

Morphological and swimming stamina differences between Yellowstone cutthroat trout (*Oncorhynchus clarkii bouvieri*), rainbow trout (*Oncorhynchus mykiss*), and their hybrids

Steven M. Seiler and Ernest R. Keeley

Abstract: We hypothesized that body shape differences between Yellowstone cutthroat trout (*Oncorhynchus clarkii bouvieri*), rainbow trout (*Oncorhynchus mykiss*), and their hybrids may influence swimming ability and thus play an important role in the invasion of nonnative rainbow trout and hybrid trout into native cutthroat trout populations. We reared Yellowstone cutthroat trout, rainbow trout, and reciprocal hybrid crosses in a common environment and conducted sustained swimming trials in order to test for genetically based morphological and swimming stamina differences. Linear and geometric morphometric analyses identified differences in body shape, with cutthroat trout having slender bodies and small caudal peduncles and rainbow trout having deep bodies and long caudal peduncles. Hybrid crosses were morphologically intermediate to the parental genotypes, with a considerable maternal effect. Consistent with morphological differences, cutthroat trout had the lowest sustained swimming velocity and rainbow trout had the highest sustained swimming velocity. Sustained swimming ability of hybrid genotypes was not different from that of rainbow trout. Our results suggest that introduced rainbow trout and cutthroat-rainbow trout hybrids potentially out-compete native Yellowstone cutthroat trout through higher sustained swimming ability.

Résumé : Nous avons émis l'hypothèse selon laquelle les différences de forme du corps entre la truite fardée de Yellowstone (*Oncorhynchus clarkii bouvieri*), la truite arc-en-ciel (*Oncorhynchus mykiss*) et leurs hybrides peuvent affecter la capacité de nage et ainsi jouer un rôle dans l'invasion des populations indigènes de truites fardées par les truites arc-en-ciel non indigènes et les truites hybrides. Nous avons élevé des truites fardées de Yellowstone, des truites arc-en-ciel et de truites issues de croisements hybrides réciproques dans un environnement commun et nous avons mené des tests de nage soutenue afin de vérifier l'existence des différences d'origine génétique dans la morphologie et dans la vigueur de nage. Des analyses morphométriques linéaires et géométriques mettent en évidence des différences dans la forme du corps: les truites fardées possèdent un corps élancé et un pédoncule caudal réduit, alors que les truites arc-en-ciel ont un corps profond et un long pédoncule caudal. Les truites des croisements hybrides ont une morphologie intermédiaire par rapport aux génotypes des parents avec un effet maternel marqué. En accord avec les différences morphologiques, les truites fardées ont la vitesse de nage soutenue la plus faible et les truites arc-en-ciel la vitesse de nage soutenue la plus élevée. La capacité de nage soutenue des génotypes hybrides ne diffère pas de celle des truites arc-en-ciel. Nos résultats indiquent que les truites arc-en-ciel introduites et les hybrides des truites fardées et arc-en-ciel peuvent potentiellement gagner la compétition avec les truites fardées indigènes à cause de leur capacité supérieure de nage soutenue.

[Traduit par la Rédaction]

Introduction

Nonnative species are commonly thought to cause declines and extinction of native species through predation, competition, or habitat alteration; however, a more severe impact may occur by hybridization with native species (Rhymer and Simberloff 1996; Sakai et al. 2001). Intraspecific hybridization, occurring between distinct populations, is thought to have played an important role in evolution of many plant taxa (Stebbins 1959) and some ver-

tebrates (Dowling and DeMarais 1993). In contrast, hybridization between species is often highly detrimental to native populations when closely related nonnative species are introduced into native species ranges and form interspecific hybrids (Rhymer and Simberloff 1996). When hybrid offspring are sterile, genetic resources are wasted and small native populations may lose significant portions of important recruitment classes. When hybrids are fertile, reproduction between first generation hybrids and parental or advanced crosses of hybrids is termed introgression (Anderson 1949).

Received 13 February 2006. Accepted 7 December 2006. Published on the NRC Research Press Web site at <http://cjfas.nrc.ca> on 30 January 2007.
J19172

S.M. Seiler¹ and E.R. Keeley. Department of Biological Sciences, Box 8007, Idaho State University, Pocatello, ID 83209, USA.

¹Corresponding author (e-mail: seilstev@isu.edu).

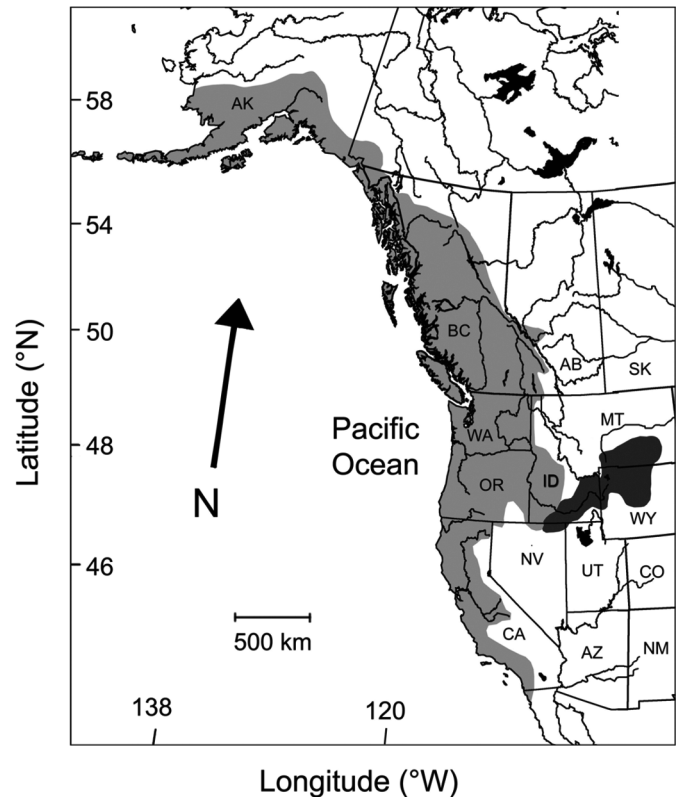
Introgression of alleles from nonindigenous species has caused rapid genomic extinction in many populations of birds, mammals, and fish (Rhymer and Simberloff 1996; Sakai et al. 2001).

In salmonid fishes, competition and hybridization with introduced species has been implicated in the demise of many native populations (Behnke 1992). In western North America, many cutthroat trout (*Oncorhynchus clarkii* spp.) populations are under pressure from introduced rainbow trout (*Oncorhynchus mykiss*; Hitt et al. 2003; Weigel et al. 2003; Rubidge and Taylor 2004). Because of ecological similarities and a recent common ancestry, rainbow and cutthroat trout are thought to compete for food and space when in sympatry (Griffith 1988) and they produce fertile offspring when they interbreed (Krueger and May 1991). Although rainbow trout are also native to western North America, spatial and temporal isolating mechanisms prevented most hybridization with cutthroat trout where their native ranges overlap (Trotter 1989). In areas where geological barriers prevented rainbow trout from moving into interior watersheds, cutthroat trout populations existed in isolation for at least 10 000 years (McPhail and Lindsey 1986; Behnke 1992). However, because of widespread propagation and introduction of rainbow trout across all of North America, rainbow trout now have reproducing populations in areas where cutthroat trout were the only native trout. Competition and hybridization with rainbow trout are recognized as key reasons for the decline of inland cutthroat trout populations (Young 1995).

