Influence of local adaptation and interstock hybridization on the cardiovascular performance of largemouth bass *Micropterus salmoides*

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Summary

In this study, the cardiovascular response to exhaustive exercise among differentiated stocks of largemouth bass *Micropterus salmoides* was compared at 10°C and 20°C to assess the level of their local adaptation. In addition, the impact that interstock hybridization had on adaptive differences was assessed using F_1 hybrids. To accomplish these assessments, four genetically distinct stocks of fish were produced using adults from two regions in the midwestern United States identified as distinct conservation management units (central Illinois, IL and southeastern Wisconsin, WI): both P_1 stocks and both reciprocal F_1 interstock hybrids. Cardiac variables (both resting and maximal) were consistently lowest for the IL×IL stock relative to the WI×WI stock and both F_1 interstock hybrids. Interestingly, however, all groups of fish were able to maintain similar levels of cardiac scope. All fish responded to exercise by increasing heart rate and decreasing stroke volume, consistent with the notion that largemouth bass modulate cardiac output *via* frequency. After exercise, cardiac variables returned to resting levels 25–35% more rapidly for IL×IL fish relative to all other groups at 20°C. At 10°C, recovery rates for both P_1 stocks were similar but more rapid than the interstock hybrids. Collectively, these results indicate that the locally adapted stock (IL×IL) exhibited cardiovascular adaptations that enabled rapid cardiovascular recovery and maintenance of low resting cardiac output and heart rate. Conversely, the translocated stock (WI×WI) and the interstock hybrids required longer for cardiovascular variables to recover after exercise and exhibited higher resting levels of cardiac output and heart rate. This study provides some of the first direct evidence of a physiological mechanism by which mixing stocks could potentially decrease fitness and illustrates the magnitude of the intraspecific diversity of cardiovascular performance.

Key words: cardiac output, heart rate, local adaptation, outbreeding, translocation, largemouth bass, *Micropterus salmoides*.

Introduction

Studies in fishes have documented that substantial genetic variation can exist within a single species (Ryman and Utter, 1987; Philipp et al., 1983), and support the notion that each population has evolved an adaptive suite of genotypic and phenotypic characters for its specific environment (Wright, 1978). These studies also show that differences in performance and fitness occur when these populations are translocated from their point of origin to a novel environment (e.g. Philipp and Whitt, 1991; Philipp and Claussen, 1995; Philipp et al., 1995; Gharrett et al., 1999). Furthermore, introgression of genetically distinct populations may result in a decrease in overall fitness and performance characteristics (e.g. growth, thermal tolerance, resistance to infectious agents) of the new population (i.e. outbreeding depression) providing further support to the concept of local adaptation (Campton, 1987; Emlen, 1991; Hallerman, 2003; Philipp et al., 2002).

To date, few studies have examined biological differences among fish stocks delineated from the results of molecular genetic analyses, and even fewer have examined the physiological variation among putative stocks or the consequences of mixing them. Theoretically, one would predict that physiological variables would differ among locally adapted and genetically distinct populations, and that mixing individuals from different populations would result in alterations in these variables that could provide a mechanistic explanation for reductions in fitness. Interestingly, there have been few attempts to evaluate population-specific differences in cardiovascular performance, or the cardiovascular consequences of hybridizing fish from different populations. This is surprising since cardiovascular performance is intimately linked to the energetics and physical performance of fish (Thorarensen et al., 1996) and serves as a sensitive and logical technique for assessing intraspecific variation (Gamperl and Farrell, 2004). Furthermore, there is a growing interest in

intraspecific variation in fish cardiovascular function (Gamperl and Farrell, 2004), and more broadly in the physiological diversity of animals (Spicer and Gaston, 1999).

In this study, we used two genetically distinct stocks of largemouth bass *Micropterus salmoides* Lacépède that were collected from different geographic regions in the upper midwestern United States to: (1) assess whether cardiovascular function differs in bass from geographic locations with different climatic conditions and (2) quantify the impact that interstock hybridization has on this important physiological characteristic. For both comparisons, we monitored the cardiovascular response of individuals to exhaustive exercise at temperatures of 10 and 20°C.

Materials and methods
Study animals

This experiment took advantage of an extensive population genetic analysis of largemouth bass *Micropterus salmoides* Lacépède (and other species of fish) throughout the upper midwest USA (Fields et al., 1997). From that molecular genetic study we were able to propose geographic boundaries for 15 putative stocks of largemouth bass in Illinois, Minnesota and Wisconsin. That stock delineation process was based on the distribution of different alleles encoded at variable protein loci, based on starch gel electrophoresis, combined with the distribution of different mitochondrial DNA haplotypes, based on variable restriction endonuclease digestion sites (Fields et al., 1997).

During fall 1997 adult largemouth bass that were fixed for alternative homozygous genotypes at a single locus (MDH-B) were selected from each of two of the proposed stocks: (1) IL: from Central Illinois: Lake Shelbyville in the Kaskaskia River Drainage within the Mississippi River Basin; Genotype=MDH–B^2B^2; (2) WI: from Southeastern Wisconsin: Big Cedar Lake in the Lake Michigan Drainage within the Great Lakes Basin; Genotype=MDH–B^1B^1.

