

The effects of acid precipitation on amphibians

BENJAMIN A. PIERCE

Department of Biology, Baylor University, Waco, TX 76798-7388, USA

Received 4 August 1992; accepted 11 September 1992

A large number of studies conclusively demonstrate that low pH conditions have important ecological effects on amphibian communities. These effects vary developmentally and depend on complex interactions with other physical, chemical, and biological parameters. Acid sensitivity varies widely within and among species of amphibians. In spite of the importance of acidity to their reproductive success and the presence of many acidic amphibian habitats, there is, at present, little direct evidence that acid precipitation has produced widespread declines in amphibian populations. However, the potential role of sublethal effects of acidity on amphibian declines has not been studied thoroughly.

Keywords: acid precipitation; acid tolerance; amphibians; embryos; frogs; salamanders; tadpoles

Introduction

Although the smallest class of vertebrates in number of species, amphibians are critical components of many natural ecosystems, a fact that is frequently unappreciated because of their secretive habits. For example, tadpoles regulate the algal composition of ponds (Dickman, 1968; Seale, 1980), and terrestrial salamanders may comprise a major portion of animal biomass in some forests (Burton and Likens, 1975). Amphibians are also a major source of nutrient flow from aquatic to terrestrial habitats.

In recent years, there has been growing interest in the ecology of amphibians in low pH environments. This research has been stimulated by concerns about the deleterious effects of acid precipitation on amphibian communities (Pough, 1976) and by recent reports of worldwide declines in amphibian populations (Barinaga, 1990; Wake, 1991). Although a link between widespread declines and acid precipitation has yet to be established, there are important reasons to believe that amphibians may be particularly vulnerable to acid rain. First, many amphibians utilize ephemeral aquatic habitats, which periodically dry and refill with fresh precipitation. These ponds typically occupy small watersheds, and the water entering them has limited contact with the soil and other buffering systems. Furthermore, the ponds themselves are frequently low in calcium and alkalinity (Freda *et al.*, 1991). For these reasons, amphibian habitats may be influenced strongly by precipitation chemistry, and may be susceptible to acidification from acid deposition.

Amphibians are also susceptible to acid precipitation because of the timing of their reproduction. Early embryological stages are most sensitive to acidity (Pierce *et al.*, 1984; Freda, 1986), and many species breed in early spring, when aquatic habitats are lowest in pH and alkalinity (Pierce *et al.*, 1984; Freda and Dunson, 1985a; Freda *et al.*,

1991). Furthermore, amphibians are particularly sensitive to many types of environmental degradation, because adults are high in the food chain, amphibian skin is highly permeable to toxic substances, and metamorphosis between the aquatic larval stage and the terrestrial adult stage is physiologically stressful (Vitt *et al.*, 1990).

In this review, I will summarize our current understanding of the ecological effects of acidity on amphibians. Only a few studies have examined the direct effects of acid precipitation on amphibians, but much research now provides a comprehensive view of the ecotoxicology of acidity for amphibians. Field studies have measured acidity of natural habitats, while laboratory and field observations provide information on the sensitivity of a number of species to low pH conditions. The interaction of acid toxicity with other environmental variables such as cations, metals, organic compounds, and temperature have been examined. Physiological mechanisms of acid toxicity have been studied at several developmental stages, and intraspecific variation, including genetic variation, in acid tolerance has been investigated. Sublethal effects of acidity, such as inhibition of growth and delay of hatching, have also been studied. Also, recent studies have examined survival and competition of amphibians in artificial ponds where pH has been experimentally manipulated. This work gives much insight into the potential ecological effects of acid precipitation on these animals.

Variation in acidity of amphibian habitats

An important, though frequently overlooked, fact is that many aquatic habitats are naturally acidic. For example, sphagnum moss and organic compounds produce acid bogs (Gorman *et al.*, 1985), and iron pyrites in the soil may oxidize to form strong mineral acids (Huckabee *et al.*, 1975). In response to this natural variation in environmental acidity, amphibians have evolved differing sensitivities to low pH conditions. While some species are relatively sensitive to acidity, others exhibit considerable tolerance. Indeed, several species, such as *Rana virgatipes* and *Hyla andersoni*, appear to be ecologically specialized for naturally acidic habitats (Gosner and Black, 1957; Freda and Dunson, 1986).

Although some aquatic habitats may be naturally acidic, combustion of fossil fuels in the past century has led to widespread deposition of acidity and concomitant acidification of previously nonacidic habitats. Thus, many species of amphibians may have been exposed to increased levels of acidity in recent times. Surveys of potential amphibian breeding habitats in North America (Dale *et al.*, 1985a; Clark, 1986a; Freda and Dunson, 1986), Britain (Aston *et al.*, 1987; Beattie *et al.*, 1991), and Europe (Leuven *et al.*, 1986) found significant numbers of sites with relatively low pH (<5). Although determining the sources of acidity in many of these cases is difficult, Pough (1976) and Cook (1983) observed drops in pH of amphibian habitats following precipitation, and Harte and Hoffman (1989) reported that pH declined in salamander-inhabited ponds following snowmelt; these observations suggest that acid precipitation does acidify amphibian breeding habitats. On the other hand, Pierce *et al.* (1984) and Freda and Dunson (1985a) found no relationship between the amount of acidity in precipitation and weekly changes in pond pH. Thus, the extent of acidification of amphibian habitats from acid deposition is difficult to ascertain. It is clear, however, that where acidic habitats occur, amphibian distribution and abundance are frequently correlated with pond pH (Gosner and Black 1957; Cooke and Frazer, 1976; Strijbosch, 1979; Clark, 1986a;

Gascon and Planas, 1986; Leuven *et al.*, 1986; Karns, 1992), suggesting that water pH plays an important role in shaping amphibian communities.

