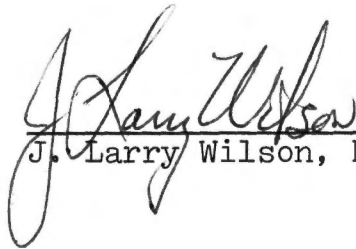


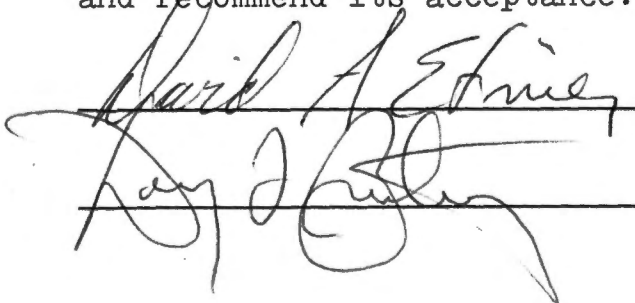
To the Graduate Council:

I am submitting herewith a thesis written by Caney Parmele Knauth entitled "An Evaluation of the Effects of Three Marking Techniques on the Growth, Behavior, and Mortality of Channel Catfish Ictalurus punctatus (Rafinesque)." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Wildlife and Fisheries Science.



J. Larry Wilson, Major Professor

We have read this thesis  
and recommend its acceptance:



Accepted for the Council:



Vice Chancellor  
Graduate Studies and Research

Thesis

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AN EVALUATION OF THE EFFECTS OF THREE MARKING TECHNIQUES ON

THE GROWTH, BEHAVIOR, AND MORTALITY OF CHANNEL CATFISH

Ictalurus punctatus (Rafinesque)

A Thesis

Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Caney Parmele Knauth

June 1977

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## ABSTRACT

Three groups of fingerling channel catfish, Ictalurus punctatus (Rafinesque), were marked using either a partial fin clip of the left pelvic fin, a cold brand using liquid nitrogen at  $-196^{\circ}\text{C}$  as a coolant, or a gun-injected spaghetti-type floy tag. Fish were observed during a 12-week period and compared to a control group for loss of marks, noticeable change in behavior, mortality attributed to the marks, and any significant effect that the marks exhibited on growth.

It was determined that the cold brand was the only marking technique producing a mark that was still retained by the fish and recognizable after a 12-week period. Behavior was greatly altered only in the group receiving the injected tag; this group also exhibited the only substantial rate of mortality attributed to the mark. Statistical analysis of the effects of the three marking techniques on growth indicated that the application of the marks had no significant effect on those fish surviving throughout the experimental period.

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## CHAPTER I

## INTRODUCTION

During the past century, as research in the field of fisheries has grown in stature and importance, the identification of an individual or group of individuals in a given study population has become an invaluable and indispensable tool.

There are recordings of very early attempts at marking fish for experimental purposes, such as that recorded by Isaak Walton in his seventeenth century book THE COMPLEAT ANGLER. He noted that Sir Francis Bacon attempted to mark fish in order to study age and homing instincts in salmon (Krumholz, 1943). Other accounts of early attempts at marking fish claim that the practice was begun by wealthy owners of riparian rights interested in discovering facts in order that they might be able to conserve the salmon and sea trout industries. However, since these early attempts were scattered and unorganized, few marked fish were ever recovered (Rounsefell and Everhart, 1953).

Records kept through the years on marked fish and their returns were originated in 1873 with what has been called the first successful tagging experiment, carried out by Charles G. Atkins. Atkins, a member of the U. S. Fish Commission, tagged Atlantic Salmon in Bucksport, Maine, keeping records on tagged fish and any fish returning to the area (Rounsefell and Everhart, 1953).

The art of marking individuals and its use as a research tool developed slowly through the years following Atkins' experiment. However, as the realization of the ability of being able to identify individuals or groups of individuals in a study population became recognized as an important tool in fishery research, more effort was channeled into actual research on new marking methods and techniques, and the improvement of those already in existence. It became evident to researchers that a good marking method should be able to supply the investigator with the appropriate information he desired in the most accurate manner and have a minimal effect on the test individual. The mark ideally would in no way affect the growth or behavior of an individual, or increase the possibility of mortality in any manner. This is an area in which much interest has been generated and in which additional research needs to be conducted.

There are several good marking methods now in use, but no one method at this time actually fulfills all the desired characteristics, that is, to mark a fish for identification for an extended time interval with minimal effect to that fish.

The purpose of this project was to evaluate the effects of three marking techniques on small channel catfish: a partial fin clip of the left pelvic fin, cold brands using liquid nitrogen at  $-196^{\circ}\text{C}$  as a coolant, and the injection of a spaghetti-type floy tag. Fish receiving these marks were observed during a 12-week period for duration of the

mark, changes in behavior, mortality attributed to the mark, and for any significant effect the mark may have had on growth of the fish.

