Diatom Community Composition and Ecological Gradients on Selected Rivers in the Eastern and Western Cape, South Africa

By

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ABSTRACT

This study sought to assess diatom community composition across ecological gradients on selected rivers in the Eastern and Western Cape Provinces, South Africa, as well as in the lowland section of the Okavango River. For South Africa data were collected over a one-year period with three-monthly or seasonal sampling conducted between spring 2014 and winter 2015. Five to ten cobbles were scrubbed and diatoms fixed with 70% ethanol to produce an end product of >20% alcohol content. In Botswana samples were collected during July and August 2014. A phytoplankton net with a mesh size of 25 μm and introduced substrate were sampled in the panhandle, Nxamaseri Floodplain and the Thamalakane River. The Hot HCl method was used to clean samples of organic material. Permanent slides were made using Pleurax as mounting agent. All information was added to the National Diatom Collection since one of the main objectives of this study was to fill the current information gap on these regions. A minimum of 400 individuals was identified per slide to produce a community composition. Multivariate statistics showed that diatom communities responded geospatially more strongly than seasonally. In South Africa the diatom communities responded at an Ecoregion Level 1. At Ecoregion Level II, catchment signatures were not strictly followed as would have been expected. Instead more localised impacts and natural fluctuations in physico-chemical changes were found to drive group formation. The influence of flow modification, such as the inter-basin transfer schemes in the Drought Corridor ecoregion, was indicated in diatom community composition. While diatoms are extremely useful as small-scale impact specific indicators and assessment tools, the results produced in this study showed that the use of diatom information for larger scale climate change impact monitoring initiatives towards sustainable freshwater resource management in future, remains untapped. Further research on the relationship between diatom community composition and more detailed environmental drivers of landscape scale ecosystem changes would greatly improve our understanding of the role diatoms play in the resilience of natural freshwater ecosystems to large-scale changes and impacts. The diatom data proved to be very robust and reliable in this study, suggesting that there exists a great resilience at the base of the food web in the highly volatile and dynamic African freshwater ecosystems. Properly functioning diatom communities could be a more important component of ecosystem resilience than currently recognised. This study also found that the exclusion of certain low abundance diatom information does not contribute to a more accurate result but instead removes valuable biodiversity information in a time when it should be promoted, protected and well documented. It is clear that in a country expected to experience severe and direct climate change induced impacts, the exclusion of diatoms when managing

freshwater sustainability and continued optimal ecosystem functioning for the delivery of associated good and services, is rather reckless. This study has provided the foundation of updated diatom information for a majority of the major rivers in the Eastern Cape in particular and some of the smaller coastal rivers in the Western Cape. This method should be applied in other regions of the country to produce diatom reference conditions, which speak directly to Ecological Reserve scale approaches in order to contribute to a more holistic approach to ecosystem management in future. In Botswana a significant difference between the community compositions of the Thamalakane River, Nxamaseri Floodplain and the Okavango Panhandle was found to be present. These differences are suspected to be caused by micro-environments with differences in nutrient load and associated water quality. These micro-habitats allowed for some species, that are more tolerant to higher nutrient loads, to be found but did not interfere with the larger scale diatom community composition in a specific geographic region. The difference between diatom communities in the panhandle and other areas in the system highlights the importance of upstream conservation in the Okavango River for continued optimal ecological functioning of the downstream Okavango Delta and its associated systems. Diatoms are able to make a considerable contribution to our current understanding of freshwater resource functioning and the consequent conservation, monitoring and management of sustainable water security for all.

OPSOMMING

Hierdie studie was daarop gemik om die samestelling van diatoom gemeenskappe oor ekologiese gradiënte van geselekteerde riviere in die Oos- en Wes-Kaap Provinsies, Suid-Afrika, asook die laagliggende gedeelte van die Okavango Rivier te assesseer. Die data is oor 'n tydperk van een jaar versamel met drie-maandelikse of seisoenale steekproefnemings wat tussen lente 2014 en winter 2015 uitgevoer is. Vyf tot tien klippe is geskrop en diatome is met 70% etanol gefiskeer om 'n eindproduk van minstens 20% te lewer. In Botswana is data versamel gedurende Julie en Augustus van 2014. ‘n Plankton net met n deurlatings ruimte van 25 μm en kunsmatige substraat is gebruik om monsters mee te versamel in die Okavango Panhandle, Nxamaseri Vloedvlakte en die Thamalakane Rivier. Die Warm HCl-metode is gebruik om organiese materiaal in monsters te verwyder. Permanente mikroskoop-skyfies is gemaak deur van Pleurax gebruik te maak as monteermiddel. Alle inligting is by die Nasionale Diatoom Versameling gevoeg aangesien een van die hoof doelstellings van hierdie studie was om die huidige inligtingsgaping oor hierdie areas te vul. ‘n Minimum van 400 individue is per skyfie geïdentifiseer om die gemeenskapsamestelling te gee. Meerveranderlike statistiese analise het getoon dat diatoom gemeenskappe in Suid Afrika eerder op ‘n Ekostreek Vlak I reageer as wat hulle seisoenaal differensieer. In teenstelling met wat verwag is het gemeeskappe op Ekostreek Vlak II is geen opvangsgebied uniekheid geopenbaar nie. In plaas daarvan is gevind dat gelokaliseerde impakte en natuurlike fluktuasies in fisies-chemiese veranderinge groepvorming bestuur. Die invloed van interbekken-waterverplasings skemas op natuurlike vloeipatrone, soos gesien is met die monsters in the Droogte Gang ekostreek was sigbaar in die diatoom gemeenskaps samestelling. Hierdie studie het daarin geslaag om diatoom gemeenskappe met Ekostreek Vlak I verbind. Gevolglik kon die studie ‘n verwysingsvoorwaarde beskrywing vir beide Vlak I en II lewer. Terwyl diatome uiters nuttig is as kleinskaalse impakspesifieke aanwysers en assesseringsinstrumente, het die resultate in hierdie studie getoon dat die gebruik van diatoom inligting vir die monitering van grootskaalse klimaatsverandering en volhoubare bestuur van varswaterhulpbronne tans grootliks onbenut bly. Verdere navorsing oor die verhouding tussen die samestelling van diatoom gemeenskappe en meer spesifieke besonderhede rakende omgewingsfaktore sal ons begrip rondom die rol van diatome in veerkragtigheid van varswater-ekosisteme baie kan bevorder. Die data was baie robuust en betroubaar in hierdie studie, wat daarop dui dat daar 'n goeie veerkragtigheid aan die basis van die voedselweb tans bestaan. Selfs met natuurlike versteurings in Afrika se hoogs dinamiese varswater-ekosisteme. Behoorlik funksionerende diatoom gemeenskappe kan 'n belangriker komponent van varswater-ekosisteem veerkragtigheid wees as wat tans bekend

is of erken word. Hierdie studie het ook bevind dat die uitsluiting van sekere laer digtheid diatoom inligting nie bydra tot 'n meer akkurate resultaat nie, maar eerder waardevolle inligting oor biodiversiteit verwyder. Suider Afrika gaan in die nabye toekoms al ernstiger en meer direkte klimaatsverandering-geïnduseerde impakte ervaar. Dit is daarom taamlik roekeloos om diatoom inligting uit varswater volhoubaarheids waarnemings te laat. Hierdie studie het die grondslag gegee vir volgehoue opdatering van diatoom inligting op verkeie riviere in die Oos- en Wes-Kaap Provinsies. Hierdie metode moet in ander streke van die land ook toegepas word sodat diatoom verwysingsvoorwaardes produseer kan word vir ander Ekostreek Vlak I en II gebiede. Sodoende kan verseker word dat diatoom inligting ingesluit word by Ekologiese Reserwe studies en in die toekoms kan bydra tot 'n meer holistiese benadering tot varswater ekostelselbestuur. In Botswana is daar gevind dat n noemenswaardige verskil in gemeenskaps samestelling bestaan tussen the Thamalakane Rivier, Nxamaseri Vloedvlakte en die Okavango Panhandle. Die verskil word toegeskryf aan die teenwoordigheid van mikro-habitats wat ‘n verskil in nutrient lading het en dus ‘n invloed om die lokale water kwaliteit het. Hierdie mikro-habits bevorder tollerante spesies wat bestand en aangepas is teen die hoër nutrient lading. Ondanks hierdie kleinskaalse veranderinge in gemeeskaps samestelling was daar nie ‘n invloed op die groterskaalse ekostreeks vlak groepering nie. ‘n Duidelike verskil tussen die verskillende streke van die Okavango Rivier en geassosieerde stelses kon nogtans waargeneem word. Hierdie geografiese stratifikasie beklemtoon die belangrike rol wat die stroom-op ekostelsel speel in die laer gedeeltes van die Okavango Rivier, veral die delta wat ‘n bewarings area van internationale belang is. Diatome kan 'n aansienlike bydrae lewer tot ons huidige begrip van varswaterhulpbron funksionering, handhawing van ekosisteem integriteit en die gevolglike bio-assessering, monitering en bestuur van volhoubare watersekuriteit vir almal.

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Introduction

CHAPTER 1 Introduction

Freshwater ecosystems today

Fresh water is the most important resource we have on the planet. Without it, no life would be possible. Yet our freshwater resources are under tremendous pressure to provide potable water for human consumption, as well as secondary needs such as food, electricity, industrial needs and aesthetic services. Over recent years wetlands have enjoyed increasing attention largely due to the establishment of the Ramsar Convention on Wetlands[1]. While this is significant progress, it is not enough in order to ensure water security and conservation. Rivers, which form part of the water providing basins together with wetlands, have received much less attention. In fact, in most urban poor communities, rivers are used as waste disposal systems. There is a great disconnect between people and rivers, especially in urban areas where water supply and demand have the largest disproportion and have the biggest impact on human well-being[2].

Globally more than a billion people do not have access to safe drinking water (WHO 2006). This is not only due to the lack of infrastructure provided by government but also due to pollution, overutilisation, degradation and exploitation of the river's ecosystem services, which ultimately leads to poor river health and an associated deterioration in water quality. When a river is continuously over-extracted and impacted it becomes so polluted and degraded that it cannot easily be restored to health (Davies and Day 1998). It becomes a managerial issue especially in densely populated areas where illness can become a huge problem. Diarrhoea for instance occurs world-wide and causes 4% of all deaths. This translates to around 2.2 million deaths annually (WHO 2000). It is therefore of utmost importance for the survival of human beings to not only understand river systems and the biodiversity they hold, but also utilise information on ecosystem requirements and projected climate impacts to successfully manage freshwater ecosystems for sustainable water quantity and quality for the future.

Climate change is one of the biggest threats to human survival modern science has had to face. Current projections for climate change impact are forecasting a serious increase in the intensity and occurrence of droughts and floods[3]. Coupled with already water-stressed environments we could see natural clean water becoming increasingly valuable and rare. In 2006 sub-Saharan Africa had the largest number of water-stressed countries in the world. At

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[1] The Convention was adopted in the Iranian city of Ramsar in 1971 and came into force in 1975. http://www.ramsar.org/ (accessed on 11 Oct 2017).
[2] http://cbc.iclei.org/project/una-rivers-life/ (accessed on 8 January 2018)
[3] http://www.csag.uct.ac.za (accessed on 8 May 2017)

the Water Scarcity in Africa: Issues and Challenges conference held in 2012, it was estimated that by 2030, as many as 250 million people in Africa could be living in areas of critical water stress. If these projections are accurate it would cause as many as 700 million people in Africa becoming displaced due to unliveable situations.

Water quality and access

According to the Blue Planet Network[4] unsafe water kills approximately 200 children every hour. They go further to state that as many as 3.4 million people die annually from water, sanitation and hygiene related causes. A much as 40% of the sub-Saharan population don't have access to a formal and improved source of water, this relates to approximately 313 million people[5]. Improving quality of water and access to sanitation can help eradicate poverty and improve human well-being tremendously (WHO 2006). The Blue Planet Network, and some other online resources which are referenced in text below, identify some of the major reasons Africa is facing a serious water related socio-economic crisis. These are:

1. Africa is an arid continent, which does not have a very high rainfall compared to surface area[2]. There are large desert areas present (Sahara and Namib) and the atmospheric conditions are simply not favourable to higher rainfall occurrence.
2. The “Scramble for Africa” left most large freshwater bodies on the continent in multi governmental management. The Nile River flows through eleven countries. Or as in the case of the Okavango River, which is shared by three countries. This makes the coordination of research and management a logistical nightmare (Tanner 2013).
3. Most of the large water bodies in Africa are polluted to some extent, with large communities living right at the water’s edge. A lack of service delivery and infrastructure directly impacts these settlements, the occupants of which are dependent on the freshwater source (WHO 2006).
4. Population distribution does not correspond to water availability. The largest concentrations of people do not live in the Congo River basin, which is the most water rich area in Africa[3].
5. The water table of the African continent is receding. With most people being dependant on surface water[3], this deficit in the groundwater will certainly exacerbate the impact of climate induced impacts on rainfall. With a lower water table, the continent will need increased rainfall to sustain sufficient surface base flow to be available during dry periods. A lower surface flow will greatly impact the availability and especially the quality

[4] http://blueplanetnetwork.org/water/ (accessed on 11 October 2017)
[5] http://www.un.org/waterforlifedecade/africa.shtml (accessed on 12 October 2017)

of the available water. Low flow concentrates and exacerbates the impact of pollutants on water quality[6].

6. A lack of education regarding water quality, conservation and management has been and remains one of the biggest social disasters related to freshwater resources[7]. Women and children in Africa are responsible for collecting water for the entire family[4]. A more hygenic environment at home allows children to spend more time at school and working on school related activities (UNICEF 2003). Usually unaware of the threats the unhealthy water poses for their families, women spend hours collecting and carrying water. Proper education regarding waste disposal, water cleaning and ecological conservation could drastically improve the way freshwater bodies are locally managed in rural Africa. Also, increased awareness for water saving would help improve conservation for future security.
7. Agricultural activities are the largest water consumer in Africa[8]. With a continent that has severe hunger and poverty, agriculture is one of the most important economic activities. However, proper management of the impacts and use of these agricultural activities should be assessed and monitored especially in terms of contingency plans for climate change impacts.

Water scarcity and development

While water scarcity is a global challenge, Southern Africa is being hit hardest by the climate induced changes in rainfall patterns[9]. South Africa is a semi-arid country that experiences regular droughts and flooding. The rainfall is unevenly distributed, with some areas in the country receiving much higher rainfall than others[9]. The availability of water had a massive impact on economic development, for Johannesburg in particular. Johannesburg is a major city not built near a large natural water resource so any development would be limited by water availability. It is only through the construction of large dams and interbasin transfers that development could occur (Van Vuuren 2012).

With water stress becoming more apparent across all areas of South Africa, even in the higher rainfall areas, the biggest driver of economic, social and environmental well-being in future will most decisively be the availability of water. Half of the water in rivers is from 8% of land area in South Africa, with only 16% of these areas currently formally protected[10]. These Strategic Water Source Areas (SWSAs) are under great pressure to ensure water security

[6] https://thewaterproject.org/water-crisis/water-in-crisis-rural-urban-africa (accessed on 20 Novemeber 2017)
[7] https://lifewater.org/blog/water-education/ (accessed on 18 November 2017)
[8] http://all-about-water-filters.com/facts-about-water-in-africa/ (accessed on 20 November 2017)
[9] http://www.csag.uct.ac.za (accessed on 9 May 2017)
[10] http://www.wwf.org.za/what_we_do/freshwater/ (accessed on 25 Septemebr 2017)

for the future[11]. In order for this to happen, future developments will have to recognise the limitations of our natural resources and develop innovative strategies to include social upliftment, education and proper ecological management.

Water managers, environmental scientists and ecologists are constantly assessing their methods and improving on tools used to assess and monitor freshwater resources. The importance of biodiversity, which underpins the functionality of an ecosystem (Campbell et el. 2009), has long been recognised and is embedded internationally into strategic objectives through the Convention on Biological Diversity[12] (CBD), Aichi Biodiversity Targets[8] and the Sustainable Development Goals[13] (SDG's). Included in these strategic objectives is SDG 6, which is aimed at providing access to clean water and sanitation for all people and SDG 13, which is focused on incorporating climate related action into resource management and development[14].

It is not only important to update our perception of water availability, but also of the importance of biological diversity. It is easier to explain why water is important, but often biologists struggle to communicate the importance of the biological component of an ecosystem service. Biodiversity is the collective term for this biological component. The planet is losing species at an unprecedented rate; this is extremely dangerous since, apart from losing ecosystem functionality, medical, technological and engineering discoveries could be losing critical organisms that may drastically improve human well-being. Nature has been the blueprint for so many designs, ideas, pharmaceutical discoveries and remedies, we are not only losing key natural services, we are also losing the possibilities of so much more.

Freshwater biodiversity

In light of the above mentioned, it could appear strange then that we are not doing more to study, assess, document and monitor the biological wealth of our freshwater ecosystems. With all the more pressing immediate water related issues and priorities, such as the availability of funding, human health and service delivery, priority is often given to reactive responses instead of precautionary adaptive strategies. In Africa, and many developing countries around the world, the funds needed to build the skills needed to provide such studies and services are often simply not feasible when there are other more immediate needs. Thankfully there have been many initiatives aimed at profiling the need for increased biodiversity conservation towards optimised ecosystem service delivery (Austin et al. 2016,

[11] https://www.sanbi.org/news/strategic-water-source-areas-are-national-assets (accessed on 20 December 2017)
[12] https://www.cbd.int/ (accessed on 20 March 2017)
[13] https://www.millennium-institute.org/isdg (accessed on 15 March 2017)
[14] For the SDG logos see Figure 85 in CHAPTER 6 Discussion.

Harrison et al. 2014, Ingram et al. 2012, Science for Environment Policy 2015). Some examples of projects and initiatives are; the Cape Action for People and the Environment[15] (C.A.P.E. Program), the Local Action for Biodiversity: Wetlands South Africa (LAB) project[16], the Urban Natural Assets for Africa: Rivers for Life[2] project, The Global Environmental Facility's Small Grants Programme[17] (SGP) and the National Freshwater Ecosystem Priority Areas project[18] (NFEPA). Most of these are driven and funded by international organisations, but together with inclusive approaches and local community buy-in, these projects are all embedded into the aim of achieving international biodiversity targets, promote urban sustainability and improve freshwater resource protection. These projects directly and indirectly support the continually developing National Aquatic Ecosystem Health Monitoring Programme and River Eco-status Monitoring Programme and aids decision makers in effectively managing South Africa's freshwater resources. Diatom studies could add a significant layer to the already well developed National Aquatic Ecosystem Health Monitoring Programme, of which the biological component is only focussed on riparian vegetation, fish and macroinvertebrates at this stage (Kleynhans and Louw 2007).

Diatoms are one of the largest and ecologically most significant groups of organisms on Earth. They occur almost everywhere that is moist; oceans, lakes, rivers, marshes, fens and bogs, damp moss and rock faces and even on the feathers of some diving birds (Atkinson 1972, Croll and Holmes 1982). Diatoms are estimated to account for as much as 20% of the global fixation of carbon, more than the entire world's tropical rainforests (Boyd et al. 2000). Mann (1999) estimated this amount to be around 20 Pg carbon fixed per year. Diatoms occur in large numbers in saline and freshwater where they form the base of the food chain. In the oceans they are estimated to contribute as much as 45% of the total primary production (Mann 1999). In addition to this diatoms are also a source of petroleum and diatomaceous earth, which is used for insolation, filtration, absorbent liquids, dynamite and mild abrasives (Legget 2017).

Diatoms occur in all freshwater bodies around the world including rivers, which is the habitat this current study sought to investigate. Diatoms are unicellular organisms although some can form colonies of different shapes, such as filaments (*Fragilaria*), fans (*Meridion*), zigzags (*Tabellaria*) and stars (*Asterionella*) (Taylor et al. 2007b). Diatoms belong to a large group known as the heterokonts which includes heterotrophs like water molds and autotrophs

[15] https://www.thegef.org/project/cape-biodiversity-conservation-and-sustainable-development-project (accessed on 10 January 2018)
[16] http://cbc.iclei.org/project/lab-wetlands-sa/ (accessed on 8 January 2018)
[17] http://www.za.undp.org/content/south_africa/en/home/operations/projects/environment_and_energy/the-gef-small-grant-programme-.html (accessed on 12 December 2017)
[18] https://www.sanbi.org/biodiversity-science/science-policyaction/mainstreaming-biodiversity/freshwater-programme (accessed on 13 December 2017)

which includes brown algae and kelp. Yellow-brown coloured chloroplasts are a typical characteristic of the heterokonts. Diatoms are classified in the Kingdom Chromista and the algal Class Bacillariophyceae. Diatom cells are enclosed in a cell wall made of silica, known as the frustule, and consist of two halves, the valves. The valves are used for the taxonomic identification of diatom species (Round et al. 1990).

Today it is estimated that more than 200 genera of living diatoms and approximately 100,000 species are known (Canter-Lund and Lund 1995). Because of the siliceous composition, diatoms are well preserved for long periods and are often used in fossil studies as well as a variety of industrial uses. One of the most noteworthy uses of diatoms is their application in environmental and earth sciences (Stoermer and Smol 1999). Diatoms are ideal for biological monitoring due to the fact that they are abundantly found in nearly all habitats. A very large number of ecologically sensitive species can occur and they leave their remains in the sediments enabling historic comparisons to be made (Dixit et al. 1992, Stoermer and Smol 1999).

Project aim and objectives

The main aim of this project was to document the diatom species of riverine habitats in the Eastern Cape Province. The National Diatom collection contains relatively few samples from this province and the addition of these would be of national biomonitoring value. This would provide a vital reference point for continued monitoring of riverine water resources in South Africa. The second aim was to assess the diatom community composition across ecological gradients which would contribute to our understanding of diatom distribution, reference conditions and continued biomonitoring. There are only three inland alluvial fans in Africa, the Okavango Delta is one of these and the only one situated in Southern Africa. This study set out to provide much needed information on the diatom ecology of the panhandle section of the Okavango River system in Botswana under the Southern African context.

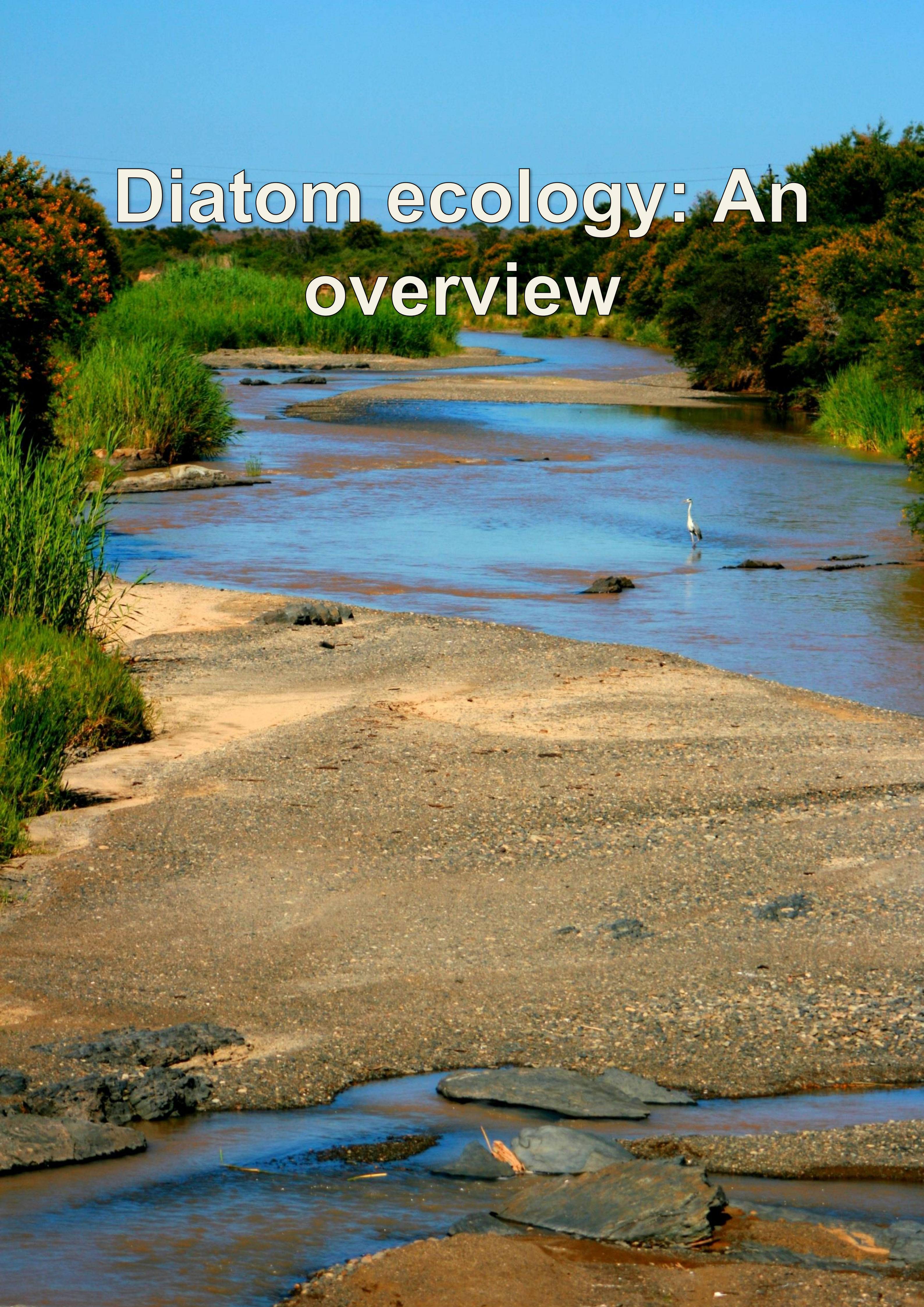
<u>The objectives that stem from these two aims were:</u>

1. Document the diatom species composition for the rivers sampled.
2. Assess the community composition across spatial ecological gradients.
3. Assess seasonal changes in diatom community composition.
4. Describe the significant species from different rivers, basins or ecological regions.
5. Assess the relationship between diatom community composition and basic physico-chemical characteristics.

The rest of the thesis is comprised of a comprehensive description of diatoms, their biology and ecology, as well as a brief history of diatom science in South Africa which is provided in

CHAPTER 2 An overview of diatom ecology and community composition. Furthermore, the chapter looks at South Africa and Botswana's freshwater resources, ecoregion classification and the role of diatoms in environmental assessments. **CHAPTER 3 Sampling methodology** provides an overview of the methods used to collect, process and analyse the data. In **CHAPTER 4 Diatom community composition across ecological gradients, South Africa**, the spatio-ecological data for the South African samples are presented. The Botswana samples were assessed separately and are presented in the form of a scientific paper for submission to the *African Journal of Aquatic Sciences* in **CHAPTER 5 Diatom community composition of the Okavango Panhandle, Botswana (draft paper prepared for AJAS)**. The discussion of the results is presented in **CHAPTER 6 Discussion** followed by **CHAPTER 7 Concluding remarks** which contains some closing and concluding remarks as well as recommendations. **CHAPTER 8 References** contains the references used in this thesis. Raw data lists and analyses, as referred to in the study chapters, are presented in the section at the back in Appendix 1-3.

Diatom ecology: An overview



CHAPTER 2 An overview of diatom ecology and community composition

Background

Diatoms are a major group of microalgae and occur in all freshwater environments on Earth. They are primary producers and play a fundamental role in the food web. Diatoms are very sensitive to small changes in water quality which makes them good monitoring tools. In South Africa diatoms are underutilised at present but a renewed interest in diatomology has seen a revival of historic information and new projects assessing diatom diversity and ecology. While South Africa has a very rich history of diatomology, much more work is needed (Harding et al. 2004). This chapter explores the past, present and possible future application of diatoms in South African freshwater monitoring by providing an overview of available information on diatom ecology and community composition.

Evolutionary history

Heterokont chloroplasts are believed to descend from red algae. This is different from the rest of the plants which descends from prokaryotes. This would suggest that diatoms originated more recently than other algae. The fossil record of heterokonts is not very extensive and it was only with the evolutionary appearance of diatoms, with their siliceous cell walls, that heterokonts started making a serious impression in the fossil records (Kooistra and Medlin 1996). The earliest fossil diatoms date back to the early Jurassic (185 million years ago). Although this is the oldest diatom fossil on record, it is believed that diatoms may have been around much earlier than this. The end-Permian mass extinction opened many niches in the marine environment and the gap between this event and the first diatom fossil could possibly be due to diatoms being unsilicified. This would mean that their early evolutionary stages are not well represented in the fossil records from this period (Medlin et al. 1997). Due to the silicification of their cell walls, diatoms are abundant in later fossil records with some large deposits found today dating back to the early Cretaceous and known as diatomite or kieselguhr[19].

Taxonomy of freshwater diatoms

Algal diversity has been estimated to be anything between 30 000 to over 1 million species. Guiry (2012) noted that this estimation has been seen to be as high as 350 million algal

[19] https://www.nobelprize.org/alfred_nobel/biographical/articles/krummel/kieselguhr.html (accessed on 10 January 2018)

species, which would mean there are around 20 times more types of algae on Earth than all other living organisms. Furthermore, Guiry (2012) stated that the wide range in estimates could be due to the declining number of taxonomists working on algae and the ever declining number of newly trained graduates entering the field. Regardless, it is clear that algae are an extremely diverse group of organisms[20]. There are more than 200 known living diatom genera and approximately 100 000 species. The World Register of Marine Species (WoRMS) alone contains around 66 000 species when duplicates are removed[21]. If this estimation is correct, there are five diatom species for every other algal species alive today (Guiry 2012).

It is therefore safe to say that diatoms are the most species rich group of all the algae. Diatoms are well known for their wide distribution and considerable role in the carbon and silicon cycles. Despite the fact that they are such an abundant, important and diverse group, the taxonomy of diatoms remains very messy and unsatisfactory for practical and conceptual classification of the group. The "Walton species concept[22]" is suspected to have been drawn too broadly, with many species remaining unrecognised within current classification. Endemic species diversity is also expected to be very underestimated due to; lack of verified information, ecological gradients and stratigraphic patterns being hard to distinguish and due to much taxonomic work being poorly documented (Mann 1999).

The algal group known as heterokonts are yet to be properly defined. Heterokonts can be treated as a division, kingdom or something in between. Consequently the diatom group within the heterokonts can be ranked as anything from class (Bacillariophyceae) to division (Bacillariophyta) (Van den Hoek et al. 1995). To add to this confusion, diatoms are sometimes referred to as Class Diatomophyceae. Older classifications divide diatoms into two orders; the centric (Centrales) and pennate (Pennales) diatoms. Round et al. (1990) suggested three classes; centric diatoms (Coscinodiscophyceae), pennate diatoms without a raphe (Fragilariophyceae), and pennate diatoms with a raphe (Bacillariophyceae).

Evolutionary descent and diversification of diatoms within their group, as well as away from the rest of the Heterokont algae, remains an unsolved mystery. To some extent much progress has been made in the past few decades regarding generic level classification within families. This progress has mainly been fuelled by electron microscopy as well as cellular structures and sexual reproduction. These were often ignored by diatomists in the past. The addition of molecular phylogenetic studies has provided valuable information; however it has

[20] http://www.algaebase.org/ (accessed on 10 January 2018)
[21] http://www.marinespecies.org/aphia.php?p=taxdetails&id=148902 (accessed 10 January 2018)
[22] The 'Waltonian species concept' is derived from morphology, genetic data, mating systems, physiology, ecology, and crossing behaviour.

yet to shed any light on the relationships of diatoms at higher classification level i.e. among families, orders and classes. Round et al. (1990) used morphology and cytology to describe genera. This has been supported by subsequent investigations including those focussed on molecular analysis. However, gene sequencing has proven that comparative analysis based on morphology, failed to shed any light on the path of evolution. The problem could be convergent evolution of morphological cell shape and structure which has been extensive in diatoms (Mann and Evans 2007). Molecular analysis has not done much better for similar reasons, including analytical difficulties with homology in rDNA sequences.

Round et al.'s (1990) three diatom classes; Coscinodiscophyceae, Fragilariophyceae and Bacillariophyceae, corresponds to three main types of valve organisation:

1. Coscinodiscophyceae - valves in which the pattern of ribs and striae (lines of pores) radiate out from a ring.
2. Fragilariophyceae - a feather-like pattern, with the ribs and striae on either side of one or two longitudinal ribs.
3. Bacillariophyceae - similar to Fragilariophyceae, except that the central strip contains a raphe system.

Although molecular work has not yet been able to shed light on the evolutionary history of diatoms, the one thing that has become evident is that the classification of Round et al. (1990) is not accurate in explaining the evolutionary history either. Primary radiation occurred in diatoms with centric shaped valves. Pennate diatoms are proven to have evolved later from centric ancestors and the Fragilariophyceae are not monophyletic[23] but rather paraphyletic[24] with respect to the Bacillariophyceae. This means that of the three groups described by Round et al. (1990), only the Bacillariophyceae is truly acceptable.

In the United States to date, 834 diatom species have been identified and published to an online page[25] aimed at providing accurate and updated ecological and taxonomical information on diatoms of the United States. While in South Africa, the work initiated by Harding et al. (2004) led to the development of a taxonomic key to the diatoms of Southern African Rivers. This key contains information on 70 genera and 286 species, including common taxa and some key endemics. Perhaps one of the key reasons this number is so

[23] Descended from a common evolutionary ancestor or ancestral group, especially one not shared with any other group.
[24] Descended from a common evolutionary ancestor or ancestral group, but not including all the descendant groups. Unlike a monophyletic group, a paraphyletic taxon does not include all the descendants of the most recent common ancestor.
[25] https://westerndiatoms.colorado.edu/species (accessed on 24 May 2017)

much lower than that of for instance the United States, is not due to lack of diversity found in Southern African Rivers, but instead due to the methods used by key pioneer diatomists working in Southern Africa and the little work that has been completed since (Harding et al. 2004).

Béla. J. Cholnoky started his work on diatoms in South Africa during the early 1950's and continued untill his death in 1972. During this time he made considerable contributions to the taxonomic information of South African diatoms, describing hundreds of taxa in 38 published articles and a book on the taxonomy and ecology of Southern and Central African diatoms (Cholnoky 1968). Due to Cholnoky not always following the requirements of the international code of botanical nomenclature implemented in 1958, many of these taxa descriptions need revising to be validated. Once validated and documented, these taxa can formally be included into the taxonomic key for Southern African Rivers. An example of this is the work done by Taylor et al. (2010) on *Cymbella kappii*.

Table 1 presents a summary of the taxonomic classification of 72 common diatom genera of the three Classes in Bacillariophyta. Although classification based predominantly on morphology and biology is currently unable to explain the evolutionary path of diatom diversification, it is still applied, especially during identification of diatoms. The unique silica cell wall and the structural characteristics displayed on it are used to distinguish diatoms from one another. The biology and morphology of diatom cells are therefore crucial characteristics for working with diatoms and, if nothing else, aids in making the identification of different diatom species more manageable and user friendly, especially when using diatoms in ecological investigations such as the current study.

Table 1 Phylogenetic arrangement of common freshwater diatom genera (Based on Round et al. 1990, taken from Cox 1996).

Kingdom	Phylum	Class	Order	Family	Genus
PLANTA	BACILLARIOPHYTA	COSCINODISCOPHYCEAE	THALASSIOSIRALES	Thalassiosiraceae	*Thalassiosira*
				Skeletonemaceae	*Skeletonema*
				Stephanodiscaceae	*Cyclotella, Stephanodiscus*
			MELOSIRALES	Melosiraceae	*Melosira*
			PARALIALES	Paraliaceae	*Ellerbeckia*
			AULACOSIRALES	Aulacosiracerae	*Aulacoseira*
			ORTHOSEIRALES	Orthoseiraceae	*Orthoseira*
			RHIZOSOLENIALES	Rhizosoleniaceae	*Urosolenia*
			CHAETOCEROTALES	Chaetocerotaceae	*Chaetoceros*
				Acanthocerataceae	*Acanthoceros*
		FRAGILARIOPHYCEAE	FRAGILARALES	Fragilariaceae	*Fragilaria, Centronella, Asterionella, Staurosirella, Staurosira, Pseudostaurosira, Punctastriata, Fragilariaforma, Martyana, Diatoma, Hanneae, Meridion, Synedra*
			TABELLARIALES	Tabellariaceae	*Tabellaria, Tetracyclus*
		BACILLARIOPHYCEAE	EUNOTIALES	Eunotiaceae	*Eunotia, Semiorbis, Peronia*
				Peroniaceae	*Aneumastus*
			MASTOGLOIALES	Mastogloiaceae	*Mastogloia, Rhoicosphenia*
			CYMBELLALES	Rhoicospheniaceae	*Anomoeoneis*
				Anomoeoneidaceae	*Placoneis*
				Cymbellaceae	*Cymbella, Encyonema, Ghomphonema*
				Gomphonemataceae	*Didymosphenia, Reimeria, Achnanthes*
			ACHNANTHALES	Achnanthaceae	*Cocconeis*
				Cocconeidaceae	*Achnanthidium*
				Achnanthidiaceae	*Eucocconeis, Cavinula*
			NAVICULALES	Cavinulaceae	*Cosmioneis*

Table 1 Continued: Phylogenetic arrangement of common freshwater diatom genera (Based on Round et al. 1990, taken from Cox 1996).

Kingdom	Phylum	Class	Order	Family	Genus
PLANTA	BACILLARIOPHYTA	BACILLARIOPHYCEAE	NAVICULALES	Cosmioneidaceae	*Diadesmis*
				Diadesmidiaceae	*Luticola, Amphipleura*
				Amphepleuraceae	*Frustulia, Brachysira*
				Brachysiraceae	*Neidium*
				Neidiaceae	*Sellaphora*
				Sellaphoraceae	*Fallacia, Pinnularia*
				Pinnulariaceae	*Caloneis, Diploneis*
				Diploneidaceae	*Navicula*
				Naviculaceae	*Gyrosigma*
				Pleurosigmataceae	*Stauroneis*
				Stauroneidaceae	*Craticula, Amphora*
			BACILLARIALES	Bacillariaceae	*Denticula, Hantzschia, Tryblionella, Nitzschia, Epithemia*
			RHOPALODIALES	Rhopalodiaceae	*Rhopalodia, Entomoneis*
			SURIRELLALES	Entomoneidaceae	*Stenopterobia*
				Surirellaceae	*Surirella, Campylodiscus, Cymatopleura*

Biology of diatoms

Habitat

Diatoms occur in all types of aquatic ecosystems and can be identified by the brown mucilaginous film on surfaces of substrates in freshwater habitats (Figure 1A). There are four different substrates that benthic diatoms occur in or on; epipelon (surface of fine-grained sediments), episammon (between sand particles), epilithon (gravel, stone and bedrock) and epiphyton) (Taylor et al. 2007a). Planktonic diatoms are free-living in the water column of slow flowing rivers and dams (Figure 1B).

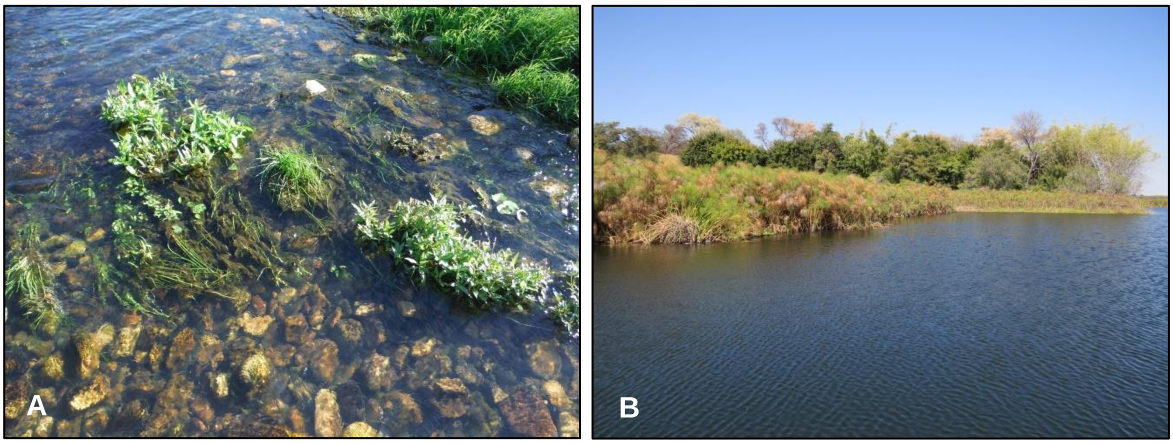


Figure 1 (A) The brown mucilaginous film on stones in a flowing river (Baviaanskloof River, Eastern Cape, South Africa). (B) Planktonic diatoms occur free-floating in the water column of slow flowing rivers such as backwaters in floodplains, wetlands and dams (The Okavango Panhandle).

Morphology

Diatom cells are unicellular eukaryotic algae with a very characteristic and unique siliceous cell wall which consists of two parts, the valves (Taylor et al. 2007b). The capsule-like cell wall is referred to as the frustule. The two valves, called thecae, are almost identical in size but the older, larger valve is known as the epivalve and the slightly smaller younger valve is known as the hypovalve. Each valve is comprised of two parts; the valve face and valve mantle. Between the two valves are girdle bands (Figure 2). There are two ways a diatom could be facing the observer; valve view and girdle view. When viewed at from the top, the diatom can either be positioned in such a way that the observer is looking at the valve face, this is valve view, or at the girdle bands on the side of the diatom, this is known as girdle view (Gell et al. 1999).

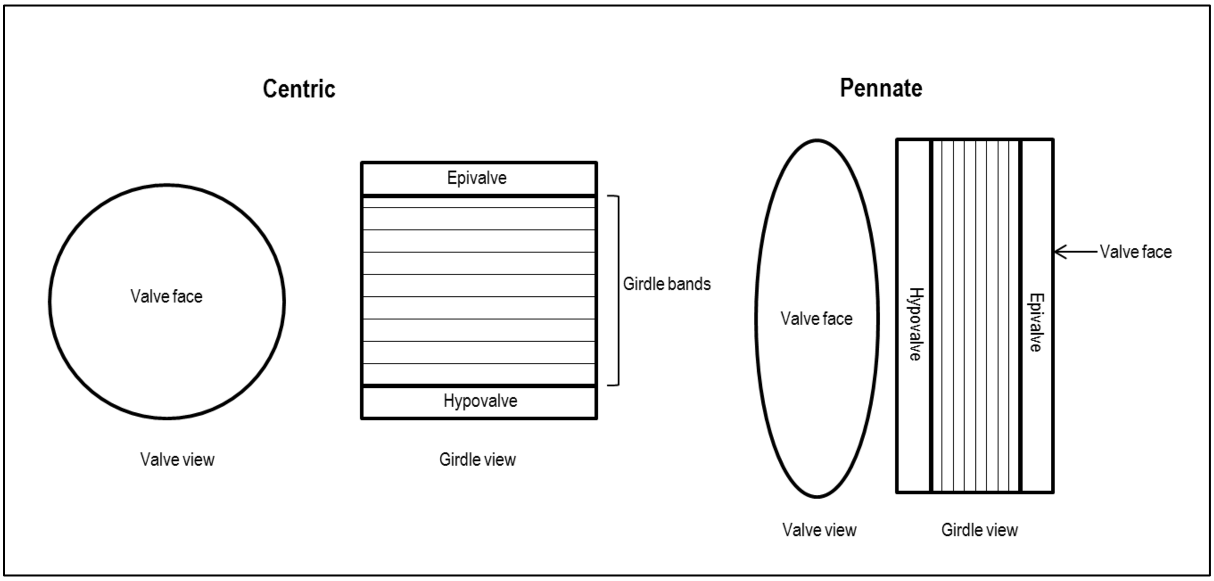


Figure 2 The difference between centric and pennate diatom frustule structure as well as the difference between valve view and girdle view. In girdle view the girdle bands are visible (Adapted from Taylor et al. 2007b).

Diatoms are divided into two groups based on the symmetry of their valve shapes. Centric diatoms have radially symmetrical valves, while pennate diatoms have bilaterally symmetrical valves. Pennate diatom shape descriptions are based along two axes; the apical and transapical (Figure 3). These two axes are the basis for how pennate diatom symmetry is described; heteropolar which means the diatom is asymmetrical around the transapical axis, isopolar meaning the diatom is symmetrical around the transapical axis and dorsiventral which means the diatom is asymmetrical around the apical axis (Gell et al. 1999).

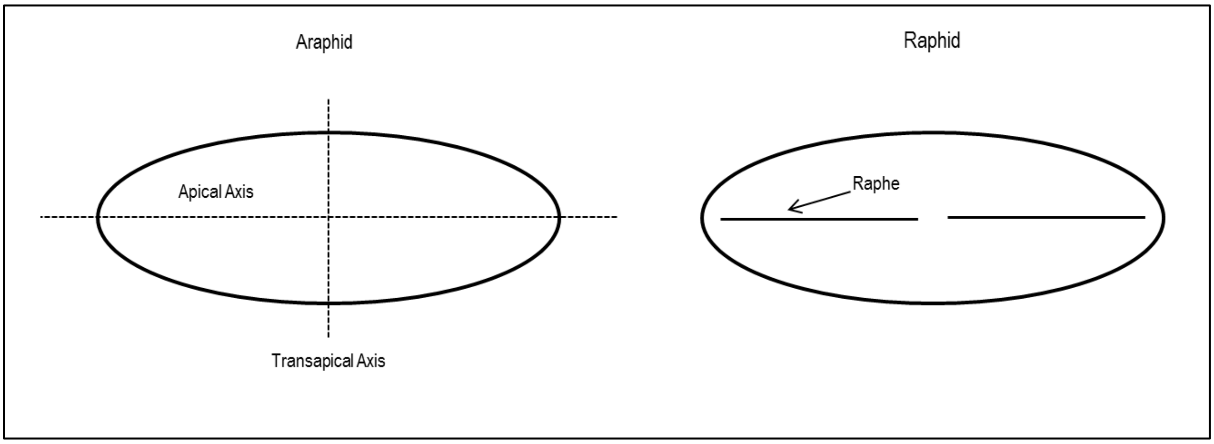


Figure 3 Pennate diatoms are described based on the apical and transapical axis. Some species do not have a raphe (araphid) and others do (raphid) (Adapted from Taylor et al. 2007b).

The raphe is a longitudinal slit-like structure on the valve from which mucilage emanates; this is referred to as Extracellular Polymeric Substance (EPS). Pennate diatoms are divided into two groups based on the presence (raphid diatoms) or absence of (araphid diatoms) a raphe. The raphe can be continuous from one point to the other, or it can be interrupted in the center of the valve (Figure 4). The raphe has proximal and terminal ends. Terminal ends are at the end of the valve and proximal ends are situated at the central area (Gell et al. 1999). Diatom valves are usually covered in punctae. The punctae are tiny pores arranged in lines which are called striae (Figure 4). The arrangement and composition of striae are key features used for diatom species identification (Gell et al. 1999).

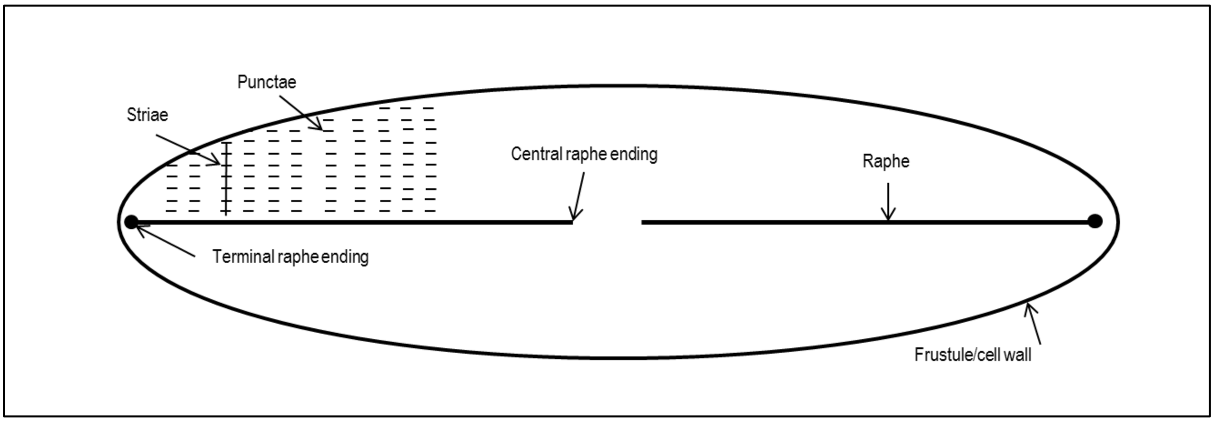


Figure 4 Basic external pennate diatom cell (adapted from Taylor et al. 2007b).

Valves can have many different variations of shapes within the pennate group (Figure 5 A-C & Figure 8 A-V). Valve apices have many variations in morphological shape (Figure 6 A-J) and these together with the different striae shapes and patterns (Figure 7 A-D) are among the characteristics on the frustule, used for taxonomic identification.

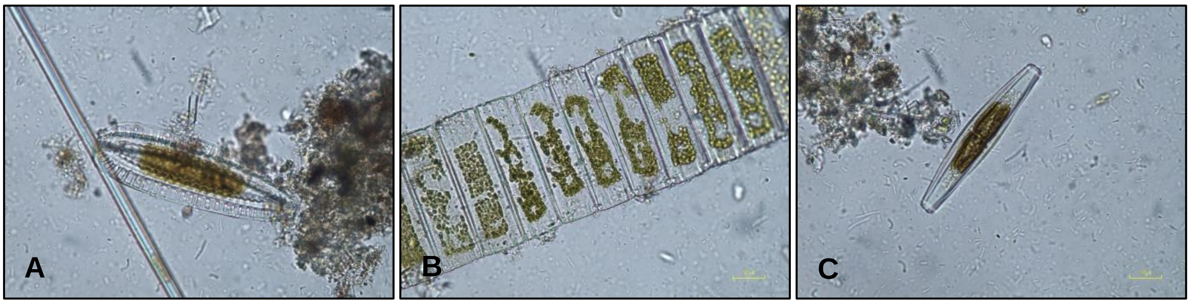


Figure 5 (A-C) Pennate diatom frustules can have a variety of shapes, sizes and external features aiding in their taxonomic identification as seen here with three very different pennate diatoms.

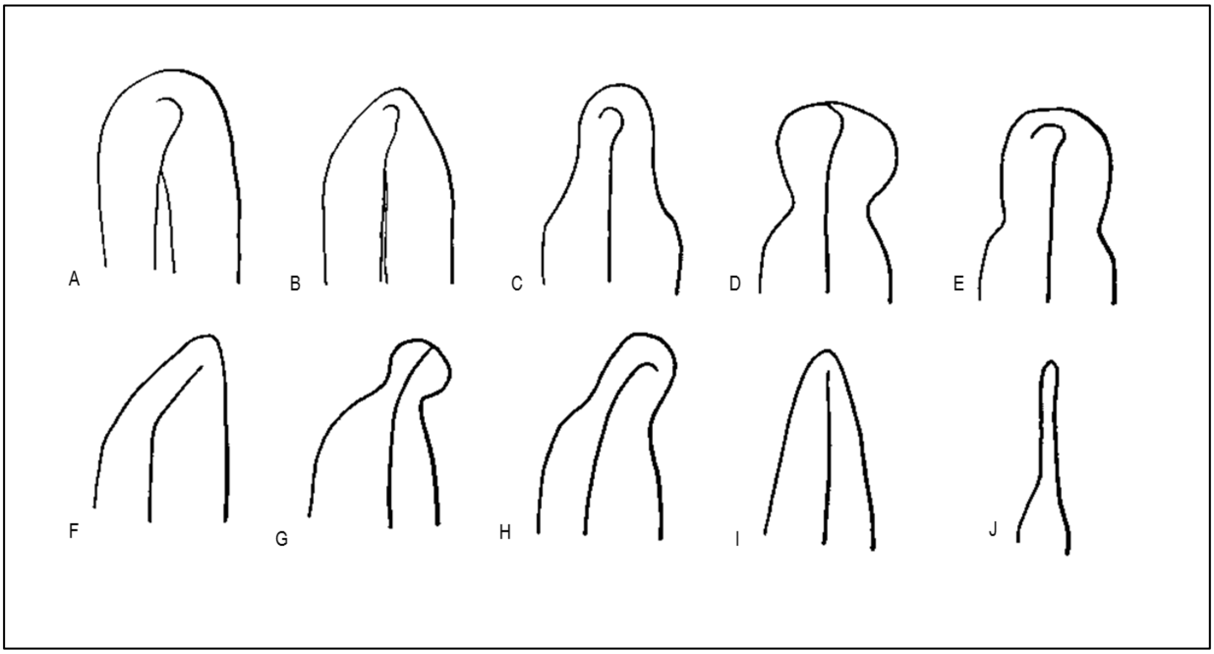


Figure 6 Valve apex variations in pennate diatoms (A) obstusely or broadly rounded; (B) cuneate; (C) rostrate; (D) capitate; (E) subcapitate; (F) sigmoidly cuneate; (G) sigmoidaly capitate; (H) sigmoidaly rostrate; (I) acutely or sharply rounded; (J) elongate (Adapted from Taylor et al. 2007b).

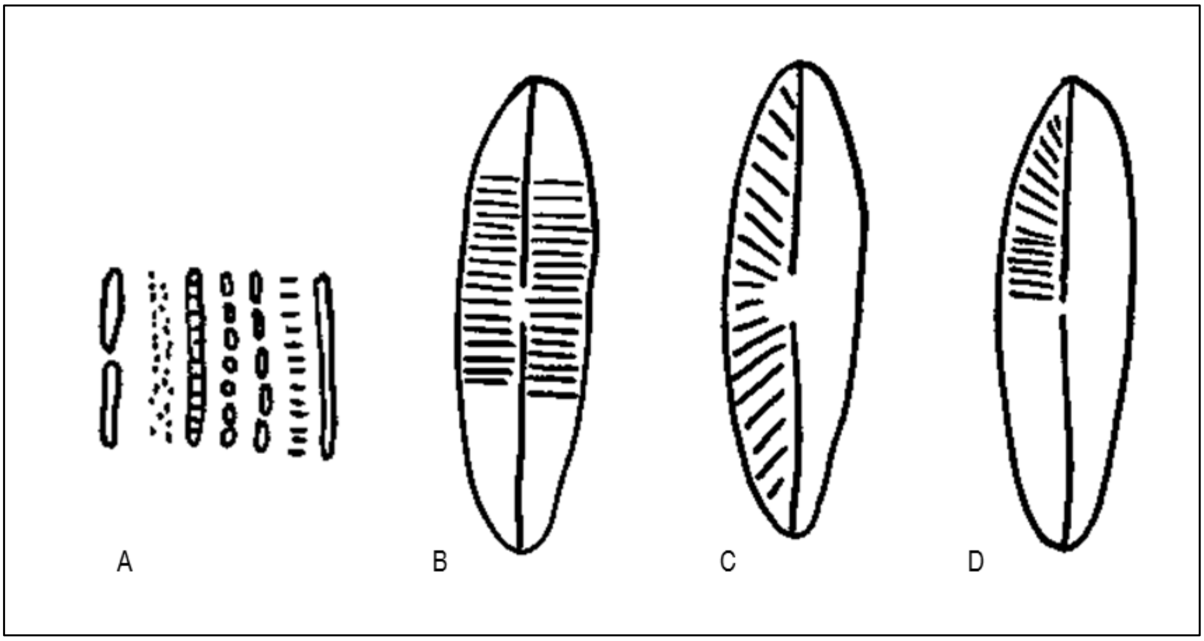


Figure 7 (A) Striae patterns and different types of striae (B) striae parallel, (C) striae radial and (D) striae parallel tending to become convergent at the ends (Adapted from Gell et al. 1999).

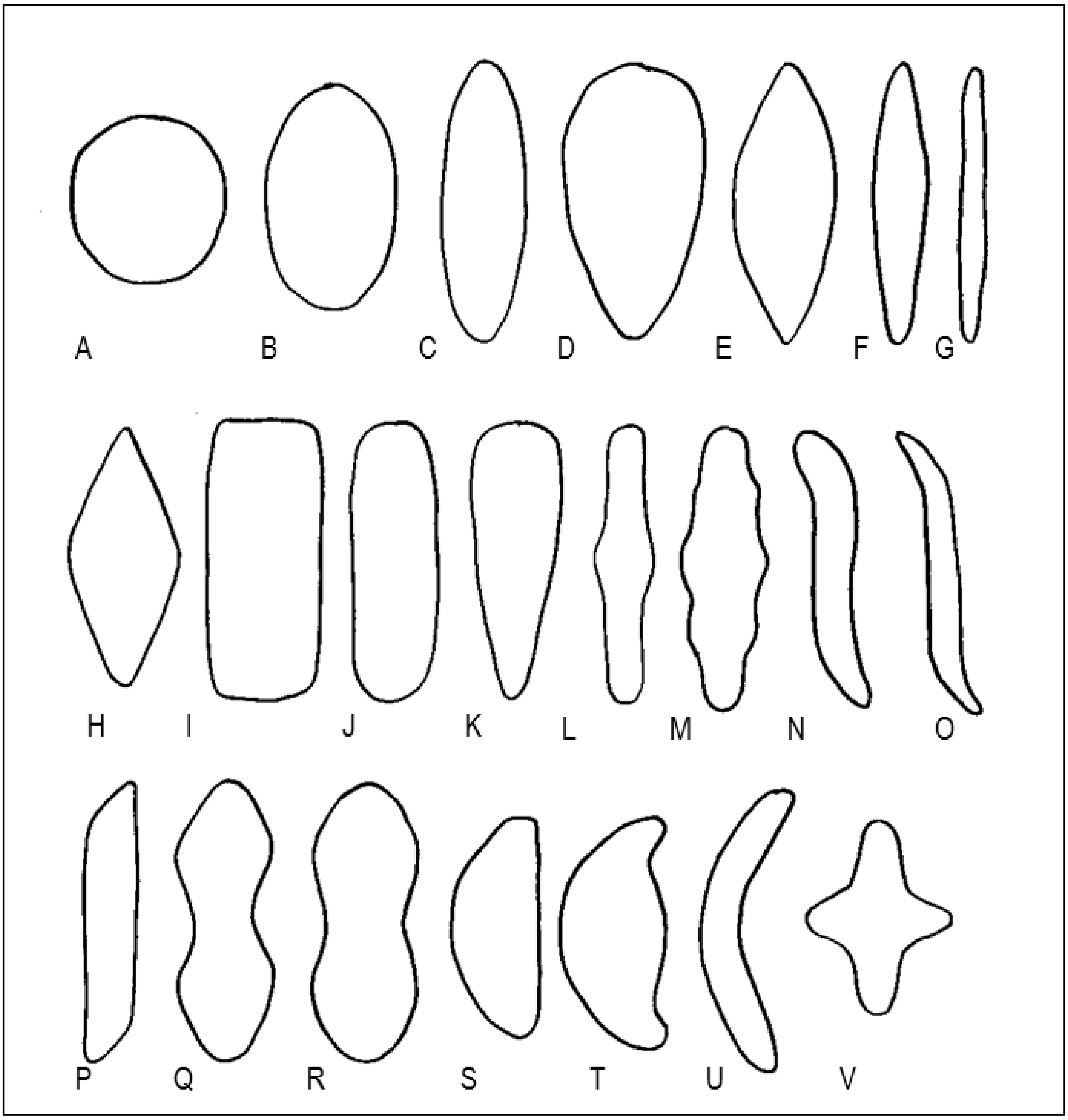


Figure 8 Valve and girdle shapes found in diatoms. (A) circular; (B) elliptical; (C) narrow elliptical; (D) ovate; (E) broadly lanceolate; (F) lanceolate; (G) narrowly lanceolate; (H) rhomboidal; (I) rectangular; (J) linear; (K) clavate; (L) linear with swollen mid-region; (M) triundulate; (N) sigmoid; (O) sigmoid lanceolate; (P) sigmoid linear; (Q) paduriform; (R) panduriform, somewhat constricted; (S) semi-circular; (T) semi-circular ventral edge swollen; (U) lunate or arcuate and (V) cruciform (Adapted from Taylor et al. 2007b).

Physiology

The diatom protoplast contains the same organelles as other eukaryotic algae i.e. a nucleus, plastids, dictyosomes, and mitochondria (Round et al. 1990). While most centric and some pennate diatoms have many chloroplasts, most pennate diatom cells only have one or two chloroplasts. The chloroplast can have a range of different shapes, which may include; C-shaped, H-shaped and lobed (Figure 9 A&B). The number of chloroplasts, the shape and position of the chloroplast in the cell are among the key characteristics used in identifying live diatoms (Cox and Shubert 2015).

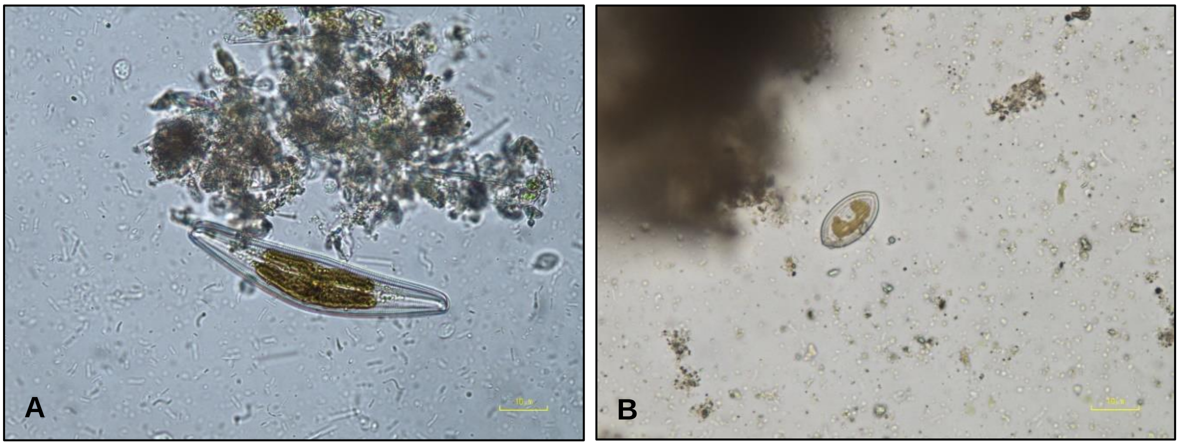


Figure 9 Chloroplasts can have different shapes and positions within the frustule. (A) Example of an H-shaped chloroplast and (B) C-shaped chloroplast, as seen in living diatoms collected in this study.

Chloroplasts are yellow-brown in colour and have four membranes containing pigments such as carotenoid fucoxanthin and β-carotene. Pigments chlorophyll a and c are used to photosynthesise. Energy is stored in the form of chrysolaminarin, being a carbohydrate and lipids which it stores in the form of oil (Round et al. 1990). The high production of lipid makes many species a wonderful source of biofuel (Kumar 2015). As one of the most important global sources of carbon fixation, diatoms are a key component of aquatic food webs. Approximately 40% of the earth's oxygen is produced by the photosynthetic processes in diatoms.

Individuals lack flagella, but they are present in male gametes of the centric diatoms. The silica which gets deposited externally in the frustule is synthesised intracellularly through polymerisation of silicic acid monomers. The exact process of depositing synthesised silica to the cell wall is still unknown and a lot of diatom gene sequencing comes from the search for this mechanism of silica deposition to the frustule (Thamatrakoln et al. 2006). Recently Javaheri et al. (2014) produced a mathematical model for analysing silicon pathways in the diatom *Thalassiosira pseudonana*. Another unique feature of diatoms is the presence of a urea cycle. Allen et al. (2011) discovered that diatoms have a functioning urea cycle, which was significant because up to that stage it was believed that the urea cycle originated in the metazoans, which only appeared several millions of years after the diatoms. Although diatoms and animals use the urea cycle for different purposes, diatoms are now seen to be evolutionary linked to animals.

Lifecycle and reproduction

Diatoms generally reproduce asexually by binary fission. The cell divides in two and the frustule splits leaving the epivalve with one daughter cell and the hypovalve with the other daughter cell. Each of these two daughter cells need to produce the other half of the frustule to form a complete diatom cell again. To do this, both daughter cells use the valve half they received as an epivalve and generate a hypovalve. As the hypovalve is always slightly smaller than the epivalve, fitting within the epivalve like a pillbox, it is found that the average cell size in a colony gets smaller after every division cycle (Figure 10). When the diatom cell reaches a critical point in cell size it becomes equipped to undergo sexual reproduction. Sexual reproduction is therefore used as a method to restore cell size to its original and optimal state (Round et al. 1990).

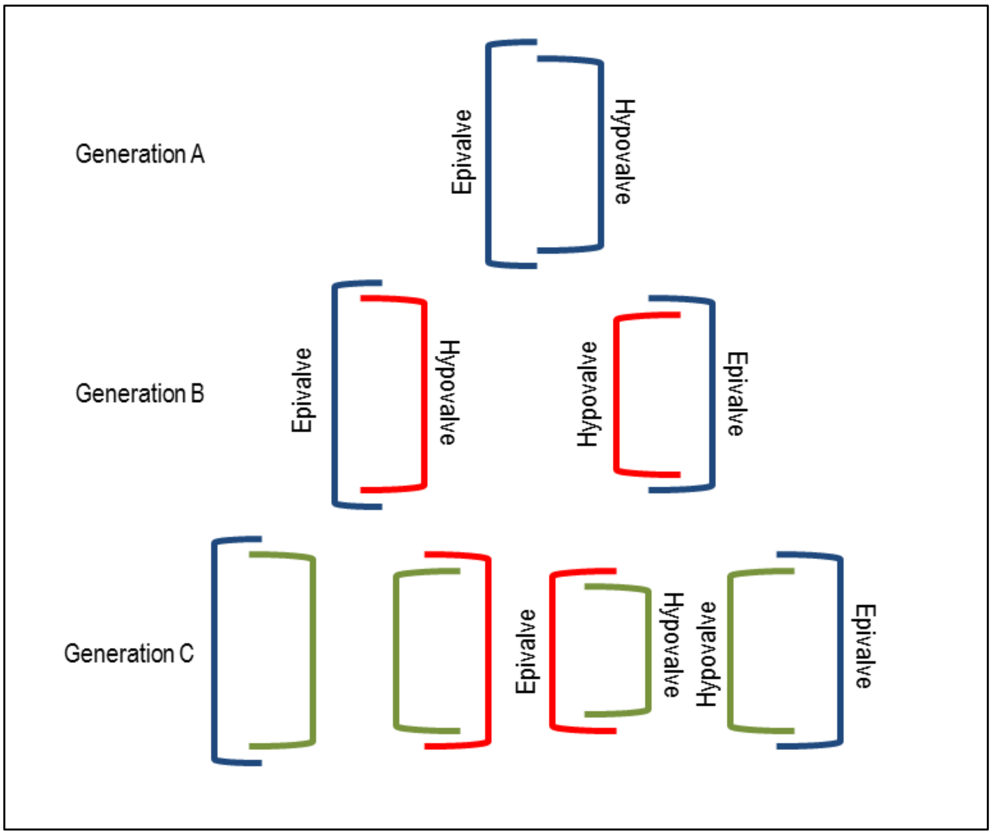


Figure 10 Vegetative cell division or asexual reproduction leads to a reduction in average cell size in diatom populations from one generation to the next.

In Pennate diatoms sexual reproduction (Figure 11) occurs by means of binary fission or with valves pairing up and gametes migrating to fuse. Pennate diatoms are usually isogamous which means the male and female gametes are identical, while centric diatoms are normally oogamous which means they produce motile male and immotile female gametes (Round et al. 1990). The gametes fuse to form a zygote. The zygote discards the silica theca and grows into a large sphere, the auxospore, which is enclosed by a membrane. When the auxospore reaches maximum size it stops growing, new valves are laid down externally and a new generation is produced.

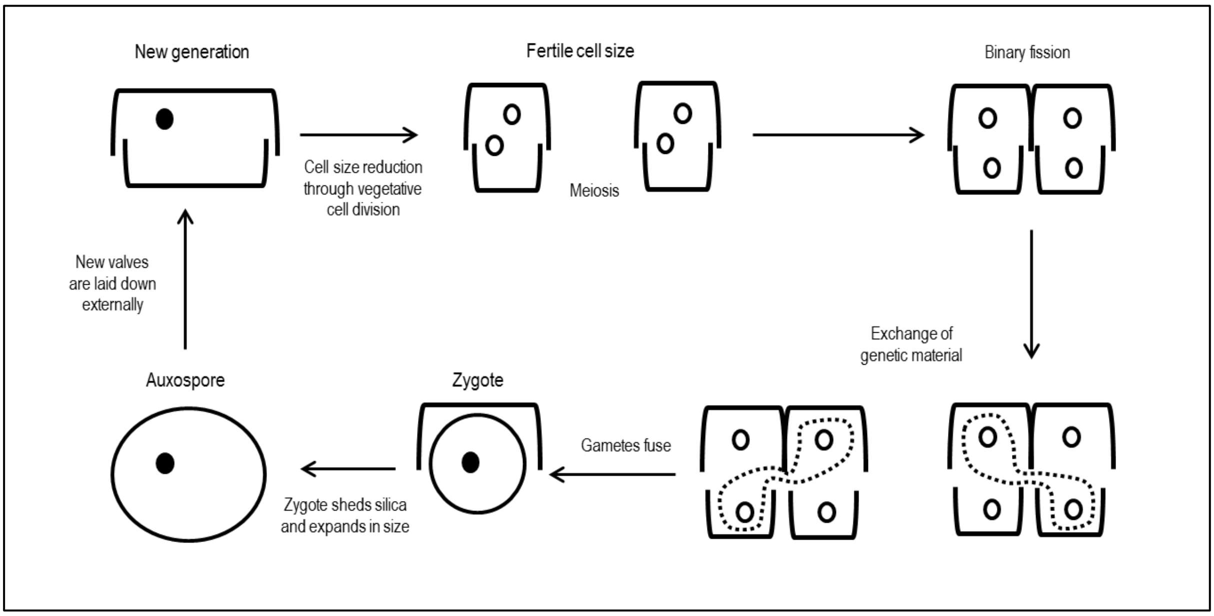


Figure 11 Sexual reproduction in pennate diatoms. Fertile cells undergo meiosis and binary fission through which a zygote and then auxospore is produced. The auxospore is capable of growing in size and so restoring the average cell size in a population (Adapted from Round et al. 1990).

The history of diatomology

The very first certain record of diatoms was made by an English country gentlemen in 1703 viewing pondweed roots (*Lemna*) through a microscope. His diagrams and descriptions referring to pretty branches adhering and floating in the water was most likely what we now refer to as *Tabellaria flocculosa* (Round et al. 1990). The first formal description of a diatom in scientific literature was only done in 1783 by the Danish naturalist Otto Friedrich Müller and it was of colonial *Bacillaria paradoxa*.

The history of diatoms and ecological studies can be separated into three eras (Stoermer and Smol 1999):

1. Exploration 1830 – 1900

During this period work on diatoms was largely descriptive and based on the discovery of new taxa. The main aim was describing their biology, lifecycles, geographic and temporal distributions. The era of exploration for diatoms is not over and new information regarding species descriptions continues.

2. Systematisation 1900 – 1970

During this era the large volume of information was reduced to manageable and useful contexts. This was the period in which indices were starting to develop and they are continually improved and simplified to this day. Reducing the clutter of information to simplify tools for decision makers and managers are a continuing goal of ecological diatomology.

3. Objectification – 1970 – present

The technological advances have enabled tools to be applied in a way that more accurately assesses the variables influencing diatom community ecology. Measuring the changes quantitatively and with great precision is proving to be of great use to environmental managers and decision makers. In the context of climate change and increased threats to sustainable potable water resources, the ever increasing developing tools for descriptive and predictive diatom ecological community based assessments are proving very exciting and powerful for water management.

South African diatomology

South Africa has a long history of diatomology (Harding and Taylor 2011) and one of the most comprehensive diatom collections in the world. This collection was previously housed at the Council for Scientific and Industrial Research (CSIR) offices in Durban but was moved to the North-West University in recent years where it is curated by Dr. Jonathan Taylor. The majority of historic information contained within this collection was collected by four botanists during the early to mid-20th centuries. They were Drs. BJ. Cholnoky, M. Giffen, REM. Archibald and FR. Schoeman. But long before these collections were made, the South African diatom flora was receiving attention from as early as 1845 (Harding et al. 2004). This work was continued into the 19th century by Fritsch, Rich and later the already mentioned mid-20th century diatomists. One of these early collectors was Malcolm Giffen who published valuable information on the Hogsback freshwater species in the 1960's (Taylor 2004). Sadly most Southern Cape samples collected by these early diatomists have gone missing, and are no longer contained within the National Diatom Collection leaving a considerable information gap in terms of reference condition, verification of historic information and assessment of environmental change in current and future monitoring initiatives.

Since these early collections were made many of the freshwater systems in South Africa have changed. Inter-basin transfer schemes, dams, channels and abstraction schemes are examples of the more permanent infrastructural developments which have occurred on freshwater ecosystems. The early collections contained within the National Diatom Collection provide a valuable historic snapshot of what our natural systems looked like before these developments were made (Harding et al. 2004).

Apart from significant contributions in terms of information relating to historic species distribution and taxonomic information, early diatomists greatly contributed towards the development of the use of diatoms as tools to assess water quality. Cholnoky (1968) adapted the Thomasson (1925) community analysis to determine water quality and obtained good results but the method was too complex and ended up being a forerunner for modern autecological indices, which are more accurate and less complex for the user. Archibald (1972) tried to use species diversity to assess water quality. The diversity approach failed to be a good indicator of water quality even though the approach was a parallel development to that of European countries at the time. Schoeman (1976) used indicator groups to try and assess water quality. This approach was based on the Thomasson method used by Cholnoky but a simplified version. This simplified method divided diatoms into four groups based on their ecological requirements and using these as an indication of trophic status in running water. In 1979, Schoeman tested a new approach developed by Lange-Bertalot (1979) which was based on a "saprobian" classification system. Schoeman (1979) found a very good correlation between species composition and water quality in the Hennops River but unfortunately this was the end for studies using diatoms as bio-indicators in South Africa. The next time diatoms were to be assessed as indicators of water quality in South Africa would be by Bate et al. (2002).

