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Response of *Gammarus pulex* and *Baetis rhodani* to springtime acid episodes in humic brooks

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HIGHLIGHTS

• G. pulex and B. rhodani were exposed to a gradient of pH and Ali in humic brooks.
• Low pH and high Ali reduced the Na body content, which was linked to mortality.
• As acidity increased, most body base cations decreased, while no Al accumulated.
• Ca accumulated in G. pulex and Mg accumulated in B. rhodani as the pH decreased.
• Overall, pH > 5.7–6.0 and Ali < 15–20 μg/L sustains healthy salmonid prey populations.

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ABSTRACT

While chronic acidification of water bodies has been steadily decreasing, episodic acidification continues to affect stream biology by temporarily decreasing pH and mobilizing aluminum. These events are becoming more common as climate change renders more frequent and intense storms and flooding. Throughout Scandinavia, the effects of acidification have been mitigated by liming since the 1980s, but remediation efforts can now be reduced. While transient acidity may reduce fish populations, also other species in streams are affected. In this in-stream study, two macro-invertebrates (*Gammarus pulex* and *Baetis rhodani*), both known as salmonid prey organisms, were exposed to snowmelt in six humic brooks with a natural gradient of pH and inorganic monomeric Al (Al_i). We hypothesize that acid toxicity thresholds can be defined using lethal (mortality) and sublethal (changes in body elemental content) metrics. Periodic observations were made of mortality and whole body concentrations of base cations (BC: Ca, Mg, Na and K) and metals (Al, Fe, Zn and Mn). Mortality increased dramatically at pH < 6.0 and Al_i > 15 μg/L for *G. pulex* and at pH < 5.7 and Al_i > 20 μg/L for *B. rhodani*. No accumulation of Al was found. The invertebrate body Na concentration decreased when pH dropped, suggesting that osmoregulation in both species was affected. In contrast to general BC pattern, Ca concentration in *G. pulex* and Mg concentration in *B. rhodani* increased when pH decreased. Although Al_i strongly correlates to pH, the Al composition of soil and bedrock also influences Al availability, potentially contributing to toxic Al_i episodes. The estimated values calculated in this study can be used to improve water quality criteria and as thresholds to adjust doses of lime compared to old recommendations in ongoing liming programs. Such adjustments may be critical since both Al_i and pH levels have to be balanced to mitigate damage to recovering stream ecosystems.

1. Introduction

Sulfur emissions and associated acid deposition have been reduced over several years in northern Europe and North America (Vestreng et al., 2007). This reduction has resulted in improved water quality as concentrations of SO_4 have decreased while pH and alkalinity have increased (Skjelkvale et al., 2005; Stoddard et al., 1999). While concurrent records of biological recovery are scarce, significant data are beginning to emerge (Kowalik et al., 2007; Murphy et al., in press; Wright et al., 2005). Due to the slow rate at which biological systems recover, acidification will continue to be an environmental problem for several decades (Morth et al., 2005; Wright et al., 2005). As chronic acidification has decreased, episodic acid surges have become more of a threat to stream biota (e.g., Lepori and Ormerod, 2005; McCormick et al., 2009). Spring floods are typical acidic episodes, where melting snow can produce considerable pH depression in receiving streams. Aquatic species display substantial variability in their sensitivity to acid conditions, and many are particularly sensitive during the spring because of the presence of juvenile stages. Even a single acid episode, therefore, can have significant long-term consequences on the biodiversity of an aquatic community (Bradley and Ormerod, 2002).

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Low pH dissolves and solubilizes aluminum (Al) in the soil, generating inorganic monomeric Al (Al_i), which is the bioavailable and highly toxic Al-fraction (Driscoll and Schecher, 1990; Gensemer and Playle, 1999). The combined effect of Al and pH in acid toxicity can cause a positive or negative interplay depending on the water chemistry (reviewed by Havas and Rosseland, 1995). To mitigate the effects of acidification in aquatic systems, including fish mortalities (Dickson, 1978; Driscoll et al., 1980) and loss of biological diversity (Haines, 1981), Sweden began an intense lime treatment program c. 1980. The addition of lime raises the pH and depresses the concentration of Al_i, reducing associated aquatic toxicity. Additionally, when lime is applied to soil or wetlands, Al becomes less mobile and remains bound to soil particles, significantly reducing leaching to surface and/or groundwater. In total, over 4000 million SEK (480 million Euros; 630 million U.S. dollars) has been spent on liming, making this program one of the largest, from a cost standpoint, environmental efforts in Sweden. As a consequence of the declining acidification levels, the amount of lime distributed has decreased by 40% compared to the maximum amount applied in 2000. It is reasonable to assume that liming treatments will continue in the future, but with reduced doses to avoid surges of Al_i in streams with salmonid fish populations. The current empirical guidelines for liming in waters with brown trout (*Salmo trutta* L.) populations require a pH of >5.6, and a maximum Al_i concentration of <50 μg/L at any time during the year. In waters containing natural salmon (*Salmo salar*), guidelines are more restrictive, requiring pH of >6.0 and Al_i of <30 μg/L (Swedish EPA, 2010). These limits are monitored in the streams at least six times per year, with three samples taken during high flow. The Al availability varies locally, and a thorough knowledge and understanding of local characteristics is required to plan and balance the lime dose in order to avoid the concomitant pH–Al toxicity and mitigate potential effects. This site-specific understanding is even more important in the present recovery phase as surviving biota might be more sensitive after being subjected to disturbance. Recovery can be prevented or impeded by impoverished water quality (Kowalik et al., 2007; Lepori et al., 2003), biotic interactions (Ledger and Hildrew, 2005), or scarcity of acid-sensitive colonists (Monteith et al., 2005).

