



Enhanced smolt characteristics of steelhead trout exposed to alternative hatchery conditions during the final months of rearing

Gayle B. Zydlewski^{a,*}, J. Scott Foott^b, Kenneth Nichols^b,
Scott Hamelberg^c, Joseph Zydlewski^d, Björn Thrandur Björnsson^e

^aU.S. Fish and Wildlife Service, Abernathy Fish Technology Center, 1440 Abernathy Creek Road, Longview, WA 98632, USA

^bU.S. Fish and Wildlife Service, California Nevada Fish Health Center, 24411 Coleman Fish Hatchery Road, Anderson, CA 96007, USA

^cU.S. Fish and Wildlife Service, Coleman National Fish Hatchery, 24411 Coleman Fish Hatchery Road, Anderson, CA 96007, USA

^dU.S. Fish and Wildlife Service, Columbia River Fisheries Program Office, 9317 Highway 99, Suite 1, Vancouver, WA 98665, USA

^eFish Endocrinology Laboratory, Department of Zoology/Zoophysiology, Göteborg University, Box 463, S-40530 Göteborg, Sweden

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Abstract

One-year steelhead trout were reared at Coleman National Fish Hatchery, CA, USA. During the final 5 months of rearing, fish were maintained in either traditional concrete raceways or moved to a pond. Rearing conditions in the pond included a cobble bottom, structures, and surface disruption. In August, prior to splitting juveniles into raceway and pond groups, length, weight, gill Na^+ , K^+ -ATPase activity, seawater tolerance and plasma levels of growth hormone (GH) and insulin-like growth factor I (IGF-I) were measured. On November 30th and January 3rd (prior to release on January 8th), fish were sampled as in August. The pond-reared fish grew faster (0.51 g day^{-1} from August to November and 0.66 g day^{-1} from November to January) than raceway-reared fish (0.41 and 0.26 g day^{-1}) fed at similar rations ($2\% \text{ body weight day}^{-1}$) through November and when raceway fish were fed more in December. In both November and January, pond-reared fish had significantly higher plasma GH levels and tolerated seawater transfer better than fish reared in raceways. Factors such as feeding conditions, structure, and fish density in the pond that differed

* Corresponding author. Tel.: +1-360-425-6072; fax: +1-360-636-1855.

E-mail address: gayle_zydlewski@fws.gov (G.B. Zydlewski).

from raceway conditions may have provided naturalization of the hatchery environment resulting in more physiologically competent smolts.

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1. Introduction

For most salmonid populations, hatcheries play an integral part in mitigation and supplementation efforts. In the Pacific Northwest of the United States, attempts are being made to rear salmonids under conditions that better mimic those conditions experienced by wild fish. The assumption is that fish reared in this manner will have physiological and behavioral characteristics more consistent with those observed in the wild. The guiding principle of current hatchery management is to produce high quality fish that are “behaviorally, morphologically and physiologically indistinguishable from their wild counterparts” (Northwest Power Planning Council, 1999).

Hatchery conditions generally consist of high fish density rearing environments starting from incubation. Most hatcheries will reduce the number of times they have to handle fish by moving them directly from incubators to raceways. Raceways typically have a uniform environment constructed of concrete. Fish are usually stocked into raceways at a fish density of approximately 300 fish m⁻³. In the Pacific Northwest, juvenile salmonids (i.e. primarily steelhead trout, *Oncorhynchus mykiss*; stream-type chinook salmon, *O. tshawytscha*; and coho salmon, *O. kisutch*) are reared through smolting and released into stream environments.

Preliminary studies have been conducted to determine whether standard hatchery conditions can be modified to produce hatchery fish that are more similar to their wild counterparts. Natural Rearing Enhancement Systems (NATURES) have been implemented at an experimental scale. NATURES conditions include cover (suspended camouflage netting), structure (fir trees), and substrate (pea gravel pavers). Evidence suggests NATURES conditions may increase fitness of released chinook salmon (Maynard et al., 1995, 1996). To date, increased fitness has been measured as increased survival within freshwater upon release (in-stream survival) and enhanced cryptic coloration. Preliminary studies reveal that NATURES conditions, in combination, result in increased post-release survival of chinook salmon in-stream. However, the overall effects of NATURES rearing have not been evaluated at full production scale, furthermore, the effects of more natural rearing conditions on the physiological condition (i.e. smolt development) of fish being released from these semi-natural environments has not been evaluated.

