APPENDIX B: DIATOM ASSESSMENT OF THE ORANGE RIVER MAIN STEAM AND ASSOCIATED TRIBUTARIES.

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B1 BACKGROUND AND INTRODUCTION

Diatoms are of great ecological importance because of their role as primary producers, and form the base of the aquatic food web. They usually account for the highest number of species among the primary producers in aquatic systems (Leira, 2005). Diatoms are photosynthetic unicellular organisms and are found in almost all aquatic and semi-aquatic habitats.

Diatoms are a siliceous class (*Bacilariophyceae* of the phylum *Bacilariophyt*a) of algae. A remarkable aspect of diatoms is their silicon dioxide cell walls. The cell walls are perforated and ornamented with many holes, which are arranged in defined and unique patterns. Identification is based on the nature of these perforations as well as their orientation and densities.

Recent studies, as well as studies in progress, have identified diatoms as useful organisms to include in the suite of biomonitoring tools currently used in South Africa (Bate *et al.*, 2002; De la Rey *et al.*, 2004; Taylor, 2004) both for assessments of current water quality and for establishing historical conditions in rivers in South Africa (Taylor *et al.*, 2005a).

Diatoms have been shown to be reliable indicators of specific water quality problems such as organic pollution, eutrophication, acidification and metal pollution (Rott, 1991; Tilman *et al.*, 1982; Dixit *et al.*, 1992; Cattaneo *et al.*, 2004), as well as for general water quality (AFNOR, 2000). The reasons why diatoms are useful tools for biomonitoring are listed by Round (1993):

- Diatoms have a universal occurrence throughout all rivers;
- Field sampling is rapid and easy;
- Cell cycle is rapid and they react quickly to perturbation;
- Diatoms are relatively insensitive to physical features in the environment;
- Cell counting by microscopic techniques is rapid and accurate;
- Cell numbers per unit area of substratum are enormous, making random counts excellent assessments of diatoms;
- The ecological requirements of diatoms are in many cases better known than those of any other group of riverine organisms;
- Permanent records can be made from every sample;
- Diatoms do not have specific food requirements, specialised habitat niches, and are not governed to a major extent by stream flow.

The specific water quality tolerances of diatoms have been resolved into different diatom-based water quality indices, used around the world. In general, each diatom species used in the calculation of the index is assigned two values; the first value reflects the tolerance or affinity of the particular diatom species to a certain water quality (good or bad) while the second value indicates how strong (or weak) the relationship is. These values are then weighted by the abundance of the particular diatom species in the sample. The diatom index used in the present study is known as the Specific Pollution sensitivity Index (SPI; Coste in CEMAGREF, 1982), one of the most extensively tested indices in Europe.

Diatom-based water quality indices have recently been evaluated and implemented in South Africa (Taylor, 2004; River Health Programme, 2005). De la Rey *et al.* (2004) and Taylor (2004) showed that diatom-based pollution indices may be good bio-indicators of water quality in aquatic ecosystems in South Africa by demonstrating a measurable relationship between water quality variables such as pH, electrical conductivity, phosphorus and nitrogen, and the structure of diatom communities as reflected by diatom index scores.

The close association between diatom community composition and water quality allows for inferences to be drawn about water quality.

B2 METHODS

B2.1 AVAILABLE DATA

B2.1.1 Orange River main stem

Three sets of data were available for this study and included:

- Samples collected during 2005 as part of Orange River study on assistant pollutants on sediment. Samples were taken from Douglas to the Orange Rive Mouth.
- Samples collected during April June 2008 and during August-September 2009 as part of a water quality monitoring and status quo assessment study of the Orange-Senqu River and associated tributaries (DWA, 2009).
- Samples taken at the EFR sites as part of this study during 2010.

Figure B1 presents the diatom samples taken in the Orange rive main stem where:

- The samples taken during 2005 are represented by pink circles.
- Samples taken during 2008 and 2009, for April, May, June and August. As an example, a blue circle with yellow outline () labelled 3, 14 means sample 3 and 14 were sampled at the same site, but during different months. Sample 3 was taken in April and sampled again in Aug but was numbered 14.
- EFR sites are indicated by red arrows.

B2.1.2 Orange River tributaries: Caledon, Kraai and Molopo Rivers

Three sets of data were available and included:

- Samples were collected during April 2008 and August 2009 as part of a water quality monitoring and status quo assessment study of the Orange-Senqu River and associated tributaries (DWA, 2009).
- Samples taken at the EFR sites as part of this study during 2010.
- Samples collected during 2005 as part of a PhD study (De la Rey, 2008a and b) investigating the use of diatom-based biological monitoring:
 - Part 1: A comparison of the response of diversity and aut-ecological diatom indices to water quality variables in the Marico-Molopo River catchment.
 - Part 2: A comparison of the response of SASS 5 and diatom indices to water quality and habitat variation. Diatoms at four RHP sites were sampled during May, July and September 2005. The EFR M8 was sampled during April 2010. This site is a wetland and three subsamples were taken from the wetland for assessment.

Figure B2 presents the diatom samples taken in the Kraai and Caledon River tributaries where:

- Samples were taken during 2009, for April 2008 and August 2009. As an example, a blue circle with yellow outline () labelled 3, 14 means sample 3 and 14 were sampled at the same site, but during different months. Sample 3 was taken in April and sampled again in Aug but was numbered 14.
- EFR sites are indicated by red arrows.

Figure B3 presents the samples taken in the Molopo River where:

- Samples taken during 2005 are represented by MOL1 4.
- Samples taken during April 2010 are presented on the Google Earth image.

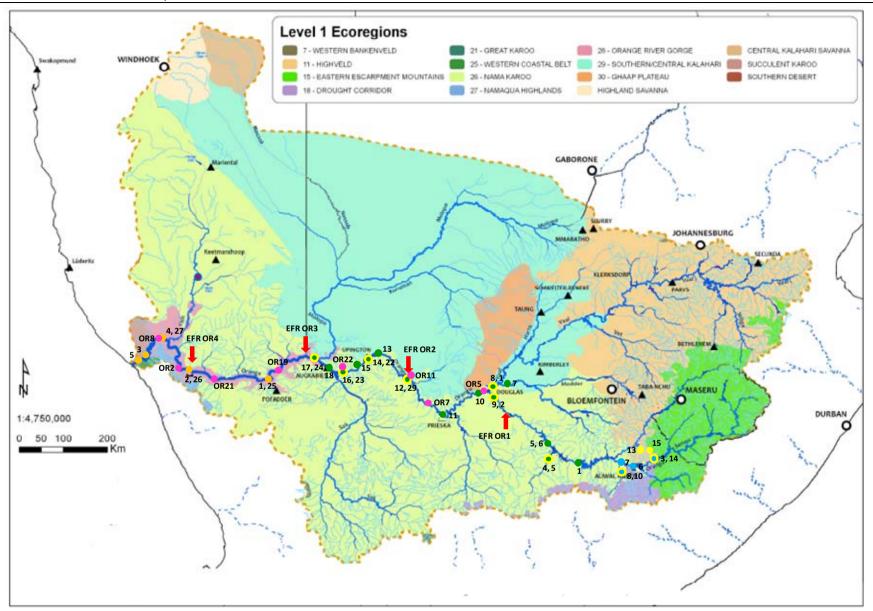
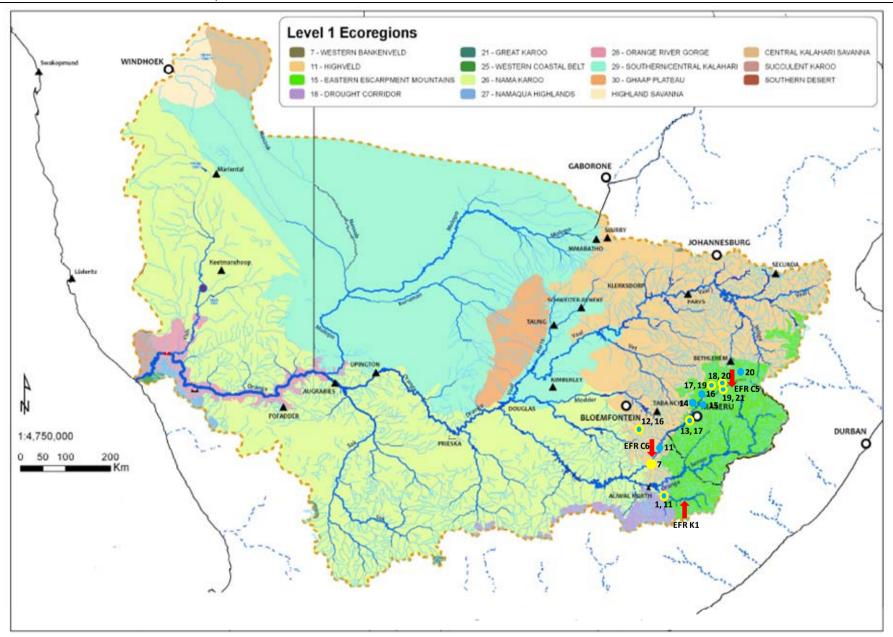
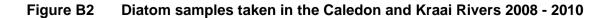
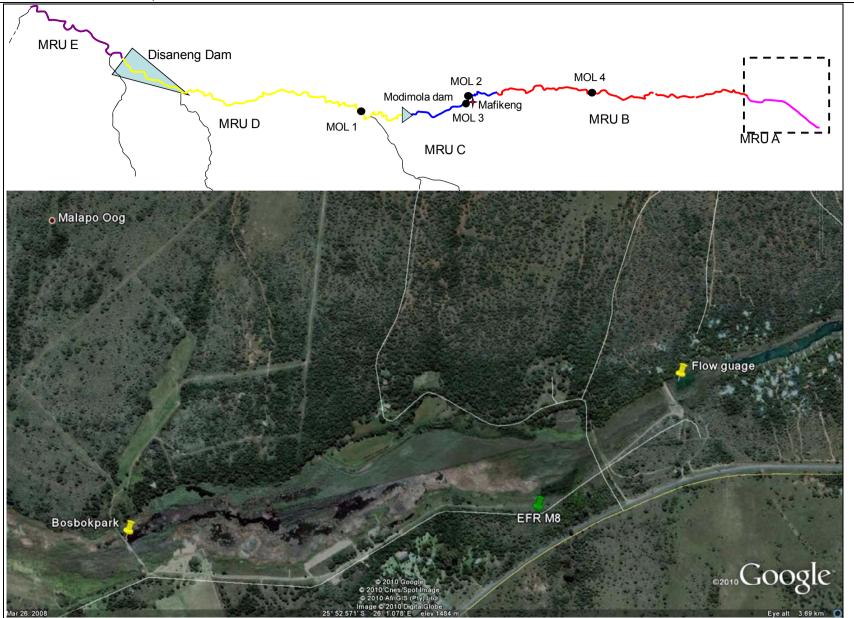


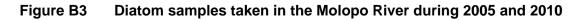
Figure B1 Diatom samples taken in the Orange rive main stem during 2005, 2008 - 2010





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B2.2 SAMPLING

Epilithon¹ and Epiphyton² were sampled as outlined Taylor *et al.* (2005b) and Taylor *et al.* (2007a). These methods were designed and refined as part of the Diatom Assessment Protocol (DAP), a Water Research Commission (WRC) initiative. Taylor *et al.* (2007a), have based the method manual on several key documents including Kelly *et al.* (1998), CEN (2003), DARES (2004) and Taylor *et al.* (2005b). Diatom samples were taken at the site by scrubbing the substrate with a small brush and rinsing both the brush and the substrate with distilled water. Samples were taken from five or more cobbles (diameter > 64, ≤265 mm) at the site where cobbles were available and the flows allowed sampling.

