

**A STUDY ON PERIPHYTON
AS INDICATOR OF WATER-QUALITY
IN REGULATED RIVERS**

by

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DECLARATION

I declare that the thesis hereby submitted by me for the PhD degree at the University of the Free State is my own independent work and has not previously been submitted by me at another university/faculty. I further more cede copyright of the thesis in favour of the University of the Free State.

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SUMMARY

In the central part of South Africa, where the average rainfall is 400–600 mm/yr and evaporation far exceeds precipitation, it is important to monitor the limited freshwater resources that are available and to keep the aquatic environment in an acceptable state.

Excessive nutrients (N and P) lead to algal blooms and deterioration of other aquatic biota as the water quality declines. Biological monitoring methods and programmes have been instrumental in the management and monitoring of the health of aquatic ecosystems. Various biomonitoring indices have been developed, using fish, benthic macroinvertebrates, phytoplankton and periphyton (including Bacillariophyta), as site- or non-site-specific indicators of water quality.

Periphyton forms the foundation of many food webs. It is adaptable to the availability of a habitat and is directly affected by changes in water quality. In unregulated rivers, “normal” flow patterns and disturbance regimes shape the benthic community composition, while in regulated rivers the “unpredictability” of flow (as an example) adds extra stress to the ecosystem.

The overall objective of this study was to determine the position of periphyton (as a group) as a biomonitoring tool and which of its components would be best suited as indicators of water quality. This study was carried out over two periods of 24 months each at two sites on the Modder River and one on the Renoster Spruit. The sites were selected because SASS5 (standard benthic macroinvertebrate index of water quality) could be carried out on “stones in current”, as this is the preferred habitat for sampling periphyton.

The physical and chemical factors sampled were temperature (°C), turbidity (NTU), flow (m/s), dissolved oxygen (mg/l and % saturation), electrical conductivity ($\mu\text{S}/\text{cm}$) and total dissolved salts (mg/l), pH, redox potential (mV), nutrients (including dissolved inorganic phosphorus and nitrogen) and

chlorophyll-a. The biotas sampled were periphyton, phytoplankton and benthic macroinvertebrates. Statistical analyses were carried out on all sampled data.

Correlations and patterns between the periphyton values and the physical, chemical and biological conditions were investigated. The effect of seasonality on the periphyton and the influence of hydrological phases (dry and wet periods) on the periphyton were examined.

Results indicate that the composition of the periphyton is shaped by seasons. An increase of Bacillariophyta was found during winter, and Chlorophyta and Cyanophyta during summer. The increase of flow during wet periods had a negative effect on the biovolume of periphyton, as fewer filamentous and colonial algae were present during the wet period. The cell and chlorophyll-a concentration also decreased because of dislodgement during high flow. Even though the nutrients had an influence on all the periphytic algal components, the best correlations were found with the periphytic chlorophyll-a concentration.

The periphyton composition and concentration were compared to the biomonitoring indices used on the sampled rivers and sites, namely FRAI (fish) and SASS5 (benthic macroinvertebrates), as well as phytoplankton.

To conclude, it was found that periphyton could be used as a biomonitoring indicator in the monitoring and management of water quality. However, as the standard biomonitoring indices operate on different spatial scales and measures, the best results would be obtained if all, or a combination of indices, were used.

Key words: periphyton, epilithic, biomonitoring, water quality, chlorophyll-a, regulated rivers, SASS5, seasonal, hydrological phases, indicator indices.

OPSOMMING

In die sentrale deel van Suid-Afrika, waar die gemiddelde reënval 400–600 mm/jaar is en verdamping die neerslag ver oorskry, is dit belangrik om die beperkte varswaterbronne wat daar is te monitor en die akwatiese omgewing in 'n goeie toestand te hou.

Oormatige voedingstowwe (N en P) lei tot algobloei en die verswakking van ander waterbiota namate die gehalte van die water afneem. Die ontwikkeling van biologiese moniteringsmetodes en programme is instrumenteel om die gesondheid van water-ekosisteme te bestuur en te monitor. Verskeie biomoniteringsindekse is ontwikkel met die gebruik van vis, bentiese makroinvertebrate, fitoplankton en perifiton (insl. Bacillariophyta), as plek- of nie-plekspesifieke aanwysers van watergehalte.

Perifiton vorm die grondslag van baie voedselwebbe. Dit is aanpasbaar by die beskikbaarheid van 'n habitat en word direk geraak deur die verandering in die waterkwaliteit. In ongereguleerde riviere vorm "normale" vloei- en versteuringspatrone die samestelling van die bentiese gemeenskap, terwyl die "onvoorspelbaarheid" van vloei (as 'n voorbeeld) ekstra stremming op die ekosisteem in gereguleerde riviere plaas.

Die oorhoofse doel van hierdie studie was om die posisie van perifiton (as 'n groep) as 'n biomoniteringsinstrument te ondersoek, asook watter van die perifitonkomponente die geskikste is as indikatore van watergehalte. Hierdie studie is oor twee periodes van 24 maande elk uitgevoer, op twee plekke op die Modderrivier en een op die Renosterspruit. Die terreine is gekies omdat SASS5 (standaard bentiese makroinvertebraatindeks van waterkwaliteit) gedoen kan word met "klippe in die stroom". Klippe is die voorkeurhabitat vir perifiton-versameling.

Die fisiese en chemiese faktore wat versamel is, is temperatuur (°C), troebelheid (NTU), vloei (m/s), opgeloste suurstof (mg/l en % versadiging), elektriese geleiding ($\mu\text{S}/\text{cm}$) en totale opgeloste soute (mg/l), pH, redokspotensiaal (mV),

voedingstowwe (insluitende opgeloste anorganiese fosfor en stikstof) en chlorofil-a. Die versamelde biotas was perifiton, fitoplankton en bentiese makroinvertebrate. Statistiese ontledings is op al die versamelde data uitgevoer.

Korrelasies en patrone tussen die perifitonwaardes en dié van die fisiese, chemiese en biologiese faktore is ondersoek. Die effek van die seisoene op die perifiton en die invloed van hidrologiese fases (droë en nat periodes) op perifiton is ondersoek.

Resultate dui aan dat die samestelling van die perifiton deur seisoene gevorm word. 'n Toename van Bacillariophyta is gedurende die winter, en Chlorophyta en Cyanophyta gedurende die somer waargeneem. Die toename van vloei gedurende nat periodes het 'n negatiewe uitwerking op die biovolume van perifiton – daar was minder filamentagtige en kolonie-vormende alge teenwoordig tydens die nat tydperk. Daar is ook 'n afname in die sel- en chlorofil-a konsentrasie deur middel van verdrywing. Al het die voedingstowwe 'n invloed op al die perifitiese algkomponente, is die beste korrelasie gevind met die perifitiese chlorofil-a konsentrasie.

Die perifitonsamestelling en -konsentrasie is ook vergelyk met die biomonitoringsindekse wat op die steekproef-riviere en -terreine gebruik word, naamlik FRAI (vis) en SASS5 (bentiese makroinvertebrate), asook met fitoplankton.

Om af te sluit, is daar gevind dat perifiton as 'n biomonitoringsindikator in die monitering en bestuur van watergehalte gebruik kan word. Omdat die standaard biomonitoringsindekse egter op verskillende ruimtelike skale werk en verskillende aspekte meet, sal die beste resultate verkry word indien alle, of 'n kombinasie van indekse, saam gebruik word.

Sleutelwoorde: perifiton, epilities, biomonitoring, waterkwaliteit, chlorofil-a, gereguleerde riviere, SASS5, seisoenaal, hidrologiese fases, indikatorindekse.

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LIST OF ABBREVIATIONS

°	degree(s)
%	percentage
ASPT	average score per taxa
biovol.	biovolume(s)
BW	Bishop's Weir
°C	degrees centigrade
chl- <i>a</i>	chlorophyll- <i>a</i> / phytoplanktonic chlorophyll- <i>a</i>
conc.	concentration(s)
corr.	correlation
CEM	Centre for Environmental Management , UFS
CPOM	coarse particulate organic matter
DO	dissolved oxygen (diss. O ₂)
DIN	dissolved inorganic nitrogen
DIP	dissolved inorganic phosphorus
diss.	dissolve(d)
Eq.	equation(s)
FAII	Fish Assemblage Integrity Index
FFG	functional feeding group
Fig.	figure(s)
FRAI	Fish Response Assessment Index
FPOM	fine particulate organic matter
GAI	Geomorphological Assessment Index
g/l	gram per litre
HAI	Hydrological Driver Assessment Index
IBI	Index of Biotic Integrity
IHI	Index of Habitat Integrity
l	litre
m/s	metres per second
m ³ /s	cubic metres per second
MC	Modder above Confluence
MIRAI	Macroinvertebrate Response Assessment Index

$\mu\text{g}/\text{cm}^2$	micrograms per square centimetre
mg/m^2	milligrams per square metre
$\mu\text{g}/\ell$	micrograms per litre
mg/ℓ	milligrams per litre
$\mu\ell$	microlitre
$\mu\text{m}^3/\text{cm}^2$	cubic micrometres per square centimetres
$\mu\text{S}/\text{cm}$	micro Siemens per centimetre
$\text{m}\ell$	millilitre
mV	millivolts
n	number of samples
N	nitrogen
NBPAE	National Biomonitoring Programme for Aquatic Ecosystems
n.d.	no date
neg.	negative
NH_4^+	ammonium
nm	nanometre
NO_3^-	nitrate
NTU	Nephelometric turbidity unit
O_2	oxygen
P	phosphorus
p	probability
PAI	Physico-Chemical Driver Assessment Index
PCA	Principle Component Analysis
$pchl-a$	periphytic chlorophyll- <i>a</i>
PO_4^+	phosphate
ppt	parts per thousand
r	correlation coefficient
r^2	coefficient of determination
RCC	river continuum concept
RHP	river health program
RVI	Riparian Vegetation Index
SASS5	South African scoring system version 5
SD	standard deviation
SDC	serial discontinuity concept

SIC	stones in current
SP	Sannaspos
STW	sewage treatment works
TDS	total dissolved salts
TSS	total suspended solids
UFS	University of the Free State
VEGRAI	Riparian Vegetation Index
WQI	Water Quality Index

CHAPTER 1

INTRODUCTION

1.1 BACKGROUND

1.1.1 WATER AVAILABILITY AND DISTRIBUTION

The average rainfall in central South Africa is 400–600 mm/yr, being higher in the east and lower in the west. Except for a small part of the South-Western Cape and a few high parts of the Drakensberg, evaporation exceeds precipitation (DWAF, 1986; Davies & Day, 1998). The high evaporation rate causes rain to evaporate soon after it reaches the ground. However, the drier areas experience great inter-seasonal and inter-annual variability in rainfall and resultant flow regime.

South Africa might experience further water stress in later years as the population continues to grow. It is therefore important to monitor the quality of the water, as we have to try to keep the aquatic environment in an accepted state.

1.1.2 POLLUTION AND EUTROPHICATION

Rivers and streams are not only open ecosystems, but are greatly influenced by the interaction with bordering systems, as energy is exchanged between water bodies and the terrestrial environment surrounding them (Valett *et al.*, 1994).

Any substance, whether natural, cultural or agricultural, that degrades the quality of water, renders itself as a pollutant (Giller & Malmqvist, 1998; Allan & Castillo, 2007). Pollutants can be from non-point sources or point sources, and can have

either a short-term or a long-term degrading effect on the aquatic environment (Ouyang *et al.*, 2006; Monteagudo, *et al.*, 2012; Tundisi & Tundisi, 2012).

Excessive nutrient inputs (especially phosphorus and nitrogen) lead to eutrophication (mostly cultural), even more so in areas with denser human activities (Dodds, 2002). As nitrogen and phosphorus are the leading nutrients in plant and algal growth, algal growth (little or excessive), composition and distribution can be indicators of pollution and the trophic status of the water ecosystem (Bowman *et al.*, 2005; Greenwood & Rosemond, 2005).

In South Africa, eutrophication first caught attention in the 1970s, and became a top-three water quality problem since. Eutrophication in South Africa is mostly caused by cultural eutrophication of point and non-point source origin (Rossouw *et al.*, 2008).

Various studies have been done on the pollution and eutrophication of the Modder River, whether it was for the health of the aquatic ecosystem, water quality for domestic or agricultural use, or human health (Grobler & Toerien, 1986; Jagals & Grabow, 1996; Koning & Roos, 1999; Koning *et al.*, 2000; Oberholster *et al.*, 2009; Nyenje *et al.*, 2010).

1.1.3 BIOMONITORING

Biological monitoring of aquatic systems has become a standard practice worldwide as a management tool, with emphasis on stream monitoring programmes and environmental impact assessments for over 50 years (Rosen, 1995; Atazadeh *et al.*, 2007).

In 1996, the Department of Water Affairs and Forestry (DWAF), the Water Research Commission (WRC) and the former Department of Environmental Affairs and Tourism (DEAT) initiated the National Biomonitoring Programme for Aquatic Ecosystems (NBPAE) in South Africa (Bate *et al.*, 2002).

Through the years, the indices (e.g. for fish and macroinvertebrates) were developed, redeveloped, extended and improved to the current ones used in the River Health Programme (RHP) and EcoStatus Classification (Bate *et al.*, 2002; Kleynhans, 2007). Some are site specific and others are non-site specific (Kleynhans, 2007, Taylor *et al.*, 2007a; De la Rey *et al.*, 2008). Through biomonitoring, we obtain knowledge of the state of our water ecosystems (rivers, lakes, reservoirs) and with that the first step is taken towards establishing efficient management systems that are essential for maintaining the quality of aquatic ecosystems (Douterelo *et al.*, 2004).

Various biomonitoring protocols have been developed using periphyton measuring algal biovolume (cell density and chlorophyll-*a* (chl-*a*)) and biodiversity (species composition) (Rosen, 1995; Larson & Passy, 2005). Diatoms have been used as a site-specific index or water quality indicator, but diatoms require specialist knowledge and are time consuming to analyse. Periphyton as a group is considered a better choice for general application where diatom specialists are not available.

1.1.4 PERIPHYTON

The term “periphyton” is loosely synonymous to the term “benthic algae” (Stevenson, 1996), and refers to algae that grow on substrates beneath the water surface. Periphyton as a primary producer is the foundation of many food webs (Matthaei *et al.*, 2003; Zalack *et al.*, 2006) and can grow on various substrates if enough light and nutrients are available (Allan & Castillo, 2007; Tundisi & Tundisi, 2012). Periphyton forms colonies or filaments that can be visible to the naked eye, or can remain single-celled (MDEQ, 1999).

Periphyton's metrics (concentration, biovolume and chl-*a*) are affected by various physical and chemical factors (Stevenson & Bahls, 1999; Biggs *et al.*, 1999), and each of the periphytic metrics are affected differently by these factors. These various physical and chemical factors change along the

longitudinal zones of a river and influence the composition, the abundance and distribution of periphyton (Ward, 1992; Figueroa-Nieves *et al.*, 2006).

Dams alter lotic systems by regulating the flow from the natural rate. Effects may include altered thermal regimes, flow-pattern changes and changes to the disturbance regimes. These changes may result in changes in the downstream benthic communities compared to those of similar, unregulated streams (Chester & Norris, 2006).

Bacillariophyta and periphyton have been used as indicators, monitoring rivers and streams all over the world for more than 50 years (Rosen, 1995; Lavoie *et al.*, 2008). In the United States of America, some states (Montana, Kentucky and Oklahoma) developed their own diatom and periphyton indices based on various metrics (Rosen, 1995). In South Africa, comprehensive studies of Bacillariophyta were done by Cholnoky after 1952. Bate looked at the ecological aspects of Bacillariophyta assemblages during the 1990s and the application of the European Bacillariophyta index to South African conditions. During this period, Harding initiated further diatom studies, which produced protocol sets. After 2000, the applications of numerical indices were tested on South African rivers, which were later adapted and improved by Taylor and Harding (Bate *et al.*, 2002; Taylor *et al.*, 2007a).

Soft, non-diatom, periphytic algae (including Cyanophyta) have been investigated as bioindicators by various researchers (Douterelo *et al.*, 2004; Fetscher *et al.*, 2014) and described in soft-based metrics (Hill *et al.*, 2000; Porter *et al.*, 2008), while other described indices that only comprise soft algae (Schneider & Lindstrøm, 2009; Schneider & Lindstrøm, 2011). A few researchers have published on the performance of diatoms vs. non-diatom algae as bioindicators (Schneider *et al.*, 2012; Fetscher *et al.*, 2014).

1.2 MOTIVATION AND OBJECTIVES OF THIS STUDY

1.2.1 MOTIVATION

As in other parts of the world, water quality and quantity in South Africa, a known arid country, are becoming increasingly relevant. As the South African population increases, so does the demand for potable water. This, with the increasing water demand of the industrial and agricultural sectors and environmental water requirements set by the government, has led to the development of water quality indices and biomonitoring tools.

The biomonitoring tools used in South Africa today were developed on perennial rivers. However, a large part of South Africa is situated in low-rainfall areas and the rivers do not flow throughout the year. Most of the smaller rivers are also in a regulated state; this includes water release from dams, inter-river water transfer for agricultural purposes, etc.

With diatoms (part of periphyton) being such a specialised field and the regulated state of the rivers in the central South Africa, this study investigates a more accessible method to measure the water quality in regulated rivers by means of periphyton as a group (diatom and soft, non-diatom algae) and its various metrics (concentration, biovolume and chlorophyll-*a*).

1.2.2 PROBLEM STATEMENT

Can periphyton be used as an independent water quality indicator, or in conjunction with other biomonitoring indices in the biomonitoring of regulated rivers in central South Africa? In addition, which of the periphytic algal metrics are best suited to represent the periphytic community for comparison with other indices?

1.2.3 HYPOTHESES

In dam and flow-regulated rivers:

- ◆ *Null hypothesis* – Periphyton cannot be used as a water quality indicator on its own or in conjunction with other indices.
- ◆ *Hypothesis 1* – Periphyton can be used as water quality indicator on its own.
- ◆ *Hypothesis 2* – Periphyton can be used in conjunction with other biomonitoring indices.
- ◆ *Hypothesis 3* – Periphyton can be used as a preliminary water quality indicator for further water quality testing or biomonitoring.

1.2.4 OBJECTIVES

This study aims to:

- ◆ Determine which of periphyton cell concentration, biovolume and chlorophyll-a concentration is a more sensitive indicator of water quality.
- ◆ Establish if there is any correlation between periphyton measure and those of other biological, chemical and physical factors.
- ◆ Determine which environmental conditions influence the periphyton community
 - ◆ during different seasons, and
 - ◆ during different hydrological conditions.
- ◆ Establish possible water quality indicators among the periphyton present in the rivers at the chosen sites.

1.2.5 DELIMITING THE STUDY

- ◆ *Sampling frequency*: In the two months between routine sampling dates, much can happen in terms of factors that control or inhibit the growth and colonisation of periphyton.

- ◆ *Size of substrate*: The smaller the substrate, the more complicated the method of sampling periphyton is. Smaller substrates are more easily disturbed (swept away, rolled over) with little increase in current velocity.
- ◆ *Disturbance of substrate*: As sampled rivers are regulated via dams, canals and other media, the sites experience regular (more) disturbance other than natural flooding and droughts.

1.2.6 THEORETICAL AND METHODOLOGICAL APPROACH

Three accessible sites were chosen with flow over riffles or bedrock (stones in current) that could be used as sampling substrates for epilithic periphyton. Various physical and chemical aspects of the water were sampled (e.g. temperature, flow, pH, nutrients, etc.) according to prescribed methods, along with periphytic samples for the analysis. The periphytic algal concentration, biovolume and chlorophyll-a concentration were identified as the different periphytic algal components to use in the study.

All the data were processed and analysed through various prescribed methods and programmes to obtain the best possible results.

1.2.7 THESIS OUTLINE

Chapter 1 serves as a background to the study by providing a basic understanding of the worldwide availability and distribution of water, and water in South Africa. A brief summary is provided of eutrophication, biomonitoring and the potential role of periphyton as an indicator of water quality.

Chapter 2 contains literature reviews on river structure and processes, as well as on periphyton where it fits into the biomonitoring process, and the influences of physical and chemical factors (water quality) on periphyton.

Chapter 3 presents the study area and sites (Sannaspos, Bishop's Weir and Modder above Confluences) used in the study, as well as the methodology used to sample and analyse the various water and periphytic samples.

Chapter 4 is the first of the results chapters. This chapter investigates the relationship and correlations between the periphytic algal concentration, biovolume and chlorophyll-a concentration at the different sites. It also investigates influences and relationships of physical and chemical factors on the various mentioned periphytic algal components.

Chapter 5 examines the effect of seasonality on the periphyton through the relationship and effect of the physical and chemical factors of flowing water, as well as to determine if one or more of the factors are the main driving force for seasonality.

Chapter 6 explores the possibility that the rivers and sites are influenced by river regulation through dam releases or agricultural water canals. It investigates the hydrological differences between dry and wet periods, and whether it has an effect on the periphytic algal concentration, biovolume and chlorophyll-a concentration, and the difference in algal composition.

Chapter 7 compares the methods and results of the various biomonitoring indices with those of periphyton at the three study sites. The sampling methodology and the results obtained according to the level of ecosystem health are described specific to each of the sampling sites of this study.

Chapter 8 contains the final discussion and conclusions on the results (Chapters 4 to 7), relevant to periphyton as indicator of water quality in regulated rivers in central South Africa.

CHAPTER 2

LITERATURE REVIEW

2.1 WORLDWIDE WATER AVAILABILITY

Water, like the air we breathe, is a very common substance, which we take for granted. It vented upwards through volcanoes, fumaroles and geysers from deep within the earth during 5 000 million years of geological history. Throughout time, the water accumulated on the surface of the earth, to the extent that it covered 70% of the surface with a mean depth of 3.8 km. Over the millennia, the distribution and abundance of water in the form of rain had created forest and deserts at will. On several occasions during the past 2 million years, water in the form of glacial ice changed continents by damming lakes, depressing land, shaping valleys, recharting river courses, depositing mounds of gravel and lowering the levels of the world's oceans by more than 10 m (Vallentyne, 1974).

Water means food and survival on land when it comes at the right time; if not, it leads to famine. Water is also the chemical basis of life, "*a universal requirement for the origin and persistence of life*" (Schindler & Vallentyne, 2008).

In nature, water is not evenly distributed. Inland water covers only two percent of the earth's surface (Wetzel, 2001) and fresh, liquid water forms a small proportion of that (<1%). Of the last-mentioned small proportion, about a third ($4 \times 10^6 \text{ km}^3$) is surface water and the rest ($1.1 \times 10^7 \text{ km}^3$) is groundwater (Davies & Day, 1998).

