EFFECTS OF TEMPERATURE ON SURVIVAL AND GROWTH OF WESTSLOPE CUTTHROAT TROUT AND RAINBOW TROUT: IMPLICATIONS FOR CONSERVATION AND RESTORATION

by

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ABSTRACT

Westslope cutthroat trout *Oncorhynchus clarkii lewisi* have declined throughout their native range in the Northern Rockies and were considered for listing under the federal Endangered Species Act. Water temperature is widely regarded as playing a key role in determining their persistence, but specific lethal levels and thermal optima for this cutthroat trout subspecies had not been precisely defined. This laboratory study used the acclimated chronic exposure method to determine tolerances and thermal optima of westslope cutthroat trout and rainbow trout *Oncorhynchus mykiss*, a potential nonnative competitor now occupying much of the former range of westslope cutthroat trout. Rainbow trout had a distinct survival advantage over westslope cutthroat trout at warmer temperatures. The ultimate upper incipient lethal temperature (temperature at which 50% of the population survives for 60-d) of rainbow trout (24.3°C; 95% CI, 24.0 - 24.7°C) was 4.7°C higher than that of westslope cutthroat trout (19.6°C; 95% CI, 19.1 – 19.9°C). In contrast, the optimum growth temperature for westslope cutthroat trout (13.6°C; 95% CI, 10.3 - 17.0°C) over the 60-d test period was very similar to that of rainbow trout (13.1°C; 95% CI, 6.8 - 18.2°C), although rainbow trout grew better over a wider range and at higher temperatures than did westslope cutthroat trout. The upper lethal and optimum growth temperatures for westslope cutthroat trout are in the lower range among most salmonids. The higher upper temperature tolerance of rainbow trout and its greater ability for growth at warmer temperatures may account for its increased occurrence at lower elevations than cutthroat trout. Water quality standards setting maximum daily temperatures from 13-15°C, near the optimum growth temperature, would ensure suitable thermal habitat to maintain the persistence of westslope cutthroat trout populations. In addition, survival and growth parameters indicated in this study can be used with stream temperature modeling to predict suitable habitat for westslope cutthroat trout, as they may be particularly susceptible to increases in stream temperature associated with climate change. Such predictions of habitat suitability will be vital in prioritizing conservation efforts with respect to reintroduction and translocation of westslope cutthroat trout.

INTRODUCTION

Historically, westslope cutthroat trout *Oncorhynchus clarkii lewisi* ranged widely over western Montana, Idaho, northwestern Wyoming, portions of eastern Oregon and Washington, and southern Alberta (Behnke 1992; Shepard et al. 2003). As with many other cutthroat and native trout species in western North America (e.g., Lahontan cutthroat trout *O. c. henshawi*, Bonneville cutthroat trout *O. c. utah*, bull trout *Salvelinus confluentus*), westslope cutthroat trout now persist in only a small portion of their native range. The species is listed as a “species of special concern” in Montana (MNHP 2004), and was recently evaluated for listing as a federally threatened species under the Endangered Species Act (USFWS 2003). Westslope cutthroat trout currently occupy about 59% of their presumed historical range (Shepard et al. 2003) with persisting populations mainly confined to colder, headwater portions of streams that they previously occupied entirely (Hanzel 1959). Protection of these isolated, remnant populations is crucial because of presumed unique adaptations to local habitats (Liknes and Graham 1988). Individuals from remaining populations could be used for propagation, reintroductions, and translocations (Sloat et al. 2005).

Leading threats to the persistence of remaining westslope cutthroat trout populations are habitat degradation, hybridization with nonnatives such as rainbow trout *Oncorhynchus mykiss*, displacement by nonnatives, and competition with nonnatives (Hanzel 1959; Liknes and Graham 1988; Behnke 1992; Shepard et al. 1997). Effects of habitat degradation include increased water temperature in streams and rivers resulting from timber harvesting, water diversions, and loss of riparian areas (Brown and Krygier 1970; Cushing and Allan 2001; Poole and Berman 2001). Non-introgressed westslope cutthroat populations are now rare (Allendorf and Leary 1988; Allendorf et al. 2004) occurring in only 10% of currently occupied habitats (Shepard et al. 2003). Much of their historical distribution is now occupied by rainbow trout, rainbow trout x cutthroat trout hybrids, brook trout *Salvelinus fontinalis*, and brown trout *Salmo trutta* (Hanzel 1959), leading to the current situation of westslope cutthroat trout restricted primarily to higher elevation, steeper gradient streams (Griffith 1988; Bozek and Hubert 1992; Dunham et al. 1999; Shepard 2004) often associated with colder water temperatures, a pattern common to many now-rare native salmonids in western North America (Behnke 1992). Water temperature is considered a key element influencing the persistence of native cutthroat trout populations because of their current confinement to headwater reaches (Paul and Post 2001).

Increasing water temperatures may influence the persistence of westslope cutthroat trout populations by affecting their distribution, survival, and growth. Water temperature affects the geographic distribution of fishes (Brett 1956; Welch et al. 1998) with increasing temperatures leading to a shift toward warmwater adapted species replacing those species adapted to cold water in lower elevations (Keleher and Rahel 1996; Cushing and Allan 2001). Because most fish lack a means of maintaining an independent body temperature, increased water temperatures can be lethal, and sublethal temperatures can alter metabolism, growth (Brett 1956; Medvick 1979; Thomas et al. 1986; Mallet et al. 1999; Kestemont and Baras 2001), and competitive interactions (De Staso and Rahel 1994; Taniguchi et al.1998).

Mortality associated with extreme temperatures is a result of the combination of temperature and exposure time (Brett 1952). The physiological mechanisms causing death at high temperatures are not entirely understood but appear to be related to underlying factors such as the increase in oxygen consumption by fishes and corresponding decrease in dissolved oxygen solubility at high temperatures, and the decreased capacity to sustain vital energy reserves and electrolyte concentrations. Ultimately, death at high temperatures is caused by osmoregulatory failure (Wedemeyer 1996).

Increased water temperature also influences fish survival indirectly by controlling metabolic function, resulting in the sensitivity of growth to temperature (McCullough et al. 2001). The response of fish growth to temperature generally follows a pattern whereby growth and food consumption increase with rising temperature to an optimum level, followed by a decrease with increasing temperature as metabolism exceeds energy intake (Hokanson et al.1977; Brett 1979; Brett et al. 1982; Ojanguren et al. 2001; McCullough et al. 2001; Selong et al. 2001; Meeuwig et al. 2004). The optimum growth temperature often coincides with physiological optimums such as metabolic scope, maximum swimming speed, and maximum oxygen debt tolerance (Brett et al. 1969) as well as behavioral optimums such as temperature preference and competitive ability (Jobling 1981; Cunjak and Green 1986; Sauter et al. 2001). Fish competing within their optimal range typically have a distinct advantage over species outside their optimal range (Cunjak and Green 1986).

Water temperature is thought to play a significant role in the competitive interactions between fishes as differences in the physiological optimum and tolerance of species can have large ecological effects (De Staso and Rahel 1994; Taniguchi et al 1998; Reese and Harvey 2002). For example, brook trout were competitively dominant over Colorado River cutthroat trout *Oncorhynchus clarkii pleuriticus* at 20°C, but neither species showed clear dominance at 10°C, indicating that brook trout may have a competitive advantage at warmer water temperatures (De Staso and Rahel 1994). Similarly, when brook and bull trout are in sympatry, brook trout suppress feeding and growth of bull trout at temperatures above 14°C, again indicating brook trout may have a competitive advantage at water temperatures above this level (T. E. McMahon, Montana State University, unpublished data).

In most coldwater drainages in western North America, remaining cutthroat trout subspecies occupy isolated headwaters whereas lower elevation streams and rivers are now occupied by mixed nonnative assemblages comprised of rainbow trout, brook trout, and brown trout. In addition, many remaining populations of native cutthroat trout are heavily introgressed with rainbow trout, forming the dominant assemblage structure at moderate elevation sites (Carmichael et al. 1993; Kruse et al. 2000; Hitt et al. 2003). Paul and Post (2001) posed the 'elevation refuge hypothesis' for native salmonids now confined to headwaters, contending that cutthroat trout have a competitive advantage in these colder, steeper gradient sites. Temperature is thought to be the major underlying factor for these elevational distributional differences among native and nonnative salmonids, but temperature requirements of many native species, and for the now widely distributed cutthroat x rainbow trout hybrids, are unknown.

Thermal protection standards are needed to protect and restore remaining populations of native, cold-water species, particularly in light of climate change (McCullough 1999) because even small shifts in temperature can have significant effects on salmonid distribution (Welch et al. 1998). It is necessary to determine the optimal physiological ranges and the thermal tolerance of fishes to develop water quality standards. Species within the family Salmonidae have among the lowest thermal tolerance of temperate-zone fishes, although variation exists among species with respect to the maximum temperature for survival and optimum growth temperature (Brett 1952; Hokanson et al. 1977; Selong et al. 2001). The thermal requirements of westslope cutthroat trout are largely unknown because the upper lethal and optimum growth temperatures have not been previously determined. Information on the thermal requirements of westslope cutthroat trout may aid in determining thermal protection standards for remaining populations, defining suitable habitat for conservation and restoration efforts, and predicting presence or absence in areas where the distribution of westslope cutthroat trout is currently unknown.

My main objective was to characterize the thermal requirements of westslope cutthroat trout, specifically the ultimate upper incipient lethal temperature (UUILT, temperature at which 50% of population survives indefinitely; Fry 1947; Fry 1971) and optimum growth temperature (temperature corresponding to peak growth), and to compare these survival and growth optima to those of a main nonnative competitor, rainbow trout. Additional objectives were to characterize the thermal requirements of first-generation westslope cutthroat x rainbow trout hybrids, and determine the influence of water temperature on competitive interactions of westslope cutthroat trout with rainbow trout and brook trout.

I hypothesized that westslope cutthroat trout would have a lower ultimate upper incipient lethal and optimum growth temperature than rainbow trout. If correct, this could explain the increased distribution of rainbow trout at warmer, lower elevations whereas westslope cutthroat trout tend to inhabit cooler, higher elevation areas. I also hypothesized that hybrids would have an ultimate upper incipient lethal and optimum growth temperature intermediate between westslope cutthroat and rainbow trout. If correct, this could help explain the increased success of hybrids in nature where both westslope cutthroat and rainbow trout occur. Additionally, growth of westslope cutthroat trout would be suppressed when in sympatry with rainbow trout and when in sympatry with brook trout. The optimum growth temperature for westslope cutthroat trout would also be at lower temperatures in sympatry than in allopatry. If correct, this could explain the competitive advantage that nonnative trout are thought to have over westslope cutthroat trout at warmer temperatures.

METHODS

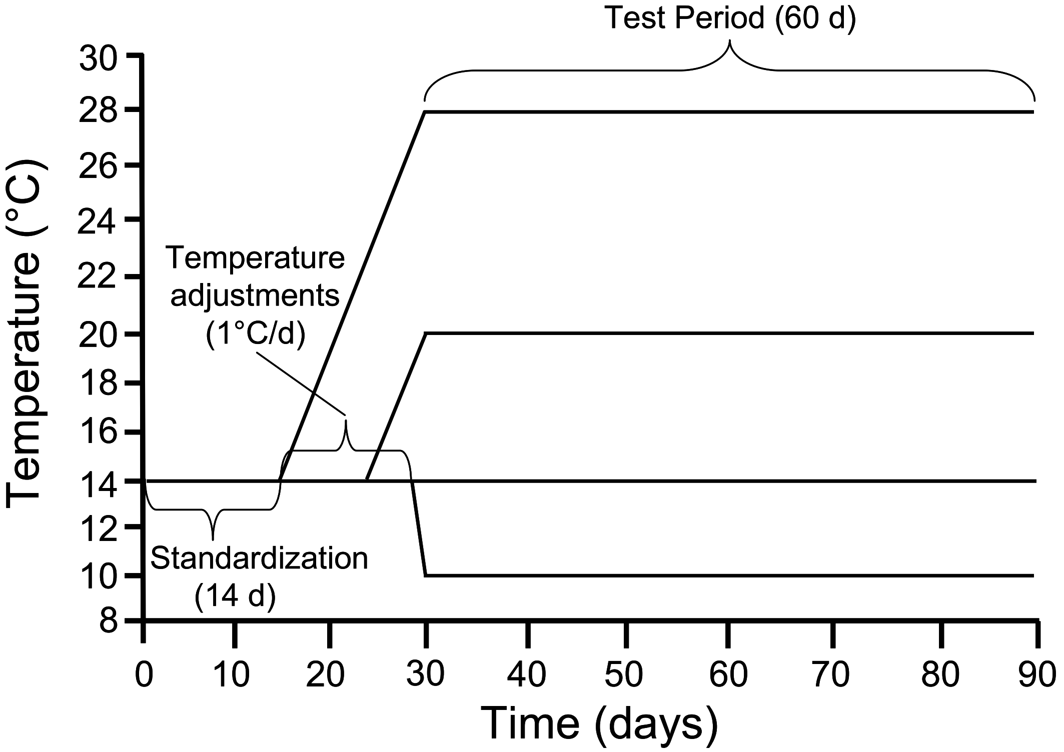
A thermal test facility housed at the U.S. Fish and Wildlife Service Bozeman Fish Technology Center (BFTC), used previously to determine the optimum and upper lethal temperatures of bull trout (Selong et al. 2001), was employed in this study to simultaneously assess fish growth and survival at different temperatures for prolonged periods. Westslope cutthroat trout used in experiments were obtained either as eggs or age-0 fish from a wild stock at Rogers Lake, Montana, or as age-0 fish from a wild stock maintained at the Westslope Trout Company, a private fish hatchery near Ronan, Montana. Rogers Lake westslope cutthroat trout were derived from the MO12 broodstock maintained at Washoe Park State Fish Hatchery near Anaconda, Montana. All rainbow trout were obtained as eggs from the Fish Lake strain maintained at the U.S. Fish and Wildlife Service Ennis National Fish Hatchery (ENFH) near Ennis, Montana. Westslope cutthroat x rainbow trout hybrids were produced by fertilizing roughly 11,000 westslope cutthroat trout eggs from Rogers Lake with rainbow trout milt from the Fish Lake strain at ENFH. Brook trout were obtained as eggs from the Crystal Lakes Hatchery, a private fish hatchery near Eureka, Montana. All eggs and fry were transported to the BFTC and held until testing. Fish were reared near 12°C and fed to excess daily with an automatic belt feeder.

