

Investigation into metal contamination of the Berg River, Western Cape, South Africa

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Abstract

A recent decline in water quality of the Berg River, Western Cape, South Africa, has led to the investigation into the degree of metal pollution in the river system. This study was conducted over a period of one year, from May 2004 to May 2005. The nitric acid digestion technique was used to extract metals from water, sediment and biofilm samples collected at various points (Site A – agricultural area, Site B – informal settlement and Site C – Newton pumping station) along the Berg River. Metal concentrations were determined using inductively coupled plasma atomic emission spectrometry (ICP-AES). The highest mean metal concentrations recorded were as follows; water samples, 6 mg·l⁻¹ for Al, 14.6 mg·l⁻¹ for Fe and 18.8 mg·l⁻¹ for Mn; sediment samples, 17 448.8 mg·kg⁻¹ for Al and 26 473.3 mg·kg⁻¹ for Fe; biofilm samples, 876.8 mg·l⁻¹ for Al and 1 017.5 mg·l⁻¹ for Fe. The increased availability, or noteworthy incidence of Al and Fe, could be due to the leaching of metals into the river water from waste and household products associated with the informal settlement and the subsequent settling on sediment. No guidelines were available for metals in biofilms. The highest recorded concentrations in water were for Site C (agricultural area). Recorded concentrations in water fluctuated throughout the study period for most of the metals analysed, but Al and Fe were consistently above the recommended guidelines as stipulated by the Department of Water Affairs and Forestry and the Canadian Council of Ministers of the Environment.

Keywords: metals, river systems, sediment, biofilm, inductively coupled plasma atomic emission spectrometry (ICP-AES)

Introduction

Metals occur in less than 1% of the earth's crust, with trace amounts generally found in the environment (Alloway, 1995a). When these concentrations exceed a stipulated limit (South African Bureau of Standards, 2001; World Health Organisation, 1991), they may become toxic to the surrounding environment. Sources of metal contamination include industrial and medical waste (Dorigo et al., 2004), pesticides, petroleum by-products (Mowat and Bundy, 2001), household products, as well as urban and pharmaceutical waste (Brooks et al., 2003). Domestic and household sources of metal contamination generally occur as a result of corrosion of metal plumbing fittings, galvanised roofs and wire fences [zinc (Zn), cadmium (Cd)], and healthcare products, such as Zn- or selenium- (Se) containing shampoos and Zn-containing baby creams (Alloway, 1995b). Silver paint containing aluminium (Al), Al-coated roofs, saucepans and utensils (Friberg et al., 1986), are also possible sources of contamination.

Natural watercourses can also be contaminated with micro-organisms, which inhabit the natural environment in the form of planktonic organisms and sessile biofilms. Under favourable conditions, micro-organisms will generally form a biofilm on any surface exposed to an aqueous environment. These biofilms can be defined as a community of attached microbial cells organised within extracellular polymer matrices (EPS). This EPS assists in the bacterial survival by providing protection against metals, predation and environmental fluctuations, and also provide increased resistance against antimicrobial agents

(Decho, 1990). Biofilms are advantageous in that they encapsulate toxic molecules, such as metals (Costerton et al., 1978), by providing a substrate for them to adhere to, thereby limiting the diffusion of biocides and other toxic molecules across the EPS (De Beer et al., 1994; Huang et al., 1995). This implies that the attached biofilm communities may then be employed in the removal of toxins from aqueous systems, as the biofilms are able to concentrate and bind ions from the passing water (Neu et al., 1992).

The presence of metals in biofilms isolated from the Elbe River in Germany was determined in a study by Friese et al. (1997). They found fractions of several elements from stones and plates incubated in the river. These elements included potassium (K), calcium (Ca), chromium (Cr), manganese (Mn), lead (Pb), nickel (Ni), copper (Cu), zinc (Zn) and iron (Fe). Factors contributing to microbial biodiversity and the tolerance of certain organisms to metals depend on the type of attachment material (e.g. glass, stones, leaves, rocks) the biofilm is isolated from, the age of the biofilm and the concentration of the EPS. In addition, research has indicated that the predominant metals present in streams and lakes are Zn, Pb, Cu, Fe and Mn (Geesey et al., 1992; Nelson et al., 1996).

Metal concentrations in environmental samples, may be determined using different analytical techniques. These techniques can be divided into single element analysis, using atomic absorption spectrometry (AAS) and simultaneous multi-element analysis, using ICP-AES and X-ray fluorescence spectrometry (XRF). Vermeiren et al. (1990) investigated ICP-AES as a method of determining the presence of the metals Cd, Pb, Cu and Zn in natural waters. Certain samples were enriched with specific concentrations of the metals, while others were not. Comparison of the results obtained for the enriched and non-enriched river water samples indicated that the technique is accurate in the determination of metal concentrations.