Successful transformation to the smolt stage (smolting) involves a suite of physiological, morphological, and behavioral changes associated with migration and seawater (SW) entry. Smolting of wild salmonids is characterized by seasonal increases in gill Na⁺,K⁺-ATPase activity (Rodgers et al., 1987; Ewing and Rodgers, 1998; Beckman et al., 2000), plasma growth hormone (GH) levels (Prunet et al., 1989; Young et al., 1989; Tanguy et al., 1994; McCormick et al., 1995; Björnsson, 1997), plasma insulin-like growth factor-I (IGF-I) levels

(Beckman et al., 2000), and increased salinity tolerance (Yada et al., 1991; Shrimpton et al., 1994a). There is evidence that these physiological changes are necessary for the migratory performance and survival of smolts. Brown trout (*Salmo trutta*) smolts that did not fully exhibit physiological changes had reduced long-term survival rates, reduced return rates and reproductive success (Morán et al., 1991). Therefore, physiological smolt characters may be used as an indirect measure of preparedness for migration and SW entry and ultimately as predictors of survival and return rates of hatchery fish.

In general, salmonids reared in the hatchery environment are larger and have depressed physiological expression of smolting. Rearing conditions have been shown to affect physiological development of smolting. For example, gill Na^+, K^+ -ATPase activity is significantly lower in hatchery Atlantic salmon, *Salmo salar* (McCormick and Björnsson, 1994), brown trout (Sundell et al., 1998; Nielsen et al., 1999) and coho salmon (Shrimpton et al., 1994a,b). Gill Na^+, K^+ -ATPase activity is usually correlated with SW tolerance and hatchery fish generally have depressed SW tolerance (e.g. coho salmon; Shrimpton et al., 1994a). Similarly, hatchery-reared fish generally have a diminished smolt-related increase in plasma GH (McCormick and Björnsson, 1994; Sundell et al., 1998).

Emigrating steelhead trout from Sacramento River, CA tributaries have been captured migrating downstream from December to April at 14.0–20.0 cm (Nobriga and Cadrett, 2001). There is little baseline information particularly on smolt development of this threatened (National Marine Fisheries Service, 1998) population of steelhead trout. However, genetic evidence indicates that this population is very distinct from other Pacific Coast populations (Behnke, 1992). The distinctive genetic characteristics of this population are likely due to the extreme environmental conditions in the region; low summer flows contribute to temperatures that exceed the thermal limit of steelhead trout. These conditions probably have promoted strong local adaptation and inhibited gene flow with other Pacific Coast populations (Busby et al., 1996). These adaptations are typified by the migration timing of smolts that occurs in the winter months at this geographic location. In contrast, smolting occurs in spring for more northerly populations of steelhead trout (e.g. Zugg, 1981). Based on available data, steelhead released in Battle Creek (a tributary of the Sacramento River) are similarly sized to naturally produced smolts and emigrate during a narrower time frame tightly correlated with time of release, January–February (Nobriga and Cadrett, 2001).

Efforts to reduce the impact of hatchery conditions on steelhead smolt development are being implemented at the U.S. Fish and Wildlife Service Coleman National Fish Hatchery (NFH) on Battle Creek in Anderson, CA. Conventionally reared fish are held in 4.6×46 -m concrete raceways. For the last 5 months of rearing, however, a group of steelhead trout (approximately 90,000) is transferred to a pond. The pond has a cobble bottom, structures and surface disruption. Fish are fed by casting traditional pelleted feed from behind a screen and natural food items are likely more available than in raceways. It is predicted that the more natural conditions found in the pond may produce smolts that have physiological characters that allow them to deal with the life history change of downstream migration and seawater entry. Smolt parameters would be closer to those documented in wild fish, i.e. be physiologically more competent than traditionally reared hatchery fish. The effect of rearing conditions on physiological characters for smolting has yet to be evaluated.

The objective of this study was to determine the effectiveness of a semi-natural rearing environment on the physiological smolt conditions (fitness upon release) of steelhead trout. Specifically, differences between osmoregulatory physiology and the GH/IGF-I axis were compared for pond- and raceway-reared smolts.

2. Materials and methods

2.1. Fish

Steelhead trout returning to Battle Creek (a tributary of the Sacramento River), Anderson, CA, USA in January 2000 were spawned one female to one male. Fish were reared in ozone-treated river water. Eggs were reared in Heath (Flex-a-Lite Consolidated, Tacoma, WA) trays, moved to indoor tanks until they reached approximately 2 g, then moved to raceways as parr in May or June 2000. Experimental fish were reared in raceways until September. On September 11th, 90,000 fish were moved to the pond and on September 13th and 14th, 36,000 and 40,000 fish were distributed into two experimental raceways (raceways 11 and 12). The weight of the fish was 16 ± 4.5 g (mean \pm S.D.).

The pond was approximately 906 m^3 (12.2 m wide \times 61 m long \times 1.2 m deep) and the raceways were 127 m^3 (4.6 m wide \times 46 m long \times 0.6 m deep). The pond had a cobble bottom, structure (three 220-l barrels, two or three 20-cm round PVC pipe sections approximately 1 m long bundled together, and four 1-m square PVC lattice structures), and sprinklers to disperse the water surface. Feeding in the pond was accomplished by casting feed from behind a screen to minimize visual contact. Pond fish were fed three to four times per day before 12:00. Raceways had traditional concrete bottoms with no structure or surface disruption. Raceway fish were fed by hand in clear view three to four times per day before 12:00.