The general test protocol for all experiments was similar to that outlined in Selong et al. (2001). A flow-through thermal testing system provided constant water flow, high dissolved oxygen concentrations, and metabolite flushing. Water supplied from cold (8°C) and warm (22°C) springs, and water heated with three 40,000-BTU water heaters, was mixed to achieve test temperatures from 8°C to 30°C. Water from the spring sources was aerated using pure oxygen and a diffusing stone, passed through a de-gassing column, and mixed in 12 separate head tanks. From the head tanks, water was supplied to 36, 75-L aluminum test tanks (120 x 35 x 25 cm) at a flow rate of 3 L/min. All connecting pipes and tank surfaces were covered with foam insulation to reduce atmospheric heat loss.

To simultaneously assess long-term growth and survival over varying temperatures, I employed the acclimated chronic exposure (ACE) thermal test method. The ACE method was developed as an alternative to standard short term upper lethal temperature methods (critical thermal maxima or CTM; incipient lethal temperature or ILT) that employ rapid temperature changes (Zale 1984; Selong et al. 2001). The ACE method measures growth and survival over prolonged time periods (60 d), incorporating more natural temperature changes and exposure times, thus allowing assessment of growth and survival over varying temperatures (Selong et al. 2001). In addition, the ACE method allows calculation of LD_{50} (median resistance time; lethal dose to 50% of the population) at various temperatures to allow prediction of median survival times based on temperature.

In accord with the ACE method, 50 fish were haphazardly selected, placed in each of 36 test tanks, and acclimatized to 14°C water for at least 14 days (standardization period) in each experiment. Fish were distributed so that sizes in each tank were similar. Three replicates were tested at each temperature. After the standardization period, water temperature in each tank was raised or lowered 1.0°C per day until the desired test temperature was reached (Figure 1).

Figure 1. Acclimated chronic exposure schedule.



Temperature adjustments were staggered such that all tanks reached final treatment temperatures on the same day. After final treatment temperatures were reached (one to sixteen days depending on treatment temperature), fish were held at a constant temperature (test period) for 60 days. On day 1 and day 60 of the test period, fish in each tank were counted and weighed in bulk to compute an initial and final weight per fish in each tank. Additionally, on day 60, all remaining fish from each tank were sacrificed and length and weight data recorded for each individual.

Fish were fed daily using an automatic belt feeder that supplied a constant feed ration over a 12-h period from about 0800 to 2000 hours. Fish were fed amounts in excess of satiation, as indicated by the presence of excess feed in tanks after a 12-h feeding cycle. Tanks were cleaned daily and mortalities removed, weighed (g), and measured (total length in mm). Temperature, dissolved oxygen concentration, and percent gas saturation were measured daily in each head tank using a YSI 55 meter and adjustments in the amount of pure oxygen diffused into the system were made to ensure adequate dissolved oxygen concentration in each tank. Dissolved oxygen varied from 6.1 to 12.2 mg/L over the course of the entire study with a mean concentration of 8.3 mg/L. Total gas saturation varied from 75.3 to 103.0% over the course of the study with a mean percent saturation of 84.7%. Oxygen saturation varied from 84.0 to 122.7% over the course of the study with a mean percent saturation of 100.6% (Table 1). Oxygen saturation was calculated for each experiment based on dissolved oxygen concentration and temperature measured daily (Appendix A). Dissolved oxygen and oxygen saturation levels throughout the study were within optimal levels for fishes (> 5 mg/L DO, > 80% O_2 saturation; Piper et al. 1982).

Table 1. Dissolved oxygen concentration, total gas saturation, and oxygen saturation in each experiment. Mean values and range of minimum and maximum values based on all tanks in each experiment.

		Dissolved oxygen concentration (mg/L)		Total gas saturation (%)		Oxygen saturation (%)	
Species	Experiment	Mean	Varied from	Mean	Varied from	Mean	Varied from
WCT	1	8.3	6.5 to 11.4	84.4	77.1 to 97.6	99.9	88.8 to 114.9
	2	7.5	6.5 to 9.2	80.2	75.3 to 87.5	95.4	84.0 to 104.1
	3	8.8	6.5 to 11.2	88.5	77.0 to 101.0	105.3	91.7 to 119.8
RBT	4	7.8	6.1 to 10.5	81.7	77.0 to 90.0	97.2	85.7 to 119.8
	5	8.9	6.6 to 12.2	88.9	78.0 to 103.0	105.1	92.7 to 122.7

Temperature was recorded hourly with an Onset Optic StowAway® Temp logger placed in each test tank. All thermographs were calibrated after the final experiment and a correction factor was used to determine mean temperatures experienced by fish in each tank over the 60-d test period. Thermographs were calibrated by placing them for one hour in a tank supplied with water of the equivalent test temperature. The temperature was recorded every five minutes by the thermograph and also using a Fisher Scientific calibrated total immersion mercury thermometer. Thermograph readings were then compared to temperatures recorded with the mercury thermometer to determine a correction factor for each thermograph. Mean daily temperature fluctuation throughout the study varied from 0.30 to 1.1°C with maximum daily fluctuations varying from 1.1 to 3.7°C (Table 2). Temperature fluctuations greater than 1°C were rare and were associated with low water flow during experiments that caused fluctuations in the amount of cold and warm water mixing in the head tanks, thus creating temperature fluctuations. All water supplied to the test facility originates in cold and warm springs north of the Bozeman Fish Technology Center. Unexpected changes in the supply of this water to the thermal test system caused substantial flow variation and subsequent atypical daily temperature fluctuations.

Table 2. Mean and range of minimum and maximum daily temperature fluctuations in each experiment.

		Daily temperature fluctuation (°C)	
Species	Experiment	Mean	Varied from
WCT	1	0.4	0.0 to 2.8
	2	0.3	0.0 to 1.1
	3	0.8	0.0 to 2.4
RBT	4	0.3	0.0 to 2.7
	5	1.1	0.0 to 3.7

Natural light entering the system from outside was supplemented with overhead halogen lighting. Supplemental lighting correlated with the natural photoperiod and was monitored throughout the duration of the project with an Onset StowAway® Light Intensity Logger. Photoperiod over the duration of the experiments averaged about 13-h light and 11-h dark.

Westslope cutthroat trout survival and growth

Three experiments were conducted to determine the survival and growth of westslope cutthroat trout at temperatures from 8°C to 30°C. In experiment one, survival and growth of westslope cutthroat trout was assessed at 8, 10, 12, 14, 16, 18, 20, 22, 24, 26, 28, and 30°C. To refine the upper lethal temperature for westslope cutthroat trout, survival and growth were tested in experiment two at water temperatures of 13, 15, 20, 21, 22, and 23°C. Only six temperatures were tested in the second experiment because of insufficient numbers of test fish. Westslope cutthroat trout used in the first two experiments were from the Rogers Lake strain.

In experiments one and two, a fixed ration was provided that was determined afterwards to be less than satiation consumption at some test temperatures. A reduced ration could lead to an optimum growth temperature lower than expected compared to feeding in excess of satiation (Brett et al. 1969; Brett 1971). Therefore, an additional experiment (number three) was conducted to determine growth of westslope cutthroat trout under feeding conditions known to be in excess of satiation. Survival and growth were tested following the general test protocol at 8, 12, 14, 16, 20, and 24°C; six temperatures were tested in the final westslope cutthroat trout experiment because of insufficient numbers of test fish. Westslope cutthroat trout used in this experiment were obtained from the Westslope Trout Company (Ronan, Montana) because of unavailability of fish from the Rogers Lake strain. Feed consumption was measured to ensure rations in excess of satiation were provided. Uneaten feed and feces were removed from each tank the morning following a 12-h feeding period, once a week. Feed and feces were separated using a 2-mm sieve, and remaining uneaten food material was dried overnight at 100°C, weighed, and subtracted from the total amount fed to determine daily food consumption in grams. A control with known quantities of feed and feces was conducted in each tank and a correction factor determined to correct for moisture loss. Feed levels were then adjusted weekly to provide feed availability in excess of consumption at each test temperature. Growth results are reported only for the final experiment at satiation ration because a reduced ration was fed in the first two experiments, whereas survival results were combined over all three experiments. Feed differed among experiments because of availability; FinStater 2014 was used in experiments one and two, and Cutthroat Trout Grower was used in experiment three. Ingredient composition and energy of feeds differed slightly (Appendix B).

Rainbow trout survival and growth

Two experiments were conducted to determine survival and growth of rainbow trout at temperatures from 8°C to 28°C. In the initial rainbow trout experiment (number four), survival and growth were assessed at 8, 10, 12, 14, 16, 18, 20, 22, 24, 26, and 28°C. It was later determined that the fixed ration provided in this experiment was less than satiation consumption at some test temperatures. Therefore, an additional experiment (number five) was conducted to determine growth of rainbow trout at satiation rations following the feeding protocol described above. Survival and growth were tested simultaneously with the final cutthroat experiment (in allopatry) at 8, 10, 14, 16, 20, and 24°C. Rainbow trout used in both experiments were from the Fish Lake strain from Ennis National Fish Hatchery. Feed differed among experiments because of availability; Silvercup Trout Food was used in experiment four whereas Cutthroat Trout Grower was used in experiment five, with ingredient composition and energy of feeds differing slightly (Appendix B).

Westslope cutthroat x rainbow trout hybrid survival and growth

No experiments with hybrids were possible because of extremely low hatching success. Within two days of fertilization, roughly one-third of all hybrid eggs had died. Mortality continued at a high rate through hatching and after hatching only a few hundred hybrids remained. These numbers were insufficient to run a full experiment thus, the upper lethal and optimum growth temperatures of hybrids could not be determined. Poor survival of first-generation westslope cutthroat x rainbow trout hybrids has been reported in other studies of cutthroat x rainbow trout hatching success (Leary et al. 1995; Allendorf et al. 2004).

Influence of temperature on competition

Experiments to determine the influence of water temperature on competition between brook and rainbow trout with westslope cutthroat trout were not conducted because of insufficient numbers of fish to conduct experiments. Westslope cutthroat trout obtained for use in these experiments (age-0 fish from the Westslope Trout Company) suffered from a chronic *Hexamita salmonis* infection that persisted despite multiple and varied prophylactic treatments. *Hexamita salmonis* is a small, mobile protozoan with eight flagella found in the intestines of salmonids. Infection can result in poor growth and increased mortality, especially in small fish. The recommended treatment for *Hexamita salmonis* is 3% Epsom salt concentration added to the food and given for three days (Warren 1991; Winton 2001). Westslope cutthroat trout were first diagnosed with *Hexamita salmonis* by the U.S. Fish and Wildlife Service Fish Health Laboratory in Bozeman, Montana, after mortalities began to increase in February 2004. Over the course of the next six months, fish were given the recommended treatment five times, which suppressed the disease for a short period, followed by another outbreak. Finally, fish were given a bath treatment with the antibiotic Metronidazole for three days in July 2004. Mortality after treatment declined for several days, before increasing to pre-treatment levels. In addition, fish were diagnosed with *Ichthyobodo* (*Costia)* twice during this time, which likely contributed to ongoing mortality. *Ichthyobodo* is a small (5 x 12 micrometers) protozoan that infects the gills and skin of fish, causing listlessness, decline in appetite, and increased mortality (Warren 1991; Winton 2001). *Ichthyobodo* was first diagnosed from routine skin scrapes in April and July of 2004. After both diagnoses, fish were given the recommended treatment of 150-200 parts per million formalin bath for one hour. In August 2004, it was determined that these westslope cutthroat trout obtained for use in competition experiments were too compromised by disease to be used in future thermal studies and the fish were sacrificed to prevent disease spread to other tanks at the Bozeman Fish Technology Center.

Brook trout obtained for use in competition experiments also began showing increased mortality in October 2004. Several routine skin scrapes revealed no external parasites. A sample taken to the USFWS Fish Health Lab revealed swollen gills, minor fin erosion, and small numbers of bacteria on fin edges, but no sign of any serious disease infection. However, brook trout mortality continued to increase, greatly reducing numbers of available fish. It was deemed that because of high mortality and potentially undiagnosed disease, fish health was likely compromised and competition experiments with brook trout were not initiated.

DATA ANALYSES

Survival

Temporal patterns in survival were assessed by plotting mean daily survival rates over the 60-d test period (Dickerson and Vinyard 1999; Johnstone and Rahel 2003). Daily survival at temperatures tested in more than one experiment was averaged among experiments to determine the mean daily survival at each temperature. The ultimate upper incipient lethal temperature (UUILT) was determined by plotting survival (%) of a species in all experiments against temperature using logistic regression (Johnstone and Rahel 2003). A regression line and its confidence interval (95%) were constructed and resulting parameter estimates used to determine the 60-d UUILT (LD_{50} at 60 days). Upper confidence intervals were bound at 100% survival where necessary. The LD_{50} (median resistance time; time to 50% survival) over shorter time periods (1, 3, 7, 15, 22, 30, 37, 45, and 52 d) were derived in a similar fashion and plotted against temperature using an exponential decay regression in Sigma Plot 8.0 (Sigma Plot 2002). The 95% confidence intervals of the 60-d UUILT data were compared among experiments to determine whether different fish sizes in each experiment affected survival.

Significant differences in survival between westslope cutthroat and rainbow trout were determined by non-overlapping 95% confidence intervals of the survival (%) against temperature data. Differences in survival between temperatures within a species were compared using a one-way analysis of variance (ANOVA). Survival (%) was arcsine transformed to normalize the data and was analyzed using a General Linear Model (GLM) ANOVA and Tukey's multiple comparison test in Minitab 14.0 (Minitab 2000) with temperature as the main effect and significance level $\alpha \leq 0.05$.

