A DIURNALLY FLUCTUATING THERMAL SYSTEM FOR STUDYING THE EFFECT OF TEMPERATURE ON AQUATIC ORGANISMS

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A diurnally fluctuating thermal system for studying the effect of temperature on aquatic organisms[1]

Abstract—An open, flow-through system with diurnally fluctuating temperature regimes has been designed to study growth and developmental dynamics of benthic macroinvertebrates. Animals are kept in five sets of plastic troughs and trays arranged in a longitudinal series and separated by polyethylene-lined warming pools. Five diurnally oscillating temperature regimes of various magnitudes are produced depending on pool size. The magnitudes of the temperature pulse are quite predictable for each regime.

Fluctuating temperatures are characteristic of most natural lotic systems. This diurnal and seasonal thermal diversity has been physically characterized (Macan 1958; Edington 1966), but its biological and ecological implications have been poorly evaluated. Past research on aquatic species has emphasized constant temperature laboratory systems. Few, if any, studies can be cited where species were maintained under quasi-natural conditions with respect to substrate, flow, light, and food resources and in which fluctuating temper-

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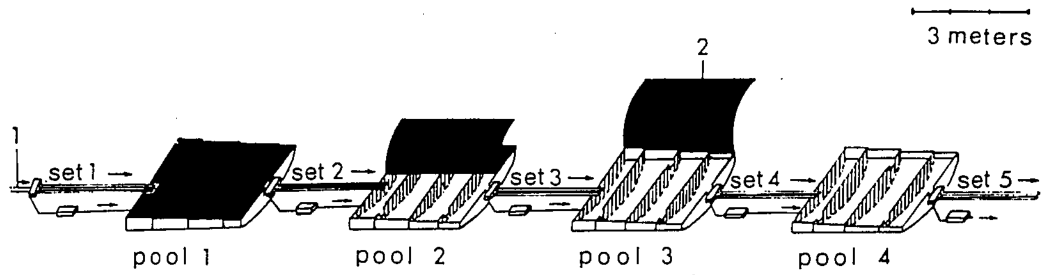

Fig. 1. Overview of entire system with polyethylene covers gradually removed to illustrate pool construction. 1—Spring overflow pipe; 2—black polyethylene cover.

ature was the important experimental variable.

Warren and Davis (1971) studied fish growth at elevated oscillating temperatures by the addition of artificial heat to ambient stream water; Edington and Hildrew (1973) observed caddisfly growth and respiration in a recirculated fluctuating system produced by electric heating and cooling devices. In contrast, the system described here provides an open, flow-through design with fluctuating temperature regimes of variable magnitudes. The capture and transfer of solar heat produces the fluctuating water temperatures. The system was designed for use with benthic macroinvertebrates, particularly insects.

The experimental system was constructed in an open meadow in Chester County, Pennsylvania. Water (11.5–13.0°C) from a spring was piped into ten plastic (PVC) troughs (ea. 3 m × 14.5 cm × 8.5 cm max depth), positioned as a longitudinal series of five sets of two. Thermal heterogeneity among sets was provided by holding pools between successive sets of troughs (Fig. 1). The pools (ea. 3 m × 2 m × 50 cm max depth) were made by widening, deepening, and damming an existing water channel. A lining of 4-mille black polyethylene retained about 3,600 liters of water in each pool; a layer of the same plastic was spread over the surface in direct contact with the water and fastened to boards at the perim-

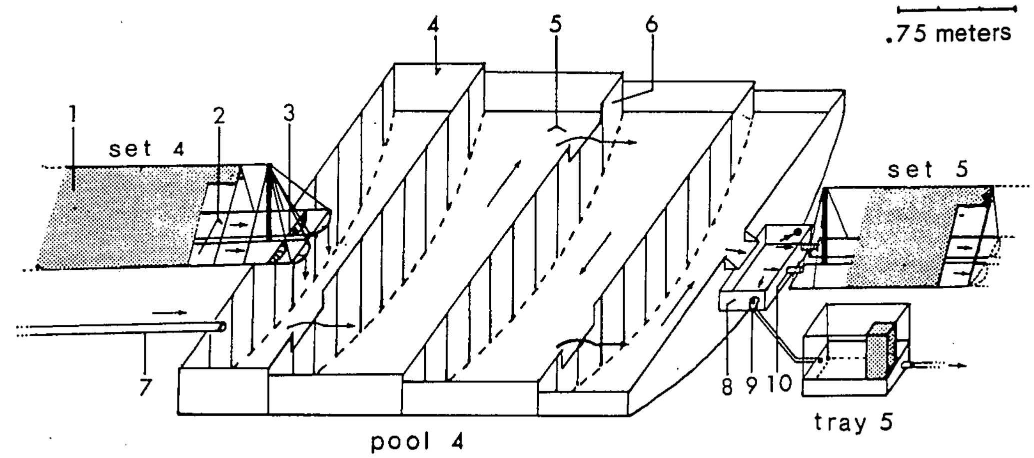

Fig. 2. Expanded view of trough set 4, pool 4, and trough set 5. 1—Mosquito netting; 2—troughs; 3—screen; 4—perimeter board; 5—polyethylene lining; 6—baffle; 7—PVC pipe; 8—plastic reservoir; 9—spigot tap; 10—overflow pipe.