The WI fish came from a less temperate environment with mean annual temperatures (1971–2000) of 7.5°C and 63.4 days with maximum temperatures <0°C. The IL fish experienced mean annual temperatures (1971–2000) of 11.3°C and 33.9 days with maximum temperatures <0°C. In the spring of 1998, we stocked adult fish (10 males and 12 females, each with the appropriate homozygous genotype) from one or the other stock into each of eight 0.04 ha clay-lined, earthen ponds at the Illinois Natural History Survey Aquatic Research Field Laboratory in Champaign, IL, USA to produce four distinct, genetically tagged experimental stocks: each of the two pure parental (P_1) stocks (IL×IL and WI×WI), as well as both reciprocal F_1 interstock hybrids (WI×IL and IL×WI). In the fall of 1998, production ponds were drained. Fingerlings from each stock were given differential fin clips for external identification and then stocked into a series of ponds in a set of common garden experiments designed to assess their relative survival and growth, as well as their relative fitness (lifetime reproductive success) after they matured. Fish fed on natural invertebrate forage, as well as fathead minnows *Pimephales promelas* and juvenile bluegill *Lepomis macrochirus* produced in the ponds. The artificial ponds were substantially smaller than the lentic systems where fish were originally captured; however, the ponds contained substrates and vegetation typical of natural Midwestern systems and were subject to seasonal variation in temperature and photoperiod.

Cardiovascular assessments

Twice during fall 2000 (i.e. when ambient water temperatures corresponded to 10 and 20°C), some individuals from each stock were seined from the ponds and held in raceways for 1 week prior to experimentation. Although water temperatures were relatively stable for a period of approximately 2 weeks prior to experimentation, they did vary up to 3°C on a diel basis. During the experiments, water temperatures were controlled so that they varied by no more than 1°C. All experiments were conducted between 10:00 h and 16:00 h. Fish were exposed to the natural photoperiods of Champaign, Illinois during residency in pond and raceway environments, as well as during experimentation. As a result, fish were field-acclimated to conditions in central Illinois. Experiments were conducted between 18 September and 4 December, 2000.

Detailed descriptions of the surgical procedure are provided elsewhere (Schreer et al., 2001; Cooke et al., 2003b). Briefly, each fish was anaesthetized prior to surgery with 60 p.p.m. clove oil (emulsified with ethanol, 9:1 ethanol: clove oil) for approximately 8 min, at which point the fish had lost equilibrium and was non-responsive. Once anaesthetized, each fish was placed on its side on a wetted sponge. The anaesthetized state was maintained during surgery by irrigating the gills with water containing a maintenance concentration of anesthetic (30 p.p.m. clove oil). Connective tissue surrounding the aorta was carefully removed, and a flexible silicone cuff-type Doppler flow probe (sizes from 0.9 to 1.2 mm) subminiature 20 MHz piezoelectric transducer: Iowa Doppler Products, Iowa City, IA, USA) was placed around the aorta. The cuff was secured around the vessel using a single suture, and the lead wire leading to the probe was then sutured to the body of the fish at several locations.

Following surgery, individual fish were placed immediately into a 70 liter tank (50 cm×50 cm) and monitored closely until they had regained equilibrium. Fish were allowed to recover from surgery and to acclimate to the tank for at least 18 h (Cooke et al., 2003b). A darkened area covering approximately 30% of the tank provided cover and ensured that the fish were not disturbed by general laboratory activity. The experimental tanks were continuously supplied with pond water at either 10 or 20°C. To elicit exercise, fish were chased around the tank by hand (Cooke et al., 2003b) until they reached exhaustion; a standard technique in fish physiology (Kieffer, 2000). At this point the fish would no longer swim and would lose equilibrium. Cardiac variables were recorded continuously for at least 1 h prior to the exercise period (the resting period),

during the exercise period, and for at least 6 h post-exercise (the recovery period).

Following experimentation, fish were killed with an overdose of anesthetic (180 p.p.m. clove oil), and a *post-mortem* calibration was conducted to convert Doppler shift (in V) to actual blood flow (ml min^{-1}; Cooke et al., 2003b). The bulbous arteriosus was catheterized with tubing (PE 120) and a constant infusion pump (Harvard Apparatus, South Natick, MA, USA) was used to perfuse anticoagulated blood (2 g sodium oxalate + 0.4 g sodium chloride + 10 ml distilled H_2O l^{-1} pig's blood) through the aorta and partial gill arches to maintain output pressure. This procedure permitted the calibration of the probes over a range of flow rates encompassing the rates recorded during the trials. Reference flow rates were analyzed with linear least-squares regression (mean r^2=0.971). The ventricles were patted dry and weighed to the nearest 0.001 g and were mass corrected for the size of the fish to represent relative ventricular mass (RVM). To examine the temperature dependence of stock-specific resting cardiac variables, we calculated Q_{10} rates (Schmidt-Nielsen, 1997).

Data collection and analysis

A flowmeter (545C-4 Directional Pulsed Doppler Flowmeter: Bioengineering, The University of Iowa, Iowa City, IA, USA) and a digital strip-chart recorder (LabVIEW, Version 4.0.1, National Instruments Corporation, Austin, TX, USA) were used to record real time data on cardiac output. Heart rate (f_H) was determined by counting the number of heart beats (flow trace peaks) over a 60 s period and stroke volume (V_S) was calculated as cardiac output ($\dot{Q}$)/f_H.