Historically, Yellowstone cutthroat trout (*Oncorhynchus clarkii bouvieri*) were the only trout species within the upper Snake River drainage in Wyoming, Idaho, Utah, and Nevada and the Yellowstone River drainage in Wyoming and Montana (Fig. 1; Varley and Gresswell 1988; Behnke 1992). Rainbow trout were stocked to supplement a sport fishery in many of these drainages, and over the past 20 years, increasing numbers of rainbow trout and hybrid trout have raised concerns regarding the persistence of native Yellowstone cutthroat trout populations (Schrader and Gamblin 1996; Jaeger et al. 2000). Unlike most inland cutthroat trout, native rainbow trout populations are often sympatric with other indigenous salmonids that compete for food and space with them (Quinn 2005). Such co-existing populations of stream-dwelling salmonids are thought to co-occur because of habitat partitioning between species (Hartman 1965). Because native cutthroat trout often decline or disappear when rainbow trout are introduced (Jaeger et al. 2000; Hitt et al. 2003; Weigel et al. 2003), it is likely that cutthroat trout lack the competitive abilities that evolved in salmonids experiencing similar co-occurring competitors.

For invasive species to become established and proliferate when introduced into new habitats, they must possess characteristics that allow them to out-compete ecologically similar native species. Swimming ability is an important correlate of fitness in fishes (Taylor and Foote 1991; Hawkins and Quinn 1996) and may play an important role in the spread of rainbow trout genes into Yellowstone cutthroat trout populations. Because body shape is strongly related to swimming ability in fishes (Lindsey 1978; Webb 1984), differences in body shape and therefore swimming ability may influence the decline in native fish species by introduced species and

Fig. 1. Historic distribution of Yellowstone cutthroat trout (*Oncorhynchus clarkii bouvieri*, dark shading) and rainbow trout (*Oncorhynchus mykiss*, light shading) in western North America.



their hybrids. In stream ecosystems, salmonids feed primarily on drifting aquatic invertebrates by swimming to hold position in the current. The most profitable foraging areas are found where the supply of invertebrates is high relative to the cost of maintaining position in the current (Hughes and Dill 1990). Because the abundance of drifting invertebrates is thought to be positively related to the velocity of water, individuals with the ability to sustain swimming at higher velocities may benefit by foraging and gaining energy more efficiently than those with weaker abilities.

To test for inherent differences in swimming stamina and morphology between native and invasive salmonid fishes, we raised Yellowstone cutthroat trout, rainbow trout, and their reciprocal hybrids in a common garden experiment. First, we conducted swimming stamina trials to determine whether differences in sustained swimming ability exist between parental and hybrid trout. Secondly, we examined whether differences in body shape distinguishing parental and hybrid genotypes were related to any differences in sustained swimming ability. Given the decline of previously allopatric cutthroat trout after the introduction of nonnative rainbow trout, we predicted that Yellowstone cutthroat trout would have lower swimming stamina than rainbow trout and their hybrids. Although few studies have directly compared individual morphology and swimming performance in fishes (but see Boily and Magnan 2002; Ojanguren and Brana 2003), salmonid fish from higher velocity streams tend to have larger paired fins and deeper bodies than those from slower streams or standing water (Swain and Holtby 1989; Keeley

et al. 2005). Hence, if cutthroat trout are poorer competitors because of morphologically linked differences in swimming ability, we expect them to have shorter paired fins and shallower bodies than rainbow or hybrid trout.

Materials and methods

Collection and rearing of experimental animals

On 23 April 2004, we collected Yellowstone cutthroat trout and rainbow trout gametes to create experimental populations. Cutthroat trout eggs and sperm were collected from Henry's Lake, located near Island Park, Idaho. Each spring, the Henry's Lake hatchery collects gametes from wild cutthroat trout as they prepare to spawn. Rainbow trout eggs and sperm were collected from the Hayspur Hatchery, Hayspur, Idaho, from a captive population of rainbow trout. All gametes were placed in coolers with ice and transported to Idaho State University.

To create cohorts of juvenile fish to be used in our experiment, we divided the clutches from each female in half. To minimize the possibility of using an infertile male, we then combined sperm from two conspecific males and used half to fertilize a conspecific egg lot and half to fertilize a heterospecific egg lot. This process created experimental cohorts of cutthroat trout, maternal cutthroat hybrid (cutthroat trout eggs crossed with rainbow trout sperm), maternal rainbow hybrid (rainbow trout eggs crossed with cutthroat trout sperm), and rainbow trout. We created four replicate cohorts of each genotype (16 total populations). Within each replicate, hybrid offspring were maternal or paternal half-siblings to the pure parental crosses.

Each cohort was incubated in an upwelling incubator supplied with water from a common reservoir. A slow input of dechlorinated city water ($\sim 2.5 \text{ L}\cdot\text{min}^{-1}$) maintained water temperature at $12.5 \text{ }^\circ\text{C}$ ($\pm 0.5 \text{ }^\circ\text{C}$) and ensured high water quality. Overflow from incubators was collected in a common drain, filtered, and pumped back to the reservoir. The temperature of the freshwater supply slowly increased during the third and fourth week of incubation and development, reaching a maximum temperature of $13.5 \text{ }^\circ\text{C}$. On 24 May 2004, a water chilling unit was added to the common reservoir, which maintained water temperature at $12.5 \text{ }^\circ\text{C}$ ($\pm 0.5 \text{ }^\circ\text{C}$) for the remainder of experiment. We removed dead eggs daily and did not detect a qualitative difference in mortality between genotypes. After hatching and development to the exogenously feeding stage, up to 100 trout from each cross were transferred to rearing channels.

The rearing system consisted of four channels constructed out of plywood lined with fiberglass and coated with non-toxic aquaculture paint. Each channel was divided into four compartments by screens to create a total of 16 rearing compartments measuring 109 cm long \times 36 cm wide. Water was supplied from the common reservoir to each channel ($\sim 3 \text{ L}\cdot\text{min}^{-1}$). Channel depth was maintained at 24 cm by a standpipe drain. Outflow from all channels was pooled, filtered, and pumped back to the header reservoir. Although we did not expect position within channels to influence development in a population, we accounted for any potential position effect by assigning each replicate cohort of a genotype to one of the 16 compartments such that each genotype appeared only once in each channel and in every possible posi-

tion across the array of channels. Over the first month of feeding, trout were fed hatchery feed three times a day until satiation and twice daily thereafter (Biodry 1000, Bio-Oregon Inc., Warrenton, Oregon). Uneaten food and waste were removed several times a week. Water quality remained high throughout rearing; periodic tests never detected build-up of nitrogenous wastes.