## CHAPTER II

### LITERATURE REVIEW

#### Fin Clip

Fin clipping is a well known and very popular method of marking fish that has been in use for many years. There has been extensive research on this method involving the clipping or removal of a single fin to the removal of various combinations of fins including both the paired and single fins. According to Stott (1968), the paired fins, especially the pelvics, are the fins usually selected for clipping, as was the case in this study.

Stolte (1973) stated in his work with coho salmon that removal of the left pelvic fin did not significantly affect either the survival or growth of the fish. Similar findings were reported by Patrick and Haas (1971). In their work with musky fingerlings, they stated that the removal of the left pelvic fin apparently did not affect either the growth or the survival of the fish held in a hatchery pond for two months.

Churchill (1963) found clipping of the left pectoral or the left pelvic fin did not affect the chance of survival or growth of walleye fingerlings averaging 79 mm in total length stocked in a lake in Wisconsin. Brynildson and Brynildson (1967) reached similar conclusions, finding removal of a pelvic fin or a pectoral fin did not significantly

affect the survival or growth of brown trout marked when 76 to 127 mm in length over a span of two years.

Stauffer and Hanson (1969) found the clipping of both the right pectoral and right pelvic fin did not affect the survival, growth, or the condition of rainbow trout held in a hatchery pond for 14 months. Shetter (1951) found removal of either pectoral fin, the dorsal and adipose fin, or the right pelvic fin did not significantly affect the survival or growth of lake trout fingerlings held in a pond for one year with predator brook, brown, rainbow, and lake trout.

Horak (1969) found removal of both pelvic fins, the dorsal, both pectoral, both pelvic and pectoral, the anal, or the adipose fin did not significantly affect the stamina of rainbow trout ranging from 84 to 94 mm in length, thus having a probable negligible effect on survival. He did find, however, that the removal of the caudal fin did reduce stamina in the fish, thus having a probable effect on survival. He advised caution when removing all paired fins since a reduction in stamina could possibly result from that action.

Coble (1971) found clipping of the left pectoral, right pelvic, both pelvics, or the anal fin did not significantly affect growth of young smallmouth bass over a one-year period. However, fin clipping did appear to significantly affect the survival of the fish in his study.

Shetter (1950) found clipping of the right pectoral, dorsal and adipose fins, or the right pelvic fin did not significantly affect the survival or growth of lake trout

fingerlings held in a pond from 2.0 to 4.5 years. However, he did find that clipping of the left pectoral did significantly lower growth and survival of these fish. Coble (1967) also stated that clipping of the left pectoral reduced growth. In his study with yellow perch held in a pond for approximately one year, he clipped either the left pectoral, right pelvic, both pelvics, or the anal fin. The left pectoral clip was the only fin clip affecting growth, and survival was lower only in those fish receiving the anal fin clip.

#### Cold Brand

Cold branding is a relatively new method of marking fish first used by Fujihara and Nakatani (1967) and Everest and Edmundson (1967). The first use of liquid nitrogen as a coolant was reported by Mighell (1969). A majority of the research on this method of marking fish has been centered on the development of the branding technique and tools used to establish a reliable mark, and not on the actual effect to the fish.

Laird et al. (1975) did a histopathological study over a 16-week period to investigate the effect of freeze branding using liquid nitrogen as a coolant on juvenile salmon averaging 130 mm in length. They concluded that this method of marking salmonid fish provides a brand which is legible for at least 16 weeks and fulfills many of the requirements of an ideal marking system. They also stated this technique

caused relatively little damage to the fish. Smith (1973), also working with salmonids, found no significant effects on mortality or any visual difference in behavior or noticeable distress attributed to marking coho, chinook, and sockeye salmon fry measuring 32 to 48 mm in length with either hot or cold brands. Brands remained legible up to four months.

Turner et al. (1974) found that branding using liquid nitrogen was a very successful method for the purpose of identification of rainbow trout. They stated mortality was very low, attributing what mortality there was to handling and anaesthetizing of the fish and not to the brand itself. The authors stated that some brands were still identifiable two years after the fish were released in a cold water reservoir.

Brauhn and Hogan (1972) found freeze branding of channel catfish with liquid nitrogen produced an identifiable mark for up to 120 days. The authors listed advantages of this method as normal behavior after branding, no epidermal puncture, and high visibility of the initial mark.

