

**ENVIRONMENTAL WATER QUALITY MONITORING
FOR RICHARDS BAY MINERALS:
SMELTER SITE AREA**



Report for 2010/2011

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Executive summary

This document reports on a programme monitoring environmental water quality of surface waters in the vicinity of the Smelter Site of Richards Bay Minerals. Data reported were collected during winter of 2010 and summer of 2011. Aquatic ecological health indices were calculated for each site based on water quality data, habitat quality, macroinvertebrate and diatom taxa sampled.

Indices for each site are presented in Table 1. A provisional overall aquatic ecological health assessment for each of the sites assessed is included. (Note: there is no health index for the habitat score (IHAS) as it was designed to interpret the South African Scoring System (SASS) results and it is included here for that reason). It must be noted that the methods used to provide the subsequent categories are largely based on expert opinion and assessment of the available data. In addition, the boundary values for the categories are based on the default values provided by the ecological Reserve method and require site-specific refinement.

Table 1. A summary of main index score results to provide an overall assessment for each of the sites.

Site	ASPT	IHAS	Water Quality	Diatoms	Overall ecological health assessment
1	Poor	54	Good/Fair	Natural	Good/Fair
7	Fair	65	Fair/Good	Fair	Fair
10	Poor	51	Good/Fair	Fair	Fair
11	Good/Fair	55	Good/Fair	Fair	Fair/Good
12	Fair	61	Good	Natural	Good
13	Good	53	Good/Fair	Fair	Good/Fair
14	Fair	57	Fair/Good	Fair	Fair

Overall, the scores indicate that the Mpisini and Mdibi Rivers were in good condition at the top of their reaches, and that ecological health decreased as one moved downstream. The slight increase in ecological health from Site 10 to Site 11 can possibly be attributed to the receiving waters of the Manzamnyama River between these two sites. It is not possible to determine whether the decrease in ecological health class from Good at Site 12 to Fair at Site 14 is due to the input of water from the Mpisini and Manzamnyama Rivers which drain the smelter complex or from impacts of human habitation occurring between Sites 13 and 14. This decrease is quite small in either case.

The low macroinvertebrate ASPT values recorded at upstream “reference” Sites (1 and 12) are related to poor availability of sampling habitat. Low water levels reduced the marginal vegetation available, gravel/sand/mud has always been limited at these sites and stones-in-current biotope (with which more sensitive macroinvertebrates are usually associated) is absent. High diatom index scores and good water quality results suggest that these sites are indeed in good condition.

As diatoms are less affected by habitat availability they probably provide a better indication of the water quality impacts at sampled sites. The results from the diatom community assessment indicate a clear degradation of water quality between Sites 1 and 7 on the Mpisini River. The smelter complex is situated immediately upstream of site 7, and as there appears to be limited impact from human settlements, it is possible that this water quality impairment may be related to the vicinity of the smelter.

Total taxa collected at Site 11 during the limited sampling in autumn 2010 and spring 2011 and during the comprehensive sampling in winter 2010 and summer 2011 ranged between 12 and 19, with winter being the taxa richest season and summer the taxa poorest (Table 2). The average number of taxa collected during the assessment year was 15.5 and the overall total number of taxa per year was 25. A list of species collected at Site 11 during four sampling trips in different seasons is supplied in Appendix 6.

Table 2. Total taxa per season and average taxa per year collected at Site 11.

Year Month Season	2010 Aug winter	2010 Nov spring	2011 Feb summer	2011 May autumn
Total taxa per season	19	14	12	17
Average taxa per year	15.5			
Total taxa per year	25			

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1 Introduction

The Unilever Centre for Environmental Water Quality, based within the Institute for Water Research at Rhodes University, was appointed by Richards Bay Minerals (RBM) to undertake environmental water quality monitoring of the surface waters in the vicinity of the RBM Smelter Site during winter 2010 and summer 2011.

Richards Bay Minerals is situated in northern KwaZulu-Natal, producing titania slag, pig iron, rutile and zircon through processes of dune mining, mineral separation, smelting and beneficiation. The RBM Smelter Site is adjacent to the KwaBonambi State Forest and is situated within a larger afforested area. The area around the Smelter Site and the Tisand Mineral Lease area is drained by several small streams which flow into the Mdibi River and ultimately into Lake Mzingazi (Figure 1).

There is concern that RBM activities at the Smelter Site may impact the ecological health of the surrounding rivers. Contaminants from the RBM Smelter premises can reach the rivers either directly, via surface water run-off to the rivers (e.g. from pollution incidents, via effluent pipes or rainfall run-off), or indirectly, via groundwater contamination. The natural drainage from the RBM Smelter Site is towards the Mpisini and Manzamnyama Rivers, which drain into the Mdibi River, which subsequently flows into Lake Mzingazi.

The specific tasks identified, to successfully undertake the comprehensive biomonitoring in winter 2010 and summer 2011, are:

1. Undertake aquatic macroinvertebrate biomonitoring at the 7 identified sites.
2. Undertake diatom biomonitoring at the 7 identified sites.
3. Undertake a habitat assessment (IHAS) at the 7 identified sites.
4. Undertake water quality monitoring of the following parameters: nutrients (specifically, Total Inorganic Nitrogen (TIN) and Soluble Reactive Phosphorus (SRP)) and chlorophyll-*a* analysis (of phytoplankton and periphyton), turbidity, electrical conductivity, dissolved oxygen (DO), biological oxygen demand (BOD), water temperature and pH at each of the 7 identified sampling sites.

In addition to the comprehensive biomonitoring at 7 identified sites, limited biomonitoring (SASS only) and basic on-site water quality analysis at Site 11 (confluence of the Mpisini and Manzamnyama Rivers) was undertaken during autumn 2010 and spring 2011.

The specific tasks identified to successfully undertake the limited biomonitoring are:

1. Undertake on-site SASS biomonitoring at Site 11
2. Undertake a habitat assessment (IHAS) at Site 11
3. Measure turbidity, electrical conductivity/TDS, dissolved oxygen, water temperature and pH at Site 11

2 Methods and materials

2.1 Sampling sites

Biomonitoring and water quality sampling was undertaken at seven sites: 1, 7, 10, 11, 12, 13, 14 (Figure 1). This was part of the comprehensive biomonitoring undertaken during winter (August 2010) and summer (February 2011), including macroinvertebrate and diatom biomonitoring, IHAS assessments and an extended water quality analysing programme including field measured parameters (such as pH and EC) and laboratory analysed parameters (such as SRP and BOD). Additionally limited biomonitoring was undertaken at Site 11 during spring (November 2010) and autumn (May 2011), excluding diatom biomonitoring and laboratory analysed parameters. The biotopes sampled at each site are depicted in the individual site summaries section, along with a description of each site (Tables 6-12).

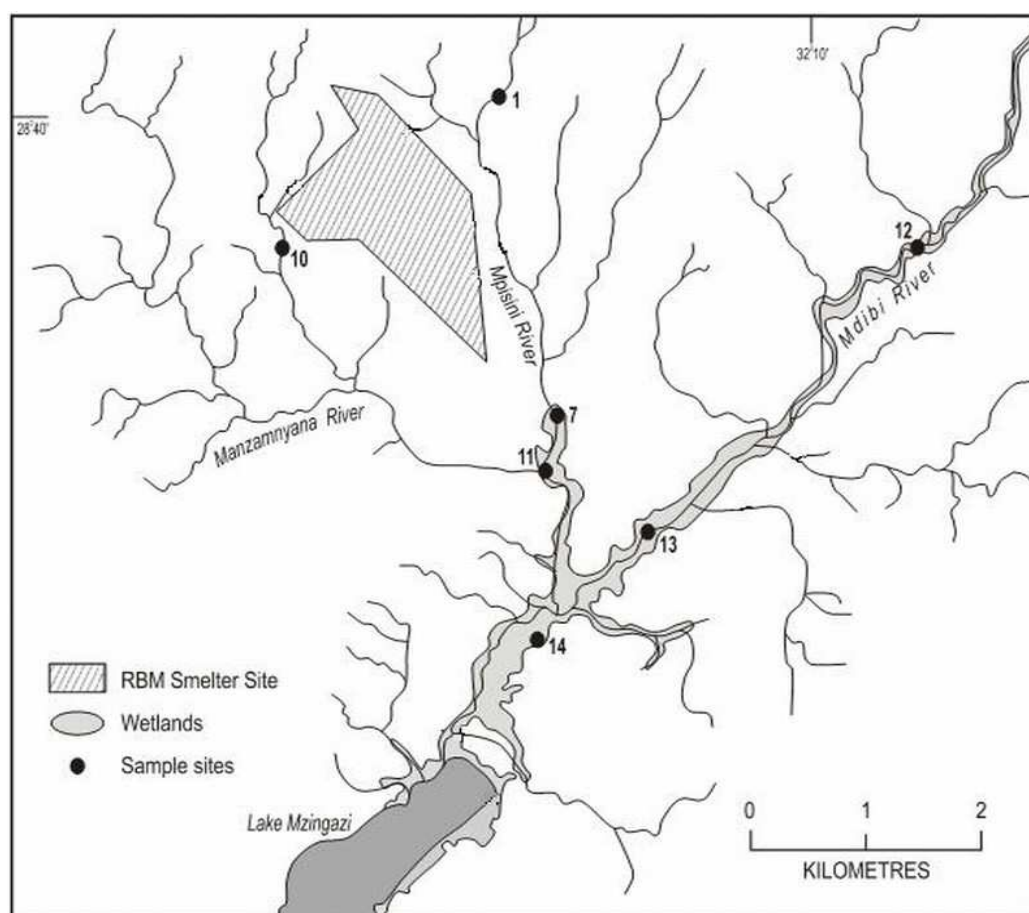


Figure 1. Monitoring points sampled in the Smelter Site area.

2.2 Water quality assessment

Water samples were collected during the winter (August 2010) and summer (February 2011) surveys at each of the biomonitoring sites and kept at 4°C for analysis at the Institute for Water Research, Rhodes University, Grahamstown. The following parameters were measured: NO₂-N, NO₃-N, NH₄-N and PO₄-P (soluble reactive phosphorus - SRP). The nitrogen data were combined to obtain Total Inorganic Nitrogen (TIN) concentrations. Assessing only

dissolved nutrient status (TIN and SRP) can lead to an incorrect conclusion regarding the nutrient enrichment of the water body. Dissolved nutrients are directly available for uptake by plants, consequently during active plant growth periods the concentrations of these nutrients will be a poor indicator of nutrient enrichment. The measurement of algal biomass (periphyton and phytoplankton) using chlorophyll-*a* concentration provides additional information when assessing the level of nutrient enrichment as algae cause most of the problems associated with nutrient enrichment (Palmer et al. 2004). Periphyton and phytoplankton samples were collected following methods described by Holm-Hansen and Riemann (1978) and analysed for chlorophyll-*a* concentrations, as an additional indication of nutrient status of the surface waters. Additional water samples were collected to assess biochemical oxygen demand (BOD). This test determines the amount of dissolved oxygen used by the aerobic microorganisms that decompose organic waste matter in the water. It is therefore used as a measurement of the presence of certain types of organic pollutants in water. In addition, the greater the BOD the smaller the amount of dissolved oxygen available in the river to other organisms. The standard method for a 5-day test (BOD₅) was used (APHA 1992). In addition to the parameters described above, on-site measures of dissolved oxygen (DO), electrical conductivity (EC), temperature and pH (using appropriate hand-held meters) were recorded during all four biomonitoring exercises (winter, spring, summer, and autumn).

Where appropriate, water quality data were interpreted using the default benchmark boundary values for ecological health as provided for in the ecological Reserve methodology for water quality assessments (DWA 2008).

2.3 Biomonitoring

Habitat assessment

A habitat assessment was undertaken at each site, using the Integrated Habitat Assessment System (IHAS; McMillan 1998). IHAS was initially developed for use with SASS4 (i.e. to adjust the SASS4 score). It provides a useful assessment of the habitat available at a site as the diversity of macroinvertebrates can be influenced by availability of biotopes and physical characteristics of the river, and surrounding land-use impacts.

Macroinvertebrate sampling

At each of the sites SASS5 samples were taken from available biotopes and scored accordingly (Dickens and Graham 2002). During the comprehensive biomonitoring a further two samples from each of the biotopes were collected (replicate samples) and all samples were preserved in 80% ethanol once the SASS evaluation was complete. The standard SASS protocol (described in Dickens and Graham (2002) as well as the standard data sheet) was utilised to collect the SASS samples as well as replicate samples. All samples were further enumerated at the UCEWQ-IWR laboratories, providing accurate counts for each of the taxa for data analysis (macroinvertebrate community assessment). For each site the SASS score was divided by the total number of families sampled in order to obtain the Average Score per Taxon (ASPT) (Dickens and Graham 2002). ASPT scores were classified according to default boundary values for ecological Reserve categories as an estimation of ecological health (DWA 2008).

