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AN EVALUATION OF FLUORESCENT ELASTOMER FOR MARKING KILLIFISH, *EPIPLATYS BIFASCIATUS* AND *EPIPLATYS SPILARGYREIUS* IN ABUNDANCE AND GROWTH STUDIES

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ABSTRACT

We evaluated the subdermal injection of fluorescent elastomer for tagging two sympatric aplocheilids, *Epiplatys bifasciatus* and *E. spilargyreius*, in pond and stream conditions. The smallest size that could be injected without mortality was 28 mm TL. The Visible Implant Elastomer (VIE) tag did not affect the growth or survival of adults of these fishes. Marks were retained throughout the 70 days of the pond experiment. The best position for injection of VIE in these fishes is the belly. VIE is most appropriate for mark-recapture population size estimation and movement studies. The tag could not be used for growth studies of these species because they become juveniles at about 21 mm TL, two weeks after hatching, and become sexually mature at about 26 mm TL, while the smallest tangible specimen was 28 mm TL.

Key words: Tagging, Visible Implant Elastomer, mark-recapture, aplocheilids, killifish.

INTRODUCTION

According to Bryant (2000), obtaining precise and accurate estimates of fish abundance in streams continues to challenge fishery biologists, despite the development of sophisticated mathematical models. Estimation of the population number by mark-recapture experiments is based on the assumption that the proportion of marked fish in a random sample is the same as the proportion of a known number of marked fish in the population. Few traditional techniques for marking fish can be readily applied to small fish in experimental studies that require the identification of individual fish (Dewey and Zigler, 1996). In Nigeria, the mark-recapture of fish by the fin-clipping method has been used to study the population dynamics of *Fundulopanchax gardneri* (Boulenger, 1911) in the Delimi River in Jos, Plateau state (Anadu *et al*., 1986).

The order Cyprinondontiformes, represented around the Kainji Lake by two aplocheilids, *Epiplatys bifasciatus* (Steindachner, 1881) and *E. spilargyreius* (Duméril, 1861) and one poeciliid, *Aplocheilichthys normani* (Ahl, 1928), was known to occur in the River Niger (Daget, 1962; Bank *et al*., 1965). However, with the absence of extensive swamp formed at the margin of the impounded lake, these species have disappeared in the new lentic environment. In 2001, *Epiplatys bifasciatus* and *E. spilargyreius* were discovered in a small grassy stream (approximately 1.9km long) flowing into the lower western portion of the lake at a latitude of 9^0 53' 45"N and a longitude of 04^0 33' 14"E near Monai village, a few kilometers from the Kainji dam. This stream is inundated to half its length annually by the lake, during which the two species are found directly in the lake. A preliminary investigation showed that similar nearby streams did not harbor these fish species. However around the lake, *E. bifasciatus* is found in two perennial riparian rivers (Shagwa and Wara Rivers) flowing into the lake on its northeastern portion. It seems the lake has constituted a geographical barrier that prevents the two populations from dispersing and colonizing adjourning streams. The Monai stream apparently represents an isolated habitat where both species are found together, with possible inbreeding depression and interspecific hybridization. Presently, this stream is threatened by human activities such as rice farming, fish cropping by pool draining in the dry season, grazing, deforestation of the catchments of the stream and pollution. This work is part of a broad study of the ecology and conservation of the two populations. The tagging experiment was carried out to estimate the population size of the two species in order to determine their minimum viable population in the Monai stream.

MATERIALS AND METHODS

Mark-recapture and removal methods require closed populations; therefore, sample reaches were selected to minimize emigration or immigration during the sampling period. The sample reaches ranged between 10 to 50m and every experiment usually lasted between one and four days. A capture occasion involved the netting of both *E. bifasciatus* and *E. spilargyreius* using a scoop-net with an opening diameter of 30 cm and a net basket (2 mm mesh size) of 45 cm length, for 30 ± 5 minutes. All fish (> 28 mm TL) from the first sample in an experiment were marked with Visible Implant Elastomer tags to serve as the mark sample in a single-census Peterson mark-recapture estimate (Ricker, 1975). The recapture sample was collected during one 30 minutes sampling occasion 3-4 days after the fish were released. Both the total number per species and the proportion of marked fish per species was recorded.

Visible Implant Elastomer (VIE) produced by Northwest Marine Technology, Inc., Washington, U.S.A. was used for the experiment. The fluorescent elastomer tag is a two component silicone polymer that is combined just before use. It is available in red, yellow green and orange and has been used on small *Lepomis macrochirus* (34 mm TL) (Dewey and Zigler, 1996). Because of the difficulty in handling these small fish, a hand-net (mesh size 100 micron) was used to immobilize them. Marking was done by injecting a small amount (1mm) of elastomer under the skin on the side of the belly using a $^3/_{10}$ cc syringe.

The elastomer was previously examined in a laboratory study to evaluate its retention and effects on growth and survival of *E. bifasciatus* and *E. spilargyreius*. We tagged about 12 to 15 newly matured specimens (28 - 36 mm TL), which is the smallest size that can be tagged successfully without any mortality as a result of handling and injection injury. The fish were released into small earthen ponds with a size of 4 x 4 m from 10th June, 2003 to 20th August, 2003 (70 days). To evaluate the effect of the tagging on the growth of *E. bifasciatus* and *E. spilargyreius*, the total length (TL, in mm) and weight (in mg) of 12 to 15 of the marked and unmarked (control) individuals of each species were compared using t-tests at the beginning and at the end of the experiment.