Water is not evenly distributed across the surface of the earth's major continents. An example of this is in South America where the total groundwater runoff is the highest, nearly twice per area than those of other continents, while Africa has the

lowest groundwater runoff. South America also has the highest evaporation per area (Wetzel, 2001).

2.2 WATER IN SOUTH AFRICA

The climate in South Africa ranges from a few relatively humid parts (>500 mm/yr) in the east, with semi-arid to hyper-arid areas in the western part. The average rainfall in central South Africa is 400–600 mm/yr, but vast areas of the country receive much less. Except for the South-Western Cape and a few high-altitude areas in the Drakensberg, the evaporation exceeds the precipitation by far in many other parts of the country. In Gauteng, the industrial heartland, the evaporation is double that of the rainfall, while in the Lower Orange River Valley it is an extraordinary ten times the amount of the rainfall. South Africa does not have a water surplus, as most of the rain evaporates soon after it has reached the ground and re-enters the atmospheric phase of the water cycle (DWAF, 1986; Davies & Day, 1998).

The rainfall in South Africa is highly seasonal. It occurs at different times of the year in the different climatic regions (Tyson, 1986). During winter, it rains in the western parts, leaving the southern interior (incl. the Karoo) in a rainy shadow. In summer, the rain normally falls in the east and in the north, while the south and west endure long, dry periods (Davies & Day, 1998).

With freshwater a limiting resource, combined with the increasing population growth, the demand for water increases (domestic, agricultural and industrial). Thus, by the mid-21st century, one might experience a water demand higher than the resource can provide (Davies & Day, 1998; García-Rodríguez *et al.*, 2007; **Fig. 2.1**). Inter-basin water transfer schemes have already been constructed to transfer water to areas in need. Two of these schemes, the largest in South Africa, are the Orange-Fish-Sunday and the Lesotho Highlands Schemes (Pallett, 1997). When there is no more water available in South Africa, water will have to come from neighbouring countries via transfer schemes.

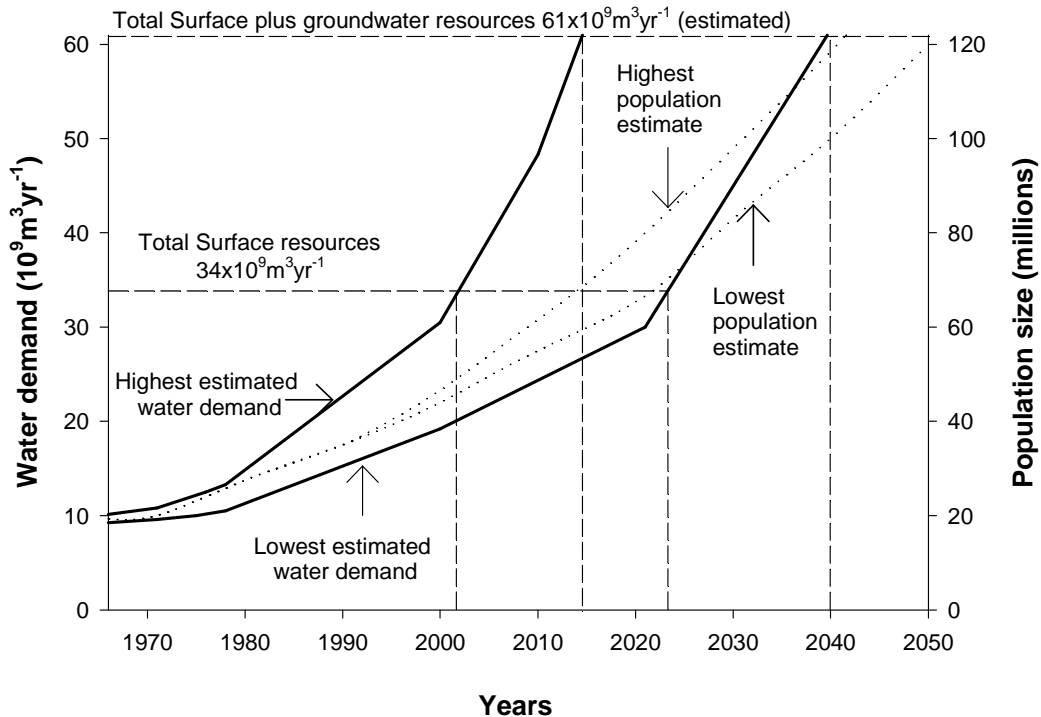


Figure 2.1: The relationship between demand for water and size of the human population of South Africa. The two dotted curves represent the fastest and slowest estimated rates of population growth. The two solid curves are the highest and lowest estimates of the amount of water needed to satisfy human requirements (redrawn from Davies & Day, 1998).

Various water transfer schemes exist on the Modder River. The Caledon-Modder Transfer Scheme and Mazelspoort Scheme are found in the upper part of the Modder River. Water is transferred from the Caledon River to the upper Modder River. This water is treated at the Rustfontein Dam and Mazelspoort Weir for the use (mainly for domestic use) by Bloemfontein and Botshabelo population. Annual estimated losses in this system add up to 6.09 million m^3 (ORASECOM, 2013a).

The lower part of the Modder River is included in the Orange-Riet Water Scheme as part of the Scholtzburg Irrigation District, and is mainly used for irrigation (ORASECOM, 2013a).

2.3 STRUCTURE OF RIVER ECOSYSTEMS

“*There is no such thing as the typical river system*” (Closs *et al.*, 2004), but people who live in the same area will have the same idea of a river. Rivers and streams are closely linked to their watersheds or catchments. They can never be considered by themselves (Horne & Goldman, 1994) and are essential to surface-water ecosystems (Wetzel, 2001). It not only includes hydrology, diversity of habitat and channel diversity, sediments and solutes and biota (Allan & Castillo, 2007), but also the interaction between these physical, chemical and biological processes (Bere & Tundisi, 2010).

Running water has various characteristics that differentiate it from lake ecosystems:

- ◆ it comprises freshwater environments that flow and are called *lotic systems*;
- ◆ water flows in one direction along a gradient;
- ◆ the relative residence time of running water is very short;
- ◆ the minimal flow (slowest current) in a river is about the maximum flow one would find in a lake;
- ◆ flowing water has high energy, which affects the stream morphology, sedimentation, the physics and chemistry of the water, and the biology of the inhabiting organisms;
- ◆ the further downstream in a river, the more complex it becomes, as every disturbance along the way influences the organisms and processes further downstream; and
- ◆ the above mentioned all form a continuum, known as the river continuum concept (RCC) (Horne & Goldman, 1994; Davies & Day, 1998; Giller & Malmqvist, 1998; Wetzel, 2001; Bere & Tundisi, 2010).

2.3.1 MORPHOMETRY AND MORPHOLOGY

Rivers (large, with low gradients at low elevations) and streams (small, with steep gradients) are made up by various tributary streams that form a drainage

basin/catchment area. As two of the same-order streams meet, they create a next higher-level order. The confluence with a smaller-order stream does not influence the stream order of the main stream (Horne & Goldman, 1994; Wetzel, 2001).

Streams usually do not flow in straight lines over long distances, but meander with bends along the contours of the landscape. Channels may branch off to connect again lower downstream, especially in the lower reaches of the river (Giller & Malmqvist, 1998).

2.3.2.1 Longitudinal zonation/profile of rivers

Downstream from the headwater of a river almost everything changes (Giller & Malmqvist, 1998), namely an increase in size, the volume of water in the river, increase in catchment area, physical and chemical characteristics, etc. These variables have attracted river ecologists' attention since the early 1900s (Ward, 1992). This longitudinal profile can be used to 'classify' rivers according to certain characteristics (Giller & Malmqvist, 1998). Bigger drainage area potentially equals a higher flow (Church, 1992).

The profile zones are roughly erosion (headwaters/mountain stream), transfer (middle reaches/foothills) and sediment deposition (lower river/floodplains) (Allan & Castillo, 2007). The ratio between the zones differs from river system to river system.

2.3.1.1.1 Headwaters

This part of a river starts small, with drips and trickles. The sources of this origin include ice and snow melts, rainfall runoff and springs/groundwater (Dodds, 2002; Moss, 2010). As soon as all these trickles and rivulets merge, a stream is born.

Headwater in mountainous streams mostly flows over cobbles and boulders (incl. bedrock) with a steep gradient (Church, 1992). It is also in these zones that pools (incl. rock pools), riffles and runs are mostly found.

The water is usually clear (high radiation) – even with a sandy bottom – low temperatures (higher temperatures if in temperate or tropical regions) and with high oxygen concentrations. The streams are shallow and mostly fast flowing. The water usually has low nutrients if there is no pollution, and therefore the autochthonous primary production is mainly periphyton (e.g. epilithic), when streams are not shaded.

Other food/energy sources for organisms are mostly allochthonous, coarse particulate organic matter (CPOM), such as twigs, flowers, bark, leaves, etc., which are washed in with sediment during runoff, blown in from adjoining terrestrial vegetation or falls from the riparian canopy (Allan & Castillo, 2007; Moss, 2010).

2.3.1.1.2 Middle reaches

According to Moss (2010), the meandering of streams are “*the channels’ attempt to accommodate higher flows*”, while it lengthens to expand for volume increase. These middle reaches form the transition between the headwater and the lower river, where sediment is transferred.

As tributaries join the main stream in the foothills, the river widens, while the slope gets a bit gentler (Davies & Day, 1998). As the stream widens, there is less shade from the riparian canopy. The water is also less pure because of mineral leaching, abiotic processes and activities from upstream living organisms.

There are still pools, riffles and some rapids in this reach. Most riffle-like areas are also in the form of runs or glides. The pools have a more sandy than rocky bottom, as sediment and debris accumulate (Church, 1992).

The primary production still consists of periphyton (if water is clear enough over habitat) and some phytoplankton. Another food/energy source is fine particulate organic matter (FPOM) transported from upstream (Horne & Goldman, 1994).

2.3.1.1.3 Lower river

In this part of the river, the meandering continues as it widens over a plateau or coastal plain, while other tributaries join (Davies & Day, 1998). As the river grows larger (Moss, 2010), it changes from being erosive to depositional.

As the tributaries join, the discharge increases, but the current speed is reduced as the gradient decreases. Sediment and other debris are deposited, smothering the stones and boulders on the bottom, leaving a sandy and silty bottom substrate (Davies & Day, 1998). Towards the ocean, the substrate becomes smoother/finer and muddier.

Because of the sedimentation, riffles are (mostly) absent in these muddy, debris-loaded waters (Horne & Goldman, 1994). As the water is somewhat warmer than in the upstream parts, less oxygen dissolves in the water from the atmosphere. The oxygen in these parts is low, as there is a high uptake from the vast amount of organisms (Davis & Day, 1998).

More nutrients and salts are dissolved in the water washed downstream and coming from dead plant and animal material (FPOM) (Davies & Day, 1998). As the river is wide with little shade and more turbid, the autochthonous primary production is done by true phytoplankton (Horne & Goldman, 1994).

2.3.1.1.4 River continuum concept (RCC)

All the zones combined form a delicate continuum from headwaters to the ocean, called the river continuum concept (RCC). The RCC describes “*the*

structure and function of communities along a river system" (Vannote *et al.*, 1980). Giller and Malmqvist (1998) describe the RCC as to "*include a number of predictions of longitudinal changes along the continuum*".

The major concepts that are associated with a river include (Calow, 1992; Cummins, 1992; Davies & Day, 1998; Giller & Malmqvist, 1998; Dodds, 2002; Lampert & Sommer, 2007):

- ◆ flow regime and gradient;
- ◆ autochthonous vs. allochthonous (CPOM/FPOM) production (**Fig. 2.2 & Fig. 2.3**);
- ◆ periphyton vs. phytoplankton as primary production (**Fig. 2.2**);
- ◆ nutrient spiralling;
- ◆ relationship between invertebrate functional feeding groups (FFGs; **Fig. 2.2**); and
- ◆ photosynthesis/respiration ratio (P:R) (**Fig. 2.3**).

An idealised summary of the RCC is displayed in **Table 2.1**.

The continuity of the RCC is a good concept, but disturbances from within and outside the river system breaks it up, as Ward and Stanford (1983) explain with their serial discontinuity concept (SDC). Some of these factors can be natural or cultural:

- ◆ when lower-order tributaries join the main river (Lampert & Sommer, 1998);
- ◆ natural features, such as geology and gradient changes (Calow, 1992);
- ◆ building of dams, reservoirs and impoundments (Ward & Stanford, 1983);
- ◆ pollution of various kinds (Ward & Stanford, 1983); and
- ◆ land management (Calow, 1992).

The Modder River is described by Seaman *et al.* (2001) not to consist of an successive series of geomorphological zones as described in an idealised RCC, and that approximately 90% of the river belongs to the Lowland (sand bed or floodplain) zone.

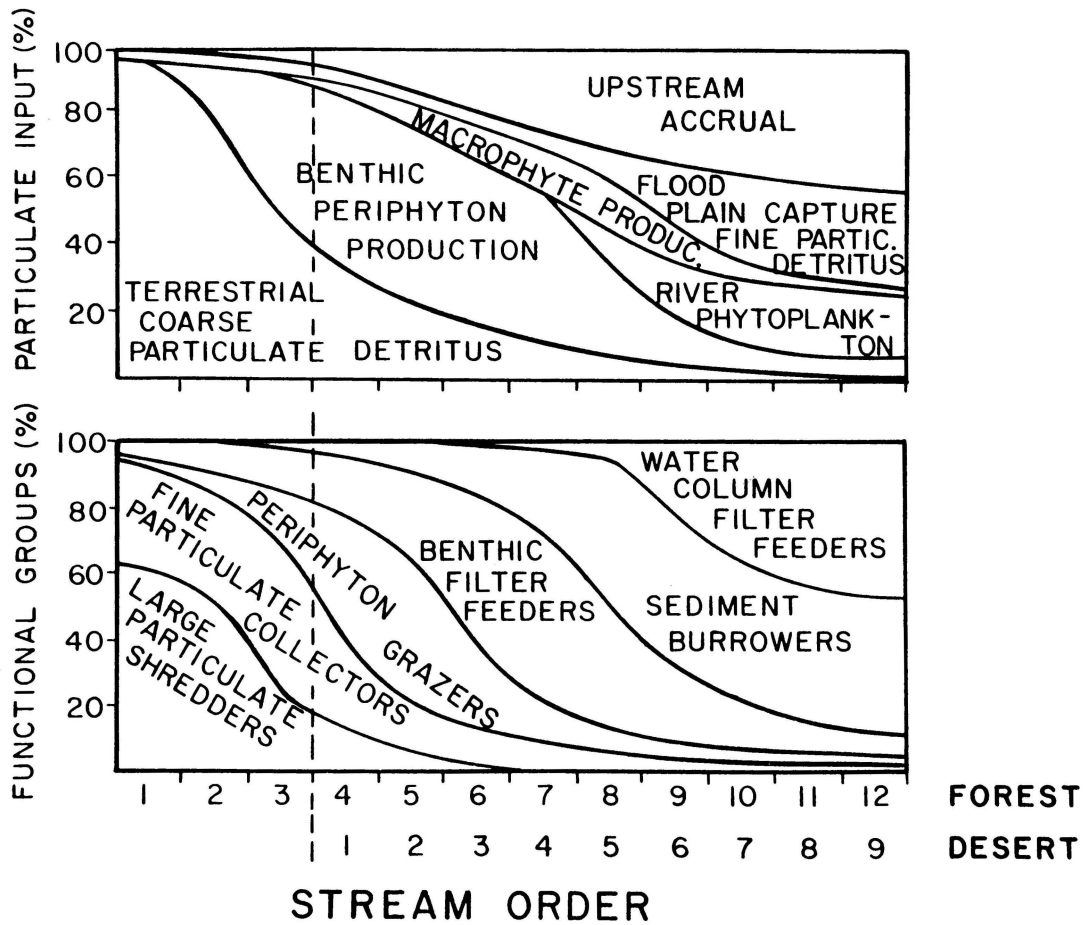


Figure 2.2: Expected changes of the relationship in the particulate organic matter and the functional feeding groups along a river system (Minshall *et al.*, 1985).

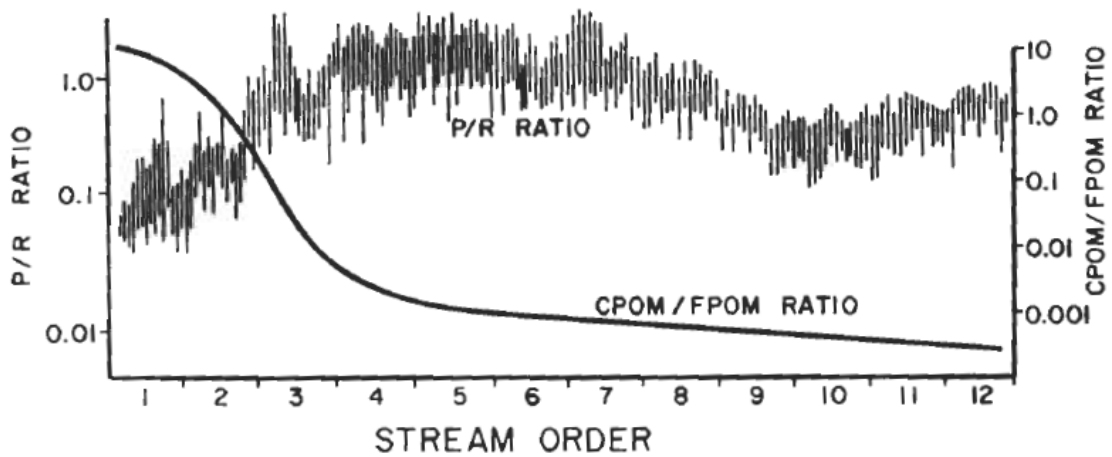


Figure 2.3: A hypothetical P/R and CPOM/FROM ratio along the river continuum (Vannote *et al.*, 1980).

Table 2.1: Idealised features (summary) of the river reaches interpreted from the river continuum concept (redrawn from Ward, 1992).

	River reaches		
	Headwater	Middle	Lower
Temperature	Cool, low amplitude	High amplitude	Moderate amplitude
P/R	<1.0	>1.0	<1.0
Energy source	Terrestrial detritus	<i>In situ</i> PP*	Transport detritus
Bottom light	Low	High	Low
Nutrient availability	Low	High	Low
Attached algae	Sparse	Abundant	Sparse
Submerged angiosperms	Absent	Abundant	Sparse
Plankton	Absent	Absent	Present
Leaf litter	Abundant	Sparse	Negligible
Invertebrates:			
Shredders	Co-dominant	Rare	Absent
Collectors	Co-dominant	Co-dominant	Dominant
Grazers	Sparse	Co-dominant	Absent
Predators	Low	Low	Low
Fish fauna	Cool-water invertivores	Piscivores and invertivores	Planktivores and bottom feeders
Environmental heterogeneity	Low	High	Low
Biotic diversity	Low	High	Low

* PP = primary production

2.3.2 PHYSICAL FACTORS

Unlike lentic systems, the factors/characteristics of lotic systems are less stable and they differ, depending on which reach/zone they are in. This is true for the physical, chemical and biological factors.

Flow, turbidity and temperature are the physical components of river water, and can be seen as a driving force behind the other factors.

2.3.2.1 Flow

Rivers flow in one direction, which distinguishes them from other aquatic systems (Callow & Potts, 1996; Lampert & Sommer, 2007). Except during prolonged dry periods, water flow is never stable and flow may change every hour during rainy seasons (Moss, 2010). River regulation also plays a role in the flow regime of a river. This can be seen in the flow of the Orange River, where flow regulation diminishes extreme high-flow and low-flow events below the Gariep Dam (Davies & Day, 1998).

Flow is determined by discharge/runoff, gradient and width of streams (Horne & Goldman, 1994), and therefore shows seasonal and daily variations. Flowing water transports sediment and thereby plays a role in the turbidity of the water (Closs *et al.* 2004). Except for light, all vertical gradients are eliminated by the constant mixing of streams (Lampert & Sommer, 2007).

The mean flow of a river is at 60% of its depth below the surface (Davies & Day, 1998; Giller & Malmqvist, 1998; Moss, 2010) and it is approximately 80–90% of the velocity of the surface (Giller & Malmqvist, 1998).

Depending on the velocity of the stream flow, it also influences the substrate size (Allan & Castillo, 2007). Corresponding with the structuring of the streambed are the organisms that colonise them. The higher the flow, the more adaptations the organisms must have to remain in/on the habitat (Lampert & Sommer, 2007). For all organisms that remain stationary, the nutrients that are not utilised immediately are lost due to flow, but on the other hand, new nutrients constantly flow past.

2.3.2.2 Turbidity

Turbidity is a visual property of water and implies a reduction or lack of clarity, which results from the presence of suspended particles or suspensoids (Wetzel,

2001; Dallas & Day, 2004). Included in suspended solids are tiny clay and silt, dead particulate organic matter and living organisms (Davies & Day, 1998), coming from soil erosion and the re-suspension of sediments (Wetzel, 2001), as well as algal and colloidal materials (Allan & Castillo, 2007).

Changes in turbidity can be a natural seasonal variation linked to flow (rain runoff, etc.; Dallas & Day, 2004) or agricultural activities, etc. With rainfall, turbidity increases naturally in rivers (Davies & Day, 1998; Dodds, 2002) and, depending on the water speed, the size in suspension varies (Davies & Day, 1998). The size of the stream order also influences turbidity: the bigger the stream order, the higher the turbidity (Wetzel & Ward, 1992).

There is a close positive relationship between turbidity and suspended solids (Walling & Webb, 1992), $1 \text{ NTU} \approx 1 \text{ mg/l}$ total suspended solids (TSS). However, the relationship is not always linear; it all depends on the size and quantity of the sediment or suspended solids (Ryan, 1991; Dallas & Day, 2004).

Turbidity in undisturbed rivers is much lower with much higher transparency than its disturbed equivalent (Wetzel & Ward, 1992). Before extreme land clearance and agriculture, the inorganic and organic turbidity was much lower (Reynolds, 1992). Other potential sources of turbidity are erosion of land surfaces, construction of dams and roads, and anthropogenic processes (industries, mining, sewage, etc.). Because of the above-mentioned, turbidity is relatively common in most South African rivers (Dallas & Day, 2004), especially when the very fine, suspended particles are electrically charged (Davies & Day, 1998).

Higher turbidity decreases the light penetration, as the suspended particles scatter solar radiation and therefore lower the photosynthesis rate and thus the primary production (Wetzel & Ward, 1992; Horne & Goldman, 1994; Wetzel, 2001; Dallas & Day, 2004) as well as the growth of periphyton and phytoplankton (Reynolds, 1992; Walling & Webb, 1992; Giller & Malmqvist, 1998). In New Zealand, it was found that a turbidity of 5 NTU can decrease primary production between 3% and 13% (Dallas & Day, 2004).

