PHYSIOLOGICAL EFFECTS OF SIMULATED TAGGING STRESS IN STRIPED BASS, *MORONE SAXATILIS*

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Introduction

Because of the major sport fishing interest in striped bass (*Morone saxatilis*) in the Sacramento-San Joaquin estuary, the California Department of Fish and Game (CDFG) annually assesses the number of spawning individuals. Spawning numbers are estimated by sampling and tagging the anadromous migrants in the estuary and in the Sacramento River with gill nets and fyke traps, respectively, by CDFG personnel (Collins 1982, White 1986). Sampled fish are placed in a canvas sling for tagging with numbered disc-dangler tags (Chadwick 1963), some of which carry rewards for return by anglers.

Striped bass are apparently stressed by the sampling and tagging operation because dead (tagged) striped bass were observed along estuarine and riverine banks near sampling sites, especially when water temperatures $\geq$20°C (D. Fenner, CDFG, personal communication). Past work has shown that observed dead fish may account for only a small percentage of the total mortality (Hill 1983). A differential mortality of tagged and untagged individuals significantly biases the population size estimates (Arnason and Mills 1987). We conducted two investigations, one in the field and one in the laboratory, to test hypotheses that differences in capture method provoke different physiological stresses in wild striped bass which are captured and tagged using CDFG procedures. The laboratory study also allowed measures of delayed (24 h post-stress) mortality.

Methods

The first investigation was conducted on board the CDFG sampling vessels where we sampled venous blood from fish right out of the gill net or fyke trap. Blood samples were collected with a heparinized syringe and needle, and we immediately measured blood O_2 and CO_2 tensions and pH using calibrated IL instrumentation thermostatted to fish temperature (15-18°C). In addition, we determined hematocrit by centrifugation and fixed a blood sample for [hemoglobin]. Remaining blood was centrifuged for subsequent measurements of plasma [HCO_3^-], [Na^+], [K^+], [Cl^-], [lactate], [glucose], [protein], and osmolality, as described earlier (Hopkins and Cech 1992).

The second investigation was conducted in the laboratory. Striped bass were purchased from a commercial aquaculturist and held in a round, outdoor holding tank (4 m diameter) for >12 mo. Holding tank received a continuous inflow of air-equilibrated U.C. Davis well water and continuous aeration. Fish were moved in groups of 10-15 to 1.3 m diameter, insulated fiberglass tanks, also equipped with continuous water inflows and aeration, in the laboratory for 1-3 wks acclimation to 22°C for physiological experiments. Throughout holding, fish were fed either Silver Cup or Biodiet fish pellets 3 times weekly. Fish were anesthetized with buffered (5 mM/l $NaHCO_3$) and salted (10 ppt NaCl) MS-222 solutions and chronically cannulated (dorsal aorta) with PE-60 polyethylene tubing (Nikinmaa et al. 1984). Fish were allowed to recover 3 d in 180 l, insulated fiberglass tanks. Initially tanks had 5 mM/l $NaHCO_3$ and 10 ppt NaCl which washed out in 4-6 h by a continuous freshwater inflow.

Three blood samples were drawn via the cannula from each fish (mean wet weight = 2.277 kg) and analyzed for several physiological variables. The first blood sample was a resting sample, taken after full recovery from surgery but before the fish was disturbed. The second sample was taken immediately after the simulated stress: either gill net, gill net + water, fyke trap, fyke trap + water, or controls (5-9 fish per group). Gill net and fyke trap stress were "worst-case scenarios" as observed aboard the tagging vessels: both included 3 min dry deck time (dry plastic ice chest in laboratory) before simulated tagging in a canvas sling. Gill net stress was simulated by using the identical (6 cm bar mesh) CDFG gill netting in the recovery tank. Gill netted fish would often struggle in the tank as they attempted to free themselves. Fyke trap stress was simulated by inducing fish to swim into a cage of plastic coated wire and be exposed to a mild current produced by a submersible pump. Proposed stress alleviation to both of these stresses was produced by adding water to the deck time (10 ppt salted water in ice chest) and to the simulated tagging (10 ppt salted water pumped through a tube into mouth of fish to keep gills ventilated). Control fish were sampled via cannulae at the appropriate time intervals, but were otherwise undisturbed in their recovery tanks. The third sample was taken 24 h after the fish had been stressed with subsequent stress recovery in fresh water.