RESULTS

The elastomer tag did not affect the growth and survival of the two killifishes in the pond experiment. During the experiment tags were not shed by the fish. The result is shown in table I, where the initial mean length and weight of *E. bifasciatus* was 31.04 mm TL and 166.18 mg respectively, while that of *E. spilargyrieus* was 32 mm TL and 251.79 mg respectively. After 72 days, the final mean length and weight of tagged *E. bifasciatus* was 48.77 mm TL (± 4.2) and 707.55 mg (± 188.72) respectively, while that of the control was 50.57 mm TL (± 1.618) and 807 mg (± 78.17). The final mean length and weight of tagged *E. spilargyrieus* was 47.857 mm TL (± 4.017) and 664.71 mg (± 158.67), while that of the control was 48.63 mm TL (± 1.56) and 640.27 mg (± 90.79) respectively. The t-test at $P > 0.05$ shows that there is no significant difference in the length and weight of the tagged and untagged fishes of both species. Tagging visibility reduced significantly, both to the naked eye and the LED blue light, as a result of the beautiful color of these killifishes which masks the tag. However, this was overcome by placing the fishes in turbid water for 24 hours, during which time the fish become less intense in color which makes the tag more visible (as shown in Figure 1). The best place to inject the VIE is the belly which is lighter in color than other parts of the body.

DISCUSSION

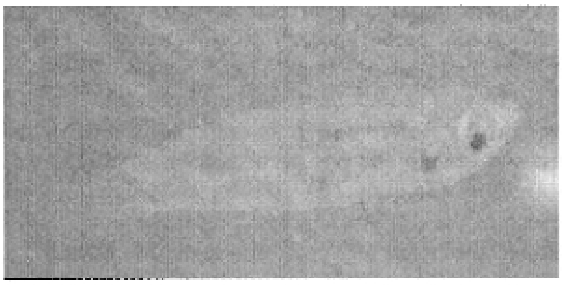
The ability to identify animals, either individually or as part of a group, is of fundamental importance in many biological studies (Parker *et al.*, 1990; Haines *et al.*, 1991; Hammer and Blankenship, 2001; Woods and Keith, 2004). Some characteristics that need to be considered when selecting a marking technique include the effect on survival, behaviour, and growth, the retention of the mark, the stress during marking and handling, and the cost of applying the mark (Wydoski and Emery, 1983).

The present study concludes that VIE has no significant effect on the growth of *E. bifasciatus* and *E. spilargyreius*. Neither did its application result in a high mortality when properly applied on fish from at least 28 mm TL. Tags were generally easy to apply when the fish are handled with a fine mesh size net (100 micron) and not handled directly.

For tagging to be an effective tool, tags must be retained by the animal and easily detectable by observers (Woods and Keith, 2004). In this study no tags were lost, which is consistent with other studies (Dewey and Zigler, 1996, Hale and Gray, 1998, Woods and Keith, 2004). A

TableI I: Mean initial and final size and weight of tagged and control specimens of *E. bifasciatus* and *E. spilargyreius*.

	Initial TL (mm)	Final TL (mm)		Initial weight (mg)	Final weight (mg)	
		Tagged	Control		Tagged	Control
E. bifasciatus	31.04	48.77 ± 4.2	50.57 ± 1.61	166.18	707.55 ± 188.7	807.00 ± 78.17
t-test at α 0.05			0.18277			0.18474
E. spilargyreius	32.00	47.85 ± 4.01	48.63 ± 1.56	251.79	664.71 ± 158.6	640.27 ± 90.7
t-test at α 0.05			0.6387			0.7198



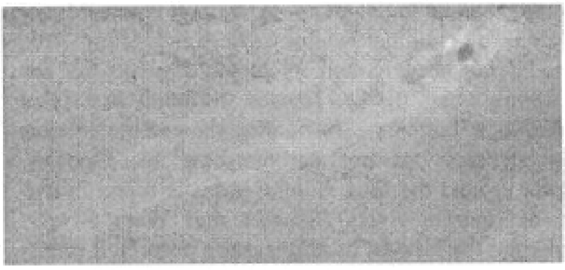


Figure 1: Two tagged *E. bifasciatus* (in turbid water, to reduce the masking influence of their intense colors).

retention rate of over 90% is commonly reported: 98 - 99% in *Lepomis macrochirus* after six months (Dewey and Zigler, 1996), 94-99% in rainbow trout after 24-30 days (Hale and Gray, 1998), 100% in the seahorse *Hippocampus intestinalis* after seven months (Woods and Keith, 2004). In this study, the smallest tagable fish was 28 mm TL. *Epiplatys bifasciatus* and *E. spilargyreius* are small sized fishes with a short life span. They become juveniles at about 21 mm TL, which is two weeks after hatching, and become sexually mature at about 26 mm TL. This means that VIE cannot be used for growth studies, but will rather be effective for mark-recapture studies of mature individuals of these species.

For marking these beautiful, colorful fishes we advise to use orange, yellow and red, since these easily contrast with the olive-green color of the fish.

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