Effects of Constant Temperatures and Diel Temperature Fluctuations on Specific Growth and Mortality Rates and Yield of Juvenile Rainbow Trout, *Salmo gairdneri*

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Specific growth and mortality rates of juvenile rainbow trout (*Salmo gairdneri*) were determined for 50 days at seven constant temperatures between 8 and 22°C and six diel temperature fluctuations (sine curve of amplitude ±3.8 deg C about mean temperatures from 12 to 22°C). For constant temperature treatments the maximum specific growth rate of trout fed excess rations was 5.12%/day at 17.2°C. An average specific mortality rate of 0.35%/day was observed at the optimum temperature and lower. At temperatures in excess of the growth optimum, mortality rates were significantly higher during the first 20 days of this experiment than the last 30 days. The highest constant temperature at which specific growth and mortality rates became equal (initial biomass remained constant over 40 days) was 23°C. The upper incipient lethal temperature was 25.6°C for trout acclimated to 16°C. A yield model was developed to describe the effects of temperature on the living biomass over time and to facilitate comparison of treatment responses. When yield was plotted against mean temperature, the curve of response to fluctuating temperatures was shifted horizontally an average 1.5 deg C towards colder temperatures than the curve of response to constant temperature treatments. This response pattern to fluctuating treatments indicates that rainbow trout do not respond to mean temperature, but they acclimate to some value between the mean and maximum daily temperatures. These data are discussed in relation to establishment of criteria for summer maximum temperatures for fish.

Key words: constant temperature, fluctuating temperature, specific growth rate, specific mortality rate, yield, lethal temperature, zero net biomass, rainbow trout, thermal criteria

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Nous avons déterminé les taux de croissance et de mortalité spécifiques de jeunes truites arc-en-ciel (*Salmo gairdneri*) sur une période de 50 jours à sept températures constantes entre 8 et 22°C et avec six fluctuations nycthémérales de températures (courbe de sinus d'amplitude de ±3.8 deg C autour de températures moyennes de 12 à 22°C). A des traitements à température constante, le taux de croissance spécifique maximal de truites arc-en-ciel pourvues d'un excédent de nourriture est de 5.12%/jour à 17.2°C. On observe un taux de mortalité spécifique moyen de 0.35%/jour à la température optimale et à des températures plus basses. A des températures supérieures à la température optimale de croissance, les taux de mortalité sont nettement plus élevés pendant les 20 premiers jours de cette expérience que pendant les derniers 30 jours. La température constante la plus haute à laquelle les taux de croissance et de mortalité sont égaux (la biomasse initiale demeurant constante pendant plus de 40 jours) est de 23°C. La température supérieure du début des mortalités est de 25.6°C pour des truites acclimatées à 16°C. Un modèle de rendement a été élaboré pour décrire les effets de la température sur la biomasse vivante avec le temps et pour faciliter la comparaison des réponses au traitement. Si l'on porte le rendement contre la température moyenne, la courbe de réponse à des températures variables se déplace horizontalement sur une moyenne de 1.5 deg C vers des températures plus froides par rapport à la

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courbe de réponse à des traitements à températures constantes. Ce genre de réponse à des traitements variables indique que les truites arc-en-ciel ne réagissent pas à la température moyenne mais s'acclimatent à une valeur quelconque entre les températures quotidiennes moyenne et maximale. Nous analysons ces données en regard de l'établissement de critères touchant les températures maximales estivales pour les poissons.

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VARIOUS thermal criteria have been proposed to protect aquatic organisms (Ohio River Valley Water Sanitation Commission 1956; U.S. Department of the Interior 1968). Generally these criteria take the form of a maximum temperature that shall not be exceeded in summer months, and provide a limitation on the rate of temperature change or rise above ambient receiving water temperature. More recently the NAS/NAE (1972) recommended short-term maximum temperatures that shall not be exceeded to prevent heat death of aquatic organisms. A lower weekly average temperature (one-third the range between the optimum temperature and the ultimate upper incipient lethal temperature) was recommended to safeguard growth and activity of the desired species.