Diatom assessment

Diatom samples were collected from hard substrates (vegetation, wood, brick or rock) on site and fixed in 20% ethanol for transport. If no suitable hard substrate was present, diatoms were sampled from the sediment surface. Samples were prepared for examination using the potassium permanganate and hot hydrochloric acid method recommended by Taylor et al. (2007a). Cleaned frustules were mounted in Pleurax on microscope slides and examined at 1000× magnification using bright field and phase contrast optics. Only whole frustules in valve view were used for identification. One hundred frustules per slide were identified.

Where possible, diatoms were identified to species level. Morphospecies were assigned where identification to species level was not possible and these were maintained throughout the analysis. All diatom counts were converted to proportional abundance before analysis. Abundances were used to calculate IPS (Coste in Cemagref 1982), a diatom-based index of general water quality that has been tested for use in South Africa (Taylor et al. 2007b, 2007c) and has been successfully applied in KwaZulu-Natal (Taylor pers. comm.). The new Biological Diatom Index (BDI-2006, Coste et al. 2009), a general pollution index, was also calculated. The older BDI index on which the new version is based has also been tested and used in South Africa (Taylor et al. 2007b, 2007c). Both indices use a large number of taxa in inferring water quality. IPS and BDI-2006 scores were rescaled to give a maximum of 20 as per common convention. Water quality classes were assigned to IPS index data after Eloranta and Soininen (2002) and to BDI-2006 index data after Prygiel and Coste (2000) (Table 3).

Table 3. Water quality classes for the IPS index (Eloranta and Soininen 2002) and the BDI-2006 index (Prygiel and Coste 2000).

Class	IPS value	BDI-2006 value
High	>17	BDI≥17
Good	15-17	17>BDI≥13
Moderate	12-15	13>BDI≥9
Poor	9-12	9>BDI≥5
Bad	<9	BDI<5

Previous reports (Muller et al. 2007, Gordon et al. 2008) used an index based on expert opinion as doubt existed as to the applicability of indices derived in Europe in a region where tropical taxa might be encountered. However, as the IPS index has been successfully applied in the region (Taylor, pers. comm.) and the BDI-2006 index contains a number of tropical taxa (Coste et al. 2009), and as the majority of taxa encountered in previous surveys are included in the two diatom indices, these indices will be used for sample classification in this report. For continuity with previous reports, sample classifications based on expert opinion are also derived for each sample. The sample classifications based on the expert opinion approach are derived by using environmental preferences of common taxa as presented by Taylor et al. (2007d) and Van Dam et al. (1994) to infer the ecological health of the site. Using this information, samples are scored according to the scheme presented in Table 4. Sites are ranked according to scores assigned according to the above scheme. Where sites fall between classes, intermediate scores are assigned e.g. 4 represents a classification of Good, and 2 represents a classification of Poor.

Only taxa that were well represented in each sample were used to infer water quality class, as these will best indicate the prevailing and recent water quality. For the purposes of this analysis, dominant taxa are the one taxon with the greatest abundance in the sample. Where other taxa have abundances not less than 10% less than the dominant taxon, they are classed as co-dominant. Other taxa that are less common than the dominant taxa and that make up 10% or more of the sample are classed as subdominant and are used to infer water quality. Taxa present in lower quantities are only used in this analysis where information from dominant and subdominant taxa is insufficient for site classification.

Table 4. Classification system for expert opinion-based diatom index

Class	Environmental preferences of common taxa	Score
High	Samples where all or most taxa found are characteristic of unpolluted oligotrophic to mesotrophic water with low to moderate levels of electrolytes. Dominant taxa must be typical of these conditions.	5
Good		4
Moderate	Dominant taxa not consistently indicative of clean conditions, and the sample has taxa typical of clean and stressed condition.	3
Poor		2
Bad	Most taxa present are tolerant of at least moderate levels of pollution, or typical of eutrophic or osmotically stressed conditions.	1

For the overall diatom classification of sites, the expert opinion-based classification described above was combined with IPS and BDI-2006 using weight of evidence to derive an overall sample classification. This overall diatom classification was adjusted to the ecological reserve categories in order to provide an overall assessment of aquatic ecological health (Table 5).

Table 5. Alignment of diatom classification system and ecological Reserve categories

Diatom classification	Ecological Reserve categories
High	Natural
Good	Good
Moderate	Fair
Poor	Poor
Bad	

Statistical analysis

Statistical analyses of macroinvertebrate community structure was undertaken using non-metric multi-dimensional scaling (NMDS) provided for in the PRIMER V5 programme (Clarke and Warwick 2001). The NMDS ordination plot represents the similarity of abundances of family level taxa between samples. Statistical analysis was conducted using the analysis-of-similarity test (also provided for by the PRIMER v5 programme). In addition to the significance value, the Global R value indicates the degree to which the samples are similar or dissimilar. An R value of 1 indicates complete separation of groups, whereas an R value near 0 implies little or no segregation.

Analysis of variance and post-hoc testing of diatom scores was undertaken using R 2.11.0 with base and stats packages (R Development Core Team 2010). Calculation of alpha diversities and ordination using non-metric multidimensional scaling were undertaken using the package vegan 1.17-2 (Oksanen et al., 2010). Hypotheses relating to the differences in diatom community structure between sites and seasons were explored using the function adonis, which undertakes non-parametric multivariate analyses of variance (after Anderson 2001). Square root transformation was used on all taxa and all taxa were used in ordinations and community analysis.

3 Results and discussion of comprehensive biomonitoring

In the sections below, the water quality, habitat and biological monitoring data collected are reported and discussed. Individual site summaries detailing specific site information observed by the samplers, water quality parameters measured and biological monitoring results are presented in Tables 6-12. Raw data are presented in Appendices 1-6.

3.1 Water quality assessment

Water temperatures recorded at sampling sites showed little variation (Figure 2A), with summer being the warmer season at all sites (Figure 2B).

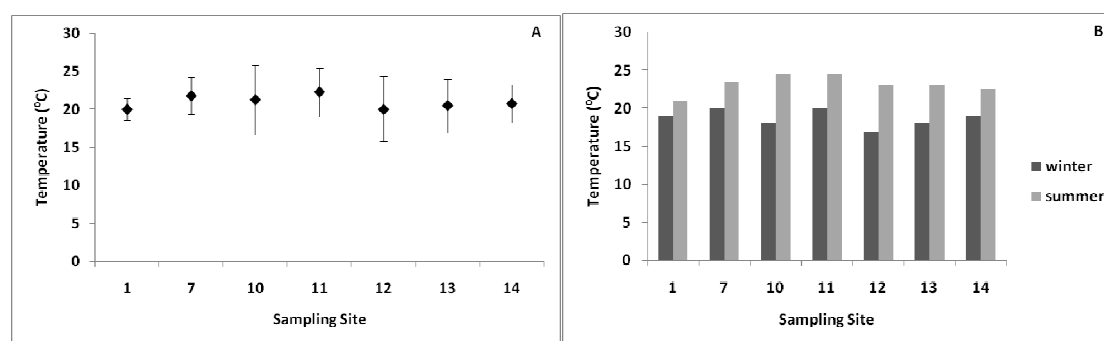


Figure 2A-B. A: Mean (with standard deviation) temperature values measured at sampling sites over two seasons. B: Present seasonal values measured at each site.

Due to a faulty pH meter, pH measurements during summer 2011 could not be undertaken. Seeing that seasonal differences have been negligible during recent monitoring programmes (Gordon et al. 2008, Gordon et al. 2010) measured pH levels during the winter survey were considered representative for both seasons. Differences in measured pH were small, with the uppermost sites on each river being slightly more acidic (Figure 3A). All sites, however, were classed either Natural or Good. Electrical conductivity at all uppermost sites were classified as being Good, with average EC values at remaining sites being categorized as Fair (Figure 3B). Seasonal variations in EC levels were negligible (Figure 3D). Generally, river water at downstream sites had higher EC values compared with upstream sites. The higher EC levels at Site 7 appears to be related to the proximity of the Smelter Site as no other cause for the increase could be identified. Remaining sites downstream of the Smelter are surrounded by human settlements and thus it is difficult to determine relative contributions of these two land uses to increased EC.

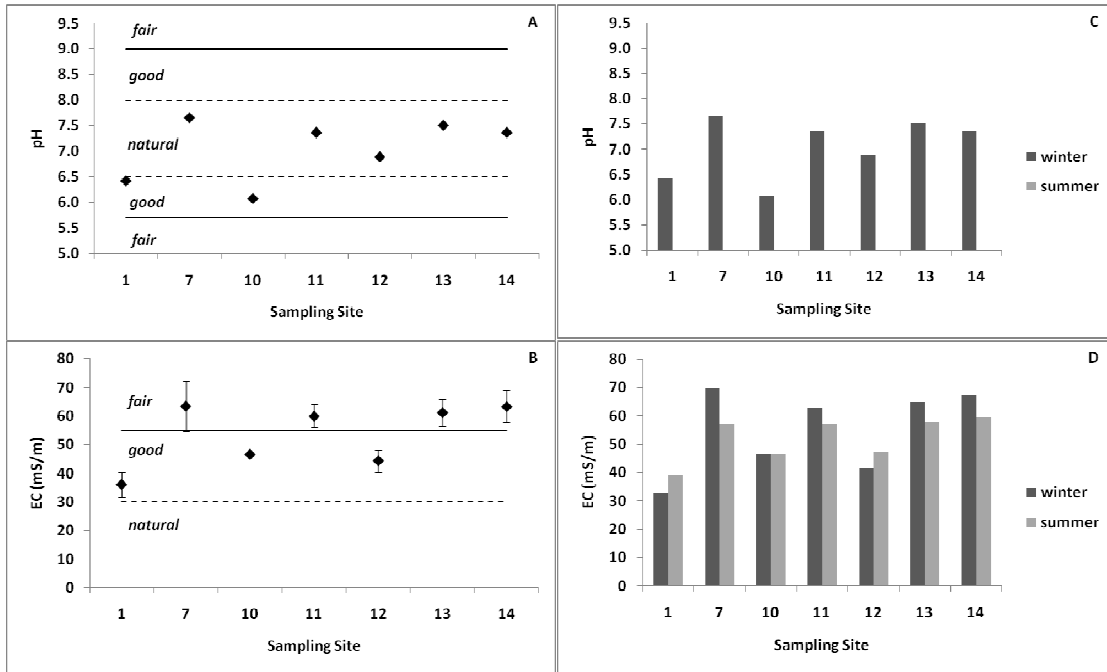


Figure 3A-D. A-B: Mean (with standard deviation) pH and electrical conductivity (EC) values measured at sampling sites over two seasons. Default ecological categories based on ecological Reserve determination methodologies are superimposed on the graphs. C-D: Present seasonal values measured at each site.

Average DO was considerably lower at Site 10, falling within the Poor ecological category (Figure 4A), whereas all other sites were classified as Fair. DO levels at all uppermost sites (Sites 1, 10, and 12) were lower than at downstream sites and in general higher in summer than in winter (Figure 4B). The low DO measured at Site 10 could possibly be attributed to the large proportion of ground water feeding the wetland lake immediately upstream and limited surface flow. The values for the 5-day biological oxygen demand (BOD) test were all below the detection limit of 2mg/L.

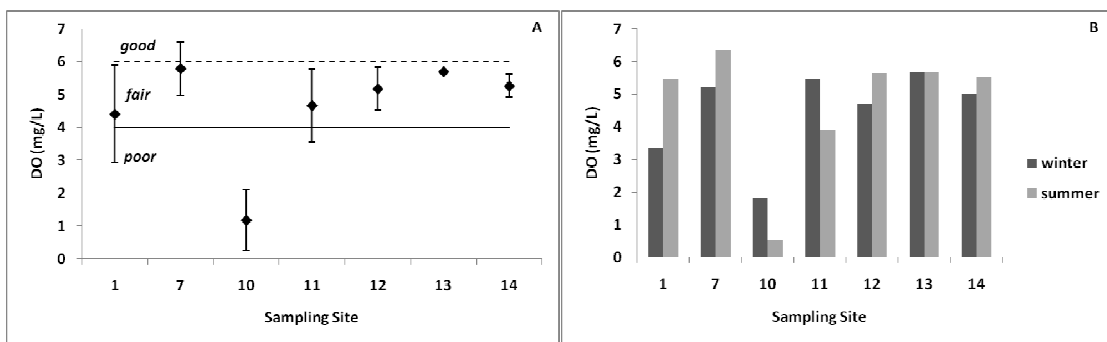


Figure 4A-B. A: Mean (with standard deviation) DO (dissolved oxygen) values measured at sampling sites over two seasons. Default ecological categories based on ecological Reserve determination methodologies are superimposed on DO graph. B: Present seasonal values measured at each site.

