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Effects of Acid Precipitation on *Daphnia magna*

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Pollutants derived from fossil fuel combustion and precipitated from the atmosphere have substantially increased in the past decades (DOVLAND et al. 1976, LIKENS 1976, LIKENS & BORMANN 1974). These materials, precipitated in such industrialized areas as southeastern Canada, have caused considerable alterations in aquatic ecosystems (GORHAM 1961). Precipitation over most of the eastern United States is presently 10 to 500 times more acidic than is natural (LIKENS 1976, LIKENS et al. 1977). Most affected aquatic ecosystems contain oligotrophic waters in regions of thin poorly buffered soils (N.A.S. 1978, JEFFERIES et al. 1979). One example of the effect of acid precipitation is seen at Woods Lake in the Adirondack Mountain Region of New York, which has been found to be extremely acidic with measurements occasionally falling below pH 4.0 (PUGLIESE 1978). At one time the lake was naturally well stocked with fish, but their population has declined sharply. Attempts to restock the lake have been unsuccessful and are thought to be related to the low pH values.

Zooplankton are an important link in food chains of aquatic ecosystems and their disappearance or decline could drastically affect trophic relationships. Declines in zooplankton density in response to acid precipitation have been reported (ALMER et al. 1974, HENDREY & WRIGHT 1976, HENDREY et al. 1976, LIEVESTAD et al. 1976, SPRULES 1975). DAVIS & OZBURN (1969) reported short term survival of *Daphnia pulex* between pH 4.3 and 10.4; however, its potential for reproduction was limited to a fairly narrow range. ANDERSON (1944) noted the advantages of using daphnia as test organisms, and ANDERSON et al. (1948) concluded that *Daphnia magna* was representative of other abundant zooplankton in sensitivity to toxic substances.

MATERIALS AND METHODS

D. magna (obtained from Connecticut Valley Biological Supply Co., Southampton, MA) was grown in river water (Quinapoxet River, Holden, MA) in 39-L plexiglass aquaria. The temperature was maintained at 18-20° C and the tanks were illuminated by room light with a daily photoperiod of 16h light: 8h dark. Cultures were fed one tablespoon of Fleischmann's yeast (BOND 1934), suspended in 100 mL of river water, per aquarium. Feeding was repeated when cultures, slightly milky after feeding, had become clear. Mild bubbling aeration was provided at all times and dissolved oxygen remained at or above 80% saturation.

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For acute tests, ten D. magna, 14 ± 1 d old were placed in 200 mL of river water in 250-mL beakers. The pH of each sample was adjusted with 0.01N sulfuric acid or 0.01M NaOH to 3.5, 3.8, 4.0, 4.2, 4.5, 4.7, 4.9, 5.6 and 5.9 (with the aid of Corning Model 610A portable pH meter and #4332-8 Corning combination pH electrode). Test chambers were covered with a pane of glass to minimize evaportaion and were placed in a constant temperature room (20 ± 1° C). The number of D. magna surviving after 15 and 30 min, and 1, 2, 4, 8 and 24 h were determined by the procedure of ANDERSON (1944). Complete immobilization or death was the endpoint. The pH at 24 h was determined and the average pH for each sample was calculated. The initial and final pH values never differed by more than 0.22 units.

For chronic tests, ten D. magna were placed in 200 mL of river water in a 250-mL beaker. The pH of seven samples was adjusted to 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, and 7.5 with 0.01N sulfuric acid or 0.01M NaOH. Test chambers were covered with a pane of glass to minimize evaporation and placed in a constant temperature room (20 ± 1° C) for the three week test period. The pH of each sample was read and recorded. If the pH varied more than 0.05 units, a daily readjustment was made. Survivors were counted each day. Complete immobilization or death was the endpoint. Any young produced were counted daily and discarded. The D. magna in each test chamber were fed 1 mL of the yeast suspension on the first day and every nine days thereafter. Chronic studies were performed with daphnia of two age groups: 4 ± 1 d and 7 ± 1 d old to ascertain the effect of age on pH tolerance.