Growth

Using data from experiments three and five, relative growth rate (G, expressed as %) was calculated according to the formula: $G = [(Y_2 - Y_1) / (Y_1 * t)] * 100$, where Y_1 is the initial mean weight and Y_2 the final mean weight in grams of fish in each tank, and t is the time period of the experiment (Ricker 1979). Relative growth rate was used because it accounts for the initial size of fish, and allowed comparison of growth among fish of different initial sizes. Growth and temperature data were used to develop growth curves for each species using quadratic regression in Sigma Plot 8.0. A regression line and its confidence interval (95%) were constructed and the resulting parameter estimates used to determine the optimum growth temperature for each species. The optimal growth range for each species was determined as the range encompassing the peak growth between the lower and upper 95% confidence interval of the regression line (Selong et al. 2001). Significant differences in growth between the two species were indicated by non-overlapping 95% confidence intervals of growth data. Differences between temperatures within a species were compared using a one-way GLM ANOVA and Tukey’s multiple comparison test with temperature as the main effect and significance level of $\alpha \leq 0.05$.

Feed Consumption and Conversion Efficiency

Feed consumption and conversion efficiency were determined for both species using data from the final experiments (three and five). Relative consumption (C, expressed as % body weight) was calculated according to the formula: C = consumption (g) per fish / body weight (g) per fish * 100 (Galarowicz and Wahl 2003). Relative consumption was determined on day 30 and day 60 and values averaged to derive an average daily relative consumption per fish over the 60-d experiment. Feed conversion efficiency (FE) was calculated according to the formula: FE = daily growth (g) per fish / daily feed consumed (g) per fish (Galarowicz and Wahl 2003; Barrows and Hardy 2001; Selong et al. 2001). Daily feed consumption and feed conversion efficiency were compared among temperatures and between species with a GLM two-way ANOVA and Tukey’s multiple comparison test with species, temperature, and species*temperature as the main effects. Significance level for all statistical testing was $\alpha \leq 0.05$.

Relative Condition

Relative condition of both species was determined at 8, 12, 14, and 16°C from length and weight data collected at the end of the final experiments (three and five). A random sample of 15 fish from each species and temperature group (except 12 rainbow trout at 12°C) from 100 to 180 mm total length (TL) were selected without replacement to obtain a representative sample from both species. Total length and weight data for each species were then $\log_{10}$ transformed and fitted to a linear regression model (Table 3). The resulting weight-length regression was used to calculate a relative standard weight (Ws′) (Anderson and Neumann 1996) for fish of each species from 100 to 180 mm TL that were not used in the random sample. A relative condition factor was then calculated for each fish using the formula: $Kn = (W / Ws') * 100$, where W is the weight (g) of an individual fish and Ws′ is the length-specific relative standard weight (Le Cren 1951; Anderson and Neumann 1996). Relative condition factor was compared among temperatures for each species with a GLM ANOVA and Tukey's multiple comparison test with temperature as the main effect and significance level of $\alpha \leq 0.05$.

All length and weight data for each species at 8, 12, 14, and 16°C were $\log_{10}$ transformed and fitted to a linear regression model (Table 3). The resulting equations for each temperature were then used to compare the predicted weight of an average size fish (westslope cutthroat trout, 123 mm; rainbow trout, 193 mm) across temperatures.

Table 3. Weight-length regression equations for westslope cutthroat (WCT) and rainbow trout (RBT) at 8, 12, 14, and 16°C, and all temperatures combined.

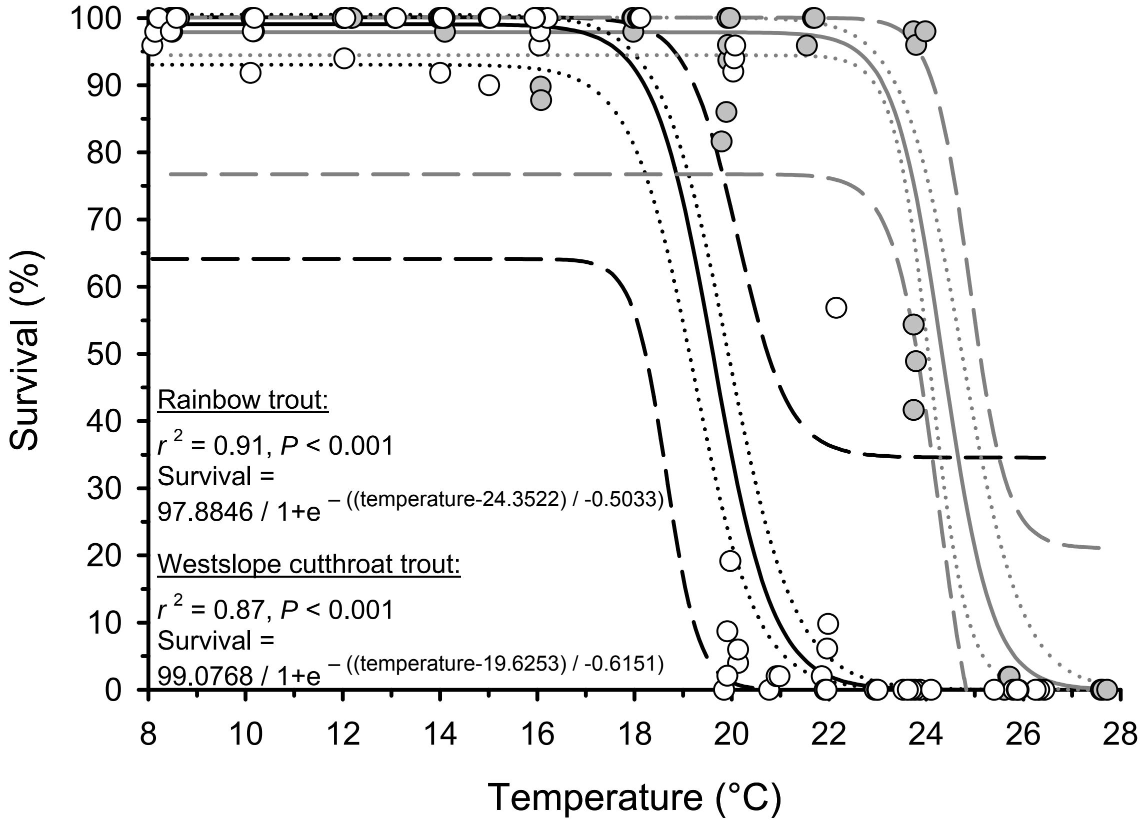
Species	Temperature	Weight-length regression equation
WCT	All	Log_{10} weight = -5.9024 + (3.4305 * Log_{10} length)
	8°C	Log_{10} weight = -5.5895 + (3.2727 * Log_{10} length)
	12°C	Log_{10} weight = -5.4756 + (3.2212 * Log_{10} length)
	14°C	Log_{10} weight = -5.1468 + (3.0717 * Log_{10} length)
	16°C	Log_{10} weight = -5.5413 + (3.2578 * Log_{10} length)
RBT	All	Log_{10} weight = -5.2212 + (3.1180 * Log_{10} length)
	8°C	Log_{10} weight = -5.1810 + (3.1057 * Log_{10} length)
	12°C	Log_{10} weight = -5.5507 + (3.2654 * Log_{10} length)
	14°C	Log_{10} weight = -5.3370 + (3.1715 * Log_{10} length)
	16°C	Log_{10} weight = -5.3859 + (3.1911 * Log_{10} length)

RESULTS

Survival

Survival of juvenile westslope cutthroat trout and rainbow trout was greater than 82% over the range of test temperatures from 8°C to 18°C (Figure 2). Survival of both species declined significantly at higher temperatures (ANOVA; westslope cutthroat trout $F = 43.74$, $P < 0.001$; rainbow trout $F = 42.84$, $P < 0.001$) although rainbow trout showed much greater tolerance to warmer temperatures than westslope cutthroat trout. Westslope cutthroat trout survival declined significantly at temperatures ≥ 20°C (Tukey's; $P < 0.001$), whereas rainbow trout survival declined significantly at 26°C and above ($P < 0.001$). Plots of survival by temperature also showed that survival of westslope cutthroat trout was significantly lower than that of rainbow trout from 20°C to 24°C, as indicated by non-overlapping 95% confidence intervals of the data (Figure 2). The predicted ultimate upper incipient lethal temperature for westslope cutthroat trout (19.6°C; 95% CI, 19.1 - 19.9°C) was 4.7°C lower than for rainbow trout (24.3°C; 95% CI, 24.0 - 24.7°C) (Figure 2). Variation in survival relative to temperature did exist among experiments. Variation in survival was very low (< 10% coefficient of variation (CV); Table 4) at temperatures from 8°C to 18°C but increased markedly (37.3 to 177.2% CV) as temperatures approached the upper limit for each species.

Figure 2. Survival (%) of age-1 westslope cutthroat trout (white circles, black line) and rainbow trout (grey circles, grey line) over 60 days in relation to temperature. Each point represents the survival in an individual tank at a given temperature. Dotted lines indicate the 95% confidence interval of the regression line and dashed lines indicate the 95% confidence interval of the data.



Survival of both species was strongly related to exposure time, but rainbow trout exhibited much greater tolerance to prolonged exposure to temperatures ≥ 20°C (Figure 3). At 20°C, westslope cutthroat trout survival was high (> 90%) for 30 days, followed by a sharp decline, and then stabilized until the end of the experiment. In contrast, rainbow trout survival at 20°C remained high (> 82%) throughout the experiment. Similarly, at 22°C and 24°C westslope cutthroat trout survival was initially high (>80%) for about one week before declining sharply, whereas rainbow trout survival at these temperatures remained high (> 70%) throughout the experiment (Figures 3 and 4). A marked difference in survival between the species occurred at 26°C. Westslope cutthroat trout mortality was high (99%) during ramping, few fish survived to reach the test temperature, and all fish perished by day three of the experiment (Figure 4). However, rainbow trout survival remained high for about one week at 26°C before it declined steadily and stabilized at 2% for the remainder of the experiment.

Table 4. Mean survival and coefficient of variation for westslope cutthroat trout and rainbow trout at each test temperature.

Westslope cutthroat trout			Rainbow trout		
Temperature (°C)	Mean Survival (%)	*Coefficient of Variation (%)	Temperature (°C)	Mean Survival (%)	*Coefficient of Variation (%)
8	98.3	1.6	8	100.0	0.0
10	96.6	4.4	10	99.3	1.2
12	99.0	2.5	12	100.0	0.0
13	100.0	0.0	14	99.7	0.8
14	98.6	3.4	16	96.3	6.1
15	96.7	6.0	18	99.3	1.2
16	98.9	1.7	20	92.9	8.1
18	100.0	0.0	22	98.6	2.4
20	35.8	123.0	24	72.8	37.3
21	1.3	86.6	26	1.3	86.6
22	12.5	177.2	28	0.0	0.0
23	0.0	0.0			
24	0.0	0.0			

*Coefficient of variation = (standard deviation / mean) * 100

Figure 3. Daily mean survival (%) of age-1 westslope cutthroat trout (solid line) and rainbow trout (dashed line) during 60-d exposure to 20°C and 22°C.

