Life at the edge: Benthic invertebrates in high altitude Andean streams

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Chapter 3

Metal leaching and altitude confine benthic macroinvertebrate community composition in Andean streams

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Abstract

Andean streams drain metal-rich bedrock and are subjected to an extreme altitude gradient, which may create highly selective conditions for life. The aim of the present study was therefore to evaluate the simultaneous effects of metals and altitude on benthic macroinvertebrate community composition in Andean streams. Polluted sites were characterized by high metal concentrations and low pH, and high altitude sites by high ultraviolet-B radiation and low concentration of dissolved organic matter. Canonical Correspondence Analysis indicated that the patterns in faunal composition were best explained by metal pollution followed by altitude, with dipterans and collembolans occurring mostly under harsh conditions of high altitude and high metal levels. Interaction between metals and altitude was most evident at polluted sites. It is concluded that in Andean streams metal leaching from igneous rock and altitude may be important factors confining benthic macroinvertebrate communities reducing their numbers and changing their composition towards specialized taxa.

Introduction

The tropical Andes encompass vast areas with altitudes above 4000 m, creating environments that potentially challenge the survival and persistence of biota. With increasing altitude environmental conditions, such as water temperatures, oxygen levels, nutrient concentrations and solar ultraviolet-B radiation (UV-B, 280–320 nm) become more extreme and consequently, the diversity of aquatic communities in mountain streams exhibits a decline towards the summits (Vinebrooke & Leavitt, 1999; Rostgaard & Jacobsen, 2005; Jacobsen & Marin, 2007). These correlated factors act together on alpine biota and the integrated effect may be more important than the effects of the single factors. Accordingly, vertical zonations of species distribution and variations in community structure have been better explained by altitude than by small-scale factors associated with the specific habitat (Jacobsen, 2003; Finn & Poff 2005). Yet, life at the highest altitudes (> 4000 m) under this unique suite of environmental conditions has seldomly been studied.

Environmental pollution may add further stress to life at high altitude. In the Andes metal pollution is caused by mining, but in addition, the natural weathering of metal-rich bedrock produces a continuous leaching of metals and acid drainage into streams, affecting water quality and benthic communities (Ministerio de Energía y Minas, 1998). The few studies evaluating the effects of increased acidity and metal mixtures in high altitude tropical (Loayza-Muro et al., 2010; van Damme et al., 2008) and temperate streams (Courtney and Clements, 2000), have shown a reduction of invertebrate abundance and sensitive taxa richness, and a significant shift in community composition towards more tolerant taxa (Gerhardt et al., 2004). Indirect effects include smothering of the streambed by metal oxyhydroxide precipitates, restricting available habitats for benthic fauna, impoverishing food quality, and modifying interactions between functional feeding groups (O'Halloran et al., 2008).

Since it remains unknown if metals and altitude shape communities in high mountains as single independent stressors or as one combined, 'multi-stress' selective force, the aim of this study was to examine the simultaneous effect of metals and high altitude on benthic macroinvertebrate community composition in Andean streams of the Cordillera Blanca, Peru.

Materials and methods

Study sites

In Central-Northern Peru (Ancash region), the Cordillera de los Andes comprise two parallel mountain ranges, the eastern Cordillera Blanca and the western Cordillera

Negra, which run along the Santa River. Below the permanent snow-line in the Cordillera Blanca, between 3700–4400 m a.s.l., slopes have been modified for small agriculture and cattle rearing. Streams in this area are fast flowing, with substrate consisting of gravel, pebble and cobbles in runs and riffles. They show transparent waters, a very sparse macrophyte growth and are almost completely unshaded due to the natural absence of trees or bushes growing on the banks, particularly above 4000 m a.s.l.