To determine recovery times, traces for $\dot{Q}$, f_H and V_S, adjusted to resting (100%), were plotted for each fish and evaluated visually. A fish was considered to be recovered when values returned to resting and became stable (within 10 % of resting values; Schreer et al., 2001; Cooke et al., 2003b). Maximal disturbance was determined as the greatest change in a cardiac variable (either positive, >100% basal or negative, <100% basal) during the recovery period. Data were visually

assessed for normality using quantile plots and homogeneity of variance using residual plots (SYSTAT, V8.0, SAS Institute). The premise of all analyses was to test two null hypotheses: (1) that there were no differences in cardiovascular performance among molecularly defined P_1 stocks and (2) there were no differences in performance of F_1 hybrids compared to the P_1 stocks. All tests were conducted using two-way analysis of variance with stock being the main effect and water temperature the secondary effect (JMPIN, V4.01, SAS Institute). We used planned contrasts to examine where specific differences of interest occurred. All values reported are means ± S.E.M. Tests were considered significant at P=0.05.

Results

Although fish used in the experiments were all age-2 and generally of similar size, both length and mass were observed to vary with temperature and by stock (Tables 1, 2). RVM was generally similar among stocks at both temperatures (Tables 1, 2). However, an interaction revealed that at 20°C IL×WI fish had higher RVM than all other stocks.

Resting cardiac function

Cardiac output and heart rate values were significantly lower at 10 than 20°C for all four stocks, whereas stroke volume, differed significantly between 10 and 20°C only for the IL×WI stock (Table 2, Fig. 1). Resting cardiac output for the IL×IL stock was consistently lower than for the other three stocks. The only departure from this pattern was that at 20°C, resting cardiac output rates were similar between both P_1 stocks. Heart rate followed a similar pattern to cardiac output, with the IL×IL fish exhibiting the lowest resting heart rates; however, fewer stock-specific differences were noted (Fig. 1). Resting stroke volume values did not vary significantly among stocks (Table 2, Fig. 1). Q_{10} values for resting cardiac output and heart rate ranged from 1.33 to 1.65, and were highest for IL×IL fish (Fig. 1). Resting stroke volume Q_{10} values ranged from 0.89 to 1.01, with values being highest (i.e. close to 1) for both P_1 stocks (Fig. 1).

Table 1. *Meristics and characteristics for stocks of largemouth bass at two water temperatures*

Temperature (°C)	Stock	N	Total length (mm)	Mass (g)	RVM	Chase time (s)	Scope for $\dot{Q}$ (ml min^{-1} kg^{-1})	Scope for f_H (beats min^{-1})
10	IL×IL	12	177±3	63±4	0.06±0.005	142±8	8.4±1.6	25.9±1.7
10	IL×WI	9	186±2*	75±4*	0.08±0.004	145±11*	11.0±1.7	28.1±1.8
10	WI×IL	8	186±2*	72±2*	0.08±0.007	151±11	11.6±2.2*	22.0±3.4
10	WI×WI	11	182±1*	65±2	0.07±0.004	156±10*	11.7±1.5*	24.0±2.3
20	IL×IL	9	176±2[a]	66±6[a]	0.07±0.009[a]	145±16	7.8±1.7	25.3±3.3
20	IL×WI	8	201±2*,[b]	96±3*,[b]	0.08±0.004[b]	112±11*	11.3±4.7	28.2±1.7
20	WI×IL	9	179±2*,[a]	64±3*,[a]	0.07±0.003[a]	135±14	7.2±0.8*	27.6±1.9
20	WI×WI	8	175±3*,[a]	62±4[a]	0.06±0.006[a]	117±14*	7.2±1.0*	27.7±3.2

RVM, relative ventricular mass.

All values are means ± 1 S.E.M.

Asterisks indicate significantly different (P<0.05) values within a stock at the two different temperatures. Dissimilar letters indicate significantly different (P<0.05) values between stocks at each temperature. Additional details on statistical tests are given in Table 2.