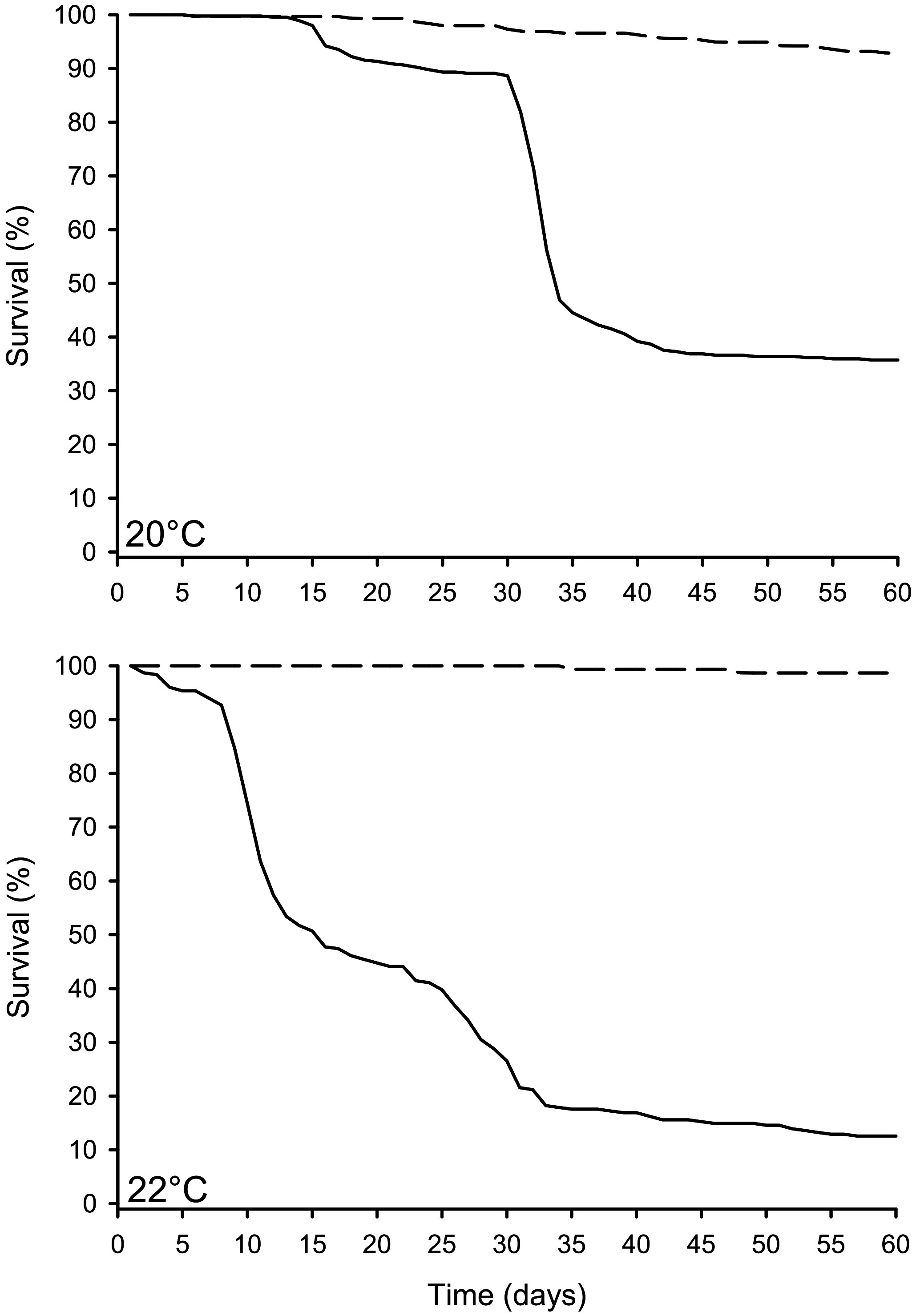
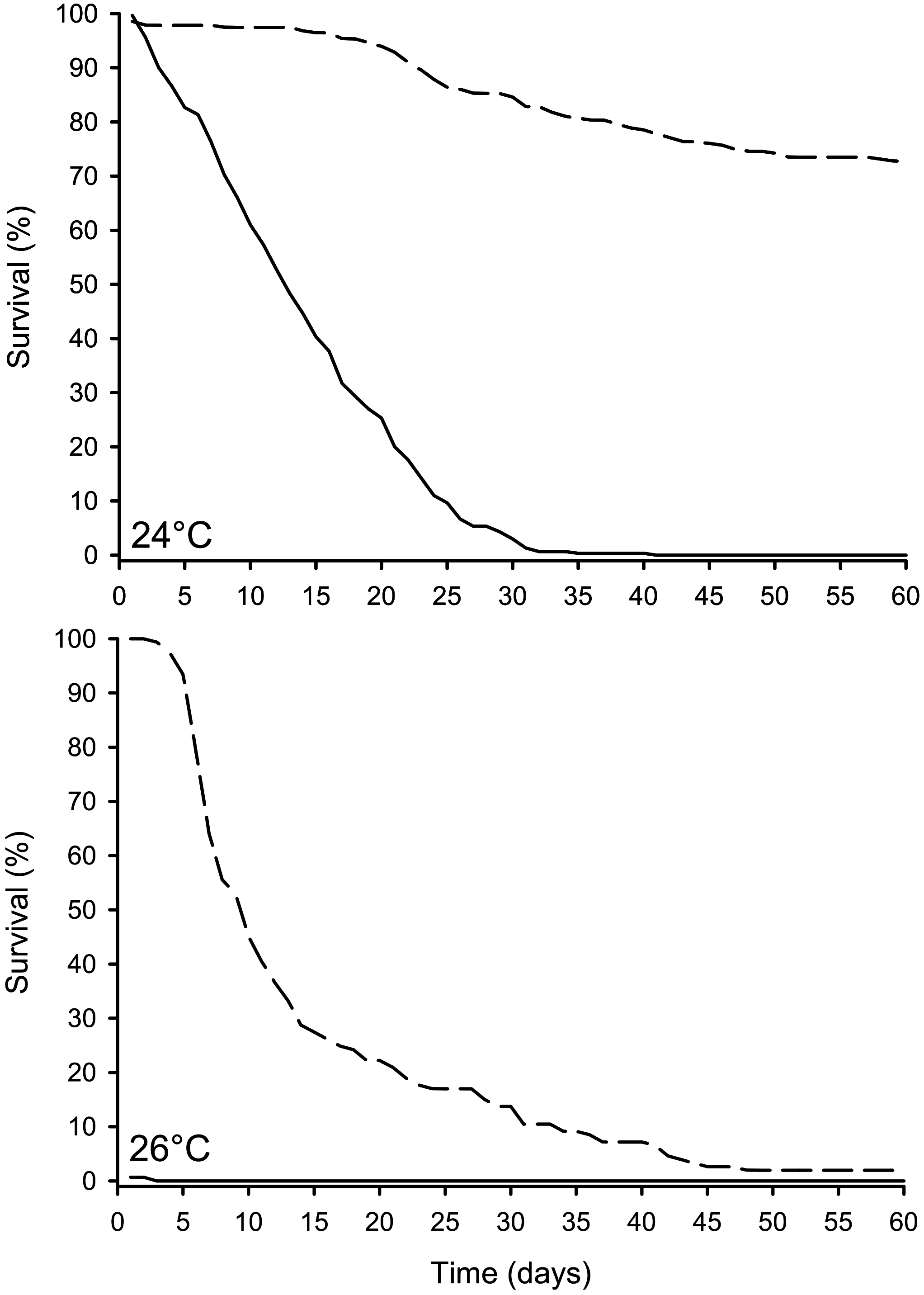
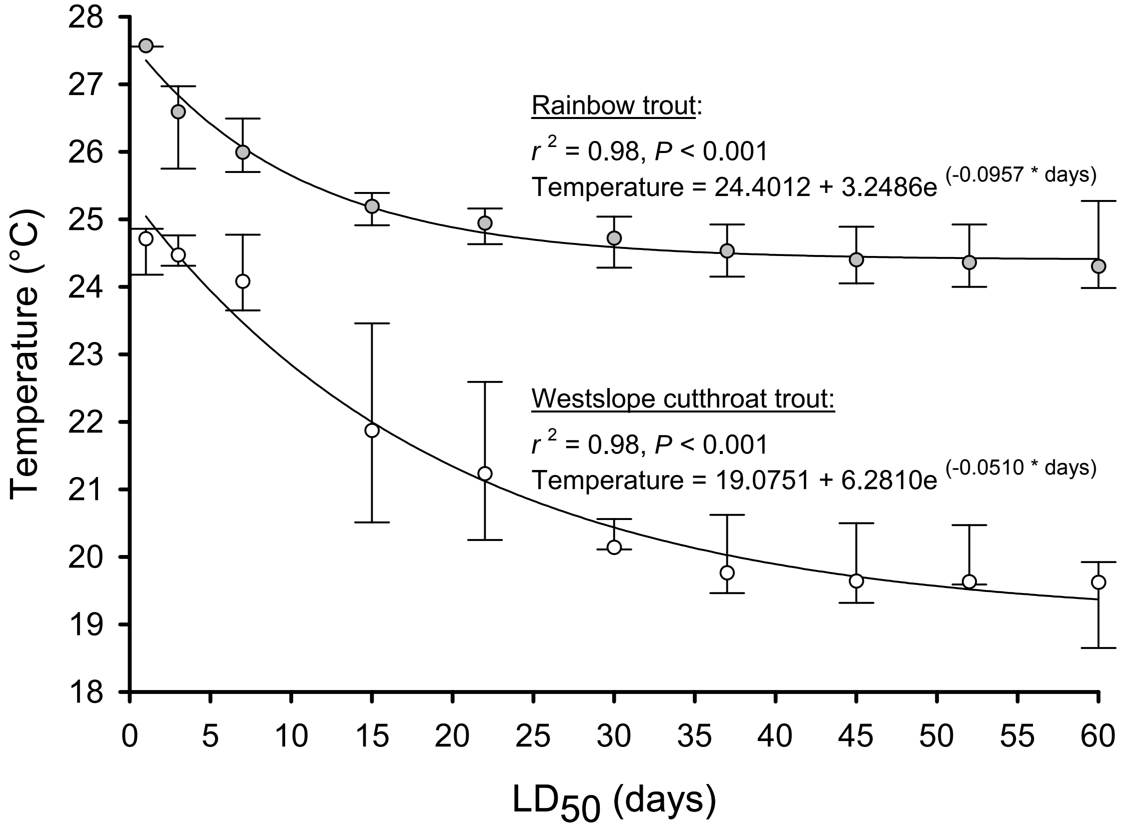


Figure 4. Daily mean survival (%) of age-1 westslope cutthroat trout (solid line) and rainbow trout (dashed line) during 60-d exposure to 24°C and 26°C.



Plots of calculated LD_{50} in relation to temperature using exponential decay regression models revealed a highly significant association between LD_{50} and temperature for both westslope cutthroat trout ($r^2 = 0.98$, $P < 0.001$) and rainbow trout ($r^2 = 0.98$, $P < 0.001$). The LD_{50} for westslope cutthroat trout was significantly lower than for rainbow trout at all temperatures (non-overlapping 95% confidence intervals of the data) (Figure 5).

Figure 5. Survival in relation to temperature for age-1 westslope cutthroat trout (white circles) and rainbow trout (grey circles). Each circle represents the temperature for the median survival time (LD_{50}). Error bars indicate the 95% confidence interval of the data.



The initial weight of westslope cutthroat trout and rainbow trout used in thermal tolerance experiments differed significantly among experiments (Table 5) (ANOVA; $F = 1233.19$, $P < 0.001$). Westslope cutthroat trout in the second experiment were significantly larger than in experiments one and three (Tukey's; $P < 0.001$), whereas no significant size difference existed between fish in experiments one and three (Tukey's; $P = 0.71$). Rainbow trout in experiment five were significantly larger than in experiment four (Tukey's; $P < 0.001$). Because of these size differences, the possible influence of fish size on thermal tolerance was examined by comparing upper lethal temperatures among experiments.

Table 5. Initial mean weight (g/fish) (±SE) for westslope cutthroat trout and rainbow trout in thermal tolerance experiments. Values within each species with different letters are significantly different ($P < 0.001$) among experiments (Tukey's).

Species	Experiment	Initial mean weight (g/fish)
WCT	1	9.4(0.24) ^{A}
	2	19.3(0.33) ^{B}
	3	8.8(0.15) ^{A}
RBT	4	13.9(0.25) ^{A}
	5	38.3(0.58) ^{B}

Fish size appeared to influence thermal tolerance of westslope cutthroat trout and rainbow trout. Thermal tolerance of larger fish was significantly lower than that of their smaller counterparts (non-overlapping 95% confidence intervals of the data), though differences among westslope cutthroat trout were variable between experiments with small fish (Figure 6). In addition, larger fish tended to succumb to mortality sooner than smaller fish during exposure to the same temperature (Figure 7).

Figure 6. Ultimate upper incipient lethal temperature (UUILT) of age-1 westslope cutthroat trout (white) and rainbow trout (grey) in relation to fish size and strain. Each point represents the UUILT of fish of different initial weights in each experiment. Error bars indicate the 95% confidence interval of the data and values with different letters are significantly different.

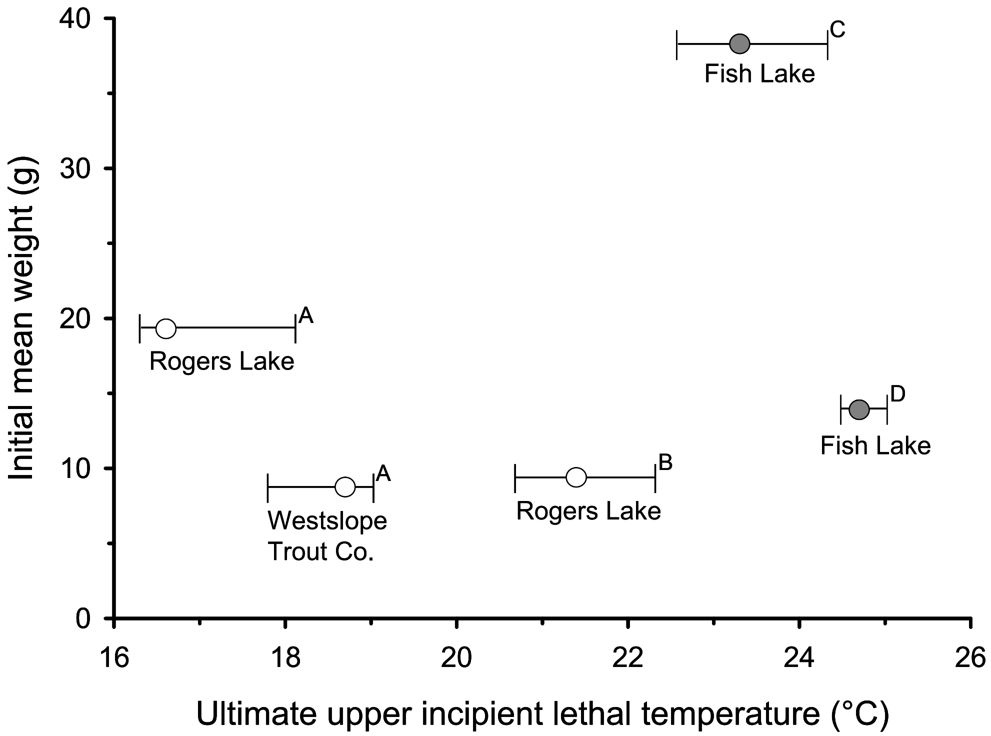
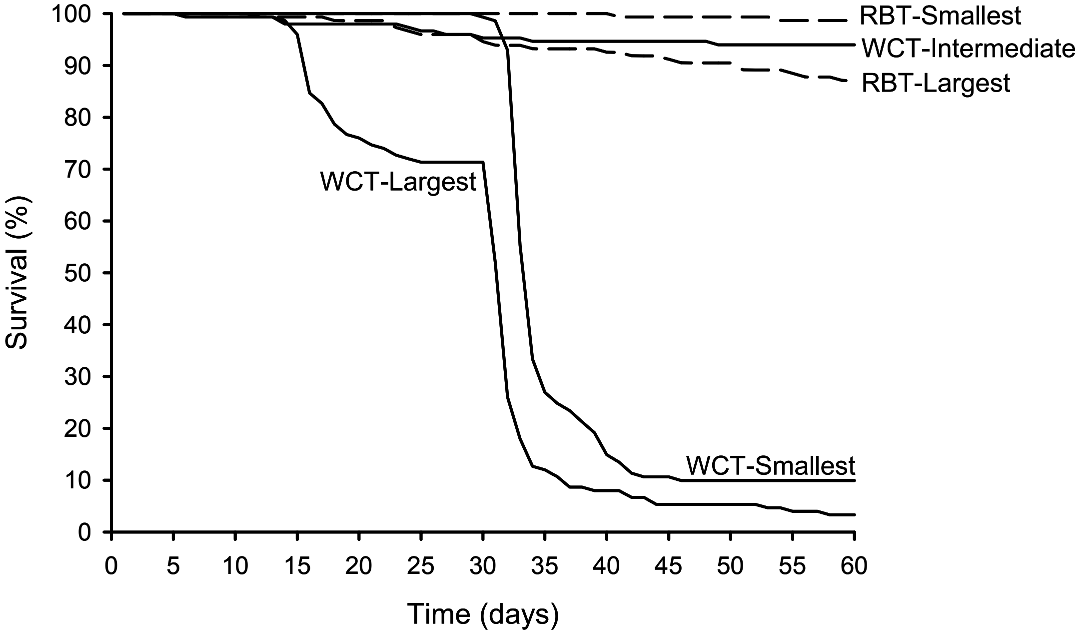