Suspended solids interfere with the feeding of aquatic invertebrates, as they settle and cover periphyton, and they can cause death by smothering (Ryan, 1991). Turbidity also affects fish in the following ways: interfering with gill function, covering spawning habitat, and the decrease of primary production leads to the reduction of food availability and interferes in the hunting of predatory fish (Closs *et al.*, 2004; Dallas & Day, 2004).

2.3.2.3 Temperature

The temperature of river water has been studied for centuries, dating back to measurement of water temperatures on the Nile River around 1799 to 1801 (Webb *et al.*, 2008). Webb *et al.* (2008) further state that temperature is an important property (physical) of flowing water, as it influences various other aspects of water, e.g. dissolved oxygen concentration, turbidity through sediment displacement, salinity, and metabolic rates/biology of aquatic organisms (Macan, 1958; Biggs, 1996; Davies & Day, 1998; Torgersen *et al.* 2001; Allan & Castillo, 2007); not to speak of its economic, agricultural and recreational importance (Webb *et al.* 2008). In short, river temperature can be seen as major controller of aquatic life (Smith, 1972) and is one of the ways energy affects water bodies (Roos & Pieterse, 1994).

Atmospheric temperature has the most important direct influence on running/flowing water (Smith & Lavis, 1975; Rosa *et al.*, 2013), as much as 86% (Mohseni & Stephan, 1999), as well as in determining the temperature of groundwater (Ward, 1985). The water temperature of running water can be influenced by various other interrelated factors besides atmospheric temperatures (**Fig. 2.4**). That includes topography, stream discharge and streambed composition (Williams & Boorman, 2012).

Temperature is not normally 'visible', except under extreme conditions (freezing or boiling). Water temperature is the lowest in the upland and warmer in the lowland of most streams, even arctic streams (Horne & Goldman, 1994) thus

corresponding with the longitudinal gradient (Lampert & Sommer, 2007). Small streams are warmed more rapidly by sunlight than large rivers are (Lampert & Sommer, 2007), while slow-flowing rivers may have a difference in temperature between that on the surface and the temperature at the bottom (Giller & Malmqvist, 1998). However, the forming of thermal density stratification in rivers is very rare (Wetzel, 2001).

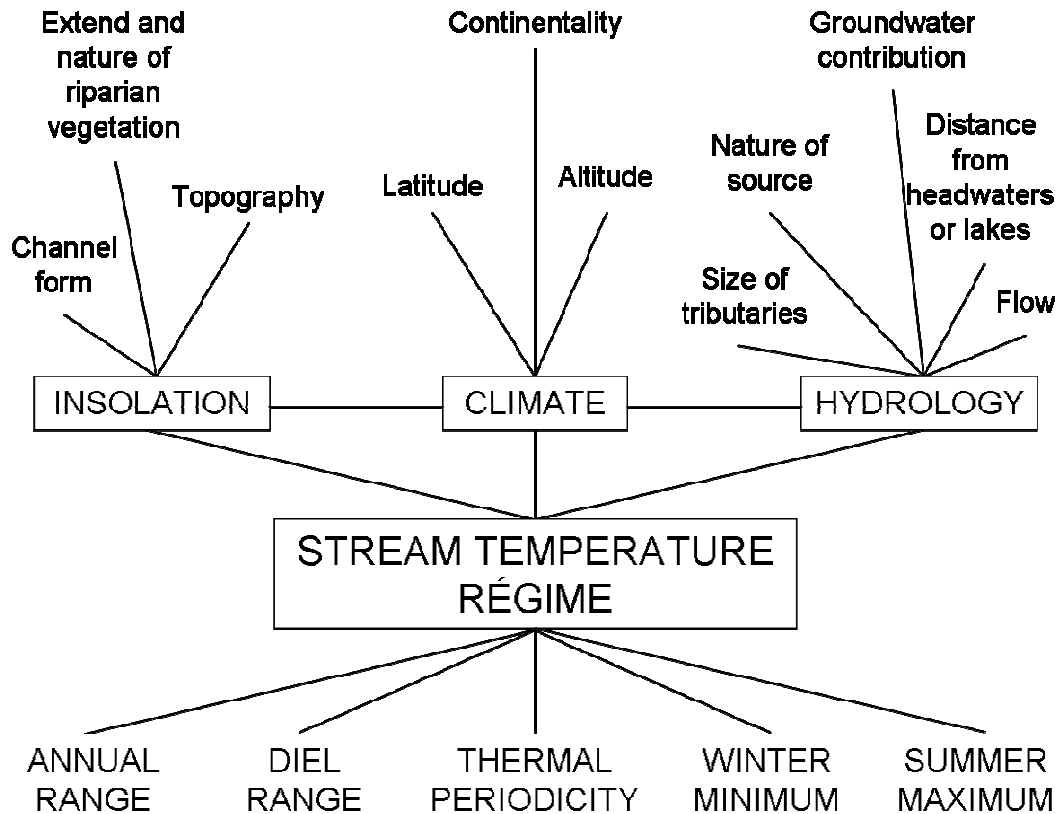


Figure 2.4: The major interrelated factors determining river temperature regime (drawn from Giller & Malmqvist, 1998).

The main source of the water in the stream is important, regardless of whether it is from snowfall, rainfall runoff or groundwater (Smith & Lavis, 1975). Furthermore, the percentage contribution of groundwater, the flow and the discharge of river/stream play a role in temperature regimes (Ward, 1985).

The seasonal and daily temperature variations are a result of atmospheric influence (Gordon *et al.*, 2004; Allan & Castillo, 2007) and solar radiation (Tundisi & Tundisi, 2012). The amount of shading also influences the water's

temperature (Giller & Malmqvist, 1998), but even then, seasonal and daily variations exist (Horne & Goldman, 1994). If there is a groundwater input, the temperature can be stable or constant (Allan & Castillo, 2007).

Jakob *et al.* (2003) and Uehlinger *et al.* (2003) all state that flow can change the temperature of water, especially if it is downstream from a dam. Other anthropogenic/man-induced changes that can modify/influence the temperature of rivers include thermal pollution and activities that alter the riparian vegetation of the catchment (Ward, 1985).

Human activities in the catchment can cause flow reduction, which may lead to the increase of water temperature as pools form or the river is impounded. The influence of dams on its downstream river is controlled by water release; if it is from the warmer epilimnion, the water temperature will increase, while if the released water is from the colder hypolimnion, it will decrease (Horne & Goldman, 1994; Allan & Castillo, 2007). Reduced flow between water releases over long periods may increase the retention time of the river below a dam and thus increase the heat absorption of the water (Allan & Castillo, 2007).

In South Africa, ecologists took notice of the importance of the temperature regime in rivers and the effect it has on the aquatic ecosystem (Dallas & Rivers-Moore, 2012). It was therefore recommended that it should be taken into consideration when hydrological assessments are done for Ecological Reserves.

Alterations in the riparian vegetation mostly occur around deforestation and agricultural activities. With deforestation, when the canopy of a stream is removed, the water is exposed to more solar radiation and water temperatures increase (Giller & Malmqvist, 1998; Webb *et al.*, 2008). Similarly, clearing of the riparian zone occurs when animals graze away the vegetation and expose the water to increased solar radiation. The clearing of the riparian vegetation further results in more sediment being deposited in the water through erosion, wind or rainwater runoff (Allan & Castillo, 2007; Webb *et al.*, 2008).

Temperature and turbidity (suspended solids/sediments) are closely linked. Williams (2006) explains that the higher the turbidity, the higher the heat absorption of the surface water, as suspended particles absorb and scatter sunlight (Paaijmans *et al.*, 2008). With a river constantly flowing and mixing (Giller & Malmqvist, 1998), the entire river-water column temperature increases (especially in a shallow river). On the other hand, heat energy is also lost through reflection from the water surface, evaporation, back radiation from the water, etc. (Wetzel, 2001).

Macan (1958) has further found that the highest water temperatures occur when the sun shines after rain. Two effects drive this: the high humidity linked to the rain prevents the cooling of the surface water (no evaporation) and runoff flows into the stream from the warm soil (Wiley & Seelbach, 1997).

Not only does water temperature play a role in the availability of dissolved oxygen in the water; with higher temperatures the dissolvability of oxygen decreases and vice versa (Horne & Goldman, 1994), but it influences other characteristics like suspended solids content, and chemical and biochemical reactions. It also affects the distribution, evolution and ecology of aquatic organisms (Walling & Webb, 1992; Allan & Castillo, 2007).

In poikilothermic freshwater, plants, animals and their internal processes (photosynthesis, respiration, digestion, muscle action) are influenced by temperature. It also influences egg development, emergence time, larval growth rates, adult size and fecundity (Davies & Day, 1998; Giller & Malmqvist, 1998).

2.3.3 CHEMICAL FACTORS

The chemical composition of freshwater is very variable in rivers, more than in lakes (Allan & Castillo, 2007). It also varies over time in association with the discharge regime and as seasons change. It can be overshadowed by the physical characteristics in relation to the distribution of biota (Horne & Goldman, 1994).

2.3.3.1 Oxygen (O₂)

Of all the gasses that may dissolve in water (including nitrogen and carbon dioxide), oxygen is definitely the most significant, as it is essential to all aquatic organisms for survival. The concentration of dissolved oxygen in water is controlled by physical, chemical and biological factors, and it represents either a source or a sink (Walling & Webb, 1992; Davies & Day, 1998; Tundisi & Tundisi, 2012).

The atmosphere is the main source for oxygen, but not the only one (Horne & Goldman, 1994). The air consists of about 21% oxygen per volume or 300 mgO₂/ℓ of air, while dropping at 0 °C and at sea level to only 14.6 mgO₂/ℓ (Horne & Goldman, 1994; Allan & Castillo, 2007). Thus, air pressure influences oxygen solubility as follows: oxygen is more soluble at high pressure and less soluble at low pressure (low pressure is associated with rainfall; SAWS, n.d.(a); Davies & Day, 1998; Wetzel, 2001).

The solubility of oxygen in water is not only determined by the temperature of the water, as solubility is less in warmer water (Horne & Goldman, 1994; Davies & Day, 1998; Giller & Malmqvist, 1998; Allan & Castillo, 2007), but also by the normal flow of water (Davies & Day, 1998). As water flows/tumbles over rocks and boulders, it incorporates air bubbles from the atmosphere. Oxygen in small turbulent streams with little pollution will have oxygen concentrations near saturation (Allan & Castillo, 2007). The water residence time and depth also affect the oxygen balance (Walling & Webb, 1992). The last-mentioned is one of the influences on the longitudinal change in oxygen levels in the water, as oxygen concentration in the water decreases along the river's length (Davies & Day, 1998).

Salinity is another factor to affect the oxygen concentration in water; the higher the salinity, the lower the solubility of oxygen in the water (Whipple & Whipple, 1911; Dallas & Day, 2004; UW-GB, 2005).

Added to the above-mentioned impacts on the solubility of oxygen in water are respiration and photosynthesis; that is, the consumption and production of oxygen, respectively.

All animals and plants respire 24 hours a day and some bacteria also use oxygen as part of their metabolic processes. The most common is when organic matter is decomposed by bacteria. The more organic matter in a system, the more oxygen is consumed during decomposition (Ryan, 1991; Horne & Goldman, 1994; Davies & Day, 1998; Allan & Castillo, 2007). When great quantities of organic matter enter a system via rain runoff or through a polluting point (sewage), the over-utilization of oxygen by the bacteria can cause oxygen depletion in the process of decomposition (Dallas & Day, 2004), which could cause other animals to die of a shortage of oxygen.

Additionally, oxygen is also a product of photosynthesis by higher plants (aquatic vegetation) and algae (Horne & Goldman, 1994; Davies & Day, 1998). In great quantities they (more likely algae) can cause oxygen super-saturation (>100%) during daylight (Walling & Webb, 1992; Dallas & Day, 2004). This in itself can be lethal/toxic to fish and other aquatic organisms (Dallas & Day, 2004). However, it was found that thick filamentous algal mats could decrease the dissolved oxygen by as much as 50% through respiration at night (Walling & Webb, 1992).

2.3.3.2 pH

The geology, atmospheric influences and biotic activities determine the pH of natural water (Davies & Day, 1998; Dallas & Day, 2004). Most freshwater systems have a pH of around 6–8 (neutral), and are relatively well buffered. In very pure water, the pH can change vary rapidly (Dallas & Day, 2004).

The concentration of hydrogen ions (H^+) determine the pH (Dallas & Day, 2004), $H_2O \leftrightarrow H^+ + OH^-$ and $pH \approx -\log_{10}[H^+]$ (Allan & Castillo, 2007). The pH is in

logarithmic 10 scale; therefore, a decline in 1 pH unit leads to a tenfold increase in hydrogen ion concentration (Allan & Castillo, 2007) and the water becomes more acidic (Davies & Day, 1998) and vice versa.

The link between pH and free carbon dioxide is that at low pH (<4) all CO₂ is free; at neutral (6–8) most CO₂ is in the form of HCO₃⁻; and at high pH (>10) it is in the form of CO₃²⁻ (Dallas & Day, 2004).

Natural acidification can occur, but anthropogenic influences via acid rain and effluent from mines mostly enhance this phenomenon. The toxicity (chemical species) of many elements is determined by pH, e.g. the mobilisation of aluminium with the lowering of pH (<6 pH; Dallas & Day, 2004). With extreme photosynthesis, because of eutrophication or under natural conditions, very high pH can occur in lentic waters (Dallas & Day, 2004), as all the free CO₂ is consumed. In lotic water, anthropogenic causes (e.g. chemical pollution) can result in high pH.

2.3.3.3 Total dissolved salts (TDS)/electrical conductivity (EC)

Total dissolved salts (TDS) are the total amount of dissolved material in water, while electrical conductivity (EC) is a measure of the property of the water sample to conduct an electrical current (amount of dissolved ions) (Davies & Day, 1998; Allan & Castillo, 2007). Salinity is used as the “*chemical term for the sum concentration of all the ionic constituents dissolved in inland water*” (Wetzel, 2001).

The ions that form the bulk of the TDS/EC are calcium (Ca²⁺), magnesium (Mg²⁺), sodium (Na⁺) and potassium (K⁺) as the major cations, with chloride (Cl⁻), sulphate (SO₄²⁻), bicarbonate (HCO₃⁻) and carbonate (CO₃²⁻) as the major anions (Davies & Day, 1998; Allan & Castillo, 2007).

TDS and EC in water are directly proportional to each other (Allan & Castillo, 2007), with a factor ranging from 5.5–7.5, depending on the system (DWAf,

1996a). In South Africa, the equation is commonly used with 6.6 (**Eq. 2.1**) as the factor (Dallas & Day, 2004).

$$\text{TDS (mg/}\ell\text{)} = \text{EC (mS/m at } 25^{\circ}\text{C)} \times 6.6 \quad \text{Eq.2.1}$$

Geology and weathering of rocks play a major role in the ionic composition (Allan & Castillo, 2007), as different types of geology give rise to different ions (Wetzel, 2001), in association with flow variation.

The TDS in rainwater are very low (diluted), only a few milligrams per litre (Allan & Castillo, 2007). As runoff volumes increase with increased flow in rivers, the TDS concentrations decrease (Webb & Walling, 1992). The TDS of rivers increase as they flow downstream (Gordon *et al.*, 2004), while saline groundwater in-stream springs are also a source for increased concentrations.

Other environmental influences include precipitation, evaporation, change in elevation, temperature, wind direction and speed, and type of vegetation (Wetzel, 2001). Anthropogenic influences include saline industrial effluent, irrigation and the reuse of water (Dallas & Day, 2004).

The average TDS concentration in river water is about 100 mg/ℓ (Allan & Castillo, 2007) and in rivers with minor significant pollution about 120 mg/ℓ (Webb & Walling, 1992). In South Africa the lowest values recorded were around 10–27 mg/ℓ and the highest around 84 020 mg/ℓ (Dallas & Day, 2004). TDS concentrations influence the abundance and distribution of organisms, as most have salt tolerance levels and ranges (Gordon *et al.*, 2004).

2.3.3.4 Nutrients

Various elements are required for normal growth and reproduction by organisms; of these, phosphorus (P) and nitrogen (N) are the most important (Davies & Day, 1998). Other nutrients include carbon (C) and silicon (Si) (Horne & Goldman,

1994). Primary producers rely on the surrounding water to supply them with the necessary nutrients (Allan & Castillo, 2007). Only a few elements are toxic (at very high concentrations) for organisms (Davies & Day, 1998).

Heterotrophs get their nutrients through the food they ingest; thus, their energy is usually limited by carbon (Allan & Castillo, 2007).

Autotrophs are often limited by the availability of nutrients. Heavily utilised macronutrients include N, P, potassium (K), calcium (Ca) and magnesium (Mg). Some micronutrients are also needed (in small amounts), of which Si is essential for Bacillariophyta (Allan & Castillo, 2007).

N and P are mostly the limiting factors for autotrophs, as they influence primary productivity, especially in benthic algal population. The N:P ratio needed by plants/algae is 7:1 by weight and 16:1 by element. If the N:P is >10 (by weight), then P is the limiting nutrient, and <10 it is N. When the PO_4^{3-} is very low, one uses the N:P, otherwise one can use $^1(\text{NO}_3^- + \text{NH}_4^+):\text{PO}_4^{3-}$ (or DIN:DIP) (Horne & Goldman, 1994; Allan & Castillo, 2007).

2.3.3.4.1 Phosphorus (P)

Phosphorus is a required nutrient in cell processes and a part of DNA (Davies & Day, 1998). It is therefore a key nutrient in primary production as it can limit growth (Dodds *et al.*, 2002).

As phosphorus has no gas form/phase in the atmosphere, the PO_4^{3-} found in natural waters is usually >1 $\mu\text{g}/\ell$ (below detection limit) (Dodds *et al.*, 2002). It leaches in small amounts from rocks and soils (Giller & Malmqvist, 1998), and is thus the least abundant of all the macronutrients (Wetzel, 2001). However, plants do not need that much P to start growing. Eutrophication may occur at low levels of P (**Table 2.2**). Sharpley and Rekolainen (1997) have found less

¹ Formulas and acronyms will be described in Sections 2.3.3.4.1 and 2.3.3.4.2.

than 0.01 mg/ℓ dissolved P to be enough while according to Hesketh and Brookes (2000), Oliver and Ganf (2000), less than 0.02 mg/ℓ total P. Thus, according to Van Ginkel (2008), biological productivity only needs a small ratio fraction of P to sustain itself.

Table 2.2: The ranges of the various trophic levels of dissolved inorganic phosphorus (DIP) and nitrogen (DIN) concentrations (DWAF, 1996a).

Trophic levels	DIP (mg/ℓ)	DIN (mg/ℓ)
Oligotrophic	≤0.005	≤0.2
Mesotrophic	>0.005 – 0.025	>0.2 – 0.5
Eutrophic	>0.025 – 0.025	>0.5 – 1.5
Hypertrophic	>0.25	>1.5

Inorganic P is mostly in the form of phosphate ions (PO_4^{3-}) in stream water (Davies & Day, 1998; Allan & Castillo, 2007) as dissolved nutrients or attached to inorganic particles (Webb & Walling, 1992) such as sediment. High concentrations of soluble reactive phosphate (SRP) or dissolved inorganic phosphate (DIP) are seldom found in non-polluted water, as they are mostly absorbed by plants (Davies & Day, 1998; Allan & Castillo, 2007).

Sources of phosphorus are the erosion of rock and sediments, decaying organic matter (e.g. canopy leaves, aquatic plant and animal waste), runoff, domestic, agricultural and industrial wastes, including fertilizer, manure, sewage, etc. (Horne & Goldman, 1994; Allan & Castillo, 2007).

Phosphate concentrations increase with an increase in flow (Webb & Walling, 1992), while low oxygen concentration plays a primary role in the release of P from sediments (Wetzel, 2001), as part of the buffer system that occurs in rivers and streams regarding phosphate adsorption and desorption (Horne & Goldman, 1994).

Phosphorus uptake is connected to the metabolism of organisms and external influences, for example, algae and plants take up more P than necessary and maintain a steady growth rate through this action (Wetzel, 2001).

2.3.3.4.2 Nitrogen (N)

The commonest form of nitrogen (N) is N_2 gas, as the atmosphere comprises 78% N_2 . N_2 is less soluble than O_2 in water, but the atmosphere is still a source, even though the triple bond N_2 is difficult for organisms to use, as <2% is available to organisms (Wetzel, 2001; Dodds, 2002). Nitrogen is also abundant in nature and is used in the biochemical processes of organisms (Davies & Day, 1998; Allan & Castillo, 2007).

In water, N is usually in the form of nitrate (NO_3^-), nitrite (NO_2^-) and ammonium (NH_4^+) (Davies & Day; 1998; Allan & Castillo, 2007). In the nitrogen cycle, the different forms of nitrogen are converted to each other through various processes and bacteria (Webb & Walling 1992; Dodds, 2002; Allan & Castillo, 2007), depending on which form is needed.

The two most important forms of N in natural water are NO_3^- and NH_4^+ . Ammonium is found in neutral to acidic water (Dodds, 2002), and is more readily taken up than NO_3^- (Allan & Castillo, 2007); therefore, the preferred form of N to plants (Horne & Goldman, 1994). High NO_2^- concentrations are usually associated with the presence of sewage and can be toxic (Dodds, 2002). It forms part of the dissolved inorganic nitrogen (DIN), but is normally in low concentrations.

Sources of N come from the atmosphere (N_2), precipitation, N fixation, terrestrial input (groundwater, runoff, fertilizer) and decomposition of organic matter (detritus, sewage, manure) (Allan & Castillo, 2007).

Nitrate ions move more easily through soils and are lost through runoff from the land, while NH_4^+ is retained by soils similar as in the case of PO_4^{3-} (Horne &

Goldman, 1994); thus, one of the reasons why NO_3^- concentration increases with the increase in flow (Webb & Walling, 1992).

Some bacteria (incl. photosynthetic heterotrophic bacteria) and Cyanophyta can turn N_2 into NH_4^+ (N fixation; though it is energy costly) and thus favours N-limiting conditions (Wetzel, 2001; Allan & Castillo, 2007). Nitrogen limitation of primary production is not as common as P limitation (Wetzel, 2001).

2.3.4 BIOLOGICAL FACTORS

Many different organisms live in water, and in freshwater (lentic and lotic alike) the visible organisms are fish, macroinvertebrates, algae (phytoplankton and periphyton) and zooplankton, while many others are not visible.

Most organisms can be classified according to their specific habitat (Horne & Goldman, 1994). Few of the organisms live in or on the benthic part of a river, of which periphyton are the primary producers and provide food to the macroinvertebrates. Fish harvest the macroinvertebrates (Giller & Malmqvist, 1998) as well as the periphyton (**Table 2.3**). For example, Cyprinid fish are grazers and eat epilithic and epiphytic algae (Reynolds, 1992).