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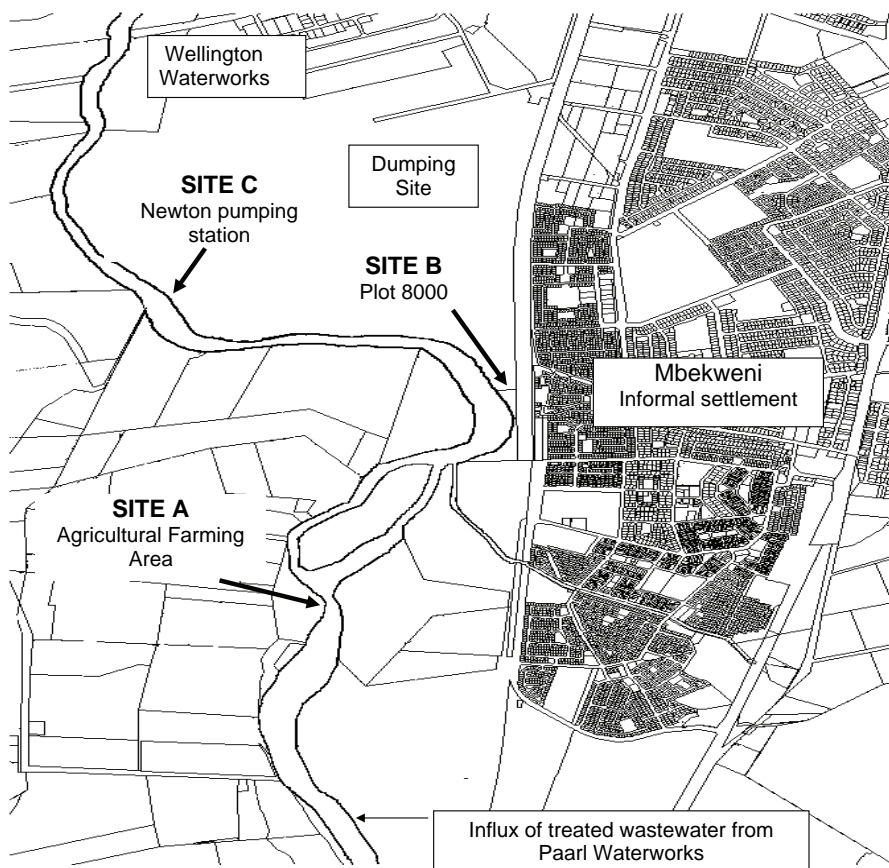


Figure 1

Map of the Berg River indicating the different sampling points: Site A, indicates the agricultural farming area; Site B, indicates Plot 8000, close to the informal settlement of Mbekweni; and Site C, the Newton Pumping station

An increase in urbanisation has led to an upsurge in informal settlements in the Western Cape, South Africa, where the inhabitants of these settlements experience a distinct lack of resources. Inadequate sanitation behaviour and a lack of adequate sewage disposal facilities may lead to the leaching of potentially harmful substances, from waste, household products, etc. into the environment. The Berg River was specifically selected due to the increased incidence of pollution recorded within the area in recent times. This increased pollution is of great concern, as the Berg River serves as a water source to towns, cities, rural communities, farms and recreational users in the area (River Health Programme, 2004). The objective of this study was to identify the predominant metals, which occur within the river water, sediment and the biofilm samples isolated from the Berg River (Paarl) in the Western Cape, South Africa.

Materials and methods

Site description

Sampling commenced in May 2004 and was conducted over a period of one year until May 2005. Experimental work entailed collecting samples along the Berg River (Western Cape) after 1, 5, 9, 17, 21, 25, 33, 37, 41, 45 and 49 weeks from the commencement of the study (Fig. 1). As indicated in Fig. 1, the samples were collected from Site A (agricultural

farming area) and Site B, known as Plot 8000 (at the informal settlement of Mbekweni). Stormwater drainage pipes from the communities in the settlement discharge into the river at Site B. A 3rd site, Site C (the Newton pumping station), serves as an inlet of storm drainage water and wastewater into the river from the residential area of Newton as well as certain areas of Mbekweni.

Sampling

Water samples (200 ml) were collected in sterile 250 ml narrow-mouth square polypropylene bottles (Cole-Palmer Instrument Company). The sediment samples were collected in 250 ml plastic containers and consisted of a combination of five different sub-samples (± 15 cm deep) collected at different locations in a defined perimeter. The biofilm samples were obtained by collecting various materials, such as glass, leaves, stones, rocks, etc. (~100 g) along each of the different sampling points and storing them in sterile whirl-pack bags. The samples were stored at 4°C during transport.

Sonication of collected biofilm samples

Bacterial suspensions were removed from rocks, leaves, glass and stones collected from each representative site, by sonication. Collected material samples (~100 g) were sonicated for 10 min in 30 ml sterile distilled water in a UMC5 sonication bath (Instrulab, Inc.). The procedure was repeated at least twice, with fresh sterile d.H₂O added after each sonication step. The sonicated samples were combined resulting in a total of 60 ml bacterial suspension. The biofilm suspension obtained was used for further analysis.

Metal concentrations in sediment, biofilm and water samples

To determine the concentrations of Al, Zn, Cu, Fe, Pb, Ni and Mn, sediment samples (0.500 to 0.600 g; dry mass), biofilm suspensions (5 ml) and water samples (5 ml) were digested with 10 ml 55% nitric acid at 40°C for 60 min and then at 120°C for 180 min, using a Grant dry-block heater. A blank (control) of 10 ml 55% nitric acid was analysed along with the collected samples, to check for possible contamination. The samples were cooled to room temperature, filtered with Whatman filter paper No 6 into 20 ml volumetric flasks, made up to a volume of 20 ml with distilled water and subsequently filtered for a second time using 0.45 µm cellulose nitrate ultrafiltration membrane filters (Whatman). Metal concentrations were determined using ICP-AES analysis according to the procedure outlined in Saleh et al. (2000).