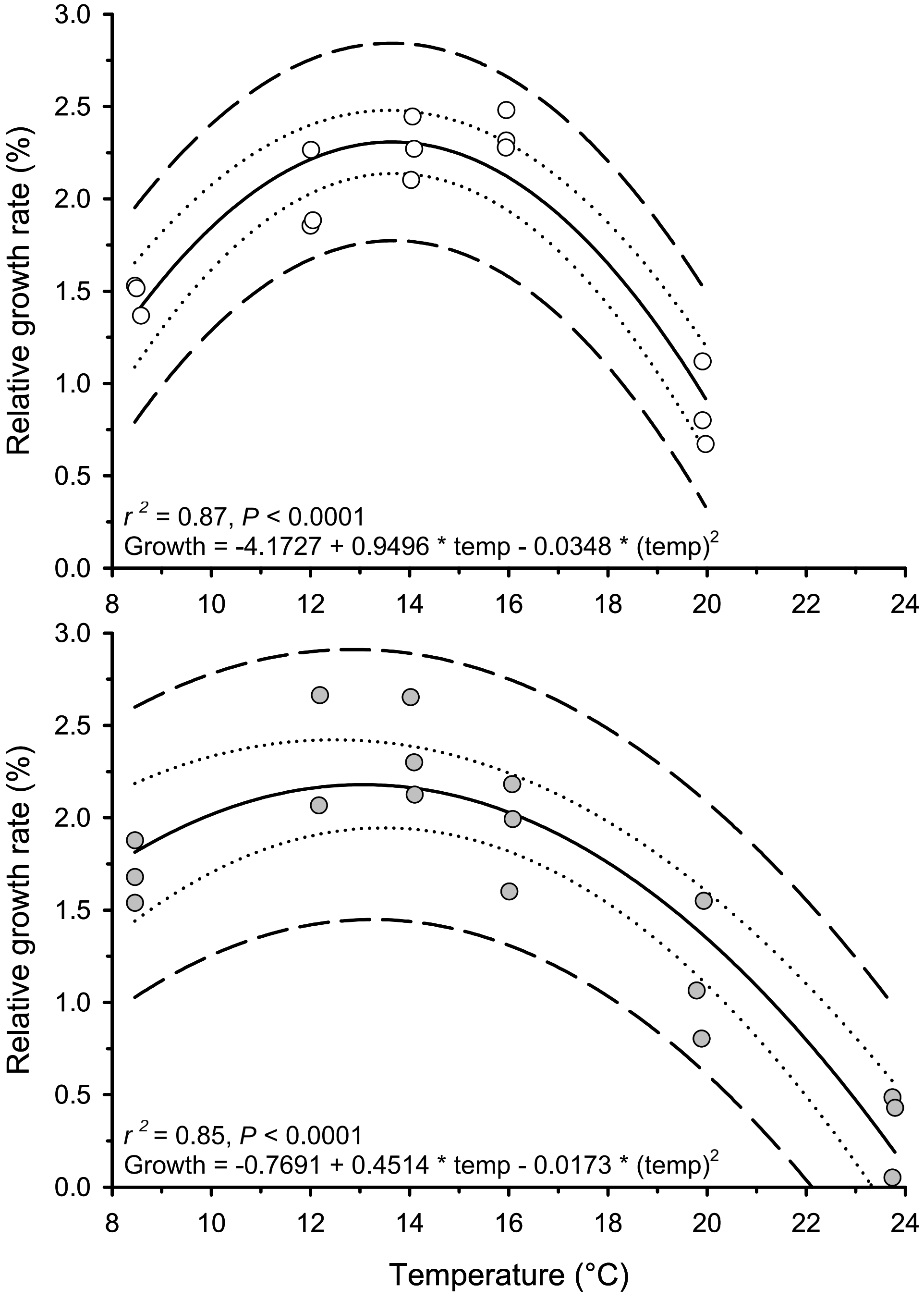
Figure 7. Daily mean survival (%) of small, intermediate, and large age-1 westslope cutthroat trout (solid) and rainbow trout (dashed) during 60-d exposure to 20°C.



Growth

Growth of juvenile westslope cutthroat trout in 60-d experiments varied significantly with temperatures from 8°C to 20°C (Figure 8). Optimum growth (2.31%) occurred at 13.6°C, with an optimal growth range, based on 95% confidence intervals of the regression line, from 10.3°C to 17.0°C. Growth was statistically similar at 12, 14, and 16°C (Tukey's; $P > 0.17$), but was significantly reduced at 8°C and 20°C ($P < 0.03$).

Figure 8. Growth of age-1 westslope cutthroat trout (top) and rainbow trout (bottom) over 60 days in relation to temperature. Each circle represents the relative growth rate (%) per tank with three tanks tested at each temperature. Dotted lines indicate the 95% confidence interval of the regression line and dashed lines indicate the 95% confidence interval of the data.



Rainbow trout showed a slightly lower optimum growth rate (2.17%) and optimum growth temperature (13.1°C) than westslope cutthroat trout (Figure 8), but had a much wider optimal growth range (6.8°C to 18.2°C). Growth of rainbow trout was similar from 8°C to 16°C (Tukey's; $P > 0.14$), and declined significantly at temperatures ≥ 24°C ($P < 0.008$). However, rainbow trout may grow faster than westslope cutthroat trout at temperatures above 20°C. Rainbow trout grew even at temperatures as high as 24°C (0.11%), whereas no westslope cutthroat trout survived long enough at 24°C to assess growth (Figure 8).

Feed Consumption and Conversion Efficiency

Relative feed consumption for both species followed a typical pattern whereby consumption increased to a peak, and decreased at increasing temperatures. For westslope cutthroat trout, consumption peaked at 16°C at 1.13% of body weight consumed per day. Peak consumption was similar for rainbow trout (1.08%) but occurred at 14°C (Table 6). Two-way ANOVA indicated a significant temperature effect ($F = 5.22$, $P = 0.005$) on consumption, but no significant species effect ($F = 2.54$, $P = 0.128$) or species * temperature interaction ($F = 2.83$, $P = 0.053$). Westslope cutthroat trout consumption was similar from 8°C to 14°C (Tukey's, $P > 0.24$), was significantly greater at 16°C ($P = 0.01$), and significantly less at 20°C ($P = 0.02$). In contrast, rainbow trout consumption did not vary significantly from 8°C to 20°C ($P > 0.42$).

Table 6. Feed consumption (±SE), and feed conversion efficiency (±SE) for westslope cutthroat (WCT) and rainbow trout (RBT). Values within each species with the same letter are not significantly different among temperatures and values significantly different between species within the same temperature are indicated within a "+" (ANOVA, Tukey's).

Temperature	Feed consumption (% body weight)		Feed conversion efficiency (g growth / g consumed)	
°C	WCT	RBT	WCT	RBT
8	0.73(0.04) ^{A}	0.86(0.07) ^{A}	1.27(0.05) ^{A}	1.17(0.03) ^{A}
12	0.87(0.04) ^{A}	1.03(0.05) ^{A}	1.33(0.06) ^{A}	1.15(0.04) ^{A}
14	0.99(0.03) ^{A}	1.08(0.05) ^{A}	1.27(0.03) ^{A}	1.15(0.02) ^{A}
16	1.13(0.06) ^{B}	0.92(0.10) ^{A}	1.13(0.03) ^{A}	1.07(0.02) ^{A}
20	0.75(0.10) ^{C}	0.93(0.09) ^{A}	0.45(0.08) ^{B}	0.80(0.06) $^{B+}$

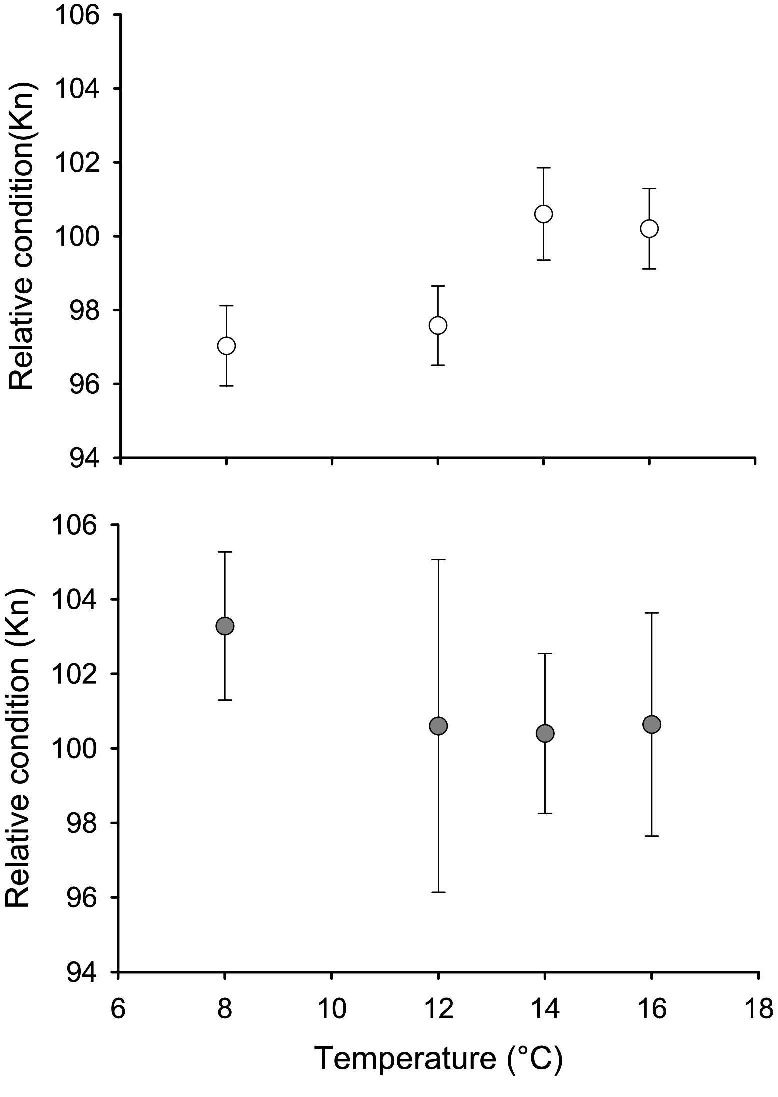
Feed conversion efficiency for westslope cutthroat and rainbow trout followed a similar pattern (Table 6). Two-way ANOVA results indicated a significant temperature effect (F = 61.75, P < 0.001) on feed conversion efficiency with no significant species effect (F = 0.48, P = 0.49), but a significant species * temperature interaction (F = 10.41, P < 0.001). Westslope cutthroat trout efficiency remained relatively constant at most temperatures, varying from 1.13 to 1.33 (P > 0.50), but declined significantly by about half (0.45) at 20°C (P < 0.001). Likewise, rainbow trout feed conversion efficiency was similar at temperatures from 8°C to 16°C (P > 0.85) but decreased significantly at 20°C (P < 0.001). Feed conversion efficiency was similar for both species at temperatures from 8°C to 16°C, but at 20°C rainbow trout had nearly double the feed conversion efficiency of westslope cutthroat trout (0.80 versus 0.45; *P* = 0.001).

Relative Condition

The relationship between relative condition and temperature was not consistent between species. Relative condition of westslope cutthroat trout varied significantly with temperature (ANOVA; *F* = 9.85, *P* < 0.001) but generally remained high (97.0 to 100.6) at all temperatures. Relative condition was similar from 8°C to 12°C (Tukey's; *P* > 0.90), increased significantly at 14°C (*P* < 0.001), and remained high at 16°C. In contrast, there were no significant differences in rainbow trout relative condition among temperatures (*P* = 0.29) with values greater than 100 at all temperatures (100.4 to 103.3) (Figure 9).

The predicted weight of average length westslope cutthroat trout and rainbow trout followed a similar pattern. Predicted weights of average length westslope cutthroat trout (123 mm) increased to a peak at 14°C, and decreased slightly at 16°C. Average length westslope cutthroat trout had a predicted weight of 17.8 g at 8°C, 18.1 g at 12°C, 18.7 g at 14°C, and 18.5 g at 16°C. Therefore, the predicted weight of average size westslope cutthroat trout increased by 1.4% from 8°C to 12°C, by 3.7% from 12°C to 14°C, and decreased by 1.3% from 14°C to 16°C. In contrast, predicted weights of average length rainbow trout (193 mm) decreased linearly with increasing temperature. Average length rainbow trout had a predicted weight of 82.7 g at 8°C, 81.8 g at 12°C, 81.6 g at 14°C, and 80.8 g at 16°C. Thus, the predicted weight of average size rainbow trout decreased by 1.1% from 8°C to 12°C, by 0.2% from 12°C to 14°C, and by 0.9% from 14°C to 16°C.