Table 2.3: The fish species in the Modder River that most probably have an influence on the periphyton, either through direct or indirect feeding or disturbance while feeding (Skelton, 2001; ²*pers. comm.* M.F. Avenant, 2013).

Fish species	Trophic level	Fish species	Trophic level
<i>Labeobarbus aeneus</i> ^{*†}	Herbivore/ Carnivore	<i>Tilapia sparrmanii</i> (?) [*]	Omnivore
<i>Labeo capensis</i> [*]	Herbivore	<i>Barbus anoplus</i>	Carnivore
<i>Labeo umbratus</i> [*]	Herbivore	<i>Labeobarbus kimberleyensis</i>	Carnivore
<i>Barbus paludinosus</i> [*]	Omnivore	<i>Pseudocrenilabrus philander</i>	Carnivore
<i>Clarias gariepinus</i>	Omnivore	<i>Gambusia affinis</i> (E)	Carnivore
<i>Cyprinus carpio</i> (E) [*]	Omnivore	<i>Gambusia affinis</i> (E)	Carnivore

* = an influence on periphyton by feeding; † = different trophic level in different life stages; ? = eat algae/plants but not sure if they eat periphyton; carnivores = include feeding on macroinvertebrates; E = exotic.

2.3.4.1 Macroinvertebrates

Macroinvertebrates play an important ecological role in lotic ecosystems. They dominate the trophic level between primary production and fish in rivers and streams (Mehari *et al.*, 2014), which include organisms from various phyla: Porifera (freshwater sponges), Platyhelminthes and Annelida (worms), Mollusca (snails) and Arthropoda (crustaceans and insects) (Dodds, 2002; Tundisi & Tundisi, 2012). Orders and families within these phyla are included in the South African Scoring System (SASS) for macroinvertebrates (an aquatic biomonitoring tool) (Dickens & Graham, 2002).

These organisms, which live on the epibenthos, all have modified body structures to keep them from being washed away by the current (Horne & Goldman, 1994). Some of the adaptations include living under stones, having hooks, grapples, claws or “lifelines”, streamline bodies or sticky secretions (Davies & Day, 1998).

² Fish Expert, Centre for Environmental Management, UFS

Macroinvertebrates fall into six functional feeding groups (FFGs), namely shredders, collectors, filter feeders (phytoplankton), grazers/scrapers (periphyton), predators and macrophyte piercers (Horne & Goldman, 1994; Wetzel, 2001; Dodds, 2002; Allan & Castillo, 2007). The habitat of an organism is determined by the FFG in which it falls, as well as where in a river system (RCC) it will occur (Horne & Goldman, 1994).

The grazers/scrapers mostly feed on periphyton by scraping the substrate surface, and therefore are mostly only present in headwater and middle reaches where the water is not too turbid for the growth of periphyton (Cummings, 1992; Davies & Day, 1998). Of all these species, insects dominate this group (Horne & Goldman, 1994). The organisms involved include aquatic oligochaetes (that eat algae), snails (some of the major consumers) and crabs (Dodds, 2002). The insects also include various orders and families that have grazer/scrapers present (Giller & Malmqvist, 1998; Wetzel, 2001; Dodds, 2002; Allan & Castillo, 2007; Tundisi & Tundisi, 2012), namely:

- ◆ Ephemeroptera (mayflies) = *Heptageniidae, *Baetidae, Ephemenellidae, *Caenidae, *Leptophlebiidae.
- ◆ Trichoptera (caddisflies) = *#Glossosomatidae, Helocopsychidae, Mollannidae, Odontoceridae, Goreridae, *Leptoceridae.
- ◆ Lepidoptera (aquatic caterpillars).
- ◆ Diptera (flies & midges) = *Chironomidae, *Tabanidae.
- ◆ Coleoptera (water beetles) = *Elmidae, *Psephenidae.
- ◆ Hemiptera (bugs) = *Corixidae.

(* = in SASS as indicators; # = only in the South-Western Cape; Dickens & Graham, 2002)

Grazing by invertebrates not only has an effect on the periphyton numbers, but also on the community structure, as they can be selective to what they graze on (Lampert & Sommer, 2007).

2.3.4.2 Periphyton (periphytic algae)

Periphyton (benthic algae or phytobenthos) are primary producers that exploit streams successfully as habitat (Biggs, 1996; MDEQ, 1999; Matthaei *et al.*, 2003; Tundisi & Tundisi, 2012). Graba *et al.* (2013) have found epilithical periphyton to be a major source of primary production. They can be divided into two groups: macroalgae (the long filamentous strands such as *Cladophora* and *Spirogyra*) and microalgae (mostly single cells coating the substrate such as Bacillariophyta and other soft algae) (Arnwine & Sparks, 2003; Lebkuecher *et al.*, 2014).

Periphyton is also an important component in lotic systems, as it not only influences the carbon and nutrient cycles, but also the composition of the invertebrates (Boston & Hill, 1991). It is therefore the foundation of many food webs (Stevenson & Bahls, 1999; Allan & Castillo, 2007; Graba *et al.*, 2013; Hart *et al.*, 2013) of lotic systems, because grazing a two-dimensional periphyton mat is mechanically more efficient than filtering phytoplankton from a three-dimensional environment (Ghosh & Gaur, 1998; Asaeda & Hong Son, 2000).

Further discussion on periphyton is in **Section 2.6**.

2.4 WATER POLLUTION & EUTROPHICATION

2.4.1 POLLUTION

Pollution is the most obvious problem associated with inland waters. Pollution is the “*degradation of natural systems by the addition of harmful substances*”, “*harmful material that makes an environment less fit for the organisms that occupy it*” (Davies & Day, 1998). Pollution can be from natural events/causes or it can be a result of human activity (Giller & Malmqvist, 1998).

Harmless substances can be harmful in large quantities (e.g. sodium chloride – NaCl), and at the same time harmful substances (e.g. aluminium – Al) are not necessarily harmful when occurring in small amounts (Davies & Day, 1998).

The deterioration of aquatic ecosystems is mostly the result of pollution that decreases the water quality (Allan & Castillo, 2007), but can also result from human pressure causing extensive regulation (Gallardo *et al.*, 2011). In natural water, any change in quality implies pollution (Giller & Malmqvist, 1998). There are three steps of pollution impact: the primary being when the effect is immediate, e.g. pollutants from point source. Secondary pollution impact alters the food chain, and as a tertiary step, the long-term effects can change the chemical composition or even modification in the species composition (Tundisi & Tundisi, 2012).

Humans affect almost all running water profoundly and thereby alter the scope and influence of species interaction in the food web (Hildrew, 1992). Some of these water pollution sources include acid rain (from atmospheric pollution), runoff and storm water runoff, leaks from fuel storage tanks, seepage from chemical dumps, treated and untreated sewage discharge, thermal pollution (from cooling water), fertilizers, herbicides and insecticides (Horne & Goldman, 1994; Davies & Day, 1998; Dallas & Day, 2004; Allan & Castillo, 2007; Tundisi & Tundisi, 2012; Mehari *et al.*, 2014). Depending on the source of the pollution, it can either be point or non-point source pollution (Giller & Malmqvist, 1998).

2.4.2 EUTROPHICATION

Eutrophication is “*the process of an ecosystem becoming more productive by nutrient enrichment stimulating primary producers*” (Dodds, 2002) or “*the alteration of the production*” (Wetzel, 2001), and can be natural or cultural (Horne & Goldman, 1994; Davies & Day, 1998). It is caused by excessive inputs of pollutants (esp. nitrogen & phosphorus) (Tundisi & Tundisi, 2012).

Natural eutrophication takes place over a long period (over thousands of years), or with or after catchment disturbances like volcanic disruptions (Horne & Goldman, 1994; Dodds, 2002). It can occur naturally in lakes and rivers as they mature (Davies & Day, 1998). Cultural eutrophication is more common, especially in countries with higher densities of human activities (Dodds, 2002).

Municipal sewage is the oldest contributor to cultural eutrophication, as domestic wastewater is very rich in nutrients (detergents are high in PO₄). Other sources are urban runoff, agricultural practices, deforestation, road building, etc. (Horne & Goldman, 1994; Giller & Malmqvist, 1998; Dodds, 2002; Walsh & Wepener, 2009). Carbon, oxygen, hydrogen, sulphur, potassium, nitrogen and phosphorus are among the required nutrients for normal growth and reproduction of plants. N and P are the most important nutrients when it comes to the enrichment (eutrophication) of water.

In streams the relationship between the water column nutrients and benthic algal biomass are not the same as those of nutrients and plankton in lakes (Dodds, 2002). In rivers it leads to higher productivity than usual (Horne & Goldman, 1994), while in lakes, with the higher retention time, the nutrients accumulate after the productivity capacity has been reached (Tundisi & Tundisi, 2012).

The trophic classification of streams is based on suspended and attached algae, and nutrient concentrations (Dodds, 2002). When periphyton is unconstrained by light limitation, it responds to the increase in nutrients by coating rocks and other surfaces with biofilms of mostly filamentous and single cell Bacillariophyta in spring, as well as some Chlorophyta in summer. The development and spreading of nuisance algae and blooms are caused by excessive nutrients in water systems (Graham *et al.*, 2009; Schneider *et al.*, 2013).

If a river already has sufficient P and N to support plant growth, eutrophication of the river is unlikely to increase the growth of macrophytes (Fox, 1992); it may increase the macrophytes' growth in oligotrophic rivers.

Specific physical and chemical factors affect aquatic organisms differently (Dodds, 2002), as eutrophication can change the diet of species as well as the length of the food chain (Giller & Malmqvist, 1998; Lampert & Sommer, 2007) through change in the ecological community structure and biodiversity loss (Rossouw *et al.*, 2008).

Toxic Cyanophyta blooms may be the result of huge amounts of excess nutrients, especially through cultural eutrophication (Dodds, 2002; Rossouw *et al.*, 2008; Fetscher *et al.*, 2014). These toxins can enter food chains and cause harm to the health of animals and humans (Lin *et al.*, 2008; Fetscher *et al.*, 2014). Bacterial decay of organic matter uses a lot of oxygen and can deplete the water drastically (Webb & Walling, 1992; Allan & Castillo, 2007), especially in eutrophic waters, where the excess plant growth leads to an extremely high decay of organic material.

The control of eutrophication can be costly (Dodds, 2002); the removal of nitrogen (NO_3 , $\text{NO}_2 + \text{NH}_4$) is more expensive than phosphorus removal, as nitrogen is more soluble.

2.5 BIOMONITORING

As the impact of human activities accumulates in the environment, the reliance on physical and chemical indicators has started to shift towards using measures of ecological condition and general water quality. Various biomonitoring indices were developed using biological indicators, of which algae were part, but the physical and chemical methods were kept to contribute to a more meaningful assessment (McCormick & Cains, 1994; Yagow *et al.*, 2003; Bere & Tundisi, 2010; Marzin, 2012). Periphyton, fish and macroinvertebrates are mainly used in the assessment and monitoring of water quality and biological integrity of streams (Griffith *et al.*, 2005). It was also found that a combination of the three major freshwater biological communities (e.g. macroinvertebrates, fish and algae) is used in biological assessment (Yagow *et al.*, 2006).

2.5.1 ORIGIN (SOUTH AFRICAN)

In the past, many methods were available for the bioassessment of the integrity of aquatic systems (De la Rey *et al.*, 2004). However, it was not until 1996 that the National Biomonitoring Programme for Aquatic Ecosystems (NBPAE) was initiated by the then Department of Water Affairs and Forestry (DWAF), the Water Research Commission (WRC) and the former Department of Environmental Affairs and Tourism (DEAT). The objective was to design a monitoring programme to monitor the health of aquatic ecosystems throughout the country and to provide information that can be used by water resource managers to manage water systems (Bate *et al.*, 2002; De la Rey *et al.*, 2008).

Chemical water quality tests were first used as monitoring methods in South Africa with biological indices added later (Taylor *et al.*, 2007b). Back then, arrays of biological indices were tested for practical use and interpretation. These indices included the South African Scoring System (SASS, based on macroinvertebrates), the Index of Biotic Integrity (IBI, based on fish), the Riparian Vegetation Index (RVI), and a suite of secondary indices, the Hydrological Index, the Water Quality Index (WQI) and the Geomorphological indices (Bate *et al.*, 2002).

2.5.2 DIFFERENT TOOLS AND THEIR CONCEPTS

Since the establishment of the NBPAE, the indices were redeveloped, extended and improved to include the current tools that are used in the River Health Programme (RHP) and EcoStatus Classification:

- ◆ Macroinvertebrate Response Assessment Index (MIRAI), which includes SASS5 for macroinvertebrates (a site-specific indicator) – determines the ecological category of the invertebrates by integrating the ecological requirements of the taxa in a community with their response to modified habitat conditions (Thirion, 2007).

- ◆ Fish Response Assessment Index (FRAI) – an assessment index that is based on environmental intolerances and the preferences of the reference fish assemblage and the species responses on environmental determinants or drivers (Kleynhans, 2007).
- ◆ Riparian Vegetation Index (VEGRAI) – an index describing the riparian vegetation status in both the reference and present status; it compares the differences between the states and then measures the vegetation response to the impact regimes (Kleynhans *et al.*, 2007).
- ◆ Index of Habitat Integrity (IHI) – refers to the maintenance of a balanced physical-chemical and habitat quality, which are comparable to that of the characteristics of regional habitats in their natural state (Kleynhans *et al.*, 2009).
- ◆ Hydrological Driver Assessment Index (HAI) – refers to the change in the hydrological regime from the reference condition to the present, and the ecological response to such change in terms of volume modification and flow variation (Kleynhans *et al.*, 2005).
- ◆ Physico-Chemical Driver Assessment Index (PAI), which includes phytoplankton (a non-site-specific indicator) – a tool used to determine the present state of the water quality (physical and chemical factors) for specific sites of resource units (Kleynhans *et al.*, 2005).
- ◆ Geomorphological Assessment Index (GAI) – evaluation and interpretation of observed changes in the condition of the channel at a study site caused by anthropogenic impacts, as the impacts may cause a shift from the reference condition (Rowntree, 2013).

2.5.3 SITE-SPECIFIC VS. NON-SITE-SPECIFIC INDICES

Some of the indices can be described as site-specific indices (condition of specific site), and others as site non-specific indices (condition upstream of a site).

Site-specific indices include SASS5 (macroinvertebrates) as a function of the habitat and the functionality of the habitat at sites sampled (De la Rey *et al.*,

2008), and periphyton (incl. Bacillariophyta) as a direct function of the physical and chemical composition of water (Taylor *et al.*, 2007a). An example of a non-site-specific index is FRAI (fish), which is an indication of the health and integrity of a river reach (catchment) and the habitat availability (Kleynhans, 2007).

In many instances, these are all used in conjunction with one other to get a better idea of river health. With the River Ecoclassification and Ecostatus Determination, various models are used in the Multi Criteria Decision Making Approach (MCDA) (Kleynhans & Louw, 2007) and were used for an ORASECOM project (ORASECOM, 2013b).

2.6 PERIPHYTON/DIATOMS

As mentioned in **Section 2.3.4.2**, periphyton is primary producers and the foundation of many food webs (Stancheva *et al.*, 2012). Lampert & Sommer (2007) describe it as “*the community of microalgae growing on submerged surfaces*”, while Azim *et al.* (2005) state that periphyton is a classical limnological term referring to the “*microfloral community living attached to the surface of submerged objects in water*”.

Periphyton grows on virtually any substrate that receives light and can sustain an algal community (Allan & Castillo, 2007; Tundisi & Tundisi, 2012). Depending on how substrates are grouped, periphyton can be divided into four to six categories according to the substrate or microhabitat they occupy (Porter *et al.*, 1993; Wetzel, 2001; Allan & Castillo, 2007; Lampert & Sommer, 2007; Tundisi & Tundisi, 2012):

- ◆ epilithic = attached to rocks, bedrock or other hard surfaces;
- ◆ epidendric/epixylic = attached to submerged tree limbs and roots, or on other wood surfaces;
- ◆ epiphytic = attached to submerged aquatic plants or macroalgae/filamentous periphyton;
- ◆ epipellic = associated with fine streambed sediments;
- ◆ episamic = associated with coarse streambed sediments; and

- ◆ epizooic = attached to aquatic animals.

Periphyton isolates and consumes inorganic nutrients and labile organics, in this manner helping purify stream and river water (Biggs, 1996). These organisms also stabilise substrata and serve as habitat for many other organisms (Stevenson & Bahls, 1999). Periphyton also has an influence on sediment composition (Schneider *et al.*, 2012). The accumulation of algae on a solid surface is a function of reproduction, death, grazing, immigration and emigration (Ghosh & Gaur, 1998). Periphytic algae can form colonies or filaments that are visible to the naked eye, or they can remain single-celled, microscopic plants that are only visible to the naked eye in their accumulated growth (MDEQ, 1999).

However, in stable-flowing, enriched streams they can proliferate, causing water management problems. The filamentous Chlorophyta, *Ulothrix* (in cold waters) and *Cladophora*, often present the greatest nuisance. In less enriched streams, stalked and tube-dwelling Bacillariophyta can also be a problem, especially species of Gomphonemaceae and Cymbellaceae (Biggs, 1996).

The combination of different algal groups forms different macroscopic communities. Some Chlorophyta develops streaming beds of filaments that can be up to several metres long in low-density areas, and often detach and float near the surface. These floating mats can then become entangled in branches and vegetation, or around cobbles protruding from the bottom of the stream, where they often continue to thrive. By contrast, Bacillariophyta tend to form thick gelatinous mats that consist mainly of polysaccharides. The mats may reach a thickness of at least a centimetre, completely smothering the substrata. In some streams, they can look like whitish-grey sewage fungus (Biggs, 1996). The epilithic periphytic algal community comprise a large proportion of Bacillariophyta (Walsh & Wepener, 2009).

As periphyton assemblages are attached to substrata, their characteristics are affected by physical, chemical and biological disturbances that occur in the stream reach during the time in which the assemblage developed (Stevenson & Bahls, 1999; Biggs *et al.*, 1999).

The rapid and straightforward ability of the physical, chemical and bacteriological variables to detect change in the environment is the reason they form the basis of water quality monitoring. However, they provide little to no information of the changes in or on the biological communities and are usually costly when a full range of analysis are done. Thus, biological indicators, such as macroinvertebrates and Bacillariophyta (including in periphyton), have been used to provide a means to measure the effects of physical-chemical influences on the natural environment (Vis *et al.*, 1998).

The characteristics of periphyton and their response to changes in water quality are not always the same. Various environmental factors such as substratum, temperature, light and current regimes, nutrient levels and grazing influence the periphyton community. Temperature, light and nutrients drive the potential for primary production, while the development of biomass is influenced by grazing and velocity. This complex interplay of factors shapes the periphyton community in its accrual and losses, and thereby the ability to detect water-quality changes from periphyton (Biggs, 1988; Kelly & Whitton, 1995; Biggs, 1996; Ghosh & Gaur, 1998, Vis *et al.*, 1998).

However, seasonal and successional changes in species composition complicate the interpretation of community response to pollution. Therefore, combinations of environmental variables are more successful in establishing reliable relationships with biomass and diversity (Vis *et al.*, 1998). Bacillariophyta presence is more likely in winter and Cyanophyta in summer (Horne & Goldman, 1994).

The most common algal division, which includes periphyton, is the Bacillariophyta, Cyanophyta and Chlorophyta. Other divisions that also include some periphyton are Euglenophyta, Chrysophyta and Rhodophyta (Horne & Goldman, 1994; Yagow *et al.*, 2006; Allan & Castillo, 2007). These algal groups have one common denominator; they all contain the chlorophyll-a pigment (Gregor & Maršálek, 2004; Kilroy *et al.*, 2013).

The resident nature of periphyton makes its existence sensitive to changes in the quality of water (Arnwine & Sparks, 2003). It is said that the main factors contributing to the accrual or loss of periphyton are nutrient levels and light availability. Temperature ranges, flow regimes, habitat availability and grazing activities also play a major role in the distribution and composition of the periphytic community at any moment (Biggs, 1988; Biggs, 1996; Ghosh & Gaur, 1998).

2.6.1 PHYSICAL INFLUENCES ON PERIPHYTON

Hydro-physical factors are important in the benthic community functionality. Physical disturbances have a significant effect on the function and structure of periphyton communities; it can control growth and loss (Nikora *et al.*, 1998; Luttenton & Baisden, 2006).

2.6.1.1 Flow and velocity

Flow in various magnitudes influences the function and structure of the lotic ecosystem (Uehlinger *et al.*, 2003; Hart *et al.*, 2013) and the integrity of its biotic communities (Navarro-Llácer *et al.*, 2010). It contributes to periphyton loss as a function of disturbance, whether through scouring during a flood or high flows, sediment abrasion, or through movement of substrates (Nikora *et al.*, 1998; Graba *et al.*, 2013). Flood disturbance not only causes spatial variability, but also temporal variability in benthic communities, including the periphyton (Mosisch & Bunn, 1997; Biggs *et al.*, 1999). Flows can change biomass, taxonomic composition and distribution on habitats. Floods also have an indirect effect on periphyton as they disturb the macroinvertebrates that graze on them (Biggs *et al.*, 1998a; Jakob *et al.*, 2003; Larson & Passy, 2012; Graba *et al.*, 2013).

Epilithic periphyton communities attached to bedrock or boulders are sometimes dislodged during high flows. The periphyton that is not dislodged during such an

event is then falls victim to be scoured by sand and gravel that flow past at high velocity (Biggs & Thomsen, 1995; Mosisch & Bunn, 1997). In this manner, entire algal communities can be destroyed during a flood event. Taxonomic change is therefore a function of current velocity (Passy, 2001; Hart *et al.*, 2013).

Depending on the magnitude, frequency and duration of a flood event, the long-term effect may differ between algal communities. Not only do they destroy the communities; they can also delay or halt the recovery and succession of such communities (Mosisch & Bunn, 1997; Passy, 2001). Total recovery from a pre-flood state can take from a few months to a year, depending on the system. The recovery depends on the viability of light and nutrients (Uehlinger *et al.*, 2003). This is especially true for river stretches downstream from dams in regulated rivers with frequent releases to fulfil irrigation requirements, domestic demand, etc. (Jakob *et al.*, 2003). If it is a sandy river system, a lot of scouring from sediment takes place and leads to the loss of periphyton.

During low flood frequencies, the periphytic community taxa are denser and more complex, while during high flood frequencies the periphytic communities comprise more low-growing taxa (Biggs *et al.*, 1998a). There are also less filamentous algae in systems with high flood frequencies than those with low flood frequencies.

Studies (Ghosh & Gaur, 1998) show that as pools change to runs and runs to riffles, not only the algal density changes, but there is also a change in the algal composition. The depth of the water column is also important (Biggs, 1996; Matthaei *et al.*, 2003)

To stimulate and increase primary productivity in lotic systems, the current over periphytic bed surfaces needs to be a minimum of 5 cm/s (0.05 m/s) (Gore, 1996).