Total inorganic nitrogen (TIN) was highest at the uppermost site on the Manzamnyama River (Site 10) and at the reference site (Site 1) in winter (Figure 5A), both of which were classified as Fair/Good. TIN levels at the uppermost sites (Sites 1, 10, and 12) were higher than at downstream sites during the winter survey. TIN levels in summer were below the detection limit at all sites (Figure 5C). Soluble reactive phosphorus (SRP) concentrations were mostly classed as Good except at Sites 7, 11 and 14 (Fair category), which are the most downstream sites assessed (Figure 5B). SRP levels were increasing from the uppermost sites to downstream sites and decreasing from the upstream Site 7 to the downstream Site 14, which lead to the conclusion, that the source of SRP would be situated between Site 1 and Site 7 on the Mpisini River. SRP levels were higher in summer at Sites 7 and 11 and lower in summer than in winter at Site 14 (Figure 5D).

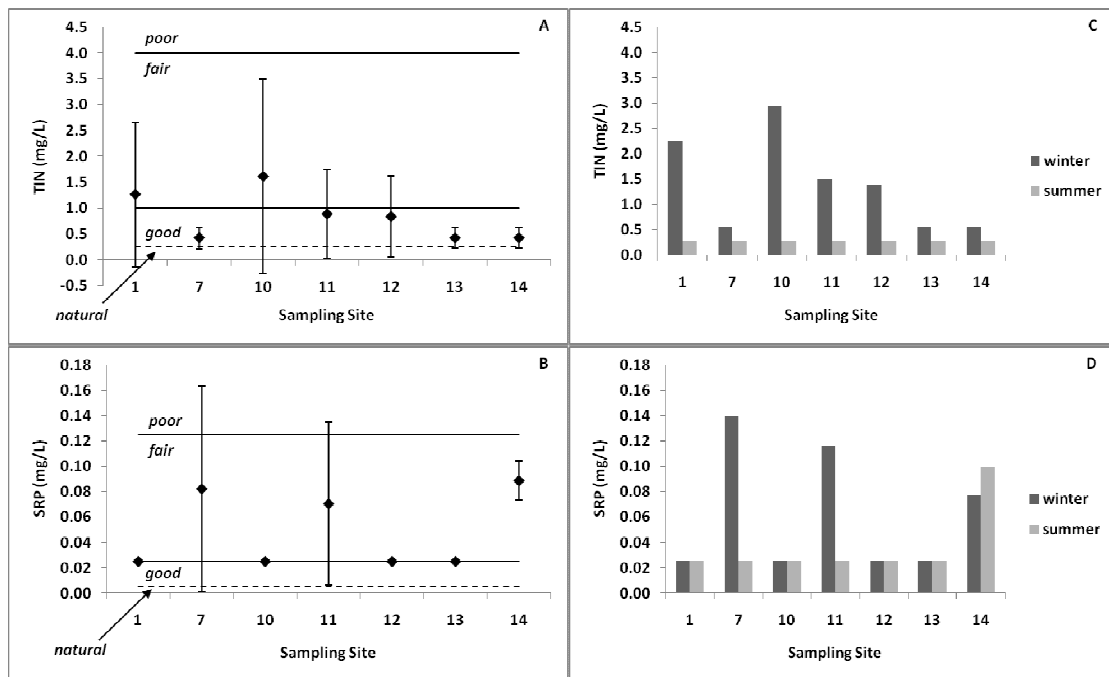


Figure 5A-D. A-B: Mean (with standard deviation) TIN (total inorganic nitrogen) and SRP (soluble reactive phosphorus) values measured at sampling sites over two seasons. Default ecological categories based on ecological Reserve determination methodologies are superimposed on the graphs. C-D: Present seasonal values measured at each site.

Phytoplankton chlorophyll-*a* concentrations at all sites were classed as Natural (Figure 6A), with no significant seasonal variations (Figure 6C). Vegetation previously submerged and used to sample periphyton at Site 14 was exposed and dry during summer 2011, so no samples could be collected at this site. Periphyton chlorophyll-*a* concentrations were lower at all upstream sites than at downstream sites. Values were measured above 450mg/cm² at Site 14 in winter 2010, which will be observed for trends in future (Figure 6B). Large seasonal variations were observed at most sites (Figure 6D).

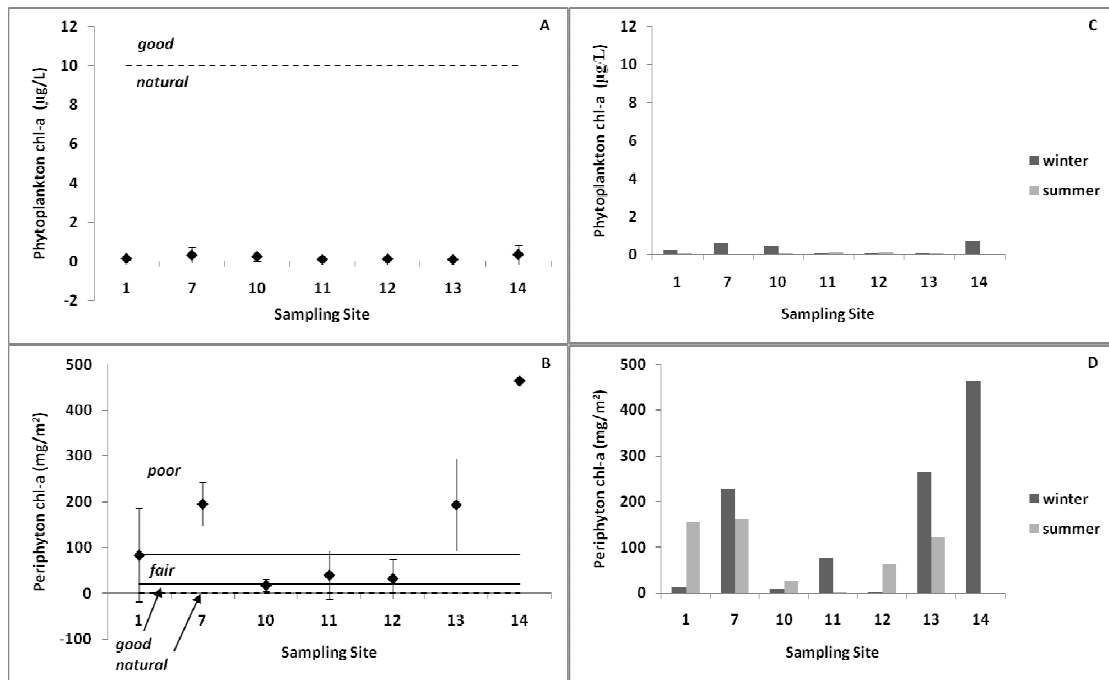


Figure 6A-D. A-B: Mean (with standard deviation) TIN (total inorganic nitrogen) and SRP (soluble reactive phosphorus) values measured at sampling sites over two seasons. Default ecological categories based on ecological Reserve determination methodologies are superimposed on the graphs. C-D: Present seasonal values measured at each site.

3.2 Habitat assessment

Overall, IHAS scores were low due to the absence of a stones biotope at all sampling sites (Figure 7A). Seasonal variability was negligible (Figure 7B). Further reasons for low IHAS scores include Site 10 being characterized by a pool with slow moving water which is regularly disturbed by cattle, Sites 1, 11 and 12 being badly affected by low flows which reduced available vegetation sampling habitat and Site 13 having reduced gravel/sand/mud (GSM) biotope.

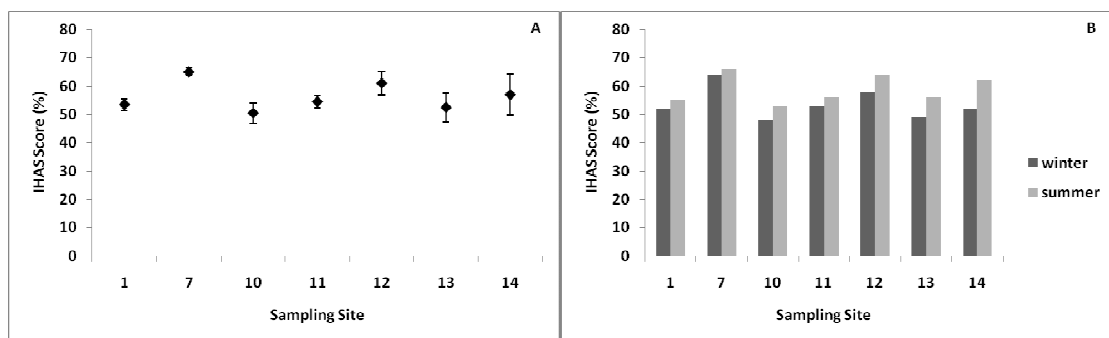


Figure 7A-B. A: Mean Integrated Habitat Assessment System (IHAS) score (with standard deviation) per sampling site. B: Seasonal differences in IHAS recorded at each site.

3.3 Macro-invertebrate assessment

3.3.1 SASS assessment

At each site, results from vegetation and GSM biotopes were combined to provide overall SASS scores, number of families and ASPT values (Figure 8A-C). SASS scores and number of taxa were usually lower at upstream sites compared to downstream sites except at Site 13 which was lower than Site 12 (Figure 8A-B) possibly reflecting impact of surrounding residential land use. SASS scores and number of taxa were considerably higher in winter than in summer at Sites 10, 11, and 14 (Figure 8D).

ASPT values were, however, more consistent seasonally (Figure 8F). The ASPT score is generally considered to be the least variable of the SASS assessment scores and thus preferred when assessing river health. ASPT scores were classed as Poor at Sites 1 and 10 (uppermost sites), Fair at Sites 7, 12, and 14, on the Good/Fair boundary at Site 11 and Good at Site 13 (Figure 8C).

Poor habitat due to low water levels is the most likely cause for the low SASS and ASPT scores at Sites 1 and 10. Additionally, Site 10 has naturally low DO values, which will impact the ability of organisms to survive at this site. Along the length of the Mdibi ASPT values increased from Fair at Site 12 (reference site) to Good at Site 13. However, after input of the combined waters from the Manzanmyama and Mpisini Rivers, the ASPT class drops to a Fair class, as observed in previous sampling occasions. However, the effect of the water from the Manzanmyama and Mpisini Rivers on the ASPT score cannot be distinguished from the potential effects caused by dense human settlements upstream of Site 14. It should be noted that the interpretation of the ASPT score as an ecological health class was undertaken using the benchmark boundary values, and these default values may not be appropriate for rivers in the Natal Coastal Plain Ecoregion. Site-specific refinement of benchmark boundary values is warranted, and is currently being undertaken through Technology and Human Resources for Industry Programme (THRIP) funding from the National Research Foundation (NRF).

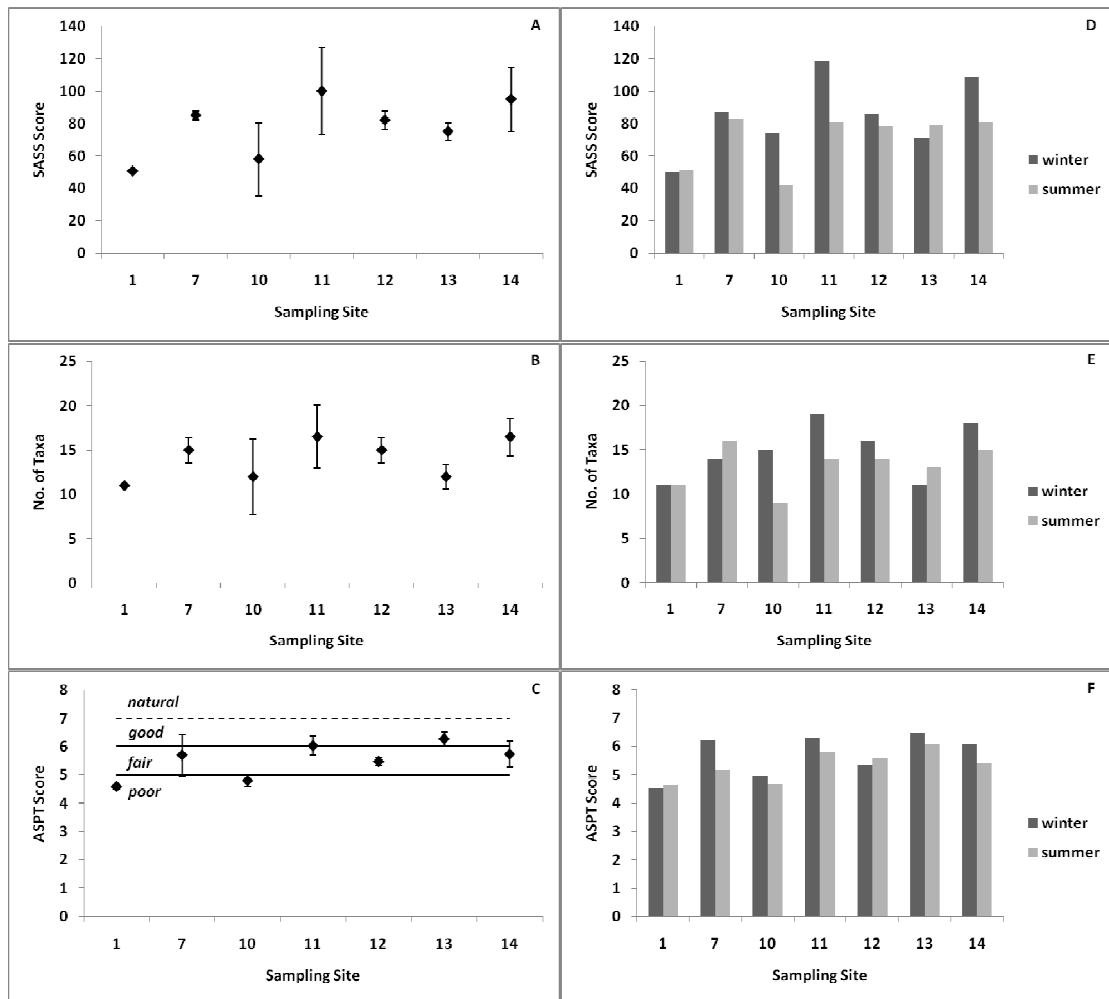


Figure 8A-F. A-C: Mean (with standard deviation) South African Scoring System (SASS) scores, number of families/taxa sampled and Average Score Per Taxon (ASPT) values determined for sites over two seasons. Default ecological categories for ASPT scores based on ecological Reserve determination methodologies are superimposed on graph C. D-F: Seasonal variation in SASS, number of taxa and ASPT values at each site.

