

LABORATORY STUDIES ON TOLERANCE OF
AQUATIC INSECTS TO HEATED WATERS¹ARDEN R. GAUFIN² AND STEPHEN HERN

University of Montana Biological Station, Big Fork, Montana 54911

ABSTRACT

The mature larvae of fifteen species of aquatic insects (Diptera, Ephemeroptera, Plecoptera, and Trichoptera) and the scud (Amphipoda) were tested to determine their relative sensitivity to heated waters under laboratory conditions. The temperature at which 50% died after 96 hours ($TL_{m,96}$) was recorded as the lethal temperature. This ranged from 11.7 C for the torrential stream mayfly, *Cinygmula par Eaton*, to 32.6 C for the snipefly, *Atherix variegata* Walker.

By 1980, it is estimated that around 200 billion gallons of cooling water will be needed daily, about one-sixth of the nationwide annual runoff, to meet projected steam electric power station needs based on once-through cooling (Pitcon, 1960). Water used for cooling purposes in industrial processes may be so hot and in such quantity that it may substantially raise the temperature of a receiving stream. Limited quantities of warm water, however, may produce desirable changes in selected localized situations. The requirements of the organisms in a stream must be known before realistic water quality standards can finally be adopted for their protection.

Literature concerning the effects of heated waters on aquatic insects is limited in extent and comparability. The effects of heated effluents on aquatic life have been reviewed in two recent comprehensive bibliographies, Kennedy and Mihursky (1967) and Raney and Menzel (1967). The effects of heated discharges on water quality and assimilation, aquatic organisms, and water uses have been thoroughly reviewed by Parker and Krenkel (1969). The temperature requirements of fish and other aquatic life were reviewed by Tarzwell (1968). Nebeker and Lemke (1968) tested the relative sensitivity of twelve species of aquatic insects to heated water in the laboratory. The lethal temperature at which 50% of the test specimens died after 96 hours exposure ($TL_{m,96}$) ranged from 21 C for winter stoneflies to 33 C for dragonflies. An excellent review of temperature effects on aquatic insects was presented by Trembley (1965). Studies conducted by the Philadelphia Academy of Science (Patrick, 1968) on the effects of heated water on the insect fauna of the Potomac River have shown significant reductions in the diversity and numbers of organisms below a steam electric power plant. Coutant (1962) found substantial reductions in the volume and num-

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²Professor of Zoology, University of Utah.

bers of macroinvertebrates in the Delaware River in sections receiving heated water.

This report summarizes the results of acute, short-term 96-hour tests (TL_{96}) used in screening 15 species of aquatic insects to determine their relative sensitivity to heated water. The 96-hour TL_{96} (Standard Methods, 1960) was used as a measure of effect in these tests. Further long-term studies are contemplated dealing with the effects of temperature on the reproduction, molting, emergence patterns, feeding rates, and long-term survival of aquatic insects.

MATERIALS AND METHODS

Test chambers consisted of oblong stainless steel tanks 90 cm long, 18 cm wide, and 17.5 cm deep. Similar tanks were utilized by Nebeker and Lemke (1968) in their studies on the tolerance of aquatic insects to heated waters at the National Water Quality Laboratory at Duluth, Minnesota. Fiberglass screening was employed to subdivide the tanks into three test cages 15 cm long, 17.5 cm wide, and 11 cm deep. Rocks were placed at the bottom of each cage to form a natural substrate for the aquatic organisms. The fresh water source was introduced at the forward end of the tank, which gradually slopes 7.5 cm to the overflow drain.

Five chambers were employed for temperature testing and one for a control, with the control maintained at the initial acclimation temperature. The oblong tanks were used as artificial streams where various water flows could be maintained with a stream of water and with paddle wheels.

The water used for all testing and for the holding tanks was obtained from the University of Montana Biological Station water system. This water originates in a spring, is chlorine-free, and has a constant temperature of $6.4\text{ C} \pm 0.1\text{ C}$. The pH is 7.8 ± 0.1 . Total hardness is near 135 ppm and the CO_2 varies from 1 to 2 ppm (CO_2 and total hardness expressed as ppm of CaCO_3). The dissolved oxygen level is consistently 100% of saturation or higher.