The South African Diatom collection contains historic records from the following rivers that were sampled during 2014 - 2015 forming part of the present study; Bloukrans, Fish and some Southern Cape Rivers. The level of information however is very variable in terms of space and time (Harding et al. 2004).

The South African Diatom collection was in almost permanent disuse since the early 1990's but the recently renewed interest in diatomology has seen some interesting publications and projects arising, which included the utilisation and revival of the South African Diatom Collection. One such example is a Water Research Commission (WRC) funded project (Harding et al. 2004) during which information on 70 genera and 286 species from South

Africa were added to an online diatom identification key. This key includes cosmopolitan species as well as some known endemics.

Diatoms as environmental indicators

Freshwater habitats are complex systems, especially rivers and streams where biophysical components change rapidly and could have great impacts on the functioning of ecosystems. In ecological studies, many different aspects of physical, chemical and biological aspects need to be measured in order to determine drivers of change, direction of condition and how to effectively manage these changes. Measuring every aspect of chemical and physical attributes of river ecosystems would be impractical, costly and extremely time consuming. Monitoring the biological components of freshwater habitats, a spectrum of responses can be integrated into a result, with lower costs and time spent collecting information. Biomonitoring of species with different lifespans and habitat preferences allows the observer to form an integrated result, based on the responses of the biological components to the changes of physico-chemical aspects associated with the ecosystem. Another good reason to monitor the biological constituents of an ecosystem is by realising the importance of biodiversity (Campbell et al. 2009). Many programmes today are aimed at effectively managing and conserving biodiversity and consequently its ecological functioning and associated ecosystem services (Stoermer and Smol 1999).

Diatoms are one of the most species-rich components of rivers and streams. They are important genetic resources and form a very large and important component of the biodiversity in these habitats[26]. Diatoms are excellent indicators of change due to the fact that they have the shortest generation times of all biological indicators. Diatoms have high sensitivity to physical, chemical and biological changes in the habitat, which together with their rapid lifecycles mean their response to changes is also rapid. They occur in large diverse numbers in all freshwater habitats which provide more significant statistical results and the information can be stored for long periods for future analysis or long term monitoring records.

Diatoms and pH

Diatoms have been found to be very good indicators for acidity with the very first link between diatoms and pH made by Hustedt (1939) and later by Stoermer and Smol (1999). Hustedt found that diatoms had different pH preferences or tolerances and consequently

[26] Genetic resources are one of the three headline objectives of the Convention on Biological Diversity (CBD). This means that the benefits arising from genetic diversity should be equitably shared and accessible while being utilised in a sustainable manner. Issues of rights, origin, access and informed consent regarding genetic resources are contained within Article 15 of the CBD.

proposed that diatoms could be classified based on this relationship into five groups. This classification system has proven very useful over the years and is still used to this day with the exception of his description of diatoms with weaker relationships to pH as being indifferent. This group has since been modified to be referred to as circumneutral. Below is the updated classification of diatoms based on their preference or tolerance to pH:

1. Alkalibiontic: pH values >7
2. Alkaliphilous: pH about 7 with largest distribution at pH>7
3. Circumneutral: Occurs at pH < and > 7
4. Acidophilous: pH about 7 with the largest distribution at pH < 7
5. Acidobiontic: pH values <7 with optimum distribution at 5.5 or below

This classification system is not without its limitations and when used to reconstruct historic acidity information, there are some problems associated with using this system. However, in terms of assessing naturally acidic environments, such as the Garden Route Rivers in South Africa with high levels of tannins present, it could be a useful guide for indication of rapid changes in water chemistry. Although the use of diatoms as indicators of acidification has predominantly been for lentic systems, they are increasingly being used as indicators of acidity in lotic ecosystems (Stoermer and Smol 1999). More recently an updated classification system by Van Dam et al. (1994), which is derived from Hustedt (1939), is used when describing diatoms' pH sensitivity.

Diatoms and eutrophication

Eutrophication is the process of aquatic ecosystems becoming more nutrient rich. This usually occurs when phosphorus (P) and nitrogen (N) concentrations increase and subsequently leads to an increase in available organic nutrients. This elevates the productivity in the system (Wentzel 1983). Eutrophication can be caused by natural disturbances or anthropogenic impacts. Natural drivers of eutrophication include among others fire, erosion, climate change and periods of drought. Anthropogenic drivers of eutrophication are usually agricultural activities, excessive extraction of surface flow and sewage inputs.

Eutrophication is more often than not an indication of anthropogenic disturbance and leads to major changes in the primary production (which increases) as well as biochemical and biological cycles. Eutrophication influences all levels of the food web and the changes could be so severe that entire communities die out. An example of this is provided by Stoermer and Smol (1999) on how the change in N:P ratio in a freshwater body can lead to a shift in primary production from diatoms to larger algae such as cyanobacteria. These larger algae outcompete diatoms for N. Eutrophication has impacts beyond the ecological functioning of the freshwater body it occurs in. Water abstracted from eutrophied sources is more expensive to process for human consumption (Misra and Mani 1993). The toxins in eutrophic water are dangerous for humans and livestock and the algal breakdown process may lead to metal contamination of drinking water (Abdel-Raouf et al. 2012). All of these are very costly problems to deal with and eutrophication therefore becomes a very important environmental management issue, especially in areas under climate change induced risk and water security and service delivery pressures such as the case with the African continent in particular.

Diatoms are good indicators of eutrophication due to specific species having specific water chemistry requirements. Diatom communities respond rapidly to changes in water chemistry and therefore are early indicators of environmental change and specifically also eutrophication. Due to diatoms being photoautotrophic organisms they are sensitive to changes in light and nutrient availability (Stoermer and Smol 1999).

Diatoms and hydrology

Hydrological conditions influence the water levels in lotic ecosystems. Water levels influence diatom communities, due to the water volume and flow rate impacting nutrient availability in these ecosystems. River ecosystems have natural flow cycles which are usually seasonally induced, although anthropogenic impacts such as abstraction, dam construction and channel modifications also impact on the natural hydrological flow pattern (Stoermer and Smol 1999). However, due to the fact that most lentic systems are fed by upstream lotic systems, it is important to take into account that diatom communities are responding to changes in hydrological seasonality (nutrient levels are impacted directly by flow), and could therefore be used as indicators and biomonitoring tools to assess the impact of changes in natural hydrological patterns.

During low flow seasons the carrying capacity[27] of rivers declines and substrate together with organic material is deposited in the active channel. During high flow periods debris and fine sediment gets washed downstream. This deposition of organic material, whether it is natural or anthropogenic, influences the biochemical cycle as discussed above in eutrophication (extreme cases). Natural fluctuations should not impact the natural biological and biochemical cycles too drastically. However, with climate change induced droughts and

[27] In this context carrying capacity refers to the size of substrate the active channel is able to transport downstream and not the new water resource carrying capacity (WRCC) concept emerging to calculate the maximum population size a resource can sustainably sustain (Kang and Zu 2012).

floods increasingly working in on both impacted and pristine ecosystems, the impacts could become exacerbated making it an important environmental monitoring aspect of sustainable potable water availability.

Diatoms and conductivity

For diatoms to be effective indicators of environmental change there needs to be sufficient silicate present in the ecosystem. Stoermer and Smol (1999) noted the importance of diatoms used as good indicators of eutrophication may also thrive in brackish waters. Due to these species having a wide conductivity tolerance and both oligotrophic and eutrophic conditions in brackish waters will have a very high conductivity. The high conductivity is caused by high levels of salt ions. Typical eutrophic sensitive species are therefore not good indicators for eutrophication in brackish systems.

Instead, the cell size volume ratio has been found to be a more accurate indication of eutrophication in brackish systems. Larger diatoms have smaller surface to volume ratios than smaller species which has an advantage to nutrient uptake in brackish environments. Small *Nitzschia* and *Amphora* species were found to be able to out-compete larger species with N and P addition to the systems (Sundbäck and Snoeijs 1991). In addition to diatom cell size being a driver in community composition, an overall decrease in number of diatom taxa has been found to be an indication of eutrophication in the Baltic Sea (Stoermer and Smol 1999).

Diatoms and climate change

Temperature is a key regulating factor of biological processes. Natural climatic changes or seasonal changes have always impacted the biota on the planet. In freshwater ecosystems, communities are adapted to the hydrological regime, seasonal availability of nutrients, habitat and even dispersal methods. The recent changes in climate patterns have been driven by anthropogenic activities such as the burning of fossil fuels (DEA 2012). The addition of habitat loss and modification increases the pressure on ecosystems, especially in confined habitats such as freshwater corridors.

The gradual increase in atmospheric temperate is expected to have the greatest climatic impact on Africa (Williams 2015). Increased frequency and intensity of drought and rainfall spells are expected, which in turn means an expected increase in regularity and severity of flooding and drought. With water being a vital resource for human existence the pressures associated with sustainable potable water quality and availability is an increasing concern for environmental and urban resource managers world-wide, especially in developing countries (DEA 2014). River watersheds are vital sources of sustainable water provision in South

Africa, which are the areas that form the focus of this study. Effective and comprehensive monitoring of these resources are therefore of utmost importance. Diatoms play a very important role in detection of early changes in water chemistry and the consequent changes in biochemical and biological cycles as discussed above (Anderson 2000, Bate et al. 2002). Increasing demand from water users, agricultural activities and nutrient inputs could lead to severe changes in ecological functioning and resilience in river ecosystems. In light of all these threats freshwater ecosystems would more effectively be monitored by including diatom community information to our already existing and well established river monitoring tools. Diatoms act as an early warning and detection mechanism; this could provide valuable information for decision makers to sustainably manage water resources and associated developments in a climate unsure future.

Diatoms as environmental assessment tool

Diatoms are excellent freshwater monitoring tools and are very useful for assessing general water quality or in monitoring a specific aspect of water quality such as eutrophication and acidity (European Guidance Standard, 2003). Diatoms provide a range of information for diagnosing change in river ecosystems. Below is a summary of these diagnostic attributes provided by diatoms as described by Harding et al. (2004) and 20 years prior by Schoeman and Hayworth (1984):

- Diatoms have a broad range of tolerance along a gradient of aquatic productivity.
- Individual species have specific water chemistry requirements.
- Diatoms have one of the shortest generation times of all biological indicators.
- Diatoms provide early warnings signs of increased pollution and habitat restoration.
- Diatoms are sensitive to changes in nutrient concentrations.
- Each taxon has specific optimum and tolerance for nutrients (N and P).
- Communities have a high species diversity which increases the confidence of results.
- Diatoms respond rapidly to eutrophication.
- Lack of physical dispersal barriers ensures little lag time between perturbation and response of diatoms.
- Diatoms’ siliceous cell walls provide historical information long after they have died and been deposited in the sediment.
- Diatoms are ideal for reference condition tracking and climate change monitoring and ecosystem functioning, before development such as dams etc.
- The taxonomy of diatoms is well documented and studied due to the easily identifiable morphology and consistency of characteristics.

- Diatoms occur in stream beds even when dry, which enables sampling at different times of year.
- Diatoms are easy to collect, prepare, observe and store for reference purposes.
- There is a considerable volume of tried and tested ecological-associative information available.
- Diatoms are suitable for biodiversity analyses.
- OMNIDIA and other software developments exist to aid interpretive processes.

The most sensitive response of diatoms to environmental changes is at community level (Stoermer and Smol 1999). Brinkhurst (1969) was one of the first authors to criticise the indicator species approach and instead advocated for a community-based approach. Frequency of occurrence (relative abundance %) of a species is a better indication for species richness, as it presents the relationship between the numbers of individuals counted per species in a sample in relation to the total number of individuals counted per sample (Stoermer and Smol 1999). The presence or absence of indicator species has become more sophisticated over the years, by now looking at benthic communities and their relative abundances and distribution instead of indicator species alone (Davis 1995). Community composition provides a much more integrated reflection of all the chemical impacts happening at a specific test site.

The use of diatoms for water quality assessment has not been without criticism. In particular, there has been concern regarding accurate species identification and the fact that diatom sampling is not a rapid assessment tool such as the South African Scoring System (SASS5). The problem of needing specialised taxonomic knowledge of diatom species, is ironically similar to the early development stages of the use of macroinvertebrates for water quality assessment in South Africa (Dickens and Graham 2002) and even more so when compared to the processes that followed critique in Europe (Harding et al. 2004).

It was proposed that the problem could be solved by simplifying identification keys and only using a limited number of taxa (Descy and Coste 1991). Prygiel et al. (2002) did just that by publishing a simplified identification key as a guide for the identification of freshwater diatoms in French inland waters. This guide was produced to support the Biological Diatom Index (BDI) developed by Lenoir and Coste (1996) for national river water quality monitoring in France. In Great Britain, a similar guide was developed by Kelly (2000) while at the same time in South Africa Taylor et al. (2007b) was developing guide manuals with the same purpose of simplifying and providing access to these simplified diatom keys.

It may also be possible to use diatoms not only as a diagnostic tool but instead to assess general water quality at a specific sampling site by using genus level data. This would mean that the diatom identification key would be much more user friendly and could be used successfully by technicians not possessing specialised diatom knowledge. The indices used would then also need to be simplified, which is exactly what Coste and Ayphassorho (1991) did when they developed the Generic Diatom Index (GDI). The GDI has since been found to be extremely accurate and highly comparable to the Specific Pollution Sensitivity Index (SPI) by researchers such as Wu and Kow (2002) who found the genus level index approach to produce very good results in Taiwan. They developed a Taiwanese Generic Index which was found to have strong correlations with other diatom indices used for water quality assessments. It is worth noting that the Taiwanese Generic Index only uses information of six genera and their ratio of occurrence. Although this approach would produce a much less detailed diagnostic of possible causes for changes in water quality, it is still very useful as early indicator of changes in general water quality.

The use of diatom protocols for water quality assessment has become more developed and refined, in order to effectively include diatoms into biomonitoring programmes and projects. An example of this is the European Guidance Standard which issued a protocol on diatom sampling for rivers in 2003. This European Guidance Standard covered aspects of sampling and pre-treatment relevant to assessment of water quality using benthic diatoms (European Guidance Standard 2003). Using diatoms as indicators for trophic status has become an increasingly popular tool for river assessments and management practices. Some of the more well-known and used indices are the Biological Diatom Index (BDI) which is very popular in France and the European Diatom Database (EDDI) which is used more widely (Harding et al. 2004). The inclusion of a diatom assessment index, has for instance become mandatory for all large freshwater development's environmental assessments and monitoring efforts. The World Bank now expects all funded water related developments to include algal assessments for example as in the case of the Cheves Hydro Project in Peru, a few years ago (Norconsult and Southern Waters 2012).

There are many different diatom indices, all contained within the OMNIDIA data-base computer software which also serves as an indicator calculation tool (Lecointe et al. 1993). OMNIDIA and its index calculations have been tested across the world on a wide variety of river ecosystems, hemispheres and different levels of impact. The indices and consequently also the OMNIDIA software have been found to be extremely reliable and is therefore the most acceptable and commonly used method for biomonitoring. Most of the indices make use of two values. These values are assigned to a diatom species as follows; the first is a value that reflects the sensitivity or tolerance to a certain water quality either good or bad, the second is descriptive of the strength of this sensitivity/tolerance. These two values are then brought into context with the relative abundance (number of individuals of a species relative to the total number of individuals identified) of a species within a sample[28].

The European Guidance Standard is a protocol that broadly divides diatom assemblages in five water quality classes (Table 2). The guide is also a standardisation tool for diatom collection, preparation and processing.

Table 2 The European Guidance Standard 2003 water quality classes and associated diatom species (Sourced from Harding et al. 2004).

Excellent	Good	Moderate	Poor	Bad
Achnanthes peragallii	*Achnanthes minutissima*	*Achnanthes conspicua*	*Actinocyclus normanii*	*Achnanthes delicatulum*
Achnanthes dauii	*Cymbella sinuata*	*Amphora pediculus*	*Amphora romana*	*Amphora veneta*
Achnanthes subatomoides	*Cymatopleura elliptica*	*Cocconeis pediculus*	*Bacillaria paradoxa*	*Navicula accommoda*
Cymbella mesinaum	*Cymbella affinis*	*Cyclotella pseudostelligera*	*Cyclostephanos dubius*	*Navicula arvensis*
Denticular mesodon	*Cymbella lanceolata*	*Fragilaria brevistriata*	*Cyclostephanos invisitatus*	*Navicula atomus*
Denticular tenuis	*Diatoma vulgaris*	*Fragilaria pulchella*	*Cyclotella atomus*	*Navicula cuspidate*
Diatom hiemale	*Fragilaria capucina*	*Gomphonema parvulum*	*Cyclotella meneghiniana*	*Navicula halophilia*
Eunotia exugua	*Frustulia vulgaris*	*Gomphonema truncatum*	*Gomphonema pseudoaugur*	*Navicula minuscule*
Fragilaria arcus	*Gomphonema olivaceum*	*Gyrosigma attenuatum*	*Hantzschia abundans*	*Navicula molestiformis*
Frustulia rhomboids	*Gomphonema acuminatum*	*Navicula capitatoradiata*	*Hantzschia amphioxys*	*Navicula pymaea*
Gomphonema oliviaceum	*Gomphonema minutum*	*Navicula viridula*	*Navicula goeppertiana*	*Navicula saprophilia*
Gomphonema rhombicum	*Navicula ignota*	*Rhoicosphenia abbreviata*	*Navicula hungarica*	*Navicula veneta*
Meridion circulare	*Navicula radiosa*	*Thalassiosira brahmaputrae*	*Navicula mutica*	*Nitzschia capitellata*
Pinnularia gibba	*Nitzschia dissipata*		*Navicula pupula*	*Nitzschia frustulum*
Stauroneis phoenicenteron	*Sellaphora bacillum*		*Navicula subminuscula*	*Nitzschia umbonata*
			Nitzschia filiformis	
			Nitzschia palea	
			Surirella ovalis	

However, no one group of organisms is ideal for biomonitoring purposes (Kelly 2002). If the aim of biomonitoring is to detect environmental changes and maintain ecological integrity of a functioning ecosystem, then a holistic approach to detecting the perturbations caused by humans is a more suitable approach. It is therefore unfortunate that South Africa has yet to include diatoms into monitoring protocols.

Diatoms are able to provide information on specific changes in water quality while the impact of these changes and changes to the physical habitat is better reflected by invertebrates and fish (McCormick and Cairns 1994). The ability of diatoms to provide detailed early indications of changes in water quality is due to their position in the food web. They are often at the

[28] http://wgbis.ces.iisc.ernet.in/energy/water/paper/diatoms_from_aquatic_habitats/indices.htm

base of aquatic food-webs which means they are the first to be impacted by changes in physico-chemical impacts (Cox 1996).

Diatoms make good biomonitoring tools for many reasons, most of which has already been discussed. When such tools are being developed (which have been initiated by WRC work of Harding and Taylor 2011) one concern has been the transfer of information between the Northern and Southern Hemispheres (Round 1991). Diatoms are well known for their cosmopolitan distribution however it has been found that some endemics exist. These endemics are to a large extent not well studied and their ecological ranges and tolerances are still unanswered questions in South Africa. This information gap is made worse by the lack of recent information from many areas in South Africa, especially the Eastern Cape. Although much work has been done to update historic records and data sets, much more work is needed to update and complete the spectrum of information needed in South Africa before the South African Diatom Index (SADI) can be implemented nationally.

Kelly (1998) suggested that geographical location plays no role in the absence or presence of a species but instead that community dynamics are driven by environmental variables at a specific site. This concept is known as "sub-cosmopolitan" and Kelly (1998) continued to explain that diatoms could therefore occur anywhere as long as their environmental requirements are met. Results obtained by De la Rey et al. (2004) supports this theory for "sub-cosmopolitan" distribution in South African rivers and concluded that 97% of the species collected on the Mooi River system (North West Province) were cosmopolitan. These results were translated from the Northern to the Southern Hemisphere with the Specific Pollution Sensitivity Index (SPI) values fully accounted for by physical and chemical variables. De la Rey et al. (2004) concluded that future studies need to cover a larger range of geographic areas and variables to further the development and testing of numerical indices, as indicators of water quality, such as the SPI and SADI.

SASS5 and SADI

South African Scoring System (SASS)

Aquatic invertebrates are very good indicators of water quality in river ecosystems. They respond rapidly to changes and have a wide range of sensitivity to changes in water quality. Their life-cycles are rapid enough to make them very good indicators of short to medium term impacts. Chutter (1998) developed a bio-assessment index tool, the South African Scoring System (SASS), which is currently in its fifth revised form and commonly known as SASS5. This method is applied across South Africa and more recently in other Southern African countries for which a SASS5 species relevant version has been created. SASS5 works well for indicating water quality when determining EcoStatus or current river condition. SASS5 does however have some limitations with regards to being used for biomonitoring of specific changes in water quality (Harding et al. 2004) and the need for adequate substrate availability and diversity of biotopes. Below is a summary of these:

- Flow volume, habitat availability and diversity as well as food availability influence the distribution of macroinvertebrates.
- They are not good indicators of water quality disturbances caused by pesticides and herbicides.
- Some species have uneven distribution patterns and can only be regarded as absent when a high number of samples have been collected.
- Temperature, altitude and latitude are known to influence distribution patterns.
- Community composition may vary greatly longitudinally down the length of a river.
- Seasonal variations of certain species make sampling efforts less accurate during certain times of the year.
- Some lotic species may be carried downstream to areas where they do not naturally occur.
- In South Africa, there is still a great need for more taxonomic work to be completed on aquatic macroinvertebrates.
- Anthropogenic disturbances such as the presence of a flow gauging weir, bridge or dams can impact the biotopes immediately upstream and downstream producing inaccurate SASS5 results.

South African Diatom Index (SADI)

The South African Diatom index (SADI) was developed by Harding and Taylor (2011) as part of a Water Research Commission (WRC) funded project which aimed at providing a diatom water quality index which is specific to South Africa River characteristics. This meant processing 768 individual samples and incorporating known endemic and ecological inferences into the index. The index is created as an extension/upgrade addition to the already existing OMNIDIA software and was based on the same platform as the European SPI. This allows for existing software and indices to be more accurate when used for South African Rivers, since SPI has one of the largest species libraries of all diatom indices currently in use (Harding and Taylor 2011). Due in large to a lack of information, the Western Cape could not be included in the development of the SADI and to a great extent only historical information from the Eastern Cape (contained within the South African Diatom Collection) was used. This is a great opportunity for future ecological riverine diatom assessments since the Western Cape and parts of the Eastern Cape provinces are well known for their high level of endemism[29,30].

Before a diatom index system can be developed for a region, information on the dominant diatom species, where they are located and the water quality information of the location need to be collected (Bate et al. 2004). However, since diatoms are sub-cosmopolitan in their distribution, the necessary ecological tolerance information is already captured in the British and European indices. This makes the use or modification of diatom indices for other parts of the world much simpler since the foundation of these indices such as OMNIDIA software, the Trophic Diatom Index (TDI) and the Specific Pollution Sensitivity Index (SPI), are already well developed (Harding and Taylor 2011). A summary of the most common diatom indices for specific impacts and the corresponding sensitive and tolerant genera are provided in Table 3.

Table 3 Summary of the most common impact specific indices and the corresponding sensitive and tolerant indicator genera[31].

	Salinity Index (freshwater)	pH (acid sensitivity) Index	Oxygen requirement	Sensitivity to N compounds	Trophic status	Sensitivity to Saprobity	Sensitivity to Desiccation
Sensitive genera	*Achnanthes* *Aulacoseira* *Cymbella* *Eunotia* *Gomphonema* *Neidium* *Pinnularia* *Stauroneis*	*Amphora* *Caloneis* *Cocconeis* *Cyclotella* *Diatoma* *Diploneis* *Fragilaria* *Nitzschia* *Surirella*	*Achnanthes* *Cymbella* *Diploneis* *Eunotia* *Fragillaria* *Gomphonema* *Neidium*	*Achnanthes* *Aulacoseira* *Cyclotella* *Cymbella* *Diatoma* *Eunotia* *Neidium* *Pinnularia* *Stauroneis* *Surirella*	*Achnanthes* *Aulacoseira* *Cymbella* *Eunotia* *Neidium* *Pinnularia* *Stauroneis*	*Achnanthes* *Aulacoseira* *Cocconeis* *Cymbella* *Eunotia* *Neidium* *Pinnularia* *Stauroneis* *Surirella*	*Aulacoseira* *Cocconeis* *Cyclotella* *Diatoma* *Fragillaria* *Neidium* *Surirella*
Tolerant genera	*Amphora* *Mastogloia* *Nitzschia* *Surirella*	*Eunotia* *Neidium* *Pinnularia*	*Cocconeis* *Cyclotella* *Nitzschia*	*Amphora* *Cocconeis* *Melosira* *Nitzschia*	*Amphora* *Cocconeis* *Diatoma* *Nitzschia*	*Amphora* *Cyclotella* *Diatoma* *Melosira* *Nitzschia*	*Diploneis* *Eunotia* *Hantzschia* *Pinnularia* *Stauroneis*

Recently there has been increasing number of studies aimed at testing the accuracy and applicability of international diatom indices for specific impacts on South African rivers. Holmes and Taylor (2015) found that diatom indices were useful for biomonitoring semi-arid

[29] https://www.westerncape.gov.za/text/2005/12/04_biodiversity_optimised.pdf (accessed on 17 September 2017)
[30] https://www.worldwildlife.org/ecoregions/at1202 (accessed on 17 September 2017)
[31] http://www.deltaenvironmental.com.au/management/Lab_methods/Generic_indices.htm (accessed on 16 September 2017)

areas. Their study focussed on sites in the upper reaches of the Great Fish River, Eastern Cape (upstream of the town Cradock) and showed significant impact from many decades of agricultural activity. The dominant taxa for these upper areas on the Great Fish River were identified as; *Amphora pediculus*, *Craticula buderi*, *Fragilaria biceps*, *Nitzschia frustulum*, *Nitzschia paleacea*, *Planothidium lanceolatum* and *Rhopalodia gibba*, all of which are considered to be pollution tolerant taxa.

The River Health Programme

The Department of Water Affairs and Forestry (DWAF), the Water Research Commission (WRC) and the Department of Environmental Affairs (DEAT) initiated the National Biomonitoring Programme for Aquatic Ecosystems (NBPAE) in order to address shortcomings in standard physical and chemical monitoring methods for monitoring South African freshwater resources (Hohls 1996). The River Health Programme formed part of the NBPAE and included the biomonitoring of fish, macroinvertebrates and riparian vegetation (Kleynhans and Louw 2007).

The National Aquatic Ecosystem Health Monitoring Programme is a national programme managed by Resource Quality Services with support from the Water Research Commission, CSIR and various regional and provincial authorities. The oldest component of the National Aquatic Ecosystem Health Monitoring Programme is the River Eco-status Monitoring Programme (REMP), formerly the River Health Programme (RHP). The REMP replaced the RHP in 2016 and is a component of the National Aquatic Ecosystem Health Monitoring Programme (NAEHMP) (DWAF 2016).

The lack of modern day diatom information from the Eastern and Western Cape could pose problems if the South African Diatom Index (SADI) would in future be incorporated into the National Aquatic Ecosystem Health Monitoring Programme, since these regions are well known for a generally higher level of endemism compared to other parts of South Africa[32,33]. The first monitoring project which included the diatom index was in the Crocodile (west) and Marico Water Management State of the Rivers report (SoR; RHP 2005) and proved very successful (Taylor et al. 2007c), which led to diatoms being included into the RHP methodology (DWAF 2008a). However, in order to make the SADI applicable across South African Rivers, the index would have to undergo an accreditation process (Harding and Taylor 2011). This process faces two problems at present: 1) trained persons with the adequate broader knowledge on diatom literature and experience for collecting and

[32] https://www.westerncape.gov.za/text/2005/12/04_biodiversity_optimised.pdf (accessed on 17 September 2017)
[33] https://www.worldwildlife.org/ecoregions/at1202 (accessed on 17 September 2017)

identifying diatom samples are needed (Harding and Taylor 2011), and 2) a complete and wide range of tested information on diatoms is needed to initiate baseline work within a national REMP framework. The Department of Water Affairs has already initiated the training of technicians for the implementation of SADI (Harding and Taylor 2011) and it is therefore very important if not urgent to establish a baseline data set for the rivers of the Western and Eastern Cape.

The Ecological Reserve

The Ecological Reserve is an eight-step process which monitors the water quality and quantity of freshwater resources in South Africa. The aim is to establish the flow requirements needed to maintain the instream and riparian biota for optimal ecosystem functioning (Harding and Taylor 2011). EcoClassification is an integral part of Ecological Water Resource Monitoring and includes the REMP and Ecological Reserve. EcoClassification is the determination and categorisation of the Present Ecological State (PES) of a river, based on the assessment of various biophysical attributes, compared to the natural or reference condition (Kleynhans and Louw 2007). The EcoClassification process is a multi-disciplinary approach applied during the Reserve Determination process in order to gain a holistic understanding of the deviation of the PES from the reference condition. The state of a river is expressed by means of two biophysical components; drivers and biological responses. The drivers of river condition are; water quality, geomorphology and hydrology. The drivers inform the habitat template for a river. The biological responses are measured by monitoring fish, invertebrates and riparian vegetation as a biological expression of the response to the habitat template (Kleynhans and Louw 2007).

EcoClassification is expressed as an Ecological Category ranging from A-F, where A represents natural condition and F represents critically modified conditions. Each component or reach is assigned a category and these categories are integrated to attain the EcoStatus of a river. The EcoStatus of a river defines the ability of that river to support natural flora and fauna and relates directly to the capacity of that system to provide ecosystem goods and services (Kleynhans and Louw 2007).

The use of diatoms in Ecological Reserve studies has been virtually non-existent in South Africa. The first time diatoms were used was in the Joint Maputo River Basin Water Resource Study in 2007 (Harding and Taylor 2011). The SPI was applied to indicate general water quality but no interpretive results were derived from the diatoms and they were not included in the EcoClassification process (Koekemoer and Taylor 2007). The diatom results did however correlate very well with the physico-chemical results as was also the case with the Knysna and Swartvlei Reserve Determination during 2006 and 2007 (DWAF 2008b).

More recently there have been a couple of Reserve studies that included diatoms into the EcoClassification process. These were; the Integrated Vaal River System, Upper Vaal Management Area, the Inkomati River System and the Mokolo River system (Harding and Taylor 2011). During these processes multiple sampling efforts were conducted to provide a time-integrated indication of unmeasured chemical conditions, which meant diatoms were sampled during fish and macroinvertebrate sampling as well as during physico-chemical sampling. The ecological category was based on the SPI score and results from 11 other indices (run with OMNIDIA software) were used for interpretive results.

It was found that diatom community composition responded very rapidly to low and high flows. This sometimes led to differences occurring between the physico-chemical results and the diatom index scores. Index scores were influenced by pioneer species and those that thrive in well-oxygenated water; as would be the case during flooding/high flow periods. This elevated occurrence of certain species influenced the index scores. However, this was overcome by looking at the less dominant community components. Diatom information proved very useful especially in places where water quality data were not available or of poor quality (Harding and Taylor 2011).

South African freshwater resources

South Africa is a water-scarce country situated at the southernmost tip of the African continent. South Africa is ranked as the world's third most biologically diverse country in the world[34]. The Western Cape Province is home to two globally recognised biodiversity hotspots; the Cape Floristic Region and the Succulent Karoo[27]. In addition to climate related pressures on water resources such as drought, the country is also classified as developing which means urban poverty, overpopulation and pressure on service delivery are some of the challenges facing freshwater resources (King and Pienaar 2011). Agricultural impacts are increasing due to an increased demand for products leading to more intensive farming practices with higher water demands and often cases of higher agricultural runoff into freshwater systems.

Economic growth and primary industry practices are a necessity even though the impact of these industries on water resources cannot be overlooked. Pollution, under-capacitated waste water treatment systems and lack of service delivery, especially in urban informal communities that are situated close to surface water, are increasing threats to potable and sustainable water provision. The integrated impacts on South Africa's freshwater resources only support and magnify the importance of properly implemented water management

[34] https://www.westerncape.gov.za/text/2005/12/04_biodiversity_optimised.pdf (accessed on 5 January 2018)

strategies. With Africa expected to experience a significant increase in urbanisation, population growth and the biggest impact of a globally changing climate, the mitigation of these and associated impacts on sustainable management and utilisation of water resources will become increasingly important in order to ensure sustainability and climate resilience.

Climate

South Africa has a range of climatic conditions from Mediterranean in the South-Western corner of the country, to temperate conditions on the interior plateau stretching into sub-tropical climate in the North-Eastern corner. In the North-Western corner, there is also a small area which has a desert climate. Rainfall normally occurs in the summer months of November to March however the Mediterranean region experiences winter rainfall which stretches from June to August. Natural variation in these patterns is common and every decade changes in flooding and drought patterns occur throughout Southern Africa. The scale at which these local climate cycles occur are in hundreds of years, if not thousands. However, the impact of industrial and development activities in the past two centuries are changing these natural variations in climate and are expected to continue increasing in severity of variations experienced across the globe[35].

The global climate is being changed due to the increased concentration of greenhouse gasses (GHG) being released into the atmosphere. This causes an overall increase in global temperatures which in turn influences the natural climate cycle by speeding up the natural changes which would have in the past taken hundreds of years to occur[36]. The more GHC are released, the more rapidly the climate changes and this cycle increases in intensity at an exponential rate causing the climate to move toward more extreme weather patterns[37].

Although these extreme circumstances are extrapolated projections at the moment, the impacts of a globally changing climate can already be seen and felt at ground level, especially by the marginalised and urban poor communities of developing countries such as those of Africa. It is alarming to consider that the current climate change projections are showing that Africa will be the continent hardest hit by climatic extremes. This will expose millions of urban poor and already vulnerable people to serious risks and vulnerability in terms of securing resources such as potable water and sustainable environmental practices for food, energy and livelihood security (Midgley et al. 2002).

[35] http://www.weathersa.co.za/learning/climate-questions/42-what-is-climate-change (accessed on 8 May 2017)
[36] http://media.csag.uct.ac.za/faq/qa_3impacts.html (accessed on 8 May 2017)
[37] http://www.enviropaedia.com/topic/default.php?topic_id=123 (accessed on 8 May 2017)

South Africa's mean air temperature is expected to increase by 2°C in the next century. This may not seem like a notable change but the impact this increase in temperature will have on rainfall patterns will be huge. It is expected to cause a drop in annual rainfall in the western parts of the country and an overall increase in annual rainfall in the eastern parts. The increase in rainfall will not be spread over the usual rainfall season but instead occur in a few heavy storms increasing the likelihood of floods. In the western areas, the drop in rainfall will mean droughts will be more severe and occur more regularly[26].

While humans are better equipped to a certain extent to adapt to these changes, especially through increased knowledge and adaptation strategies, plants and animals will not be able to adapt quickly enough to fast changing climate patterns due to the evolutionary time scale. This poses serious threats to healthy ecosystem functioning and the provision of goods and services by these resources. South Africa's climate response strategy has been strongly focussed on climate mitigation and renewable energy development (DEA 2007, 2011, 2014) however more recently a strong shift in focus has been occurring towards adaptation strategy for projected impacts in South Africa (DEA 2016). The question still remains, how can adaptation strategies serve to protect and maintain ecosystem services when rapid changes in climate and demand threatens the resilience and sustainability of these natural resources? Water is one of the planet's most valuable resources. While there is a lot of water on the planet, only 0.3% is available for human use in surface lakes, rivers and wetlands[38]. With an increasing global population, the pressure on water resources also increase and with the additional pressure of climate change, there has never been a more important time to effectively understand and manage this valuable resource.

Water management

Strategic Water Source Areas

Strategic Water Source Areas (SWSAs) are areas that provide a higher than average annual runoff in comparison to the standard or average provided in a geographical region. These areas are of great interest since they sustain the ecological functioning of the freshwater resources and thereby contribute substantially to maintaining water quantity and quality as well as supporting developmental needs (Figure 12). In South Africa, Lesotho and Swaziland it was found that 8% of land area classifies as SWSAs providing 50% of all freshwater[39]. These areas are some of the most important ecological infrastructure in these countries since it supports most of the built infrastructure, industry and water related service provision.

[38] https://www.nationalgeographic.org/media/earths-fresh-water/ (accessed on 5 January 2018)
[39] https://www.sanbi.org/news/strategic-water-source-areas-are-national-assets (accessed on 20 December 2017)

Degradation of SWSAs could lead to a disproportionate impact on water quality and quantity since these areas also contribute disproportionately to the freshwater resource sustainability. The ecosystems downstream from these high yield areas would be severely impacted in terms of sustainability of growth and development as well as potable water provision (Driver et al. 2011).

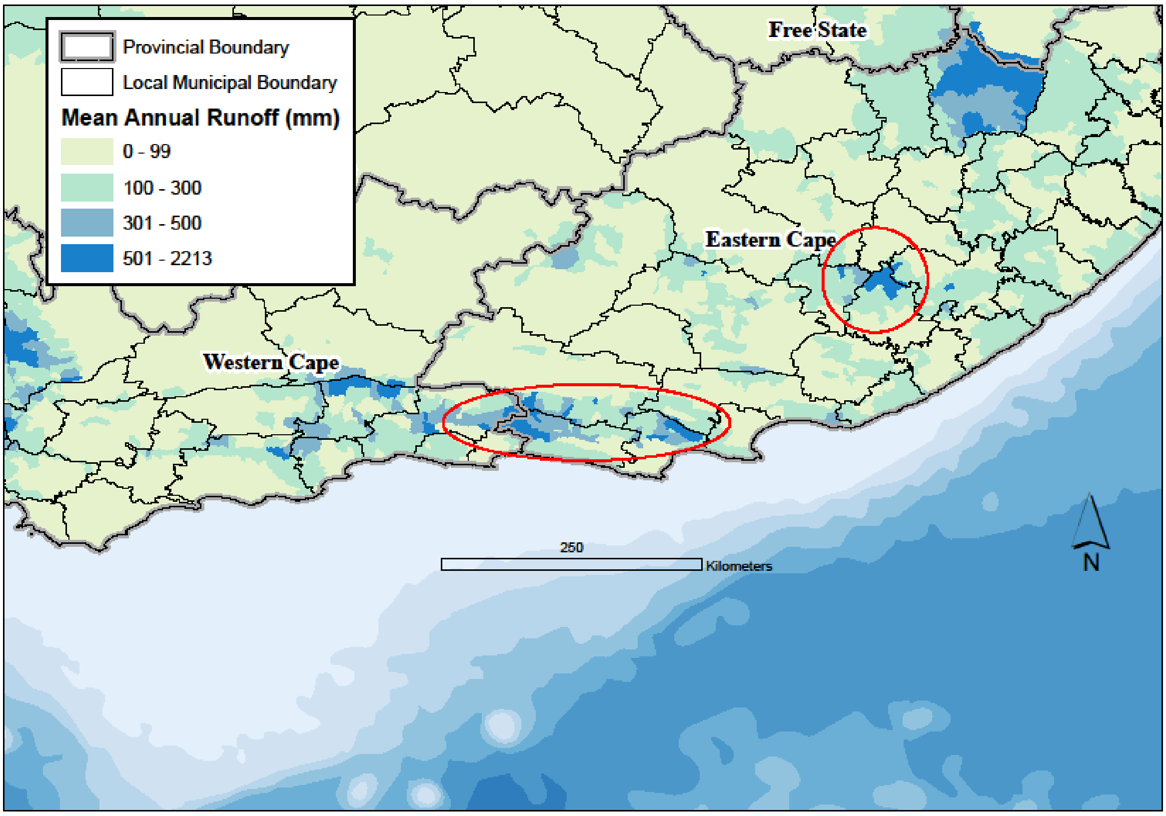


Figure 12 Map indicating the areas with highest water yield. In the Eastern Cape Province two strategic water source areas stand out indicated by the red polygons. Both these areas were incorporated into the sampling area of this study. (Map modified and produced by Luke Moore).

Water Management Areas

In terms of section 5 of the National Water Act 36 of 1998 (NWA 1998) the Minister of Water and Sanitation must establish and publish a National Water Resource Strategy (NWRS). The first NWRS was published in 2004. As part of the second NWRS nine new Water Management Areas (WMAs) were established (NWA 2016). These were; Limpopo, Olifants, Vaal, Orange, Mzimvubu-Tsitsikamma, Breede-Gouritz, Berg-Olifants, Inkomati-Usuthu and Pongola-Mtamvuna. This current study focussed on rivers that fall within the Mzimvubu-Tsitsikamma WMA (Figure 13).

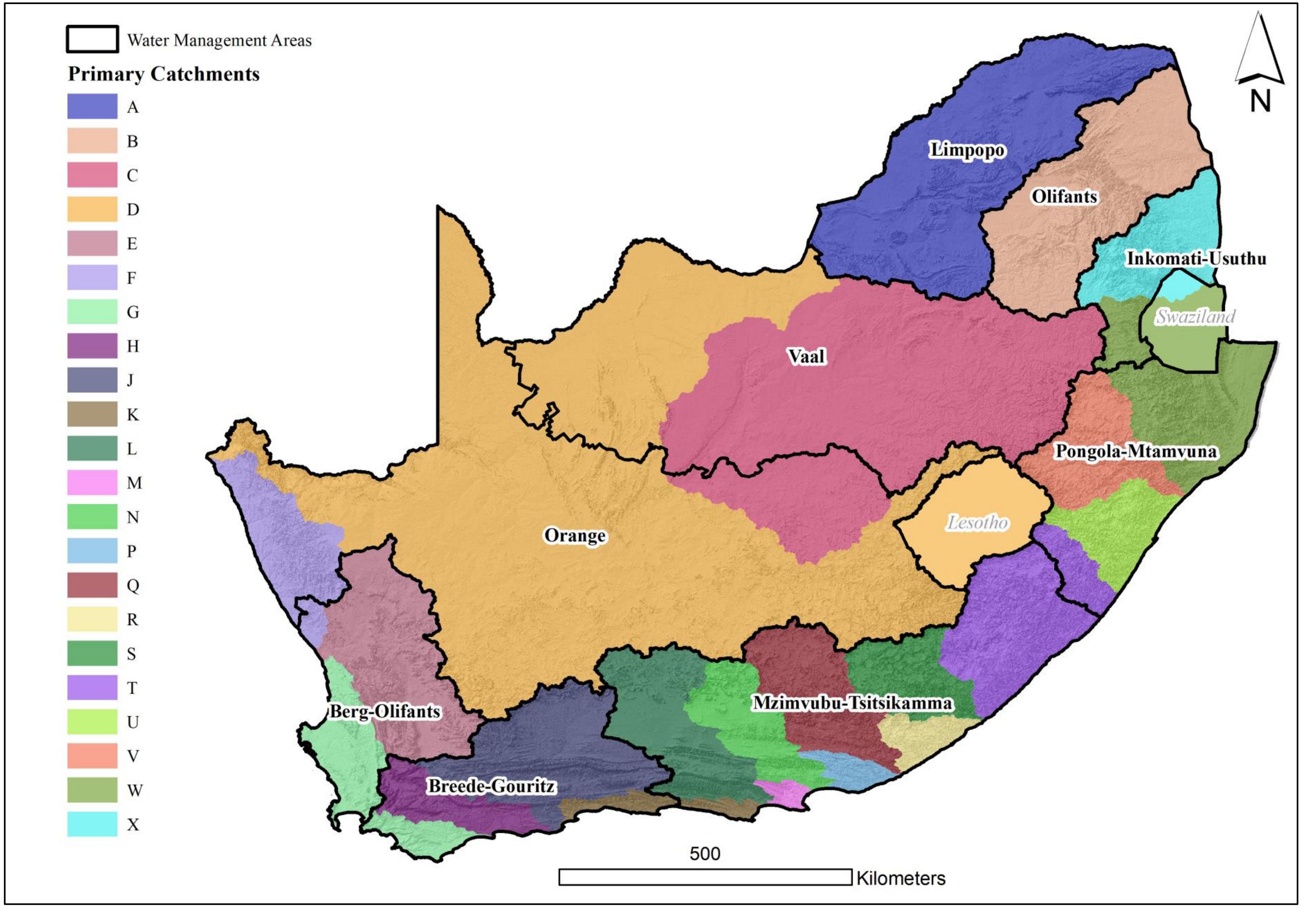


Figure 13 The main drainage regions of South Africa (shaded in different colours) and the nine Water Management Areas shown in black outline. (Image modified and produced by Luke Moore).

River ecoregion classification

River ecoregion classification allows for rivers to be grouped together based on their similarities in a top-down nested hierarchy (Kleynhans et al. 2005). While water management areas are based on decision makers and resource managers within water boards spread across the country, ecoregional classification is aimed at informing these management areas to improve protection and monitoring efforts of the Ecological Reserve. Chapter 3 of the National Water Act of South Africa (NWA 1998) states that the Ecological Reserve should be protected in order to ensure sustainable development and management of our freshwater resources. In order to achieve this, the freshwater resource should be delineated and quality objectives should be well defined.

Bioregions were developed as part of the River EcoStatus Monitoring Programme (REMP) which is the rivers component of the National Aquatic Ecosystems Biomonitoring Programme (NAEBP). Bioregions are based on a bottom-up approach. In contrast to this approach, ecoregion classification uses a top-down hierarchy system based on different levels of data. This means that at different levels within this hierarchy the rivers grouped together will be expected to have similar attributes based on the information used during assessment. The level of detail increases as the hierarchical level increases. The purpose of ecoregional classification is to conceptualise and simplify the information for ecological water requirements (DWAF 2016).

Kleynhans et al. (2005) identified 31 Level I ecoregions for South Africa, Lesotho and Swaziland by using Omernik's (1987) approach (Figure 14). The Level 1 assessment made use of physiography, climate, rainfall, geology and potential natural vegetation. Omernik’s method was also used to classify Level 1 and 2 ecoregions in the USA. Potapova and Charles (2002) studied diatom community composition across spatial and ecological gradients of these two ecoregions and found that there were three distribution patterns present. The first was between upland, mostly fast-flowing oligotrophic systems and slower flowing eutrophic systems. The second distribution gradient was found to be linked to pH and the third and most obvious gradient was found to be latitude and altitude driven temperature changes. No such study of ecological gradients and diatom community has been conducted for South African Rivers. The current study was aimed at assessing community composition across certain ecological gradients on rivers of the Eastern and Western Cape provinces.

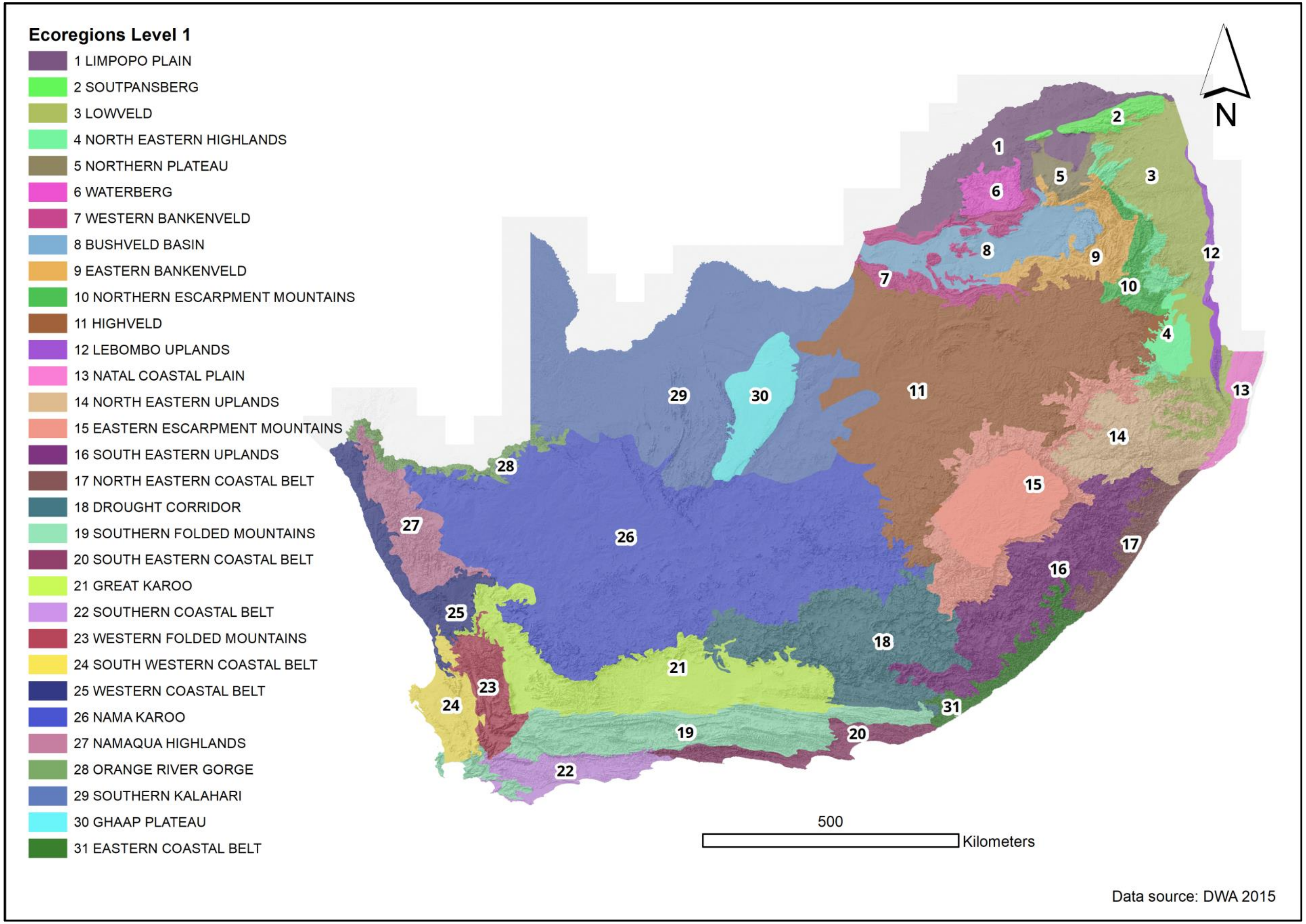


Figure 14 Ecoregions of South Africa, Lesotho and Swaziland as produced by Kleynhans et al. (2005) and published by the Department of Water Affairs and Forestry, South Africa. (Map modified and produced by Luke Moore)

Rivers sampled during this study fell into three Level 1 ecoregions; Ecoregion 18 – Drought Corridor, Ecoregion 19 – Southern Folded Mountains and Ecoregion 20 – South Eastern Coastal Belt.

Drought Corridor (DC)

The Drought Corridor Ecoregion has a generally low mean annual precipitation with moderately high, to high levels of variation in annual precipitation experienced. Drainage density is low in most areas with stream frequency being described as low to medium high in some areas. The area covers 62 675 km^2 and the mean annual temperature is moderate to high. The rainfall season is usually late to very late summer. The dominant vegetation types are the South Eastern Mountain Grassland and Eastern Mixed Nama Karoo. One of the prominent rivers in this ecoregion is the Great Fish River (Kleynhans et al. 2005).

Southern Folded Mountains (SFM)

The Southern Folded Mountains Ecoregion has a generally low mean annual precipitation but towards the South this turns into moderate to high. The variation in annual precipitation is moderate to high with moderate levels of drainage density and medium to high stream frequency. The ecoregion is 49 395 km^2 in size and mean annual temperatures are considered moderate but also low in some places. The rainfall season is usually very late summer to early winter. The dominant vegetation types are; Mountain Fynbos, Grassy Fynbos and Little Succulent Karoo. Other distinctive types of vegetation found in this ecoregion are; Fynbos, Karoo, Renosterveld and Thicket types. The most prominent river in this ecoregion is the Gouritz River. Other rivers of regional significance are; the Gamtoos, Kromme, and Breede rivers (Kleynhans et al. 2005).

South Eastern Coastal Belt (SECB)

The South Eastern Coastal Belt Ecoregion has a moderate to high mean annual precipitation and the drainage density is low to medium. Variation in annual precipitation is low to moderate and the stream frequency can be described as low to medium with some limited places being medium to high. The rainfall season is all year round with slightly higher precipitation occurring late summer to winter. The mean annual temperature is moderate to moderately hot and the ecoregion covers an area of 13 085 km^2. The dominant vegetation types are Afromontane Forest and Mesic Succulent Thicket. Other prominent vegetation types occurring in this ecoregion are; Fynbos, Renosterveld, Grassland, and Thicket vegetation. The Swartkops and Keurbooms rivers flow through this region (Kleynhans et al. 2005).

The Okavango River

Due to the lack of undisturbed lowland rivers in South Africa, the closest available alternative was found to be the Okavango River in Botswana. For this reason the Okavango River was under the Southern African context.

The Okavango River is the main artery of water supply for Botswana in Southern Africa. Water originates in the highlands of Angola 1200 meters above sea level and runs down towards Namibia before entering Botswana in the North-West at Mohembo. Two main tributaries join close to the border between Angola and Namibia, the Cuito and Cubango rivers, to form the Okavango River (also known as the Kavango River in Namibia). In the most North-Western parts of the Okavango River basin average annual rainfall is 1300 millimetres (Figure 15). This declines at a steady rate as the river moves south and east to where the annual average rainfall is only 450 millimetres (Mendelsohn et al. 2010).

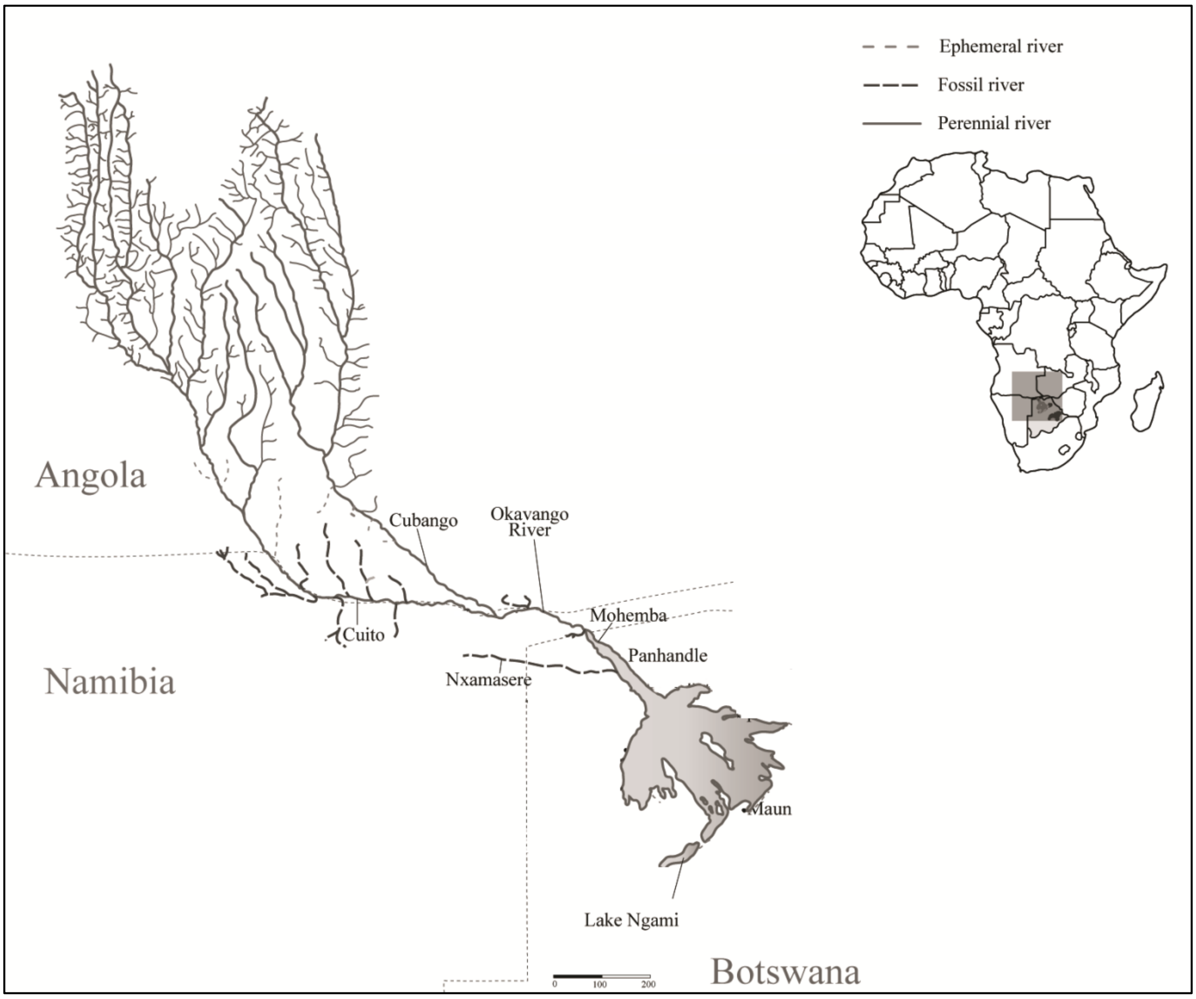


Figure 15 The Okavango River System from its origin in the highlands of Angola, through Namibia and into the Kalahari Desert in Botswana (Adapted from Van As et al. 2012).

The Okavango Panhandle and Delta lie near the lowest part of the Kalahari Basin. The Kalahari Basin is a sea of sand, the remains of an ancient ocean from 550-700 million years ago (Haddon 2005). The Basin stretches from the Northern Cape in South Africa, through most central parts of Botswana, through Angola and into the Democratic Republic of Congo (Van As et al. 2012). The lower reaches of the Okavango River are therefore located in a semi-arid region. Rainfall upstream of Botswana is crucial for the sustainability of ecological functioning in the panhandle and delta river systems. Since the Okavango River is one of the biggest sources of freshwater in Botswana, it is a lifeline to many local people, animals and plants. The river provides food, building material, freshwater and many other services and goods to local villages, towns and cities (Figure 16).

The Okavango Delta (wetland section downstream of the panhandle) has been listed as one of the most important wetlands in the world by Junk et al. (2006). And more recently is has been declared a World Heritage site (UNESCO 2014). The Okavango Delta and its upstream components (panhandle) are of critical importance for securing people's livelihoods under increasing threats from climate change and increased resource demands in Namibia and Angola. Although there are some agricultural and economic developments proposed and already present upstream of Botswana, most interests lie outside the basin which consequently has left the basin, at present, in pristine condition (Mendelsohn et al. 2010).

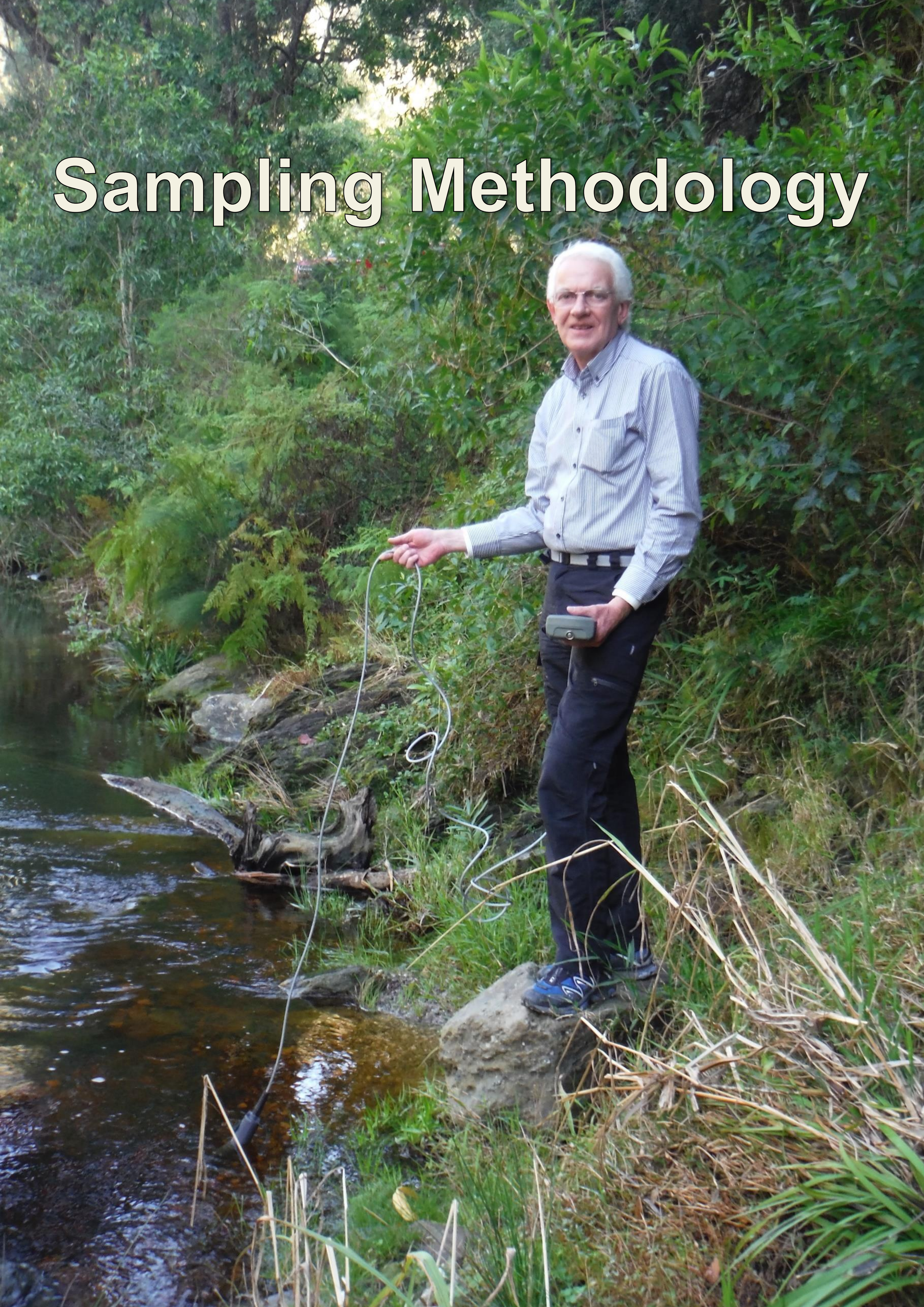
The larger mammals in the lower sections of the Okavango River play a key role in optimal ecological functioning of the river and associated systems. Hippopotami create channels in the papyrus beds and together with other larger species contribute to the surface paths which maintain the distribution of water in the delta. The annual flooding is the most important resource of water in Botswana, and this has been found to vary greatly from year to year depending on the rainfall in the Angolan highlands, which accounts for 49% of variation experienced downstream. Only 21% of inundation is driven by localised rainfall, which is to be expected since the Okavango River flows inland into a low rainfall area (Mendelsohn et al. 2010).

Figure 16 (A) Local fisherman catching fish in the Okavango River. (B) School girl drinking water from a public tap in a local village, also a place (C) where children play and come together. (D) A woman and children washing and doing laundry in the river. (E) The women collect reeds from the river to use as thatch roofing for huts.

The water quality is exceptionally good with the highest conductivity reading still being similar to that of bottled water. This however also means that there are not a lot of nutrients present in the inflowing water. Most of the potassium, nitrogen and phosphorus that do enter the panhandle are very quickly absorbed by papyrus and phragmites reeds. The seasonal and occasionally inundated areas of the delta downstream rely on nutrients carried down the river and deposited in the sediment which accumulates over long periods. The biggest local contributors to nutrients are dust, faeces and micro-organisms. Dust accounts for 40% of all nitrogen and 60% of phosphorus added to the delta. Bacteria and prokaryotic organisms fix nitrogen into forms useful to other living organisms. Larger mammals grazing on the terrestrial landscapes deposit faeces into the river especially during drinking (Mendelsohn et al. 2010).

The seasonal inundation plays a very important role in the release of nutrients. As important as flooding is for the maintenance of ecological functions, so are the drying periods. Peat in the permanent swamp areas contains a very high volume of nutrients from decomposed roots, charcoal and other detritus. However, only through burning and drying can these nutrients become readily available to other organisms in the Okavango Delta. Another interesting source of nutrients lays hidden in the seasonal and occasional floodplains in the form of dormant eggs. Algal blooms occur often when the first flood waters move into the dry seasonal and occasional channels. Soon after this the zooplankton hatch and form swarms which are followed by fish and larger predators. The flooding or wetting of seasonally inundated peat swamp land allows for nutrients previously not available to become available to the aquatic ecosystem. This cycle is repeated over again when the flood recedes and plant, animal, faecal and dust nutrients are deposited in the sediments lying dormant till the next flooding cycle returns (Mendelsohn et al. 2010).

Sampling Methodology



CHAPTER 3 Sampling methodology

Data collection

The National Diatom Collection, presently housed at North-West University, lacks specimens from the Western and Eastern Cape Provinces in South Africa and therefore site selection was based on the addition of samples to the national collection from these provinces. When possible, sites were located on or near Department of Water and Sanitation (DWS) gauging weirs (Figure 17A & B) which allowed for information collected during this study to serve as an addition to already long standing national biomonitoring records.

Information collected from sample sites at the time of sampling were; a) physical characteristics such as channel width and depth, substrate calibre and percentage shading and b) physico-chemical variables including dissolved oxygen, conductivity, pH, salinity and temperature.

Figure 17 A typical Department of Water and Sanitation (DWS) gauging weir, (A) situated on the Groot River upstream of the town Patensie and (B) on the Volkers River upstream of the Darlington Dam.

For diatom collection, the methods described by Taylor et al. (2007a) were used. This manual, the product of a WRC funded project (TT 281/07), describes the collection, preservation, preparation and analysis needed to accurately work with diatoms from Southern Africa. Taylor et al. (2007a) prescribed the use of cobbles and boulders as the preferred substratum for diatom sampling in a riverine habitat. This corresponds to prescribed practices throughout the world.

In South Africa, it has been found that sampling during mid-winter to early spring in summer rainfall areas are ideal, since during low-flow periods diatom communities are well-developed on the submerged substrates (Taylor et al. 2007a). However, for comparative ecological analysis a seasonal sampling effort was conducted across all sites when possible. For this purpose, a once every threemonthly sampling effort was conducted over a period of one calendar year, spring 2014 to winter 2015.

In addition to the South African sampling a six week sampling effort during 2014 was made on the lowland section of the Okavango River in Botswana. This was done in order to assess the diatom communities associated with a low nutrient lowland riverine habitat. In Southern Africa, most lowland rivers are impacted by anthropogenic activities such as urbanisation, agricultural land-use and commercial development which make comparative ecological assessments difficult between upstream and downstream environments.

Site selection

This study focussed on rivers in South Africa and Botswana in Africa (Figure 18). The Eastern Cape Province and eastern parts of the Western Cape Province in South Africa and the Okavango Basin in Botswana were selected as areas of investigation for this study. The Eastern and Western Cape sites were selected due the lack of information in the National Diatom Collection, and the Okavango River basin in Botswana was selected due to the little information available from pristine lowland river diatom communities in Southern Africa. The Okavango River in Botswana is one of the few places where lowland rivers are still in a pristine condition.

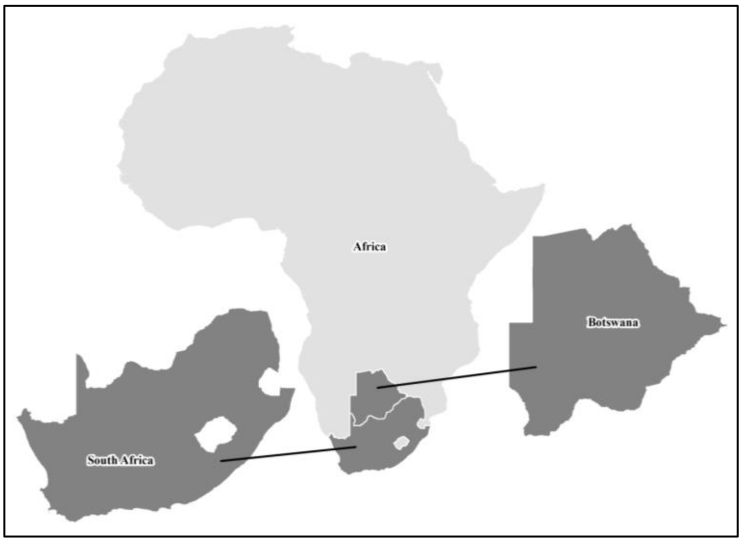


Figure 18 Sites selected for this study were situated in South Africa and Botswana, Africa (Map produced by Luke Moore).

South Africa

Site selection was, as far as possible, based on the location of DWA gauging weirs on rivers in the Eastern and Western Cape (Figure 19). The existence of long-term hydrological and ecological data meant that the newly collected data can be added to these longstanding biomonitoring data sets.

The following criteria were used as a guide for site selection:

1. Rivers upstream of the gauging weir had to be in a relatively undisturbed condition.
2. The riparian vegetation as well as the river bank as a whole should be intact and in a relatively undisturbed condition, as sediment inputs and embeddedness influences water quality (as shown by SASS5).
3. The sampled substrate size/calibre was cobbles which had to be present at the site, and more importantly, in the active channel (Figure 20A).
4. The site had to be located within traveling distance from the main roads for repetitive seasonal sampling efforts.
5. A variety of riverine habitats were selected (for instance mountain stream and upper foothill sites) across different biomes for ecological and hydrological diversity (Figure 20B-D).

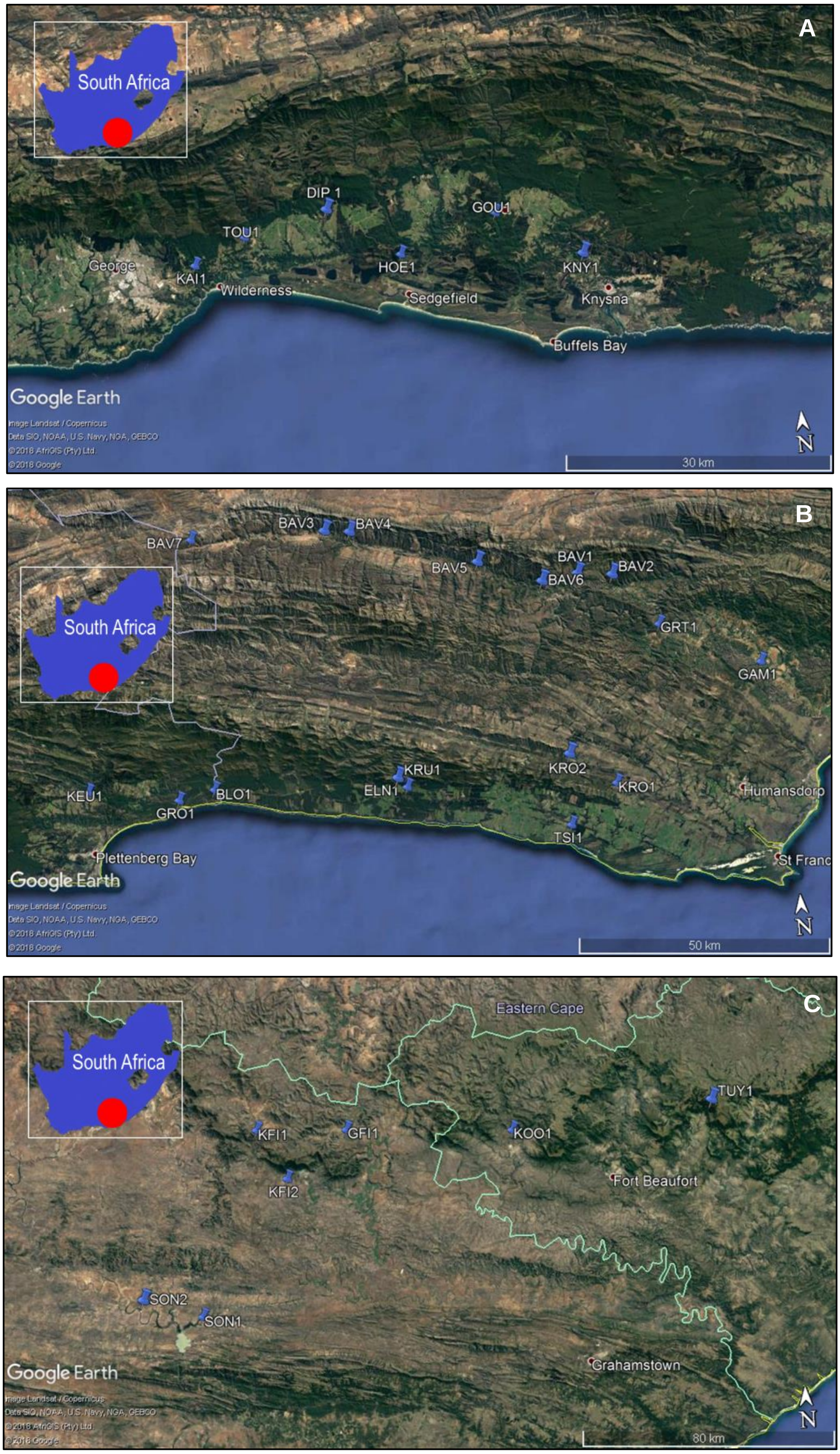


Figure 19 (A – C) Location of sampled sites in the Eastern and Western Cape Provinces, South Africa. (Modified from Google Earth 8 March 2017). (See Table 4 for location acronyms)

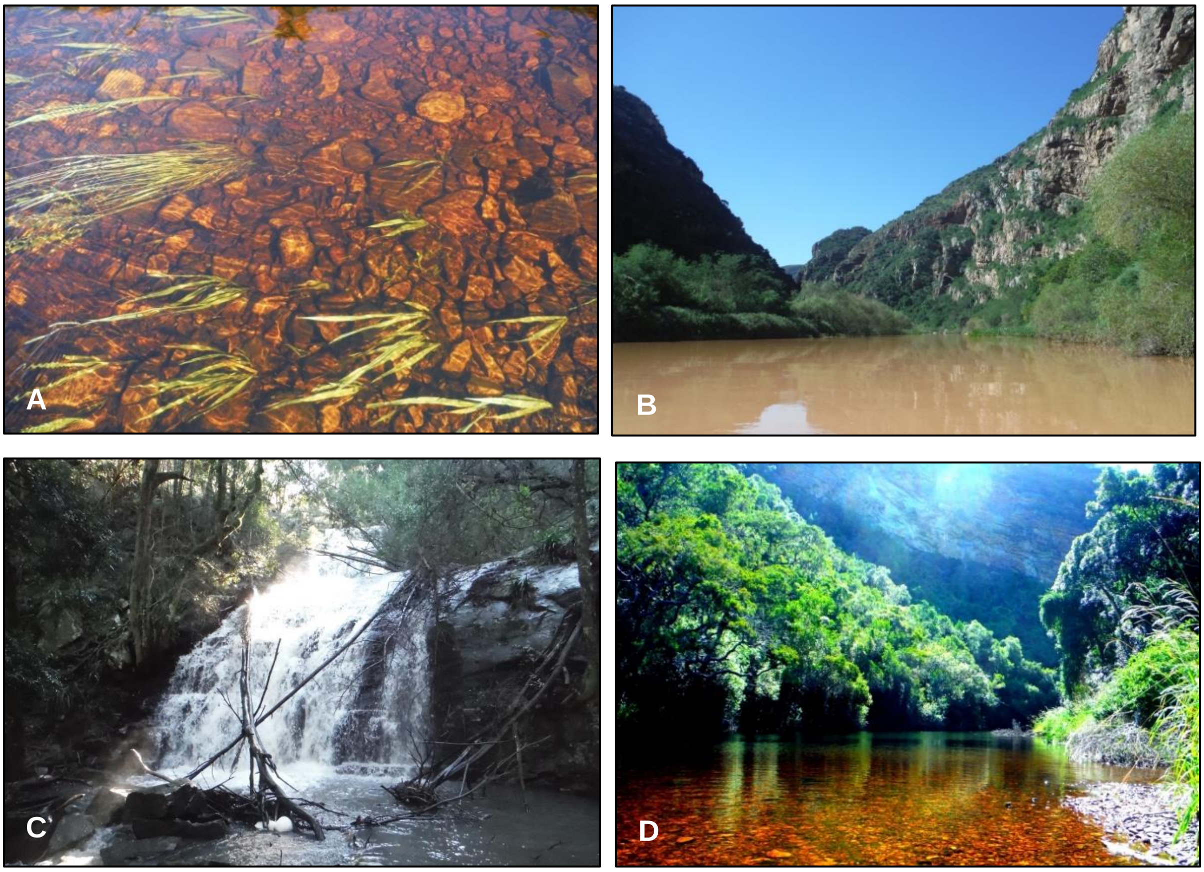


Figure 20 Sites had to have (A) cobble sized substrate present in the active channel. A variety of hydrological habitats were selected such as (B) the slow flowing Groot River near Patensie, (C) a mountain stream (Tyume River site) and (D) the Groot River near Nature's Valley which is a pristine coastal river.