Fujihara and Nakatani (1967) found that cold brands using ethanol (100%) and dry ice at  $-80^{\circ}\text{C}$  appeared to cause no undue stress to the fish and no mortality was experienced. They felt that cold branding is a less traumatic method for marking fish, producing a legible mark for up to four months. Mighell (1969) stated that liquid nitrogen at  $-196^{\circ}\text{C}$  is a better coolant than that used by Fujihara and Nakatani (1967), since ice and mucus did not accumulate on the brand-

ing tool. Thus, the tool did not have to be cleaned regularly in order to keep the brands from being distorted. He also found that this method did not affect behavior or cause any mortality in 4000 coho salmon averaging 77 mm in fork length held two months in a raceway after branding. The mark was easily recognized at the end of the test period, and the author stated that he had eight sockeye salmon retaining the brand for up to 14 months.

Refstie and Aulstad (1975) did experience mortality in their work branding rainbow and sea trout with liquid nitrogen. The fish experienced large wounds which the authors attributed to applying the branding tool to the fish for too long a period. The authors also branded salmon and did not experience the same results as with the rainbow and sea trout, explaining that for the salmon a 3-second exposure time obviously was not too long. The authors found that after a year, at least 87 to 92% of the fish could be identified. Raleigh et al. (1973) stated an application time of 1.5 to 2.0 seconds to be optimal when cold branding using liquid nitrogen as a coolant on both trout and centrarchids. The authors found that a branding time of 3 to 5 seconds would usually result in a blurred brand or loss of dermal tissues over the branded area resulting in open sores, especially in the finely scaled species.

### Spaghetti-Type Floy Tag

There are many different types of information-carrying tags developed over the years which can be attached to fish in an assorted number of techniques and locations (Rounsefell and Kask, 1943). Thorson (1967) and Dell (1968) described a new fish tag and cartridge-fed applicator. Dell (1968), in preliminary work with this new tag and tagging device, tagged 200 rainbow trout ranging in length from 260 to 410 mm in the dorsal musculature just below the dorsal fin. After two months, the author saw no gross shortcomings of the tag with tags firmly attached and tagging wounds minimal. He also stated that these tags could be applied uniformly and precisely even by inexperienced personnel.

Greenland and Bryan (1974) tagged large channel catfish with an FD-67 spaghetti-type floy tag and tagging device as described by Dell (1968) and Thorson (1967) and held them in a pond for 12 weeks for observation. By the end of the first six weeks, 70% of the FD-67 tags had been shed by the fish; at the end of the 12-week test period, 90% of the tags were lost. The authors attributed this loss to the lack of being able to secure a solid attachment of the internal anchor of the floy tags to the interneurals of the fish. This was due to the short length of the connecting filament between the internal anchor of the tag and the information-carrying tube, thus not allowing the anchor to open up behind the interneurals of the channel catfish for a solid attachment.

Keller (1971) used FD-67 spaghetti-type floy tags injected with a tagging gun to tag small brook trout ranging from 99 to 165 mm in total length. Of 75 brook trout tagged and stocked as fall fingerlings, only 10% retained their tags till the following August. Of 81 trout ranging from 140 to 213 mm tagged and stocked as spring yearlings, only 31% retained their tags till the following August. Fish were observed with lesions where tags had been injected under the dorsal fin and had worked loose. Some trout were observed with a portion of the internal anchor stem protruding through the skin. The author stated that in instances where tags were lost and wounds had healed, a jagged scar was left on the fish's side. The author performed another experiment in which he tagged the fish closer to the dorsal fin. After two months, 98% of the fish still retained their tags and tagging wounds were healed or healing in most of the test fish. The author concluded that satisfactory retention of floy tags by small brook trout was apparently dependent on tag placement to assure engagement with the interneurals.

Stobo (1972) used spaghetti-type tags and an applicator as described by Dell (1968) to tag yellow perch. He stated the tags did cause open wounds in the fish, but caused no differential mortality due to the size of the fish. He also stated tag retention was good, and mortality caused by the tags was insignificant. The author concluded that the tagging method is satisfactory for use on yellow perch.

Carline and Brynildson (1972) found the use of FD-67 tags initially retarded growth of 10-month-old brook trout but had little effect on the trout thereafter. They stated survival of the tagged trout was similar to a control group, and tag losses ranged from 2.0 to 5.7% over a seven-month period. Results of this study were questionable, however, in that both the control and the tagged groups received different fin clips, thus adding another variable to their research results.

Roberts et al. (1973 a, b, c) did histopathological studies on tagging lesions in newly tagged salmon parr, the chronic tagging lesions in returning adult fish, and secondary infections associated with tagging. The authors stated the lesion caused by tagging is quite a severely traumatic insult to the young fish. They also stated that providing secondary infection does not supervene, there is no evidence that the lesion caused by tagging has any obvious effect on the behavior or development of the fish.