Effects of acidity on embryos

Field and laboratory studies have demonstrated that acidity has direct toxic effects on amphibian embryos and larvae, but the sensitivity and effects vary developmentally. Early developmental stages of amphibians appear to be most sensitive (Pierce *et al.*, 1984). Because fertilization occurs externally in most frogs, the fertilization process may be particularly vulnerable to low pH. Even in salamanders, where internal fertilization is the general rule, sperm are deposited externally as spermatophores, which may be exposed to acidity in the environment. Schlichter (1981) examined the effects of low pH on fertilization of *Rana pipiens* eggs and concluded that fertilization was reduced below pH 6.3. These findings have been questioned (Dale *et al.*, 1985b; Freda, 1986) because Schlichter used buffers that were later shown to be toxic. Thus, the extent to which reduced sperm motility at low pH might affect fertilization is unclear. Beattie *et al.* (1992) found that the rate of fertilization of Common Frog eggs increased significantly after lime was added to an acidic pond, but the overall rate of fertilization was high even in acidic ponds. Also, Karns (1983) reported high rates of fertilization of eggs of several frogs in bog water with pH as low as 4.2.

Several laboratory studies have demonstrated that acidity is toxic to amphibian embryos (Gosner and Black, 1957; Pough and Wilson, 1977; Tome and Pough, 1982; Freda and Dunson, 1985c). At very low pH, embryological development is disrupted and usually ceases within hours (Freda and Dunson, 1985c). At more moderate pHs, development continues but the fluid-filled space surrounding the embryo (the perivitelline space) shrinks, and the embryo becomes constricted within the egg capsule (Gosner and Black, 1957; Tome and Pough, 1982; Dunson and Connell, 1982; Freda and Dunson, 1985c); this phenomenon has been termed the 'curling defect.' At moderate pHs, embryos frequently complete development but are unable to hatch successfully from the egg capsules. The cause of this failure to hatch is unknown, but it may be due to physical changes in the egg capsule, failure of the perivitelline space to expand, or inhibition of the hatching enzyme (Urch and Hedrick, 1981; Dunson and Connell, 1982). Those embryos that do hatch successfully at low pH often exhibit developmental abnormalities as a result of developing in the constricted perivitelline space (Gosner and Black, 1957; Pierce *et al.*, 1984; Freda and Dunson, 1985c). Another sublethal effect of acidity on embryonic stages is delay in hatching time (Pierce and Sikand, 1985). Predation on amphibian eggs is also affected by low pH conditions; Henrikson (1990) concluded that predation on the eggs of some species may increase in acidified lakes because fish populations decline, allowing increases in invertebrates and salamanders that prey on amphibian eggs.

Similar to observations made in the laboratory, low hatching success and characteristic developmental abnormalities associated with low pH have been observed in acidic ponds (Pough, 1976; Gascon and Planas, 1986; Portnoy, 1990). Absence of particular species, population size, number of egg masses, and hatching success in the field have been associated with pond acidity (Hagström, 1981; Karns, 1983; Clark, 1986a,b; Freda and Dunson, 1986; Gascon and Planas, 1986). Transplantation studies, in which embryos were moved from neutral to acidic ponds, also demonstrated the toxic effects of acidity

under natural conditions (Freda and Dunson, 1986; Albers and Prouty, 1987; Portnoy, 1990). Experimental increases in pH (produced by liming) of two acidic ponds in northern England raised hatching success of *Rana temporaria* embryos from 0% to 69% and from 22% to 93% (Beattie *et al.*, 1992). In these field studies, it is important to keep in mind that pH is intercorrelated with other chemical and biological parameters, and thus the direct causes of mortality and population abundance are difficult to determine. Nevertheless, the results of field studies are entirely consistent with laboratory studies that demonstrate direct toxicity of acidity to amphibian embryos.

The toxic effects of acidity on amphibian embryos are complex and are influenced by other environmental factors that vary in time and space. These other factors include temperature (Pough and Wilson, 1977; Punzo, 1983), calcium concentration (Freda and Dunson, 1985c; Dale *et al.*, 1985a; Gascon *et al.*, 1987; Cummins, 1988), sodium concentration (Freda and Dunson, 1985c), concentration of other ions in the water (Freda and Dunson, 1985c), concentration of dissolved organic compounds in the water (Freda *et al.*, 1989), and metals (reviewed by Freda, 1991). Many of these factors covary with pH, and their interaction with acidity can be complex. For example, aluminum solubility increases as pH drops; thus, low pH waters frequently contain elevated levels of aluminum. However, aluminum speciation also changes with pH, and aluminum toxicity depends upon the form of aluminum present in the water. The interaction of acid toxicity and aluminum toxicity is also species dependent. With some species under some conditions, aluminum increases sensitivity of amphibian embryos to acidity (Clark and Hall, 1985; Freda and McDonald, 1989; Freda *et al.*, 1989; Freda, 1991), whereas with other species or different conditions, aluminum can actually reduce the toxic effects of acidity (Clark and LaZerte, 1987; Freda and McDonald, 1989; Freda *et al.*, 1989).