Table 2. *Analysis of variance summaries provided for all tests performed*

Parameter	Source	F	d.f.	P
Total length	Stock	12.5	3	<0.001
	Temp	0.1	1	0.777
	Stock×Temp	4.7	3	0.006
Mass	Stock	12.6	3	<0.001
	Temp	0.9	1	0.337
	Stock×Temp	4.4	3	0.008
RVM	Stock	2.1	3	0.124
	Temp	0.5	1	0.486
	Stock×Temp	3.8	3	0.016
Resting				
$\dot{Q}$	Stock	7.3	3	<0.001
	Temp	113.4	1	<0.001
	Stock×Temp	0.3	3	0.838
f_H	Stock	5.6	3	<0.001
	Temp	146.5	1	<0.001
	Stock×Temp	0.2	3	0.890
V_S	Stock	1.5	3	0.229
	Temp	5.4	1	0.023
	Stock×Temp	1.4	3	0.264
Chase duration	Stock	0.7	3	0.574
	Temp	6.3	1	0.014
	Stock×Temp	1.4	3	0.265
Maximum				
$\dot{Q}$	Stock	5.5	3	0.002
	Temp	22.9	1	<0.001
	Stock×Temp	0.5	3	0.717
f_H	Stock	2.7	3	0.047
	Temp	67.0	1	<0.001
	Stock×Temp	0.8	3	0.498
V_S	Stock	0.2	3	0.928
	Temp	7.3	1	0.009
	Stock×Temp	1.2	3	0.306
Scope				
$\dot{Q}$	Stock	1.3	3	0.291
	Temp	4.3	1	0.042
	Stock×Temp	1.2	3	0.310
f_H	Stock	0.6	3	0.587
	Temp	1.6	1	0.207
	Stock×Temp	0.7	3	0.547
Recovery time				
$\dot{Q}$	Stock	14.6	3	<0.001
	Temp	0.2	1	0.674
	Stock×Temp	7.0	3	0.040
f_H	Stock	14.3	3	<0.001
	Temp	0.2	1	0.631
	Stock×Temp	7.1	3	0.031
V_S	Stock	8.2	3	<0.001
	Temp	0.1	1	0.809
	Stock×Temp	3.4	3	0.032

For all statistical analyses the main effect was stock and the secondary effect was water temperature (Temp).
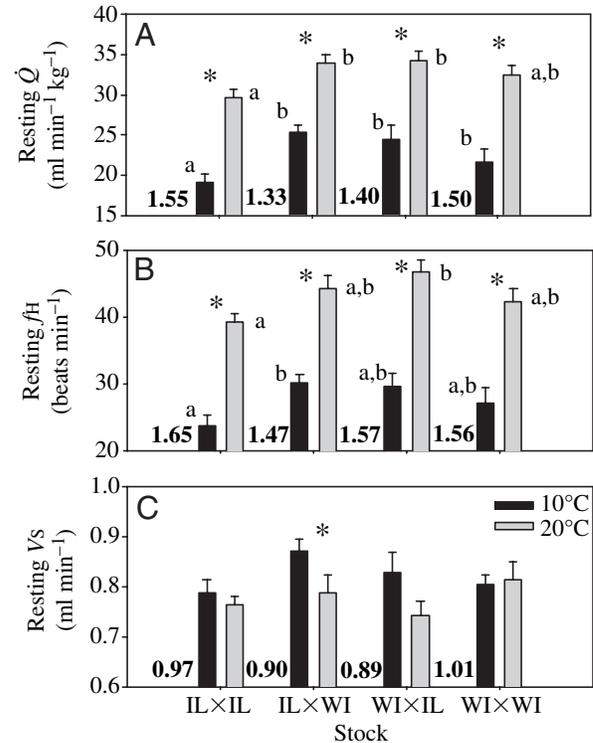
For other abbreviations, see text.

Fig. 1. Effects of water temperature and stock on the resting cardiac performance of largemouth bass. (A) Cardiac output; (B) heart rate; (C) stroke volume. Asterisks indicate significantly different ($P<0.05$) values within a stock at the two different temperatures. Dissimilar letters indicate significantly different ($P<0.05$) values between stocks at each temperature. Bold numbers represent stock-specific Q_{10} values for all three cardiac variables. Additional details on the statistical tests are given in Table 2. Values are means ± 1 s.e.m. Sample sizes are provided in Table 1.

Maximum cardiac function and scope

IL×WI and WI×WI fish both exhausted more rapidly at 20°C than at 10°C, whereas there was no temperature-specific difference among other stocks (Tables 1, 2). There was, however, a strong but nonsignificant trend towards the IL×IL fish taking longer to exhaust than the other stocks at 20°C (Table 2). In general, maximal cardiac output and heart rate were significantly higher at 20°C than at 10°C, whereas stroke volume was only significantly different among IL×IL and WI×WI fish across the two temperatures (Table 2, Fig. 2). Maximal cardiac output values were lower for IL×IL fish than for IL×WI fish. For heart rate, the only significant difference detected was between IL×IL and WI×IL at 20°C (Fig. 2). Maximal change in stroke volume did not differ among stocks at either temperature (Table 2, Fig. 2). Scope for cardiac output at 10°C for both WI×IL and WI×WI were higher than at 20°C (Tables 1, 2). No other differences were noted for cardiac scope (Tables 1, 2).