These estimates of optimum temperatures for growth, activity, and survival are estimates derived from constant temperature exposures. To apply these criteria to natural surface waters, an implicit assumption is made that the "average" temperature in constant and fluctuating temperature regimes gives the same response and common estimate of the optimum end point. The validity of this assumption must be tested since many fish experience large temperature changes in their natural environment. Needham and Jones (1959) reported diel fluctuations of 12 deg C in a trout stream. Brett (1971) described the diel vertical migrations of underyearling sockeye salmon, *Oncorhynchus nerka*, in Babine Lake, British Columbia. Young sockeye move into 17°C surface waters to feed at dusk and retreat to 5°C hypolimnetic waters at daylight. Spigarelli (1975) observed that Lake Michigan salmonids moved into thermal plumes; they did not acclimate to, or stay at, discharge temperatures for extended periods of time. These observations illustrate the prevalence of daily temperature changes in the fish's life history.

The study reported here was conducted at the Environmental Research Laboratory-Duluth as part of the continuing program to determine thermal requirements of important freshwater fishes. The purpose was to test the assumption that the average fluctuating temperature causes the same biological response as an equal constant temperature. The long-term effects of various constant temperatures and diel temperature fluctuations on specific growth and mortality rates of juvenile rainbow trout, *Salmo gairdneri*, are described. Observed growth and mortality rates are converted into a yield model to facilitate comparison of responses to both test conditions. The observed results are discussed in relation to establishment of thermal criteria for fish.

Materials and Methods

Experimental Tanks

Twenty-six 19-ℓ aquaria were used. A drain at the downstream end maintained water depth at 12.5 cm and 10 ℓ in each tank. Duplicate tanks were placed in a circulating water bath (57 × 40.5 × 15 cm deep) for each fluctuating temperature treatment. Fluorescent lighting controlled by a timer switch, adjusted weekly, followed the seasonal day-length cycle at Duluth, Minn. Light intensity at the water's surface among all tanks averaged 3.6 lm (range 0.5–10.0 lm).

Water Supply

Test water was obtained directly from Lake Superior (Hokanson et al. 1973). The water flowed through the following plumbing materials: polyvinyl chloride (PVC) pipes and valves into a stainless steel headbox, rigid plastic manifold, and stainless steel coils with Tygon® tubing connectors. Flow rate was controlled by glass capillary tubing, and aerated water was directed to the bottom of the aquarium by a standpipe. Flow rates were maintained at 287 ± 23 ml/min (mean ± 1 SD, range 176–332 ml/min). Dissolved oxygen concentrations were maintained at 91 ± 6% (mean ± 1 SD, range 68–107%) of air saturation as determined from 275 grab samples. Duplicate tanks were sampled alternately 3 times weekly. The pH varied from 7.2 to 7.7 in 55 samples.

Temperature Control

Two systems controlled temperatures. The test water flowed through stainless steel heat exchangers immersed in the water bath for fluctuating temperature adjustment. Temperature was regulated by a controller that operated two solenoid valves and flow of hot or cold water into the bath. Temperature information was contained in a dual cam programmer with cam speed set for a 24-h revolution. Plexiglass cams were cut to achieve the desired diel temperature cycle (24-h periodicity). Nominal treatments were programmed to follow a sine curve with amplitude of ± 4 deg C about the daily mean temperature. The minimum temperature was set at 0400 CDT, and the maximum temperature was set at 1600 CDT. Constant temperature control was achieved by thermostatic regulation of immersion heaters in a headbox which dispersed test water through a manifold system to experimental chambers.

Thermal treatments consisted of seven constant temperatures between 8 and 22°C and six diel temperature fluctuations with daily means between 12 and 22°C (Fig. 1). Multipoint thermographs were calibrated daily with a mercury thermometer to the nearest 0.1 deg C. The reported grand average temperature of each treatment was computed from daily means derived from hourly values. The maximum variation (± 2 SD, N = 50) of grand average constant temperatures was ± 0.6 deg C and for fluctuating temperatures ± 0.8 deg C. The maximum of fluctuating temperatures was controlled within ± 0.9 deg C and the minimum within ± 1.8 deg C.

Experimental Fish

Approximately 4800 rainbow trout eggs were obtained from two Lake Superior stock females. The eggs, fertilized May 23, 1972, were reared in a Heath incubator and held initially at 6–8°C. Eyed embryos were held at 9–12°C until hatching on July 5, 1972. Total hatch of eggs was 87%.