The test organisms, except for species of *Simulium*, *Hexagenia*, *Atherix*, and *Gammarus*, were collected from Rock Creek, a trout stream located southeast of Missoula, Montana. *Simulium* and *Hexagenia* were collected from Mud Creek, a slow-flowing meadow creek, *Atherix* from the Clark's Fork of the Columbia River, and *Gammarus* from a spring-fed pond near Big Fork, Montana. All test organisms were mature larvae. The test organisms were placed in large, vigorously aerated, fiberglass holding tanks for a minimum of three days prior to testing. Fresh water was added at a rate of 3 to 5 liters per minute to insure a constant temperature and a fresh water supply.

Desired temperatures in the test chambers were obtained by manual regulation of mixing faucets. Temperatures were allowed to stabilize

TABLE 1. Temperatures (°C) at which 50% of the test species died after 96 hours exposure (TL_m⁹⁶), Big Fork, Montana, 1968-69.

Species tested	Test 1	Test 2	Test 3	Mean TL ₃	Average group TL _m
Diptera					
<i>Atherix variegata</i> Walker	32.6	32.2	—	32.4	28.7
<i>Simulium</i> sp.	25.0	25.2	—	25.1	
Trichoptera					
<i>Parapsyche elsis</i> Milne	21.8	21.6	—	21.7	26.5
<i>Limnephilus ornatus</i> Banks	24.5	25.0	—	24.75	
<i>Neothrema alicia</i> Banks	25.8	26.0	—	25.9	
<i>Drusus</i> sp.	27.2	27.4	—	27.3	
<i>Brachycentrus occidentalis</i> Banks	29.7	—	—	29.7	
<i>Hydropsyche</i> sp.	30.0	30.1	—	30.05	
Plecoptera					
<i>Isogenus aestivalis</i> (Needham and Claassen)	16.0	16.3	—	16.15	22.55
<i>Pteronarcella badia</i> (Hagen)	24.4	24.6	24.2	24.4	
<i>Pteronarcys californica</i> Newport	28.0	26.4	26.2	27.0	
Ephemeroptera					
<i>Cinygmula par</i> Eaton	11.7	—	—	11.7	18.82
<i>Ephemerella doddsi</i> Needham	15.4	15.5	—	15.45	
<i>Ephemerella grandis</i> Eaton	21.5	—	—	21.5	
<i>Hexagenia limbata</i> Guerin	26.1	27.1	—	26.6	
Amphipoda					
<i>Gammarus limnaeus</i> Smith	14.5	14.6	—	14.55	14.55

over a period of 24 hours to insure uniformity. If the system remained stable during this 24-hour period, the test was initiated.

Experimentation began with an initial series of temperatures usually ranging from 10 to 25 C. The specimens were placed in an aerated water bath, and the temperature gradually raised (2 to 4 C per hour) to the appropriate test temperature before they were transferred to the test chambers. This procedure was followed to insure against nebulous results induced either by thermal "shock" from immediate transfer from one temperature to another or by the complete acclimation that can accompany a very gradual increase in temperature.

In the test chambers the paddle wheels created a turbulence and helped maintain a dissolved oxygen level of 100% saturation or higher. A liberal fresh water supply was provided (at least 2 liters per minute) for the removal of toxic waste. Temperature values were taken at least four times daily and if any value varied by more than 0.5 C, the test was discarded. If any of the control organisms died, the test was terminated.

The temperature at which 50% of the organisms died was obtained

by a modification of the straight line graph interpolation method as outlined in Standard Methods (1960).

RESULTS

Late instar larvae of 15 species of aquatic insects and one species of amphipod were tested to determine their tolerance of high water temperatures. A marked difference in sensitivity was apparent (Table 1) in the different species. A mayfly, *Cinygmula par* Eaton, died at 11.7 C and was the most sensitive of all the species tested. This species is found in very cold clear mountain streams in Montana. The fresh water shrimp, *Gammarus limnaeus* Smith, proved to be surprisingly sensitive to temperature increases, exhibiting a 96-hour TL_m of only 14.5 C. *Ephemerebella doddsi* Needham, a small, widely distributed mayfly characteristic of cold turbulent streams in the Intermountain Region, was also very sensitive, with a TL_m value of 15.4 C. A lotic species of mayfly, *Hexagenia limbata* Guerin, was much more tolerant than other mayflies tested with a TL_m of 26.6 C.