Dissolved oxygen content of samples in acute tests after 24 h and samples in chronic tests after 1, 2, and 3 weeks was determined with the use of a YSI D.O. probe (Yellow Springs Instrument Co., Yellow Springs, Ohio). Total alkalinity and hardness of samples in acute tests after 24 h and samples in chronic tests after 1, 2, and 3 weeks were determined by direct titration (Hach Chemical Co., Loveland, CO).

RESULTS

The effect of pH on the survival of D. magna (14 d) is shown in Figure 1. In this acute test (24 h) over 85% of the organisms survived if the pH was 4.5 or higher. At pH values of 4.2 or lower survival was always less than 50%.

Figure 2 illustrates chronic survival in relation to time at different pH values for two different age groups (4 and 7 d) of D. magna. The mean pH for each sample can be found in Table 1. While survival at pH 4.5 and 5.0 was greater than 85% during the acute tests, survival at these pH values during chronic studies fell to zero after three week exposures. At pH 5.0 there was no survival of 4 d D. magna at the end of one week exposure, whereas at the same pH, 7 d D. magna showed 35% survival. During the chronic studies reproduction was observed with 4 d D. magna at pH values of 5.5 and greater. In the older age group (7 d) reproduction was observed at pH values of 5.0 and greater.

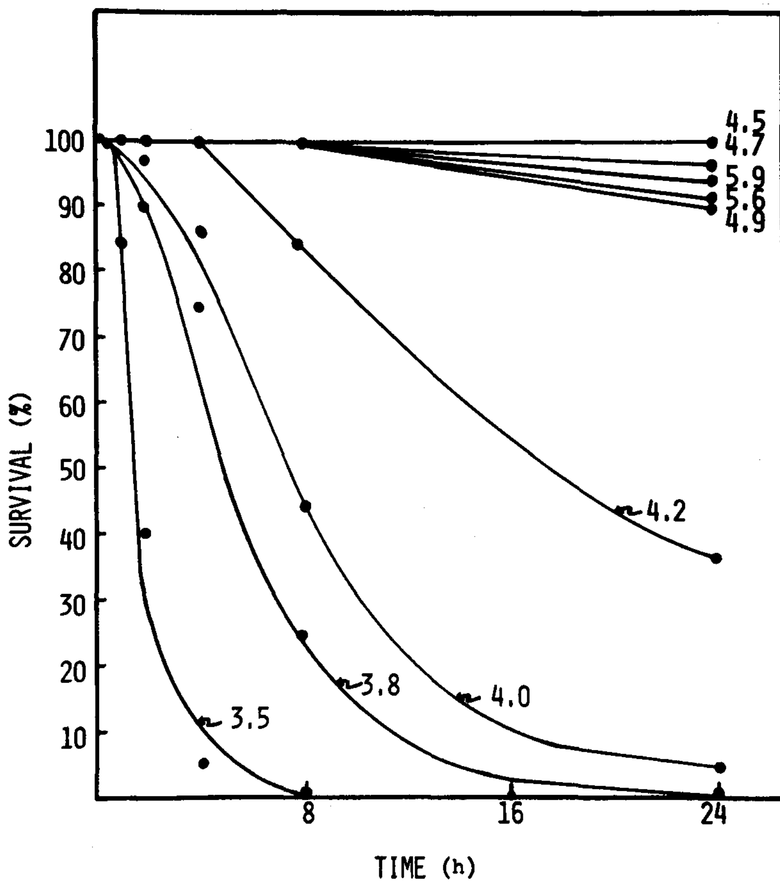

Figure 1. The effect of pH on acute survival of D. magna. pH values are listed with each response curve.

Figure 3 illustrates survival versus pH for the acute test after 24 h and for the chronic tests after three weeks. Linear regression analysis ($p \geq 0.05$) performed on data obtained from three experimental runs of the acute test established that a linear relationship exists between pH 3.8 and 4.5. From this straight line the estimated 24 h TL_m was determined to be pH 4.2. Figure 3 also demonstrates that chronic tests performed on 4 and 7 d D. magna reveal that their three week TL_m's occur at higher pHs. The 7 d organisms are more tolerant to low pH values than the 4 d organisms.