All fish were fed Silver Cup (Nelson & Sons, Murray, UT) pelleted feed. In September and October, fish in the raceways and pond were fed at a rate of 1.8–2.0% body weight day^{-1} . In November, feeding rate was decreased due to decreases in water temperature, fish were fed at 1.2–1.3% body weight day^{-1} . In December, feeding rates were increased to 2.0% and 2.8% body weight day^{-1} in the pond and raceways, respectively. Based on November fish sampling, raceway fish were smaller than pond-reared fish, therefore, December feeding rates were adjusted to be higher in the raceways in an attempt to produce fish of the same size upon release. Water temperatures in the raceways and pond were within 0.8 °C. Water temperature decreased from an average of 16.1 °C in August to 7.1 °C in December (Fig. 1). Water from Battle Creek was passed through an ozone treatment plant, into one-pass raceways and then through the pond. Water flows through the raceways were lower ($25.7 \text{ l min}^{-1} \text{ m}^{-3}$) than through the pond ($33.5 \text{ l min}^{-1} \text{ m}^{-3}$). Both rearing units had natural lighting.

2.2. Fish handling

Juvenile steelhead trout were sampled on three occasions: August 28th 2000, November 30th 2000, and January 3rd 2001. In August, fish were sampled from raceways before

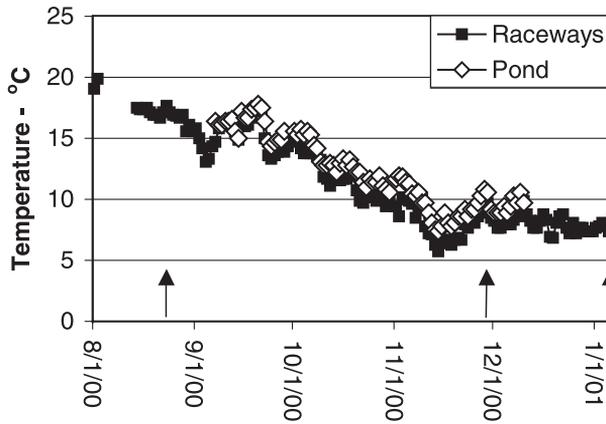


Fig. 1. Water temperature profiles for raceways and pond rearing units. Arrows indicate sampling dates: August 28th 2000, November 30th 2000, and January 3rd 2001.

experimental fish were transferred to either the pond or the experimental raceways. In November and January, fish were sampled from experimental raceways and the pond. On all occasions, fish were captured using a 2.4-m diameter cast net with 0.64-cm mesh. Fish were rapidly transferred to a 19.4-l bucket with 125 mg l⁻¹ MS-222 (pH = 7.0; buffered with 25 mg l⁻¹ NaHCO₃) and 4 l of raceway or pond water. Fish were measured (nearest 0.1 cm) and weighed (nearest 0.1 g). In August, fish were bled by severing the caudal peduncle and blood collected into heparinized capillary tubes. In November and January, blood was collected from the caudal vein using heparinized syringes, put in tubes and centrifuged in order to obtain plasma within 5 min. The plasma was frozen on dry ice and stored at -80 °C until analysis. Gill biopsies were taken from all fish, placed in SEI-buffer (300 mM sucrose, 20 mM Na₂EDTA, 50 mM imidazole, pH 7.3), put on dry ice then stored at -80 °C until analysis. A total of 40 fish were sampled for length, weight, and gill biopsy; blood was taken from 20 of these fish for plasma hormone analyses.

An additional 12 fish were netted from each rearing unit and placed into aerated 132-l aquaria with 26–28‰ SW (26‰ in August, 27.5‰ in November, 27‰ in January) at 8–15 °C (15 °C in August, 10.9 °C in November, and 8 °C in January) for 24-h SW challenge. After 24 h, survival was noted and plasma obtained and stored as described above.

2.3. Plasma and tissue analysis

Plasma from pond- and raceway-reared fish ($n=20$) was analyzed for plasma GH and IGF-I. Plasma GH levels were analyzed using the RIA method described by Björnsson et al. (1994). Plasma IGF-I levels were assessed using the RIA method by Moriyama et al. (1994). Gill Na⁺,K⁺-ATPase activity was measured using the methods outlined by McCormick (1993).

For SW-challenged fish, plasma osmolality was measured using a Wescor Model 5520 vapor pressure osmometer. Plasma sodium levels were measured using a Jenway Model PFP7 flame photometer. Plasma chloride levels were determined colorimetrically at 450

nm using Sigma Chemical Company Diagnostics procedure No. 461 modified for a microplate reader.