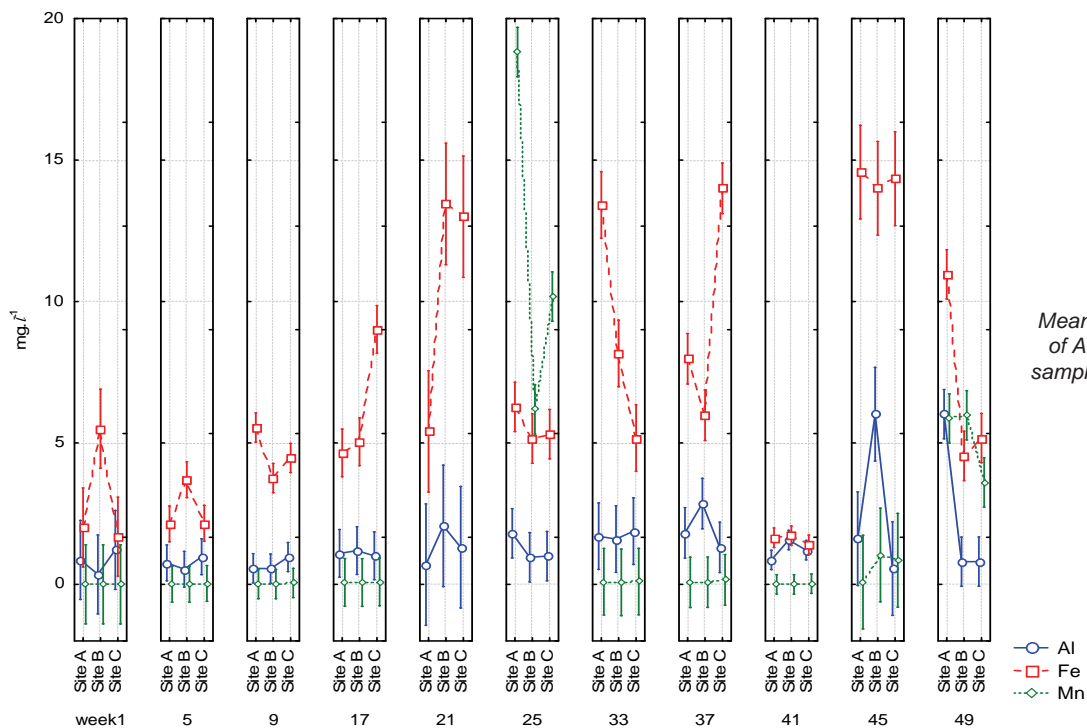


Figure 2A
Mean metal concentrations
of Al, Fe and Mn in water
samples from the Berg River

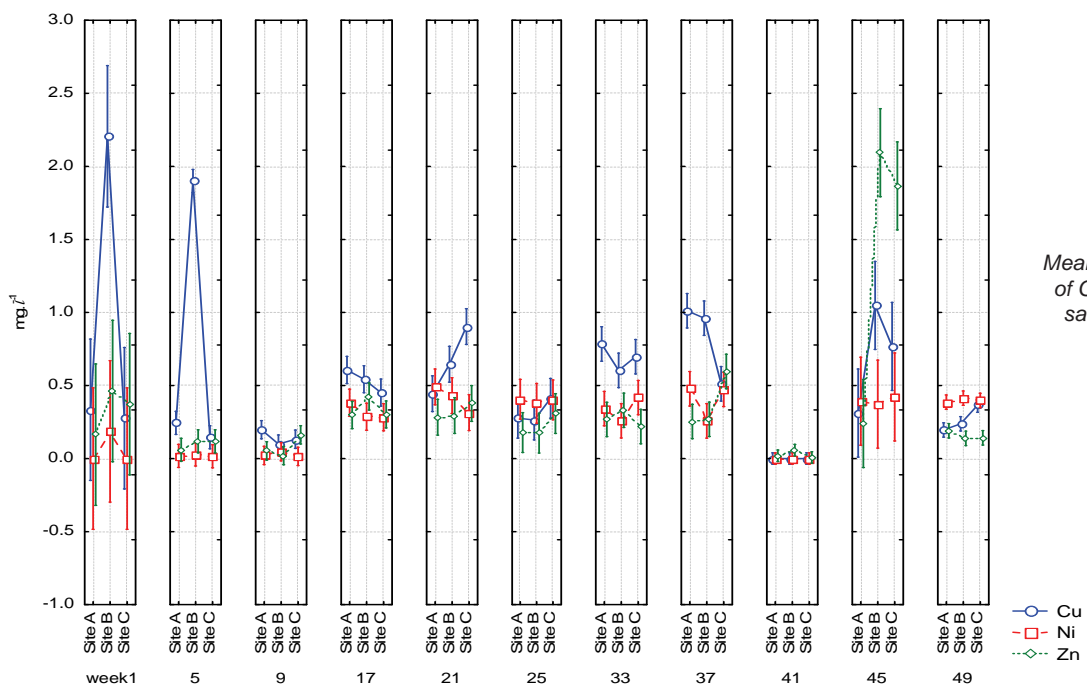


Figure 2B
Mean metal concentrations
of Cu, Ni and Zn in water
samples from the Berg
River

Statistical analysis

The results presented are the averages of five repeats for each particular sampling point at the different sampling sites. For statistical analysis, dry mass (0.500 to 0.600 g) and volume (5 mL) were taken into consideration for the calculation of the final metal concentrations in a given sample. Repeated measures ANOVA (RMAs) were performed on all data obtained as outlined in Dunn and Clark (1987), using Statistica™. In each RMA, the residuals were analysed to determine whether they were normally distributed. In all hypothesis tests a significance level of 5% was used as standard.

