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Research Paper

Bioaccumulation trends of arsenic and antimony in a freshwater ecosystem affected by mine drainage

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Environmental context. The food web behaviours of As and Sb are poorly understood. We compare As and Sb bioaccumulation in a contaminated freshwater ecosystem. Metalloid accumulation decreased with increasing trophic level. Bioprecipitated minerals in microbial mats represent a direct route of uptake (by ingestion) of metalloids to tadpoles, which contained the highest concentrations ever reported. We demonstrate food web bioaccumulation, but not biomagification, of As and Sb. We also report an unexpectedly high tolerance of tadpoles to metalloid toxicity.

Abstract. We compared As and Sb bioaccumulation and biomagnification when these metalloids co-occurred at varying environmental concentrations in a stream and wetlands near a contaminated mine site in Idaho (USA). We measured As and Sb concentrations in water and substrate samples, and in tissues of organisms representing several trophic levels. Bioaccumulation of both As and Sb was observed in stream organisms with the following trend of bio-diminution with increasing trophic level: primary producers > tadpoles > macroinvertebrates > trout. We also note reductions in metalloid concentrations in one of two stream remediation reaches engineered within the past 17 years to ameliorate metalloid contamination in the stream. Several wetlands contained thick microbial mats and were highly populated with boreal toad tadpoles that fed on them. The mats were extremely contaminated (up to 76 564 mg kg^{-1} As and 675 mg kg^{-1} Sb) with amorphous As- and Sb-bearing minerals that we interpret as biogenic precipitates from geomicrobiological As- and Sb-cycling. Ingested mat material provided a direct source of metalloids to tadpoles, and concentrations of 3867 mg kg^{-1} (As) and 375 mg kg^{-1} (Sb) reported here represent the highest whole body As and Sb levels ever reported in living tadpoles. The bulk of tadpole metalloid burden remained in the gut despite attempts to purge the tadpoles prior to analysis. This study adds to a number of recent investigations reporting bioaccumulation, but not biomagnification, of As and Sb in food webs. Moreover, our results suggest that tadpoles, in particular, may be more resistant to metalloid contamination than previously assumed.

Additional keyword: tadpoles.

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Introduction

Mining and smelting are major sources of trace metal contamination in freshwater systems. The toxic metalloids arsenic and antimony, either individually or in combination, have caused adverse environmental effects in the vicinity of contaminated mines around the world.[1–8] Arsenic is a ubiquitous poison and environmental contaminant and Sb, a toxic element of emerging environmental concern, is increasingly mined for a variety of industrial applications.[9] Both elements are classified as pollutants of priority interest by the U.S. Environmental Protection Agency (EPA), which sets the maximum contaminant level (MCL) for As and Sb in drinking water at <10 and <6 μg L^{-1} respectively.[10] The two metalloids occur in the same group of the periodic table and they exhibit similar, but not necessarily identical, geochemical and toxicological properties that vary with chemical form and oxidation state. In natural waters, these metalloids are present primarily as the pentavalent oxyanions arsenate (As^V) and antimonate (Sb^V) in oxygenated settings, or as trivalent arsenite (As^{III}) and antimonite (Sb^{III}) under anoxic conditions.[9,11] Arsenic and Sb are chalcophilic elements that frequently co-occur in sulfidic mineral phases, such as arsenopyrite (FeAsS) or stibnite (Sb_2S_3), associated with hydrothermal ores.[9,12] The oxidative weathering and dissolution of As- or Sb-bearing sulfide minerals in subaerially exposed mine waste is a common point source for contamination of aquatic ecosystems.

The behaviour of As and Sb in aquatic ecosystems is complex, with the former element being better studied than the latter. Both metalloids can bioaccumulate in freshwater food chains but they are not known to bio-magnify, and in some cases they are reported to undergo bio-diminution with increasing trophic level.[5,6,8,13–15] The primary routes of As and Sb uptake into the food chain are through direct contact of organisms with

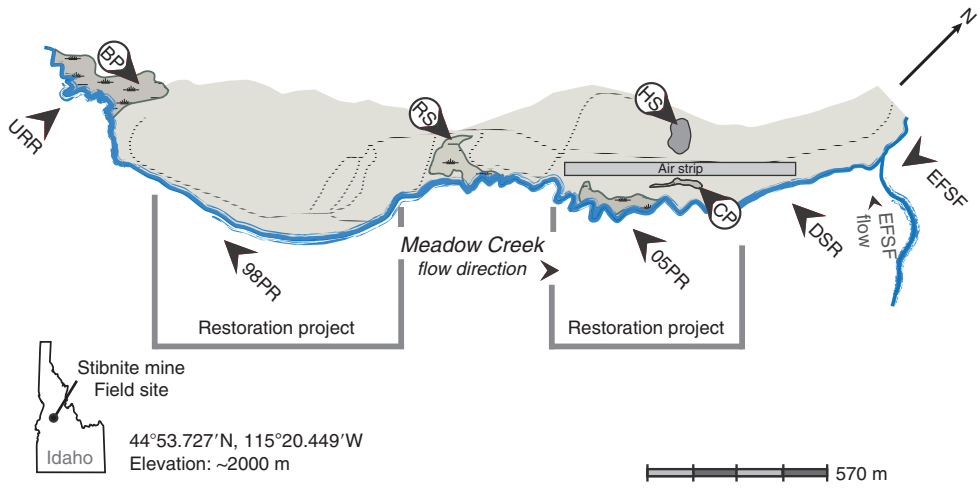

Fig. 1. Map of the Stibnite Mine field site showing the extent of mine tailings (shaded area), restoration projects and wetlands. Dotted lines are service roads. Sampled reaches in this study are labelled with site name abbreviations (see *Field Site* in the *Experimental* section). Sites GH, OP and MPW are not within map range, and are located ~1 km northeast of the tailings.

contaminated water and sediment, or through the consumption of contaminated autotrophs (e.g. biofilm, algae and macrophytes) by invertebrates.[5,6,16] Conflicting reports exist regarding the comparative bioaccumulation of As and Sb. Fu et al.[8] reported higher bioaccumulation of As than Sb from water to algae, water to fish, and terrestrial soil to earthworms around an active Sb mine. Telford[5] also reported greater uptake of As than Sb in aquatic plants in the vicinity of a gold and Sb mine. However, consistently higher bioaccumulation of Sb (compared to As) has been observed in bryophytes, benthic macroinvertebrates, and fish in a river ecosystem affected by a historic realgar mine.[6] In general, there have been few and conflicting studies documenting the environmental behaviour of Sb, or the bioconcentration of As and Sb when both metalloids are present.

The primary objective of this study is to compare As and Sb bioaccumulation and biomagification in aquatic organisms from several trophic levels when these metalloids co-occur at varying environmental concentrations. Our study site consisted of a freshwater creek (Meadow Creek) and wetlands near the As- and Sb-contaminated Stibnite Mine in central Idaho (Fig. 1). We assessed As and Sb concentrations and redox speciation in surface water, and concentrations in sediment and stream or wetland biota including: riparian tree leaves, biofilm, algae, submergent macrophytes, benthic macroinvertebrates, frog and toad tadpoles and predatory fish (trout). We sought to determine if As and Sb bioaccumulate or biomagnify in stream or wetland food chains, and to draw inferences with respect to pathways of biological As and Sb uptake in the ecosystem. We note particularly elevated metalloid concentrations in frog and toad tadpoles resulting from the ingestion of microbial mats that contain abundant microbiologically precipitated As- and Sb-bearing minerals. In 1998 and 2005 stretches of Meadow Creek adjacent to the mine tailings were engineered in an attempt to ameliorate toxic metalloid concentrations. Therefore, as a secondary objective, we compare our water, sediment and biota concentrations of As and Sb with values reported pre-remediation, to evaluate the effectiveness of these different restoration strategies in reducing the metalloid burden in the stream ecosystem.