3.3.2 Macroinvertebrate community assessment

An analysis of similarity of enumerated family-level macroinvertebrate data collected during winter 2010 and summer 2011 showed that all sites were significantly different from one another. The NMDS plot showed that Site 10 was considerably different from the remaining sites, while Site 11 was to a lesser degree different from Sites 1, 12, 13 and 14 (Figure 9).

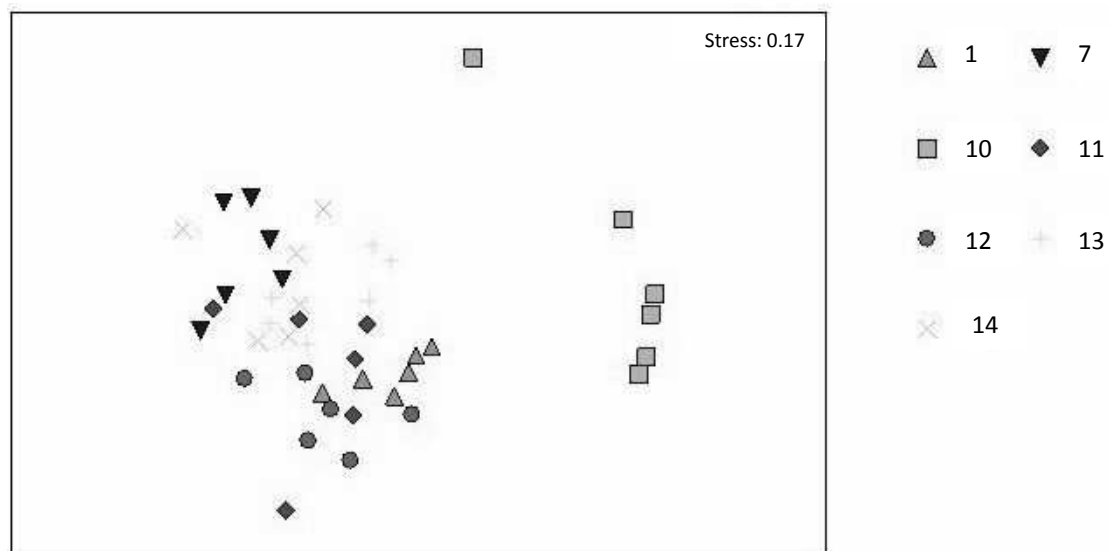


Figure 9. Non-metric multi-dimensional scaling plot of enumerated macro-invertebrate data, analysed according to location sampled.

3.4 Diatom assessment

3.4.1 Diatom indices

Mean index values for each site are presented in Figure 10. Raw data underlying these plots are presented in Appendix 1. General trends between sites are largely the same across all indices, although inter-site variation and the range across scores are greater for the score based on expert opinion.

In general, Site 12, upstream in the Mdibi River, had the best water quality according to diatom-based metrics (Figure 10). Site 1, upstream in the Mpisini, was also rated as fairly clean (being in a good to high class) according to the IPS index and classification based on expert opinion. The discrepancy between expert opinion ratings and IPS and/or BDI-2006 scores are a result of the predominance of unusual or unknown taxa in upstream sites. Taxa such as these are not used when deriving IPS (ecological information on genera does contribute to this score) or BDI-2006 values, although they contribute to expert opinion ratings (see below). For example, in the sample from Site 1 collected in August 2010, all the dominant and subdominant taxa are unusual or unknown, and, as a result, 84% of the diatoms in the sample do not contribute to IPS or BDI-2006 scores. IPS and BDI-2006 scores are as a result based on taxa that made up, at most, 4% of the diatoms in this sample.

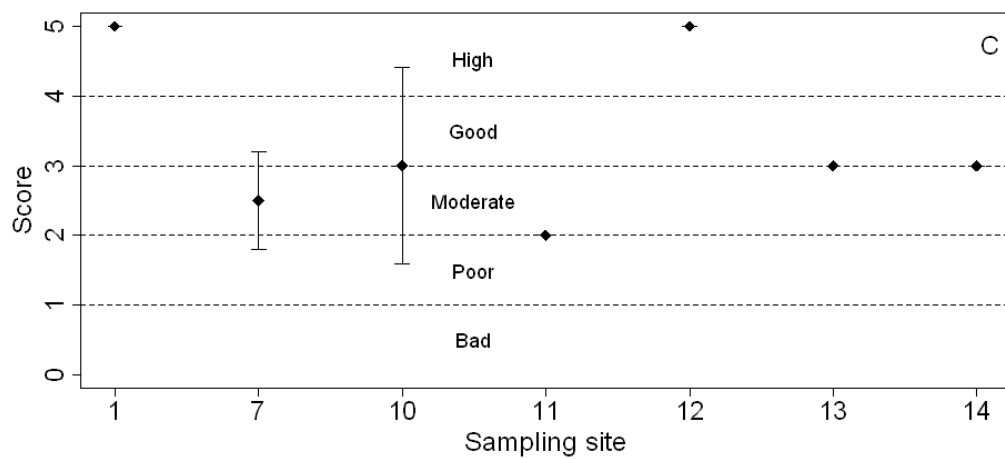
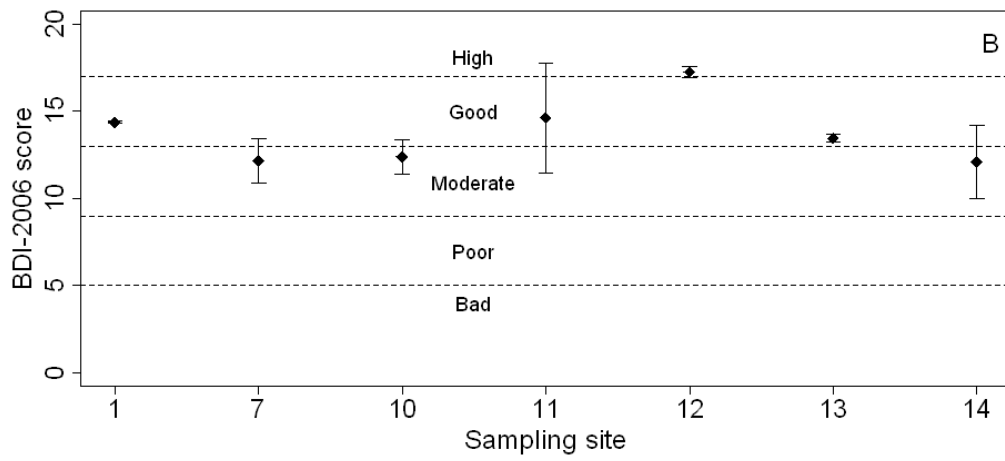
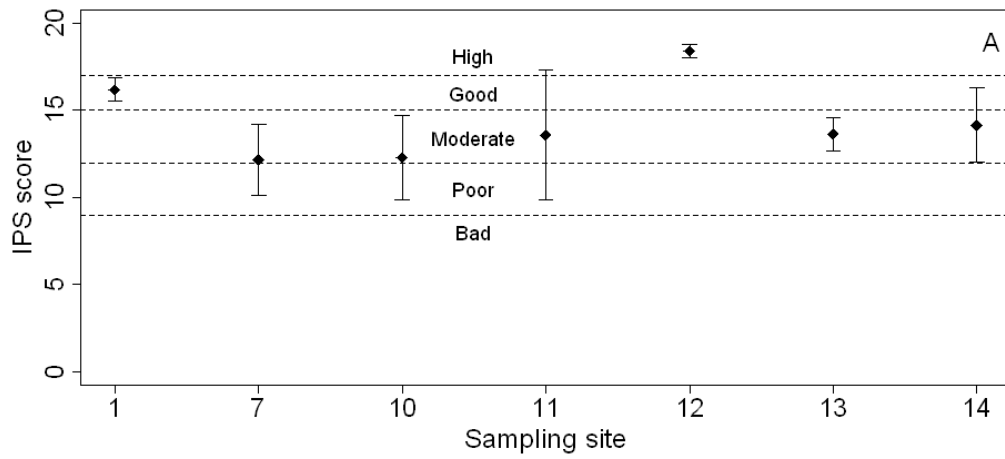


Figure 10A-C. Mean (with standard deviation) scores of the three diatom indices used in this study. A: IPS index; B: BDI-2006 index; C: Score based on expert opinion.

The occurrence of rare, unusual or unknown taxa in samples from sites with lower water quality is of minor importance as taxa from these samples tend to be largely cosmopolitan and rare or unusual taxa and contribute to a far lesser extent to diatom community structure. However, the presence of taxa not used in IPS or BDI-2006 index derivation still plays some role (e.g. *Achnanthes oblongella*, making up 21% of the sample from Site 11 during February 2011, and 9% of the Site 11 sample from August 2010, is not used in the calculation of the BDI-2006 index).

Consistent inter-site differences in IPS and BDI-2006 scores over the period sampled were not detected during statistical analysis (IPS $p=0.212$; BDI-2006 $p=0.153$). This result was unexpected, but appears to be a result of inflated error variance owing to relatively large changes with time between samples from some sites. These changes do not indicate a consistent seasonal pattern, however, as season was less significant than site as a driver of IPS and BDI-2006 index scores (IPS $p=0.999$; BDI-2006 $p=0.876$).

Differences between sites in the index based on expert opinion were statistically significant ($p=0.017$), although, again, seasonal changes were not ($p=0.689$). Post-hoc testing indicated that Site 11 was significantly worse than Sites 1 and 12 ($p=0.029$ for both). Although not significant at the 5% level, Site 7 was also worse than Sites 1 and 12 overall ($p=0.064$ for both). Results indicating that Site 7 has low inferred water quality are consistent with previous years (Gordon et al. 2008, 2010). However, the overall site diatom index values for Site 11 are lower than in previous years. This is a largely function of the particularly low water quality inferred from the sample collected from Site 11 during August 2010. The sample from February 2011 indicated good water quality according to the IPS and BDI-2006 indices, but poor water quality according to the index based on expert opinion. This disagreement is a function of differing interpretations of the water quality indicated by the dominant taxon, *Gomphonema pumilum*.

4.3.2 Description of diatoms sampled at each site

Site 1 (Upper Mpisini)

August 2010

This sample contained 13 taxa and had a Shannon diversity of 1.66. The site's diatom flora was dominated by *Achnanthes pulviscula*, with two unknown species of *Brachysira* present in significant numbers. No knowledge on the specific ecological requirements of these taxa is available. However, these taxa have been found to be associated with good quality water in the Smelter region (Gordon et al. 2008, 2010) and are commonly dominant in upstream reference sites. In addition, *Brachysira* species typically are found in oligotrophic, electrolyte-poor water, and many prefer a somewhat acid pH. Of the remaining taxa found to make up more than 1% of the diatom population, most are typical of clean or oligotrophic water.

February 2011

This sample contained 15 taxa and had a Shannon diversity of 1.62. The same two unknown *Brachysira* species that were common in August 2010 at this site are co-dominant in this sample. For the reasons given above, these can be taken as indicative of clean water. Most of the remaining taxa are typical of good water conditions.