B2.3 ANALYSIS

Preparation of diatom slide followed the Hot HCl and KMnO₄ method as outlined in Taylor *et al.* (2007a). A Nikon Eclipse E100 microscope with phase contrast optics (1000x) was used to identify diatom valves on slides. The aim of the data analysis was to count diatom valves to produce semiquantitative data from which ecological conclusions can be drawn (Taylor *et al.*, 2007a). Schoeman, (1973) and Battarbee (1986) concluded that a count of 400 valves per slide is satisfactory for the calculation of relative abundance of diatom species and this range is supported by Prygiel *et al.* (2002), according to Taylor *et al.* (2007a). Therefore a count of 400 valves per sample or more was counted and the nomenclature followed Krammer and Lange-Bertalot (1986-91). Diatom index values were calculated in the database programme OMNIDIA (Lecointe *et al.*, 1993) for epilithon data in order to generate index scores to general water quality variables.

B2.4 DATA ANALYSIS

B2.4.1 Application of SPI water quality class boundaries

The water quality class boundaries for the SPI have been set by several authors for rivers in Sweden, Finland, Belgium and France. Class boundaries set by Eloranta and Soininen (2002) was used in the RHP (2005) study. For Reserve studies the class boundaries, set by Prygiel and Coste (2000), which are applicable to low and mid-altitude areas in Western Europe, and the class limits set by Eloranta and Soininen (2002) were considered to be too high for the different water quality ECs dealt with in Reserve studies. The Prygiel and Coste (2000) class boundaries were adapted for Reserve studies to accommodate boundary ECs and applied during the interpretation of the results.

The class boundaries for Eloranta and Soininen (2002) and Prygiel and Coste (2000) are provided in Table B1 and the adjusted class boundaries used in the current Reserve studies is provided in Table B2.

¹ Algae growing on rock or stone surfaces.

² Algae growing on macrophyte surfaces.

Table B1Class limit boundaries for the SPI according Eloranta and Soininen (2002) and
Prygiel and Coste (2000)

Prygiel and Coste (2000)	Class	Eloranta and Soininen (2002) Index score		
Index score	01055			
17 ≤ SPI ≤ 20	High quality	>17		
13 ≤ SPI < 17	Good quality	15 – 17		
9 ≤ SPI < 13	Moderate quality	12 – 15		
5 ≤ SPI < 9	Poor quality	9 - 12		
SPI < 5	Bad quality	<9		

Table B2 Adjusted class limit boundaries for the SPI index applied in current Reserve studies

SPI score	Class	Ecological Category
>17.3		А
16.8 – 17.2	HIGH QUALITY	A/B
13.3 – 16.7	GOOD QUALITY	В
12.9 – 13.2	GOOD QUALITY	B/C
9.2 – 12.8	MODERATE	С
8.9 – 9.1	QUALITY	C/D
5.3 - 8.8	POOR QUALITY	D
4.8 - 5.2	POORQUALITY	D/E
< 4.8	BAD QUALITY	E

B2.4.2 Diatom based Ecological classification

Ecological characterisation of the samples was based on Van Dam *et al.* (1994). This work includes the preferences of 948 freshwater and brackish water diatom species in terms of pH, nitrogen, oxygen, salinity, humidity, saprobity and trophic state as provided by OMNIDIA (Le Cointe *et al.*, 1993).

B2.5 TERMINOLOGY

Terminology used in this specialist appendix is outlined in Taylor *et al.* (2007a) and summarised below.

Trophy	
Dystrophic	Rich in organic matter, usually in the form of suspended plant colloids, but of a low nutrient content.
Oligotrophic	Low levels or primary productivity, containing low levels of mineral nutrients required by plants.
Mesotrophic	Intermediate levels of primary productivity, with intermediate levels of mineral nutrients required by plants.
Eutrophic	High primary productivity, rich in mineral nutrients required by plants.
Hypereutrophic	Very high primary productivity, constantly elevated supply of mineral nutrients required by plants.
Mineral content	
Very electrolyte poor	< 50 µS/cm
Electrolyte-poor (low electrolyte content)	50 - 100 μS/cm
Moderate electrolyte content	100 - 500 μS/cm
Electrolyte-rich (high electrolyte content)	> 500 µS/cm
Brackish (very high electrolyte content)	> 1000 µS/cm
Saline	6000 μS/cm
Pollution (Saprobity)	
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Unpolluted to slightly polluted	BOD <2, O ₂ deficit <15% (oligosaprobic)		
Moderately polluted	BOD <4, O ₂ deficit <30% (β -mesosaprobic)		
Critical level of pollution	BOD <7 (10), O ₂ deficit <50% (β-ά-mesosaprobic)		
Strongly polluted	BOD <13, O ₂ deficit <75% (ά-mesosaprobic)		
Very heavily polluted	BOD <22, O ₂ deficit <90% (ά-meso-polysaprobic)		
Extremely polluted	BOD >22, O ₂ deficit >90% (polysaprobic)		

B2.6 CONFIDENCE IN DATA AVAILABILITY

The confidence in data availability for each EFR site is provided in Table B

Site	Data Availability	Confidence
EFR O1	No data were available for the EFR site specifically except one sample taken during 2010 EFR site visit. Diatom sample collection during 2008 and 2009 US and DS of site was available along with <i>in situ</i> water quality data.	3
EFR O2	Site specific diatom data were available from sample collection during 2005, 2008 – 2009 as well as data from sample collected during EFR site visit. Diatoms were taken during 2005, 2008 - 2009 across the reach, along with measured <i>in situ</i> water quality measurements.	3.5
EFR O3	No data were available for EFR site specifically except one sample taken during 2010 EFR site visit. Diatom sample collection during 2008 and 2009 US and DS of site was available along with <i>in situ</i> water quality data.	3
EFR O4	Site specific diatom data were available from sample collection during 2008 – 2009 as well as data from sample collected during EFR site visit. Three diatom samples were taken during 2005, 2008 - 2009 across the reach, along with measured <i>in situ</i> water quality measurements.	3.5
EFR C5	No data were available for the EFR site specifically except one sample taken during 2010 EFR site visit. Good information available from diatom sample collection during 2008 and 2009 across the reach and tributaries, along with <i>in situ</i> water quality measurements.	3.5
EFR C6	No data were available for the EFR site specifically except one sample taken during 2010 EFR site visit. Less samples taken during 2008 – 2009 across the reach than EFR C5.	3
EFR K7	No data were available for the EFR site specifically except one sample taken during 2010 EFR site visit. Diatom sample collection during 2008 and 2009 US and DS of site was available along with <i>in situ</i> water quality data.	3
EFR M8	Diatom data collected during 2005 as part of a PhD study (De la Rey, 2008). Diatoms at four RHP sites were sampled during May, July and September 2005. The EFR site was sampled during April 2010.	2

B2.7 REPORT OUTLINE

The Results are provided for the Orange River main stem, Kraai River, Caledon River and Molopo River respectively.

B2.7.1 Orange River main stem

Section B3 provides the results of the samples pertaining to the Orange River main stem. The Orange River was divided into ten different reaches based on the occurrence of dams within the system and Management Resource Units (MRU) (Ref). The diatom reaches are outlined below:

- Reach 1: Lesotho border to Gariep Dam.
- Reach 2: Orange River downstream of Gariep Dam to Van der Kloof Dam. This is also MRU A.
- Reach 3: Orange River downstream of Van der Kloof Dam to confluence with Vaal River. This is also MRU B.
- Reach 4: Upstream of Vaal/Orange River confluence. Falls within MRU B.
- Reach 5: Vaal/Orange River confluence to Prieska. Falls within MRU B.
- Reach 6: Prieska to Boegoeberg Dam. Same delineation as MRU C.
- Reach 7: Boegoeberg Dam to Augrabies Falls. Falls within MRU D.
- Reach 8: Augrabies Falls to Vioolsdrift. Falls within MRU E.

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- Reach 9: Vioolsdrift to Fish River confluence. Falls within MRU F.
- Reach 10: Fish River confluence to Estuary.

B2.7.2 Kraai, Caledon and Molopo Rivers

Section B4 to B6 provides the results of the the samples pertaining to the Kraai, Caledon and Molopo Rivers. Reaches were based on the Management Resource Units delineated for the Reserve study as outlined in (ref).

B3 ORANGE RIVER DIATOM RESULTS

B3.1 REACH 1 - LESOTHO BORDER TO GARIEP DAM

B3.1.1 Sample 3 and 14: Upstream of Kraai River confluence, just below Lesotho border

This site was sampled during April 2008 (Sample 3) and August 2009 (Sample 14). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below.

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
3	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Continuously high	6.9	D
14	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	14.9	В

The **April** 2008 sample was dominated by species preferring high organic levels (EOMI, ESBM, FSAP and MAPE). These species are usually abundant in sewage effluent. The diatom community was dominated by *Nitzschia* species indicating a highly polluted water body with readily available nutrients (Cholnoky 1968; van Dam *et al.*, 1994). Salinity was elevated at this site and this is evident by the dominance of NIFR that usually occurs in electrolyte rich to brackish waters and are tolerant of highly polluted waters (Taylor *et al.*, 2007b). Pollution tolerant valves made up 64% of the count indicating that this site was highly polluted.

The **August** 2009 sample indicated a marked improvement in biological water quality. Organic pollution levels were reduced to a great extent. The sample was dominated by CPLA and RUNI indicating that nutrients were still problematic at this site and that turbidity was elevated.

B3.1.2 Sample 13 and 15: Sterkspruit and Kornetspruit

These two samples were taken during August 2009. The sites were located in smaller tributaries of the Upper Orange River. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
13	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Elevated	13.6	В
15	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	13.3	В

The presence of *Achnanthidium* species in both samples indicated well oxygenated waters. However both samples had a high occurrence (5% dominance) of RSIN and ESBM as well as CPLE indicating that nutrient enrichment may have been problematic at this site at times. Pollution levels were low at Site 13, however pollution levels were substantially higher at site 15.

B3.1.3 Sample 6 and 7: Orange River

Sample 6 was located at Aliwal North while sample 7 was located above Gariep Dam. No diatom results could be generated as the diatom counts were non-viable due to high flows.

B3.1.4 Sample 8 and 10: Stormbergspruit at Burgersdorp

This site was sampled during April 2008 (Sample 8) and August 2009 (Sample 10). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
8	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Periodically elevated	9.3	C/D
10	Circumneutral	Eutrophic	Fresh-brackish	Low	Periodically elevated	5.9	D

These samples were characterised by species preferring elevated nutrient levels as well as organics. During August 2009 pollution levels increased drastically and it was evident that anthropogenic activities were impacting this site. Sewage seemed to be the primary pollution problem at this site.

B3.2 OVERALL ASESSMENT FOR REACH 1

The upper tributaries of the Orange River are in a good condition, with well oxygenated waters. These tributaries were in a B EC although it seems that nutrient input from surrounding farming activities may be problematic at times. Stormbergspruit below Aliwal North has elevated nutrient and organic levels. Pollution levels are very high at times as 79% of the August 2009 sample was dominated by pollution tolerant valves. As the samples in the main stem of the Orange River were non-viable it estimated that this reach is in a C EC and is characterised by elevated phosphate and organically bound nitrogen levels. Organic pollution is also problematic while salinity levels are elevated at times (calcium-based salinity).

B3.3 REACH 2 - ORANGE RIVER DS OF GARIEP DAM TO VAN DER KLOOF DAM (MRU A)

B3.3.1 Sample 1: Gariep Dam wall

This site was sampled during May 2008. The SPI score and ecological classification according to Van Dam *et al.* (1994) are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
1	Alkaliphilous	Eutrophic	Brackish-fresh	Moderate	Continuously high	7.8	D

The sample was dominated by AMSA which prefers organically enriched eutrophic waters as well as NIFR that usually occurs in electrolyte rich to brackish waters and are tolerant of highly polluted waters. Organically bound nitrogen was continuously elevated. Pollution levels were very high as the percentage pollution tolerant valves in the sample count were 69%.