2.4. Statistical analyses

In all cases, analyses were run on ranked data because the data failed to meet the assumptions of parametric statistics. Two-way analysis of variance (ANOVA) and one-way ANOVA on ranks were conducted for all parameters as appropriate. Significance was followed by post hoc analysis of ranked data using the Tukey test. Raceways 11 and 12 were good replicates in most cases. Statistics were run for pooled data for raceways unless this was not appropriate (as indicated in tables or figures). Spearman rank order correlations were conducted between length and all measured factors to determine if observed differences may be due to size. Spearman rank order correlations were also conducted for GH on condition factor and gill Na^+, K^+ -ATPase activity (pooled for November and January, as well as rearing units) to explain the relative influence of GH on smolting.

3. Results

For all samples, raceways 11 and 12 were good replicates for the following parameters: length, weight, condition factor, plasma IGF-I, plasma osmolality (of SW-challenged fish), and gill Na^+, K^+ -ATPase activity. Data were pooled for these raceways for comparisons of the previous factors. In November only, raceways 11 and 12 differed for plasma GH, plasma Na^+ and plasma Cl^- (of SW-challenged fish) and data were not pooled for ANOVAs for these parameters. When raceways were pooled for comparisons, statistical differences for raceway and pond were maintained.

3.1. Size and condition factor

Growth and condition factor of fish in both rearing units are given in [Table 1](#). Pond-reared fish were larger (both length and weight) than raceway-reared fish in November and January ([Table 1](#)). Size distributions in the raceways and pond were unimodal in November and January. Effects of date and rearing unit on length and weight were both significant. The effect of date on length was independent of rearing unit ($p=0.061$). However, the effect of date on weight was dependent on rearing unit ($p=0.011$).

From August to November (94 days), fish in the pond grew 0.51 g day^{-1} , while the fish from the raceways grew 0.41 g day^{-1} . From November to January, fish in the pond gained 0.66 g day^{-1} , while those from the raceways gained 0.26 g day^{-1} .

In November, pond-reared fish had significantly lower condition factor than those reared in the raceways. However, condition factor was significantly higher for pond-reared fish in January. In both rearing units, condition factor decreased from November to January. Significant differences for condition factor were primarily due to date alone ($p<0.001$). The effect of rearing unit was not significant ($p=0.871$), however, there was a significant interaction between date and rearing unit for condition factor ($p=0.001$).

Table 1
Post hoc results of two-way ANOVA comparing length, weight, and condition factor

	Length (cm)			Weight (g)			Condition factor ($\times 100 \text{ g cm}^{-3}$)		
	August	November	January	August	November	January	August	November	January
Pond	–	17.4 ^a (14.1–20.3)	19.6 ^b (15.2–22.5)	–	62.1 ^a (30.2–100.2)	84.4 ^b (53.3–147.3)	–	1.18 ^a (1.05–1.87)	1.14 ^b (0.94–1.96)
Raceway	10.7 (8.3–12.9)	16.3 ^{a*} (12.7–19.4)	17.8 ^{b*} (13.0–21.2)	14.0 (6.6–26.8)	52.7 ^{a*} (24.9–90.9)	61.4 ^{b*} (17.9–111.0)	1.13 (0.90–1.27)	1.22 ^{a*} (1.04–1.43)	1.11 ^{b*} (0.82–1.31)

Values are median and range (in parentheses). Comparisons are made between rearing units (pond and raceway) within sampling dates (differences denoted by asterisk on raceway value) and between dates (November and January) within rearing units, differences denoted by letters. Sample size for raceway and pond is 80 and 40, respectively. Significant differences are $p < 0.05$.

3.2. Plasma GH and IGF-I levels

Plasma GH levels increased in both groups over the course of the study. Levels were nearly threefold higher in pond-reared fish than raceway-reared fish in both November and January (Fig. 2). There was no significant interaction between date and rearing unit ($p=0.407$), but both date and rearing unit effects were significant ($p<0.001$). Length was not correlated with GH levels in November or January ($p=0.869$ and $p=0.960$, respectively), whereas GH levels were weakly correlated with gill Na^+, K^+ -ATPase activity (positive, $p=0.037$, correlation coefficient=0.208) and condition factor (negative, $p<0.001$, correlation coefficient=−0.494).

Plasma IGF-I levels increased in both groups over the course of the study. Levels did not differ between groups in November, but plasma IGF-I levels were lower in pond-reared fish than raceway-reared fish in January (Fig. 3). There was a significant interaction between date and rearing unit ($p=0.030$) where rearing unit ($p<0.001$) had more of an effect on changes in IGF-I than date ($p=0.461$). In November, length and IGF-I levels were negatively correlated ($p=0.047$, correlation coefficient=−0.285), but this correlation was not significant in January ($p=0.464$). GH and IGF-I (all values pooled) were positively correlated (Spearman rank order correlation coefficient=0.222, $p=0.009$).