Eight sampling sites were selected between 3,087–4,079 m a.s.l. in the Cordillera Blanca, four being located in the Quilcayhuanca catchment and four in the Rúrec catchment (Figure 1). In both catchments a reference and a naturally polluted stream were selected at low (3,040 and 3,087 m a.s.l.) and high altitude (3,998 and 4,079 m a.s.l.). The reference sites had their source in clean lagoons or springs with a very low metal background, while the naturally polluted sites had their origin in separate gorges characterized by metal rich bedrock. All sites were sampled on the 10 and 11 of July (dry season) and on the 27 and 28 of November 2010 (rainy season), and on the 6 and 7 of March (rainy season) and on the 13 and 14 of July 2011 (dry season).

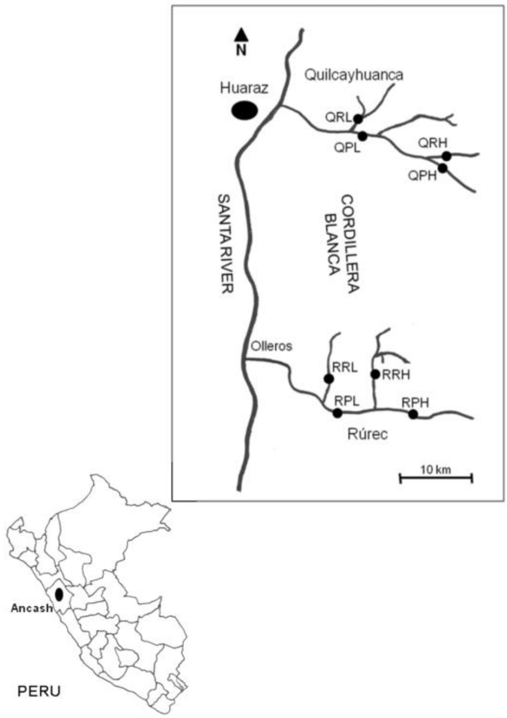

Figure 1. Map of the sampling area, indicating the sampling sites in the streams in the Quilcayhuanca and Rúrec catchments in the Cordillera Blanca. Labels for streams are as follows: QRL = Quilcayhuanca Reference Low, QPL = Quilcayhuanca Polluted Low, QRH = Quilcayhuanca Reference High, QPH = Quilcayhuanca Polluted High, RRL = Rúrec Reference Low, RPL = Rúrec Polluted Low, RRH = Rúrec Reference High, RPH = Rúrec Polluted High. High, 3,998 - 4,079 m a.s.l.; Low, 3,040 - 3,087 m a.s.l.

Physical chemical characteristics

Measurements of pH, temperature (C°), conductivity and dissolved oxygen were performed at each sampling site using a WTW Multi 340i instrument equipped with SenTix® 41-3, TetraCon® 325-3 and CellOx® 325-3 probes (Weilheim, Germany). Solar ultraviolet-B irradiance (280–315 nm) was measured at the water surface with a Delta Ohm HD 2302.0 photo-radiometer and a LP 471 UVB probe with a quartz cosine corrector (Padua, Italy). UV-B irradiance was measured every two days during July (dry season) and November (rainy season), from 10:00 h to 14:00 h under full sun conditions, and the maximum values were averaged. Water samples for analysis of dissolved organic carbon (DOC) were filtered through 0.45 μm membranes in the field, acidified with HCl, and stored in amber glass bottles at 4°C. The concentration of DOC was determined within 24 h using a Shimadzu TOC-5000 analyzer (Columbia, ML, U.S.A.). Stream depth was calculated from four measurements at each of three parallel cross-sections with a calibrated stick. Mean current velocity was obtained with a mechanical flow-meter (Wildlife Supply Company, Buffalo, NY, USA). Discharge was calculated as the average of the three products of mean current velocity, mean depth and stream width at three cross-sections. For determining hardness, water samples were taken with 500-mL glass bottles below the water surface, kept at 4°C in a Styrofoam box, and analyzed using standard methods (Clesceri et al., 1998). Water samples for total metals analysis were taken with 1-L polypropylene bottles, preserved with 10 N HNO_3 and analyzed by ICP-ES (induced-coupled plasma emission spectroscopy) (U.S. EPA, 1994). Samples for determining hardness and metals were taken in triplicate at each sampling site.