Swimming stamina

As an estimate of swimming performance, we measured sustained swimming ability using a Blazka-type swimming chamber (Brett 1964). The swim chamber consisted of a clear acrylic tube (30 cm long \times 9 cm diameter) contained within a larger tube (47 cm long \times 24 cm diameter). An impeller powered by a variable-speed $\frac{1}{2}$ HP DC motor cycled water through the central chamber. Each test fish was contained within the swim chamber by $1 \text{ cm} \times 1 \text{ cm}$ screening on its ends. To ensure laminar flow through the swim chamber, we attached an 8 cm long plastic grid with $2 \text{ cm} \times 2 \text{ cm}$ openings to the upstream end of the central chamber. During all trials, water pumped into the chamber from the rearing system reservoir maintained temperature at $12.5 \text{ }^\circ\text{C}$ ($\pm 0.5 \text{ }^\circ\text{C}$) and near saturation with oxygen.

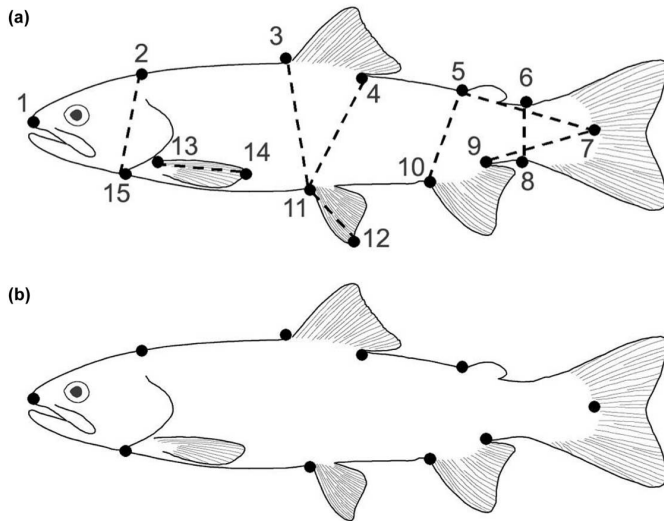
On the day before trials, one trout from each of the four genotypes was haphazardly selected from a randomly chosen stream channel. Each trout was measured (fork length, $\pm 1 \text{ mm}$), placed into a holding tube, and returned to its rearing channel. Holding tubes were sections of polyvinyl chloride (PVC) pipe matching the dimensions of the swim tunnel. Isolation allowed the trout to acclimate to a confinement similar to the swim chamber and assured that no test fish ate for 24 h before the swim trial. On the following day, trout were individually tested for sustained swimming velocity.

Sustained swimming velocity trials were conducted between 24 November 2004 and 23 December 2004. We did not expect differences in body size to influence absolute sustained swimming velocity across the size range of fish tested in this study (range for each genotype: $78\text{--}98 \text{ mm}$ fork length). However, to account for size differences, we corrected the absolute water velocity ($\text{cm}\cdot\text{s}^{-1}$) that each fish encountered to body lengths per second ($\text{bl}\cdot\text{s}^{-1}$). Trials were initiated by placing individual trout in the swim tunnel for 5 min without flow, followed by 30 min at $4 \text{ bl}\cdot\text{s}^{-1}$. After this acclimation period, velocity was increased by $0.5 \text{ bl}\cdot\text{s}^{-1}$ every 10 min until the test fish could not maintain its position in the chamber and became pinned against the back screen. A mild electrical stimulus (1 volt, $\frac{1}{2}$ amp) was used to encourage fish resting on the back screen to continue swimming. When a test fish became pinned against the back screen and could not be stimulated to swim any longer, the time elapsed from the last velocity increase was recorded, flow was turned off, and the fish was removed from the chamber. Each trout was euthanized, placed on its right side with fins extended, photographed for morphometric comparisons, and preserved in 10% formalin.

We compared swimming velocity among genotypes using analysis of variance (ANOVA). The response variable, critical swimming velocity (U_{crit} , $\text{bl}\cdot\text{s}^{-1}$) was calculated by the following formula:

$$U_{\text{crit}} = V + V_i(T_f/T_i)$$

Fig. 2. (a) Locations for 15 landmarks identified on each trout photograph. Broken lines indicate the head depth (2–15), body depth 1 (3–11), body depth 2 (4–11), caudal depth 1 (5–10), caudal depth 2 (6–8), caudal length 1 (5–7), caudal length 2 (7–9), pelvic fin (11–12), and pectoral fin (13–14). These nine measurements are used in the linear morphometric analysis. (b) Landmarks used in the geometric analysis of body shape.

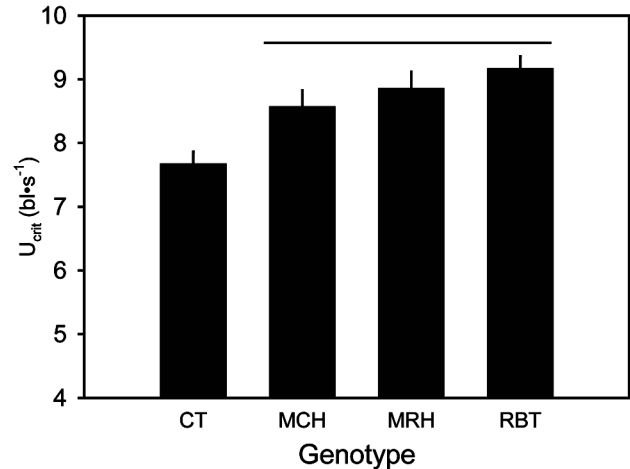


where V is the highest velocity at which a fish swam for the entire time increment ($\text{bl}\cdot\text{s}^{-1}$), V_i is the velocity increment ($0.5 \text{ bl}\cdot\text{s}^{-1}$), T_f is the time elapsed between the final velocity increase and fatigue (min), and T_i is the time between velocity increases (10 min). Because the cross-sectional area of the largest fish was $<5\%$ of the cross-sectional area of the tunnel, we did not consider solid blocking effects that sometimes occur when fish of a given size swim in relatively small chambers (Bell and Terhune 1970). When we found significant differences in mean U_{crit} values, we identified which genotypes differed using Tukey's test ($\alpha = 0.05$).

Morphological comparisons

To compare body morphology between genotypes, 15 landmarks (Fig. 2a) were digitized onto the photograph of each trout using tpsDig (Rohlf 2005a). A scale bar included in each photograph allowed landmark locations (x and y coordinates) to be translated into linear measurements using the Pythagorean theorem. Nine measurements were chosen to compare linear morphological characters of body depth, caudal depth, caudal length, and paired fin length (Fig. 2a). To correct for size-related differences across individuals, we regressed each morphological characteristic against fork length. All measurements were \log_{10} -transformed to ensure that we met the assumption of homogeneity of variance, and we visually examined the residuals from all analyses by plotting them against body size to ensure that we met this assumption. Because of the large number of pairwise comparisons of slopes, we could not assume that the slope describing the relationship between each morphological trait and body size was equal across all genotypes. We used separate within-group slopes (groups = genotypes) to adjust each

Fig. 3. Mean sustained swimming velocity (body lengths per second ($\text{bl}\cdot\text{s}^{-1}$); +1 standard error) of trout genotypes. Means sharing the line do not differ significantly (Tukey's test, $\alpha = 0.05$). Acronyms: CT, cutthroat trout (*Oncorhynchus clarkii bouvieri*); MCH, maternal cutthroat hybrid; MRH, maternal rainbow hybrid; RBT, rainbow trout (*Oncorhynchus mykiss*).