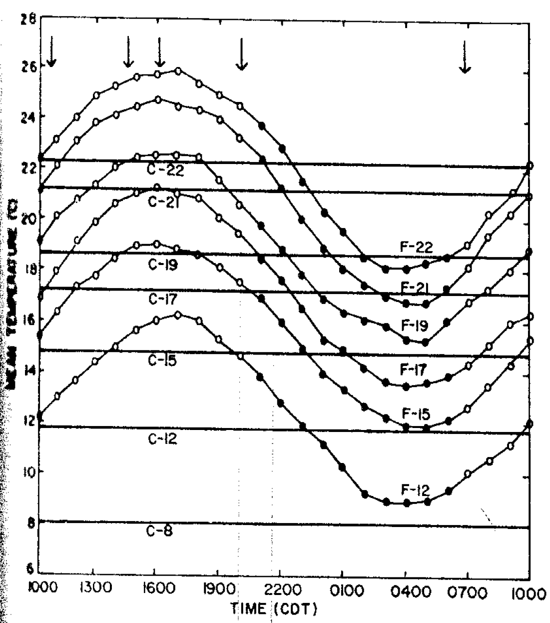

FIG. 1. Mean temperature plotted against time of day in hourly intervals for seven constant (C) and six diel fluctuating (F) treatments. Average amplitude of fluctuating treatments was ±3.8 deg C. ○, daylight hours and ●, hours of darkness for August 25. Arrows represent the median time of five ½-h feeding periods.

Alevins were transferred to two fiberglass tanks (150 × 27 × 35 cm deep) partially covered with black plastic. Each tank was provided with a water flow of 2–4 ℓ/min. Water temperature was increased gradually to 16°C when alevins commenced feeding on July 14. Trout were fed EWOS salmon and trout starter twice a day. Total survival of alevins was 89%.

Rainbow trout, acclimated to 16°C, were randomly distributed to each test tank between 1000 and 1315 CDT, August 1. Fifty trout were placed in each tank for a total of 100 per treatment. A random sample of an additional 50 trout averaged 30.2 mm (range 25.5–34.0 mm) total length and 245 mg (range 126–365 mg) wet weight. This common estimate of initial size applies to all treatment groups.

Growth

The trout were fed a varied diet consisting primarily of Glencoe Mills and Oregon Moist trout pellets. Fish were fed to satiation 5 times daily. Median feeding times were 0645, 1025, 1415, 1600, and 2000 CDT (Fig. 1).

Growth was followed for 50 days by placing a nine-celled sampling grid in each experimental tank every 10 days and selecting a random sample of about one-fifth the number of survivors (i.e. 10 or fewer fish). All sampled trout were preserved in 10% buffered formalin, blotted damp-dry, and wet weights were recorded to the nearest milligram.

Graphical inspection of the growth data showed that growth rates were constant over the 50-day period so that the weight at time t could be expressed as

$$W_t = W_o^{e^{g_j t}}$$

where g_j is the instantaneous growth rate for the jth experimental treatment group. However, since each treatment group started out with a common estimate of initial weight (W_o), treatments were not independent of one another. It is more efficient to recast the problem as one in multiple linear regression and force the regression lines through a common origin so that g_j could be estimated in the linearized form.

$$\ln W_t = \ln W_o + g_1 x_1 + g_2 x_2 + \dots + g_k x_k$$

where $x_j = t$ if the observations come from the jth treatment group and $x_j = 0$ for all other treatment groups. The parameters $\ln W_o$, g_1, g_2, ..., g_k and their standard errors were computed by using the BMDO3R multiple linear regression program described in Dixon (1970). Specific growth rates were calculated as $g_j \times 100$. Differences between regression coefficients were tested as suggested by Steel and Torrie (1960).

Mortality

The upper incipient lethal temperature of rainbow trout acclimated to 16°C was determined 10 days before start of the growth study to set the maximum for thermal treatment. Subsamples of 10 trout were transferred abruptly into each experimental tank set at constant temperatures of 24.5, 25.4, and 26.3°C. They were not fed for 24 h before transfer or during this test. Percentage survival was recorded, and the 24- and 96-h median tolerance limits (TL50) were derived for duplicate tests.

Deaths of rainbow trout were recorded daily during the growth experiment. Daily instantaneous mortality rates (i) were derived (Ricker 1958) at 5–10-day intervals because mortality rates were not constant. Mortality rates were assumed to be constant over short time intervals so that i was estimated using the linearized equation

$$(L_nN_o - L_nN_t)/(t_t - t_o)$$

where N_t is the number of survivors at time t and N_o is the initial number of fish at time t_o. Specific mortality rates were calculated as $i \times 100$.