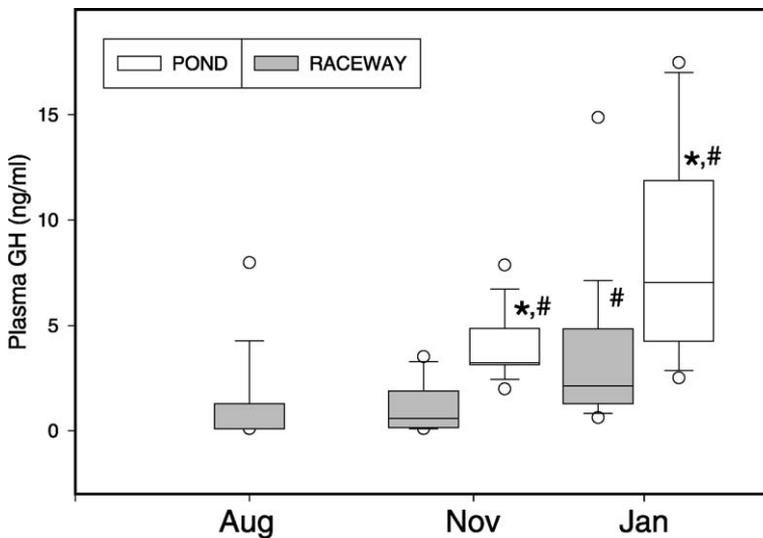


Fig. 2. Box plots of median, 5th and 95th percentiles (circles), for plasma Growth Hormone of steelhead reared in pond and raceway environments from September to January. Differences between baseline levels in August and levels in November and January are denoted with #. Differences between raceway and pond within November and January are denoted with *. Sample size for raceway and pond is 40 and 20, respectively. Differences between pond and raceway in November are for pond vs. raceway 11 only (when pooled, the statistical differences are maintained).

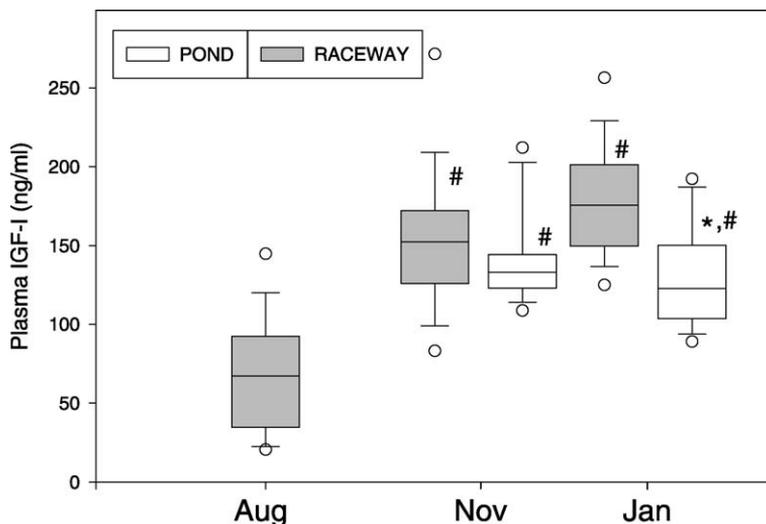


Fig. 3. Box plots of median, 5th and 95th percentiles, for plasma Insulin-like Growth Factor-I of steelhead reared in pond and raceway environments from September to January. Differences between baseline levels in August and levels in November and January are denoted with #. Differences between raceway and pond within November and January are denoted with *. Sample size for raceway and pond is 40 and 20, respectively.

3.3. SW tolerance and gill Na^+ , K^+ -ATPase activity

In August (raceway samples only), survival after 24-h SW challenge was 88% (21/24 fish). In November, 24-h SW challenge survival of fish reared in the pond and raceways was 100% (12/12 and 24/24, respectively). In January, survival following 24-h SW exposure of fish reared in the pond was 100% (12/12), compared to 96% (23/24) for fish reared in raceways.

Plasma Na^+ and plasma Cl^- , plasma Na^+ and plasma osmolality, and plasma Cl^- and plasma osmolality were all positively correlated (correlations coefficients: 0.462, 0.710, 0.622, respectively, and $p < 0.0001$ for all correlations). Plasma ion levels and plasma osmolality were used as three measures of the same aspect of SW tolerance.

In January, fish from the pond had significantly greater SW tolerance than fish reared in raceways. Osmotic perturbations were greater for SW challenged raceway-reared fish than SW challenged pond-reared fish in January (Table 2); plasma Na^+ and plasma osmolality were 15% and 7% higher, respectively. In November, plasma Na^+ was 6% higher in SW challenged raceway fish than in pond-reared fish while there were no differences for plasma osmolality. There were no differences for rearing unit or date for plasma Cl^- . There was a significant interaction between date and rearing unit ($p = 0.013$) where date had less of an effect ($p = 0.101$) than rearing unit ($p = 0.017$) on plasma osmolality. Plasma osmolality was negatively correlated with length in November and January ($p = 0.018$, correlation coefficient = -0.393 , and $p < 0.001$, correlation coefficient = -0.589 , respectively).

