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TEMPERATURE BEHAVIOR OF THE BLUESPOTTED SUNFISH ENNEACANTHUS GLORIOSUS (HOLBROOK), WITH AN EVALUATION OF THE INTERPRETATION OF THERMAL BEHAVIOR DATA[1]

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ABSTRACT: The bluespotted sunfish, *Enneacanthus gloriosus* (Holbrook), had a final temperature preferendum of 31.5 C. Fish acclimated to 30 C avoided 36 C. The analysis of acute temperature preference data and the relationship between final temperature preferendum and upper avoidance temperatures are discussed.
(KEY TERMS: bluespotted sunfish, *Enneacanthus gloriosus*; temperature preference; final temperature preferenda; analysis of acute preference data.)

INTRODUCTION

The bluespotted sunfish, *Enneacanthus gloriosus* (Holbrook) was confined originally to the lowlands of the Atlantic slope between southern New York and extreme western Florida (Lee and Gilbert, in press). Subsequently, it was introduced into Lake Ontario (Werner, 1972) and the upper Susquehanna River drainage (Denoncourt, *et al.*, 1975). Casterlin and Reynolds (1979, 1980) reported on the thermoregulatory behavior and diel activity patterns of individuals from Harveys Lake in northeastern Pennsylvania, while Reynolds and Casterlin (1980) discuss the temporal relationships between activity and thermoregulatory pattern in this species. The purpose of this paper is to report temperature preference and avoidance data on specimens of the bluespotted sunfish collected from Maryland's Eastern Shore, and to discuss the analysis and interpretation of acute temperature behavior responses.

METHODS AND MATERIALS

One- to two-year old bluespotted sunfish were collected from Mason Creek (tributary of Choptank River) near Bridgetown, Maryland, in late summer and fall in 1977 and 1978. All fish were collected by seine and immediately transported to holding tanks located in the Appalachian Environmental Laboratory, Frostburg, Maryland. Fish were held for three days at their capture temperature, then acclimated to one of the following temperatures at a rate which did not exceed 1^{o}C/day; 6, 12, 18, 24, 30, and 33 C. Fish were held at their respective acclimation temperatures (± 1^{o}C) for a minimum of five days before testing. Natural photoperiod was maintained using Vitalites, and constant aeration maintained oxygen levels between 90 and 100 percent saturation.

Preference Tests

Temperature preference trials were conducted in a horizontal trough patterned after the description of Meldrim and Gift (1971). The aluminum trough (3.6 x 0.2 x 0.25 m) used in these experiments was described by Hall, *et al.* (1978a), and Hall, *et al.* (1978b), and provided a temperature gradient of approximately 16^{o}C, which ranged above and below the respective acclimation temperature.

Six fish were tested individually at each acclimation temperature and introduced into the trough at the acclimation temperature. Following a 40-minute orientation period, the water temperature at the organisms position was recorded each minute for 20 minutes. The mean of the 20 observations was deemed the acute preferred temperature for that particular test (Stauffer, *et al.*, 1975).

The relationship between acclimation temperature and preference temperature was explored via linear regression (Stauffer, *et al.*, 1976), quadratic equation (Cherry, *et al.*, 1977), log transformation, and third and fourth degree polynomials. Stepwise regression analysis was used to determine which model best described the data. The best model was solved for the point at which acclimation temperature equaled preference temperature. This was deemed the final temperature preferendum (Fry, 1947).

Avoidance Tests

After analysis of the temperature preference data, avoidance trials were conducted at the acclimation temperature closest to the final temperature preferendum. The temperature avoidance trials were conducted using the avoidance apparatus described by Melisky, *et al.* (1978), which was modified from that used by Meldrim and Gift (1971), Cherry, *et al.* (1977), and Stauffer, *et al.* (1976). The unit consisted of two subtroughs which drain at the center. Water from two circulating

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water baths entered at opposite ends of each subtrough, which allowed the investigator to provide a fish with a choice of two temperatures. To correct for positional effects, water from the same bath supplied opposite ends of the two subtroughs. The avoidance unit was enclosed in a small room and fish movement was monitored via one-way mirrors. Constant illumination was supplied by Vitalites.