However, there were a few exceptions to this, for instance the Baviaanskloof Wilderness Area (Figure 21A & B), which had no gauging weir data available.

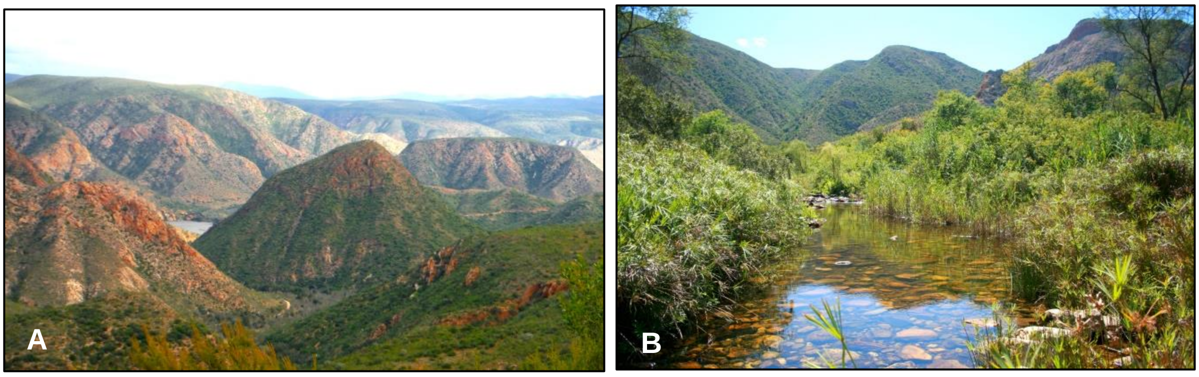


Figure 21 Some rivers were harder to access in narrow valleys (A) with no DWS gauging weir information available, (B) such as the pristine upper tributaries of the Kouga River in the Baviaanskloof.

Study sites

In September 2014 a desktop scouting exercise was undertaken to identify possible suitable sites for this study. There were many sites to choose from initially, but during preliminary inspection of all possible sites during the sampling field trip in October 2014, only the best suited sites were selected. This first sampling field trip served as an opportunity for on-site assessment of the ecological condition and logistics associated with using a specific site in this study. Some sites were found to be very remotely located and repeat sampling would be impractical. Others were found to be in a less than ideal ecological condition due to disturbances such as bank modification and over extraction of water. Examples of this was seen on the Sundays River (Figure 22A) which was in a less than desirable condition lower down near the town of Kirkwood and the Krom River near the Kouga Dam (Figure 22B). The DWS gauging weir was situated downstream of the dam where there was hardly any flow present which made the hydrological impact too large to regard this site as relatively undisturbed or suitable for repeat sampling.

During the sampling period, spring 2014 to winter 2015, certain rivers changed more drastically than others (Figure 23A-F). In some cases, seasonal rivers had flow after good rains. In these cases, the rivers were sampled for completeness of records from the Eastern Cape Province (Figure 24A & B). In other cases, sites were either dry or the bank so modified after flooding events that accessing the sites had become impossible. Table 4 provides a summary of the sampling sites of the study in South Africa, including the specific rivers, site coordinates and the name of the DWS gauging weir associated with the site (if present).

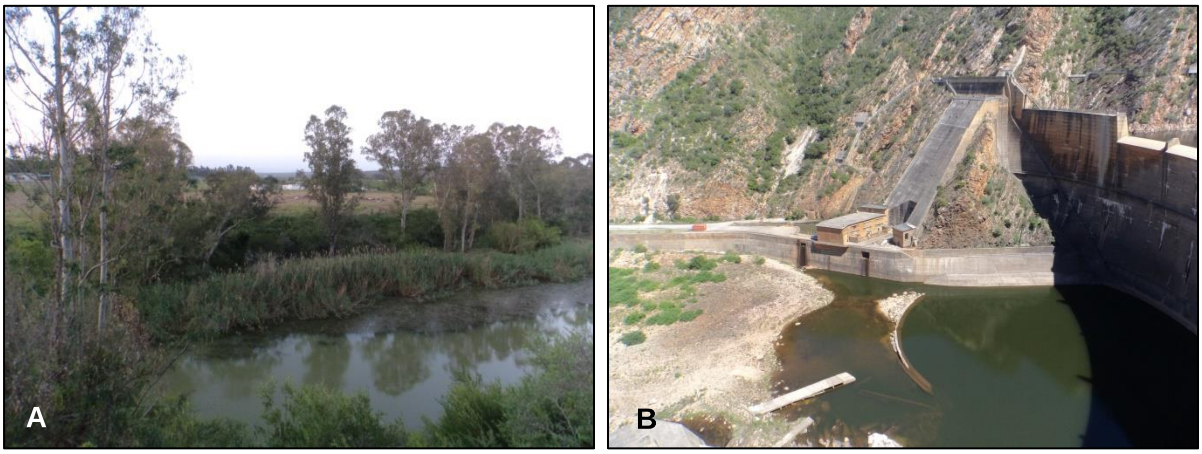


Figure 22 Sites visited during the exploration and first sampling trip in October 2014. (A) The Sundays River near the town of Kirkwood and (B) the Kouga Dam wall and immediate downstream where there was no sustainable flow present.

Figure 23 The Volkers River during (A) January 2015 and then again during (B) April 2015 sampling. (C) The Gamtoos River during October 2014 and again (D) during April 2015 when the river was flooding over the low-water access bridge. (E) The Hoogekraal River during October 2015 and again (F) during April 2015 when Prof Jo van As had to offer assistance accessing the active channel to collect cobbles for sampling.

A total of 29 sites were sampled across the Eastern Cape with some of the sites crossing into the Western Cape Province. Of these, 13 were sampled during every season (total of four repeat samples). Five sites were inaccessible during one of the four sample efforts, another five sites were only sampled twice due to their remote location as was the case with some sites located in the Baviaanskloof. The rest of the samples were only sampled once due to unexpected flow present (Figure 24A & B).

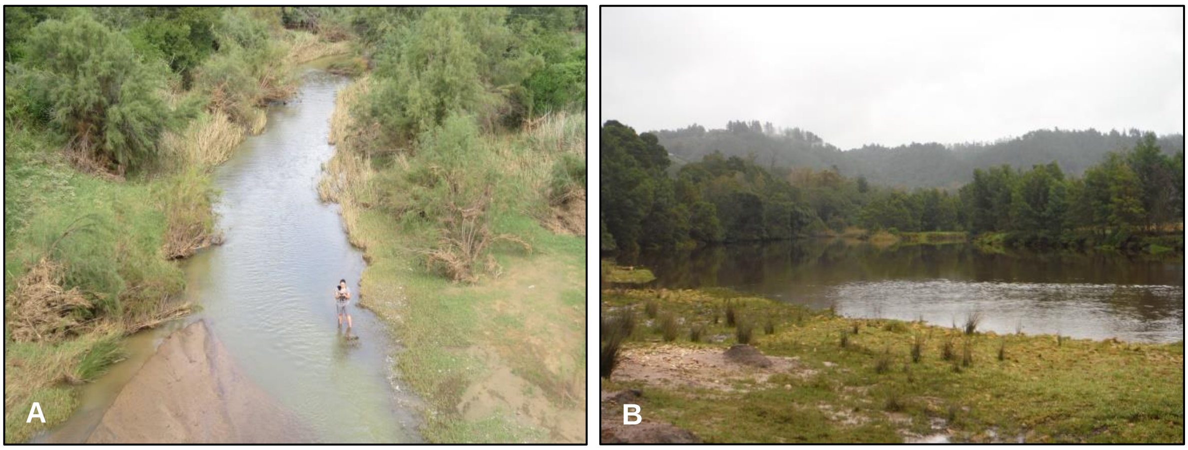


Figure 24 The Sundays River (A) and the lower Knysna River (B) were sampled during April 2015 due the presence of substantial seasonal flow.

Table 4 Summary of sites selected in South Africa for this study. Presented below is information on the drainage basin, river name, GPS coordinates, Department of Water and Sanitation (DWS) weir nearest to the site, sample type and dates for sampled sites.

Drainage Basin	River/channel	Site ID	Closest DWS Weir	Latitude	Longitude	Sample type	Sampling date			
							Spring 2014	Summer 2015	Autumn 2015	Winter 2015
Keiskamma River	Tyume River	TYU1	R1H014	-32.605031°	26.967091°	Epilithon	n/a	7/1/2015	3/4/2015	26/7/2015
Great Fish River	Koonap River	KOO1	Q9H002	-32.634009°	26.307115°	Epilithon	25/10/2014	8/1/2015	3/4/2015	27/7/2015
	Great Fish River	GFI1	Q5H004	-32.605650°	25.752022°	Epilithon	24/10/2014	9/1/2015	Inaccessible	Inaccessible
	Little Fish River	KFI1	Q8H005	-32.591194°	25.451025°	Epilithon	24/10/2014	9/1/2015	4/4/2015	27/7/2015
		KFI2	Q8H002	-32.733391°	25.543284°	Epilithon	24/10/2014	9/1/2015	Inaccessible	Inaccessible
Sundays River	Volkers River	SON1	N2H009	-33.107759°	25.228941°	Epilithon	24/10/2014	10/1/2015	2/4/2015	29/7/2015
	Sundays River	SON2	N2H007	-33.078417°	25.014940°	Epilithon	No flow	No flow	2/4/2015	No flow
Gamtoos River	Bokkraal River (Kouga River tributary)	BAV1	n/a	-33.646681°	24.442213°	Epilithon	22/10/2014	11/1/2015	1/4/2015	30/7/2015
	Wit River (Groot River tributary)	BAV2	n/a	-33.653831°	24.516115°	Epilithon	22/10/2014	11/1/2015	1/4/2015	Inaccessible
	Baviaanskloof	BAV3	n/a	-33.532286°	23.911548°	Epilithon	Inaccessible	10/1/2015	Inaccessible	Inaccessible
	Baviaanskloof	BAV4	n/a	-33.537918°	23.964973°	Epilithon	Inaccessible	11/1/2015	Inaccessible	30/7/2015
	Baviaanskloof	BAV5	n/a	-33.611543°	24.231419°	Epilithon	Inaccessible	11/1/2015	Inaccessible	30/7/2015
	Baviaanskloof	BAV6	n/a	-33.655008°	24.365275°	Epilithon	Inaccessible	11/1/2015	Inaccessible	30/7/2015
	Baviaanskloof	BAV7	n/a	-33.508316°	23.636284°	Epilithon	Inaccessible	Inaccessible	Inaccessible	29/7/2015
	Groot River	GRT1	L7H006	-33.730862°	24.617945°	Epilithon	22/10/2014	12/1/2015	31/3/2015	1/8/2015
	Gamtoos River	GAM1	L9H001	-33.812238°	24.829759°	Epilithon	22/10/2014	12/1/2015	31/3/2015	1/8/2015
Krom River	Krom River	KRO1	K9H001	-34.006116°	24.499108°	Epilithon	21/10/2014	n/a	n/a	n/a
		KRO2	n/a	-33.965342°	24.392384°	Epilithon	n/a	12/1/2015	31/3/2015	1/8/2015
Tsitsikamma River	Tsitsikamma River	TSI1	K8H005	-34.073671°	24.394398°	Epilithon	21/10/2014	12/1/2015	31/3/2015	1/8/2015
Elandsbos River	Elandsbos River	ELN1	K8H002	-33.981256°	24.050248°	Epilithon	18/10/2014	12/1/2015	29/3/2015	6/8/2015
Kruis River	Kruis River	KRU1	K8H001	-33.981331°	24.021132°	Epilithon	18/10/2014	12/1/2015	29/3/2015	Inaccessible
Bloukrans River	Bloukrans River	BLO1	K7H001	-33.955285°	23.637539°	Epilithon	18/1/2014	13/1/2015	29/3/2015	6/8/2015
Groot River	Groot River	GRO1	n/a	-33.968231°	23.559605°	Epilithon	18/10/2014	13/1/2015	29/3/2015	6/8/2015
Keurbooms River	Keurbooms River	KEU1	K5H019	-33.938538°	23.366401°	Epilithon	20/10/2014	13/1/2015	29/3/2015	Inaccessible
Knysna River	Knysna River	KNY1	K5H003	-34.007330°	23.014911°	Epilithon	n/a	n/a	27/3/2015	Inaccessible
Goukamma River	Goukamma River	GOU1	n/a	-33.947669°	22.919372°	Epilithon	19/10/2014	14/1/2015	27/3/2015	3/8/2015
Hoogekraal River	Hoogekraal River	HOE1	K4H001	-33.979889°	22.799666°	Epilithon	19/10/2014	14/1/2015	27/3/2015	3/8/2015
Touws River	Touws River	TOU1	K3H005	-33.946747°	22.612169°	Epilithon	20/10/2014	Inaccessible	27/3/2015	Inaccessible
Kaaimans River	Kaaimans River	KAI1	K3H001	-33.970266°	22.548267°	Epilithon	20/10/2014	14/1/2015	27/3/2015	3/8/2015

Site descriptions

Sites selected for this study were situated across a wide range of ecological gradients. Some river basins originating in the dry Eastern Karoo interior of South Africa which meant the water was murky and had a higher pH compared to the coastal rivers which originate in the mountain ranges running along the Garden Route of South Africa. These short coastal rivers in the Garden Route are known to have red-brown coloured water due to the tannins present in the water, causing the water to have a relatively low pH and therefore being more acidic. Below follows a brief description of the different sites sampled during this study.

Keiskamma River

The Tyume River was selected as an appropriate river to sample in this basin due to the upper reaches being relatively undisturbed. The Tyume River originates in the forested Amatola Mountains and flows down through the Tyume Valley on the Eastern side of the town of Hogsback until it joins the Keiskamma River below the Binfield Park Dam[40]. The Tyume is one of the major tributaries of the Keiskamma River which enters the Indian Ocean at Hamburg between East London and Port Alfred[41]. The only historic information on this river basin comes from Malcolm Giffen who sampled the Tyume River during 1948 – 1951 (Giffen 1984). The Tyume site was situated near the end of the Amathole hiking trail (Figure 25A) and is classified as a mountain stream site. The canopy cover was dense and the geomorphology typical of that associated with mountain streams with boulders, bedrock and fast flowing waterfalls and cascades being present at the site. The cobbles, below the waterfall, were sampled (Figure 25B).

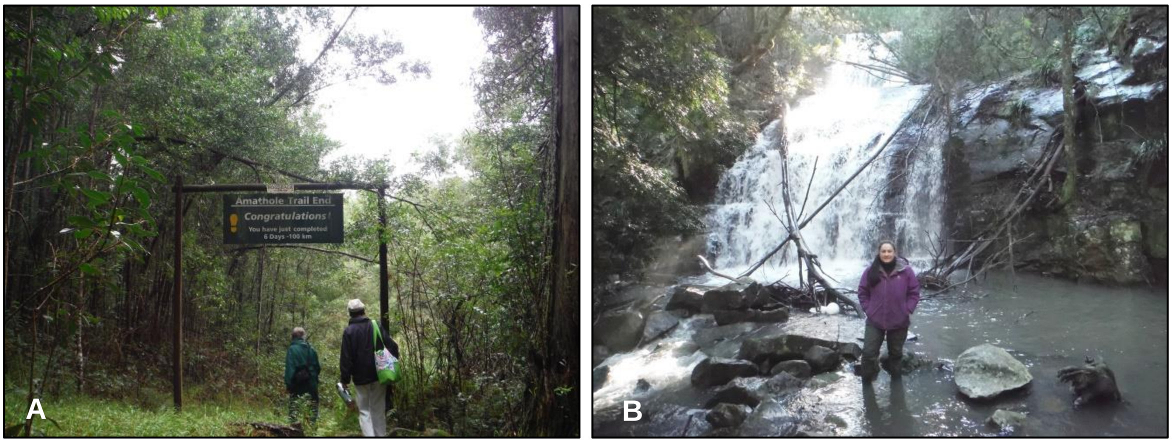


Figure 25 The Tyume River site was situated near the end of the Amathole Hiking Trail (A) which has typical geomorphological characteristics associated with mountain stream sites (B) such as boulders, bedrock and fast flowing waterfalls and cascades.

[40] https://en.wikipedia.org/wiki/Tyhume_River accessed on 1 April 2017
[41] http://www.sa-venues.com/things-to-do/easterncape/keiskamma-river-mouth/ accessed on 1 April 2017

Great Fish River

Four sites were selected in this river basin. One site on the Koonap River, two sites on the Small Fish River and one site on the Great Fish River were identified during the exploratory sampling trip of October 2014.

1. Koonap River

The Koonap River is a prominent tributary of the Great Fish River. The Koonap River flows down from the Katberge Mountains through sheep farms and past informal settlements before entering the historic Eastern Cape town of Adelaide. Sampling as far upstream of these impacts was therefore a priority for this study (Figure 26A). During winter sampling in July 2015 the water levels were very high and the bridge, which is usually completely clear of the water, had the water level threatening to overflow the road (Figure 26B).

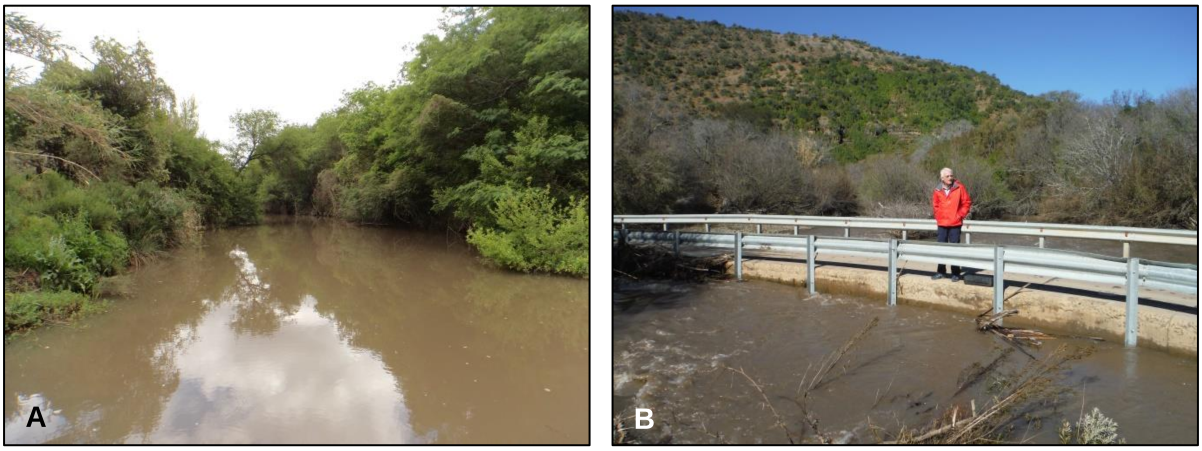


Figure 26 The Koonap River upstream of Adelaide. (A) The site on the Koonap River selected for sampling and (B) the water level on the bridge immediately downstream of the site during floods in winter 2015.

2. Small Fish River

Two sites were identified on the Small Fish River for sampling. One site was situated just upstream of the town of Somerset East and the second site was situated on a tributary of the Small Fish River higher up in the basin towards the town of Cradock. The lower site, KFI 2, was downstream of a bridge (Figure 27A) and the upper site, KFI 1, was situated on a tributary upstream but still had agricultural impacts present (Figure 27B & C). Both sites had impacts and could not be described as pristine, however with the possible impact of inter-basin transfer occurring on the other sites within the Great Fish River basin it was decided to sample the Klein Fish River for comparative purposes. During April 2015 the Small Fish River experienced low flows, especially KFI 1 (Figure 27D).

Figure 27 Two sites were selected on the Small Fish River. (A) Site KFI 2 upstream of the town of Somerset East. (B) Site KFI 1 was situated higher up in the basin although agricultural impacts were still present as seen here in the valley just downstream of the site. (C) Site KFI 1 was situated on a tributary of the Small Fish River. (D) During April 2015 it was very dry and only little flow was present on KFI 1.

3. Great Fish River

The Great Fish River stretches 644 kilometres through the Eastern Cape Province. Although the river was once one of the largest in the Eastern Cape, the abstraction of water for agricultural activities in its basin was not sustainable which led to the underground Orange-Fish Tunnel (Oviston) to be constructed in 1966 (DWAF 1975). The tunnel opened in 1975. This 83 km tunnel, which is the third longest water transfer tunnel in the world, transfers water from the Orange River basin at the Gariep Dam to the Great Fish River[42]. Water is transferred into the Teebusspruit and then into the Great Brak River from where it is fed into the Great Fish River upstream of the town Cradock. This inter-basin transfer scheme which

[42] https://plak.co.za/moreinfo/19526/orange-fish-river-tunnel accessed on 1 April 2017

mixes the water of two different watersheds could have serious environmental impacts. The Orange-Fish tunnel also feeds water to the Sundays River through a secondary transfer scheme from the Great Fish River upstream of the town Somerset East. One site on the Great Fish River was selected for sampling downstream of the town of Cradock (Figure 28A). Agricultural impacts were evident and meant that the site could not be described as pristine (Figure 28B).

Figure 28 The site selected on the Great Fish River (A) as far upstream as possible. The impact of agricultural activities as seen in the density of the reeds present at the site (B) would later (in the sampling period) make it impossible to access the active channel for sampling.

Sundays River

Beyond the inter-basin transfer scheme between the Orange River and the Great Fish River, there is also another inter-basin transfer scheme between the Great Fish River and the Sundays River just upstream of the town of Somerset East (Figure 29A). Water is transferred from the Great Fish River, just upstream of Somerset East, into the Sundays River basin by means of water channels (Figure 29B) and delivers water to the Volkers River, which the sampled site was situated on (Figure 29C), from where it runs down to the Darlington Dam (Figure 29D). The Sundays River upstream of the Darlington Dam is seasonal and only had water present during March 2015 at which time the river was sampled. For seasonal repeat sampling purposes a site on the Volkers River was selected on a DWS gauging weir (Figure 29C). Archie Archibald recorded the diatom species of the Great Fish and Sundays Rivers in the 1960's. Unfortunately he did not specify abundances or dominant species (Holmes and Taylor 2015).

Figure 29 (A) A lesser known inter-basin transfer scheme between the Great Fish River and the Sundays River. (B) Water is transferred from the Great Fish River by means of water channels. (C) The Volkers River sample site immediately upstream of the DWS gauging weir. (D) The Darlington Dam which is seen in the valley below and forms the uppermost perennial point of the Sundays River.

Gamtoos River

The Gamtoos River originates once the Groot River and Kouga Rivers merge just below the Kouga Dam[43]. This joining of the two tributaries occurs upstream of the town of Patensie and can be seen here in Figure 30A, which was visible from the access road during high water levels in July 2015. Numerous sites were selected in this river basin most of which were located within the Baviaanskloof wilderness area. Due to the remoteness there were no DWS gauging weirs present in the wilderness area (Figure 30B), and the road has completely deteriorated leaving a very treacherous gravel single-way mountain pass which is only accessible to 4x4 vehicles. For this reason complete sampling of the Baviaanskloof was only conducted twice, during January and July 2015 (Figure 30C-D).

[43] https://en.wikipedia.org/wiki/Gamtoos_River accessed on 5 April 2017

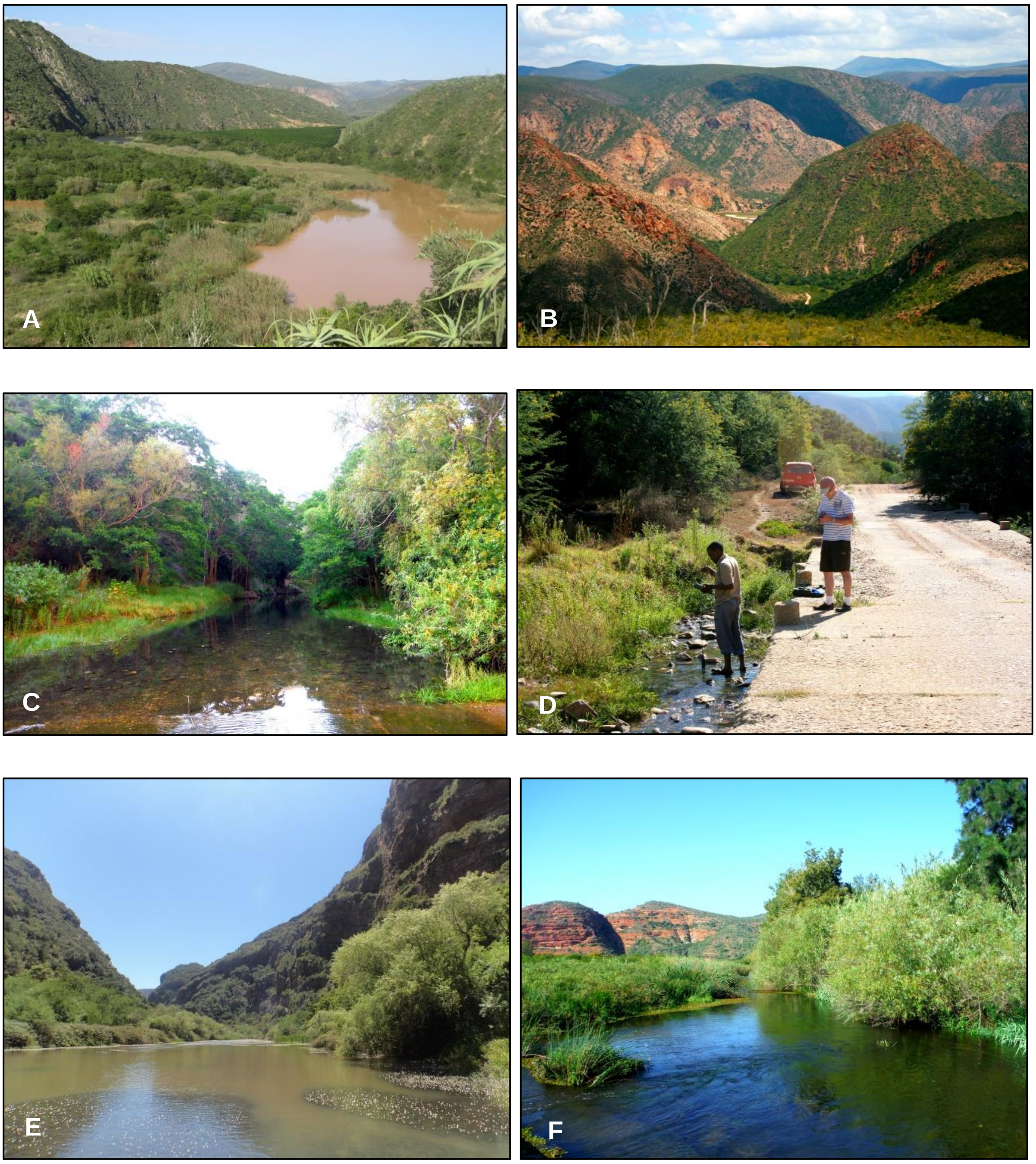


Figure 30 The Gamtoos River. (A) The confluence of the Groot (from valley to the right) and Kouga rivers (from valley to the left) which then becomes the Gamtoos River. (B) The Baviaanskloof is a narrow valley with tributaries of the Groot and Kouga rivers which are in pristine condition. (C) A typical site in the Baviaanskloof (BAV 4). (D) Sampling where the valley is slightly wider as to avoid surprise run-ins with Rhino and Buffalo during January 2015. (E) The Groot River site upstream of the confluence of the Kouga and Groot rivers. (F) The Gamtoos River site situated in the citrus farming community downstream of the confluence and near the town of Patensie.

The outer most sites were sampled as often as possible during sampling trips and these included two sites in the wilderness area, the Groot River upstream of the joining with the Kouga River (Figure 30E) and the Gamtoos River site which was situated downstream of the town Patensie (Figure 30F). The Kouga River could not be sampled due to the impact of the Kouga Dam just upstream of the DWA gauging weir but sample site BAV 1 was situated on a tributary of the Kouga River upstream of the Kouga dam in the wilderness area.

Krom River

The Krom River was originally expected to be sampled near the Churchill Dam north-east of Port Elizabeth, however the condition of the site close to the DWS gauging weir was not optimal for sampling due to severe flow alterations and embeddedness (Figure 31A). Instead a site was selected more upstream near the town of Kareedouw (Figure 31B).

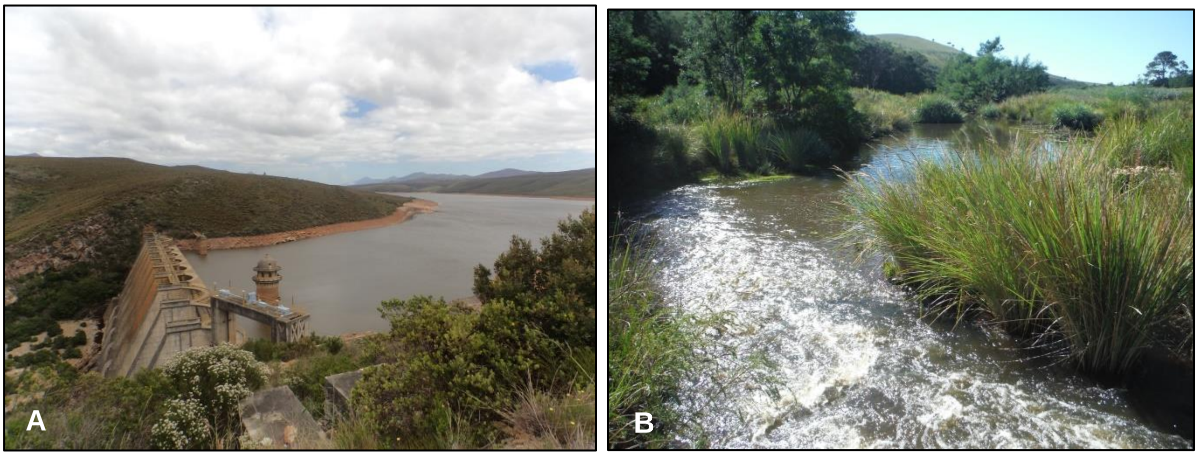


Figure 31 The Krom River site (A) originally planned to be sampled at the Churchill Dam but, the site was moved upstream (B) to be situated near the town of Kareedouw instead.

Tsitsikamma River

One site on the Tsitsikamma River was selected in the agricultural communities where the river had good flow. The entire Tsitsikamma River basin has been severely dammed by farms mostly for the irrigation of livestock feed. The Tsitsikamma basin is well known for the dairy farms located in the area. Although it is expected that there might be some instream impacts visible in the diatom communities from the surrounding agricultural activities, the site was selected where the river would have perennial flow, instream cobble substrate present throughout the sampling period (Figure 32A) and where there was less immediate impact to be seen such as bank disturbances, dams and the presence of livestock (Figure 32B).

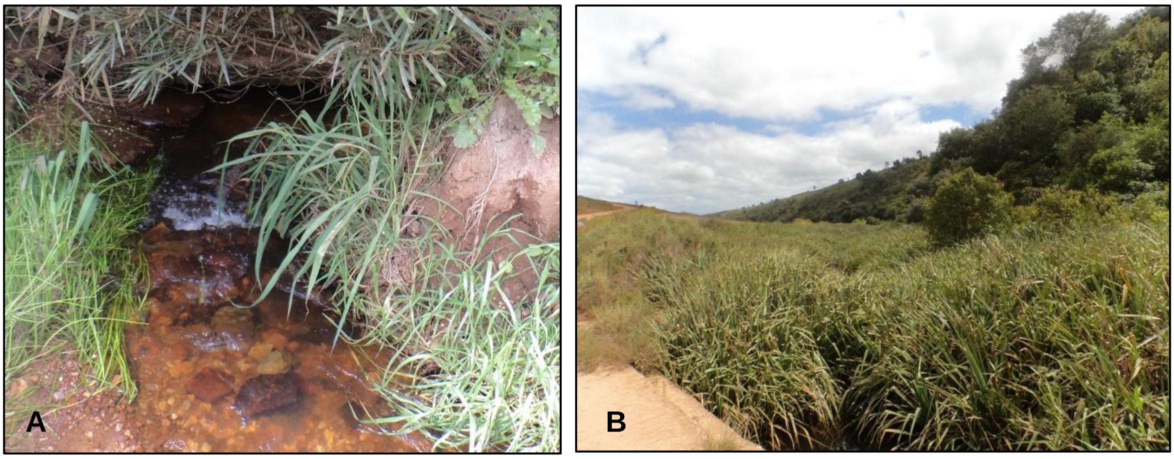


Figure 32 The site on the Tsitsikamma River (A) had perennial flow and was situated (B) in a section of the river with the least immediate agricultural impacts.

Elandsbos River

The Elandsbos River is a short coastal river originating in the Tsitsikamma Mountains and runs for large parts through the Tsitsikamma National Park. The river mouth is situated on the Otter Hiking Trail in the Marine Protected Area situated along the coast of the Tsitsikamma National Park[44]. The site was situated immediately upstream of the DWS gauging weir and on a rapid (Figure 33A & B).

Figure 33 The Elandsbos River site. Situated immediately upstream of the DWS gauging weir (A) and on a rapid (B).

[44] http://www.otterhiking.com/html/otter_hiking_trail.html accessed on 7 April 2017

Kruis River

The Kruis River is impacted by forestry in the upper basin, invasive species on the banks and bank modifications due to holiday accommodation immediately up and downstream of the site. The site was situated just upstream of a DWS gauging weir (Figure 34A). During the sampling period the site changed dramatically. From being shaded and relatively sheltered from the bank modifications happening downstream, severe disturbance on the immediate banks of the site was seen from January 2015 onwards. Clearing of invasive species in the riparian vegetation could be seen (Figure 34B).

Figure 34 The site on the Kruis River (A) was slightly impacted but became more so during the sampling period. (B) Removal of riparian vegetation and debris from upstream forestry was carried down to the site from January 2015 onwards.

Bloukrans River

The Bloukrans River (Figure 35A-D) is an iconic river in the Garden Route and for a very long time the only way to cross the deep valley was by means of the Bloukrans Pass which is one of the mountain passes built by Andrew and Thomas Bain[45]. Today the mountain pass is degraded and no longer open to the public due to lack of maintenance[46]. The site on the Bloukrans River was selected immediately upstream of the DWS gauging weir. Apart from some invasive species present in the valley and in the riparian zone, the river is in pristine condition. The Bloukrans River acts as the boundary between the Eastern and Western Cape provinces. All sites discussed below are therefore situated in the Western Cape and all sites above were situated within the Eastern Cape Province.

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[45] https://www.mountainpassessouthafrica.co.za/find-a-pass/item/716-the-bain-legacy.html accessed on 6 April 2017

[46] https://www.mountainpassessouthafrica.co.za/find-a-pass/western-cape/item/133-bloukrans-pass-r102-garden-route.html accessed on 6 April 2017

Figure 35 The Bloukrans River site. (A) Mr. Luthando Bopheka and Mr. David Mitchell assisted with data collection. (B) The mountain pass is no longer maintained but remains one of the most historic roads in South Africa. (C) The site was situated on a rapid and immediately upstream of a DWS gauging weir. (D) The road closure, and consequent sheltering of this valley from the public, allows for other species such as baboons to re-settle in the valley.

Groot River

The Groot River had no DWS gauging weir however it was found to be in pristine condition and was easily accessible, which led to the decision to include this site in this study (Figure 36A). The Groot River originates in the Witberg Mountains and runs for large parts through the Tsitsikamma National Park[47]. The site was situated close to the coastal town of Nature's Valley and upstream of the Groot River Estuary (Figure 36B).

47 http://www.nvra.co.za/assets/PDF/SANPARKS%20estuary%20guide.PDF accessed on 7 April 2017

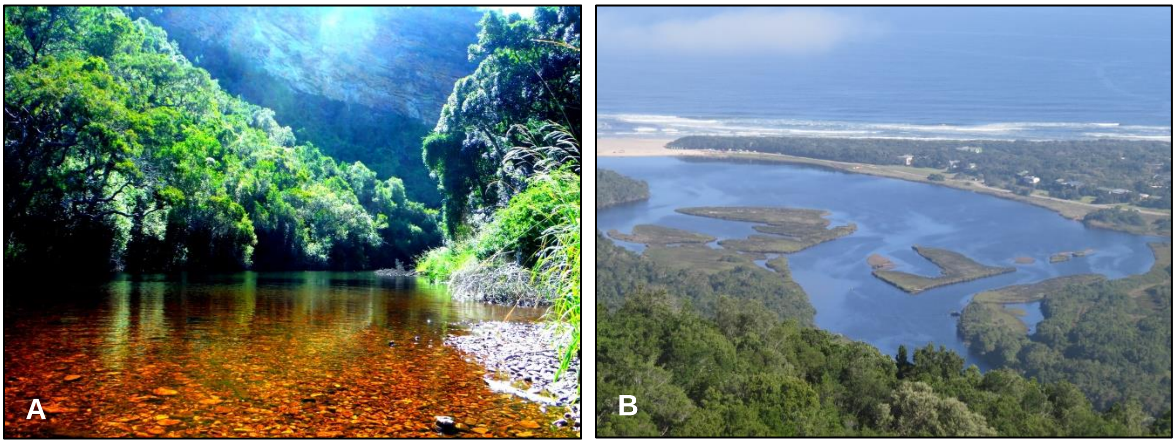

Figure 36 The Groot River is a pristine coastal river and the site (A) was selected upstream of the (B) Groot River Estuary at the town of Nature's Valley in the Tsitsikamma National Park.

Keurbooms River

The Keurbooms River is one of the larger rivers flowing through the Garden Route. The river ends in a very large estuary at the town Keurbooms River. The river runs through the Keurbooms River Nature Reserve which is a World Heritage site[48]. The site (Figure 37A) was situated upstream of a DWS gauging weir (Figure 37B) and next to the Plettenberg Bay Game Park. There were some invasive species present in the riparian zone and the bridge, which had been washed away a couple of years before this study commenced, served as a reminder of just how large the Keurbooms River is, especially at full capacity during flooding periods.

Figure 37 The Keurbooms River is one of the larger rivers in the Garden Route. (A) The site was situated upstream of a (B) DWS gauging weir near the Plettenberg Bay Game Park.

[48] http://www.capenature.co.za/reserves/keurbooms-river-nature-reserve/ accessed on 2 April 2017

Knysna River

The Knysna River is heavily impacted by historic forestry activities[49] throughout most parts of the basin. This river was therefore not selected as an appropriate sample river for this study, however during the April 2015 high flows the lower section of the river was sampled (Figure 38A). Lower sections of South African rivers are usually greatly impacted by anthropogenic disturbance but this small section upstream of the Knysna Estuary was less impacted and had sufficient flow for sampling after the floods (Figure 38B).

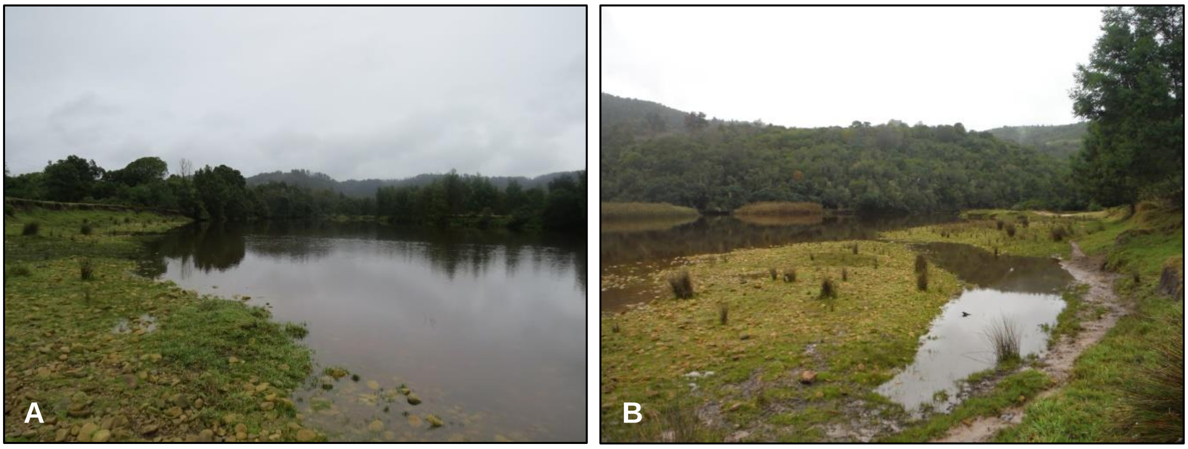

Figure 38 The Knysna River was too impacted to be sampled throughout this study, however during the flooding of April 2015 the river was sampled (A) in the lower reaches. (B) Downstream view of the site.

Goukamma River

The Goukamma River site was not situated on a DWS gauging weir but was selected due to its relatively undisturbed condition (Figure 39A).

Hoogekraal River

The Hoogekraal River site was situated upstream of a DWS gauging weir. The site was in a relatively undisturbed condition, however there were some forestry activities found to be present higher up in the basin (Figure 39B).

[49] https://en.wikipedia.org/wiki/Knysna accessed on 6 Aprill 2017

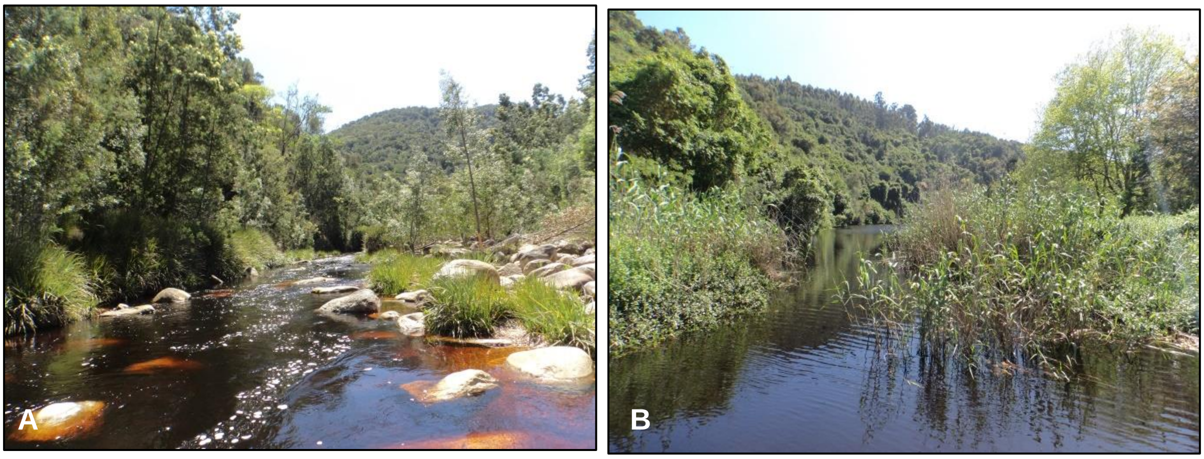

Figure 39 The Goukamma River site (A) to the north of the town of Wilderness in the Western Cape and (B) the Hoogekraal River site upstream of the DWS gauging weir.

Touws River

The Touws River site was only sampled twice during the study (Figure 40A). These were during October 2014 and April 2015. During January and July 2015 the site was assessed and found to be unsafe for sampling due to various external influences such as vagrants being present. During April 2015 the water level was very high and collecting stones in the flooded river proved to be a hard task (Figure 40B).

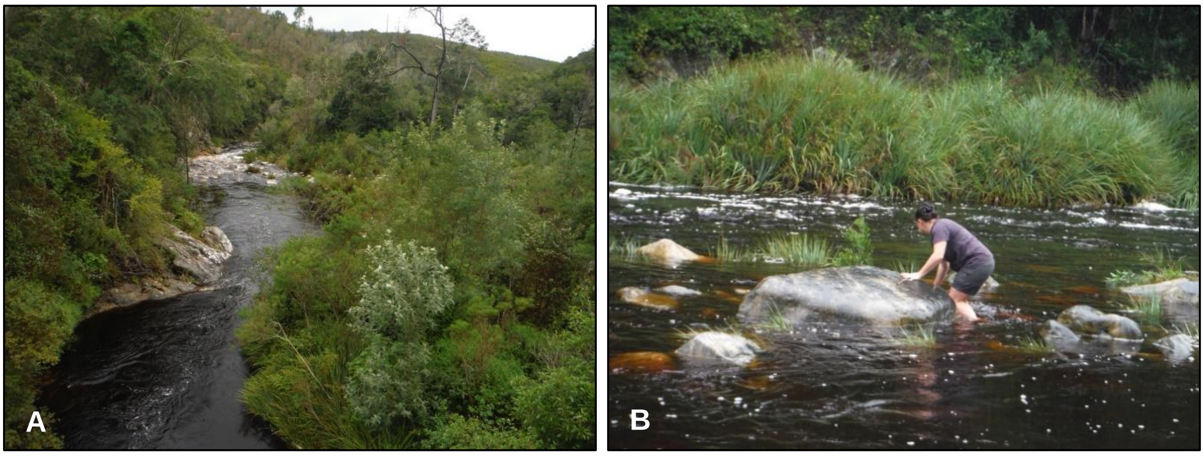

Figure 40 The Touws River was (A) sampled twice at the selected site. (B) During April 2015 the river was in flood.

Kaaimans River

The Kaaimans River flows from the Outeniqua Mountains near the city of George in the Western Cape to where it exits into the Indian Ocean between the towns of Wilderness and

Victoria Bay. The Kaaimans River is in pristine condition (Figure 41A) and the site was situated immediately upstream of the DWS gauging weir along the Seven Passes road built by Thomas Bain in the 1950s[50]. During April 2015 sampling the river was in flood (Figure 41B).

Figure 41 The Kaaimans River site (A) during October 2014 and (B) during flooding in April 2015.

Sampling and site set-up

The standard method for data collection is based on a site set-up of 10 m perpendicularly to the channel and the collection of 5 - 10 cobble sized (6 – 25 cm) stones from the centre of the active channel (Taylor et al. 2007a). This eliminates the influence of seasonal water levels at the outer edges. During low-flow periods the carrying capacity of a river declines which can lead to fine sediment being deposited especially on the outer edges of the active channel. This is simply due to the velocity of flow (usually) being higher instream, and therefore also the carrying capacity. Lower flow on the outside edges also leads to dead benthic organisms to build up rather than being flushed downstream as would be the case in the center of the active channel. This sedimentation build-up of benthos organisms (epipelon and epipsammon) was avoided by sampling riffles, runs and glides where water was continuously flowing over the selected cobbles and collecting stones from the center of the active channel. Figure 42 below provides a diagrammatic representation of the site set-up for sample collection.

[50] https://en.wikipedia.org/wiki/Kaaimans_River accessed on 5 April 2017

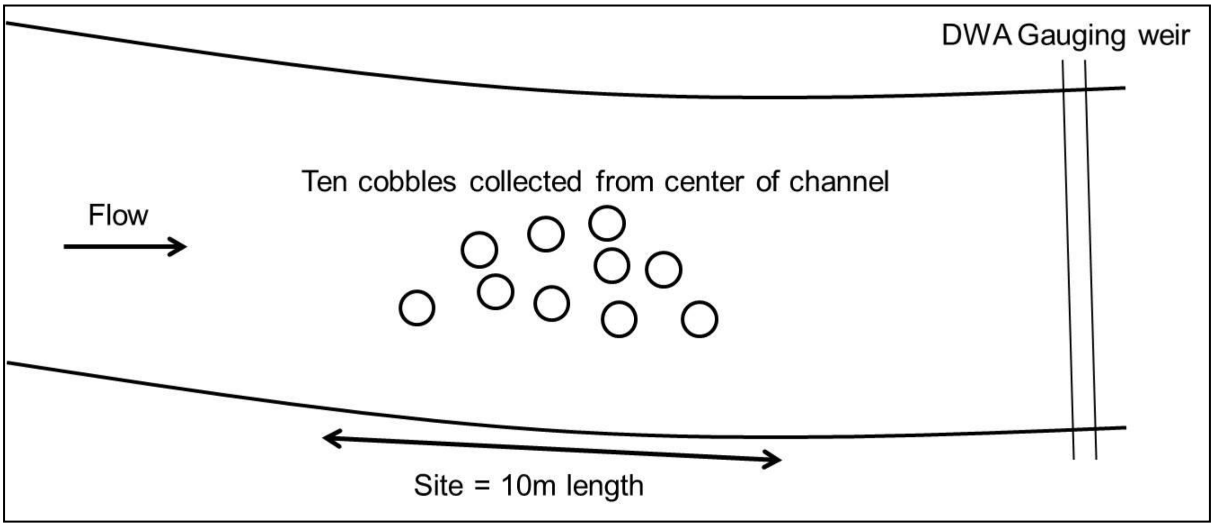

Figure 42 Site setup with five to ten (maximum) cobbles sampled per site which are taken from the center of the active channel.

The stones ranged in size but all fell within the boundaries of the cobble sediment calibre as described by the Wentworth scale (Gordon et al. 1992). The cobbles were scrubbed, on the water exposed side only (top), with a clean toothbrush into a white tray (similar to the trays used for SASS5) (Figure 43A & B). A small volume of distilled water was used to wash dislodged frustules from the cobbles, approximately 50 ml. All stones collected at a site were scrubbed into the same white tray concentrating the diatoms into one sample (Figure 43C). The liquid was then poured into a glass McCartney specimen bottle and topped up with ethanol (70%), to produce a fixed sample of >20% ethanol end product (Figure 43D). A handheld water meter (HANNA HI9828) was used to measure the temperature, pH, conductivity and dissolved oxygen in the water at time of sampling (Figure 43A & B). This was especially important for sites with no DWS gauging weir present. Location was recorded by means of a handheld Geospatial Position System (GPS).

Figure 43 An example of (A) a cobble being scrubbed (B) using gloves and a clean toothbrush in order to avoid between-site sample contamination. (C) All cobbles were scrubbed at the site at time of sampling and into the same white tray to concentrate the diatoms into one sample. (D) The sample was stored in a glass McCartney sample bottles with an end fixed ethanol >20% solution. Physico-chemical parameters were sampled with (E) a handheld water meter (HANNA HI9828). (F) The temperature, pH, conductivity, salinity and dissolved oxygen of the water were recorded at time of sampling for all sites on each sampling occasion.

Botswana

In Southern Africa, and most parts of the world, lowland undisturbed rivers are very rare to find. The reason for this is that anthropogenic activities are typically better suited to the low gradient of valleys and open floodplains. The Okavango River system in Botswana (Ramsar site) is an exception to this (Figure 44). The Okavango Delta was declared a UNESCO World Heritage Site on 22 June 2014 (UNESCO 2014). The conservation and preservation of the delta (wetland downstream of the panhandle) is recognised internationally.

Figure 44 The Okavango Delta was declared a World Heritage Site in 2014.

The panhandle forms the upper part of the delta. Here the river still has a channel shape (Figure 45), before fanning out into the hand-shaped wetland towards Maun in the south-east. At Maun the Thamalakane fault line acts as a collection barrier directing the water towards Lake Ngami (Mendelsohn et al. 2010). During sampling in July and August 2014 the panhandle section of the Okavango River and the Thamalakane River downstream were selected and sampled.

Study sites

Sites varied from faster flowing side channels at Askiesbos to the slower flowing Samochima Lagoon near the Krokovango farm, 35 km from Shakawe. In addition to the panhandle, a fossil river was added to the sample diversity. The Nxamaseri Floodplain (Figure 46) used to be a mighty tributary of the Okavango River, today this floodplain is seasonally inundated with water pushing in from the panhandle downstream when water levels are high. When the water level starts receding again in the panhandle the fossil river bed is an oasis of wildlife scattered between pools of varying colour as the microscopic life scrambles for quick successional generations before the water disappears completely. Downstream of the delta, the Thamalakane River was sampled at Maun.

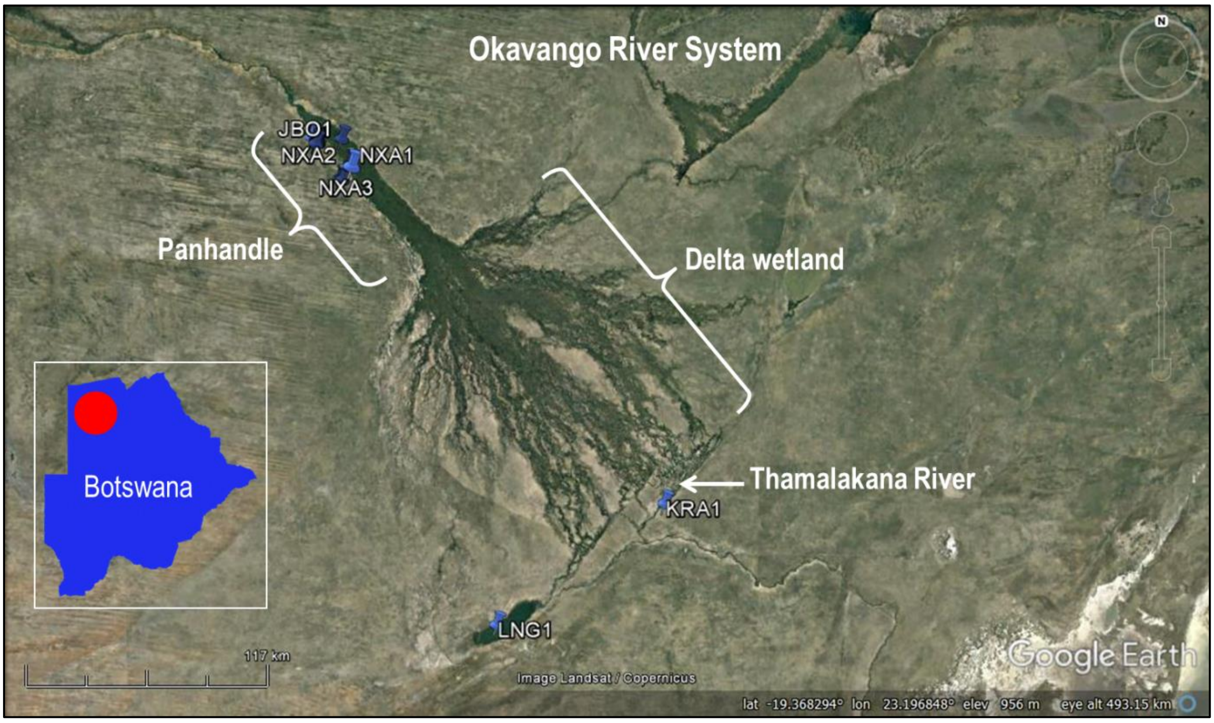

Figure 45 Sites sampled in Botswana were situated in the panhandle section of the Okavango River and on the Thamalakane River. (Modified from Google Earth, 8 March 2017).

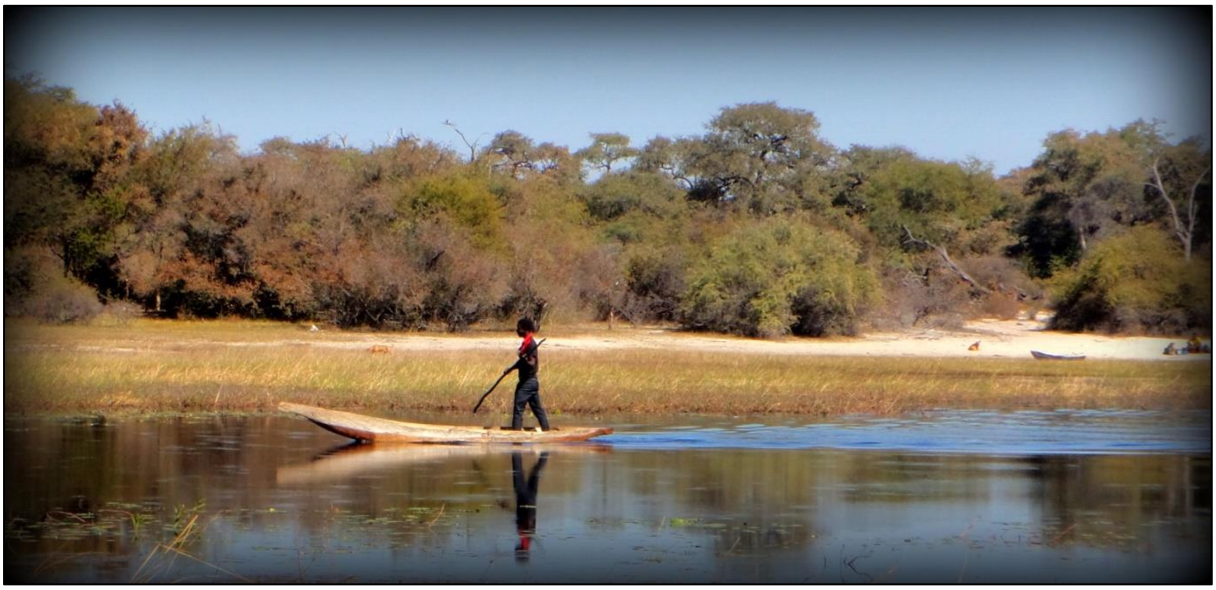

Figure 46 A local in his Makoro on the Nxamaseri floodplain in Botswana.

Table 5 provides a summary of sites sampled in Botswana during this study together with the river they were on and the coordinates and type of samples collected. In total 10 samples were collected during July and August 2014. These 10 samples were spread across six areas. Three were located in the Samochima Lagoon near the Krokovango farm and another three samples were situated in the Nxamaseri Floodplain. One sample was collected from each of the following sites; a side channel of the Samochima Lagoon, an artificial wetland system on Krokovango Farm, the Thamalakane River at Maun and the Ngaringe Channel.

Table 5 Summary of sites selected in Botswana on the Okavango drainage basin for this study including the river name, GPS coordinates, sample type and sample dates.

Basin	River/channel	Site	Latitude	Longitude	Sample type	Sampling date	Zone
Okavango	Askiesbos Channel	ASK3	18.428253°	21.89433°	Introduced substrate	19/08/2014	Lower Foothills
	Thamalakane River	KRA1	19.965841°	23.465230°	Epiphyton from Snail shells	26/08/2014	Upper Foothills
	Krokovango Wetland	KRO1	18.433394°	21.893449°	Introduced substrate	19/08/2014	Lower Foothills
	Samochima Lagoon	LAG15	18.428569°	21.895688°	Introduced substrate	20/08/2014	Lower Foothills
	Samochima Lagoon	LAG21	18.428875°	21.898372°	Phytoplankton	17/08/2014	Lower Foothills
	Samochima Lagoon	LAG3	18.427099°	21.903041°	Introduced substrate	20/08/2014	Lower Foothills
	Ngaringe Channel	NGA1	18.413784°	22.009710°	Phytoplankton	27/07/2014	Lowland River
	Nxamaseri Floodplain	NXA2	18.599290°	22.025201°	Epiphyton and phytoplankton	24/07/2014	Lower Foothills
	Nxamaseri Floodplain	NXA4	18.590054°	22.013133°	Epiphyton and phytoplankton	4/08/2014	Lower Foothills

Site Description

All the selected sites fell within the Okavango River and its associated systems. During a six week period over July and August 2014 the Aquatic Ecology Research Group at the University of the Free State (UFS) visited the Okavango River in Botswana. During this time the samples for this study were collected from various sites spread across channel, river, lagoon and floodplain type systems. Below follows a brief description of these different systems and sites selected for sampling.

Askiesbos Channel

The Askiesbos Channel (Figure 47A & B) runs parallel to the main panhandle river channel. The Askiesbos Channel is separated from the main river channel by papyrus vegetation and a narrow sand bank. The side channel is narrow and contains dense aquatic vegetation. Introduced substrate was used to sample this site which is discussed in more detail in Sampling and site set-up on page 95.

Figure 47 (A) The Askiesbos Channel was sampled (B) by means of introduced substrate.

Krokovango Crocodile Farm

The Aquatic Ecology Research Group (UFS) makes use of the Leseding research camp on the Krokovango Crocodile Farm (Figure 48A). The research camp is equipped with an on-site temporary aquatic laboratory for field analysis of aquatic samples ranging over a wide spectrum of research fields (Figure 48D). The Aquatic Research Group (Figure 48C) has also been involved with the construction of an artificial wetland (Figure 48B) on the crocodile farm which was sampled during a fieldtrip in 2014. The crocodile farm made for a perfect location to conduct fieldwork from since it is situated on the banks of the Samochima Lagoon. This location allows for the UFS's research boats to be launched from (Figure 52A & B) and provides a convenient location to do day trips from and bring back samples to the aquatic field laboratory for processing (Figure 48D).

Figure 48 Krokovango Crocodile Farm (A) in the panhandle of the Okavango Delta was sampled at an artificial wetland on the property. (B) Here the marker white pole is clearly visible showing the location of the introduced substrate. (C) The UFS's Aquatic Ecology Research Group at the research camp during the 2014 fieldtrip. (D) The temporary aquatic laboratory at the Leseding research camp at the Krokovango Crocodile Farm in Botswana.

Ngaringe Channel

The advantage of having access to a boat was that we were able to venture deeper into the panhandle section of the Okavango River to sample areas a bit further afield (Figure 49A). The Ngaringe Channel was one such place which was sampled on an exploratory daytrip from the camp. The channel was hidden between thick papyrus and the sampling effort included a floodplain on an island situated in the channel (Figure 49B).

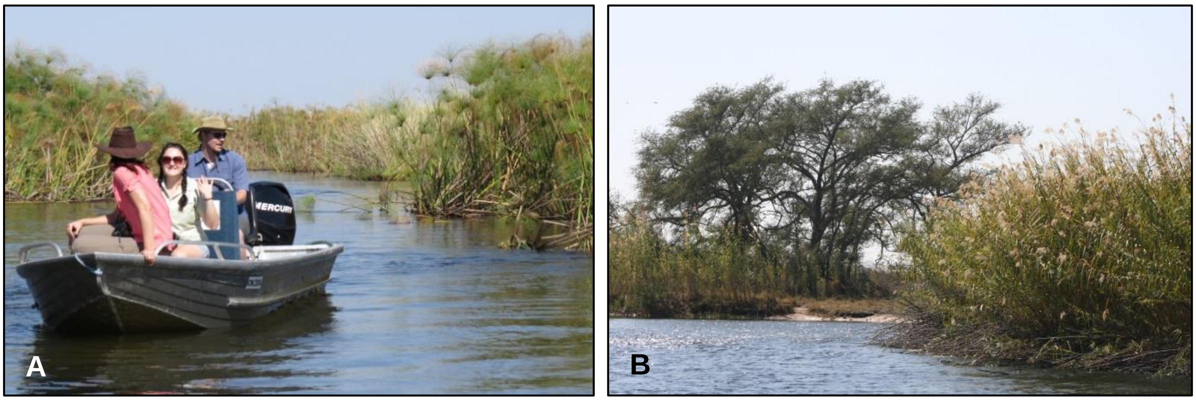

Figure 49 The fieldwork boat (A) and (B) the Ngaringe Channel.

Nxamaseri Floodplain

The Nxamaseri Floodplain was situated to the south-east of Samochima Lagoon and the research camp. Thousands of years ago this floodplain used to be a large tributary of the Okavango River. Today it is a seasonally inundated floodplain receiving water from the panhandle section of the Okavango River during annual flooding periods. Water is pushed into the fossil river bed from the Okavango River and when the flood level goes down in the Okavango River so does the water level in the Nxamaseri Fossil River leaving isolated pools behind. These pools start to dry and have been seen to turn different colours as the phytoplanktonic life scramble in their successional stages ultimately producing eggs that can withstand the seasonality associated with this system. The dormant eggs are deposited in the fossil river bed until the next seasonal flood fills the floodplain with water initiating another cycle of successional events. Various pools in this fossil river bed were sampled varying in size and depth (Figure 50B & C). A phytoplankton net and turkey baster were used to collect diatoms from the Nxamaseri Floodplain (Figure 50A).

Figure 50 The Nxamaseri Floodplain was sampled using a phytoplankton net and (A) turkey baster. (B) Some inundated areas were deep and very large, while others were drying rapidly turning into smaller (C) pools. Here Prof. Liesl van As is seen collecting water quality readings with a hand-held water meter.

Thamalakane River

The Thamalakane fault line, which forms the most south-westerly end of the African Rift Valley, acts as a collection barrier for the water in the Okavango Delta. Water is collected into the Thamalakane River which flows in a south-western direction through Maun (Figure 51A & B) and then enters Lake Ngami to the south. This river was sampled at Maun by means of a phytoplankton net and turkey baster.

Figure 51 (A) The Thamalakane River runs through Maun at the most south-eastern end of the Okavango Delta. (B) The seasonal flood push the river into the properties situated directly on the river bank.

Samochima Lagoon

The Samochima Lagoon is situated on the panhandle section of the Okavango River and it also serves as a launching place for the boats (Figure 52A). The Krokovango Crocodile Farm borders on the lagoon, which made it a very convenient sampling location since it was close to the camp and was large enough to be viewed as part of the panhandle main channel since the only thing separating it from the main channel of the panhandle is a drifting raft of papyrus (Figure 52B & C). Samochima Lagoon is teaming with wildlife such as crocodiles, fish eagles and hippopotami (Figure 52D). Introduced substrate and phytoplankton were collected in the Samochima Lagoon. Introduced substrate was attached to the papyrus (Figure 52E) and had to be marked with GPS coordinates (Figure 52F) since retrieving the samples after a few weeks would be difficult if their location had not been marked properly. In a few cases (less than 5) introduced substrate could not be located again. This could have been due to a number of reasons including the detachment of the introduced substrate, the movement of the drifting papyrus or the density of the papyrus making it very difficult to find the attached Petri dishes. Samples were distributed across the lagoon so as to maximise the diversity of microhabitats sampled. These included the deeper papyrus wall between the lagoon and the main river channel as well as the outer edges where the lagoon had slower flow and shallower water levels present.

Figure 52 The Krokovango Crocodile Farm serves as a good place to launch the research boat from (A) and made for a (B) good sampling location. Samochima Lagoon is only separated from the main channel by a drifting raft of papyrus (C). (D) Special care had to be taken when putting out or retrieving samples to avoid predators such as this crocodile. (E) Introduced substrate was attached to the papyrus from the boat. (F) The location of introduced substrate was marked with a GPS.

Sampling and site set-up

Site set-up was conducted differently for each type of sample collected. Due to the lack of larger substrate in the lower reaches of the river, a phytoplankton net with a mesh size of 25 µm was used to collect phytoplankton from the slow flowing open water areas (Figure 53A & B).

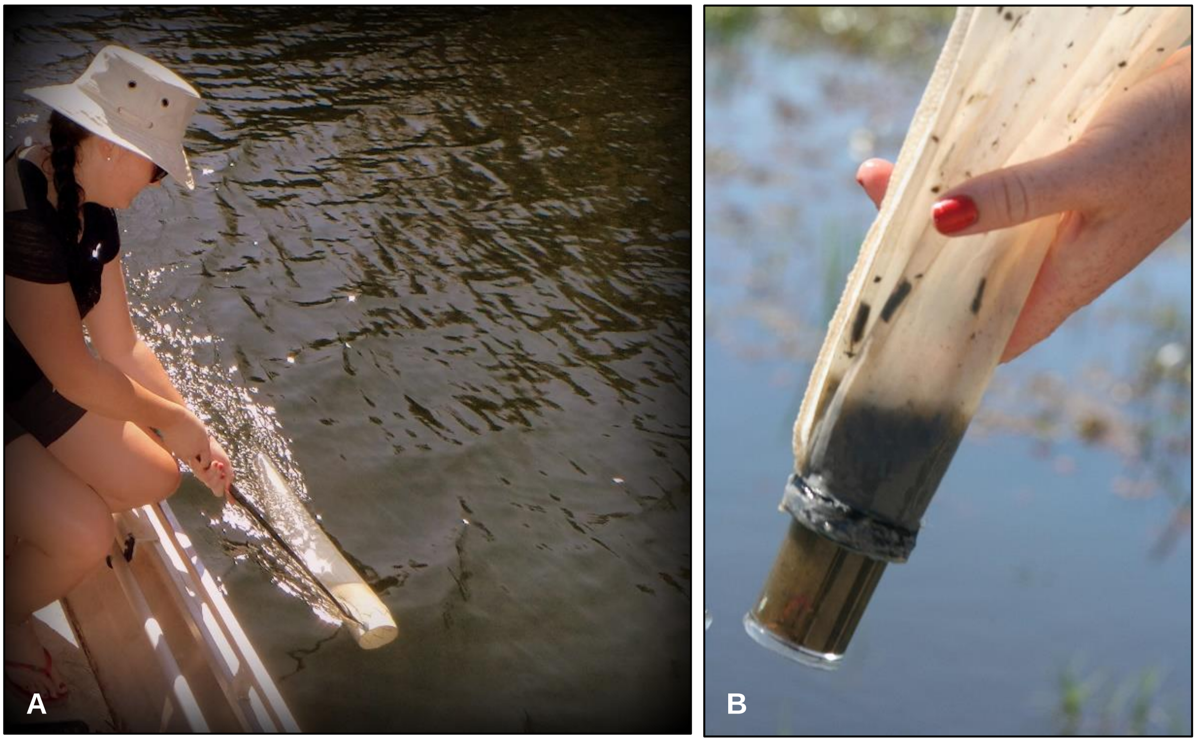

Figure 53 Due to lack of substrate and time to cultivate colonies in some cases a phytoplankton net (A) was used to collect phytoplankton samples such as on the Samochima Lagoon. (B) The net collects and concentrates the sample into a smaller collection bottle which can then be fixed with ethanol for analysis.

In order to sample the non-free floating diatom species, epiphyton was sampled directly from the macrophytic plants or by means of introduced substrate. Introduced substrate consisted of Petri dishes that were attached to the macrophytes (Figure 54A & B). These dishes were left to be colonised by the diatoms for a period of 4-6 weeks. After this time, the dishes were removed and cleaned with a toothbrush, according to the prescribed method by Taylor et al. (2007a) of scrubbing stones.

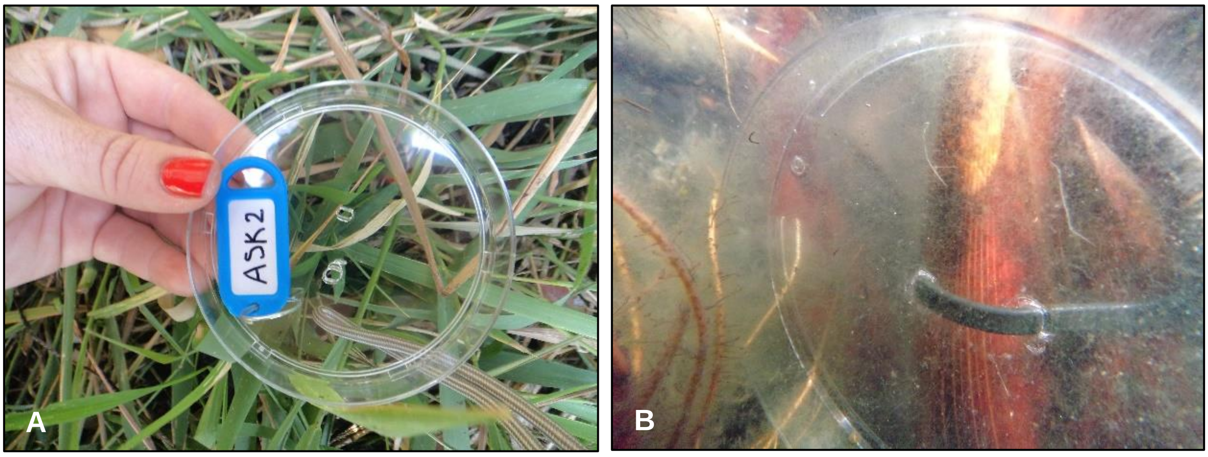

Figure 54 (A) Introduced substrate was used and (B) attached to macrophytic plants to colonise for 4-6 weeks.