In November, perturbation in plasma Na^+ of SW-challenged fish was less for fish from the pond than those from the raceways. This difference was maintained in January

Table 2
Post hoc results of two-way ANOVA for plasma osmolality, plasma Na⁺, and plasma Cl⁻ of seawater-challenged steelhead

	Plasma osmolality (mmol kg ⁻¹)			Plasma Na ⁺ (mmol l ⁻¹)			Plasma Cl ⁻ (mEq l ⁻¹)		
	August	November	January	August	November	January	August	November	January
Pond	–	375 (338–388)	365 (343–409)	–	160 ^a (148–172)	168 ^b (156–200)	–	134 (125–153)	132 (125–144)
Raceway	417 (354–484)	368 ^a (332–437)	392 ^{b*} (364–530)	191 (166–216)	169 ^{a*} (148–201)	194 ^{b*} (169–252)	151 (136–163)	132 (118–166)	136 (121–171)

Values are median and range (in parentheses). Comparisons are made between rearing units (pond and raceway) within sampling dates (differences denoted by asterisk on raceway value) and between dates (November and January) within rearing units, differences denoted by letters. Sample size for raceway and pond is 24 and 12, respectively. Significant differences are $p < 0.05$.

For the difference between the raceway and pond November comparison of plasma Na⁺ and Cl⁻, the difference is between pond and raceway 11 only, raceway 12 is not significantly different from pond (when pooled, the statistical differences are maintained).

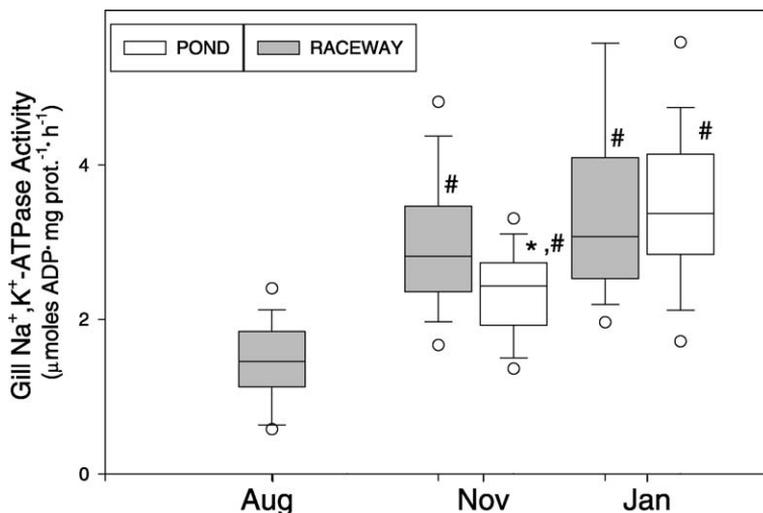


Fig. 4. Box plots of median, 5th and 95th percentiles, for gill Na^+,K^+ -ATPase activity of steelhead reared in pond and raceway environments from September to January. Differences between baseline levels in August and levels in November and January are denoted with #. Differences between raceway and pond within November and January are denoted with *. Sample size for raceway and pond is 80 and 40, respectively.

when plasma Na^+ of fish reared in the pond was significantly lower than for fish reared in raceways. No significant interaction between date and rearing unit was observed for perturbations of plasma Na^+ or plasma Cl^- of SW-challenged fish. Both date and rearing unit had significant effects on plasma Na^+ ($p < 0.001$ for both), but not for plasma Cl^- ($p = 0.499$ and 0.953). Plasma Na^+ was negatively correlated with length in November and January (correlation coefficient = -0.385 , $p = 0.022$ and correlation coefficient = -0.636 , $p < 0.001$, respectively). Plasma Cl^- was negatively correlated with length in January (correlation coefficient = -0.496 , $p = 0.004$) but not in November ($p = 0.140$).

Increases in gill Na^+,K^+ -ATPase activity occurred from November to January in both rearing units. In November, fish in the pond had significantly lower gill Na^+,K^+ -ATPase activity than those in the raceways (Fig. 4). This difference was not maintained in January when levels for the pond and raceways were similar. There was a significant interaction between date and rearing unit ($p = 0.002$) where both date ($p < 0.001$) and rearing unit ($p = 0.048$) had significant effects on gill Na^+,K^+ -ATPase activity. Length and gill Na^+,K^+ -ATPase activity were not correlated in November or January ($p = 0.367$ and $p = 0.754$, respectively).