One fish (total of six trials) was placed in each subtrough with the water from both baths set at the acclimation temperature. After an initial 40-minute orientation period, the amount of time fish spent on each half of each subtrough was recorded for 10 minutes. At the end of 10 minutes, the side of trough labeled "higher temperature side" (Table 1) was raised 3°C above the acclimation temperature. Fish movement was again monitored for 10 minutes, and the number of seconds spent on the "lower temperature side" was recorded. The experiment was continued by raising both water baths 3°C and repeating the monitoring procedure. This process was continued until the lower temperature side and the upper temperature side were at 39 C and 41 C, respectively.

TABLE 1. Mean Time the Bluespotted Sunfish (N = 6) Spent on Each Side of the Avoidance Trough When Acclimated to 30 C.

Lower Temperature Side		Higher Temperature Side	
Temperature	Time	Temperature	Time
30*	366.8	30	233.2
30	381.8	33	218.2
33	503.7	36**	96.3
36	577.7	39	22.9

*Control.
**Avoidance temperature.

A two-way, factorial analysis of the variance table was constructed using the six fish and the higher of the two temperature choices as classes as described by Stauffer, *et al.* (1976). The time in seconds the fish spent on the lower temperature side was recorded in each block of the table. Since temperature intervals were a "fixed effect" and the particular fish groups were "random effects" the mean square for the fixed temperature effect was tested by the interaction term "fish x temperature interval." The mean square and degrees of freedom for the interaction term was then used to perform a Duncan's multiple range test for the fixed effect. The point at which significantly ($P \leqslant 0.05$) more time was spent on the lower temperature side during an experimental run than on the lower temperature side during the control run was deemed the avoidance temperature.

RESULTS

Bluespotted sunfish preferred temperatures higher than their acclimation temperatures between 6 and 30 C (Figure 1). Stepwise regression analysis indicated that the data were best explained by the quadratic equation: $P = -0.0158\ A^2 + 1.014\ A + 15.25$, where P = preferred temperature and A = acclimation temperature. The above equation explained 80 percent of the raw data (N = 36). The final temperature preferendum was determined to be 31.5 C by solving the above equation for the point where P = A.

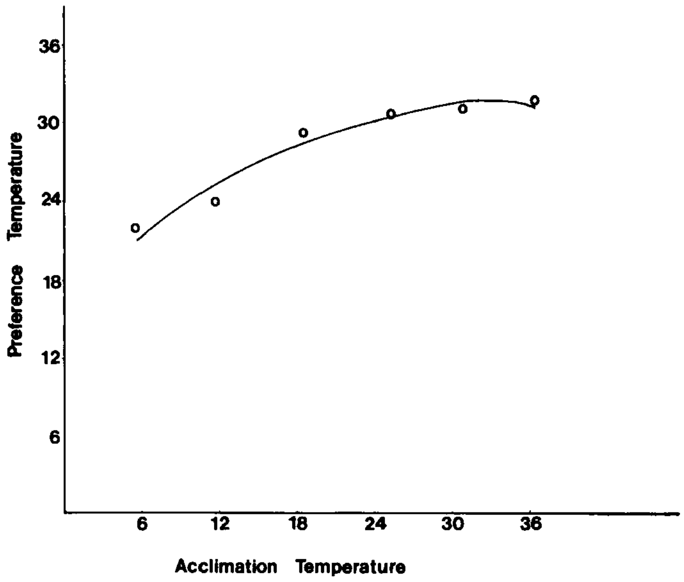

Figure 1. Relationship Between Temperature Preference and Acclimation Temperature for the Bluespotted Sunfish, *Enneacanthus gloriosus* (P = preference temperature; A = acclimation temperature).