All samples were fixed in glass McCartney specimen bottles and topped up with ethanol (70%) to produce a fixed sample of >20% ethanol end product. Physico-chemical parameters were sampled with a handheld water meter to measure the temperature, pH, salinity, conductivity and dissolved oxygen in the water on the day of sampling. Altitude and sample position was obtained by means of a handheld Geospatial Position System (GPS).

Sample processing

Sample cleaning

Archiving the information and data processing was a very important step in this project since one of the main aims was to expand the National Diatom Collection, hosted and curated at North-West University in South Africa. The recommended technique for processing the diatom samples is the Hot HCL and $KMnO_4$ method (Figure 55A & B) (henceforth referred to simply as the Hot HCl method). This method cleans or removes the organic material in a sample producing a solution suitable for making permanently mounted microscope slides. Samples were preserved and stored according to the prescribed methods of Taylor et al. (2007a).

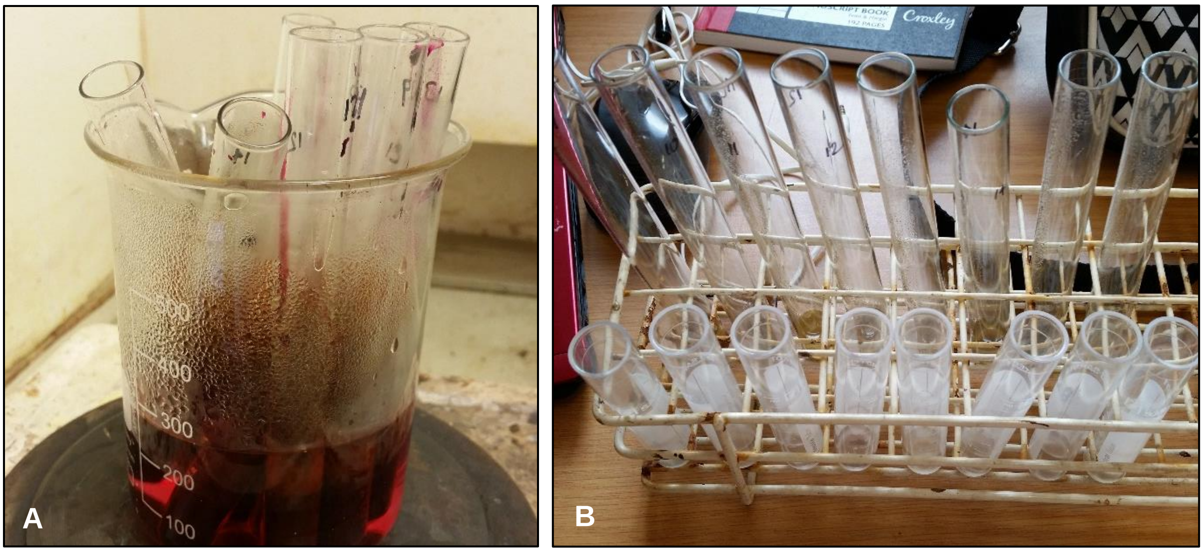

Figure 55 The Hot HCl method for cleaning diatom samples using a (A) hot plate, fume cabinet and (B) heat resistant test tubes.

Hot HCl method (modified from Taylor et al. 2007a)

1. Shake the sample well and pour a small sub-sample of approx. 1 ml into a glass test tube (depending on the concentration of the material). Mark the test tube clearly with the sample number in at least two different places using a permanent marker.
2. Add the same volume of the sub-sample (approx. 1 ml) saturated potassium permanganate ($KMnO_4$) solution to the test tube.
3. Place test tube in a heat resistant beaker which has 200ml boiling water in.
4. Place heat resistant beaker containing test tube/s on a hot plate at low heat.
5. Let the solution bubble (not boil) at low heat for 30 minutes.
6. Remove the heat resistant beaker from the hot plate and, in a fume cabinet, add 1 ml (same volume as sub-sample) of concentrated HCl (32%), very slowly into the test tube.
7. Place the heat resistant beaker back on the hot plate with test tube/s.
8. Leave on low heat till sample changes colour from brown to light yellow. Usually the solution will have a yellowish colour when clear.
9. When oxidation is complete, or the solution has changed colour from dark brown to yellowish, add 2 drops of hydrogen peroxide to test if the oxidation process is complete. If there is still organic material present bubbling will occur, in which case the solution should be left longer on the stove and retested in a couple of minutes.
10. The test tube/s are allowed to cool.

11. Before pouring the diatom and acid samples from the test tube, the tubes are vigorously swirled, the aim of the rotary movement is to re-suspend the diatoms, whilst causing small stones and heavier sand particles to fall to the bottom of the tube.
12. The samples are rinsed by centrifuging with distilled water at 2500 rpm for 10 minutes.
13. After centrifugation the supernatant is decanted and washing is repeated a further 2 times or until the sample is circumneutral. The supernatant should be poured off in a single movement, and care should be taken not to lose any diatom material. It is better to repeat the washing an extra time rather than pouring off too much of the supernatant and loosing diatom material in the process.
14. After the last wash, the cleaned material is poured into small glass storage vials bearing the necessary sample information (Figure 56A). It is important to store diatom samples in glass as opposed to plastic vials, as glass releases silica, which counters the dissolution of diatom valves.
15. Alternatively, the excess acid and soluble chlorides can be washed out by a series of timed decantations. The test tube is filled with distilled water, mixed thoroughly and the solution is allowed to settle overnight after which the clear supernatant is decanted.
16. After each decantation, the remainder is swirled to get it into suspension and the beaker is again filled with distilled water.
17. This is repeated until the suspension is clear and it is circumneutral.

Permanent slides

For the preparation of permanent slides (Figure 56B) Pleurax is prescribed as a preferred mounting agent (Taylor et al. 2007a). Pleurax helps to increase the contrast of external structures on the frustule by altering the refraction of light when viewed with a compound microscope. This assists with the accurate identification of individual diatoms. The permanent slides were made with standard microscope slides and a round 18 mm cover slip. Slides were labelled with all the appropriate information which included the site name, date of collection, mounting agent, GPS coordinates, collector’s name, type of sample and river name. The extra cleaned sample liquid was preserved at a > 20% ethanol/volume concentration, for future referencing (Figure 56A & B). Two permanent slides were made of the diatoms collected at each sample site, one of which was added to the National Diatom Collection which is housed in the Division Botany, School of Environmental Science and Development, Potchefstroom Campus, North-West University. The other was stored at the Aquatic Ecology Research Laboratory in the Department Zoology and Entomology, Bloemfontein Campus, University of the Free State (Figure 56C & D).

Figure 56 (A) 4 ml glass bottles were used to store cleaned samples in. (B) A Vortex mixer ensures the sample is well mixed before placing a small amount on a cover slip for slide-making. (C) An example of a permanent slide made using Pleurax as mounting agent and (D) labelled with all necessary information.

Identification of diatoms

In order to identify the diatoms in a live or fixed sample (permanent slide) a course was attended in Scotland during 2014. The course was held at the Field Studies Council's Kindrogan Center (Figure 57C). The Freshwater Algae Identification Course was presented by Dr. Eileen Cox and Prof. Elliot Shubert (Figure 57A & F) and provided an introduction to the identification of freshwater algae (Figure 57D & E). Training included informal lectures (Figure 57A), field collection from a variety of sites (Figure 57B) and microscope work (Figure 57E). Through generous funding from the National Research Foundation (NRF) of South Africa it was possible to attend this course which made a significant contribution to the success of this study. This course provided the necessary training to collect and identify freshwater diatoms accurately.

Figure 57 During July 2014 a Freshwater Algae Identification Course was attended in Scotland. (A) Dr. Eileen Cox was one of the course presenters and offered one-on-one training (B) with diatom sampling and the identification of live diatoms. (C) The course was held at Kindrogan and offered an opportunity to learn how to identify (D) diatoms and gain supervised experience with the (E) microscopy skills needed. (F) The course was presented by Prof. Elliot Shubert and Dr. Eileen Cox and attended by 5 students from around the globe.

On return to South Africa some of the seasonal South African sampling sites were analysed in the field. A field laboratory was set-up during sampling trips of April and June 2015 (Figure 58A & B).

Figure 58 During the seasonal sampling trips of April and June 2015 a field laboratory was set up in order to observe live samples. (A) Proff. Jo and Liesl van As working on the field microscopes in the Baviaanskloof after a day of sample collection during April 2015. (B) The field laboratory at Nature's Valley during June 2015.

During 2016 a week in Dr. Jonathan Taylor's laboratory at the North-West University aided the verification of diatom identifications made to date and any problems that may have arisen during the sample processing and permanent slide making. This time spent working in Dr. Taylor's laboratory was invaluable in ensuring the samples were processed properly.

Frustule counting

When frustule counting was done, the slides at that stage, were provided with only a number and not the label which contains site specific information. This was done so that when the species were identified the location and possible ecology of the site was unknown. This enabled for an objective compilation of community composition to be done. For identification of all species "An Illustrated Guide to Some Common Diatom Species from South-Africa" produced by Taylor et al. (2007b) was used. For any additional information needed towards correctly identifying a species the following resources were also consulted; Cox (1996), Kelly (2000), Bate et al. *(*2004), Janse van Vuuren et al. (2006) and online resources such as the online Algae database called Algaebase (Guiry and Guiry 2017).

Diatoms were counted by using the microscope field as the defined area and moving to adjacent fields while taking care not to count the same valves twice. A community composition for each site was obtained by counting at least 400 individuals on a slide

(Prygiel et al. 2002, Schoeman 1979 and Taylor et al. 2007a). Counts were made using a Nikon microscope with phase contrast optics (1000x magnification). Broken valves were also included into the count when more than 50% of the valve was intack and positive identification was possible. When a valve was in girdle view and it was possible to positively identify the species it was included into the count. Data were entered into Microsoft Excel sheets and then processed using PRIMER multivariate statistics software.

Statistical analysis

Multivariate statistics in PRIMER (V6, Clarke and Gorley 2006) was used for community scale comparisons as well as biological and environmental correlation assessments. Multivariate statistics are more suited to community based analyses, as this makes inter-community comparisons possible by using the same standard species list. The species list was based on a total collection synthesis. See raw data sheets in Appendix 1. By combining sample data for each site it was possible to compare differences between sites across ecological gradients which allowed for possible distribution and community patterns to be exposed.

Data were imported into PRIMER as abundance data with a zero indicating the absence of a record for that species on a site. The data were then put through an overall transform of square root to ensure that less abundant species were not neglected during analyses. A Bray-Curtis similarity was applied to produce a resemblance matrix. This matrix was analysed using an MDS to produce a configuration plot. In some cases, a Cluster analysis was also performed to further assess the relationships between samples.

The strength of relationships and patterns exposed by MDS and Cluster were tested by running an ANalysis Of SIMilarity (ANOSIM) analysis. The ANOSIM test is similar to an ANOVA, which tests a hypothesis; however unlike the ANOVA an ANOSIM evaluates a dissimilarity matrix instead of raw data. In this case the resemblance matrix with the predicted groupings as produced by MDS and Cluster were used (Clarke 1993). ANOSIM produces two values; a P value that represents the level of significance and an R value that represents the strength of the groupings. The R value usually varies between 0 and 1. A value closer to 1 indicates high separation while a value closer to 0 indicates no separation between groups assigned to samples. Significance level (P) is expressed as a percentage (%). According to Clarke and Gorley (2006) the R value is more important than the P value since one could have a significant P value but the strength of the relationship (R) or effect of this grouping on community distribution patterns can then not be regarded as important if the R value is low.

Once it was established that the groupings were significant and strong, a SIMPER analysis was used to produce species level information pertaining to these groupings. SIMPER provides the percentage similarity and dissimilarity between factors/groups within the data (Clarke and Gorley 2006). It also provides the variables/species responsible for these groupings along with the percentage contribution of the species to the similarity. A cumulative percentage is provided to a combination of species which explains for instance 90% of the similarity found within a group.

In an attempt to link these groupings to environmental data a Bio-ENV analysis was used. Bio-ENV calculates rank correlations (Spearman's) between the matrix of biological and accompanying environmental data to find the best possible suite/combination of environmental variables to explain the biological structure (Clarke and Ainsworth 1993). In the test histogram, bars represent the null hypothesis (no relationship between species and any combination of environmental variables) with the dotted line representing the real rank correlation coefficient. If this dotted line is larger than the bars then the null hypothesis can be rejected at $p < 1\%$. In order to assess the change and relationship between environmental data a PCA ordination was used. A PCA is similar to an MDS ordination but is more suited to environmental data. All graphs and ordinations were saved as metafiles and imported into Microsoft Word 2010.

The Harvard system was used for referencing and in cases where online resources had no author the webaddress was referenced directly in footnotes. Where possible all resource material and sources were added to the reference list.

Photos used in thesis were either taken by the author or are used courtesy of the Aquatic Ecology Research Group, University of the Free State.


Diatom community
Composition

CHAPTER 4 Diatom community composition across ecological gradients, South Africa

Community composition

Permanent slides were analysed with a minimum of 400 individuals identified per slide. A complete species list as recorded by sample is provided in Appendix 1 – Raw data: Species lists by Ecoregion, River, Site and Sample. In most cases the individuals were successfully identified to species level. However, in some cases the angle, quality of the valve or the obstruction of other frustules made it difficult to identify an individual with 100% certainty. In these cases, they were identified up to genus level and provided with a unique species number (sp). In total 36 873 individuals were identified across 84 sites and 24 rivers. In total 215 species were identified from these samples and 99 individuals were only identified up to genus level. Table 6 is a summary of data collected during this study and provides a short summary on the ecological requirements of the individuals identified.

Table 6 A summary of all species identified from samples collected during this study and the sites where they were collected. This species list was compiled from 84 samples collected from 24 rivers over four seasons during 2014 and 2015. Ecological information is adapted from Taylor et al. (2007b) see Table 4 for location abbrevations.

Species	Ecological preference and the sites they were identified from during this study
Achnanthes abundans (synonym for *Psammothidium abundans*)	No ecological data available at present also see *Psammothidium abundans*. KEU1_4
Achnanthes crassa	Found in alkaline streams and slow flowing waters. TSI1_2, GRT1_1, GRT1_2, KOO1_1
Achnanthes linearoides	Found in circumneutral to slightly acidic, oligotrophic waters constantly supplied with oxygen. BAV5_2
Achnanthes oblongella	This species is found in small, circumneutral, oligotrophic, electrolyte-poor streams. DIP1_1, ELN1_1, ELN1_3, GOU1_3, GRO1_1, GRO1_2, GRO1_3, GRO1_4, HOE1_1, HOE1_2, HOE1_3, HOE1_4, KAI1_1, KAI1_2, KAI1_3, KAI1_4, KEU1_1, KEU1_2, KEU1_3, KNY1_3, TOU1_1, TSI1_1, TSI1_2, TSI1_3, TSI1_4, BAV1_2, BAV1_3, BAV1_4, BAV2_1, BAV2_2, BAV2_3, BAV3_2, BAV4_2, BAV4_4, BAV5_2, GAM1_3, GRT1_1, KRO2_3, KRO2_4, GFI1_1, GFI1_2, KFI1_2, KFI2_2, KOO1_1, SON1_3, SON2_3, TUY1_2, TUY1_3, TUY1_4
Achnanthes standerii	This species is **endemic** to South Africa and is found in well-oxygenated, oligotrophic slightly acidic fresh water. BAV2_1
Achnanthes subaffinis	Found in oligotrophic streams and slow flowing water. KEU1_3, BAV1_4, BAV4_2, BAV4_4, BAV6_2, KRO1_1, KRO2_2, KRO2_3, KRO2_4
Achnanthes swazi	This **endemic** South African species is found in clean well-oxygenated oligotrophic fresh water. TSI1_1, BAV5_4, KRO1_1, KRO2_4, TUY1_3
Achnanthidium affine	Found in clean well-oxygenated oligotrophic, alkaline, calcareous, fresh water with moderately elevated electrolyte content. KRO2_2, KOO1_3
Achnanthidium biasolettianum	Found in calcareous oligo- to mesotrophic water with moderate to elevated electrolyte content. BLO1_4, BAV2_1
Achnanthidium eutrophilum	Found in well-oxygenated eutrophic fresh waters. Tolerant only to slight or moderate pollution. ELN1_3, GRO1_4, KAI1_3, KEU1_2, TSI1_1, TSI1_3, BAV2_3, BAV5_4, GAM1_2, GAM1_3, GRT1_2, KRO2_4, KFI2_1

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Achnanthidium exiguum	This cosmopolitan species has very wide ecological amplitude. It is also able to grow under very low light and can tolerate temperatures of up to 40°C. The optimum growth conditions for this taxon are alkaline water with moderate to elevated electrolyte content. GAM1_4, GRT1_1, KOO1_4
Achnanthidium macrocephalum	Found in calcareous, oligo- to mesotrophic fresh waters. BLO1_4, BAV5_4
Achnanthidium minutissimum	Found in well-oxygenated, clean, fresh waters. Usually attached to a substratum by a short mucilage stalk. BLO1_1, GRO1_2, HOE1_1, KNY1_3, TOU1_3, BAV1_2, BAV1_3, BAV1_4, BAV2_2, BAV3_2, BAV4_2, BAV5_2, GAM1_1, GRT1_3, KRO1_1, KFI1_2, KOO1_1, SON1_3, SON2_3, TUY1_4
Achnanthidium straubianum	Found in calcareous, mesotrophic to eutrophic fresh waters. ELN1_1, GRO1_3, KEU1_3
Adlafia bryophila	Uncertain ecological requirements, predominantly in clean waters but not necessarily oligotrophic. Aerophilous, frequently found on intermittently wet bryophytes. GRO1_2, BAV4_4
Amphora coffeaeformis	A cosmopolitan species found in waters with high electrolyte content and in brackish and saline inland waters. KOO1_1
Amphora copulata	A cosmopolitan species found in waters with moderate electrolyte content, sometimes occurring in brackish habitats. HOE1_4
Amphora fontinalis	A **rare species** possibly associated with acidic waters. BLO1_4
Amphora inariensis	Probably cosmopolitan, occurring in oligotrophic waters with moderate electrolyte content. KFI2_1, KOO1_1
Amphora montana	A cosmopolitan species found in alkaline waters, rarely becoming dominant. KOO1_4
Amphora normannii	A cosmopolitan aerophilic species found in mountain regions and associated with wetland biotopes. BAV1_4
Amphora ovalis	A cosmopolitan species found in waters with moderate electrolyte content, extending into brackish and saline inland waters. KFI2_1
Amphora pediculus	A cosmopolitan species found in waters with moderate electrolyte content and tolerating critical levels of pollution. This species may be epiphytic on other algae, including diatoms. ELN1_4, GAM1_1, GAM1_2, GRT1_1, GFI1_1, GFI1_2, KFI2_2, SON2_3
Amphora strigosa	A cosmopolitan species abundant in saline habitats. GRO1_3
Amphora veneta	A cosmopolitan species found in waters with elevated electrolyte content, tolerating critical to very heavy pollution. HOE1_4
Asterionella formosa	Widely distributed in the plankton of eutrophic lakes and rivers. Cells are attached by the larger basal pole to form stellate colonies. KRO1_1, KRO2_2
Aulacoseira granulata	Found in both benthos and plankton of eutrophic rivers and lakes. TSI1_4, KOO1_4, SON1_1
Aulacoseira subartica f. *subborealis*	Occurs in alkaline, eutrophic lakes and rivers with moderate electrolyte content. KRO1_1
Bacillaria paradoxa	A cosmopolitan species occurring in electrolyte-rich and brackish waters, usually near the coast. BAV1_3, BAV4_2, BAV6_2, BAV6_4, GAM1_1, GAM1_2, GAM1_3, GRT1_3, KFI2_1, KOO1_3
Brachysira brebissonii	A cosmopolitan species found in acidic, oligotrophic, electrolyte-poor waters. A good indicator for naturally acidic water with no anthropogenic impacts. KAI1_3, TOU1_1, TOU1_3, KRO1_1
Brachysira wygaschii	A cosmopolitan species occurring in oligotrophic, electrolyte-poor waters.
Brachysira zellensis	Cosmopolitan species found in oligotrophic water with a low electrolyte content. GOU1_3
Caloneis molaris	Cosmopolitan, ecology uncertain. KOO1_3
Capartogramma crucicula	A tropical to subtropical diatom species. BAV3_2, KRO2_3
Cocconeis engelbrechtii	This **endemic** South African species occurs in alkaline inland waters with highly elevated electrolyte concentrations (i.e. brackish waters). KNY1_3, BAV4_4, BAV6_4, GRT1_1, KOO1_1
Cocconeis pediculus	A cosmopolitan epiphytic species occurring in waters of a moderate to high electrolyte content, including brackish conditions. GAM1_1, GRT1_2, KFI2_1, KFI2_2, KOO1_1, KOO1_4, SON1_4
Cocconeis placentula	Occurring in mesotrophic to eutrophic flowing and standing waters. Found in abundance on plants, wood and stone. HOE1_2, KNY1_3, TSI1_1, BAV1_3, BAV1_4, BAV2_1, BAV2_3, BAV3_2, BAV4_2, BAV4_4, BAV5_4, GAM1_1, GAM1_3, GAM1_4, GRT1_1, GRT1_3, GFI1_2, KFI2_1, KOO1_1, KOO1_2, KOO1_3, SON1_1, SON1_2, SON1_3, SON1_4, TUY1_4
Cocconeis placentula var. *euglypta*	Similar to the nominate variety. HOE1_4, BAV6_2, GRT1_4, GFI1_1, KFI2_2
Cocconeis placentula var. *lineata*	Similar to the nominate variety but occurring in oligotrophic waters. DIP1_1, BAV1_2, BAV2_2, BAV5_4, BAV6_2, GAM1_2, GRT1_2, GRT1_4, KRO2_2, KOO1_4, SON1_1, TUY1_2, TUY1_3, TUY1_4

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Craticula accomoda	A common characteristic indicator species for high levels of pollution. Found in strongly organically polluted waters, in particular effluent from sewage treatment works. It has a scattered occurrence in oligo- to eutrophic waters. TSI1_1, SON1_2
Craticula ambigua	A cosmopolitan usually epipelic species found in moderately to very electrolyte rich, eutrophic waters, resistant to critical and strong levels of pollution. KFI1_1
Craticula buderi	Cosmopolitan distribution, common in fresh waters with moderate to elevated electrolyte content (e.g. calcareous streams). Found in a wide range of trophic conditions and occurs in mine effluent. KEU1_2, TSI1_3, BAV1_4, GAM1_1, GAM1_2
Craticula halophile	A cosmopolitan species commonly occurring in salt springs and standing waters with a high to very high electrolyte content. HOE1_1
Craticula molestiformis	A cosmopolitan species occurring in electrolyte rich, often heavily polluted water including sewage effluent. GOU1_1, TSI1_3, BAV4_4
Craticula vixnegligenda	Distribution uncertain, associated with electrolyte poor, mesotrophic to eutrophic waters. BAV2_1, KRO1_1, KRO2_2
Ctenophora pulchella	Occurring in brackish inland waters and may also be found in waters impacted by industrial activities and mining. HOE1_3
Cyclostephanos dubius	A euplanktonic species found in inland waters with elevated chloride concentration as well as calcareous, alkaline waters. GAM1_1, GFI1_2, SON1_1
Cyclotella meneghiniana	This taxon has a cosmopolitan distribution in the benthos and plankton of eutrophic, electrolyte rich rivers, streams and lakes. DIP1_1, HOE1_1, TSI1_4, BAV4_2, BAV5_2, GAM1_1, GAM1_2, GAM1_3, GAM1_4, GRT1_1, GRT1_2, GRT1_4, KRO2_3, KRO2_4, KFI2_1, SON1_3
Cyclotella ocellata	This taxon occurs in mesotrophic to eutrophic waters with an elevated pH (optimum pH 8.4). GRO1_1
Cymatopleura solea	A cosmopolitan species found in eutrophic waters with moderate to high electrolyte content sometimes found in brackish biotopes. Favouring alkaline waters. An epipelic and epiphytic species found in the littoral zone. GRT1_1
Cymbella aspera	A cosmopolitan species found in oligotrophic waters with moderate electrolyte content. Attached to the substratum by dichotomous mucilage stalks. BAV1_3, BAV3_2, BAV4_2, BAV4_4, BAV5_2, GRT1_2, GRT1_3, GRT1_4
Cymbella neocistula	A cosmopolitan epiphytic and epilithic species found in circumneutral to slightly alkaline, mesotrophic waters with moderate to high electrolyte content. KFI1_4, KOO1_3
Cymbella tumida	A cosmopolitan species found in oligo- to mesotrophic waters with moderate electrolyte content. Occurs in the littoral zone of standing and flowing waters. BAV6_4, GAM1_1, KOO1_3, KOO1_4
Cymbella turgidula	A cosmopolitan species found in oligotrophic to mesotrophic, alkaline waters with moderate electrolyte content. KOO1_1
Cymbopleura naviculiformis	Cosmopolitan species found in oligotrophic to mesotrophic water with a low to moderate electrolyte content. KRO2_4
Denticula subtilis	A cosmopolitan species found in electrolyte-rich and brackish waters. BLO1_4, HOE1_4
Diadesmis confervacea	A cosmopolitan species found in range of waters, including eutrophic, electrolyte rich and extremely polluted waters. TOU1_1, BAV3_2, BAV4_2, BAV5_2, BAV5_4, GAM1_2, GRT1_2
Diadesmis contenta	A cosmopolitan species found in small bodies of oligotrophic acidic water such as morasses, wetlands and small streams. Also found in biotopes with greatly reduced light intensity. HOE1_4, TUY1_4
Diatoma vulgaris	Found in mesotrophic to eutrophic waters with average electrolyte content. The cells are joined at the corners forming zig-zag colonies. BAV5_4
Diploneis elliptica	A cosmopolitan species occurring in oligotrophic standing waters, especially those with moderate electrolyte content. KNY1_3, GAM1_1, GAM1_3
Diploneis oblongella	Found in well-aerated clean or mildly polluted water with moderate electrolyte content. GAM1_2
Diploneis subovalis	A tropical freshwater diatom species, found in standing waters and occasionally in flowing waters. Occurs in water with moderate to elevated electrolyte content. BAV1_4, KRO2_4
Discotella stelligera	Found in freshwater in the plankton of inland rivers and lakes. KRO1_1, KOO1_1
Encyonema mesianum	A cosmopolitan montane species found in weakly acidic waters. ELN1_3, KEU1_3, BAV2_3, GAM1_2
Encyonema minutum	A cosmopolitan species found in oligotrophic waters with moderate electrolyte content. BLO1_1, BLO1_3, ELN1_1, KAI1_3, TOU1_1, TOU1_3, TSI1_3, KRO2_3, KRO2_4, KOO1_4, SON1_1
Encyonema neogracile	A cosmopolitan species found in oligotrophic, electrolyte-poor waters. GRO1_1, KEU1_1, KEU1_2, BAV2_2, KRO2_4
Encyonema silesiacum	A cosmopolitan species found in standing and flowing oligo- to eutrophic waters and may tolerate strongly polluted conditions. ELN1_1, ELN1_2, BAV7_4
Encyonema ventricosum	A cosmopolitan species found in alkaline well-oxygenated waters. BAV6_4

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Encyonopsis leei var. *sinensis*	This taxon was described from a single site in China, but is known from numerous localities in South Africa. It occurs in slightly acidic, oligo- to mesotrophic waters with low to moderate electrolyte content. GRO1_4, GAM1_1, KOO1_1
Encyonopsis raytonensis	This **endemic** species has only been recorded from South Africa. Found in acidic, well-oxygenated waters. DIP1_1
Encyonopsis subminuta	Cosmopolitan species found in calcareous water with moderate electrolyte content. Requires an oxygen-rich environment. KRO2_3, KOO1_1
Eolimna minima	Cosmopolitan, found in a wide range of waters including heavily polluted biotopes. May possibly be associated with organic detritus. DIP1_1, HOE1_4, KEU1_1, TSI1_4, BAV1_2, KRO2_3
Eolimna subminuscula	A cosmopolitan species common in electrolyte-rich, strongly polluted rivers and flowing waters. BAV2_1, GAM1_4, KFI1_2, SON1_1
Epithemia adnata	A cosmopolitan species found in both flowing and standing waters of moderate to high electrolyte content. Also extending into brackish biotopes. Tolerant to elevated water temperatures. DIP1_1, BAV1_2, BAV1_3, BAV1_4, BAV2_2, BAV6_4, GRT1_1, GRT1_2, GRT1_3, KFI1_1, KFI1_2, KFI1_3, KFI1_4, KFI2_1, KFI2_2, KOO1_1, KOO1_4, SON1_4
Epithemia sorex	A cosmopolitan species found in both flowing and standing waters of moderate to high electrolyte content. Also extending into brackish biotopes. HOE1_2, BAV6_2, GRT1_1, GRT1_2, GRT1_4, KFI1_1, KFI1_2, KFI1_4, KFI2_1, KFI2_2
Eunotia bilunaris	Found in acidic, flowing or standing waters with low electrolyte content. GRO1_1, GRO1_2, GRO1_4, KAI1_2, KAI1_3, KNY1_3, TOU1_1, TOU1_3
Eunotia exigua	Found particularly in oligotrophic electrolyte-poor and extremely acidic habitats (acidobiontic). GOU1_2, GOU1_4, GRO1_1
Eunotia flexuosa	Occur in oligotrophic, standing or slow flowing waters. BLO1_3, ELN1_1, ELN1_2, ELN1_3, GOU1_3, GRO1_2, GRO1_3, HOE1_1, HOE1_4, KAI1_1, KEU1_1, KEU1_3, KRU1_1, KRU1_3, TOU1_1, TSI1_2, TSI1_4, BAV2_1, BAV4_2, BAV5_2, BAV5_4, KRO2_3
Eunotia formica	Found in standing or slow flowing dystrophic to oligotrophic waters with average electrolyte content, although this has not been confirmed. ELN1_4, KAI1_4, KNY1_3, TOU1_3, BAV1_4, BAV3_2, BAV4_4, BAV6_4
Eunotia incisa	Occurs in upland streams in acidic (acidobiontic), oligotrophic, electrolyte-poor waters. BLO1_3, ELN1_3, ELN1_4, GOU1_3, GOU1_4, GRO1_1, GRO1_2, GRO1_3, GRO1_4, HOE1_3, KAI1_1, KAI1_2, KAI1_3, KAI1_4, KEU1_1, KRU1_3, TOU1_3, TSI1_1, BAV1_4, KRO2_3
Eunotia minor	Occurs in circumneutral waters, in pools and springs. An acidophilous species. BLO1_2, BLO1_3, BLO1_4, ELN1_1, ELN1_2, ELN1_3, ELN1_4, GOU1_1, GOU1_2, GOU1_3, GRO1_1, GRO1_2, GRO1_3, GRO1_4, HOE1_1, HOE1_2, HOE1_4, KAI1_3, KAI1_4, KEU1_3, KNY1_3, KRU1_1, KRU1_2, KRU1_3, TOU1_1, TOU1_3, TSI1_2, TSI1_3, TSI1_4, BAV2_2, BAV2_3, BAV3_2, BAV4_2, BAV4_4, BAV5_2, BAV5_4, BAV7_4, GRT1_2, GRT1_3, GRT1_4, KRO2_2, KRO2_3, KRO2_4, KFI1_2, SON1_2, SON1_3
Eunotia pectinalis var. *undulata*	Found in circumneutral to weakly acidic, electrolyte-poor waters. ELN1_2, ELN1_4, KRU1_1, BAV4_4
Eunotia rhomboidea	Found in oligotrophic, electrolyte-poor waters. ELN1_3, KAI1_2, KRU1_1, KRU1_3, BAV1_4
Fragilaria biceps	This cosmopolitan taxon is found in the benthos of rivers and lakes and is easily suspended in the plankton due to its relatively large surface area. Often found in mesotrophic to eutrophic waters. Living cells are usually apically attached to a substratum by a mucilage pad or free living. BLO1_4, DIP1_1, GRO1_1, GRO1_2, HOE1_3, HOE1_4, KEU1_1, KEU1_2, TOU1_3, TSI1_2, TSI1_3, BAV1_3, BAV1_4, BAV2_1, BAV2_2, BAV2_3, BAV3_2, BAV4_2, BAV4_4, BAV5_2, BAV5_4, BAV6_4, BAV7_4, GAM1_1, GAM1_4, GRT1_2, GRT1_2, GRT1_3, GRT1_4, KRO1_1, KRO2_2, KRO2_3, KRO2_4, GFI1_1, KFI1_1, KFI1_3, KFI2_1, KOO1_3, KOO1_4, SON1_3, SON2_3
Fragilaria capucina	This benthic cosmopolitan taxon is found in circumneutral, oligo- to mesotrophic waters with moderate electrolyte content. KEU1_3, TSI1_2, BAV1_2, BAV1_3, BAV2_3, BAV4_2
Fragilaria capucina var. *rumpens*	A cosmopolitan benthic taxon in oligo- to mesotrophic fresh waters. ELN1_2, GRO1_4, KEU1_1, BAV1_4, BAV2_1, BAV4_4
Fragilaria capucina var. *vaucheriae*	Wide ecological range, not clearly defined. DIP1_1, GRT1_1, KRO2_4, GFI1_1, KOO1_4, TUY1_3, TUY1_4
Fragilaria crotonensis	This cosmopolitan taxon is found in the plankton of lakes and standing water bodies. Occurs in oligotrophic to weakly eutrophic, slightly alkaline freshwater with moderate electrolyte content. BAV5_4
Fragilaria parasitica	A cosmopolitan benthic taxon found in mesoeutrophic, circumneutral waters. Often found attached to other algae, including other diatoms. BAV4_4
Fragilaria parasitica var. *constricta*	Cosmopolitan benthic species occurring in mesotrophic to eutrophic, circumneutral waters. Often found attached to other algae, including other diatoms. BAV4_2, KOO1_1

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Fragilaria tenera	This cosmopolitan taxon is found in the benthos of rivers and lakes and is easily suspended in the plankton due to its relatively large surface area. Often found in mesotrophic to eutrophic waters. BAV5_2
Fragilaria ulna	This cosmopolitan taxon is found in the benthos of rivers and lakes and is easily suspended in the plankton due to its relatively large surface area. Often found in mesotrophic to eutrophic, alkaline waters. BLO1_4, GRO1_1, TSI1_4, BAV1_2, BAV1_3, BAV3_2, BAV6_4, TUY1_2
Frustulia crassinervia	A cosmopolitan species occurring in oligotrophic standing waters, especially those with a low electrolyte content. GRO1_1, GRO1_4, KEU1_2
Frustulia rostrata	Found in acidic standing or flowing waters also associated with bryophytes. ELN1_3, ELN1_4, KAI1_2, KNY1_3
Frustulia saxonica	A cosmopolitan species occurring in dystrophic, acidic, electrolyte-poor waters. BLO1_1, BLO1_2, BLO1_3, BLO1_4, ELN1_1, ELN1_2, ELN1_3, ELN1_4, GOU1_3, GRO1_1, GRO1_2, GRO1_4, HOE1_1, HOE1_2, HOE1_4, KAI1_2, KAI1_3, KEU1_1, KEU1_2, KEU1_3, KRU1_1, KEU1_3, TOU1_1, TOU1_3, TSI1_2, BAV1_2, BAV1_3, BAV1_4, KRO2_3, KRO2_4, GFI1_2
Frustulia vulgaris	A cosmopolitan species with wide ecological amplitude. Occurring in fresh to slightly brackish water habitats. Also found ranging from oligotrophic to highly polluted waters. BLO1_1, BAV6_4, KRO2_4
Geissleria decussis	A cosmopolitan species found in eutrophic, unpolluted or moderately polluted waters with average or slightly elevated electrolyte content. BAV1_4
Gomphonema acuminatum	A cosmopolitan species found circumneutral to weakly alkaline waters. Tolerant of slight or moderate pollution. Attached to the substratum by a mucilage stalk. GRO1_1, KRO2_3
Gomphonema affine	A tropical/sub-tropical species tolerant of elevated electrolyte concentrations. BAV3_2, BAV6_2, KRO2_3, KRO2_4
Gomphonema aff. *gracile*	This taxon is able to tolerate extremely polluted conditions and is found in abundance in mining effluent. KRO2_2
Gomphonema aff. *lagenula*	Ecology unknown. BLO1_4, BAV1_3, BAV2_1, BAV3_2, KRO2_4
Gomphonema angustum	Cosmopolitan species commonly occurring in oligotrophic waters. Found to also occur in a wide range of pH and electrolyte concentrations, including calcium-rich waters. TSI1_2, BAV1_3
Gomphonema capitatum	Ecological requirements similar to *G. truncatum*. KEU1_2
Gomphonema clavatum	A cosmopolitan montane species found in oligotrophic waters but tolerating high electrolyte content. BAV5_2
Gomphonema insigne	A cosmopolitan species found in electrolyte-rich waters. BAV6_4, KOO1_1
Gomphonema italicum	Similar to *G. truncatum*, also found in slightly eutrophic habitats. GAM1_3
Gomphonema lagenula	A poorly delineated form little is known of the ecology. KRO2_2
Gomphonema laticollum	A sub-cosmopolitan species, found in slightly eutrophic habitats. Cells of *G. laticollum* are attached to substrata by dichotomous mucilage stalks. BAV5_4
Gomphonema minutum	A cosmopolitan species found in eutrophic waters but not tolerant to more than moderate levels of pollution. Attached to a substratum by dichotomous mucilage stalks. GAM1_1, GAM1_2
Gomphonema parvulum	A cosmopolitan species which is very widespread in a range of waters, from small pools to lakes and rivers and generally considered to be tolerant of extremely polluted conditions. BLO1_4, DIP1_1, ELN1_1, HOE1_4, TSI1_2, BAV1_3, BAV1_4, BAV2_2, BAV3_2, BAV4_4, BAV7_4, GAM1_2, GAM1_3, GRT1_1, GRT1_2, GRT1_4, GFI1_1, KFI1_2, KFI2_1, KFI2_2, KOO1_4, SON1_1, SON1_2, SON1_3
Gomphonema pseudoaugur	A cosmopolitan species found in mesotrophic to eutrophic waters but not tolerant of more than critical levels of pollution. Attached to the substratum by dichotomous mucilage stalks. BAV5_4
Gomphonema pumilum var. *rigidum*	A cosmopolitan species found in mesotrophic to eutrophic waters with moderate electrolyte content. Not tolerant of more than critical levels of pollution. BAV6_4
Gomphonema truncatum	A cosmopolitan species found in oligotrophic waters with elevated electrolyte content but not tolerant of more than moderate pollution. BAV1_3, BAV2_1, BAV2_3
Gomphonema venusta	This species was **described from samples from South Africa** by three American researchers in Passy *et al.* 1997. This species occurs very commonly in the northern and central parts of South Africa. Found in circumneutral to weakly alkaline, oligotrophic to mesotrophic waters with a low to moderate electrolyte content. BAV1_2, BAV2_1, BAV2_2, BAV2_3, SON2_3
Gyrosigma acuminatum	A cosmopolitan species found in electrolyte-rich to brackish waters. May tolerate critical levels of organic pollution. GAM1_3
Gyrosigma attenuatum	This species is unable to tolerate critical levels of pollution. GAM1_4
Gyrosigma rautenbachiae	This species has been found in abundance in standing and slow flowing, brackish inland waters impacted by industrial pollutants. GAM1_1, GRT1_4, KRO2_4, KFI2_1
Gyrosigma scalproides	This species appears to be able to grow in limited light and is often found in turbid waters. HOE1_3, HOE1_4, KOO1_3

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Hantzschia amphyoxys	A cosmopolitan species favouring periodically dry habitats, including soils and rock crevices. Widespread in a range of rivers, but probably washed in from soils. GRT1_1
Hippodonta hungarica	May be the first record of this species in South Africa. One other record for this species occuring in Africa was made for Ghana in: Smith, Smith & Nii Yemoh Annang 2015) HOE1_1
Lemnicola hungarica	Occurs in weakly alkaline waters with moderate to elevated electrolyte content and may occur in critically polluted waters. Found mostly as an epiphyte, commonly found associated with *Lemna* spp. (duckweed). GOU1_3, BAV5_2
Luticola acidoclinata	A cosmopolitan species found in oligo- to dystrophic, circumneutral to slightly acidic, electrolyte-poor waters. BAV1_3
Luticola goeppertiana	A cosmopolitan species commonly occurring in electrolyte-rich, subaerial environments as well as in heavily polluted waters. KNY1_3, SON2_3
Luticola kotschyi	Found in thermal waters, as well as waters with elevated electrolyte content. BAV2_3
Melosira varians	This cosmopolitan taxon is found in both the benthos as well as the plankton and becomes particularly abundant in eutrophic, occasionally slightly brackish, waters. GRO1_1, HOE1_1, HOE1_4, TSI1_4, BAV4_2, BAV4_4, BAV5_4, BAV6_2, GAM1_1, GAM1_2, GAM1_3, GAM1_4, GRT1_4, GFI1_2, KFI1_3, KFI2_1, KOO1_3, KOO1_4
Navicula angusta	A cosmopolitan species, found in weakly acidic, oligotrophic, clean, un-impacted, electrolyte-poor waters. A good indicator for these conditions. KRO2_4
Navicula antonii	Cosmopolitan, found in eutrophic to hypereutrophic waters with moderate to high electrolyte content. Tolerant of strongly polluted conditions. A good indicator for such anthropogenic impacts. GFI1_2
Navicula arvensis var. *maior*	A cosmopolitan species found in waters with moderate to elevated electrolyte content. BAV2_1
Navicula capitatoradiata	Cosmopolitan, found in eutrophic waters, fresh waters with high electrolyte content as well as in brackish waters. Tolerant of critical levels of pollution. BAV4_4, KFI2_1, SON1_4
Navicula cincta	A (possibly) cosmopolitan species found in oligotrophic, calcareous waters. BAV3_2
Navicula cryptocephala	A cosmopolitan species with very wide ecological amplitude. Occurring in weakly acidic, oligotrophic, electrolyte poor waters and also in weakly alkaline, eutrophic, moderately electrolyte-rich waters. Tolerant to critical levels of pollution. KRO2_3
Navicula cryptotenella	A cosmopolitan species, very common in South Africa. Occurs in all freshwater biotopes which range between oligo- to eutrophic. Tolerant only of moderately polluted conditions. BAV1_4, BAV6_4, GAM1_1, KFI2_2
Navicula cryptotenelloides	A cosmopolitan species found in mesotrophic to eutrophic calcareous streams and lakes. GRO1_3
Navicula erifuga	A cosmopolitan species found in eutrophic, brackish waters or those with very high electrolyte content. Tolerant of critical levels of pollution. TSI1_3, KFI1_4
Navicula germainii	Cosmopolitan, found in eutrophic waters, tolerant of critical levels of pollution. BAV4_4
Navicula libonensis	A cosmopolitan species, found in eutrophic, electrolyte-rich waters and able to tolerate critical or occasionally even heavier pollution levels. KFI1_3
Navicula longicephala	Cosmopolitan in eutrophic, electrolyte-rich to brackish waters. Tolerant of critical levels of pollution. GRO1_3
Navicula radiosa	A cosmopolitan species occurring in a wide variety of waters ranging from humic, weakly acidic, oligotrophic, electrolyte-poor waters to strongly alkaline, eutrophic, calcareous waters. This species is however, very sensitive to organic pollution. BLO1_4, HOE1_2, BAV1_2, BAV1_3, BAV1_4, BAV2_1, BAV3_2, BAV5_2, BAV6_2, GAM1_4, GRT1_1, GRT1_2, GRT1_3, KRO2_3, KFI1_1, KFI2_1
Navicula ranomafenensis	Common species in acidic, oligotrophic, clean waters. KRO1_1
Navicula recens	Cosmopolitan species, found in large eutrophic rivers with elevated electrolyte content, also found in brackish waters. Tolerant to critical levels of pollution. Free living or in mucilage tubes. BLO1_4, TSI1_1, TSI1_4, BAV2_2, BAV7_4, GRT1_3, KRO2_4, KFI1_1, KOO1_3
Navicula reichardtiana	A cosmopolitan species common in eutrophic, moderately electrolyte-rich and particularly in calcareous waters. Tolerant of critical levels of pollution, a good indicator of these conditions. KRO2_3
Navicula rhynchocephala	Cosmopolitan, found in oligo- to eutrophic freshwaters with low to moderate electrolyte content. Tolerant of critical levels of pollution, but living preferentially in clean waters. HOE1_1, HOE1_2, BAV4_2
Navicula riediana	A common species in South Africa found in alkaline, eutrophic, electrolyte-rich waters. HOE1_3, TSI1_4, KFI1_4
Navicula rostellata	A cosmopolitan eutrophic species. Tolerant of critical levels of pollution. KOO1_3
Navicula tripunctata	Cosmopolitan, free living and in mucilage tubes. A good indicator of eutrophic waters with moderate to high electrolyte content. Tolerant to critical levels of pollution. HOE1_2, HOE1_3, GFI1_1, KFI2_1

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Navicula veneta	Cosmopolitan, common in heavily eutrophied, electrolyte-rich to brackish water. Very pollution tolerant, often the dominant species in industrially impacted waters. BLO1_4, BAV5_4, SON1_2
Navicula zanonii	A tropical to sub-tropical species, found commonly in alkaline waters in South Africa. BAV4_4, SON2_3
Navicymbula pusilla	Cosmopolitan, found oligo- to eutrophic waters with a moderate to high electrolyte content, especially waters with higher Ca- and Cl-salinity. KNY1_3
Nitzschia acicularis	A planktonic as well as epipelic species found in eutrophic waters with moderate to high electrolyte content. Tolerant of strong pollution but not of extremely polluted conditions. DIP1_1, GRO1_2, KOO1_4
Nitzschia archibaldii	A cosmopolitan species found in circumneutral, slightly to moderately polluted waters with moderate electrolyte content. Reported to be tolerant of Pb and Zn. GFI1_1
Nitzschia aurariae	A cosmopolitan species occurring in electrolyte-rich waters and sporadically in other types of waters. Found commonly in effluent waters from gold mines. GOU1_3, BAV1_4, KFI2_2, KOO1_3, SON1_3
Nitzschia capitellata	A widespread species occurring in electrolyte-rich and brackish waters. Tolerant of extremely polluted conditions. TUY1_4
Nitzschia clausii	A cosmopolitan species found in brackish coastal waters as well as in electrolyte-rich inland waters. In large rivers systems, this species may be associated with industrial effluents and is tolerant of strongly polluted conditions. TSI1_3, TSI1_4
Nitzschia closterium	A cosmopolitan species usually found in the plankton of brackish waters but extending into other brackish biotopes such as wetlands. Also found in saline inland waters. GRO1_4, HOE1_4, KAI1_3, BAV1_3, BAV1_4, BAV2_1, BAV4_2, BAV5_4, BAV7_4, GRT1_4, KRO2_4, KFI1_1, KFI1_4, KFI2_2
Nitzschia desertorum	A cosmopolitan species found in electrolyte-rich and brackish inland waters. GAM1_4
Nitzschia dissipata	A cosmopolitan species found in waters of moderate to high electrolyte content, not present in waters of low electrolyte content. KEU1_2, KNY1_3, KFI1_1, KFI2_1
Nitzschia dissipata var. *media*	A cosmopolitan species found in waters of moderate to high electrolyte content, not present in waters of low electrolyte content. GRT1_2
Nitzschia etoshensis	Occurs in electrolyte-rich to saline waters. GRT1_1
Nitzschia filiformis	A cosmopolitan species found in waters of moderate to high electrolyte content also extending into brackish waters. Tolerant of strongly polluted conditions, but not of critical levels of pollution. Often found occurring in mucilage tubes. ELN1_3, GRO1_2, HOE1_1, HOE1_3, KAI1_4, TSI1_2, TSI1_3, BAV1_3, BAV1_4, BAV2_3, BAV7_4, GAM1_1, GRT1_2, GRT1_3, GRT1_4, KRO2_4, GFI1_1, GFI1_2, KFI1_1, KFI2_1, KOO1_3, KOO1_4
Nitzschia frustulum	A cosmopolitan species found in electrolyte-rich and brackish waters. Tolerant of fluctuations in osmotic pressure and of critical levels of pollution. BAV6_2, GAM1_2
Nitzschia gracilis	A cosmopolitan species found in eutrophic, electrolyte-rich waters but not tolerating more than moderately polluted conditions. BLO1_4, GOU1_3
Nitzschia intermedia	Found in the littoral zone of large eutrophic rivers and lakes with moderate to high electrolyte content. This species does not tolerate more than critical levels of pollution. KFI2_1, KOO1_4
Nitzschia iremissa	Little is known about the ecology of this species, but it is thought to be tolerant of elevated levels of pollution. ELN1_1, GRO1_3, KFI2_2
Nitzschia liebertruthii	A cosmopolitan species occurring in very electrolyte-rich to brackish waters. KNY1_3, BAV2_3, BAV3_2, GAM1_1, GAM1_3, KFI1_1, KFI2_2, SON1_1
Nitzschia linearis	A cosmopolitan species with a wide ecological range, favouring circumneutral, oxygen rich waters of moderate to high electrolyte content. Tolerant of moderately polluted conditions. BLO1_4, KFI2_1
Nitzschia linearis var. *subtilis*	A cosmopolitan species with a wide ecological range, favouring circumneutral, oxygen rich waters of moderate to high electrolyte content. Tolerant of moderately polluted conditions. KFI1_4
Nitzschia littorea	A cosmopolitan species usually found in coastal waters but also occurring in brackish inland biotopes influenced by mining effluent. BLO1_4
Nitzschia microcephala	A cosmopolitan species usually found in electrolyte-rich waters with critical levels of pollution. Tolerant of changes in osmotic pressure. GFI1_1, KFI1_1, KFI2_1
Nitzschia nana	A cosmopolitan species found in brackish and electrolyte-rich waters, able to tolerate changes in osmotic pressure but found in moderately polluted waters only. KRO2_4
Nitzschia palea	A cosmopolitan and very commonly occurring species found in eutrophic and very heavily polluted to extremely polluted waters with moderate to high electrolyte content. BLO1_4, GRO1_4, HOE1_3, TOU1_3, TSI1_2, TSI1_4, BAV1_3, BAV4_2, BAV4_4, BAV6_2, GAM1_1, GRT1_3, GRT1_4, KRO2_4, KFI1_1, KFI1_4, KOO1_1, KOO1_3, SON1_2, SON1_3, SON1_4
Nitzschia pusilla	A cosmopolitan species found in a variety of eutrophic waters as well as on damp earth. Not tolerant of pollution. BLO1_4, KNY1_3

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Nitzschia recta	A cosmopolitan species common in a variety of water types but not tolerating more than moderately polluted conditions. GRO1_2
Nitzschia sigma	A cosmopolitan species found in eutrophic, electrolyte-rich inland waters and extending into brackish estuarine and coastal biotopes. BAV4_2, BAV7_4
Nitzschia sublinearis	Found in slightly to moderately polluted, electrolyte-rich waters. BAV6_2
Navicula symmetrica	Cosmopolitan in eutrophic, electrolyte-rich waters. Tolerant of strongly polluted conditions. KOO1_3
Nitzschia umbonata	A common species in eutrophic electrolyte rich waters and tolerating extremely polluted conditions. BAV1_4, BAV5_2, BAV7_4, GRT1_2, KRO2_4, KOO1_2, SON1_1, SON1_4, SON2_3
Nitzschia communis	Cosmopolitan species found in electrolyte-rich and brackish waters. Tolerant of extremely polluted conditions. BAV2_3
Pinnularia divergens	A montane species occurring in acidic, oligotrophic, electrolyte-poor waters. KFI1_3
Pinnularia gibba	A cosmopolitan species found in waters with low to moderate electrolyte content, especially in springs and small streams. BLO1_4
Pinnularia microstauron var. *rostrata*	A cosmopolitan species found in clean, circumneutral, oligotrophic waters with low electrolyte content. ELN1_1, GRO1_4
Pinnularia subcapitata	Cosmopolitan, found in oligotrophic electrolyte poor waters. BLO1_1, ELN1_3, GRO1_1, GRO1_2, HOE1_1, HOE1_4, BAV1_3, BAV2_3, BAV7_4
Pinnularia viridiformis	Cosmopolitan, one of the most common *Pinnularia* species, in oligo- to mesotrophic waters with low to moderate electrolyte content. HOE1_2, SON1_1
Pinnularia viridis	A cosmopolitan species found in circumneutral, oligo- to mesotrophic waters with low to moderate electrolyte content. BAV4_2
Placoneis dicephala	A cosmopolitan species found on sediments in a range of waters, tolerant of moderate pollution. BAV2_3
Placoneis elginensis	A cosmopolitan species found in range of waters, especially unpolluted to slightly polluted dystrophic waters. GRT1_1
Placoneis placentula	A cosmopolitan species found in unpolluted or slightly polluted waters with moderate to high electrolyte content. BAV1_3
Planothidium engelbrechtii	Found abundantly in saline inland waters with very high electrolyte content. Capable of tolerating critical to very heavy organic pollution. BLO1_3, BLO1_4, HOE1_1, HOE1_2, HOE1_3, KRU1_1, TOU1_1, TSI1_2, BAV1_3, BAV2_3, BAV3_2, BAV4_4, BAV6_2, GAM1_2, KFI2_1
Planothidium frequentissimum	A common species in standing and flowing, circumneutral to alkaline waters with a moderate to high electrolyte content. Capable of tolerating critically polluted conditions. DIP1_1, ELN1_2, BAV1_4, BAV3_2, BAV4_2, BAV4_4, BAV5_2, BAV7_4, GRT1_4, KFI1_1, KFI1_2, KFI1_4, TUY1_3
Planothidium rostatrum	Occurring in circumneutral to alkaline waters with low to moderate electrolyte content. More often attached to plants than stones. BLO1_1, ELN1_1, ELN1_4, GOU1_1, GRO1_1, GRO1_3, BAV5_4, GRT1_2, GRT1_3
Pleurosigma elongatum	Cosmopolitan, found in brackish inland waters. KFI1_1, KFI2_1
Pleurosigma salinarum	Cosmopolitan, found in brackish and saline inland waters. GAM1_3, KFI1_4, KFI2_2
Psammothidium abundans	Previously described as a sub-Antarctic endemic. The species has sporadically been found in South Africa, Australia and Scotland. More recently it was also found to occur in European Rivers. KEU1_1
Pseudostaurosira brevistriata	This taxon is found in clean alkaline fresh waters ranging from oligotrophic to eutrophic. KNY1_3, BAV2_1, GAM1_2, GRT1_1, SON1_1
Reimeria sinuata	A cosmopolitan aerophilic species found in montane biotopes, mosses, springs and streams. GRT1_4, SON1_1
Reimeria uniseriata	This species is found in alkaline, mesotrophic to eutrophic waters with moderate electrolyte content. *R. uniseriata* seems to be able to grow in conditions of reduced light penetration (i.e. high turbidity). GFI1_1, GFI1_2, KFI2_2, KOO1_1, KOO1_2, KOO1_3, KOO1_4, SON1_2, SON1_3, SON1_4
Rhopalodia gibba	A cosmopolitan species found in standing and slow flowing waters, especially springs, of moderate to high electrolyte content. BAV6_2, BAV7_4, GRT1_1, GRT1_2, GFI1_1, KFI1_1, KFI1_2, KFI1_3, KFI1_4, KFI2_1, KFI2_2, KOO1_4, SON1_4, TUY1_3
Rhopalodia gibberula	A cosmopolitan species found in waters of moderate to high electrolyte content. Tolerant of elevated water temperatures. GRT1_1, GRT1_2, KFI2_1, KFI2_2
Rhopalodia operculata	A cosmopolitan species found in waters of moderate to high electrolyte content. Also found in thermal mineral springs. BLO1_1, TSI1_3, BAV1_3, BAV2_3, KFI1_1, KFI1_4
Sellophora pupula	A cosmopolitan species found in a broad spectrum of electrolyte-rich waters with some populations found under strongly polluted conditions. KEU1_1, BAV2_3, BAV3_2, BAV6_2

Table 6 continued: A summary of all species identified from samples collected during this study, ecological information and the sites where they were collected.

Species	Ecological preference and the sites they were identified from during this study
Sellophora seminulum	A cosmopolitan species found in range of waters, including eutrophic, electrolyte-rich and extremely polluted waters. GRT1_1
Simonsenia delognei	A cosmopolitan species found in electrolyte-rich and brackish waters. Also found in soils and is tolerant of osmotic fluctuations. GOU1_1, KOO1_1
Stauroneis anceps	Widespread in all types of water. GRO1_4
Stauroneis smithii	A cosmopolitan species reported variously from electrolyte poor water as well as from eutrophic waters with moderate electrolyte content. KRO2_3
Staurosira construens	This taxon occurs in standing waters of a good quality. GAM1_2
Staurosira elliptica	Found in the benthos of electrolyte-rich fresh or brackish waters. HOE1_3, KAI1_3, KNY1_3, BAV4_4, BAV5_2, BAV5_4, BAV6_2, GAM1_1, GAM1_2, GAM1_3, GAM1_4, GRT1_1, GRT1_2, GRT1_3, GRT1_4, GFI1_2, KFI1_4
Staurosirella pinnata	Found in clean waters with moderate to high electrolyte content. GAM1_3
Stephanodiscus agassizensis	A planktonic species found in eutrophic rivers and lakes with an elevated electrolyte concentration and turbidity. GFI1_1, SON1_1, SON1_4
Surirella angusta	A cosmopolitan species found in eutrophic waters with moderate electrolyte content. HOE1_4, BAV4_2
Surirella brebissonii	A cosmopolitan species found in waters of moderate to high electrolyte content also extending into brackish waters. KFI2_1
Tabellaria flocculosa	This taxon flourishes in electrolyte-poor, oligotrophic, circumneutral or slightly acidic waters. The cells are linked at the corners forming zigzag colonies which may be attached or planktonic in lakes, pools and streams. ELN1_2, ELN1_3, GOU1_3, GRO1_1, GRO1_2, GRO1_3, GRO1_4, HOE1_2, KAI1_3, KEU1_1, KEU1_2, KEU1_3, KNY1_3, TOU1_1, TOU1_3, TSI1_2, TSI1_3, BAV1_3, BAV2_1, BAV2_2, BAV2_3, GRT1_2, KRO1_1, KRO2_3, KRO2_4, GFI1_2, SON1_4
Tabularia fasciculata	A cosmopolitan species with broad ecological tolerance. It does however appear that this taxon favours moderately to high electrolyte concentrations. Has also been reported from critically polluted industrial wastewater. BLO1_1, BLO1_4, ELN1_1, GOU1_4, GRO1_1, BAV2_3, GAM1_1, GAM1_3, GRT1_1,KRO2_3, KRO2_4, GFI1_1, KFI1_2, KFI2_1
Thalassiosira weissflogi	A halophilic riverine species. GAM1_1, GAM1_3, GAM1_4, GFI1_1
Tryblionella apiculata	A cosmopolitan species found in electrolyte-rich waters. Tolerant of strongly polluted conditions. HOE1_2, BAV7_4, KFI2_2, KOO1_1, KOO1_3, SON1_1
Tryblionella calida	A cosmopolitan species commonly occurring in eutrophic waters with elevated electrolyte content. Favours standing waters. BAV4_2
Tryblionella coarctata	A cosmopolitan species found in brackish waters. KOO1_3
Tryblionella hungarica	A cosmopolitan species found in waters with high electrolyte content to brackish waters. Tolerant of strongly polluted conditions. HOE1_4, GRT1_3, GRT1_4, KFI1_1
Tryblionella levidensis	A cosmopolitan species found in waters ranging from those with moderate electrolyte content to electrolyte-rich and brackish waters. Tolerant of strongly polluted conditions. HOE1_3, GAM1_2, GRT1_2, GRT1_3, KFI2_1, KFI2_2

When 84 samples from 24 rivers (Appendix 1 – Raw data: Species lists by Ecoregion, River, Site and Sample) were compared the results produced 10 different groups (Figure 59). The analysis was based on species abundance data with a Bray-Curtis similarity performed before the cluster analysis was run. Outlier sites were removed and a 40% similarity was used as a minimum for group formation. Except in the case of Group 10, where GFI1_2 and GAM1_3 separated out at a value just less than 40%.

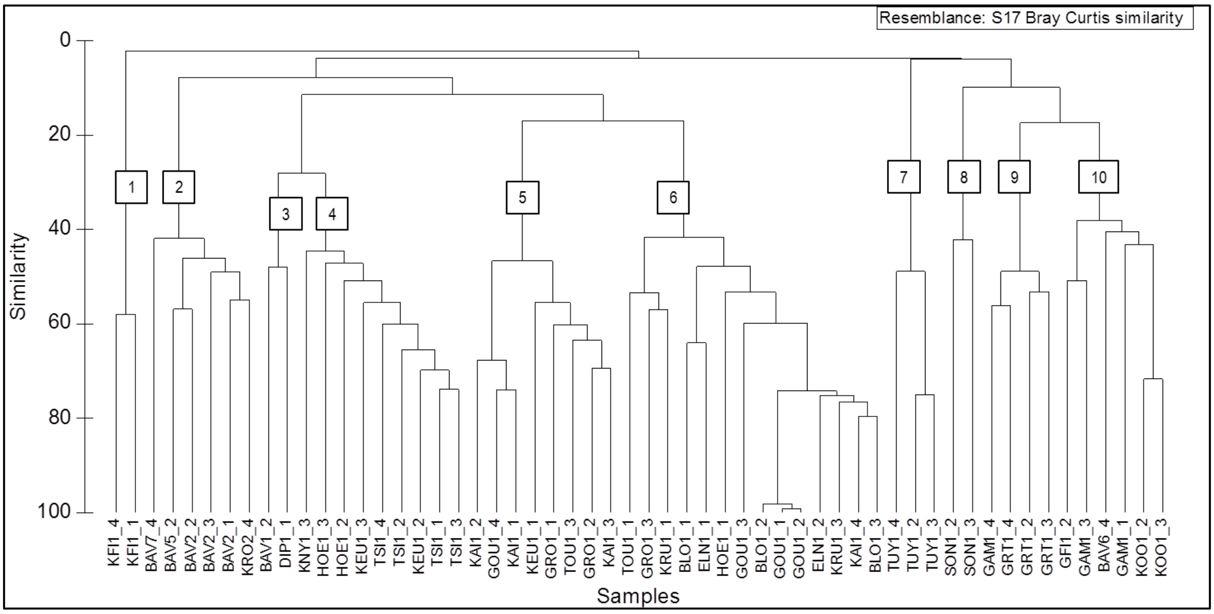

Figure 59 Cluster diagram of community composition compared across all sites and season sampling efforts. (see Table 4 for abbreviations)

The statistic showed the formation of 10 distinct groups. There was no clear separation between seasonal samples, instead the groupings showed a spatial separation between sites in different geographical regions. This is clearly seen with KFI1, BAV2, TSI1, KAI1, GOU1, TUY1, SON1, GRT1 and KOO1 which showed strong similarity at a site level (more than one sample from the same site falling within the same group).

Group one consists of KFI1 sites and **group two** contains sites from BAV2, BAV7, BAV5 and KRO2. These groupings all seem to be either at a site level or a regional level as is the case with KRO2 which is the neighbouring catchment of the Baviaanskloof in which all other group two sites are located. **Group three** consists of a BAV1 and the DIP1 sites**. Group four** is quite large with KNY1, HOE1, KEU1 and all four TSI1 sites falling within this group. All sites are within the South Eastern Coastal Belt ecoregion.

Group five contains three KAI1 sites as well as two GRO1 sites and one sample each from the GOU1, KEU1 and TOU1 sites. Once more all these sites fall within the South Eastern Coastal Belt ecoregion which could explain the similarity in community composition. **Group six** is by far the largest grouping of samples. This group contains one sample from the TOU1, HOE1, GRO1 and KAI1 study sites. Two samples from sites KRU1, ELN1 and BLO1 and three samples from GOU1, all of which fall within the South Eastern Coastal Belt ecoregion.

Group seven consists solely of samples collected from the TUY1 site. This makes sense since the Amathole was the most north-eastern site sampled during this study, and is situated high up in the catchment (mountain stream) within the Amathole Mountains. **Group eight** is similar to group seven with respect to being solely comprised of samples collected from one sampling site; SON1 in this case. This is interesting since the SON1 site is situated on the Volkers River which is used as a water transfer channel between the Orange River catchment and the Sundays River catchment. More detailed analysis of the community composition might shed some light on the possible impact this transfer scheme could be having on the diatom community and river's ecology.

Group nine is composed of GRT1 samples and one GAM1 sample. This is not too peculiar since the GAM1 site is situated upstream of GRT1 and just upstream of the confluence with the Kouga River which together with the Groot River (GRT1) forms the Gamtoos River (GAM1) upstream of the town of Patensie. The GAM1 site is just downstream of Patensie. All sites fall within the Southern Folded Mountains ecoregion.

Group ten contains the other GAM1 samples which grouped together with a sample from GFI1 and BAV6 respectively and the KOO1 sites. A bit of a diverse grouping however these sites at first glance seem to be somewhat comparable with regards to ecological characteristics. They are all wider rivers flowing through sandy or drier parts of their regions. These sites are spread across two ecoregions, the drought corridor and the Southern Folded Mountains. More detailed assessment is needed to ascertain which environmental drivers and more importantly which species were responsible for this similarity grouping to form.

An MDS ordination supported the Ecoregion Level stratification (Figure 60). Seasons were coloured and ecoregions were labelled number one to three. The ordination shows no apparent colour grouping but does show a grouping by number. The sites numbered with a one grouped more towards the right side and the sites with a number three grouped more towards the left of the ordination.

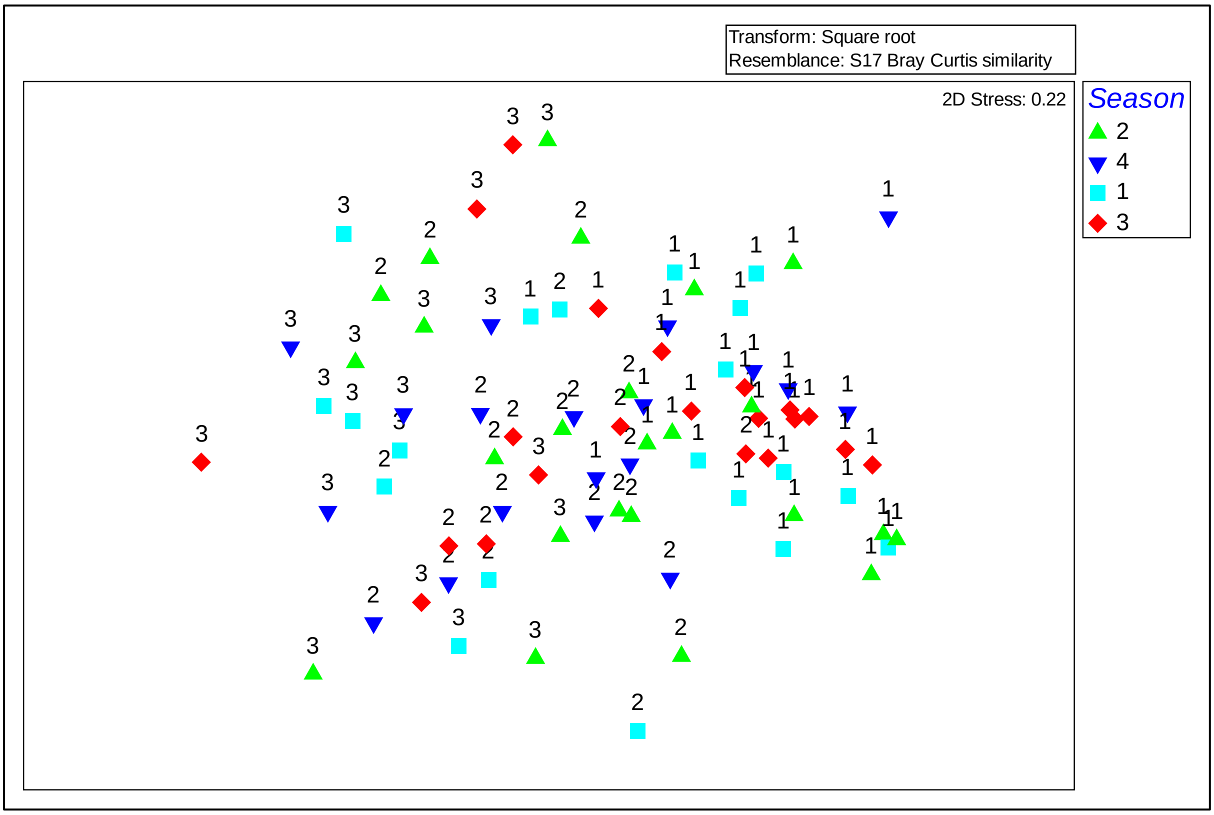

Figure 60 Non-metric MDS ordination of all samples across four seasons and three ecoregions. The colours are by season; turquoise – spring, green – summer, red – autumn and blue – winter. Sample labels are numbered as follows 1 – South Eastern Coastal Belt, 2 – Southern Folded Mountains and 3 – Drought Corridor.

The result obtained was confirmed by an ANOSIM test (Figure 61). The sample statistic or Global R value was 0.538 (the strength of the factors on the samples). An R value close to 1 indicates high separation and closer to 0 indicates no separation between factors. 0.538 is a satisfactory indication that there is a strong separation at Ecoregion Level. The P value was 0.001 (significance of the differences between factors). The black line in the graph below (Figure 61) is not overlaying the permutation of the similarity matrix (multiple grey bars on the left) and the result therefore shows significant difference.

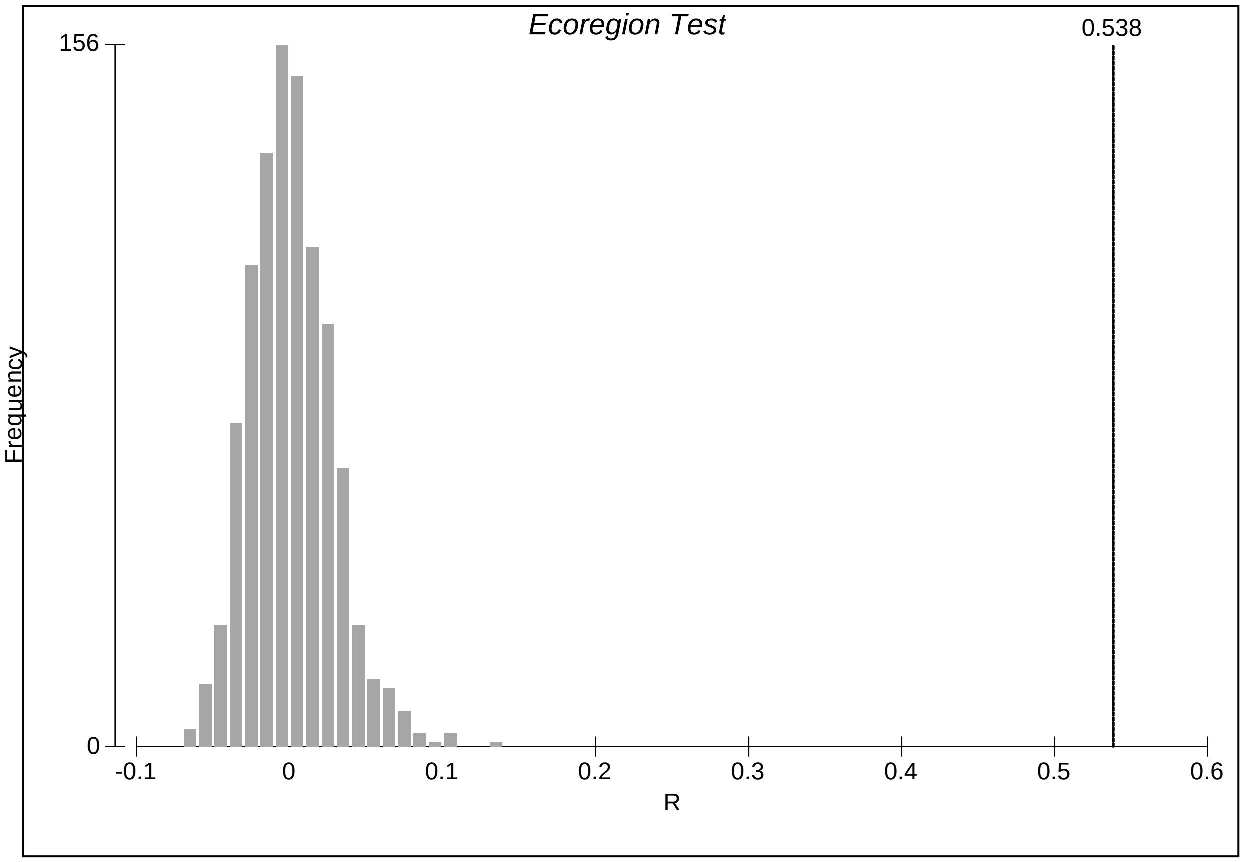

Figure 61 ANOSIM test for the significant differences in species composition between identified groups. In this case pre-selected grouping was applied at Ecoregion Level I.

If the diatom community composition responds at an Ecoregion Level, the sample sites should group according to the regions when all abundance data from the different sampling efforts are added together, thus removing the seasonality from the data sets. A total species list was compiled for each site which contained total species abundance data. Cluster analysis and a MDS ordination confirmed that diatom communities grouped at an Ecoregion Level (Figure 62 and Figure 63). SON2 was an outlier, which makes sense since this sample was taken from a seasonal section of the Sondags River during the annual flow in April 2015.