Cardiac recovery

Cardiac recovery times for IL×WI fish varied by temperature for heart rate and cardiac output, but in the opposite direction to what was observed for WI×WI fish
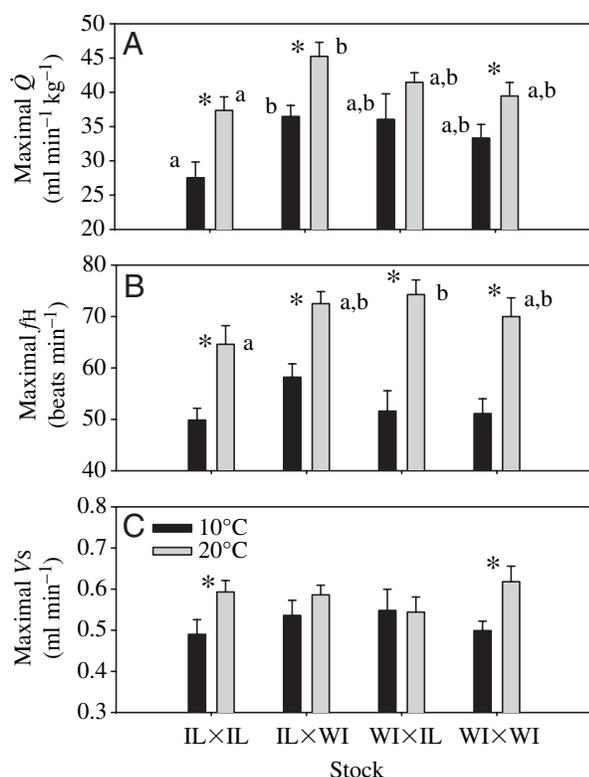
Fig. 2. Effects of water temperature and stock on the maximal cardiac performance of largemouth bass. (A) Cardiac output; (B) heart rate; (C) stroke volume. Asterisks indicate significantly different ($P<0.05$) values within a stock at the two different temperatures. Dissimilar letters indicate significantly different ($P<0.05$) values between stocks at each temperature. Additional details on the statistical tests are given in Table 2. Values are means ± 1 S.E.M. Sample sizes are provided in Table 1.
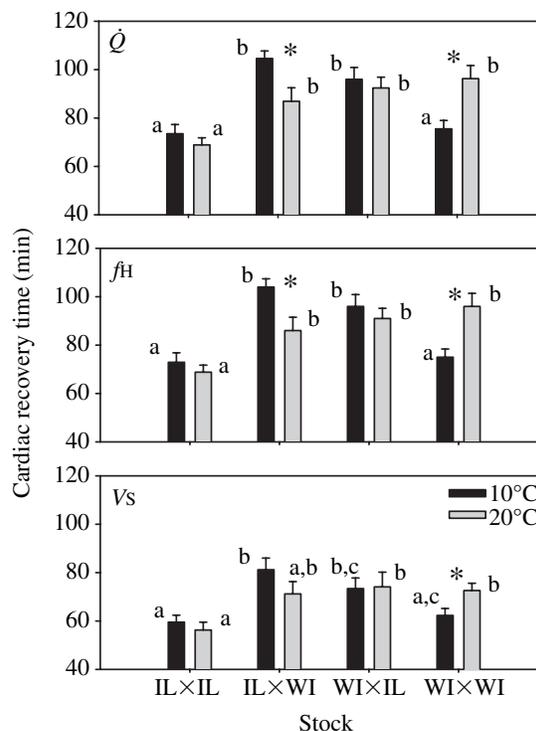

Fig. 3. Effects of water temperature and stock on the recovery of cardiac variables of largemouth bass following exhaustive exercise. Asterisks indicate significantly different ($P<0.05$) values within a stock at the two different temperatures. Dissimilar letters indicate significantly different ($P<0.05$) values between stocks at each temperature. Additional details on the statistical tests are given in Table 2. Values are means ± 1 S.E.M. Sample sizes are provided in Table 1.

(Fig. 3). At 10°C, cardiac output recovered similarly quickly for both P_1 stocks, but significantly more slowly, for both hybrids. At 20°C, cardiac output for the IL×IL stock recovered more rapidly than all other stocks. The same temperature-specific patterns of recovery observed for cardiac output were also observed for heart rate (Tables 1, 2, Fig. 3). Stroke volume at 10°C recovered most rapidly for both P_1 stocks, although the WI×WI recovery rate did not differ from the WI×IL stock (Table 2, Fig. 3). At 20°C, the IL×IL stock recovered more rapidly than the WI×IL and WI×WI stock.

Discussion

There is a paucity of research on population-specific differences in the cardiovascular physiology of fishes (see Gamperl and Farrell, 2004). However, in well-studied species such as rainbow trout *Oncorhynchus mykiss*, cardiovascular variables and responses vary widely among studies perhaps reflecting population level differences (e.g. Thorarensen et al., 1996; Brodeur et al., 2001) that clearly exist for swimming performance and metabolic rate (Taylor and McPhail, 1985; Lee et al., 2003). Formal analyses of population level

differences in cardiovascular performance are limited to two recent accounts for largemouth bass (Cooke et al., 2003b) and rock bass *Ambloplites rupestris* (Bunt et al., 2004), but neither study was designed primarily to assess intra-specific variation in a controlled manner. For the first time, we present data from a common garden experiment designed to explicitly evaluate the role of local adaptation on basic cardiovascular function using two genetically distinct stocks of largemouth bass and their reciprocal hybrids.