Results and discussion

Metals in water

Comparisons of the mean metal concentrations in water are represented in Figs. 2A and B. The highest mean metal concentrations were recorded for Mn (18.8 mg·L⁻¹), Fe (14.6 mg·L⁻¹) and Al (6 mg·L⁻¹) during weeks 25, 45 and 49, respectively, at Site A. The concentrations of Al and Fe were significantly higher than the recommended guidelines of Al and Fe in freshwater systems as stipulated by the Department of Water Affairs and Forestry (DWA, 1996) and the Canadian Council of Ministers

of the Environment (CCME, 2001) (Table 1). Overall the mean concentrations for Al ranged from 0.4 mg·ℓ⁻¹ during Week 1 at Site B to 6 mg·ℓ⁻¹ during Week 49 at Site A, and for Fe from 1.7 mg·ℓ⁻¹ during Week 1 at Site C to 14.6 mg·ℓ⁻¹ during Week 45 at Site A (Table 1). The particular agricultural area, situated near the Berg River in the Western Cape uses the following pesticides and insecticides for the treatment of their crops: Acrobat® MZ, Copperflo, Cosavet, Folpan, Karathane, Mancozeb™, Phosguard™. The increased concentrations of Al could be due to the use of Phosguard™, of which Al oxide is a component (Seachem, 2006). Phosguard™ is a porous adsorbent that removes phosphates, silicates, and other nutrients that support algae growth in this particular agricultural area.

Manganese concentrations fell within the recommended quality guideline range for most of the sampling period with the exception of Weeks 25, 45 and 49, where increased concentrations ranging from 3.6 mg·ℓ⁻¹ to 18.8 mg·ℓ⁻¹ at Sites C and A (Fig. 2), respectively, were recorded. Manganese is a major component of pesticides such as Mancozeb™ and Maneb. Mancozeb™, then in turn, makes up 60% of the fungicide, Acrobat® MZ, which is used to manage early and late blight in potatoes (Acrobat®, 2005). Mancozeb, or manganese-zinc ethylene bis (dithiocarbamate), is a fungicide used for the treatment of plant diseases in the Western Cape. Its usage in agriculture amounts to 1 343 x 10³ kg/a (Vermeulen et al., 2001). Contamination of

the water could be due to excessive use or improper discarding of these pesticides into the river (Agency for Toxic Substances and Disease Registry) (ATSDR, 2000). As previously mentioned, Site A (Fig. 1) is situated in an agricultural area and is the site at which the highest concentrations of metals were recorded. Run-off from a nearby farm enters the river at this site, which could account for the increase in Mn concentrations.

The recorded concentrations of Cu, Ni and Zn in water samples varied throughout the study period (Table 1). Lead could not be detected in any of the analysed samples. The concentrations of Cu, Ni and Zn fluctuated above and below the recommended guidelines of DWAF (1996) and the CCME (2001). The mean concentrations recorded for Ni were higher than the recommended concentrations during Weeks 17, 21, 25, 33, 37, 45 and 49. The recorded mean Cu and Zn concentrations fell within the recommended guidelines for Week 41 only (Table 1). The increases in Zn could also be as a result of Mancozeb™ use, as Zn is a component of this pesticide. The increased metal concentrations could also possibly be due to recurring anthropogenic episodes associated with the Berg River during the sampling period (Barnes, 2003).

Metals in sediment

Comparisons of the mean concentrations for Al and Fe are presented in Fig. 3. Results for Cu, Mn, Ni, Pb, and Zn are presented in Tables 2 and 3. No recommended sediment quality guidelines for Al, Fe, Mn, Pb, Ni, Cu and Zn were available from DWAF (1996) and guidelines for Cu and Zn only were available from the Canadian Council of Ministers of the Environment (CCME, 2001). Significant differences ($p < 0.05$) in the concentrations for Al and Fe were observed for the sediment samples. The highest mean concentration (mg·kg⁻¹) of 17 448.8 mg·kg⁻¹ was recorded for Al during Week 1 at Site B (Fig. 3). In comparison to the results obtained for Week 1, no significant differences in concentrations ($p > 0.05$) of Al were recorded from Weeks 5 to 49, where mean concentrations ranged from 353.2 mg·kg⁻¹ at Site A to 2 687.7 mg·kg⁻¹ at Site C, both during Week 5. The highest mean concentrations of 21 035 mg·kg⁻¹ and 26 473.3 mg·kg⁻¹ were recorded for Fe in Weeks 1 and 5, at Sites B and C, respectively. In comparison to the results obtained for Weeks 1 and 5, no significant ($p > 0.05$) differences in concentrations of Fe were recorded for Weeks 9 to 49, where the mean concentrations ranged from 700 mg·kg⁻¹ to 7 014.9 mg·kg⁻¹, during Weeks 33 and 45, at Sites A and C, respectively.

TABLE 1
Concentrations obtained in water of the Berg River compared to recommended safe concentrations as stipulated by the Department of Water Affairs and Forestry (1996) and the Canadian Council of Ministers of the Environment Quality Guidelines (2001)

Metal	Recommended safe concentrations as stipulated by DWAF (1996) (mg·ℓ ⁻¹)	Environmental quality guidelines as stipulated by CCME (2001) (mg·ℓ ⁻¹)	Mean metal concentrations obtained in water (mg·ℓ ⁻¹)
Al	0.1 – 0.15	0.005 – 0.1	0.4 – 6
Cu	0.002 – 0.012	0.002 – 0.004	0 – 2.2
Fe	N/A	0.3	1.4 – 14.6
Mn	1.3	N/A	0 – 18.8
Ni	N/A	0.025 – 0.15	0 – 0.5
Pb	N/A	0.001 – 0.007	0 – 0
Zn	0.036	0.03	0.01 – 2.1

N/A = Data not available

TABLE 2
Mean metal concentrations of Cu, Mn and Ni recorded in sediment samples for the different sampling weeks