BAV1 grouped with the South Eastern Coastal Belt (SECB) sites even though it is classified as being situated within the Southern Folded Mountains (SFM) ecoregion. BAV6, GRT1 and GAM1 all grouped with the Drought Corridor (DC) ecoregion instead of the SFM ecoregion to which it is assigned.

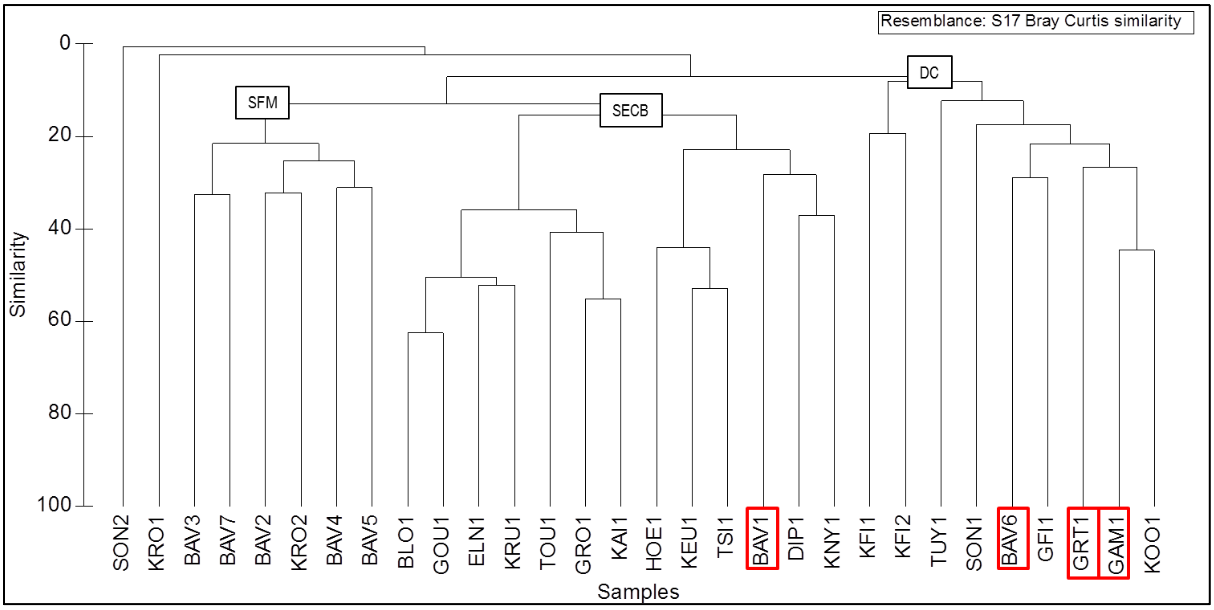

Figure 62 Cluster diagram of community composition comparison based on total species abundance per site. Ecoregions indicated by SFM – Southern Folded Mountains, SECB – South Eastern Coastal Belt and DC – Drought Corridor.

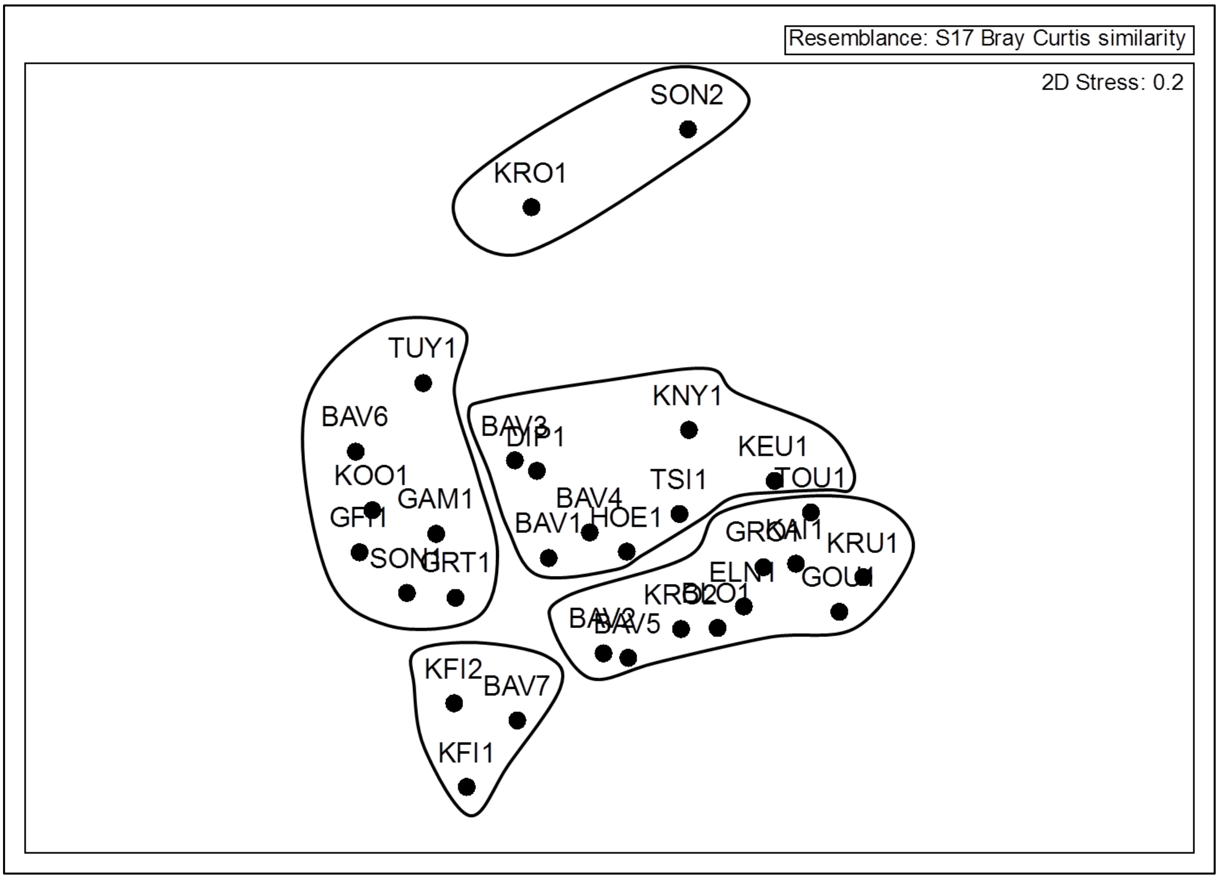

Figure 63 Non-metric MDS ordination of community composition comparison based on total species abundance per site.

A SIMPER test was applied to identify the species responsible for the identity of these groups. A SIMPER test compares samples within the prescribed groups and identifies the species responsible for the formation of these groupings. Below are tables to summarise the results obtained from SIMPER analyses for each ecoregion (Table 7-9). Also contained within the tables are frequencies of occurrence data for each of the ecoregion indicator species. Frequency of occurrence data were obtained through mathematical calculations and are provided in Appendix 3 – Relative Abundance and Frequency of occurrence data at different scale. These calculations take into account:

- the number of sites the species occurred on,
- the abundance of the species in the ecoregion and
- the abundance of the species per site.

The calculations produced two values; **T** which represents the percentage the particular species contributed to the total abundance recorded across all sampled sites in the particular sample size and **F** the percentage of frequency with which the species occurred.

The average similarity between samples in the **South Eastern Coastal Belt (SECB**) was found to be around 29%. The main species responsible for the identity or formation of this group were; *Eunotia minor*, *Achnanthes oblongella*, *Eunotia incisa*, *Frustulia saxonica*, *Eunotia flexuosa* and some other less abundant species listed in the table below (Table 7). The likelihood of finding *Eunotia minor* in a sample collected from a river in the SECB is 79%. *Eunotia minor* accounted for 28% of all species identified from sites sampled in the SECB.

For the **Southern Folded Mountains (SFM**) ecoregion the species responsible for the identity and formation of this group were found to be; *Fragilaria biceps*, *Eunotia minor*, *Nitzschia closterium*, *Achnanthes oblongella*, *Gomphonema parvulum*, *Planothidium frequentissimum* and a variety other species as seen in Table 8 below. The SFM had an average similarity of 27%. The most likely species to be found when identifying individuals from samples collected in the SFM was *Fragilaria biceps* at a percentage frequency of occurrence of 81%.

Species responsible for the identity of the **Drought Corridor ecoregion** are; *Cocconeis placentula*, *Cocconeis placentula* var. *lineata*, *Gomphonema parvulum*, *Rhopalodia gibba*, *Achnanthes oblongella*, *Achnanthidium minutissimum*, *Staurosira elliptica* and quite a few more are listed in Table 9 (see below). The average similarity within this group was around 23%. The DC ecoregion had the largest number of species contributing to group identity.

This ecoregion also had the lowest contribution % for top contributing species which means there is a bigger focus on combination of species than the presence of one or two specific species. *Cocconeis placentula* was the species with the highest percentage frequency of occurrence at 50%, which was much lower than the species with highest percentage frequency of occurrence in the SFM and SECB.

Table 7 SIMPER and frequency of occurrence data for sites situated in the South Eastern Coastal Belt ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

South Eastern Coastal Belt							
Average similarity: 28.76							
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
Eunotia minor	15.91	7.69	1.12	26.74	26.74	28	79
Achnanthes oblongella	12.05	4.69	1.01	16.31	43.05	19	68
Eunotia incisa	10.24	3.77	0.9	13.09	56.14	14	47
Frustulia saxonica	7.78	3.32	0.96	11.56	67.7	7	68
Eunotia flexuosa	3.18	1.33	1.05	4.63	72.33	1	45
Tabellaria flocculosa	3.16	0.81	0.67	2.83	75.16	2	45
Fragilaria biceps	3	0.74	0.62	2.56	77.72	1	32
Planothidium engelbrechtii	2.56	0.72	0.61	2.52	80.24	1	26
Gomphonema parvulum	3.63	0.69	0.41	2.42	82.66	3	13
Achnanthidium minutissimum	2.46	0.69	0.69	2.41	85.06	1	21
Eolimna minima	2.1	0.42	0.36	1.46	86.52	1	11
Achnanthidium eutrophilum	2.71	0.36	0.37	1.26	87.78	2	16
Eunotia formica	2.04	0.34	0.32	1.17	88.95	1	13
Nitzschia palea	1.87	0.32	0.45	1.11	90.06	1	18

Table 8 SIMPER and frequency of occurrence data for sites situated in the Southern Folded Mountains ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

Southern Folded Mountains							
Average similarity: 27.04							
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum%	T%	F%
Fragilaria biceps	13.38	7.11	2.05	26.29	26.29	12	81
Eunotia minor	6.75	2.66	2.01	9.83	36.13	3	54
Nitzschia closterium	5.31	2.27	1.07	8.4	44.53	2	31
Achnanthes oblongella	4.93	2.23	1.3	8.25	52.78	3	54
Gomphonema parvulum	5.21	2.07	0.51	7.65	60.43	3	42
Planothidium frequentissimum	4.11	1.97	0.71	7.29	67.72	2	27
Achnanthidium minutissimum	5.1	1.49	0.68	5.52	73.24	4	38
Diadesmis confervacea	3.8	0.89	0.48	3.28	76.51	2	23
Eunotia flexuosa	2.91	0.66	0.76	2.45	78.96	1	19
Achnanthidium eutrophilum	2.94	0.56	0.48	2.09	81.05	1	23
Nitzschia filiformis	2.29	0.52	0.45	1.94	82.99	2	35
Cymbella aspera	1.9	0.51	0.48	1.9	84.89	0	31
Planothidium engelbrechtii	2	0.42	0.42	1.55	86.43	1	23
Navicula radiosa	0.91	0.32	0.76	1.18	87.61	1	46
Cocconeis placentula	1.15	0.31	0.48	1.15	88.76	6	50
Achnanthes subaffinis	3.03	0.29	0.26	1.08	89.84	2	31
Navicula recens	0.9	0.22	0.48	0.81	90.65	0	19

Table 9 SIMPER and frequency of occurrence data for sites situated in the Drought Corridor ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

Drought Corridor							
Average similarity: 22.63							
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
Cocconeis placentula	10.19	3.42	1.18	15.12	15.12	10	50
Cocconeis placentula var. *lineata*	8.57	2.1	0.73	9.28	24.4	13	25
Gomphonema parvulum	4.7	1.51	1.34	6.68	31.07	3	45
Rhopalodia gibba	5.8	1.43	0.99	6.32	37.39	9	50
Achnanthes oblongella	2.98	1.09	0.93	4.8	42.19	1	50
Achnanthidium minutissimum	4.66	1.04	0.7	4.6	46.79	3	25
Staurosira elliptica	7.5	0.95	0.41	4.21	51	2	10
Fragilaria biceps	3.65	0.94	1.19	4.14	55.14	1	40
Epithemia adnata	4.37	0.86	0.77	3.79	58.93	5	45
Nitzschia palea	4.85	0.82	0.54	3.63	62.55	6	35
Reimeria uniseriata	5.02	0.72	0.36	3.16	65.72	8	50
Navicula radiosa	2.71	0.6	0.57	2.65	68.36	1	10
Nitzschia filiformis	3.28	0.56	0.69	2.46	70.82	1	30
Melosira varians	2.95	0.53	0.7	2.35	73.17	1	25
Fragilaria capucina var. *vaucheriae*	2.96	0.39	0.32	1.74	74.91	2	20
Tabularia fasciculata	1.87	0.39	0.59	1.71	76.63	0	15
Cocconeis placentula var. *euglypta*	3.03	0.36	0.28	1.58	78.21	3	10
Epithemia sorex	3.09	0.35	0.35	1.54	79.76	3	25
Cocconeis pediculus	1.9	0.33	0.5	1.47	81.23	2	20
Bacillaria paradoxa	1.48	0.29	0.54	1.26	82.49	0	10
Navicula recens	1.97	0.28	0.41	1.24	83.73	1	10
Amphora pediculus	2.2	0.28	0.42	1.23	84.96	2	20
Nitzschia microcephala	1.68	0.22	0.29	0.98	85.94	1	15
Cyclotella meneghiniana	1.73	0.2	0.43	0.89	86.83	0	10
Planothidium frequentissimum	1.68	0.16	0.26	0.69	87.52	1	25
Achnanthidium eutrophilum	2.06	0.15	0.24	0.67	88.19	1	5
Nitzschia closterium	1.89	0.14	0.25	0.62	88.81	2	15
Pseudostaurosira brevistriata	2.1	0.14	0.28	0.61	89.42	0	5
Eunotia minor	1.1	0.14	0.29	0.61	90.03	0	15

This information is valuable, since it places emphasis on the community composition rather than the presence or absence of a specific indicator species. The comparison between ecoregions is summarised in Table 10. There were a number of species that played an important role in group identity across all three ecoregions. However, there were more species only indicated in two or especially one ecoregion. This showed that some species, in combination with others, were significant in all three ecoregions, whereas other species were more likely to play a role in community identity in one specific ecoregion. In the SECB 42% of the species only contributed to this specific ecoregion, for SFM it was found to be 29% and for the DC it was the case for 55% of the species. Species specifically important to the community identity of the SECB were; *Eolimna minima*, *Eunotia formica*, *Frustulia saxonica* and *Tabellaria flocculosa*. For the SFM ecoregion species important for community identity were; *Achnanthes subaffinis*, *Cymbella aspera* and *Diadesmis confervacea*. For the DC ecoregion species important for community identity were; *Epithemia adnate*, *Epithemia sorex*, *Fragilaria capucina* var. *vaucheriae*, *Melosira varians*, *Pseudostaurosira brevistriata*, *Reimeria uniseriata*, *Rhopalodia gibba*, *Staurosira elliptica* and *Tabularia fasciculate*. The species that played a role in all three ecoregions were; *Achnanthes oblongella*, *Achnanthidium eutrophilum*, *Achnanthidium minutissimum*, *Eunotia minor*, *Fragilaria biceps* and *Gomphonema parvulum.*

In terms of community composition, there was no apparent separation between seasons however, when the environmental water quality data (Appendix 2 – Raw data: Measured water quality paramaters) were analysed using a PCA ordination, a clear seasonal separation could be seen (Figure 64). This was expected, as water quality parameters change when flow and air temperature change. As expected an increase in temperature was associated with a decrease in dissolved oxygen and vice versa. The vertical plane is strongly associated with temperature and oxygen, while the horizontal plane portrays an increase and decrease in pH, salinity and conductivity.

When the same diagram is re-coloured to show ecoregion, a clear ecoregion gradient can be seen moving in the horizontal plane. The Drought Corridor (DC) ecoregion showed higher conductivity, pH and salinity while the South Eastern Coastal Belt (SECB) ecoregion showed lower readings of pH, salinity and conductivity (Figure 65).

Table 10 Summary of species responsible for group identity, across the three ecoregions (SECB, SFM, DC). Grey indicates that the species was an important component for community identity as indicated by SIMPER results.

Species	SECB	SFM	DC	Species	SECB	SFM	DC
Achnanthes oblongella	■	■	■	*Fragilaria biceps*	■	■	■
Achnanthes subaffinis		■		*Fragilaria capucina* var. *vaucheriae*			■
Achnanthidium eutrophilum	■	■	■	*Frustulia saxonica*	■		
Achnanthidium minutissimum	■	■	■	*Gomphonema parvulum*	■	■	■
Amphora pediculus			■	*Melosira varians*			■
Bacillaria paradoxa			■	*Navicula radiosa*		■	■
Cocconeis pediculus			■	*Navicula recens*		■	■
Cocconeis placentula		■	■	*Nitzschia closterium*		■	■
Cocconeis placentula var. *euglypta*			■	*Nitzschia filiformis*		■	■
Cocconeis placentula var. *lineata*			■	*Nitzschia microcephala*			■
Cyclotella meneghiniana			■	*Nitzschia palea*	■		■
Cymbella aspera		■		*Planothidium engelbrechtii*	■	■	
Diadesmis confervacea		■		*Planothidium frequentissimum*		■	■
Eolimna minima	■			*Pseudostaurosira brevistriata*			■
Epithemia adnate			■	*Reimeria uniseriata*			■
Epithemia sorex			■	*Rhopalodia gibba*			■
Eunotia flexuosa	■	■		*Staurosira elliptica*			■
Eunotia formica	■			*Tabellaria flocculosa*	■		
Eunotia incisa	■			*Tabularia fasciculata*			■
Eunotia minor	■	■	■				

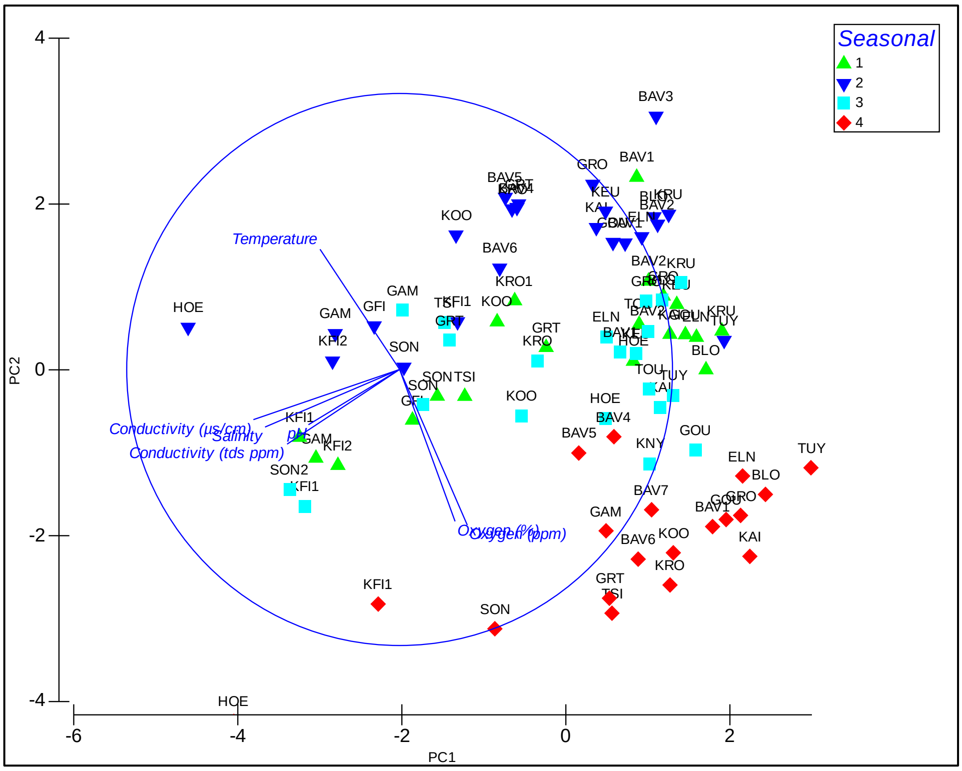

Figure 64 PCA ordination of water quality data collected from all sites sampled over the period of one calendar year (four seasons). Season 1 = spring, 2 = summer, 3 = autumn and 4 = winter.

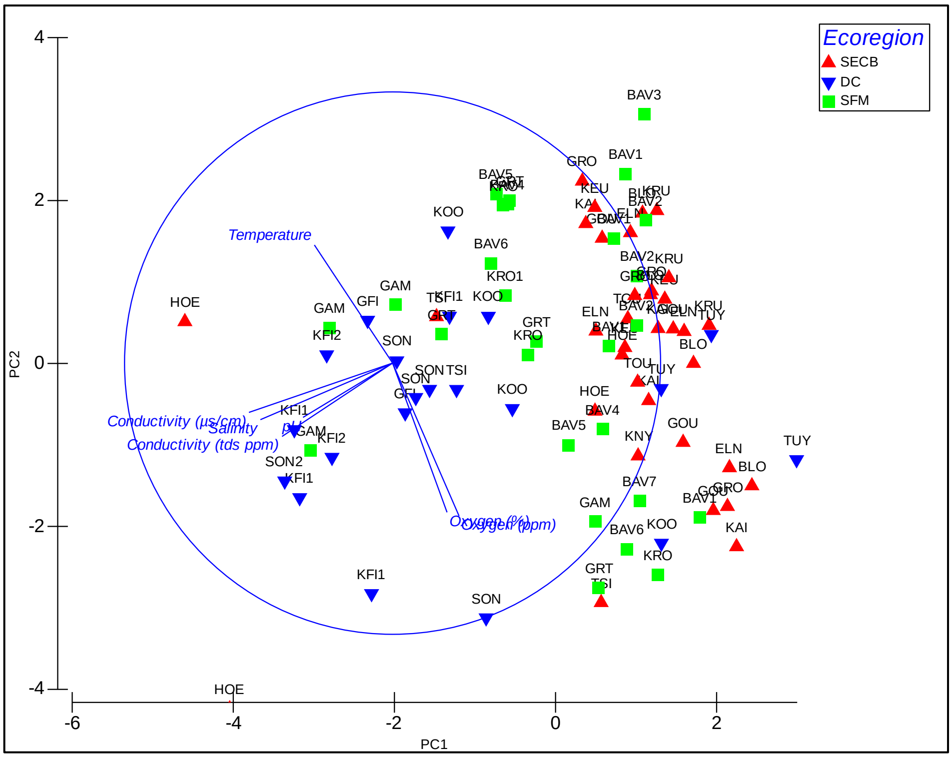

Figure 65 PCA ordination of water quality data for all sites sampled across three ecoregions.

Based on these results, species level data were analysed further at the Ecoregion Level.

The South Eastern Coastal Belt (SECB)

It has been found that smaller resolution differences at a temporal scale gets lost and dissolved in the volume and resolution of analysis. For that reason seasonal level data were used for this smaller scale analysis, to see if temporal changes had a significant impact on the community composition of sites, within each ecoregion. A four group separation occurred as seen in the Cluster diagram below (Figure 66). **Group one** consisted of the majority or all samples of the Bloukrans, Kruis, Elands and Goukamma rivers. **Group two** was composed of samples of the Kaaimans, Groot and Touws rivers. **Group three** consisted of samples collected from the Hoogekraal, Tsitsikamma, Keurbooms and Knysna rivers. **Group four** had one sample of the Hoogekraal, Diep and Bloukrans rivers each.

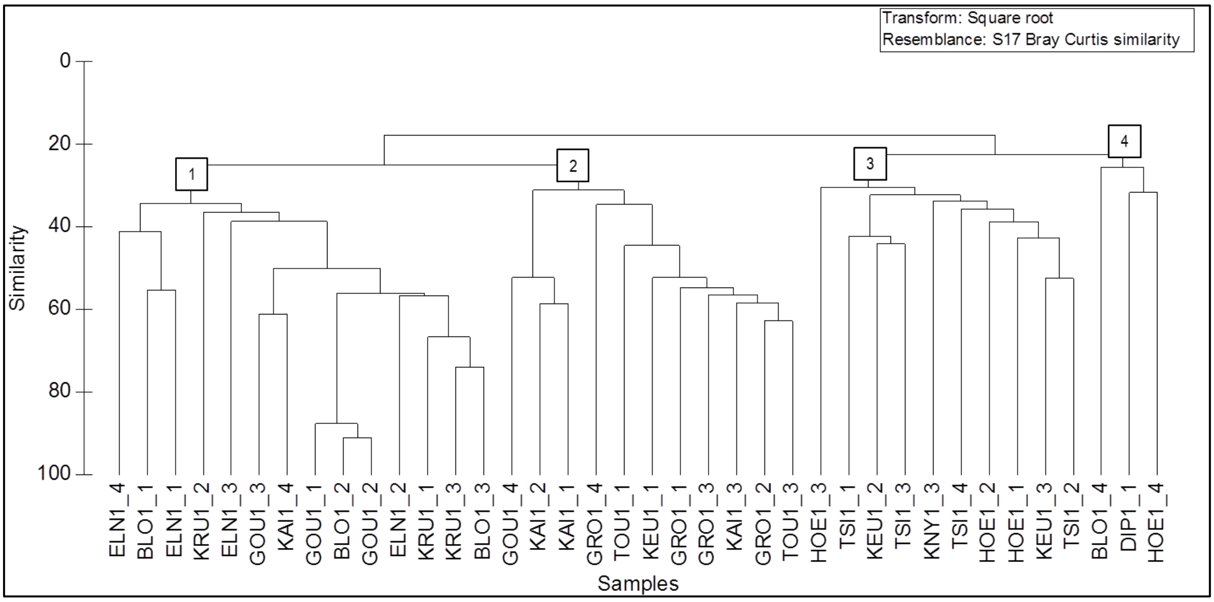

Figure 66 Cluster diagram of seasonal data for sites situated in the South Eastern Coastal Belt ecoregion. Groups are indicated by the numbers one to four.

This pattern of spatial (river) instead of temporal separation was supported by an MDS ordination (Figure 67) and an ANOSIM test which produced a significant result with Global R value of 0.745. A SIMPER analysis identified the species responsible for this grouping with their percentage contribution to the group identity. This data are summarised in Table 11 below. In Figure 69 and Figure 70 light micrograph images of the important species for group identity within the SECB are presented together with their valve dimensions and other information important for species identification.

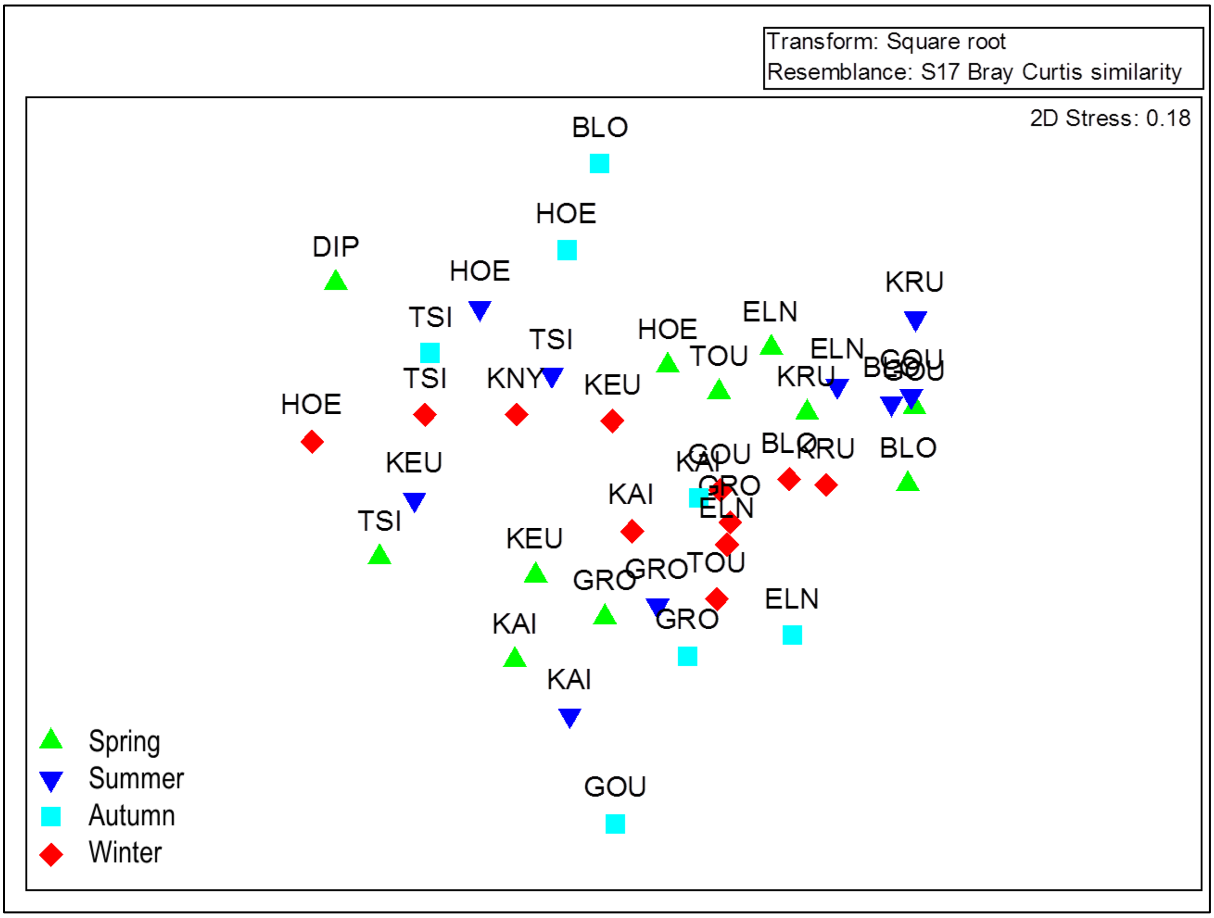

Figure 67 Non-metric MDS ordination of seasonal samples collected from rivers situated in the South Eastern Coastal Belt ecoregion.

For group one, the similarity was 45% and the species responsible for community identity were; *Eunotia minor*, *Frustulia saxonica* and *Eunotia incisa* (Table 11). For group two the similarity was 41% and the species were identified to be; *Eunotia incisa*, *Frustulia saxonica*, *Achnanthes oblongella*, *Eunotia minor* and *Tabellaria flocculosa*. Group three had a group similarity of 34% and the species responsible for community identity were; *Achnanthes oblongella*, *Eunotia minor*, *Planothidium engelbrechtii*, *Frustulia saxonica*, *Fragilaria biceps* and *Tabellaria flocculosa*. Group four had an average similarity of 27% and the species identified for community identity were; *Gomphonema parvulum*, *Achnanthes oblongella*, *Fragilaria biceps*, *Eolimna minima*, *Denticula subtilis* and *Eunotia minor*.

Table 11 SIMPER and frequency of occurrence data for species responsible for group formation in the South Eastern Coastal Belt ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

	Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
Group1 Average similarity: 44.53%	*Eunotia minor*	15.43	35.97	2.18	80.77	80.77	63	100
	Frustulia saxonica	3.32	3.41	0.86	7.66	88.43	5	71
	Eunotia incisa	3.16	2.17	0.43	4.87	93.3	6	43
	Species	**Av.Abund**	**Av.Sim**	**Sim/SD**	**Contrib%**	**Cum.%**	**T%**	**F%**
Group 2 Average similarity: 40.73%	*Eunotia incisa*	12.27	18.2	1.4	44.67	44.67	41	91
	Frustulia saxonica	6.64	6.18	0.91	15.16	59.83	15	82
	Achnanthes oblongella	4.51	4.42	0.91	10.85	70.69	7	82
	Eunotia minor	5.34	4.08	0.69	10.02	80.7	11	64
	Tabellaria flocculosa	4.35	3.98	0.9	9.77	90.47	6	73
	Species	**Av.Abund**	**Av.Sim**	**Sim/SD**	**Contrib%**	**Cum.%**	**T%**	**F%**
Group 3 Average similarity: 34.28%	*Achnanthes oblongella*	15.26	23.68	3.96	69.09	69.09	52	100
	Eunotia minor	5.29	4.19	0.86	12.21	81.29	10	70
	Planothidium engelbrechtii	1.84	0.91	0.5	2.65	83.95	2	50
	Frustulia saxonica	1.83	0.87	0.46	2.53	86.48	2	50
	Fragilaria biceps	1.91	0.87	0.5	2.53	89.01	2	50
	Tabellaria flocculosa	1.21	0.76	0.58	2.21	91.22	1	60
	Species	**Av.Abund**	**Av.Sim**	**Sim/SD**	**Contrib%**	**Cum.%**	**T%**	**F%**
Group 4 Average similarity: 27.45%	*Gomphonema parvulum*	10.7	11.88	9.53	43.3	43.3	27	100
	Achnanthes oblongella	7.68	6.16	1.25	22.44	65.73	17	100
	Fragilaria biceps	4	2.76	1.2	10.05	75.78	5	100
	Eolimna minima	3.07	2.08	0.58	7.57	83.35	3	67
	Denticula subtilis	2.72	1.5	0.58	5.47	88.82	3	67
	Eunotia minor	2.45	1.3	0.58	4.74	93.56	2	67

A PCA ordination of water quality parameters showed a clear differentiation between winter and summer samples, especially with regards to temperature and oxygen. More interestingly the PCA ordination also showed a separation between different rivers within the ecoregion Figure 68).

Group one, as produced by the Cluster analysis, showed a lower conductivity, salinity and pH, when compared to sites that were in group three and four. Group two, which was situated between group one and group three and four in the Cluster diagram, also showed this pattern with water quality parameters. The water quality and species data seem to be following the same pattern, which suggests that species differentiation between groups is linked to differences in water quality characteristics.

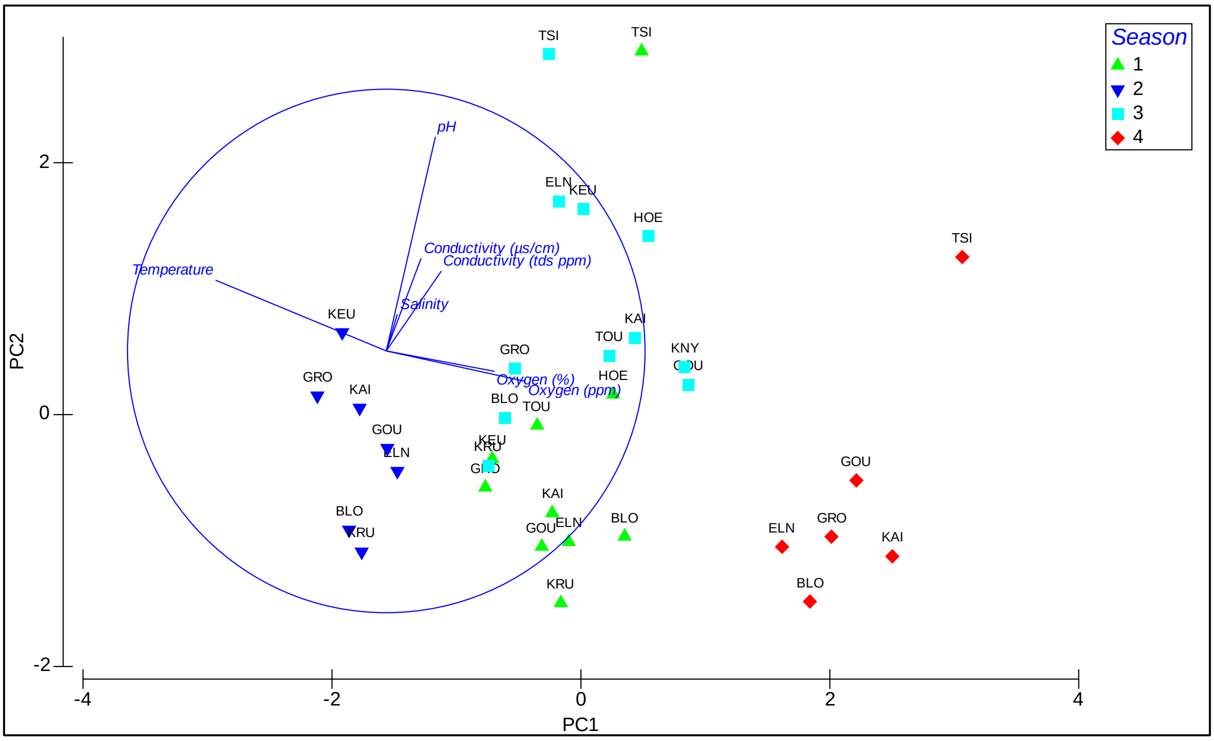

Figure 68 PCA ordination depicting the relationship between water quality parameters across samples in the South Eastern Coastal Belt ecoregion. Temporal changes (seasonal sampling) are depicted in the colouring as follows; green = spring, blue = summer, turquoise = autumn and red = winter.

In order to test this relationship, a BEST analysis was conducted. BEST analysis tests for relationships between biological and environmental data that could best explain the biological patterns. The analysis found that pH was the strongest water quality parameter associated with community composition in the South Eastern Coastal Belt ecoregion (Table 12). The global test sample statistic was Rho = 0.39.

Table 12 BEST results for South Eastern Coastal Belt ecoregion.

Variables	Correlation
pH	0.390
pH and Conductivity (µs/cm)	0.386
pH and Salinity	0.367
pH, Salinity and Conductivity (µs/cm)	0.367

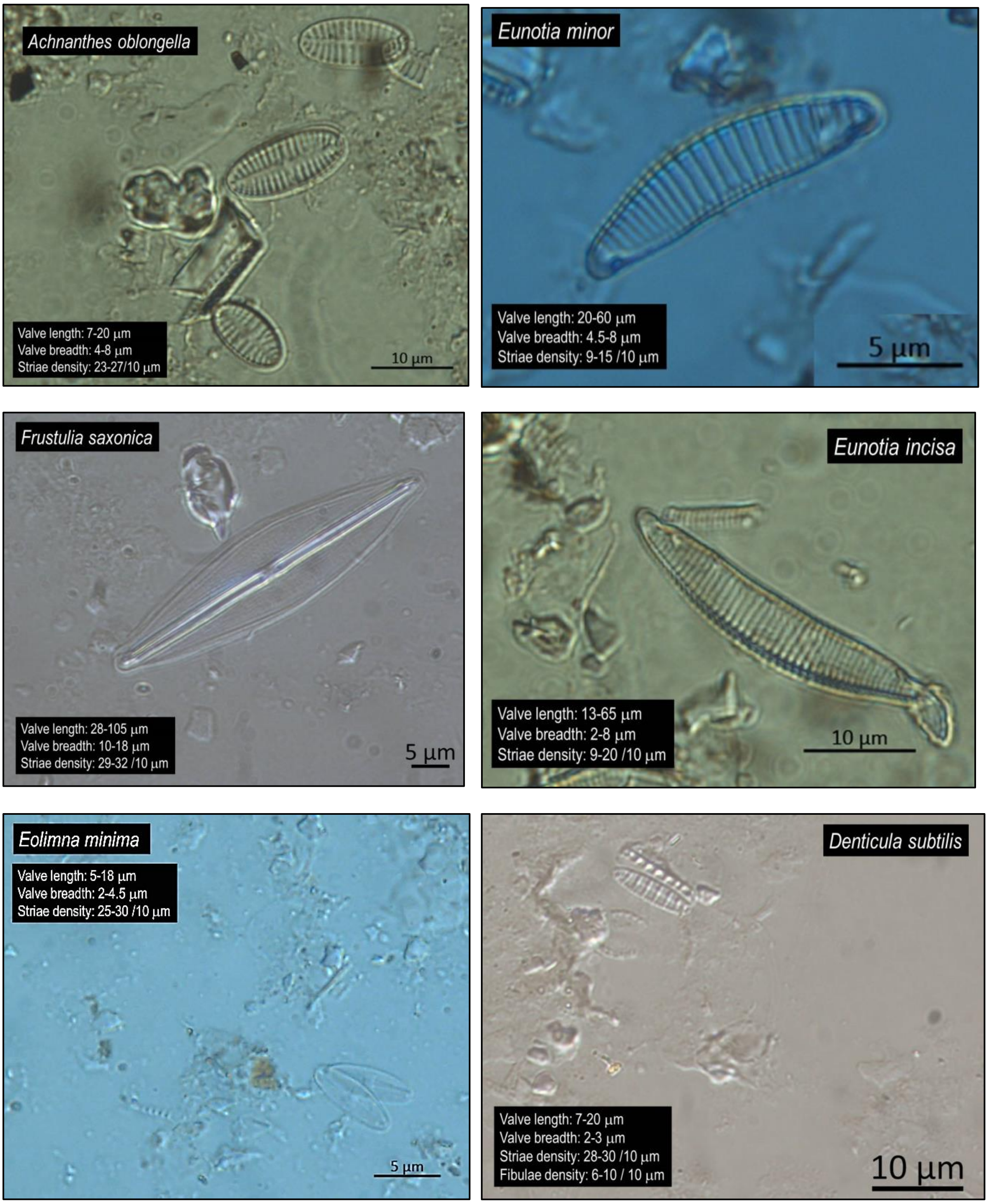

Figure 69 Species found to be important for diatom community composition in the South Eastern Coastal Belt ecoregion in South Africa.

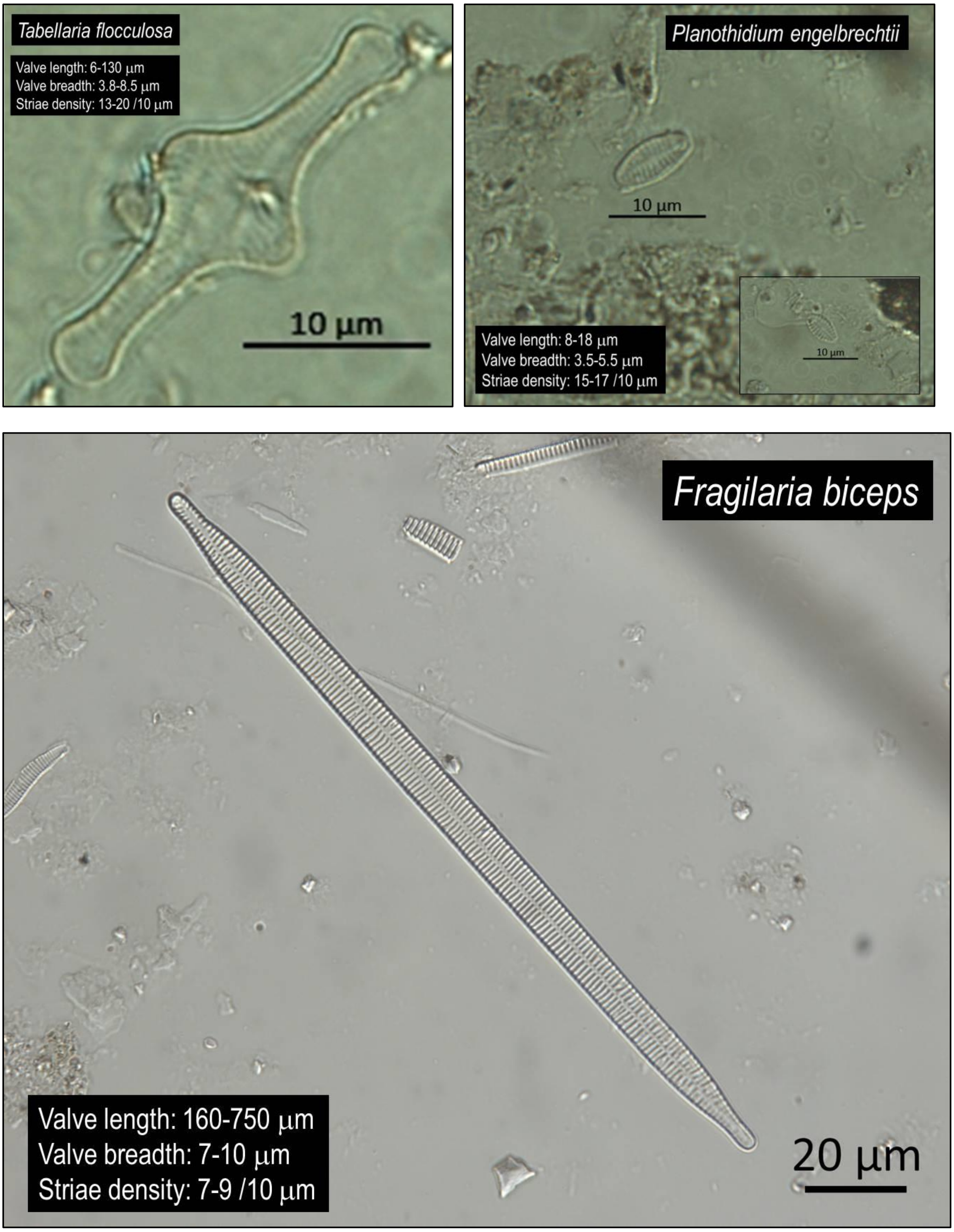

Figure 70 Species found to be important for diatom community composition in the South Eastern Coastal Belt ecoregion in South Africa.

Southern Folded Mountains (SFM)

Cluster analysis for the Southern Folded Mountain ecoregion produced three distinct groups, at a >20% similarity, as seen in Figure 71. There was no seasonal separation present and instead a spatial pattern is seen in the MDS ordination presented in Figure 72.

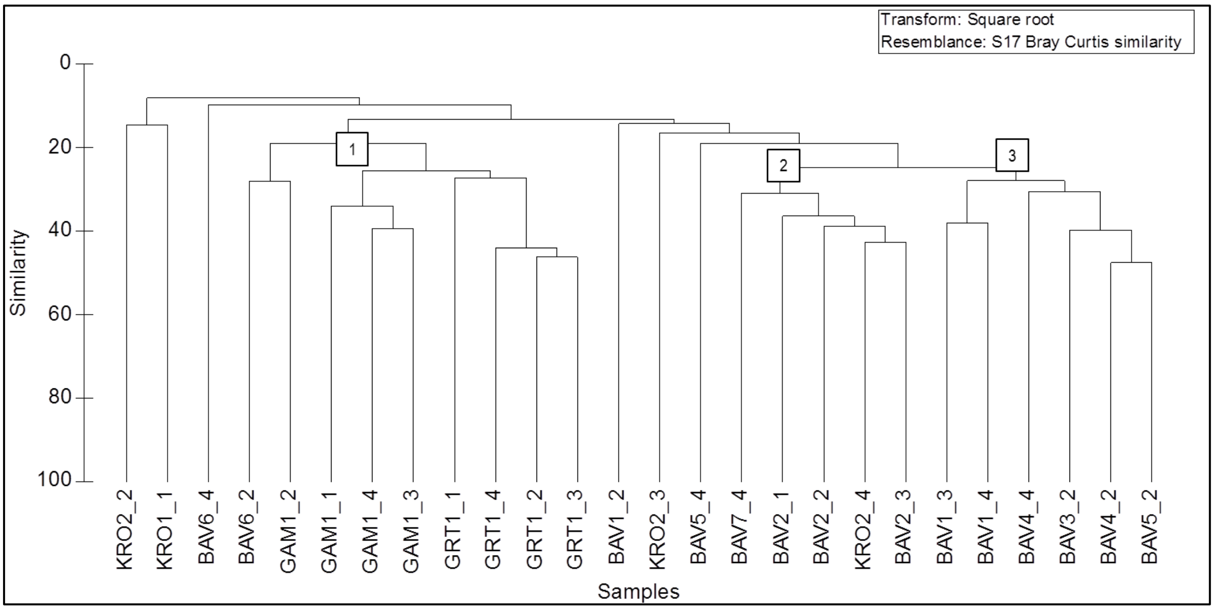

Figure 71 Cluster diagram of seasonal data for sites situated in the Southern Folded Mountains ecoregion.

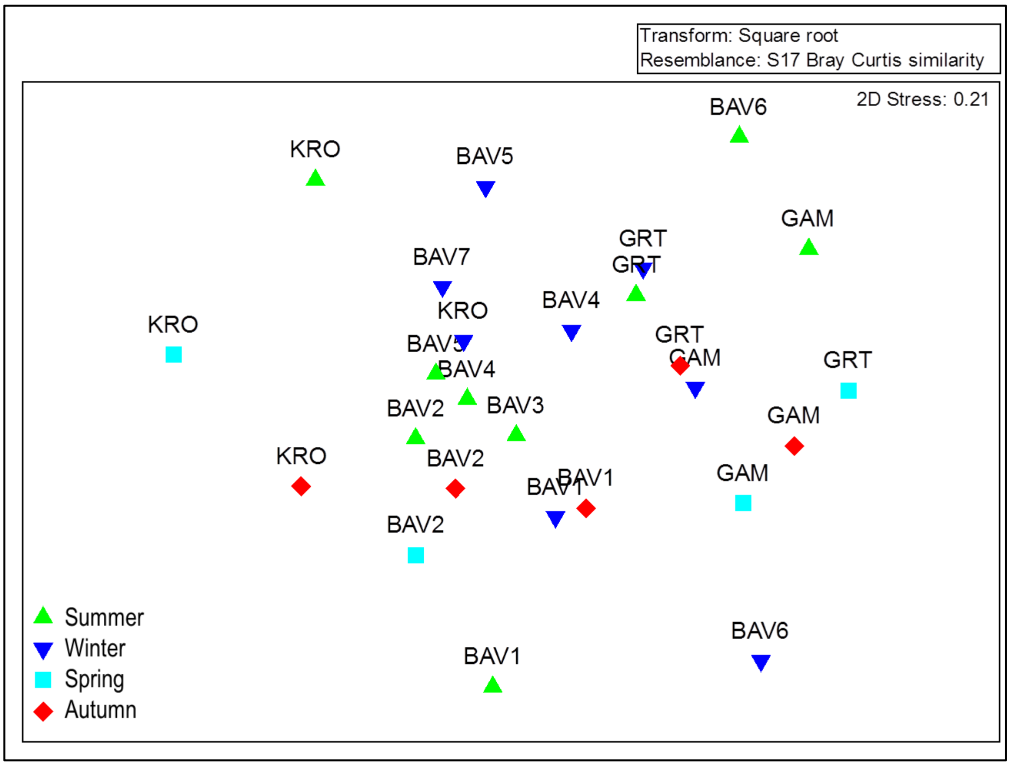

Figure 72 Non-metric MDS ordination of seasonal samples collected from rivers situated in the Southern Folded Mountains ecoregion.

Group one consisted of all the Gamtoos and Groot River sites with the addition of a Baviaanskloof site. **Group two** contained all the Wit River sites (BAV2) as well as a site on the Krom River and a very upstream site on the Baviaanskloof River. **Group three** consisted of most of the Bokkraal River sites (BAV1) and four Baviaanskloof River sites inside or close to the conservancy area. The Baviaanskloof area seemed to group separate from the rivers outside the kloof and conservancy area. To test the strength of these groupings an ANOSIM was run which produced a significant result with Global R value of 0.507. SIMPER analysis identified the species responsible for this grouping with their percentage contribution to the group identify. The data are summarised in Table 13. Photo plates of the important species for group identity within the SFM are presented in Figure 74-79 together with their valve dimensions and other information important for species identification.

For group one the prominent species responsible for group formation were; *Staurosira elliptica*, *Cocconeis placentula*, *Cyclotella meneghiniana* and *Melosira varians*. The group had an average similarity of 26%. Group two was found to be characterised by; *Fragilaria biceps*, *Nitzschia closterium*, *Achnanthes oblongella* and *Gomphonema venusta.* The group had an average similarity of 35%. In group three the average similarity was 32% and the prominent species responsible for group identity were; *Fragilaria biceps*, *Achnanthes oblongella*, *Achnanthidium minutissimum* and *Planothidium frequentissimum*.

A PCA ordination of water quality parameters showed a clear differentiation between winter and summer samples, especially with regards to temperature and oxygen (Figure 73). The PCA ordination also showed a separation between different rivers within the ecoregion. The majority of the Groot and Gamtoos River samples (Cluster group one) lie more to the right of the ordination which indicates a higher pH, salinity and conductivity measured at these sites, while at the same time most of the Baviaanskloof sites (Cluster groups two and three) lay more towards the left of the ordination which indicates that lower salinity, pH and conductivity was measured at time of sampling. The water quality and species data seem to be following the same pattern, which could suggest that species differentiation between these groups are linked to differences in water quality characteristics.

This relationship was tested using BEST analyses (Table 14). The analysis found that pH and salinity were the strongest water quality parameters associated with community composition in the Southern Folded Mountains ecoregion. Global test sample statistic was Rho = 0.189 which is much lower compared to the South Eastern Coastal Belt ecoregion.

Table 13 SIMPER and frequency of occurrence data for species responsible for group formation in the Southern Folded Mountains ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

	Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
Group 1 Average similarity: 25.78%	*Staurosira elliptica*	10.78	10.53	1.7	40.85	40.85	29	100
	Cocconeis placentula	4.78	2.22	0.54	8.62	49.47	10	56
	Cyclotella meneghiniana	2.26	1.71	1.08	6.62	56.08	2	78
	Melosira varians	2.47	1.41	0.69	5.46	61.54	2	67
	Cocconeis placentula var. *lineata*	3.8	1.25	0.37	4.85	66.4	8	44
	Fragilaria biceps	2.65	1.22	0.57	4.73	71.13	4	56
	Navicula radiosa	1.98	0.95	0.55	3.68	74.81	2	56
	Nitzschia filiformis	2.66	0.91	0.41	3.52	78.34	4	44
	Gomphonema parvulum	1.84	0.76	0.53	2.95	81.28	2	56
	Bacillaria paradoxa	1.36	0.72	0.6	2.81	84.09	1	56
	Nitzschia palea	1.79	0.48	0.38	1.86	85.95	2	44
	Epithemia sorex	0.89	0.34	0.41	1.31	87.26	0	44
	Epithemia adnata	1.15	0.27	0.3	1.04	88.3	1	33
	Tabularia fasciculata	1.17	0.25	0.3	0.98	89.28	1	33
	Thalassiosira weissflogi	0.69	0.24	0.3	0.92	90.21	0	33
Group2 Average similarity: 35.36%	Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
	Fragilaria biceps	13.38	17.49	9.62	49.46	49.46	38	100
	Nitzschia closterium	4.96	3.21	0.62	9.08	58.55	9	60
	Achnanthes oblongella	3.87	3.07	1.04	8.69	67.24	4	80
	Gomphonema venusta	3.84	2.29	0.62	6.47	73.71	5	60
	Eunotia minor	3.24	2.23	0.91	6.31	80.02	3	80
	Tabellaria flocculosa	2.31	1.62	0.95	4.57	84.59	2	80
	Nitzschia filiformis	2.75	1.41	0.6	4	88.59	3	60
	Achnanthidium eutrophilum	2.72	0.76	0.32	2.16	90.75	4	40
Group3 Average similarity: 32.06%	Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
	Fragilaria biceps	7.39	6.35	4.09	19.82	19.82	14	100
	Achnanthes oblongella	4.39	4.25	3.34	13.26	33.08	4	100
	Achnanthidium minutissimum	6.03	3.67	0.96	11.45	44.52	12	83
	Planothidium frequentissimum	3.72	2.93	1.25	9.13	53.66	4	83
	Cymbella aspera	2.48	1.94	1.17	6.05	59.71	2	83
	Gomphonema parvulum	3.97	1.79	0.73	5.57	65.28	6	67
	Cocconeis placentula	3.2	1.68	0.97	5.25	70.53	4	83
	Eunotia minor	2.96	1.49	0.74	4.66	75.19	3	67
	Diadesmis confervacea	3.36	1.4	0.48	4.37	79.56	5	50
	Achnanthes subaffinis	2.37	0.82	0.46	2.56	82.12	3	50
	Nitzschia closterium	1.76	0.77	0.48	2.42	84.53	1	50
	Navicula radiosa	1.53	0.77	0.7	2.39	86.93	1	67

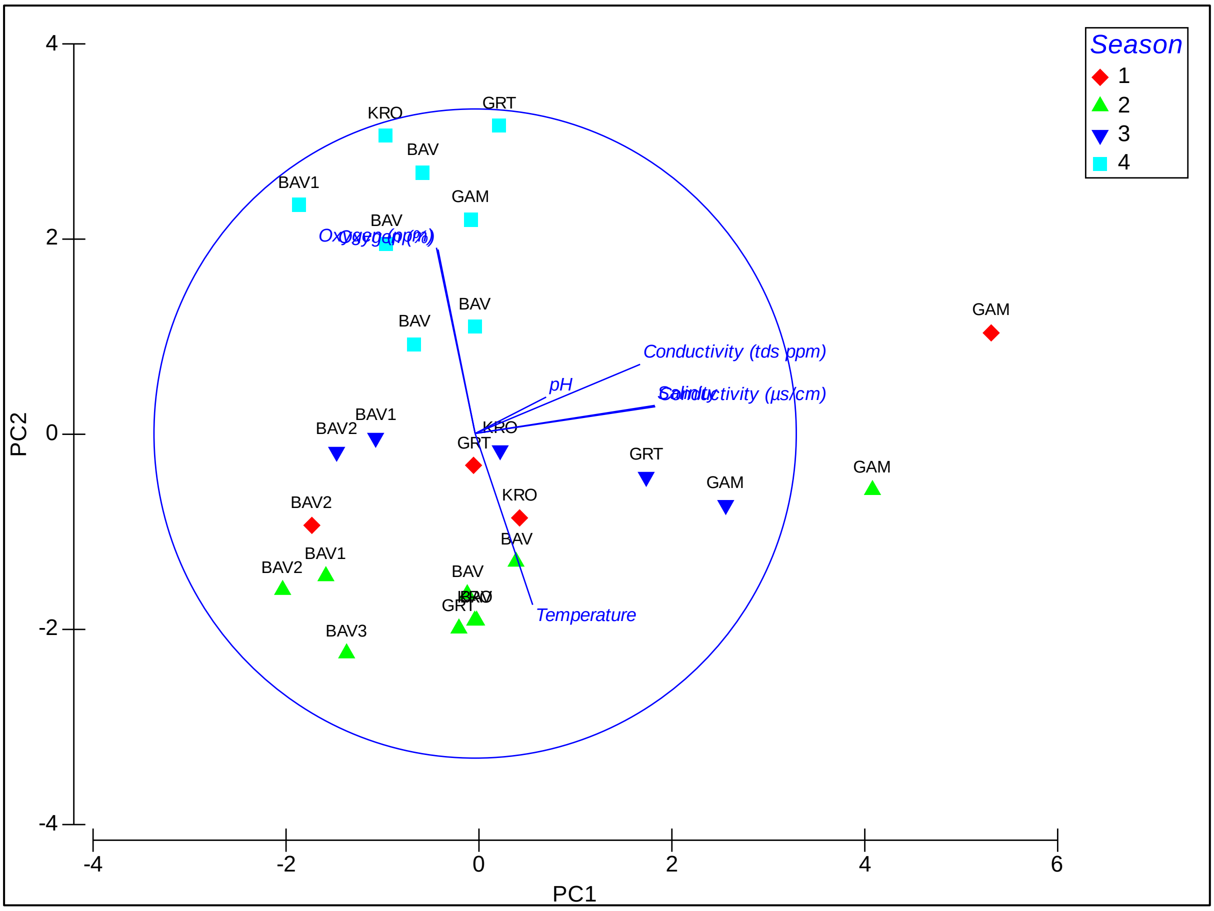

Figure 73 PCA ordination depicting the relationship between water quality parameters across samples in the Southern Folded Mountains ecoregion. Temporal changes (seasonal sampling) are depicted in the colouring as follows; red = spring, green = summer, blue = autumn and turquoise = winter.

Table 14 BEST results for Southern Folded Mountains ecoregion.

Variables	Correlation
Salinity and pH	0.189
pH and Conductivity (µs/cm)	0.188
pH, Salinity and Conductivity (µs/cm)	0.180
pH	0.162

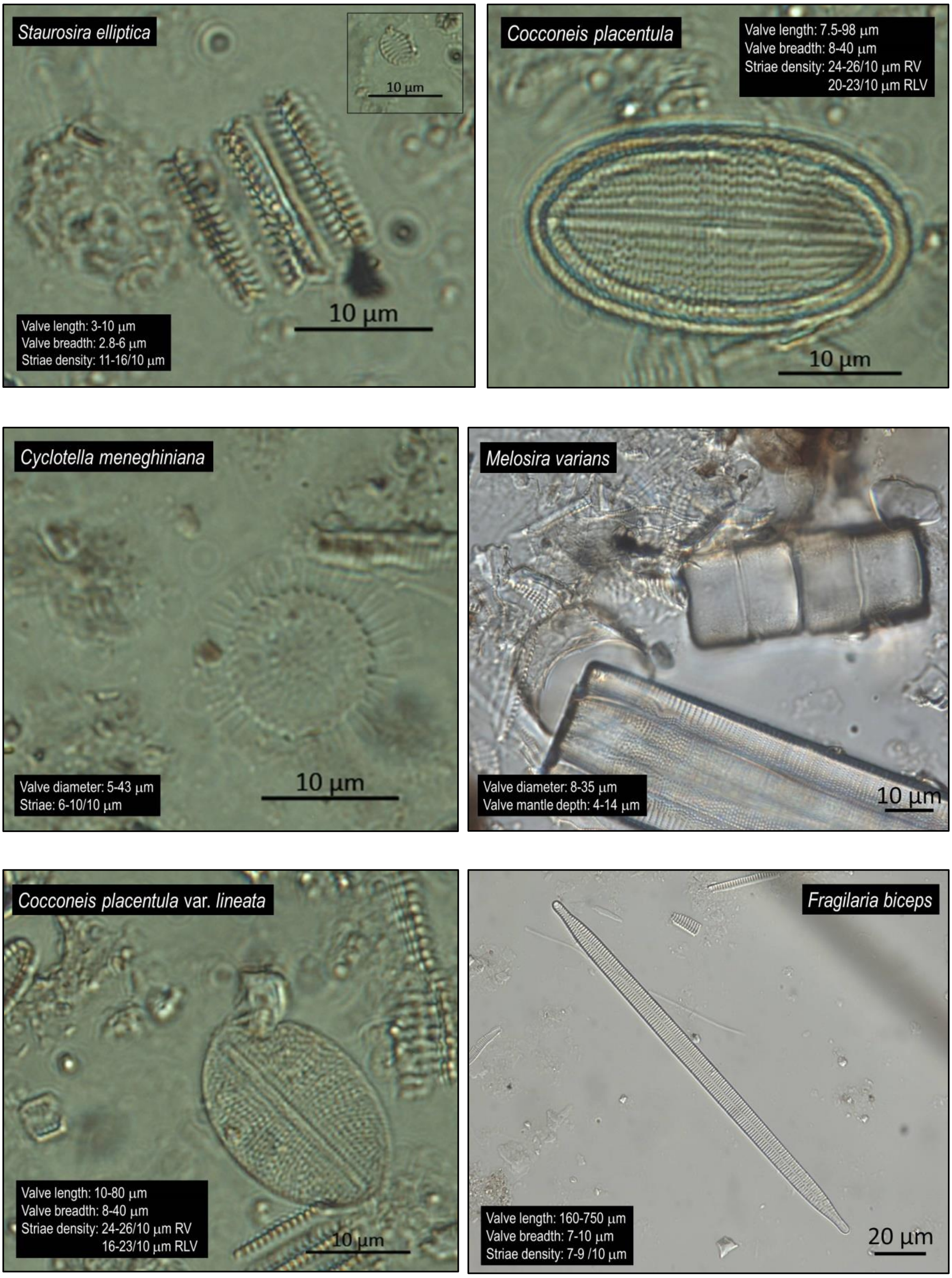

Figure 74 Species found to be important for diatom community composition in the South Southern Folded Mountains ecoregion in South Africa

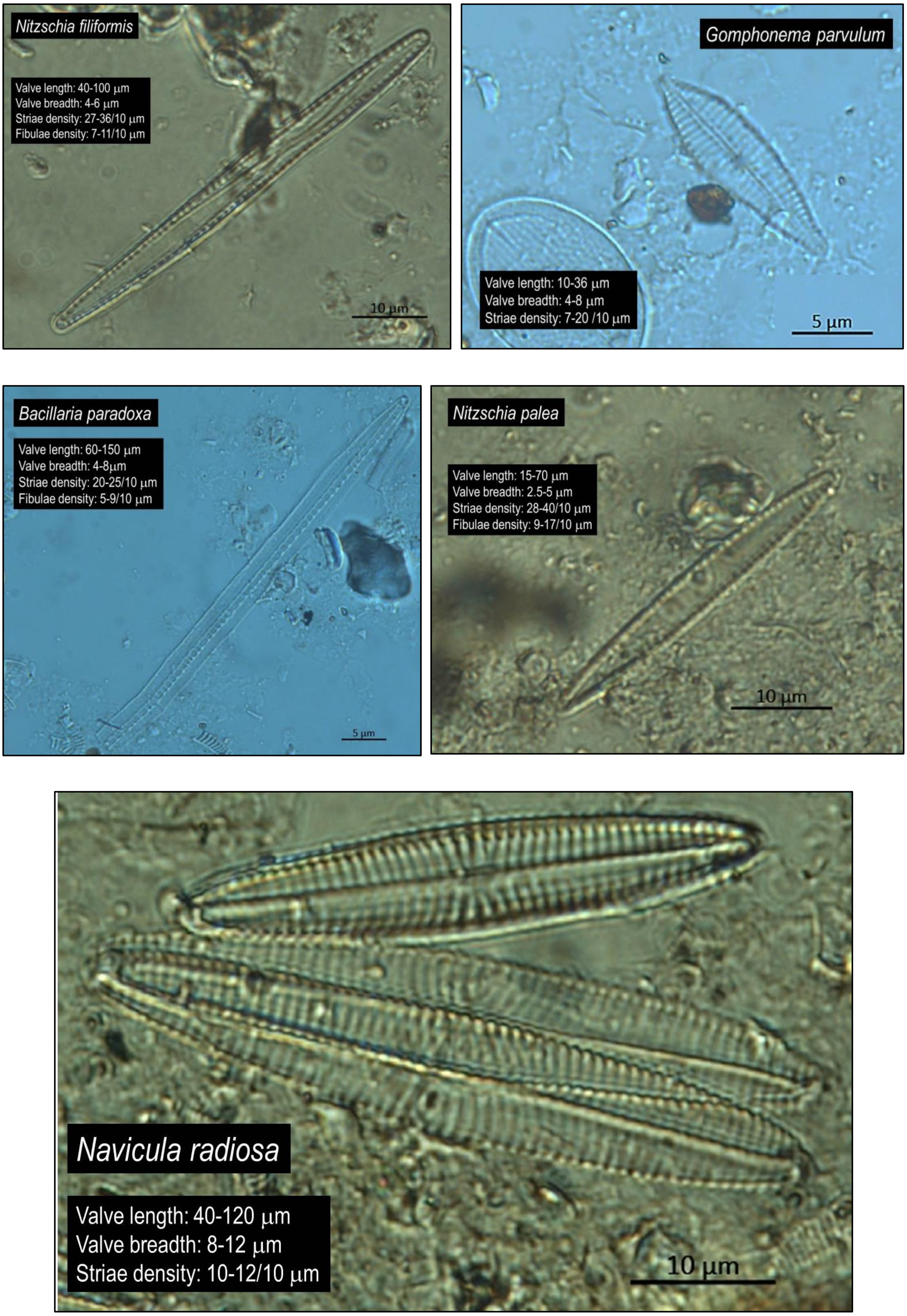

Figure 75 Species found to be important for diatom community composition in the South Southern Folded Mountains ecoregion in South Africa.

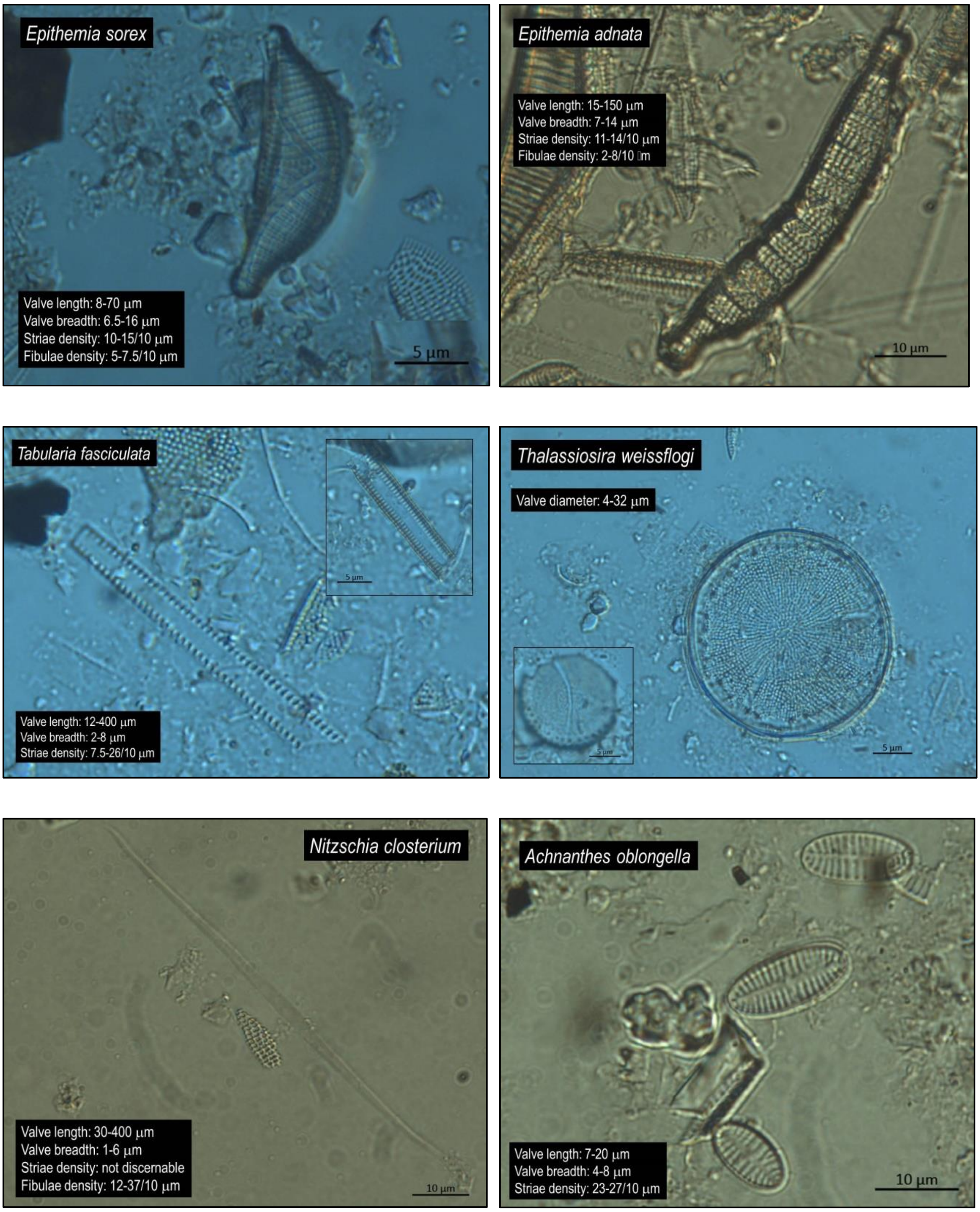

Figure 76 Species found to be important for diatom community composition in the South Southern Folded Mountains ecoregion in South Africa.

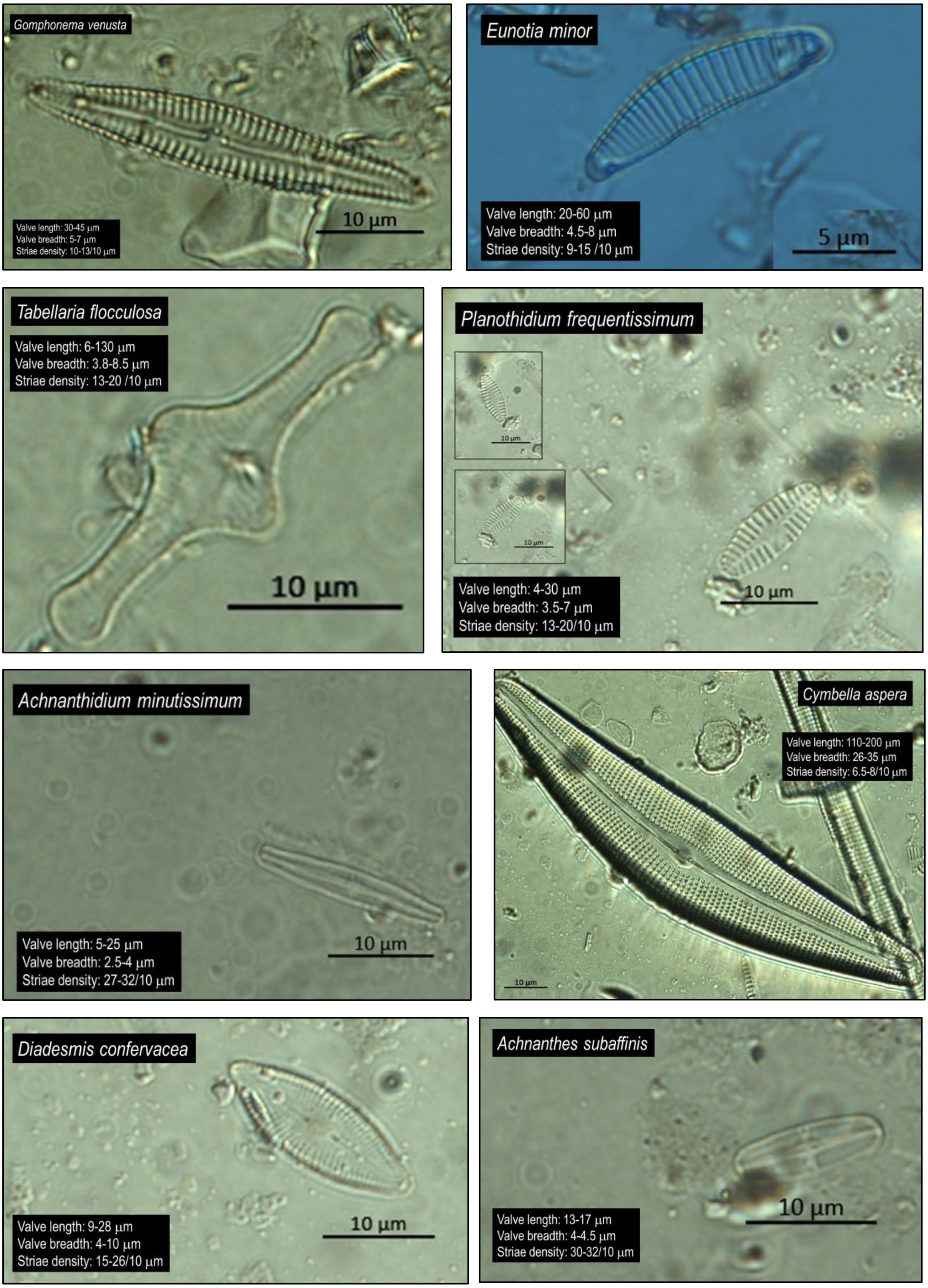

Figure 77 Species found to be important for diatom community composition in the South Southern Folded Mountains ecoregion in South Africa.

