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# DELAYED MORTALITY AND PHYSIOLOGICAL STRESS RESPONSES IN TOURNAMENT-CAUGHT LARGEMOUTH BASS (<u>Micropterus</u> <u>salmoides</u>)

A Thesis

Presented for the

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Degree

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Warmest regards go to my family, who have continued to support and encourage me throughout my graduate studies.

## ABSTRACT

Largemouth bass (Micropterus salmoides) from 12 fishing tournaments during May 1988 through November 1989 (N = 457) were examined to evaluate delayed mortality and to characterize the physiological stress responses displayed by tournament-caught bass. Delayed mortality was highly significantly ($P \leq 0.01$) related to the effects of water temperature and differing reservoirs. The effects of differing tournament organizations were also significant ($P \leq 0.05$), but duration of live well confinement displayed no significant effects upon delayed mortality. Fish held in the live well for different periods of time displayed differences in plasma glucose levels which were near significance at the $P \leq 0.05$ level. Fish held for longer than 4 hours seemed to display slightly lower glucose values than fish held less than 4 hours. Plasma cortisol concentrations were significantly ($P \leq 0.05$) correlated with the relative weights of individual fish. No other significant physiological stress responses to tournament-related stressors were apparent as indicated by the blood plasma indicators of chloride, glucose, or cortisol concentrations.

TABLE OF CONTENTS

## LIST OF TABLES

LIST OF FIGURES

# CHAPTER I

## INTRODUCTION

The increasing numbers of largemouth bass fishing tournaments have both fishermen and fisheries managers concerned over the impact tournaments may have on the fishery as a resource. Although most tournaments have incorporated a policy of releasing fish once they have been weighed, some mortality may still occur for several days following the tournament.

This delayed mortality has been estimated to range from 0-43% (May 1973; Welborn and Barkley 1974; Archer and Loyacano 1975; Moody 1975; Seidensticker 1975; Warnecke 1986; Schramm et al. 1987; Bryan 1988; Bennett et al. 1989). The higher mortalities were associated with tournaments which were held in warmer water temperatures. Schramm et al. (1987) found prerelease and total mortality were significantly related to water temperature but not significantly related to air temperature, cumulative solar radiation, number of fishing teams, mean catch per team, or mean weight per fish. However, postrelease (delayed) mortality was not found to be significantly related to any of the measured variables. The water temperatures observed in their study were from 17-30 C, with a mean of 25 C. Unpublished studies at the University of Tennessee, by

Dr. J. L. Wilson, indicated delayed mortality may increase markedly in water temperatures greater than those observed during reported mortality studies. Since water temperatures in excess of 30 C are common in local reservoirs during late summer, a time when many bass tournaments are conducted, this may be cause for concern.

The duration of time a fish is held in the live well has also been suggested as having an effect upon the survival of released fish (Seidensticker 1975), due to confinement-induced stress and/or poor water quality within the live well. Carmichael et al. (1984a,b) documented physiological stress responses in hatchery-reared largemouth bass exposed to confinement and/or poor water quality. Because fish are caught and held in live wells for a period of time which may be as long as 13 hours in some tournaments, there may be some difference in the stress responses and mortality of bass held for differing durations before weigh-in. The stress responses observed by Carmichael were differentially affected by the temperature to which the bass had been acclimated. This seems to suggest bass might react to the similar stresses of tournament-related handling differently at different temperature regimes.

Plumb et al. (1988), in a simulation of tournament conditions, observed higher delayed mortality rates among fish experiencing warmer postrelease water temperatures.

In addition, they found that fish which had been released immediately experienced significantly lower delayed mortality than those fish held in the live well for 3-9 hours. It appears that live well confinement induced stress to a point which impaired the survival of the simulated "tournament-caught" bass. If this is the case, differing periods of live well confinement may also have different effects upon the survival and stress responses of tournament-caught fish.

The objectives of the present study were to examine delayed mortality and the associated physiological stress responses of tournament-caught largemouth bass. Specific objectives included: (1) assessing the effects of water temperature, duration of live well confinement, relative weight, tournament location, and differing handling procedures on the survival of bass, and (2) characterizing the physiological stress responses of bass subjected to the aforementioned stressors. Using this information, recommendations may be made to modify tournament procedures in order to enhance the survival of released fish.

# CHAPTER II

## METHODS

Mortality data and blood samples were collected from fish caught during 12 bass tournaments on four Tennessee Valley Authority reservoirs. However, mortality data from one tournament was not used in the analyses, due to the possible loss of moribund bass during the holding period following weigh-in. Reservoirs sampled included the Fort Loudon-Tellico complex and Cherokee, Douglas, and Watts Bar reservoirs.

Largemouth bass, *Micropterus salmoides*, were collected at tournaments in 1988 beginning in May and continuing through October, and July through November of 1989. Smallmouth bass, *M. dolomieu*, were not included in the sampling because their thermal preferences and tolerances are thought to be different than those of largemouth bass (Coutant 1975).

Tournament weigh-in procedures varied between sponsoring groups, but all operated under similar rules regarding angler conduct. Only artificial lures were used, and anglers were encouraged to ensure the survival of fish through penalties, which were assessed for weighing-in dead fish. Tournament duration ranged from 8 to 13 hours. The type of tournament varied from large (over 150 boats) to small (approximately 10 boats) events.

Anglers transported fish from their boat live wells to the weigh-in area in plastic bags filled with water. The actual weigh-in procedure, during which the fish were out of water, generally lasted 2-3 minutes. However, during some tournament weigh-ins fish were out of the water for considerably longer periods of time. Some of the larger tournaments possessed the resources to employ highly organized weigh-in procedures and to utilize holding tanks in which fish were allowed to "recover" following weigh-in, before being released into the reservoir. The weigh-in procedures of smaller tournaments were generally less structured, and fish were released into the reservoir immediately after weigh-in.