Embryos from different species of amphibians display considerable variation in their tolerance to acidity (Table 1). While some species, such as *Ambystoma* salamanders, appear to be relatively sensitive to low pH conditions, other species such as *Rana virgatipes*, *Rana sylvatica*, and *Hyla femoralis* are relatively tolerant. Species that occur in naturally acidic habitats generally have greater tolerance than species that occur in nonacidic habitats. For example, the Carpenter Frog, *Rana virgatipes*, is associated with acidic sphagnum bogs on the coastal plain of North America (Conant, 1975), and embryos of this species display high tolerance to low pH (Gosner and Black, 1957). There are no obvious phylogenetic patterns in embryological tolerance, with the exception that *Ambystoma* salamanders appear to be more sensitive than frogs. However, comparison of acid tolerance among species is complicated by considerable within-species variation in acid sensitivity and by different methodologies used to assess tolerance in different species. Thus, caution should be exercised in drawing conclusions about the nature of species differences in acid tolerance.

In addition to among-species differences in acid tolerance, some studies have reported within-species variation in embryo tolerance. For example, reports of acid tolerance for spotted salamanders vary widely from population to population (Pough, 1976; Cook, 1983; Freda and Dunson, 1985c; Clark 1986b; Albers and Prouty, 1987; Clark and LaZerte, 1987; Blem and Blem, 1989; Portnoy, 1990). However, in many cases, these differences were observed by different investigators using different methods. Because differences in methodology can influence the effects of acidity, these population differences are difficult to interpret. Nevertheless, studies of several species have examined population differences using uniform methods and have detected geographic variation in

Table 1. Acid tolerance of amphibian embryos

Species	pH at which approximately 50% mortality occurs	References
FROGS		
<i>Acris gryllus</i>	4.6	Gosner and Black (1957)
<i>Bufo americanus</i>	4.0	Freda and Dunson (1985c)
<i>Bufo boreas</i>	4.4–4.5	Corn <i>et al.</i> (1989)
<i>Bufo bufo</i>	4.5	Leuven <i>et al.</i> (1986)
<i>Bufo calamita</i>	4.0–4.7	Beebee and Griffin (1977) Beebee (1986)
<i>Bufo canorus</i>	4.6	Bradford <i>et al.</i> (1992)
<i>Bufo punctatus</i>	4.0	Pierce (1991)
<i>Bufo woodhousei</i>	4.0–4.2	Karns (1983) Freda and Dunson (1985)
<i>Hyla andersoni</i>	3.8	Gosner and Black (1957)
<i>Hyla crucifer</i>	4.2	Gosner and Black (1957)
<i>Hyla femoralis</i>	3.4	Warner <i>et al.</i> (1991)
<i>Hyla gratiosa</i>	4.2	Warner <i>et al.</i> (1991)
<i>Hyla versicolor</i>	4.3	Gosner and Black (1957)
<i>Microhyla ornata</i>	3.7	Padhye and Ghate (1988)
<i>Pseudacris nigrata</i>	4.1	Gosner and Black (1957)
<i>Pseudacris triseriata</i>	4.8	Corn <i>et al.</i> (1989)
<i>Rana arvalis</i>	3.8–4.0	Leuven <i>et al.</i> (1986) Andren <i>et al.</i> (1988) Andren <i>et al.</i> (1989)
<i>Rana catesbeiana</i>	4.3	Gosner and Black (1957)
<i>Rana clamitans</i>	4.1	Gosner and Black (1957)
<i>Rana dalmatina</i>	4.5	Andren <i>et al.</i> (1988)
<i>Rana esculenta</i>	4.5	Leuven <i>et al.</i> (1986)
<i>Rana muscosa</i>	4.4	Bradford <i>et al.</i> (1992)
<i>Rana palustris</i>	4.4	Gosner and Black (1957)
<i>Rana pipiens</i>	4.0–4.5	Freda and Dunson (1985c) Corn <i>et al.</i> (1989)
<i>Rana sylvatica</i>	3.75–4.3	Pierce and Harvey (1987) Corn <i>et al.</i> (1989)
<i>Rana sphenoccephala</i>	4.1	Gosner and Black (1957)
<i>Rana temporaria</i>	4.3	Leuven <i>et al.</i> (1986)
<i>Rana virgatipes</i>	3.8	Gosner and Black (1957)
<i>Scaphiopus intermontanus</i>	4.3	Pierce (1991)
<i>Xenopus laevis</i>	3.5–4.0	Tome and Pough (1982) Dunson and Connell (1982)
SALAMANDERS		
<i>Ambystoma maculatum</i>	4.5–5.0	Freda and Dunson (1985c) Blem and Blem (1989)
<i>Ambystoma tigrinum</i>	5.6	Harte and Hoffman (1989)
<i>Ambystoma jeffersonianum</i>	4.5	Pough and Wilson (1977) Freda and Dunson (1985c)
<i>Ambystoma texanum</i>	4.2–5.0	Punzo (1983) Pierce and Wooten (1992b)

acid tolerance (Clark and LaZerte, 1987; Pierce and Harvey, 1987; Andren *et al.*, 1989; Tyler-Jones *et al.*, 1989; Karns, 1992). Thus, acid tolerance of amphibian embryos clearly varies from population to population. Some studies found that geographic variation in acid tolerance of embryos was correlated with the levels of acidity in the pond (Andren *et al.*, 1989; Karns, 1992); more tolerant embryos were found in acidic environments, suggesting that embryos had adapted to acidity. In contrast, other studies found no association between acid tolerance of embryos and the pH of the pond (Tome and Pough, 1982; Cook, 1983; Clark, 1986b; Pierce and Harvey, 1987).

One possible cause of intraspecific differences in acid tolerance is genetic variation. The long-term effects of acidity on amphibian communities depend on the ability of amphibians to adapt successfully to low pH conditions; the presence and nature of genetic variation in acid tolerance will strongly influence their capacity for adaptation. Variation in acid tolerance among egg clutches of the same species has been widely reported in amphibians (Pough, 1976; Tome and Pough, 1982; Pierce and Sikand, 1985; Clark and LaZerte, 1987; Corn *et al.*, 1989; Tyler-Jones *et al.*, 1989), and these differences have been interpreted frequently as evidence of genetic variation in embryo acid tolerance. However, among-clutch and among-population differences in acid tolerance may also result from nongenetic maternal factors and acclimation to acidity; genetic differences can be assessed only through controlled crosses.

