

NOTES AND COMMENTS

A note on recent advances in the genetic characterization of Tilapia stocks in Lake Victoria Region

WILSON MWANJA, PAUL FUERST, AND LES KAUFMAN

WILSON MWANJA
Department of Zoology,
Makerere University,
P.O.Box 7062,
Kampala, Uganda.

LES KAUFMAN
Boston University, Marine Programme (Correspondence Author).

INTRODUCTION

Oreochromis esculenta, the original "ngege" is virtually extinct in Lake Victoria, and is limited to satellite lakes and reservoirs in the greater Lake Victoria region. *Oreochromis variabilis* can still be found in Lake Victoria and some satellite lakes in the Kyoga System, but in small numbers and only at a few localities (WANDERA and KAUFMAN, unpub. data). Little is known about the influence that species translocations have had on the genetic structure of these crucial fishery species, and even the source of the parent stocks for the introductions remain obscure. Genetic variability was examined within and among allopatric populations of three species in the tilapiine genus *Oreochromis*: *O. esculentus* (endemic to Lakes Victoria and Kyoga), and two exotic species introduced to Lake Victoria in the late 1950's to supplement the failing fisheries for native tilapiines, *O. niloticus* and *O. leucostictus*.

MATERIALS AND METHODS

Population samples were obtained (>10 individuals/species/locality) from Lake Victoria and eight satellite Lakes in the Victoria basin: Lakes Nabugabo, Kayugi, Kayanja, and Manywa in the Nabugabo System; Lake Kanyaboli in the Yala-Nzoia System, and Lakes Mburo, Kachira, and Kijanebalola in the Koki Lakes System. Of these, only Nabugabo contained Nile Perch (OGUTU-OWHAYO, 1993). Small bits of muscle tissue were removed immediately upon capture. Only those individuals that could be identified unambiguously in the field were taken for the study.

The tissues were placed in 95% ethanol in sample vials, and the alcohol changed after 1hr. DNA was extracted from the muscle tissue samples using a standard phenol/chloroform extraction procedure (SAMBROOKE, 1982). DNA samples were amplified through PCR using a Perkin-Elmer thermocycler using single short arbitrary 10-mer oligonucleotide primers. Amplification products were separated by electrophoresis in a 1.6% synergel agarose gel, stained with ethidium bromide, and viewed under ultraviolet light. Individual species were analyzed for species-specific markers (bands that occurred exclusively among individuals of a particular species, Table 1). Gametic diversity (Table 2) was calculated after LYNCH and MULLIGAN (1994), which is specific to the analysis of gene structure using RAPD data. Gene introgression (Table 4) was estimated in terms of the proportion of RAPD alleles characteristic of a given taxon that appeared in congener populations. Cladograms (using maximum parsimony) were constructed with the aid of the program PAUP 3.1, with analyses conducted under highly stringent conditions. The outgroup chose *Tilapia zillii*, a tilapiine cichlid assumed to be phylogenetically basal to the general *Oreochromis* and *Sarotherodon* based on the work of Trewavas and others (TREWAVAS, 1983).

RESULTS

All species exhibited a relatively high number of species-specific alleles, with *O. leucostictus* exhibiting the highest number followed by *O. niloticus*, and *O. esculentus* with the least

(Table 1). *O. niloticus* exhibited the highest mean within population gene diversity, and *O. esculentus* the lowest (Table 2). *O. esculentus* exhibited the highest degree of population subdivision, but statistically it did not differ significantly in this regard from *O. niloticus*, both of which displayed remarkably high levels of population distinctness.

O. leucostictus was unusual in its low degree of population subdivision, and *T. zillii* for its relatively high within-population genetic diversity (Table 3). All six of the *O. esculentus* populations examined exhibited evidence of *O. niloticus* alleles (Table 4). The most highly introgressed population was that of Lake Mburo, and the purest was Lake Kanyaboli. The three Nabugabo satellite Lakes and Lake Kachira showed similar, moderately high levels of introgression from *O. niloticus* into *O. esculentus*. Gene introgression from *O. esculentus* into *O. niloticus* was generally lower than the reverse. Lake Victoria *O. niloticus* showed little evidence of *O. esculentus* alleles, though Lake Nabugabo, where *O. esculentus* has been extirpated, displayed surprisingly high levels of introgression and retention of *O. esculentus* alleles.

Table 1. Total number of population-specific bands within the three species of the genus *Oreochromis*.

| | <i>O. esculentus</i> | <i>O. niloticus</i> | <i>O. leucostictus</i> |
|-----------------------------------|----------------------|---------------------|------------------------|
| <i>Population</i> | | | |
| Lake Kanyaboli | 6 | — | — |
| Lake Manywa | 4 | — | — |
| Lake Kijanebalola | 4 | — | — |
| Lake Kayugi | 9 | — | — |
| Lake Kayanja | 2 | — | — |
| Lake Kachira | 5 | 6 | 21 |
| Lake Mburo | 13 | 7 | 9 |
| Lake Victoria | — | 5 | 8 |
| Lake Nabugabo | — | 7 | 9 |
| Lake Edward | — | 9 | — |
| Lake Albert | — | 5 | — |
| <i>Species (total)</i> | 43 | 39 | 47 |
| <i>Total bands</i> | 140 | 115 | 105 |
| <i>Proportion of unique bands</i> | 0.31 | 0.34 | 0.45 |

Table 2. Estimates of mean within-populations gene diversity (H_w) for three species belonging to the genus *Oreochromis*.