B3.3.2 Sample 4 and 5: Seekoei River at DWA weir

This site was sampled during May 2008 (Sample 4) and August 2009 (Sample 5). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
4	Alkaliphilous	Meso- eutrophic	Fresh-brackish	Continuously high	Very little	11.8	С
5	Alkaliphilous	Unknown	Fresh-brackish	Continuously high	Elevated	13.4	В

The **April** 2008 sample was dominated by *Fragilaria* species which included FELL, FPIN and FCON. FELL is tolerant to moderate pollution but occurs in electrolyte rich or brackish waters (Taylor *et al.*, 2007b). FPIN tolerates moderate pollution (CRATICULA, 2010)³ while FCON occurs in standing waters of good quality. The presence of NIFR at 20% abundance indicated elevated salinity levels. Pollution levels were low at this site but there was some indication of industrial related impacts.

The **August** 2009 sample indicated that the biological water quality was good and had improved from April. The dominant genus was *Sellaphora*, but no identification could be made to species level. The community composition indicated a decrease in salinity, however the nutrient levels were somewhat higher. Pollution levels were very low at the site.

From the results of the samples it is evident that the biological water quality of the Seekoei River is good. Flows are very low at time with the river experiencing no flow conditions at times. Salinity may be problematic at times, and it is evident that under no flow conditions water temperatures become elevated.

B3.3.3 Sample 5 and 6: Vanderkloof Dam

This site was sampled at the Dam wall (Sample 5) and downstream of the dam (Sample 6) during May 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
5	Circumneutral	Hypereutrophic	Fresh- brackish	Very low	Continuously high	13.2	B/C
6	Alkaliphilous	Meso-eutrophic	Fresh- brackish	Continuously high	Very little	13.8	В

The biological water quality of Vanderkloof Dam is mainly impacted by agriculture, with waters consisting of continuously high nutrient levels. Pollution levels were overall low. Sample 5 was dominated by AMSA and the endemic, *E. archibaldii* indicating eutrophic waters with elevated electrolyte content (Taylor *et al.*, 2007b). Sample 6, which is located in the Orange River below the dam wall was in a B condition. Although *E. archibaldii* was still dominant there was a lower occurrence of species with a preference for elevated nutrient levels and it is assumed that multi-level releases are made from the dam or that pollutants settle to the bottom of the dam (longer residence time) and therefore the water entering the Orange River from the Vanderkloof Dam is of improved quality.

³ http://craticula.ncl.ac.uk/EADiatomKey/html/index.html

B3.4 OVERALL ASSESSMENT FOR REACH 2 (MRU A)

The diatoms indicated that Gariep Dam was eutrophic with elevated nutrient and salinity levels. The Seekoei River was in a good – moderate condition, which experiences no flow conditions at times. The water entering Vanderkloof Dam was mainly impacted by agriculture. The water was eutrophic and salinity levels were elevated. The Orange River in this reach has an estimated biological water quality of B/C. Nutrient, organic and salinity levels are elevated in this reach at times, however releases from Gariep Dam may dilute some of the pollution inputs in the Orange River between these two dams, depending on the type of release.

B3.5 REACH 3 - ORANGE RIVER DS OF VANDERKLOOF DAM TO CONFLUENCE WITH VAAL RIVER (MRU B)

B3.5.1 EFR O1

The EFR site is located at Hopetown and was sampled during June 2010. This site falls within MRU B. The SPI score and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR O1	Circumneutral	Indifferent	Fresh	Continuously high	Very little	15.7	В

Achnanthidium rivulare (ADRI) Potapova & Ponader was the dominant species in this sample and prefers well oxygenated waters and tolerates moderate nutrient and salinity levels (Taylor *pers. comm.*, 2010). The diatom community was also dominated by the presence of AMIN which occurs in well oxygenated waters (Taylor *et al.*, 2007b). These species are pioneer species and colonize rapidly and are also indicators of anthropogenic impact. This species had a dominance of 19% indicating low levels of disturbance (Acs, 2004). The high occurrence of this species also indicates elevated flows. Pollution tolerant species made up 4% of the sample indicating low pollution levels.

B3.5.2 Sample 2 and 9: Orange River at Marksdrift

This site is located approximately 75 km downstream of EFR O1 and 14 km upstream of the Vaal River confluence, within MRU B. This site was sampled during May 2008 (Sample 2) and August 2009 (Sample 9). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
2	Circumneutral	Meso- eutrophic	Fresh-brackish	Continuously high	Very little	15.3	В
9	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	13.4	В

The **May** 2008 sample was dominated by a variety of *Achnanthidium* species which prefer well oxygenated waters with high oxygen content. There was a high occurrence of FNAN and FCAP indicating that oxygenation was good and organically bound nitrogen levels were low (Taylor *et al.*, 2007b). Algal growth could have been present as these species usually attach to algae. It is suspected that the sample was taken under higher flow conditions than the August 2009 sample,

and that elevated flows may have had a dilution effect on pollution as presence of pollution tolerant valves in the count was estimated at 0.2%

The **August** 2009 sample indicated that flows were lower than in May 2008 as there was a drastic decline in *Achnanthidium* species. The diatom count was dominated by ESBM and NDIS. ESBM is common in strongly polluted electrolyte rich waters while NDIS indicates calcium based salinity (Taylor *et al.*, 2007b). Although the site was in a B EC the high occurrence of *Nitzschia* species indicate a highly polluted water body with readily available nutrients (Cholnoky 1968; van Dam *et al.*, 1994). Salinity levels were more elevated than in May 2008. Pollution tolerant valves also increased to 8% in the count.

B3.6 OVERALL ASSESSMENT FOR REACH 3

The main land use within this reach is irrigation farming. The reach is influenced by hydro-electric operation from Vanderkloof Dam. The water entering Vanderkloof Dam was mainly impacted by agriculture as the diatoms indicated that the water was eutrophic and salinity levels were elevated. However, due to top releases, the biological water quality entering the Orange River from this dam was a B and these releases have a dilution effect on pollution impacts. Further downstream up to the confluence with the Vaal River impacts are attenuated by increased flows. The biological water quality is in a B condition with calcium based salinity and elevated nutrients being problematic at times in this reach. The results may be overestimated due to the size of the Orange River and the large flow volumes, which may have a dilution effect of pollution.

B3.7 REACH 4 - UPSTREAM OF VAAL/ORANGE RIVER CONFLUENCE (MRU B)

B3.7.1 Sample 7: Riet River

This site was sampled during May 2008 in the Riet River below the Modder River confluence. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
7	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Continuously high	8.9	C/D

The diatom composition was characterised by species with a preference for electrolyte rich to brackish conditions e.g. PSAL, SMST, and SIDE. Nutrients are continuously elevated and organics are high. The percentage pollution tolerant valves present in the count were 32.5% indicating high pollution levels. This site is also impacted by the Modder River. Agricultural activities are the main impact in this river reach. Water temperatures may be elevated at times.

B3.7.2 Sample 8 and 1: Douglas bridge, Vaal River

This site was sampled during May 2008 (Sample 8) and August 2009 (Sample 1). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
8	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	9.9	С

1				1				4
	1	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	12.1	С

The **May** 2008 sample was dominated by ADEU, which prefers well oxygenated waters and eutrophic conditions with moderate pollution levels (Taylor *et al.*, 2007b). NIFR also dominated indicated elevated salinity. Overall the biological water quality was moderate (C EC) and the diatom community was indicative of waters impacted by a suite of anthropogenic activities. Salinity and nutrient levels were moderate.

The biological water quality during **August** 2009 was also moderate. Oxygen levels improved from May 2008. The dominant species was AMSA indicating eutrophic conditions. The presence of CPLA in high abundance indicates elevated nutrients. Pollution levels at these sites were moderate to low with most of the species tolerating moderately polluted conditions.

B3.8 OVERALL ASESSMENT FOR REACH 4 (MRU B)

The Vaal main stem entering the Orange River is of moderate biological water quality (C EC) and indicative of waters impacted by a suite of anthropogenic activities. The Riet River is impacted by farming activities and may contribute at times to elevated salinity, nutrient and organic levels in the Vaal main stem.

B3.9 REACH 5: VAAL/ORANGE RIVER CONFLUENCE TO PRIESKA (MRU B)

Two samples fall within this reach and is discussed below.

B3.9.1 Sample 10: Katlani

Site 10 is 22 km downstream of the Vaal River confluence and was sampled during May 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
10	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Very little	12.6	С

The sample was dominated by the centric species COCE. This species is found in the plankton of rivers, in meso- to eutrophic waters with an elevated pH (optimum pH 8.4) (Taylor *et al.*, 2007b). ENMI is found in oligotrophic waters with moderate electrolyte content (Taylor *et al.*, 2007b). There was a high occurrence (but not dominant) of AMIN indicating elevated flows providing well oxygenated waters. There were a wide range of diatoms indicating industrial impacts although these species were in low abundance.

B3.9.2 Sample 11: Prieska bridge

The site was sampled during May 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
11	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Continuously high	12.2	С

The diatom community was dominated by CPLE, which has a broad ecological range and is found in most running waters except where nutrients are low or acidic conditions prevail (Fore and Grafe, 2002) and AMSA which favours organically enriched eutrophic waters (Taylor *et al.*, 2007b). NIFR and RUNI also occurred in high abundance indicating elevated salinity levels and eutrophic conditions. RUNI is also an indicator of high turbidity levels (Taylor *et al.*, 2007b). This site is impacted by anthropogenic activities that include industry and sewage effluent. Organics and nutrient levels are high and may be problematic at this site.

B3.10 OVERALL ASSESSMENT FOR REACH 5

The biological water quality is a C. Although the water quality at Prieska is mainly due to the result of urban activities, the rest of the reach seems to have low nutrient levels as well as organics. Elevated flows attenuate pollution inputs in this reach.

B3.11 OVERALL ASSESSMENT FOR MRU B (REACH 3 – 5)

The biological water quality of MRU B is estimated as being in a C EC. Biological water quality in Reach 3 is primarily hydroelectric operations of Vanderkloof Dam. Reach 4 represents water entering the Orange River system from the Vaal River. Biological water quality is impacted in the Orange River downstream of the Vaal River confluence due to the accumulative effects of anthropogenic activities in the Vaal River catchment. MRU B deteriorates from a B EC before the confluence to a C EC after the confluence and the overall biological water quality for this reach is set at a C EC.

B3.12 REACH 6: PRIESKA TO BOEGOEBERG DAM (MRU C)

B3.12.1 Sample OR7

This site was sampled in July 2005 and is situated 33 km downstream of Prieska within MRU C. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 7	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Elevated	13.6	В

The sample was dominated by CTUM, CKPP, ECPM and FCVA. According to Taylor *et al.* (2007b) CTUM occurs in oligo- to mesotrophic water with moderate electrolyte content while ECPM requires oxygen rich environments of moderate electrolyte content. Generally the biological water quality was good with no serious pollution.

B3.13 OVERALL ASSESSMENT FOR REACH 6 (MRU C)

The reach is mostly an inaccessible gorge and there are little known activities. Only one sample in this reach was taken during 2005. The biological water quality is a B EC for this reach (low confidence). The diatom assessment indicates waters of good quality and low pollution levels. However this site is far downstream from Prieska and may not reflect the water quality related impacts of this Town.