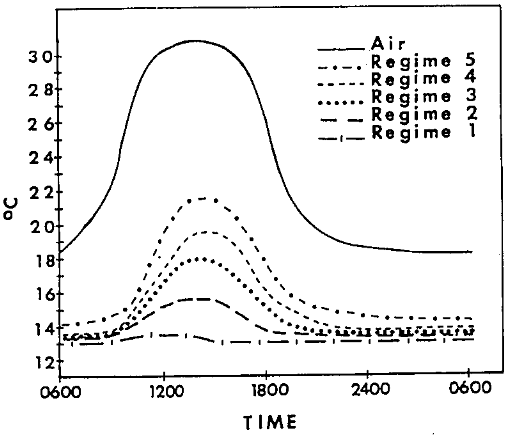

Fig. 3. Continuous 24-h recording of air and water temperatures for each experimental regime during a typical summer day.

Table 1. Average thermal conditions of the five experimental regimes during a 6-week summer period, 1975. Standard deviation of the mean is in parentheses.

Regime	Maximum	Minimum	Avg change
1	12.98(0.52)	12.51(0.19)	0.46
2	14.87(1.05)	12.87(0.22)	2.00
3	17.40(1.35)	12.74(0.34)	4.66
4	18.55(1.58)	12.81(0.40)	5.74
5	20.30(1.99)	13.38(0.77)	6.92

eter. Four wooden baffles, positioned before lining the pool, provided maximum circulation. The average retention time for water passing through a pool between two trough sets was 75 min. Water leaving a given pool entered a plastic reservoir (45 × 24 × 12 cm) equipped with two 1.5-inch (3.7 cm) OD overflow pipes and two spigot taps (Fig. 2). Overflow pipes supplied the next set of troughs, while the taps channeled water into plastic rearing trays. Baglike screens filtered the water as it left each trough and tray.

Warm summer air and direct exposure to sunlight provided natural heating for the system. The large black surface and long retention time of each pool were conducive to capture and transfer of solar heat. Daily excursions of solar intensity and air temperature produced diurnally oscillating temperature regimes (Fig. 3). Thermal maxima and minima were at times of day approximating natural conditions in the White Clay Creek. First order tributaries to the White Clay system originate as spring outflows (11.5–13.0°C) and temperature increases with distance downstream in summer. The maximum temperature of trough set 5 was typical of local fourth order streams. The remaining trough sets represented thermal conditions at points along the natural first through fourth order temperature gradient. The magnitudes of the temperature pulses were quite predictable for each regime (Table 1).

Temperature regimes similar to these can be obtained with much smaller holding pools if the black cover is removed and the water directly exposed to air and sunlight. This may result, however, in rapid, uncontrollable algal blooms and colonization of pools by undesirable animals. Such small, open systems can be recommended only for short term studies (<7 days). Open pools are rejuvenated by extensive cleaning or the addition of a new liner. Pool liners can be replaced without interrupting an experiment by laying polyethylene on the pool surface and allowing water to flow into it: old water is forced out as the new liner fills. This procedure leaves most of the algal mat, leaves, etc. trapped between the old and new liner rather than flowing into the experimental trough.

Maintenance during an experiment includes removal of pool surface water after occasional rains and cleaning of screens at the outflow of troughs and trays. Daily cleaning of screens is required when animals are small (200–400-μm mesh), but several days may elapse between cleanings as mesh size is increased. Occasional shading may be necessary if algal production becomes excessive.

The system described has operated successfully for 3 years. No significant differences were observed for any of the following chemical constituents: nitrates, nitrites, ammonia, phosphates, chlorides, iron, manganese, potassium, sodium, pH, or total alkalinity; this facilitates interpreting temperature effects. Detailed 24-h studies of dissolved oxygen show maximum differences at dawn when all five trough sets were at their lowest temperature with only 2°C range between them. Average dissolved oxygen concentrations at 0600 hours were 8.70, 8.85, 9.10, 9.40, and 9.55 mg liter^{-1} for sets one through five. These differences, which can be explained by accrual of oxygen during exposure in the troughs, were not considered significant for the experiments performed thus far. The quantity of oxygen also seems sufficient. Analyses of oxygen utilization and ventilation rates as a function of dissolved oxygen concentration for many aquatic invertebrates suggest that dissolved oxygen levels above 8–9 mg liter^{-1} have minimal effects on increased energy output for oxygen regulation.

Rheophilic species were reared in the plastic troughs with a substrate sequence (bottom to surface) of sand, gravel, pebbles, and rubble. Troughs were seeded with algae from local streams and slopes adjusted to equilibrate surface velocity. Water entered each trough at a rate approaching 0.5 liters s^{-1}. All five sets of troughs were monitored continuously by two multiple pen thermographs.

