# Allozyme comparisons of fish used in aquaculture in South Africa

Characterization, conservation, selection based on molecular markers and selection as a result of cryopreservation of semen

Herman van der Bank Geneticist

### Introduction

Inland aquaculture in South Africa is effectively limited to two species, rainbow trout and sharptooth catfish (Table 1). In a non drought year the annual output of trout would peak 1000 tons, and 1500 tons for catfish. Aquaculture research in South Africa decreased drastically since the demise of the National Aquaculture Programme of the Foundation for Research Development in 1989/90. This is the reason why recent production statistics for many species are not available. However, it is heartening that interest in aquaculture is once again flaring, and that the private sector has been funding some research projects. Research on triploidy, heat tolerant trout and cold tolerant tilapia strains were undertaken at the University of Stellenbosch, and the aquaculture potential of a catfish hybrid (Heterobranchus longifilis X Clarias gariepinus) was evaluated at Rhodes University. Although

the hybrid was reported to be imminently suited to aquaculture, no commercial production has commenced.

Species:	1988	1989	1990	1991	1992	1993
African catfish (C. gariepinus)	137	203	1500	1150	n.a.	n.a.
Rainbow trout (O. mykiss)	600	620	950	1220	990	1000
Tilapia (O. mossambicus)	20	20	30	40	n.a.	n.a.
Carp (C. carpio)	5	10	20	35	n.a.	n.a.
Ornamental fish	2.5	3.5	3.9	4.2	5.5	4.8

Table 1
South African aquaculture production statistics in metric tons
(HECHT and BRITZ, 1993; BRITZ, personal communication, 1997).
n.a. = not available.

The following discussion is a summary of results obtained for the characterization, conservation, selection based on molecular markers or selection as a result of cryopreservation of semen of trout, catfish and other species used in inland aquaculture in South Africa.

#### Rainbow Trout

Rainbow trout is an alien species, first imported for angling purposes from England in 1897. This species can be regarded as a success from angling and aquaculture points of view, but it had a detrimental effect on a number of indigenous species. Trout products include fresh, smoked whole, sliced smoked fillets, gravelax, paté, traviar and terrine. Eyed trout eggs are also exported to the Northern Hemisphere during their summer months (local winter), and proved to perform exceptionally well. The demand for South African trout eggs abroad exceeds the current production thereof.

It is interesting that trout production figures increased by 49% between 1989 and 1991, but hardly changed since 1992 when 990t were produced (Table 1). Trout farmers use between 720 and 750t of feed to produce 430t of trout; the gate selling price average R 12/kg and the total value is estimated at R 11.6 million.

Genetic variation studied in nine rainbow trout populations in South Africa showed average heterozygosity values of 4.5-7.5% and these relatively high levels can be attributed to the historical mixing of strains (VAN DER BANK *et al.*, 1992b). Growth rate differences related to genotypic variation, whilst food conversion rate and survival performance results did not seem to be related to heterozygosity values. The trout were obtained from Brink (in HECHT and BRITZ, 1993), who was able to improve the production performance of local trout strains (compared to control groups) to 6.5-14%, average 10.2%, in a subsequent study. Trout normally cannot tolerate summer temperatures higher than 20°C, but we have an eastern Cape strain which tolerates temperatures as high as 26°C.

# Sharptooth Catfish

Catfish products include fresh and smoked fillets, and tinned catfish. The waste (gut, etc.) is used as an additional protein source for farmed catfish and the bones are used to produce bone meal to supplement their diets. In addition, the skins are used to produce leather for wrist watch straps, gloves, handbags, etc., and the

pituitary gland (hypophysis) is used to induce spawning of various fish species.

There was an 87% increase of catfish production from 1989 to 1990, followed by a 23% drop over the period 1990 to 1991 and it can be expected that there has been little development since the last survey (Table 1). Drought and marketing problems were responsible for the decrease in production. However, a potential increase to ca. 5,000-6,000t per annum can be achieved (projection based on established production capacity of the present catfish farming community) and rapid progress can be anticipated because hatchery techniques have been mastered, good rainfall has occurred 1996. since and market constraints have reduced (HECHT and BRITZ, 1993).

Genetic variation in two commercially used (domesticated) and a wild population of catfish were compared and we obtained an average mean heterozygosity value of 5% for the latter population, but very little (0.3%) and much more (7.6%) for the two domesticated populations respectively (VAN DER BANK *et al.*, 1992a).