Pierce and Sikand (1985) conducted genetic crosses among Wood Frogs, *Rana sylvatica*, from several ponds, and measured the tolerance of embryos and larvae resulting from these crosses. There were large differences in the acid tolerances of embryos from different clutches, but only limited evidence for significant differences among the progeny of different males, which would indicate a direct genetic influence on acid tolerance, since males contribute only a set of paternal genes to their progeny. In contrast, there were large differences among the progeny of females, indicating that maternal factors strongly influence acid tolerance of embryos. Similar results were obtained for embryos of *Rana pipiens* (Corn *et al.*, 1989). Genetic crosses among Wood Frogs from two different ponds in Connecticut (Pierce and Harvey, 1987) produced no evidence that genes influenced geographic variation in embryonic acid tolerance of this species.

Effects of acidity on larvae

Amphibian larvae are more tolerant of acidity than embryos, and typically do not experience acid-stressed mortality until the pH approaches 4 or drops below (Pierce *et al.*, 1984; Freda and Dunson, 1985b). Within species, there is little correlation between the acid tolerance of embryos and larvae (Pierce and Sikand, 1985).

Both laboratory and field studies demonstrate that amphibian larvae lose sodium at low pH; death results when approximately 50% of the body sodium is lost (Freda and Dunson, 1984, 1985b). Sodium loss at low pH is due to inhibition of sodium influx and greatly increased sodium efflux. Increased calcium concentration in the water slows loss of sodium and increases survival time. Tadpoles collected from acidic ponds have lower body sodium, chloride, and water concentrations and display elevated sodium efflux (Freda and Dunson, 1985b). Interestingly, species with lower initial body sodium content display less sodium efflux and higher survival in low pH conditions than species with higher initial sodium content (Freda and Dunson, 1984; Warner *et al.*, 1991).

As in embryos, aluminum interacts with acid toxicity in larval amphibians and may

lead to increased mortality, depressed growth, and small size at metamorphosis (Cummins, 1986; Freda and McDonald, 1989; Freda *et al.*, 1989; Freda, 1991). As with embryos, the interactions of aluminum and acidity are complex and are species dependent.

Amphibian larvae exhibit considerable interspecific (Freda and Dunson, 1984) and intraspecific (Pierce and Sikand, 1985; Pierce and Harvey, 1987) differences in acid tolerance. The maternal effects that play an important role in determining intraspecific variation in embryo acid tolerance are not observed in larvae; instead, direct, genetic factors appear to influence acid tolerance (Pierce and Sikand, 1985).

Although amphibian larvae typically exhibit acid-stressed mortality only at very low pH, acidity may have important nonlethal effects at considerably higher pH. One nonlethal effect with important ecological consequences is inhibition of growth (Freda and Dunson, 1985b; Ling *et al.*, 1986; Cummins, 1986; 1989). Growth of amphibian larvae is inhibited even with short-term (three day) exposure to acidity (Pierce and Montgomery, 1989). Individual variation in larval growth response to low pH has a strong genetic component (Pierce and Wooten, 1992a; Wooten, 1992). Larval growth rates are important ecological variables in amphibian communities, influencing the timing of metamorphosis, susceptibility to predation, competition with other amphibian larvae, size at metamorphosis, survival in the terrestrial environment, and even the timing and success of future reproduction (Wilbur and Collins, 1973; Travis, 1980; Berven, 1982; Berven and Gill, 1983; Brodie and Formanowicz, 1983; Smith, 1987). Considering the importance of larval growth rates for amphibian ecology, acidity may have its most pronounced effect on amphibian populations through inhibition of larval growth.

Several recent studies have examined the interaction of acidity and biotic factors such as density and competition in artificial, replicated populations (Cummins, 1989; Sadinski, 1991; Warner *et al.*, 1991; Sadinski and Dunson, 1992). For example, Warner *et al.* studied the interaction of pH and density on survival and growth of tree frogs in replicated artificial ponds consisting of polyethylene cattle tanks filled with 580 litres of lake water and a sample of plankton, microinvertebrates, and leaf litter from local ponds. Embryos of two species were placed in the ponds, *Hyla gratiosa* (a relatively acid-sensitive species) and *Hyla femoralis* (a more acid-tolerant species). In both species, survival was decreased by low pH. Interestingly, low pH increased the susceptibility of *Hyla gratiosa* to the adverse effects of high density (lengthened larval period and decreased size at metamorphosis). However, low pH had no interactive effect on the adverse results of high density in *Hyla femoralis*, the more acid-tolerant species. Thus, sublethal effects of acidity can potentially alter the structure of amphibian communities.

Effects of acidity on adults

Most research has focused on the effects of acidity on embryological and larval stages of amphibians – it has been generally assumed that adults are relatively well buffered from the effects of acidity and, since most adult amphibians are terrestrial, they would not be exposed to low pH conditions. Recent studies of terrestrial salamanders in North America, however, indicate that this is not the case: soil pH plays an important role in the distribution of these animals, and salamanders exposed to low pH soil exhibit many of the same symptoms exhibited by acid-stressed aquatic larvae.