Yield

Yield is defined as the living biomass at any given instant equivalent to the concept of standing crop of field investigations, or the change in stock size (Ricker 1958). A yield model was developed because we sampled fish without replacement, which does not allow one to follow the living biomass as a function of thermal treatments. A hypothetical test population was created, which allowed the observance of changes in living biomass over time and facilitated comparisons of both constant and fluctuating temperature regimens by evaluating one response (yield) instead of two (constant growth rate and variable mortality rate). The yield (Y_t) at time t may be expressed as

$$Y_t = N_t \cdot W_t = N_o \cdot P_t \cdot W_o e^{gt}$$

where N_t, N_o, W_t, and W_o were previously defined, and P_t is the probability of surviving until time t. The value of P_t is estimated by taking the product of the observed proportion of survivors over each 5–10-day sampling interval. The fate of a hypothetical test population is described given an initial number (N_o = 10,000) of rainbow trout and a common estimate of average initial weight (W_o = 227 mg). Yield (kilograms) was estimated at 5–10-day intervals for each treatment from corresponding estimates of growth and mortality rates.

Thermal Requirements

Growth

The growth of rainbow trout from sample estimates and fitted regression lines is illustrated in Fig. 2 for two treatments. Mean sample weights did not always fall below the fitted regression line in the later sampling intervals, and deviation from regression showed no consistent trend that could be attributed to sampling error. Therefore, growth rates were assumed to be constant for all treatments in the 50-day test period.

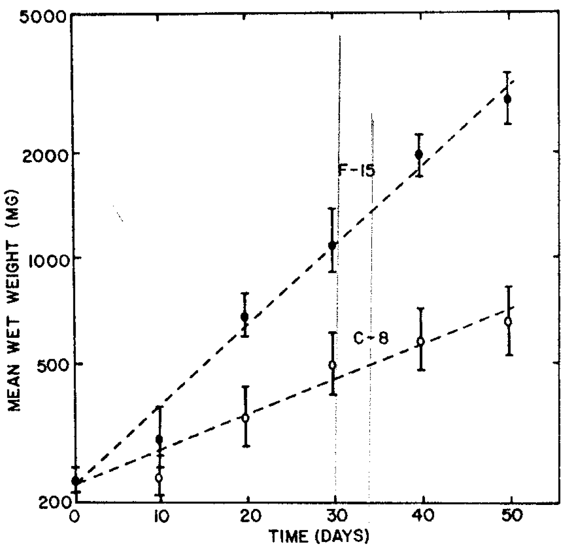

FIG. 2. Semilogarithmic plot of mean wet weights of rainbow trout (*Salmo gairdneri*) sampled at 10-day intervals for two treatments (C-8, constant 8°C; F-15, fluctuating temperature, mean 15°C). Duplicate samples from each treatment were similar; therefore, they were combined to provide a single estimate of mean weight. Vertical lines are 95% confidence limits of sample mean weights. Dashed lines are computed regressions for overall growth at each treatment. The common origin (W_o) for all treatments was 5.4237 (antilog 227 mg).

The data and computed specific growth rates for each treatment are presented in Table 1. The specific growth rates and corresponding 95% confidence limits are plotted against mean daily temperature in Fig. 3. Growth rates increased with increasing temperature, reaching a maximum of 5.12%/day at C-17 and 5.24%/day at F-15. The optimum range for growth for constant treatments was 17.2–18.6°C and for mean fluctuating treatments (amplitude ± 3.8 deg C), 15.5–17.3°C. Growth rates at higher temperatures declined with increasing temperature. Growth at the highest fluctuating treatment (F-22) was not significantly different from zero ($P < 0.10$, t-test). Although corresponding constant and fluctuating treatment means were not significantly different in the design of this study, the trends in treatment differences were consistent (Fig. 3). When the data were plotted the growth curve at fluctuating temperatures was generally shifted to the left of the growth curve at constant temperatures. Thus growth of rainbow trout appears to be accelerated at fluctuating treatments when the mean temperature is below the constant temperature optimum for growth and retarded by mean fluctuating temperatures above the constant temperature optimum.

TABLE 1. Specific growth rates of rainbow trout (*Salmo gairdneri*) for each treatment and tests of significant differences. The common initial size for all treatments was 245 mg, and the origin of the multiple linear regression was 5.4237 (antilog 227 mg).

Nominal[a] treatment	Mean temp (°C)	No. fish	Growth period (*days*)	Specific growth rate (%/*day*) Slope	±SD
C-8	8.1	94	50	2.29	0.148
C-12	11.8	65[b]	50	3.66	0.172
C-15	14.8	86	50	4.36	0.152
C-17	17.2	85	50	5.12[c]	0.153
C-19	18.6	94	50	4.83[c]	0.148
C-21	21.2	61	50	4.49	0.169
C-22	22.3	37	50	3.94	0.194
F-12	12.5	86	50	4.23	0.154
F-15	15.5	86	50	5.24[c]	0.155
F-17	17.3	79	50	4.86[c]	0.164
F-19	19.1	81	50	4.58	0.157
F-21	21.0	18	40	4.48	0.274
F-22	22.2	5	7	−2.12[d]	2.642

[a]Constant (C) and fluctuating (F) treatments.