The sample groups consisted of fish that had been in the live well for 4 hours or longer and those that had been in the live well for less than 4 hours. Fishermen were asked to mark bass they caught, indicating the duration of live well confinement an individual fish had experienced. For 1988 tournaments, fishermen were asked to mark fish by clipping the right pelvic fin of fish confined in the live well less than 4 hours, and to clip the left pelvic of fish confined greater than 4 hours. Some fishermen who caught small numbers of fish did not clip fins, but were able to remember the time the fish were caught, and subsequently, the fins were clipped by research personnel. In 1989 tournaments, fish were marked by fishermen inserting colored safety pins through the mandible of the bass. The different colors corresponded to the duration of live well confinement the fish had experienced. The pelvic fins were then clipped by research personnel to signify the live well confinement group to which the fish belonged.

These live well-confined groups were compared, when possible, to a control group of fish collected at the time of the tournament. Bass obtained from the reservoirs by 220-V AC-current electrofishing served as the controls. These fish were kept in a holding tank for as long as 1 and 1/2 hours before blood samples were taken. Controls were marked by placing a notch in their anal fin, and they were placed in the net cage with the tournament-caught bass. Control groups were obtained in nine tournaments, but sample sizes were small in some of the tournaments.

Bass were collected prior to being released into the reservoir, after having been through all of the handling procedures of a particular tournament. Elapsed time, from the point at which fish were removed from the boat live wells to release in the reservoir (or experimental holding tank), ranged from approximately 10 to 30 minutes. The fish were held in a 400-L fiberglass tank, with a flow-through of reservoir water, prior to being anesthetized for collection of blood samples; holding periods ranged from 1 minute to 1 hour. The weight and total length of each fish were also recorded.

Delayed Mortality

All of the marked fish were held in a floating net cage, measuring 4.8 m long x 2.4 m wide x 2.4 m deep, where they remained for up to 12 days. The net was made of 2.5 cm bar mesh nylon. Fish were transported to the holding cage by boat in coolers or by truck in a bed-mounted fiberglass transport tank. Transport distances ranged from 100 m to 2 km. The holding cage was located in a boat slip at the marina nearest the weigh-in site. Depths where the net cages were placed ranged from 3-15 m.

For the first seven days after the tournament, daily measurements of water temperaure and dissolved oxygen were recorded at the surface and at depths of 1 and 2 m in the net cage. A Yellow Springs Instrument Co. Model 51B oxygen meter was used to determine temperature and dissolved oxygen levels. Mortalities were recorded, noting each fish's length and the treatment group to which they belonged. Observations were made every other day after Day 7, including Day 12 when the surviving fish were released into the reservoir. The treatment group of each released fish was recorded, along with its length.

Delayed mortality was calculated as the percentage of fish in each group that died after being held in the net cage. Arcsine transformations of mortality percentages were performed prior to statistical analysis, in order to minimize bias introduced from 100% mortality or survival. Mean holding water temperatures were measured for each tournament. The relationship between transformed mortality rate and the effects of mean water temperature, duration of live well confinement, differing reservoirs, and differing sponsoring groups was examined.

The statistical model used the fixed effects of the variables temperature, temperature squared, reservoir, tournament group, and duration of live well confinement. The variable temperature squared was included to test the possibility of temperature relating to mortality in a non-linear fashion. Random effects were also included in the model. Individual tournaments nested within a reservoir and live well confinement duration in individual tournaments nested within a reservoir were considered as random variables. Temperature and temperature squared were the only continuous variables, the others were discrete.

Statistical analyses for these variables were performed using the General Linear Mixed Models program

(GLMM), developed under the Southern Regional Project S-189 of the USSES (University Statisticians of Southern Experiment Stations). Some of the variables were dropped from the final model due to a lack of significant effects upon the mortality estimates. The variables water temperature, reservoir, tournament organization, and live well confinement duration were used to obtain the mortality estimates derived from the final model.

It was not possible to include relative weight (Wr) as a variable within the model; since individual fish were not identified after experiencing mortality, the relative weights of dead fish were unknown. The model used only allows for the inference of main effects upon mortality. Due to limitations of the available data set, the presence and examination of interactions among different variables were not examined.

Physiological Stress

Bass were anesthetized by immersion in a cooler filled with 20 L of an aqueous solution of tricaine methanesulfonate at a concentration of 325 ppm. Fish became completely quiescent in 1-3 minutes, and were then removed from the solution. They were placed ventral side up in a V-shaped plexiglas trough, which minimized the amount of hand contact they received during the bleeding process. Using a 21-gauge heparinized syringe which was

3.8-cm long, approximately 1.0 cc of blood was withdrawn from the hemal arch of each bass. The hemal arch was tapped by inserting the needle into the ventral midline between the anal fin and the caudal peduncle as described by Strange (1985).

Blood samples were transferred to a 1-ml polystyrene centrifuge tube, capped, and kept in an ice bath prior to centrifugation. The samples were centrifuged in a Fisher Model 59 centrifuge for 5 minutes at 2500 G. The plasma supernatant was pipetted (with diSPo Pasteur-type soda lime glass pipets) into a Corning Science Products 1.2-ml cryovial which remained in an ice bath during transport to the laboratory. At the laboratory, vials were stored in a freezer at a temperature of -20 C for later analysis. Plasma samples were allowed to thaw at room temperature prior to all analyses.

Plasma samples were analyzed to determine the concentrations of several indicators of sublethal stress. Plasma chloride and glucose concentrations were examined in an effort to estimate the extent of secondary stress effects expressed by the fish. Cortisol concentrations were examined as an indication of primary stress effects. Depressions in the plasma chloride concentration and elevations of plasma glucose and cortisol concentrations are considered to be indications of stress in fish (Mazeaud et al. 1981).