Since we expected the naturally polluted sites to contain a mixture of metals with potential additive effects, we calculated the cumulative criterion unit (CCU), which is a cumulative measure for all metals at a specific site, allowing examining the relationships between benthic community structure and metal concentrations (Clements et al., 2000). The CCU is defined as the ratio between the stream metal concentration and the U.S. Environmental Protection Agency (EPA) criterion value for toxicity, summing the ratios for all metals measured at a specific site (U.S. EPA, 1986): $CCU = \sum m_i/c_i$, where m_i is the total recoverable metal concentration (dissolved and suspended fractions) and c_i is the criterion value for the *i*th metal. The criterion value is based on U.S. EPA guidelines on critical concentrations, which may harm aquatic organisms when exceeded. CCU values are scaled as follows: < 1.0, no adverse effects; 1.0–2.0, adverse effects; 2.0–10.0, significant mortality to sensitive species and altered benthic community composition expected; > 10.0, extremely toxic (Clements et al., 2000). Because water hardness affects the toxicity and bioavailability of some metals, criterion values for Cd, Cu, Pb and Zn were modified according to (U.S. EPA, 1986) to account for variation in water hardness between streams.

For Al, Fe and Mn no adjustment was needed and we followed the EPA criterion values (U.S. EPA, 1986).

Invertebrate sampling

At each sampling site, six Surber samples (each 20 cm^2, mesh size 250 μm) were collected randomly, three from gravel-pebble substratum and three from stones in different microhabitats over representative sections, including stagnant water along the banks. All samples were preserved in 70% (v/v) ethanol, and sorted in the laboratory with the use of a Zeiss Stemi DV4 stereomicroscope (Göttingen, Germany). Since knowledge on Peruvian stream fauna, and South American streams in general is scarce, insects could only be identified with certainty to the family level and most non-insects to order or class, using taxonomical keys (Roldán, 1996; Domínguez and Fernández, 2009). This relatively coarse level of taxonomic resolution considers the high correlation between family richness of insects at individual stream sites and species richness, and may allow comparative analyses of community structure and detecting effects of pollution on benthic communities (Loayza-Muro et al., 2010; Vanderklift, 1996). In addition, the relative abundance of Ephemeroptera, Trichoptera and Plecoptera (%EPT) was calculated because these groups are generally considered to be sensitive to environmental pollution.

Canonical correspondence analysis

We selected Canonical Correspondence Analysis (CCA) based on the idea that benthic faunal assemblages most likely show non-linear relationships to the environment (ter Braak and Šmilauer, 2002). CCA was performed to examine the effects of altitude, metal pollution, and the interaction of these two factors on community composition. Prior to the CCA, the family counts were log-transformed to reduce the effects of highly abundant taxa (ter Braak and Šmilauer, 2002). The CCA analysis was performed applying default options of the CCA function in the vegan package (Oksanen et al., 2011) in R (R Development Core Team, 2011). The significance of the three canonical axes was assessed through permutation tests as implemented in the anoca.cca function of this same package, applying a maximum of 9999 permutations.

Results

Characterization of sampling sites

The UV-B radiation level at 4000 m was two-fold that at 3000 m in the dry and rainy seasons (Figure 2, Supplementary data Table 1), while water temperature, dissolved oxygen, stream discharge and water flow were similar. DOC concentration at 4000 m a.s.l.