Several early studies provided some evidence that terrestrial salamanders may avoid

low pH substrates (Vernberg, 1955; Mushinsky and Brodie, 1975). However, Wyman and Hawksley-Lescault (1987) and Wyman (1988) first provided comprehensive data showing correlations between soil pH and the presence of some, but not all, terrestrial amphibians. Subsequent studies showed that amphibian density and the number of amphibian species are positively correlated with soil and humus pH (Wyman and Jancola, 1992). Laboratory studies of the Red Backed Salamander, *Plethodon cinereus*, demonstrated that it avoids low pH substrates when given a choice, and it experiences reduced respiration and growth when reared on low pH substrates. Like aquatic larvae, most terrestrial salamanders lose sodium and water at low pH (Frisbie and Wyman, 1991). Unfortunately, similar studies have not been conducted on adult stages of aquatic frogs and salamanders.

Assessing the impact of acid rain on amphibians

The studies reviewed here indicate that amphibians are susceptible to the toxic effects of acidity, and they are potentially affected by acidification of their habitats. In spite of the fact that much of this research was stimulated by concerns about acid rain, only a few studies have attempted to assess directly the effects of acid precipitation on amphibian communities.

Bradford *et al.* (1992) examined the effects of low pH and aluminum on two species of amphibians, *Rana muscosa* and *Bufo canorus*, which are found at high elevations in the Sierra Nevada of California. These two frogs were of interest because they have experienced dramatic declines in the past two decades, and their breeding sites are extremely low in acid neutralizing capacity (Bradford, 1991; Bradford *et al.*, 1992). Acidification of breeding habitats had been proposed as a possible cause for declines in these two species. Laboratory studies showed that *Rana muscosa* was more tolerant of acidity than *Bufo canorus*, but mortality of embryos and larvae in both species occurred only at pHs well below the most extreme pHs of surface waters in the Sierra Nevada (Bradford *et al.*, 1992). Field studies of potential amphibian breeding habitats in the Sierra Nevada found no significant differences in pH, acid neutralizing capacity, or electrical conductivity between sites with amphibians and those without (D.F. Bradford, personal communication). These data suggest that acid precipitation is not currently causing direct mortality in high elevation populations of amphibians in the Sierra Nevada, although declines may have occurred through sublethal effects.

Corn and Vertucci (1992) also examined the vulnerability to acid precipitation of six species of amphibians occurring in the Rocky Mountains of western North America. Assessment of vulnerability was based on amphibian occurrence, water chemistry of amphibian breeding habitats, and estimated atmospheric deposition at 57 localities. Only one locality possessed water chemistry and sulphate deposition levels that placed it at risk for acidification, and two species of the amphibians were found there. Habitats with amphibian populations did not differ in sensitivity or acidic precipitation from those that had recently lost populations. Laboratory studies (Corn *et al.*, 1989) found that amphibians from the area could tolerate pHs measured in the field during the breeding season. The conclusion of these studies was that acid deposition poses little widespread risk to amphibian populations in the Rocky Mountains, although a few local populations close to point sources of emissions might be at risk (Corn and Vertucci, 1992).

Freda and Dunson (1985a) studied 74 temporary ponds in Pennsylvania and found that

14% of amphibian breeding sites were acidic enough to cause direct mortality to *Ambystoma* embryos. Another 16% were acidic enough to inhibit growth rates. However, no correlation was found between acid deposition and pond pH or alkalinity, and the investigators were unable to determine the relative contribution of acid precipitation and natural acidity to the acidification of these ponds. Many of these ponds were extremely vulnerable to acidification from acid rain because of their very low alkalinity.

Over an eight-year period, Blem and Blem (1989; 1991) observed severe declines in the number of Spotted Salamanders' egg masses laid in many ponds in eastern Virginia; at the same time they noted reduced survivorship of salamander eggs and larvae in this area. For example, detailed study of 218 ponds between 1988 and 1990 (Blem and Blem, 1991) revealed that the number of salamander egg masses declined by 24%. Although the pH of ponds with egg masses was significantly higher than those without egg masses, the pH of ponds where reproduction failed during the study period (number of egg masses declined by 80% and/or all egg masses failed to produce viable larvae) was not significantly different from the pH of ponds with successful reproduction. Furthermore, some acidic ponds continued to support substantial numbers of egg masses. Detailed chemical analysis of water samples from the ponds suggested that high aluminum, copper, and zinc, and low silicon were associated with declines in the number of egg masses over the three-year period. Thus, there was no evidence that acidification was responsible for the observed declines in reproductive success.

Beebee *et al.* (1990) collected extensive data on several ponds at heathland sites in Britain where Natterjack Toads, *Bufo calamita*, have declined over the past 50 years. Two ponds previously used by the toads were now too acidic for successful reproduction. From water analysis and diatom, macrophyte, metal, and soot particles in sediment cores, the authors concluded that atmospheric pollution had been the most significant factor in recent acidification of these two ponds, although land use changes and natural processes may have also contributed to acidification. Acidification most likely led to historical declines of toads at these two sites and now precludes successful reproduction. However, the extent to which acidification has caused toad declines throughout much of its range in Britain is uncertain.

Recognizing the paucity of long-term studies of the effects of acid precipitation on amphibian communities, Freda *et al.* (1991) have recently proposed a logistically reasonable, cost-efficient scheme for detecting changes in amphibian populations associated with acid precipitation. Their plan incorporates chemical analysis of pond samples, annual censuses of egg masses of sensitive species, transplant experiments, and sampling of larval survival. Widespread adoption of this plan would provide badly needed data on the relationship between acidification and amphibian declines.