Site 7 (Lower Mpisini)

August 2010

This sample contained 26 taxa and had a Shannon diversity of 2.92. *Gomphonema parvulum*, *Navicula symmetrica* and *Cymbella tumida* are codominant in this sample, although the abundances of these taxa are not particularly high. *Gomphonema parvulum* is a cosmopolitan taxon that is highly pollution tolerant, and *Navicula symmetrica* is typical of eutrophic, electrolyte-rich waters and is highly pollution tolerant. *Cymbella tumida*, on the other hand, is typical of oligo- to mesotrophic conditions with moderate electrolyte levels. Other taxa present are typical of waters with at least moderate electrolyte levels, with some taxa typical of meso- to eutrophic water.

February 2011

This sample contained 18 taxa and had a Shannon diversity of 2.23. *Nitzschia fonticola*, *Nitzschia perminuta* and *Cocconeis placentula* were codominant. *Nitzschia fonticola*, though typical of water with moderate to high electrolyte levels, is not typically found in water that is more than slightly polluted. *Nitzschia perminuta* is likewise relatively pollution-intolerant but is found in fresh-brackish water. *Cocconeis placentula* however is typical of meso-eutrophic waters. The remaining taxa comprise a mix of those tolerant to high levels of nutrients and/or electrolytes, and some pollution intolerant taxa.

Site 10 (Upper Manzamnyama)

August 2010

This sample contained 15 taxa and had a Shannon diversity of 1.31. The sample was heavily dominated by an unknown species of *Brachysira* also common at Site 1. Most remaining taxa are tolerant of moderate electrolyte levels and varying degrees and types of pollution.

February 2011

This sample contained 30 taxa and had a Shannon diversity of 3.03. Along with the major increase in number of taxa, a distinct change in community structure was noted. *Navicula erifuga* was codominant with *Nitzschia palea* in this sample. The former is highly pollution tolerant, and typical of eutrophic conditions with high electrolyte levels. The latter is highly cosmopolitan and is also typical of eutrophic water with moderate to high levels of electrolytes. With few exceptions, the remaining taxa are indicative of high nutrient and/or electrolyte levels.

Site 11 (Confluence)

August 2010

This sample contained 26 taxa and had a Shannon diversity of 2.77. *Bacillaria paradoxa* was dominant in this sample, with *Navicula symmetrica* subdominant. *Bacillaria paradoxa* is a cosmopolitan taxon typical of electrolyte-rich or brackish water. *Navicula symmetrica* is highly pollution tolerant and common in electrolyte-rich and eutrophic water. Other taxa present in larger abundances include taxa common in electrolyte-rich and/or eutrophic water, as well as taxa better known from oligotrophic and electrolyte-poor conditions.

February 2011

This sample contained 13 taxa and had a Shannon diversity of 1.60. *Gomphonema pumilum* was dominant in this sample, with *Achnanthes oblongella* subdominant. *Gomphonema pumilum* is typical of meso- to eutrophic conditions with moderate electrolyte levels. *Achnanthes oblongella*, on the other hand, is usually found in circumneutral, oligotrophic, electrolyte-poor water. Other taxa present in small quantities are generally typical of polluted, especially eutrophic, waters.

Site 12 (Upper Mdibi)

August 2010

This sample contained only 6 taxa and had a Shannon diversity of 0.95. *Achnanthes oblongella* was dominant in the sample, with *Achnanthes pulviscula* subdominant. Dominant and subdominant taxa, as well as remaining taxa, are all indicative of good water quality with low levels of electrolytes and nutrients.

February 2011

This sample contained 10 taxa and had a Shannon diversity of 1.75. An unknown species of *Eunotia* was dominant, and one of the unknown *Brachysira* species found at Site 1 as well as *Eunotia bilunaris* were subdominant. As noted above, *Brachysira* species typically are found in oligotrophic, electrolyte-poor and often acidic water. *Eunotia* species too are generally typical of oligotrophic, electrolyte-poor and usually acidic water. *Eunotia bilunaris* is no exception as it is typical of electrolyte-poor, acidic water. Remaining taxa are generally typical of oligotrophic conditions, with an acidic to neutral pH, and a range of electrolyte concentrations.

Site 13 (Middle Mdibi)

August 2010

This sample contained 13 taxa and had a Shannon diversity of 1.50. *Achnantheidium saprophilum* was dominant in this sample, and another *Achnantheidium* species, *Achnantheidium minutissimum*, was subdominant. The former is typical of organically enriched or eutrophic water, while the latter is an indicator of well-oxygenated, clean water. Remaining taxa are for the most part typical of circumneutral, oligotrophic, electrolyte-poor water, however, some more pollution tolerant taxa are present.

February 2011

This sample contained 18 taxa and had a Shannon diversity of 2.45. *Cocconeis placentula* dominated this sample, while *Achnanthes oblongella* and *Gomphonema angustatum* were subdominant. *Cocconeis placentula* is typical of meso- to eutrophic waters. *Achnanthes oblongella* is associated with circumneutral, oligotrophic, electrolyte-poor water, while *Gomphonema angustatum* may occur over a range of acidity and electrolyte levels, but is only common in oligotrophic water. Remaining taxa contain several associated with clean water, but also a number of taxa common in eutrophic conditions.

Site 14 (Lower Mdibi)

August 2010

This sample contained 16 taxa and had a Shannon diversity of 2.22. *Achnantheidium saprophilum* was dominant in the sample, with *Gomphonema pumilum* var. *rigidum* and *Achnantheidium minutissimum* subdominant. As in the August 2010 sample from Site 13, this combines two different *Achnantheidium* species, one associated with eutrophic water and one with clean water. *Gomphonema pumilum* var. *rigidum* is commonly associated with meso- to eutrophic conditions and moderate electrolyte levels, but is not considered pollution-tolerant. Many of the remaining taxa would be considered indicative of clean water; however, a few are normally associated with high nutrient and/or electrolyte levels.

February 2011

This sample contained 12 taxa and had a Shannon diversity of 1.91. The great majority of taxa encountered were species of *Gomphonema*. *Gomphonema extentum* was dominant in this sample, and *Gomphonema lagenula*, *Gomphonema angustatum* and *Gomphonema parvulum* were all subdominant. *Gomphonema extentum* is often associated with moderate levels of pollution. Little is known of the ecology of *Gomphonema lagenula*. *Gomphonema angustatum* is cosmopolitan, but common only in oligotrophic water, and tolerant of a range of acidity and electrolyte levels. *Gomphonema parvulum* is found in range of conditions and is extremely pollution tolerant. Remaining taxa include those commonly associated with clean water as well as polluted conditions.

4.3.3 Diatom community analysis

The ordination of samples based on diatom abundance is presented in Figure 11. Effects of site and season on diatom community structure were statistically significant ($p=0.003$ and $p=0.044$ respectively). In general, sites with higher inferred water quality are to the left of the plot, while sites with lower inferred water quality are to the right. This is not absolute, however, as some sites with similar inferred water quality (e.g. Site 10 winter sample) have a clearly different community structure and are fairly widely dispersed along NMDS axis 1 from others with similar water quality (Site 13 summer sample and Site 14 winter sample).

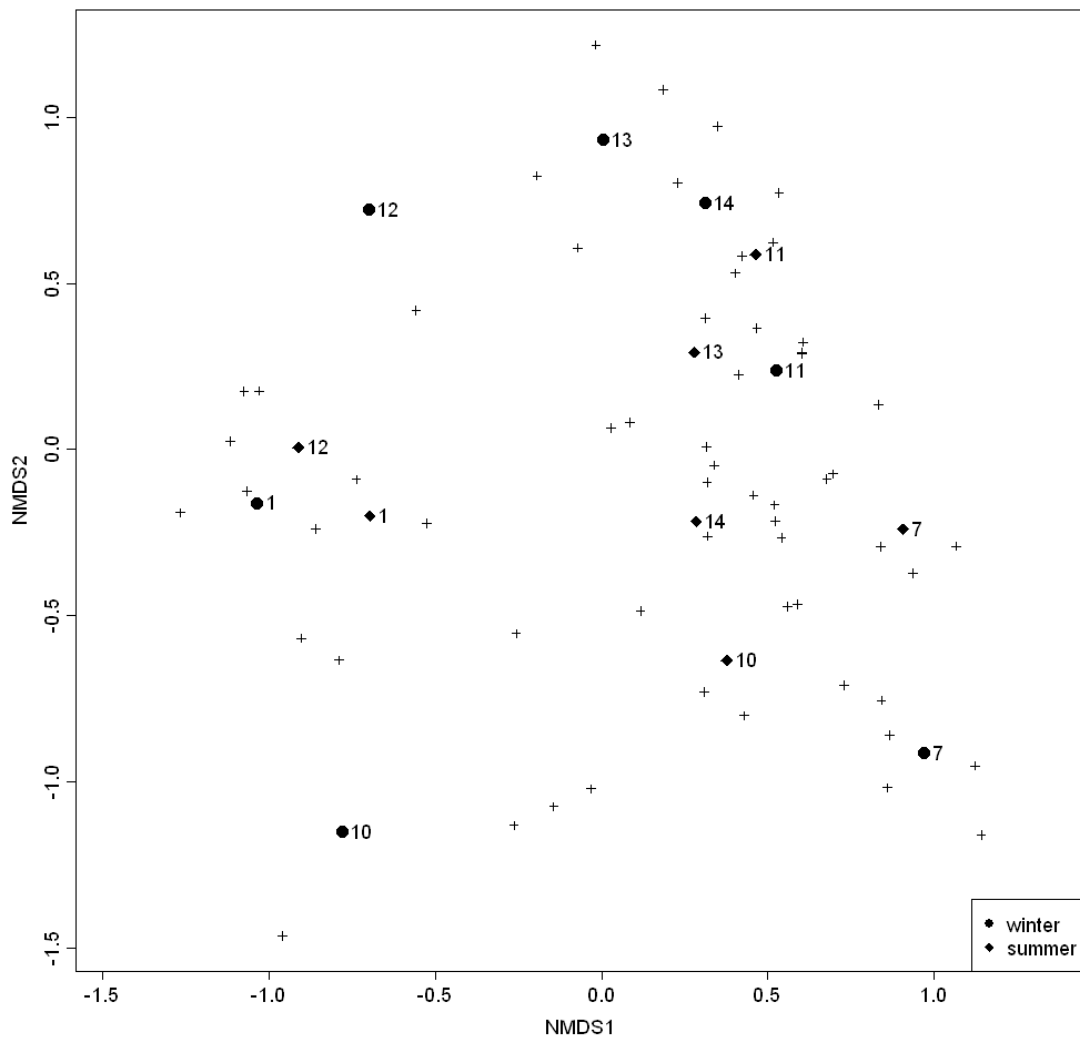


Figure 11. NMDS ordination plot of Bray-Curtis distances between all samples assessed, based on square-root transformed diatom abundances (stress: 18.4). Sites are labeled, species indicated by +.

Downstream Sites 11, 13 and 14 are grouped in top right quadrant of Figure 11, indicating that they have similar diatom community structure. Sites 1 and 7 in the Mpisini are widely separated in Figure 11, indicating a major change in community structure along the relatively short stretch of river separating the two sites. Finally, Site 10, the upstream reference site in the Manzanmyama River, has a distinctly different diatom community to other upstream reference sites. However, upstream reference Sites 1 and 12 are fairly similar in terms of diatom community structure, despite being located in different rivers (Mpisini and Mdibi respectively). This suggests that the water quality in Mdibi and Mpisini Rivers may differ from the Manzanmyama. Alternatively, if a site further upstream than Site 10 on the Manzanmyama is available, it should be checked to determine whether it might be a more appropriate as a reference site.

A simple seasonal trend is less apparent in Figure 11, though variation between sites in diatom community structure appears greater in winter than in summer. Certain sites showed large seasonal changes in community composition (most notably at Site 10). However, others, most notably Site 1, varied little in diatom community structure between seasons.

3.4.4 Overall diatom assessment

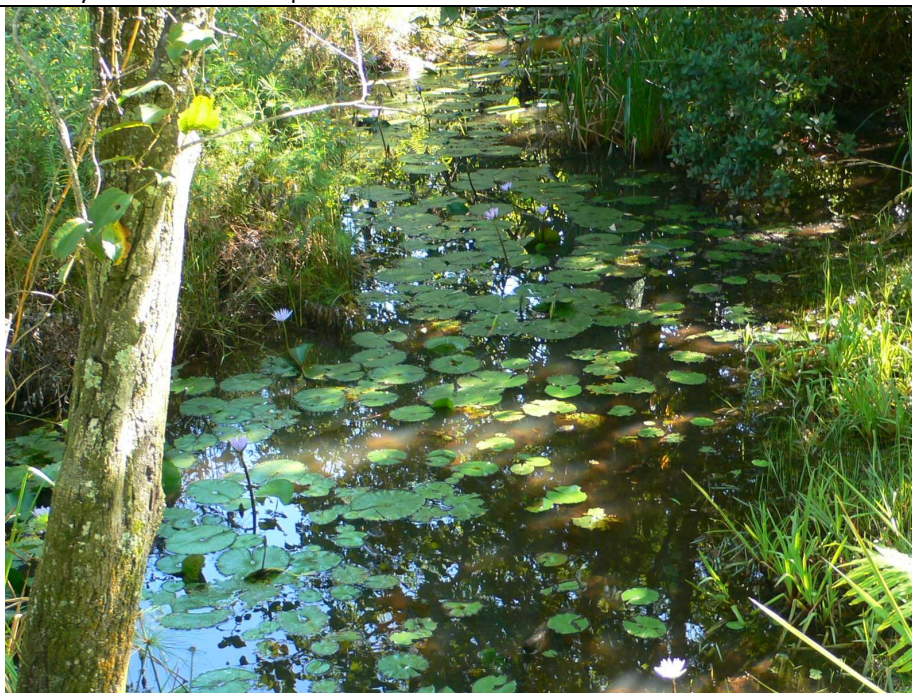
Overall weight-of-evidence site classifications for Sites 10, 13 and 14 decreased from good to moderate compared to results from the previous year (Gordon et al. 2010). The reason for this is not known, however, but low water levels in the current study may have decreased dilution rates in the rivers. The score from Site 11 decreased over this period as well but this did not result in a changed classification. Scores from upstream Sites 10 and 12 were not affected in this way.