was half that at 3000 m a.s.l. and decreased in the dry season. Conductivity and hardness were higher and pH was lower in the polluted streams compared to the reference streams. Likewise, the concentrations of all metals and CCU values were higher in the polluted streams than in the reference streams (Supplementary data Table 2), and in most of the cases they increased with altitude and from the rainy to the dry season (Figure 3). The metal concentrations at the polluted sites (e.g. Al, 4.83 mg/L; As, 0.028 mg/L; Fe, 58.8 mg/L; Mn, 1.17 mg/L; Ni, 0.11 mg/L; Zn, 0.278 mg/L) ranged from 2 (Cu) to 588 (Fe) times those at the reference sites, indicating a high degree of natural leaching. At these sites, the streambed was smothered by orange precipitates and encrusted layers, most likely comprising iron oxyhydroxides. The mean CCU ranged from 1.04 to 94.81, with all polluted sites showing values higher than 10.0, meaning potentially significant mortality to sensitive species and altered benthic community composition. Although the CCU at reference sites exceeded 1.0, indicating metal pollution, the large differences with CCU values at polluted sites allowed separating the two site categories.

Relationship between benthic macroinvertebrate community composition and physical-chemical characteristics

A total of 28 families of aquatic insects and 10 other invertebrate taxa were identified (Supplementary data Table 3). Among the insects, Diptera (11), Coleoptera (7) and Trichoptera (4) were represented by the largest number of families. The number of individuals and the number of taxa did not differ between reference sites at 3000 and 4000 m (Table 1). In contrast, the number of individuals, number of taxa, and %EPT were much lower at the polluted sites than at the reference sites. Fewest taxa survived under the harshest conditions, the polluted sites during the dry season.

The CCA ordination revealed that the patterns (Figure 4A CCA axis 1) in macroinvertebrate family composition were best related to pollution. The families Hirudidae, Oligochaeta (Annelida), Ephydridae, Chironomidae (Diptera), Ptilidae (Coleoptera), Isotomidae, Sminthuridae, Hypogastruridae (Collembola) and Acari persisted at high pollution levels (left side of CCA axis 1), suggesting a lower sensitivity towards metal contamination, and probably also to low pH. In contrast, Baetidae (Ephemeroptera), Hydrobioscidae, Hydroptilidae, Limnephilidae (Trichoptera), Dixidae, Empididae (Diptera), Scirtidae (Coleoptera), Hydracarina (Acari) and Planariidae (Turbellaria) appeared more sensitive to metal pollution, since these taxa were mostly arranged at the opposite (right) side of CCA axis 1. Altitude related mostly to the second CCA axis (Figure 4A). Heptageniidae (Ephemeroptera), Leptoceridae (Trichoptera), Blephariceridae, Simuliidae, Tipulidae (Diptera), Dytiscidae, Gyrinidae (Coleoptera) and Lymnaeidae (Gasteropoda) were more abundant better at low elevations (low side of CCA axis 2), whereas Perlidae (Plecoptera), Ceratopogonidae, Elmidae, Psychodidae, Tabanidae (Diptera), Staphylinidae (Coleoptera) and Amphipoda, Ostracoda, Copepoda (Crustacea)

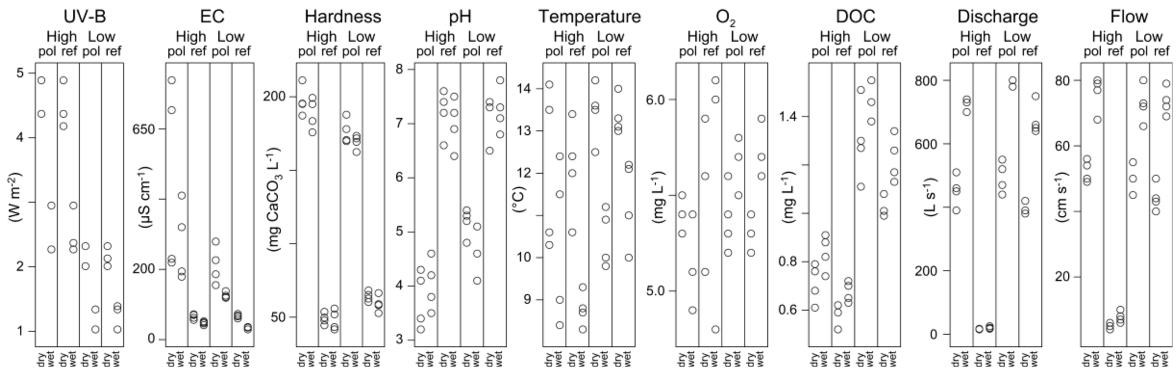

Figure 2. Dot plots of the environmental properties repeatedly measured at eight sampling sites. High, 4000 m a.s.l.; Low, 3000 m a.s.l.; Pol, polluted; Ref, reference.