Figure 78 Species found to be important for diatom community composition in the South Southern Folded Mountains ecoregion in South Africa.

Drought Corridor (DC)

When all the samples in the Drought Corridor ecoregion were analysed, by means of a Cluster analysis, five groupings at >18% similarity were produced (Figure 79). **Group one** consisted of all the Tyume River sites, while **group two** contained one Volkers River sample and one Koonap River sample. **Group three** consisted solely of Small Fish River samples. **Group four** consisted of two Volkers River samples. **Group five** contained the majority of Koonap River sites, one Volkers River and one Great Fish River site each. This grouping makes sense, since the Tyume River is situated in the Amathole Mountains and in one of South Africa's Strategic Water Source Areas. This area receives a higher than usual volume of annual rainfall and these mountains have been associated with higher levels of biodiversity than the surrounding areas. It is therefore not too surprising that this site would separate from the rest of the Drought Corridor samples.

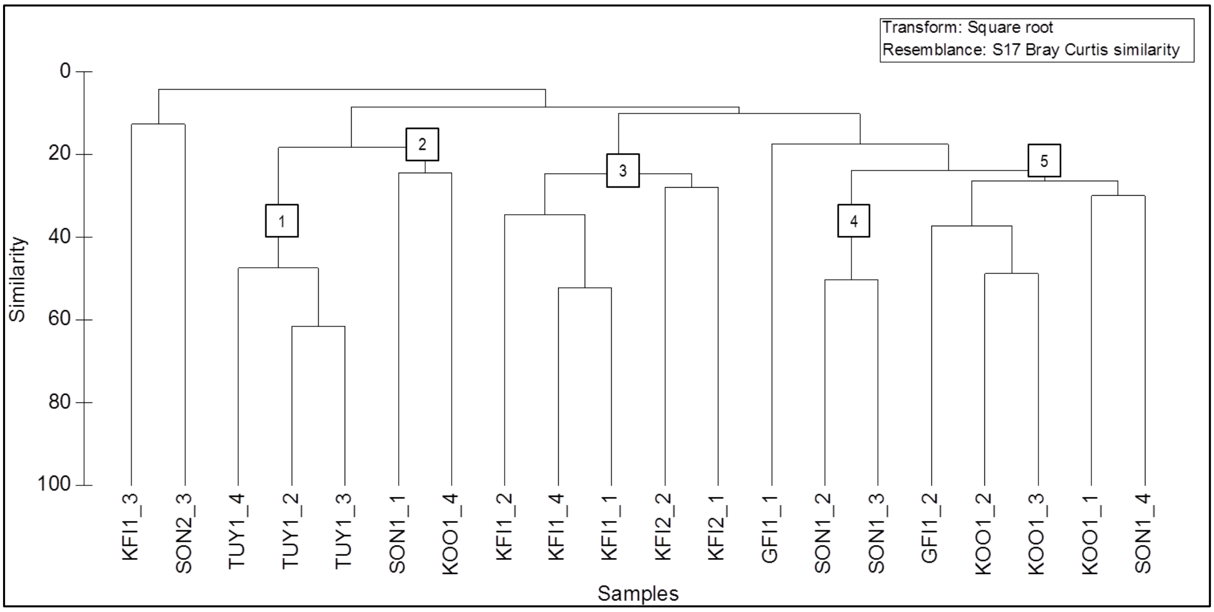

Figure 79 Cluster diagram of seasonal data for sites situated in the Drought Corridor ecoregion.

The Small Fish River separating from the Great Fish River is interesting, since the Small Fish River is a tributary of the Great Fish River, but the Great Fish River is also used as a transfer channel for water from the Orange River Basin. The majority of the Koonap River sites grouped together. Most of the Volkers and Great Fish River sites grouped closer to the Koonap River than the Small Fish River. This could be an indication that something other than the inter-basin transfer scheme is underpinning community composition. No clear seasonal pattern was discernible. Instead the pattern presented spatially with a clear separation between TUY and KFI sites from the rest (Figure 80).

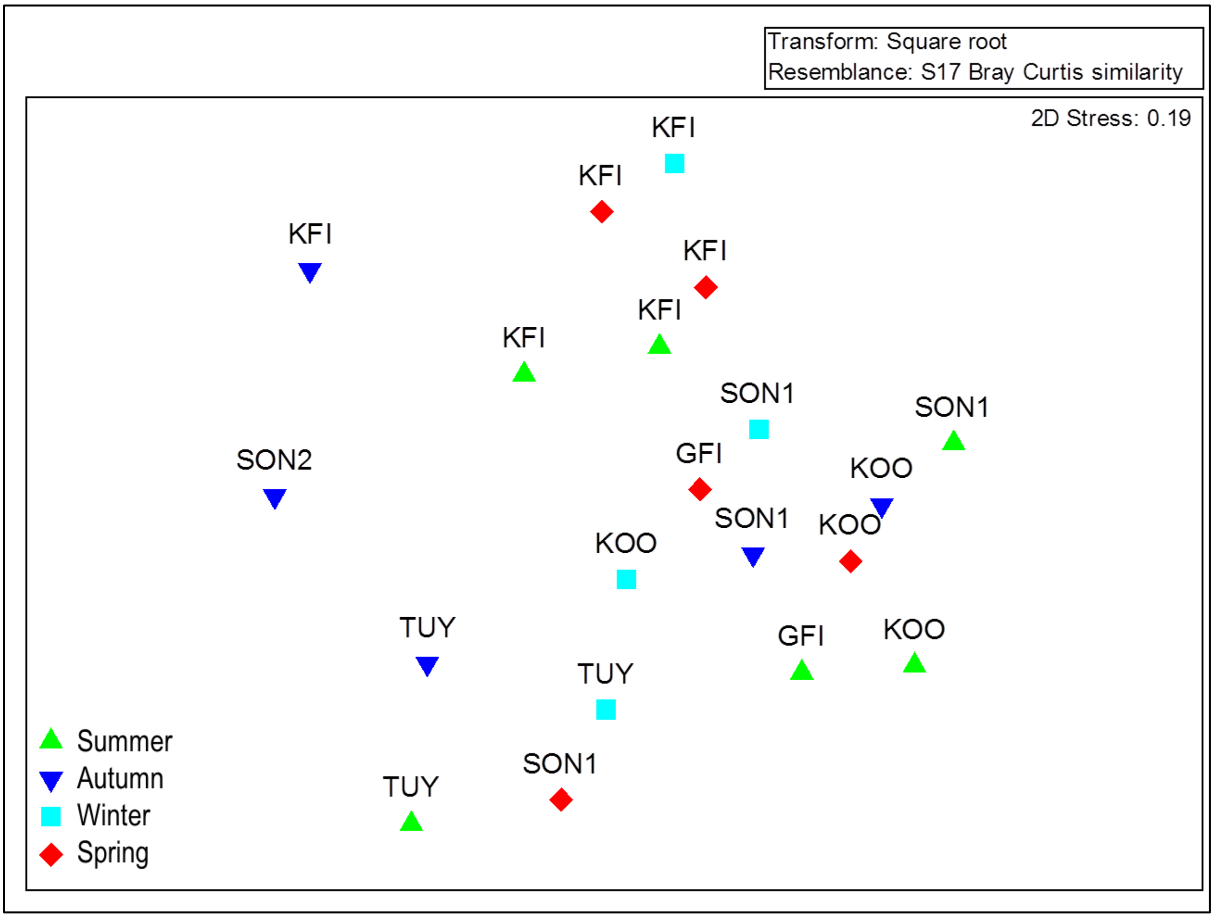

Figure 80 Non-metric MDS ordination of seasonal samples collected from rivers situated in the Drought Corridor ecoregion.

This pattern of spatial instead of temporal separation was tested using an ANOSIM which produced a very significant result with Global R value of 0.827. A SIMPER analysis identified the species responsible for these groupings, with their percentage contribution to the group identity. This data together with the frequency of occurrence for each of these species are summarised in Table 15 below. **Group one** had an average similarity of 52% with *Cocconeis placentula* var. *lineata*, *Achnanthes oblongella* and *Planothidium frequentissimum* identified as the species responsible for group identity. For **group two** the following species were identified as responsible for group identity; *Cocconeis placentula* var. *lineata*, *Aulacoseira granulata* and *Gomphonema parvulum*. This group had an average similarity of 24%. **Group three** was characterised by the following species; *Rhopalodia gibba*, *Epithemia sorex*, *Epithemia adnate*, *Nitzschia closterium* and some others as listed in Table 15. The average group similarity was 30%. **Group four** was found to have an average similarity of 50% and to be characterised by; *Nitzschia palea*, *Gomphonema parvulum*, *Reimeria uniseriata* and *Cocconeis placentula*. The average similarity of **group five** was found to be 31% and the community was characterised by *Reimeria uniseriata* and *Cocconeis placentula*. Photo plates of the important species for group identity within the DC are presented in Figure 82-85 together with their valve dimensions and other information important for species identification.

Table 15 SIMPER and frequency of occurrence data for species responsible for group formation in the Drought Corridor ecoregion. Average abundance (Av.Abund) per zone, Average similarity of species abundance (Av.sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%), Cumulative contribution (Cum%), Percentage relative abundance (T%), Percentage frequency of occurrence (F%).

Group	Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%	T%	F%
Group 1 Average similarity: 51.98%	*Cocconeis placentula* var. *lineata*	17.12	38.33	2.89	73.73	73.73	71	100
	Achnanthes oblongella	3.46	7.17	9.73	13.79	87.52	3	100
	Planothidium frequentissimum	4.37	4.71	0.58	9.06	96.58	7	67
Group 2 Average similarity: 24.27%	*Cocconeis placentula* var. *lineata*	7.53	10.19	0	41.98	41.98	13	100
	Aulacoseira granulata	6.21	7.35	0	30.29	72.27	9	100
	Gomphonema parvulum	4.03	4.65	0	19.16	91.43	4	100
Group 3 Average similarity: 29.58%	*Rhopalodia gibba*	10.36	10.84	1.88	36.65	36.65	26	100
	Epithemia sorex	6.62	6.14	2.67	20.75	57.4	11	100
	Epithemia adnata	6.61	4.83	1.22	16.31	73.71	15	100
	Nitzschia closterium	3.75	2.14	0.62	7.24	80.95	5	60
	Gomphonema parvulum	2.53	0.97	0.61	3.27	84.23	3	60
	Planothidium frequentissimum	1.12	0.5	0.62	1.68	85.9	1	60
	Rhopalodia gibberula	1.05	0.43	0.61	1.47	87.37	0	60
	Pleurosigma elongatum	1.55	0.42	0.32	1.42	88.79	1	40
	Navicula radiosa	2	0.4	0.32	1.36	90.15	2	40
Group 4 Average similarity: 50.13%	*Nitzschia palea*	13.06	18.82	0	37.54	37.54	43	100
	Gomphonema parvulum	7.58	14.65	0	29.23	66.77	13	100
	Reimeria uniseriata	5.29	8.89	0	17.74	84.51	7	100
	Cocconeis placentula	4.57	3.88	0	7.74	92.26	6	100
Group 5 Average similarity: 30.99%	*Reimeria uniseriata*	9.68	14.99	3.35	48.37	48.37	22	100
	Cocconeis placentula	10.75	13.58	1.52	43.81	92.18	31	100

A PCA ordination of water quality parameters showed a clear differentiation between winter and summer samples, especially with regards to temperature, oxygen and in this ecoregion also pH. The PCA ordination showed a separation between different rivers within the ecoregion (Figure 81). The Tyume River samples (Cluster group one) were concentrated to the right of the ordination, which indicates low conductivity and salinity. The Small Fish River samples were concentrated more towards the left which indicates a higher conductivity and salinity. The winter samples in this ecoregion had a higher pH associated with their respective water quality readings.

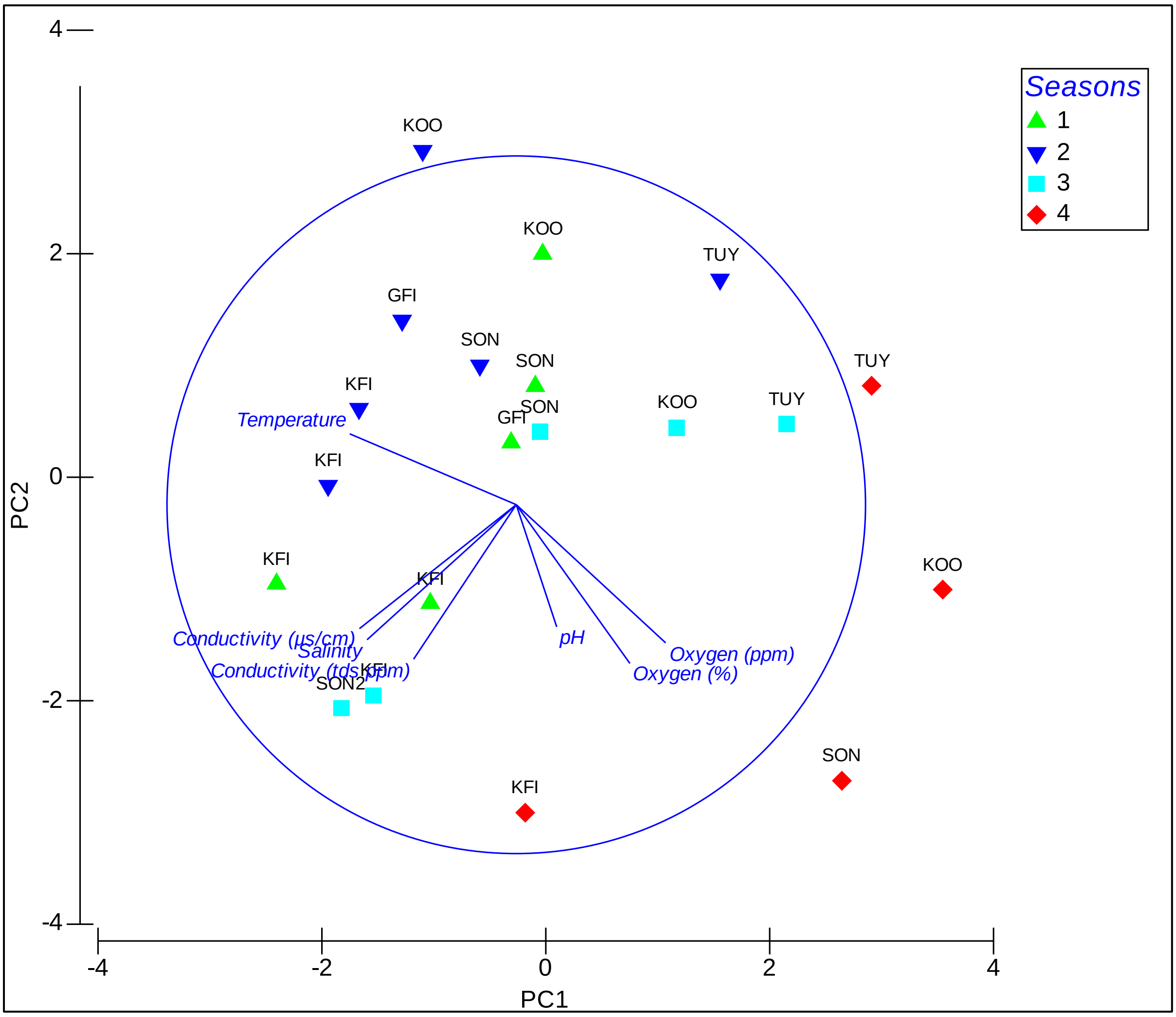

Figure 81 PCA ordination depicting the relationship between water quality parameters across samples in the Drought Corridor ecoregion. Temporal changes (seasonal sampling) are depicted in the colouring as follows; green = spring, blue = summer, turquoise = autumn and red = winter.

This relationship was tested using BEST analyses (Table 16). The analysis found that salinity was the strongest water quality parameter associated with community composition in the Drought Corridor ecoregion. Global test sample statistic was Rho = 0.49 which was the highest for all ecoregions sampled.

Table 16 BEST results for the Drought Corridor ecoregion

Variables	Correlation
Salinity	0.490
Salinity and Conductivity (µs/cm)	0.481
Conductivity (µs/cm)	0.476
Salinity, Conductivity (µs/cm) and Conductivity (tds ppm)	0.466

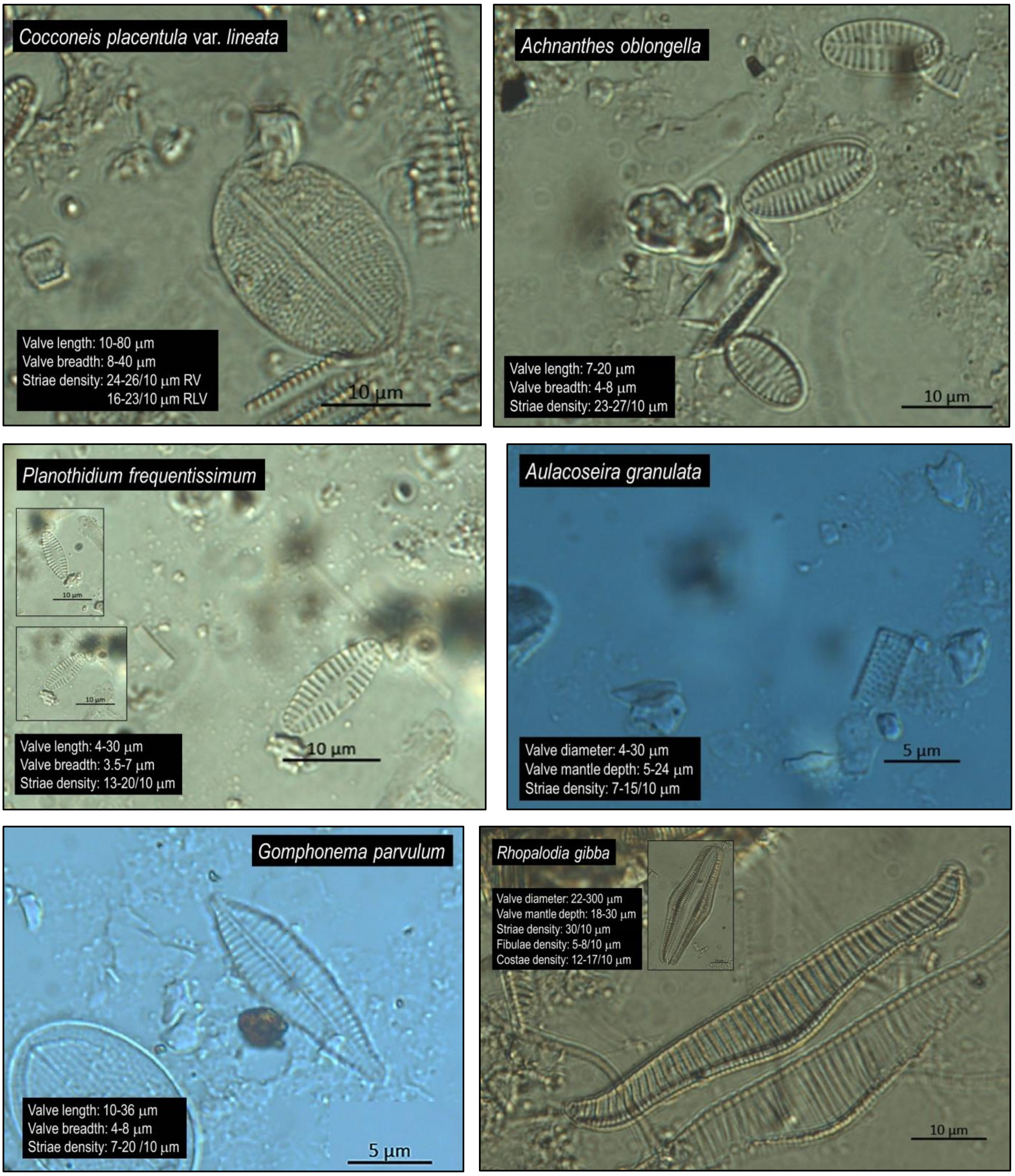

Figure 82 Species found to be important for diatom community composition in the South Drought Corridor ecoregion in South Africa.

Figure 83 Species found to be important for diatom community composition in the South Drought Corridor ecoregion in South Africa.

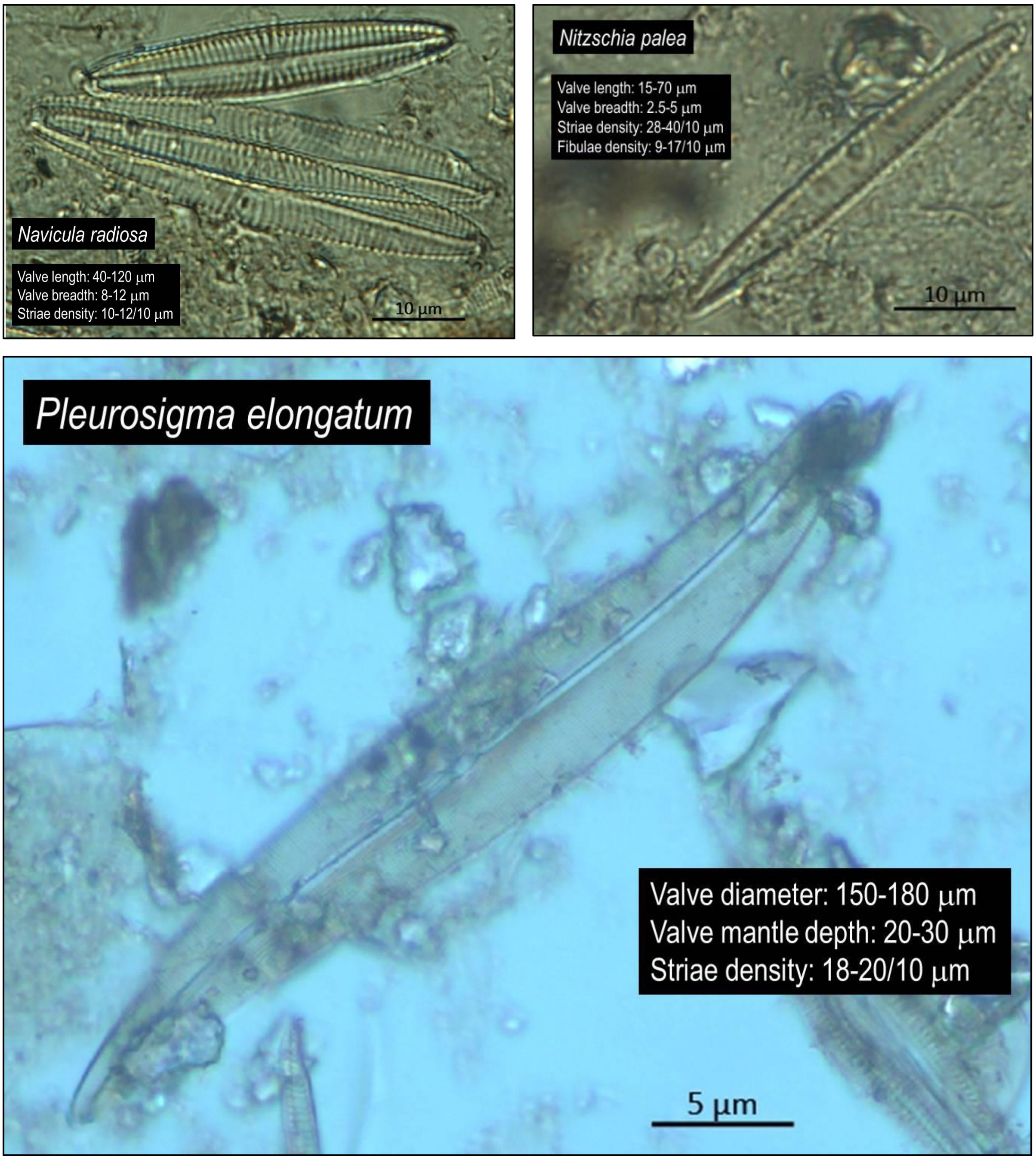

Figure 84 Species found to be important for diatom community composition in the South Drought Corridor ecoregion in South Africa.

Diatoms of the Okavango Panhandle

CHAPTER 5 Diatom community composition of the Okavango Panhandle, Botswana (draft paper prepared for AJAS)

Diatom community composition of the panhandle section of the Okavango River, Botswana

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Abstract

The project sought to document the diatoms of the panhandle section of the Okavango River. During 2014 diatoms were sampled from a variety of sites in the Okavango Panhandle. The diatom community showed little variation between different sites in the panhandle. There was, however, a significant difference between the community compositions of the Thamalakane River, Nxamaseri Floodplain and the Okavango Panhandle. These differences are suspected to result from micro-environments with differences in nutrient load and associated water quality. The species characterising the communities were; *Eunotia formica*, *Eunotia minor*, *Asterionella formosa* and *Fragilaria biceps*. The main physico-chemical drivers of community composition were salinity, temperature and pH. Micro-environments were found to exist within the larger lowland river habitat such as the panhandle, Okavango River. These micro-habitats allowed for some species, that are more tolerant to higher nutrient loads, to be found but did not interfere with the larger scale diatom community composition in a specific geographic region. The difference between diatom communities in the panhandle and other areas in the system highlights the importance of upstream conservation in the Okavango River for continued optimal ecological functioning of the downstream Okavango Delta and its associated systems.

Keywords: Algae, Conservation, Lowland River, Micro-environments, Physico-chemical drivers, Water quality

Introduction

Freshwater habitats are complex systems, especially rivers and streams where biophysical components change rapidly and could have great impacts on the functioning of ecosystems (Davies and Day 1998). Biodiversity is the most important component of well-functioning ecosystems and the sustainability of the associated ecosystem services and goods they deliver (Campbell et al. 2009). Many programs today are therefore aimed at effectively managing and conserving the biodiversity component of ecosystems and consequently its functioning and associated ecosystem services (Stoermer and Smol 1999, Campbell et al. 2009).

The Okavango Panhandle and Delta lie near the lowest part of the Kalahari Basin, which is a desert, the remains of an ancient ocean from 550-700 million years ago that stretches from the Northern Cape in South Africa, through most central parts of Botswana, through Angola and into the Democratic Republic of Congo (Haddon 2005). The lower reaches of the Okavango River lies in a semi-desert region. Rainfall upstream of Botswana is crucial for the sustainability of ecological functioning in the panhandle and delta river systems. Since the Okavango River is one of the biggest sources of freshwater in Botswana, it is a lifeline to many local people, animals and plants. The river provides food, building material, freshwater and many other services and goods to local villages, towns and cities.

The Okavango Delta (wetland section most downstream) has been listed as one of the most important wetlands in the world by Junk et al. (2006) and more recently it was declared a World Heritage site (UNESCO 2014). The lower portion of the Okavango River therefore received considerable attention and protection with little to no management and conservation efforts being made upstream in the panhandle section. The Okavango Delta and all its upstream components are of critical importance for securing people's livelihoods under increasing threats from climate change and increased resource demands in Namibia and Angola (Mendelsohn et al. 2010).

The water quality is exceptionally good with the highest conductivity reading (just below 200 μs/cm) being similar to that of bottled water (West et al. 2015). This, however, also means that there are not a lot of nutrients present in the inflowing water. Most of the potassium, nitrogen and phosphorus which do enter the panhandle are very quickly taken up by *Phragmites mauritianus* and *Cyperus papyrus* reeds which create micro-environments. The seasonal and occasionally inundated areas of the delta downstream rely on nutrients carried down the river and deposited in the sediment that accumulates over long periods. The biggest local contributors to nutrients are dust, faeces and micro-organisms (Mendelsohn et al. 2010). Microalgae influence the water chemistry (such as the pH) through photosynthesis and respiration (Mackay et al. 2012). Diatoms in particular are big contributors to nitrogen fixation globally and in many instances form the base of the aquatic ecosystem food web.

The Kavango Zambezi (KAZA) Trans-frontier Conservation Area is the largest in the world, spanning an area of 520 000 km^2, comparable to the size of France (Peace Parks Foundation 2017). While the park includes the Okavango Delta, the panhandle is excluded. National Geographic's Okavango Wilderness Project is one of many projects, which emphasize the importance of upstream conservation for downstream sustainability. Unfortunately, this project, as with many others, is either focused on the Namibian or Angolan sections of the river leaving the panhandle behind to be forgotten in Okavango River conservation efforts (Quammen 2017).

Diatoms are one of the most species-rich components of rivers and streams. They are important genetic resources and form a very large and important component of the biodiversity in these habitats. Diatoms have high sensitivity to physical, chemical and biological changes in the habitat, which together with their rapid lifecycles, mean their response to changes is also rapid (Taylor 2004). Junk et al. (2006) described the Okavango Delta as mesotrophic, which makes sense with the deposition of nutrients occurring downstream in slow flowing lowland regions of a river basin. However, due to the wide open water channels, it is expected that the papyrus beds create micro-environments forming important nutrient traps. A biodiversity comparison of seven globally important wetlands (Junk et al. 2006) found that a wetland's biodiversity represented the character of the region in which it occurred. The Okavango Wetland is therefore relatively low in diversity, nutrients and productivity as associated with the semi-arid region it occurs in. This has been documented by many studies, one of which is West et al. (2015) who showed that the water quality remained relatively unchanged from the panhandle right down to the bottom of the delta in Maun. This would suggest that diatom community composition, which is very closely associated with changes in water quality, would be homogenous across the panhandle and wetland section of the Okavango River.

Material and Methods

During July and August 2014 data were collected from the lowland section of the Okavango River in Botswana. This was done in order to assess the diatom communities associated with a low nutrient lowland riverine habitat situated immediately upstream of an important World Heritage site, the Okavango Delta. In Southern Africa, most lowland rivers are impacted greatly by degradation caused by anthropogenic activities such as urbanization, agricultural land-use and commercial development making natural ecological assessments difficult. The Okavango River, which is not significantly impacted, was therefore a very suitable location to assess the diatom community composition of a lowland river, especially in the context of conservation status and continued resilience of the downstream components of the river. This study was therefore aimed at documenting and assessing the diatom communities from the panhandle section of the Okavango River to establish a reference condition and highlight the importance of conservation of diversity upstream of an aquatic system as sensitive and important as a World Heritage site.

Study area

The Okavango River is the main artery of water supply for Botswana in Southern Africa. Water originates in the highlands of Angola 1200 meters above sea-level and runs down towards Namibia before entering Botswana in the north-west at Mohembo. Two main tributaries join close to the border between Angola and Namibia, the Cuito and Cubango Rivers, to form the Okavango River (also known as the Kavango River in Namibia). In the most north-western parts of the Okavango River basin average annual rainfall is 1300 millimeters. This declines at a steady rate as the river moves south and east to where the annual average rainfall is only 450 millimeters (Mendelsohn et al. 2010).

Data collection was done from eight different sites with ten different samples being processed (Table 1). Sites varied from faster flowing side channels at Askiesbos to the slower flowing Samochima Lagoon near the Krokovango farm, 35 km from Shakawe. In addition to the panhandle, a fossil river was added to the sample diversity. The Nxamaseri Floodplain use to be a large tributary of the Okavango River, today this floodplain is seasonally inundated with water pushing in from the panhandle downstream when water levels are high. When the water level starts going down again in the panhandle the fossil river bed is an oasis of wildlife scattered between pools of varying color as the microscopic life scrambles for quick successional generations before the water disappears completely. Downstream of the delta, the Thamalakane River was sampled at Maun.

Table 1: Summary of study sites sampled during 2014. Site code, location, type of sample and river section the site was situated in.

River	Section	Area	Lat	Long	Site code	Sample type
Okavango	Panhandle	Samochima Lagoon	18° 25.626'S	21° 54.182'E	LAG3	Introduced
			18° 25.714'S	21° 53.741'E	LAG15	Introduced
			18° 25.714'S	21° 53.741'E	LAG21	Phytoplankton
			18° 25.695'S	21° 53.660'E	ASK3	Introduced
		Man-made Wetland	18° 26.004'S	21° 53.607'E	KRO1	Introduced
		Main Channel	18° 24.827'S	22° 0.583'E	NGA1	Phytoplankton
		Nxamaseri Floodplain	18° 35.957'S	22° 1.512'E	NXA2	Epiphyton
			18° 35.957'S	22° 1.512'E	NXA2_1	Phytoplankton
			18° 35.403'S	22° 0.788'E	NXA4	Epiphyton and phytoplankton
	Delta	Thamalakane River	19° 57.910'S	23° 27.849'E	KRA1	Epiphyton

Data collection

This study considers four different substrates diatoms occur in or on. They are; epipelon (surface of sediments), epipsammon (between sand particles), epilithon (gravel, stone and bedrock) and epiphyton (on macrophytic plants). Planktonic diatoms (potamoplankton) are free-living in the water column of slow flowing rivers and dams. This study used two methods to collect data, one was by means of introduced substrate (epilithon) and the second was with a plankton net (phytoplankton). Site set-up was conducted differently for each type of sample collected. Due to the lack of larger substrate in the lower reaches of the river, a plankton net with a mesh size of 25 µm was used to collect phytoplankton from the slow flowing open water areas. In order to sample the attached diatom species, epiphyton was sampled directly from the macrophytic plants or by means of introduced substrates. Introduced substrates consisted of petri dishes that were attached to the macrophytes. These dishes were left to be colonised by the diatoms for a period of 4-6 weeks. After this time, the dishes were removed and cleaned with a toothbrush, according to the prescribed method by Taylor et al. (2007a) for scrubbing stones.

All samples were fixed in glass McCartney specimen bottles and topped up with ethanol (70%) to produce a fixed sample of > 20% ethanol end product. Physico-chemical parameters were sampled with a handheld water meter (HANNA HI9828) to measure the temperature, salinity, pH, conductivity and dissolved oxygen in the water on the day of sampling.

Data Analysis

Identification of diatom species was done using a Nikon compound microscope, with phase contrast optics (1000x magnification), in an optical field as the defined area and successively moving to adjacent fields, while taking care not to count the same valves twice. The guide to common diatom species of South Africa (Taylor et al. 2007b) was used for taxonomic identifications as well as online resources if verification was needed. Community composition for each site was obtained by counting at least 400 individuals on a slide to obtain an accurate community representation (Schoeman 1979, Prygiel et al. 2002 and Taylor et al. 2007a). Broken valves were also included in the count when more than 50% of the valve was intact and enabling positive identification. When a valve was in girdle view and it was possible to positively identify the species, it was included into the count. Data were entered into Microsoft Excel sheets and then processed using PRIMER 6 statistics software (V6, Clarke and Gorley 2006). Multivariate statistics in PRIMER 6 was used for community scale analyses.

Results

In total 4486 individuals were identified across 10 sites. In total 69 species were identified from these samples and 16 individuals were only identified up to genus level (Table 2).

Table 2: Species list for individuals identified from sites sampled in Botswana during 2014. ASK – Askiesbos Channel, KRA – Thamalakane River, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain. Letter x – species present at site, abundance 1 - 5%, asterisc (*) – abundance > 5% and hyphen (-) – abundance < 1%.

	NGA1	KRO1	ASK3	KRA1	LAG3	LAG15	LAG21	NXA4	NXA2	NXA2_1
Achnanthes oblongella							-			
Achnanthidium eutrophilum		x								
Achnanthidium exiguum						-	x			
Achnanthidium macrocephalum							x*			
Achnanthidium minutissimum	x		x*							
Asterionella formosa	x*	x	x*	x*	x	x*	x*		x	
Aulacoseira ambigua								x*		x
Caloneis bacillum sensu lato										-
Caloneis sp1								-		
Craticula acidoclinata				-						
Craticula cuspidata								x		
Craticula halophila									x	
Craticula molestiformis					-					
Craticula vixnegligenda							-			
Cyclotella meneghiniana		-	x		-					
Diadesmis confervacea				x*						
Encyonema mesianum	-	x*	x	x	x					
Encyonema minutum						x		x		-
Encyonema neogracile	-									
Eunotia bilunaris			-							-
Eunotia flexuosa	-	-	x	-	-	x				x
Eunotia formica	x*		x*		x*	x*	x*	x*	x*	x
Eunotia minor	x*	x*	x*	-	x*	x*	x*		-	-
Eunotia pectinalis var. *undulata*	x*		x	x	x	x	x	x		
Eunotia rhomboidea			-		x*					
Fragilaria biceps	x*		x		x*	x*	x*		x*	
Fragilaria capucina								x		
Fragilaria nanana										x*
Fragilaria ulna var. *acus*				x*						
Frustulia saxonica	-	x	x*		x	-	x			
Gomphonema aff. *gracile*	x*			x*				x*	x*	x*

Table 2: Continued: Species list for individuals identified from sites sampled in Botswana during 2014 continued.

Gomphonema affine		x*	x*				x*	x*	x*	x*
Gomphonema contraturris		x	x		x	x				
Gomphonema globiferum	-								-	
Gomphonema gracile					x*	x*		x*		
Gomphonema insigne		x*								
Gomphonema parvulum		x*	x*	x	x*	x*	-		x*	-
Luticola goeppertiana	x									
Mayamaea atomus					-					
Melosira varians						x	x			
Navicula radiosa		x					-			
Navicula recens				x						
Navicula sp1						-				
Nedium sp1							-			
Nitzschia acicularis				x*		x				
Nitzschia amphibia				x*		x		x*	x	-
Nitzschia closterium	x*	-		x						
Nitzschia filiformis				-						
Nitzschia sp1							-			
Nitzschia umbonata		-		x						
Pinnularia divergens				-						
Pinnularia gibba								-	x	
Pinnularia subbrevistriata		x								
Pinnularia subcapitata									-	
Pinnularia viridiformis								x*		
Pinnularia viridis							-			
Placoneis clementis		-	-							
Placoneis placentula	-									
Pseudostaurosira brevistriata	x									-
Sellophora pupula						-	x	x	-	
Sellophora seminulum				-						
Sellophora stroemii									x*	
Stauroneis anceps						-			-	-
Stauroneis phoenicenteron				-						
Stauroneis sp1								x		
Staurosira construens								x*		-
Staurosira elliptica			-			x*	x*			x*
Staurosirella pinnata						x*				
Tabularia fasciculata		x	x	x	x*	x*	x*	x*	x*	x*

A Cluster analysis was conducted on all sample data. The result is presented below in Figure 1. From the diagram it is clear that some community differences existed between the panhandle section of the Okavango River, the Thamalakane River downstream and the seasonal Nxamaseri Floodplain. KRA1 (Thamalakane River) separated from all the other samples collected.

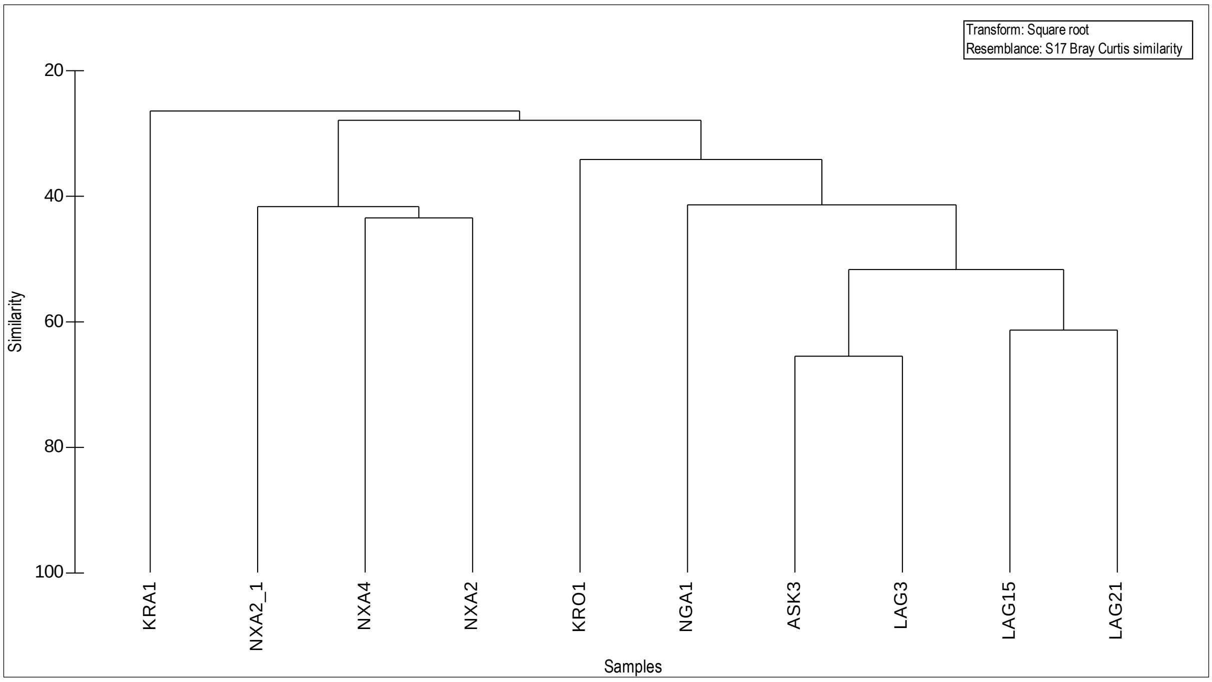

Figure 1 Cluster diagram of sampled sites. ASK – Askiesbos Channel, KRA – Thamalakane River, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

The NXA2 and NXA4 sites grouped together with more than 40% similarity between sites. The same was true for the panhandle sites with the exception of KRO1 (Krokovango Wetland). A non-metric MDS ordination confirmed these results as presented in Figure 2. Here the different geographic regions have been included as shaded factors to show the clear differentiation geographically of sampled sites.

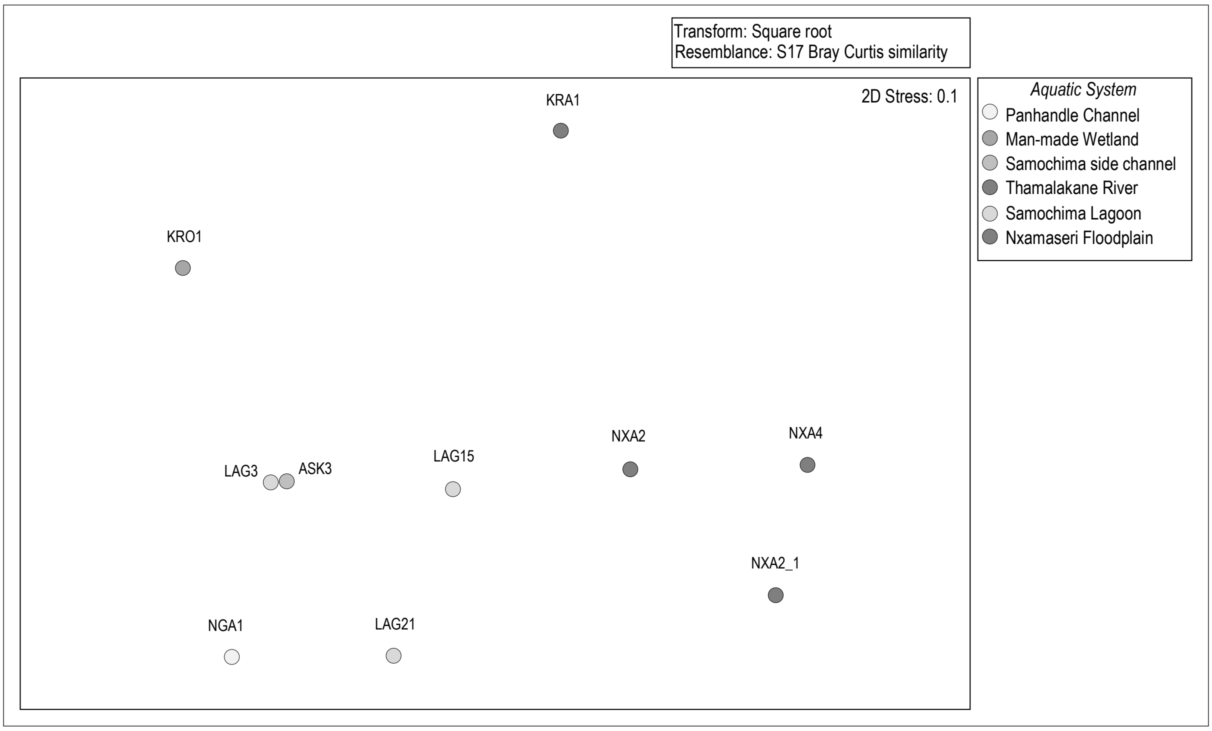

Figure 2: Non-metric MDS ordination of Bray-Curtis similarity between sites. ASK – Askiesbos Channel, KRA – Thamalakane River, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

The results obtained were also confirmed by an ANOSIM test. The sample statistic or Global R value was 0.812, which confirms that a strong separation was found between the factors as applied based on the non-metric MDS and Cluster result. SIMPER was used to identify the species responsible for community composition for the Okavango Panhandle and Nxamaseri Floodplain sites (Thamalakane River only has one site that cannot be used to produce statistically significant results here). Table 3 below presents the results for the panhandle sites. Sites within this section of the Okavango River showed an average of 44.70% similarity. It was found that *Eunotia minor*, *Eunotia formica*, *Asterionella formosa* and *Fragilaria biceps* were the species most responsible for community identity with a cumulative 52.28% similarity between samples of the Okavango Panhandle. Apart from these *Gomphonema parvulum*, *Tabularia fasciculata*, *Frustulia saxonica* and *Eunotia pectinalis* var. *undulata* were found to each contribute more that 5% to community composition.

Table 3 SIMPER data for sites situated in the panhandle section of the Okavango River, Botswana. Average abundance (Av.Abund), Average similarity of species abundance (Av.Sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%) and Cumulative contribution (Cum%).

Panhandle of Okavango River	Average similarity within group: 44.70%				
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Eunotia minor	7.37	7.81	9.33	17.46	17.46
Eunotia formica	6.47	6.26	1.35	14.00	31.46
Asterionella formosa	4.95	5.33	3.62	11.93	43.39
Fragilaria biceps	4.52	3.97	1.22	8.89	52.28
Gomphonema parvulum	5.71	3.43	0.91	7.68	59.96
Tabularia fasciculata	3.81	3.27	1.24	7.32	67.28
Eunotia pectinalis var. *undulata*	4.64	3.21	1.35	7.17	74.45
Frustulia saxonica	3.27	2.94	3.50	6.57	81.02
Gomphonema affine	3.52	1.49	0.48	3.33	84.35
Encyonema mesianum	2.58	1.48	0.67	3.31	87.67

The SIMPER results for the Nxamaseri Floodplain sites are presented in Table 4. The sites situated on the Nxamaseri Floodplain showed a 42.31% average similarity. The species responsible for community identity were found to be; *Tabularia fasciculata*, *Gomphonema* aff. *gracile* and *Gomphonema affine*. These species accounted for 63.11% of the community identity on these sites. Apart from these *Eunotia formica* and *Nitzschia amphibia* were each found to contribute more than 5% to community composition. The average dissimilarity between these groups was found to be 73.61%.

SIMPER results for the dissimilarity comparison between the panhandle and Nxamaseri Floodplain is presented in Table 5. The species that were found to be responsible for the dissimilarity between these two sampled regions were; *Fragilaria ulna* var. *acus*, *Gomphonema* aff. *gracile*, *Eunotia formica*, *Eunotia minor*, *Diadesmis confervacea*, *Navicula recens*, *Nitzschia amphibia*, *Fragilaria biceps* and *Nitzschia acicularis* accounting for

50,17% of the total dissimilarity. *Gomphonema* aff. *gracile*, *Eunotia minor*, *Nitzschia amphibia* and *Nitzschia acicularis* occurred in both regions, but with a considerable difference in abundance. The rest of the species had low contribution with percentages below 5%.

Table 4 SIMPER data for sites situated in the Nxamaseri Floodplain, Botswana. Average abundance (Av.Abund), Average similarity of species abundance (Av.Sim), Similarity to standard deviation ratio (Sim/SD), Contribution % to the similarity (Contrib%) and Cumulative contribution (Cum%).

Nxamaseri Floodplain	Average similarity within group: 42.31				
Species	Av.Abund	Av.Sim	Sim/SD	Contrib%	Cum.%
Tabularia fasciculata	9.31	11.18	14.41	26.42	26.42
Gomphonema aff. *gracile*	6.69	8.32	7.73	19.67	46.09
Gomphonema affine	5.60	7.20	19.75	17.01	63.11
Eunotia formica	4.45	4.62	3.22	10.91	74.01
Nitzschia amphibia	3.22	2.89	1.92	6.84	80.85
Aulacoseira ambigua	3.59	2.03	0.58	4.79	85.64
Gomphonema parvulum	2.95	0.98	0.58	2.32	87.96
Encyonema minutum	1.72	0.93	0.58	2.20	90.16

Table 5 SIMPER data for average dissimilarity between the panhandle section of the Okavango River and the Nxamaseri Floodplain, Botswana. Average abundance (Av.Abund), Average similarity of species abundance (Av.Diss), Similarity to standard deviation ratio (Diss/SD), Contribution % to the similarity (Contrib%) and Cumulative contribution (Cum%).

Dissimilarity	Panhandle	Nxamaseri	Average dissimilarity: 73.61%			
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Fragilaria ulna var. *acus*	0.00	13.27	8.69	18.05	11.81	11.81
Gomphonema aff. *gracile*	0.78	7.87	4.62	3.84	6.27	18.08
Eunotia formica	6.47	0.00	4.19	1.99	5.69	23.77
Eunotia minor	7.37	1.41	3.91	1.95	5.32	29.09
Diadesmis confervacea	0.00	5.20	3.41	18.05	4.63	33.71
Navicula recens	0.00	4.80	3.14	18.05	4.27	37.98
Nitzschia amphibia	0.44	5.20	3.14	3.99	4.26	42.24
Fragilaria biceps	4.52	0.00	2.95	1.73	4.00	46.24
Nitzschia acicularis	0.53	4.90	2.89	3.19	3.92	50.17
Gomphonema parvulum	5.71	4.12	2.66	1.23	3.61	53.78
Gomphonema affine	3.52	0.00	2.31	0.85	3.14	56.92
Frustulia saxonica	3.27	0.00	2.12	1.87	2.88	59.8
Nitzschia closterium	1.29	3.74	2.08	4.36	2.83	62.63
Eunotia pectinalis var. *undulata*	4.64	2.45	2.03	0.82	2.76	65.39
Staurosira elliptica	2.85	0.00	1.78	0.72	2.42	67.8
Nitzschia umbonata	0.17	2.65	1.62	6.43	2.20	70
Eunotia rhomboidea	2.40	0.00	1.54	0.61	2.09	72.09
Gomphonema gracile	2.21	0.00	1.43	0.64	1.94	74.03
Encyonema mesianum	2.58	2.65	1.37	2.55	1.86	75.89
Sellophora seminulum	0.00	2.00	1.31	18.05	1.78	77.67
Gomphonema contraturris	1.83	0.00	1.19	1.21	1.61	79.28
Asterionella formosa	4.95	6.56	1.12	1.14	1.52	80.8

Table 5 SIMPER data for average dissimilarity between the panhandle section of the Okavango River and the Nxamaseri Floodplain, Botswana continued.

Dissimilarity	Panhandle	Nxamaseri	Average dissimilarity: 73.61%			
Species	Av.Abund	Av.Abund	Av.Diss	Diss/SD	Contrib%	Cum.%
Tabularia fasciculata	3.81	4.80	1.11	0.98	1.51	82.31
Achnanthidium minutissimum	1.48	0.00	0.95	0.59	1.30	83.61
Melosira varians	1.15	0.00	0.72	0.64	0.98	84.58
Achnanthidium macrocephalum	1.09	0.00	0.69	0.41	0.94	85.53
Craticula acidoclinata	0.00	1.00	0.66	18.05	0.89	86.42
Nitzschia filiformis	0.00	1.00	0.66	18.05	0.89	87.31
Pinnularia divergens	0.00	1.00	0.66	18.05	0.89	88.2
Stauroneis phoenicenteron	0.00	1.00	0.66	18.05	0.89	89.09
Gomphonema insigne	0.93	0.00	0.64	0.41	0.87	89.96
Staurosirella pinnata	0.93	0.00	0.57	0.41	0.77	90.73

Physico-chemical water parameters are presented below in Figures 3 – 5. Salinity was excluded due to the very low values measured with little to no variation found between readings. NXA4 had the highest oxygen reading and LAG 15 the lowest (Figure 3). pH readings were very consistent with readings varying between slightly above 7 to slightly below 6. The lowest pH was measured at NGA1. Oxygen (%) once more had the highest reading at NXA4 and the lowest at LAG15 (Figure 4). Temperature showed little variation with all readings being just below 20°C. The highest conductivity was measured at NXA4 at 130 μs/cm and the lowest conductivity was measured at Lag 15 with a conductivity of less than 20 μs/cm (Figure 5). Sites KRO1, LAG21 and NXA2 had moderate conductivity readings of around 60 μs/cm. All these readings are very low compared to other systems.

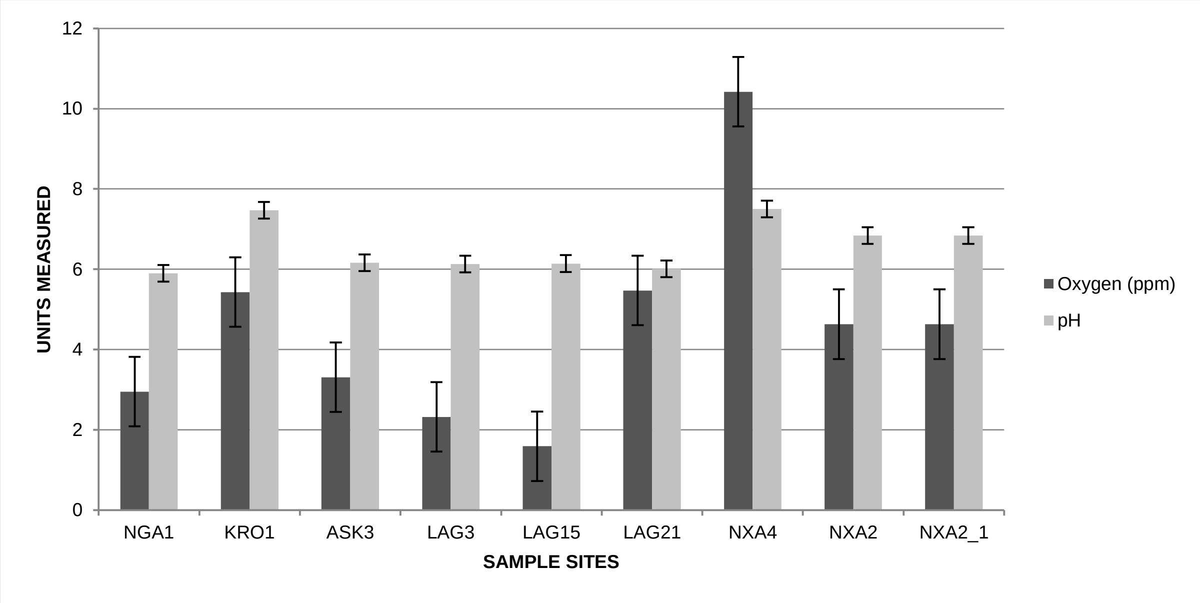

Figure 3: Oxygen (ppm) and pH readings measured between various sites in the Okavango Panhandle and the Nxamaseri Floodplain during 2014. ASK – Askiesbos Channel, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

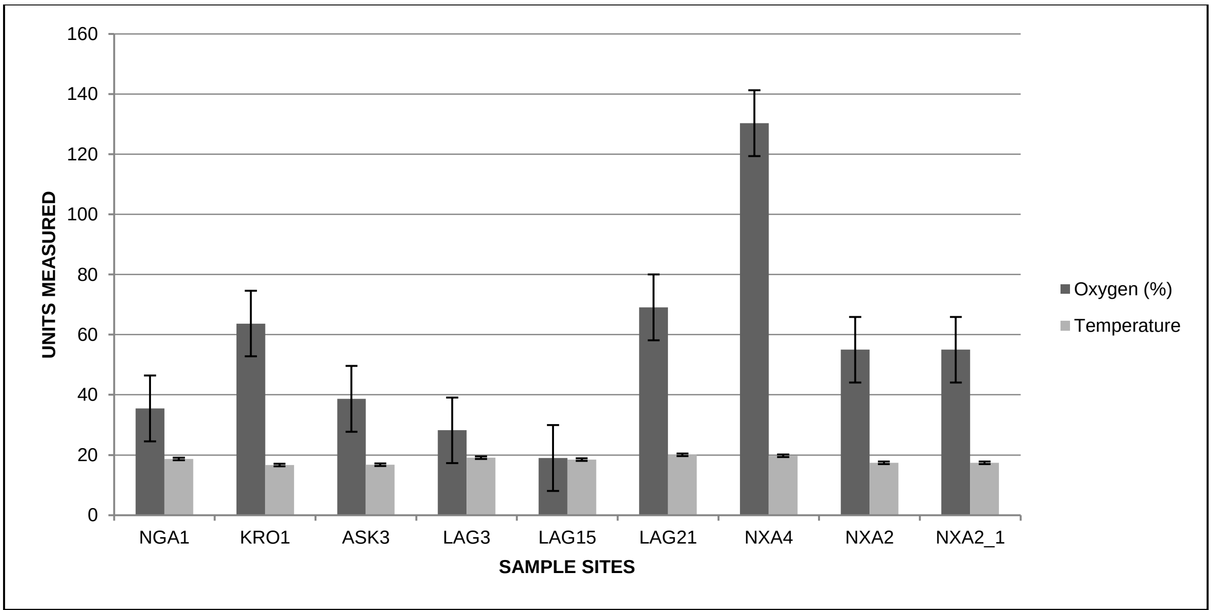

Figure 4: Oxygen (%) and temperature (°C) readings measured between various sites in the Okavango Panhandle and the Nxamaseri Floodplain during 2014. ASK – Askiesbos Channel, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

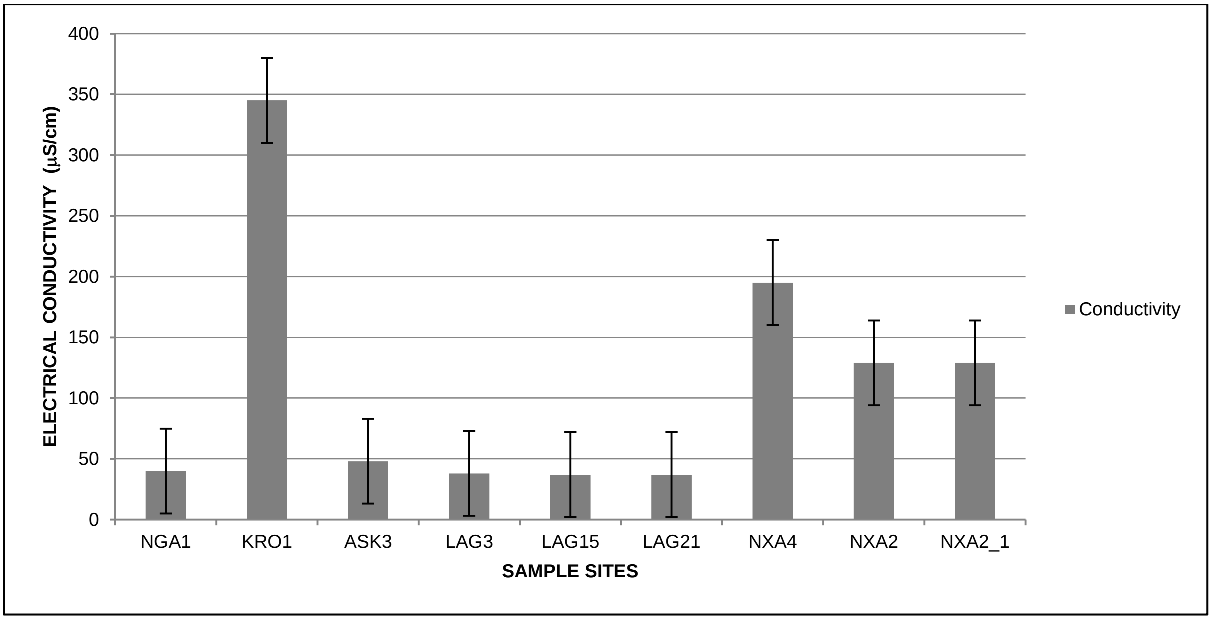

Figure 5: Conductivity (µs/cm) readings measured between various sites in the Okavango Panhandle and the Nxamaseri Floodplain during 2014. ASK – Askiesbos Channel, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

A PCA ordination of the water quality parameters measured. The ordination clearly shows the difference in water quality between the Thamalakane River, Okavango Panhandle and the Nxamaseri Floodplain sites at the time of sample collection (Figure 6).

A BEST analysis was performed to test the relationship between physico-chemical water quality data collected and the diatom community composition patterns. The analysis found that salinity, temperature and pH were the strongest water quality parameters associated with community composition in the Okavango Panhandle in Botswana (Table 6). The global test sample statistic was Rho = 0.66 with the correlation for the best combination found to be 0.66.

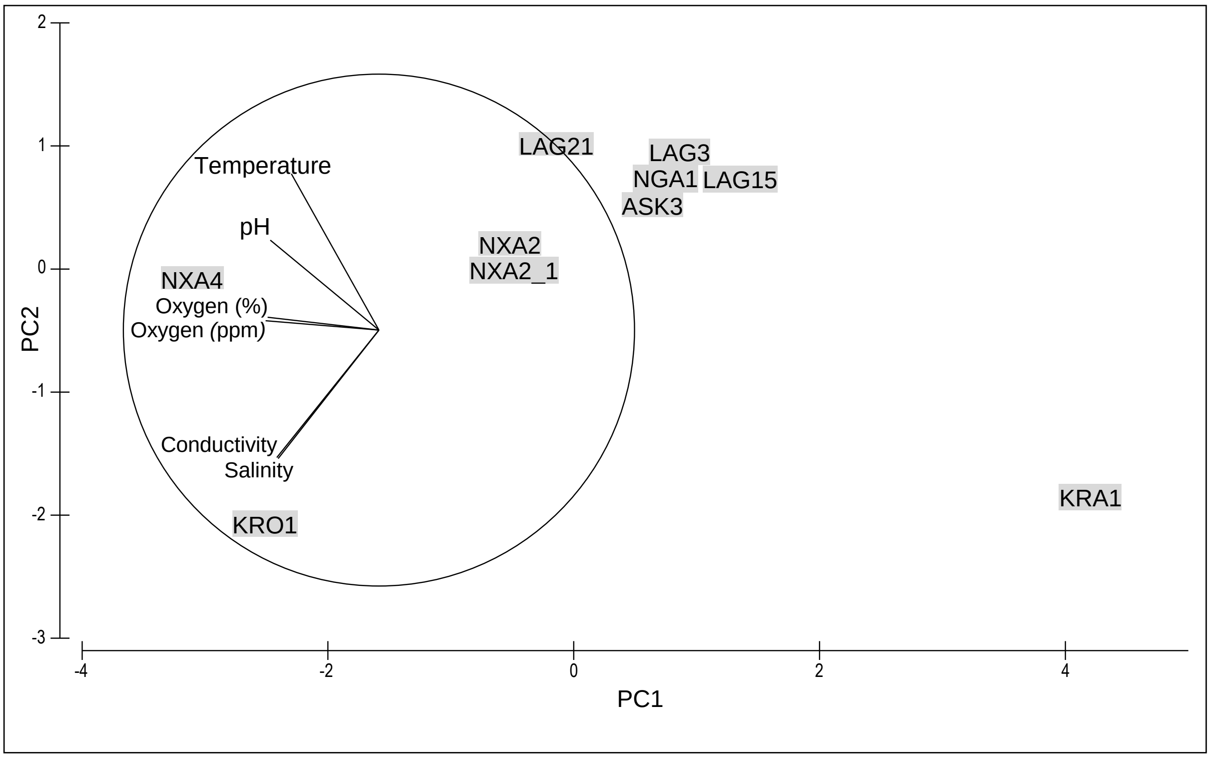

Figure 6: PCA ordination of physico-chemical as measured at time of sampling. ASK – Askiesbos Channel, KRA – Thamalakane River, KRO - Krokovango Wetland, LAG – Samochima Lagoon, NGA - Ngaringi Channel and NXA – Nxamaseri Floodplain.

Table 6 BEST analysis for diatom community composition and physico-chemical water quality parameters.

Variables	**Correlation**
Salinity, Temperature and pH	0.660
Salinity and pH	0.648
Conductivity, Temperature and pH	0.645
Salinity and Temperature	0.644
Salinity, Temperature, Conductivity and pH	0.642
Conductivity and Temperature	0.6280
Conductivity and pH	0.615
Salinity, Conductivity and pH	0.608
pH	0.602
Salinity, Conductivity and Temperature	0.59

Discussion

From a basin conservation point of view, the exclusion of the panhandle section from large-scale conservation initiatives makes no sense. The migration corridor is diverted around the panhandle via the Zambezi River to facilitate migration into the Caprivi (known as the Zambezi Region, since 2013) instead of just following the river upstream directly. The only possible explanation for this approach is the uncertainty that exists in the panhandle regarding development and conservation. The panhandle is an unmonitored rural dilemma, with small culturally complex and diverse communities living scattered across the landscape with little to no monitoring of impacts on the river system present. It is therefore impossible to predict the consequences of an animal corridor straight up through the panhandle. Possibly we could expect to see increased poaching, increased difficulty to maintain veterinary fences, increased human/animal conflicts and increased unmonitored development and unplanned informal settlements. Because the area is already neglected when it comes to landscape development planning and integrated conservation strategies, it would make sense that rather than attempt to address the disarray, migration routes were instead diverted away from this region. This may have made sense during the establishment of large scale conservation priorities, however, if the Okavango Delta is to remain a pristine wildlife haven, the conservation of upstream river sections should be prioritized in order to conserve the biodiversity it contains and the unique communities and the interactions that preserve the integrity of the downstream portion of the river.

Some of the sampled areas were better suited to phytoplankton sampling and others were more suited for the introduction of substrates. Because the aim of this study was primarily to document the communities present in the panhandle, sampling was not limited to one specific method but rather aimed at including all biotopes or micro-environments present. When all the data were compared, no sampling method difference was seen in the community composition, but instead a regional separation occurred.

The panhandle section of the Okavango River showed significant difference in community composition when compared to the downstream Thamalakane River and the Nxamaseri Floodplain. The KRO1 site was situated on a man-made wetland on a crocodile farm and was therefore expected to show some difference in community composition from the rest of the samples in the Okavango Panhandle, which it did. This is most likely due to the elevated nutrient load present in the localised wetland system which is aimed at dealing with waste water from the crodocile farm. Despite this difference in functionality the community still showed a close comparability to the other panhandle samples collected. The panhandle diatom community identity was driven by *Eunotia minor, Eunotia formica*, *Asterionella formosa* and *Fragilaria biceps*. *Eunotia minor* is known to occur in circumneutral waters while *Eunotia formica* has been described to occur in slow flowing dystrophic to oligotrophic waters, with slightly elevated electrolyte content (Taylor et al. 2007b). In contrast to this *Asterionella formosa* and *Fragilaria biceps* are more commonly known to occur either attached or free-living in the plankton of meso-eutrophic rivers. However, we know that the water is not eutrophic in the panhandle and in fact it has a very low nutrient content as shown in the conductivity measured at sampled sites (Figure 5). A possible explanation is the micro-environments created by floating papyrus beds. Papyrus acts as a nutrient trap and forms highly productive environments for algae and insects in this otherwise vast aquatic desert. It is no wonder then that papyrus is such an integral part of the Okavango River's ecological functioning. Together with larger mammals, which keep channels open by forging paths through the papyrus, the integrity of the papyrus could be more important than presently acknowledged for the continued functioning of the Okavango River in the panhandle and downstream in the delta. If it were not for these micro-environments, which act as nutrient factories, the Okavango River in Botswana would not be able to sustain aquatic life. It makes sense then that both circumneutral and slightly eutrophic diatom species would be important role players in the panhandle.

In comparison, the Nxamaseri Floodplain's diatom community composition was driven by *Gomphonema* aff. *gracile* and *Gomphonema affine. Gomphonema affine* is a subtropical species tolerant of elevated electrolyte content and *Gomphonema* aff. *gracile* is tolerant of extremely polluted conditions (Taylor et al. 2007b). Considering that there are cattle and donkeys roaming the floodplain and the increased microscopic productivity within the floodplain as water starts to recede, it would make sense that more tolerant species would be more dominant in such an extreme environment. To survive in the Nxamaseri Floodplain a species needs to face desiccation for long periods, and then competition when the next flood arrives. It is an environment of extremes. The river bed is situated in a semi-arid region which is situated in the subtropics and is trampled by livestock and other animals for large parts of the year. While the same water flows in the floodplain that does in the panhandle, this significant difference in diatom community composition shows what a big impact a change in water volume and quality can make to the composition of a community at the base of the food-web. This was seen clearly in the water quality data where the Thamalakane River site had much lower temperature, dissolved oxygen, conductivity, salinity and pH than the Nxamaseri Floodplain site NXA4 (Figure 3–5). This is expected since NXA4 was situated in an isolated smaller pool upstream of NXA2, which was much larger in size and had still been connected to the panhandle active channel not too long ago (Figure 6). This supports the occurance of more tolerant species at the Nxamaseri sites.

The dissimilarity in community composition between the Okavango Panhandle and Nxamaseri Floodplain were driven by the following species; *Fragilaria ulna* var. *acus*, *Gomphonema* aff. *gracile*, *Eunotia formica*, *Eunotia minor*, *Diadesmis confervacea*, *Navicula recens*, *Nitzschia amphibia*, *Fragilaria biceps* and *Nitzschia acicularis* which accounted for 50,17% of the total dissimilarity. *Gomphonema* aff. *gracile*, *Eunotia minor*, *Nitzschia amphibia* and *Nitzschia acicularis* occurred in both regions, but with a considerable difference in abundance (Table 5).

The use of diatoms in pulse-wetlands and lower river reaches such as in the panhandle is less developed but similar to what Mackay et al. (2012) found. This study also identified a geochemical stratification between permanently inundated and seasonally flooded communities. Not much information exists about the biodiversity of semi-arid wetland and associated lower reach ecosystems. Perhaps due to the overall lower diversity associated with these areas (Junk et al. 2006). It is a tragedy that these unique environments are not more prioritised for conservation and research initiatives, especially since the intricate interactions between the diversity that does occur is often very dynamic, well adapted and unique. There is no way in predicting how much neglect or unmonitored impact will be too much. Due to the lack of information and general knowledge on the biodiversity and intricate functioning of the panhandle, it is hard to predict what increased impacts could mean for the overall functioning and sustainability of the Okavango River and the Okavango Delta downstream specifically. This is worrying when looking at the real threat posed by climate-induced impacts, increased upstream consumption and development, flow alternations associated with possible hydro dam development and increased localized utilization and the lack of any real conservation and monitoring at present. This study produced a baseline diatom reference condition, a snapshot of the importance of micro-environments created by nutrient trapping papyrus and other reeds. This study was also able to show how small scale changes in water quality could have significant impacts on diatom community composition and possible associated ecosystem functioning.

Conclusion

A wetland's biodiversity is greatly dependant and influenced by its regional climate and hydrological regime (Junk et al. 2006). The Okavango Wetland is situated in a semi-arid region and the overall biodiversity is therefore also relatively lower than would be expected from a wetland situated on a subtropical latitude. Like most semi-arid areas, the ecological interactions are very complex and evolved over many thousands of years to be well-equipped with the low nutrient load and the seasonal pulse flooding. The micro-environments created by papyrus beds and seasonally inundated island floodplains are therefore extremely important for nutrient capturing and primary production in an otherwise aquatic desert.

Diatoms play a vital role in the nutrient cycling and availability at the bottom of the food web. Their diversity is relatively low as expected for the Okavango River in Botswana but the panhandle community composition was still uniquely distinguishable from other associated systems of the lower Okavango River. The Nxamaseri Floodplain, which is an adjacent system and annually inundated with panhandle water, was significantly different from the panhandle diatom community composition due to localised impacts. Diatom communities are well known to respond rapidly to small changes in water quality, which was documented here showing how fast a community can change over short distance and with relatively small changes in water quality. This highlights the importance of conservation of biodiversity upstream for the continued optimal ecological functioning of the downstream Okavango Delta as an important RAMSAR and World Heritage site. Conservation efforts should include the upstream catchment to ensure that unplanned development and utilisation do not continue to be unmonitored. Small impacts can have drastic effects on diatom community composition, which play an important role in ecosystem functioning.