B3.14 REACH 7: BOEGOEBERG DAM TO AUGRABIES (MRU D)

B3.14.1 Sample 12 and 29: Boegoeberg

This site is located at EFR O2 and was sampled during May 2008 (Sample 12) and August 2009 (Sample 29). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
12	Circumneutra I	Hyper eutrophic	Fresh- brackish	Continuousl y high	Elevated	12.9	B/C
29	Alkaliphilous	Eutrophic	Fresh- brackish	Continuousl y high	Very little	14.8	В

The sample taken in **May** 2008 was dominated by AMSA which favours organically enriched eutrophic waters and STMI usually found in strongly polluted waters with high electrolyte content (Taylor *et al.*, 2007b). ENMI was also dominant and is found in oligotrophic waters with moderate electrolyte content (Taylor *et al.*, 2007b). Organics and salinity may have been problematic at this site.

The **August** 2009 sample was dominated by ENMI as well as *Achnanthidium* species indicating well oxygenated waters (Taylor *et al.*, 2007b). Pollution levels were very low. STMI occurred at 16% abundance indicating that pollution did impact this site but due to the high abundance of *Achnanthidium* species, flows were elevated at this site diluting the pollution related impacts.

B3.14.2 EFR O2

The SPI score and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR O2	Alkaliphilous	Eutrophic	Fresh- brackish	Continuousl y high	Elevated	13.4	В

This site was sampled during **June** 2010 and the dominant species present in the sample were *Cymbella* species. CTGL and CTUM occurred at 12% and 6% dominance respectively while an unknown *Cymbella* species occurred at 10% dominance. CTGL and CTUM occur in oligo- to mesotrophic waters with moderate electrolyte content (Taylor *et al.*, 2007b). ENMI and GVNU were also in high abundance and have similar preferences as CTUM and CTGL (Taylor *et al.*, 2007b). The diatom community indicates that nutrients are elevated at times as well as salinity.

B3.14.3 Sample OR 11

This site was sampled in **July** 2005 and is at the same location as EFR O2. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 11	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Elevated	10.6	С

The sample was dominated by CINV, AMSA, CDUB and FELL. The dominance of CDUB indicates elevated chloride concentrations (Taylor *et al.*, 2007b). Nutrients were elevated at this site and salinity may have been problematic at times.

B3.14.4 Sample 13: Orange River at Gifkloof, 22 km US of Upington

The site was sampled during May 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
11	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	12.4	С

The sample was dominated by CPST, RCUR and STMI. STMI is usually found in strongly polluted waters with high electrolyte content while RCUR is found in electrolyte rich as well as brackish waters and is tolerant of critical levels of pollution (Taylor *et al.*, 2007b). No ecological information was available on CPST other than occurring in moderate water quality. Salinity levels may be problematic at this site.

B3.14.5 Sample 14 and 22: Orange River, at Upington waterworks

This site was sampled during May 2008 (Sample 14) and August 2009 (Sample 22). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
14	Alkaliphilous	Meso-eutrophic	Fresh- brackish	Continuously high	Very little	13.6	В
22	Alkaliphilous	Meso-eutrophic	Fresh- brackish	Continuously high	Very little	15.1	В

The sample in **May** 2008 was dominated by AMSA, ENMI, CPST and CPLA. AMSA favours organically enriched eutrophic waters and CPLA has a broad ecological range and is found in most running waters except where nutrients are low or acidic conditions prevail (Taylor *et al.*, 2007b). ENMI was also dominant and is found in oligotrophic waters with moderate electrolyte content (Taylor *et al.*, 2007b).

The **August** 2009 sample was dominated by ENMI, *Sellaphora* species and FPIN. Biological water quality was good and the majority of the species present at this site had a preference for good – moderate water quality.

As this site is located at the WWTW results may be skewed and not be representative of the impacts incurred by Upington.

B3.14.6 Sample 15: Kanon eiland

The site was sampled during May 2008 and is situated approximately 25 km downstream of Upington. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
15	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Elevated	13.1	B/C

The sample was dominated by CPST, FPIN and FELL. FELL is tolerant to moderate pollution but occurs in electrolyte rich or brackish waters (Taylor *et al.*, 2007b). FPIN tolerates moderate pollution (CRATICULA, 2010)⁴. Water quality was good although nutrient levels were elevated at times. There was some deterioration in biological water quality during 2008 in the section between Upington and Kanon eiland downstream. Nutrient levels increased in this section; however, the deterioration was not serious.

B3.14.7 Sample 16 and 23: Orange River at Neusberg weir

This site was sampled during May 2008 (Sample 14) and August 2009 (Sample 22). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
16	Alkaliphilous	Eutrophic	Fresh- brackish	Moderate	Elevated	11.9	С
23	Alkaliphilous	Eutrophic	Fresh- brackish	Continuously high	Elevated	14.6	В

The **May** 2008 sample was dominated by ADEU, CPST, and NLBT. NLBT occurs in very electrolyte rich to brackish waters while ADEU prefers well oxygenated waters and eutrophic conditions with moderate pollution levels (Taylor *et al.*, 2007b). 23% of the valves were pollution tolerant indicating that pollution levels were elevated. Organically bound nitrogen was elevated at times and salinity may be problematic at times.

The **August** 2009 sample had very low occurrences of pollution tolerant valves and the site improved from a C to a B EC. Dominant species included ADEU, ENMI, *Sellaphora* species, FPIN, DVUL and CPLE. Nutrients were elevated at this site while salinity and organics were moderate and did not seem to be problematic. It was evident that the agriculture in the area was impacting on the site to a certain degree.

B3.14.8 Sample OR 22

This site was sampled in **July** 2005 and is situated 4 km US of OR 20. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 7	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	10.7	С

The sample was dominated by CDUB, FTEN and RCUR. As with OR 11, chloride concentrations were elevated at this site. RCUR occurred in high abundance and is found in electrolyte rich as well as brackish waters and is tolerant of critical levels of pollution (Taylor *et al.*, 2007b). Nitrogen levels were elevated at times and there was evidence of organic pollution.

⁴ http://craticula.ncl.ac.uk/EADiatomKey/html/index.html

B3.14.9 Sample 18: Khamkirri

The site was sampled during **May** 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
18	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	12.7	С

The sample was dominated by ADEU, CPST and APED. APED is tolerant of critical levels of pollution (Taylor *et al.*, 2007b). Organically bound nitrogen levels were elevated and salinity may have been problematic at times.

B3.15 OVERALL ASESSMENT FOR REACH 7 (MRU D)

The biological water quality fluctuated between a B and C EC during 2005, 2008 – 2009, and 2010. It is evident that there is a gradual deterioration within the reach from Boegoeberg Dam to Augrabies. Nutrient levels are elevated throughout the reach and agriculture seems to be the major impact in this reach. Chloride concentrations were problematic during July 2005 in this reach. Although elevated at times organic pollution does not seem to be a major problem in this reach. Nutrients were elevated for all sampling years indicating continuous impact, while salinity may be problematic at times. The EC for this reach is a B/C.

B3.16 REACH 8: AUGRABIES FALLS TO VIOOLSDRIFT (MRU E)

B3.16.1 Sample 17 and 24: Orange River, Blouputs

This site was sampled during May 2008 (Sample 17) and August 2009 (Sample 24). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
17	Alkaliphilous	Eutrophic	Fresh- brackish	Continuously high	Elevated	12.7	С
24	Alkaliphilous	Unknown	Fresh- brackish	Continuously high	Elevated	14.4	В

The **May** 2008 sample was dominated by CINV, ESBM, NDIS, and NDME. ESBM occurs in electrolyte rich, strongly polluted water and CINV prefers waters of elevated electrolyte content. NDIS and NDME indicate calcium based salinity (Taylor *et al.*, 2007b).

From the **August** 2009 sample it was evident that salinity levels decreased substantially as there were very little species present with an affinity for electrolyte rich conditions. Dominant species included FGEO, PSBV, FPIN and *Sellaphora* species. According to Taylor *et al.* (2007b) PSBV occurs in moderately polluted waters as does FPIN (CRATICULA, 2010)⁵.

⁵ http://craticula.ncl.ac.uk/EADiatomKey/html/index.html

B3.16.2 EFR O3

The SPI score and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR O3	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	13.3	B/C

This site was sampled during **June** 2010 and the dominant species present in the sample were STMI, CPLA, SAGA and ESBM. STMI is usually found in strongly polluted waters with high electrolyte content, while SAGA is found in eutrophic waters with elevated electrolyte content and turbidity (Taylor *et al.*, 2007b). The presence of CPLA in high abundance indicates elevated nutrients. ESBM is common in electrolyte rich waters which are strongly polluted (Taylor *et al.*, 2007b). Pollution tolerant species made up 12% of the sample indicating that organic pollution was elevated at this site. The variable of concern at this site is nutrients and organics.

B3.16.3 Sample OR 19

This site was sampled in July 2005 and is approximately 90 km DS of EFR O3. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 19	Alkaliphilous	Eutrophic	Fresh-brackish	Continuously high	Elevated	11.4	С

The sample was dominated by CINV, CDUB, AMSA, FBRE, FTEN and FELL. The dominance of CDUB indicates elevated chloride concentrations (Taylor *et al.*, 2007b). Nutrients were elevated at this site and organic pollution may have been problematic at times.

B3.16.4 Sample 1 and 25: Orange River, Pella

This site was sampled during June 2008 (Sample 1) and August 2009 (Sample 25). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
1	Circumneutral	Eutrophic	Fresh- brackish	Moderate	Elevated	11.8	С
25	Alkaliphilous	Eutrophic	Brackish-fresh	Moderate	Continuousl y high	10.7	С

The **June** 2008 sample was dominated by CSPT, FPIN, STMI, and FTEN. STMI usually found in strongly polluted waters with high electrolyte content while FTEN is found in meso- to eutrophic waters and FPIN is found in clean waters with high electrolyte content (Taylor *et al.*, 2007b). Organically bound nitrogen levels were elevated and salinity levels were high.

The **August** 2009 sample was dominated by FGEO, NIFR, FPIN, *Sellaphora* species and ESOR. According to Taylor et al. (2007b), ESOR occurs in electrolyte rich to brackish conditions while NIFR also occurs in these conditions. Nutrient enrichment had increased and salinity levels were

high. Organic pollution levels were high with pollution tolerant species making up 39% of the sample.

B3.16.5 Sample OR 21

This site was sampled in July 2005 and is approximately 50 km US of EFR O4. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 21	Alkaliphilous	Meso- eutrophic	Fresh-brackish	Continuously high	Very little	10.8	С

The sample was dominated by FELL, FBRE, CDUB, FTEN, RCUR, and, AMSA. The dominance of CDUB indicates elevated chloride concentrations while RCUR is found in electrolyte rich as well as brackish waters and is tolerant of critical levels of pollution (Taylor *et al.*, 2007b). Salinity was elevated at this site and organic pollution may be problematic at times.

B3.17 OVERALL ASESSMENT FOR REACH 8 (MRU E)

The biological water quality for this reach is set at a C EC. During July 2005 chloride concentrations were problematic. Although elevated at times organic pollution does not seem to be a major problem in this reach, although during 2009 organic pollution increased drastically at Pella. Nutrients were elevated for all sampling years (except at OR 21 during 2005) indicating continuous impact, while salinity may be problematic at times. The EC for this reach is a C.

B3.18 REACH 9: VIOOLSDRIFT TO FISH RIVER CONFLUENCE (MRU F)

B3.18.1 Sample 2 and 26: Orange River, Vioolsdrift weir

This site was sampled during June 2008 (Sample 2) and August 2009 (Sample 26). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
2	Alkaliphilous	Meso-eutrophic	Fresh- brackish	Continuously high	Elevated	13.4	B/C
26	Alkaliphilous	Unknown	Fresh- brackish	Continuously high	Elevated	13.0	B/C

The sample in **June** 2008 was predominantly dominated by *Fragilaria sundayensis*. Other dominant species included STMI, FPIN and FELL. Organic nitrogen levels were periodically elevated at this site. There was evidence of organic pollution and elevated salinity.