[b]One aquarium overflowed and 28 trout escaped on August 15, 1972.

[c]Slope not significantly different from optimum at F-15 ($P < 0.05$, *t*-test).

[d]Slope not significantly different from zero ($P < 0.10$, *t*-test).

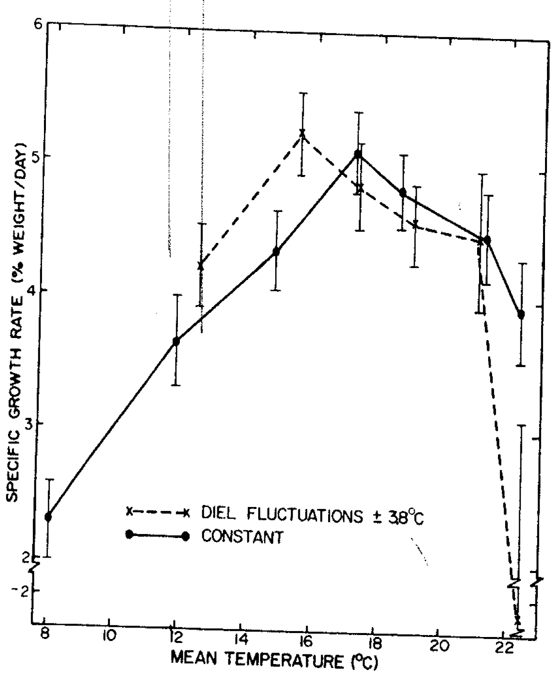

FIG. 3. Effect of constant temperatures and diel temperature fluctuations on specific growth rate of rainbow trout fed excess rations. Vertical lines are 95% confidence limits of the regression coefficient.

MORTALITY

The upper incipient lethal temperature (101 h) of the sample of rainbow trout acclimated to 16°C was 25.6°C. The mean 24-h TL50 was 25.7°C. All trout survived at 24.5°C for 101 h. The upper incipient lethal temperature was exceeded for 3 h daily only in the highest fluctuating treatment (F-22).

The average specific mortality rate was 0.36%/day (range 0–2.11%/day) at temperatures within the optimum range and lower (Table 2). Death occurred primarily among smaller individuals and apparently was related to stress from social interactions. Only at treatments in excess of the physiological optimum did we observe increased mortality rates during the first 20 days of this experiment. Mean lengths of dead fish at higher temperatures were similar to those of surviving fish. Mortality rates at temperatures above the respective optima were not constant, but generally reached a maximum in the first 10 days of the study. Thereafter mortality rates declined reaching an average for all treatments of 0.18%/day (range 0–1.25%/day) after 20 days. In the highest fluctuating treatment (F-22), mortality rate was high, 42.8%/day, during the first 7 days; consequently the fish did not survive to the end of the first sampling interval. Mortality rates generally were higher among the fluctuating treatment groups than among the corresponding constant treatment groups for the same time period. Apparently mortality rates of rainbow trout at elevated temperatures were also influenced by the thermal experience above the mean daily temperature.

YIELD

Based on the yield model, yield of 10,000 rainbow trout (average initial wet weight 227 mg) fed an excess daily ration was computed and plotted against time for three treatments (Fig. 4). The population maintenance level represents the initial weight of 2.27 kg for this hypothetical population and is the expected yield whenever the specific growth and mortality rates are equal over a given time interval. The response to C-19 represents a fast growing population with good survival rates. After 40 days, 9429 fish survive with an average wet weight of 1562 mg. The response to C-8 represents a slow-growing population with good survival rates. After 40 days, 9468 fish survive with an average wet weight of 567 mg. The yield at F-21 declined to a low of 0.69 kg after 10 days because the specific mortality rate exceeds the growth rate during this time interval. The yield increased to 1.96 kg after 40 days when

TABLE 2. Specific mortality rate (percent per day) of rainbow trout for different thermal treatments and time intervals.