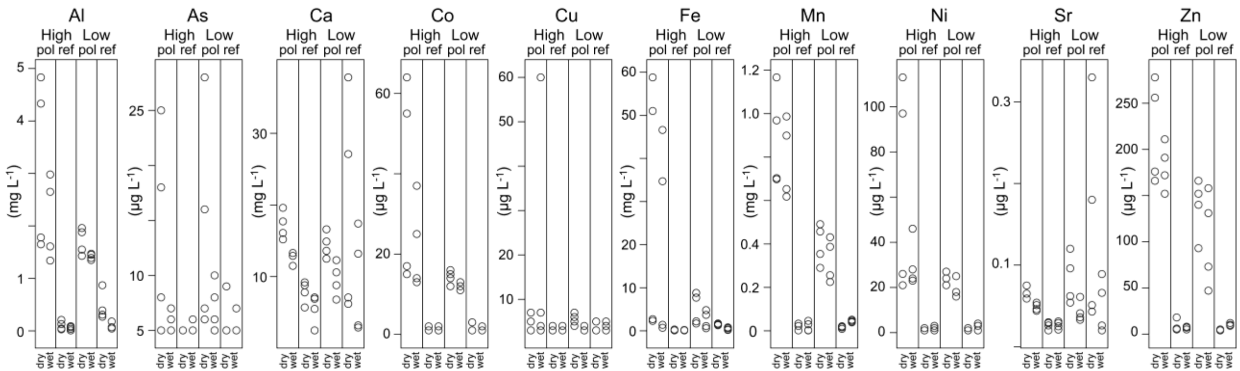

Figure 3. Dot plots of the metal concentrations repeatedly measured at eight sampling sites. High, 4000 m a.s.l.; Low, 3000 m a.s.l.; Pol, polluted; Ref, reference

Table 1. Number of individuals, number of taxa, and relative abundance of individuals belonging to Ephemeroptera, Plecoptera and Trichoptera (%EPT) at the eight sampling sites in the Cordillera Blanca area, Peru. Status: Pol, polluted; Ref, reference. Months: J, July; D, December; M, March. Year: 1, 2010; 2, 2011.

Catchment	Altitude	Status	Month	Number of individuals	Number of taxa	%EPT
Quilcayhuanca	3,998 m	Pol	J1	54	6	1.9
			D1	47	8	2.1
			M2	39	5	0
			J2	89	8	2.2
		Ref	J1	930	22	10.3
			D1	993	13	9.7
			M2	671	11	12.5
			J2	764	14	13.6
	3,040 m	Pol	J1	95	7	2.1
			D1	83	8	2.4
			M2	92	8	2.2
			J2	78	6	0
		Ref	J1	1403	18	8.3
			D1	1208	10	7.4
			M2	1059	13	7.0
			J2	1499	17	7.5
Rúrec	4,079 m	Pol	J1	45	4	0
			D1	25	5	0
			M2	20	4	0
			J2	23	5	0
		Ref	J1	466	11	8.2
			D1	161	13	10.6
			M2	132	15	13.6
			J2	429	14	10.0
	3,087 m	Pol	J1	18	4	0
			D1	12	5	0
			M2	63	7	0
			J2	61	7	0
		Ref	J1	1108	15	6.7
			D1	390	14	6.7
			M2	469	10	7.0
			J2	1454	18	5.9