The **August** 2009 sample was dominated by FGEO, FTEN, FPIN and *Sellaphora* species. The site remained stable although salinity and organic input had slightly deteriorated.

B3.18.2 EFR O4

The SPI score and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR O4	Alkalibiontic	Eutrophic	Fresh- brackish	Moderate	Elevated	11.4	С

This site was sampled during **June** 2010 and the dominant species present in the sample were STMI, CPLA, SAGA and ESBM. STMI is usually found in strongly polluted waters with high electrolyte content, while SAGA is found in eutrophic waters with elevated electrolyte content and turbidity (Taylor *et al.*, 2007b). The presence of CPLA in high abundance indicates elevated nutrients. ESBM is common in electrolyte rich waters which are strongly polluted (Taylor *et al.*, 2007b). Pollution tolerant species made up 17% of the sample indicating that organic pollution was elevated at this site and had increased from EFR O3. The variable of concern at this site is nutrients. *Coscinodiscus devius* was present at this site indicating that salinity is of major concern at this site. This species is a marine species and was not noted in any of the other samples during 2005, 2008 – 2010. There is a possibility that this site could be influence by backup from the Orange River mouth, although this is unlikely. The presence of this species in a fresh water system is of major concern and indicates critical impact. These species could however also be present due to the hot springs in the area (Dr Hugo Bezuidenhout, *pers. comm.*) and further monitoring will be needed to clarify this issue.

B3.18.3 Sample OR 2

This site was sampled in July 2005 and is approximately 34 km DS of EFR O4. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
OR 2	Alkaliphilous	Meso- eutrophic	Fresh-brackish	Continuously high	Very little	10.5	С

The sample was dominated by FELL and FBRE. There were only 4 species in this sample and therefore an accurate analysis is very difficult. The two dominant species however did indicate that salinity levels were high at this site.

B3.19 OVERALL ASSESSMENT FOR REACH 9 (MRU F)

The biological water quality of this reach is a C EC. Elevated nutrient levels are of concern as well as salinity. Although still to be verified the presence of *Coscinodiscus devius* indicates that salinity levels have increased drastically since 2009 and is of major concern.

B3.20 REACH 10: FISH RIVER CONFLUENCE TO ESTUARY

B3.20.1 SAMPLE 4 AND 27: Sendlingsdrift

This site was sampled during June 2008 (Sample 4) and August 2009 (Sample 27). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
4	Alkaliphilous	Hypereutrophic	Fresh- brackish	Moderate	Elevated	10.1	С

27	Alkaliphilous	Eutrophic	Fresh- brackish	Continuousl y high	Continuousl y high	10.1	С

During **June** 2008 the sample was dominated by *Fragilaria sundayensis*, NPAL, SBRE, CPST and STMI. NPAL occurs in eutrophic and heavily to extremely polluted waters while SBRE can occur in brackish waters (Taylor *et al.*, 2007b). Organics seemed elevated along with nutrients.

During **August** 2009 the sample was dominated by FGEO, ENMI, *Sellaphora* species, NIFR, FTEN and AMSA. The site remained stable although there was a sharp increase in nutrient levels as well as an increase in salinity.

B3.20.2 Sample OR 8

This site could not be assessed as the valve count was not viable due to high flows.

B3.20.3 Sample 3: Brandkaros

This site was sampled in June 2008. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
3	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	12.4	С

The sample was dominated by *Fragilaria sundayensis*, FELL, FPIN, SAGA and STMI. Nutrients are elevated at this site. The presence of SAGA indicated elevated turbidity (Taylor *et al.*, 2007b).

B3.20.4 Sample 5: Alexander Bay

This site was similar to Sample 3 – Brandkaros. The dominant species were the same and the EC resulted in a C EC with a SPI score of 10.9.

B3.21 OVERALL ASSESSMENT FOR REACH 9

The EC of this reach is a C EC. Nutrient levels are elevated at times.

B4 KRAAI RIVER DIATOM RESULTS

B4.1 SAMPLE 1 AND 11: UPSTREAM OF EFR K7

This site was sampled during April 2008 (Sample 1) and August 2009 (Sample 11). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
1	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Continuously	8.9	C/D
11	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	13.1	B/C

The **April** 2008 sample was dominated by species preferring high organic levels and elevated salinity (NDRA, NACI, NIFR and RUNI). The presence of RUNI in high abundance is an indication of elevated turbidity (Taylor *et al.*, 2007b). Salinity was elevated at this site and this is evident by the dominance of NIFR that usually occurs in electrolyte rich to brackish waters and are tolerant of highly polluted waters. Pollution tolerant valves made up 65% of the count indicating that this site was highly polluted. Nutrients and organics are problematic.

The **August** 2009 sample indicated a marked improvement in biological water quality. Organic pollution levels reduced to a great extent with the presence of pollution tolerant valves decreasing to 8%. Nutrient and organic loads decreased but salinity was still elevated. NZLT was the most abundant species while NDIS was also dominant indicating calcium-based salinity (Taylor *et al.*, 2007b). *Cocconeis* species were present in abundances above 5% indicating elevated nutrient levels as these species have a broad ecological range and is found in most running waters except where nutrients are low or acidic conditions prevail (Taylor *et al.*, 2007b). The presence of RUNI in high abundance indicates high turbidity and nutrient levels.

B4.2 EFR K7

EFR K7 was sampled during July 2010. The SPI score and ecological classification according to Van Dam *et al.* (1994) are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR K7	Alkaliphilous	Eutrophic	Fresh-brackish	Continuousl y high	Elevated	12.6	С

The sample was dominated by FELL indicating high electrolyte content extending into brackish conditions (Taylor *et al.*, 2007b). As with the August 2009 sample NDIS was dominant indicating calcium-based salinity. Although CPLA and *Nitzschia* species were not dominant the presence of these species indicates elevated nutrients and strongly polluted waters. Pollution levels were low at the site. Turbidity levels seemed to recede as no indicators of turbid conditions were present. There is evidence that water temperatures may be elevated at time.

B4.3 OVERALL ASSESSMENT OF THE KRAAI RIVER

From the range of samples assessed it is evident that organic pollution and elevated nutrient levels are problematic in this reach. Calcium-based salinity is present and it seems that the river is very turbid at times. The overall EC of this reach in terms of biological water quality is a C.

B5 CALEDON RIVER DIATOM RESULTS

B5.1.1 EFR C5: MRU A/B

EFR C5 is located in MRU A/B upstream of the Little Caledon confluence. The site was sampled during July 2010 and the SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR C5	Alkaliphilous	Mesotrophic	Fresh-brackish	Continuously high	Elevated	14.2	В

The dominant species were NDIS, CPLA, CPLE, FCVA, RUNI and *Gomphonema* species. The occurrence of these species in high abundance indicates elevated nutrient levels as well as elevated turbidity and calcium-based salinity (Taylor *et al.*, 2007b). The physico-chemical data indicated that salinity and nutrient levels are naturally elevated. Pollution levels are elevated at this site than at site 20 and organic pollution may be problematic. The biological water quality however seemed in a good condition as the reach upstream of the site is situated in the Golden Gate Nature Reserve.

B5.1.2 Sample 20: Little Caledon: MRU B

This site was sampled during April 2009. The site is in the upper reaches of the little Caledon, in the Golden Gate National Reserve upstream of Clarence and enters the Caledon River in MRU B. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
20	Alkaliphilous	Mesotrophic	Fresh-brackish	Moderate	Elevated	14	В

A high abundance of CPLE occurred indicating elevated nutrient levels. The dominance of NANT indicated anthropogenically impacted waters as this species is a good indicator of these conditions (Taylor *et al.*, 2007b). GMIN is found in eutrophic waters, however this species does not tolerate more than moderate pollution (Taylor *et al.*, 2007b) and indicates that this site had low pollution levels. Elevated nutrient levels may be due to animal activities in the area. The biological water quality at this site was overall in a good condition, which is expected as the site is located in the Golden Gate Reserve.

B5.1.3 Sample 18 and 20: Little Caledon at Caledonpoort: MRU B

This site was sampled twice; during April 2008 (Sample 18) and August 2009 (Sample 20). The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
18	Alkaliphilous	Eutrophic	Fresh-brackish	Moderate	Elevated	13.2	B/C
20	Alkaliphilous	Eutrophic	fresh-brackish	Moderate	Elevated	14.2	В

The April and August sample was dominated by CPLE indicating elevated nutrient levels. During **April** 2008, EOMI, and GPUM also dominated while NTPT and CMOL were present in high abundance although not dominant. The presence of these species indicates elevated organics in the system. The **August** 2009 sample indicated increased calcium-based salinity as well as elevated turbidity levels.

From the samples assessed it seems as if the Little Caledon River has good biological water quality (B EC). In the upper reaches of the river nutrients and organics are elevated. While the lower reaches are similar, turbidity and salinity become elevated as well.

B5.1.4 Sample 19 and 21: Caledon River at Caledonpoort: MRU B

The site is situated in MRU B at the confluence with the Little Caledon River and was sampled during April 2008 and August 2009. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
19	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Periodically elevated	7.9	D
21	Alkaliphilous	Eutrophic	fresh-brackish	Moderate	Elevated	14.3	В

The **April** 2008 sample indicated that the site was heavily impacted as the dominant species present in the sample were EOMI, MAPE and FSAP. These species are of the most pollution tolerant species and it is evident that organic pollution levels were very high. 53.2% of the sample consisted of pollution tolerant species, and pollution levels were critical. A possible source of the pollution could be pesticide use as the main land use in the area is agriculture and subsistence farming.

The **August** 2009 indicated good biological water quality. The dominant species present were CPLE, EOMI and NANT. These species indicate anthropogenically impacted waters with elevated nutrient and organic levels. The dominance of RSIN indicated a recent flushing event that aleviated some of the pollution levels in the river. The impact of fertilizer and pesticide use is of concern at this site, and that these activities impact the mainstem Caledon, as pollution levels in the Little Caledon are low and play an important role of attenuating the effects of farming practices in the Caledon River.

B5.1.5 Sample 17 and 19: Caledon at Grootspruit road bridge: MRU B

This site is situated in MRU B above the Meulspruit confluence. The results of the April 2008 (Sample 17) and August 2009 (Sample 19) samples are provided below along with the SPI scores and ecological classification according to Van Dam *et al.* (1994):

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
17	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	12.9	B/C
19	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	16.3	В

Dominant species in the **April** 2008 sample was similar to the previous sites discussed and included EOMI, NTPT, NDIS, GPAR and CINV. NTPT is a good indicator of eutrophic conditions while GPAR and EOMI prefer organic rich conditions (Taylor *et al.*, 2007b). Calcium-based salinity is elevated. These waters are typical of agriculturally impacted waters.

Conditions were improved during **August** 2009 and the dominant species was NTPT, NDIS and DVUL indicating eutrophic water with elevated electrolyte and organic pollution.

B5.1.6 Sample 16: Meulspruit above Meulspruit Dam: MRU B

This site was sampled during April 2009. The site is in the Meulspruit above the Meulspruit Dam. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
16	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Periodically elevated	10.5	С

The site was dominated by EOMI, SSEM, NPAL and *Nitzschia* species. Surrounding agriculture is impacting on this site and from the dominant species it was evident that organic pollution was high as there was a 29% presence of pollution tolerant species in the sample. The high occurrence of *Nitzschia* species indicated a highly polluted water body with readily available nutrients (Cholnoky 1968; van Dam *et al.*, 1994).