Our data illustrate that when translocated to novel environments, there are substantial differences in the cardiovascular performance of these largemouth bass relative to locally adapted individuals in the recipient population. Furthermore, when these two stocks were mixed, the interstock hybrids performed either similarly to the Wisconsin stock or more poorly than both P_1 stocks. These data suggest that interstock hybrids did not experience heterosis (i.e. hybrid vigor; Emlen, 1991; Thornhill, 1993) and instead exhibited performance indicative of a brake down of co-adapted gene complexes (Templeton, 1986; Hallerman, 2003) even at the F_1 generation. These data emphasize the fact that cardiovascular performance characteristics exist in locally adapted populations and that outbreeding occurs when fish from

different populations are mixed. In Illinois, locally adapted bass generally exhibited the lowest resting cardiac activity, the lowest maximal cardiac activity in response to exhaustive exercise, and recovered almost 25–35% faster than other stocks examined. Interestingly, at the lower temperature (10°C), the northern P_1 stock from Wisconsin recovered in the same time period as did the more southern P_1 stock (IL×IL). At the warmer temperature (20°C), however, the northern stock took longer to recover than the southern stock. Data presented here provide clearer evidence than has been previously reported for largemouth bass using swimming performance and respirometry (i.e. Cooke et al., 2001) that the translocation of fish and their subsequent hybridization with locally adapted stocks has physiological consequences that may have direct fitness implications, since cardiovascular performance is a determinant of aerobic swimming ability (Keen and Farrell, 1994; Kolok and Farrell, 1994).

When we evaluated resting cardiac activity, cardiac output and heart rate values for IL×IL fish were consistent with previous studies on largemouth bass (Cooke et al., 2003b) and were approximately 30% less than both of the interstock hybrids. The elevated resting cardiac output and heart rate of the WI×WI fish and in particular, the interstock hybrids, could result in greater myocardial energetic costs, but this would be minimal in the overall energetic budget as myocardial metabolism is generally less than 4.5% of total metabolic rate (Farrell and Jones, 1992). The maximal values for cardiac output and heart rate were also frequently higher for the interstock hybrids than the P_1 stocks, potentially yielding additional energy costs. However, without further analyses of oxygen uptake, transport and use, it is not possible to determine if the bioenergetic consequences of these cardiovascular performance differences are ecologically meaningful. Despite having higher resting rates and maximal rates, the scope for cardiac output and heart rate were similar among all four stocks. The interstock hybrids, therefore, maintained scope, but likely did so with a higher cost because they were operating at higher resting rates. Largemouth bass (Cooke et al., 2003a,b) and other centrarchid fishes (e.g. smallmouth bass *Micropterus dolomieu*, Schreer et al., 2001; black crappie *Pomoxis nigromaculatus*, Cooke et al., 2003a) are frequency modulators, responding to performance challenges typically by increasing heart rate and decreasing stroke volume. In this study, the pattern of largemouth bass being frequency modulators was maintained in the translocated stock (WI×WI) as well as the interstock hybrids and was similar to that of the IL×IL fish.

When exposed to a performance challenge such as exhaustive exercise, recovery times provide information on the duration that individual fish are forced to deal with the challenge, precluding them from allocating resources to other functions. In our study, recovery times were consistently about 25–35% longer for both interstock hybrids than for the Illinois stock. Previous work on largemouth bass revealed that cardiac recovery time was remarkably consistent across temperatures when exposed to 150 s of exercise and 30 s of air (~135 min

for $\dot{Q}$ and f_H and ~110 min for V_s; Cooke et al., 2003b). In the current study, the largemouth bass cardiac recovery times for translocated and interstock hybrids (~110 min for $\dot{Q}$ and f_H and ~70 min for V_s) were more variable than those of Cooke et al. (2003b). Although these were more rapid recovery times than the previous research (i.e. Cooke et al., 2003b), in the current study fish were not exposed to air, which is an additional stressor. Furthermore, when contrasted with the relatively rapid recovery of the locally adapted Illinois stock, the recovery times of the Wisconsin stock and interstock hybrids are indicative of major physiological differences in how they respond to exercise. During this period of lengthened recovery, interstock hybrids could exhibit reduced digestion (Randall and Daxboeck, 1982) and food intake (Smart, 1981; Barton and Schreck, 1987), negatively affecting acquisition of new energy resources. Furthermore, because cardiovascular performance is strongly correlated with metabolic rate, when the fish are at an elevated stage on their scope, fish may have less scope for activity available to escape predators or deal with other stressors (Priede, 1977, 1985). Although cardiac performance is a factor in determining oxygen consumption and metabolic rate, there are also other factors that we did not measure here that could exacerbate or mitigate the consequences of intraspecific variation in cardiovascular performance.

Evidence suggesting that inbreeding and outbreeding depression may be manifested as alterations in cardiovascular performance in higher organisms comes from selective breeding experiments with mice *Mus domesticus* (e.g. Mattson, 2001) or observations of high rates of cardiac abnormalities in some inbred organisms such as Florida panther *Felis concolor coryi* (Roelke et al., 1993). To our knowledge, however, no information on the cardiovascular consequences of inbreeding or outbreeding currently exist for teleost fish (Waldman and McKinnan, 1993). The only research that may be relevant is the assessment of cardiovascular performance in transgenic and domesticated fish. Triploid brown trout *Salmo trutta* appeared to have reduced factorial metabolic scope relative to diploid fish (Altimiras et al., 2002), while domesticated rainbow trout reared in a hatchery did not exhibit appreciable differences in cardiac morphometrics or performance relative to wild fish (Gamperl and Farrell, 2004). While these studies did evaluate the cardiovascular consequences of selective breeding, they did not, however, evaluate the effects of interstock hybridization. The approach used in our study differs in that it provides a possible proximal mechanism for reductions in fitness that can result from such hybridization. Because there were minimal differences in cardiac morphometrics among the treatment groups we examined, the variation in cardiac performance may represent disruption in humoral/neurological regulation and/or efficiency of enzymatic and other cellular processes, consistent with the breakdown of co-adapted gene complexes (Templeton, 1986).