Biggs *et al.* (1998b) have found that as velocity increases, there is a gradient to the loss of periphyton; firstly, it is the long filamentous algae with velocities faster

than 0.3 m/s. At this stage, the biomass loss is greater than the density loss from the substrate.

The second algal group that is lost as the velocity increases is erect, large-celled taxa, leaving only the smaller, tightly adhered taxa – and the most resilient to survive – as not only density, but also diversity is lost. Stalked/short filamentous Bacillariophyta prefer a water velocity of 0.4 to 0.6 m/s (intermediate velocities). With higher stream velocity, one will find lower Bacillariophyta density (Asaeda & Hong Son, 2000).

Increase in flow and velocity not only decreases the periphyton biomass by removal, but also plays an indirect role in the increased maintenance of the biomass by increasing the nutrient transfer to the algal mats, stimulating growth (Biggs *et al.*, 1998b; Larson & Passy, 2012).

2.6.1.2 Turbidity and light

Light as a principal factor determines whether biofilms will be autotrophic (algae) or heterotrophic (fungi or bacteria). Biofilms that are deprived of light either fall below the photic zone through the water, turbidity or sediment coverage (Burns & Ryder, 2001). Weather is also an important factor. Several days of rain or cloud cover will also reduce algal growth (Arnwine & Sparks, 2003). As all periphyton contains chlorophyll-*a*, it needs light to photosynthesise (Gregor & Maršálek, 2004; Matheson *et al.*, 2012)

If light deprivation is caused by flooding through increased turbidity (Wiley & Seelbach, 1997), it is temporary, except when stones and rocks are overturned with the periphyton facing downwards (Burns & Ryder, 2001).

If shallow photic zones have variable water levels, they are usually dominated by heterotrophs (e.g. macroinvertebrates, fungi), while with flow regulations when water levels stabilises/levels out, the photic zone is usually dominated by autotrophic biofilms (e.g. periphyton) (Burns & Ryder, 2001).

When a stream is heavily shaded (headwater streams and mid-sized rivers), either by riparian cover or topography, the availability of light can be the key factor of periphyton growth, as 95% of the sunlight can be blocked off if it is a full canopy cover (Mosisch *et al.*, 2001; Roberts *et al.*, 2004; Hillebrand, 2005). As the canopy vegetation is removed by an activity (logging, wild fires, etc.) the productivity in the newly unshaded stream will increase, provided the nutrient concentrations are abundant (Mosisch *et al.*, 2001).

Increased light or solar radiation not only increases periphyton biomass, but also changes the composition of the periphyton assemblage. Bacillariophyta require less light than filamentous Chlorophyta as a group; thus, in shaded parts of a river in the same reach, Bacillariophyta will be more abundant or dominant than in unshaded parts (Mosisch *et al.*, 2001; Hillebrand, 2005). It can be argued that one would find less filamentous Chlorophyta in more turbid water than in clearer water. Bourassa and Cattaneo (2000) have found that the proportion of Cyanophyta is higher in un-enriched streams that are shaded, while Bacillariophyta and filamentous Chlorophyta are more abundant in unshaded streams.

Light cannot only be limited by riparian cover or turbidity; it can also be caused by self-shading. When nutrients are abundant (eutrophication), high density on the periphyton mat can cause the longer/taller algae to overshadow the smaller/shorter species (Nijboer & Verdonschot, 2004; Hillebrand, 2005). This may lead to a decrease in photosynthesis rates and have a negative impact on biomass (Guasch *et al.*, 2003)

Light seems to be the major limiting factor, especially when nutrients are abundant, or at least secondarily limiting (Mosisch *et al.*, 1999), as periphyton cannot survive without photosynthesis. This is important to the Modder River as most of the river channel is classified as alluvial (Seaman *et al.*, 2001).

2.6.1.3 Temperature

Temperature interacts with periphyton (and other aquatic organisms) to influence the metabolism and growth rates (Biggs, 1996; Rosa *et al.*, 2013) and thereby ultimately also the community composition (Biggs, 1988). Temperature is also one of the driving forces behind the seasonal change, or an indicator of thermal alterations in the aquatic ecosystem (Kishi *et al.*, 2005).

Studies done by Kishi *et al.* (2005) show that the change in temperature not only changed the growth rate of periphyton, but also the rate it was consumed by herbivores. At low temperatures (3–6 °C), the periphyton growth rate was low, as was the grazing rate. Low temperatures reduce the metabolic rates of grazers, thus less feeding on periphyton (Giller & Malmqvist, 1998). At 12 °C, there was an abundance of periphyton as fish predation reduced the grazers that feed on periphyton. When predation of the grazers is stopped, the periphyton abundances decrease under the increase of the grazing effect (Kishi *et al.*, 2005).

Temperature not only influences the growth rate of periphyton, but also the composition of the periphyton community found. Various studies (Biggs, 1996; Vis *et al.*, 1998; Flynn *et al.*, 2002) have shown increases in periphyton growth during spring and summer months, with Bacillariophyta dominating the community during spring and filamentous algae (Chlorophyta and/or Cyanophyta) during the late summer and sometimes even during fall.

2.6.1.4 Habitat

The available habitat determines the type of periphyton (see **Section 2.6**) and the community composition. The stability of the habitat/substrate also plays a role (Biggs, 1996), as well as the habitat size (Arnwine & Sparks, 2003). Shifting sandy river bottoms are not suitable. Stable colonising substrate and gravel need to be bigger than 2 cm for periphyton to grow on.

Substrata are unlikely to move when flow increases, act as refugia for periphyton during the high-flow events (Matthaei *et al.*, 2003).

2.6.2 CHEMICAL INFLUENCES ON PERIPHYTON

Various studies have shown that water chemistry, including salinity and pH, has an effect on the species composition of periphytic algae (Potapova & Charles, 2002).

2.6.2.1 pH

The optimal water pH for most aquatic organisms lies between 6.0 and 8.5, with a few specialised organisms, low in diversity, living outside these ranges. In eutrophic systems, high pH is associated with high salt levels, high nutrient levels, or night anoxia, whereas low pH values lead to an abundance of heavy metals and high sulphur levels (DWAF, 1996a; Sabater & Admiraal, 2005).

As pH can influence the nutrient levels in a system, it can change the algal composition from P-limited algae (Bacillariophyta, Chlorophyta) to N-limited algae (cyanobacteria) when P is added to a system with a high N:P ratio (Sabater & Admiraal, 2005). Schneider *et al.* (2013) have found that in periphyton, Bacillariophyta reach maximum richness around 6.9, while the other algae reach maximum richness at about 6.4.

2.6.2.2 Total dissolved salts (TDS)/electrical conductivity (EC)

It is not only nutrients that often occur in high concentrations in polluted rivers, but other ions that can also have an effect on the total salinity levels (Potapova & Charles, 2002). The increase of salinity levels of an aquatic ecosystem may lead to a change in periphyton species composition if it is severe enough (Potapova & Charles, 2003).

The EC range of water associated with pollution from urban areas is between 300 and 1 500 $\mu\text{S}/\text{cm}$ (Newall & Walsh, 2005). An EC above 1 000 $\mu\text{S}/\text{cm}$ is seen as the boundary between freshwater and brackish water (Potapova & Charles, 2003).

2.6.2.3 Nutrients and eutrophication

Nitrogen (N) and phosphorus (P) play a major role in eutrophication (Dodds *et al.*, 1997; Zimmo *et al.*, 2004), whether it is natural or cultural. In waters flowing through urban and agricultural areas, pollution can come from point and non-point sources (Nijboer & Verdonschot, 2004).

Excess nitrogen (N) and phosphorus (P) from eutrophication sources lead to algal abundance, and cause ecostatus and maintenance problems of ecosystem integrity to aquatic systems (Bowman *et al.*, 2005). As eutrophication increases, especially in nutrient-poor rivers, so does the periphyton. It can be said that nutrients have a positive effect on the accrual rates of the algal cells (Greenwood & Rosemond, 2005).

The relationship between nutrient supply and periphyton in rivers and streams is more complex than that of phytoplankton and nutrients in lakes (Bernhardt & Likens, 2004). Different from lakes and phytoplankton, flow and light (canopy shading) plays an important role in the growth and primary production of periphyton in rivers.

In the absence of overwhelming dominance by physical and biological factors, it is found that in rivers nitrogen and phosphorus are limiting factors to periphyton growth and abundance (Mosisch *et al.*, 2001; Bowman *et al.*, 2005; Carr *et al.*, 2005). Other studies have found that both the concentration and form of N and P available in the water have various effects on the periphyton changes (Dodds *et al.*, 2002).

The limiting nutrient is mostly P, but sometimes it can be N (Mosisch *et al.*, 2001; Dodds *et al.*, 2002). The preferred N:P ratio for each algal species differs (Bowman *et al.*, 2005). P limiting occurs when the N:P ratio is >10 and N limiting when it is <10 (Rosemond *et al.*, 1993). Luttenton and Lowe (2006) have found that algal biovolume decreases at a low N:P ratio (1:5 and 1:7.5).

Different periphyton species have different tolerance levels for nutrients. As nutrient concentrations and ratios change, so does the periphyton community (Greenwood & Rosemond, 2005). Organic enrichment changes periphyton composition, and with enough light it favours filamentous species. In the presence of high P and low N, certain cyanobacteria can convert atmospheric N₂ to ammonia and amino acids (Burns & Ryder, 2001). High nutrient concentrations are associated with high Bacillariophyta species diversity. As P becomes limiting, it leads to a loss in diversity (Snyder *et al.*, 2002).

The increase in nutrients also enhances new habitat development. *Cladophora* grows very fast at high nutrient concentrations, and thus becomes a potential habitat for Bacillariophyta (epiphytes) and macroinvertebrates (Nijboer & Verdonschot, 2004).

The effect of nutrients is not only on the periphyton species diversity and biomass, but also on the chlorophyll-*a* concentration (Tank & Dodds, 2003). Greenwood and Rosemond (2005) have found that the periphyton chl-*a* (pchl-*a*) concentration increases with nutrient enrichment just as the periphyton biomass does, but not always at the same rate. They have also found a seasonal change in pchl-*a* concentration, where spring concentrations are higher than in summer and autumn, but it appears to be linked to pchl-*a* concentration produced per cellular unit, rather than a change in biovolume or cell density.

Nuisance periphyton algal biomass is seen as more than 100 mg/m² chl-*a* (10 µg/cm² chl-*a*) by some authors (Dodds *et al.*, 2002; Ponader *et al.*, 2007) and by other as >150 mg/m² chl-*a* (15 µg/cm² chl-*a*) (Dodds *et al.*, 1998).

2.6.2.3.1 Phosphorus

The periphyton mat is in direct contact with the water and can rapidly extract P from it. They can extract P more rapidly than that lost to soils and macrophytes; therefore, they respond first to the increase in P loading (McCormick & Stevenson, 1998). In using P for growth, periphyton recycles the P and influences its retention time in aquatic systems (Dodds, 2003).

Phosphorus is the principal nutrient in limiting primary production of periphyton (e.g. growth and biomass), especially during spring (Hillebrand & Sommer, 2000). The change in P concentration can thus also change the species composition and diversity (McCormick *et al.*, 2001). In unshaded streams, the major proportion of P is present in the epilithic periphyton (Nijboer & Verdonschot, 2004).

2.6.2.3.2 Nitrogen

The nitrate (NO_3^-) concentration in natural rivers has an average of 0.1 mg/l NO_3^- (Nijboer & Verdonschot, 2004). The pollution source of NO_3^- in ground and surface water is mostly from wastewater outfall and agricultural runoff (Zimmo *et al.*, 2004). The average NO_3^- concentrations in rivers influenced by agriculture can be around 10 mg/l (Nijboer & Verdonschot, 2004).

Some studies have found that during summer, for total and Chlorophyta biovolumes (Luttenton & Lowe, 2006), N can be a limiting factor in enriched water (Hillebrand & Sommer, 2000).

Ammonium (NH_4^+) can be toxic for fish at concentrations higher than 0.5 mg/l (Zimmo *et al.*, 2004), while less herbivorous fish may also further lead to more algae.

2.6.2.3.3 Other

Silicon (Si) can be limiting for Bacillariophyta at times when water is highly enriched with N and P (Hillebrand & Sommer, 2000).

A relatively low concentration of pollutant metals from sources such as wastewater effluent can have an adverse effect on aquatic ecosystems (Ridge & Sedlak, 2004), including periphyton.

If nutrients could just keep flowing downstream, nutrients would have no effect on the river when there is no uptake by any plants (Nijboer & Verdonschot, 2004).

2.6.3 BIOLOGICAL INFLUENCES ON PERIPHYTON

2.6.3.1 **Grazing**

The secondary benthic trophic level is filled with grazers and scrapers (e.g. gastropods, insect larvae and herbivorous fish) as they ingest periphyton, the primary trophic level (Matthaei *et al.*, 2003; Liess & Hillebrand, 2004). Aquatic insects may be the dominant group, but herbivorous fish and crayfish can also have a major impact on the epilithic periphyton in temperate water (Barbee, 2005). Herbivore fish includes species such as *Labeo capensis* and *Tilapia sparmanii* (Skelton, 2001).

Biggs (1996) has found that the lower grazing activities by macroinvertebrates during spring can account for part of the increase in periphyton biomass. Grazing not only has an effect on the biomass of the periphyton, but also on the structure and composition of the periphyton community (Barbee, 2005). This includes the main algal groups and not just the species composition (Burns & Ryder, 2001). Smaller invertebrates graze mostly on Bacillariophyta, while the

larger ones (mostly large insect larvae) graze on filamentous Chlorophyta (Anderson *et al.*, 1999; Hillebrand, 2005).

Thus, while grazers may control nuisance periphyton in enriched streams (Anderson *et al.*, 1999), some Cyanophyta and *Cladophora* (Chlorophyta) may contain chemical toxins/excretions to prevent grazing (Dudley *et al.*, 1986).

As the number of periphytic herbivores increases because the periphyton increases, it is always best to do algal surveys together with those of macroinvertebrates (Arnwine & Sparks, 2003).

2.6.4 RIVER CONTINUUM CONCEPT AND RIVER REGULATING

Proposed in the River Continuum Concept (RCC) is a downstream pattern of increasing periphyton biomass from headwater to mid-catchment reaches as streams unite, channels widen and riparian shading is reduced (Biggs, 1996). In the lower reaches of a river, the biomass decreases again as a function of light reduction in deeper waters and increased turbidity.

With the natural nutrient gradient of the trophic status that runs along the river continuum, the oligotrophic conditions are mostly in springs and in upstream parts of a river, while eutrophic areas occur in large rivers and their downstream parts, depending on the catchment features (e.g. land use) (Rott *et al.*, 2003).

However, if such downstream trends can be interrupted by localised features, the RCC pattern can be reversed in non-forested catchments. In catchments with localised nutrient inputs and/or frequent disturbances, periphyton communities may display quite different patterns (Biggs, 1996).

Dams and weirs form barriers in river systems, resulting in the change in the natural flow regime, turbidity, temperature and chemistry of the river downstream from that point. These changes, in turn, affect the seasonal and annual patterns of the river biota. Their longitudinal position is especially important in a river, as

this affects how much of the natural processes will be impacted (Palmer & O'Keeffe, 1990).

The Modder River falls mostly in the Lowland geomorphological zone with an alluvial channel type; however, some other zones and channel types do exist in the river system. One of the first bigger weirs was built at Sannaspos in 1896 for domestic water use of Bloemfontein (through pipeline). In 1904, the Mazelspoort Weir was built and later a purification plant (domestic use). Mocke's Dam was built in 1913, upstream from Mazelspoort Weir, as a water-holding reservoir (domestic and agricultural use). In 1955, the Rustfontein Dam (domestic use and water holding reservoir) was built and in 1970, the Krugersdrift Dam (agricultural use), to mention a few of the structures helping with river regulation. Two water transfer schemes also link into the Modder River system; the Modder-Novo water scheme (mostly for domestic water use) in the upper reaches of the river and the Orange-Riet River water scheme (mostly for agricultural use) in the lower reaches of the river (Seaman *et al.*, 2001).

CHAPTER 3

METHODOLOGY

3.1 SITE SELECTION CRITERIA

Sampling sites were selected from biomonitoring sites already in use by the Centre for Environmental Management (CEM) at the University of the Free State and on the basis that South African Scoring System (SASS – macroinvertebrate sampling) could be done with “stones in current” or “bedrock in current”, as stones are the preferred habitat to sample periphyton. The SASS5 method is also a site-specific method and can be useful in comparing results.

Other criteria for the sites were that they should be accessible and cover a range of water quality conditions.

A number of initial sites were selected, but some were discarded, as too few results were obtained. This was because the sites were either mostly dry during the first sampling period and/or flooded during the second sampling period for extended periods. River regulation played a role in this aspect.

The following sites, Sannaspos, Modder above Confluence and Bishop’s Weir, were the final selected sites, as ample sampling was done for both periods. They cover low to medium and highly polluted water, and are comparative in structure, upstream weirs and bridges. In regulated rivers, these structures are inevitable, as large portions of the river have either a sandy bottom or consist of a series of pools, natural or as a result from damming of river by series of small weirs. Bridges and weirs were usually built (in the past) on sites that would have been suitable for sampling macroinvertebrates or periphyton.

3.2 STUDY SITES

The Modder River has a relative small catchment area of approximately 7 960 km, in the central part of the Free State Province, and with a mean annual runoff of about $184 \times 10^6 \text{ m}^3$ (Koning & Roos, 1999). Only the specific sites and surroundings (**Fig. 3.1**) are shown in the maps at each site description. The entire 1:50 000 Topographical Map Sheets are shown in **Appendix A**, as well as where sites are located in relation to the map. Coordinates are given as Lat/Lon hddd°mm'ss.s" (WGS84). **Table 3.1** shows various descriptions of the areas surrounding the study sites.

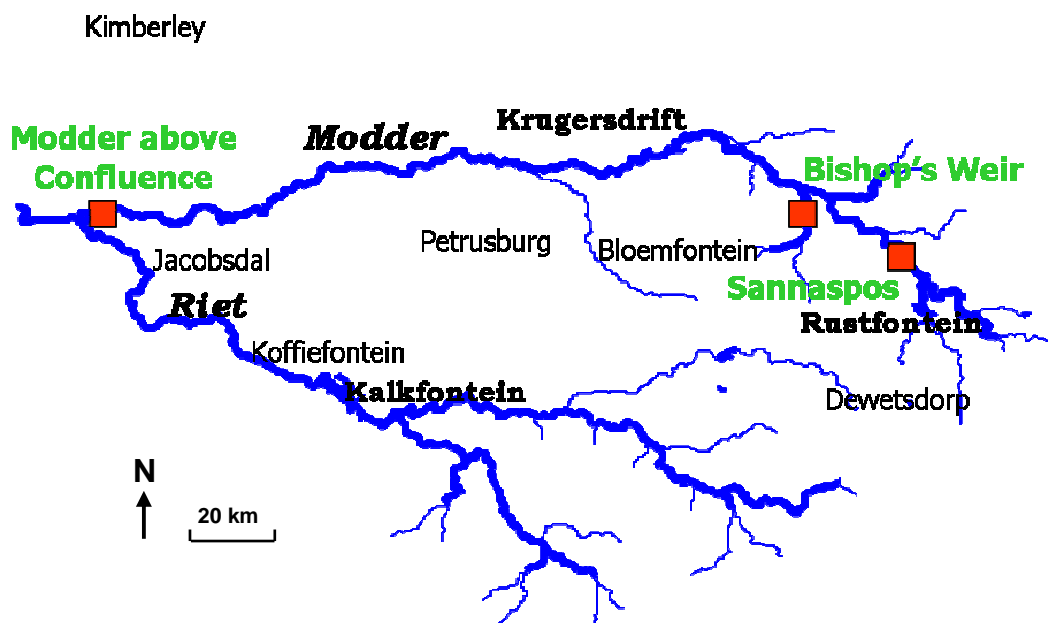


Figure 3.1: A map showing the rivers and study sites.

Table 3.1: Descriptions of the study sites and surrounding areas ([†]DCM, 2000; [§]DWA, 2005; [#]Mucina & Rutherford, 2006).

Site	River	Province	Ecoregion [§]	Water Management Areas* Quaternary Catchment	Biome [#]	Bioregion [#]	
Sannaspos	Modder River	Free State	Highveld (11.03)	Upper Orange	C52B	Grassland	Highveld Alluvial vegetation (AZa5)
Modder above Confluence	Modder River	Northern Cape	Southern Kalahari (29.02)	Upper Orange	C52L	Grassland	Upper Gariep Alluvial vegetation (AZa4)
Bishop's Weir	Renoster Spruit	Free State	Highveld (11.10)	Upper Orange	C52F	Grassland	Highveld Alluvial vegetation (AZa5)

Photographs of study sites in various states (“normal”, dry, flood, etc.) are in **Appendix B** (e.g. **Fig. B.1**).

3.2.1 MODDER RIVER

The origin of the Modder River is near Dewetsdorp, from where it flows in a northwesterly direction towards the Soetdoring Nature Reserve. From there it flows in a west–south-west direction, where it converges with the Riet River (**Fig. 3.1**). The Modder River does not consist of a succession of geomorphological zones; approximately 90% of the river belongs to the Lowland zone (sand bed or floodplain) (Seaman *et al.*, 2001). There are several dams and weirs on the Modder River, which makes it highly regulated.

3.2.1.1 Sannaspos (SP) (-29°09'40.0"S, 26°34'19.0"E)

This site is situated 40 km outside Bloemfontein, approximately 100 m downstream from the N8 Bloemfontein-Thaba Nchu road and train bridges constructed across the Modder River and a weir (built in 1896). This site is located downstream of the Rustfontein Dam (built in 1955 for mostly domestic water use) and the confluence of the Klein Modder and Modder Rivers (Seaman *et al.*; 2001). In summer, thousands of swallows nest on the underside of the bridge and their droppings fall into the river. The upper part of the Modder River is connected to Knellpoort Dam through the Caledon-Modder Transfer Scheme (Novo Transfer Scheme; built in late 1990's). This site also receives water from the Botshbelo sewage treatment works (STW) via the Klein Modder River.

The river is approximately 2–5 m wide and 0.25–0.5 m deep. The banks are covered with vegetation (shrubs as well as trees and grass) and large areas of rock. The river itself runs over bedrock, with very few loose rocks in the stream (**Fig. 3.2**). This site falls within the Lowland geomorphological zone classification with an alluvial channel type (Seaman *et al.*, 2001). The general velocity is slow to medium, depending on the season. Reeds are as abundant at this site as at the other sampling sites. From this point in the river, the vegetation alongside the river increases. There are visible signs of erosion on the riverbanks at this site. From aerial images, it is visible that agriculture (dry-land cultivation and livestock farming) has an impact on the surrounding area (**Fig. 3.3**).



