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Arsenic in stream waters is bioaccumulated but neither biomagnified through food webs nor biodispersed to land

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ABSTRACT

Human activities such as mining have contributed substantially to the increase of metals in aquatic environments worldwide. These metals are bioaccumulated by aquatic organisms and can be biomagnified along trophic webs. The dispersal of contaminants from water to land has been little investigated, even though most aquatic invertebrates in streams have aerial stages. We used field and laboratory approaches to investigate the effects of arsenic pollution on stream invertebrate assemblages, and its bioaccumulation, biomagnification and trophic transfer from aquatic to terrestrial environments by emergent insects. We conducted the study in an arsenic-impacted stream (40 μg L^{-1} As at the most polluted site) and a reference stream (0.3 μg L^{-1} As). Invertebrate abundance and richness were lowest at the most impacted site. Arsenic in biofilm and in invertebrates increased with the arsenic content in the water. The highest arsenic accumulators were bryophytes (1760 μg g^{-1}), followed by the biofilm (449 μg g^{-1}) and shredder invertebrates (313 μg g^{-1}); predators had the lowest arsenic concentration. Insects emerging from water and spiders along streambanks sampled from the reference and the impacted stream did not differ in their body arsenic concentrations. In the laboratory, the shredder *Sericostoma vittatum* had reduced feeding rates when exposed to water from the impacted stream in comparison with the reference stream (15.6 *vs.* 19.0 mg leaves mg body mass^{-1} day^{-1}; $p < 0.05$), but they grew faster in the polluted water (0.16 ± 0.04% day^{-1} *vs.* 0.05 ± 0.01% day^{-1}, $p < 0.05$). *S. vittatum* exposed to contaminated stream water accumulated arsenic from food, not through contact with water. We concluded that although arsenic is bioaccumulated, mainly by food ingestion, it is not biomagnified through food webs and is not transported from the aquatic to terrestrial environment when insects leave the stream water.

1. Introduction

As a consequence of human activities, many natural and artificial compounds are being released into the environment. Many such compounds enter the food webs and profoundly harm the environment and humans. Some pollutants are accumulated by living organisms, particularly by primary producers (Ancion et al., 2010; Varun et al., 2012; Vithanage et al., 2011). When the concentration of pollutants is higher in the organisms than in their environment, there is a possibility of biomagnification through food webs, particularly affecting higher trophic levels.

If highly mobile organisms bioaccumulate pollutants, they can disperse these pollutants away from the source of contamination. Most invertebrates in low-order streams are insects with an aerial stage, and therefore capable of transporting pollutants to land (Mogren et al., 2013). We investigated the potential movement of a pollutant from water to land by insects, using arsenic as the model. Metals and metalloids are common in nature, but can be toxic even at low concentrations (Gall et al., 2015). Biomagnification of Cd, Zn, Hg and Se was observed in freshwaters, but the mobility of other elements through food webs remains unclear (Cui et al., 2011; Ikemoto et al., 2008; Juncos et al., 2016; Kehrig et al., 2013; Painter et al., 2016).

Arsenic is a toxic trace element that occurs naturally in the environment. Arsenic in water may originate from rocks by weathering (Alonso et al., 2014; Wang and Mulligan, 2006) and volcanic activity (Juncos et al., 2016). Human activities, including industry, municipal wastes and particularly mining, are important sources of arsenic to the environment (Liu et al., 2010; Subhani et al., 2015). Fungicides, herbicides, insecticides and phosphate fertilizers are also potential sources of arsenic contamination (Ghaeni et al., 2015; Wang and Mulligan, 2006; Wang et al., 2015). All organisms can take up arsenic (as arsenite) because it is chemically similar to phosphate (Wang and

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Mulligan, 2006). Arsenic is toxic to humans and other animals, causing neurological disorders, liver and kidney diseases, cancer, gastrointestinal disturbances and other health problems (Subhani et al., 2015).

Aquatic primary producers such as phytoplankton, periphyton, bryophytes, macrophytes (Culioli et al., 2009) and macroinvertebrates accumulate arsenic from water and sediment (Awrahman et al., 2015; Magalhães et al., 2015; Rahman et al., 2012), causing reduced survival (Canivet et al., 2001; Gaete and Chávez, 2008), growth (Marsden and Rainbow, 2004) and reproduction (Mogren et al., 2012). In terms of community structure, high concentrations of trace elements, including arsenic, reduce the abundance and richness of macroinvertebrates (Doi et al., 2007; Mayer-Pinto et al., 2010; Qu et al., 2010; Rhea et al., 2006) and potentially impair their functional role in the ecosystem. Understanding the arsenic pathway in biological systems is therefore important for environmental management and restoration.

We used field and laboratory approaches to investigate (1) at community level, the effects of arsenic pollution on macroinvertebrate density, richness, and community composition; (2) routes of arsenic incorporation into consumer bodies; and (3) arsenic mobility in food webs, using the stable-isotope approach, and its transfer to the terrestrial environment. We hypothesized that (1) because of its toxicity, arsenic will cause decreases in invertebrate density and richness; (2) at sub-lethal levels, consumers will incorporate arsenic both from contaminated food and directly from water contact; and (3) because of contaminated food ingestion, arsenic concentration will increase through food webs, and eventually arsenic will be transferred to the terrestrial environment with the emergence of aquatic-insect consumers.