B5.1.7 Sample 15: Below Ficksburg: MRU B

This site was sampled during April 2009. The site is below Ficksburg in RU B. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
15	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Periodically elevated	6.1	D

The impact of Ficksburg was evident from the diatom community composition. Dominant species included CMOL, EOMI, FSAP and MAPE and is characteristic of unsewered, industrialised waters (Taylor *et al.*, 2007b). 65% of the sample count was pollution tolerant species indicating that organic pollution is severe at this site and nutrient and salinity levels are high.

B5.1.8 Sample 14: Mopeli: MRU B

This site was sampled during April 2009. The site is below Ficksburg in RU B. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
14	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Periodically elevated	8.3	D

The dominant species were similar to upstream sites within the Caledon River with EOMI and a variety of *Nitzschia* species being dominant. Organic pollution was high (64% pollution tolerant species present in sample) and nutrient levels were problematic.

B5.1.9 Sample 13 and 17: Caledon below Maseru: MRU B

This site was sampled in April 2008 and August 2009 and is situated in MRU B. Details regarding the ecological classification according to Van Dam *et al.* (1994) are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
13	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Periodically elevated	6.5	D
17	Alkaliphilous	Eutrophic	Fresh-brackish	Low	Continuously elevated	7.1	D

This site is situated below Maseru and the diatom community composition indicates waters that are impacted severely by anthropogenic activities. The waters were characteristic of sewage and industrial related impacts. The **April** 2008 sample was dominated by EOMI, CMLF, FSAP, MAAT and TPSN. TPSN indicates high salinity as it is a halophilic species while the rest of the species indicate high organic and nutrient loading (Taylor *et al.*, 2007b). Pollution levels were critical during April 2008. The **August** 2009 sample was dominated by EOMI, ESBM and MAPE. The SPI score slightly, although within the same EC and pollution levels improved markedly.

B5.2 OVERALL ASESSMENT FOR CALEDON MRU B

The upper reaches of the Caledon within MRU B has good water biological water quality. This is mostly due to little anthropogenic activities in this reach and the contribution of clean waters from the Little Caledon. From the Meulspruit confluence there is a gradual deterioration in biological water quality of the Caledon River. Agriculture seems to be the major impact on the Caledon, and it is evident that the use of fertilizers and pesticides are impacting the river. Biological water quality in the Meulspruit is moderate and organic pollution and elevated nutrient levels are present.

The impacts of Ficksburg and Maseru have an accumulative effect on the Caledon and the biological water quality deteriorates rapidly. Sewage and industrial effluent are the major impact between these two towns. As farming intensifies below Maseru the impact of this land use becomes evident. The Caledon is characterised by high – critical pollution levels and nutrient and phosphate levels are elevated most of the time in the lower reaches of MRU B. Of concern is increasing salinity levels as well as high turbidity levels. Salinity seems to be mainly calcium-based. The overall biological water quality was determined to be a C.

B5.2.1 Sample 11: Caledon at Tienfontein pumpstation: MRU C

Site 11 was sampled during April 2008 and is in MRU C upstream of EFR C6. No results could be generated for this site as the slide was non-viable, most probably due to high flows.

B5.2.2 EFR C6: MRU D

This site was sampled during July 2010 and is in MRU D. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the sample is provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
EFR C6	Alkaliphilous	Mesotrophic	Fresh-brackish	Fairly high	Very little	19.2	А

The diatom community was dominated by the presence of *Achnanthidium* species, especially ADBI which occurs in oligo- to mesotrophic water with moderate to elevated electrolyte content (Taylor *et al.*, 2007b). It is assumed that flows were very high during sampling, as *Achnathidium* species are pioneer species and colonize rapidly. They are also indicators of anthropogenic impact. This species had a dominance of 81% indicating high levels of disturbance (Acs, 2004). The high occurrence of this species also indicates elevated flows or a recent flushing event and as this site

is below Welbedacht Dam elevated flows due to releases are highly likely. There were very little other species present in the sample, and therefore the SPI is not a true reflection of the usual conditions at the site. Other species present were NTPT, *Cocconeis* spp. NDIS and MAPE, indicating that nutrients, organics and salinity are variables that could impact the site at lower flows as these species occurred in all upstream sites at higher abundance. It is expected that this site is in a C EC or lower but due to elevated flows or regular flushing events the effects of local impacts are diluted.

B5.2.3 Sample 12 and 16: Leeuspruit at Hobhouse: MRU D

This site was sampled in April 2008 and August 2009 and is situated in Leeuspruit which is a tributary of the Caledon River and enters the Caledon in MRU D. Details regarding the ecological classification according to Van Dam *et al.* (1994) are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
12	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	14.4	В
16	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Elevated	13.7	В

During **April** 2008 the biological water quality was good. The main activity in the area is agriculture and it seems as if pesticide and fertilization use is impacting heavily at this site as organic pollution is severe and salinity is a major concern at this site. Dominant species were NCLA, NIFR and NDIS is indicative of problematic salinity levels while the dominant species, NSSY and NPAL indicate organic pollution.

The biological water quality during August was similar to April 2008. Organic pollution and salinity was still very problematic with dominant species, NDIS, APEL and NDIS, indicating salinity related problems (calcium-based) and EOMI and NLIB, indicating severe organic pollution.

It is important to note that organic pollution levels were critical at this site during both sampling occasions and that elevated flows may account for the good biological water quality scores.

B5.3 OVERALL ASESSMENT FOR CALEDON MRU C and D

SAMPLE 11 in MRU C was non-viable and therefore no high confidence results for MRU C could be generated. It is assumed that high flows were the reason for non-viable diatom slides as this site is situated at Tienfontein pumpstation. The diatom assemblage at EFR C6 in MRU D also indicated elevated flows and although the SPI score indicated the site was in an A EC, this might not be a true reflection of current conditions. The dominant species is an indicator of anthropogenic disturbance and at an abundance of 88% this site is exposed to high levels of disturbance. Other species present indicated that nutrients, organics and salinity are variables that could impact these sites, but due to releases from Welbedacht Dam these impacts are ameliorated. It is estimated that the biological water quality condition of MRU C and D is in a C EC, although this is a low confidence determination.

B5.4 OVERALL ASESSMENT FOR CALEDON RIVER

The biological water quality for the Caledon River was set at an overall C/D. The major determinants for this assessment was the high levels of organic pollution throughout reaches A and B, and elevated nutrient levels in the lower sections of MRU B. The prevalence of calcium-

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based salinity also contributed to the overall assessment. It should be noted that turbidity can get excessively high at times in this reach.

B6 MOLOPO RIVER DIATOM RESULTS

B6.1 EFR M8: MRU 8

During April 2008 EFR M8 was sampled at three different places within the wetland. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for the two samples are provided below:

Sample	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
Bosbokpark	Alkaliphilous	Mesotrophic	Fresh-brackish	Continuously high	Elevated	17.4	А
Flow gauge	Circumneutral	Oligotrophic	Fresh	Continuously high	Very small	17.3	А
Molo Masu	Alkaliphilous	Eutrophic	Fresh-brackish	Fairly high	Very small	9	С

Bosbokpark and the Flow gauge had a similar community assemblage with dominance of *Achnanthidium* species which included ADBI, AMIN, ACHS and ACAF. These species prefer clean well oxygenated waters (Taylor *et al.*, 2007b). These two sites had high SPI scores indicating very good water quality. Oxygen levels were high and nutrient and organic levels were low. Molo Masu was more impacted than the other two sites within the wetland, and this may be due to a very localised impact (e.g. groundwater feed, or pipeline) or the wetland ameliorates polluted water entering the wetland very well. Dominant species included NAGW, MSMI, ESOR and NDRA. MSMI usually occurs in brackish biotopes while NAGW and NDRA occur in trophic waters (Taylor *et al.*, 2007b). Based on the species composition of all three samples salinity and nutrients are elevated at times but overall the samples represent a well functioning wetland. It could be that surrounding agriculture is impacting to some extent on the wetland due to elevated salinity and nutrient levels at times although the wetland ameliorates these impacts.

The biological water quality was determined as an A/B EC.

B6.2 MOL 1: (D4MOLO-BUHRM) MRU B

During 2005 RHP site D4MOLO-BUHRM was sampled during July 2005 as part of a PhD study. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for this site is provided below:

Date	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
May 2005	Alkaliphilous	Meso-eutrophic	Fresh- brackish	Continuousl y high	Elevated	18.9	А

The SPI score was very high (18.9) indicating very good water quality conditions. Dominant species included AMIN, ELAC and NRCH. AMIN is a pioneer species and colonize rapidly and they are also indicators of anthropogenic impact. This species had a dominance of 39% indicating low levels of disturbance (Acs, 2004). The dominance of this species also indicates elevated flows or a recent flushing event and therefore the SPI score may not be a true reflection of the current conditions. NRCH was also dominant and this species is common in eutrophic, and especially calcareous waters and tolerant of critical pollution levels (Taylor *et al.*, 2007b). It is also an indicator species for these conditions. Other species present in low abundance indicates that organic, nutrient and salinity levels may become elevated and problematic at this site.

It is very difficult to provide an overall biological water quality EC to MRU B based on one survey. As the diatom community indicates the probability of elevated flows at the time of sampling the SPI score may not be a true reflection of conditions at the site. Due to the type of land use in the area and the presence of pollution tolerant species, albeit in low abundance the biological water quality for this reach was determined to be a B/C although the confidence in this assessment is very low.

B6.3 MOL 2 (D4MOLO-MAFIK) AND MOL 3 (D4MOLO-LOMAN) MRU C

B6.3.1 MOL 2 (D4MOLO-MAFIK)

During 2005 RHP site D4MOLO-Mafik was sampled during May, July and September 2005 as part of a PhD study. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for this site during the relative months are provided below:

Date	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
May 2005	Circumneutral	Hyper eutrophic	Fresh- brackish	Low	Continuous	1.5	Е
Jul 2005	Alkaliphilous	Eutrophic	Fresh- brackish	Low	Continuous	1.5	Е
Sep 2005	Alkaliphilous	Eutrophic	Fresh- brackish	Low	Periodically elevated	3.9	Е

This site is situated in the Mafikeng residential area. It is expected that the biological water quality at any site within an urban area will be low due to various anthropogenic activities. The three samples were very similar and there was a notable absence of species with a preference for moderate – good water quality. The species present are typical of urban waters. Dominant species included NPAE and NVEN in high abundance while the rest of the species are tolerant to high – critical levels of pollution, salinity and nutrient levels. The SPI scores were similar for May and July and improved slightly during September. At SPI scores this low the water is critically polluted which will impact severely on biota. Major impacts relate to sewage and industrial effluent and therefore organic pollution as the % pollution tolerant valves present in the May and September sample were higher than 80% indicating critical levels of pollution. As the SPI scores were relatively constant from May to Sep indications are that impacts are continuous and that the river does not recover from these impacts at this site. This site is critically impacted.

B6.3.2 MOL 3 (D4MOLO-LOMAN)

During 2005 RHP site D4MOLO-Lodim was sampled during May, July and September 2005 as part of a PhD study. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for this site during the relative months are provided below:

Date	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
May 2005	Alkaliphilous	Eutrophic	Fresh- brackish	Moderate	Elevated	5.5	D
Jul 2005	Alkaliphilous	Eutrophic	Fresh- brackish	Low	Periodically elevated	6.5	D
Sep 2005	Alkaliphilous	Eutrophic	Fresh- brackish	Low	Elevated	3.9	Е

This site is situated downstream of MOL 2 in Mafikeng. The biological water quality ranged between a D and E EC during May to September 2005. Dominant species included APED, EOMI, NAMP, NPAL, GPAR and NVEN. These species indicate organic related impacts which are urban related. This site is slightly better than the upstream site and there are indications of some

recovery from water quality related impacts. Pollution tolerant biota would occur under these conditions, although they would be stressed.