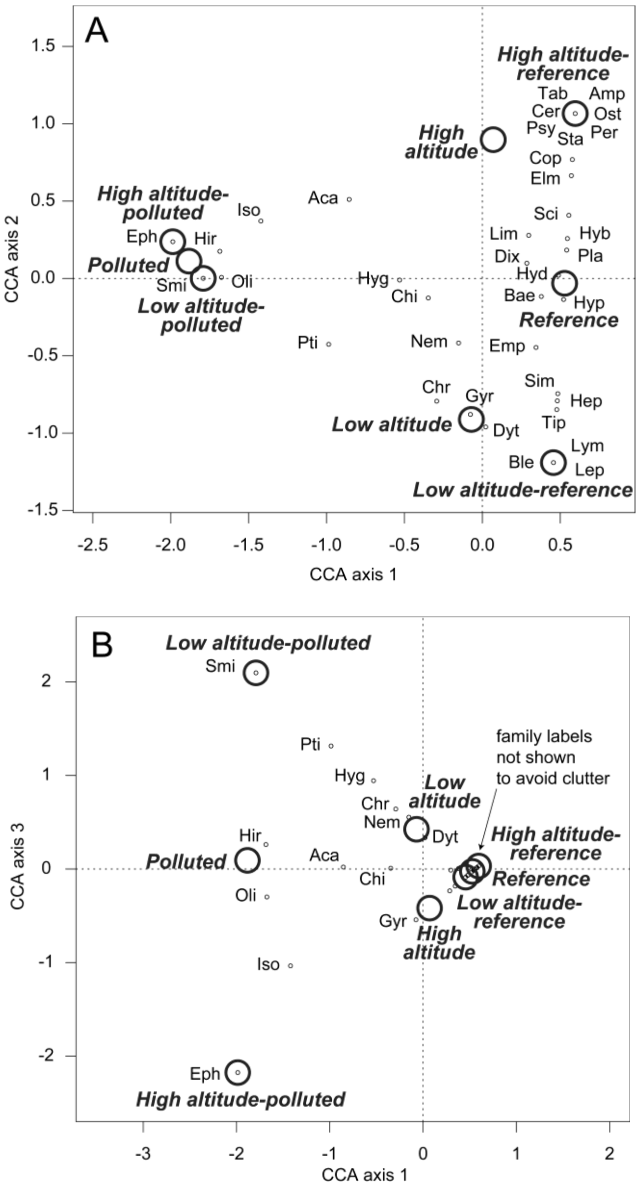

Figure 4. Canonical correspondence analysis (CCA) ordination diagrams of faunal assemblages (A: axes 1 and 2; B: axes 1 and 3) showing families (small circles) and the centroid scores of the altitude and pollution factors and their interaction (large circles), on the basis of 32 samples from eight sites (Figure 1). The eigenvalues were 0.45, 0.29, and 0.07 for axis 1, axis 2 and axis 3, respectively. All axes were significant ($p < 0.005$ for axes 1 and 2, and $p = 0.025$ for axis 3). Family codes appear in Supplementary data Table 3.

persisted at high altitudes.

The third axis of the canonical ordination (Figure 4B) allowed separating the interactive effect of altitude and metals from the single effect of metals on benthic community composition. Ephydridae and Isotomidae occurred mostly at polluted sites at high altitude, whereas Sminthuridae and Ptilidae were mostly found in polluted streams at low altitudes.

Discussion

High altitude Andean streams encompass a suite of unique environmental conditions that potentially challenge the survival of aquatic biota. Indeed, the canonical ordination suggested that metals and altitude had a strong influence on benthic macroinvertebrate community composition in Andean streams.