References

- Albers, P.H. and Prouty, R.M. (1987) Survival of Spotted Salamander eggs in temporary woodland ponds of coastal Maryland. *Environ. Pollut.* **46**, 45–61.
- Andren, C., Henrikson, L., Olsson, M. and Nilson, G. (1988) Effects of pH and aluminum on embryonic and early larval stages of Swedish Brown Frogs *Rana arvalis*, *R. temporaria*, and *R. dalmatina*. *Holarct. Ecol.* **11**, 127–35.
- Andren, C., Marden, M. and Nilson, G. (1989) Tolerance to low pH in a population of Moor Frogs, *Rana arvalis*, from an acid and a neutral environment: a possible case of rapid evolutionary response to acidification. *Oikos* **56**, 215–23.

- Aston, R.J., Beattie, R.C. and Milner, A.G.P. (1987) Characteristics of spawning sites of the Common Frog (*Rana temporaria*) with particular reference to acidity. *J. Zool., Lond.* **213**, 233–42.
- Barinaga, M. (1990) Where have all the froggies gone? *Science* **247**, 1033–4.
- Beattie, R.C., Aston, R.J. and Milner, A.G.P. (1991) A field study of fertilization and embryonic development in the Common Frog (*Rana temporaria*) with particular reference to acidity and temperature. *J. Appl. Ecol.* **28**, 346–57.
- Beattie, R.C., Aston, R.J. and Milner, A.G.P. (1992) Embryonic and larval survival of the Common Frog (*Rana temporaria* L.) in acidic and limed ponds. *Herpetol. J.* **26**, 356–60.
- Beebee, T.J. (1986) Acid tolerance of Natterjack Toad (*Bufo calamita*) development, *Herpetol. J.* **1**, 78–81.
- Beebee, T.J.C., Flower, R.J., Stevenson, A.C., Patrick, S.T., Appleby, P.G., Fletcher, C., Marsh, C., Natkanski, J., Rippey, B. and Battarbee, R.W. (1990) Decline of Natterjack Toad in Britain: palaeoecological, documentary and experimental evidence for breeding site acidification. *Biol. Conserv.* **53**, 1–20.
- Beebee, T.J.C. and Griffin, J.R. (1977) A preliminary investigation into Natterjack Toad (*Bufo calamita*) breeding site characteristics in Great Britain. *J. Zool. Lond.* **181**, 341–50.
- Berven, K.A. (1982) The genetic basis of altitudinal variation in the Wood Frog, *Rana sylvatica*. I. An experimental analysis of life history traits. *Evolution* **36**, 962–83.
- Berven, K.A. and Gill, D.E. (1983) Interpreting geographic variation in life history traits. *Am. Zool.* **23**, 85–97.
- Blem, C.R. and Blem, L.B. (1989) Tolerance of acidity in a Virginia population of the Spotted Salamander, *Ambystoma maculatum* (Amphibia: Ambystomatidae). *Brimleyana* **15**, 37–45.
- Blem, C.R. and Blem, L.B. (1991) Cation concentration and acidity in breeding ponds of the Spotted Salamander, *Ambystoma maculatum* (Shaw) (Amphibia: Ambystomatidae), in Virginia. *Brimleyana* **17**, 67–76.
- Bradford, D.F. (1991) Mass mortality and extinction in a high-elevation population of *Rana muscosa*. *J. Herpetol.* **25**, 174–7.
- Bradford, D.F., Swanson, C. and Gordon, M.S. (1992) Effects of low pH and aluminum on two declining species of amphibians in the Sierra Nevada, California. *J. Herpetol.*, in press.
- Brodie, E.D., Jr. and Formanowicz, D.R. Jr. (1983) Prey size preference of predators: differential vulnerability of larval amphibians. *Herpetologica* **39**, 67–75.
- Burton, T.M. and Likens, G.E. (1975) Salamander populations and biomass in the Hubbard Brook Experimental Forest, New Hampshire. *Copeia* 541–6.
- Clark, K.L. (1986a) Distributions of anuran populations in central Ontario relative to habitat acidity. *Water Air Soil Pollut.* **30**, 727–34.
- Clark, K.L. (1986b) Responses of Spotted Salamander, *Ambystoma maculatum*, populations in central Ontario to habitat acidity. *Can. Field Nat.* **100**, 463–9.
- Clark, K.L. and Hall, R.J. (1985) Effects of elevated hydrogen ion and aluminum concentration on the survival of amphibian embryos and larvae. *Can. J. Zool.* **63**, 116–23.
- Clark, K.L. and LaZerte, B.D. (1987) Intraspecific variation in hydrogen ion and aluminium toxicity in *Bufo americanus* and *Ambystoma maculatum*. *Can. J. Fisheries Aquatic Sci.* **44**, 1622–8.
- Cook, R.P. (1983) Effects of acid precipitation on embryonic mortality of *Ambystoma* salamanders in the Connecticut Valley of Massachusetts. *Biol. Conserv.* **27**, 77–88.
- Cooke, A.S. and Frazer, J.F.D. (1976) Characteristics of newt breeding sites. *J. Zool., Lond.* **178**, 223–36.
- Conant, R. (1975) *A Field Guide to Reptiles and Amphibians of Eastern North America*, 2nd Edition. Boston, MA: Houghton Mifflin.
- Corn, P.S. and Vertucci, F.A. (1992) Descriptive risk assessment of the effects of acidic deposition on Rocky Mountain amphibians. *J. Herpetol.*, **23**, 361–9.