Figure 3.2: Sannaspos.

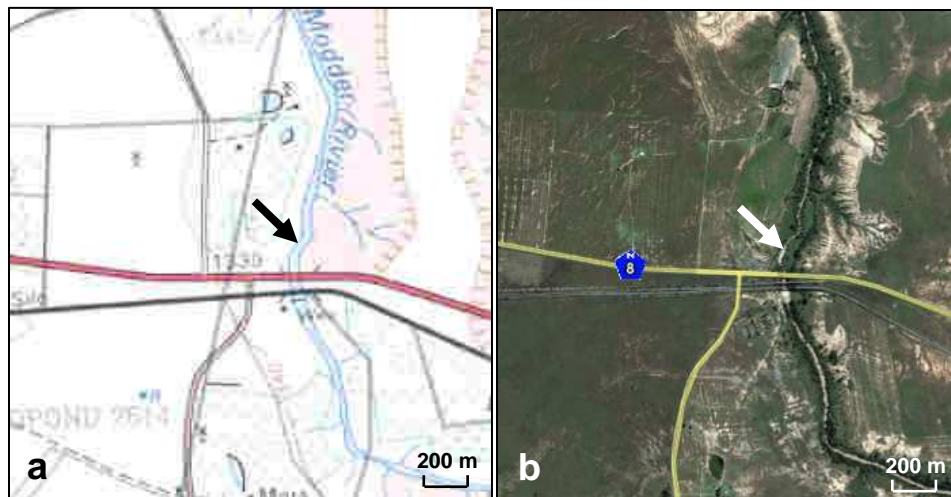


Figure 3.3: Location of Sannaspos site – (a) 1:50 000 Topographic Map Sheet no: 2926BA Sannaspos (CD:SM, 2002) and (b) aerial photograph (Google Earth, 2007).

Fig. B.1 shows the study site (where sampling was done) during “normal” flow at various times of the year throughout the study period. **Fig. B.2, B.3** and **B.4** show the site while dry, with higher than “normal” flow and during flooding.

Sampling was done neither when the site was dry, nor when it was in flood. At sampling times, the flooding was mostly associated with water released from the Rustfontein Dam.

Fig. B.5 shows sampled rocks covered with periphyton, while **Fig. B.6** shows filamentous algae on the bedrock of the stream during May 2004. It was common to find the filamentous algae at this site during the May sampling, as the flow is usually lower during that time of year (lower rainfall) and water temperatures are not yet very low.

3.2.1.2 Modder above Confluence (MC) (-29°01'42.0"S, 24°38'18.0"E)

This site is situated on the Modder River before it flows into the Riet River. The site is situated downstream of the Nico Smit Bridge and approximately 20 m downstream of a gauging weir. Above the Nico Smit Bridge, a big weir also receives water from the Orange-Riet River Water Scheme (mostly for agricultural water use). This site also falls within the Lowland geomorphological zone classification with an alluvial channel type (Seaman *et al.*, 2001). The stream has an average width of 2–5 m and depth of 0.25–0.5 m. The general stream velocity is slow to medium. The banks of the river slopes less than 30° and are covered with reeds and grass (**Fig. 3.4**). The aerial image shows that agriculture (central pivot and flood irrigation) has an impact on the surrounding area (**Fig. 3.5**).

Fig. B.7 shows the site (cobble bed) under “normal” flow conditions, while **Fig. B.8, B.9, B.10** and **B.11** show the site under low and high low conditions, as well as under no-flow and flood conditions. No sampling was done when there was no flow, nor when it was flooded. The flooding was either associated with rainfall or excess water from the Orange/Riet canal system.

Fig. B.12 shows rocks/cobble with periphyton on it that was sampled, as well as rocks in the run (where sampling was done) and in the transition zone (Sep 04) between the pool and the run.



Figure 3.4: Modder above Confluence.

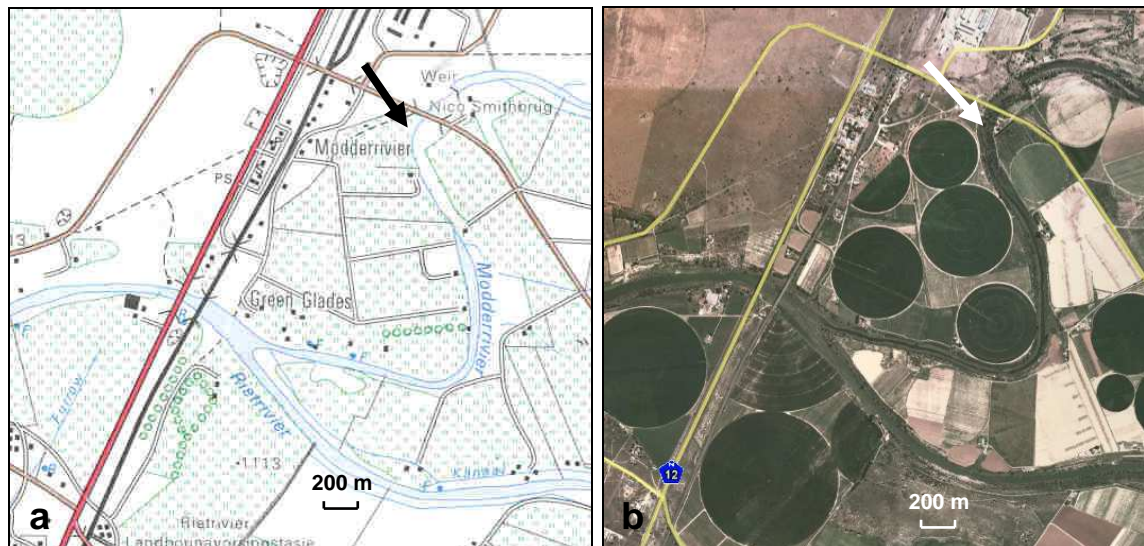


Figure 3.5: Location of Modder above Confluence site – (a) 1:50 000 Topographic Map Sheet no: 2924BA Modderrivier (CD:SM, 2002) and (b) aerial photograph (Google Earth, 2007).

3.2.2 RENOSTER SPRUIT

The Renoster Spruit (**Fig. 3.1**) is a tributary of the Modder River, and has several weirs on it. It originates southeast of Mangaung and carries water that includes runoff through urban and industrial areas, as well as treated wastewater. Just upstream from where it crosses the S376 road, the Bloem Spruit (a tributary that originates in the mid-western part of Bloemfontein) joins and its water sources are mainly the same as those of the Renoster Spruit.

3.2.2.1 Bishop's Weir (BW) (-28°58'00.0"S, 26°19'09.0"E)

The site is situated on the middle reaches of the Renoster Spruit at Shannon Valley on the eastern outskirts of Bloemfontein. It is located directly downstream of the weir, a concrete road bridge and a steel train bridge over the river, 2 km upstream from the confluence of the Renoster Spruit and the Modder River at Glen. The origin of the water in the Renoster Spruit also includes two sewage treatment works (STW); the Sterk STW in the upper Renoster Spruit and the Bloemspruit STW on the lower Bloem Spruit (tributary of the Renoster Spruit). This site falls within the Foothills (cobble bed) geomorphological zone classification with a fixed boulder bed channel type (Seaman *et al.*, 2001).

The average stream width is 5–10 m and the depth 0.5 m (**Fig. 3.6**). The stream flows in a series of rapids (SIC biotope) over a dolerite outcrop into a pool. A small amount of gravel is found between the rocks. The flow is mostly moderate to high. The general velocity is medium to fast. Banks slope less than 30° and are 80% covered by rocks, trees, shrubs and grasses. The main sources of impact on the Renoster Spruit are livestock farming (dairy) and effluent from sewage treatment works on the Renoster Spruit and the Bloem Spruit. Extensive fish kills at the site have occurred in the past (Seaman *et al.*, 2011). From aerial images, it is visible that agriculture (dry-land cultivation and irrigation) has an impact on the surrounding area (**Fig. 3.7**).

Fig. B.13 shows the site at “normal” flow condition. **Fig. B.14** and **B.15** show the site during low and high flow condition. This site was neither dry, nor in flood during any of the sampling times over the study period. **Fig. B.16** shows the periphyton on the sampled rocks/small boulders. Note the thick periphyton mat on the rocks, regardless of the season.



Figure 3.6: Bishop’s Weir.

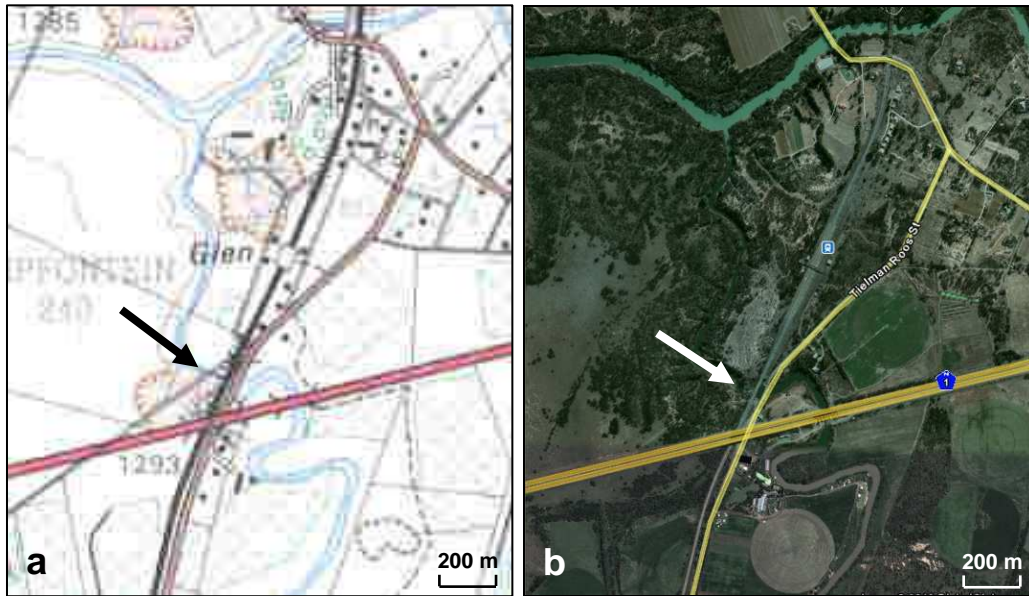


Figure 3.7: Location of Bishop's Weir site – (a) 1:50 000 Topographic Map Sheet no: 2826CD Glen (CD:SM, 2002) and (b) aerial photograph (Google Earth, 2007).

3.3 MATERIALS & METHODS

3.3.1 SAMPLING

Sampling in the Modder River system took place every two months for a period of 47 months, i.e., June 2003 to May 2005 and January 2006 to November 2007, to co-inside with benthic macroinvertebrate (SASS) sampling. The initial study had been planned for a two-year period, after which it was decided to add an additional two years to increase the statistical significance of the data.

In situ measurements were made and subsurface samples (1 litre) were taken from shore and brought to the laboratory for chemical analysis. Samples were kept on ice and stored in a refrigerator ($\leq 5-0$ °C; for two days) until the analyses could be done in the laboratory. The water samples for analysis by the Institute for Groundwater Studies (UFS) were stored, according their specifications, in a refrigerator.

The water used for chlorophyll-a analyses were filtered directly after the laboratory was reached, as well as the preservation of the water to be used for algal composition assemblage.

For the phytoplankton samples, subsurface water was used, while for the periphyton a known surface area was cleared for both the chlorophyll-a concentration and the algal composition assemblage determination (revised from Porter *et al.*, 1993 and Stevenson & Bahls, 1999).

Photographs of some equipment are shown in **Appendix C** (example: **Fig. C.1**).

3.3.2 PHYSICAL & CHEMICAL

The water temperature (°C), concentration of dissolved oxygen (mg/l), percentage of oxygen saturation (O₂ %), electrical conductivity (µS/cm) and total dissolved salts (ppt or g/l) were measured *in situ*, from the shore, with a YSI Model 85 oxygen, conductivity, salinity and temperature meter. Electrical conductivity serves as an indicator of the dissolved salts in the water.

As described in Vos (2002), turbidity, a measurement of the concentration of suspended organic, inorganic and biological material in the water (clarity), was determined with an Aqua Lytic Turbidimeter AL 1000 and is expressed as Nephelometric Turbidity Unit (NTU).

The pH and Redox potential (mV) was measured *in situ*, using Eutech Instruments CyberScan pH 110 meter (pH/mV/°C/°F with RS232). Redox potential may indicate extreme chemical conditions, eutrophication and anaerobic conditions.

Atmospheric temperature and rainfall data for the Bloemfontein (station – 0261516B0) and Kimberley (station – 0290468A9) area for 2003-2007 were obtained from the South African Weather Service (SAWS, n.d.(b&c)).

Flow (m/s) was measured 4/10s from the bottom of the water column depth (where periphyton was sampled) by using an OTT Z30 Counter attached to an OTT Small Current Meter C2. To determine the flow velocity, the following formula was used:

$$\text{Prop Speed (N)} = \frac{\text{Revolutions}}{\text{Time(s)}} \quad \text{Eq. 3.1}$$

$$\text{Flow Velocity (V)} = (N \times 0.226) + 0.024 \quad \text{if } N \leq 0.7, \text{ and}$$

$$\text{Flow Velocity (V)} = (N \times 0.2547) + 0.004 \quad \text{if } N \leq 9.79.$$

The Grid References were taken by a Garmin: eTrex Vista Personal navigator GPS.

Dissolved reactive *ortho*-phosphate ($\text{PO}_4\text{-P}$; indication of eutrophication) (as described in Vos, 2002) was determined using 100 ml GF/C filtered water, together with the Stannous Chloride Method, as described in APHA (2005). Ammonium molybdate reacts with stannous chloride, whereby molybdophosphoric acid is formed and reduced by stannous chloride to intensely coloured molybdenum blue. Absorbance* was read at 690 nm.

The following salts were measured by the Institute for Groundwater Studies (UFS) using an Inductively Coupled Plasma (ICP) Spectrometer: Calcium (Ca^{2+}), Magnesium (Mg^{2+}), Sodium (Na^{1+}), Potassium (K^{1+}), Phenolphthalein alkalinity (Palk), Total alkalinity (Talk), Chloride (Cl^{1-}), Nitrate-nitrogen ($\text{NO}_3\text{-N}$), Phosphate (PO_4^{2-}), Sulphate (SO_4^{2-}), and Ammonium-nitrogen ($\text{NH}_4\text{-N}$). All mentioned variables were used to determine total dissolved salts (TDS). Phosphate was used as dissolved inorganic phosphorus (DIP), and $\text{NO}_3\text{-N}$ plus $\text{NH}_4\text{-N}$ was used as dissolved inorganic nitrogen (DIN).

*All absorbances were determined with the VIS-7220 spectrophotometer for the determining of the unknown concentrations, by plotting it against a standard curve of known concentrations for each of the analyses.

Determination of chlorophyll-a concentration

As described in Vos (2002), the use of chlorophyll-a as an index of trophic status is because it is normally the most abundant and important pigment in phytoplankton cells. Thus, measurements provide a convenient estimation of the algal biomass (Walmsley, 1984). Chlorophyll-a in this study was measured using a modified method described by Sartory and Grobbelaar (1984), and Stevenson and Bahls (1999), and involved filtering a known volume of water through a GF/C filter paper, whereafter the filter paper was suspended in 10 ml 95% ethanol overnight (>12h). The absorbance was measured at 750 nm and 665 nm. After adding 100 µl of 0.3 N HCl, the absorbance was again measured after 2 minutes at 665 nm with a VIS-7220 spectrophotometer.

The following formula was used:

$$\text{Chlorophyll-a in extract (mg/l)} = (A_{665} - A_{665a}) \times 28.66 \text{ where:} \quad \text{Eq. 3.2}$$

A_{665} = absorbance of ethanol extract at 665 nm before it was acidified minus absorbance at 750 nm.

A_{665a} = absorbance of the acidified ethanol extract at 665 nm minus the absorbance at 750 nm.

The concentration of chlorophyll-a in the original sample:

$$\text{Concentration } (\mu\text{g/l}) = \frac{\text{concentration of extract} \times 10 \text{ ml (extract volume)}}{\text{volume of sample in litre}}$$

Eq. 3.3

For the phytoplankton (in the free-flowing water), the formula was used as is, but for the periphyton (epilithic) it was further converted to $\mu\text{g}/\text{cm}^2$.

$$(y \mu\text{g}/\ell \times \text{volume of sample in litre}) \div \text{known surface area cm}^2 = z \mu\text{g}/\text{cm}^2$$

$$\text{e.g. } (10 \mu\text{g}/\ell \times 0.1 \ell) \div 7.5 \text{ cm}^2 = 0.13 \mu\text{g}/\text{cm}^2$$

Eq. 3.4

3.3.3 BIOLOGICAL

Collecting periphyton

Periphyton (epilithic) was sampled by selecting flat-surfaced instream rocks, stones or bedrock that are representative of the algal coverage present. The flow velocity of the river was also taken into consideration, as the flow in the middle of a river is faster than closer to the riverbank.

Samplings were done on the same rocks or in the same area with each visit. A known area (7.5 cm^2 ; inside area surface of a tube with rubber O-ring) was then scraped off with a stainless-steel bristle brush for each of the periphytic chlorophyll-*a* and algal samples. The water was then abstracted by a syringe with a rubber tube attached to its tip (**Fig. C.1**; revised from Porter *et al.*, 1993 and Stevenson & Bahls, 1999). The chlorophyll-*a* sample was filled up to 100 ml and the algal composition sample up to 50 ml (to be used as constant, known volumes in calculations).

Algal genera identification and enumeration

As described in Vos (2002), the dominant algal genera (phytoplankton & periphyton) were identified (to genus level only) with an inverted Zeiss Light Microscope after fixation with formaldehyde (final concentration of 2%) and placed in a sedimentation chamber according to the Utermöhl method for at least 24 hours (Wetzel & Likens, 1991). The number of a specific algal genera was

determined in a known volume of water, counting the individuals (cells, filaments and colonies) occurring in 20 blocks of known dimensions (**Fig. C.2**). The result was multiplied by a constant to obtain the total concentration. Algal genera were determined as a percentage of the total community.

For the phytoplankton (in the free-flowing water), the formula was used as is, but for the periphyton (epilithic) it was further converted to cells/cm².

$$(y \text{ cells/ml} \times \text{algal preserved volume in ml}) \div \text{known surface area cm}^2 = z \text{ cells/cm}^2$$

e.g. $(100 \text{ cells/ml} \times 50 \text{ ml}) \div 7.5 \text{ cm}^2 = 667 \text{ cells/cm}^2$ **Eq. 3.5**

Algal biovolume calculations

The biovolume of each genus was calculated by using the formulas suggested in Hillebrand *et al.* (1999) and The Academy of Natural Science (2011) webpage. The dimensions for the calculations were obtained by measuring the algae (various representative samples from 2003 to 2007 were re-prepared during July and August 2011) under the inverted Zeiss Light Microscope with a calibrated grid in one of the ocular lenses (**Fig. C.2**). Between five (5) and twenty (20) measurements were taken for each genera, depending on the abundance present in the samples.

The average Bacillariophyta (diatom) genera dimensions were compared to the average dimensions measured by Taylor *et al.* (2006), and the rest of the genera to dimensions found on Protist information server (2003) or The Academy of Natural Science (2011) web pages. A few algae were not observed for measurement, and average dimensions were used as found mentioned in the above-mentioned sources.

Macroinvertebrates

SASS4 & SASS5 were done by Ms Marie Watson (Centre for Environmental Management, UFS) as part of a biomonitoring project on the Modder, Riet and Caledon Rivers. The samplings were done at the same sites and on the same days as for this study.

South African Scoring System version 4 & 5 (SASS4 & 5)

Dallas (1995) and Chutter (1994) describe SASS4 as a scoring system based on benthic macroinvertebrates, where a sensitivity/tolerance score is allocated to each taxon according to its known tolerance to water quality conditions. The higher the SASS score, the more sensitive is the taxon.

For the SASS4 method in biomonitoring, the total SASS4 score for each site is calculated by adding all the taxon scores, and the Average Score per Taxon (ASPT) is calculated by dividing the Total SASS4 Score by the number of taxa found at the site. Both these scores are considered when determining water quality impairment (Dallas, 1995).

SASS5 (**Fig. C.3**) is a revised version of SASS4 (Dickens & Graham, 2002) and has been used by the River Health Programme since May 2001. **Table 3.2** can be used to interpret the scores obtained during sampling.

Table 3.2: SASS5 and ASPT values as an indication of biotic conditions developed for the Highveld (adjusted from Thirion, 2003).

SASS5	ASPT	Condition
>120	>5.5	Excellent
91-120	5.0-6.0	Very Good
71-90	4.5-5.5	Good
61-70	4.5-5.5	Fair
30-60	variable	Poor
<30	variable	Very Poor

Fish

The FAIL (old index) and FRAI indices for fish were done by the Centre for Environmental Management (UFS) as part of a biomonitoring project on the Modder, Riet and Caledon Rivers. The fish were sampled as a once off, each year. Each of the study sites were included in the various reaches used for their FAIL and FRAI data.

3.3.4 STATISTICAL ANALYSIS

Statistical analyses and graphs were done with ³PRIMER (v6; Primer-E Ltd), SigmaPlot (v7; Systat Software Inc) & Microsoft Office Excel 2003.

In the Box plots (SigmaPlot) the box represents the 25th–75th percentiles with the 50th (median) as a solid line and the mean as a dotted line. The whiskers show the 10th and 90th percentiles, while the outliers show those outside the 10th and 90th percentiles.

³ Plymouth Routines In Multivariate Ecological Research.

The Pearson correlation coefficients between the various environmental factors and periphytic algal components were calculated in PRIMER and interpreted as follows:

Correlation coefficient (r) & coefficient of determination (r^2)

- ◆ If the correlation value is +1 or -1 ($r^2 = 1$) it is a perfect relationship.
- ◆ If the correlation value is close to +1 or -1 ($r^2 = 1$) it is a strong relationship, usually beyond 0.7 or -0.7 ($r^2 = 0.49$).
- ◆ If the correlation value is closer to +0.5 or -0.5 ($r^2 = 0.25$) it is an average to moderate (>0.6 or >-0.6 ; $r^2 = 0.36$) relationship.
- ◆ If the correlation value is greater than 0.3 or smaller than -0.3 ($r^2 = >0.09$) it is a weak relationship.
- ◆ If the correlation value is smaller than 0.3 or greater than -0.3 ($r^2 = <0.09$) it is negligible.

Probability (p)

- ◆ If the p -value is <0.001 (very small), it is statistically highly significant.
- ◆ If the p -value is between 0.05 and 0.001 (but not close to 0.05), it is considered statistically significant.
- ◆ If the p -value is close to 0.05, it is considered marginally significant.
- ◆ If the p -value is >0.05 (but not close to), it is considered insignificant.