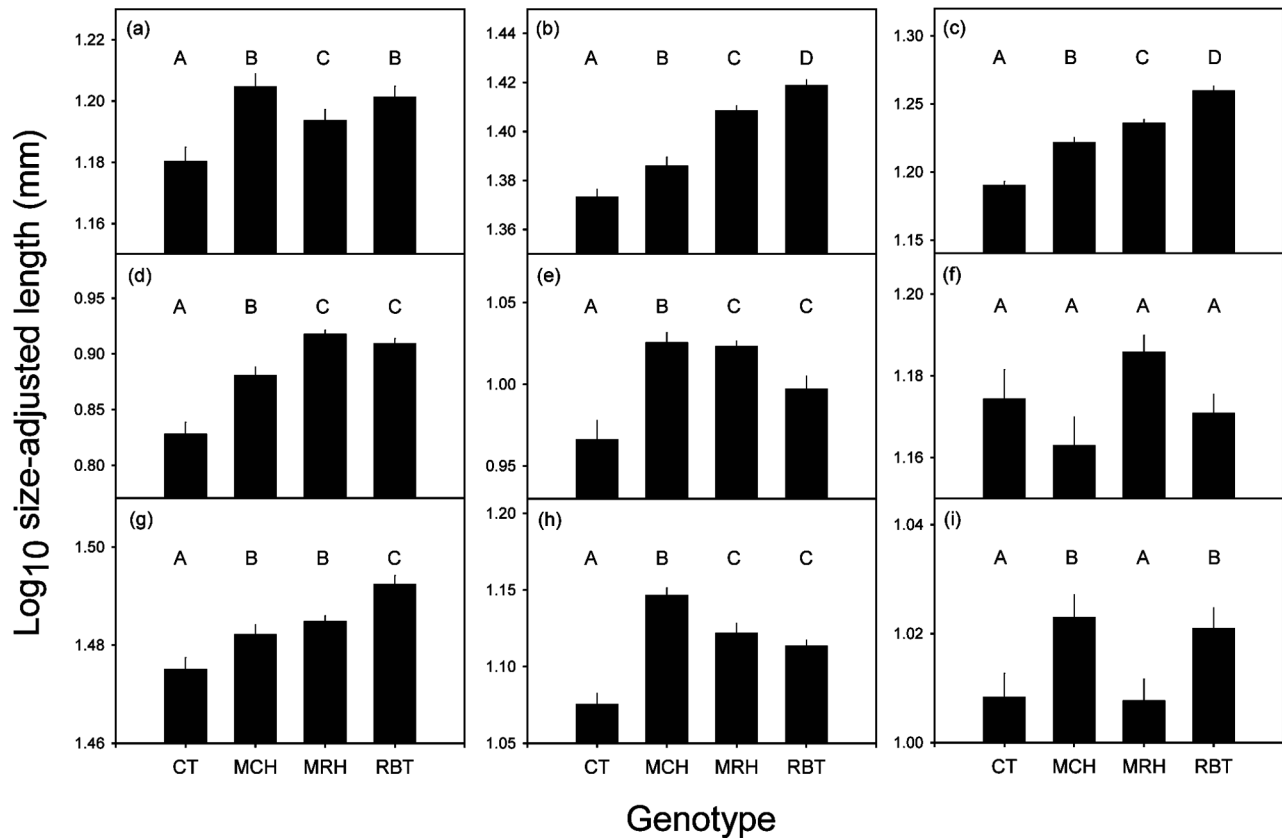
trait to a common body size based on the method of Thorpe (1976).

In addition to the linear morphometric technique, we also compared body shape using geometric morphometrics (cf. Zelditch et al. 2004). For this analysis, we used a subset of 10 landmarks, which were spread across the body of each trout at easily identifiable, stationary points (Fig. 2b). Geometric morphometric techniques quantify the spatial covariation of each landmark relative to all other landmarks and allow visualization of shape through thin plate spline diagrams. The landmark locations from each trout were used to create geometric shape variables (partial warp scores and uniform shape components) using tpsRelw (Rohlf 2005b).

We used multivariate analysis of variance (MANOVA) of the nine size-adjusted linear characters to assess body shape differences among genotypes, followed by ANOVAs to compare how individual traits differed across genotypes. When differences were detected on individual traits, we used post-hoc Tukey's tests to identify which genotypes differed. We conducted a separate MANOVA to assess whether the genotypes differed based on the geometric shape variables.

To summarize shape differences across the linear measurements and across the geometric shape variables, we used principal component analyses (PCA). By using size-adjusted linear characters in the PCA, principal axes represent variation in the relative size of linear characters (Keeley et al. 2005). PCA on geometric shape variables, commonly called a relative warps analysis (Rohlf 1993), allowed us to summarize differences in body shape and graphically examine implied morphological changes along principal axes using tpsRelw (Rohlf 2005b). We used ANOVA to compare differences in mean factor scores among genotypes for the linear and geometric analyses, and we used Tukey's tests to examine differences in mean factor scores for pairwise comparisons. For both PCAs, we examined principal axes with eigenvalues greater than 1 (Quinn and Keough 2002); however, we only considered axes which differentiated at least

Fig. 4. Comparisons of mean size-adjusted linear measurements (± 1 standard error) for (a) head depth, (b) body depth 1, (c) body depth 2, (d) caudal depth 1, (e) caudal depth 2, (f) caudal length 1, (g) caudal length 2, (h) pelvic fin length, and (i) pectoral fin length. For each measurement, means that do not share letters are significantly different (Tukey's test, $p < 0.05$). Acronyms: CT, cutthroat trout (*Oncorhynchus clarkii bouvieri*); MCH, maternal cutthroat hybrid; MRH, maternal rainbow hybrid; RBT, rainbow trout (*Oncorhynchus mykiss*). All measurements are based on \log_{10} -transformed data.



one genotype in further analyses. Finally, we tested whether morphological scores from PCA of linear characters and geometric shape variables were related to swimming ability by regressing U_{crit} scores from each trout on its corresponding factor score from the first principal axis.