- Corn, P.S., Stolzenburg, W. and Bury, R.B. (1989) Acid precipitation studies in Colorado and Wyoming: interim report of surveys of montane amphibians and water chemistry. *US Fish Wildlife Service Biol. Rep.* 80(40.26).
- Cummins, C.P. (1986) Effects of aluminium and low pH on growth and development in *Rana temporaria* tadpoles. *Oecologia* **69**, 248–52.
- Cummins, C.P. (1988) Effect of calcium on survival of *Rana temporaria* embryos at low pH. *Funct. Ecol.* **2**, 297–302.
- Cummins, C.P. (1989) Interaction between the effects of pH and density on growth and development in *Rana temporaria* L. tadpoles. *Funct. Ecol.* **3**, 45–52.
- Dale, J.M., Freedman, B. and Kerekes, J. (1985a) Acidity and associated water chemistry of amphibian habitats in Nova Scotia. *Can. J. Zool.* **63**, 97–105.
- Dale, J.M., Freedman, B. and Kerekes, J. (1985b) Experimental studies of the effects of acidity and associated water chemistry on amphibians. *Proc. Nova Scotia Inst. Sci.* **35**, 35–54.
- Dickman, M. (1968) The effect of grazing by tadpoles on the structure of a periphyton community. *Ecology* **49**, 1188–90.
- Dunson, W.A. and Connell, J. (1982) Specific inhibition of hatching in amphibian embryos by low pH. *J. Herpetol.* **16**, 314–15.
- Freda, J. (1986) The influence of acidic pond water on amphibians: a review. *Water Air Soil Pollut.* **30**, 439–50.
- Freda, J. (1991) The effect of aluminum and other metals on amphibians. *Environ. Pollut.* **71**, 305–28.
- Freda, J., Cavdek, V. and McDonald, D.G. (1989) Role of organic complexation in the toxicity of aluminum to *Rana pipiens* embryos and *Bufo americanus* tadpoles. *Can. J. Fisheries Aquatic Sci.* **47**, 217–24.
- Freda, J. and Dunson, W.A. (1984) Sodium balance of amphibian larvae exposed to low environmental pH. *Physiol. Zool.* **57**, 435–43.
- Freda, J. and Dunson, W.A. (1985a) The effect of acidic precipitation on amphibians breeding in temporary ponds of Pennsylvania. *US Fish Wildlife Service Biol. Rep.* 80(40.22).
- Freda, J. and Dunson, W.A. (1985b) Field and laboratory studies of ion balance and growth rates of rapid tadpoles chronically exposed to low pH. *Copeia* 415–23.
- Freda, J. and Dunson, W.A. (1985c) The influence of external cation concentration on the hatching of amphibian embryos in water of low pH. *Can. J. Zool.* **63**, 2649–56.
- Freda, J. and Dunson, W.A. (1986) Effects of low pH and other chemical variables on the local distribution of amphibians. *Copeia* 454–66.
- Freda, J. and McDonald, D.G. (1989) Effects of aluminum on the Leopard Frog *Rana pipiens*: life stage comparisons and aluminum uptake. *Can. J. Fisheries Aquatic Sci.* **47**, 210–16.
- Freda, J., Sadinski, W.J. and Dunson, W.A. (1991) Long term monitoring of amphibian populations with respect to the effects of acidic deposition. *Water Air Soil Pollut.* **55**, 445–62.
- Frisbie, M.P. and Wyman, R.L. (1991) The effects of soil pH on sodium balance in the Red-backed Salamander, *Plethodon cinereus* and 3 other terrestrial salamanders. *Physiol. Zool.* **64**, 1050–68.
- Gascon, C. and Planas, D. (1986) Spring pond water chemistry and the reproduction of the Wood Frog, *Rana sylvatica*. *Can. J. Zool.* **64**, 543–50.
- Gascon, C., Planas, D. and Moreau, G. (1987) The interaction of pH, calcium and aluminium concentrations on the survival and development of Wood Frog (*Rana sylvatica*) eggs and tadpoles. *Ann. R. Zool. Soc. Belg.* **117**, 189–99.
- Gorman, E., Eisenreich, S.J., Ford, J. and Santelmann, M.V. (1985) Chemistry of bog waters. In Stumm, W. (ed) *Chemical Processes in Lakes*. New York: Wiley.
- Gosner, K.L. and Black, I.H. (1957) The effects of acidity on the development and hatching of New Jersey frogs. *Ecology* **38**, 256–62.