## CHAPTER III

## METHODS AND MATERIALS

Channel catfish fingerlings, Ictalurus punctatus (Rafinesque), were obtained from a population being held in cages in the influent canal at the Bull Run Steam Plant in Melton Hill Reservoir, Oak Ridge, Tennessee. They were transported to the fishery facility and held in galvanized steel tanks until the study was initiated. They were treated with formalin at 15 ppm to reduce the possible incidence of disease due to stress (Avault and Allen, 1970).

Three circular plastic pools were used for the experiment, each measuring approximately 2.5 m in diameter by 0.5 m in height. Each pool was fitted with 16 dividers made of small mesh netting with wooden borders. Each divider was connected to the outside rim of the pool and to a circular section of hardware cloth in the center. An agitator was placed in the center of the hardware cloth cylinder to allow water agitation to be distributed equally from the center of the pool. Thus, each pool was divided into 16 equal sections holding approximately 142 L of water per section which was freely circulated throughout the entire pool. The pools were filled and the water was allowed to age prior to the introduction of fish.

The three experimental pools contained a total of 48 sections which allowed space for 12 replicates for each of the three marks tested and the control group. A partial fin

clip of the left pelvic fin, freeze branding using liquid nitrogen at  $-196^{\circ}\text{C}$  as a coolant, and a gun-injected spaghetti-type floy tag as described by Dell (1968) were the marks to be evaluated in this study. The experiment was designed to determine what effects these marks would have on growth, behavior, and mortality of the fish. The fish were also observed for duration of marks over the 12-week test period.

The partial fin clips were performed by clipping off the distal half of the left pelvic fin at a right angle to the fin rays with a small, sharp pair of scissors. Care was taken not to clip the fin too close to the body; this lessened the chances for infection and disease.

Cold brands were applied using liquid nitrogen at  $-196^{\circ}\text{C}$  as a coolant. The liquid nitrogen was stored in a 0.5 L thermos bottle with a 10 mm hole punched in the top to allow for the evaporation of gas and to control possible build-up of pressure. The branding tool used was a brass rod approximately 180 mm in length fitted with a cork at one end for handling and to enable the tool to fit loosely in the thermos of coolant when not in use (Figure 1). The tip of the rod was shaped so as to give a brand in the shape of the letter O. The brands were applied on the fish's right side under the dorsal fin (Figure 2). The branding tool was held firmly against the fish for a period of two seconds.

The tags used in this experiment were FD-67 spaghetti-type floy tags injected by a tag applicator as described by



Figure 1. Branding and Tagging Equipment

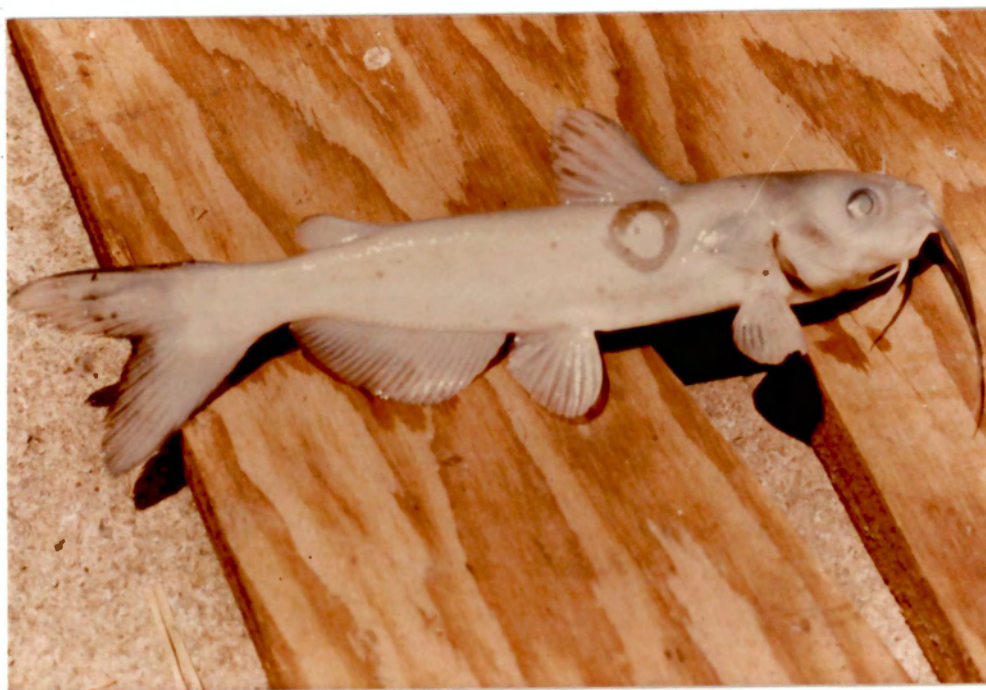


Figure 2. Cold Brand

Dell (1968) and Thorson (1967) (Figure 1). The tag used consisted of an information-carrying vinyl tube measuring approximately 57 mm in length and a T-bar internal anchor measuring 10 mm in length which was connected to the tube by a slender filament measuring approximately 19 mm in length. The tags were injected into the fish's right side under the dorsal fin as described by Dell (1968) in an attempt to have the T-bar positioned in such a manner as to solidly engage with the interneurals of the fish.