Experimental

Field site

The Stibnite Mine (also referred to as the Yellow Pine Mine) is located in Valley County, Idaho within the Payette National Forest (44°53.727′N, 115°20.449′W; Fig. 1). The mine operated intermittently from 1931 to 1995 to extract gold, antimony and tungsten ore. Arsenic, Sb, cyanide (CN^-), and other toxic metals leached from mine tailings into nearby Meadow Creek where it flows through 3.2 km of tailings that were piled in a 0.64-km^2 area near the mine. Between the 1980s and 2000, concentrations of As and CN^- in the creek consistently exceeded the EPA aquatic chronic level and often exceeded the aquatic acute level for stream dwelling organisms.[17] From 1995 to 1999, elevated levels of As, Sb, Hg and CN^- were documented in Meadow Creek water, sediment, fish and nearby wetlands (Table 1).[18] In 2001, the EPA proposed that the mine area be added to the National Priorities List as a potential 'Superfund' site. The South Fork of the Salmon River and its tributaries, including Meadow Creek, are designated critical habitat for three species listed under the *US Endangered Species Act*: Snake River chinook salmon (*Oncorhynchus tshawytscha*), Snake River steelhead (*Oncorhynchus mykiss*) and Columbia River bull trout (*Salvelinus confluentus*).

In 1998, the USDA Forest Service (USFS), EPA and responsible mining companies engineered a stretch of Meadow Creek adjacent to the tailings in an attempt to ameliorate toxic metalloid concentrations in the stream. They excavated a 1.4-km-long, straight flowing, low gradient stream channel in Meadow Creek to a depth below the mine tailings, and installed a sand filter between the tailings and the stream. The streamside was then armoured with rock substrate. A second project, completed in 2005, diverted the stream channel around the tailings in a 1.6-km-long reach downstream from the 1998 project. The 2005 reach was constructed with greater attention to natural substrates, meanders and gradients, and included a riparian zone between the stream and mine tailings that was constructed with transplanted soil and tree seedlings. We sampled biotic and abiotic components of five reaches of Meadow Creek near the tailings (Fig. 1): (1) an upstream reference reach (URR) that was not affected by the tailings,

Table 1. Aqueous As and Sb concentration (μg L^{-1}) ± standard deviation and pH levels in surface water from Meadow Creek and wetland sites

n is the number of samples analysed. En-dashes represent no measurement taken. Meadow Creek Sites: an upstream reference reach (URR) that was not affected by the tailings, the 1998 restoration project reach (98PR), the 2005 restoration project reach (05PR), a non-remediated reach downstream of the mine tailings (DSR) and a tributary stream, the East Fork of the South Fork of the Salmon River (EFSF)

Site	Date	*n*	pH	As	Sb
Meadow Creek flow direction ↓					
URR	2011	3	6.8	<2[A]	3.1 ± 0.03
	2012	2	8.3	<2[A]	4.0 ± 2.8
98PR	2010	1	8.3	<2[A]	<1.4[A]
	2011	4	7.8	7.9 ± 2.0	3.8±0.1
	2012	2	8.6	<2[A]	<1.4[A]
05PR	2010	1	9.3	40.4	4.9
	2011	3	7	35.6 ± 0.4	7.9 ± 0.7
	2012	2	8.6	27.9 ± 3.3	6.7 ± 1.7
DSR	2010	1	9.3	41.6	6
	2011	3	6.8	36.4 ± 1.3	9.9 ± 0.4
	2012	2	8.4	28.1 ± 4.2	6.1 ± 0.5
EFSF	2011	3	–	9.3 ± 0.1	3.9 ± 0.4
	2012	1	–	10.3	9.6
Wetland					
Channel pond	2010	1	8.7	147.1	324.5
	2011	3	7.8	138.4 ± 1.0	453.8 ± 16.4
	2012	2	8.2	168.2 ± 10.5	206.8 ± 149.4
Heap Seep	2010	2	10.4	27 373.3 ± 1639.3	1127.9 ± 35.5
	2011	4	8.6	18 788.9 ± 169.2	239.1 ± 5.3
	2012	1	9.4	10 870	1042
Glory Hole	2010	1	8.1	64.9	21.2
	2011	3	6.4	53.7 ± 2.8	21.0 ± 1.3
	2012	2	8.1	70.6 ± 9.0	20.7 ± 2.0
Mine Pit wetland	2010	2	7.7	191.3 ± 7.2	115.5 ± 3.9
	2011	3	6.3	23.8 ± 1.7	68.6 ± 2.4
	2012	2	7.9	104.4 ± 10.4	164.4 ± 23.7
Office Pond	2012	2	7.4	395.7 ± 94.2	28.0 ± 6.5
Red Seep	2012	1	8	1243.1	1034.7
Boulder Pond	2012	2	8.3	21.5 ± 6.8	11.9 ± 0.9

[A]Below current instrument detection limit (As, <2 μg L^{-1}; Sb, <1.4 μg L^{-1}).

(2) the 1998 restoration project reach (98PR), (3) the 2005 restoration project reach (05PR), (4) a non-remediated reach downstream of the mine tailings (DSR) and (5) a tributary stream, the East Fork of the South Fork of the Salmon River (EFSF).

Low lying areas around the tailings contain ephemeral and permanent ponds, as well as groundwater springs that emerge along the edge of the tailing's piles. We sampled seven of these wetlands (Fig. 1) representing a range of different habitats and communities. We assigned descriptive designations to these locations as follows: (1) Channel Pond (CP), a small, permanent pond between Meadow Creek and the tailings, located in the pre-restoration stream channel of the 05PR reach, (2) Heap Seep (HS), a seasonally flooded, shallow (5–20 cm deep) impoundment that receives direct drainage from the largest tailings heap, (3) the flooded mine pit known locally as the Glory Hole (GH), through which Meadow Creek flows before leaving the mine area, (4) the Mine Pit Wetland (MPW), a marsh immediately downstream of the mine pit, (5) Office Pond (OP) a small permanent pond near the location of the mining company office but outside of the immediate tailings area, (6) Red Seep (RS), a small spring that emerges from the edge of the tailings and is coloured bright red from precipitated realgar mineral (As_2S_3) and (7) Boulder Pond (BP), a larger pond (1100 m^2) located at the upstream end of the mine tailings piles.

