

Allozyme variation in a Johnston's topminnow, *Aplocheilichthys johnstoni*, population from the Zambezi River system

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Abstract

Twenty five specimens of *Aplocheilichthys johnstoni* (Günther, 1893) were collected from the Cuando River in the Zambezi River system. Protein electrophoresis was used to analyse the genetic structure of this population. Seven of the 20 loci studied, (15% using the 95% criterion) revealed polymorphism. The heterozygosity value obtained ($H_o = 0.050$) compare favourably to those recorded for other fish species. Observed allele frequencies deviated from expected Hardy-Weinberg proportions at the **EST-3**, **GPD-1** and **GPI-2** protein coding loci. The results from the genetic analysis of *A. johnstoni* are discussed in relation to its role in mosquito larval control.

Introduction

The Aplocheilichthyinae (African lampeyes) comprise approximately nine genera of which two genera, *Aplocheilichthys* and *Hypsopanchax* are found in Southern Africa. The genus, *Aplocheilichthys*, is characterised by their bright, glossy white or blue eyes and therefore they are sometimes referred to as "lampeyes" (Skelton, 1993). Johnston's topminnow, *A. johnstoni*, is recognised as an aquarium species as well as for mosquito larval control. They are small (<50 mm total length) and keep to shallow, densely vegetated habitats. They primarily utilise the upper 10cm of the water where they feed on insect larvae, daphnia and other small invertebrates. Their anterodorsally located mouths enable them to feed on neustonic organisms and they are therefore extremely vulnerable to the spraying of insecticides (especially those aimed at the killing of mosquito larvae) and other pollutants (Kleynhans, 1986). Johnston's topminnows are serial spawners and eggs are laid on vegetation. The eggs are not drought resistant and excessive water extraction poses a threat to the survival of the species (Kleynhans, 1986).

The distribution of Johnston's topminnow in Southern Africa ranges from the Cunene, Okavango, Zambezi, Pungwe and Busi Rivers. Isolated populations of *A. johnstoni* are also known from the Marico, Notwane, Crocodile and Levuvhu Rivers (Limpopo River system). Further north, the species is found in the Zambian-Zaire and Kasai-Zaire River systems, the catchment areas of Lake Malawi and Lake Rukwa and east-coast rivers of Tanzania (Bell-Cross, 1972; Kleynhans, 1986). It is the presence of these isolated populations of *A. johnstoni* that is of particular interest to conservationists. The biological and commercial potential of this species served as motivation for this study.

Materials and methods

Twenty five specimens of *A. johnstoni* were collected from the Cuando River, Upper Zambezi River system (18°07'38"S,

23°22'51"E). Reference specimens were donated to the J.L.B. Smith Institute for Ichthyology, Grahamstown (RUSI 61856). They were captured using electro-narcosis and whole fish were frozen in liquid nitrogen (-196°C) and stored at -40°C until electrophoresis. Each specimen was homogenised in 1ml distilled water prior to electrophoresis. The samples were prepared as described in Engelbrecht and Mulder (1999) and analysed by means of horizontal starch gel-electrophoresis. The buffer systems used are described in Table 1. A total of 13 enzyme systems were screened using the enzyme-staining methods of Harris and Hopkinson (1976) and Hillis and Moritz (1990). The methods of Shaklee et al., (1990) were followed for the interpretation of gels and locus nomenclature. Statistical analysis was performed using the BIOSYS-1 programme of Swofford and Selander (1981). The statistical calculations included the following: the percentage of polymorphic loci ($P_{0.95}$), average observed (H_o) and expected heterozygosity (H_e) per locus and allele frequency deviations from expected Hardy-Weinberg proportions using the χ^2 -test for goodness of fit. Levene's correction for Hardy-Weinberg equilibrium was used in order to take the small population size into account (Levene, 1949).

Results

Seven of the 20 loci analysed in *A. johnstoni* revealed polymorphism. The loci screened, enzyme commission numbers and buffer systems used for each protein analysed are listed in Table 1. The allele frequencies for polymorphic loci, percentage of polymorphic loci using the 0.95 criterion and average observed (H_o) and expected (H_e) heterozygosities are presented in Table 2. The percentage of polymorphic loci ($P_{0.95}$) was calculated at 15% and the observed heterozygosity estimate was $H_o = 0.050$. Allele frequencies at the **EST-3**, **GPD-1** and **GPI-2** loci deviated significantly from expected Hardy-Weinberg proportions (Table 2). A deficit of heterozygotes were also observed at these loci (Table 2).

Discussion

According to Nei (1987), one of the main objectives of population genetics is to describe the amount of genetic variation in populations and then to study the maintenance of this variation. Analysis of the

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