On June 18, 1976, fish were randomly selected and anaesthetized by placing them in a solution of six drops of Hypno Fish Calmer per 3.8 L of water. The fish were then weighed, measured, and marked appropriately, except in the control group which received no mark, and placed in their designated pool section. This procedure was followed until all fish had been appropriately marked and distributed. The length and weight ranges for each of the experimental groups at the time of marking are listed in Table 1.

Each individual fish during the experimental period was fed Purina sinking trout chow at a rate of 2% of its body weight six days a week.

Fish were weighed and measured every three weeks to gather data for statistical analysis using an analysis of covariance to establish if the marks employed had any significant effect on growth of the fish over the 12-week experimental period (Snedcor, 1956). At the same time, feeding schedules were adjusted to allow for the growth of the fish

Table 1. Size of Experimental Groups at Time of Marking

| Test Group | Range in Length<br>in Millimeters | Mean<br>Length | Range in Weight<br>in Grams | Mean<br>Weight |
|------------|-----------------------------------|----------------|-----------------------------|----------------|
| Fin Clip   | 130-179                           | 148            | 13.6-37.8                   | 23.5           |
| Cold Brand | 125-184                           | 155            | 14.6-44.7                   | 27.3           |
| Tags       | 126-185                           | 155            | 17.4-47.2                   | 27.9           |
| Control    | 137-198                           | 160            | 17.0-60.2                   | 30.2           |

during the three-week period.

Uneaten food and waste material were siphoned from the pools twice weekly. Also, each pool received a 757 L change of aged water twice a week.

A daily record of the water temperature was kept throughout the experiment. Regression analysis of this data was performed at the end of the study period to determine if temperature variation during the experiment was a significant variable.

## CHAPTER IV

## RESULTS

## Duration of Marks

Clipping of the distal portion of the left pelvic fin was an easily identifiable mark when first applied. Observation of the fish after the first three-week period indicated the fins had regenerated approximately  $1/3$  of that portion which had been removed. Also, it was observed that the regenerated portion of the fin was showing a definite tendency toward the rounded characteristics of the pelvic fin as they were before the mark was applied.

Observation of the fish after six weeks (half-way through the experimental period) indicated that the partially clipped pelvic fins had regenerated approximately  $2/3$  of the clipped portion. They were rounded, having the same appearance as the original fin before clipping. The only difference between the paired pelvic fins of the test fish after six weeks appeared to be that the clipped fin was slightly shorter than the opposite pelvic fin. There was no obvious bending of the fin rays in the regenerating fin along the line of incision where the fin had been originally clipped.

At the end of nine weeks, fish were again observed, and it was found that all the partially clipped left pelvic fins had completely regenerated and could no longer be dif-

ferentiated from the opposite unclipped fin.

Cold brands using liquid nitrogen at  $-196^{\circ}\text{C}$  left an easily identifiable and very distinct white mark when first applied to the fish due to the freezing of mucus and epithelial tissue in the branding area (Laird et al., 1974). The mark disappeared, however, when the fish were first placed in the water due to the thawing of the frozen area of the brand. After 24 hours, the brand had reappeared and it was very distinct as a dark-colored circle on the fishes' sides within 48 hours.

Observation of the fish after the first three-week period indicated very little change had occurred in the marks; cold brands were still very distinct and easily recognized. However, they did exhibit a slight change in that some brands had developed small areas of a somewhat lighter appearance than the normally dark color of the brand.

After six weeks, cold brands were still very easily recognized and quite clear. However, larger areas of the cold brand were becoming somewhat lighter in color and a few brands were beginning to show signs of very slight distortion.

After nine weeks, the brands exhibited definite signs of fading. They were lighter in color and becoming less distinct; however, they were still easily recognized. The brands had become much more distorted as the fish had grown. Those fish exhibiting faster growth in this test group showed much more distortion of the brand than those

growing at a slower rate.

At the end of the 12-week test period, cold brands were still recognizable. The brands were somewhat distorted but still legible, and brands were much lighter in color than when originally applied to the fish.