Sample collection

Samples were collected during the summers of 2010 through 2012, except for EFSF, OP, RS and BP samples which were collected in 2012 only. Surface water samples from each location were collected for As and Sb concentration and speciation analysis by filtering through a 0.45-μm nylon syringe filter (Fisher, Co., Hampton, NH, USA) into 5-mL evacuated tubes (Vacutainer, BD Company, Franklin Lakes, NJ, USA) to prevent contact with air during storage.[19] Water samples for As and Sb speciation were preserved by adding 50 μL of 0.125 Methylenediaminetetraacetic acid (EDTA).[20] Water samples were collected approximately 15 cm below the air–surface interface. Sediment samples (0–15 cm below the sediment–water interface) were collected using a garden trowel and sealed in completely filled glass jars. Water and sediment samples were stored on ice in the dark for transportation to the laboratory, where they were stored at 5 °C and analysed within one week of collection. Biological samples included the following primary producers: riparian tree leaves (alder, *Alnus* sp.; and willow, *Salix* sp., the main source of allochthonous primary production in the stream), submergent macrophytes (mosses and algal streamers) and biofilm (a mixture of attached microalgae, bacteria and fine inorganic detrital material referred to as periphyton). Tree leaves and macrophytes were collected in sealable

plastic bags. Biofilm was collected by scraping a 3.45-cm^2 area of rock and collecting scraped material and rinse water onto Whatman 0.45-µm glass fiber filter paper (GE Healthcare Bio-Sciences, Pittsburgh, PA). Benthic macroinvertebrates were dislodged from substrates, collected in a Surber sampler (Morris and Lee, Inc., Yulee, FL, USA), and Tricoptera (*Arctopsyche* sp.) and Ephemeroptera (*Drunella* sp.) larvae (primarily filterer and shredder–gatherer functional feeding groups respectively) were placed in sealable plastic bags. Predatory stream-dwelling fish (rainbow trout, *Oncorhynchus mykiss*), stream-dwelling Rocky Mountain tailed frog tadpoles (*Ascaphus montanus*; primarily an algavore), pond-dwelling toad tadpoles (Boreal toad, *Anaxyrus boreas*) and Columbia spotted frog tadpoles (*Rana luteiventris*) were collected using nets. All biological samples were transported on ice to the laboratory and then frozen until analysis. Live tadpoles collected in 2012 were held in source water (2 L) for 24 h in the laboratory after collection, in an attempt to purge tadpole gut content prior to freezing and subsequent analysis.[21]

Sample preparation and analysis

All acids and reagents used were trace metal grade. Water samples for total As and Sb determination were acidified (2 %) using nitric acid (HNO_3) prior to measurement by inductively coupled plasma–mass spectrometry (ICP-MS) or inductively coupled plasma–optical emission spectroscopy (ICP-OES). Instruments were calibrated daily with National Institute of Standards and Technology (NIST) traceable standards. A second-source NIST-traceable standard was analysed after every tenth sample to verify accuracy within 10 % standard error. A deionised water blank was also run after every tenth sample to monitor baseline drift. Water samples from the site that were spiked with known concentrations of As and Sb were used to verify that there was no interference to the detection of these elements from the sample matrix. Surface water samples for determination of As and Sb redox speciation were measured by high performance liquid chromatography with inductively coupled plasma–mass spectroscopy (HPLC-ICP-MS).[7,20] Sediment samples (~10 g wet weight) for the determination of total As and Sb concentration were oven dried (65 °C for 72 h) and ground using a mortar and pestle. Dried and ground sediment (0.5 g) was digested using microwave assisted digestion with a 3 : 1 (v/v) mixture of concentrated HNO_3 and HCl according to US EPA Method 3052.[22] Digestions were filtered and diluted prior to As and Sb analysis by ICP-OES.

Biological samples were thawed and rinsed using deionised water to remove residual sediment and source water. Rinse water was dried from the exterior of specimens using lint-free tissue paper and initial (wet) sample weight was recorded. For riparian plants, whole leaves were analysed. For submergent macrophytes we analysed roots and shoots together. Biofilm samples were processed and digested on the filters used for sample collection. For macroinvertebrates, tadpoles and fish, whole body samples were analysed. To examine tadpole gut metalloid concentrations, samples from CP, HS and OP ponds were thawed, rinsed and dissected under a stereoscope for the removal, digestion and analysis of the intestinal tract (extending from bottom of the esophagus to the anus). Biological samples were freeze-dried using a *Labconco FreeZone* (Labconco Co., Kansas City, MO, USA) 4.5-L freeze drying system for 48 h and reweighed to obtain the sample dry weight. Samples were acid digested by refluxing with a mixture of concentrated HNO_3 and HCl on a hotplate at 50 °C for up to five days until samples were completely dissolved.[23] The acid was then evaporated at 70 °C, the residue was reconstituted in 10 mL of 2 % HNO_3 and filtered (0.45 µm) prior to As and Sb analysis. Elemental values were normalised to sample dry weight. The biological concentration factor (BCF) was calculated for all biological samples as the average metalloid concentration in the organism divided by the average concentration in the water.[6]

Identification of minerals in microbial mats and tadpole intestinal tracts was conducted by electron microprobe (EMP) analysis. Prior to analysis, samples were freeze-dried for 48 h and homogenised using a mortar and pestle. Microprobe samples were mounted on polished (6 µm) graphite rods in a sample–ethanol mixture and carbon coated prior to analysis.

Analytical

Elemental analyses in 2010 were conducted using a Perkin–Elmer ELAN II ICP-MS (Perkin–Elmer, Inc, Waltham, MA, USA). Analyses in 2011 and 2012 utilised a Perkin–Elmer ELAN 6000 ICP-MS or a Varian VISTA-MPX ICP-OES (Agilent Technologies Inc., Santa Clara, CA). Arsenic and Sb speciation were measured in water by HPLC-ICP-MS using a Perkin–Elmer Series 200 HPLC interfaced with a Perkin–Elmer ELAN II ICP-MS with chromatography conditions described in Kulp et al.[19] Amorphous mineral identification was conducted using a JOEL 8900 Superprobe electron microprobe (JEOL Co., Freising, Germany).

Results

Stream results

Surface water and sediment

Average As concentrations in stream water ranged from <2 to 41.6 µg L^{-1}, whereas Sb concentrations occurred in the range of <1.4 to 9.9 µg L^{-1} (Table 1). Average background water concentrations (URR) were below instrument detection limits for As during 2011 and 2012, with corresponding Sb concentrations of 3.1–4.0 µg L^{-1}. Downstream from the reference site, we found progressive increases in As and Sb concentrations (Fig. 2). Both elements were slightly elevated in the 98PR reach in 2011compared to the URR, but remained below the drinking water MCL (<10 and <6 µg L^{-1} respectively) and were below detection limits in other years. However, further downstream in the 05PR and DSR reaches, we found elevated As (27.9–41.6) and Sb concentrations (4.9–9.9 µg L^{-1}). Compared to historic (pre-1999) values, dissolved As and Sb concentrations in the 98PR reach were lower in all three years of our study (Table 1 and Fig. 2a). Further downstream (i.e., 05PR and DSR reaches) there was no significant post-remediation reduction in dissolved As, but Sb concentrations were reduced by as much as 71 % compared to historic data. Measurements of dissolved As and Sb redox speciation in surface water samples collected in 2010 show that the pentavalent ions (As^V and Sb^V) were the dominant valence states in solution, and in most stream reaches they were the only forms of the metalloids present (Table 2). However, in the 05PS and DS reaches, As^{III} accounted for 30–37 % of the total measured aqueous As. Antimonate (Sb^V) was the predominant aqueous Sb species, with Sb^{III} detected only in minor amounts (5.0 %) at 05PS.