Figure 9. Average relative condition (Kn) of westslope cutthroat trout (top) and rainbow trout (bottom) after 60-d exposure to constant temperature. Error bars represent the 95% confidence interval of the mean.



DISCUSSION

Rainbow trout had a distinct survival advantage over westslope cutthroat trout at warmer temperatures. Westslope cutthroat trout were able to survive temperatures greater than 22°C for only short periods, whereas rainbow trout had greater than 90% survival over 60 d at 22°C, and a UUILT nearly 5°C higher than that of westslope cutthroat trout. Rainbow trout also showed a much greater thermal resistance than westslope cutthroat trout as evidenced by a markedly greater difference in LD_{50} as exposure time increased. The LD_{50} of westslope cutthroat trout was 2-3°C lower than that of rainbow trout up to 7-d exposure, but increased to near 5°C difference at exposure time of 37 d and greater. Thus, rainbow trout were able to withstand higher temperatures for longer periods than westslope cutthroat trout.

A delay in mortality of westslope cutthroat trout occurred at 20°C, with survival greater than 50% until day 34, at which point survival dropped rapidly to low levels. This indicates that at 20°C, there is a time threshold that westslope cutthroat trout can survive and once this threshold is surpassed, survival quickly declines below the critical level of 50% of the population. Traditional incipient lethal temperature experiments lasting only seven days lack the ability to determine these longer term thresholds, which may be critical for survival in nature as seasonal high temperatures often last longer than seven days. In addition, for westslope cutthroat trout the UUILT for 7 days was 4°C higher than for 60 days. Similarly, for rainbow trout the 7-d UUILT was 2°C higher than the 60-d UUILT. Therefore, 7-d tolerance tests may fail to determine delayed mortality and may overestimate the lethal temperature for fishes. Future thermal tolerance and optima experiments should be conducted for 60 days using the ACE method, to obtain more ecologically relevant thermal criteria for fishes.

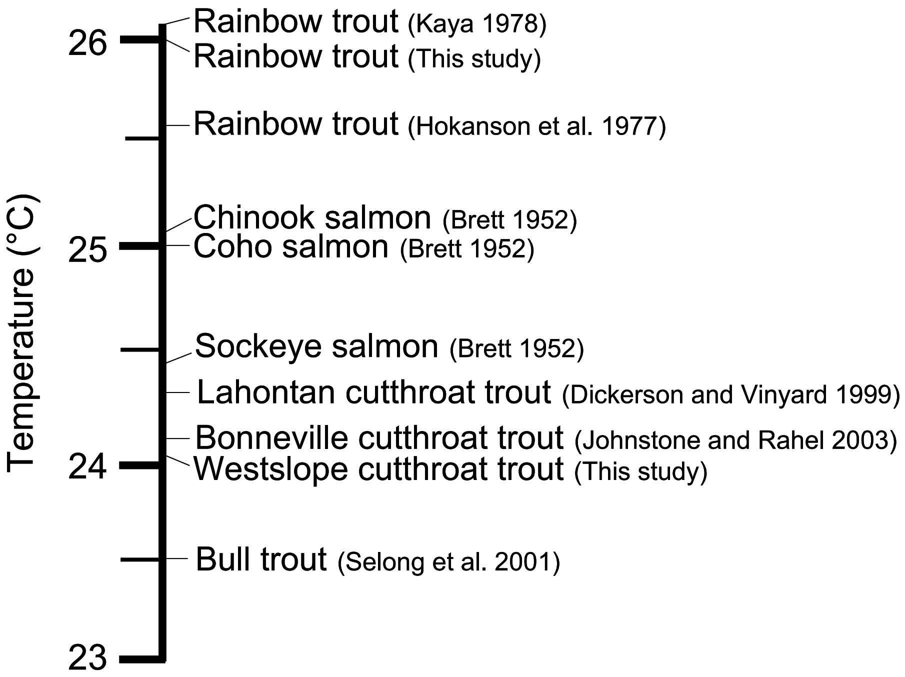
However, upper lethal temperatures based on 7-d exposure periods can be useful in comparing tolerance of westslope cutthroat trout to those of other salmonids (Table 7). The 7-d UUILT for westslope cutthroat trout (24.1°C, this study) is near the lower range reported for salmonids. Other species with UUILTs near 24°C include Lahontan cutthroat trout (24 - 25°C, Dickerson and Vinyard 1999), Bonneville cutthroat trout (24.2°C, Johnstone and Rahel 2003), and bull trout (23.5°C, Selong et al. 2001). Species at the upper end of the thermal tolerance range for salmonids include sockeye salmon *Oncorhynchus nerka* (24.4°C, Brett 1952), coho salmon *O. kisutch* (25.0°C, Brett 1952), and chinook salmon *O. tschawytscha* (25.1°, Brett 1952) (Figure 10). Rainbow trout have one of the highest upper temperature tolerances among salmonids (26.0°C; this study), which was similar to that reported in previous ILT temperature tolerance testing (26.2°C, Kaya 1978; 25.6°C, Hokanson et al. 1997).

Table 7. Ultimate upper incipient lethal temperatures (UUILT), and optimum growth temperatures for various salmonids. All UUILT values are based on 7-d test periods for comparison.

Species	7-d UUILT (°C)	Optimum growth temp. (°C)	Reference
Bonneville cutthroat trout *Oncorhynchus clarkii utah*	24.2		Johnstone and Rahel 2003
Brook trout *Salvelinus fontinalis*		14.0	T.E. McMahon, unpub. data
Brown trout *Salmo trutta*		16.9	Ojanguren et al. 2001
Bull trout *Salvelinus confluentus*	23.5	13.2	Selong et al. 2001
Chinook salmon	25.1		Brett 1952
Oncorhynchus tschawytscha		19.0	Brett et al. 1982
Coho salmon *Oncorhynchus kisutch*	25.0		Brett 1952
Lahontan cutthroat trout	24.0-25.0		Dickerson and Vinyard 1999
Oncorhynchus clarkii henshawi		12.0	Meeuwig et al. 2004
Lake trout *Salvelinus namaycush*		12.5	Edsall and Cleland 2000
Rainbow trout	25.6	17.2	Hokanson et al. 1977
Oncorhynchus mykiss	26.2		Kaya 1978
	26.0	13.1	This study
Sockeye salmon	24.4		Brett 1952
Oncorhynchus nerka		15.0	Brett et al. 1969
Westslope cutthroat trout *Oncorhynchus clarkii lewisi*	24.1	13.6	This study

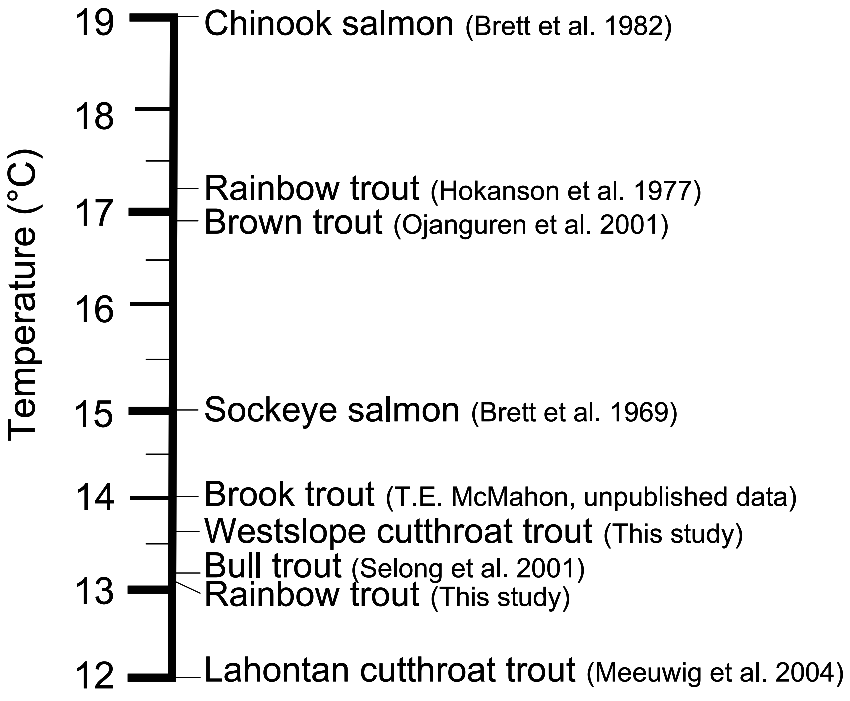
Fish size may influence thermal tolerance of fishes at different temperatures. In this study, fish size did appear to influence thermal tolerance as larger juveniles tended to have reduced tolerance compared to their smaller counterparts. Such instances of larger, older fish being more sensitive to warmer temperatures than younger fish has been previously recognized (Benfey et al. 1997; Selong et al. 2001; Meeuwig et al. 2004). Therefore, to adequately determine the ultimate upper incipient lethal temperature and optimum growth temperature of fishes, testing of several sizes of fish is recommended in order to develop temperature tolerance survival curves applicable to all juveniles of a species.

Figure 10. Range of 7-d ultimate upper incipient lethal temperatures for various salmonids.



Despite very different upper thermal tolerances, westslope cutthroat trout and rainbow trout had surprisingly similar optimum growth temperatures. However, my results indicate that rainbow trout may have a growth advantage at temperatures above the optimal range for both species. As with upper temperature tolerance, it appears that two distinct groups exist with respect to optimum growth temperatures for salmonids. Salmonids comprising the group with lower optima include Lahontan cutthroat trout (12°C - 13°C, Meeuwig et al. 2004), bull trout (13.2°C, Selong et al. 2001) westslope cutthroat trout (13.6°C, this study), and brook trout (14.0°C, T.E. McMahon, Montana State University, unpublished data). In contrast, sockeye salmon (15°C, Brett et al. 1969), brown trout (16.9°C, Ojanguren et al. 2001), and chinook salmon (19°C, Brett et al. 1982) constitute a group with higher growth optimums (Figure 11).

Figure 11. Range of optimum growth temperatures for various salmonids.



According to this study, rainbow trout would belong to the group of salmonids with lower optimums. However, optimum growth for rainbow trout in this study occurred at a much lower temperature (13.1°C) than previously reported for the species (17.2°C, Hokanson et al. 1977) (Figure 11). This difference may be attributable to considerably smaller fish used by Hokanson et al. (1977) than were used in this study (mean length, 30 mm versus 153 mm, respectively), as larger, older fish tend to prefer and have lower optimum temperatures than smaller, younger fish (Kwain and McCauley 1978; Meeuwig et al. 2004).

Differences in the strain of rainbow trout tested could also account for differing optimum growth temperatures. However, whether physiological differences among strains exist has been inconclusive (Myrick and Cech 2000; Imsland et al. 2001; Larsson et al. 2005). Strain related differences in swimming ability occur in coho salmon (Taylor and McPhail 1985); sockeye salmon (Taylor and Foote 1991), and rainbow trout (Thomas and Donahoo 1977), but differences in thermal tolerance (Kaya 1978), conversion efficiency, oxygen consumption, and swimming performance (Myrick and Cech 2000) of rainbow trout strains have not been observed in other studies. It remains to be seen whether strain related differences in growth of rainbow trout exist and further research in this area would be beneficial.

Based on optimal growth temperatures established in this study, the fundamental niche for westslope cutthroat trout, defined by Christie and Regier

(1988) as -3°C to +1°C of the optimum growth temperature, is from 10.6°C to 14.6°C. Similarly, the growth ranges reported in this study generate a fundamental niche for rainbow trout from 10.1°C to 14.1°C. The high degree of overlap between the fundamental niches of these species indicates the potential for competition in areas where the species co-occur. Based on the information from this study, the two species may be equally competitive from 12°C to 16°C, but rainbow trout may gain a growth advantage at temperatures above this range. However, differences in growth may become more evident when two species are in sympatry (Gunckel et al. 2002; T.E. McMahon, Montana State University, unpublished data). Future research on growth of westslope cutthroat trout and rainbow trout in sympatry is needed to determine possible influence of water temperature on competition.

As with other species feed conversion, both westslope cutthroat trout and rainbow trout became less efficient at temperatures above peak growth temperatures as more energy was used to meet increasing metabolic demands and less was converted into growth (McCarthy et al. 1998; Selong et al. 2001). Interestingly, rainbow trout feed efficiency was greater than westslope cutthroat trout at 20°C, indicating an enhanced ability to convert food into growth at warmer temperatures. This enhanced efficiency by rainbow trout may explain the observed growth advantage over westslope cutthroat trout at warmer temperatures.

Relative condition of fishes typically increases with temperature to some optimum (Filbert and Hawkins 1995), and decreases as temperatures reach extremes for the species (Kocovsky and Carline 2001). Relative condition of westslope cutthroat in this study appeared to be highest near the optimum growth temperature for the species. However, the relationship between relative condition and temperature for rainbow trout was equivocal as condition did not vary significantly with temperature. Inclusion of temperatures nearer the upper limit for growth of rainbow trout may have elicited a greater response in relative condition. Furthermore, the influence of temperature on parameters such as relative condition, feed efficiency, and growth may become more prominent at rations less than satiation (Brett et al. 1969; Miglavs and Jobling 1989; Van Ham et al. 2003). Future thermal studies to determine the effect of reduced ration on relative condition, feed efficiency, and growth of westslope cutthroat trout and rainbow trout over a range of temperatures would be beneficial.

The ACE Methodology

The ACE technique has some important advantages over traditional methods for determining thermal requirements of aquatic ectotherms. The short term nature of traditional methods, and the limited ability for test organisms to properly acclimate, may limit the ecological relevance of these techniques compared to the ACE method (Selong et al. 2001). However, this study has revealed that the long term nature of the ACE method may, in turn, present logistical difficulties. The length of the experiments (~90 d) substantially limits the number that can be conducted in a year. Obtaining eggs from wild parentage stocks is also problematic as they may only be available during limited time periods, unlike more domesticated stocks which have been selected for longer duration breeding. This necessitates holding test fish for long periods between experiments, making it difficult to keep fish size constant among different experiments. However, as noted, it may be beneficial to conduct tolerance and growth experiments with different size fish in order to develop criteria more suitable to all juveniles of the species.