The results obtained on loss of the injected spaghetti-type floy tag in this experiment were quite discouraging. This test group could be broken down into three subgroups for description of tag retention during the experiment. The first subgroup, comprised of  $1/3$  of the total test group, contained those fish which had shed their tags within three weeks after marking and survived throughout the experiment. All tagging wounds had healed although there remained a large jagged scar at the site of tag injection. The second subgroup (also comprising  $1/3$  of the total test group) contained those fish which shed their tags within the first three-week period of the experiment and died as a result of the trauma of tagging and development of ulcers at the injection site. The third subgroup was comprised of the remaining  $1/3$  of the test fish and contained those fish which died as a direct result of the trauma of tagging and ulcers attributed to the injection of the tags but which still retained the tags at death. This author feels that had the fish in the third subgroup lived, they too would have shed their tags within the first three weeks since they exhibited the same symptoms as those fish in the second subgroup which shed their tags before dying.

No tags were shed within the first week of the experiment. It was found, however, that a portion of the internal anchor of the tag had pierced the skin causing large ulcers to develop at the site. This ulceration was aided by the fish's continual rubbing of the irritated area on the bottom of the experimental pool. Eventually, both ends of the internal anchor were freed from the flesh due to the movement of the tag and the rubbing actions of the fish. This resulted in the tag being attached to the flesh only by the small, slender, connecting filament of the tag (Figure 3).



Figure 3. Tagging ulcer with tag still loosely attached by the slender connecting filament of the vinyl tube to the internal anchor.

This attachment was soon broken, leaving a large ulcer which continued to irritate the fish.

Those fish able to shed their tags without causing too much damage to themselves were soon able to recover. The tagging wound healed quickly and left a jagged scar which could easily be identified throughout the rest of the experimental period.

### Mortality

Mortality experienced in this experiment was distributed throughout the three experimental pools. No mortality was experienced during the 12-week experimental period in the test group receiving the partial fin clip, and only one fish in the group receiving the cold brands died as a direct result of the mark received by that fish. That fish died in the fifth week of the experiment as a result of infection of the branding area.

Of the test group receiving the injected tags, only 33% survived the entire study period. Mortality in this test group was directly attributed to the application of the gun-injected tags and the ulcers which developed due to these tags. This test group was the only one having a higher rate of mortality than the control group.

### Behavior

Behavior of fish in the test groups was compared by direct observation to the actions of the fish in the control

group in the experimental environment. The control group would position themselves near the bottom at the center of the pool facing outwards into the open area of their compartment. They would move out of this position only to feed, or for an occasional slow swim around their pool section area and then return to their original position at the center of the pool.

All fish in the test group receiving the partial clip of the left pelvic fin were found to exhibit similar behavior to that of the control group throughout the entire experimental period. The test group receiving the cold brands also exhibited this same behavior pattern throughout the 12-week period with the exception of the one fish which developed an infection in the branding area and later died. This fish was observed swimming erratically around the enclosure, occasionally rubbing the branded area on the bottom of the pool. As the infection of the branded area worsened, the fish would perform this action much more frequently. Eventually the fish became very sluggish and, before dying, swam in a very erratic manner, sometimes swimming from the bottom to the top of the pool and around the compartment on its side.

The fish in the test group receiving the injected spaghetti-type floy tags behaved as did the other test groups when first placed in the pools. However, during the first week of the experiment, they began to swim around their particular pool sections, often rubbing the tag injection site on the bottom of the pool. The continual rubbing action

caused ulcers to form at the injection site; in most cases, the internal anchor protruded through the fishes' skin. Those fish shedding their tags without inflicting too much damage to themselves then took on the behavioral pattern as exhibited by the control group. However, those fish which had large ulcers as a result of shedding or attempting to shed the injected tags continued to behave in an erratic manner, continuously rubbing their ulcerated sides on the pool bottom. This increased the severity of the wounds and eventually resulted in the death of the fish.

#### Growth

An analysis of covariance was run on the surviving fish in the four test groups. It was found that, at the .05 level, there was no significant difference in growth between these groups for the 12-week period.

Water temperatures ranged from a low of  $20.5^{\circ}\text{C}$  to a high of  $26.0^{\circ}\text{C}$  with a mean water temperature of  $24.0^{\circ}\text{C}$  during the 12-week period. Regression analysis revealed this temperature variation had no significant effect on the results obtained for growth comparisons.

## CHAPTER V

## DISCUSSION

As stated in the introduction, a mark, ideally, should in no way affect the growth or behavior of a fish, nor should it increase the possibility of mortality of an individual. The mark should be easily identified by the investigator, supplying all the information required throughout a desired time interval without loss of that mark. Two other characteristics that are always considered when deciding on what marking method is best suited for a particular project are the ease with which the selected mark can be applied to the fish whether in the lab or field, and the expense involved.

## Fin Clip

Fin clipping, as stated by Stott (1968), is the most popular method of marking fish. The results of this investigation indicated that the use of a partial clip of the left pelvic fin is a useful batch marking technique for small channel catfish ranging in size from 130 to 179 mm in total length when identification of groups of fish is required for projects of very short duration lasting up to six weeks. After six weeks, positive identification of small fin-clipped channel catfish in the field would be a difficult and questionable task even for a trained observer. The partial fin clip had no significant effect on the growth of the fish under

the experimental conditions and no mortality was experienced; in addition, there was no noticeable alteration of the fish's behavioral patterns.