Both the Okavango Delta and the panhandle are important lifelines for the animals and especially the people of Botswana. With most rural villages in the panhandle and delta directly dependant on the river for their livelihoods, the conservation and sustainable management of this area is of utmost importance and urgency if sustainability is to be ensured for the future well-being of Botswana and its people.

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Discussion

CHAPTER 6 Discussion

Diatom phylogeny enjoyed attention for many years before it came to a somewhat standstill. It was only when Round et al. (1990) re-examined the phylogeny and morphological descriptions that the evolutionary history was truly fully questioned again. This could have been due to the fact that evolutionary biology had experienced an increase in popularity since the development and refinement of genetic analyses. Even in modern times, we are still not able to explain the evolutionary history of diatoms by means of genetic analyses or morphological descriptive phylogeny. What is clear is that diatoms evolved more recently than other algae and that they are linked to animals as well as plants. This not only makes them a very unique and interesting piece of the history of life on Earth, but also amplifies their evolutionary place within the food-web and fundamental role within freshwater ecosystem functioning and for that matter also in the marine environment.

This study was able to successfully link freshwater diatom communities with specific ecological regions within a landscape, as well as show that diatom community information is quite robust when applied at a larger ecological scale. The results produced in this study corresponds to those of Potapova and Charles (2002) who produced the first study that linked diatom community composition to Ecoregion Level I descriptions of the USA. Potapova and Charles (2002) found that communities linked closely to pH and temperature. In this study a similar link between diatom community composition and pH was identified. Temperature played a less important role and instead conductivity and salinity were identified as key physico-chemical drivers of community composition. This difference could possibly be due to a difference in specific Ecoregion Level I characteristics. Eutrophication and oligotrophic systems separated significantly from one another in Potapova and Charles's study. The same was found to be true for South African Ecoregion Level I scale diatom community differentiation with the DC and SECB showing the highest separation in community composition.

Diatoms have been used, for the most part, in the past for smaller scale assessments. The development of various indices have been a useful addition to water quality management, however, it seems as though the only value of diatoms in assessment and monitoring sciences, in Southern Africa anyway, have been deduced to being applied as diagnostic entities for specific impacts. Diatom indices originate from Europe where most rivers are modified, impacted and channelised. While these indices have been successfully implemented on a variety of systems across the world since their development, it would seem as though most of the freshwater diatom work is concerned with one of two questions;

can the diatom indices be successfully applied in the Southern Hemisphere and how can we simplify it even more, to make it increasingly user-friendly, cost effective and fast, without losing too much depth of information?

While these are extremely important aspects of diatom science today, it would appear that taxonomy and larger scale ecological work on diatoms as a group have been somewhat less pronounced, especially in countries outside of Europe. Perhaps this is due to the fact that funding such studies requires a good motivation for application towards sustainability and freshwater security in future. Especially in developing countries, where funding is limited and service provision and impact monitoring take higher priority. Whatever the reason may be, the fact remains that we are not yet fully knowledgeable on the ecological character of diatoms.

Diatoms are well known to be cosmopolitan, with the sub-cosmopolitan theory stating that theoretically a diatom species should be able to occur in any river as long as the ecological requirements/preferences are met (Kelly 1998). The water quality, or rather physico-chemistry, related ecological requirements of different diatom species are well studied and documented, as has been illustrated in Chapter two, however, the landscape scale ecological gradients that drive biological distribution on different natural freshwater ecosystems are not yet well defined or examined. While river catchment signatures have been described for macroinvertebrates and riparian vegetation (King and Schael 2001), no such question has been asked regarding diatom distribution. Perhaps this is due to the fact that diatoms are assumed to be cosmopolitan and should therefore not adhere to any signature distribution patterns associated with larger aquatic biota? Perhaps the reason for the occurrence of endemic diatom species, which in itself is questioned because of the sub-cosmopolitan theory, is due to ecological gradients acting on the physical character of a river ecosystem, thus driving the specific ecological characteristics found in a specific river catchment and consequently directly influencing diatom distribution and the associated food web associated with the river catchment.

Perhaps then a more appropriate description for cosmopolitan species could be tolerant species with the less abundant or rare species instead being described as habitat specific species. Tolerant species are found in higher abundances across a broader spectrum of ecological requirements, such as Ecoregion Level 1 classification. More sensitive or habitat specific species respond rapidly to small scale changes and could be responsible for the seasonality seen in community composition within a catchment. However, these seasonal changes are not so extensive as to make the community composition unrecognisable with that of the rest of the sites for a specific Ecoregion Level I. Both the tolerant and habitat specific species are important for small scale diagnostics as extracted from OMNIDIA and other index calculating packages. There are thus multi-layered approaches, which need to be considered when setting out to answer questions regarding diatom ecology.

If this layered approach to freshwater ecosystem functioning is to be used as a classification, assessment and monitoring tool and in associated managerial and decision-making processes, it is very clear that scale is currently the single biggest constraint when working with diatoms. Diatom indices do not currently effectively address the question of scale. While extremely useful and effective at small scale for impact diagnosis and monitoring, diatoms have been underutilised, or dare I say, ignored when it comes to large scale climate change monitoring initiatives of freshwater river systems. With rivers being the largest provider of potable freshwater on Earth, it is important to look at diatoms as impact monitoring and potentially diagnostic tools also at larger landscape scale. This has and is already being done very effectively in marine environments, especially in the Arctic regions, where scientists are studying and monitoring diatom communities as information hubs for present and past climatic conditions (Anderson 2000, Cermeño et al. 2013, Roberts, Jones et al. 2015, Soppa et al. 2016, Virta and Soininen 2017). It would make sense then to utilise diatoms with the same efficiency in freshwater systems.

Africa's river systems have been underestimated by European explorers, naturalists and scientists since the discovery and colonisation periods. The ecological dynamics of Africa's freshwater ecosystems are quite robust and diverse, sometimes changing dramatically over short distances. The geography and climate contribute to the highly dynamic character with slight changes in climate causing large scale changes in the functioning of these ecosystems. Many countries have very monotonous river ecosystems to monitor and manage; such is the case with Australia for instance. While Australia is also relatively new to the use of diatoms in ecological monitoring, their rivers are not as diverse or dynamic as those found in Africa, and southern Africa. This also applies specifically to the Okavango River in Botswana, which in terms of water quality and geomorphology, can be quite monotonous from where it enters the country down into the lowest parts of the delta (West et al. 2015). When working on rivers and associated biodiversity in southern Africa, this possibly dynamic and diverse character of freshwater ecosystems should be taken into consideration, especially for developmental projects, which could have adverse and exacerbated effects on water security if not sustainably implemented.

Rivers are self-regulating entities. They have the ability to adjust and/or adapt to natural disturbances within the system. An example of this is seen within the Western Cape (Mediterranean climate) where reproductive strategies of riparian vegetation species are adapted to the winter flooding patterns (Magoba 2014). Not only is the positioning of a specific species on the river bank important for reproduction success, their timing with regards to the production of seed has been linked to the natural flow cycle (Reinecke 2013). Flash flooding in the winter is utilised as a tool for seed dispersal. When taking this one type of adaptation to natural fluctuation into account, it can be expected that the same could be possible at the base of the food chain in these systems. The rivers are able to cope well with natural disturbance. This robustness was seen in the diatom communities in the Eastern and Western Cape provinces of South Africa.

Community composition did not show significant difference between seasonal sampling efforts, but instead separated at an Ecoregion Level. There are exceptions to this such as was the case with the GAM1 site which had samples in group nine and group ten of the intitial Cluster assessment (Figure 59). Also, GOU1 samples were spread across group five and group six. Both these sites were situated slightly lower down the catchment. GAM1 is an upper foothill zone site and GOU1 is classified as a lower foothill zone site. There might be other impacts driving these slight differences in seasonal sampling as is usually the case during hydrological changes. The slightest anthropogenic impact could be exacerbated by a seasonal change in flow. Possible causes can only truly be clarified with a more in depth assessment of possible environmental drivers lower down the catchment and their relationship with diatom community composition.

While there may have been small scale changes in community composition between seasons, samples were still able to group with other samples of their specific ecoregion. Interestingly enough, at a large scale the diatoms did not adhere to a river catchment signature pattern, instead responding to a larger ecological gradient (Ecoregion Level), while at smaller scale (within ecoregions) there were small separations appearing, but these were also not always at a specific catchment level. This could also be due to the scale of the current study being too large to see the formation of such smaller scale patterns. This nonconformity to catchment signatures (unlike that demonstrated by other freshwater associated biota) could mean that while diatoms respond very rapidly at a very small scale to small changes in physico-chemical characteristics, at the same time they do not respond so dramatically as to make them unreliable large scale ecological components of the specific landscape’s biotic blueprint. This could potentially make them very effective biotic monitoring tools at a larger ecological scale than most other freshwater associated biota. In a climate change context (large scale driven impacts) the first responders at the biotic level could very well be diatoms. This is extremely interesting, since we have no real information on diatom evolutionary history and we know diatoms are not able to migrate between catchments. The dispersal questions associated with this robustness of diatom community information is something worth exploring further in future studies.

In contrast to what is commonly assumed about diatoms being very sensitive and selective to environmental change, this study found diatoms, at community level, to be very robust and resilient to natural changes in flow and the associated changes in water chemistry (seasonality). It is important to note that none of the sites used in this study, including the Okavango River in Botswana, were severely impacted by industrial or other anthropogenic disturbances. Some rivers such as the Great Fish, Tsitsikamma, Krom and Gamtoos rivers had agricultural activities happening upstream of the sample sites, but the overall ecological integrity of the sites were still intact and one could expect them to be considered to be in good condition (Kleynhans and Louw 2007). So, based on the natural fluctuation within these systems, diatoms showed a response, but not dramatically. This is surprising considering some of these rivers showed considerable seasonal change, especially with regards to flow. During most of the spring and summer samples the rivers had low flow, but in winter and autumn especially, almost all rivers were in flood. This clearly influenced light penetration, nutrient availability, sediment load, as well as river width and depth as seen from field observations made at the times of sampling.

No seasonal sampling was conducted in Botswana and future work on this could provide necessary information on the influence of the annual flooding in the lower sections of the river on diatom community composition. The lower Okavango River receives most of its water from rain in the Angolan highlands. The water reaches the Namibian and Botswana border in October and continues to flood down till March annually. Samples in this study were collected during July and August which was when the flood waters were receding. This created the isolated pools in Nxamaseri Floodplain. Since the flood is known to carry valuable nutrients into the panhandle and delta systems the productivity at the base of the food web could very well be quite different during peak flooding.

One noteworthy observation made by the author was that no diatom species information needed to be excluded in order to obtain accurate multivariate statistical community-based results. Often in ecological studies, including that of diatoms, the authors remove low abundance species from data sets (<1% abundance) (Holmes and Taylor 2015, Stevenson et al. 2006). This was not done in the current study, and when compared to analyses run with removed information, identical results were obtained. While this could be regarded as support for the continued removal of low abundance species, the contrary could also be said. By including the low abundance species, it lowers the preparation time on data sheets and promotes an inclusive approach to the assessment of biodiversity related information. It also ensures that all community information gets documented properly for future use. One of the biggest problems with the Eastern Cape data is that the little historic information available does not have any abundance information included in the documents and in some cases low abundance species are hardly documented at all (Archibald 1981, Giffen 1984). In light of the current biodiversity concerns expressed globally, it would almost seem unethical to not include all components of the biodiversity of a diatom sample, if only for proper recording purposes. This would indeed be a victory for the promotion of the effective, comprehensive and respectful utilisation and conservation of biotic assets as described by the Strategic Plan for Biodiversity 2011-2020, associated Aichi Biodiversity Targets and the Sustainable Development Goals 2030[51] (Figure 85).

Figure 85 The 17 Sustainable Development Goals (SDG's) adopted by world leaders in September 2015 to transform the world into a better place for all by 2030. (Available online from the United Nations Department of Public Information[52]).

As a consequence to statistical analyses indicating a relationship between diatom community composition and Ecoregion Level I classification, a smaller scale investigation into diatom community composition was made within the three sampled ecoregions. This means that the current study not only produced a diatom community reference description

[51] For more information regarding the sustainable development goals visit: https://sustainabledevelopment.un.org/?menu=1300 (accessed on 10 January 2018)
[52] http://www.un.org/sustainabledevelopment/wp-content/uploads/2016/10/UN-Guidelines-for-Use-of-SDG-logo-and-17-icons.October-2016.pdf (accessed on 10 January 2018)

for Ecoregion Level I, but consequently also a reference condition for diatoms of Ecoregion Level II. Ecoregion Level II descriptions for South Africa were not yet concluded by the time this study came to an end. It will be interesting to see how the diatom community description produced in this study compares to the Ecoregion Level II information produced for South Africa. These descriptions will hopefully aid in the inclusion of diatoms into larger scale ecological monitoring initiatives towards a more holistic approach to freshwater resource management and decision-making for the better of all South Africans as included in the National Water Act of 1998 (NWA 1998).

For **Ecoregion Level I** it was found that the main species responsible for group identity were as follows; SECB – *Eunotia minor*, *Achnanthes oblongella* and *Eunotia incisa*, for SFM – *Fragilaria biceps*, *Eunotia minor*, *Nitzschia closterium*, and *Achnanthes oblongella* and for the DC – *Cocconeis placentula*, *Cocconeis placentula* var. *lineata*, *Gomphonema parvulum*, *Rhopalodia gibba*, *Achnanthes oblongella* and *Achnanthidium minutissimum*. According to the European Guidance Standard (2003) water quality classes, *Gomphonema parvulum* is considered to be an indication of moderate water quality, however, *Gomphonema parvulum* is also known to be tolerant of extremely polluted water conditions (Taylor et al. 2007b). This wide ecological description most probably contributes to its distribution being described as cosmopolitan. Theoretically all diatoms are cosmopolitan. Therefore perhaps a better description would be a very tolerant species therefore found regularly and in a wide variety of habitats. This is one example highlighting the need for more detailed ecological assessments and associated descriptions. This will become increasingly important in the next couple of decades as the pressure on freshwater resources increases (UNESCO 2014).

Cocconeis placentula and *Fragilaria biceps* are described as occurring in mesotrophic to eutrophic waters (Taylor et al. 2007b). This is, however, not necessarily accurate for the rivers of the DC ecoregion since these rivers, on average, had a higher conductivity, higher turbidity and were found to be more brackish in nature. As described in Diatoms and conductivity in CHAPTER 2 An overview of diatom ecology and community composition, diatoms that are good indicators of eutrophication generally have a wide conductivity tolerance and both eutrophic and oligotrophic conditions in brackish waters have a high conductivity (Taylor et al. 2007b). Since the drought corridor is characterised mostly by wide, turbulent and brackish rivers a better indication for eutrophication in these systems would be the number of taxa present and the size of diatoms.

In contrast to *Cocconeis placentula*, both *Cocconeis placentula* var. *lineata* and *Achnanthes oblongella* prefer oligotrophic water. This makes sense when brought into context with *Cocconeis placentula* and *Fragilaria biceps* not necessarily being an indication of high nutrient load as explained above. *Rhopalodia gibba* and *Nitzschia closterium* prefer brackish water with moderate to high electrolyte content while *Achnanthidium minutissimum* is described as having a preference for well-oxygenated and clean water (Taylor et al. 2007b). *Eunotia minor* is known to occur in circumneutral waters, while *Eunotia incisa* prefers upland streams of an acidic, oligotrophic and electrolyte-poor nature. This makes *Eunotia incisa* well suited to being a prominent driver of community identity in the SECB of which the rivers are characterised by tannin-loaded acidic waters, flowing over a short distance to the coast with rapid changes in slope. These were mostly mountain stream or upper catchment geomorphological classification as described by Rowntree et al. (2000) and therefore lower nutrient availability than associated with slower flowing lowland river sections (Dollar and Rowntree 2003).

Above is only a discussion on the species responsible for at least more than 50% of the community identity. There are more species associated with the group identity as presented in the various SIMPER tables; Table 7, Table 8 and Table 9 in CHAPTER 4 Diatom community composition across ecological gradients. This means that while a species can be a prominent contributing factor in community identity in one ecoregion, it could be a less prominent contributor in another. Species found to play a role in community identity in all three ecoregions were; *Achnanthes oblongella*, *Achnanthidium eutrophilum*, *Achnanthidium minutissimum*, *Eunotia minor*, *Fragilaria biceps* and *Gomphonema parvulum*. The SECB had 14 species responsible for community identity, the SFM had 17 species identified and the DC had 29 species contributing to the community identity. The SFM had the lowest percentage species overlapping with the two other ecoregions. Only 29% of the species responsible for community identity played a community identity role in the SECB and DC.

The DC had the largest number of species contributing to community composition; this could be explained due to the fact that the DC is a very dynamic, harsher environment with many rivers in the region only presenting seasonal flow. However, some of these have experienced flow alterations. An example of this is found in the Sundays River, which has been turned into a perennial river by the Fish-Sundays inter-basin transfer scheme (van Vuuren 2012). Water is transferred from the Orange River into the Great Fish River via the Oviston tunnel, then at the town of Somerset East water is transferred from the Great Fish River into the Little Fish River (below sampled sites) and then to the Volkers River, which flows into the Darlington Dam, from where it is released downstream into the Sundays River. The Sundays River only has a short seasonal branch upstream of the Darlington Dam, and presumably the Volkers River would also be seasonal in flow if it was not for the addition of Orange River water via the Great Fish River. The high diversity in diatom species contributing to community identity could therefore also be due to the significant influence the

Orange River basin water has had on the ecological functioning of many important rivers in this ecoregion over the past 40 years. Further studies and more detailed assessments are needed to truly understand the impact inter-basin transfers have on the diatom communities and associated ecosystem functioning.

It is interesting to note that even though some rivers have been completely modified with regards to their ecological functioning, the diatom community still showed Ecoregion Level I separation. This again supports the robustness of diatom information at larger scales. The high level of diversity in species important for community identity could also be due to the fact that the inter-basin transfer scheme has been operational for 40 years and the diatom communities have adapted to these impacts, but now have a more complex community composition. Could it be said then that these rivers are more or less vulnerable to climate impacts? Should less water become available for redistribution from the Orange River Basin (van Vuuren 2012) due to climate impacts, how resilient would these already impacted rivers be? Perhaps they would not be as vulnerable as would usually be expected from an already impacted catchment. These are all very real threats that water redistribution initiatives, such as this one, will be faced with if the predicted impacts of climate change start impacting the South African strategic water source areas.

At **Ecoregion Level II** the **SECB** separated into four groups. The data presented in The South Eastern Coastal Belt (SECB) section of CHAPTER 4 Diatom community composition across ecological gradients, shows that unlike what would be expected, data did not separate seasonally or according to catchment signatures. Instead the data presented a much more scattered pattern. The Touws, Groot and Tsitsikamma rivers were the only rivers of which all samples separated into the same groups. The other rivers had sites occurring in more than one group. This indicates a potential seasonal influence impacting at this level of assessment. However, samples collected from different sites but the same season did not group together. Thus, flooding in itself could not explain diatom community diversity. Instead physico-chemical drivers were found to be associated with diatom community composition. These four groups were found to be most closely linked to a difference in pH. This was seen in the data (see Appendix 2 – Raw data: Measured water quality paramaters) with the Tsitsikamma River showing a range in pH measured over a one year sampling period of 1.14, the Touws River site showed a 0.67 range in pH and the Groot River had a range of 0.86 in pH. The Keurbooms River had a range of 2.34 and the Elands River showed a range of 2.51 in pH. This may not look like much, but considering that pH is an exponential value, the change is quite considerable especially in an ecoregion where pH is indicated to play a prominent role in diatom community composition.

When looking at the physico-chemical parameters and the group formation for SECB, group three and group four samples had a higher conductivity and salinity than measured at other river sites. The conductivity was generally very low in this ecoregion, probably due to most sites being in upper geomorphological regions which mean the higher slope does not allow for nutrient deposition. The lowest conductivity, 32 μs/cm, was recorded on the Kruis River during autumn, which makes sense, since this was when the rivers were flooding. Therefore it should be taken into consideration when a higher conductivity is associated with some of the samples. It was still not exceptionally high, except for the case in the Hoogekraal River that had two readings that were very high compared to the other samples. These were 1007 μs/cm and 2274 μscm during summer and winter, respectively. When the mouth of the Swartvlei Estuary at Sedgefield has not been breached and the water level rises and becomes perched, it often backs brackish water from within Swartvlei into the Hoogekraal River. The sampled site was close enough to the Swartvlei that this may have caused the elevated electrical conductivity values.

Conductivity was found to be the other contributing physico-chemical parameters, in conjunction with pH, which could be linked to community composition in the SECB. The SECB had lower pH readings than the other two ecoregions, with the lowest pH recorded at the Bloukrans and Kruis rivers during summer (low flow conditions) with a value of 4.35. All the sites had higher pH readings during autumn when most rivers were in flood. Seasonality has a more apparent impact on diatom communities at Ecoregion Level II scale, but only due to prominent natural changes in flow and the associated impact it has on pH and conductivity.

The relationship between diatoms and pH is well documented such as seen with the Hustedt classification system (Hustedt 1939). It would therefore be expected that the prominent diatom species responsible for community differences between the four different groups would be acidobiontic (pH < 7 with optimum at 5.5 or below) or acidophilous (pH = 7 or below). Studies from China (Liu et al. 2011) and Canada (Hargan et al. 2015) found the genus *Eunotia* to be very abundant in acidic environments. The genus *Eunotia* had representatives identified in all four groups contributing to community identity. For all four groups, *Eunotia minor* was indicated and for groups one and two *Eunotia incisa* was also indicated as a contributing species in group identity. *Eunotia minor* is described as being acidophilous with *Eunotia incisa* being acidobiontic. Both prominent species were common in acidic environments that make them perfectly suited as important role players in community identity in the SECB.

Achnanthes oblongella and *Tabellaria flocculosa* are acidophilous while *Frustulia saxonica* is described as being acidobiontic. *Planothidium engelbrechtii* and *Fragilaria biceps* are common in waters with high electrolyte content or a higher conductivity. This is well suited to group three, where these species play an important role in group identity. Group three was the group more closely linked to increased conductivity readings. Another species associated with group three and four's community identies was *Gomphonema parvulum* that is a tolerant species to increased conductivity and very polluted waters (Taylor et al. 2007b).

Group four also had two other species involved with community identity that were not indicated in any of the other groups. These were *Eolimna minima* and *Denticula subtilis*. *Eolimna minima* is associated with organic detritus and can occur in a very wide range of ecological conditions, including heavily polluted waters. *Denticula subtilis* is often found in brackish waters with high electrolyte content (Taylor et al. 2007b). Both species are well suited for the environment they were found in this study. A gradient of physico-chemical parameters is present from left to right in the cluster ordination in Figure 66 (see Chapter 4). Samples grouped towards the left of the ordination had a low pH and conductivity while samples more towards the right had a higher pH and conductivity.

The **DC** had five groups forming as seen in Figure 79 in section Drought Corridor (DC) of CHAPTER 4 Diatom community composition across ecological gradients. The Small Fish River sites grouped together almost entirely to form group three and similarly the majority of Tyume River sites grouped together to form group one. The majority of the Koonap and Great Fish rivers' samples grouped together to form group five, while group four consisted of two samples collected from the Volkers River. Group two consisted of one sample on the Volkers River and one from the Koonap River.

Catchment signatures (see Figure 19 and Table 4) appear to be a little more prominent in this ecoregion, with a clearer separation between the Tyume River (Keiskamma River catchment), Koonap and Great Fish rivers (Great Fish River catchment), Small Fish River (also Great Fish River catchment) and the Sundays River (Sundays River catchment) being apparent than what was found for the SECB. The complete separation of the Small Fish River is interesting since it would be expected for these samples to group together with the Koonap and Great Fish rivers, if catchment signatures were in fact perfectly adhered too. The species distributions across the five groups were found to be most closely linked to electrical conductivity. When looking at the recorded physico-chemical readings at time of sampling in Appendix 2 – Raw data: Measured water quality paramaters it is apparent that the DC rivers had a more consistently higher conductivity recorded than in other ecoregions. These reading are still extremely low when compared to heavily impacted environments. An increase in conductivity in conjunction with the high pH readings recorded in this ecoregion, makes it safe to say that this ecoregion is characterised by brackish waters with a higher electrolyte content and consequently a higher conductivity.

When looking at the species identified as important role players in group formation within the DC, the ecological conclusion regarding physico-chemical character was confirmed. Compared to the nine species important for group formation within the SECB, the DC had 15 species contributing to group formation. For an ecoregion, which at first glance would appear more monotonous than the SECB with its many short coastal catchments, the DC had more groups forming and a more diverse assemblage of species responsible for diatom community composition. This would suggest that the semi-arid, often seasonally water fed and more homogenous landscape of the DC is no indication on how the rivers function or possibly react to change. Another possibility is that interbasin transfer schemes have greatly impacted on the diatom diversity and made the communities more diverse and complex for an otherwise more homogenous ecoregion. This was seen in Botswana where the semi-arid river had lower diversity in community composition despite the variation between areas within the system. It would therefore have been expected for the DC to show less complex community composition than was found to be the case. Compared to what Holmes and Taylor (2015) found in the upper reaches of the Great Fish River this study did not identify *Amphora pediculus* as a dominant species in the DC. This is good since *Amphora* species are well known to outcompete other larger species for N and P when there is eutrophication present in brackish rivers. They are also good indicators for pH since they are sensitive to changes in acidity (Sundbäck and Snoeijs 1991).

The most diverse group was group three with nine species contributing to group identity. Among these were; *Rhopalodia gibba*, *Epithemia sorex* and *Epithemia adnate*. *Rhopalodia gibba* is known to occur in slow flowing water with elevated electrolyte content. For *Epithemia sorex* and *Epithemia adnate* the ecological requirements are similar to that of *Rhopalodia gibba*, with the exception that this species is also well known to occur in brackish environments. All groups had one, two or all of the following species indicated in group formation; *Gomphonema parvulum*, *Cocconeis placentula* var. *lineata*, *Cocconeis placentula*, *Achnanthes oblongella* and *Nitzschia closterium*. These have been discussed above in Ecoregion Level I descriptions specifically with regards to their relationship to brackish and often consequent elevated conductivity readings.

Groups one and three also had *Planothidium frequentissimum*, which is a very tolerant species to polluted conditions. Apart from this it is also found to occur regularly in slow flowing, circumneutral to alkaline waters with a slightly elevated electrolyte content. In group two the centric diatom *Aulacoseira granulata* was indicated as an important role-player in group identity. This diatom is known to occur in eutrophic lakes and rivers. Groups four and five had *Reimeria uniseriata* as a key role player in community identity. This species is found in alkaline water of mesotrophic to eutrophic nature with moderate electrolyte content and is also able to tolerate high turbidity (Taylor et al. 2007b).

Group three had *Rhopalodia gibberula*, *Pleurosigma elongatum* and *Navicula radiosa* as important role players in community composition. *Rhopalodia gibberula* is found in water with elevated electrolyte content and often temperatures. It is worth mentioning that even though the results did not find a significant relationship between water temperature and species distribution, the water temperatures recorded in the DC were higher than recorded in other ecoregions.

Pleurosigma elongatum occurs on brackish inland rivers, as do *Navicula radiosa*, however, the latter is very sensitive to organic pollution. Therefore, *Navicula radiosa* can occur on a very wide range of pH and conductivity, but not in polluted or organically enriched eutrophic waters (Taylor et al. 2007b). This is a clear indication that the elevated conductivity in the DC is for the most part presumably not caused by organic inputs through agricultural activities in the area, but may instead be due to the brackish nature of the waters in this ecoregion. This is important to take into consideration when using diatoms as impact assessment tools in this region, as well as monitoring larger scale changes in ecological integrity.

The **SFM** had only three groups forming as presented in Figure 71 of the Southern Folded Mountains section within CHAPTER 4 Diatom community composition across ecological gradients. Group one consisted of samples collected from the Gamtoos and Groot rivers together with one sample from the Baviaanskloof River (BAV6_2). Groups two and three consisted of Baviaanskloof samples and one sample collected from the Krom River (KRO2_4). Group two consisted mostly of sites situated just upstream (BAV2, BAV7 and KRO2) or downstream of the Baviaanskloof wilderness area with BAV7 only showing seasonal flow during winter and being the furthest upstream at the entrance to the kloof. This group also contained a sample from the Krom River, which is situated in the neighbouring kloof, as well as the most downstream Baviaanskloof site BAV2, which is just upstream of the wilderness gate towards Patensie. This site (BAV2) is on the Wit River, situated on the eastern side of Bergplaas, a natural watershed within the wilderness area. This grouping thus makes sense, with the rest of the Baviaanskloof samples situated between BAV7 and BAV2 grouping together in group three.

A clear geospatial separation between upstream inside the Baviaanskloof and downstream outside the Baviaanskloof is seen. This could be due to many reasons, one being the possible anthropogenic impacts of agricultural activities downstream of the Baviaanskloof wilderness area. The Groot River site was not far downstream of the wilderness area and not yet significantly impacted on by agricultural activities of the predominantly citrus farming active in the Patensie area. The Groot and Gamtoos rivers are wider, more turbid and situated within the upper foothills and transitional geomorphological zones, while the Baviaanskloof sites were mostly mountain streams or transitional.

The SFM diatom species distribution was found to be most closely linked to a combination between pH and salinity. This makes sense since the SFM is situated between the SECB and DC, forming a transitional region as such between the coastal and drier inland region. Therefore, while the Baviaanskloof area is sheltered, pristine and densely vegetated with steep streams feeding into a meandering clear stream at the bottom of the valley, the Groot River, and consequently also the Gamtoos River (the Gamtoos is formed by the Kouga and Groot rivers joining upstream of Patensie), is fed by water entering the kloof at the easterly end from the northern DC region. This water is well known to be very brackish and joins with the water released from the Kouga Dam to form the Gamtoos River. While there are clearly many different impacts acting on this system, the pristine less brackish waters of the Baviaanskloof River (a tributary of the Groot River) separated from the Groot and Gamtoos rivers.

With the exception of two autumn samples (BAV1_3 and BAV2_3), the Baviaanskloof samples consistently had a pH just above or well below 7. The readings were not very acidic, with a minimum pH of 5.67 measured at BAV2_2. The highest pH recorded was 8.38, also on BAV2_3. The drastic change in pH at BAV2 occurred between low summer flow and the flooding of the autumn. The Groot and Gamtoos rivers had higher pH readings with the lowest recorded during winter and the highest during spring in the Gamtoos River (Appendix 2 – Raw data: Measured water quality paramaters). While the Baviaanskloof sites showed a general increase in pH with an increase in flow, the Groot and Gamtoos showed the opposite with pH decreasing with an increase in flow. This could be due to agricultural runoff entering the river during the flood periods.

With respect to salinity, the Groot and Gamtoos Rivers had slightly higher readings that also consequently meant that the conductivity recorded on these rivers were higher than that of the Baviaanskloof sites. This could be due to the impact of irrigation in the surrounding farmed area. Higher levels of evaporation may leave higher concentrations of salts behind. In total 28 species were found to contribute to the identity of group formation in the SFM. This is higher than for the SECB (nine) and the DC (15) combined. As a transitional ecoregion situated between the other two ecoregions, it could be expected that the SFM would have a combination of species important for group identity from both. Group one had 15 species identified to be important for the community identity. Of these 15 species, seven were also indicated in one or more groups of the DC and only one species (*Fragilaria biceps*) was found to be indicated in the SECB. The remaining seven species were; *Cyclotella meneghiniana*, *Melosira varians*, *Staurosira elliptica*, *Nitzschia filiformis*, *Bacillaria paradoxa*, *Tabularia fasciculata* and *Thalassiosira weissflogi*. *Staurosira elliptica* and *Bacillaria paradoxa* is described as usually occuring in electrolyte-rich brackish water. *Cyclotella meneghiniana* is found in eutrophic electrolyte-rich waters while *Melosira varians* is often found in eutrophic slightly brackish waters. *Nitzschia filiformis* is known to occur in brackish waters with a moderate to high electrolyte content and is also tolerant to polluted conditions (Taylor et al. 2007b). *Tabularia fasciculata* is found in waters with elevated electrolyte content but is also known to have a broad ecological tolerance while *Thalassiosira weissflogi* is described as a halophilic[53] riverine species. *Thalassiosira weissflogi* was found in three of the four samples collected from the Gamtoos River. The salinity was not exceptionally high with readings of 0.6 ppt, 0.42 ppt and 0.13 ppt. However, compared to other salinity readings in this ecoregion, 0.6 was the highest measurement. The Hoogekraal River in the SECB had the highest recorded salinity reading of 1.72. The difference between rivers like the Hoogekraal, with sometimes higher salinity, was that their pH was often much lower than that of the Gamtoos River. It makes sense, that pH and salinity would be equal role-players in this ecoregion when it comes to species distribution.

It is clear, from a species point of view that group one was most closely comparable to the DC (brackish waters with elevated electrolyte content) than to the SECB. In addition, there seems to be an indication of eutrophication that could very well be possible due to the agricultural activities present, especially in the Gamtoos River valley, however, as discussed in the DC, this is not always true in brackish waters. A clear preference towards a higher pH was found to be present in group one. The Groot River originating in the DC, north of the Baviaanskloof, has a note-worthy impact on the lower reaches of this catchment with many species important for community identity overlapping with those found in the DC. The overpowering influence on diatom community composition from the one tributary (Groot River) over the other (Kouga River) could be due to the Kouga Dam upstream of the confluence and the controlled release of water from the dam instead of free flow. This highlights the importance of appropriate ecological flow assessments before, during and after development of blue-green infrastructure, since channel modification could lead to significant ecological changes, even at the base of the food web as shown here. With the added pressures of agricultural and other anthropogenic impacts associated with lower

[53] A halophilic organism is one that prefers high salt concentrations.

situated river reaches, the conservation and sustainable management of the pristine wilderness area upstream will only become increasingly important in order to ensure water security for downstream economic development and consumption.

Group two had eight species contributing to group identity. Of these eight species, three were similar to species indicated in the SECB (*Fragilaria biceps*, *Eunotia minor* and *Tabellaria flocculosa*), one was indicated also in the DC (*Nitzschia closterium*) and one species was indicated in both the DC and SECB (*Achnanthes oblongella*). The remaining species were; *Gomphonema venusta*, *Nitzschia filiformis* and *Achnanthidium eutrophilum*. *Gomphonema venusta* was described from South Africa by Passy *et al.* (1997) and was only found regularly in the northern and central parts of the country. This species is known to occur in circumneutral to weakly alkaline waters with a low to moderate electrolyte content and of oligotrophic to mesotrophic nature. *Achnanthidium eutrophilum* is found in eutrophic well-oxygenated water and only tolerant to slight to moderate pollution.

Based on the species responsible for group identity, this group has more species in common to the SECB than to the DC, however, not by a large margin. It seems to be a good mixture of different species with wide tolerances for the most part and well suited to the diverse group of sites included in this group. BAV 2 was expected to be more relatable to the rest of the inner Baviaanskloof wilderness area, but instead grouped with a site that is on the opposite end of the kloof and another in the neighbouring kloof. This eliminates the catchment signatures theory from playing a role with group formation in the SFM. Instead the species diversity and combinations seem more diverse than in the SECB and DC with localised impacts playing a larger role in community composition than could be shown in the other ecoregions. The samples indicate that these sites were in good condition, with varying pH readings recorded, but with no extremely brackish or acidic conditions present. Instead of an elevated conductivity, the presence of some species suggests low to moderate nutrient levels.

Group three had a total of 14 species contributing to group identity. Of these, five were similar than to species indicated in the DC (*Planothidium frequentissimum*, *Gomphonema parvulum*, *Cocconeis placentula*, *Nitzschia closterium* and *Navicula radiosa*), three were similar to those indicated in the SECB (*Fragilaria biceps*, *Eunotia minor* and *Planothidium engelbrechtii*) and one species was indicated in both the SECB and DC (*Achnanthes oblongella*). The remaining species were; *Achnanthidium minutissimum*, *Cymbella aspera*, *Diadesmis confervacea*, *Achnanthes subaffinis* and *Eunotia formica*.

Achnanthidium minutissimum is found in clean, well-oxygenated waters, while *Achnanthes subaffinis* is known to occur in oligotrophic waters, as is *Cymbella aspera* with the additional preference to moderate electrolyte-rich content. In contrast to this *Diadesmis confervacea* is tolerant of polluted, eutrophic and electrolyte rich waters. *Eunotia formica* has been recorded to occur in slow flowing dystrophic[54] to oligotrophic waters with moderate electrolyte content but this is not well tested at this stage.

When the frequency of occurrence data for species were examined it was found that in most cases the frequency and the abundance with which species occurred could very closely be related to the role a species played in community identity and group formation. There were, however, some exceptions to this; in Ecoregion Level I within the DC ecoregion (Table 9 see Chapter 4) it was found that firstly the most frequently occurring species (F) had a 50% frequency compared to the 81% and 79% for the SFM and SECB respectively. Secondly the most frequently occurring species did not always represent the highest percentage relative abundance (T). In the DC ecoregion *Cocconeis placentula* had a 50% frequency of occurrence but a mere 10% relative abundance. Compared to this *Cocconeis placentula* var. *lineata* had a mere 15% frequency of occurrence, but a 13% relative abundance. Another example is seen with *Gomphonema parvulum* that had a 45% frequency of occurrence but contributed a mere 3% to relative abundance in samples. This emphasises the importance of community composition in ecological reference condition descriptions and biomonitoring initiatives, regardless of the scale, instead of the presence or absence of single species.

The panhandle section of the Okavango River in Botswana showed a considerable difference in community composition when compared to the downstream Thamalakane River and the Nxamaseri Floodplain. The panhandle diatom community identity was driven by *Eunotia minor*, *Eunotia formica*, *Asterionella formosa* and *Fragilaria biceps*. *Asterionella formosa* and *Fragilaria biceps* are known to occur in meso-eutrophic rivers (Taylor et al. 2007b). The Okavango River in Botswana is not eutrophic; in fact the conductivity is extremely low. Instead the above-mentioned species are expected to have dominated the panhandle community due to the micro-environments created by papyrus beds, which trap nutrients causing a slightly elevated nutrient load. Also, some aquatic plants such as *Nymphaea* (water lily) actively pump nutrients from their roots out into the surrounding water column.

The Okavango Wetland is situated in a semi-arid region and the overall biodiversity is therefore also relatively lower than would be expected from a wetland situated on subtropical latitude (Junk et al. 2006). Like most semi-arid areas, the ecological interactions are very complex and well-equipped to deal with seasonal pulse flooding. The micro-environments

[54] Dystrophic refers to brown acidic water with low oxygen levels and often associated with low biodiversity.

created by papyrus beds and seasonally inundated island floodplains are very important for nutrient capturing and primary production in an otherwise aquatic desert. The role of this aquatic system and it associated ecosystem services for the productivity in a semi-arid region such as Botswana cannot be overstated. Upstream conservation of the panhandle section, as with upland rivers in South Africa, is very important to ensure downstream sustainability. While South Africa have few to no undisturbed lowland river ecosystems, this lowland system in Botswana remains relatively undisturbed and should be conserved for the continued functionality and integrity of the Okavango Delta downstream as well as for the continued provision of ecosystem services and goods to people directly dependant on the river system.

While the use of indicator species may have a place in certain conservation contexts, too often it is seen that too much focus is placed on one species. This overstates the importance of the individual and neglects to acknowledge the importance of biodiversity and a holistic ecosystem approach. Ecosystem functioning can be compared to a game of Jenga[55]. Many blocks can be removed and the tower will stay standing, but remove one wrong block and the whole tower comes tumbling down. This is referred to in Resource Quality Objectives as the Threshold of Potential Concern (TPC). Ecosystem functioning and integrity are not based on the survival of one species. Species going extinct is a natural part of evolution and the history of life on this planet. Instead the more important questions are why they are currently disappearing so fast and what impact the removal of a species (block) could have on the ecosystem as a whole (tower)?

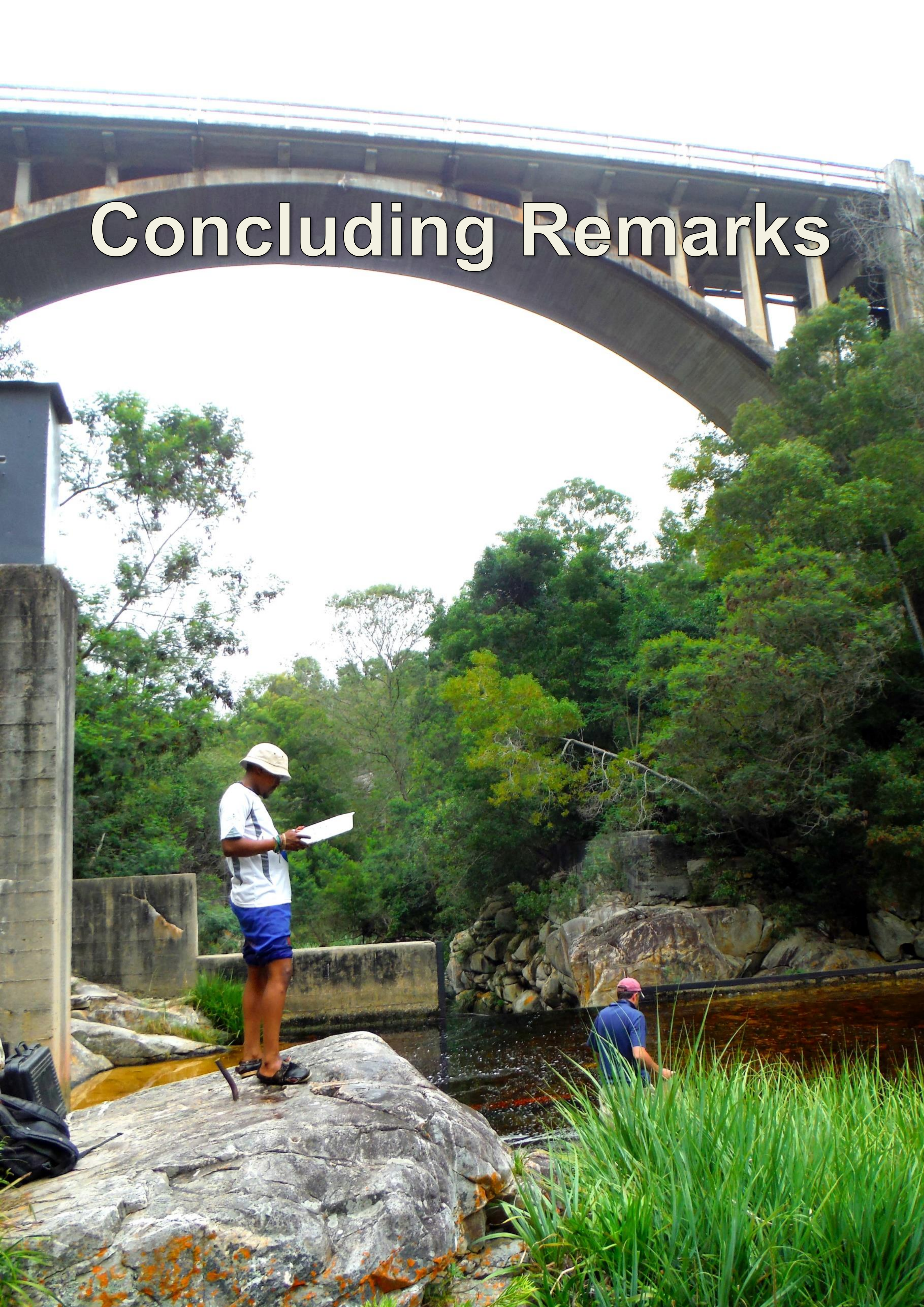
The fast-pased unregulated removal or loss of species from the biodiversity component of an ecosystem could have disastrous effects when not caused by natural drivers of evolution, but instead is accelerated by anthropogenic impacts. Currently some large scale examples of this include deforestation and other forms of habitat loss (could be the result of excessive abstraction of water from a stream/river), excessive greenhouse gas emissions, irresponsible utilisation and exploitation of natural resources such as blue-green infrastructure and pollution due to increased waste production and ineffective strategies for responsible disposal.

Diatom communities responded at various scales and across various ranges of physico-chemical properties. Diatoms could therefore be used in a wide range of studies to provide depth to our current understanding of Southern Africa's freshwater resource functioning. It is our responsibility to protect and sustainably utilise these resources for future generations

[55] Wooden rectangular blocks stacked in rows of three per row. Players remove one block per turn; the first player to remove a block which brings the tower down looses the game.

and improved human well-being, especially in areas projected to experience the most severe, direct impacts under a changing climate. While much more work is needed in order to fully develop our knowledge on the potential uses of diatoms in larger scale climate related freshwater resource monitoring initiatives, the results obtained in this study would suggest that diatoms are well capable of contributing significantly as early detectors of larger scale impacts, as well as potential indicators of resilience. No species information needs to be removed in order to produce reference conditions or statistical assessments and the combination of species was found to be more important than the presence or absence of one specific species. While small scale impact specific assessments are expected to be influenced by seasonality, as seen in the natural changes occurring even on undisturbed rivers, for larger scale monitoring initiatives diatoms can be sampled at any time of the year, as these small scale natural fluctuations should not impact the community composition and change its ecoregion identity. The possibility for such extensive and versatile application of diatoms only reaffirms the importance of their role in ecosystem functioning and why these organisms cannot be excluded or ignored when assessing or monitoring changes on freshwater ecosystems.

Concluding Remarks



CHAPTER 7 Concluding remarks

The use of the term ‘endemic diatom species’ is perhaps indefensible. Finlay et al. (2002) argued that it is impossible to prove that microorganisms do not occur elsewhere in the biosphere. Describing a diatom species as endemic is then not realistic, since diatoms are believed to have a cosmopolitan distribution although this is often contested and a subject requiring more research. Similarly it could be questionable to refer to a species as dominant in a sample when its dominance is a mere expression of community composition driven by environmental conditions. The use of dominant species only creates confusion and discrimination when describing a community identity or reference condition for a river, basin or ecoregion. Instead a dominant species should be viewed as an indicator of environmental change which is expressed as relative abundance at a specific moment in time.

An additional possible problem with the use of terminology such as dominance, is the associated elevated role or importance in the ecosystem functioning which is inaccurately associated with this terminology. Often science will try to produce evidence of dominant species, also referred to as indicator or keystone species, and their important roles in a particular ecosystem, in order to promote conservation status or sell the relevance or importance of a specific line of research. It is therefore not important what a specific diatom species is classified as, but it is far more important to uncover and understand the role this species play and how they respond as a community to larger scale ecological changes. A reference condition is very useful when monitoring the response an ecosystem has to environmental pressures, such as climate change. Without a diatom blueprint for our rivers and continued research, we are incapable of fully understanding these ecosystems, how they function and how they respond to change, thus making us incapable of preparing, mitigating and adapting to the expected impacts we will face in a climate change impacted future.

South African diatomology is experiencing a renewed revival, but many more studies and collection efforts are needed before we can fully utilise the information for decision-making purposes. The study on the Eastern and Western Cape, including the Okavango Panhandle in Botswana, produced a large quantity of data, which will now be housed at and accessioned into the South African National Diatom Collection. While the main goal was to document and assess the community composition for the Eastern Cape Rivers, it is also important that these data not become stagnant. These data could serve as the foundation for many reference conditions. It represents a snapshot of ecological health and biodiversity wealth for the Eastern and southern Cape, which could be used to inform future studies on large scale change, potential drivers and specific impacts on freshwater resources. It is recommended that these data be entered into the OMNIDIA software package and specific pollution and impact scores be generated in order to strengthen our baseline knowledge and current records on the condition of these rivers. Further studies into the specific drivers and associated community responses of ecoregions are needed to better understand the implication shifting weather patterns hold for reliable water quality and availability in future.

While diatom data has been shown to be very robust and versatile for possible application in Southern Africa as part of freshwater monitoring, much more information is needed. The differences in community composition between the three ecoregions i.e. South Eastern Coastal Belt, Southern Folded Mountains and the Drought Corridor were significant. It was also found that in the lower reaches of the Okavango River a spatial differentiation could be made between diatom communities. If all diatom species are cosmopolitan and community composition is driven solely by local environmental impacts, then why do ecoregions seem to show a significant difference in community composition? What environmental differences exist between these regions that so strongly influence diatom communities? We know from the ecoregions description process that geomorphology, climate, rainfall, geology and potential natural vegetation were used for Level I ecoregion descriptions. In other words, Ecoregion Level I is based, for the most part, on abiotic attributes and general river character, such as physico-chemical properties seems to be very responsive to this classification produced for Level I. This study was able to link these environmentally driven physico-chemical characteristics to the base of the food web in river ecosystems. What we do not know for certain, is what specific aspect within the suite of variables influencing the physico-chemical character of or within an ecoregion is the most prominent drivers of community segregation. Unfortunately, this was outside the scope of this study and more detailed chemical water analyses in future studies would greatly aid our understanding of impact driven responses at a larger geographic scale.

Now that the current study, only the second of its kind internationally and the first for Africa, has confirmed the link between diatom community composition and Level 1 ecoregion classification, it is very important if not urgent to delve deeper into the underpinning diatom-based drivers of landscape scale ecological patterns in freshwater ecosystems. It is only through increased knowledge, understanding and commitment that we will be able to ensure potable and sustainable availability of freshwater in Southern Africa in the future. Diatoms are a fundamental part of this knowledge puzzle. While one diatom species does not make the community, similarly one biotic component of the ecosystem does not make the ecosystem.

It is the hope of the author to see continued and increasing interest for diatom-based river work to be conducted in South Africa, as well as the rest of Africa. This would enable the freshwater science community to better understand these ecosystems and their associated diatom communities, predict their responses and provide decision makers, policy makers and environmental managers with the necessary information to ensure that Southern Africa, as a semi-arid region, has a future in which sustainable development is not hampered or limited by the availability of good quality freshwater. Diatoms should be an integrated part of all freshwater studies and students in the field of freshwater and ecological sciences should be well capacitated to deal with the collection and interpretation of algal and specifically diatom information.

Biodiversity is critical to ecosystem services and should be treated as such. It is good to see so many international initiatives aimed towards documenting, preserving and increasingly utilising biodiversity. While the realisation of the wealth of biodiversity is important, it is also important to remember that while it is a commodity, it can only be properly utilised when it is fully studied and understood. Cities and regions cannot be expected to sustainably manage, develop and utilise green economy when they are not fully equipped with the scientific knowledge to do so. It is the responsibility of freshwater scientists, educators and environmental assessors to work towards this common goal. It is only through increased collaboration and communication that we will truly be able to achieve improved sustainability and the consequent improvement of human livelihoods. On a continent expected to experience the largest increase in urbanisation, urban poverty and climate impacts, there has never been a more exciting time to be an environmental scientist. Nor has there ever been a more important time for inclusive interdisciplinary approaches to solving environmental problems and answering ecological questions regarding freshwater resource management.

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Appendices

APPENDICES

Appendix 1 – Raw data: Species lists by Ecoregion, River, Site and Sample

Table 17 Species list and abundance data as recorder for the South Eastern Coastal Belt ecoregion sites sampled.

Ecoregion	South Eastern Coastal Belt Ecoregion																																					
River	Bloukrans				Di ep	Elands				Goukamma				Groot				Hoogekraal				Kaaimans				Keurbooms			Knys na	Kruis			Touws		Tsitsikamma			
Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Achnanthes abundans	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthes crassa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	0	0
Achnanthes oblongella	0	0	0	6	16 8	8	0	6	0	0	0	26	0	24	4	8	1 4	84	22 7	20 3	58	71	37	69	53	12 3	31 6	22 2	180	0	0	0	5	0	32 0	28 2	30 0	25 6
Achnanthes subaffinis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	92	0	0	0	0	0	0	0	0	0	0
Achnanthes swazi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0
Achnanthidium affine	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5
Achnanthidium eutrophilum	0	0	0	0	0	0	0	24 0	0	0	0	0	0	0	0	0	3 0	0	0	0	0	0	0	16	0	0	10	0	0	0	0	0	0	0	29	0	21	0
Achnanthidium macrocephalum	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium minutissimum	16	0	0	0	0	0	3	0	0	0	0	0	0	0	12	0	0	37	0	0	0	6	0	0	0	0	0	0	7	0	0	0	1	13	0	0	0	0
Achnanthidium sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium sp3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium straubianum	0	0	0	0	0	9	0	0	0	0	0	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0	0	9	0	0	0	0	0	0	0	0	0	0
Adlafia bryophila	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora copulata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora fontinalis	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora pediculus	0	0	0	0	0	0	0	0	34	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora sp1	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0
Amphora strigosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora veneta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	South Eastern Coastal Belt Ecoregion																																					
River	Bloukrans				Diep	Elands				Goukamma				Groot				Hoogekraal				Kaaimans				Keurbooms			Knysna	Kruis			Touws		Tsitsikamma			
Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Aulacoseira granulata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Brachysira brebissonii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	4	11	0	0	0	0
Brachysira wygaschii	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Brachysira zellensis	0	0	0	0	0	0	0	0	0	0	0	13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Caloneis sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Cocconeis engelbrechtii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	15	0	0	0	0	0	0	0	0	0
Cocconeis placentula	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	97	0	6	0	0	0	0	0	0	0	15	0	0	0	0	0	6	0	0	0
Cocconeis placentula vae euglypta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	130	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cocconeis placentula var. *lineata*	0	0	0	0	79	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Craticula accomoda	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0
Craticula buderi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0	0	0	19	0
Craticula halophila	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Craticula molestiformis	0	0	0	0	0	0	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11	0
Craticula sp1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Craticula sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Ctenophora pulchella	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cyclotella meneghiniana	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	8
Cyclotella ocellata	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cymbella sp4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Denticula subtilis	0	0	0	12	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	22	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Diadesmis confervacea	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0
Diadesmis contenta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Diploneis elliptica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Diploneis oblongella	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Diploneis subovalis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema mesianum	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Encyonema minutum	3	0	0	1	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	6	22	0	0	4	0
Encyonema neogracile	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	1	4	0	0	0	0	0	0	0	0	0	0	0
Encyonema silesiacum	0	0	0	0	0	1	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema sp3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	South Eastern Coastal Belt Ecoregion																																					
River	Bloukrans				Diep	Elands				Goukamma				Groot				Hoogekraal				Kaaimans				Keurbooms			Knysna	Kruis			Touws		Tsitsikamma			
Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Encyonopsis leei var. *sinensis*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	62	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonopsis raytonensis	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Eolimna minima	0	0	0	0	27	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	16	0	0	0	0	23	0	0	0	0	0	0	0	0	0	0	0	58
Epithemia adnata	0	0	0	0	56	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Epithemia sorex	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia bilunaris	0	0	0	0	0	0	0	0	0	0	0	0	0	16	2	0	23	0	0	0	0	0	4	3	0	0	0	0	29	0	0	0	8	11	0	0	0	0
Eunotia exigua	0	0	0	0	0	0	0	0	0	0	2	0	4	21	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia flexuosa	0	0	8	0	0	1	17	6	0	0	0	6	0	0	11	23	0	10	0	0	31	47	0	0	0	2	0	2	0	2	0	6	13	0	0	2	0	11
Eunotia formica	0	0	0	0	0	0	0	0	65	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	2	0	77	0	0	0	0	1	0	0	0	0
Eunotia incisa	0	0	37	0	0	0	0	49	110	0	0	66	390	153	235	81	33	0	0	6	0	310	260	230	70	123	0	0	0	0	0	17	0	164	32	0	0	0
Eunotia minor	231	430	340	9	0	161	315	111	43	420	420	240	0	11	33	155	82	213	22	0	19	0	0	95	316	0	0	83	66	218	60	301	97	74	0	54	24	17
Eunotia pectinalis	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia pectinalis var. *undulata*	0	0	0	0	0	0	38	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0	0
Eunotia rhomboidea	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0	0	17	0	0	0	0	0	0	54	0	75	0	0	0	0	0	0
Eunotia sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	26	0	0	0	0	0	0	0	0	0	0	0
Fallacia sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria biceps	0	0	0	51	15	0	0	0	0	0	0	0	0	3	2	0	0	6	0	20	1	0	0	0	0	1	3	0	0	0	0	0	0	2	0	64	6	0
Fragilaria capucina	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	9	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	1	0	0
Fragilaria capucina var. *rumpens*	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	21	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria capucina var. *vaucheriae*	0	0	0	0	15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria ulna	0	0	0	1	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	42
Frustulia crassinervia	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	29	0	0	0	0	0	0	0	0	0	56	0	0	0	0	0	0	0	0	0	0	0
Frustulia rostrata	0	0	0	0	0	0	0	34	2	0	0	0	0	0	0	0	0	0	0	0	0	0	79	0	7	0	0	0	12	0	0	0	0	0	0	0	0	0
Frustulia saxonica	7	2	43	35	0	19	12	6	37	0	0	22	0	69	54	89	1	8	2	0	9	0	4	52	0	129	53	23	0	128	0	12	217	136	0	4	0	0
Frustulia sp3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Frustulia sp4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Frustulia vulgaris	22	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema acuminatum	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema affine lagenula	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

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River	Bloukrans				Diep	Elands				Goukamma				Groot				Hoogekraal				Kaaimans				Keurbooms			Knysna	Kruis			Touws		Tsitsikamma			
Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Gomphonema angustum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0
Gomphonema capitatum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0
Gomphonema parvulum	0	0	0	102	58	56	0	0	0	0	0	0	0	0	0	0	0	0	0	0	207	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0
Gomphonema sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyrosigma scalproides	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Lemnicola hungarica	0	0	0	0	0	0	0	0	0	0	0	86	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Luticola goeppertiana	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Melosira varians	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	4	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	10
Navicula cryptotenelloides	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula erifuga	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Navicula longicephala	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula radiosa	0	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	57	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula recens	0	0	0	42	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	22	0	0	2
Navicula rhynchocephala	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula riediana	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	27	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	7
Navicula sp10	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp12	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	33	0	0	0	0	0
Navicula sp13	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	27	0	0	0	0	0
Navicula sp20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	8	0
Navicula sp21	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0
Navicula tripunctata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	51	11	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula veneta	0	0	0	17	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicymbula pusilla	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	19	0	0	0	0	0	0	0	0	0
Nitzschia acicularis	0	0	0	0	1	0	0	0	0	0	0	0	0	0	9	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia aurariae	0	0	0	0	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia capitellata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia clausii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	1
Nitzschia closterium	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	15	0	0	0	22	0	0	17	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia dissipata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	1	0	0	0	0	0	0	0	0	0

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Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Nitzschia filiformis	0	0	0	0	0	0	0	3	0	0	0	0	0	0	4	0	0	8	0	30	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	2	22	0
Nitzschia gracilis	0	0	0	1	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia iremissa	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia liebertruthii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Nitzschia linearis	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia littorea	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia palea	0	0	0	25	0	0	0	0	0	0	0	0	0	6	0	0	51	0	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	8	0	11
Nitzschia pusilla	0	0	0	58	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0	0	0	0	0	0	0
Nitzschia recta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	18	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sigma	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	9
Nitzschia sp13	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0
Nitzschia sp19	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0
Nitzschia sp20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0
Nitzschia sp21	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp22	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp23	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	26	0	0	0	0	0	0	0	0	0
Pinnularia gibba	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia microstauron var. *rostrata*	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	36	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia sp1	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
Pinnularia subcapitata	14	0	0	0	0	0	0	1	0	0	0	0	0	6	1	0	0	1	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia viridiformis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Planothidium engelbrechtii	0	0	8	65	1	0	0	0	0	0	0	0	0	0	0	0	0	23	36	10	0	0	0	0	0	0	0	0	6	24	0	0	13	0	0	4	0	0
Planothidium frequentissimum	0	0	0	0	3	0	37	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Planothidium rostratum	95	0	0	0	0	147	0	0	145	2	0	0	0	15	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pleurosigma sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0
Psammothidium abundans	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	South Eastern Coastal Belt Ecoregion																																					
River	Bloukrans				Diep	Elands				Goukamma				Groot				Hoogekraal				Kaaimans				Keurbooms			Knysna	Kruis			Touws		Tsitsikamma			
Species	BLO1_1	BLO1_2	BLO1_3	BLO1_4	DIP1_1	ELN1_1	ELN1_2	ELN1_3	ELN1_4	GOU1_1	GOU1_2	GOU1_3	GOU1_4	GRO1_1	GRO1_2	GRO1_3	GRO1_4	HOE1_1	HOE1_2	HOE1_3	HOE1_4	KAI1_1	KAI1_2	KAI1_3	KAI1_4	KEU1_1	KEU1_2	KEU1_3	KNY1_3	KRU1_1	KRU1_2	KRU1_3	TOU1_1	TOU1_3	TSI1_1	TSI1_2	TSI1_3	TSI1_4
Pseudostaurosira brevistriata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0
Rhopalodia operculata	27	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0
Sellophora pupula	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0
Simonsenia delognei	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Stauroneis anceps	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Staurosira elliptica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	6	0	0	0	0	3	0	0	0	0	0	0	0	0	0
Surirella angusta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Surirella sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tabellaria flocculosa	0	0	0	0	0	0	1	1	0	0	0	3	0	84	47	53	34	0	1	0	0	0	0	4	0	36	9	12	7	0	0	0	29	28	0	1	1	0
Tabularia fasciculata	1	0	0	33	0	4	0	0	0	0	0	0	6	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tabularia sp2	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella apiculata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella hungarica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella levidensis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0
Tryblionella sp3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0

Table 18 Species list and abundance data as recorder for the Southern Folded Mountains ecoregion sites sampled.

Ecoregion	Southern Folded Mountains Ecoregion																										
River	Bokkraal			Wit			Baviaanskloof								Gamtoos				Groot				Krom			
Species	BAV1_2	BAV1_3	BAV1_4	BAV2_1	BAV2_2	BAV2_3	BAV3_2	BAV4_2	BAV4_4	BAV5_2	BAV5_4	BAV6_2	BAV6_4	BAV7_4	GAM1_1	GAM1_2	GAM1_3	GAM1_4	GRT1_1	GRT1_2	GRT1_3	GRT1_4	KRO1_1	KRO2_2	KRO2_3	KRO2_4
Achnanthes crassa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	20	0	0	0	0	0	0
Achnanthes linearoides	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthes oblongella	67	10	41	27	44	22	26	22	7	19	0	0	0	0	0	0	8	0	2	0	0	0	0	0	18	8
Achnanthes sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	0	0	0	0	0
Achnanthes standerii	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthes subaffinis	0	0	9	0	0	0	0	31	32	0	0	8	0	0	0	0	0	0	0	0	0	0	16	3	93	9

Ecoregion	Southern Folded Mountains Ecoregion																									
River	**Bokkraal**			**Wit**			**Baviaanskloof**								**Gamtoos**				**Groot**				**Krom**			
Species	**BAV1_2**	**BAV1_3**	**BAV1_4**	**BAV2_1**	**BAV2_2**	**BAV2_3**	**BAV3_2**	**BAV4_2**	**BAV4_4**	**BAV5_2**	**BAV5_4**	**BAV6_2**	**BAV6_4**	**BAV7_4**	**GAM1_1**	**GAM1_2**	**GAM1_3**	**GAM1_4**	**GRT1_1**	**GRT1_2**	**GRT1_3**	**GRT1_4**	**KRO1_1**	**KRO2_2**	**KRO2_3**	**KRO2_4**
Achnanthes swazi	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	56	0	8	1
Achnanthidium affine	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	156	0	0
Achnanthidium biasolettianum	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium eutrophilum	0	0	0	0	0	52	0	0	0	0	16	0	0	0	0	46	2	0	0	4	0	0	0	0	0	41
Achnanthidium exiguum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	6	0	0	0	0	0	0	0
Achnanthidium macrocephalum	0	0	0	0	0	0	0	0	0	0	31	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium minutissimum	4	2	56	0	11	0	197	59	0	31	0	0	0	0	145	0	0	0	0	0	20	0	21	0	0	0
Adlafia bryophila	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora normannii	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora pediculus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	3	0	0	8	0	0	0	0	0	0	0
Asterionella formosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	7	3	0	0
Aulacoseira subartica f. subborealis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	174	0	0	0
Bacillaria paradoxa	0	1	0	0	0	0	0	1	0	0	0	5	4	0	3	7	4	0	0	0	13	0	0	0	0	0
Brachysira brebissonii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	60	0	0	0
Capartogramma crucicula	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Cocconeis engelbrechtii	0	0	0	0	0	0	0	0	11	0	0	0	102	0	0	0	0	0	28	0	0	0	0	0	0	0
Cocconeis pediculus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0	0	1	0	0	0	0	0	0
Cocconeis placentula	0	38	57	4	0	2	4	3	3	0	0	0	170	0	190	0	152	68	22	0	16	0	0	0	0	0
Cocconeis placentula var. *euglypta*	0	0	0	0	0	0	0	0	0	0	0	87	0	0	0	0	0	0	0	0	0	1	0	0	0	0
Cocconeis placentula var. *lineata*	61	0	0	0	16	0	0	0	0	0	1	147	0	0	0	164	0	0	0	41	0	8	0	2	0	0
Craticula buderi	0	0	16	0	0	0	8	0	0	0	0	0	0	0	34	23	0	0	0	0	0	0	0	0	0	0
Craticula molestiformis	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Craticula vixnegligenda	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	8	32	0	0
Cyclostephanos dubius	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Cyclotella meneghiniana	0	0	0	0	0	0	0	2	0	2	0	0	0	0	4	27	19	7	6	4	0	3	0	0	2	6
Cymatopleura solea	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0
Cymbella aspera	0	2	0	0	0	0	9	9	17	11	0	0	0	0	0	0	0	0	0	1	3	1	0	0	0	0
Cymbella sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Cymbella sp3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Cymbella tumida	0	0	0	0	0	0	0	0	0	0	0	0	2	0	2	0	0	0	0	0	0	0	0	0	0	0
Cymbopleura naviculiformis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1

Ecoregion	Southern Folded Mountains Ecoregion																									
River	**Bokkraal**			**Wit**			**Baviaanskloof**								**Gamtoos**				**Groot**				**Krom**			
Species	**BAV1_2**	**BAV1_3**	**BAV1_4**	**BAV2_1**	**BAV2_2**	**BAV2_3**	**BAV3_2**	**BAV4_2**	**BAV4_4**	**BAV5_2**	**BAV5_4**	**BAV6_2**	**BAV6_4**	**BAV7_4**	**GAM1_1**	**GAM1_2**	**GAM1_3**	**GAM1_4**	**GRT1_1**	**GRT1_2**	**GRT1_3**	**GRT1_4**	**KRO1_1**	**KRO2_2**	**KRO2_3**	**KRO2_4**
Diadesmis confervacea	0	0	0	0	0	0	26	35	0	84	55	0	0	0	0	1	0	0	0	2	0	0	0	0	0	0
Diatoma vulgaris	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0	0	0	0	0	0	0	0	0	0	0
Diploneis elliptica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	3	0	0	0	0	0	0	0	0	0
Diploneis oblongella	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	19	0	0	0	0	0	0	0	0	0	0
Diploneis subovalis	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Discotella stelligera	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	100	0	0	0
Encyonema mesianum	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0
Encyonema minutum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0	0	0	32	1
Encyonema neogracile	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Encyonema silesiacum	0	0	0	0	0	0	0	0	0	0	0	0	0	29	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema sp1	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema sp2	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema sp4	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonema ventricosum	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0
Encyonopsis leei var. *sinensis*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Encyonopsis subminuta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	112	0
Eolimna minima	33	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	28	0
Eolimna subminuscula	0	0	0	28	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0
Epithemia adnata	62	60	11	0	1	0	0	0	0	0	0	0	21	0	0	0	0	0	6	8	26	0	0	0	0	0
Epithemia sorex	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	7	7	0	1	0	0	0	0
Eunotia flexuosa	0	0	0	0	14	0	0	7	0	37	37	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0
Eunotia formica	0	0	40	0	0	0	1	0	42	0	0	0	28	0	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia incisa	0	0	25	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	28	0
Eunotia minor	0	0	0	0	26	15	6	33	45	8	36	0	0	2	0	0	0	0	0	8	1	9	0	17	167	34
Eunotia pectinalis	0	0	0	0	0	0	0	0	15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia rhomboidea	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria biceps	0	45	33	170	206	150	14	55	38	212	31	0	3	192	4	0	0	101	0	21	14	12	4	28	2	180
Fragilaria capucina	67	36	0	0	0	2	0	22	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria capucina var. *rumpens*	0	0	23	54	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria capucina var. *vaucheriae*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	105	0	0	0	0	0	0	17
Fragilaria crotonensis	0	0	0	0	0	0	0	0	0	0	34	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	Southern Folded Mountains Ecoregion																									
River	**Bokkraal**			**Wit**			**Baviaanskloof**								**Gamtoos**				**Groot**				**Krom**			
Species	**BAV1_2**	**BAV1_3**	**BAV1_4**	**BAV2_1**	**BAV2_2**	**BAV2_3**	**BAV3_2**	**BAV4_2**	**BAV4_4**	**BAV5_2**	**BAV5_4**	**BAV6_2**	**BAV6_4**	**BAV7_4**	**GAM1_1**	**GAM1_2**	**GAM1_3**	**GAM1_4**	**GRT1_1**	**GRT1_2**	**GRT1_3**	**GRT1_4**	**KRO1_1**	**KRO2_2**	**KRO2_3**	**KRO2_4**
Fragilaria parasitica	0	0	0	0	0	0	0	0	112	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria parasitica var. *constricta*	0	0	0	0	0	0	0	70	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria tenera	0	0	0	0	0	0	0	0	0	19	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria ulna	32	17	0	0	0	0	1	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0
Frustulia saxonica	3	2	21	0	0	6	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	14	12
Frustulia sp1	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Frustulia sp2	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Frustulia vulgaris	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	1
Geissleria decussis	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema acuminatum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Gomphonema affine	0	0	0	0	0	0	12	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0	0	0	1	7
Gomphonema affine gracile	0	0	0	0	0	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0	0	0	24	0	0
Gomphonema affine lagenula	0	36	0	8	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	19
Gomphonema angustum	0	98	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema clavatum	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema insigne	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema italicum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	54	0	0	0	0	0	0	0	0	0
Gomphonema laticollum	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema minutum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	46	9	0	0	0	0	0	0	0	0	0	0
Gomphonema parvulum	0	24	8	0	9	0	119	0	27	0	0	0	0	148	0	24	7	0	2	34	0	3	0	0	0	0
Gomphonema parvulum var. *lagenula*	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	182	0	0
Gomphonema pseudoaugur	0	0	0	0	0	0	0	0	0	0	77	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema pumilum	0	0	0	0	0	0	0	0	0	0	0	0	44	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema sp1	0	0	0	0	0	0	0	28	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema sp3	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema truncatum	0	8	0	1	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema venusta	102	0	0	30	64	33	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyrosigma acuminatum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0
Gyrosigma attenuatum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
Gyrosigma rautenbachiae	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	3	0	0	0	1
Gyrosigma sp1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	Southern Folded Mountains Ecoregion																									
River	Bokkraal			Wit			Baviaanskloof								Gamtoos				Groot				Krom			
Species	BAV1_2	BAV1_3	BAV1_4	BAV2_1	BAV2_2	BAV2_3	BAV3_2	BAV4_2	BAV4_4	BAV5_2	BAV5_4	BAV6_2	BAV6_4	BAV7_4	GAM1_1	GAM1_2	GAM1_3	GAM1_4	GRT1_1	GRT1_2	GRT1_3	GRT1_4	KRO1_1	KRO2_2	KRO2_3	KRO2_4
Gyrosigma sp2	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyrosigma sp3	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyrosigma sp4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0
Hantzschia amphyoxys	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0
Lemnicola hungarica	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Luticola acidoclinata	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Luticola kotschyi	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Melosira varians	0	0	0	0	0	0	0	10	7	0	38	1	0	0	8	41	39	11	0	0	0	6	0	0	0	0
Navicula angusta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	16
Navicula arvensis var. maior	0	0	0	20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula capitatoradiata	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula cincta	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula cryptocephala	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	8	0
Navicula cryptonella	0	0	11	0	0	0	0	0	0	0	0	0	2	0	10	0	0	0	0	0	0	0	0	0	0	0
Navicula germainii	0	0	0	0	0	0	0	0	16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula radiosa	1	12	9	1	0	0	1	0	0	3	0	23	0	0	0	0	0	8	35	2	8	0	0	0	3	0
Navicula recens	0	0	0	0	4	0	0	0	0	0	0	0	0	2	0	0	6	0	0	0	32	0	0	0	0	4
Navicula reichardtiana	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0
Navicula ranomafenensis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	19	0	0	0
Navicula rhynchocephala	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp1	0	0	0	0	0	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp16	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp18	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	19	0	0	0
Navicula sp2	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp22	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	0
Navicula sp6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	0	0	0	0
Navicula sp7	0	0	0	0	0	0	0	0	0	0	0	26	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp8	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Navicula veneta	0	0	0	0	0	0	0	0	0	0	20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula zanonii	0	0	0	0	0	0	0	0	47	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	Southern Folded Mountains Ecoregion																									
River	Bokkraal			Wit			Baviaanskloof								Gamtoos				Groot				Krom			
Species	BAV1_2	BAV1_3	BAV1_4	BAV2_1	BAV2_2	BAV2_3	BAV3_2	BAV4_2	BAV4_4	BAV5_2	BAV5_4	BAV6_2	BAV6_4	BAV7_4	GAM1_1	GAM1_2	GAM1_3	GAM1_4	GRT1_1	GRT1_2	GRT1_3	GRT1_4	KRO1_1	KRO2_2	KRO2_3	KRO2_4
Nitzschia aurariae	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia closterium	0	13	12	83	0	0	0	12	0	0	13	0	0	61	0	0	0	0	0	0	0	3	0	0	0	62
Nitzschia desertorum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0
Nitzschia dissipata var. media	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0
0N0itzschia etoshensis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5	0	0	0	0	0	0	0
Nitzschia filiformis	0	17	16	0	0	21	0	0	0	0	0	0	0	36	7	0	0	0	0	38	101	26	0	0	0	10
Nitzschia frustulum	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	21	0	0	0	0	0	0	0	0	0	0
Nitzschia liebertruthii	0	0	0	0	0	19	4	0	0	0	0	0	0	0	1	0	4	0	0	0	0	0	0	0	0	0
Nitzschia nana	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Nitzschia palea	0	3	0	0	0	0	0	3	21	0	0	78	0	0	1	0	0	0	0	0	11	9	0	0	0	2
Nitzschia sigma	0	0	0	0	0	0	0	11	0	0	0	0	0	22	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp14	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp17	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0
Nitzschia sp18	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Nitzschia sp3	0	0	0	0	0	0	0	0	0	0	0	13	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Nitzschia sp5	8	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp6	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp9	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sublinearis	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia umbonata	0	0	1	0	0	0	0	0	0	2	0	0	0	1	0	0	0	0	0	2	0	0	0	0	0	1
Nitzscia communis	0	0	0	0	0	42	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Ntzschia sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Orthoseira sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0
Oxyneis sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
Pinnularia sp3	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia subcapitata	0	2	0	0	0	2	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia viridis	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Placoneis dicephala	0	0	0	0	0	20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Placoneis elginensis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0