Overcompensation for a loss of genetic variation was achieved at the latter population (since the owner uses crosses between various wild and domesticated stocks) and the opposite holds for the other domesticated population (i.e. he uses the progeny to produce the next batch, thereby inducing inbreeding). The use of domesticated stocks to start new hatcheries may have negative implications for conservation because the escape of an access number of domesticated catfish into the wild (e.g. due to dam walls destruction as a result of heavy rains) could detrimentally affect the survival of progeny after introducing uncommon alleles.

GROBLER et al. (1992) have determined that a significant difference exists between the frequencies of some alleles in fast and slow growing groups of catfish. The feasibility of genetic selection for rapid growth in *Clarias gariepimus* was tested by VAN DER WALT et al. (1993b), who found noticeable differences between various genotypes. For example, one group increased its initial mass

advantage over another group from 105% at 30 days to 115% at 90 days.

A study by VAN DER WALT et al. (1993a) confirmed that the alleles which correlated to growth increase were similar between those obtained for the selected South African population and that of the population from The Netherlands, which were subjected to many generations of mass selection.

The important difference between the results from these two studies is that the South African catfish were less inbread, and are therefore better suited as candidates for aquaculture (to combat morphological irregularities associated with inbreeding). This was confirmed by GROBLER and VAN DER BANK (1994), who concluded that phenotypic variation is positively correlated to heterozygosity for different weight and length groups of catfish.

The effect of cryopreservation and various cryodiluents on allozyme variation in F<sub>1</sub> progeny of African catfish were demonstrated by VAN DER BANK and STEYN (1992). They have shown that significant differences of allele frequencies from expected Hardy-Weinberg proportions occurred in offspring obtained by using cryopreserved milt, compared to the control group produced by using fresh semen. These differences related to different cryodiluents and fertility as a result thereof. These selective qualities of cryopreservation techniques may have far reaching implications. For example, VAN DER BANK and STEYN (1992) used similar cryodiluents and freezing rates used by commercial institutions for livestock and humans, and selection of specific catfish genotypes were favoured by using these cryodiluents. Thousands of women annually make use of sperm banks and if the technique used for cryopreservation is found to favour specific sperm in humans also, it could have obvious ethical implications. However, VAN DER WALT et al. (1993c) have shown that the freezing rate used to induce cryopreservation is an important factor to reduce selection for specific allele combinations because an appropriate freezing rate minimises this effect. Therefore, an optimal freezing rate would be ideal to conserve the natural resources for future utilisation.

## Other Inland species

VAN DER BANK (in press) reported results obtained for tilapias. Tilapias are very popular as table fish; tilapia production increased from 11 to 40 metric tons from 1988 to 1991 (Table 1) and currently the demand exceed the supply thereof in southern Africa. Furthermore, due to the pressure on marine stock, it is expected that the demand for freshwater fish would increase. Despite these facts, very few fish farms have been developed.

Isozyme studies were predominantly used to assess genetic variation and differentiation of tilapia species in southern Africa (VAN DER BANK and FERREIRA, 1987a,b; LIZEMORE et al., 1989; VAN DER BANK et al., 1989; OOSTHUIZEN et al., 1993, in press; VAN DER BANK, 1994). We are now also involved in studies regarding allozyme variation in domesticated nile crocodile (Crocodylus niloticus), and VAN DER BANK (1995) and VAN DER BANK and VAN DER BANK (1995) studied allozyme variation in two freshwater mussel species. These results were obtained to characterize populations, and it can be used in subsequent studies for selection of suitable stocks for aquaculture. This need was identified because alternate protein sources should be investigated due to the ever increasing human population growth world wide. In addition to the food sources mentioned above, aquaculture holds other benefits. For example, in South Africa one person is employed per 2,9 tons of trout and catfish (HECHT and BRITZ, 1993).

#### Conclusion

No sensible long-term management or conservation plan can be implemented without a proper, initial understanding of the amount and pattern of genetic variation within the species. For instance, it would not have been possible to improve production characteristics and to maintain high performance levels without the abovementioned studies to characterize the taxa and to monitor effects of cryopreservation, management, directed selection and conservation efforts.

#### References

GROBLER (J.P.), VAN DER BANK (F.H.), 1994 — Allozyme polymorphism and phenotypic variation in African catfish (Clarias gariepinus). Comp. Biochem. Physiol. 107B: 111-116.

GROBLER, (J.P.), DU PREEZ (H.H.), VAN DER BANK (F.H.), 1992 — A comparison of growth performance and genetic traits between four selected groups of African catfish (*Clarias gariepinus* BURCHELL 1822). Comp. Biochem. Physiol. 102A: 373-377.