Examples of indirect effects caused by acidity that likewise deserve study are the predator–prey interactions of salmonid fishes and macroinvertebrates. Like fish, benthic organisms are affected by acidification (Okland and Okland, 1986). In crustacean species, acid stress is known to significantly decrease osmolality and hemolymph Na^+ concentrations. In insects, acid stress tends to disrupt the ion balance by reducing major ion concentrations (Johnson et al., 1993) rather than affecting internal pH; acid-tolerant species may have a greater ability to maintain the crucial physiological Na balance (Harrison, 2001). Respiration can also contribute to the regulation of external pH changes, but the relative importance of various tissues and organs involved in pH regulation may differ quantitatively between species (Cooper, 1994). Crustacea and Bivalvia also lose ions, although they can simultaneously increase their Ca levels to buffer acid stress by mobilizing ions from their exoskeleton, shell and mantle (Johnson et al., 1993).

The crustacean *Gammarus pulex* and the mayfly *Baetis rhodani* are known to be sensitive to acidity and are used in several biotic indices (Murphy et al., 2013). *B. rhodani* has also been used to monitor the success of liming treatment (Ahlstrom and Johansson, 2010; Lingdell and Engblom, 1995). Both of these macro-invertebrates are salmonid prey (Macneil et al., 1999) and are considered here as indicators of stream habitat that is well-suited for salmon and trout. While there are early short-term invertebrate studies that have simulated acid episodes and liming (e.g., McCahon and Pascoe, 1989; Ormerod et al., 1987), the tolerance of *G. pulex* and *B. rhodani* has been primarily examined in waters that have low levels of organic material (e.g. Lepori and Ormerod, 2005; Merrett et al., 1991; Raddum and Fjellheim, 1987). Few studies have combined in situ quantitative assessment of bioavailable Al (Al_i) and the response of these two representative species when exposed to episodic acid stress in humic waters. Improved instrumentation and refined analytical methods have resulted in the ability to inexpensively detect low concentrations of bioavailable and toxic Al_i and elemental analyses for small sample sizes of biota, thus increasing the number of analyses that can be performed. Furthermore, while earlier studies were designed to detect an effect of pH and Al on mortality or drift, and possibly Al accumulation in tissues, they were not designed to identify a no effects concentration (NEC) at which water becomes toxic. Such information can be critical in designing a tolerance gradient along which lime doses can be reduced in order to prevent acid toxicity to salmonid prey which could jeopardize the recovering stream ecosystems. It is also important to focus on biological recovery in the context of the EU Water Framework Directive (EC, 2000) as differences between undisturbed and disturbed conditions need to be established (Schneider and Lindstrom, 2011).

In this study, acid toxicity thresholds of two key organisms, *G. pulex* and *B. rhodani*, were estimated. In addition to assessing mortality, body concentrations of base cations (Ca, Mg, Na and K) and metals (Al, Fe, Mn and Zn) were measured to document loss or accumulation and they were also related to mortality. These toxicity data can be used to improve the instruments for monitoring the effects of liming, and to enable balanced reductions of lime doses. We used in-stream exposures in humic streams with a natural Al_i and pH gradient representative of a spring flood. We hypothesized that acid toxicity thresholds can be quantified and defined by mortality and changes in the whole body elemental content.

2. Material and methods

2.1. Study area

Field exposures were performed in six brooks during two spring floods: 3 April to 7 May 2001 (*G. pulex*) and 21 March to 26 April 2002 (*B. rhodani*). The exposures in 2002 were conducted in parallel with exposures of brown trout (*S. trutta* L., Andrén and Rydin, 2012). The brooks are located on coniferous hills in a base-poor region in central Sweden (see Fig. A in Suppl. data); the water was characterized as soft and humic. Catchment geology consisted of granite and gneiss bedrock overlain by a thin layer of till, rendering low buffering capacity. The catchment areas were small (2–7 km^2) and dominated by spruce and pine forests (74–97%) with minor wetlands (2–26%) upstream from the exposure sites. Although some foresting activities occur in the watersheds, the brooks are relatively pristine. However, until the year 2000, lime was spread downstream of the headwater Havssvalgsb. (site 4070; see Table 1 and Fig. A Suppl. data). Some of this lime may have accumulated in the lake 2.5 km upstream of Örvallsb. (site 4250), and impacted the water quality of the stream if strong undercurrents re-suspended and dissolved the lime from the sediment. The brooks sampled for this study were selected based on their water quality (Andrén, 1995), to represent different levels of acidity during the snow melt. Waters of different quality (e.g., acidic, and more neutral or limed water) did not converge or mix near the experimental cage sites, resulting in stable exposure environments.

2.2. Experimental design, invertebrate collection, and exposure

One exposure container was used per stream and species. Approximately 60 individuals of *G. pulex* or 100 individuals of *B. rhodani*, respectively, were placed in the appropriate container. Test organisms were not fed during the exposures. The exposure containers were constructed from 5 L polypropylene jugs with 1 dm^2 openings covered with a 0.5 mm stainless steel mesh net. Disposable plastic 5 mL pipettes with enlarged, diagonal openings at the tip were used to gently transfer individual invertebrates between containers and sample canisters. Invertebrates used in the study were collected using a kick net in two nearby streams where each species was