Plasma chloride concentrations were determined with the use of Sigma Diagnostics Corporation's colorimetric procedure No. 460, which is based upon a mercuric thiocyanate reaction. Sigma Diagnostics colorimetric procedure No. 510 was used in the analysis of glucose concentrations. This procedure is based on glucose oxidase/peroxidase reactions with o-dianisidine serving as a chromogenic indicator. The procedures were performed as prescribed with one exception: the amount of unknown and reagent used in glucose analysis was 3/5 of the amount prescribed. This change in procedure was approved by a Sigma Diagnostics technical representive during a telephone conversation, and allowed for the conservation of both plasma sample and reagent. Colorimetric quantifications were made with a Bausch and Lomb Spectronic 2000 spectrophotometer. Concentrations were determined by calculation from a regression of the absorbances of known standards.

The determination of cortisol concentration was made with the use of Diagnostic Products Corporation's Coat-A-Count cortisol assay. In the Coat-A-Count radioimmunoassay, cortisol labeled with the radio-isotope Iodine-125 competes with the cortisol in the plasma sample for binding sites on the interior of a polypropylene test tube, which is coated with a cortisol-specific antibody. After a fixed duration of competitive binding, the reaction is stopped by decanting the tubes, and the amount of Iodine-125 left in the tube is quantified by a gamma counter. The amount of radioactively-labeled cortisol in the tube is inversely proportional to the concentration of the unknown sample, and the sample concentration is calculated from a calibration curve using known sample concentrations. The antibody used to coat the tube is highly specific for cortisol, with very low cross-reactivity to other compounds commonly found in fish plasma.

Statistical analyses of blood parameters were performed using the General Linear Mixed Models program (GLMM), developed under the Southern Regional Project S-189 of the USSES (University Statisticians of Southern Experiment Stations). Glucose and cortisol concentration values were transformed to the natural log of the values prior to analyses. Chloride concentration values were not transformed.

The statistical model used the fixed effects of the variables temperature, temperature squared, relative weight of individual fish, reservoir, tournament group, and duration of live well confinement. The variable temperature squared was included to test the possibility of temperature relating to the observed physiological stress responses in a non-linear fashion. Random effects were also included in the model. Individual tournaments nested within a reservoir and live well confinement duration in individual tournaments nested within a reservoir were considered as random variables. Relative weight, temperature and temperature squared were the only continuous variables, the others were discrete.

The model used only allows for the inference of main effects upon the physiological stress responses of the bass. The stress responses observed by Carmichael et al. (1984a) were differentially affected by the temperature to which bass had been acclimated. This seems to suggest bass might react to the similar stresses of tournament-related handling differently at different temperature regimes. However, limitations inherent to the available data set prevent examination of interactions among the different variables.

## CHAPTER III

## RESULTS

### Delayed Mortality

The observed delayed mortalities are presented in Table 1. Nine tournaments had higher delayed mortalities for fish experiencing live well holding times greater than 4 hours. Two had higher mortalities for fish held less than 4 hours. Mortality rates varied between tournament groups. Generally, those tournaments with the most efficient weigh-in procedures exhibited higher survival rates for released fish. Water temperatures ranged from 15-33 C for the 12 tournaments. A trend toward higher mortalities was observed with increased temperatures.

Fish were held for 12 days following the 1988 tournaments. Only 4 fish which survived past day 5 had died before day 12. In addition, no bass died after day 7. Therefore, in 1989 tournaments, fish were held for only seven days. During the holding periods following the two 1988 tournaments on Cherokee Reservoir, storms destroyed the net cages after day 5. Consequently, mortality rates for these tournaments reflect 5-day holding periods. The mortality rates for the 1989 tournament held on Cherokee were considered unreliable due to the possible loss of several moribund bass; these

Table 1. Delayed mortality (%) and mean water temperatures experienced by tournament-caught largemouth bass. Sample size (N) is within parentheses.

| Date | Mean Temp. C | Mortality rate (%) In live well < 4 hours | In live well > 4 hours | Control |
|---|---|---|---|---|
| 5-21-88 | 22.1 | 20 (5) | 50 (12) | - |
| 6-4-88 | 26.6 | 0 (9) | 0 (13) | 0 (4) |
| 6-18-88 | 27.3 | 20 (20) | 52 (31) | 0 (11) |
| 7-31-88 | 30.1 | 93 (15) | 84 (37) | 100 (5) |
| 8-7-88 | 30.6 | 50 (16) | 63 (24) | 18 (17) |
| 9-10-88 | 25.2 | 11 (9) | 33 (9) | - |
| 10-8-88 | 20.2 | 17 (6) | 10 (30) | 0 (2) |
| 7-1-89 | 25.8 | - | - | - |
| 8-5-89 | 27.0 | 71 (24) | 83 (23) | 100 (3) |
| 9-9-89 | 26.3 | 90 (10) | 92 (22) | 0 (5) |
| 10-15-89 | 17.8 | 20 (10) | 14 (21) | 0 (2) |
| 11-4-89 | 16.3 | 0 (7) | 0 (12) | 0 (1) |

rates were not included in the analyses. At the end of the holding periods for each tournament, bass unaccounted for were considered to be surviving fish.

Statistical analysis revealed no significant effects on delayed mortality for the variable of time in the live well (Table 2). However, there were highly significant effects ($\underline{P} \leq 0.01$) observed for the variables of water temperature and differing reservoirs. Mortality increased significantly as water temperature increased. There were also significant differences in the mortality observed on different reservoirs. Significant differences ($\underline{P} \leq 0.05$) were observed between the delayed mortality estimates of different tournament organizations.