Results

Swimming stamina

We completed 25 swimming trials with 25 fish from each genotype for a total of 100 trials. The average (± 1 standard error, SE) number of swimming increments was 8.16 (0.43) for Yellowstone cutthroat trout, 9.84 (0.51) for maternal cutthroat hybrids, 10.40 (0.56) for maternal rainbow hybrid, and 11.08 (0.41) for rainbow trout. When we compared sustained swimming stamina, we found a significant difference in sustained swimming velocity across the four genotypes (ANOVA, $F_{[3,96]} = 7.05$, $P < 0.001$; Fig. 3). Tukey's test revealed that cutthroat trout had lower mean U_{crit} than all other genotypes. Although both hybrid groups had mean U_{crit} scores intermediate to those of cutthroat trout and rainbow trout, we found no differences between either hybrid group and rainbow trout (Fig. 3). As expected, U_{crit} was not correlated with fork length across genotypes (each genotype, $F_{[1,24]} \leq 1.16$, $P \geq 0.2$), lending support to our assumption

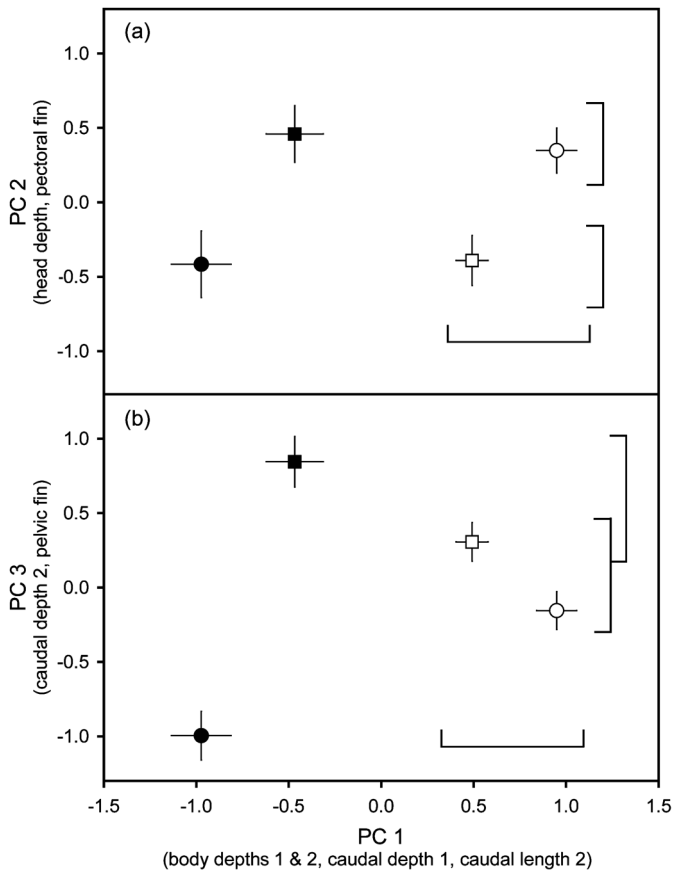
that the body length – water velocity correction removed any effect of size on absolute swimming velocity.

Morphological comparisons

Based on our morphological analysis of linear measurements, we found a significant difference among genotypes (MANOVA, Wilk's $\lambda = 0.0657$, $F_{[27,257.65]} = 14.70$, $P < 0.0001$). When we compared each size-adjusted linear character across the four classes of trout, we found that cutthroat trout had the shallowest distance on all five measures of depth (Figs. 4a–4e). We did not find any differences in caudal length 1 (Fig. 4f); however, cutthroat trout had the shortest caudal length 2 measurements (Fig. 4g). This apparent discrepancy can be explained by increased body depth found in rainbow and hybrid trout, which leads to longer distance between landmarks 7 and 9 than between landmarks 5 and 7 (Fig. 2). Cutthroat trout had the shortest pelvic fins (Fig. 3h) and shortest pectoral fins, which were similar to maternal rainbow hybrids (Fig. 3i).

When we summarized the variation in linear measurements using PCA, we found that all four genotypes were separated based on three principal axes (Fig. 5). The first three principal axes had eigenvalues greater than 1, explaining 66.4% of the total variation (Table 1). None of these axes was correlated with body length (for all correlations, $r^2 \leq$

Fig. 5. Mean principal component scores (± 1 standard error) from a PCA of nine size-adjusted measurements for (a) principal component (PC) 1 versus PC 2 and (b) PC 1 versus PC 3. On each axis, means that share brackets do not differ significantly (Tukey's test, $\alpha = 0.05$). Symbols: ●, cutthroat trout (*Oncorhynchus clarkii bouvieri*); ■, maternal cutthroat hybrid; □, maternal rainbow hybrid; ○, rainbow trout (*Oncorhynchus mykiss*).



0.0008, $P \geq 0.78$), verifying that the size-correction method effectively removed any effect of size across the test fish.

Principal component (PC) 1 summarized variation in body depths 1 and 2, caudal depth 1, and caudal length 2 (Table 1) and was the most effective at distinguishing differences in size-adjusted linear measurements among genotypes (ANOVA, $F_{[3,99]} = 77.95$, $P < 0.0001$; Fig. 5a). Based on PC 1, cutthroat trout had shallow bodies and short caudal peduncles, whereas rainbow trout had deep bodies with longer caudal peduncles. Hybrid genotypes were intermediate, with maternal cutthroat hybrids more similar to cutthroat trout and maternal rainbow hybrids more similar to rainbow trout. PC 2 summarized variation in head depth and pectoral fin length (Table 1) and identified differences between the genotypes with a paternal effect (ANOVA, $F_{[3,99]} = 9.20$, $P < 0.0001$; Fig. 5a); cutthroat trout and maternal rainbow hybrid trout had shallower head depth and shorter pectoral fins than maternal cutthroat hybrids and rainbow trout. PC 3 summarized variation in caudal depth 2 and pelvic fin length (Table 1) and identified differences among genotypes (ANOVA, $F_{[3,99]} = 12.29$, $P < 0.0001$). Based on PC 3, cutthroat trout had shallower caudal peduncles and shorter pelvic fins than the other genotypes (Fig. 5b).

The morphometric analysis with geometric shape variables provided results similar to those from the size-adjusted linear measurements. A MANOVA of the partial warp scores detected significant differences among genotypes (Wilk's $\lambda = 0.0441$, $F_{[45,244.38]} = 10.10$, $P < 0.0001$). Principal component scores from axis 1 and 2 from the PCA of the geometric shape variables summarized 35.4% of the variation in landmark locations. Across both axes, we found a significant effect of genotype (PC 1, $F_{[3,99]} = 37.59$, $P < 0.0001$; PC 2, $F_{[3,99]} = 9.66$, $P < 0.0001$). When we graphically interpreted the implied shape changes for these axes, we found that PC 1 describes similar shape changes as PC 1 from the analysis of linear characters. Increasing scores on the first axis describe a deepening of the fish across the entire body (Fig. 6). Based on PC 1, cutthroat trout had the shallowest bodies, rainbow trout had the deepest bodies, and hybrid genotypes had intermediate body depth (Fig. 6). PC 2 describes a slight elongation of the head owing to a shift in landmarks 1, 2, and 15 (Fig. 6). Based on PC 2, cutthroat trout had more elongated heads than the other genotypes (Fig. 6).

Swimming performance in relation to morphology

When we combined swimming performance and body shape from individual fish, we found that body shape explained a significant proportion of the variation in critical swimming velocity. We found a positive relationship between U_{crit} scores and PC 1 from the linear measurements ($F_{[1,99]} = 11.11$, $P = 0.001$; Fig. 7a) and PC 1 from the geometric shape variables ($F_{[1,99]} = 7.69$, $P = 0.007$; Fig. 7b). Higher sustained swimming velocities are related to the combination of deeper bodies, larger fins, and larger caudal peduncles found in rainbow trout and hybrid trout.