Time (weeks)	Metals (mg·kg ⁻¹)								
	Cu			Mn			Ni		
	A	B	C	A	B	C	A	B	C
1	5.0±4.5	74.0±0	8.2±2.1	3.0±0.5	11.0±0	11.0±0.9	5.0±0.2	44.0±0	5.0±0.1
5	4.0±1.6	3.8±1.8	12.0±2.9	5.0±0.6	7.5±0.8	70.0±5.8	5.0±0.3	5.0±0.4	5.0±0.1
9	2.0±0.9	3.2±1.8	4.4±2.2	3.0±0.5	8.4±1.1	12.0±3.4	4.0±0.4	5.0±0.5	8.0±0.3
17	2.0±0.1	5.9±2.5	9.9±2.1	7.0±0.6	9.4±0.6	12.0±0.7	5.0±0.1	5.0±0.2	5.0±0.4
21	3.0±0.6	3.3±0.4	2.9±0.4	5.0±1.3	8.6±0.5	13.0±2.5	0.0±0.0	0.0±0.0	6.0±0.5
25	1.0±1.7	12.0±1.9	11.0±3	8.0±0.4	3.9±0.3	13.0±0.9	0.0±0.0	0.4±0.0	0.0±0.0
33	5.0±1.3	4.9±0.9	6.7±2.2	3.0±0.2	4.1±0.4	5.9±0.4	2.0±0.3	2.0±0.4	1.0±0.3
37	7.0±0.9	6.3±0.8	7.4±1.2	7.0±0.9	5.1±0.5	15.0±2.3	2.0±0.4	2.0±0.3	3.0±0.2
41	5.0±0.8	4.9±1.3	4.5±0.2	9.0±0.7	4.4±0.4	19.0±1.4	0.0±0.0	0.0±0.0	3.0±0.3
45	7.0±0.9	6.7±1.6	6.9±0.4	11.0±0.6	16.0±3.8	43.0±7.2	4.0±0.6	5.0±0.5	1.0±0.7
49	4.0±0.7	2.9±0.7	7.8±4.9	7.0±0.2	12.0±1.4	44.0±1.4	4.0±0.4	4.0±0.3	5.0±0.4

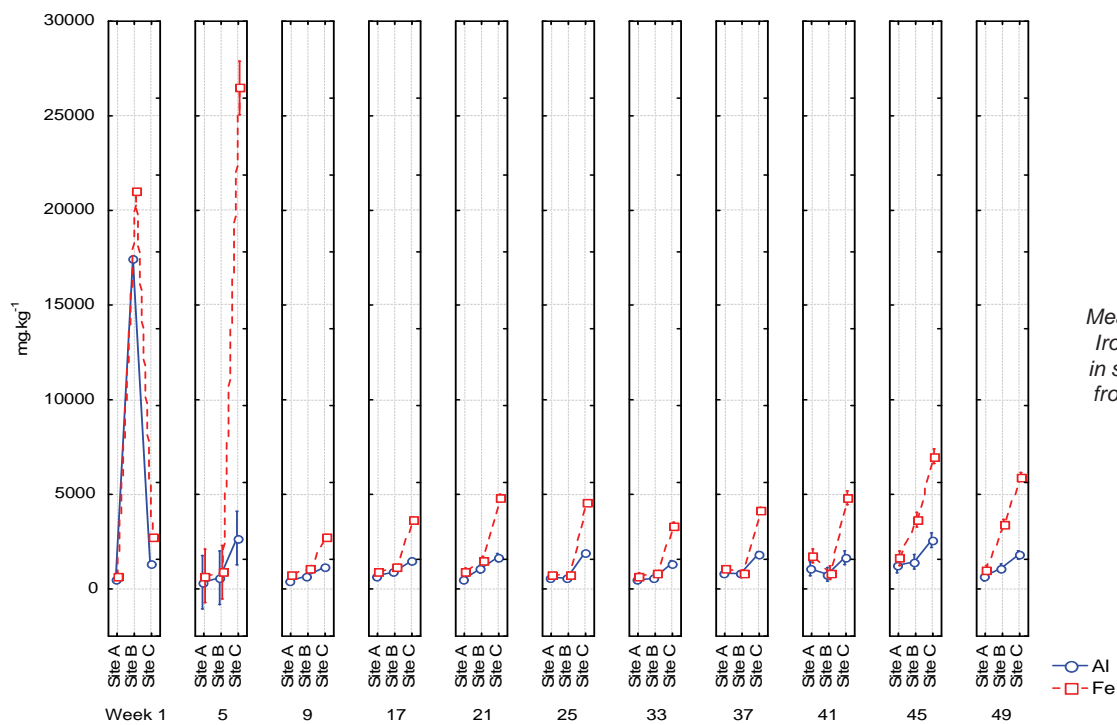


Figure 3
Mean Aluminium and Iron concentrations in sediment samples from the Berg River