Nominal[a] treatment	Mean temp (°C)	Time interval (*days*)						
		0–5	5–10	10–15	15–20	20–30	30–40	40–50
C-8	8.1	0.20	0.20	0	0	0.34	0	0.51
C-12	11.8	0	0.21	0	0.41	0	0	1.25
C-15	14.8	0	0.23	0.30	0	0	0	0
C-17	17.2	0.66	0.45	0.60	0.31	0	0	0
C-19	18.6	0.61	0	0.21	0	0.18	0	0
C-21	21.2	2.49	4.58	1.19	0	0.54	0	0
C-22	22.3	3.61	13.62	1.33	0.70	0	0	0.57
F-12	12.5	0	0.21	0.56	0.57	0.20	0.31	0
F-15	15.5	0	1.06	0.85	0	0.19	0.29	0
F-17	17.3	0.63	0.66	2.11	0.32	0	0	0.65
F-19	19.1	0.84	1.57	0.60	0	0.20	0.31	0
F-21	21.0	11.49	21.29	4.46	0	0.74	0	—
F-22	22.2	46.05	34.66	—	—	—	—	—

[a]Constant (C) and fluctuating (F) treatments.

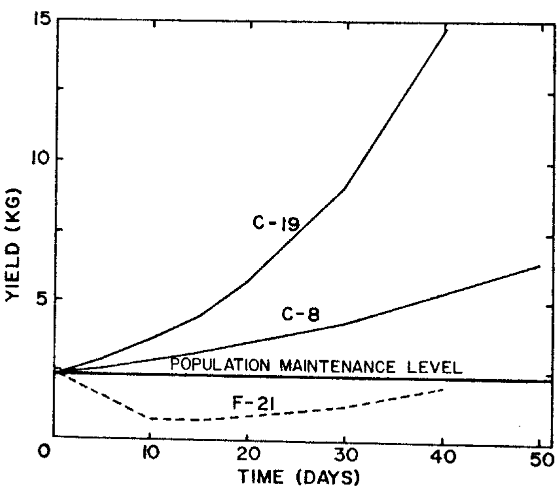

FIG. 4. Yield of 10,000 rainbow trout (initial wet weight 227 mg) at 10-day intervals for three treatments. The population maintenance level represents the initial weight of the test population and the expected yield when the specific growth and mortality rates are equal.

the mortality rate declined. This population consisted of 1442 fish with an average wet weight of 1360 mg.

The yield of this hypothetical rainbow trout population at different time periods and thermal treatments is shown in Table 3. To compare responses to constant and fluctuating treatments, the 40-day yield is plotted against mean daily temperature in Fig. 5. A maximum yield of 15.87 kg was produced at C-17, and 15.96 kg at F-15 after 40 days. Yields declined at lower temperatures because of reduced growth rates and at higher temperatures because of reduced growth rates and increased mortality rates. When the specific growth rate equals the specific mortality rate the population experiences no change in weight (zero net biomass gain). The maximum temperature at which a population of rainbow trout can maintain its initial weight for 40 days is a constant temperature of 23°C and a fluctuating mean temperature (± 3.8 deg C amplitude) of 21°C. In graphic presentation the yield in response to fluctuating temperatures was generally shifted towards colder temperatures than the curve of response to constant temperatures. A graphical estimate of the average shift was 1.5 deg C which was constant for all time intervals.

Implications for Thermal Criteria

Development of water quality criteria involves the identification of graphical end points that have particular physiological and ecological significance (Brett 1971; NAS/NAE 1972). Criteria are not designed to produce maximum growth of fish, but to protect a balanced and indigenous fauna of direct importance to man. The physiological optimum and upper incipient lethal temperature of a species is used directly in derivation of summer-limiting temperatures for aquatic life. Measures of physiological performance and activity in constant temperature studies point to an optimum of 16–18°C for rainbow trout (Lichtenheld 1966; Lavrovsky 1968; McCauley and Pond 1971; Dickson and Kramer 1971). The upper incipient lethal temperature (25.6°C) of this stock of rainbow trout was almost identical to those reported for other stocks of this species (Bidgood

TABLE 3. Yield (kilograms) of 10,000 rainbow trout (initial wet weight 227 mg) for different thermal treatments and time periods.