4. Discussion

Physiological characteristics of pond-reared fish indicated that they were better prepared for SW entry (and perhaps downstream migration) at the time of release. The

physiological parameters examined in this study traditionally have been used as indicators of smolting in hatchery and wild fish (Zaugg, 1981; Stefansson et al., 1991; Rodgers et al., 1987; Björnsson et al., 1989, 1995; Prunet et al., 1989; Young et al., 1989; Yada et al., 1991; Shrimpton et al., 1994a; McLeese et al., 1994; Solbakken et al., 1994; Schmitz et al., 1994; Tanguy et al., 1994; McCormick et al., 1995; Björnsson, 1997; Ewing and Rodgers, 1998; Beckman et al., 2000). It should be noted however, that these parameters are not being used as an indicator of survival. This is a topic that is actively being discussed in the literature (Zaugg, 1989; Virtanen et al., 1991; Beckman et al., 1999). In this study, differences in parameters are being used as an indication of physiological and developmental status of fish produced from two rearing units. Comparison at three discrete time points is used to infer differences in stages and physiological performance of these fish. Data collected from returning adults of the two rearing groups in this study, which were tagged with unique coded wire tags, will allow future assessment of smolt parameters as indicators of survival in this stock of steelhead trout.

One of the most indicative physiological characters associated with smolting is SW tolerance, the ability to tolerate rapid changes in environmental osmolality. Better SW tolerance in January, 15% lower plasma Na^+ and 7% lower plasma osmolality, of pond-reared fish may provide an advantage to those fish upon release. Osmotic perturbations, the result of poor SW tolerance, have been demonstrated to affect schooling behavior and reduce escape success from predators of Atlantic salmon during downstream migration (Handeland et al., 1996). Pond-reared fish may have been better prepared for downstream migration and SW entry than those reared in raceways.

One of the most striking physiological differences between pond-reared and raceway-reared fish was the significantly higher levels of plasma GH in pond-reared fish demonstrated in both November and January. A number of endocrine systems are involved in changes associated with smolting, but GH is thought to be the primary mediator of environmental information for smolt development (Björnsson, 1997; Dickhoff et al., 1997). GH and IGF-I have been shown to increase during smolting (Dickhoff et al., 1997) and be related to upregulation of gill Na^+, K^+ -ATPase activity and increased SW tolerance (McCormick, 1995). The relationship between plasma levels of GH and IGF-I is not fully understood for smolting salmonids, although a positive relationship between growth and both GH and IGF-I has been noted (McCormick et al., 2000). The direct relationship between GH and IGF-I observed in this study is consistent with the understanding that most of the plasma IGF-I levels in vertebrates is a result of direct stimulation of the liver by circulating GH (Yakar et al., 1999; Sjögren et al., 1999). As a result, circulating GH and IGF-I levels are usually correlated. However, further studies of the GH-IGF-I axis are necessary before conclusions can be drawn for fish. That withstanding, it is important to note that for both these endocrine factors, levels for pond-reared and raceway-reared fish differed significantly.

While SW tolerance and GH levels were different for the different rearing units there were no differences in pond and raceway-reared groups for gill Na^+, K^+ -ATPase activity. Gill Na^+, K^+ -ATPase activity, a traditional measure of smolting salmonids, has been used as an indirect indicator of SW tolerance, and smolt development. In both pond and raceway-rearing, a smolting-related increase in this enzyme was observed, providing evidence that these groups were undergoing smolting in a parallel time frame. However,

gill Na^+, K^+ -ATPase activity was not significantly correlated with plasma osmolality or plasma Na^+ of the SW-challenged fish in this study. The numbers of fish measured in this study (12 per group) may not have been adequate to measure such a correlation. There is conflicting evidence concerning the use of gill Na^+, K^+ -ATPase as a measure of smolting in steelhead. [Zaugg \(1981\)](#) found a positive association between gill Na^+, K^+ -ATPase and migration in steelhead trout, however, [Ewing et al. \(1984\)](#) did not. The lack of differences between the rearing units is likely a result of this weak association between SW tolerance and gill Na^+, K^+ -ATPase in this species.

A smolting-related decline in condition factor was observed for both pond- and raceway-reared fish. Decreased condition factors have been associated with smolting and have been linked to increases in metabolic rate, lipid use and length growth, especially of the caudal peduncle ([Hoar, 1988](#); [Winans and Nishioka, 1987](#)). Also, feeding hatchery fish high-energy pelleted diets may mitigate the degree of drop in condition factor traditionally observed in wild fish. Condition factor differed between rearing units in November and January. Lower condition factor observed in raceway-reared fish in January may reflect differences in feeding regime experienced through December.

During the last 2 months of the study, growth rate of the fish in the pond was 2.5-fold greater than that of the fish in the raceways. Rapid growth may have resulted from a greater proportion of the population reaching a size or growth threshold for smolting than those in the raceways. This difference may be explained by a number of aspects of the semi-natural environment: food type available, relative stress levels, and differences in rearing density. Regardless, growth rate appears to be a better predictor of migratory performance than size, particularly in chinook salmon ([Beckman et al., 1998](#)). Rapid growth during smolting of chinook salmon has been associated with increased smolt performance and survival to adult ([Beckman et al., 1999](#)).