Time (weeks)	Metals (mg·l ⁻¹)									
	Cu			Mn			Ni			
	Sites	A	B	C	A	B	C	A	B	C
1		1.0±0.2	1.7±0.2	2.0±0.5	2.6±0.9	1.4±0.1	9.0±0.8	0.4±0.1	0.3±0.0	0.3±0.0
5		1.0±0.1	0.2±0.0	1.0±0.1	2.2±0.2	1.1±0.1	11.0±0.3	0.5±0.0	0.4±0.0	0.7±0.0
9		1.0±0.1	0.7±0.2	1.0±0.2	4.9±0.3	2.0±0.1	32.0±1.4	0.6±0.0	0.6±0.0	0.9±0.0
17		1.5±0.2	0.8±0.1	1.0±0.1	17.0±3.1	8.4±1.3	9.0±1.2	19.0±41.0	0.6±0.0	0.7±0.1
21		0.0±0.0	0.5±0.1	0.0±0.0	1.4±0.4	4.6±0.8	8.0±0.5	0.0±0.0	0.0±0.0	0.0±0.0
25		1.0±0.1	0.7±0.1	1.0±0.2	71.0±17.0	1.7±0.2	6.0±0.4	1.0±0.1	0.4±0.0	0.5±0.0
33		0.3±0.0	0.4±0.1	0.0±0.0	0.7±0.0	1.4±0.2	3.0±0.3	0.0±0.0	3.0±5.8	0.0±0.0
37		2.0±0.2	0.4±0.0	1.0±0.1	13.0±1.5	2.2±0.5	9.0±1.2	0.5±0.0	0.0±0.0	0.1±0.0
41		1.0±0.0	0.4±0.1	0.0±0.1	3.4±0.4	1.6±0.2	4.0±0.2	0.0±0.0	0.0±0.0	0.0±0.0
45		1.0±0.1	0.6±0.1	0.0±0.1	1.2±0.1	1.8±0.4	5.0±0.8	0.7±0.0	1.0±0.1	0.4±0.0
49		1.0±0.1	0.7±0.0	0.0±0.1	0.8±0.0	1.3±0.2	4.8±0.1	0.6±0.1	0.4±0.0	0.3±0.0

The Agency for Toxic Substances and Disease Registry (ATSDR, 1995) reported that Al comprises 8% of the earth's crust, with dissolved Al and Fe primarily derived from soils (Neal et al., 1997). In addition, Al forms complexes with organic matter in soil, which could contribute to elevated concentrations (Tipping et al., 1991).

Effluent from a nearby stormwater drain enters the river at Site B, where the highest concentration of Al was recorded in Week 1. The high concentrations of Al and Fe in the sediment samples could partly be due to the leaching of metals from housing materials and household products, utilised by inhabitants of the informal settlements, into the river. In addition, the galvanised sheeting used to build informal dwellings is primarily composed of Fe. When pure iron reacts readily with oxygen and moisture in the environment, a red or brown ferric oxide coating is formed, which destructively corrodes the galvanised sheeting. Excess Fe in aqueous environments can be the cause of chronic and acute health effects, but according to the Department of Water Affairs and Forestry, accidental Fe poisoning is rare (DWAF, 1996).

Tables 2 and 3 represent the mean concentrations for Cu, Mn, Ni, Pb and Zn. Manganese, Ni and Pb concentrations were significantly low throughout the study period with the highest mean metal concentrations for Mn, Ni and Pb recorded at: 70 mg·kg⁻¹ in Week 5 at Site C; 44 mg·kg⁻¹ in Week 1 at Site B and 23 mg·kg⁻¹ in Week 45 at Site B (Tables 2 and 3), respectively. No recommended sediment quality guidelines for Pb, Mn and Ni were available from DWAF (1996) and the CCME (2001).

The highest mean Cu concentration of 74 mg·kg⁻¹ was recorded at Site B, during the first sampling week (Table 2). This result for Cu was higher than the recommended environmental quality guideline of 35.7 mg·kg⁻¹ in freshwater sediment as specified by the CCME (2001). Thereafter, during Weeks 5 to 49, concentrations for Cu decreased significantly ($p < 0.05$), to levels lower than the guideline (CCME, 2001).

The highest mean Zn concentration of 395 mg·kg⁻¹ was recorded at Site B, during Week 1 (Table 3). This result was higher than the recommended Canadian sediment quality guide-