Similarly to dissolved As, sedimentary As concentrations increased in a downstream direction through the tailings area (Table 3). Sediment Sb concentrations were not elevated above the range typically reported for uncontaminated sediments.[9]

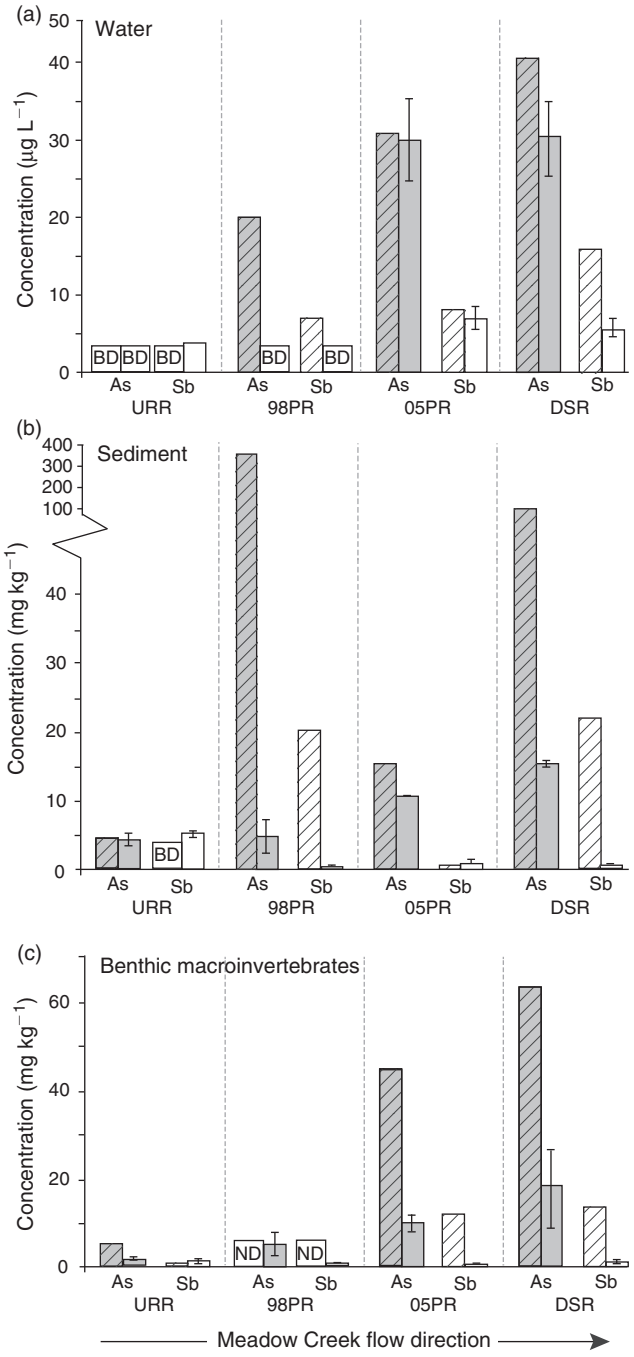

Fig. 2. A comparison of As and Sb concentrations in water (a), sediment (b) and benthic macroinvertebrates (c). Samples collected in 2012 (solid bars) and between 1996 and 1999[18] (patterned bars). BD, below detection; ND, not determined. Error bars represent standard deviation.

Sedimentary As and Sb concentrations were highly variable among all reaches in the pre-remediation data set, possibly reflecting localised inputs of tailings into the creek channel prior to its diversion (Table 3). The 98PR and DSR reaches had notably lower concentrations of sediment-associated As and Sb in 2012 compared to those measured pre-remediation (Fig. 2b). Sediment metalloid concentrations in the 05PR reach were low pre-remediation (15 and 1.0 mg kg^{-1} for As and Sb respectively) and exhibited little change between pre-1999 and 2012 measurements.

Stream primary producers

Arsenic concentrations in submerged macrophytes (i.e. vascular submergent plants, mosses and algae) and biofilm averaged 6.6 ± 0.6 mg kg^{-1} (dry weight, DW) in the URR section and were between 3.1 ± 1.8 and 46.8 ± 11.6 mg kg^{-1} (mean ± standard deviation) in the downstream reaches (Table 4). The Sb concentration of all vascular plants was <4 mg kg^{-1}. Average As concentrations were 19.9 ± 6.2 mg kg^{-1} in biofilm and 5.8 mg kg^{-1} in algal streamers from the URR. Algae and biofilm exhibited a wide range in downstream reaches with the highest concentrations observed in the 05PR reach (Table 4).

Both metalloids were below detection in alder and willow leaves from the URR, but were elevated in leaves collected from downstream reaches, as well as the EFSF tributary stream (Table 4). Concentrations of both As and Sb were less than 6 mg kg^{-1} in all leaves except for willow leaves from the 05PR riparian zone, which contained an average As concentration of 29.3 ± 2.8 mg kg^{-1}. Metalloid concentrations were higher in willow compared to alder leaves, with the exception of Sb in the 98PR reach.

Stream macroinvertebrates

Benthic macroinvertebrates contained As levels <2.3 mg kg^{-1} and Sb levels <1.5 mg kg^{-1} in the URR and the EFSF (Table 5). Macroinvertebrates from sites adjacent to the tailings area (98PR, 05PR and DSR) had elevated levels of both metalloids, with DSR macroinvertebrates exhibiting the greatest concentrations (up to 66.4 mg kg^{-1} for As and 5.3 mg kg^{-1} for Sb). No difference in As or Sb uptake was observed between the two insect genera. In stream reaches where pre-remediation data were available, As and Sb concentrations in benthic macroinvertebrates were reduced compared to the historic data (Fig. 2c). The insect genera sampled in the earlier dataset are not known.

Stream tadpoles and trout

Arsenic concentrations in juvenile trout were not elevated in the downstream reaches of Meadow Creek relative to the URR reference site (Table 5). Antimony was below detection in fish from all reaches.

Tailed frog tadpoles, which primarily graze on periphyton or biofilm, contained much higher metalloid concentrations than macroinvertebrates or trout (Table 5). Tadpoles in the URR had an average whole-body As concentration of 6.7 ± 0.30 mg kg^{-1} and an average Sb concentration of 0.16 ± 0.003 mg kg^{-1} in 2011 samples. Metalloid concentrations in the 2012 tadpoles were all below detection in the URR. The highest metalloid concentrations in tailed frog tadpoles (637.4 ± 173.2) were measured at the 98PR site in 2011. Whole body As concentrations in tadpoles were elevated ($>80.8 \pm 29.5$ mg kg^{-1}) in all reaches adjacent to the mine tailings (98PR, 05PR and DSR) compared to URR concentrations. Antimony concentrations in tadpoles from the downstream reaches measured up to 7.7 ± 3.6 mg kg^{-1} in 2011 but were notably lower in 2012 (Table 5).