The dissolved oxygen levels of the samples remained at or above 5.4 mg/L during these tests.

Total alkalinity, expressed as mg/L $CaCO_3$, ranged from 1.6 at pH 4.5 to 23.1 at pH 7.5 for the chronic studies. Total alkalinity at pH 3.5 was zero. Hardness expressed as mg/L $CaCO_3$, averaged

TABLE

INITIAL pH	MEAN pH 4 d	MEAN pH 7 d
4.5	4.70	4.78
5.0	5.10	5.29
5.5	5.69	5.64
6.0	6.06	6.09
6.5	6.30	6.51
7.0	6.75	6.80
7.5	6.98	7.13

TABLE 1. Mean pH values for chronic test samples employing 4 d and 7 d D. magna.

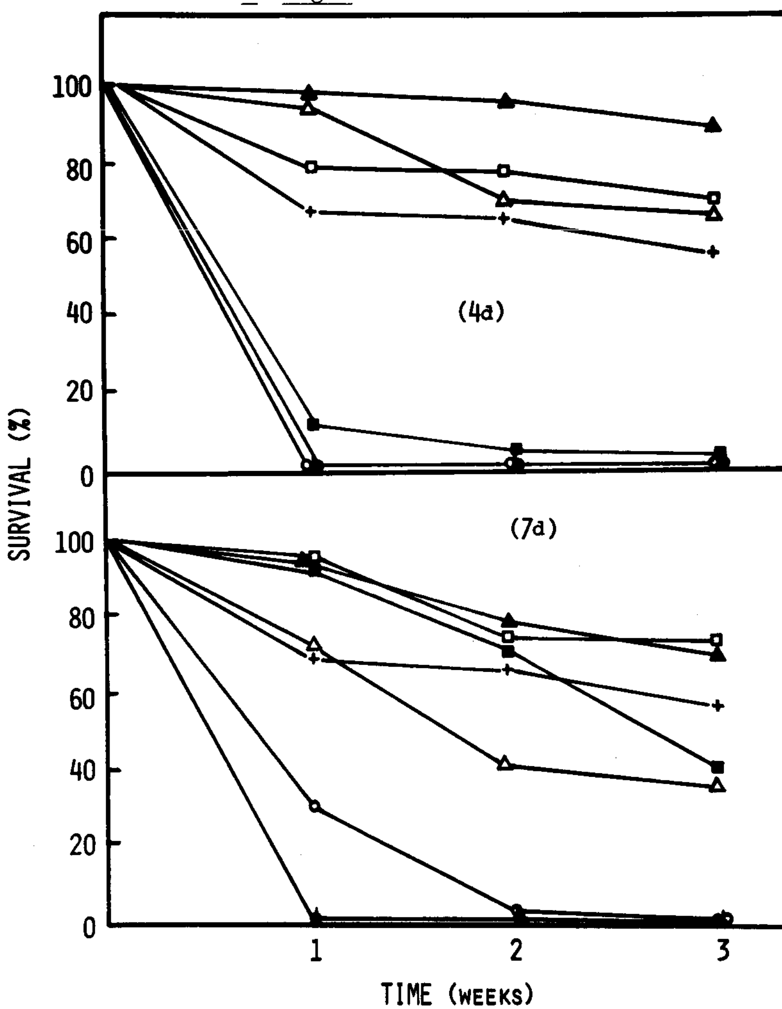

Figure 2. Chronic effects of pH on D. magna. pH 4.5 (●), pH 5.0 (O), pH 5.5 (■), pH 6.0 (□), pH 6.5 (+), pH 7.0 (▲), and pH 7.5 (Δ).