The major impact on this reach is Mafikeng and the anthropogenic related impacts one would find in an urban setting. The two sites are located in Mafikeng and the diatom communities indicate a highly impacted river reach. MOL 2 is critically impacted while MOL 3 is slightly improved with MOL 3 being slightly less organically polluted as the pollution tolerant valves decreased to 40% during May and September but increasing to 63% in July. The overall biological water quality for MRU C is a D/E with organics and nutrients being very problematic. Toxicants are expected.

B6.4 MOL 4 (D4MOLO-MODIM) MRU B

During 2005 RHP site D4MOLO-MODIM was sampled during May, July and September 2005 as part of a PhD study. The SPI scores and ecological classification according to Van Dam *et al.* (1994) for this site during the relative months are provided below:

Date	рН	Trophy	Salinity	Oxygen	Nitrogen metabolism	SPI	EC
May 2005	Alkaliphilous	Hypereutrophic	Fresh- brackish	Low	Continuous	5.3	D
Jul 2005	Alkaliphilous	Eutrophic	Brackish-fresh	Moderate	Continuous	7.2	D
Sep 2005	Alkaliphilous	Indiferent	Fresh- brackish	Continuousl y high	Elevated	12.2	С

This site is below Modimola Dam and the May and July sample was dominated by AMIN indicating elevated flows that may have been due to releases from the dam. NIFR occurred as dominant species in all three samples while NSSY and NPAL were dominant in the September sample. September saw an improvement in biological water conditions mainly due to a decrease in organic pollution as pollution tolerant valves made up 20% of the sample while organic pollution levels were very high during May and Jul (more than 70%). Of concern is that although flows were elevated during May and Jul organic pollution was extremely high indicating that the water quality in Modimole Dam may not be good. It seems that organics and nutrients as well as salinity may be problematic in this reach.

Based on this sample the biological water quality of MRU C was a C/D although the assessment is of low confidence.

B6.5 OVERALL BIOLOGICAL WATER QUALITY OF THE MOLOPO

Physico-chemical data is scare for this system and the confidence in this assessment is low as no seasonal data was available. The major impact in this river is Mafikeng and the river is critically impacted in MRU C. Organic pollution is critical while nutrient are a major concern. There is a systematic deterioration in the River towards Modimole Dam. Modimole Dam receives bad water quality although this is an observation as no physico-chemical data was available for the dam. Below Modimole Dam biological water quality conditions improve slightly, probably due to turnover time in the dam. Although no data was available for the reaches below D4MOLO-MODIM the overall biological water quality for the Mokolo was set as a D EC.

B7 SPECIES LISTS

The species lists of the samples collected during the 2010 EFR study is provided below.

B7.1 ORANGE RIVER MAIN STEM

Species	Abbr.	EFR O1	EFR O2	EFR O3	EFR O4
Achnanthes species	ACHS	10	17	13	0
Achnanthidium biasolettianum (Grunow) Lange-Bertalot	ADBI	8	8	0	0
Achnanthidium eutrophilum (Lange-Bertalot) Lange-Bertalot	ADEU	3	10	0	0
Achnanthidium rivulare Potapova & Ponader	ADRI	198	0	0	0
Achnanthes lanceolata ssp. frequentissima var. rostrata (Oestrup) Round & Bukhityarova	ALAR	0	2	0	1
Planothidium frequentissima (Lange-Bertalot) Round & Bukhityarova	ALFR	2	0	0	0
Achnanthes minutissima Kützing	AMIN	77	9	0	1
Achnanthes minutissima var. saprophila Kobayasi & Mayama	AMSA	2	3	3	0
Amphora ovalis (Kützing) Kützing	AOVA	0	1	0	0
Amphora pediculus (Kützing) Grunow	APED	0	2	4	0
Aulacoseira granulata var. angustissima (O Müller) Simonsen	AUGA	0	7	0	3
Aulacoseira granulata (Ehrenberg) Simonsen	AUGR	0	4	2	4
Cyclostephanos dubius (Fricke) Round	CDUB	0	3	8	17
Craticula halophila (Grunow) DG Mann	CHAL	0	1	0	0
Cyclostephanos invisitatus (Hohn & Hellerman) Theriot, Stoermer & Håkansson	CINV	0	0	17	13
Cymbella kappii Cholnoky	CKPP	1	0	0	0
Cyclotella meneghiniana Kützing	CME N	0	0	0	3
Craticula molestiformis (Hustedt) Lange-Bertalot	CMLF	1	0	0	0
Cyclotella ocellata Pantocsek	COCE	1	5	0	1
Coscinodiscus sp.	COSS	0	0	0	10
Cocconeis pediculus Ehrenberg	CPED	0	1	0	0
Cocconeis placentula Ehrenberg	CPLA	2	14	95	21
Cocconeis placentula var. euglypta (Ehrenberg) Grunow	CPLE	0	10	6	0
Cyclotella pseudostelligera Hustedt	CPST	0	0	1	0
Cymbella turgidula Grunow	CTGL	2	49	0	0
Cymbella tumida (Brébisson) Van Heurck	CTUM	1	27	0	0
Cymbella species	CYMS	0	42	0	0
Diatoma vulgaris Bory	DVUL	0	1	0	0
Encyonopsis minuta Krammer & Reichardt	ECPM	0	28	1	1
Encyonopsis microcephala (Grunow) Krammer	ENC M	2	0	0	0
Encyonopsis leei var. sinensis Metzeltin & Krammer	ENLS	7	0	1	0
Encyonema minutum (Hilse) DG Mann	ENMI	5	4	0	0
Eolimna minima (Grunow) Lange-Bertalot	EOMI	6	0	0	0
Eolimna subminuscula (Manguin) Lange-Bertalot	ESBM	2	1	35	24
Fragilaria biceps (Kützing) Lange-Bertalot	FBCP	0	12	0	0
Fragilaria capucina Desmazières	FCAP	0	5	0	1
Fragilaria capucina var. vaucheriae (Kützing) Lange-Bertalot	FCVA	0	3	0	0
Fragilaria geocollegarum Witkowski	FGEO	0	0	0	10
Fragilaria pinnata Ehrenberg	FPIN	0	0	4	0
Fragilaria tenera (WM Smith) Lange-Bertalot	FTEN	0	3	0	0
Fragilaria ulna var. acus (Kützing) Lange-Bertalot	FUAC	0	5	0	4
Gomphonema affine Kützing	GAFF	0	1	0	1

Species	Abbr.	EFR O1	EFR O2	EFR O3	EFR O4
Gomphonema angustatum (Kützing) Rabenhorst	GAN	1	0	0	0
Geissleria decussis(Hustedt) Lange-Bertalot	G GDEC	0	0	0	4
Gomphonema minutum (Agardh) Agardh	GMIN	5	0	2	0
	GOM	18			
Gomphonema species	S		19	2	1
Gomphonema parvulum (Kützing) Kützing	GPAR	0	3	0	1
Gomphonema pseudoaugur Krammer	GPSA GPU	1	0	0	0
Gomphonema pumilumvar. rigidum Reichardt & Lange-Bertalot	M	2	0	0	0
Gomphonema venusta Passy, Kociolek & Lowe	GVNU	10	29	0	0
Hippodonta capitata (Ehrenberg) Lange-Bertalot Metzeltin & Witkowski	HCAP	0	1	0	0
Melosira varians Agardh	MVAR	0	1	0	0
Navicula amphiceropsis Lange-Bertalot & Rumrich	NAAM	0	7	0	0
Navicula adamantiformis Archibald	NADF	0	0	0	11
Nitzschia agnewii Cholnoky	NAG W	1	0	0	0
Nitzschia amphibia Grunow	NAMP	0	1	0	0
Navicula antonii Lange-Bertalot	NANT	0	1	0	0
Navicula species	NASP	2	0	0	5
Navicula cryptocephala Kützing	NCRY	3	0	0	0
Navicula cryptotenella Lange-Bertalot	NCTE	1	3	5	1
Nitzschia dissipata (Kützing) Grunow	NDIS	1	2	0	0
Nitzschia draveillensis Coste & Ricard	NDRA	1	0	0	0
Nitzschia fonticola Grunow	NFON	1	1	0	2
Navicula heimansioides Lange-Bertalot	NHM S	9	0	0	0
Nitzschia frustulum (Kützing) Grunow	NIFR	0	6	7	1
Navicula libonensis Schoeman	NLIB	0	2	3	0
Nitzschia linearis (Agardh) W Smith	NLIN	1	3	1	4
Nitzschia linearisvar. subtilis(Grunow)	NLSU	0	0	1	0
Navicula microrhombus (Cholnoky) Schoeman and Archibald	NMC B	0	0	5	9
Nitzschia palea (Kützing) W. Smith	NPAL	0	0	0	3
Nitzschia recta Hantzsch	NREC	0	0	0	4
Navicula schroeteri Meister	NSHR	1	0	0	0
Nitzschia sinuata var. tabellaria (Grunow) Grunow	NSIT	0	0	0	1
Navicula schroeteri var. symmetrica (Patrick) Lange-Bertalot	NSSY	0	3	0	0
Navicula tenelloides Hustedt	NTEN	0	1	2	2
Nitzschia umbonata (Ehrenberg) Lange-Bertalot	NUM B	1	0	0	0
Nitzschia species	NZSS	2	11	2	24
Placoneis dicephala (W Smith) Mereschkowsky	PDIC	0	1	0	3
Rhoicosphenia curvata (Kützing) Grunow	RCUR	1	0	0	0
Rhopalodia gibba (Ehrenberg) O Müller	RGIB	0	1	0	1
Reimeria sinuata (Gregory) Kociolek & Stoermer	RSIN	2	0	0	0
Reimeria uniseriata Sala Guerrero & Ferrario	RUNI	0	2	8	0
Stephanodiscus agassizensis Håkansson & Kling	SAGA	0	4	60	110
Surirella angusta Kützing	SANG	0	4	0	0
Stephanodiscus hantzschii Grunow	SHAN	0	0	3	4
Sellaphora seminulum (Grunow) DG Mann	SSEM	5	0	0	1
Stephanodiscus minutulus (Kützing) Cleve and Möller	STMI	1	12	98	89
Tryblionella apiculata Gregory	ΤΑΡΙ	0	0	0	4
Tryblionella hungarica (Grunow) DG Mann	THUN	0	0	1	0
Thalassiosira pseudonana Hasle & Heimdal	TPSN	0	0	7	0

Species	Abbr.	EFR O1	EFR O2	EFR O3	EFR O4
Thalassiosira weissflogii (Grunow) Fryxell & Hasle	TWEI	0	0	3	0