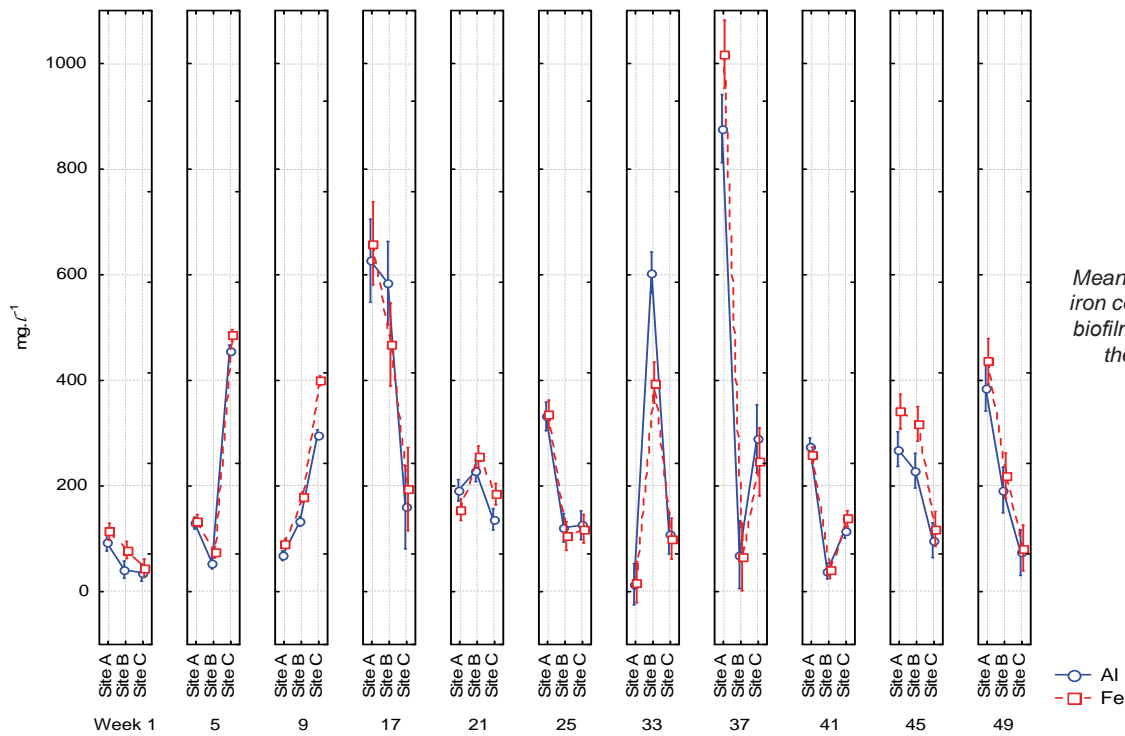


Figure 4
Mean aluminium and iron concentrations in biofilm samples from the Berg River

lines of $123 \text{ mg}\cdot\text{kg}^{-1}$ (CCME, 2001). During the subsequent sampling weeks the mean Zn concentrations ranged from 4 to $36 \text{ mg}\cdot\text{kg}^{-1}$ (Table 3), which falls within the accepted CCME guidelines. During Week 1 of sampling the region experienced very low rainfall (6 mm). In the following weeks, the recorded rainfall ranged between 52.2 mm and 154 mm. The lower rainfall could imply that the metals were not readily available in the water column, impeding metal transfer from site to site and increasing metal accumulation in the sediment.

Metals in biofilm

No recommended biofilm quality guidelines were available from DWAF (1996) and the Canadian Council of Ministers of the Environment (CCME, 2001). Biofilms are layers of organisms, organic matter and inorganic material, organised within EPS (Decho, 1990). These organisms attach and develop on biologically active or non-active surfaces (Stickler, 1999).

Comparisons of the mean concentrations for Al and Fe are presented in Fig. 4. Results for Cu, Mn, Ni, Pb, and Zn are represented in Tables 4 and 5. The highest metal concentrations recorded in the biofilm samples ($\text{mg}\cdot\text{l}^{-1}$), were for Al and Fe, as in the case of sediment. Throughout the entire study period, Fe was consistently present in elevated concentrations in water, sediment and biofilm samples when compared to the other metals analysed for. The recorded results for Fe and Al fluctuated throughout the entire study period. The mean concentrations recorded for Al ranged from $14.1 \text{ mg}\cdot\text{l}^{-1}$ during Week 33 to $876.8 \text{ mg}\cdot\text{l}^{-1}$, during Week 37, both at Site A (Fig. 4). In addition, the mean Fe concentrations in biofilm samples ranged from $18 \text{ mg}\cdot\text{l}^{-1}$ during Week 33 to $1017.5 \text{ mg}\cdot\text{l}^{-1}$ during Week 37, again both at Site A (Fig. 4).

As indicated above, an increase in the concentrations of Al and Fe in the biofilm samples was observed at Site A in Week 37. For the same time period, a corresponding study investigating microbial contamination of the river revealed a microbial count in sessile samples of 3.9×10^7 organisms. This increased number

of micro-organisms could have facilitated the Al and Fe accumulation in the biofilm. It is well recognised that micro-organisms have developed unique means of resistance to specific metals (Roanne and Pepper, 2000), the mechanisms of which are not yet fully understood. Extracellular polymeric substances contain various constituents such as polysaccharides, proteins, nucleic acids, lipids or humic substances (Mayer et al., 1999). Decho (1990) showed that the EPS assists in the bacterial survival by providing protection against predation and environmental fluctuations.

Tables 4 and 5, represents the metal concentrations recorded in biofilm samples for Cu, Mn, Ni, Pb and Zn. These metal concentrations were consistently lower than the concentrations recorded for Al and Fe. The highest mean metal concentrations for Cu, Mn, Ni, Pb and Zn were recorded at: $2 \text{ mg}\cdot\text{l}^{-1}$ in Weeks 1 and 37 at Sites C and A; $71 \text{ mg}\cdot\text{l}^{-1}$ in Week 25 at Site A; $19 \text{ mg}\cdot\text{l}^{-1}$ in Week 17 at Site A; $1.6 \text{ mg}\cdot\text{l}^{-1}$ in Week 5 at Site C and $8.4 \text{ mg}\cdot\text{l}^{-1}$ in Week 25 at Site A, respectively (Tables 4 and 5).

Kröpfel et al. (2006) studied metal accumulation by biofilms in the Tisza River, Hungary. The authors utilised different substrates to cultivate biofilms, with the aim of investigating the applicability of biofilms as biomonitors. The different substrates used were andesite, polished granite, Plexi-glass, granite and polycarbonate. The biofilms were cultivated for six weeks, after which, the samples were analysed using total reflection x-ray fluorescence (TXRF) for elemental analysis. The concentrations of essential elements and heavy metal pollutants (Cu, Ni, Pb and Zn) were highest in biofilms on polished granite or granite (rocks and stones) (Kröpfel et al., 2006). In the present study, various substrates such as rocks, leaves, glass and stones were collected as representative biofilm samples from the Berg River. Biofilms are currently being studied as biomonitors in ecological research, but it is important to note that as yet no guidelines exist for biofilms in aquatic systems.