Nominal[a] treatment	Mean temp (°C)	Time (*days*)						
		5	10	15	20	30	40	50
C-8	8.1	2.52	2.79	3.13	3.51	4.27	5.37	6.42
C-12	11.8	2.72	3.24	3.89	4.57	6.59	9.51	12.10
C-15	14.8	2.82	3.47	4.24	5.28	8.16	12.62	19.50
C-17	17.2	2.83	3.58	4.49	5.71	9.62	15.87	26.48
C-19	18.6	2.80	3.56	4.49	5.71	9.09	14.73	23.87
C-21	21.2	2.51	2.49	2.94	3.68	5.46	8.56	13.41
C-22	22.3	2.31	1.42	1.62	1.90	2.83	4.19	5.87
F-12	12.5	2.80	3.42	4.11	4.94	7.39	10.94	16.70
F-15	15.5	2.95	3.63	4.52	5.87	9.73	15.96	26.94
F-17	17.3	2.80	3.45	3.96	4.97	8.08	13.13	20.01
F-19	19.1	2.73	3.18	3.88	4.87	7.55	11.58	18.30
F-21	21.0	1.60	0.69	0.69	0.86	1.25	1.96	—
F-22	22.2	0.20	0.10	—	—	—	—	—

[a]Constant (C) and fluctuating (F) treatments.

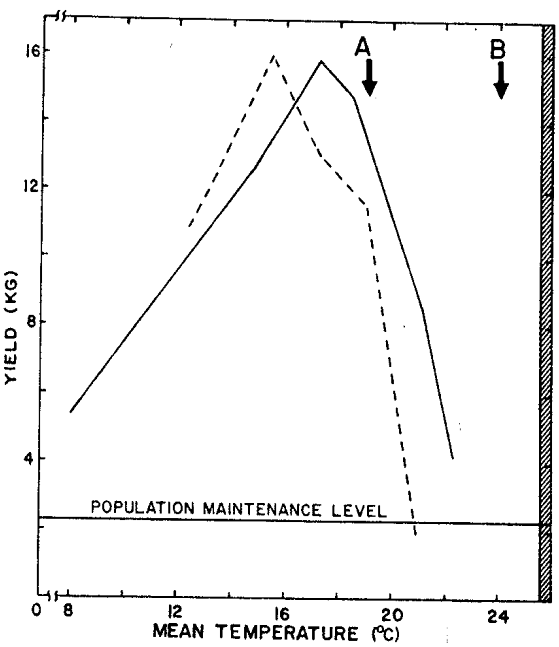

FIG. 5. Effect of constant temperatures (solid line) and diel temperature fluctuations (dashed line) on the 40-day yield of 10,000 rainbow trout (initial mean weight 227 mg). The shaded area represents the zone of resistance in excess of the upper incipient lethal temperature. Recommended water-temperature criteria for rainbow trout are defined by arrows (National Academy of Sciences 1972): (A) maximum weekly average temperature and (B) short-term maximum for survival.

and Berst 1969; Alabaster 1964). Mortality rates of rainbow trout fed to satiation increased significantly above the physiological optimum temperature. Other authors have also reported increased mortality rates in growth experiments at apparently sublethal temperatures (Strawn 1961; Allen and Strawn 1968; Brett et al. 1969). Mortality rates of rainbow trout were higher than one might expect as 100% survived temperatures within 1 deg C of their upper incipient lethal temperature in short-term tests where fish were not fed 24 h before or during exposure to lethal temperatures.

Temperature criteria recommended for rainbow trout waters by the NAS/NAE (1972) are indicated by arrows in Fig. 5. The maximum weekly average temperature (19°C, point A) is set at one-third of the range between the optimum temperature reported in earlier studies and the ultimate upper incipient lethal temperature. The maximum temperature (24°C, point B) for short-term survival is based on a 24-h TL50 minus 2 deg C for fish acclimated to the mean temperature (19°C). This 2 deg C "safety factor" was estimated by Coutant (1973). Care should be exercised in the application of a formula derived from short-term entrainment studies to the long-term effects of a modified temperature regime over a wider area of the receiving water. The harvestable yield depends on environmental effects on population growth and mortality over time. The maximum temperature at which a rainbow trout population can be expected to maintain its weight for 40 days was a constant temperature of 23°C and a fluctuating mean temperature (± 3.8 deg C) of 21°C. The temperature range from 21 to 23°C has also been reported by various authors as representing the upper limits of rainbow trout distribution (Burton and Odum 1945; Horak and Tanner 1964; Pierce 1966; McCauley and Pond 1971). Apparently temperatures selected by rainbow trout in their natural habitat are below the upper zero net gain value whereby a sustained yield will occur. The criteria of the National Academy of Sciences applied to our test conditions would allow a 13% reduction from maximum 40-day yield for constant temperature exposures and a 27% reduction from maximum yield for fluctuating temperature regimes.