Physiological stress

No significant differences were found in plasma chloride concentrations for bass held at different water temperatures, on different reservoirs, among different tournament groups, or fish exposed to differing durations of live well confinement (Table 3). Also, the relative weights of fish, a measure of overall health, demonstrated no significant relationship with chloride concentration. Tournament-caught bass, which experienced live well confinement, demonstrated chloride values similar to those of the electroshocked controls.

Table 2. Analysis of significance for the relationships of delayed mortality to selected tournament conditions. Asterisks denote significant differences ( * denotes $\underline{P} \leq 0.05$; ** denotes $\underline{P} \leq 0.01$).

| Variable | Test Statistic | P |
|---|---|---|
| Water temperature (C) | 21.0 | 0.0002 ** |
| Lake | 8.8 | 0.0007 ** |
| Tournament group | 3.0 | 0.035 * |
| Time in the live well | 0.6 | 0.56 |

Table 3. Analysis of significance for the relationships of plasma chloride concentrations to selected tournament conditions.

| Variable | Test Statistic | P |
|---|---|---|
| Time in the live well | 1.10 | 0.33 |
| Water temperature (C) | 0.90 | 0.34 |
| Relative weight | 0.33 | 0.57 |
| Tournament group | 0.62 | 0.68 |
| Lake | 0.29 | 0.83 |

Differences near significance at the $\underline{P} \leq 0.05$ level were observed between the glucose values of fish exposed to live well confinement durations greater than 4 hours, and those held in the live well for shorter durations (Table 4). Fish held longer than 4 hours seemed to experience depressed glucose concentrations at warmer water temperatures, suggesting an interaction between the effects of water temperature and live well confinement. Meanwhile, glucose concentrations of electroshocked controls demonstrated values which were not statistically different than either of the live well confined tournament groups. No significant effects on glucose concentrations were found for the variables of water temperature, reservoir, tournament group, or relative weight.

Cortisol concentrations of tournament-caught bass were not significantly related to water temperature, differing reservoirs, tournament group effects, or differing durations of live well confinement (Table 5). Electroshocked controls exhibited cortisol values similar to tournament-caught bass. The relative weights of bass, within a specific reservoir and across all reservoirs, were found to have a significant effect upon the cortisol concentration of sampled fish ($\underline{P} \leq 0.05$). There was a negative correlation between relative weight and the

Table 4. Analysis of significance for the relationships of plasma glucose concentrations to selected tournament conditions.

| Variable | Test Statistic | P |
|---|---|---|
| Time in the live well | 2.80 | 0.06 |
| Tournament group | 1.73 | 0.13 |
| Water temperature (C) | 0.13 | 0.72 |
| Lake | 0.21 | 0.89 |
| Relative weight | 0.01 | 0.92 |

Table 5. Analysis of significance for the relationships of plasma cortisol concentrations to selected tournament conditions. Asterisks denote significant differences ( * denotes $P \leq 0.05$).

| Variable | Test Statistic | P |
|---|---|---|
| Relative weight (intra-reservoir) | 5.11 | 0.024 * |
| Tournament group | 0.99 | 0.43 |
| Lake | 0.69 | 0.56 |
| Time in the live well | 0.56 | 0.57 |
| Water temperature (C) | 0.22 | 0.64 |

cortisol concentration; fish with higher relative weights expressed significantly lower cortisol values.

CHAPTER IV

# DISCUSSION

## Delayed Mortality

A highly significant relationship was observed between delayed mortality and water temperatures (Figure 1). Delayed mortality was greatest at warmer water temperatures. Treatment groups from tournaments with mean water temperatures below 18 C experienced delayed mortality rates between 0-20 %. When mean water temperatures were above 30 C, delayed mortality rates of tournament-caught bass ranged from 50-93 %.

Delayed mortality rates of largemouth bass for the 11 tournaments were similar to those previously reported, at similar temperatures (May 1973; Welborn and Barkley 1974; Moody 1975; Seidensticker 1975; Schramm et al. 1987; Bryan 1988). However, the previous studies have generally been conducted at water temperatures cooler than those observed during this study. Water temperatures as warm as 33 C were observed during this study, while no previous study experienced temperatures greater than 30 C, and most were less than 25 C (May 1973; Welborn and Barkley 1974; Archer and Loyacano 1975; Seidensticker 1975; Schramm et al. 1985; Schramm et al. 1987; Bennett et al. 1989). Consequently, some of the delayed mortality estimates are higher than those found