The results of the physical chemical analyses revealed naturally occurring low pH levels and high metal concentrations at the polluted sites, which may be explained by the dominance of the Chicama formation at the upper sections containing metamorphic sedimentary rocks characterized by pyrite. Pyrite (Fe_2S) oxidation is the dominant reaction in the proglacial zone, generating protons and lowering the pH below 4. As a result, igneous rocks are readily weathered, resulting in high metal solute levels in the water (Burns, 2010). This may explain the high aluminum and iron concentrations in the metal-rich streams, which are present as plagioclase and biotite minerals forming the granodiorite batholith in this area. Also the highly correlated nickel, cobalt, strontium and zinc contents from the Cordillera Blanca batholiths (Rivera et al., 2008) are likely mobilized by increased acidity.

In the streams, the low pH of the leachates increased the bioavailability of the metal ions, enhancing the detrimental effects on aquatic organisms (Courtney and Clements, 2000). Also, the presence of stable orange precipitates and encrusted layers comprising iron oxyhydroxides smothered the streambed, impoverishing food and substrate quality, restricting available habitats for benthic fauna and modifying interactions between functional feeding groups (Courtney and Clements, 2000; O'Halloran et al., 2008). Metal-contaminated substrates may have negatively influenced the composition of benthic communities, producing chronic toxicity and inhibiting colonization by sensitive taxa. A similar effect has been described for several mayfly taxa preferring clean, highgradient streams with coarse substrata and a low degree of sedimentation (Courtney and Clements, 2002). The structuring role of metals was similar to that seen in other scenarios where increased metal levels and decreasing pH coincided with decreased numbers of individuals and families (Gerhardt et al., 1993; Löhr et al., 2006). Since metal concentrations are usually highly correlated in polluted environments, effects on community composition were likely caused by all metals jointly. This was indicated by the high CCU values at the polluted sites (ranging from 11.32 to 94.81), largely exceeding the cutoff of 10.0, which represents metal mixtures causing mortality and altering community structure (Clements et al., 2000). The high CCU values were reflected by much lower numbers of individuals and taxa, and by a strong shift in family composition between reference and polluted sites. The canonical ordination identified crustaceans, ephemeropterans, trichopterans, dipterans and coleopterans as metal- and acid-sensitive groups mainly present at reference sites, and more tolerant dipterans and collembolans associated with polluted sites. This was conform our expectation, since the sensitivity of EPT taxa towards acid pH and metals has been well documented in field surveys in acid mine areas (Gerhardt et al., 2004; van Damme et al., 2008; Loayza-Muro et al., 2010) and experimental microcosms (O'Halloran et al., 2008). Although it is difficult to sort out the effect of individual metals or acid pH on the observed assemblage responses due to their strong correlation, our results suggested that the leachate of igneous rocks produced substantial shifts in community composition towards more tolerant taxa, particularly during the dry season. The differences in benthic community composition observed particularly between reference and metal-impacted streams might further be attributed to the interaction between DOM, UV-B and heavy metals, since organic matter simultaneously reduces metal bioavailability and penetration of UV-B in the water column (Kelly et al., 2001; Clements et al., 2008; Kashian et al., 2004). However, under the elevated sunlight conditions in the high Andes, it is likely that the DOM became photochemically unstable, significantly lowering metal complexation and the mitigation of metal toxicity (Brooks et al., 2007). Solar radiation also causes photobleaching of DOM degrading the chromophores that absorb light, and losing as much as half of its UVR absorption capacity (Zepp et al., 2007). Hence, the low levels of DOC in high altitude metal-impacted streams and its eventual photodegradation may have well left benthic communities more exposed to both metals and UV-B radiation.