Discussion

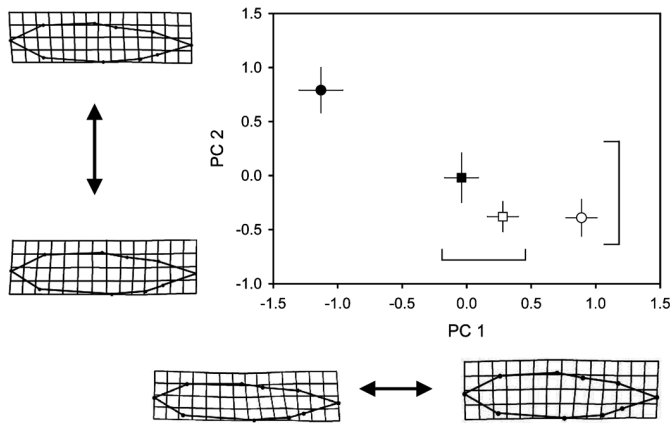
Our study provides evidence for differences in maximum sustained swimming speed and morphology between Yellowstone cutthroat trout, rainbow trout, and their hybrids. Although rainbow and cutthroat trout have similar body plans, our data show that rainbow trout have deeper bodies and larger paired fins than cutthroat trout. Morphological differences in salmonid fishes are often related to swimming and feeding ecology (McLaughlin and Grant 1994; Keeley et al. 2005), and sustained swimming ability is often measured as a correlate of fitness because it influences several components of fitness, including habitat use and the ability to undergo extensive migrations (Brett 1964; Taylor and McPhail 1985a; Taylor and Foote 1991). We found that individuals with the highest swimming stamina were those with the deepest bodies and largest fins. Hence, even subtle differences in morphology appear to provide an advantage in sustained swimming ability to rainbow trout and hybrid trout.

Although co-occurring species sometimes hybridize, intermediate morphological or behavioral characteristics often put hybrid individuals at a competitive disadvantage (Arnold 1997). Such selective disadvantages are thought to be a primary mechanism in preventing introgression between species. In contrast, natural isolating mechanisms may not be present when one species is moved outside of its native range and can interbreed with an indigenous species. If hy-

Table 1. Loading coefficients from a principal component analysis with varimax rotation of size-adjusted linear measurements of experimentally reared genotypes.

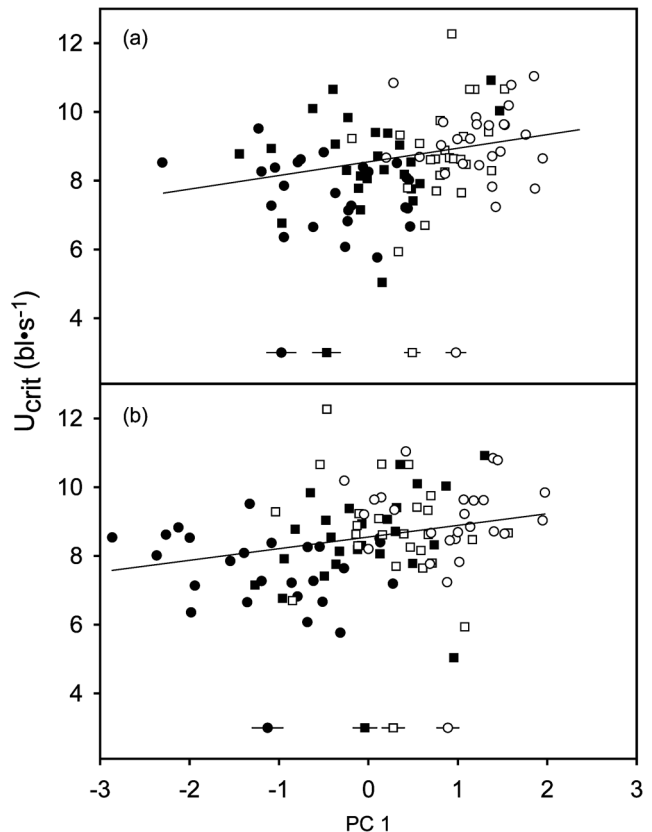
Morphological character	Rotated component loadings				
	Landmarks	PC 1	PC 2	PC 3	
Head depth	2–15	0.178	0.774	0.125	
Body depth 1	3–11	0.894	-0.110	0.174	
Body depth 2	4–11	0.787	0.164	0.364	
Caudal depth 1	5–10	0.734	0.098	0.376	
Caudal depth 2	6–8	0.324	0.088	0.632	
Caudal length 1	5–7	0.399	-0.359	-0.342	
Caudal length 2	7–9	0.833	0.161	-0.026	
Pelvic fin length	11–12	0.106	-0.008	0.809	
Pectoral fin length	13–14	0.011	0.843	-0.051	
Eigenvalue		3.4	1.5	1.0	Total
Variation (%)		38.2	16.7	11.4	66.4

Fig. 6. Mean principal component scores (± 1 standard error) from a principal component analysis (PCA) of geometric shape variables. Thin-plate spline deformation grids show shape changes implied by low and high scores on each axis. On each axis, means that share brackets do not differ significantly (Tukey's test, $\alpha = 0.05$). Symbols: ●, cutthroat trout (*Oncorhynchus clarkii bouvieri*); ■, maternal cutthroat hybrid; □, maternal rainbow hybrid; ○, rainbow trout (*Oncorhynchus mykiss*).



brid individuals possess greater competitive abilities than one of the parental forms, it is likely that hybrids will not be at a selective disadvantage. Our study supports this contention: first generation hybrids had intermediate morphological characteristics and demonstrated swimming abilities that were higher than cutthroat trout. Therefore, unless strong selection occurs against cutthroat-rainbow hybrids at another component of their life cycle, they are likely to be important competitors with native cutthroat trout. Past studies of hybridization between rainbow trout and coastal cutthroat trout indicate similar shape differences and swimming performance (Hawkins and Quinn 1996), as well as no reduced survivorship of hybrid individuals from fertilization to exogenous feeding (Hawkins and Foote 1998). Hence, higher swimming ability of introduced rainbow trout and cutthroat-rainbow hybrids may be especially detrimental to inland cutthroat trout species and is consistent with the pattern of rapid replacement of cutthroat trout populations by rainbow

Fig. 7. (a) The relationship between sustained swimming velocity and body shape as defined by PC 1 from the principal component analysis (PCA) of nine linear measurements. The equation of the line is U_{crit} (body lengths per second ($bl \cdot s^{-1}$)) = $8.566 + 0.408 \times PC 1$, $r^2 = 0.11$, $p = 0.001$. (b) The relationship between sustained swimming velocity and body shape as defined by PC 1 from the PCA of geometric shape variables. The equation of the line is U_{crit} ($bl \cdot s^{-1}$) = $8.566 + 0.364 \times PC 1$, $r^2 = 0.08$, $p = 0.003$. Symbols: ●, cutthroat trout (*Oncorhynchus clarkii bouvieri*); ■, maternal cutthroat hybrid; □, maternal rainbow hybrid; ○, rainbow trout (*Oncorhynchus mykiss*). Symbols above the x axis indicate the mean PC 1 score (± 1 standard error) for each genotype.



trout and hybrid swarms (Jaeger et al. 2000; Hitt et al. 2003; Weigel et al. 2003).