Furthermore, I found that of great concern is the increased likelihood for disease outbreaks resulting from long holding periods, as several planned experiments could not be completed because of instances of disease. If a disease outbreak occurs during a test year, this may delay running an experiment by a full year. Wild stocks are likely to be less disease resistant than hatchery-bred stocks (Winton 2001), further adding to the potential for disease outbreaks. The many advantages of the ACE method outweigh potential disadvantages, but planning of future studies with wild salmonids should include steps to minimize disease risks by isolating test fish and frequent fish health assessments and disease monitoring for early detection and control of disease outbreaks.

Ecological and Management Implications

Temperature has been shown to be a major factor affecting the distribution of fishes, especially coldwater salmonids (Paul and Post 2001; Sloat 2001; Dunham et al. 1999). For example, distribution of westslope cutthroat trout in the Madison River, Montana was associated with maximum daily stream temperatures less than 16°C, whereas rainbow trout distribution was associated with maximum daily stream temperatures less than 18°C (Sloat 2001). The ability of rainbow trout to survive prolonged exposure to high temperatures (> 20°C) and to grow over a wide range of temperatures, may account for the increased occurrence of rainbow trout at lower elevations and warmer temperatures. In contrast, the lower thermal tolerance and narrower growth range of westslope cutthroat trout may further explain their increasing distribution in cooler, headwater streams, where colder water temperatures may allow them to remain more competitive with nonnative salmonids more tolerant of warmer temperatures.

Temperature may also interact with other factors to influence the distribution and persistence of salmonids. Elevated water temperatures may exhibit stress on fishes, decreasing their predator avoidance (Deacutis 1978; Marine and Cech 2004) and compromising immune system function which increases disease susceptibility (Noga 1996). Increased water temperature may also influence competition by shifting the advantage to species with physiological optimums at higher temperatures (De Staso and Rahel 1994).

Water quality standards addressing water temperature can be implemented in streams that are critical to the persistence of remaining westslope cutthroat trout populations. Thermal criteria for westslope cutthroat trout from this study is in accord with Coffin and Cowan (1995) and indicates that daily stream temperatures not exceed 20°C for prolonged survival of westslope cutthroat trout. However, standards based on growth criteria in this study may better reflect suitable thermal habitat for the long-term persistence of westslope cutthroat trout. For instance, bull trout occurrence was relatively low (< 50%) at temperatures near the lethal limit for the species, but became much greater (75%) at temperatures near the optimum growth temperature for the species (Dunham et al. 2003). Therefore, water quality standards setting maximum daily temperatures from 13-15°C, near the optimum growth temperature, would ensure suitable thermal habitat to maintain the persistence of westslope cutthroat trout populations.

Furthermore, the ability to predict the temperature fish can survive based on exposure time is valuable in managing at-risk populations and setting water quality criteria regarding stream temperature. This can now be predicted for westslope cutthroat trout and rainbow trout based on the relationship between LD_{50} and temperature derived in this study. For example, if a stream is known to have temperatures greater than 23°C lasting for several days, one can predict that the majority of westslope cutthroat trout could only survive for nine days. Temperature has been identified as an important factor influencing the success of translocations of cutthroat trout (Rio Grande cutthroat trout *Oncorhynchus clarkii virginalis* and Greenback cutthroat trout *Oncorhynchus clarki stomias*) (Harig and Fausch 2002; Young and Guenther-Gloss 2004). Therefore, using the magnitude and duration of high temperatures in streams, managers can now determine whether reintroduced westslope cutthroat trout could successfully survive in potential translocation sites.

The thermal criteria found in this study could be used with stream temperature data to predict presence of westslope cutthroat trout in nature. For example, using fish sampling data and U.S. Geological Survey temperature data from streams throughout the United States, the presence or absence of 13 warm-water species was predicted with greater than 90% accuracy (Scheller et al. 1999). Similarly, point water temperature and fish presence data from streams throughout Montana were used to predict the probability of occurrence of westslope and Yellowstone cutthroat trout. The predicted probability of occurrence for cutthroat trout was best modeled by a second-order polynomial and was highest, about 91%, at water temperatures of 11.2°C, but dropped to less than 40% at temperatures $\geq$ 20°C (Brad Shepard, Montana Fish, Wildlife and Parks, personal communication).

Potential impacts of climate change and land use practices on the distribution of westslope cutthroat trout can be predicted based on thermal criteria for the species. For instance, distribution patterns for salmonids in the Rocky Mountain region have a strong relationship to elevation and latitude such that distribution is limited to areas where mean July air temperatures are less than 22°C (Keleher and Rahel 1996). With projected warming by 1 to 5°C in mean July air temperatures, the geographic area containing suitable habitat for salmonids in the Rocky Mountain region is projected to decrease by 17 to 72%. In addition, land use practices that alter temperature and amount of woody debris, pool habitat, and fine sediment in streams may affect the distribution of westslope cutthroat trout by facilitating displacement by nonnatives such as brook trout (Shepard 2004). Therefore, defining and protecting suitable habitat for westslope cutthroat trout is vital for their persistence. Survival and growth parameters indicated in this study can be used with stream temperature modeling to predict suitable habitat for westslope cutthroat trout (Sloat et al. 2005), as they may be particularly susceptible to increases in stream temperature associated with climate change. Such predictions of habitat suitability will be vital in prioritizing conservation efforts with respect to reintroduction and translocation of westslope cutthroat trout.

Recommendations for Future Research

Now that upper lethal and optimum growth temperatures have been established for westslope cutthroat trout, future research should focus on additional factors (e.g., ration) which may interact with water temperature to affect survival and growth. Thermal studies on different strains of westslope cutthroat trout and rainbow trout would be beneficial in providing a conclusive answer as to whether strain influences the temperature for optimum growth of a species. In particular, the rather large difference in peak growth of rainbow trout in our study compared to previous studies suggests the potential for such strain differences. In addition, thermal studies to determine the influence of competition with nonnative salmonids (rainbow and brook trout) on growth of westslope cutthroat trout could further explain the dominance of these species at warmer water temperatures. Lastly, determining the thermal requirements of other salmonids that have declined throughout their native range (e.g., Yellowstone cutthroat trout) can help fisheries managers in protecting at-risk populations and defining suitable thermal habitat for restoration.

REFERENCES CITED

Allendorf, F. W., and R. F. Leary. 1988. Conservation and distribution of genetic variation in a polytypic species, the cutthroat trout. Conservation Biology 2:170-184.

Allendorf, F. W., R. F. Leary, N. P. Hitt, K. L. Knudsen, L. L. Lundquist, and P. Spruell. 2004. Intercrosses and the U.S. Endangered Species Act: should hybridized populations be included as westslope cutthroat trout? Conservation Biology 18:1203-1213.

Anderson, R. O., and R. M. Neumann. 1996. Length, weight, and associated structural indices. Pages 447-482 *in* B. R. Murphy and D. W. Willis editors. Fisheries techniques, second edition. American Fisheries Society, Bethesda, Mayland.

APHA (American Public Health Association), American Water Works Association, and Water Environment Federation. 1992. Standard methods for the examination of water and wastewater, 18th edition. APHA, Washington, D.C.

Barrows, F. T., and R. W. Hardy. 2001. Nutrition and Feeding. Pages 493-558 *in* G. A. Wedemeyer, editor. Fish hatchery management, second edition. American Fisheries Society, Bethesda, Maryland.

Behnke, R. J. 1992. Native trout of western North America. American Fisheries Society, Monograph 6, Bethesda, Maryland.

Benfey, T. J., L. E. McCabe, and P. Pepin. 1997. Critical thermal maxima of diploid and triploid brook charr, *Salvelinus fontinalis*. Environmental Biology of Fishes 49:259-264.

Bozek, M. A., and W. A. Hubert. 1992. Segregation of resident trout in streams as predicted by three habitat dimensions. Canadian Journal of Zoology 70:886-890.

Brett, J. R. 1952. Temperature tolerance in young Pacific salmon, genus *Oncorhynchus*. Journal of the Fisheries Research Board of Canada 9:265-322.

Brett, J. R. 1956. Some principles in the thermal requirements of fishes. The Quarterly Review of Biology 31:75-87.

Brett, J. R., J. E. Shelbourn, and C. T. Shoop. 1969. Growth rate and body composition of fingerling sockeye salmon, *Oncorhynchus nerka*, in relation to temperature and ration size. Journal of the Fisheries Research Board of Canada 26:2363-2394.

Brett, J. R. 1971. Energetic responses of salmon to temperature. A study of some thermal relations in the physiology and freshwater ecology of sockeye salmon (*Oncorhynchus nerka*). American Zoologist 11:99-113.

Brett, J. R. 1979. Environmental factors and growth. Pages 599-675 *in* W.S. Hoar, D.J. Randall, and J.R. Brett, editors. Fish Physiology, volume 8. Academic Press, New York.

Brett, J. R., W. C. Clarke, and J. E. Shelbourn. 1982. Experiments on thermal requirements for growth and food conversion efficiency of juvenile chinook salmon *Oncorhynchus tshawytscha*. Canada Fisheries and Marine Service Technical Report 1127.

Brown, G. W., and J. T. Krygier. 1970. Effects of clear-cutting on stream temperature. Water Resources Bulletin 6:1133-1139.

Carmichael, G. J., J. N. Henson, M. E. Schmidt, and D. C. Morizot. 1993. Introgression among Apache, cutthroat, and rainbow trout in Arizona. Transactions of the American Fisheries Society 122:121-130.

Christie, G. C., and H. A. Regier. 1988. Measures of optimal thermal habitat and their relationship to yields for four commercial fish species. Canadian Journal of Fisheries and Aquatic Sciences 45:301-314.

Coffin, P. D., and W. F. Cowan. 1995. Lahontan cutthroat trout (*Oncorhynhus clarki henshawi*) recovery plan. U.S. Fish and Wildlife Service Region 1, Portland, Oregon.

Cunjak, R. A., and J. M. Green. 1986. Influence of water temperature on behavioural interactions between juvenile brook charr, *Salvelinus fontinalis*, and rainbow trout, *Salmo gairdneri*. Canadian Journal of Zoology 64:1288-1291.

Cushing, C. E., and J. D. Allan. 2001. Streams, their ecology and life. Academic Press. San Diego, California.

De Staso, J., and F. J. Rahel. 1994. Influence of water temperature on interactions between juvenile Colorado River cutthroat trout and brook trout in a laboratory stream. Transactions of the American Fisheries Society 123:289-297.

Deacutis, C. F. 1978. Effect of thermal shock on predator avoidance by larvae of two fish species. Transactions of the American Fisheries Society 107:632-635.

Dickerson, B. R., and G. L. Vinyard. 1999. Effects of high chronic temperatures and diel temperature cycles on the survival and growth of Lahontan cutthroat trout. Transactions of the American Fisheries Society 128:516-521.

Dunham, J. B., M. M. Peacock, B. E. Rieman, R. E. Schroeter, and G. L. Vinyard. 1999. Local and geographic variability in the distribution of stream-living Lahontan cutthroat trout. Transactions of the American Fisheries Society 128:875-889.

Dunham, J., B. Rieman, and G. Chandler. 2003. Influences of temperature and environmental variables on the distribution of bull trout within streams at the southern margin of its range. North American Journal of Fisheries Management 23:894-904.

Edsall, T. A., and J. Cleland. 2000. Optimum temperature for growth and preferred temperatures of age-0 lake trout. North American Journal of Fisheries Management 20:804-809.

Filbert, R. B., and C. P. Hawkins. 1995. Variation in condition of rainbow trout in relation to food, temperature, and individual length in the Green River, Utah. Transactions of the American Fisheries Society 124:824-835.

Fry, F. E. J. 1947. Effects of the environment on animal activity. University of Toronto Studies, Biological Series 55. Publication of the Ontario Fisheries Research Laboratory 68:1-62.

Fry, F. E. J. 1971. The effect of environmental factors on the physiology of fish. Pages 1-98 *in* W.S. Hoar and D.J. Randall, editors. Fish physiology, volume 6. Academic Press, New York.

Galarowicz, T. L., and D. H. Wahl. 2003. Differences in growth, consumption, and metabolism among walleyes from different latitudes. Transactions of the American Fisheries Society 132:425-437.

Griffith, J. S. 1988. Review of competition between cutthroat trout and other salmonids. American Fisheries Society Symposium 4:134-140.

Gunckel, S. L., A. R. Hemmingsen, and J. L. Li. 2002. Effect of bull trout and brook trout interactions on foraging habitat, feeding behavior, and growth. Transactions of the American Fisheries Society 131:1119-1130.

Hanzel, D. A. 1959. The distribution of the cutthroat trout (*Salmo clarki*) in Montana. Proceedings of the Montana Academy of Sciences 19:32-71.

Harig, A. L., and K. D. Fausch. 2002. Minimum habitat requirements for establishing translocated cutthroat trout populations. Ecological Applications 12:535-551.

Hitt, N. P., C. A. Frissell, C. C. Muhlfeld, and F. W. Allendorf. 2003. Spread of hybridization between native westslope cutthroat trout, *Oncorhynchus clarki lewisi*, and nonnative rainbow trout, *Oncorhynchus mykiss*. Canadian Journal of Fisheries and Aquatic Sciences 60:1440-1451.

Hokanson, K. E. F., C. F. Kleiner, and T. W. Thorslund. 1977. Effects of constant temperatures and diel temperature fluctuations on specific growth and mortality rates and yield of juvenile rainbow trout, *Salmo gairdneri*. Journal of the Fisheries Research Board of Canada 34:639-648.

Imsland, A. K., A. Foss, and S. O. Stafansson. 2001. Variation in food intake, food conversion efficiency and growth of juvenile turbot from different geographic strains. Journal of Fish Biology 59:449-454.

Jobling, M. 1981. Temperature tolerance and the final preferendum-rapid methods for the assessment of optimum growth temperatures. Journal of Fish Biology 19:439-455.