Fish acclimated to 30 C avoided 36 C water (Table 1). During the avoidance trials, all fish died or lost equilibrium when water temperatures in both sides of the unit were 39 C and higher. Two fish lost equilibrium when exposed to 36 and 39 C near the end of the trial. Therefore, although no specific tests were conducted, one can estimate that the critical thermal maximum is probably between 36 C and 39 C. If given a choice, the bluespotted sunfish will avoid 36 C water.

DISCUSSION

With the exception of a paper by Casterlin and Reynolds (1979), and Reynolds and Casterlin (1980) no additional information concerning temperature regulated behavior or thermal tolerance of this species could be found. Casterlin and Reynolds (1980) reported a final temperature preferendum of 28.5 C ± 0.2°C for populations from Harveys Lake, Pennsylvania, and indicated that there were no significant differences between day and night responses. Differences between the values of the final temperature preferendum in the two studies may be related to differences in techniques or to population differences. [Casterlin and Reynolds (1970), used an electronic shuttle box in which the fish controlled the water temperature.] Through the use of the electronic shuttle box, the final temperature preferendum is measured directly; while the techniques used in this study measured acute responses, so that the final temperature preferendum must be estimated by extrapolation. However, the relatively close agreement between the two studies would appear to indicate that the final temperature preferendum is a real and measurable phenomenon; especially considering the small standard deviation reported by Casterlin and Reynolds (1979) and the fact that the quadratic equation explained 71 percent of the raw data reported in this study.

These data are particularly interesting in light of a recent study by Hall, *et al.* (1978b). Hall, *et al.* (1978b), found that northern populations of white perch demonstrated a cooler final temperature preferendum than southern populations. The cooler final temperature preferendum of the bluespotted sunfish from Harveys Lake may be related to genetic differences which are associated with geographical variations.

Recently, it has been debated (see Panel Discussion in Richards, *et al.*, 1977) whether acute temperature preference data should be predicted by linear or cuvilinear techniques, or simply by visual estimation. Visual estimation is simpler and many times provides a good estimate of the final temperature preferendum when used in conjunction with a theoretical line with a slope of 1 C. Fry (1947) defined final temperature preferendum as the point where the preference-acclimation curve intersected a line with a slope of 1. Solving a model for the point where P = A has the same effect. However, there are obvious drawbacks to the use of visual estimates if data from different populations, or data generated using different techniques are to be statistically compared. Additionally, investigator bias may be inadvertently introduced when estimates are used. Simple models of the form P = m A + b have been used (Stauffer, *et al.*, 1975, 1976), in the past. For some species, these models are particularly useful, and they provide a means of comparing different data sets for those portions of the curve with a constant slope. However, these models may tend to overestimate the final temperature preferendum, since the slope of the response curve of many species tends to decrease at the higher acclimation temperatures (see Reynolds and Casterlin, 1979, 1980). Based on numerous laboratory studies, it appears that the quadratic equation most often best describes the relationship between acclimation temperature and preferred temperature. In an attempt to achieve the best fit, data should also be examined through the use of higher order polynomials and a technique such as stepwise regression which can determine when the addition of another variable into the model does not significantly explain more of the data. Once the "best" model is found, it may be further tuned through the use of nonlinear least squares procedures.

The relationships between the avoidance and preference data should be noted. As stated, avoidance experiments were conducted at the acclimation temperature (30 C) closest to the final temperature preferendum (31.5 C). When fish were given a choice between 30 C and 33 C, no detectable avoidance behavior occurred. However, when offered a choice between 33 C and 36 C, fish spent significantly more time at 33 C. Therefore, it would appear that for the bluespotted sunfish, the upper avoidance temperature is slightly higher than the final temperature preferendum. Similar trends can be found for most North American species if one refers to the compilation of preferences and avoidance data prepared by Coutant (1977).

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