The drop in inferred water quality between Site 1 and Site 7 has been noted in previous reports (Gordon et al. 2008, 2010), as has the change in diatom community structure between these sites. This change has been associated with increased pH and conductivity in the past, and, in the current study, Site 7 is seen to have increased pH, conductivity and, in winter, elevated levels of phosphate (as SRP) compared to Site 1. The Mpisini River between these sites is approximately 3 km long, and runs adjacent to the RBM Smelter Site. Several drains from the Smelter Site that lead into the Mpisini are found in this stretch. In the past, flow from these drains has been found to be rare (Muller and Gordon 2005). In addition, two small ephemeral streams drain forested land to the east and enter the Mpisini between Sites 1 and 7. Smelter Site runoff, as surface flow or possibly via the groundwater, and/or input from these streams, is the most probable cause of the decrease in water quality along the Mpisini between Site 1 and Site 7. Further and more detailed monitoring of changes in water quality between these sites is recommended to identify the cause of the change.

In the past, water quality as inferred from diatom community structure has been found to improve from Site 7 on the Mpisini to Site 11, at the confluence of the Mpisini and Manzamnyama Rivers (Gordon et al. 2008, 2010). This has been attributed either to recovery along the Mpisini, or dilution of Mpisini water by Manzamnyama water at Site 11, or both. In the current study, Site 11 was not found to have consistently improved water quality in the winter sample according to all diatom indices (there was some disagreement between indices derived from the summer sample). The reason for this is not known, and further sampling will be required to establish whether this may indicate a trend. Other physicochemical parameters indicate that this site is in a fair to good class, while diatom indices rate this site as poor to good. Upstream sites on the Mpisini and Mdibi Rivers (Sites 1 and 12) returned the best inferred water quality results. Water at these sites was relatively acidic with low dissolved salt levels, and good levels of dissolved nutrients. Site 10, upstream on the Manzamnyama River, had lower water quality as inferred from diatom population structure. Site 10 has comparable pH, conductivity and phosphate levels to upstream Sites 1 and 12, but higher nitrogen (as TIN) levels. Nitrogen levels at Site 10 decreased from winter 2010 to summer 2011, which correlates with decreased diatom index values. In addition, dissolved oxygen levels were particularly low at Site 10.

Inferred water quality degrades with distance downstream such that sites lower on the rivers surveyed can be classed as being in a moderate to good state. This decrease in inferred water quality occurs where rivers pass through settled areas and it is likely that this has some impact. It was noted during assessment of samples collected during this survey that alpha diversity and diatom indices were to a certain extent inversely correlated. This was not to the extent that alpha diversity could be used to infer water quality, but it is of interest to note that sites with high water quality generally had relatively few taxa present and were dominated by one or a few taxa. While dominants varied considerably between site and season, a small group of taxa were found to be characteristic of clean, upstream sites in this area.

Table 6. Summary of Site 1 on the Mpisini River.



Site description: This site is upstream of the Smelter Site and chosen as a possible reference site. During winter 2010, water levels were the lowest observed at this site to date. GSM biotope is dominated by mud with limited sand available, usually covered with leaf litter.

Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).

T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
20.0	6.4	4.4	36.0	1.25	0.025	0.14	83.26
	Good	Fair	Good	Fair	Good	Natural	Fair

Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).

ASPT	IHAS	Diatoms	Water quality
4.6	54	4.3	Good/Fair
Poor		Natural	

Overall ecological assessment: Good/Fair

Table 7. Summary of Site 7 on the Mpisini River.



Site description: This site is immediately downstream of the Smelter Site. The surrounding land use is forestry (there is no impact from human settlements). Vegetation and GSM biotopes provide good sampling opportunities. This is the only site which contains gravel and limited stones (sampling of these areas is included in the GSM biotope). This is a cattle drinking site.

Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).

T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
21.8	7.7	5.8	63.6	0.29	0.082	0.32	194.76
	Natural	Fair	Fair	Good	Fair	Natural	Poor

Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).

ASPT	IHAS	Diatoms	Water quality
5.7	65	2.8	Fair/Good
Fair		Fair	

Overall ecological assessment: Fair

Table 8. Summary of Site 10 on the Manzanymyama River.



Site description: This site consists of a deep wetland lake (upstream of picture) which gradually becomes shallower (pictured above) before flowing very slowly into a wetland. Surrounding land use is forestry with the Smelter Site in close proximity. There are no impacts from human settlements. Vegetation biotope is sampled in the wetland lake, consisting of marginal vegetation (reeds and grass) and aquatic vegetation. GSM biotope is sampled in the shallower part of the lake and consists of sand and anoxic mud. The GSM is regularly disturbed by cattle passing through and drinking.

Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).

T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
21.3	6.1	1.2	46.6	1.60	0.025	0.25	17.52
	Good	Poor	Good	Fair	Good	Natural	Good

Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).

ASPT	IHAS	Diatoms	Water quality
4.8	51	3.0	Good/Fair
Poor		Fair	

Overall ecological assessment: Fair

Table 9. Summary of Site 11 at confluence of the Mpisini and Manzanmyama Rivers.


							
<p>Site description: The site is within a forest at the confluence of the Mpisini and Manzanmyama Rivers, downstream of the Smelter Site. Surrounding land use is forestry with no effects from human settlements. There is very limited vegetation biotope available for sampling, particularly during the lower flows. Vegetation sampled usually consists of marginal vegetation leaves that dip into the water, root wads and twig snarls. GSM biotope consists of sand and mud. The sand biotope has become limited with low flows during the spring and autumn surveys and disturbance from cattle in summer 2011.</p>							
<p>Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).</p>							
T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
22.3	7.4	4.7	60.1	0.87	0.07	0.10	39.46
	Natural	Fair	Fair	Good	Fair	Natural	Fair
<p>Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).</p>							
ASPT		IHAS		Diatoms		Water quality	
6.0		55		2.8		Good/Fair	
Good/Fair				Fair			
<p>Overall ecological assessment: Fair/Good</p>							

Table 10. Summary of Site 12 on the Mdibi River


							
<p>Site description: This is the uppermost site on the Mdibi River and tentatively proposed as a reference site for this river. Surrounding land use is forestry and some limited human settlement. The GSM biotope consists of mud and leaf litter decay with, at times, limited sand. Low water levels and tree felling made it impossible to sample upstream in August 2010. Therefore sampling took place under the bridge further downstream than usual.</p>							
<p>Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).</p>							
T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
20.0	6.9	5.2	44.4	0.82	0.025	0.12	32.11
	Natural	Fair	Good	Good	Good	Natural	Fair
<p>Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).</p>							
ASPT		IHAS		Diatoms		Water quality	
5.5		61		5.0		Good	
Fair				Natural			
<p>Overall ecological assessment: Good</p>							

Table 11. Summary of Site 13 on the Mdibi River.


							
<p>Site description: The site is located downstream of a bridge culvert. Surrounding land use includes forestry and settlements. Vegetation biotope is dominated by reed stalks and leaves, although there are some aquatic plants available. The vegetation biotope was limited due to low water levels for sampling during 2010/2011. GSM biotope is limited, usually consisting of some mud and sand which is covered by thick shredded leaf litter.</p>							
<p>Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).</p>							
T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
20.5	7.5	5.7	61.3	0.42	0.025	0.09	192.84
	Natural	Fair	Fair	Good	Good	Natural	Poor
<p>Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).</p>							
ASPT		IHAS		Diatoms		Water quality	
6.3		53		3.3		Good/Fair	
Good				Fair			
<p>Overall ecological assessment: Good/Fair</p>							

Table 12. Summary of Site 14 on the Mdibi River.



Site description: This is the lowermost site on the Mdibi River, situated upstream from Lake Mzingazi. Surrounding land use is subsistence forestry and human settlements. Vegetation biotope usually consists of marginal reeds, grasses and aquatic plants, however during both seasons there was very limited marginal vegetation due to low flows. GSM consists of good sand and mud sampling biotope.

Water quality parameters: (T: temperature; DO: dissolved oxygen; EC: electrical conductivity; TIN: total inorganic nitrogen; SRP: soluble reactive phosphorous; (water quality categories based on the ecological Reserve methodologies for water quality are provided below the values where relevant).

T (°C)	pH	DO (mg/L)	EC (mS/m)	TIN (mg/L)	SRP (mg/L)	Chlorophyll- <i>a</i>	
						Phytoplankton (µg/L)	Periphyton (mg/m ²)
20.8	7.4	5.3	63.4	0.42	0.089	0.35	463.59
	Natural	Fair	Fair	Good	Fair	Natural	Poor

Biological and water quality indices: summary of the main index scores (ASPT: average score per taxon; IHAS: integrated habitat assessment system; diatoms; and water quality) (ecological health categories are largely based on those used for ecological Reserve determinations).

ASPT	IHAS	Diatoms	Water quality
5.7	57	3.3	Fair/Good
Fair		Fair	

Overall ecological assessment: Fair

4 Results and discussion of limited biomonitoring

In the sections below, water quality, habitat and biological monitoring data collected at Site 11 are reported. Data from the limited sampling undertaken during autumn 2010 and spring 2011 are presented and data from the comprehensive sampling undertaken during winter 2010 and summer 2011 are included for comparative purposes.

4.1 Water quality assessment

Water temperatures recorded at Site 11 showed variation of about 5°C (Figure 12A), with spring and summer being the warmer seasons. Due to a faulty pH meter, pH was not measured during summer 2011. Measurements during remaining seasons were well within the natural boundary (Figure 12B). Electrical conductivity was classified as Fair during all seasons, with values bordering on the Good class (Figure 12C). Dissolved oxygen concentrations ranged from Fair in winter and spring to Poor in summer and Good in autumn (Figure 12D).

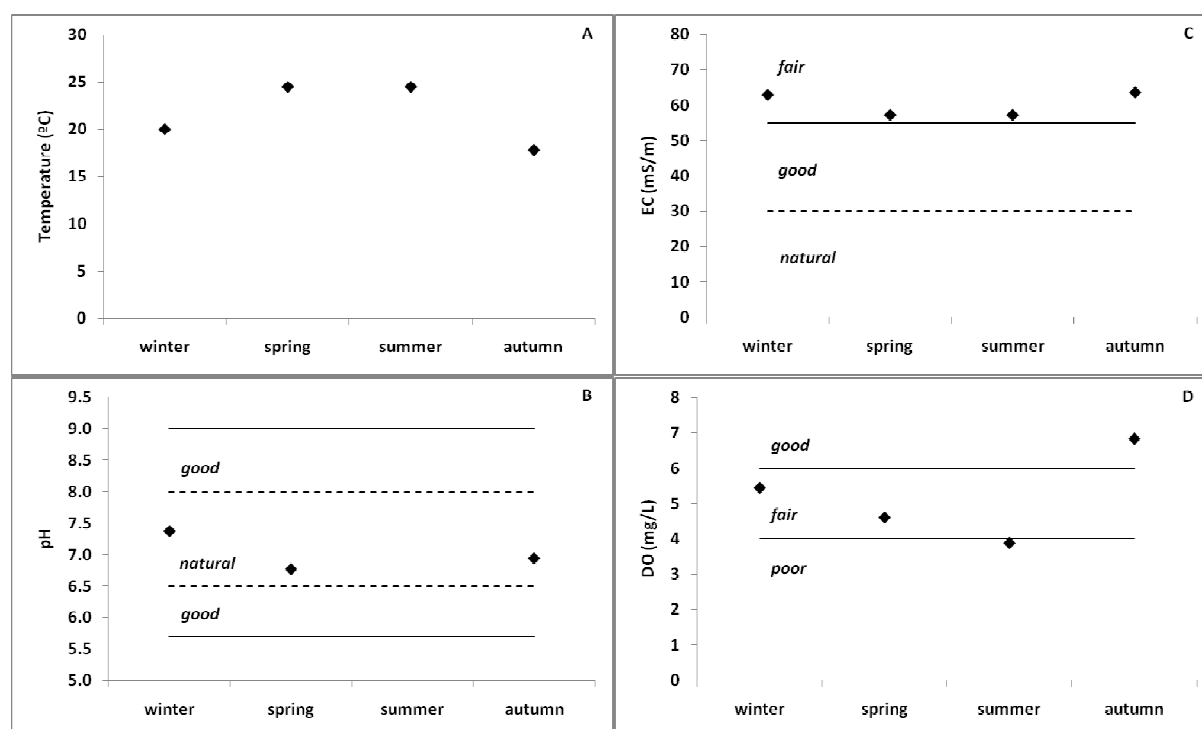


Figure 12A-D. Temperature, pH, electrical conductivity (EC), and dissolved oxygen (DO) values measured at sampling Site 11 over four seasons. Default ecological categories based on ecological Reserve determination methodologies are superimposed on the graphs.