In addition to providing insight into the fundamental physiological diversity and intraspecific cardiovascular performance of fish, this study also has implications for conservation scientists. Our findings suggest that molecular

genetic techniques are able to delineate populations that have sufficiently different physiologies to warrant separate management and conservation activities (e.g. Nielsen, 1995). The current study supported previous findings (i.e. fitness, Philipp and Claussen, 1995; swimming performance, Cooke et al., 2001) in that the northern stock of largemouth bass (WI×WI) did not perform as well in Illinois as the Illinois stock. For largemouth bass, therefore, local adaptations in cardiovascular performance exist among populations that occur within a relatively small geographic region so translocations should me minimized. Finally, our results illustrate the negative consequences of mixing fish from different locally adapted populations. Future efforts focusing on fish from the F_2 generation will provide additional insight into intra-specific cardiovascular performance and the potential long-term consequences of fish translocation practices.

References

Altimiras, J., Axelsson, M., Claireaux, G., Lefrancois, C., Mercier, C. and Farrell, A. P. (2002). Cardiorespiratory status of triploid brown trout (*Salmo trutta*) during swimming at two acclimation temperatures. *J. Fish Biol.* **60**, 102-116.

Barton, B. A. and Schreck, C. B. (1987). Metabolic cost of acute physical stress in juvenile steelhead. *Trans. Amer. Fish. Soc.* **116**, 257-263.

Brodeur, J. C., Dixon, D. G. and McKinley, R. S. (2001). Assessment of cardiac output as a predictor of metabolic rate in rainbow trout. *J. Fish Biol.* **58**, 439-452.

Bunt, C. M., Cooke, S. J., Schreer, J. F. and Philipp, D. P. (2004). Effects of incremental increases in silt load on the cardiovascular performance of riverine and lacustrine rock bass, *Ambloplites rupestris*. *Environ. Poll.* **128**, 437-444.

Campton, D. E. (1987). Natural hybridization and introgression in fishes: methods of detection and interpretation. In *Population Genetics and Fishery Management* (ed. N. Ryman and F. Utter), pp. 161-192. Seattle, WA: University of Washington Press.

Cooke, S. J., Kassler, T. W. and Philipp, D. P. (2001). Physiological performance of largemouth bass related to local adaptation and interstock hybridization: implications for conservation and management. *J. Fish Biol.* **58A**, 248-268.

Cooke, S. J., Grant, E. C., Schreer, J. F., Philipp, D. P. and Devries, A. L. (2003a). Low temperature cardiac response to exhaustive exercise in fish with different levels of winter quiescence. *Comp. Biochem. Physiol.* **134A**, 157-165.

Cooke, S. J., Ostrand, K. G., Bunt, C. M., Schreer, J. F., Wahl, D. H. and Philipp, D. P. (2003b). Cardiovascular responses of largemouth bass to exhaustive exercise and brief air exposure over a range of water temperatures. *Trans. Amer. Fish. Soc.* **132**, 1154-1165.

Emlen, J. M. (1991). Heterosis and outbreeding depression: a multilocus model and application to salmon production. *Fish. Res.* **12**, 187-212.

Farrell, A. P. and Jones, D. R. (1992). The heart. In *Fish Physiology*, vol. 12A (ed. W. S. Hoar, D. J. Randall and A. P. Farrell), pp. 1-88. San Diego: Academic Press.

Fields, R., Desjardins, M., Hudson, J., Kassler, T., Ludden, J., Tranquilli, J., Toline, C. and Philipp, D. P. (1997). *Genetic Analyses of Fish Species in the Upper Midwest*. Final Report to the Minnesota and Wisconsin Departments of Natural Resources. Three volumes, 576 pp.

Gamperl, A. K. and Farrell, A. P. (2004). Cardiac plasticity in fishes: environmental influences and intraspecific differences. *J. Exp. Biol.* **207**, 2539-2550.

Gharrett, A. J., Smoker, W. W., Reisenbichler, R. R. and Taylor, S. G. (1999). Outbreeding depression in hybrids between odd- and even-broodyear pink salmon. *Aquaculture* **173**, 117-129.

Hallerman, E. M. (2003). Coadaptation and outbreeding depression. In *Population Genetics: Principles and Practices for Fisheries Scientists* (ed. E. M. Hallerman), pp. 239-259. Bethesda, MD: American Fisheries Society.

Keen, J. E. and Farrell, A. P. (1994). Maximum prolonged swimming speed and maximum cardiac performance of rainbow trout acclimated to two different water temperatures. *Comp. Biochem. Physiol.* **108A**, 287-295.

Kieffer, J. D. (2000). Limits to exhaustive exercise in fish. *Comp. Biochem. Physiol.* **126A**, 161-179.