(Rumsey, 2009; Rumsey, 2010)

The data for the Principal Component Analyses were typed as environmental in PRIMER, whereupon the data were normalised. The normalised data were then used to calculate the Euclidean distance or PCA after which the graphs were drawn.

CHAPTER 4

RELATIONSHIPS BETWEEN PERIPHYTON ALGAL COMPONENTS AND PHYSICAL AND CHEMICAL CONDITIONS

4.1 INTRODUCTION

Periphyton is sensitive to a change in water quality (McCormick & Stevenson, 1998) and reacts to it. Some algae grow in length (filamentous algae), while others simply divide. Then there is also the difference in algal cell size (macro- vs. microalgae) (Arnwine & Sparks, 2003) to take into consideration, not only between genera, but also between different species in the same genus (Lavoie *et al.*, 2006).

If a sample assemblage comprises of mixed species, large numbers of small-sized species may only make up a small part of the total biovolume, while larger species may have a higher biovolume-to-numbers ratio (Hillebrand *et al.*, 1999). Algae are also known to be able to adjust their intracellular chlorophyll-*a* concentration, depending on the environment (Von Schiller *et al.*, 2007).

This chapter investigates the multivariate relationships between the periphytic algal components and the physical and chemical factors that drive them. The data for the sites were calculated individually and combined to seek the best results.

4.2 RESULTS

Additional processed data tables are in **Appendix D.1**. Tables and figures in **Appendix D** are numbered as, e.g. **Fig. D.1**.

4.2.1 SANNASPOS

Sannaspos (SP; $n = 21$; **Fig. 4.1**) had very strong linear and statistically highly significant correlations between the total periphytic algal concentrations (cells/cm^2) and biovolume ($\mu\text{m}^3/\text{cm}^2$) ($r^2 = 0.762$; $p = <0.001$), periphytic chlorophyll-a (pchl-a; $\mu\text{g}/\text{cm}^2$) concentrations and biovolume ($r^2 = 0.678$; $p = <0.001$), and periphytic algal and the pchl-a concentrations ($r^2 = 0.679$; $p = <0.001$).

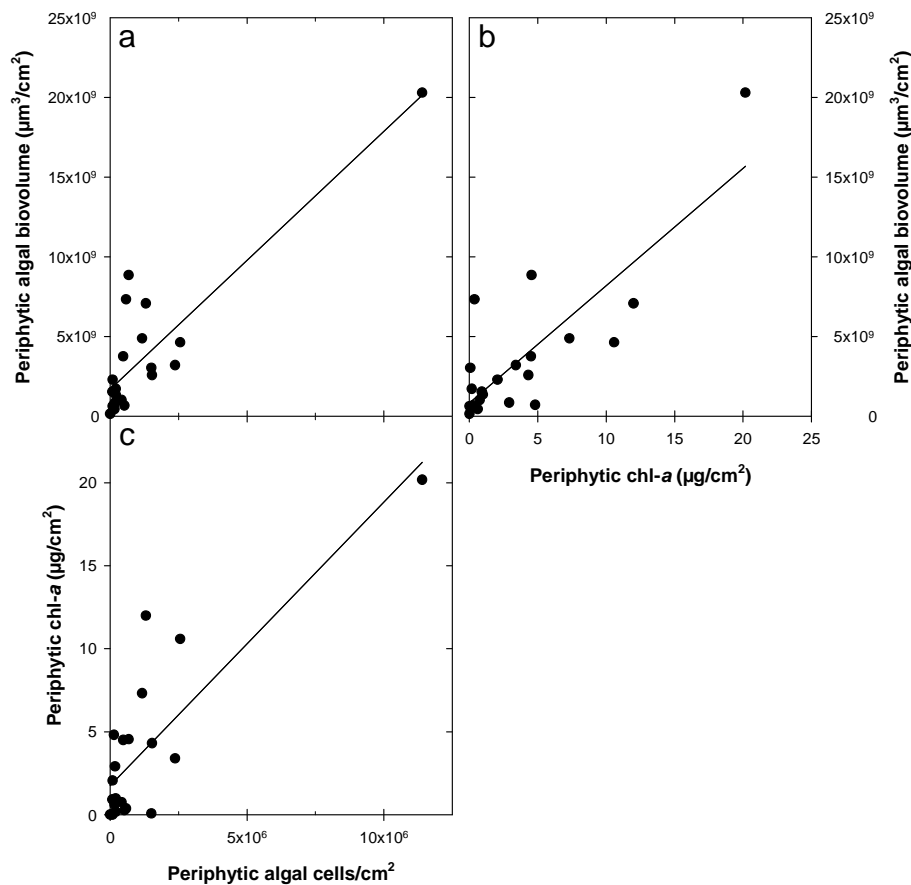


Figure 4.1: The linear relationships between periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration at Sannaspos (SP) over the period from May 2003 to November 2007.

At SP, the dominant division was Bacillariophyta, followed by the Chlorophyta, Euglenophyta and Cyanophyta (**Table 4.1**). The median percentages between

the divisions' biovolume are closer together than those of the concentration are. The total pchl-*a* concentration had strong and statistically highly significant correlations with the periphytic algal concentration of the Bacillariophyta ($r^2 = 0.736$; $p = <0.001$), Chlorophyta ($r^2 = 0.523$; $p = <0.001$) and Euglenophyta ($r^2 = 0.470$; $p = <0.001$), as well as the periphytic algal biovolume of the Bacillariophyta ($r^2 = 0.752$; $p = <0.001$) and Euglenophyta ($r^2 = 0.463$; $p = <0.001$).

Table 4.1: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Sannaspos, percentage of the periphytic algal concentration and biovolume.

	Periphytic algal divisions (%)							
	Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
	conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Min	0.00	0.00	11.33	9.14	0.00	0.00	0.00	0.00
Max	32.56	37.82	97.221	97.31	85.71	90.86	8.00	65.63
Mean	6.60	4.31	72.45	42.98	19.27	29.10	1.53	23.60
Median	2.78	2.02	75.00	34.84	16.67	13.06	0.76	22.75
SD	9.08	8.37	20.19	28.54	21.30	31.49	2.28	23.68

n = 21

At SP, the physical and chemical factors had weak or negligible correlations with the periphytic algal concentration (**Table 4.2; Fig. D.1a**). Regarding the nutrients, the dissolved inorganic phosphorus (DIP) and dissolved inorganic nitrogen (DIN) had moderate (insignificant) and strong (statistically significant) relationships, respectively, with the periphytic algal concentration (**Fig. D.1b**). The periphytic algal biovolume (**Table 4.2; Fig. D.2**) showed weak and negligible correlations with the physical and chemical factors. DIP and DIN correlations, although stronger, have both a weak (insignificant) to moderate (statistically significant) relation with the periphytic algal biovolume. The physical and chemical factors had either weak or negligible correlations with the pchl-*a* concentration (**Table 4.2; Fig. D.3**), while the DIP and DIN concentration had weak (insignificant) to average (statistically significant) correlations with the pchl-*a* concentration.

Table 4.2: The coefficient of determination (r^2) and probability (p) values at Sannaspos between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)	
	r^2	p	r^2	p	r^2	p
Temperature	(-) 0.140	<0.1	(-) 0.114	<0.1	(-) 0.192	<0.1
Diss. oxygen	0.018	>0.1	0.027	>0.1	0.172	>0.1
Redox	(-) 0.092	>0.1	(-) 0.062	>0.1	(-) 0.030	>0.1
pH	5.09×10^{-3}	>0.1	0.012	>0.1	5.94×10^{-3}	>0.1
Turbidity	(-) 0.021	>0.1	(-) 0.070	>0.1	(-) 0.030	>0.1
Flow	(-) 0.015	>0.1	(-) 0.025	>0.1	(-) 3.32×10^{-3}	>0.1
DIP	0.153	<0.1	0.104	<0.1	0.203	<0.1
DIN	0.302	<0.01	0.131	<0.01	0.237	<0.01
TDS	0.032	>0.1	0.062	>0.1	0.116	>0.1

n = 21; (-) = indicates a negative correlation.

4.2.2 BISHOP'S WEIR

At Bishop's Weir (BW; n = 25; **Fig. 4.2**), the periphytic algal concentrations and biovolume ($r^2 = 5.29 \times 10^{-3}$; $p = >0.1$) and pchl-a concentrations and biovolume ($r^2 = 0.025$; $p = >0.1$) had no significant linear correlation between them, while the periphytic algal concentration and the pchl-a concentration ($r^2 = 0.207$; $p = <0.05$) had an average correlation with statistical significance.

Once again, at the BW site, the Bacillariophyta was the dominant periphyton division, followed by the Chlorophyta (**Table 4.3**). Here the divisions' algal biovolumes medians (percentages) are also much closer than those of the algal concentration are. The divisions that had the best correlations with the total pchl-a concentration were the Bacillariophyta ($r^2 = 0.143$; $p = <0.1$) and Chlorophyta ($r^2 = 0.126$; $p = >0.1$) algal concentrations with weak, insignificant

correlations, and the Bacillariophyta biovolume ($r^2 = 0.285$; $p = <0.02$) with an average correlation and statistical significance.

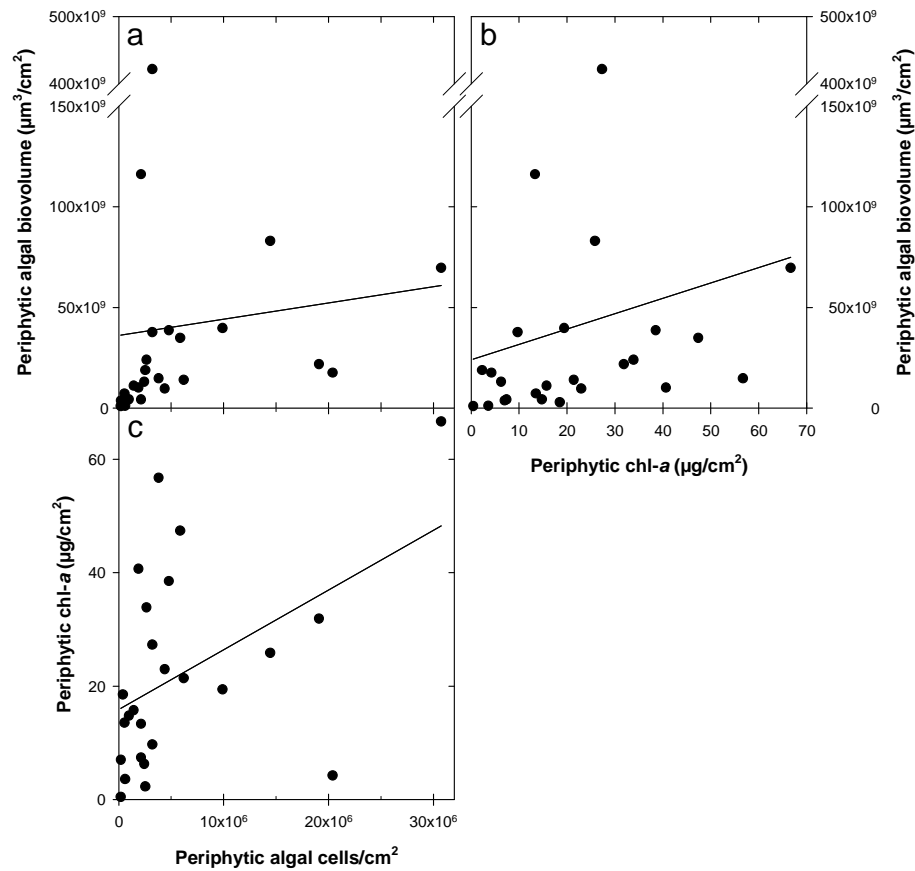


Figure 4.2: The linear relationships between periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration at Bishop's Weir (BW) over the period from May 2003 to November 2007.

Table 4.3: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Bishop's Weir, percentage of the periphytic algal concentration and biovolume.

	Periphytic algal divisions (%)							
	Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
	conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Min	0.00	0.00	5.26	1.48	0.79	0.28	0.00	0.00
Max	38.46	40.85	97.50	94.85	81.34	98.44	6.10	73.91
Mean	7.65	6.02	67.76	43.43	23.17	31.91	1.36	17.26
Median	2.00	0.88	75.61	38.61	11.11	24.26	0.64	13.65
SD	11.68	12.06	26.14	28.48	25.63	29.95	1.78	21.30

n = 25

The periphytic algal concentrations had only weak and negligibly insignificant correlations with the physical and chemical factors (**Table 4.4; Fig. D.4a**). The DIN had a weak, insignificant correlation with the periphytic algal concentration (**Table 4.4; Fig. D.4b**). The physical and chemical factor correlations with the periphytic algal biovolume (**Table 4.4; Fig. D.5a**) were all weak or negligible and insignificant. With the nutrients (DIN & DIP; **Table 4.4; Fig. D.5b**) the correlation with the periphytic algal biovolume was negligible and insignificant. The pchl-a concentration (**Table 4.4; Fig. D.6a**) had moderate (statistically significant) correlations with temperature (neg. corr.), while the DIN and DIP concentrations had weak (statistically significant) and negligible (insignificant) correlations, respectively (**Table 4.4; Fig. D.6b**).

Table 4.4: The coefficient of determination (r^2) and probability (p) values at Bishop's Weir between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)	
	r^2	p	r^2	p	r^2	p
Temperature	(-) 0.090	>0.1	7.87×10^{-3}	>0.1	(-) 0.359	<0.01
Diss. oxygen	0.031	>0.1	(-) 0.012	>0.1	0.196	<0.05
Redox	0.119	>0.1	0.042	>0.1	0.104	>0.1
pH	(-) 8.25×10^{-3}	>0.1	(-) 0.061	>0.1	(-) 0.81×10^{-3}	>0.1
Turbidity	0.023	>0.1	(-) 0.017	>0.1	(-) 6.61×10^{-3}	>0.1
Flow	(-) 0.048	>0.1	(-) 0.039	>0.1	(-) 0.092	>0.1
DIP	0.78×10^{-3}	>0.1	0.014	>0.1	0.067	>0.1
DIN	0.119	>0.1	(-) 0.022	>0.1	0.212	<0.05
TDS	(-) 0.018	>0.1	(-) 0.21×10^{-3}	>0.1	0.025	>0.1
DIN:DIP	0.099	>0.1	(-) 0.061	>0.1	0.174	<0.1

n = 25; (-) = indicates a negative correlation.

4.2.3 MODDER ABOVE CONFLUENCE

At Modder above Confluence (MC; n = 19; **Fig. 4.3**), the periphytic algal concentrations and biovolume ($r^2 = 0.106$; $p = >0.1$), and pchl-a concentrations and biovolume ($r^2 = 0.090$; $p = >0.1$) had no linear significant correlations, while the periphytic algal concentration and the pchl-a ($r^2 = 0.582$; $p = <0.001$) had a strong, statistically highly significant correlation.

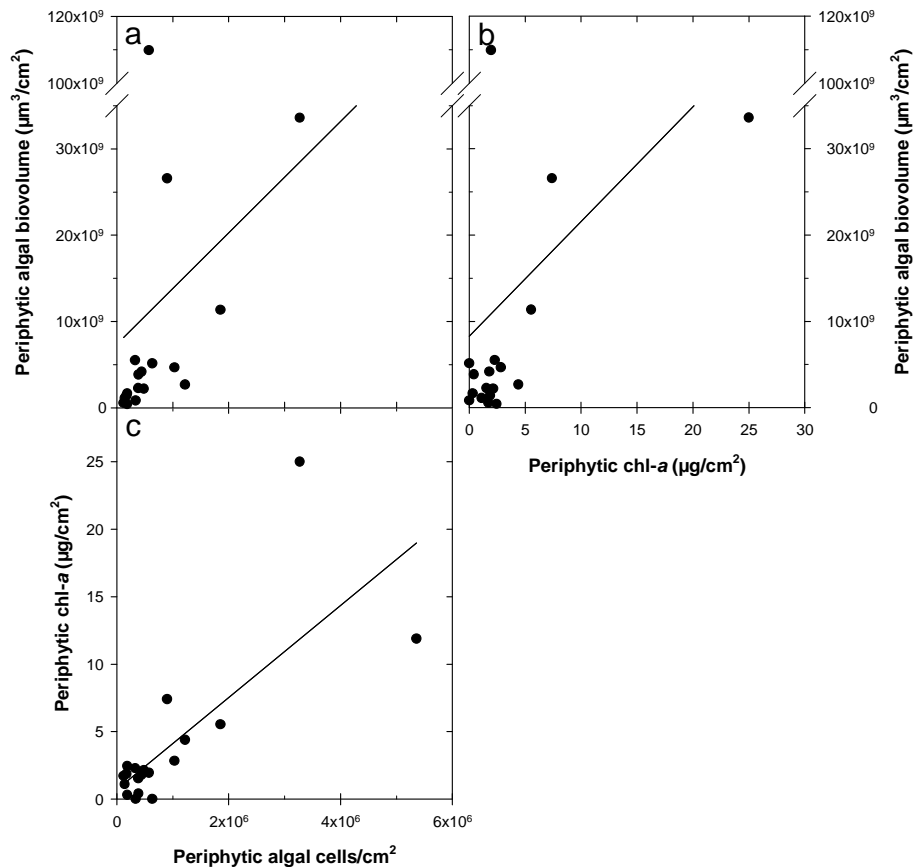


Figure 4.3: The linear relationships between periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration at Modder above Confluence (MC) over the period from May 2003 to November 2007.

At the MC, the Bacillariophyta were the dominant division of the periphytic algae, followed by the Chlorophyta (**Table 4.5**). The difference between the various divisions' median percentage of the periphytic algal biovolumes was also closer than that of the percentage concentrations, especially the Bacillariophyta and Chlorophyta. The Bacillariophyta were the only division that had any correlation with the pchl-a concentration. Both the Bacillariophyta periphytic algal concentration ($r^2 = 0.632$; $p = <0.001$) and biovolume ($r^2 = 0.737$; $p = <0.001$) had strong, statistically highly significant correlations with the pchl-a concentration.

Table 4.5: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Modder above Confluence, percentage of the periphytic algal concentration and biovolume.

	Periphytic algal divisions (%)							
	Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
	conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Min	0.00	0.00	18.76	0.37	0.00	0.00	0.00	0.00
Max	47.37	34.15	100.00	100.00	65.62	94.34	32.58	63.30
Mean	10.30	4.67	67.62	39.72	18.42	35.06	3.32	20.37
Median	2.27	0.53	64.93	32.41	13.16	34.21	1.49	11.66
SD	14.11	8.60	23.50	30.85	16.85	33.30	7.33	22.88

n = 19

The physical and chemical factors had no significant correlations with any of the periphytic algal components (**Table 4.6; Fig. D.7a, D.8a & D.9a**), neither did the nutrients (DIP & DIN; **Table 4.6; Fig. D.7b, Fig. D.8b & Fig. D.9b**). The pchl-a concentration had a strong (statistically highly significant) correlation with the DIN:DIP ratio.

Table 4.6: The coefficient of determination (r^2) and probability (p) values at Modder above Confluence between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)	
	r^2	p	r^2	p	r^2	p
Temperature	(-) 0.116	>0.1	(-) 0.051	>0.1	0.111	>0.1
Diss. oxygen	0.032	>0.1	0.62×10^{-3}	>0.1	0.019	>0.1
Redox	0.023×10^{-3}	>0.1	(-) 7.55×10^{-3}	>0.1	0.060	>0.1
pH	(-) 5.28×10^{-3}	>0.1	(-) 0.082	>0.1	0.038	>0.1
Turbidity	(-) 0.029	>0.1	(-) 0.027	>0.1	0.068	>0.1
Flow	0.018	>0.1	2.72×10^{-3}	>0.1	0.020	>0.1
DIP	(-) 0.039	>0.1	(-) 0.053	>0.1	0.018	>0.1
DIN	7.22×10^{-3}	>0.1	(-) 4.05×10^{-3}	>0.1	0.153	<0.1
TDS	1.25×10^{-3}	>0.1	0.025	>0.1	0.094	>0.1
DIN:DIP	0.122	>0.1	0.015	>0.1	0.589	<0.001

n = 19; (-) = indicates a negative correlation.

4.2.4 COMBINED SITES

It was found that the combined data of all the sites (SP, BW and MC; n = 65; **Fig. 4.4a**) showed no significant linear correlation ($r^2 = 0.039$; $p = >0.1$) between the total periphytic algal concentrations and biovolume. The pchl-a concentration and biovolume had a weak but statistically significant correlation ($r^2 = 0.094$; $p = <0.02$; **Fig. 4.4b**), while a statistically highly significant and moderate linear correlation ($r^2 = 0.372$; $p = <0.001$; **Fig. 4.4c**) was found between the periphytic algal concentration and the pchl-a concentration.

The Bacillariophyta were the dominant division found with the combined periphytic data, followed by the Chlorophyta with both the periphytic algal concentration and biovolume (**Table 4.7**). The Cyanophyta and the Euglenophyta showed lower presence.

The total pchl-a concentration had statistically highly significant and moderate to strong linear correlations to the Bacillariophyta concentration ($r^2 = 0.355$; $p < 0.001$) and biovolume ($r^2 = 0.495$; $p < 0.001$), respectively. The divisions had weak or negligible correlations.

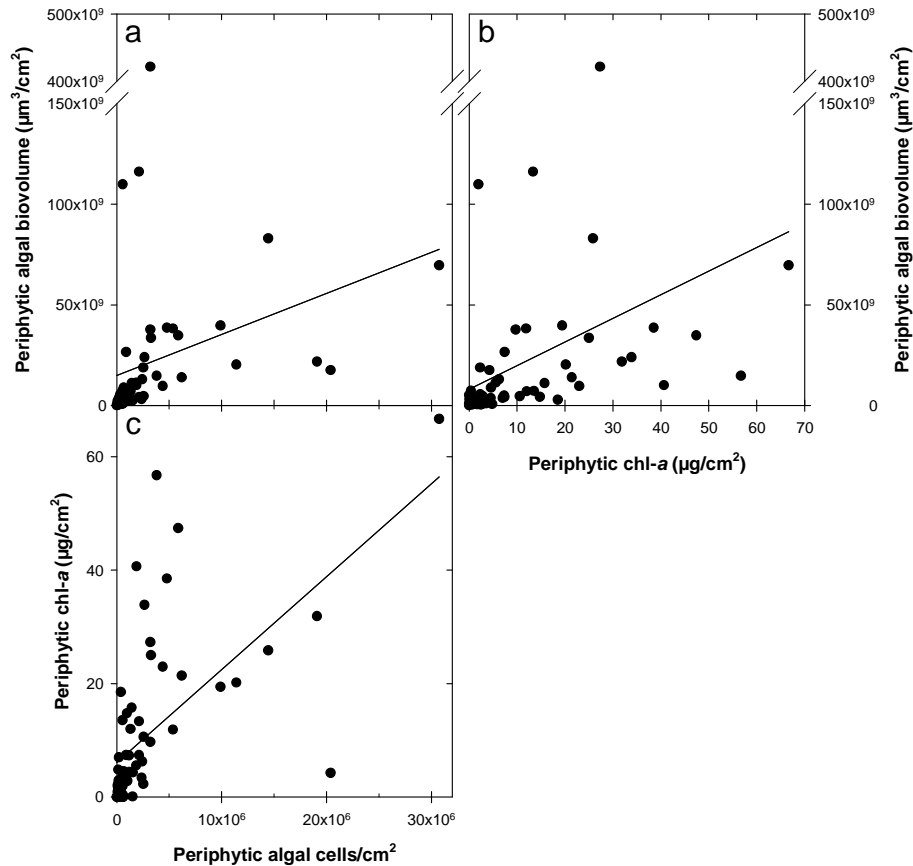


Figure 4.4: The linear relationships between the combined periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007.