Slack water species were maintained in plastic trays (45 × 24 × 12 cm). Water flow was regulated by adjustable reservoir spigots to yield a tray turnover time of 2 min. Water exiting a tray passed through screening and into the PVC overflow pipe which emptied into the next pool.

Growth experiments for each insect species were started by placing a known number of newly hatched larvae into test chambers at each thermal regime: the number depended on the species but never exceeded typical saturation densities observed in the field. The initial population biomass per chamber was determined by dry weight of a random subsample of the "seed" individuals. Each chamber was subsampled at regular intervals to provide data on population growth (biomass) and to minimize density effects on the growing organisms. Emerging adults were trapped in mosquito netting above each trough and tray (Fig. 2). Experiments ended after all individuals had metamorphosed into adults.

Embryonic development of aquatic insects has also been studied with this system. Eggs were kept in glass vials partially submerged in the plastic reservoirs at the head of each trough set at densities low enough to minimize oxygen depletion. The 100 ml of filtered (0.45 μm) stream water in each vial stayed within ±0.2°C of incubation temperatures except during rapid temperature changes, when a 5–10-min lag period was observed. Surface diffusion of oxygen into the vials kept levels at saturation for the temperature.

Data for embryonic and larval development of *Sigara alternata* (Say) at five fluctuating temperatures (Table 2, Fig. 4) indicate that average temperature is inadequate to describe thermal conditions in fluctuating environments. The stimulatory effects of brief pulses into higher temperatures are shown by the decrease in embryonic developmental time and the increase in larval growth rate with increased magnitude of the thermal fluctuation. The response of this species to fluctuating temperatures represents a response pattern more general for several aquatic insects (Sweeney unpublished data) and for other insects (Huffaker 1944; Messenger and Flitters 1959). The physiological effect of a given temperature is affected by the length of exposure and the range of temperatures associated with it over a 24-h period. For example, the developmental physiology of a species is different at a constant 15°C from that at a 10–20°C fluctuation about a 15°C temperature. These considerations are critical both to understanding natural temperature relations and to establishing thermal limits for protecting stream communities.

The use of the apparatus is not restricted to the warm months. Short term (6–8 weeks) midwinter studies are possible since the spring water in winter is warm relative to ambient stream water. For these studies the holding pools are shaded with plywood held at a 30-degree angle over the surface. The 12°C water becomes progres-

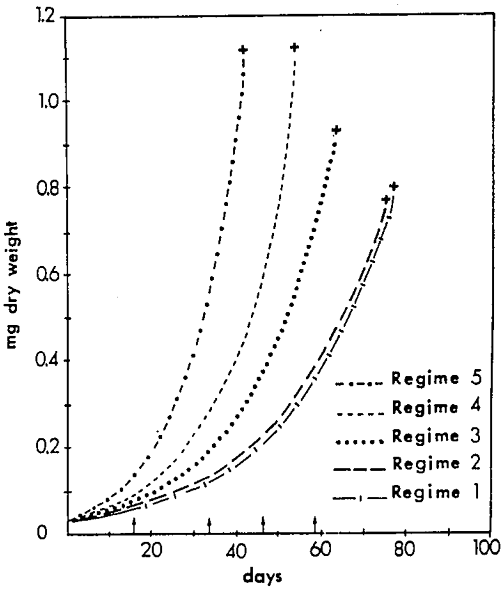

Fig. 4. Larval growth curves for *Sigara alternata* at five fluctuating temperatures. Sampling intervals are represented by arrows; coordinates for mean body size at metamorphosis and developmental days to first adult are depicted by plus signs. The exponential curve ($Y = ae^{bX}$) yielded an $R^2 > 0.90$ for all five data sets.

sively colder as it passes through the pools. Coldest temperatures, at 0600 hours, coincide with minimum air temperatures.

The temperatures reached with this apparatus are determined by natural environmental temperatures. Groundwater temperatures determine the coldest available experimental regime during the summer and the warmest winter temperatures. Since first order streams in a given area will never be any colder (summer) or warmer (winter) than ambient groundwater, this would impose a restriction only for studies at seasonally unnatural temperatures.

The apparatus provides the possibility of studying the significance of diurnal and seasonal temperature fluctuations to such ecologically important processes as egg development, growth, metabolism, emergence, etc. The experimental variable is presented as it is in the field. The apparatus is inexpensive and requires limited maintenance once established. Temperature regulation is automatic; thermographs only require winding and chart changes once a week. The entire system is gravity fed, spring and solar driven: an electrical power source is unnecessary. The flow-through design and diurnally oscillating temperature regimes represent a more natural approach to evaluating the complexity of steam temperature patterns than do closed constant temperature systems.

Table 2. Summary of egg development experiments for *Sigara alternata* at five fluctuating temperatures.

	Thermal regimes				
	1	2	3	4	5
Avg temp (°C)	11.49	13.28	14.13	14.44	14.86
First hatch (d)	25.0	23.0	19.0	16.0	7.0
Last hatch (d)	30.0	26.0	21.0	19.0	21.0
Avg incubation (d)	27.5	23.5	19.8	17.5	13.7
Hatch success (%)	91.1	93.3	87.5	68.3	90.0

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