B7.2 ORANGE RIVER TRIBUTARIES: KRAAI AND CALEDON RIVERS

Species	Abbr.	EFR K7	EFR C5	EFR C6
Achnanthidium affine (Grunow) Czarnecki	ACAF	0	0	18
Achnanthes species	ACHS	0	0	111
Achnanthidium biasolettianum (Grunow) Lange-Bertalot	ADBI	1	0	195
Achnanthes exigua Grunow	AEXG	4	1	0
Achnanthes minutissima Kützing	AMIN	1	4	28
Achnanthes minutissima Kützing	AMMA	0	0	1
Amphora montana Krasske	AMMO	1	0	0
Achnanthes minutissima var. saprophila Kobayasi & Mayama	AMSA	2	3	6
Amphora pediculus (Kützing) Grunow	APED	1	4	0
Craticula molestiformis (Hustedt) Lange-Bertalot	CMLF	2	0	0
Cocconeis pediculus Ehrenberg	CPED	6	19	6
Cocconeis placentula Ehrenberg	CPLA	13	26	3
Cocconeis placentula var. euglypta (Ehrenberg) Grunow	CPLE	2	4	2
Cymbella turgidula Grunow	CTGL	0	1	0
Craticula vixnegligenda Lange-Bertalot	CVIX	0	1	0
Diploneis elliptica (Kützing) Cleve	DELL	1	0	0
Diploneis smithii (Brébisson) Cleve	DSMI	0	1	0
Diatoma vulgaris Bory	DVUL	0	16	0
Epithemia adnata (Kützing) Brébisson	EADN	4	0	0
Encyonopsis minuta Krammer & Reichardt	ECPM	0	0	1
Eunotia minor (Kützing) Grunow	EMIN	0	1	0
Encyonopsis leei var. sinensis Metzeltin & Krammer	ENLS	1	10	0
Eolimna minima (Grunow) Lange-Bertalot	EOMI	0	4	0
Eolimna subminuscula (Manguin) Lange-Bertalot	ESBM	1	4	0
Epithemia sorex Kützing	ESOR	0	1	0
Fragilaria brevistriata Grunow	FBRE	4	0	0
Fragilaria capucina var. vaucheriae (Kützing) Lange-Bertalot	FCVA	1	45	19
Fragilaria elliptica (Schumann) Williams & Round	FELL	192	0	0
Fragilaria pinnata Ehrenberg	FPIN	11	0	0
Geissleria decussis(Hustedt) Lange-Bertalot	GDEC	2	0	0
Gomphonema insigne Gregory	GINS	0	5	0
Gomphonema minutum (Agardh) Agardh	GMIN	1	0	0
Gomphonema species	GOMS	10	27	0
Gomphonema parvulum (Kützing) Kützing	GPAR	0	1	0
Gomphonema parvulum f. saprophilum Lange-Bertalot & Reichardt	GPAS	0	1	0
Gomphonema pumilumvar. rigidum Reichardt & Lange-Bertalot	GPUM	0	4	2
Gomphonema truncatum Ehrenberg	GTRU	1	0	0
Gomphonema venusta Passy, Kociolek & Lowe	GVNU	0	1	0
Luticola nivalis (Ehrenberg) D.G. Mann	LNIV	0	1	0
Mayamaea atomus var. permitis (Hustedt) Lange-Bertalot	MAPE	4	0	3
Melosira varians Agardh	MVAR	2	2	0
Nitzschia acicularis (Kützing) WM Smith	NACI	0	12	0
Navicula antonii Lange-Bertalot	NANT	0	1	1
Navicula species	NASP	6	2	0
Nitzschia clausii Hantzsch	NCLA	1	0	0

Species	Abbr.	EFR K7	EFR C5	EFR C6
Navicula capitatoradiata Germain	NCPR	0	1	0
Navicula cryptocephala Kützing	NCRY	7	0	0
Nitzschia dissipata (Kützing) Grunow	NDIS	60	71	3
Navicula erifuga Lange-Bertalot	NERI	5	0	0
Nitzschia fonticola Grunow	NFON	1	0	0
Navicula gregaria Donkin	NGRE	3	6	0
Nitzschia hantzschiana Rabenhorst	NHAN	1	0	0
Nitzschia heufleriana Grunow	NHEU	2	0	0
Nitzschia frustulum (Kützing) Grunow	NIFR	2	13	0
Navicula ignota Lund	NINO	0	4	0
Navicula libonensis Schoeman	NLIB	1	0	0
Nitzschia linearis (Agardh) W Smith	NLIN	2	8	0
Nitzschia linearisvar. subtilis(Grunow)	NLSU	0	7	0
Navicula microcari Lange-Bertalot	NMCA	1	0	0
Nitzschia microcephala Grunow	NMIC	1	0	0
Nitzschia palea (Kützing) W. Smith	NPAL	2	0	0
Nitzschia perspicua Cholnoky	NPRP	0	0	0
Navicula radiosa Kützing	NRAD	1	1	0
Navicula reichardtiana Lange-Bertalot	NRCH	0	5	0
Navicula recens (Lange-Bertalot) Lange-Bertalot	NRCS	2	0	0
Nitzschia recta Hantzsch	NREC	0	18	0
Navicula tripunctata (OF Müller) Bory	NTPT	0	1	1
Nitzschia species	NZSS	16	18	0
Placoneis dicephala (W Smith) Mereschkowsky	PDIC	1	0	0
Pleurosigma salinarum (Grunow)	PSAL	7	0	0
Rhoicosphenia curvata (Kützing) Grunow	RCUR	6	0	0
Rhopalodia gibberula (Ehrenberg) O Müller	RGBL	1	2	0
Rhopalodia operculata (Agardh) Håkansson	ROPE	1	0	0
Reimeria sinuata (Gregory) Kociolek & Stoermer	RSIN	0	1	0
Reimeria uniseriata Sala Guerrero & Ferrario	RUNI	0	42	0
Stephanodiscus minutulus (Kützing) Cleve and Möller	STMI	2	0	0

B7.3 ORANGE RIVER TRIBUTARIES: MOLOPO WETLAND

Species	Abbr.	Bosbokpark	Flow guage	Molo Masu
Achnanthidium affine (Grunow) Czarnecki	ACAF	33	9	0
Achnanthes species	ACHS	18	51	0
Achnanthes crassa Hustedt	ACRA	84	49	0
Achnanthidium biasolettianum (Grunow) Lange-Bertalot	ADBI	62	24	0
Planothidium frequentissima (Lange-Bertalot) Round & Bukhityarova	ALFR	7	0	0
Achnanthes linearis (W Smith) Grunow	ALIN	9	0	0
Achnanthes minutissima var. affinis (Grunow) Lange-Bertalot	AMAF	0	6	0
Achnanthes minutissima Kützing	AMIN	58	0	1
Amphora montana Krasske	AMMO	2	0	0
Achnanthes minutissima var. saprophila Kobayasi & Mayama	AMSA	9	6	0
Achnanthes oblongella Oestrup	AOBG	1	0	0
Amphora pediculus (Kützing) Grunow	APED	0	4	0
Aulacoseira granulata var. angustissima (O Müller) Simonsen	AUGA	0	0	11
Brachysira neoexilis (Grunow) DG Mannt	BNEO	0	2	1

WP 5: Assessment of Environmental Flow requirements

Species	Abbr.	Bosbokpark	Flow guage	Molo Masu
Cyclotella atomus Hustedt	CATO	0	0	1
Caloneis bacillum (Grunow) Cleve	CBAC	0	0	1
Cyclostephanos invisitatus (Hohn & Hellerman) Theriot, Stoermer & Håkansson	CINV	0	0	5
Cymbella kappii Cholnoky	СКРР	0	2	0
Cyclotella melosiroides (Kirchner) Lemmermann	CMEL	0	0	0
Cyclotella meneghiniana Kützing	CMEN	0	0	2
Craticula molestiformis (Hustedt) Lange-Bertalot	CMLF	0	2	0
Cymbella tumida (Brébisson) Van Heurck	CTUM	0	0	2
Cymbella species	CYMS	0	1	5
	DCON	0	0	
Diploneis elliptica (K黷zing) Cleve	DELL	0	0	5
Diploneis puella (Schumann) Cleve	DPUE	0	0	5
Epithemia adnata (Kützing) Brébisson	EADN	0	1	0
Encyonopsis cesatii (Rabenhorst) Krammer	ECES	0	33	1
Encyonopsis krammeri Reichardt	ECKR	0	98	2
Eunotia minor (Kützing) Grunow	EMIN	0	1	0
Encyonopsis microcephala (Grunow) Krammer	ENCM	0	27	0
Encyonema neogracile Krammer	ENNG	0	5	0
Eolimna minima (Grunow) Lange-Bertalot	EOMI	46	8	0
Epithemia sorex Kützing	ESOR	0	0	47
Encyonopsis subminuta Krammer & Reichardt	ESUM	0	12	1
Fragilaria biceps (Kützing) Lange-Bertalot	FBCP	0	5	0
Fragilaria elliptica (Schumann) Williams & Round	FELL	0	3	1
Fragilaria nanana Lange-Bertalot	FNAN	2	1	0
Fragilaria tenera (WM Smith) Lange-Bertalot	FTEN	0	1	0
Fragilaria ulna var. acus (Kützing) Lange-Bertalot	FUAC	1	0	0
Gomphonema affine Kützing	GAFF	2	0	0
Gomphonema angustatum (Kützing) Rabenhorst	GANG	0	0	4
Gomphonema clavatum Ehrenberg	GCLA	1	0	0
Gomphonema clevei Fricke	GCLE	0	0	0
Gomphonema exilissimum Lange-Bertalot & Reichardt	GEXL	9	0	0
Gomphonema gracile Ehrenberg	GGRA	6	0	0
Gomphonema insigne Gregory	GINS	1	0	0
Gomphonema species	GOMS	11	0	4
Gomphonema parvulum (Kützing) Kützing	GPAR	7	0	4
Gomphonema parvulum f. saprophilum Lange-Bertalot & Reichardt	GPAS	0	0	1
Gomphonema lagenula Kützing	GPLA	4	0	0
Gomphonema parvulius Lange-Bertalot & Reichardt	GPPA	9	0	4
Gyrosigma rautenbachiae Cholnoky	GRAU	0	0	1
Gomphonema truncatum Ehrenberg	GTRU	0	1	0
Mayamaea atomus (Kützing) Lange-Bertalot	MAAT	1	0	2
Mastogloia smithii Thwaites	MSMI	0	0	31
Nitzschia acicularis (Kützing) WM Smith	NACI	0	0	18
Nitzschia agnewii Cholnoky	NAGW	0	0	59
Nitzschia amphibia Grunow	NAMP	0	46	1
Navicula species	NASP	0	1	0
Nitzschia capitellata Hustedt	NCPL	0	0	3
Navicula cryptocephala Kützing	NCRY	2	0	0
Navicula cryptotenella Lange-Bertalot	NCTE	0	1	2

Species	Abbr.	Bosbokpark	Flow guage	Molo Masu
Nitzschia dissipata (Kützing) Grunow	NDIS	1	0	0
Nitzschia draveillensis Coste & Ricard	NDRA	0	0	24
Nitzschia filiformis (WM Smith) Van Heurck	NFIL	0	0	14
Nitzschia hantzschiana Rabenhorst	NHAN	0	0	2
Nitzschia archibaldii Lange-Bertalot	NIAR	0	0	0
Nitzschia frustulum (Kützing) Grunow	NIFR	1	0	12
Nitzschia irremissa Cholnoky	NIRM	0	0	16
Nitzschia linearis (Agardh) W Smith	NLIN	0	0	1
Nitzschia linearisvar. subtilis(Grunow)	NLSU	0	0	1
Navicula microcari Lange-Bertalot	NMCA	0	0	4
Nitzschia palea (Kützing) W. Smith	NPAL	0	0	17
Navicula radiosa Kützing	NRAD	0	0	0
Navicula reichardtiana Lange-Bertalot	NRCH	0	0	1
Nitzschia recta Hantzsch	NREC	0	0	4
Nitzschia reversa W Smith	NREV	0	0	1
Navicula tenelloides Hustedt	NTEN	3	0	0
Nitzschia radicula Hustedt	NZRA	0	0	9
Nitzschia species	NZSS	9	0	32
Nitzschia supralitorea Lange-Bertalot	NZSU	0	0	2
Pleurosigma salinarum (Grunow)	PSAL	0	0	14
Rhopalodia gibberula (Ehrenberg) O Müller	RGBL	0	0	2
Sellaphora seminulum (Grunow) DG Mann	SSEM	1	0	0
Stephanodiscus minutulus (Kützing) Cleve and Möller	STMI	0	0	2
Tryblionella apiculata Gregory	TAPI	0	0	13
Tryblionella hungarica (Grunow) DG Mann	THUN	0	0	1
Thalassiosira pseudonana Hasle & Heimdal	TPSN	0	0	3

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