Stream bioaccumulation trends

We selected the 05PR site in 2012 to illustrate bioaccumulation trends for As and Sb in the contaminated reaches of Meadow Creek. The concentration of both metalloids generally decreased from biofilm, to tadpoles, to macroinvertebrates, to trout, although tadpoles and macroinvertebrates had similar Sb concentrations (Fig. 3a). The BCF for each organism is

Table 2. Total As and Sb concentration and redox speciation (μg L^{-1} (%)) in surface water from Meadow Creek and wetland sites in 2010 from one analysed sample per site

Values in parenthesis report the percent recovery of total As or Sb represented by each oxidation state. See *Field Site* in the *Experimental* section for site abbreviations. bd, below instrument detection limit (As, <1 μg L^{-1}; Sb, <1 μg L^{-1})

Site	Arsenic (μg L^{-1} (%))			Antimony (μg L^{-1} (%))		
	Total	As^{V}	As^{III}	Total	Sb^{V}	Sb^{III}
Meadow Creek flow direction ↓						
URR	1.84	1.8 (100)	bd	0.45	0.5 (100)	bd
98PR	1.64	1.6 (100)	bd	0.65	0.7 (100)	bd
05PR	30.2	19.1 (63.35)	11.1 (36.7)	7.96	7.6 (95.5)	0.4 (5.0)
DSR	31.3	22.1 (70.5)	9.3 (29.5)	9.55	9.6 (100)	bd
Wetlands						
CP	136.96	128.5 (93.8)	5.1 (3.7)	436.1	435.5 (99.9)	0.57 (0.1)
HS	22 730.78	21 902.2 (96.4)	350.5 (1.5)	1508.89	1503.4 (99.6)	5.54 (0.4)

Table 3. Total sedimentary mean As and Sb concentrations (mg kg^{-1}) ± standard deviation from Meadow Creek and wetland sites in 2012

URR, reference site, upstream; *n* is the number of samples analysed. See *Field Site* in the *Experimental* section for site abbreviations

Site	*n*	Arsenic	Antimony
URR	2	4.4 ± 0.8	0.6 ± 0.2
Meadow Creek			
98PR	2	4.9 ± 2.4	0.4 ± 0.3
05PR	2	11.5 ± 0.004	1.4 ± 0.3
DSR	2	15.6 ± 0.4	0.8 ± 0.01
EFSF	2	19.9 ± 4.4	0.8 ± 0.2
Wetland			
CP	2	292.6 ± 21.6	203.5 ± 42.6
HS	2	1388 ± 192.5	43.1 ± 17.1
OP	2	149.6 ± 41.9	10.4 ± 0.5
BP	1	1860	45.2
GH	1	4728	4.1
MPW	1	15 192	20.5

presented in Fig. 3b. Bioaccumulation of As is 10–100 times greater than bioaccumulation of Sb in biofilm, tadpoles and macroinvertebrates. Antimony was not detectable in trout from this stream.

Wetland results

Surface water and sediment

Several wetlands in this study were characterised by aqueous As and Sb concentrations up to three orders of magnitude greater than in Meadow Creek (Table 1). The HS site accounts for the highest of these concentrations, which ranged from 10 870 to 27 373 μg L^{-1} for As and 239.1 to 1127.9 μg L^{-1} for Sb over the three study years. Aqueous metalloid concentrations were also highly elevated at the RS and OP sites (Table 1). The CP location was unique among all sites in that it had dissolved Sb concentrations that were consistently higher than As during all three years. As^{V} and Sb^{V} were the predominant aqueous redox states of the two metalloids in the wetland surface waters, accounting for >96.3 % of dissolved As and >99 % of dissolved Sb, with the remaining balance present as As^{III} or Sb^{III} (Table 2).

Sedimentary As and Sb concentrations in wetlands were two to three orders of magnitude higher than in stream sediments (Table 3). The greatest sedimentary concentrations for As occurred at MPW, whereas the highest sedimentary Sb concentrations were observed at CP. Consistent with the dissolved aqueous concentrations reported above, the CP location was unique in having more Sb than As in sediments.

Wetland primary producers

Due to differences in sample availability, submergent macrophytes were analysed from the CP and OP ponds, whereas photosynthetic microbial mats were analysed from the CP, RS and HS sites. The results are reported in Table 6. Pond plants at CP averaged 171.6 ±11.1 mg kg^{-1} As and 24.9 ± 4.9 mg kg^{-1} total Sb (DW), whereas As and Sb concentrations in CP algae were higher at 1735 ± 129 and 47.3 ± 1.7 mg kg^{-1} respectively. Two plants collected from OP showed a high variability in As concentration (608.4 and 1244 mg kg^{-1}) but contained similar total Sb concentrations (73.0 and 83.7 mg kg^{-1}). The highest total As and Sb concentrations measured in this study occurred in microbial mats at the RS and HS locations (Table 6). The RS mat contained 76 564 mg kg^{-1} total As and 674.9 mg kg^{-1} total Sb, whereas the HS mat concentrations were 3366 ± 200 mg kg^{-1} total As and 30.2 ± 5.9 mg kg^{-1} total Sb. At the RS site, red, amorphous realgar (As_4S_4) and other As- and Sb-sulfide mineral phases are actively precipitating in microbial mats and sediments that surround the spring. Microbial mats containing flocculated As- and Sb-bearing minerals were also observed at the HS location. The precipitation of these minerals appears to be the primary mechanism of As and Sb enrichment in the sediments and mats from the pond and spring sites. Electron microprobe analyses showed that high concentrations of As in the microbial mat samples were associated with amorphous, flocculated As-sulfide minerals in the mat material (Table 7), whereas Sb was associated mainly with amorphous Fe-oxides.

Wetland tadpoles

Boreal toad tadpoles, primarily sediment grazers, were abundant in the wetlands and had extremely elevated metalloid concentrations (Table 6). Tadpoles in the HS and CP ponds exhibited whole body As and Sb concentrations higher than any previously reported for tadpoles. Tadpoles at HS displayed whole-body As concentrations ranging from 1532 ± 435 to 3043 ± 523 mg kg^{-1} (DW) and average Sb concentrations between 33.5 ± 5.3 and 75.6 ± 56.8 mg kg^{-1}. Arsenic concentrations in CP tadpoles were 786.0 ± 161.8 and 1124 ± 488.6 mg kg^{-1} in 2011 and 2012 respectively and were particularly notable for their elevated Sb concentrations of >200 mg kg^{-1} (Table 6). In 2012, tadpoles from the OP and

Table 4. Mean As and Sb concentrations ± standard deviation and range in primary producer samples (dry weight) from Meadow Creek sites
Site abbreviations: see *Field Site* in the *Experimental* section. *n* refers to the number of samples; bd, below instrument detection limit

Organism	Site	*n*	Collection years	As (mg kg^{-1})		Sb (mg kg^{-1})	
				Mean	Range	Mean	Range
Alder leaves (*Alnus* sp.)	URR	3	2012	bd		bd	
	98PR	3	2012	2.5 ± 0.4	2.0–2.7	0.9 ± 0.2	0.7–1.1
	05PR	3	2012	5.7 ± 1.3	4.9–7.2	3.1 ± 1.2	2.0–4.2
	EFSF	2	2012	2.4 ± 0.003	2.35–2.36	2.2 ± 0.2	2.0–2.3
Willow leaves (*Salix* sp.)	URR	3	2012	bd		bd	
	98PR	3	2012	3.0 ± 0.5	2.4–3.4	bd	
	05PR	2	2012	29.3 ± 2.8	27.3–31.3	5.3 ± 0.8	4.7–5.9
	EFSF	2	2012	4.4 ± 0.9	3.7–5.1	3.3 ± 0.08	3.3–3.4
Vascular submergent plants	98PR	2	2011	3.1 ± 1.8	1.8–4.3	0.33 ± 0.18	0.2–0.5
	05PR	2	2011	46.8 ± 11.6	38.5–55.1	3.8 ± 0.7	3.3–4.2
	EFSF	1	2011	43.1		1.7	
	URR	2	2012	6.6 ± 0.6	6.1–7.0	bd	
Algae	DSR	2	2011	120.0 ± 31.0	98.1–141.8	11.2 ± 1.4	10.2–12.2
	URR	1	2012	5.8		bd	
	98PR	2	2012	20.0 ± 3.1	17.7–22.1	bd	
	05PR	1	2012	30.1		bd	
	EFSF	1	2012	18.8		bd	
Biofilm	URR	3	2012	19.9 ± 6.7	13.4–26.9	2.1 ± 0.2	1.9–2.3
	98PR	3	2012	32.7 ± 8.2	26.5–42.0	1.6 ± 0.6	1.2–2.2
	05PR	2	2012	323.3 ± 56.2	283.5–363.0	3.5 ± 2.5	1.7–5.3
	EFSF	2	2012	38.7 ± 10.9	31.0–46.4	2.3 ± 0.5	1.9–2.7