Ecoregion	Southern Folded Mountains Ecoregion																									
River	Bokkraal			Wit			Baviaanskloof								Gamtoos				Groot				Krom			
Species	BAV1_2	BAV1_3	BAV1_4	BAV2_1	BAV2_2	BAV2_3	BAV3_2	BAV4_2	BAV4_4	BAV5_2	BAV5_4	BAV6_2	BAV6_4	BAV7_4	GAM1_1	GAM1_2	GAM1_3	GAM1_4	GRT1_1	GRT1_2	GRT1_3	GRT1_4	KRO1_1	KRO2_2	KRO2_3	KRO2_4
Placoneis placentula	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Placoneis sp1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Planothidium engelbrechtii	0	6	0	0	0	4	18	0	33	0	0	5	0	0	0	1	0	0	0	0	0	0	0	0	0	0
Planothidium frequentissimum	0	0	20	0	0	0	38	7	23	18	0	0	0	77	0	0	0	0	0	0	0	4	0	0	0	0
Planothidium rostatrum	0	0	0	0	0	0	0	0	0	0	39	0	0	0	0	0	0	0	0	63	32	0	0	0	0	0
Pleurosigma salinarum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0
Pleurosigma sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pseudostaurosira brevistriata	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	33	0	0	115	0	0	0	0	0	0	0
Reimeria sinuata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0
Rhopalodia gibba	0	0	0	0	0	0	0	0	0	0	0	10	0	1	0	0	0	0	2	4	0	0	0	0	0	0
Rhopalodia gibberula	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	1	0	0	0	0	0	0
Rhopalodia operculata	0	1	0	0	8	38	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Rhopalodia sp1	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sellophora pupula	0	0	0	0	0	2	1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sellophora seminulum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11	0	0	0	0	0	0	0
Stauroneis smithii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	14	0
Stauroneis sp1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Staurosira construens	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0
Staurosira elliptica	0	0	0	0	0	0	0	0	53	2	1	15	0	0	9	56	87	234	159	158	210	338	0	0	0	0
Staurosirella pinnata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0	0	0	0	0	0	0
Stephanodiscus sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
Surirella angusta	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Surirella sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tabellaria flocculosa	0	1	0	4	17	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	2	0	2	16
Tabularia fasciculata	0	0	0	0	0	15	0	0	0	0	0	0	0	0	6	0	32	0	6	0	0	0	0	0	2	12
Tabularia sp1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	11	0	0	0	0	0	0	0	0	0	0
Thalassiosira weissflogi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	5	4	0	0	0	0	0	0	0	0
Tryblionella apiculata	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella calida	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella hungarica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	5	0	0	0	0
Tryblionella levidensis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0	0	14	2	0	0	0	0	0

Table 19 Species list and abundance data as recorder for the Drought Corridor ecoregion sites sampled

Ecoregion	Drought Corridor Ecoregion																			
River	Great Fish		Small Fish						Koonap				Volkers				Sondags	Tyume		
Species	GFI1_1	GFI1_2	KFI1_1	KFI1_2	KFI1_3	KFI1_4	KFI2_1	KFI2_2	KOO1_1	KOO1_2	KOO1_3	KOO1_4	SON1_1	SON1_2	SON1_3	SON1_4	SON2_3	TUY1_2	TUY1_3	TUY1_4
Achnanthes crassa	0	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0	0	0	0
Achnanthes oblongella	2	20	0	11	0	0	0	2	1	0	0	0	0	0	36	0	1	7	10	21
Achnanthes swazi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	22	0
Achnanthidium affine	0	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0
Achnanthidium eutrophilum	0	0	0	0	0	0	93	0	0	0	0	0	0	0	0	0	0	0	0	0
Achnanthidium exiguum	0	0	0	0	0	0	0	0	0	0	0	17	0	0	0	0	0	0	0	0
Achnanthidium minutissimum	0	0	0	4	0	0	0	0	153	0	0	0	0	0	42	0	1	0	0	21
Amphora coffeaeformis	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
Amphora inariensis	0	0	0	0	0	0	2	0	1	0	0	0	0	0	0	0	0	0	0	0
Amphora montana	0	0	0	0	0	0	0	0	0	0	0	7	0	0	0	0	0	0	0	0
Amphora ovalis	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
Amphora pediculus	46	63	0	0	0	0	0	14	0	0	0	0	0	0	0	0	1	0	0	0
Aulacoseira granulata	0	0	0	0	0	0	0	0	0	0	0	25	55	0	0	0	0	0	0	0
Bacillaria paradoxa	0	0	0	0	0	0	1	0	0	0	4	0	0	0	0	0	0	0	0	0
Caloneis molaris	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Caloneis sp1	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0
Cocconeis engelbrechtii	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
Cocconeis pediculus	0	0	0	0	0	0	19	9	7	0	0	17	0	0	0	6	0	0	0	0
Cocconeis placentula	0	157	0	0	0	0	6	0	32	250	240	0	2	4	51	18	0	0	0	33
Cocconeis placentula var. *euglypta*	200	0	0	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0	0
Cocconeis placentula var. *lineata*	0	0	0	0	0	0	0	0	0	0	0	66	48	0	0	0	0	422	312	173
Craticula accomoda	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0	0	0	0	0
Craticula ambigua	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cyclostephanos dubius	0	1	0	0	0	0	0	0	0	0	0	0	25	0	0	0	0	0	0	0
Cyclotella meneghiniana	0	0	0	0	0	0	4	0	0	0	0	0	0	0	6	0	0	0	0	0
Cymbella neocistula	0	0	0	0	0	1	0	0	0	0	3	0	0	0	0	0	0	0	0	0
Cymbella sp2	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0
Cymbella tumida	0	0	0	0	0	0	0	0	0	0	4	46	0	0	0	0	0	0	0	0
Cymbella turgidula	0	0	0	0	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0

Ecoregion	Drought Corridor Ecoregion																			
River	Great Fish		Small Fish						Koonap				Volkers				Sondags	Tyume		
Species	GFI1_1	GFI1_2	KFI1_1	KFI1_2	KFI1_3	KFI1_4	KFI2_1	KFI2_2	KOO1_1	KOO1_2	KOO1_3	KOO1_4	SON1_1	SON1_2	SON1_3	SON1_4	SON2_3	TUY1_2	TUY1_3	TUY1_4
Diadesmis contenta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Discotella stelligera	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Encyonema minutum	0	0	0	0	0	0	0	0	0	0	0	4	2	0	0	0	0	0	0	0
Encyonema sp5	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Encyonopsis leei var. *sinensis*	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0
Encyonopsis subminuta	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0
Eolimna subminuscula	0	10	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0
Epithemia adnata	0	0	32	253	1	55	7	2	2	0	0	18	0	0	0	6	0	0	0	0
Epithemia sorex	0	0	10	43	0	12	81	119	0	0	0	0	0	0	0	0	0	0	0	0
Eunotia minor	0	8	0	0	0	0	0	0	0	0	0	0	0	4	4	0	0	0	0	0
Eunotia sp1	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Fallacia sp1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
Fragilaria biceps	1	0	1	0	1	0	10	0	0	0	6	32	0	0	5	0	1	0	0	0
Fragilaria capucina var. *vaucheriae*	1	0	0	0	0	0	0	0	0	0	0	22	0	0	0	0	0	0	6	108
Fragilaria parasitica var. *constricta*	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
Fragilaria ulna	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	16	0	0
Frustulia saxonica	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gomphonema insigne	0	0	0	0	0	0	0	0	107	0	0	0	0	0	0	0	0	0	0	0
Gomphonema parvulum	2	0	0	7	0	0	6	57	0	0	0	24	10	58	57	0	0	0	0	6
Gomphonema venusta	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Gyrosigma rautenbachiae	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyrosigma scalproides	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	0
Luticola goeppertiana	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Melosira varians	0	1	0	0	1	0	4	0	0	0	8	75	0	0	0	0	0	0	0	0
Navicula antonii	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula capitatoradiata	0	0	0	0	0	0	34	0	0	0	0	0	0	0	0	19	0	0	0	0
Navicula cryptonella	0	0	0	0	0	0	0	20	0	0	0	0	0	0	0	0	0	0	0	0
Navicula erifuga	0	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula libonensis	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula radiosa	0	0	12	0	0	0	43	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula recens	0	0	12	0	0	0	0	0	0	0	38	0	0	0	0	0	0	0	0	0

Ecoregion	Drought Corridor Ecoregion																			
River	**Great Fish**		**Small Fish**						**Koonap**				**Volkers**				**Sondags**	**Tyume**		
Species	**GFI1_1**	**GFI1_2**	**KFI1_1**	**KFI1_2**	**KFI1_3**	**KFI1_4**	**KFI2_1**	**KFI2_2**	**KOO1_1**	**KOO1_2**	**KOO1_3**	**KOO1_4**	**SON1_1**	**SON1_2**	**SON1_3**	**SON1_4**	**SON2_3**	**TUY1_2**	**TUY1_3**	**TUY1_4**
Navicula riediana	0	0	0	0	0	13	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula rostellata	0	0	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0
Navicula sp14	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Navicula sp15	0	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula sp17	0	0	0	0	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0
Navicula sp9	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula symmetrica	0	0	0	0	0	0	0	0	0	0	24	0	0	0	0	0	0	0	0	0
Navicula tripunctata	14	0	0	0	0	0	49	0	0	0	0	0	0	0	0	0	0	0	0	0
Navicula veneta	0	0	0	0	0	0	0	0	0	0	0	0	0	57	0	0	0	0	0	0
Navicula zanonii	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0
Nitzschia acicularis	0	0	0	0	0	0	0	0	0	0	0	22	0	0	0	0	0	0	0	0
Nitzschia archibaldii	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia aurariae	0	0	0	0	0	0	0	38	0	0	1	0	0	0	97	0	0	0	0	0
Nitzschia capitellata	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12
Nitzschia closterium	0	0	21	0	0	82	0	26	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia dissipata	2	0	0	0	0	0	39	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia filiformis	4	2	1	0	0	0	13	0	0	0	25	24	0	0	0	0	0	0	0	0
Nitzschia intermedia	0	0	0	0	0	0	12	0	0	0	0	1	0	0	0	0	0	0	0	0
Nitzschia iremissa	0	0	0	0	0	0	0	58	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia liebertruthii	0	0	3	0	0	0	0	4	0	0	0	0	1	0	0	0	0	0	0	0
Nitzschia linearis	0	0	0	0	0	0	21	0	9	0	0	0	0	0	0	0	0	0	0	0
Nitzschia linearis var. *subtilis*	0	0	0	0	0	72	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia microcephala	51	0	22	0	0	0	11	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia palea	0	0	6	0	0	47	0	0	1	0	3	0	0	270	94	39	0	0	0	0
Nitzschia sp11	0	0	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp12	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp7	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nitzschia sp8	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Nitzschia umbonata	0	0	0	0	0	0	0	0	0	1	0	0	15	0	0	60	1	0	0	0
Pinnularia divergens	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pinnularia viridiformis	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0

Ecoregion	Drought Corridor Ecoregion																			
River	**Great Fish**		**Small Fish**						**Koonap**				**Volkers**				**Sondags**	**Tyume**		
Species	**GFI1_1**	**GFI1_2**	**KFI1_1**	**KFI1_2**	**KFI1_3**	**KFI1_4**	**KFI2_1**	**KFI2_2**	**KOO1_1**	**KOO1_2**	**KOO1_3**	**KOO1_4**	**SON1_1**	**SON1_2**	**SON1_3**	**SON1_4**	**SON2_3**	**TUY1_2**	**TUY1_3**	**TUY1_4**
Planothidium engelbrechtii	0	0	0	0	0	0	8	0	0	0	0	0	0	0	0	0	0	0	0	0
Planothidium frequentissimum	0	0	1	13	0	1	0	0	0	0	0	0	0	0	0	0	0	0	44	42
Pleurosigma elongatum	0	0	13	0	0	0	17	0	0	0	0	0	0	0	0	0	0	0	0	0
Pleurosigma salinarum	0	0	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0
Pseudostaurosira brevistriata	0	0	0	0	0	0	0	0	0	0	0	0	6	0	0	0	0	0	0	0
Reimeria sinuata	0	0	0	0	0	0	0	0	0	0	0	0	57	0	0	0	0	0	0	0
Reimeria uniseriata	61	51	0	0	0	0	0	6	86	155	66	34	0	21	36	130	0	0	0	0
Rhopalodia gibba	3	0	260	92	1	206	27	43	0	0	0	1	0	0	0	73	0	0	11	0
Rhopalodia gibberula	0	0	0	8	0	0	2	1	0	0	0	0	0	0	0	0	0	0	0	0
Rhopalodia operculata	0	0	57	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Simonsenia delognei	0	0	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Staurosira elliptica	0	185	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Stephanodiscus agassizensis	23	0	0	0	0	0	0	0	0	0	0	0	187	0	0	60	0	0	0	0
Surirella brebissonii	0	0	0	0	0	0	6	0	0	0	0	0	0	0	0	0	0	0	0	0
Tabellaria flocculosa	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0	0	0
Tabularia fasciculata	7	0	0	13	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0
Thalassiosira weissflogi	39	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella apiculata	0	0	0	0	0	0	0	6	2	0	8	0	1	0	0	0	0	0	0	0
Tryblionella coarctata	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Tryblionella hungarica	0	0	1	0	0	0	5	0	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella levidensis	0	0	0	0	0	0	12	14	0	0	0	0	0	0	0	0	0	0	0	0
Tryblionella sp1	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Appendix 2 – Raw data: Measured water quality paramaters

Table 20 Water quality paramaters measured at time of diatom sampling

Site code	BLO1	ELN1	KRU1	GRO1	HOE1	GOU1	KAI1	TOU1	KEU1	TSI1	KRO1	GAM1	GRT1	BAV1	BAV2	SON1	KFI1	KFI2	GFI1	KOO1	BAV3	BAV4	BAV5	BAV6	BAV7	TYU1	KRO2	KNY1	SON2
Date Spring Sampling	18/10/2014	18/10/2014	18/10/2014	18/10/2014	19/10/2014	19/10/20147	20/10/2014	20/10/2014	20/10/2014	21/10/2014	21/10/2014	22/10/2014	22/10/2014	22/10/2014	22/10/2014	24/10/2014	24/10/2014	24/10/2014	24/10/2014	25/10/2014	No Sample	No Sample	No Sample	No Sample	No Sample	No Sample	No Sample	No Sample	No Sample
Oxygen (ppm)	10.76	9.56	9.67	8.33	9.22	9.86	9.39	8.62	9.04	8.26	6.57	8.78	8.82	4.68	7.87	8.2	7.69	9.36	8.58	6.67									
Oxygen (%)	102	95.8	96.5	88.9	91	102.8	96	91.4	99	87.8	74.2	98.6	102.9	56.8	90.5	95.5	103.7	123	106.6	81.4									
Salinity	0.04	0.04	0.03	0.09	0.07	0.05	0.05	0.06	0.03	0.23	0.14	0.6	0.08	0.03	0.03	0.22	0.63	0.47	0.29	0.11									
Conductivity (µs/cm)	90	87	72	78	150	99	113	124	74	471	290	1200	176	42	65	460	1270	957	591	242									
Conductivity (tds ppm)	45	44	36	39	75	49	57	62	37	235	154	600	88	30	33	230	635	479	295	121									
Temperature	12.98	14.36	13.95	17.76	14.07	16.33	15.69	16.77	18.91	17.21	20.02	20.8	22.44	20	20.54	20.38	23.52	23.74	21.15	20.77									
pH	4.82	4.68	4.41	4.9	5.41	4.47	4.63	5.13	5.17	7.22	5.76	8.09	7.62	6	6.19	8.56	7.77	9.11	8.39	7.98									
Date Summer Sampling	13/01/2015	12/1/2015	12/1/2015	13/01/2015	14/01/2015	14/01/2015	14/01/2015	No Sample	13/01/2015	12/1/2015	No Sample	12/1/2015	12/1/2015	11/1/2015	11/1/2015	10/1/2015	9/1/2015	9/1/2015	9/1/2015	8/1/2015	10/1/2015	11/1/2015	11/1/2015	11/1/2015	No Sample	7/1/2015	12/1/2015	No Sample	No Sample
Oxygen (ppm)	6.59	6.49	6.3	5.09	4.24	6.65	6.23		5.98	0.8		5.81	4.75	6.67	6.51	6.94	7.73	5.84	5.53	4.61	4.24	4.05	4.2	5.93		10.25	4.69		
Oxygen (%)	77.4	76	74	61.6	52.1	79.2	77		74.9	80.8		73.5	60.3	80.5	78.4	83	108.6	74.8	71.4	61.6	51.8	46.2	51	74.2		122.2	58.8		
Salinity	0.04	0.04	0.03	0.05	1.07	0.06	0.06		0.04	0.2		0.42	0.07	0.03	0.02	0.24	0.69	0.39	0.28	0.13	0.07	0.09	0.08	0.1		0.02	0.09		
Conductivity (µs/cm)	77	88	69	105	2097	122	126		81	406		869	157	74	52	505	1389	803	577	279	143	184	180	215		40	182		
Conductivity (tds ppm)	39	44	34	52	1007	61	63		41	203		434	79	37	26	253		402	288	140		92	90	108		20	91		
Temperature	22.9	20.51	20.76	23.93	24.37	22.63	24.57		25.74	20.25		25.66	25.47	22.71	22.24	21.63	25.57	22.52	23.13	25.22	20.74	17.99	22.24	24.68		17.14	24.27		
pH	4.35	4.77	4.35	4.99	6	4.63	4.81		5.68	6.08		7.28	6.59	5.98	5.67	8.99	7.96	8.85	8.5	7.16	6.21	6.42	6.65	7.13		6.03	6.29		
Date Autumn Sampling	29/03/2015	29/03/2015	29/03/2015	29/03/2015	27/03/2015	27/03/2015	27/03/2015	27/03/2015	29/03/2015	31/03/2015	No Sample	31/03/2015	31/03/2015	1/4/2015	1/4/2015	2/4/2015	4/4/2015	No Sample	No Sample	3/4/2015	No Sample	No Sample	No Sample	No Sample	No Sample	3/4/2015	31/03/2015	27/03/2015	2/4/2015
Oxygen (ppm)	8.32	8.68	7.79	8.07	12.42	16.2	12.96	11.78	9.84	5.95		5.63	6.98	9.52	9.05	8.81	9.05			10.32						10.46	8.87	16.58	9.97
Oxygen (%)	87.7	95.5	82.8	84.6	131.6	167.1	136.4	125.1	104.6	63.8		64.3	80.7	104.7	101	108.3	110.1			125.3						118.2	100.9	176.6	125.6
Salinity	0.04	0.05	0.03	0.01	0.08	0.04	0.05	0.06	0.04	0.22		0.28	0.21	0.04	0.02	0.22	0.64			0.11						0.02	0.12	0.08	0.64
Conductivity (µs/cm)	77	106	64	96	173	95	107	119	77	446		581	428	81	51	461	1276			222						39	252	172	1284
Conductivity (tds ppm)	39	53	32	48	86	48	54	59	39	223		290	214	40	26	231	638			111						20	126	86	642
Temperature	17.44	18.47	16.67	17.39	18.32	16.76	17.59	17.69	18	18.03		21.38	21.55	18.81	18.57	23.78	17.78			20.82						14.61	20.42	18.6	24.94
pH	5.5	7.19	5.28	5.76	6.54	5.92	6.09	5.8	7.51	7.04		6.88	7.13	8.05	8.38	9.26	9.37			9.09						10.12	6.55	5.49	9.22
Date Winter Sampling	6/8/2015	6/8/2015	No Sample	6/8/2015	3/8/2015	3/8/2015	3/8/2015	No Sample	No Sample	1/8/2015	No Sample	1/8/2015	1/8/2015	30/7/2015	No sample	29/7/2015	27/7/2015	No Sample	No Sample	27/7/2015	No Sample	30/7/2015	30/7/2015	30/7/2015	29/7/2015	26/7/2015	1/8/2015	No Sample	No Sample
Oxygen (ppm)	17.42	15.84		18.59	16.83	17.43	21.19			21.5		16.7	19.96	18.3		17.14	13.12			15.35		12.1	12.53	18.3	14.54	13.16	21.2		
Oxygen (%)	155.2	145.6		165.9	155.7	154.8	187.4			192		154.3	179.5	167.7		157.7	136.5			139.4		129.8	133.5	171.5	147.9	123.9	189		
Salinity	0.04	0.04		0.05	1.72	0.05	0.06			0.17		0.13	0.15	0.04		0.29	0.57			0.04		0.09	0.11	0.09	0.08	0.01	0.09		
Conductivity (µs/cm)	80	85		103	3145	111	117			353		263	317	87		385	1143			80		192	238	179	168	26	198		
Conductivity (tds ppm)	56	60		72	2274	78	82			247		184	222	61		409	800			56		135	167	126	117	18	139		
Temperature	9.79	10.53		10.32	10.82	9.47	9.33			10.41		12.57	10.41	10.81		10.18	11.83			7.91		15.27	16.27	11.27	10.83	6.87	10.26		
pH	4.72	4.99		5	6.59	5.44	4.89			6.36		6.42	6.81	6.9		10.39	8.67			10.56		5.77	6.47	7.53	6.42	6.81	6.3		

Appendix 3 – Relative Abundance and Frequency of occurrence data at different scale

Table 21 Frequency of occurrence and abundance data for the South Eastern Coastal Belt ecoregion

South Eastern Coastal Belt ecoregion	16501	T%	n= 38	F%
Eunotia minor	4660	28	30	79
Achnanthes oblongella	3070	19	26	68
Eunotia incisa	2366	14	18	47
Frustulia saxonica	1173	7	26	68
Gomphonema parvulum	427	3	5	13
Planothidium rostratrum	408	2	6	16
Tabellaria flocculosa	351	2	17	45
Achnanthidium eutrophilum	346	2	6	16
Eunotia flexuosa	198	1	17	45
Planothidium engelbrechtii	190	1	10	26
Fragilaria biceps	174	1	12	32
Eunotia rhomboidea	153	1	4	11
Eunotia formica	146	1	5	13
Frustulia rostrata	134	1	5	13
Cocconeis placentula var. *euglypta*	130	1	1	3
Eolimna minima	124	1	4	11
Cocconeis placentula	124	1	4	11
Nitzschia palea	117	1	7	18
Eunotia bilunaris	96	1	8	21
Achnanthidium minutissimum	95	1	8	21
Achnanthes subaffinis	92	1	1	3
Lemnicola hungarica	86	1	1	3
Frustulia crassinervia	86	1	3	8
Cocconeis placentula var. *lineata*	79	0	1	3
Nitzschia pusilla	70	0	2	5
Nitzschia filiformis	70	0	7	18
Navicula recens	66	0	3	8
Navicula radiosa	62	0	2	5
Encyonopsis leei var. *sinensis*	62	0	1	3
Navicula tripunctata	62	0	2	5
Epithemia adnata	56	0	1	3
Nitzschia closterium	54	0	3	8
Tabularia fasciculata	46	0	5	13
Fragilaria ulna	44	0	3	8
Eunotia pectinalis var. *undulata*	43	0	2	5
Pinnularia microstauron var. *rostrata*	42	0	2	5
Encyonema minutum	41	0	7	18
Planothidium frequentissimum	40	0	2	5
Amphora pediculus	34	0	1	3
Denticula subtilis	34	0	2	5
Navicula riediana	34	0	2	5
Navicula sp12	33	0	1	3
Achnanthidium straubianum	29	0	3	8
Rhopalodia operculata	29	0	2	5
Navicula sp13	27	0	1	3
Eunotia exigua	27	0	3	8
Eunotia sp2	26	0	1	3
Nitzschia sp23	26	0	1	3
Pinnularia subcapitata	25	0	6	16
Fragilaria capucina var. *rumpens*	25	0	3	8
Craticula buderi	24	0	2	5
Frustulia vulgaris	22	0	1	3
Navicymbula pusilla	19	0	1	3
Nitzschia recta	18	0	1	3
Navicula veneta	17	0	1	3
Melosira varians	17	0	4	11
Tryblionella hungarica	16	0	1	3
Brachysira brebissonii	16	0	3	8
Fragilaria capucina var. *vaucheriae*	15	0	1	3
Cocconeis engelbrechtii	15	0	1	3
Stauroneis anceps	14	0	1	3
Brachysira zellensis	13	0	1	3
Craticula molestiformis	13	0	3	8
Fragilaria capucina	12	0	3	8
Craticula accomoda	12	0	1	3
Craticula halophila	11	0	1	3
Achnanthes crassa	10	0	1	3
Nitzschia acicularis	10	0	2	5
Cyclotella meneghiniana	10	0	3	8
Gomphonema sp2	10	0	1	3
Navicula rhynchocephala	10	0	2	5
Staurosira elliptica	10	0	3	8
Nitzschia sp10	9	0	1	3
Encyonema neogracile	8	0	3	8
Hippodonta hungarica	8	0	1	3
Navicula sp20	8	0	1	3
Nitzschia aurariae	7	0	1	3
Nitzschia linearis	7	0	1	3
Gyrosigma scalproides	7	0	2	5
Achnanthidium sp2	6	0	1	3
Adlafia bryophila	6	0	1	3
Nitzschia sigma	6	0	1	3
Tryblionella sp2	6	0	1	3
Achnanthes swazi	6	0	1	3
Amphora sp2	6	0	1	3
Navicula sp21	6	0	1	3
Nitzschia sp15	6	0	1	3
Achnanthidium affine	5	0	1	3
Amphora copulata	5	0	1	3
Diadesmis contenta	5	0	1	3
Achnanthes abundans	4	0	1	3
Amphora veneta	4	0	1	3
Encyonema sp3	4	0	1	3
Gomphonema capitatum	4	0	1	3
Pinnularia sp1	4	0	1	3
Encyonema silesiacum	4	0	2	5
Nitzschia clausii	4	0	2	5
Achnanthidium sp3	4	0	1	3
Nitzschia capitellata	4	0	1	3
Nitzschia sp19	4	0	1	3
Pseudostaurosira brevistriata	4	0	1	3
Brachysira wygaschii	3	0	1	3
Navicula sp4	3	0	1	3
Nitzschia iremissa	3	0	2	5
Nitzschia dissipata	3	0	2	5
Nitzschia sp16	3	0	1	3
Nitzschia sp20	3	0	1	3
Achnanthidium biasolettianum	2	0	1	3
Achnanthidium sp1	2	0	1	3
Amphora fontinalis	2	0	1	3
Amphora strigosa	2	0	1	3
Diadesmis confervacea	2	0	1	3
Encyonopsis raytonensis	2	0	1	3
Gomphonema affine lagenula	2	0	1	3
Gomphonema angustum	2	0	1	3
Navicula longicephala	2	0	1	3
Navicula sp10	2	0	1	3
Nitzschia gracilis	2	0	2	5
Pinnularia sp2	2	0	1	3
Sellophora pupula	2	0	1	3
Simonsenia delognei	2	0	1	3
Tabularia sp2	2	0	1	3
Encyonema mesianum	2	0	2	5
Amphora sp1	2	0	1	3
Craticula sp2	2	0	1	3
Epithemia sorex	2	0	1	3
Fragilaria sp1	2	0	1	3
Frustulia sp4	2	0	1	3
Nitzschia sp22	2	0	1	3
Pinnularia viridiformis	2	0	1	3
Pleurosigma sp1	2	0	1	3
Tryblionella apiculata	2	0	1	3
Tryblionella sp3	2	0	1	3
Achnanthidium macrocephalum	1	0	1	3
Aulacoseira granulata	1	0	1	3
Craticula sp1	1	0	1	3
Cyclotella ocellata	1	0	1	3
Eunotia pectinalis	1	0	1	3
Gomphonema acuminatum	1	0	1	3
Navicula cryptotenelloides	1	0	1	3
Navicula sp3	1	0	1	3

Nitzschia littorea	1	0	1	3
Nitzschia sp13	1	0	1	3
Pinnularia gibba	1	0	1	3
Surirella angusta	1	0	1	3
Caloneis sp2	1	0	1	3
Ctenophora pulchella	1	0	1	3
Cymbella sp4	1	0	1	3
Diploneis elliptica	1	0	1	3
Fallacia sp2	1	0	1	3
Frustulia sp3	1	0	1	3
Luticola goeppertiana	1	0	1	3
Navicula erifuga	1	0	1	3
Nitzschia liebertruthii	1	0	1	3
Nitzschia sp21	1	0	1	3
Surirella sp1	1	0	1	3
Tryblionella levidensis	1	0	1	3

Table 22 Frequency of occurrence and abundance data for the Drought Corridor ecoregion

Drought Corridor ecoregion	8061	F%	n=20	F %
Cocconeis placentula var. *lineata*	1021	13	5	25
Cocconeis placentula	793	10	10	50
Rhopalodia gibba	717	9	10	50
Reimeria uniseriata	646	8	10	50
Nitzschia palea	460	6	7	35
Epithemia adnata	376	5	9	45
Stephanodiscus agassizensis	270	3	3	15
Epithemia sorex	265	3	5	25
Gomphonema parvulum	227	3	9	45
Achnanthidium minutissimum	221	3	5	25
Cocconeis placentula var. *euglypta*	208	3	2	10
Staurosira elliptica	186	2	2	10
Fragilaria capucina var. *vaucheriae*	137	2	4	20
Nitzschia aurariae	136	2	3	15
Nitzschia closterium	129	2	3	15
Amphora pediculus	124	2	4	20
Achnanthes oblongella	111	1	10	50
Gomphonema insigne	107	1	1	5
Planothidium frequentissimum	101	1	5	25
Achnanthidium eutrophilum	93	1	1	5
Melosira varians	89	1	5	25
Nitzschia microcephala	84	1	3	15
Aulacoseira granulata	80	1	2	10
Nitzschia umbonata	77	1	4	20
Nitzschia linearis var. *subtilis*	72	1	1	5
Nitzschia filiformis	69	1	6	30
Navicula tripunctata	63	1	2	10
Rhopalodia operculata	59	1	2	10
Cocconeis pediculus	58	1	5	25
Nitzschia iremissa	58	1	1	5
Fragilaria biceps	57	1	8	40
Navicula veneta	57	1	1	5
Reimeria sinuata	57	1	1	5
Navicula radiosa	55	1	2	10
Navicula capitatoradiata	53	1	2	10
Cymbella tumida	50	1	2	10
Navicula recens	50	1	2	10
Nitzschia dissipata	41	1	2	10
Thalassiosira weissflogi	39	0	1	5
Nitzschia linearis	30	0	2	10
Pleurosigma elongatum	30	0	2	10
Cyclostephanos dubius	26	0	2	10
Tryblionella levidensis	26	0	2	10
Nitzschia symmetrica	24	0	1	5
Tabularia fasciculata	24	0	3	15
Achnanthes swazi	22	0	1	5
Nitzschia acicularis	22	0	1	5
Navicula cryptonella	20	0	1	5
Achnanthidium exiguum	17	0	1	5
Tryblionella apiculata	17	0	4	20
Eunotia minor	16	0	3	15
Fragilaria ulna	16	0	1	5
Eolimna subminuscula	13	0	2	10
Navicula riediana	13	0	1	5
Nitzschia intermedia	13	0	2	10
Craticula accomoda	12	0	1	5
Nitzschia capitellata	12	0	1	5
Rhopalodia gibberula	11	0	3	15
Cyclotella meneghiniana	10	0	2	10
Frustulia saxonica	10	0	1	5
Nitzschia sp11	10	0	1	5
Cymbella turgidula	8	0	1	5
Navicula erifuga	8	0	1	5
Nitzschia liebertruthii	8	0	3	15
Planothidium engelbrechtii	8	0	1	5
Achnanthes crassa	7	0	1	5
Amphora montana	7	0	1	5
Encyonema minutum	6	0	2	10
Pseudostaurosira brevistriata	6	0	1	5
Surirella brebissonii	6	0	1	5
Tryblionella hungarica	6	0	2	10
Bacillaria paradoxa	5	0	2	10
Navicula sp11	5	0	1	5
Navicula sp15	5	0	1	5
Navicula sp19	5	0	1	5
Navicula sp9	5	0	1	5
Tabellaria flocculosa	5	0	2	10
Achnanthidium affine	4	0	1	5
Cymbella neocistula	4	0	2	10
Encyonopsis subminuta	4	0	1	5
Navicula antonii	4	0	1	5
Navicula sp17	4	0	1	5
Nitzschia archibaldii	4	0	1	5
Amphora inariensis	3	0	2	10
Encyonopsis leei var. *sinensis*	3	0	1	5
Gyrosigma rautenbachiae	3	0	1	5
Gyrosigma scalproides	3	0	1	5
Caloneis sp1	2	0	1	5
Cymbella sp2	2	0	1	5
Discotella stelligera	2	0	1	5
Navicula rostellata	2	0	1	5
Pleurosigma salinarum	2	0	2	10
Simonsenia delognei	2	0	1	5
Tryblionella sp1	2	0	1	5
Amphora coffeaeformis	1	0	1	5
Amphora ovalis	1	0	1	5
Caloneis molaris	1	0	1	5
Cocconeis engelbrechtii	1	0	1	5
Craticula ambigua	1	0	1	5
Diadesmis contenta	1	0	1	5
Encyonema sp5	1	0	1	5
Eunotia sp1	1	0	1	5
Fallacia sp1	1	0	1	5
Fragilaria parasitica var. *constricta*	1	0	1	5
Gomphonema venusta	1	0	1	5
Luticola goeppertiana	1	0	1	5
Navicula libonensis	1	0	1	5
Navicula sp14	1	0	1	5
Navicula zanonii	1	0	1	5
Nitzschia sp12	1	0	1	5
Nitzschia sp7	1	0	1	5
Nitzschia sp8	1	0	1	5
Pinnularia divergens	1	0	1	5
Pinnularia viridiformis	1	0	1	5
Tryblionella coarctata	1	0	1	5

Table 23 Frequency of occurrence and abundance data for the Southern Folded Mountains ecoregion

Southern Folded Mountains ecoregion	12311	T%	N= 26	F%
Fragilaria biceps	1515	12	21	81
Staurosira elliptica	1322	11	12	46
Cocconeis placentula	729	6	13	50
Achnanthidium minutissimum	546	4	10	38
Cocconeis placentula var. *lineata*	440	4	8	31
Eunotia minor	407	3	14	54
Gomphonema parvulum	405	3	11	42
Achnanthes oblongella	321	3	14	54

Nitzschia filiformis	272	2	9	35
Nitzschia closterium	259	2	8	31
Gomphonema venusta	229	2	4	15
Diadesmis confervacea	203	2	6	23
Achnanthes subaffinis	201	2	8	31
Epithemia adnata	195	2	8	31
Planothidium frequentissimum	187	2	7	27
Gomphonema parvulum var. *lagenula*	182	1	1	4
Aulacoseira subartica f. subborealis	174	1	1	4
Achnanthidium eutrophilum	161	1	6	23
Melosira varians	161	1	9	35
Achnanthidium affine	158	1	2	8
Pseudostaurosira brevistriata	150	1	3	12
Cocconeis engelbrechtii	141	1	3	12
Planothidium rostatrum	134	1	3	12
Nitzschia palea	128	1	8	31
Fragilaria capucina	127	1	4	15
Fragilaria capucina var. *vaucheriae*	122	1	2	8
Encyonopsis subminuta	112	1	1	4
Fragilaria parasitica	112	1	1	4
Eunotia formica	111	1	4	15
Navicula radiosa	106	1	12	46
Discotella stelligera	102	1	2	8
Eunotia flexuosa	101	1	5	19
Gomphonema angustum	98	1	1	4
Cocconeis placentula var. *euglypta*	88	1	2	8
Fragilaria capucina var. *rumpens*	85	1	3	12
Cyclotella meneghiniana	82	1	11	42
Craticula buderi	81	1	4	15
Gomphonema pseudoaugur	77	1	1	4
Tabularia fasciculata	73	1	6	23
Fragilaria parasitica var. *constricta*	72	1	2	8
Achnanthes swazi	68	1	4	15
Planothidium engelbrechtii	67	1	6	23
Gomphonema affine lagenula	64	1	4	15
Eolimna minima	61	0	2	8
Brachysira brebissonii	60	0	1	4
Frustulia saxonica	60	0	7	27
Fragilaria ulna	57	0	4	15
Gomphonema italicum	56	0	2	8
Gomphonema minutum	55	0	2	8
Cymbella aspera	53	0	8	31
Eunotia incisa	53	0	2	8
Navicula recens	48	0	5	19
Navicula zanonii	47	0	1	4
Rhopalodia operculata	47	0	3	12
Tabellaria flocculosa	45	0	8	31
Craticula vixnegligenda	44	0	3	12
Gomphonema pumilum	44	0	1	4
Nitzscia communis	42	0	1	4
Bacillaria paradoxa	38	0	8	31

Encyonema minutum	38	0	3	12
Fragilaria crotonensis	36	0	2	8
Nitzschia sigma	33	0	2	8
Gomphonema affine gracile	32	0	2	8
Achnanthidium macrocephalum	31	0	1	4
Gomphonema affine	31	0	4	15
Eolimna subminuscula	30	0	2	8
Encyonema silesiacum	29	0	1	4
Gomphonema sp1	28	0	1	4
Nitzschia liebertruthii	28	0	4	15
Navicula sp7	26	0	1	4
Achnanthes crassa	23	0	2	8
Navicula cryptonella	23	0	3	12
Nitzschia frustulum	22	0	2	8
Navicula arvensis var. *maior*	20	0	1	4
Navicula veneta	20	0	1	4
Placoneis dicephala	20	0	1	4
Diploneis oblongella	19	0	1	4
Fragilaria tenera	19	0	1	4
Navicula sp18	19	0	1	4
Epithemia sorex	18	0	4	15
Tryblionella levidensis	18	0	3	12
Rhopalodia gibba	17	0	4	15
Amphora pediculus	16	0	3	12
Navicula angusta	16	0	1	4
Navicula germainii	16	0	1	4
Eunotia pectinalis	15	0	1	4
Achnanthes sp1	14	0	1	4
Navicula sp5	14	0	1	4
Navicula sp6	14	0	1	4
Stauroneis smithii	14	0	1	4
Cocconeis pediculus	13	0	2	8
Nitzschia sp3	13	0	1	4
Thalassiosira weissflogi	13	0	3	12
Diatoma vulgaris	12	0	1	4
Encyonema neogracile	12	0	2	8
Staurosirella pinnata	12	0	1	4
Gomphonema truncatum	11	0	3	12
Sellophora seminulum	11	0	1	4
Tabularia sp1	11	0	1	4
Asterionella formosa	10	0	2	8
Tryblionella hungarica	9	0	2	8
Amphora veneta	8	0	1	4
Navicula cryptocephala	8	0	1	4
Nitzschia sp5	8	0	1	4
Achnanthidium exiguum	7	0	2	8
Diadesmis contenta	7	0	1	4
Navicula sp1	7	0	1	4
Nitzschia nana	7	0	2	8
Nitzschia sp17	7	0	1	4
Nitzschia umbonata	7	0	5	19
Surirella angusta	7	0	1	4

Achnanthidium biasolettianum	6	0	1	4
Amphora sp3	6	0	1	4
Craticula accomoda	6	0	1	4
Encyonema mesianum	6	0	2	8
Eunotia pectinalis var. *undulata*	6	0	1	4
Gyrosigma rautenbachiae	6	0	3	12
Gyrosigma sp4	6	0	1	4
Nitzschia sp14	6	0	1	4
Diploneis elliptica	5	0	2	8
Gomphonema sp3	5	0	1	4
Nitzschia aurariae	5	0	1	4
Nitzschia etoshensis	5	0	1	4
Nitzschia iremissa	5	0	1	4
Pinnularia subcapitata	5	0	3	12
Cymatopleura solea	4	0	1	4
Cymbella tumida	4	0	2	8
Eunotia rhomboidea	4	0	1	4
Frustulia sp1	4	0	1	4
Frustulia sp2	4	0	1	4
Frustulia vulgaris	4	0	2	8
Geissleria decussis	4	0	1	4
Gomphonema insigne	4	0	1	4
Luticola kotschyi	4	0	1	4
Navicula capitatoradiata	4	0	1	4
Navicula sp22	4	0	1	4
Nitzschia sp9	4	0	1	4
Pinnularia sp3	4	0	1	4
Pleurosigma salinarum	4	0	1	4
Reimeria sinuata	4	0	1	4
Rhopalodia gibberula	4	0	2	8
Sellophora pupula	4	0	3	12
Amphora normannii	3	0	1	4
Hantzschia amphyoxys	3	0	1	4
Lemnicola hungarica	3	0	1	4
Luticola sp1	3	0	1	4
Navicula rhynchocephala	3	0	1	4
Placoneis elginensis	3	0	1	4
Rhopalodia sp1	3	0	1	4
Tryblionella calida	3	0	1	4
Achnanthes standerii	2	0	1	4
Adlafia bryophila	2	0	1	4
Capartogramma crucicula	2	0	2	8
Cyclostephanos dubius	2	0	1	4
Cyclotella sp1	2	0	1	4
Diploneis subovalis	2	0	2	8
Encyonema ventricosum	2	0	1	4
Encyonopsis leei var. *sinensis*	2	0	1	4
Gomphonema clavatum	2	0	1	4
Gomphonema laticollum	2	0	1	4
Gyrosigma acuminatum	2	0	1	4
Navicula reichardtiana	2	0	1	4
Nitzschia desertorum	2	0	1	4

Nitzschia sp18	2	0	1	4
Nitzschia sp6	2	0	1	4
Nitzschia sublinearis	2	0	1	4
Orthoseira sp1	2	0	1	4
Staurosira construens	2	0	1	4
Achnanthes linearoides	1	0	1	4
Craticula molestiformis	1	0	1	4
Cymbella sp1	1	0	1	4
Cymbella sp3	1	0	1	4
Cymbopleura naviculiformis	1	0	1	4
Encyonema sp1	1	0	1	4
Encyonema sp2	1	0	1	4
Encyonema sp4	1	0	1	4
Gomphonema acuminatum	1	0	1	4
Gyrosigma attenuatum	1	0	1	4
Gyrosigma sp1	1	0	1	4
Gyrosigma sp2	1	0	1	4
Gyrosigma sp3	1	0	1	4
Luticola acidoclinata	1	0	1	4
Navicula cincta	1	0	1	4
Navicula sp16	1	0	1	4
Navicula sp2	1	0	1	4
Navicula sp8	1	0	1	4
Nitzschia sp2	1	0	1	4
Nitzschia sp4	1	0	1	4
Ntzschia sp1	1	0	1	4
Oxyneis sp1	1	0	1	4
Pinnularia viridis	1	0	1	4
Placoneis placentula	1	0	1	4
Placoneis sp1	1	0	1	4
Stauroneis sp1	1	0	1	4
Stephanodiscus sp1	1	0	1	4
Tryblionella apiculata	1	0	1	4

Table 24 Frequency of occurrence and abundance data for site groups within the Drought Corridor ecoregion

Drought Corridor ecoregion				
Group 1	**1269**	**T%**	**N=3**	**F%**
Cocconeis placentula var. lineata	907	71	3	100
Achnanthes oblongella	38	3	3	100
Fragilaria capucina var. vaucheriae	114	9	2	67
Planothidium frequentissimum	86	7	2	67
Cocconeis placentula	33	3	1	33
Achnanthes swazi	22	2	1	33
Achnanthidium minutissimum	21	2	1	33
Fragilaria ulna	16	1	1	33
Nitzschia capitellata	12	1	1	33
Rhopalodia gibba	11	1	1	33
Gomphonema parvulum	6	0	1	33
Diadesmis contenta	1	0	1	33
Navicula sp14	1	0	1	33
Nitzschia sp8	1	0	1	33
Group 2	**848**	**T**	**N=2**	**F%**
Cocconeis placentula var. lineata	114	13	2	100
Aulacoseira granulata	80	9	2	100
Gomphonema parvulum	34	4	2	100
Encyonema minutum	6	1	2	100
Stephanodiscus agassizensis	187	22	1	50
Melosira varians	75	9	1	50
Reimeria sinuata	57	7	1	50
Cymbella tumida	46	5	1	50
Reimeria uniseriata	34	4	1	50
Fragilaria biceps	32	4	1	50
Cyclostephanos dubius	25	3	1	50
Nitzschia filiformis	24	3	1	50
Fragilaria capucina var. vaucheriae	22	3	1	50
Nitzschia acicularis	22	3	1	50
Epithemia adnata	18	2	1	50
Cocconeis pediculus	17	2	1	50
Achnanthidium exiguum	17	2	1	50
Nitzschia umbonata	15	2	1	50
Amphora montana	7	1	1	50
Pseudostaurosira brevistriata	6	1	1	50
Eolimna subminuscula	3	0	1	50
Cocconeis placentula	2	0	1	50
Rhopalodia gibba	1	0	1	50
Tryblionella apiculata	1	0	1	50
Nitzschia intermedia	1	0	1	50
Nitzschia liebertruthii	1	0	1	50
Pinnularia viridiformis	1	0	1	50
Group 3	**2390**	**T%**	**N=5**	**F%**
Rhopalodia gibba	628	26	5	100
Epithemia adnata	349	15	5	100
Epithemia sorex	265	11	5	100
Nitzschia closterium	129	5	3	60
Gomphonema parvulum	70	3	3	60
Planothidium frequentissimum	15	1	3	60
Rhopalodia gibberula	11	0	3	60
Rhopalodia operculata	59	2	2	40
Navicula radiosa	55	2	2	40
Nitzschia palea	53	2	2	40
Nitzschia microcephala	33	1	2	40
Pleurosigma elongatum	30	1	2	40
Cocconeis pediculus	28	1	2	40
Tryblionella levidensis	26	1	2	40
Tabularia fasciculata	17	1	2	40
Nitzschia filiformis	14	1	2	40
Achnanthes oblongella	13	1	2	40
Fragilaria biceps	11	0	2	40
Nitzschia liebertruthii	7	0	2	40
Tryblionella hungarica	6	0	2	40
Pleurosigma salinarum	2	0	2	40
Achnanthidium eutrophilum	93	4	1	20
Nitzschia linearis var. subtilis	72	3	1	20
Nitzschia iremissa	58	2	1	20
Navicula tripunctata	49	2	1	20
Nitzschia dissipata	39	2	1	20
Nitzschia aurariae	38	2	1	20
Navicula capitatoradiata	34	1	1	20
Nitzschia linearis	21	1	1	20
Navicula cryptonella	20	1	1	20
Amphora pediculus	14	1	1	20
Navicula riediana	13	1	1	20
Navicula recens	12	1	1	20
Nitzschia intermedia	12	1	1	20
Nitzschia sp11	10	0	1	20
Cocconeis placentula var. euglypta	8	0	1	20
Navicula erifuga	8	0	1	20
Planothidium engelbrechtii	8	0	1	20
Cocconeis placentula	6	0	1	20
Reimeria uniseriata	6	0	1	20
Tryblionella apiculata	6	0	1	20
Surirella brebissonii	6	0	1	20
Navicula sp15	5	0	1	20
Navicula sp19	5	0	1	20
Achnanthidium minutissimum	4	0	1	20
Melosira varians	4	0	1	20
Cyclotella meneghiniana	4	0	1	20
Gyrosigma rautenbachiae	3	0	1	20
Amphora inariensis	2	0	1	20
Caloneis sp1	2	0	1	20
Staurosira elliptica	1	0	1	20
Bacillaria paradoxa	1	0	1	20
Cymbella neocistula	1	0	1	20

Amphora ovalis	1	0	1	20
Craticula ambigua	1	0	1	20
Fallacia sp1	1	0	1	20
Nitzschia sp12	1	0	1	20
Group 4	**854**	**T%**	**N=2**	**F%**
Nitzschia palea	364	43	2	100
Gomphonema parvulum	115	13	2	100
Reimeria uniseriata	57	7	2	100
Cocconeis placentula	55	6	2	100
Eunotia minor	8	1	2	100
Nitzschia aurariae	97	11	1	50
Navicula veneta	57	7	1	50
Achnanthidium minutissimum	42	5	1	50
Achnanthes oblongella	36	4	1	50
Craticula accomoda	12	1	1	50
Cyclotella meneghiniana	6	1	1	50
Fragilaria biceps	5	1	1	50
Group 5	**2224**	**T%**	**N=5**	**F%**
Cocconeis placentula	697	31	5	100
Reimeria uniseriata	488	22	5	100
Nitzschia palea	43	2	3	60
Nitzschia umbonata	61	3	2	40
Nitzschia filiformis	27	1	2	40
Achnanthes oblongella	21	1	2	40
Cocconeis pediculus	13	1	2	40
Tryblionella apiculata	10	0	2	40
Melosira varians	9	0	2	40
Epithemia adnata	8	0	2	40
Tabellaria flocculosa	5	0	2	40
Staurosira elliptica	185	8	1	20
Achnanthidium minutissimum	153	7	1	20
Gomphonema insigne	107	5	1	20
Rhopalodia gibba	73	3	1	20
Amphora pediculus	63	3	1	20
Stephanodiscus agassizensis	60	3	1	20
Navicula recens	38	2	1	20
Nitzschia symmetrica	24	1	1	20
Navicula capitatoradiata	19	1	1	20
Eolimna subminuscula	10	0	1	20
Frustulia saxonica	10	0	1	20
Nitzschia linearis	9	0	1	20
Eunotia minor	8	0	1	20
Cymbella turgidula	8	0	1	20
Achnanthes crassa	7	0	1	20
Fragilaria biceps	6	0	1	20
Navicula sp11	5	0	1	20
Navicula sp9	5	0	1	20
Cymbella tumida	4	0	1	20
Bacillaria paradoxa	4	0	1	20
Achnanthidium affine	4	0	1	20
Encyonopsis subminuta	4	0	1	20
Navicula antonii	4	0	1	20
Navicula sp17	4	0	1	20
Cymbella neocistula	3	0	1	20
Encyonopsis leei var. *sinensis*	3	0	1	20
Gyrosigma scalproides	3	0	1	20
Cymbella sp2	2	0	1	20
Discotella stelligera	2	0	1	20
Navicula rostellata	2	0	1	20
Simonsenia delognei	2	0	1	20
Tryblionella sp1	2	0	1	20
Nitzschia aurariae	1	0	1	20
Cyclostephanos dubius	1	0	1	20
Amphora inariensis	1	0	1	20
Amphora coffeaeformis	1	0	1	20
Caloneis molaris	1	0	1	20
Cocconeis engelbrechtii	1	0	1	20
Encyonema sp5	1	0	1	20
Fragilaria parasitica var. *constricta*	1	0	1	20
Tryblionella coarctata	1	0	1	20

Table 25 Frequency of occurrence and abundance data for site groups within the South Eastern Coastal Belt ecoregion

South Eastern Coastal Belt ecoregion				
Group 1	**5706**	**T%**	**N=14**	**F%**
Eunotia minor	3606	63	14	100
Frustulia saxonica	288	5	10	71
Eunotia flexuosa	46	1	7	50
Eunotia incisa	349	6	6	43
Planothidium rostratrum	389	7	4	29
Achnanthes oblongella	93	2	4	29
Eunotia rhomboidea	136	2	3	21
Frustulia rostrata	43	1	3	21
Tabellaria flocculosa	5	0	3	21
Eunotia formica	66	1	2	14
Eunotia pectinalis var. *undulata*	43	1	2	14
Planothidium engelbrechtii	32	1	2	14
Achnanthidium minutissimum	19	0	2	14
Pinnularia subcapitata	15	0	2	14
Encyonema minutum	7	0	2	14
Tabularia fasciculata	5	0	2	14
Nitzschia filiformis	4	0	2	14
Encyonema silesiacum	4	0	2	14
Achnanthidium eutrophilum	240	4	1	7
Lemnicola hungarica	86	2	1	7
Gomphonema parvulum	56	1	1	7
Planothidium frequentissimum	37	1	1	7
Amphora pediculus	34	1	1	7
Rhopalodia operculata	27	0	1	7
Frustulia vulgaris	22	0	1	7
Brachysira zellensis	13	0	1	7
Achnanthidium straubianum	9	0	1	7
Nitzschia aurariae	7	0	1	7
Pinnularia microstauron var. *rostrata*	6	0	1	7
Pinnularia sp1	4	0	1	7
Eunotia exigua	2	0	1	7
Simonsenia delognei	2	0	1	7
Amphora sp1	2	0	1	7
Fragilaria capucina var. *rumpens*	1	0	1	7
Craticula molestiformis	1	0	1	7
Nitzschia iremissa	1	0	1	7
Nitzschia gracilis	1	0	1	7
Encyonema mesianum	1	0	1	7
Craticula sp1	1	0	1	7
Eunotia pectinalis	1	0	1	7
Nitzschia sp13	1	0	1	7
Nitzschia sp21	1	0	1	7
Group 2	**4858**	**T%**	**N=11**	**F%**
Eunotia incisa	1979	41	10	91
Frustulia saxonica	751	15	9	82
Achnanthes oblongella	355	7	9	82
Tabellaria flocculosa	315	6	8	73
Eunotia minor	547	11	7	64
Eunotia bilunaris	67	1	7	64
Eunotia flexuosa	96	2	5	45
Achnanthidium minutissimum	32	1	4	36
Fragilaria biceps	8	0	4	36
Nitzschia palea	63	1	3	27
Encyonema minutum	29	1	3	27
Brachysira brebissonii	16	0	3	27

Achnanthidium eutrophilum	46	1	2	18
Nitzschia closterium	32	1	2	18
Frustulia crassinervia	30	1	2	18
Eunotia exigua	25	1	2	18
Fragilaria capucina var. *rumpens*	24	0	2	18
Planothidium rostatrum	19	0	2	18
Tabularia fasciculata	8	0	2	18
Pinnularia subcapitata	7	0	2	18
Encyonema neogracile	4	0	2	18
Frustulia rostrata	79	2	1	9
Encyonopsis leei var. *sinensis*	62	1	1	9
Pinnularia microstauron var. *rostrata*	36	1	1	9
Navicula sp12	33	1	1	9
Navicula sp13	27	1	1	9
Eolimna minima	23	0	1	9
Nitzschia recta	18	0	1	9
Eunotia rhomboidea	17	0	1	9
Stauroneis anceps	14	0	1	9
Planothidium engelbrechtii	13	0	1	9
Achnanthidium straubianum	11	0	1	9
Gomphonema sp2	10	0	1	9
Nitzschia acicularis	9	0	1	9
Staurosira elliptica	6	0	1	9
Adlafia bryophila	6	0	1	9
Nitzschia filiformis	4	0	1	9
Achnanthes abundans	4	0	1	9
Encyonema sp3	4	0	1	9
Brachysira wygaschii	3	0	1	9
Melosira varians	2	0	1	9
Nitzschia iremissa	2	0	1	9
Achnanthidium sp1	2	0	1	9
Amphora strigosa	2	0	1	9
Diadesmis confervacea	2	0	1	9
Navicula longicephala	2	0	1	9
Sellophora pupula	2	0	1	9
Craticula sp2	2	0	1	9
Fragilaria sp1	2	0	1	9
Eunotia formica	1	0	1	9
Fragilaria ulna	1	0	1	9
Craticula molestiformis	1	0	1	9
Cyclotella ocellata	1	0	1	9
Gomphonema acuminatum	1	0	1	9
Navicula cryptotenelloides	1	0	1	9
Navicula sp3	1	0	1	9

Fallacia sp2	1	0	1	9
Group 3	**4595**	**T%**	**N=10**	**F%**
Achnanthes oblongella	2390	52	10	100
Eunotia minor	479	10	7	70
Tabellaria flocculosa	31	1	6	60
Frustulia saxonica	90	2	5	50
Planothidium engelbrechtii	79	2	5	50
Fragilaria biceps	99	2	5	50
Eunotia flexuosa	25	1	4	40
Nitzschia filiformis	62	1	4	40
Achnanthidium eutrophilum	60	1	3	30
Cocconeis placentula	118	3	3	30
Nitzschia palea	29	1	3	30
Fragilaria capucina	12	0	3	30
Eunotia incisa	38	1	2	20
Eunotia formica	79	2	2	20
Achnanthidium minutissimum	44	1	2	20
Navicula recens	24	1	2	20
Navicula tripunctata	62	1	2	20
Navicula riediana	34	1	2	20
Craticula buderi	24	1	2	20
Melosira varians	14	0	2	20
Cyclotella meneghiniana	9	0	2	20
Navicula rhynchocephala	10	0	2	20
Staurosira elliptica	4	0	2	20
Nitzschia clausii	4	0	2	20
Nitzschia dissipata	3	0	2	20
Gomphonema parvulum	4	0	1	10
Frustulia rostrata	12	0	1	10
Cocconeis placentula var. *euglypta*	130	3	1	10
Eolimna minima	58	1	1	10
Eunotia bilunaris	29	1	1	10
Achnanthes subaffinis	92	2	1	10
Frustulia crassinervia	56	1	1	10
Nitzschia pusilla	12	0	1	10
Navicula radiosa	57	1	1	10
Fragilaria ulna	42	1	1	10
Encyonema minutum	4	0	1	10
Achnanthidium straubianum	9	0	1	10
Rhopalodia operculata	2	0	1	10
Eunotia sp2	26	1	1	10
Nitzschia sp23	26	1	1	10
Pinnularia subcapitata	1	0	1	10
Navicymbula pusilla	19	0	1	10

Cocconeis engelbrechtii	15	0	1	10
Craticula molestiformis	11	0	1	10
Craticula accomoda	12	0	1	10
Craticula halophila	11	0	1	10
Achnanthes crassa	10	0	1	10
Nitzschia sp10	9	0	1	10
Encyonema neogracile	4	0	1	10
Hippodonta hungarica	8	0	1	10
Navicula sp20	8	0	1	10
Gyrosigma scalproides	5	0	1	10
Achnanthidium sp2	6	0	1	10
Tryblionella sp2	6	0	1	10
Achnanthes swazi	6	0	1	10
Amphora sp2	6	0	1	10
Navicula sp21	6	0	1	10
Nitzschia sp15	6	0	1	10
Achnanthidium affine	5	0	1	10
Gomphonema capitatum	4	0	1	10
Achnanthidium sp3	4	0	1	10
Nitzschia capitellata	4	0	1	10
Nitzschia sp19	4	0	1	10
Pseudostaurosira brevistriata	4	0	1	10
Navicula sp4	3	0	1	10
Nitzschia sp16	3	0	1	10
Nitzschia sp20	3	0	1	10
Gomphonema angustum	2	0	1	10
Pinnularia sp2	2	0	1	10
Encyonema mesianum	1	0	1	10
Epithemia sorex	2	0	1	10
Frustulia sp4	2	0	1	10
Nitzschia sp22	2	0	1	10
Pinnularia viridiformis	2	0	1	10
Pleurosigma sp1	2	0	1	10
Tryblionella apiculata	2	0	1	10
Tryblionella sp3	2	0	1	10
Aulacoseira granulata	1	0	1	10
Caloneis sp2	1	0	1	10
Ctenophora pulchella	1	0	1	10
Cymbella sp4	1	0	1	10
Diploneis elliptica	1	0	1	10
Frustulia sp3	1	0	1	10
Luticola goeppertiana	1	0	1	10
Navicula erifuga	1	0	1	10
Nitzschia liebertruthii	1	0	1	10

Surirella sp1	1	0	1	10
Tryblionella levidensis	1	0	1	10
Group 4	**1342**	**T%**	**N=3**	**F%**
Achnanthes oblongella	232	17	3	100
Gomphonema parvulum	367	27	3	100
Fragilaria biceps	67	5	3	100
Eunotia minor	28	2	2	67
Frustulia saxonica	44	3	2	67
Planothidium engelbrechtii	66	5	2	67
Eolimna minima	43	3	2	67
Denticula subtilis	34	3	2	67
Eunotia flexuosa	31	2	1	33
Cocconeis placentula	6	0	1	33
Nitzschia palea	25	2	1	33
Cocconeis placentula var. *lineata*	79	6	1	33
Nitzschia pusilla	58	4	1	33
Navicula recens	42	3	1	33
Navicula radiosa	5	0	1	33
Epithemia adnata	56	4	1	33
Nitzschia closterium	22	2	1	33
Tabularia fasciculata	33	2	1	33
Fragilaria ulna	1	0	1	33
Encyonema minutum	1	0	1	33
Planothidium frequentissimum	3	0	1	33
Pinnularia subcapitata	2	0	1	33
Navicula veneta	17	1	1	33
Melosira varians	1	0	1	33
Tryblionella hungarica	16	1	1	33
Fragilaria capucina var. *vaucheriae*	15	1	1	33
Nitzschia acicularis	1	0	1	33
Cyclotella meneghiniana	1	0	1	33
Nitzschia linearis	7	1	1	33
Gyrosigma scalproides	2	0	1	33
Nitzschia sigma	6	0	1	33
Amphora copulata	5	0	1	33
Diadesmis contenta	5	0	1	33
Amphora veneta	4	0	1	33
Achnanthidium biasolettianum	2	0	1	33
Amphora fontinalis	2	0	1	33
Encyonopsis raytonensis	2	0	1	33
Gomphonema affine lagenula	2	0	1	33
Navicula sp10	2	0	1	33
Nitzschia gracilis	1	0	1	33
Tabularia sp2	2	0	1	33
Achnanthidium macrocephalum	1	0	1	33
Nitzschia littorea	1	0	1	33
Pinnularia gibba	1	0	1	33
Surirella angusta	1	0	1	33

Table 26 Frequency of occurrence and abundance data for site groups within the Southern Folded Mountains ecoregion

Southern Folded Mountains ecoregion				
Group 1	**4307**	**T%**	**N=9**	**F%**
Staurosira elliptica	1266	29	9	100
Cyclotella meneghiniana	70	2	7	78
Melosira varians	106	2	6	67
Cocconeis placentula	448	10	5	56
Fragilaria biceps	152	4	5	56
Navicula radiosa	76	2	5	56
Gomphonema parvulum	70	2	5	56
Bacillaria paradoxa	32	1	5	56
Cocconeis placentula var. *lineata*	360	8	4	44
Nitzschia filiformis	172	4	4	44
Nitzschia palea	99	2	4	44
Epithemia sorex	18	0	4	44
Achnanthidium eutrophilum	52	1	3	33
Tabularia fasciculata	44	1	3	33
Epithemia adnata	40	1	3	33
Eunotia minor	18	0	3	33
Tryblionella levidensis	18	0	3	33
Rhopalodia gibba	16	0	3	33
Amphora pediculus	16	0	3	33
Thalassiosira weissflogi	13	0	3	33
Cymbella aspera	5	0	3	33
Achnanthidium minutissimum	165	4	2	22
Pseudostaurosira brevistriata	148	3	2	22
Planothidium rostratum	95	2	2	22
Cocconeis placentula var. *euglypta*	88	2	2	22
Craticula buderi	57	1	2	22
Gomphonema italicum	56	1	2	22
Gomphonema minutum	55	1	2	22
Navicula recens	38	1	2	22
Achnanthes crassa	23	1	2	22
Nitzschia frustulum	22	1	2	22
Cocconeis pediculus	13	0	2	22
Achnanthes oblongella	10	0	2	22
Tryblionella hungarica	9	0	2	22
Achnanthidium exiguum	7	0	2	22
Planothidium engelbrechtii	6	0	2	22
Nitzschia liebertruthii	5	0	2	22
Gyrosigma rautenbachiae	5	0	2	22
Diploneis elliptica	5	0	2	22
Rhopalodia gibberula	4	0	2	22
Diadesmis confervacea	3	0	2	22
Fragilaria capucina var. *vaucheriae*	105	2	1	11
Cocconeis engelbrechtii	28	1	1	11
Navicula sp7	26	1	1	11
Diploneis oblongella	19	0	1	11
Achnanthes sp1	14	0	1	11
Navicula sp6	14	0	1	11
Nitzschia sp3	13	0	1	11
Staurosirella pinnata	12	0	1	11
Gomphonema affine	11	0	1	11
Sellophora seminulum	11	0	1	11
Tabularia sp1	11	0	1	11
Navicula cryptonella	10	0	1	11
Achnanthes subaffinis	8	0	1	11
Amphora veneta	8	0	1	11
Diadesmis contenta	7	0	1	11
Nitzschia sp17	7	0	1	11
Amphora sp3	6	0	1	11
Craticula accomoda	6	0	1	11
Gyrosigma sp4	6	0	1	11
Encyonema minutum	5	0	1	11
Nitzschia etoshensis	5	0	1	11
Planothidium frequentissimum	4	0	1	11
Cymatopleura solea	4	0	1	11
Pleurosigma salinarum	4	0	1	11
Reimeria sinuata	4	0	1	11
Nitzschia closterium	3	0	1	11
Encyonema mesianum	3	0	1	11
Hantzschia amphyoxys	3	0	1	11
Luticola sp1	3	0	1	11
Placoneis elginensis	3	0	1	11
Achnanthidium affine	2	0	1	11
Discotella stelligera	2	0	1	11
Frustulia saxonica	2	0	1	11
Eolimna subminuscula	2	0	1	11
Nitzschia umbonata	2	0	1	11

Cymbella tumida	2	0	1	11
Cyclostephanos dubius	2	0	1	11
Encyonopsis leei var. *sinensis*	2	0	1	11
Gyrosigma acuminatum	2	0	1	11
Nitzschia desertorum	2	0	1	11
Nitzschia sp18	2	0	1	11
Nitzschia sublinearis	2	0	1	11
Orthoseira sp1	2	0	1	11
Staurosira construens	2	0	1	11
Tabellaria flocculosa	1	0	1	11
Sellophora pupula	1	0	1	11
Cymbella sp1	1	0	1	11
Cymbella sp3	1	0	1	11
Gyrosigma attenuatum	1	0	1	11
Gyrosigma sp1	1	0	1	11
Navicula sp8	1	0	1	11
Nitzschia sp4	1	0	1	11
Ntzschia sp1	1	0	1	11
Oxyneis sp1	1	0	1	11
Stephanodiscus sp1	1	0	1	11
Group 2	**2394**	**T%**	**N=5**	**F%**
Fragilaria biceps	898	38	5	100
Achnanthes oblongella	101	4	4	80
Eunotia minor	77	3	4	80
Tabellaria flocculosa	39	2	4	80
Nitzschia closterium	206	9	3	60
Gomphonema venusta	127	5	3	60
Nitzschia filiformis	67	3	3	60
Navicula recens	10	0	3	60
Gomphonema parvulum	157	7	2	40
Achnanthidium eutrophilum	93	4	2	40
Rhopalodia operculata	46	2	2	40
Tabularia fasciculata	27	1	2	40
Gomphonema affine lagenula	27	1	2	40
Frustulia saxonica	18	1	2	40
Encyonema neogracile	12	1	2	40
Cocconeis placentula	6	0	2	40
Gomphonema truncatum	3	0	2	40
Pinnularia subcapitata	3	0	2	40
Nitzschia umbonata	2	0	2	40
Planothidium frequentissimum	77	3	1	20
Fragilaria capucina var. *rumpens*	54	2	1	20
Nitzscia communis	42	2	1	20
Encyonema silesiacum	29	1	1	20
Eolimna subminuscula	28	1	1	20
Nitzschia sigma	22	1	1	20
Navicula arvensis var. *maior*	20	1	1	20
Placoneis dicephala	20	1	1	20
Nitzschia liebertruthii	19	1	1	20
Fragilaria capucina var. *vaucheriae*	17	1	1	20
Cocconeis placentula var. *lineata*	16	1	1	20
Navicula angusta	16	1	1	20
Eunotia flexuosa	14	1	1	20
Achnanthidium minutissimum	11	0	1	20
Achnanthes subaffinis	9	0	1	20
Gomphonema affine	7	0	1	20
Cyclotella meneghiniana	6	0	1	20
Achnanthidium biasolettianum	6	0	1	20
Nitzschia sp14	6	0	1	20
Nitzschia iremissa	5	0	1	20
Planothidium engelbrechtii	4	0	1	20
Craticula vixnegligenda	4	0	1	20
Frustulia sp1	4	0	1	20
Frustulia sp2	4	0	1	20
Luticola kotschyi	4	0	1	20
Navicula sp22	4	0	1	20
Encyonema mesianum	3	0	1	20
Pseudostaurosira brevistriata	2	0	1	20
Nitzschia palea	2	0	1	20
Fragilaria capucina	2	0	1	20
Sellophora pupula	2	0	1	20
Achnanthes standerii	2	0	1	20
Epithemia adnata	1	0	1	20
Navicula radiosa	1	0	1	20
Achnanthes swazi	1	0	1	20
Encyonema minutum	1	0	1	20
Rhopalodia gibba	1	0	1	20
Nitzschia nana	1	0	1	20
Gyrosigma rautenbachiae	1	0	1	20
Frustulia vulgaris	1	0	1	20
Diploneis subovalis	1	0	1	20
Cymbopleura naviculiformis	1	0	1	20
Encyonema sp4	1	0	1	20
Navicula sp16	1	0	1	20
Placoneis sp1	1	0	1	20
Tryblionella apiculata	1	0	1	20
Group 3	**2829**	**T%**	**N=6**	**F%**
Fragilaria biceps	397	14	6	100
Achnanthes oblongella	125	4	6	100
Achnanthidium minutissimum	345	12	5	83
Planothidium frequentissimum	106	4	5	83
Cocconeis placentula	105	4	5	83
Cymbella aspera	48	2	5	83
Gomphonema parvulum	178	6	4	67
Eunotia minor	92	3	4	67
Navicula radiosa	25	1	4	67
Diadesmis confervacea	145	5	3	50
Eunotia formica	83	3	3	50
Achnanthes subaffinis	72	3	3	50
Planothidium engelbrechtii	57	2	3	50
Nitzschia closterium	37	1	3	50
Nitzschia palea	27	1	3	50
Fragilaria parasitica var. *constricta*	72	3	2	33
Epithemia adnata	71	3	2	33
Fragilaria capucina	58	2	2	33
Staurosira elliptica	55	2	2	33
Eunotia flexuosa	44	2	2	33
Gomphonema affine lagenula	37	1	2	33
Nitzschia filiformis	33	1	2	33
Fragilaria capucina var. *rumpens*	31	1	2	33
Craticula buderi	24	1	2	33
Frustulia saxonica	23	1	2	33
Fragilaria ulna	18	1	2	33
Melosira varians	17	1	2	33
Cyclotella meneghiniana	4	0	2	33
Nitzschia umbonata	3	0	2	33
Bacillaria paradoxa	2	0	2	33
Fragilaria parasitica	112	4	1	17
Gomphonema angustum	98	3	1	17
Navicula zanonii	47	2	1	17
Gomphonema sp1	28	1	1	17
Eunotia incisa	25	1	1	17
Fragilaria tenera	19	1	1	17
Navicula germainii	16	1	1	17
Eunotia pectinalis	15	1	1	17
Gomphonema affine	12	0	1	17
Cocconeis engelbrechtii	11	0	1	17
Nitzschia sigma	11	0	1	17
Navicula cryptonella	11	0	1	17
Gomphonema affine gracile	8	0	1	17
Gomphonema truncatum	8	0	1	17
Surirella angusta	7	0	1	17
Gomphonema sp3	5	0	1	17
Nitzschia aurariae	5	0	1	17

Nitzschia liebertruthii	4	0	1	17
Eunotia rhomboidea	4	0	1	17
Geissleria decussis	4	0	1	17
Navicula capitatoradiata	4	0	1	17
Pinnularia sp3	4	0	1	17
Amphora normannii	3	0	1	17
Lemnicola hungarica	3	0	1	17
Navicula rhynchocephala	3	0	1	17
Rhopalodia sp1	3	0	1	17
Tryblionella calida	3	0	1	17
Pinnularia subcapitata	2	0	1	17
Adlafia bryophila	2	0	1	17
Gomphonema clavatum	2	0	1	17
Rhopalodia operculata	1	0	1	17
Tabellaria flocculosa	1	0	1	17
Sellophora pupula	1	0	1	17
Capartogramma crucicula	1	0	1	17
Diploneis subovalis	1	0	1	17
Achnanthes linearoides	1	0	1	17
Craticula molestiformis	1	0	1	17
Encyonema sp1	1	0	1	17
Encyonema sp2	1	0	1	17
Gyrosigma sp2	1	0	1	17
Gyrosigma sp3	1	0	1	17
Luticola acidoclinata	1	0	1	17
Navicula cincta	1	0	1	17
Pinnularia viridis	1	0	1	17
Placoneis placentula	1	0	1	17
Stauroneis sp1	1	0	1	17