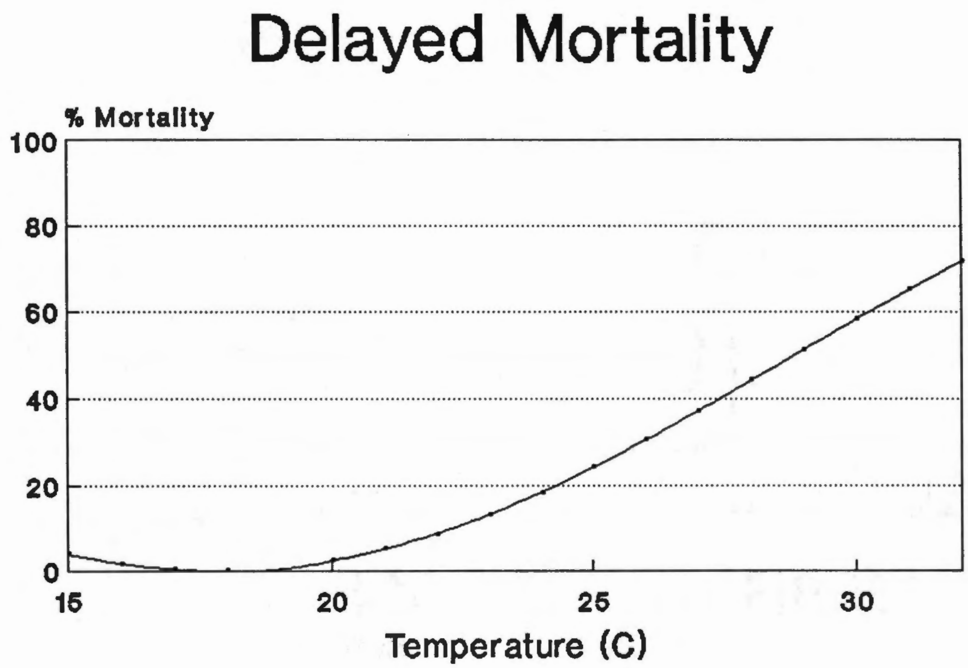


Figure 1. Delayed mortality rate (%) as related to water temperature for 11 tournaments. Mortality rate determined from regression of arcsine transformed percentages within estimates of effects derived from the statistical model.

in earlier studies.

No significant relationship between the duration of live well confinement and delayed mortality was found. Therefore, the amount of time a fish spends in the live well appeared not to have as great an effect on its survival as some of the other examined influences. This suggested that fish at cooler temperatures were able to withstand the additional stress brought about by long periods of live well confinement. At warmer temperatures, confinement stress may not have as great an effect on mortality as other factors (e.g., decreased resistance to pathogens, poor water quality, etc.).

Significant differences were exhibited in mortality between different tournament organizations (Table 6). The weigh-in methods used by individual tournament organizers were not consistent from group to group. Some of the tournaments were large highly-organized events with >150 participants. Others were much smaller "wildcat" tournaments, or club sponsored events in which the weigh-ins were conducted by the fishermen themselves. Some of the tournaments were able to utilize more efficient weigh-in procedures, which limited the amount of time individual fish were handled. This high degree of efficiency appeared to result in lower delayed mortality of tournament-caught fish. Although it was not measured in this study, the amount of time fish remained

Table 6. Estimates of mortality (%) of tournament-caught bass from 6 tournament organizations.

| Organization | Mean | Standard Error |
|---|---|---|
| A | 7.81 | 3.47 |
| B | 12.42 | 1.07 |
| C | 17.31 | 3.04 |
| D | 36.68 | 0.93 |
| E | 63.56 | 0.77 |
| F | 63.64 | 3.83 |

in weigh-in bags seemed to have a considerable effect on the survival of the fish. The use of a holding tank in which fish were allowed to recover may have resulted in lower delayed mortality as well.

Differences were observed in the delayed mortality rates of tournaments conducted upon different reservoirs (Table 7). Degraded water quality parameters, including dissolved oxygen availability and contamination from pollutants, are probably primary factors influencing mortality. Those reservoirs with the highest estimated mortality (Watts Bar, Ft. Loudon-Tellico) were mainstream Tennessee River impoundments, while the reservoir with lowest mortality estimates was an upper storage impoundment (Douglas). The mortality estimates may be correlated with the degree of potential water quality degradation to which these reservoirs are subjected.

The relative weights (Wr) of fish obtained from each of these reservoirs were analyzed to detect differences in the condition of the fish sampled from these reservoirs (Table 8). Statistical analysis revealed that fish from Douglas Reservoir had significantly higher relative weights than fish from any other reservoir. Bass from Cherokee had significantly higher Wr values than bass from any reservoir exclusive of Douglas. Meanwhile, Watts Bar and the Ft. Loudon-Tellico Reservoir complex yielded fish with the lowest Wr values, and these

Table 7. Estimates of mortality (%) of tournament-caught bass on 4 Tennessee Valley reservoirs.

| Reservoir | Mean | Standard Error |
|---|---|---|
| Douglas | 2.52 | 1.24 |
| Cherokee | 29.91 | 2.36 |
| Ft. Loudon-Tellico | 45.36 | 0.89 |
| Watts Bar | 67.85 | 2.26 |

Table 8. Analysis of relative weights (Wr) of tournament-caught bass (N = 432) on 4 Tennessee Valley reservoirs. Means for reservoirs with the same letter are not significantly different.

| Reservoir | N | Mean |
|---|---|---|
| Douglas | 83 | 94.24 a |
| Cherokee | 131 | 91.57 b |
| Ft. Loudon-Tellico | 163 | 85.03 c |
| Watts Bar | 55 | 85.73 c |

values were not significantly different from each other.

The estimates of mortality derived from the statistical model indicated that those reservoirs with lower delayed mortality estimates were the same reservoirs which yielded bass with the higher Wr values. Because indivdual bass were not identified during recordings of mortality, no statistical comparison could be made between Wr and delayed mortality. However, there is an indication that relative weight may be correlated with delayed mortality in tournament-caught largemouth bass. If this is the case, bass which were in the best "condition" when caught may have experienced better survival after release. This may also have been a component in the effect differing reservoirs had upon mortality.

Inferences made using the delayed mortality data must take into account that the fish were subjected to post-release conditions which may have been sub-optimal for survival, and consequently, mortality rates could be artificially high. However, conditions should not be greatly different than those experienced by released fish not held in the net cage.

Another factor which may increase mortality rates is the fact that blood samples were taken from each fish; this may be an additional stressor to the fish. In order to test this, blood samples were obtained in a manner similar to study methods from 11 fish collected by AC electrofishing, These fish were held in tanks in the lab for 12 days. No mortalities were observed during this time. This fact seems to indicate the stress of blood collection was not enough to cause mortality in and of itself.