Differences in growth rate resulted in size differences at the end of the experiment, with pond-reared fish being larger. Size was correlated with increased SW tolerance and may partly explain differences between pond- and raceway-reared fish. However, size was not correlated with GH in November or January, IGF-I in January or gill Na^+, K^+ -ATPase activity in November or January. Other factors associated with rearing conditions are therefore likely to influence physiological parameters of smolt development.

A density difference is likely to have affected the smolt-related parameters measured in this study. Fish density was nearly threefold lower in the pond than in the raceways (99 and 299 fish m^{-3} , respectively). High densities (e.g. raceways relative to the pond of this study) have been shown to suppress smolting of hatchery-reared coho salmon ([Schreck et al., 1985](#)). Similar assertions have been made for brown trout ([Sundell et al., 1998](#)).

Density is unlikely to explain all differences seen in this study. During the same year, fish reared in Coleman NFH raceway 8 were reared at a significantly lower density (55 fish m^{-3}) than the pond. Interestingly however, fish reared in the pond had higher levels of gill Na^+, K^+ -ATPase activity, GH, IGF-I, and better SW tolerance than fish in raceway 8 though they were the same size (Zydlewski, unpublished).

One confounding factor in comparing densities in the present study is the presence of structures in the pond. These structures may in fact increase the effective density by

attracting fish to certain areas. Additionally, negative density-dependent effects are often the result of increased ammonia levels and in this study the pond was fed with water that had already passed through raceways. Therefore, direct comparisons of density effects may be misleading.

Food type and delivery may have played a large role in growth and smolt development of fish reared in the pond. Although both rearing environments were provided with similar rations of standard hatchery food, fish in the pond may have had other food sources available to them, e.g. aquatic invertebrates. This is likely as raceway-reared fish in December were fed at a significantly higher rate but pond-reared fish still grew more quickly from December to January. The pond had aquatic vegetation and was not cleaned during the study. Conversely, raceways had no aquatic vegetation and were cleaned on a weekly basis. Also, structures, surface disruption, and potential reduction in antagonistic behavior during feeding may have allowed different feeding opportunities for fish in the pond than those reared in the raceways.

Differences in the extent of visual and physical impacts on fish during rearing are likely to be partly responsible for the significant differences in the parameters reported here. Fish reared in raceways had significantly more contact time with humans. Weekly cleaning of raceways included draining the water and physically removing waste products. Daily feeding was conducted in a visible manner. Other intermittent monitoring of raceway water quality further increased human contact. In contrast, human interaction with fish in the pond was minimal. There was no cleaning activity in the pond and feed was distributed from behind a screen. Additionally, water was sprayed over the surface of the pond, disrupting the surface and minimizing potential visual disruption of fish by humans. As a result, raceway rearing provided a more stressful condition than that in the pond. Differences in hatchery rearing conditions such as those of the present study have been shown to affect physiological parameters of smolting, e.g. cortisol dynamics (Shrimpton et al., 1994b).

Based on studies of wild salmonids, characteristics of the pond-reared fish are likely to reflect those of wild-reared fish. Since the CA Central Valley steelhead stock is listed as threatened, wild fish could not be sampled for direct comparison of physiological parameters associated with smolting. However, differences observed between fish reared in raceways and those reared in the pond are consistent with differences reported between hatchery and wild brown trout, coho salmon, and Atlantic salmon. Wild brown trout have higher levels of gill Na^+, K^+ -ATPase activity, GH, and better SW tolerance than hatchery fish (Sundell et al., 1998). Wild coho salmon generally have higher levels of gill Na^+, K^+ -ATPase activity and better SW tolerance than hatchery fish (Shrimpton et al., 1994a,b). Also, seasonal changes in condition factor are more variable in wild fish than in hatchery fish (Shrimpton et al., 1994b) as are seen in the pond fish of this study. In general, for both brown trout and coho salmon, wild fish were smaller than hatchery fish, unlike the current study or the study on Atlantic salmon (McDonald et al., 1998). Yearling wild Atlantic salmon are generally similar in size and condition as their hatchery counterparts. However, larger hatchery fish generally have been reported to have poor smolt development (Shrimpton et al., 1994a,b; McCormick and Björnsson, 1994; Sundell et al., 1998). In this study, however, larger fish had better smolt development, particularly GH levels, SW tolerance, and growth rates.

5. Conclusions

Hatchery conditions can be manipulated to enhance smolt development to produce more wild-like smolts. Rearing parameters that may accomplish this include enhanced feeding opportunities, low human interaction, low fish density, and structure. While this study has demonstrated physiological differences between fish in different rearing environments, relative importance of these factors cannot be evaluated here. The effects of specific rearing practices on smolting need to be evaluated further. Future studies need to be conducted to determine how individual changes in rearing practices (separate manipulations of feeding opportunities, fish density, and structure type) affect smolt development. Specifically, how changing hatchery rearing practices may correspond with changing smolt development. Changes within the hatchery to produce appropriate (wild-like) smolt development will likely increase migratory survival and enhance adult returns.

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