Considerable difference in susceptibility to temperature increases existed between the three species of stoneflies tested. *Isogenus aestivalis* (Needham and Claassen) was quite sensitive, 50% dying at 16 C, while *Pteronarcella badia* (Hagen) and *Pteronarcys californica* Newport, two closely related species, survived increases to 24.6 and 26.6 C respectively. Six species of caddis flies were tested and clearly reflected thermal differences in their habitat requirements. *Parapsyche elsis* Milne, which is largely restricted to cold, fast-flowing mountain streams, had a TL_m of 21.8 C while *Hydropsyche* sp. taken from a slow-flowing stream draining a marshy lake was very tolerant with a TL_m of 30.1 C. *Atherix variegata* Walker, the snipe fly, was the most tolerant of all species tested with a TL_m of 32.6 C. No dragonfly or damselfly nymphs were tested because a thick ice and snow cover coating their habitats early in the winter prevented collecting large enough numbers for testing purposes.

DISCUSSION

The rate of development and the time of emergence of aquatic insects is directly influenced by the temperature. An increase in water temperatures in the winter above 5 C might completely eliminate winter stoneflies belonging to the family Capniidae.

Many species of stoneflies, mayflies, and caddis flies emerge in late spring before stream temperatures reach high summer levels. An artificial increase in stream temperatures during the winter would very likely cause these species to develop more rapidly, emerge earlier, and be killed by cold air temperatures, and may substantially reduce the population or eliminate the species.

The stonefly *Isogenus aestivalis* and mayfly *Cinygmula par* are largely restricted to clear, cold water streams in the Intermountain

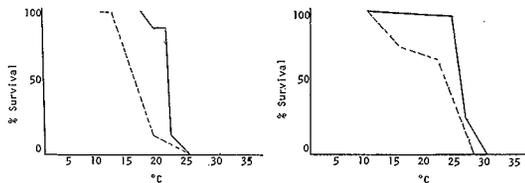


FIG. 1. Straight-line interpolation graphs of representative $TL_{m^{90}}$ s. Left: *Ephemera doddsi*, dashed line, $TL_{m^{90}} = 15.5$ C; *E. grandis*, solid line, $TL_{m^{90}} = 21.5$ C. Right: *Pteronarcys badia*, dashed line, $TL_{m^{90}} = 24.6$ C; *P. californica*, solid line, $TL_{m^{90}} = 26.6$ C.

Region and even a slight increase in water temperature may have an adverse effect on their survival. By comparison the snipe fly, *Atherix variegata*, is often found in open sections of streams which warm up during the summer months and this species is decidedly temperature tolerant.

Two of the species of stoneflies tested, *Pteronarcella badia* and *Pteronarcys californica*, are common in medium to large streams in the western United States and are comparatively temperature tolerant. These species require two and three years respectively to complete their life cycle and have become adapted to the warmer waters of late summer, which many aquatic insects avoid by emerging in the spring.

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A SCANNING ELECTRON MICROSCOPY STUDY OF STRIDULATING ORGANS IN TWO HEMIPTERA¹

FRED A. LAWSON AND JEAN CHU²

ABSTRACT

Scanning electron micrographs of the stridulatory file of two Hemiptera, *Galgupha ovalis* (Corimaelenidae) and *Microporus obliquus* (Cydnidae), are given. The scraper and adjacent surface of *G. ovalis* are also pictured.

Structures which are obviously stridulating mechanisms were discovered in *Galgupha ovalis* (Hussey) (Corimaelenidae) and in *Microporus obliquus* Uhler (Cydnidae) during investigation of other features of the wings of Hemiptera-Homoptera. These structures were discussed briefly at the subfamily level by McAtee and Malloch (1933) but their function was not recognized. Numerous species, representing genera from many parts of the world, were examined by Leston (1954, 1957) and Leston and Pringle (1963); these descriptions and summaries are parts of a more generalized coverage and were not intended to give the details presented here. The terminology used in the present paper is that of Comstock (1936). The *file* is the strigil and the *scraper* is the lima or the plectrum referred to by Leston (1954, 1957). The file is a ventral row of teeth on an enlarged basal portion of the first anal vein of the metawing in each species; the scraper is a sharp, upturned dorsal ridge on the anterior margin of the first abdominal tergum.

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²University of Wyoming, Laramie 82070.