<u>Physiological Stress</u>

Although statistical significance was observed only for the relationship between cortisol and relative weight, the relationship between glucose and live well confinement duration approached significance at the $\underline{P} \leq 0.05$ level. Additionally, nonsignificant trends were observed which suggest there may be interactions between some of the variables. However, the statistical limitations of the available data restricts any discussion of interactions to speculation. Even though some of the observed trends were not found to be significant, they may be indicative of the stress response exhibited by the fish.

The amount of variation in plasma chemistry values among fish was great, even when the effects of the measured variables were taken into consideration. This individual variation decreased the sensitivity of the statistical tests. Carmichael et al. (1984a,b) demonstrated the effectiveness of directly comparing experimentally obtained plasma chemistry values to the "normal" values of unstressed bass. By comparing the mean plasma chemistry values determined from tournament-caught bass to normal values, the inferences made from the statistical analyses may be supplemented. The normal values for hatchery-reared largemouth bass as determined by Carmichael et al. (1984a) were used in the comparison because normal values for wild largemouth bass were not available. Hatchery-reared bass should possess normal values which are not greatly different from those of wild bass. Basal plasma chloride, glucose, and cortisol values were found to be similar in both wild and hatchery-reared rainbow trout (Wydoski et al. 1976; Caldwell and Strange 1987), and largemouth bass should exhibit similar tendencies.

Water temperature displayed no significant effects upon any of the measured plasma characteristics. The effects of increasing water temperature on plasma chloride, glucose, and cortisol concentrations were variable, and seemed to be interrelated with live well confinement duration. Glucose and cortisol concentrations appeared to increase slightly in fish held for shorter periods at the warmer temperatures. But those held for longer periods appeared to exhibit decreasing concentrations at higher temperatures. The stress responses observed by Carmichael et al. (1984a,b) were differentially affected by the temperature at which the bass had been acclimated. This seems to suggest bass might exhibit different responses to stress at different temperature regimes.

Although water temperature did have a significant effect upon survival of tournament-caught largemouth bass, the observed plasma chemistry indicators of sublethal stress seemed to be only slightly affected. This suggests that physiological stress is not a direct factor in delayed mortality associated with high water temperatures. However, physiological stress due to tournament stressors may have an adverse affect upon the immune system of the fish. This may result in a decrease of a fish's ability to combat bacterial and fungal infections, which tend to be more severe at warmer water temperatures. Delayed mortality may be affected by water temperature primarily through a reduced ability to combat pathogens and/or osmoregulatory dysfunction which may occur hours after live release.

The duration of live well confinement was found to have a near significant ($\underline{P} \leq 0.05$) effect upon plasma glucose concentrations but not the other measured stress indicators, chloride and cortisol concentrations (Figure 2). Fish held in the live well for longer periods of time seemed to exhibit a more pronounced depression of glucose values at warmer water temperatures. This was

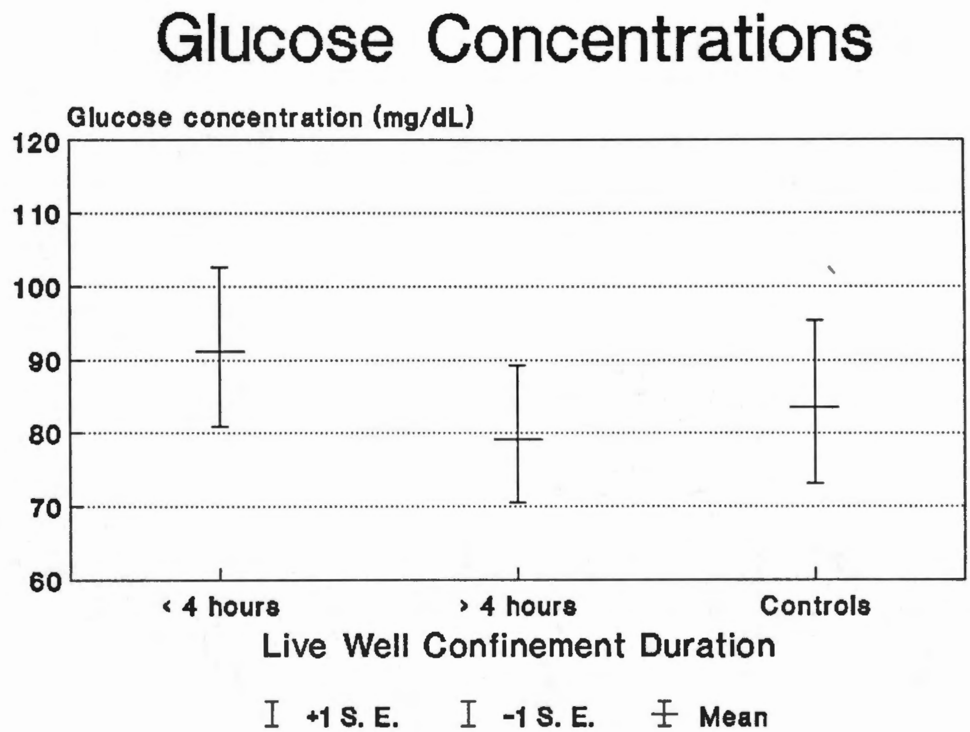


Figure 2. Glucose concentrations for 3 live well confinement duration groups. < 4 hours refers to tournament-caught bass held in the live well for less than four hours before weigh-in, while >4 refers to those held more than four hours before weigh-in. Controls were bass which were captured by electrofishing and spent no time in the live well, nor exposed to weigh-in stressors, but were exposed to experiment-related stressors.

not caused by either high water temperatures or longer periods of live well confinement alone, suggesting an interaction between the effects of live well confinement and water temperature.