| <i>Species</i> | <i>n</i> | H_w | <i>SE</i> | $Var H_w$ | $Var_I H_w$ | $Var_L H_w$ | $Var_p H_w$ |
|------------------------|----------|-------|-----------|-----------|-------------|-------------|-------------|
| <i>O. niloticus</i> | 5 | 0.213 | 0.031 | 0.00095 | 0.000038 | 0.000085 | 0.00083 |
| <i>O. esculentus</i> | 6 | 0.152 | 0.020 | 0.00042 | 0.000025 | 0.000035 | 0.00036 |
| <i>O. leucostictus</i> | 4 | 0.202 | 0.019 | 0.00034 | 0.000038 | 0.000071 | 0.00023 |

n = number of populations studied
 $Var H_w$ = sampling variance
 $Var_I H_w$ = sampling variance due to finite number of individuals
 $Var_L H_w$ = sampling variance due to finite number of loci
 $Var_p H_w$ = sampling variance due to finite number of populations

DISCUSSION

The genetic distinctness of both *O. esculentus* and *O. niloticus* populations is of an order normally associated with subspecies (Table 3). Even the comparatively low values found in *O. leucostictus* were higher than expected. We attribute this to founder effect, due either to natural or artificial seeding of these populations by a very few individuals in each case. Introgression between the endemic *O. esculentus* and the introduced *O. niloticus* is rampant. Gene flow has been predominantly, though not exclusively, from *O. escu-*

Table 3. Estimates of genetic diversity for within (H_w) and between (H_B) populations and Wright's measure of population subdivision, F_{ST} , among Tilapia populations from Lake Victoria Basin.

| <i>Species</i> | <i>n</i> | H_w | H_B | F_{ST} |
|------------------------|----------|-------|-------|----------|
| <i>O. niloticus</i> | 5 | 0.213 | 0.605 | 0.740 |
| <i>O. esculentus</i> | 7 | 0.152 | 0.613 | 0.801 |
| <i>O. leucostictus</i> | 4 | 0.202 | 0.539 | 0.686 |
| <i>Tilapia zillii</i> | 2 | 0.222 | — | — |

n = number of populations studied

lenticus into *O. niloticus*. It is distinctly possible that no pure stocks of *O. esculentus* are existent today. The best remaining hope lies in two disparate localities: the Nyumba ya Mungu Reservoir in Tanzania, where the source stock of *O. esculentus* may have been relatively pure, and various satellite Lakes of Lake Kyoga, where we have discovered astonishingly rich remnant communities resembling those of Lakes Victoria and Kyoga prior to the huge ecological changes of the past four decades. For centuries, *O. esculentus*

Table 4. Estimate of gene introgression based on proportions of species specific 'fixed' allele harbored by a congener.

| | <i>fixed alleles</i> | <i>O. niloticus</i> | <i>O. esculentus</i> |
|-----------------------|----------------------|---------------------|----------------------|
| <i>O. esculentus</i> | | | |
| <i>Lake Kanyaboli</i> | — | 6.72 | — |
| <i>Lake Kayugi</i> | — | 14.39 | — |
| <i>Lake Kayanja</i> | — | 12.94 | — |
| <i>Lake Manywa</i> | — | 13.73 | — |
| <i>Lake Kachira</i> | — | 14.11 | — |
| <i>Lake Mburo</i> | — | 35.27 | — |
| <i>O. niloticus</i> | | | |
| <i>Lake Victoria</i> | — | — | 0.91 |
| <i>Lake Mburo</i> | — | — | 6.67 |
| <i>Lake Nabugabo</i> | — | — | 21.10 |
| <i>Lake Kachira</i> | — | — | 8.20 |

was among the most prized food fishes in East Africa, and it was the staple fish on Lakes Victoria and Kyoga in pre-colonial and early colonial times (BALIRWA, 1992). This was on account not only of its abundance, but also its excellent taste, firm meat, and suitability for sun-drying. Nonetheless, *O. esculentus* was never taken up by aquaculture scientists during the "blue revolution" that led to the current popularity of other tilapiines as targets in aquaculture. Now that the species has disappeared from Lakes Victoria and Kyoga and there is a real possibility of its biological extinction, reconsideration of its status, and its future in East Africa is long overdue.

ACKNOWLEDGEMENTS

We would like to thank our colleagues of the Fisheries Research Institutes of Uganda (FIRI-Jinja) and Kenya (KMFRI-Kisumu) for all assistance accorded during this study. We are greatly indebted to fishermen for their cooperation and assistance during the collection of study specimens. This work was funded by grants from the National Agricultural Research Organization (NARO) of Uganda, the National Science Foundation (NSF) of the USA, and the Pew Fellows Program for Conservation and the Environment.

REFERENCES

Balirwa, J.S. 1992. The Evolution of the fishery of *Oreochromis niloticus* (Pisces: Cichlidae) in Lake Victoria. *Hydrobiologia*, 232: 85-89.

Lynch, M. and Mulligan, B.G. 1994. Analysis of population genetic structure using RAPD markers. *Molecular Ecology*, 3: 91-99.

Ogutu-Ohwayo, R. 1993. The effects of Nile perch, *Lates niloticus* L. on the fish of Lake Nabugabo, with suggestions for conservation of endangered endemic tilapias. *Conservation*, 7(3): 701-711.

Sambrooke, J., Fritsch, E.F., Maniatis, T. 1982. *Molecular Cloning: Laboratory Manual*. Cold Spring Harbor Press, Cold Spring Harbor, NY.

Trewavas, E. 1983. Tilapiine fishes of the Genera *Sarotherodon*, *Oreochromis*, and *Danakilla*. *British Museum (Natural History)*, London, No. 878.