HECHT (T.), BRITZ (P.J.), 1993 — Aquaculture '92. *Proc. Aquacult.* Assoc. sthn Afr. 1, 198 pp.

LIZEMORE (E.), FERREIRA (J.T.), VAN DER BANK (F.H.), 1989 — An electrophoretic and morphometric evaluation of two subspecies of *Tilapia rendalli* from southern Africa. *Madoqua*, 16(1): 35-39.

OOSTHUIZEN (J.S.), VAN DER BANK (F.H.), FERREIRA (J.T.), 1993 — Genetic distances between and evolutionary relationships for *Tilapia guinasana*, *T. rendalli* and *T. sparmanii*. *Biochem. Syst. Ecol.* 21(3): 351-362.

OOSTHUIZEN (J.S.), VAN DER BANK (F.H.), FERREIRA (J.T.), in press — A protein electrophoretic study to determine the amount and pattern

of allozyme variation in Tilapia guinasana. Madoqua.

VAN DER BANK (F.H.), 1994 — Molecular systematics of fifteen Old world cichlid species using allozyme data. *Biochem. Syst. Ecol.* 22(8): 791-801.

VAN DER BANK (F.H.), 1995 — Allozyme variation in a freshwater mussel population (Coelatura kunenensis Mousson, 1887) from southern Africa. Water S.A. 21(2): 153-157.

VAN DER BANK (F.H.), in press — Tilapia biodiversity, genetic resources conservation and sustainable use: A South African perspective. Proceedings of the Workshop on the Characterization of Ghanaian Tilapia Genetic Resources for Use in Fisheries and Aquaculture, 4-7 June. 1996. Accra Ghana.

VAN DER BANK (F.H.), FERREIRA (J.T.), 1987a.—
Electrophoretic analysis of serum transferrins and serum esterases of three morphometrical different populations of *Oreochromis mossambicus* (Peters).

S. Afr. J. Wildl. Res., 17 (1): 13-15.

VAN DER BANK (F.H.), FERREIRA (J.T.), 1987b —
Morphometric and electrophoretic analyses of three wild populations of Oreochromis mossambicus (Peters). In: Proc. World Symp. on Selection, Hybridization

and Genetic Engineering in Aquaculture. K. TIEWS (ed.) Bordeaux, France. 27-30 May 1986, Berlin, 1987 (I): 103-111.

VAN DER BANK (F.H.), STEYN (G.H.), 1992 —

The effect of cryopreservation and various cryodiluents on allozyme variation of glucose phosphate isomerase in the F<sub>1</sub> progeny of African catfish (Clarias gariepinus).

Comp. Biochem. Physiol.

103B(3): 641-643.

VAN DER BANK (F.H.), VAN DER BANK (M.), 1995 —
Enzyme polymorphism in the freshwater mussel species, Aspatharia pfeifferiana (Mollusca, Lamellibranchiata) from southern Africa. Animal Genetics 26: 31-35.

VAN DER BANK (F.H.), GRANT (W.S.), FERREIRA (J.T.), 1989 — Electrophoretically detectable genetic data for fifteen southern African cichlids, *J. Fish Biol.* 34: 465-483.

VAN DER BANK (F.H.), GROBLER (J.P.), DU PREEZ (H.H.), 1992a — A comparative biochemical genetic study of three populations of domesticated and wild African catfish (Clarias gariepinus). Comp. Biochem. Physiol. 101B: 387-390.

VAN DER BANK (F.H.), SWART (M.K.J.), FERREIRA (J.T.), 1992b. —
Genetic variation in nine rainbow trout (Oncorhynchus mykiss) populations in South Africa.
Comp. Biochem. Physiol.
103B: 495-499.

VAN DER WALT (L.D.), VAN DER BANK (F.H.), STEYN (G.J.), 1993a — Allozyme variation in domesticated African catfish (Clarias gariepinus) from the Netherlands.

Comp. Biochem. Physiol.

104B: 15-18.

VAN DER WALT (L.D.), VAN DER BANK (F.H.), STEYN (G.J.), 1993b — An association between glucose-6-phosphate isomerase phenotypes and rapid growth in the African catfish (Clarias gariepinus).

Comp. Biochem. Physiol.
104B: 765-768.

VAN DER WALT (L.D.), VAN DER Bank (F.H.), STEYN (G.J.), 1993c —
The suitability of using cryopreservation of spermatozoa for the conservation of genetic diversity in African catfish (Clarias gariepinus). Comp. Biochem. Physiol. 106A: 313-318.