Table 5. Mean As and Sb concentrations ± standard deviation and range in animal samples (dry weight) from Meadow Creek sites
Site abbreviations: see *Field Site* in the *Experimental* section. *n* refers to the number of samples; bd, below instrument detection limit

Organism	Site	*n*	Collection years	As (mg kg^{-1})		Sb (mg kg^{-1})	
				Mean	Range	Mean	Range
Benthic Macroinvertebrates	URR	5	2010	1.3 ± 0.3	0.97–1.9	0.16 ± 0.11	0.05–0.30
	05PR	6	2010	27.1 ± 16.2	20.9–53.1	0.55 ± 0.30	0.21–1.1
	DSR	6	2010	30.5 ± 21.9	8.2–66.4	1.5 ± 2.0	0.25–5.3
	EFSF	6	2010	2.1 ± 0.90	1.2–3.5	0.14 ± 0.09	0.02–0.16
	URR	5	2011	1.2 ± 0.14	0.96–1.3	0.09 ± 0.04	0.04–0.14
	98PR	5	2011	11.0 ± 6.2	2.1–12.6	1.0 ± 0.34	0.71–1.5
	05PR	5	2011	8.8 ± 6.3	3.8–19.3	1.2 ± 0.9	0.62–2.7
	DSR	5	2011	13.6 ± 8.7	6.9–27.6	3.2 ± 1.1	1.7–4.3
	EFSF	5	2011	2.4 ± 1.2	1.1–4.3	0.8 ± 0.7	0.24–1.9
	URR	2	2012	2.3 ± 0.2	0.78–1.0	0.91 ± 0.45	0.12–0.75
	98PR	3	2012	6.7 ± 3.9	3.1–10.9	0.23 ± 0.05	0.19–0.28
	05PR	5	2012	10.0 ± 1.8	7.3–11.2	0.25 ± 0.12	0.13–0.44
	DSR	5	2012	19.2 ± 8.2	7.5–26.8	0.52 ± 0.21	0.34–0.82
	EFSF	2	2012	5.2 ± 0.25	5.0–5.4	0.46 ± 0.10	0.39–0.53
Trout	URR	3	2012	3.2 ± 1.2	2.3–4.6	bd	
	98PR	1	2012	bd		bd	
	05PR	2	2012	3.9 ± 0.4	3.6–4.2	bd	
	DSR	3	2012	4.2 ± 0.9	3.2–5.0	bd	
Tadpoles (*Ascaphus montanus*)	URR	2	2011	6.7 ± 0.30	6.5–6.9	0.16 ±0.0	0.16–0.16
	98PR	2	2011	637.4 ± 173.2	514.9–759.9	7.7 ± 3.6	5.2–10.3
	DSR	2	2011	142.0 ± 74.7	89.1–198.8	7.0 ± 3.5	4.5–9.5
	EFSF	2	2011	46.9 ± 13.7	37.2–56.6	0.38 ±0.0	0.37–0.38
	URR	3	2012	bd		bd	
	05PR	3	2012	80.8 ± 29.5	59.9–101.6	0.32 ± 0.45	0–0.6
	DSR	3	2012	90.3 ± 36.7	51.2–124.2	0.31 ± 0.53	0–0.91
	EFSF	3	2012	bd		bd	

BP ponds were also measured and contained highly elevated As (>390 mg kg^{-1}) and Sb (>20 mg kg^{-1}) concentrations.

Dissection of seep and spring pond tadpoles revealed that the intestinal tracts were filled and affected with sediment and mat material, despite our attempts to purge the animals prior to analysis (Fig. 4). The majority of the concentration for both metalloids was located in the intestinal tracts of the tadpoles, which accounted for >89.8 % of whole body As and >83.8 % of

whole body Sb (data not shown). Electron microprobe analysis of HS gut material showed that the high gut As concentrations were associated with amorphous As–S–Fe minerals, whereas the Sb was associated primarily with amorphous Fe-oxides (Table 7). Table 7 reports the weight percent (wt-%) of Sb, As, S and Fe in the gut material normalised to Si, Al, K, Mg and Ca, which are associated with the clay mineral matrix of the HS sediment. The same As- and Sb-bearing amorphous mineral phases identified in the gut were also present in the HS mat samples (Table 7; Fig. 4).

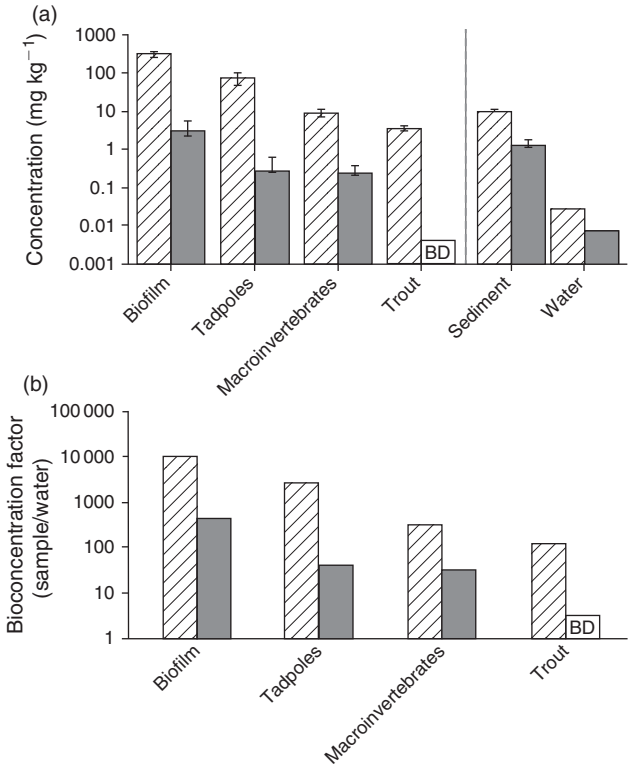

Fig. 3. A comparison of mean concentrations of As (patterned bars) and Sb (solid bars) in biofilm, tadpoles, macroinvertebrates, trout, sediment and water in the 2005 restoration project reach (05PR) reach of Meadow Creek during 2012 (a), and the bioconcentration factors of the two metalloids in each genera relative to water concentrations (b). Error bars represent standard deviation.

Discussion

Bioaccumulation of As and Sb in the stream ecosystem

The tailings piles at Stibnite Mine continue to be a source of high As and Sb concentrations in the surrounding watershed. Five to seven years post-remediation, dissolved As concentrations were still significantly elevated and exceed their respective MCL concentrations for human consumption in reaches downstream from the tailings and in the area of the flooded mine pit. Nonetheless, dissolved As concentrations in all stream reaches were below the EPA aquatic life standard (150 μg L^{-1}). Antimony concentrations were likewise below the proposed aquatic life standard of 30 μg L^{-1}. Despite meeting these aquatic life standards, metalloid concentrations in organisms downstream of the mine tailings were considerably elevated compared to upstream organisms. These findings demonstrate that As and Sb can bioaccumulate to significant levels in the food web even when aqueous concentrations are within levels deemed acceptable for wildlife.