This method of marking fish for identification is an extremely simple and rapid operation either in the lab or the field. The only equipment required to mark small catfish is an ordinary pair of sharp scissors, making the cost of this marking method extremely low.

Notable disadvantages in using the partial fin clip of the left pelvic fin as a marking technique include the fact that clipping of the fin does cause injury subjecting the fish to possible invasion of various infectious agents. Also, the fact that the duration of this mark is so very limited renders this method completely useless as a marking procedure for projects requiring reliable recognition of the fish for longer time intervals than approximately six weeks. Removal of the complete fin is one method of increasing the duration of the mark, since regeneration does not occur in fins which have been completely removed. However, investigators feel that this particular practice might possibly decrease the fish's survival capabilities in the long run.

#### Cold Brand

The results of this experiment indicated that cold branding of small channel catfish ranging in total length from 125 to 184 mm using liquid nitrogen as a coolant is a useful batch marking technique which is easily applied and

recognizable for a duration period of at least 12 weeks at temperatures ranging from 20.5°C to 26.0°C with a mean temperature of 24.0°C. This method of marking fish could also be employed for use in the identification of individual fish with applications of various numerals and/or letters and combinations of those marks applied with more complex branding equipment such as that described by Raleigh et al. (1973).

Cold brands were still easily identified at the termination of the 12-week experimental period although the brands had become less distinct and somewhat distorted, this distortion being attributed to the growth of the catfish during the test period (Raleigh et al., 1973) and (Smith, 1973). There have been reported cases of cold brands using liquid nitrogen as a coolant, which in some instances were retained from 14 months (Mighell, 1969) up to two years (Turner et al., 1974). Thus, cold brands do in fact have an obvious potential in producing identifiable marks for periods of lengthy duration, making this a valuable method of marking in those projects requiring individual identification of fish for longer periods of time.

Cold brands were found to have no significant effect on the growth of the fish under the experimental conditions employed, and no mortality or alteration of the fish's behavior was attributed to the marks when properly applied. The death of the one individual that had exhibited abnormal behavior in this test group was attributed to the infection

of the branded area most probably resulting from the improper application of the brand at the time of marking.

Refstie and Aulstad (1975) experienced similar results attributing large wounds to too long an application period of the branding tool to the fish at the time of marking.

Raleigh et al. (1973) stated that if the application period is too long, dermal tissue is destroyed, leaving an open sore on the fish. An open sore is always subject to possible infection by the invasion of some pathogen. Raymond (1974) stated that if excess pressure is applied to the branding tool the resulting cellular damage is similar to that resulting from too long an application period. This, too, can lead to the infection of the branded area possibly resulting in the death of the fish.

The use of cold brands as a method of marking fish for identification, although a more complex and variable method of marking fish than applying a partial fin clip, is still a rather simple, inexpensive, and rapid method either in the lab or the field. The costs involved in cold branding would include the liquid nitrogen and the branding apparatus employed which would be, in part, dependent on the degree of sophistication of the branding device required by the investigator. Mighell (1969) stated that the use of cold brands to mark fish should be attractive to any agency wishing to mark large numbers of fish due to its low cost, the simplicity of the devices used to mark fish, the ease with which the marks are applied, and the effectiveness of the marks

capable of being applied at very rapid rates.

This method of marking fish has major advantages over both fin clipping and tagging in that there was no noticeable alteration of the fish's performance capabilities since there is no removal of any of the fish's functional body parts. When cold brands are properly applied, there is no piercing or cutting of the skin surface of the fish, thus reducing the possibility of sites for invasion of infectious agents.

Raleigh et al. (1973) stated that the most frequent complaint against cold branding has been a high degree of variability in the quality and subsequent recognition of the mark. There are many elements involved in the process of cold branding which could lead to problems on which this complaint is based. An investigator would be wise to carry out preliminary studies to ensure that the equipment available and the technique to be used in the branding operation produce a clear, long lasting, recognizable mark.

#### Spaghetti-Type Floy Tag

The results of this experiment on the use of the injected spaghetti-type floy tags indicated that this method of marking small channel catfish ranging from 126 to 185 mm in length is questionable as to the dependability of the mark. This is directly related to the importance of the correct placement of the tags so as to get a strong, solid attachment of the internal anchor of the tags between the interneurals of the fish. Dell (1968) stated that this method of tagging

could be performed uniformly and precisely even by inexperienced personnel. However, Greenland and Bryan (1974) experienced a 90% loss of internal anchor tags from channel catfish attributed to the lack of being able to secure a solid attachment between the anchor portion of the tag and the interneurals of the fish. Keller (1971) also experienced a 90% loss of these tags in his work with small brook trout, noting that where tags had worked loose from under the dorsal fin, or were in the process of doing so, lesions had developed; this was also the case in this study with small channel catfish. Keller (1971) concluded that satisfactory retention of the tags was dependent on tag placement under the dorsal fin to assure proper engagement of the interneurals.