In addition to the pattern between morphology and sustained swimming performance that we report, other factors may play an important role in rainbow–cutthroat trout interactions. For example, in salmonid fishes, burst swimming speed is thought to be important for predator evasion (Taylor and McPhail 1985b), and the impact of propagation and rearing of salmonids in hatcheries may affect interactions between cutthroat trout and rainbow trout by promoting increased growth rates, elevated aggression, and decreased swimming ability (see review by Weber and Fausch 2003). Increased size and behavioral differences may confer a competitive advantage to hatchery-influenced rainbow trout. Across many salmonid species, larger fish have a competitive advantage in territorial competition (Grant 1990; Rhodes and Quinn 1998), with relative mass differences as small as 5% predicting the outcome of territorial competition (Abbott et al. 1985). Similarly, more aggressive trout tend to defend larger feeding territories and obtain more food by displacing conspecifics from profitable foraging locations (Grant 1990). The evidence for decreased swimming ability by hatchery-influenced salmonids provides evidence that our results may be a conservative estimate of the sustained swimming differences between Yellowstone cutthroat trout, rainbow trout, and their hybrids. Given that most, if not all, rainbow trout present within native inland cutthroat trout waterways were reared in hatcheries or are wild descendants from hatchery stocks, future studies should address how growth rate, relative size, and behavioral differences influence competitive interactions between native cutthroat trout, introduced rainbow trout, and their hybrids.

Our study provides an example in which an introduced species and hybrids between the introduced species and a native species have superior performance over the native species in an ecologically important characteristic. Few other studies of animal populations have examined the mechanisms by which introduced species and their hybrids may compete with and spread into native populations. One notable example is Rosenfield et al. (2004), who show that hybrids between Pecos pupfish (*Cyprinodon pecosensis*) and the nonnative sheepshead minnow (*Cyprinodon variegatus*) have higher swimming stamina than both parental species. Our study is unique in that it makes an additional finding: subtle differences in morphology can influence swimming performance and potentially the competitive abilities of individuals.

The spread of rainbow trout into watersheds with native cutthroat trout represents a conspicuous case of freshwater homogenization (see review by Rahel 2002) and provides an excellent case study for invasive species ecology. The native distributions of cutthroat and rainbow trout are well documented (McPhail and Lindsey 1986; Young 1995); therefore, patterns of cutthroat trout displacement can be investigated in watersheds with different histories of rainbow trout introduction and across environmental factors (cf. Hitt et al. 2003; Weigel et al. 2003). Furthermore, hybridization between native cutthroat trout and introduced rainbow trout presents a special opportunity to study a type of ecological invasion thought to have severe implications for native populations. Further studies on this system could lead to stronger

protection of native cutthroat trout populations and develop a better ecological understanding of invasive species.

Acknowledgements

This study was supported in part by NSF grants (DEB-0212445 and 0211818) awarded to ERK and Margaret Ptacek. Additional funding for the trout rearing system came from NSF EPSCoR, a NSF Dissertation Improvement grant (DEB-0508915) awarded to SMS, as well as Idaho State University through awards from the Department of Biological Sciences and the Graduate Student Research and Scholarship Committee (F01-111). Experimental crosses were made possible with help from the Idaho Department of Fish and Game. In particular, we thank Tom Frew, Damon Keen, Brad Dredge, John Mitchell, Jarrett Page, and Dan Baker. Rearing system construction, fish care, and valuable input to this project were provided by Meredith Seiler, Margaret Ptacek, Ken Rodnick, Ralph Chapman, Marjorie Matocq, Janet Loxterman, Adam Keeley, Laurie Fielder, Chad Shouse, Chris Leeseberg, Kelly Gunnell, Ryan McManamay, Trevor Knight, and Michael Sheehy. Two anonymous reviewers provided helpful comments on an earlier draft. Animal care and experimental procedures were approved the Idaho State University Animal Welfare Committee (Protocol No. 0502532).

References

- Abbott, J.C., Dunbrack, R.L., and Orr, C.D. 1985. The interaction of size and experience in dominance relationships of juvenile steelhead trout (*Salmo gairdneri*). *Behaviour*, **92**: 241–253.
- Anderson, E. 1949. *Introgressive hybridization*. John Wiley & Sons, New York.
- Arnold, M.L. 1997. *Natural hybridization and evolution*. Oxford University Press, Oxford, UK.
- Behnke, R.J. 1992. *Native trout of western North America*. American Fisheries Society, Bethesda, Md.
- Bell, W.H., and Terhune, L.D.B. 1970. Water tunnel design for fisheries research. *Fish. Res. Board Can. Tech. Rep. No. 195*. pp. 1–69.
- Boily, P., and Magnan, P. 2002. Relationship between individual variation in morphological characters and swimming costs in brook charr (*Salvelinus fontinalis*) and yellow perch (*Perca flavescens*). *J. Exp. Biol.* **205**: 1031–1036.
- Brett, J.R. 1964. The respiratory metabolism and swimming performance of young sockeye salmon. *J. Fish. Res. Board Can.* **21**: 1183–1226.
- Dowling, T.E., and DeMarais, B.D. 1993. Evolutionary significance of introgressive hybridization in cyprinid fishes. *Nature (London)*, **362**: 444–446.
- Grant, J.W.A. 1990. Aggressiveness and the foraging behaviour of young-of-the-year brook charr (*Salvelinus fontinalis*). *Can. J. Fish. Aquat. Sci.* **47**: 915–920.
- Griffith, J.S. 1988. Review of competition between cutthroat trout and other salmonids. *In Status and management of interior stocks of cutthroat trout. Edited by R.E. Gresswell. Am. Fish. Soc. Symp.* **4**: 134–140.
- Hartman, G.F. 1965. The role of behavior in the ecology and interaction of underyearling coho salmon (*Oncorhynchus kisutch*) and steelhead trout (*Salmo gairdneri*). *J. Fish. Res. Board Can.* **22**: 1035–1081.
- Hawkins, D.K., and Foote, C.J. 1998. Early survival and development of coastal cutthroat trout (*Oncorhynchus clarki clarki*),