Arsenic was present in water, sediment and biota with the following decreasing pattern of accumulation: primary producers > tadpoles > sediment > macroinvertebrates > trout > water. This is in agreement with previous reports that As is bioavailable at all trophic levels but does not bio-magnify in the food web.[5,8,14] Antimony was present at much lower concentrations than As, making the pattern of Sb accumulation in the stream difficult to discern. However, Sb concentrations were notably higher in biofilm and grazing tadpoles compared to water, sediment and other organisms. The BCF of organisms to water was considerably higher for As than Sb in all organisms. Arsenic is bioaccumulated to a greater extent than Sb into the photosynthetic base of the food web. These results are in agreement with previous reports of higher bioaccumulation of As than Sb for transfer from water to algae[8] and water to plants.[5]

Arsenic and Sb in benthic macroinvertebrates generally followed water concentrations with the 05PR and DSR reaches

Table 6. Mean As and Sb concentrations ± standard deviation and range in biological samples (dry weight) collected from wetland sites
Field Site in Experimental. *n* refers to the number of samples; bd, below instrument detection limit. For Algae–algal mat data, algae were used as the sample for CP and algal mats were used as the sample for HS and RS

Organism	Site	*n*	Collection dates	As (mg kg^{-1})		Sb (mg kg^{-1})	
				Mean	Range	Mean	Range
Submergent pond plants	CP	2	2011	171.6 ± 11.1		24.9 ± 4.9	
	OP	2	2012	926.6 ± 450.0	608.4–1244	78.4 ± 7.6	73.0–83.7
Algae–algal mat	CP	2	2012	1735 ± 129.8	1643–1827	47.3 ± 1.7	46.1–48.5
	HS	4	2012	3366 ± 200.4	3087–3558	30.2 ± 5.9	25.7–38.9
	RS	1	2012	76 564		674.9	
Tadpole whole body (*Anaxyrus boreas*)	HS	5	2010	2477 ± 378.0	2015–2777	54.8 ± 36.2	11.8–100.7
	CP	7	2011	786.0 ± 161.8	615.3–1113	283.6 ± 63.4	211.2–377.4
	HS	6	2011	1531 ± 435.6	906.2–2061	33.5 ± 5.3	25.2–41.1
	CP	5	2012	1124 ± 488.6	529.2–1815	200.4 ± 124.3	0–294.0
	HS	6	2012	3043 ± 523.7	2601–3866	75.6 ± 56.8	0–145.5
	OP	6	2012	396.3 ± 205.6	bd–417.0	21.4 ± 12.4	bd–28.6
	BP[A]	3	2012	732.3 ± 100.9	615.8–793.9	23.5 ± 3.1	21.6–27.1

[A]BP tadpoles are *Rana luteiventris*.

exhibiting the highest values, but trends were less pronounced than for the primary producers (Fig. 2). Antimony accumulation in the macroinvertebrates was quite low (average Sb concentration was <3.2 mg kg^{-1}), and the BCF of As in these organisms was higher than that of Sb by up to two orders of magnitude (Fig. 3b). The benthic macroinvertebrate larvae examined were mayflies (*Drunella* spp.), representing the shredder–gatherer functional feeding group, and filter feeding caddisflies (*Arctophyche* spp.). These feeding types are reported to accumulate higher concentrations of metals than omnivores or predators.[5,6] Likely food sources for these genera are submergent plants, algae, biofilm and riparian leaves that fall into the stream. We measured a wide range in metalloid concentrations within both genera for individuals collected from the same reach, but average concentrations of the two genera were similar in reaches where both were present. The macroinvertebrate concentrations reported in Table 4 represent data from pooled mayfly and caddisfly samples, and therefore provide an estimate of the As and Sb burden in a large proportion of the macroinvertebrate community that is available as prey for fish.

Trout did not exhibit biomagnification of As or Sb, nor did they display significantly elevated metalloid concentrations downstream compared to our reference reach (URR). However, whole body As concentrations in Meadow Creek trout ranged from 2.3 to 5.0 mg kg^{-1} (DW), slightly higher than values in fish reported from several other studies. For example, *Salmo trutta* from a mining affected river in France (Presa River) accumulated 1.92 mg kg^{-1} (DW) of As in whole body samples.[6] Telford et al.[5] reported flat-headed gudgeon (*Philypnodon grandiceps*) from a contaminated mining creek in Australia (As water concentration 46 ± 2 µg L^{-1}) to have 1.6 ± 0.4 mg kg^{-1} (DW), and in a moderately As-contaminated mining area in China, Fu et al.[8] reported As values in fish of 0.266 ± 0.109 mg kg^{-1}. However, fish muscle (*Channa striata*) from a highly contaminated pond in Thailand was reported to have As concentrations up to 22.2 mg kg^{-1}.[24] Trout did not display any discernable trends of As bioaccumulation as a function of water concentrations, possibly because they move between stream reaches.

Antimony bioaccumulation data for freshwater fish are limited. Telford et al.[5] reported that fish in their study had an average Sb value of 0.3 ± 0.3 mg kg^{-1}. The Presa River *S. trutta* samples had an average maximum Sb concentration of 0.45 ± 0.17 mg kg^{-1}.[6] In our study, trout Sb concentrations were all below detection (Table 5). We note that the fish sampled in our study were young (snout–fork length <5 cm), which may have precluded metalloid accumulation. Consequently, our values may underestimate tissue metalloid concentrations in older individuals if concentrations increase with age.

Studies on metal concentrations in tadpoles, and particularly on As and Sb concentrations, are also limited. Burger and Snodgrass[21] reported bullfrog tadpoles (*Rana catesbeiana*) from a contaminated Savannah River site (South Carolina, USA) to have average As concentrations of 3.1 ± 0.202 mg kg^{-1} (DW). Clark et al.[25] measured cricket frogs (*Acris crepitans*) with average As concentrations of 51.3 mg kg^{-1} (DW) in an As-contaminated lake. Telford et al.[5] measured As and Sb concentrations from two unidentified tadpole (*Anuran*) samples showing average concentrations of 62 ± 2 and 174 ± 10 mg kg^{-1} (DW) respectively. Arsenic and Sb concentrations in tailed frog tadpoles from the downstream sites were elevated compared to the URR, and this species had the highest metalloid concentrations in the stream food chain after the primary producers. Arsenic concentrations in tadpoles from the downstream reaches were consistently higher than those reported in previous studies, with notably high concentrations

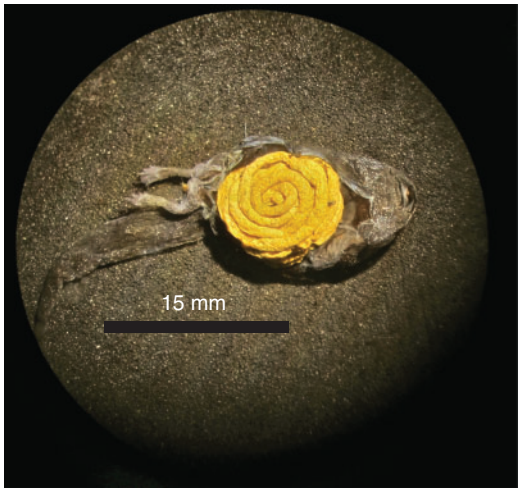

Fig. 4. *Anaxyrus boreas* tadpole under 0.7× magnification displaying ventral body cavity and affected gut containing microbial mat, sediment and flocculated As- and Sb-bearing mineral precipitates.