The apparently lowered glucose levels may, in part, have been due to metabolic consumption of glucose, which would occur at a faster rate at warmer temperatures. Since corticosteroids and catecholamines (adrenaline), which have been implicated in the production of glucose (Mazeaud et al. 1977), are metabolized at a slower rate than glucose, one would expect these to remain elevated if glucose depression were due to metabolic consumption. Some disruption of the processes responsible for production of corticosteroids and catecholamines may have occurred during the longer period of live well confinement. Glucose concentrations have been shown to drop to near basal levels in confined channel catfish near death (Strange 1980), while cortisol concentrations remained elevated at 20 C. But dying fish at 30 C exhibited depressed cortisol and glucose concentrations. Therefore, evidence exists for the possibility of temperature-related disruptions in interrenal and adrenergic stress response functions.

Bass held for short periods exhibited similar chloride concentrations at all temperature ranges, and these values were also similar to values obtained for control fish. Tournament-caught bass held in the live well longer than 4 hours seemed to exhibit slightly decreased chloride concentrations at warmer temperatures, but these values were still within previously described normal ranges. Tournament-related stressors did not appear to compromise osmoregulation prior to the point at which bass would be released into the reservoir. However, Gustaveson (1978) did not observe significant decreases of chloride concentration in hooking-stressed largemouth bass until 24 hours after the initial stressor. In other studies of the stress responses of largemouth bass (Carmichael et al. 1984a,b; Williamson and Carmichael 1986), significant decreases in chloride concentrations were observed between 2-24 hours after application of a stressor. Therefore, osmoregulatory dysfunction may still have occurred, even though the bass displayed normal chloride concentrations at the point of release.

Estimates of mean glucose concentrations of tournament-caught and control fish were within the normal ranges of hatchery-reared bass, as were chloride values. Cortisol value estimates of both control and tournament-caught bass were greater than those established as "normal" for hatchery-reared largemouth bass. Tournament-caught bass did not seem to express more severe stress responses than the controls, which were electroshocked. Electroshocking did not evoke hyperglycemic conditions in the time between the shock and blood sampling. Compared to the normal values of hatchery-reared bass, immediate increases in plasma cortisol concentrations were, however, evident after electroshocking. Similar observations have been recorded with electroshocked rainbow trout (Schreck et al. 1976; Mesa and Schreck 1989). The concentrations reported in those studies were very near the concentrations of control bass obtained here. Electroshocking of bass to obtain controls within this study induced stress which may have contributed to the mortality observed for controls in the delayed mortality assessments.

A significant negative correlation was observed between the relative weights of tournament-caught bass and cortisol concentrations. Bass with higher relative weights, a measure of the condition of the fish, displayed slightly lower cortisol values (Figure 3). No significant effects upon other plasma indicators were observed for relative weight. Giesy (1988) reported a positive correlation between condition factor (K) and cortisol concentrations in largemouth bass. He also found cortisol concentrations to be positively correlated with weight, but negatively correlated with length. Giesy observed many of these bass had been subjected to starvation stress, and hypothesized that starved bass

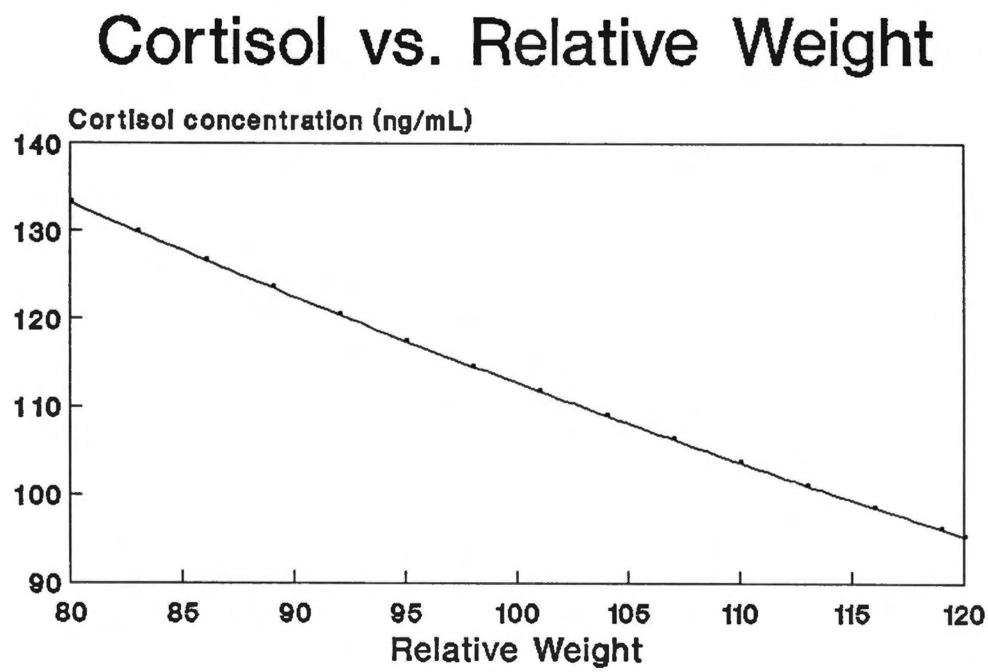


Figure 3. Relation of cortisol concentrations and relative weight of bass from 4 East Tennessee reservoirs. Regression formula determined from estimates of natural log of cortisol concentrations derived from statistical model.

exhibited different cortisol responses to capture stress than bass not subjected to starvation. The negative correlation of cortisol with length he observed may be more indicative of cortisol response in healthy fish. The indication is that larger fish, in better condition, exhibit slightly less severe stress responses as indicated by cortisol concentrations. However, the effect of this lessened response was not great enough to significantly influence plasma glucose or chloride concentrations at the point of blood sampling.