The results of this experiment indicate that before any large scale tagging operation is performed, there should be preliminary tagging experiments conducted as to the size and species of the fish to be marked to establish the proper location and technique to be used in placement of the type and size tag selected. This would better insure proper engagement of the tags to the interneurals of the fish, minimizing possibilities of the tags affecting the fish adversely. Also, it is advised that tagging personnel attempt some preliminary marking to insure proper application and correct placement of the tags.

This method of marking fish has advantages in that the tags supply a great deal of information allowing individual identification of a large number of fish for long intervals

of time. Tags can be applied rapidly either in the lab or the field. If properly applied, this method produces a long lasting, easily recognizable mark which is not likely to be overlooked by untrained observers, as may be the case with a partial fin clip or a cold brand.

The equipment needed to mark fish with an FD-67 spaghetti-type floy tag consists of a cartridge-fed applicator priced at \$65.00, and an appropriate number of FD-67 floy tags which come in cartridge form and are priced at \$110.00 per 1000 tags excluding legends or numbering. Legends or numbering can be obtained at a minimum cost of \$.03 per tag which would increase the cost of the tags at least \$30.00 per 1000. Thus, this method of marking fish can be an expensive operation, especially when marking large numbers of fish.

Results of this experiment indicated that the injection of the tags and trauma induced by the tags after their injection had no significant effect on growth in those individuals surviving throughout the experimental period. Roberts et al. (1973 b) stated that the lesion caused by tagging is quite a traumatic insult to the young fish, but providing secondary infection does not supervene, there is no evidence that the tagging lesion has any obvious effect on the behavior or development of the fish.

Mortality was extremely high in this experimental group, and fish exhibited erratic behavior, all of which was directly attributed to the tags and large lesions which developed at the tagging site.

This method of marking fish, even if properly applied, still has a major disadvantage in that application of the tag requires the breaking of the skin surface of the fish, thus affording a site for possible invasion of infectious agents. Other disadvantages of this method of marking fish are that the possibilities exist that a tag with an internal anchor and a long, trailing, information-carrying tube will have some effect on the fish in its natural environment including: (1) entanglement of the tag in vegetation, trapping the fish or enlarging tagging wounds as a result of the entanglement, (2) accumulation of algae or other vegetation on the tag affecting the fish's performance capabilities and ability to compete with other non-tagged fish or enlarging tagging wounds, and (3) making a fish more susceptible to predation as a result of the tag's appearance and possible attraction of predators.

## CHAPTER VI

## SUMMARY

1. Forty-eight channel catfish, Ictalurus punctatus (Rafinesque), ranging from 125 to 198 mm in length and 14 to 60 g in weight were randomly divided into four test groups to evaluate the effects of the following marking techniques: (1) partial fin clip of the left pelvic fin, (2) cold branding using liquid nitrogen as a coolant, and (3) an injected spaghetti-type floy tag. Fish were observed for 12 weeks for duration of marks, abnormal behavior and mortality attributed to the marks, and to determine if the marks had any significant effect on growth.

2. It was determined that the partial fin clip of the left pelvic fin had completely regenerated before the termination of the 12-week observation period, and that no abnormal behavior or mortality could be attributed to the mark. It was also statistically determined that the partial fin clip had no significant effect on the growth of the fish during the test period as compared to other test groups.

3. Cold brands using liquid nitrogen as a coolant were still recognizable after the 12-week period, but were somewhat faded and distorted due to the growth of the fish during the test period. There was no abnormal behavior or mortality observed during the experimental period with the exception of one individual which was subject to infection in the branded area. Statistical analysis of the test group receiv-

ing cold brands indicated that this method of marking had no significant effect on growth of the fish during the experimental period as compared to other test groups.

4. Two-thirds of the test group receiving spaghetti-type floy tags had shed their tags within the first three weeks of the experiment. This high rate of tag loss was attributed to the small size of the fish and the lack of a strong, solid attachment of the internal anchor of the tag between the interneurals of the fish. Behavior of tagged fish was altered as evidenced by erratic swimming and continual rubbing of the tag injection site on the bottom of the pool. Mortality was very high in this test group; only 33% of the original number survived throughout the entire experimental period. Those that survived were those fish able to shed their tags, sustaining relatively little damage to themselves in the process. Statistical analysis of the surviving fish in this group indicated that there was no significant effect on the growth of the fish as compared to other test groups.

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