4.2 SASS and habitat assessment

During each season, results from vegetation and GSM biotopes were combined to provide overall SASS scores, number of families and ASPT values (Figure 13A-C). SASS scores and number of taxa were lower in warmer seasons than during winter and autumn (Figure 13A-B). ASPT scores bordered on the Good/Fair boundary value during all seasons with winter and summer being classified as Good and spring and autumn as Fair (Figure 13C). The ASPT

score pattern was mirrored in the IHAS score pattern, suggesting available habitat may have been a driving factor in macroinvertebrate presence/absence. Overall, IHAS scores were low due to the absence of a stones biotope and seasonal variability was negligible (Figure 13D). Site 11 was also badly affected by low flows which reduce available vegetation sampling habitat, adding to the low IHAS scores. All raw data for the water quality and SASS assessments are supplied in Appendix 5.

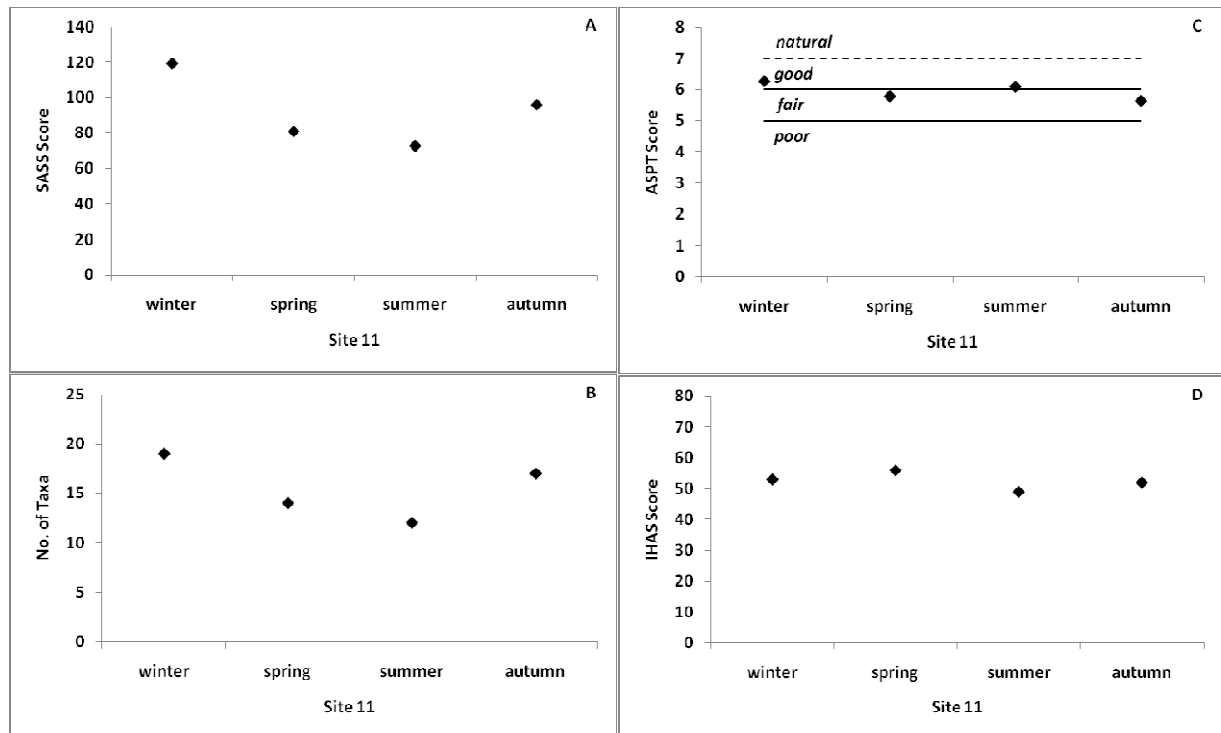


Figure 13A-D South African Scoring System (SASS) score (A), number of families/taxa sampled (B) and Average Score Per Taxon (ASPT) values (C) determined for Site 11 over four seasons. Default ecological categories for ASPT based on ecological Reserve determination methodologies are superimposed on graph C.

Total taxa collected at Site 11 per season ranged between 12 and 19, with winter being the taxa richest season and summer the taxa poorest (Table 13). The average number of taxa collected during the assessment year was 15.5 and the overall total number of taxa per year was 25. A list of species collected at Site 11 during four sampling trips in different seasons is supplied in Appendix 6.

Table 13. Total taxa per season and average taxa per year collected at Site 11.

Year Month Season	2010 Aug winter	2010 Nov spring	2011 Feb summer	2011 May autumn
Total taxa per season	19	14	12	17
Average taxa per year	15.5			
Total taxa per year	25			

5 Conclusion

A summary of the indices measured is provided in Table 1. A provisional overall aquatic ecological health assessment for each of the sites assessed is provided. It must be noted that the methods used to provide the subsequent categories are largely based on expert opinion and assessment of the available data. In addition, the boundary values for the categories are based on the default values provided by the ecological Reserve method and require site-specific refinement.

Upstream sites on the Mpisini and Mdibi Rivers (Sites 1 and 12, reference sites) returned the best water quality results. Water at these sites was relatively acidic with low dissolved salt levels, and good levels of dissolved nutrients. Site 10, upstream on the Manzamnyama River, had lower water quality as inferred from diatom population structure and water quality parameters. Macroinvertebrate taxa and abundance sampled at Site 10 were considerably different from those sampled at all other sites, as previously reported (Gordon et al. 2010). Nitrogen levels at Site 10 decreased from winter 2010 to summer 2011, which correlates with decreased diatom index values. In addition, dissolved oxygen levels were particularly low at Site 10 as reported previously (Gordon et al. 2008, Gordon et al. 2010). This site is driven by groundwater, and the resultant DO values measured there are considerably lower than at all other sites. In addition, there is poor sampling habitat for macroinvertebrates at this site.

There appears to be a water quality impact occurring between Sites 1 and 7, reflected in the diatom community assessment score and characterized by elevated EC, SRP and periphyton chlorophyll-a levels. The smelter complex is situated immediately upstream of Site 7, and as there appears to be limited impact from human settlements, it is possible that this water quality impairment may be related to the activities of the smelter. Further monitoring of changes in water quality between these sites is recommended to identify the cause of the change.

In the past, water quality as inferred from diatom community structure has been found to improve from Site 7 on the Mpisini to Site 11... This was attributed either to recovery along the Mpisini, or dilution of Mpisini water by Manzamnyama water at Site 11, or both. In the current study, Site 11 was not found to have consistently improved water quality in the winter sample. The reason for this is not known, and further sampling is recommended to establish whether this may indicate a trend.

In the current study, additional limited biomonitoring assessments were undertaken in autumn (November 2010) and spring (May 2011). Water quality parameters remained constant between seasons with the exception of DO, which was high in winter decreasing steadily until summer and had highest values in autumn. SASS scores and number of taxa followed a similar trend and ASPT scores stayed at the Good to Fair boundary. These results will be observed further during the next two years of assessment. An overview of total taxa per season and average taxa per year collected at Site 11 is provided in Table 2 in the executive summary and Table 13 in the results section.

River water from the Mpisini and Manzamnyama Rivers (which drain the Smelter Site) enters the Mdibi River between Sites 13 and 14. There is a decrease in ASPT and slight decrease in WQ between these, although diatoms remain the same. Increases in SRP between these two sites are maybe related to input from Mpisini and Manzamnyama Rivers although the impacts of human settlements, which occur between Site 13 and 14, could also be contributing to the increased nutrients at these two sites.

The biological data collected so far will go towards establishing better site-specific reference conditions, and may be useful in recalibrating the benchmark boundary values for ASPT scores to yield site-specific boundary values.

6 References

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7 Appendix

Appendix 1. Diatom index scores from sample sites in the Smelter area August 2010–February 2011 are presented below. Site classifications based on the indices are also presented.

Site	Date	IPS		BDI-2006		Expert opinion	
		Index	Class	Index	Class	Index	Class
1	Aug 2010	16.7	Good	14.4	Good	5	High
	Feb 2011	15.7	Good	14.3	Good	5	High
7	Aug 2010	10.7	Poor	11.3	Moderate	2	Poor
	Feb 2011	13.6	Moderate	13.1	Good	3	Moderate
10	Aug 2010	14.0	Moderate	13.1	Good	4	Good
	Feb 2011	10.6	Poor	11.7	Moderate	2	Poor
11	Aug 2010	10.9	Poor	12.4	Moderate	2	Poor
	Feb 2011	16.2	Good	16.9	Good	2	Poor
12	Aug 2010	18.1	High	17.5	High	5	High
	Feb 2011	18.7	High	17.0	High	5	High
13	Aug 2010	14.3	Moderate	13.6	Good	3	Moderate
	Feb 2011	13.0	Moderate	13.3	Good	3	Moderate
14	Aug 2010	15.7	Good	13.6	Good	3	Moderate
	Feb 2011	12.7	Moderate	10.6	Moderate	3	Moderate

Appendix 2. Summary of biomonitoring, water chemistry and nutrient analyses undertaken. Detection limits were as follows: Ammonium (NH₄⁺): 0.05mg/L; Nitrites (NO₂⁻): 0.01mg/L; Nitrates (NO₃⁻): 0.5mg/L; Soluble Reactive Phosphorus (SRP): 0.05mg/L. Total Inorganic Nitrogen (TIN) were calculated by adding NH₄⁺, NO₂⁻ and NO₃⁻. Values below detection limit are shown as half the detection limit (NH₄⁺= 0.025mg/L; NO₂⁻=0.005mg/L; NO₃⁻=0.25mg/L; SRP=0.025mg/L). TIN=0.280mg/L indicates that all added parameters were below the detection limit.

Site Code	River	Month	Year	Biomonitoring				Water Chemistry				Nutrients						
				Sass Score	No. of Taxa	ASPT Score	IHAS Score (%)	Temp (°C)	DO (mg/L)	pH	EC (mS/m)	NH4 (mg/L)	NO3 (mg/L)	NO2 (mg/L)	TIN (mg/L)	SRP (mg/L)	Phyto-plankton chl-a (µg/L)	Peri-phyton chl-a (mg/cm ²)
1	Mpisini	August	2010	50	11	4.55	52	19.0	3.36	6.42	328	0.050	2.163	0.041	2.254	0.025	0.187	11.204
7	Mpisini	August	2010	87	14	6.21	64	20.0	5.22	7.66	698	0.050	0.500	0.021	0.571	0.140	0.593	227.996
10	Manzamnyama	August	2010	74	15	4.93	48	18.0	1.83	6.07	467	0.050	2.845	0.051	2.946	0.025	0.423	8.304
11	Confluence	August	2010	119	19	6.26	53	20.0	5.45	7.37	629	0.050	1.415	0.025	1.490	0.116	0.085	77.012
12	Mdibi	August	2010	86	16	5.38	58	17.0	4.72	6.89	416	0.050	1.316	0.024	1.389	0.025	0.101	1.647
13	Mdibi	August	2010	71	11	6.45	49	18.0	5.70	7.51	646	0.050	0.500	0.013	0.563	0.025	0.083	264.222
14	Mdibi	August	2010	109	18	6.06	52	19.0	5.01	7.37	673	0.050	0.500	0.017	0.567	0.078	0.703	463.592
11	Confluence	November	2010	81	14	5.79	56	17.8	6.84	6.94	636							
1	Mpisini	February	2011	51	11	4.64	55	21.0	5.46		392	0.025	0.250	0.005	0.280	0.025	0.098	155.325
7	Mpisini	February	2011	83	16	5.19	66	23.5	6.37		573	0.025	0.250	0.005	0.280	0.025	0.046	161.515
10	Manzamnyama	February	2011	42	9	4.67	53	24.5	0.52		465	0.025	0.250	0.011	0.286	0.025	0.074	26.746
11	Confluence	February	2011	73	12	6.08	49	24.5	3.89		572	0.025	0.250	0.005	0.280	0.025	0.109	1.899
12	Mdibi	February	2011	78	14	5.57	64	23.0	5.64		471	0.025	0.250	0.005	0.280	0.025	0.137	62.580
13	Mdibi	February	2011	79	13	6.08	56	23.0	5.70		580	0.025	0.250	0.005	0.280	0.025	0.089	121.460
14	Mdibi	February	2011	81	15	5.40	62	22.5	5.52		595	0.025	0.250	0.005	0.280	0.100	0.002	
11	Confluence	May	2011	96	17	5.65	52	24.5	4.61	6.77	572							

Appendix 3. Summary of number of taxa found at each sampling site in both seasons (August = winter, February = summer). Averages are shown per site over three replicates.