Research has shown that the EPS layer also exhibits a high metal absorption capacity. A study by Suh et al. (1999) investigated the accumulation of Pb^{2+} into the EPS layer of pure species

Metals (mg·kg ⁻¹)						
Time (weeks)	Pb			Zn		
Sites	A	B	C	A	B	C
1	0.0±0.0	2.0±0.3	2.1±0.3	7.6±1.4	395.0±0.0	11.0±0.5
5	2.0±1.3	2.0±0.6	11.0±1.7	6.5±0.7	12.0±2.3	22.0±2.3
9	1.0±0.8	2.0±1.9	3.0±0.4	7.0±1.4	11.0±2.3	11.0±1.4
17	3.0±2.2	2.0±0.2	3.0±0.4	8.5±0.5	9.5±0.5	9.0±1.4
21	1.0±0.3	3.0±0.4	4.2±0.7	4.3±0.8	9.2±0.8	7.3±0.9
25	3.0±0.4	2.0±0.2	4.2±0.3	13.0±1.4	9.2±0.5	11.0±0.9
33	2.0±0.5	2.0±0.3	2.9±0.6	4.0±2.1	3.8±0.4	6.2±0.7
37	3.0±0.6	3.0±0.5	4.9±0.3	8.6±0.5	10.0±0.7	11.0±1.3
41	4.0±0.6	3.0±0.3	4.6±2.6	13.0±1.0	12.0±0.9	14.0±2.3
45	5.0±1.5	23±34	8.9±6.8	19.0±2.5	23.0±7.9	16.0±1.3
49	2.0±0.4	6.0±2.9	5.1±0.6	12.0±1.0	27.0±6.7	36.0±41.0

Metals (mg·ℓ ⁻¹)						
Time (weeks)	Pb			Zn		
Sites	A	B	C	A	B	C
1	0.4±0.1	0.0±0.0	0.2±0.0	2.0±0.5	2.0±0.2	1.0±0.1
5	0.4±0.0	0.2±0.0	1.61±0.1	2.0±0.2	2.0±0.1	5.0±0.2
9	1.3±0.2	0.6±0.0	1.2±0.1	2.0±0.1	3.0±0.2	6.0±0.3
17	0.3±0.0	0.9±0.1	0.5±0.1	5.6±1.0	3.0±0.5	2.0±0.3
21	1.3±0.2	0.7±0.1	0.4±0.1	0.0±0.0	2.0±0.4	1.0±0.2
25	0.0±0.0	0.2±0.0	0.3±0.1	8.4±1.0	1.0±0.1	2.0±0.2
33	0.2±0.1	0.3±0.0	0.2±0.0	0.5±0.0	1.0±0.2	1.0±0.1
37	0.7±0.1	0.2±0.0	0.7±0.1	8.0±0.1	1.0±0.1	2.0±0.3
41	0.7±0.1	0.0±0.0	0.2±0.0	3.0±0.7	1.0±0.2	1.0±0.3
45	0.9±0.1	0.6±0.1	0.1±0.1	5.0±0.6	3.0±0.5	1.0±0.1
49	1.4±0.4	0.5±0.0	0.0±0.0	6.0±1.4	3.0±0.2	1.0±0.1

Aureobasidium pullulans biofilms using transmission electron microscopy (TEM). The results of the study showed that the EPS was responsible for more than 90% of Pb²⁺ accumulation of the total Pb dissolved. The TEM microphotographs showed that the Pb²⁺ penetrated the cell wall, cell membrane and even into the inner cellular parts of the cell.

Prat et al. (1999) studied the recovery of an aquatic ecosystem after toxic mining waste was dumped into the Guadamar River Basin, Seville, Spain in April 1998. Nine sites were sampled, three of which were unaffected and designated as the control or reference sites. Sampling was carried out over a period of five months, in the form of plankton and particulate material, naturally occurring biofilms and macroinvertebrates obtained from water, riffles (stones), introduced artificial substrates (large tiles), vegetation and sediment. These samples were analysed for water quality and for arsenic (As), cadmium (Cd), antimony (Sb), thallium (Tl), Cu, Pb and Zn concentrations, as these were the most abundant heavy metals in the mine spill. Compared to the reference stations, the concentrations of metals were higher in the polluted sites, with As, Cu, Pb and Zn, being the most abundant. Except for Cd, Cu and Sb, all the other metals in biofilms from the polluted sites were found to be more than 15 times higher than in the reference stations (Prat et al., 1999). In addition, the metal concentrations recorded in the biofilm samples were 5 to 20 times higher than the recorded values in macroinvertebrates (Prat et al., 1999). This study proved that biofilms are effective in the accumulation of metals from the environment.

Conclusions

The conclusions of this study were as follows:

- Al and Fe were recorded at consistently higher concentrations than all the other metals analysed for in water, sediment and biofilm samples
- In both the sediment and biofilm samples, the concentrations of Al and Fe were significantly higher ($p < 0.05$) than Cu, Zn, Pb, Ni, and Mn
- On average, the results generated for water for Al and Fe, were higher than the quality guidelines recommended by DWAF (1996) and the CCME (2001)
- The results for Cu and Zn were higher than the recommended quality guidelines in freshwater sediment (CCME, 2001). No guidelines for Al, Fe, Mn, Pb and Ni in sediment were available.
- The highest metal concentrations were obtained in the sediment and biofilm samples, yet no freshwater guidelines for metals in sediment were available from DWAF and no guidelines for metal concentrations in biofilms were available from either DWAF or the CCME.

Future research

Future research would include the setting-up of parameters or guidelines for acceptable metal concentrations in rivers. Research at the Cape Peninsula University of Technology is

currently being conducted on the Lourens-, the Diep-, the Kuils- and the Plankenbrug Rivers, as well as the Berg River, as discussed in this article.

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