- Hagström, T. (1981) Reproductive strategy and success of amphibians in water acidified by atmospheric pollution. *Proc. Eur. Herpetol. Symp. C.W.L.P. Oxford*, 1980, pp. 55–57.
- Harte, J. and Hoffman, E. (1989) Possible effects of acidic deposition on a Rocky Mountain population of the Tiger Salamander *Ambystoma tigrinum*. *Conserv. Biol.* **3**, 149–58.
- Henrikson, B. (1990) Predation on amphibian eggs and tadpoles by common predators in acidified lakes. *Holarct. Ecol.* **13**, 201–6.
- Huckabee, J.W., Goodyear, C.P. and Jones, R.D. (1975) Acid rock in the Great Smokies: unanticipated impact on aquatic biota of road construction in regions of sulfide mineralization. *Trans. Am. Fish. Soc.* **104**, 677–84.
- Karns, D.R. (1983) Toxic bog water in northern Minnesota peatlands: ecological and evolutionary consequences for breeding amphibians. PhD Dissertation, University of Minnesota.
- Karns, D.R. (1992) Effects of acidic bog habitats on amphibian reproduction in a northern Minnesota Peatland. *J. Herpetol.*, **26**, 401–12.
- Leuven, R.S.W., den Hartog, C., Christiaans, M.M.C. and Heijligers, W.H.C. (1986) Effects of water acidification on the distribution patterns and the reproductive success of amphibians. *Experientia* **42**, 495–503.
- Ling, R.W., Van Amberg, J.P. and Werner, J.K. (1986) Pond acidity and its relationship to larval development of *Ambystoma maculatum* and *Rana sylvatica* in upper Michigan. *J. Herpetol.* **20**, 230–6.
- Mushinsky, H.R. and Brodie, E.D., Jr. (1975) Selection of substrate pH by salamanders. *Am. Midland Nat.* **93**, 440–3.
- Padhye, A.D. and Ghate, H.V. (1988) Effect of altered pH on embryos and tadpoles of the frog *Microhyla ornata*. *Herpetol. J.* **1**, 276–9.
- Pierce, B.A. (1991) Additional studies of acid sensitivity of eggs and larvae of two pothole-dwelling amphibians from Utah. Final Report to the Air Quality Division, National Park Service, US Department of the Interior.
- Pierce, B.A., Hoskins, J.B. and Epstein, E. (1984) Acid tolerance in Connecticut Wood Frogs (*Rana sylvatica*). *J. Herpetol.* **18**, 159–67.
- Pierce, B.A. and Harvey, J.M. (1987) Geographic variation in acid tolerance of Wood Frogs. *Copeia* **1987**, 94–103.
- Pierce, B.A. and Montgomery, J. (1989) Effects of short-term acidification on growth rates of tadpoles. *J. Herpetol.* **23**, 97–102.
- Pierce, B.A. and Sikand, N. (1985) Intrapopulation variation in acid tolerance of Connecticut Wood Frogs: genetic and maternal effects. *Can. J. Zool.* **63**, 1647–51.
- Pierce, B.A. and Wooten, D.K. (1992a) Genetic variation in tolerance of amphibians to low pH. *J. Herpetol.*, **26**, 422–9.
- Pierce, B.A. and Wooten, D.K. (1992b) Acid tolerance of *Ambystoma texanum* from central Texas. *J. Herpetol.* **26**, 230–2.
- Portnoy, J. (1990) Breeding biology of the Spotted Salamander *Ambystoma maculatum* (Shaw) in acidic temporary ponds on Cape Cod, USA. *Biol. Conserv.* **53**, 61–5.
- Pough, F.H. (1976) Acid precipitation and embryonic mortality of Spotted Salamanders *Ambystoma maculatum*. *Science* **192**, 68–72.
- Pough, F.H. and Wilson, R.E. (1977) Acid precipitation and reproductive success of *Ambystoma* salamanders. *Water Air Soil Pollut.* **7**, 531–44.
- Punzo, F. (1983) Effects of environmental pH and temperature on embryonic survival capacity and metabolic rates in the Smallmouth Salamander, *Ambystoma texanum*. *Bull. Environ. Contam. Toxicol.* **31**, 467–73.
- Sadinski, W.J. and Dunson, W.A. (1992) A multilevel study of low pH on amphibians of temporary ponds. *J. Herpetol.* **26**, 413–22.
- Schlichter, L. (1981) Low pH affects the fertilization and development of *Rana pipiens* eggs. *Can. J. Zool.* **59**, 1693–9.

- Seale, D.B. (1980) Influence of amphibian larvae on primary production, nutrient flux, and competition in a pond ecosystem. *Ecology* **61**, 1531–50.
- Smith, D.C. (1987) Adult recruitment in chorus frogs: effects of size and date at metamorphosis. *Ecology* **68**, 344–50.
- Strijbosch, H. (1979) Habitat selection of amphibians during their aquatic phase. *Oikos* **33**, 363–72.
- Tome, M.A. and Pough, F.H. (1982) Responses of amphibians to acid precipitation. In Haines, T.A. and Johnson, R.E. (eds) *Acid Rain/Fisheries*, pp. 245–54. Bethesda, MD: American Fisheries Society.
- Travis, J. (1980) Phenotypic variation and the outcome of interspecific competition in hylid tadpoles. *Evolution* **34**, 40–50.
- Tyler-Jones, R., Beattie, R.C. and Aston, R.J. (1989) The effects of acid water and aluminium on the embryonic development of the Common Frog, *Rana temporaria*. *J. Zool. Lond.* **219**, 355–72.
- Urch, U.A. and Hedrick, J.L. (1981) Isolation and characterization of the hatching enzyme of the amphibian *Xenopus laevis*. *Arch. Biochem. Biophys.* **206**, 424–31.
- Vernberg, F.J. (1955) Correlation of physiological and behavioral indexes of activity in the study of *Plethodon cinereus* (Green) and *Plethodon glutinosus* (Green). *Am. Midland Nat.* **54**, 382–93.
- Vitt, L.J., Caldwell, J.P., Wilbur, H.M. and Smith, D.C. (1990) Amphibians as harbingers of decay. *BioScience* **40**, 418.
- Wake, D.B. (1991) Declining amphibian populations. *Science* **253**, 860.
- Warner, S.C., Dunson, W.A. and Travis, J. (1991) Interaction of pH, density, and priority effects on the survivorship and growth of 2 species of hylid tadpoles. *Oecologia* **88**, 331–9.
- Wilbur, H.M. and Collins, J.P. (1973) Ecological aspects of amphibian metamorphosis. *Science* **182**, 1305–14.
- Wooten, D.K. (1992) The effect of low pH and genetics on growth of *Pseudacris streckeri* larvae. M.S. Thesis, Baylor University.
- Wyman, R.L. (1988) Soil acidity and moisture and the distribution of amphibians in five forests of southcentral New York. *Copeia* 394–9.
- Wyman, R.L. and Hawksley-Lescault, D.S. (1987) Soil acidity affects distribution, behavior, and physiology of the salamander *Plethodon cinereus*. *Ecology* **68**, 1819–27.
- Wyman, R.L. and Jancola, J. (1992) Degree and scale of terrestrial acidification and amphibian community structure. *J. Herpetol.*, **26**, 392–401.