The maximum UV-B levels registered in this study at the highest altitude sites (4.89 W/m) were most likely due to a naturally thinner ozone layer over low latitudes and the more direct path of solar radiation through the atmosphere near the equator (Kinzie et al., 1998). These values stand out exceeding those registered in temperate and high latitude alpine areas causing significant impairment of aquatic invertebrates in artificial streams [1.7 W/m^2 (Kiffney et al., 1997a); 2.7 W/m^2 (McNamara and Hill, 1999)], and those observed to structure natural invertebrate communities in experimental field studies [0.1 W/m^2 (Cabrera et al., 1997); 0.17 W/m^2 (Vinebrooke and Leavitt, 1999); 0.5 W/m^2 (Kiffney et al., 1997b); 1.6 W/m^2 (Kelly et al., 2003)]. Since these values are among the highest irradiances reaching the Earth's crust, they may be well considered an important driver of the altitudinal responses of benthic assemblages observed in the present study. Intense radiation may inhibit sensitive taxa either directly or indirectly by altering food resources (Kiffney et al., 1997b; Kelly et al., 2001). Simuliidae, for example, were abundant during the dry season only at low altitude sites, which agrees with previous studies describing a strong drift response and emigration of blackflies due to high UV exposure (Donahue and Schindler, 1998; Kelly et al., 2001). On the other hand, the presence of crustaceans in the reference high altitude streams may be related to the accumulation of photoprotective pigments, such as carotenoids and mycosporine-like amino acids (Rautio et al., 2009). These substances are effective solar radiation screeners providing protection against the harmful effects of UV-B radiation, especially in shallow UV-transparent water bodies as those found in the Andes at 4000 m a.s.l.

Persistence of species under high solar radiation may also be based on avoiding the high UV-B levels, related to preferences for habitats providing considerable shading, such as sediment dwelling in the case of midges or opaque case-protection for caddisflies. Similarly, aquatic vegetation may provide physical refuge from elevated UV-B radiation (Vinebrooke and Leavitt, 1999). Indeed, ephemeropterans, coleopterans and amphipods were abundant at the high altitude reference site in Quilcayhuanca, where macrophyte cover provided food and shading, and low stream discharge and flow provided also habitat stability allowing an important number of individuals and taxa. Although macrophytes may have obscured part of the blazing UV at this high site, the response of specific taxa suggests that high UV-B conditions may be considered a relevant factor producing changes in assemblage composition, particularly during the dry season coinciding with low water flow, shallow-depths and low DOC content in the water.

The altitudinal response of the benthic communities might also be partially due to stream velocities. The high altitude streams, where water flow and discharge levels were relatively low, were dominated by Amphipoda, Ostracoda, Copepoda (Crustacea), Perlidae (Plecoptera) and Elmidae (Coleoptera). Similar responses of these taxa have been recorded in other invertebrate communities (Miserendino and Pizzolón, 2003; Scheibler and Debandi, 2008).

The CCA analysis suggested that the altitudinal effect on family composition at polluted sites differed from that at reference sites. Ephydridae and Isotomidae, the taxa surviving only under harsh conditions of high altitude and high pollution levels, have been described to occur in inhospitable environments, such as alkaline or saline lakes, hot springs, crude oil pools and mud flats exposed to ultraviolet radiation (Foote, 1995; Wagner et al., 2008), and metal polluted soils (Crouau and Pinelli, 2008), respectively. They were also present at high-altitude and metal-polluted streams elsewhere in the Peruvian Andes (Loayza-Muro et al, 2010). The altitudinal effect occurring at polluted sites is probably explained by the differences in streambed composition between altitudes. At high altitudes, mud-shore habitats are typical along the margins of polluted streams and may well represent a rich food supply supporting a large array of Ephydridae (Foote, 1995), while at 3000 m polluted streams show substrates consisting of stones, pebbles and cobbles. In contrast, reference streams showed a more homogenous stony substrate composition at both altitudes.

The aim of the present study was to evaluate the simultaneous effect of metal pollution and altitude on the composition of benthic macroinvertebrate communities in Andean streams. Our results showed that the faunal composition was mostly related to pollution followed by altitude. An interaction of these factors was most evident at polluted sites, with dipterans and collembolans occurring mostly under harsh conditions of high altitude and high pollution levels. Hence it is suggested that in highland Andean streams metal pollution and altitude modulate benthic macroinvertebrate assemblages, reducing their numbers and changing their composition towards specialized taxa.

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