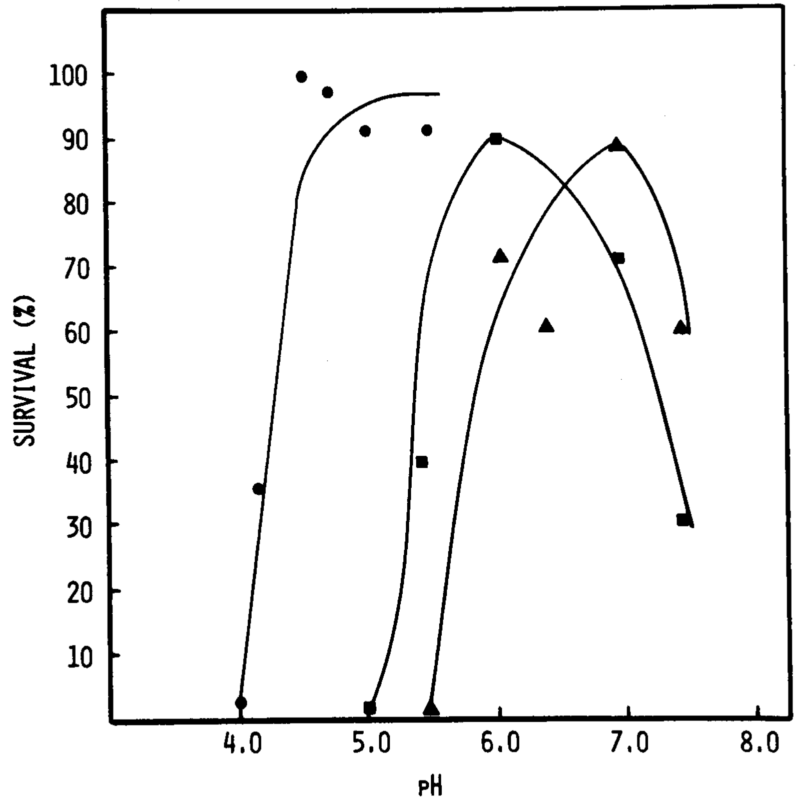

Figure 3. Acute (●, 24 h) and chronic (3 w) survival of D. magna exposed to acidic water. Chronic tests contain 4 d (▲) and 7 d (■) organisms.

22.3 mg/L. The total alkalinity and hardness of unadjusted river water averaged 11.0 mg/L and 22.0 mg/L respectively.

DISCUSSION

Acid precipitation, which has contributed to the decrease in the pH of some aquatic ecosystems to values as low as 4.0, has led to declines in zooplankton and fish populations (ALMER et al. 1974, HENDREY & WRIGHT 1976, HENDREY et al. 1976, LIEVESTAD et al. 1976, SPRULES 1975, NEVILLE 1979). Daphnia spp. are an important fish food and D. magna has been reported to be a good test organism for zooplankton in general (MAKI 1979, ANDERSON 1944). MAKI (1979) reports that a high correlation exists between the chronic response of D. magna and fish to toxic substances. DAVIS & OZBURN (1969) studied the effect of pH on D. pulex survival and found acute survival to be likely between pH 4.3 and 10.4, however, pH varied too much to perform detailed analysis of the data. Chronic studies monitored continuously were not performed. Reproduction was observed between pH 7.0 and 8.7 in samples that were observed beyond their 32 h acute test.

Our study shows that adult D. magna (14 d) will tolerate low pH for short periods of time (i.e., 24 h TL_m = 4.2). However, chronic studies indicate that continued survival is unlikely at or below pH 5.0. Younger organisms (4 d) were unable to survive at pH 5.5 as compared to 7 d organisms which showed 40% survival after three weeks. Reproduction was not observed below pH 5.5 with 4 d D. magna and below 5.0 with 7 d D. magna. The effects of pH on reproduction require further study, but our study shows that normal reproduction rates are unlikely at pH 5.0 or less.

The effects of pH on the physical and chemical parameters of the aquatic ecosystem as well as on biochemical parameters of numerous aquatic species must be further investigated. The effects of such parameters on zooplankton survival in the wild has not been established.

Decreasing pH due to acid precipitation may be critical to the survival of zooplankton such as D. magna. Disrupting an important link in an aquatic food web may lead to deleterious effects for the entire aquatic ecosystem.

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