Kolok, A. S. and Farrell, A. P. (1994). Individual variation in the swimming performance and cardiac performance of northern squawfish, *Ptychocheilus oregonensis*. *Physiol. Zool.* **67**, 706-722.

Lee, C. G., Farrell, A. P., Lotto, A. G., MacNutt, M. J., Hinch, S. G. and Healey, M. C. (2003). Effects of temperature on swimming performance and oxygen consumption in adult sockeye (*Oncorhynchus nerka*) and coho (*O. kisutch*) salmon stocks. *J. Exp. Biol.* **206**, 3239-3251.

Mattson, D. L. (2001). Comparison of arterial blood pressure in different strains of mice *Amer. J. Hypertens,* **14**, 405-408.

Nielsen, J. L. (ed.) (1995). Evolution and the aquatic ecosystem: defining unique units in population conservation. *Amer. Fish. Soc. Symp.* **17**.

Philipp, D. P. and Claussen, J. E. (1995). Fitness and performance differences between two stocks of largemouth bass from different drainages within Illinois. *Amer. Fish. Soc. Symp.* **15**, 236-243.

Philipp, D. P. and Whitt, G. S. (1991). Survival and growth of northern, Florida, and reciprocal F1 hybrid largemouth bass in central Illinois. *Trans. Amer. Fish. Soc.* **120**, 156-178.

Philipp, D. P., Childers, W. F. and Whitt, G. S. (1983). A biochemical genetic evaluation of the northern and Florida subspecies of largemouth bass. *Trans. Amer. Fish. Soc.* **112**, 1-20.

Philipp, D. P., Parker, H. R., Pasdar, M., Burkett, D. P., Claussen, J. E. and Whitt, G. S. (1995). Diversity below the species level: biochemical evidence for adaptation among largemouth bass stocks. In *Protection of Aquatic Biodiversity* (ed. D. P. Philipp), pp. 135-147. Proceedings of the World Fisheries Congress, Theme 3. New Delhi: Oxford and IBH Publishing Co. Pvt. Ltd.

Philipp, D. P., Claussen, J. E., Kassler, T. W. and Epifanio, J. M. (2002). Mixing stocks of largemouth bass reduces fitness through outbreeding depression. *Amer. Fish. Soc. Symp.* **31**, 349-363.

Priede, I. G. (1977). Natural selection for energetic efficiency and the relationship between activity level and mortality. *Nature* **267**, 610-611.

Priede, I. G. (1985). Metabolic scope in fishes. In *Fish Energetics: New Perspectives* (ed. P. Tytler and P. Calow), pp. 33-64. Baltimore: The Johns Hopkins University Press.

Randall, D. J. and Daxboeck, C. (1982). Cardiovascular changes in the rainbow trout (*Salmo gairdneri* Richardson) during exercise. *Can. J. Zool.* **60**, 1135-1140.

Roelke, M. E., Martenson, J. S. and O'Brien, S. J. (1993). The consequences of demographic reduction and genetic depletion in the endangered Florida panther. *Curr. Biol.* **3**, 340-350.

Ryman, N. and Utter, F. (eds) (1987). *Population Genetics and Fishery Management*. Seattle: University of Washington Press.

Schmidt-Nielsen, K. (1997). *Animal Physiology: Adaptation and Environment*. Cambridge, UK: Cambridge University Press.

Schreer, J. F., Cooke, S. J. and McKinley, R. S. (2001). Cardiac response to variable forced exercise at different temperatures – an angling simulation for smallmouth bass. *Trans. Amer. Fish. Soc.* **130**, 783-795.

Smart, G. R. (1981). Aspects of water quality producing stress in intensive fish culture. In *Stress and Fish* (ed. A. D. Pickering), pp. 277-293. NY: Academic Press.

Spicer, J. I. and Gaston, K. J. (1999). *Physiological Diversity and its Ecological Implications*. Oxford, UK: Blackwell Science.

Taylor, E. B. and McPhail, J. D. (1985). Variation in burst and prolonged swimming performance among British Columbia populations of coho salmon (*Oncorhynchus kisutch*). *Can. J. Fish. Aquat. Sci.* **42**, 2029-2033.

Templeton, A. R. (1986). Coadaptation and outbreeding depression. In

Conservation Biology: The Science of Scarcity and Diversity (ed. M. Soulé), pp. 105-116. Sunderland, MA: Sinauer Associates.

Thorarensen, H., Gallaugher, P. E. and Farrell, A. P. (1996). The limitations of heart rate as a predictor of metabolic rate in fish. *J. Fish. Biol.* **49**, 226-236.

Thornhill, N. W. (ed.) (1993). *The Natural History of Inbreeding and Outbreeding: Theoretical and Empirical Perspectives*. Chicago: University of Chicago Press.

Waldman, B. and McKinnan, J. S. (1993). Inbreeding and outbreeding in fishes, amphibians, and reptiles. In *The Natural History of Inbreeding and Outbreeding: Theoretical and Empirical Perspectives* (ed. N. W. Thornhill), pp. 250-282. Chicago: University of Chicago Press.

Wright, S. (1978). *Variability Within and Among Populations – Evolution and the Genetics of Natural Populations*. Volume 4. Chicago: University of Chicago Press.