2. Material and methods

2.1. Study area

We sampled two streams; a stream receiving water from an abandoned mine (Escádia Grande, Portugal; 'impacted stream'; 4 sites, A to D; 40°04′02″N, 008°06′22″W – to 40°02′11″N, 008°05′45″W) and a 'reference stream' located in the same area (site E), but not affected by arsenic (40°03′56″N, 008°06′17″W). The streams were similar in width, depth, flow, substrate and water chemistry, except for high electrical conductivity and arsenic concentration in the impacted stream and high phosphate in the reference stream (Table 1).

2.2. Arsenic effects on invertebrate assemblages in stream

We evaluated the effects of arsenic pollution (mean ~40 μg L^{-1}; 32 – 43 μg L^{-1} range of 8 measurements during four seasons in the previous year; site A) on aquatic invertebrates in terms of density, rarefied richness and community composition (Fig. 1). We collected 10 random Surber samples (0.1 m^2; 500 μm) at each stream site. Live samples were transported to the laboratory in water inside a cooler box, sorted alive on the same day, and invertebrates identified to genus according to Tachet et al. (1987), except dipterans which were identified to family level.

Table 1
Chemical and physical variables of the arsenic-impacted and reference streams (mean ± SE, n = 5; *t*-test and probabilistic p-value). Lousã Mountain, Portugal (March-April 2015). NS: non-significant.

Limnological variables	Reference	Impacted	*t*	p
Water temperature (°C)	12 ± 0	12 ± 1	0.1	NS
Conductivity (μS cm^{-1})	29 ± 0	36 ± 1	11.6	< 0.001
pH	6.7 ± 0.0	6.7 ± 0.0	0.1	NS
Dissolved oxygen (mg L^{-1})	11 ± 0	10 ± 0	1.0	NS
O_2 saturation (%)	115 ± 3	110 ± 3	1.6	NS
Nitrate (μg L^{-1})	321 ± 188	366 ± 16	0.2	NS
Phosphate (μg L^{-1})	53 ± 4	22 ± 2	7.9	< 0.001
Sulfate (μg L^{-1})	19 ± 4	31 ± 21	0.6	NS
Arsenic (μg L^{-1})	0.3 ± 0.2	39.2 ± 11.3	10.8	< 0.001

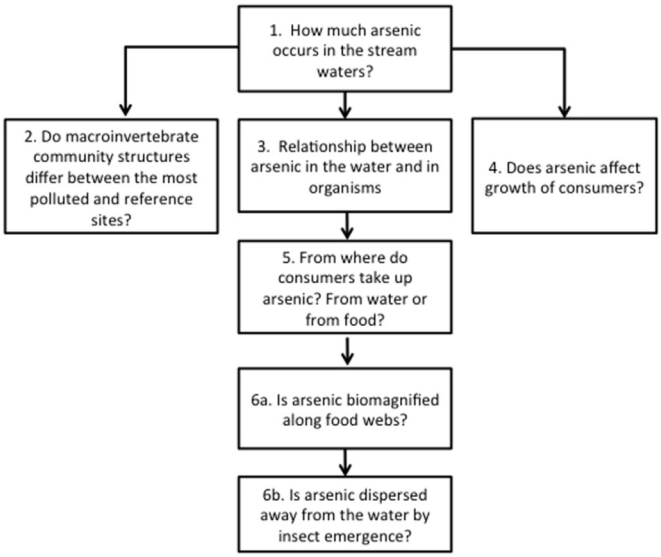

Fig. 1. Flow-chart of the experimental approach.

2.3. Relationship between concentrations of arsenic in stream water and biota

We collected water samples, biofilm and two invertebrate taxa, a scraper (*Heptagenia* sp.) and a collector (*Hydropsyche* sp.), in four stretches along the impacted stream (sites A to D; 5.3 km apart) to investigate the relationship between arsenic concentrations in the water and in organism bodies (Fig. 1). We used these two trophic groups to understand the incorporation of arsenic into food chains from biofilm and fine particulate organic matter. We did not use shredders because their density at the stream sites was low and not enough specimens were available to make similar measurements. Along the four stretches of the polluted stream, arsenic concentrations in water decreased from 40 to 2.5 μg L^{-1}; at the reference site, arsenic concentrations were 0.3 μg L^{-1}.

Biofilm was collected by delimiting a 72 cm^2 area on submerged stones (using a flask cap) and scraping the area with a toothbrush into a 30 mL plastic flask. The samples were taken to the laboratory in a cooler box and the content was passed through a pre-weighed filter (Whatman 0.45 μm GFC), dried (60 °C/48 h) and weighed. The filters were then digested in a nitric-acid and hydrogen-peroxide solution (4:1) to determine the arsenic (see below). Arsenic concentration was determined as described in 2.7.

2.4. Arsenic effect on growth and survival of a selected consumer: Sericostoma vittatum

We used the shredder *Sericostoma vittatum* Rambur, 1842 (Sericostomatidae, Trichoptera) as a model organism to assess the effects of arsenic concentration on growth in a laboratory experiment (Fig. 1). *S. vittatum* is a common North Iberian endemic species, easily maintained and previously used in laboratory experimental studies (Feio and Graça, 2000; Graça et al., 2001). We sampled larvae from a nearby stream (Candal; 40°04′44″N, 008°12′10″W). We measured the capsule width under a binocular microscope to obtain dry mass values using an equation obtained from a set of 24 specimens which were measured to the nearest 0.1 mm, oven dried (60 °C for 72 h) and weighed to the nearest 0.01 g: mass (mg) = (0.0017* size in mm) + 0.0589 (R^2 = 0.80, n = 24).

Specimens were placed individually in 250 mL cups with 100 mL of