> PUPAE TO NEEM SEED KERNEL EXTRACTS - UMAR, Abba., KELA, Santaya Larit and OGIDI, Jonathan Asadabe	403 – 406
3. SUSCEPTIBILITY OF MOSQUITO LARVAE TO CONVENTIONAL INSECTICIDES IN A TROPICAL ARID ECOSYSTEM - UMAR, Abba, KELA, Santaya Larit and ABDULRAHMAN, Hauwa Tati	407 – 409
4. FOOD AND FEEDING HABITS OF <i>Campylomormyrus tamandua</i> IN ANAMBRA RIVER, NIGERIA - NWANI, Christopher Didigwu., EYO, Joseph Effiong and UDEH, Emmanuel Fame	410 - 414
5. NEURAL TISSUE AND COMPLETE REGENERATION OF THE TAIL OF THE GEKKONID LIZARD, <i>Hemidactylus flaviviridis</i> - NDUKUBA, Patrick Ifeanyichukwu	415 – 418
6. PREVALENCE OF CAPRINE STRONGYLE INFECTION AND THE DIAGNOSTIC EFFICACY OF SOME MEDIA FOR FAECAL CULTURE AND NEMATODE LARVAL RECOVERY FROM GOAT FAECES - NWOSU, Chukwunyere Okwudiri., IWUOHA, Chimaobi Lawrence., TORRU, Churchill and MOHAMMED, Ali	419 – 421
7. OCTANOL/WATER PARTITION COEFFICIENT AND BIOACCUMULATION INDEX OF BONNY LIGHT CRUDE OIL IN CAT FISH <i>Clarias agboyiensis</i> IN LABORATORY-DOSED SEDIMENTS - UBANI, Chibuike Samuel., ONWURAH, Ikechukwu Noel Emmanuel and ALUMANAH, Eddy	422 – 425
8. LENGTH-WEIGHT RELATIONSHIP AND CONDITION FACTOR OF FOUR MORMYRID SPECIES OF ANAMBRA RIVER - NWANI, Christopher Didigwu., EZENWAJI, Henry Maduka Godfrey., EYO, Joseph Effiong and UDE, Emmanuel Fame	426 – 430
9. MOSQUITO CONTROL STRATEGIES IN ISHIAGU RURAL COMMUNITIES: IMPLICATIONS TO PUBLIC HEALTH - NDUKA, Florence Onyemachi and NWOSU, Eugene Chidebem	431 - 433
10. A MODULE FOR THERMAL PEST CONTROL IN STORED RAW MATERIALS USED IN FEED MILLS / FOOD MANUFACTURING INDUSTRIES - MGBEMENE, Chigbo Aghaegbusi., AGWUBILO Ikenna and OKAFOR, Fabian Chukwuemenam	434 – 438