Numerous studies and observations give conflicting views on effects of fluctuating temperatures. The limitation on rate of temperature change in thermal criteria is based, in part, on the knowledge that death of aquatic organisms can occur if they are transferred from one water temperature to another too quickly without suitable acclimation. Most fish species, however, can survive sudden temperature changes up to 15–18 deg C within their respective thermal tolerance limits (Hart 1947, 1952). Sublethal temperature changes have often been labeled stressful by physiologists who have observed changes in tissue function (Hughes and Roberts 1970; Wedemeyer 1973; Heinicke and Houston 1965; Coutant 1973). It is not clear whether the measured changes from short-term experiments were deleterious to fish or were merely adaptive changes in the process of thermal acclimation. Brett (1971) postulated that the diel vertical migrations of salmon to colder waters after feeding in warm surface waters may be an energy conservation mechanism in food-limited environments. In the present study, diel temperature fluctuations within the range normally selected by rainbow trout (i.e. physiological optimum and lower) were beneficial to growth. Fluctuations at mean temperatures above the physiological optimum, however, are more harmful than currently estimated from constant temperature exposures in most laboratory studies.

The observed response to fluctuating temperatures can be attributed to the differential rates of thermal acclimation to high and low temperatures. Body temperatures of alewives, *Alosa pseudoharengus*, equilibrated faster to increasing than decreasing temperature changes (Spigarelli et al. 1974). Brett (1946) acclimated fish to increasing and decreasing temperatures for various time periods. By noting changes in their lower and upper lethal temperatures, he found that they acclimated faster to higher than to lower temperatures. Heath (1963) acclimated cutthroat trout, *Salmo clarki*, to constant and cycled temperatures (10–20°C). Maximum tolerance to high temperature was observed on a 24-h acclimation cycle, and the lethal temperature of cycled trout suggests that they acclimate to the peak (20°C) rather than to the mean temperature. Fish acclimated to diel temperature fluctuations compensate for both warm and cold temperatures simultaneously and have an increased range of thermal tolerance (Feldmeth et al. 1974). Amargosa pupfish, *Cyprinodon nevadensis*, acclimated to a diel temperature cycle from 15 to 35°C had critical thermal maxima equal to those acclimated to constant 35°C and had critical thermal minima equal to those acclimated to constant 25°C (the mean of the cycle). The critical thermal minimum was changed more slowly reaching its lowest point after 22 days of acclimation compared to the maximum which was reached in less than 7 days.

The convention of plotting biological response against mean temperature has not been justified by this study. The observed shift (1.5 deg C) in response between constant and fluctuating treatments (± 3.8 deg C) is consistent between all treatments and time intervals although this difference may not be statistically different in the design of this study. The amount of shift in response to fluctuating treatments is a direct function of the amplitude of the diel cycle (Environmental Research Laboratory-Duluth, unpublished data). These data trends indicate that a shift in response greater than 1 deg C may occur when diel temperature variations exceed ± 2 deg C. A method to correct this difference in response is desirable before we can validly apply results from simplified laboratory experiments conducted at constant temperatures to field conditions with large diel temperature changes or habitat gradients. These results indicate that instantaneous response may lag behind the instantaneous temperature experience and that temperatures above the mean exert greater influence than below. As a hypothesis we will assume that under a fluctuating temperature regime an organism will acclimate to some function of its temperature exposure over time (i.e. between the median or 50th percentile and the maximum or 100th percentile of the fluctuating temperature duration). This hypothesis should be tested for different species and temperature cycle periodicities and amplitudes before one can derive the specific mathematical function to predict fish response to irregular temperature cycles.

Derivation of temperature standards must take into account the magnitude of temperature variation of the natural environment to which these criteria apply. From the foregoing discussion, the mean weekly temperature should be conservatively retracted towards the optimum end point derived from constant temperature studies (i.e. 17°C for rainbow trout) to achieve the desired degree of protection recommended by the NAS/

NAE (1972) where diel temperature variations exceed ± 2 deg C. The temperature at which the specific growth and mortality rates become equal in constant temperature experiments (i.e. upper zero net biomass gain for rainbow trout is 23°C) should set the maximum thermal limit at any time or place where a sustained yield is desired. These end points were estimated from maximum ration studies. These limits should be further retracted towards colder temperatures to safeguard natural populations receiving less than full rations (Brett et al. 1969; Fry 1972). The specific mathematical function that describes acclimation to fluctuating treatments should be derived in future studies so that results of constant temperature studies can be universally applied in setting water temperature standards.

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