Although significant differences in relative weight were observed between reservoirs, no significant reservoir effects on plasma indicators of sublethal stress were observed. However, when statistical model estimates of the mean plasma values for each reservoir are ranked due to their distance from mean baseline (nonstressed) values, reservoir rankings are consistent for each plasma indicator. Douglas Reservoir yielded the estimates closest to baseline, followed by the Ft. Loudon-Tellico complex and then Cherokee. Watts Bar consistently displayed estimated plasma values most indicative of stress. All reservoir estimates of plasma chloride concentrations fell within normal ranges; different reservoirs appeared to demonstrate no influence on osmoregulation at the point of live release. Of estimated glucose values for each reservoir, only Watts

Bar yielded an estimated value greater than the normal range. Cortisol estimates were higher than normal for every reservoir, but the Watts Bar value was twice that of the other reservoirs.

There were no significant trends observed which indicate differences in the plasma values obtained on different reservoirs, but there did seem to be a consistent trend for certain reservoirs to yield fish which expressed slightly more of a stress response. This did not appear to be linked entirely to the relative weight of fish from the reservoir, but to some other factor. Water quality, and dissolved oxygen availability in particular, may be the primary factor.

No significant relationships were found between plasma chemistry indicators of sublethal stress and differences in which group conducted the tournament. There were variations in the weigh-in and live release procedures utilized by different tournament organizations, but the varying procedures had no significant effect upon any of the measured plasma indicators. None of the estimates of chloride concentrations obtained from the statistical model were outside the normal ranges of hatchery-reared bass. Estimated glucose values were lowest for the two tournaments incorporating recuperative holding periods of bass before release into the reservoir. Of those which did not, three of four had estimates slightly greater than normal values, with the "wildcat-style" tournament yielding the highest value. While all cortisol estimates were above normal ranges, the two tournaments using recovery tanks possessed the lowest values, and the highest were associated with the wildcat-style event. One tournament which used recovery tanks added salt and quinaldine to the water (in 4% and 1% concentrations, respectively). The addition of these chemicals may have helped that group achieve the lowest estimated glucose and cortisol values. No consistencies were observed with rankings of the other groups between glucose and cortisol estimates. In addition, the rankings of groups showed no consistencies between plasma chloride estimate rankings and rankings associated with glucose or cortisol estimates. The results indicate that utilization of recuperative holding tanks after weigh-in and before release into the reservoir may slightly decrease the stress response exhibited by tournament-caught bass.

# CHAPTER V

## SUMMARY

Delayed mortality rates and water temperature were significantly related, with the highest delayed mortality occurring at the warmest water temperatures. If organizers avoid conducting tournaments when water temperatures are above 25 C (77 F), bass caught and released after weigh-in should experience survival rates greater than 75%. Conversely, tournaments held when water temperatures are greater than 30 C (86 F) may experience survival rates no better than approximately 40%. The Bass Angler's Sportsman's Society (B.A.S.S.), for example, avoids warm water temperatures by conducting tournaments on lakes at more northern latitudes during late summer. Tournament organizations which are confined to a limited geographical region, however, cannot utilize such a strategy. As such, they might choose to limit the number of tournaments they conduct, or number of fish weighed-in per fisherman, or some other strategy when water temperatures are warmer than a certain point.

The length of time fish spend in the live well before weigh-in did not have a significant impact upon the delayed mortality of bass. Additionally, as evidenced by the lower glucose values of fish confined for longer periods, the stress related to live well confinement appears not to be as severe as that related to capture. Therefore, it does not appear that tournaments should incorporate multiple weigh-ins throughout the day in order to limit the amount of time fish spend in live wells. It also appears that the design of live wells, and application of their use by tournament fishermen are suitable to ensure the health of fish at the time of weigh-in.

Delayed mortality rates between the four reservoirs were found to be significantly different, while no differences were found in the physiological stress responses of fish from different reservoirs. Some reservoirs will experience more tournament-related delayed mortality than others due to unidentified factors. But, some of the differences between reservoirs may be ascribed to the condition of fish utilized by tournaments. The fact that fish with higher relative weights exhibited a less severe cortisol stress response, and that reservoirs with lower delayed mortality rates yielded fish with higher relative weights seems to indicate that bass which have experienced better foraging opportunities may be able to better withstand the stresses associated with being subjected to tournament capture and release. Consequently, tournament organizers may choose to avoid conducting tournaments on reservoirs in which the bass are in poorer "condition".

Differences were also found in the delayed mortality rates of bass caught and released by different tournament organizations. There were no significant physiological stress responses between organizations, as observed immediately after weigh-in, indicating no significant differences in the amount of stress that fish were subjected to prior to weigh-in. This also suggests that differential mortality between tournament groups may primarily be affected by variations in the weigh-in procedures utilized by tournaments. The individual effects of different weigh-in procedures upon delayed mortality should be examined in an effort to identify the more stressful procedures. Then, recommendations for tournament protocol may be made in an effort to limit weigh-in-related delayed mortality.

There is a possibility that interactions among the measured variables may exist, although the existing data did not allow for an examination of interaction effects. Further study should examine the effects of some of these interactions. For example, there is some indication that the physiological stress response observed for glucose was differentially affected at different water temperatures. The knowledge of the existence of such an effect would enhance the inferences one might make as to the occurrence of high temperature-related disruptions in the gluco-corticoid response to stress.

Additional studies might also want to contrast the responses to tournament-related stressors experienced by largemouth bass and smallmouth bass. On some area reservoirs, smallmouth bass comprise a substantial proportion of the tournament catch. Specific management strategies employed to reduce tournament stress on largemouth bass may be inappropriate for application to smallmouth bass.

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# VITA

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