- steelhead (*Oncorhynchus mykiss*), and reciprocal hybrids. *Can. J. Fish. Aquat. Sci.* **55**: 2097–2104
- Hawkins, D.K., and Quinn, T.P. 1996. Critical swimming velocity and associated morphology of juvenile coastal cutthroat trout (*Oncorhynchus clarki clarki*), steelhead trout (*Oncorhynchus mykiss*), and their hybrids. *Can. J. Fish. Aquat. Sci.* **53**: 1487–1496.
- Hitt, N.P., Frissell, C.A., Muhlfeld, C.C., and Allendorf, F.W. 2003. Spread of hybridization between native westslope cutthroat trout, *Oncorhynchus clarki lewisi*, and nonnative rainbow trout, *Oncorhynchus mykiss*. *Can. J. Fish. Aquat. Sci.* **60**: 1440–1451.
- Hughes, N.F., and Dill, L.M. 1990. Position choice by drift-feeding salmonids: model and test for Arctic grayling (*Thymallus arcticus*) in subarctic mountain streams, interior Alaska. *Can. J. Fish. Aquat. Sci.* **47**: 2039–2048.
- Jaeger, M.W., Van Kirk, R.W., and Kellogg, T. 2000. Distribution and status of Yellowstone cutthroat trout in the Henry's Fork watershed. *Intermt. J. Sci.* **6**: 197–216.
- Keeley, E.R., Parkinson, E.A., and Taylor, E.B. 2005. Ecotypic differentiation of native rainbow trout (*Oncorhynchus mykiss*) populations from British Columbia. *Can. J. Fish. Aquat. Sci.* **62**: 1523–1539.
- Krueger, C.C., and May, B. 1991. Ecological and genetic effects of salmonid introductions in North America. *Can. J. Fish. Aquat. Sci.* **48**(Suppl. 1): 66–77.
- Lindsey, C.C. 1978. Form, function, and locomotory habits in fish. *In Fish physiology*. Vol. II. Edited by W.S. Hoar and D.J. Randall. Academic Press, New York. pp. 1–100.
- McLaughlin, R.L., and Grant, J.W.A. 1994. Morphological and behavioral differences among recently-emerged brook charr, *Salvelinus fontinalis*, foraging in slow- vs. fast-running water. *Environ. Biol. Fishes*, **39**: 289–300.
- McPhail, J.D., and Lindsey, C.C. 1986. Zoogeography of the freshwater fishes of Cascadia (the Columbia system and rivers north to the Stikine). *In The zoogeography of North American freshwater fishes*. Edited by C.H. Hocutt and E.O. Wiley. John Wiley and Sons, New York. pp. 615–637.
- Ojanguren, A.F., and Brana, F. 2003. Effects of size and morphology on swimming performance in juvenile brown trout (*Salmo trutta* L.). *Ecol. Freshw. Fish*, **12**: 241–246.
- Quinn, G.P., and Keough, M.J. 2002. *Experimental design and data analysis for biologists*. Cambridge University Press, Cambridge, UK.
- Quinn, T.P. 2005. *The behavior and ecology of Pacific salmon and trout*. University of Washington Press, Seattle, Wash.
- Rahel, F.J. 2002. Homogenization of freshwater faunas. *Annu. Rev. Ecol. Syst.* **33**: 219–315.
- Rhodes, J.S., and Quinn, T.P. 1998. Factors affecting the outcome of territorial contests between hatchery and naturally reared coho salmon parr in the laboratory. *J. Fish Biol.* **53**: 1220–1230.
- Rhymer, J.M., and Simberloff, D. 1996. Extinction by hybridization and introgression. *Annu. Rev. Ecol. Syst.* **27**: 83–109.
- Rohlf, F.J. 1993. Relative warp analysis and an example of its application to mosquito wings. *In Geometrical morphometrics*. Edited by L.F. Marcus, E. Bello, and A. Garcia-Valdecasas. Monografias Museo Nacional de Ciencias Naturales, Madrid, Spain. pp. 131–159.
- Rohlf, F.J. 2005a. tpsDig, digitize landmarks and outlines. Version 2.04. Department of Ecology and Evolution, State University of New York at Stony Brook, New York.
- Rohlf, F.J. 2005b. tpsRelw, relative warps analysis. Version 1.42. Department of Ecology and Evolution, State University of New York at Stony Brook, New York.
- Rosenfield, J.A., Nolasco, S., Lindauer, S., Sandoval, C., and Kodric-Brown, A. 2004. The role of hybrid vigor in the replacement of Pecos pupfish by its hybrids with sheepshead minnow. *Conserv. Biol.* **18**: 1589–1598.
- Rubidge, E.M., and Taylor, E.B. 2004. Hybrid zone structure and the potential role of selection in hybridizing populations of native westslope cutthroat trout (*Oncorhynchus clarki lewisi*) and introduced rainbow trout (*O. mykiss*). *Mol. Ecol.* **13**: 3735–3749.
- Sakai, A.K., Allendorf, F.W., Holt, J.S., Lodge, D.M., Molofsky, J., With, K.A., Baughman, S., Cabin, R.J., Cohen, J.E., Ellstrand, N.C., McCauley, D.E., O'Neil, P., Parker, I.M., Thompson, J.N., and Weller, S.G. 2001. The population biology of invasive species. *Annu. Rev. Ecol. Syst.* **32**: 305–332.
- Schrader, W.C., and Gamblin, M. 1996. Region 6 rivers and streams investigation, South Fork Snake River. Idaho Department of Fish and Game, Federal Aid in Sport Fisheries Restoration, Regional Fisheries Management Investigation, Job Performance Report Program F-TIR-16, Boise, Idaho.
- Stebbins, G.L. 1959. The role of hybridization in evolution. *Proc. Am. Philos. Soc.* **103**: 231–251.
- Swain, D.P., and Holtby, L.B. 1989. Differences in morphology and behavior between juvenile coho salmon (*Oncorhynchus kisutch*) rearing in a lake and in its tributary stream. *Can. J. Fish. Aquat. Sci.* **46**: 1406–1414.
- Taylor, E.B., and Foote, C.J. 1991. Critical swimming velocity of juvenile sockeye salmon and kokanee, the anadromous and non-anadromous forms of *Oncorhynchus nerka* (Walbaum). *J. Fish Biol.* **38**: 407–419.
- Taylor, E.B., and McPhail, J.D. 1985a. Variation in body morphology among British Columbia populations of coho salmon, *Oncorhynchus kisutch*. *Can. J. Fish. Aquat. Sci.* **42**: 2020–2028.
- Taylor, E.B., and McPhail, J.D. 1985b. Burst swimming and size-related predation of newly emerged Coho salmon, *Oncorhynchus kisutch*. *Trans. Am. Fish. Soc.* **114**: 546–551.
- Thorpe, R.S. 1976. Biometric analysis of geometric variation and racial affinities. *Biol. Rev.* **51**: 105–108.
- Trotter, P.C. 1989. Coastal cutthroat trout: a life history compendium. *Trans. Am. Fish. Soc.* **118**: 463–473.
- Varley, J.D., and Gresswell, R.E. 1988. Ecology, status, and management of the Yellowstone cutthroat trout. *In Status and management of interior stocks of cutthroat trout*. Edited by R.E. Gresswell. *Am. Fish. Soc. Symp.* **4**: 13–24.
- Webb, P.W. 1984. Body form, locomotion, and foraging in aquatic vertebrates. *Am. Zool.* **24**: 107–120.
- Weber, E.D., and Fausch, K.D. 2003. Interactions between hatchery and wild salmonids in streams: differences in biology and evidence for competition. *Can. J. Fish. Aquat. Sci.* **60**: 1018–1036.
- Weigel, D.E., Peterson, J.T., and Spruell, P. 2003. Introgressive hybridization between native cutthroat trout and introduced rainbow trout. *Ecol. Appl.* **13**: 38–50.
- Young, M.K. 1995. Conservation assessment for inland cutthroat trout. *US For. Serv. Gen. Tech. Rep. No. RM-256*.
- Zelditch, M.L., Swiderski, D.L., Sheets, H.D., and Fink, W.L. 2004. *Geometric morphometrics for biologists*. Elsevier Academic Press, San Diego, Calif.