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
Porifera														
Coelenterata														
Turbellaria														
Oligochaeta			1	2	1			1			1	1		
Leeches														
Amphipoda											2		1	
Potamonautidae		1	1	2							1			
Atyidae	39	44	23	27			23	36	63	39	28	35	22	36
Palaemonidae														
Hydracarina		2			2	2			1					
Notonemouridae														
Perlidae														
Baetidae	1	3	26	15	2	2	3	2	3	2	4	6	11	7
Caenidae	1	1	4	1					1		9	3		1
Ephemeroidea														
Heptageniidae														
Leptophlebiidae														
Oligoneuridae														
Polymitarcyidae														
Prosopistomatidae														
Teloganodidae														
Tricorythidae			3	1			1	6			8	3	12	1
Calopterygidae														
Chlorocyphidae														
Chlorolestidae														
Coenagrionidae	1	1	2		4	6	1	1	2	1	1			1
Lestidae														
Platycnemidae														
Protoneuridae														
Aeshnidae														
Corduliidae														
Gomphidae		1			1					1	1			2
Libellulidae					1	1								
Crambidae														
Pyralidae														

Site Year Month	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
Belostomatidae														
Corixidae														
Gerridae	1	3		1	1		1			3			2	
Hydrometridae														
Naucoridae														
Nepidae														
Notonectidae														
Pleidae				1										
Veliidae	1	1		1			1						1	1
Corydalidae														
Sialidae														
Dipseudopsidae														
Ecnomidae			2											
Hydropsychidae			13										1	
Philopotamidae			1											
Polycentropodidae														
Psychomyiidae														
Barbarochthonidae														
Calamoceratidae														
Glossosomatidae														
Hydroptilidae														
Hydrosalpingidae														
Lepidostomatidae														
Leptoceridae				1					1	3		7	1	2
Petrothrincidae														
Pisuliidae														
Sericostomatidae														
Dytiscidae					1									
Elmidae/Dryopidae						1					2			1
Gyrinidae			2					2					1	
Haliplidae														
Helodidae											1		1	
Hydraenidae														
Hydrophilidae					1									1
Limnichidae														
Psephenidae														

Site Year Month	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
Athericidae														
Blepharoceridae														
Ceratopogonidae		1			3	1					1			
Chironomidae	6	10	1	2	3	4	1	2	1	1	5	2	3	1
Culicidae					4	1								
Dixidae														
Empididae														
Ephydriidae														
Muscidae														
Psychodidae														
Simuliidae			3								1		3	
Syrphidae														
Tabanidae														
Tipulidae														
Ancylidae									1					
Bulininae														
Hydrobiidae														
Lymnaeidae														
Physidae				1										
Planorbinae												1		
Thiaridae				1			4							
Viviparidae														
Corbiculidae														
Sphaeriidae														
Unionidae														

Appendix 4. Diatom abundances from sample sites in the Smelter area (August 2010 and February 2011). All data are proportions of identified frustules in each sample. Where diatoms could not be identified, morphospecies were assigned and used in all analyses.

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
<i>Achnanthes gibberula</i>	0.01													
<i>Achnanthes oblongella</i>		0.02		0.01			0.09	0.21	0.64	0.02	0.08	0.13	0.04	0.03
<i>Achnanthes pulviscula</i>	0.39	0.03					0.03		0.27	0.01	0.03	0.05	0.07	
<i>Achnanthes subaffinis</i>			0.01											
<i>Achnantheidium affine</i>							0.01							
<i>Achnantheidium eutrophilum</i>						0.01								
<i>Achnantheidium exiguum</i>		0.01				0.04					0.02	0.03		
<i>Achnantheidium macrocephalum</i>	0.01													
<i>Achnantheidium minutissimum</i>	0.03	0.01						0.01	0.04		0.25		0.16	
<i>Achnantheidium saprophilum</i>						0.03		0.04			0.53	0.07	0.28	
<i>Amphora exigua</i>							0.02							
<i>Bacillaria paradoxa</i>							0.23							
<i>Brachysira aff. steindorfiana</i>	0.19	0.33								0.38				
<i>Brachysira sp2</i>	0.26	0.43			0.70	0.04			0.02	0.09				
<i>Brachysira styriaca</i>					0.01									
<i>Caloneis bacillum</i>						0.01								
<i>Capartogramma crucicula</i>							0.01					0.03	0.01	
<i>Cocconeis placentula</i>				0.16				0.05				0.25	0.01	
<i>Craticula halophila</i>					0.01									
<i>Cyclostephanos invisitatus</i>			0.01											
<i>Cymbella tumida</i>			0.09	0.01										
<i>Cymbopleura naviculiformis</i>					0.03									
<i>Denticula sundaysensis</i>			0.02											

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
<i>Diadsmis contenta</i>				0.02		0.01	0.07							
<i>Eunotia bilunaris</i>	0.01									0.11				0.03
<i>Eunotia flexuosa</i>							0.04							
<i>Eunotia kocheliensis</i>	0.01													
<i>Eunotia minor</i>	0.01	0.01							0.02	0.04				
<i>Eunotia sp2</i>		0.01		0.03								0.02		
<i>Eunotia sp3</i>									0.01	0.24				
<i>Fragilaria biceps</i>			0.05											
<i>Fragilaria capucina</i>			0.04											
<i>Fragilaria capucina</i> var. <i>vaucheriae</i>						0.03								
<i>Frustulia vulgaris</i>			0.01				0.02					0.01		
<i>Gomphonema</i> aff. <i>gracile</i>											0.02			
<i>Gomphonema</i> aff. <i>lagenula</i>										0.02				
<i>Gomphonema affine</i>	0.04													0.03
<i>Gomphonema angustatum</i>	0.03	0.01		0.06		0.02				0.07	0.01	0.12	0.05	0.15
<i>Gomphonema clavatum</i>											0.01			
<i>Gomphonema exilissimum</i>											0.02		0.03	
<i>Gomphonema extentum</i>														0.36
<i>Gomphonema gracile</i>			0.07											
<i>Gomphonema insigne</i>													0.08	
<i>Gomphonema lagenula</i>												0.02		0.17
<i>Gomphonema parvulum</i>		0.01	0.14	0.05		0.02					0.02		0.03	0.11
<i>Gomphonema pseudoaugur</i>														0.08
<i>Gomphonema pumilum</i>		0.01		0.04		0.01		0.53				0.04		

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
<i>Gomphonema pumilum</i> var. <i>rigidum</i>			0.04											0.17
<i>Gomphonema</i> sp7							0.01							
<i>Gomphonema venusta</i>							0.01							
<i>Gomphonema vibrio</i>		0.07												
<i>Gomphosphenia</i> aff. <i>oahuensis</i>			0.06											
<i>Lemnicola hungarica</i>											0.01			
<i>Luticola kotschyi</i>					0.01	0.05							0.04	
<i>Luticola mutica</i>					0.01									
<i>Mastogloia elliptica</i>										0.02				
<i>Mastogloia smithii</i>					0.02									
<i>Mayamaea atomus</i>						0.02								
<i>Navicula arvensis</i> var. <i>maior</i>												0.02		
<i>Navicula cryptocephala</i>			0.01	0.01			0.01							
<i>Navicula erifuga</i>				0.01		0.19		0.03				0.10		0.01
<i>Navicula gregaria</i>			0.01											0.01
<i>Navicula libonensis</i>						0.01								
<i>Navicula longicephala</i>							0.02							
<i>Navicula microcari</i>														0.01
<i>Navicula recens</i>							0.01							
<i>Navicula riediana</i>							0.01							
<i>Navicula rostellata</i>												0.01		0.02
<i>Navicula schroeteri</i>								0.08	0.03					0.03
<i>Navicula</i> sp6							0.02		0.04				0.06	
<i>Navicula</i> sp7								0.05						

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
<i>Navicula symmetrica</i>			0.11				0.10	0.01						
<i>Navicula vandamii</i>					0.04									
<i>Navicula veneta</i>								0.02				0.02		
<i>Navigiolum algeriense</i>							0.01							
<i>Nitzschia amphibia</i>			0.05	0.01	0.01				0.04					
<i>Nitzschia amplectens</i>							0.01							
<i>Nitzschia archibaldii</i>					0.04									
<i>Nitzschia clausii</i>			0.04											0.01
<i>Nitzschia desertorum</i>											0.01			
<i>Nitzschia dissipata</i>						0.04	0.05							
<i>Nitzschia filiformis</i>			0.01											
<i>Nitzschia fonticola</i>				0.26										
<i>Nitzschia frustulum</i>			0.02			0.03					0.01		0.01	
<i>Nitzschia hantzschiana</i>				0.03	0.01									
<i>Nitzschia intermedia</i>				0.01										
<i>Nitzschia linearis</i>			0.05			0.02								
<i>Nitzschia microcephala</i>					0.01	0.01								
<i>Nitzschia nana</i>	0.01	0.04			0.07							0.01		
<i>Nitzschia obtusa</i> var. <i>kurzii</i>			0.01											
<i>Nitzschia palea</i>		0.01	0.01	0.06		0.11	0.02							
<i>Nitzschia perminuta</i>				0.22				0.01						0.03
<i>Nitzschia pura</i>							0.01							
<i>Nitzschia pusilla</i>						0.06								
<i>Nitzschia</i> sp1			0.06											

Site	1		7		10		11		12		13		14	
	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011	2010	2011
	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb	Aug	Feb
<i>Nitzschia</i> sp6			0.01											
<i>Nitzschia</i> sp22							0.05							
<i>Nitzschia</i> sp23							0.05							
<i>Nitzschia supralitorea</i>							0.01							
<i>Nitzschia valdecostata</i>			0.06											
<i>Pinnularia obscura</i>													0.01	
<i>Pinnularia subcapitata</i>						0.04								
<i>Placoneis clementis</i>					0.01	0.04								
<i>Planothidium frequentissimum</i>			0.03											
<i>Rhopalodia operculata</i>								0.01						
<i>Rhopalodia rupestris</i>						0.02								
<i>Sellaphora</i> sp1					0.02	0.04								
<i>Seminavis strigosa</i>			0.02	0.02			0.03							
<i>Stauroneis pachycephala</i>	0.01						0.02							
<i>Stephanodiscus agassizensis</i>							0.01							
<i>Surirella ovalis</i>								0.01						
<i>Surirella tenera</i>		0.01												
<i>Tabularia fasciculata</i>				0.01										
<i>Tryblionella debilis</i>							0.02							

Appendix 5. Summary of biomonitoring and water chemistry at Site 11 during four seasons.

Year	Month	Season	Biomonitoring				Water Chemistry			
			Sass Score	No. of Taxa	ASPT Score	IHAS Score (%)	Temp (°C)	DO (mg/L)	pH	EC (mS/m)
2010	August	winter	119	19	6.26	53	20.00	5.45	7.37	63
2010	November	spring	81	14	5.79	56	24.5	4.61	6.77	57
2011	February	summer	73	12	6.08	49	24.50	3.89		57.2
2011	May	autumn	96	17	5.65	52	17.8	6.84	6.94	64

Appendix 6. List of taxa collected at Site 11 during 4 seasons using the SASS5 method.

Abundances are estimated as follows: 1 = 1, A = 2-10, B = 10-100.

Year Month Season	2010 August winter	2010 November spring	2011 February summer	2011 May autumn
Oligochaeta	A	A		1
Amphipoda	1	1		
Potamonautidae				A
Atyidae	B	B	B	B
Palaemonidae				1
Hydracarina	1		1	
Baetidae	A	1	A	A
Caenidae	1	A		
Tricorythidae	A	B	B	A
Chlorocyphidae	A	A		1
Coenagrionidae	1	1	1	B
Gomphidae			1	
Gerridae	A	A	A	B
Hydrometridae	1			A
Pleidae				1
Veliidae	A	1	1	A
Leptoceridae	1	1	1	1
Gyrinidae	A	A		
Ceratopogonidae	A		1	1
Chironomidae	A	A	1	A
Psychodidae				A
Tipulidae	1			
Ancylidae	1			
Planorbinae			1	
Thiaridae	B	B		B
Total taxa per season	19	14	12	17
Average taxa per year	15.5			
Total taxa per year	25			