Table 7. Electron microprobe quantitative analysis (wt-%) from Heap Seep site microbial mat material and *Anaxyrus* tadpole intestinal tract (gut) material

Analyses are normalised to remove Si, Al, K, Mg and Ca associated with the silicate-sediment matrix

Analyte	Heap Seep mat material				Heap Seep gut material			
	As–S-rich		Sb–Fe-rich		As–S–Fe-rich		Sb–Fe-rich	
	wt-%	Normalised	wt-%	Normalised	wt-%	Normalised	wt-%	Normalised
Si	2.01	–	16.25	–	1.36	–	4.07	–
Al	1.48	–	9.72	–	0.70	–	2.75	–
K	0.30	–	0.60	–	0.22	–	1.99	–
Mg	0.00	–	0.73	–	0.00	–	0.12	–
Ca	0.04	–	6.97	–	0.12	–	1.43	–
Sb	4.64	11.56	43.16	65.85	0.01	0.01	28.32	78.76
As	21.62	53.82	2.06	3.14	36.11	39.06	0.65	1.82
S	13.24	32.97	0.14	0.21	20.17	21.82	0.06	0.17
Fe	0.66	1.64	20.19	30.81	36.15	39.11	6.92	19.25
Total	44.00	100	99.82	100	94.83	100	46.32	100

(637.4 ± 173.2 mg kg^{-1}) measured in 2011 at 98PR. Antimony concentrations in the stream tadpoles, on the other hand, were much lower than those reported by Telford.[5]

Effectiveness of the remediation projects

Engineered remediation projects conducted in 1998 and 2005 have reduced metalloid concentrations in Meadow Creek water, sediment and biota compared to pre-remediation values. Data collected prior to and immediately following the first remediation project in 1998 show that dissolved metalloid and sediment concentrations in the 98PR reach have decreased by 10 fold, to uncontaminated levels, since 1999. We infer that diversion and the installation of a sand filter at this site has been effective in reducing the metalloid burden to this short reach of Meadow Creek. In contrast, the strategies used in engineering the 05PR reach appear to be less effective in reducing dissolved metalloid and sediment concentrations in that reach, or in the more downstream DSR site. This could be the result of greater ground water input into the DSR than is delivered to the 98PR reach. This is supported by our finding that the 05PR and DSR are the only reaches that contain a significant proportion (30–37 %) of dissolved As in the form of reduced As^{III}, which we interpret to indicate that reducing groundwater is a predominant source of As to these stretches. The installation of a sand filtration barrier or other similar mechanism in the 05PR could reduce contaminated groundwater flow into that reach of the stream. Comparison of metalloid concentrations in macroinvertebrates from this study to values collected prior to and immediately after the 1998 remediation project show that insect larva concentrations have decreased in the 05PR and DSR. No prior macroinvertebrate data were available for the 98PR, but macroinvertebrate As concentrations in that reach were lower than the two more downstream reaches. Antimony levels in macroinvertebrates from all reaches in 2012 were not significantly elevated above the URR values.

Occurrence of As and Sb in wetlands

The most contaminated sites in this study were wetlands that occurred around the margins of the tailings heaps. The water in several of these wetlands contains dissolved As and Sb concentrations in the parts per million (mg L^{-1}) range. These extremely contaminated waters actively precipitate As- and Sb-bearing mineral phases which become incorporated into microbial mats, algae and sediment resulting in As concentrations in the parts per thousand (g kg^{-1}) range, and Sb concentrations of hundreds of parts per million (mg kg^{-1}). Metalloid concentrations in the base of the wetland food web are therefore largely reflective of the mineral precipitates that are dispersed throughout the mats, algae and sediments, rather than of direct biological assimilation.

These contaminated mats and sediments represent a direct source of As and Sb, by ingestion, to boreal toad tadpoles. Whole body concentrations in these tadpoles were up to 75.5 times higher in As and 2.2 times higher in Sb than the highest values that, to our knowledge, have been previously reported.[5] Purging methods had little effect on the removal of contaminated intestinal contents based on observations during dissection and direct analysis of intestinal metalloid concentrations. Our findings of high metal concentrations located in the gut, along with the ineffectiveness of purging, are in agreement with previous reports.[21,26,27] Electron microprobe analyses identified the same amorphous As-sulfide phases and Sb associated with iron-oxide minerals in both the HS microbial mat and in tadpole intestinal tracts from that location. The extreme metalloid concentrations in these tadpoles are therefore associated with ingestion of flocculated As- and Sb- bearing mineral precipitates contained within microbial mats.

Several prior studies have demonstrated the precipitation of amorphous As-bearing sulfide minerals resulting from microbiological As^{V} reduction to As^{III} in sulfidic conditions.[28,29] Similarly, recent work by Kulp et al.[19] demonstrated the precipitation of amorphous stibnite mineral (Sb_2S_3) during biological Sb^{V} and sulfate reduction by sediment bacterial communities from this study site. Others have shown that two crystalline polymorphs of Sb_2O_3, senarmontite and valentinite, are precipitated during bacterial Sb^{V} reduction in the absence of sulfide.[30] We attribute the high concentration of As- and Sb-bearing mineral phases in the contaminated algal and microbial mats of the ponds to geomicrobiological As- and Sb-cycling by microorganisms in the mat community. The biologically induced precipitation of these mineral phases represents a direct mechanism of concentration for As and Sb in the base of the food web, and these minerals are actively consumed along with the mat material by tadpoles. The apparent resistance of these larval amphibians to extremely high concentrations of As and Sb in water, food and sediment reinforces the suggestion by Kerby et al.[31] that the commonly presumed high sensitivity of amphibians to trace metal contamination may not apply to all taxa.

Conclusions

Tailings derived from gold, antimony and tungsten mining activity can leach high concentrations of As and Sb to nearby streams and wetlands. Remediation efforts, such as those completed along Meadow Creek, can be effective in reducing water metalloid concentrations to levels below EPA aquatic life standards. In this study the more effective remediation design was that which incorporated water filtration (e.g. sand filters). Infiltration of groundwater appears to be a direct source of As and Sb to the remediated reach that did not include a sand filter.

Arsenic bioaccumulates at all trophic levels in the stream food web, but it does not bio-magnify and it generally diminishes with increasing trophic level. Antimony accumulates to a lesser extent than As and accumulates most readily in lower trophic levels (e.g. biofilm and plants). Extremely elevated metalloid concentrations in two species of tadpoles (one stream-dwelling and one pond-dwelling species) are caused by ingestion of algal and microbial mat material and sediment that are highly contaminated with As- and Sb-bearing minerals. These minerals are interpreted to be the biogenic products of microbiological As and Sb cycling in the mats, and are a primary source of metalloid uptake for these tadpoles. Contaminated sediments and mat material remained present in the tadpole gut even after purging the live animals by conventional methods, and account for the highest whole body As and Sb concentrations ever recorded in live tadpoles. This study adds to the growing number of investigations reporting bioaccumulation and food web diminution of As and Sb in freshwater ecosystems, and suggests that tadpoles may be far more resistant to metalloid contamination than was previously assumed. Future studies should investigate the mechanisms of unexpectedly high resistance to metalloid toxicity in these tadpoles.

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