Blood samples and data were analyzed using standard procedures. Arterial gas tensions (PO_2, PCO_2) and pH were measured immediately following sampling with a Radiometer PHM73 analyzer and electrodes. Hematocrit (packed cell volume by centrifugation 3 min at 11,500 x G), total [hemoglobin] (cyanmethemoglobin spectrophotometric determination), and [$lactate^-$] (YSI Model 27 analyzer) were also measured immediately. The remaining blood was centrifuged (5 min at 4,500 x G) and the plasma removed and frozen for subsequent measurements of [Na^+], [Cl^-], [K^+], [glucose], [protein], and total osmolality. Plasma samples were later thawed and [Na^+] and [K^+] were measured using an IL 343 flame photometer. Plasma [Cl^-] was measured using a Radiometer CMT10 chloride titrator. Plasma [glucose] were measured using the YSI model 27 analyzer. Plasma [protein] was measured using an American Optical refractometer and a B&L Spec 88 spectrophotometer. Total osmolality was measured with a Wescor 1500B vapor pressure osmometer. Data were analyzed using one-way ANOVA among treatment groups at the same time interval and repeated measures ANOVA among time intervals for each treatment group. Treatments on experimental days were randomized to avoid possible time-related effects.

Results and Discussion

Field data showed gill net-caught fish to be more acidotic and generally stressed than fyke trap-caught fish (Table 1). Gill net-caught fish showed a significantly increased venous PCO_2 and [lactate] and a significantly decreased venous pH from this mixed acidosis, compared with fyke trap-caught fish. This probably resulted from the combination of struggling (and, possibly, buccal-opercular movement constraints) in the gill net and subsequent air exposure on the deck.

Gill net-caught fish also showed significantly increased [glucose] and hematocrit values, whereas the other variables generally showed little change (Hopkins and Cech 1992). Hyperglycemia is commonly used as a stress indicator in fishes (Wedemeyer et al. 1990). Hematocrit increases from erythrocytic swelling has been previously described in striped bass after chasing-type exercise (Nikinmaa et al. 1984). Although we did not measure catecholamines, the significantly greater hematocrit in gill-netted, as compared with fyke-trapped, fish probably resulted from more severe catecholamine-induced erythrocytic swelling (Nikinmaa 1986). Differential hemoconcentration from fluid shifts can be ruled out because [hemoglobin] and [plasma protein] did not significantly differ between groups.

Table 1. Mean (SE) venous blood values of striped bass caught in gill nets or fyke traps, which differ, significantly, between the two capture methods. (From Hopkins and Cech 1992)

Variable	Gill Net (n=35)	Fyke Trap (n=40)
PCO_2 (mm Hg)	24 (2)	17 (2)
pH	7.04 (0.04)	7.32 (0.03)
Lactate (mM/l)	9.75 (0.66)	7.10 (0.37)
Glucose (mg/dl)	171.3 (11.2)	131.1 (6.2)
Hematocrit (%)	37 (1)	33 (1)

Laboratory studies showed that although resting values for all physiological variables were not statistically distinguishable among stress treatment groups, several changes occurred after stress exposures. Immediately after stress, both gill net and fyke trap fish showed significant ($p < 0.05$) arterial hypoxia (low PO_2), and hypercapnia (high PCO_2), compared with the other groups. These effects probably resulted from air exposure, because all non-control fish showed increased blood [lactate$^-$] and decreased blood pH, characterizing a struggling-related metabolic acidosis. The air-exposed treatments' increased PCO_2 (respiratory acidosis) produced deeper, mixed acidoses (lower pH). Air exposure appears to severely limit gas exchange in striped bass, presumably due to the lack of adequate support for the gill lamellae (gas exchanging structures) their consequent sticking together, and a resulting severely reduced gill surface area for gas diffusion. A presumed water shift from the extracellular compartment (including plasma) to the intracellular compartment, e.g. from increased catecholamine concentrations (Nikinmaa et al. 1984), increased hematocrit, especially in air-exposed treatments. Other variables showed little change. All physiological variables essentially returned to resting levels in recovered (24 h post-stress) fish. Three of the 9 fish exposed to gill net stress died.

In conclusion, fyke trap capture methods appear to induce less physiological stress than gill net methods for adult striped bass in fresh water. Both capture and tagging methods, as assessed by field and laboratory studies, induced blood gas disturbances, acidoses, and fluid shift responses in these fish. These responses apparently resulted from some combination of fyke trap stress, buccal or opercular constrictions from the gill net, handling, and/or by air exposure on deck and in the tagging sling. Air exposure (after capture and during tagging operations) appears to significantly impair gas exchange, leading to arterial hypoxia and hypercapnia in laboratory fish. Stress alleviation techniques, including recovery baths and artificial gill ventilatory flows to insure adequate bathing of the gills with salted (10 ppt NaCl) water, have been recommended to, and are being implemented by, CDFG.

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