Johnstone, H. C., and F. J. Rahel. 2003. Assessing temperature tolerance of Bonneville cutthroat trout based on constant and cycling thermal regimes. Transactions of the American Fisheries Society 132:92-99.

Kaya, C. M. 1978. Thermal resistance of rainbow trout from a permanently heated stream, and of two hatchery strains. Progressive Fish-Culturist 40:37-39.

Keleher, C. J., and F. J. Rahel. 1996. Thermal limits to salmonid distribution in the Rocky Mountain region and potential habitat loss due to global warming: a geographic information system (GIS) approach. Transactions of the American Fisheries Society 125:1-13.

Kestemont, P., and E. Baras. 2001. Environmental factors and feed intake: mechanisms and interactions. Pages 131-145 *in* D. Houlihan, T. Boujard, and M. Jobling, editors. Food intake in fish. Blackwell Science, London, England.

Kocovsky, P. M., and R. F. Carline. 2001. Influence of extreme temperature on consumption and condition of walleyes in Pymatuning Sanctuary, Pennsylvania. North American Journal of Fisheries Management 21:198-207.

Kruse, C. G., W. A. Hubert, and F. J. Rahel. 2000. Status of Yellowstone cutthroat trout in Wyoming waters. North American Journal of Fisheries Management 20:693-705.

Kwain, W., and R. W. McCauley. 1978. Effects of age and overhead illumination on temperatures preferred by underyearling rainbow trout, *Salmo gairdneri*, in a vertical temperature gradient. Journal of the Fisheries Research Board of Canada 35:1430-1433.

Larsson, S., T. Forseth, I. Berglund, A. J. Jensen, I. Näslund, J. M. Elliott, and B. Jonsson. 2005. Thermal adaptation of Arctic charr: experimental studies of growth in eleven charr populations from Sweden, Norway, and Britain. Freshwater Biology 50:353-368.

Le Cren, E. D. 1951. The length-weight relationship and seasonal cycle in gonad weight and condition in the perch (*Perca fluviatilis*). The Journal of Animal Ecology 20:201-219.

Leary, R. F., F. W. Allendorf, and G. K. Sage. 1995. Hybridization and introgression between introduced and native fish. American Fisheries Society Symposium 15:91-101.

Liknes, G. A., and P. J. Graham. 1988. Westslope cutthroat trout in Montana: life history, status, and management. American Fisheries Society Symposium 4:53-60.

Mallet, J. P., C. H. Persat, and P. Auger. 1999. Growth modeling in accordance with daily water temperature in European grayling (*Thymallus thymallus* L.). Canadian Journal of Fisheries and Aquatic Sciences 56:994-1000.

Marine, K. R., and J. J. Cech, Jr. 2004. Effects of high water temperature on growth, smoltification, and predator avoidance in juvenile Sacramento River chinook salmon. North American Journal of Fisheries Management 24:198-210.

McCarthy, I., E. Moksness, and D. A. Pavlov. 1998. The effects of temperature on growth rate and growth efficiency of juvenile common wolfish. Aquaculture International 6:207-218.

McCullough, D. A. 1999. A review and synthesis of effects of alterations to the water temperature regime on freshwater life stages of salmonids, with special reference to chinook salmon. U.S. Environmental Protection Agency Report EPA 910-R-99-010. Seattle, Washington.

McCullough, D. A., S. Spalding, D. Sturdevant, and M. Hicks. 2001. Summary of technical literature examining the physiological effects of temperature on salmonids. U.S. Environmental Protection Agency Report EPA-910-D-01-005. Washington, D.C.

Medvick, P. A. 1979. Growth rates of juvenile maomao, *Abudefuf abdominalis*, at constant and cyclic temperatures. Transactions of the American Fisheries Society 108:293-298.

Meeuwig, M. H., J. B. Dunham, J. P. Hayes, and G. L. Vinyard. 2004. Effects of constant and cyclical thermal regimes on growth and feeding of juvenile cutthroat trout of variable sizes. Ecology of Freshwater Fish 13:208-216.

Miglavs, I., and M. Jobling. 1989. Effects of feeding regime on food consumption, growth rates, and tissue nucleic acids in juvenile Arctic charr, *Salvelinus alpinus*, with particular respect to compensatory growth. Journal of Fish Biology 34:947-957.

Minitab. 2000. Minitab Statistical Software: release 14.0. Minitab, State College, Pennsylvania.

MNHP (Montana Natural Heritage Program), and Montana Fish, Wildlife and Parks. 2004. Montana Animal Species of Concern. Pages 1-11.

Myrick, C. A., and J. J. Cech, Jr. 2000. Temperature influences on California rainbow trout physiological performance. Fish Physiology and Biochemistry 22:245-254.

Nogas, E. J. 1996. Fish disease: diagnosis and treatment. Mosby-Year Book, Inc. St. Louis, Missouri.

Ojanguren, A. F., F. G. Reyes-Gaviláń, and F. Braña. 2001. Thermal sensitivity of growth, food intake, and activity of juvenile brown trout. Journal of Thermal Biology 26:165-170.

Paul, A. J., and J. R. Post. 2001. Spatial distribution of native and nonnative salmonids in streams of the eastern slopes of the Canadian Rocky Mountains. Transactions of the American Fisheries Society 130:417-430.

Piper, R. G., I. B. McElwain, L. E. Orme, J. P. McCraren, L. G. Fowler, and J. R. Leonard. 1982. Fish hatchery management. U.S. Fish and Wildlife Service, Washington, D.C.

Poole, G. C., and C. H. Berman. 2001. An ecological perspective on in-stream temperature: natural heat dynamics and mechanisms of human-caused thermal degradation. Environmental Management 27:787-802.

Reese, C. D., and B. C. Harvey. 2002. Temperature-dependent interactions between juvenile steelhead and Sacramento pikeminnow in laboratory streams. Transactions of the American Fisheries Society 131:599-606.

Ricker, W. E. 1979. Growth rates and models. Pages 677-743 *in* W.S. Hoar, D.J. Randall, and J.R. Brett, editors. Fish physiology, volume 8. Academic Press, New York.

Sauter, S. T., J. McMillan, and J. Dunham. 2001. Salmonid behavior and water temperature. U.S. Environmental Protection Agency Report EPA-910-D0-01-001. Washington, D.C.

Scheller, R. M., V. M. Snarksi, J. G. Eaton, and G. W. Oehlert. 1999. An analysis of the influence of annual thermal variables on the occurrence of fifteen warmwater fishes. Transactions of the American Fisheries Society 128:257-264.

Selong, J. H., T. E. McMahon, A. V. Zale, and F. T. Barrows. 2001. Effect of temperature on growth and survival of bull trout, with application of an improved method for determining thermal tolerance in fishes. Transactions of the American Fisheries Society 130:1026-1037.

Shepard, B. B., B. Sanborn, L. Ulmer, and D. C. Lee. 1997. Status and risk of extinction for westslope cutthroat trout in the upper Missouri River basin, Montana. North American Journal of Fisheries Management 17:1158-1172.

Shepard, B. B., B. E. May, and W. Urie. 2003. Status of westslope cutthroat trout (*Oncorhynchus clarki lewisi*) in the United States: 2002. Report of the Westslope Cutthroat Interagency Conservation Team. 88p.

Shepard, B. B. 2004. Factors the may be influencing nonnative brook trout invasion and their displacement of native westslope cutthroat trout in three adjacent southwestern Montana streams. North American Journal of Fisheries Management 24:1088-1100.

SigmaPlot. 2002. SigmaPlot® version 8.0. SPSS Publishing, Chicago, Illinois.

Sloat, M. R., B. B. Shepard, R. G. White, and S. Carson. 2005. Influence of stream temperature on the spatial distribution of westslope cutthroat trout growth potential within the Madison River basin, Montana. North American Journal of Fisheries Management 25:225-237.

Sloat, M. R. 2001. Status of westslope cutthroat trout in the Madison River basin: the influence of dispersal barriers and stream temperature. Master's Thesis. Montana State University, Bozeman, Montana.

Taniguchi, Y., F. J. Rahel., D. C. Novinger, and K. G. Gerow. 1998. Temperature mediation of competitive interactions among three fish species that replace each other along longitudinal stream gradients. Canadian Journal of Fisheries and Aquatic Sciences 55:1894-1901.

Taylor, E. B., and J. D. McPhail. 1985. Variation in burst and prolonged swimming performance among British Columbia populations of coho salmon, *Oncorhynchus kisutch*. Canadian Journal of Fisheries and Aquatic Sciences 42:2029-2033.

Taylor, E. B., and C. J. Foote. 1991. Critical swimming velocities of juvenile sockeye salmon and kokanee, the anadromous and non-anadromous forms of *Oncorhynchus nerka* (Walbaum). Journal of Fish Biology 38:407-419.

Thomas, A. E., and M. J. Donahoo. 1977. Differences in swimming performance among strains of rainbow trout (*Salmo gairdneri*). Journal of the Fisheries Research Board of Canada 34:304-306.

Thomas, R. E., J. A. Gharett, M. G. Carls, S. D. Rice, A. Moles, and S. Korn. 1986. Effects of fluctuating temperature on mortality, stress, and energy reserves of juvenile coho salmon. Transactions of the American Fisheries Society 155:52-59.

USFWS (United States Fish and Wildlife Service). 2003. Endangered and threatened wildlife and plants: reconsidered finding for an amended petition to list the westslope cutthroat trout as threatened throughout its range. Federal Register 68(152):46989-47009.

Van Ham, E. H., M. H .G. Berntssen, A. K. Imsland, A. C. Parpoura, S. E. Wendelaar Bonga, and S.O. Stefansson. 2003. The influence of temperature and ration on growth, feed conversion, body composition and nutrient retention of juvenile turbot (*Scophthalmus maximus*). Aquaculture 217:547-558.

Warren, J. W. 1991. Diseases of hatchery fish. Sixth edition. U.S. Fish and Wildlife Service, Pacific Region.

Wedemeyer, G. A. 1996. Physiology of fish in intensive culture systems. Chapman and Hall, New York.

Welch, D. W., Y. Ishida, and K. Nagasawa. 1998. Thermal limits and ocean migrations of sockeye salmon (*Oncorhynchus nerka*): long-term consequences of global warming. Canadian Journal of Fisheries and Aquatic Sciences 55:937-948.

Winton, J. R. 2001. Fish health management. Pages 594-611 *in* G.A. Wedemeyer, editor. Fish hatchery management, second edition. American Fisheries Society, Bethesda, Maryland.

Young, M. K., and P. M. Guenther-Gloss. 2004. Population characteristics of greenback cutthroat trout in streams: their relation to model predictions and recovery criteria. North American Journal of Fisheries Management 24:184-197.

Zale, A. V. 1984. Applied aspects of the thermal biology, ecology, and life history of the blue tilapia, *Tilapia aureau* (Pisces: Cichlidae). Doctoral dissertation. University of Florida, Gainesville, Florida.

APPENDICES

APPENDIX A

EQUATIONS FOR CALCULATING OXYGEN SATURATION

To determine the oxygen saturation based on dissolved oxygen concentration and temperature the following equations were used (APHA 1992).

Equation 1

Determine the atmospheric pressure at altitude:

$\ln P = 5.25 * \ln (1- h/44.3)$, where h = altitude in km

Equation 2

Determine the equilibrium oxygen at nonstandard pressure (C_p):

$C_p = C' * P [(1-P_{wv}/P) (1- \Theta P) / (1-P_{wv}) (1-\Theta)]$

C_p = equilibrium oxygen concentration at nonstandard pressure, mg/L

C' = equilibrium oxygen concentration at standard pressure of 1 atm, mg/L

P = nonstandard pressure, atm

P_{wv} = partial pressure of water vapor, atm, computed from:

$\ln P_{wv} = 11.8571 - (3840.70/T) - (216{,}961.00/T^2)$

T = temperature, °K, where °K = °C + 273.15

$\Theta = 0.000975 - (1.426 * 10^{-5} t) + (6.436 * 10^{-8} t^2)$

t = temperature, °C

Equation 3

Determine oxygen saturation (%) based on dissolved oxygen concentration (DO):

$\%$ saturation = $(100 * \text{DOmg/L}) / C_p$

APPENDIX B

INGREDIENT COMPOSITION OF FEEDS

Table 8. Ingredient composition of the feed FinSTARTER 2014.

Ingredient	Content (g/100g)
Corn gluten meal	33.4
Wheat gluten	20.7
Krill	10.0
Wheat flour	13.8
Fish oil	10.2
Lysine HCL	1.5
Dical	2.7
Asta, pink	0.1
Vit premix	0.5
Lecithin	2.0
TMP 9920[a]	5.0
Stay-C	0.2
Total	100.0
Related information:	
Energy (gross heat/g)	5,444.0

[a] TMP 9920; proteinated mins; % - Cu 1, Zn 2.5, Mn 2.6, flour 93.9

Table 9. Ingredient composition of the feed Silver Cup Trout Feed.

Ingredient	Content
Protein	45 %
Fat	11 %
Fiber	3 %
Ash	12 %
Moisture	< 10 %
Digestible Energy	3,850.0 kcal/kg
Vitamin A	10,000.0 IU/kg
Vitamin D	500 IU/kg
Vitamin E	250 IU/kg
Related information:	
Energy (gross heat/g)	5267.0

Table 10. Ingredient composition of the feed Cutthroat Grower 0401.

Ingredient	Content (g/100g)
Yellow corn gluten	6.6
Wheat gluten	5.3
Fish meal	27.1
Liver meal	5.2
Krill	16.0
Wheat flour	12.6
Fish oil	9.3
Lysine HCL	1.4
Di-calcium phosphate	0.7
Vitamin premix	2.5
Lecithin	8.0
TMP 9920[a]	5.0
Trace mineral premix #3	0.1
Stay-C	0.2
Total	100.0
Related information:	
Energy (gross heat/g)	5909.0

[a] TMP 9920; proteinated mins; % - Cu 1, Zn 2.5, Mn 2.6, flour 93.9

* Moisture and Lysin added