The physical and chemical factors all had weak or negligible correlations (mostly insignificant) with the periphytic algal components (**Table 4.8; Fig. D.10a, D.11a & D.12a**). The periphytic algal concentration had an average correlation (statistically significant) to the DIN concentration (**Table 4.8; Fig. D.10b**). The pchl-a concentration had moderate correlations (statistically highly significant) with DIN and DIP (**Table 4.8; Fig. D.12b**).

Table 4.7: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at the combined sites, percentage of the periphytic algal concentration and biovolume.

	Periphytic algal divisions (%)							
	Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
	conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Min	0.00	0.00	5.26	0.37	0.00	0.00	0.00	0.00
Max	47.37	40.85	100.00	100.00	85.71	98.44	32.58	73.91
Mean	8.08	5.07	69.23	42.20	20.52	31.92	1.99	20.22
Median	2.27	0.91	72.41	35.40	12.75	24.26	0.76	13.65
SD	11.63	9.89	23.32	28.79	21.74	31.04	4.32	22.35

n = 65

Table 4.8: The coefficient of determination (r^2) and probability (p) values of the combined sites between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)	
	r^2	p	r^2	p	r^2	p
Temperature	(-) 0.039	>0.1	2.20×10^{-3}	>0.1	(-) 0.087	<0.02
Diss. oxygen	0.027	>0.1	0.032×10^{-3}	>0.1	0.102	<0.02
Redox	3.48×10^{-3}	>0.1	4.60×10^{-3}	>0.1	0.49×10^{-3}	>0.1
pH	5.43×10^{-3}	>0.1	(-) 9.17×10^{-3}	>0.1	0.016	>0.1
Turbidity	(-) 0.012	>0.1	(-) 0.0132	>0.1	(-) 0.032	>0.1
Flow	0.046×10^{-3}	>0.1	(-) 1.41×10^{-3}	>0.1	2.28×10^{-3}	>0.1
DIP	0.149	<0.01	0.087	<0.02	0.393	<0.001
DIN	0.252	<0.001	7.31×10^{-3}	>0.1	0.443	<0.001
TDS	0.46×10^{-3}	>0.1	6.65×10^{-3}	>0.1	0.054	<0.1

n = 65; (-) = indicates a negative correlation.

4.3 DISCUSSION

4.3.1 SANNASPOS

At Sannaspos, the strong correlations between the total periphytic algal concentrations, the biovolumes and pchl-*a* concentrations (**Fig. 4.1**) make it possible to predict two of these, when only one is available, with some accuracy. A contributing factor was that SP had few filamentous and colonial genera present, also in smaller quantities. In addition, the only Euglenophyta genus present was *Trachelomonas*, while the other sites had other genera (big, with high volumes) in their assemblages.

As most periphyton consists of Bacillariophyta (Potapova & Charles, 2003), it was no surprise when the dominant division at SP turned out to be just that, followed by Chlorophyta (**Table 4.1**). There was less correlation between the pchl-*a* concentration and the individual divisions' concentrations and biovolumes than with those of the total periphytic algae. The vast difference in the percentage of the Bacillariophyta, Chlorophyta and Euglenophyta cell concentrations, but less in the percentage of the biovolume is a suggestion that the Chlorophyta and Euglenophyta most probably contributed more to the primary productivity at SP than the Bacillariophyta did (Lavoie *et al.*, 2006).

The physical (e.g. temperature, turbidity) and chemical (e.g. diss. O₂) factors had little to no major impact on the periphytic algal concentration, biovolume and pchl-*a* concentration, while the nutrients (DIP and DIN) had (**Table 4.2; Fig. D.1, D.2 & D.3**). The DIP concentrations had an overall median in the eutrophic ranges, while the DIN concentrations had a median within the mesotrophic range, indicating excess nutrient availability. According to the regression correlations and significance, the DIN concentration had the most significant effect on the periphytic algal components even if the correlations were at best average, with statistical significance.

4.3.2 BISHOP'S WEIR

Unlike SP, Bishop's Weir only favoured an average (statistically significant) correlation between the periphytic algal and the pchl-a concentration (**Fig. 4.2**). Here the Bacillariophyta was the dominant division (cell concentration) over the Chlorophyta (**Table 4.3**), but with the periphytic algal biovolume there was a smaller difference in percentage. This was supported by the fact that many colonial and filamentous genera that belonged to the Chlorophyta division (Hillebrand *et al.*, 1999), were found. There were also a fair number of Euglenophyta (large cells, big volumes) present in the samples.

The median percentage of the Cyanophyta at BW was lower than that of SP, which concurs with the findings of Douterelo *et al.* (2004) that Cyanobacteria abundance and diversity within the periphytic assemblage decrease with the increase of eutrophication. This is quite different from that of lentic systems (Ejsmont-Karabin & Spodniewska, 1990).

That there were no real significant correlations between the nutrient (DIP and DIN) concentrations and the periphytic algal components could be because this site had always had nutrient concentrations that were considered to be in the eutrophic to hypertrophic ranges (nutrient-saturated) (**Table 4.4; Fig. D.4, D.5 & D.6**). This may also be why only the periphytic algal and pchl-a concentrations have an average (statistically significant) correlation.

4.3.3 MODDER ABOVE CONFLUENCE

Much like BW, Modder above Confluence only had a correlation (strong) between the periphytic algal and pchl-a concentrations, although statistically highly significant, while no correlation could be found between the periphytic algal or pchl-a concentrations and the periphytic algal biovolume (**Fig. 4.3**). MC did not have as much filamentous or colonial genera (and high numbers) as BW, but more than SP did. This showed that pchl-a concentrations are an ideal factor to calculate the living biomass of a site's algal community for comparison

with other samples, whereas other algal metrics do not distinguish between living and dead matter (Wellnitz & Poff, 2006; Kilroy *et al.*, 2013).

With the Bacillariophyta (concentration and biovolume) as the only division to have a meaningful correlation (highly statistically significant) with the total pchl-a concentration, and the small difference between the periphytic biovolume percentages of the Bacillariophyta and Chlorophyta (**Table 4.5**), it seems to confirm the above-mentioned findings that a correlation relationship exists between the periphytic algal and pchl-a concentrations.

After the above-mentioned, it came as no surprise that the periphytic algal concentration and biovolume had negligibly insignificant correlations with any of the physical and chemical factors. The nutrient concentrations were moderate (median in mesotrophic ranges for both the DIP and DIN) and had no significant correlation to the pchl-a concentration at the site (**Table 4.6; Fig. D.7, D.8 & D.9**).

4.3.4 COMBINED SITES

The lack of linear correlation and significance between the combined periphytic algal concentration and biovolume may be due to the variety of algal genera present at the three sites (SP, BW & MC; **Fig. 4.4**), as the cell sizes and thereby the cell volumes not only differ between genera, but also among the individuals of a species (Lavoie *et al.*, 2006). This could also be a result of some filamentous and colonial algal genera that occurred at all the sites throughout most of the study period, e.g. *Oscillatoria*, *Melosira*, *Cladophora*, *Pediastrum* and *Scenedesmus*.

Since the Bacillariophyta (followed by Chlorophyta) were the dominant algal division at all the sites, it is understandable that it should be dominant when the algal data were combined (**Table 4.7**). The filamentous and colonial algae, with few exceptions, were mostly members of the Chlorophyta division, as seen in the median periphytic algal concentration:biovolume (c:bv) ratio, where the

Chlorophyta had a ratio of 0.526 and the Bacillariophyta a c:bv ratio of 2.046, thus explaining the small difference between the biovolume of the Bacillariophyta and Chlorophyta. The Euglenophyta had a low-median c:bv ratio of 0.056 and the Cyanophyta a high 2.499 ratio, even though both had very low cell concentrations. This indicates that the Euglenophyta cells are much bigger, compared to those of the Cyanophyta.

When combined, the physical factors cancelled one another out and showed no significant linear correlation with the periphytic algal concentration, biovolume or pchl-*a* concentration (**Table 4.8; Fig. D.10, D.11 & D.12**). As mentioned, the nutrient concentrations differed considerably between the sites, as did the periphytic algal concentration, biovolume or pchl-*a* concentrations. The nutrient concentrations had weak to average correlations with the periphytic algal concentration, and little to no correlation with the periphytic biovolume, but high moderate correlations with the pchl-*a* concentrations. This concurs with the finding that intracellular chlorophyll can fluctuate with the amount of nutrients available (Von Schiller *et al.*, 2007), even if algal cell numbers do not reflect it. This makes pchl-*a* concentration possibly the best indicator to be used as a measure of periphyton abundance reflecting the trophic state of a river (Wellnitz & Poff, 2006).

CHAPTER 5

SEASONAL TRENDS AND INFLUENCES

5.1 INTRODUCTION

Seasonal changes are an important aspect of surface water and its quality (Ouyang *et al.*, 2006), of which temperature (Kishi *et al.*, 2005) and South Africa's very high seasonal rainfall (Tyson, 1986) are some of the driving forces. These not only influence the seasonality of periphyton composition, but also the biomass and diversity (Vis *et al.*, 1998; Hillebrand & Sommer, 2000).

This chapter discusses the effect of seasons on periphyton and seasonal drivers. The seasons were divided according to the cycles observed in the atmospheric and water temperature data in summer (Nov & Jan), autumn (Mar), winter (May & Jul) and spring (Sep). The gap (missing data covered by grey area) in the graph lines from May 2005 to January 2006 is due to a six-month break in sampling. The other gaps (missing data) in the graph lines indicate when sampling was not possible due to very high or low flow conditions.

As the spring and autumn data were few (about four each per site), the multiple regressions and PCA data were only done on the summer and winter data.

5.2 RESULTS

The atmospheric temperatures showed the classical seasonal pattern in the average daily maximum and minimum data (SAWS, n.d.(b); **Fig. 5.1**), while the monthly rainfall data (SAWS, n.d.(c); **Fig. 5.2**) showed a definite seasonal pattern of a rainy season from mid-summer to autumn and little to no rain during the winter months. A higher rainfall pattern was found during the 2006 and 2007

period, compared to the years before. The monthly discharge (DWA, n.d.; **Fig. 5.3**) followed the rainfall pattern for the months with high to very high rainfall.

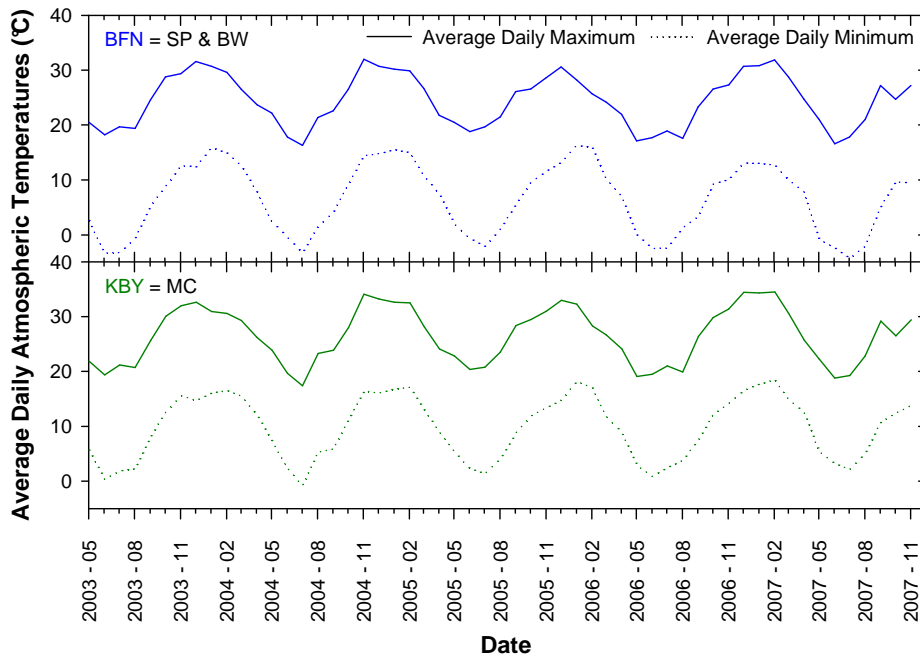


Figure 5.1: Atmospheric temperature data of Bloemfontein (BFN – 0261516B0) and Kimberley (KBY – 0290468A9) over the period from May 2003 to November 2007 (SAWS, n.d.(b)).

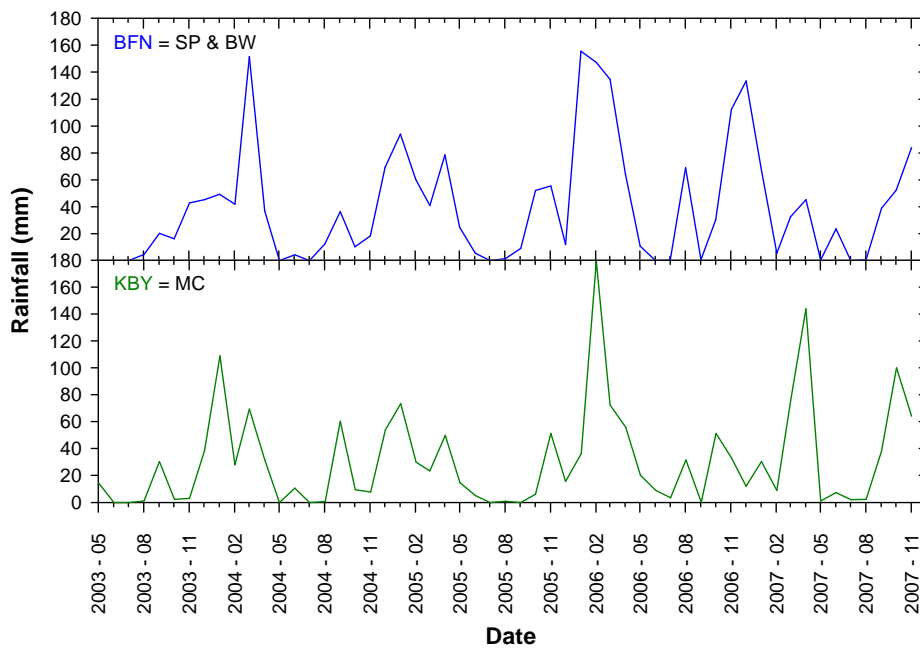


Figure 5.2: Rainfall data of Bloemfontein (BFN) and Kimberley (KBY) over the period from May 2003 to November 2007 (SAWS, n.d.(c)).

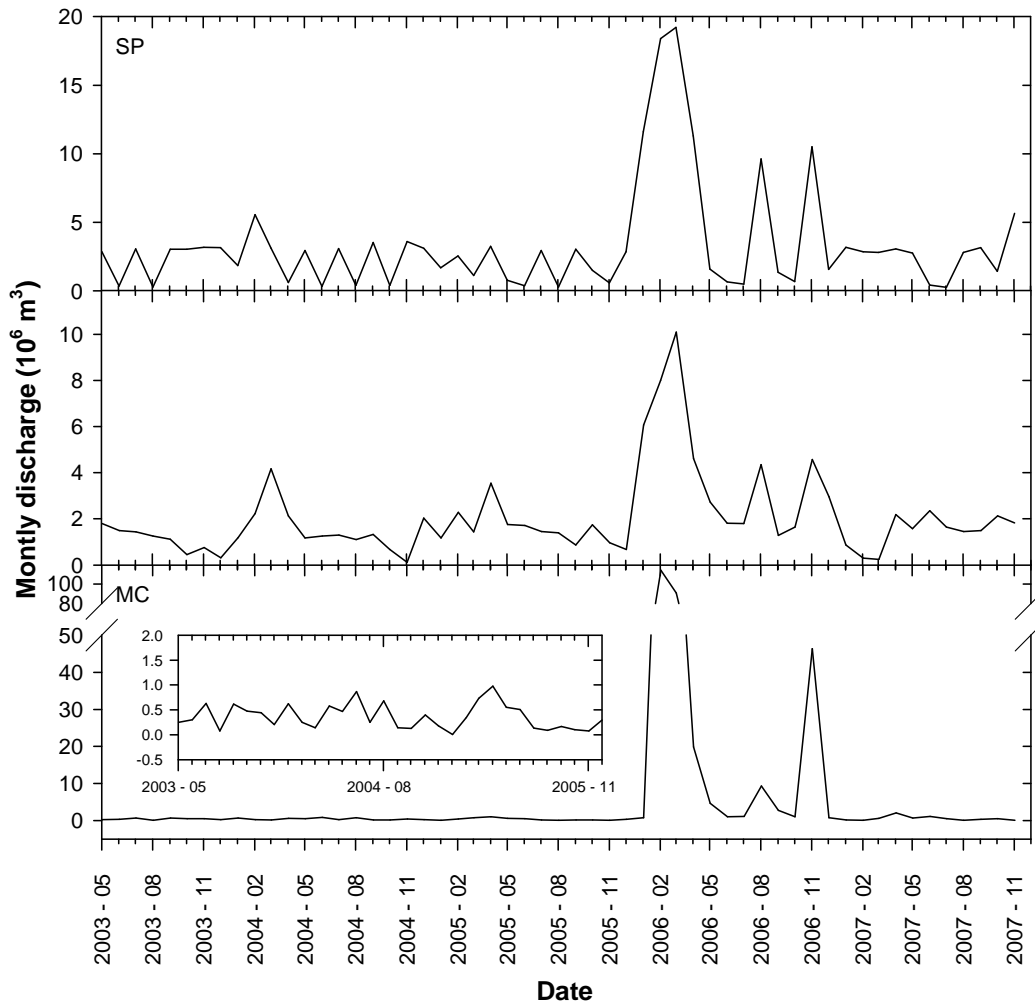


Figure 5.3: Monthly discharge (10^6 m^3) at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The small insert graph on MC is for the period May 2003 to November 2005 to overcome the dwarfing effect of February 2006 (DWA, n.d.).

Additional processed data tables are in **Appendix D.2**. Tables and Figures in **Appendix D** are numbered as, e.g. **Fig. D.13**.

5.2.1 PHYSICAL AND CHEMICAL

The water temperature (**Fig. 5.4**) at all three the study sites showed the same seasonal pattern the atmospheric temperatures had, with high summer values and low winter values – with the spring and autumn values mostly between them.

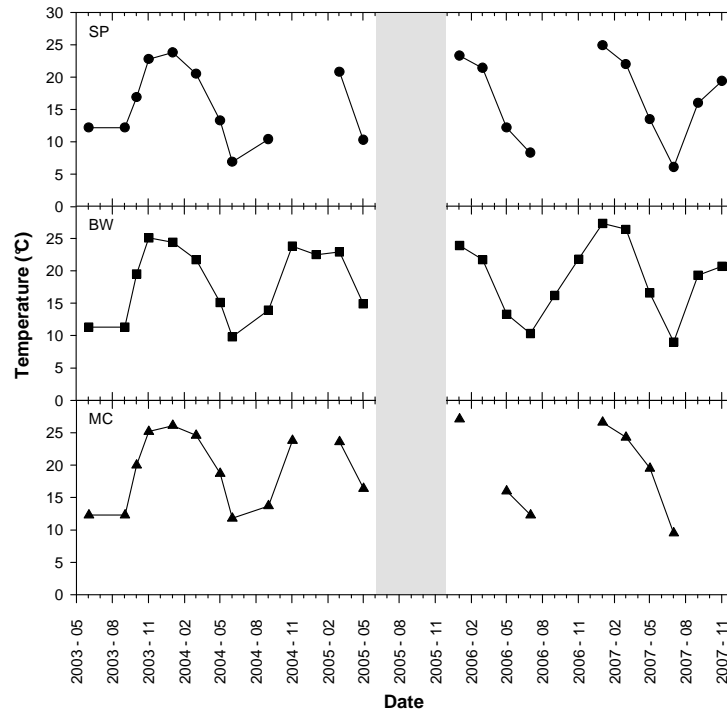


Figure 5.4: Water temperature (°C) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

The dissolved oxygen concentrations (**Fig. 5.5**) are the inverse of those of the temperature; low in the summer months and higher in the winter months. A similar seasonal pattern to that of the diss. O₂ was repeated with the pH (**Fig. 5.6**) at all three sites, although the Modder above Confluence indicated weak signs of seasonality.

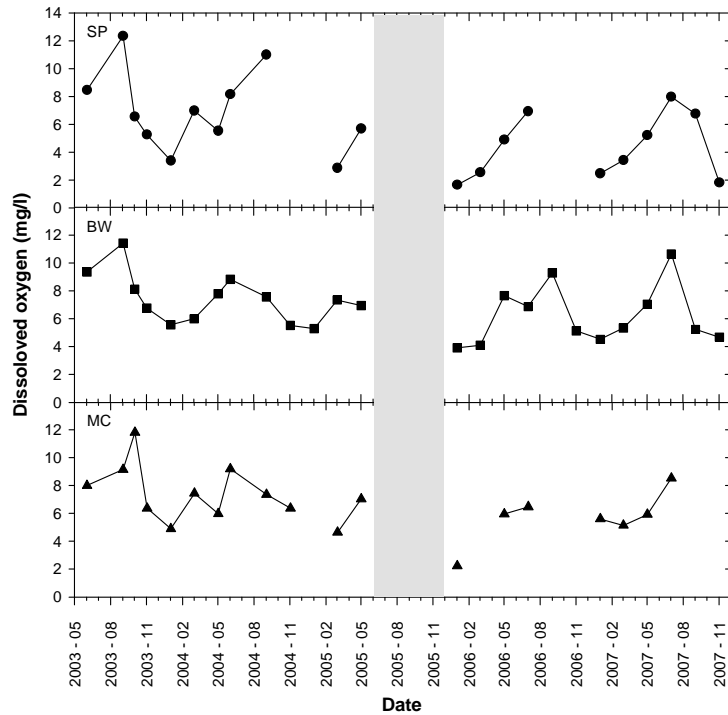


Figure 5.5: Dissolved oxygen concentration (mg/l) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

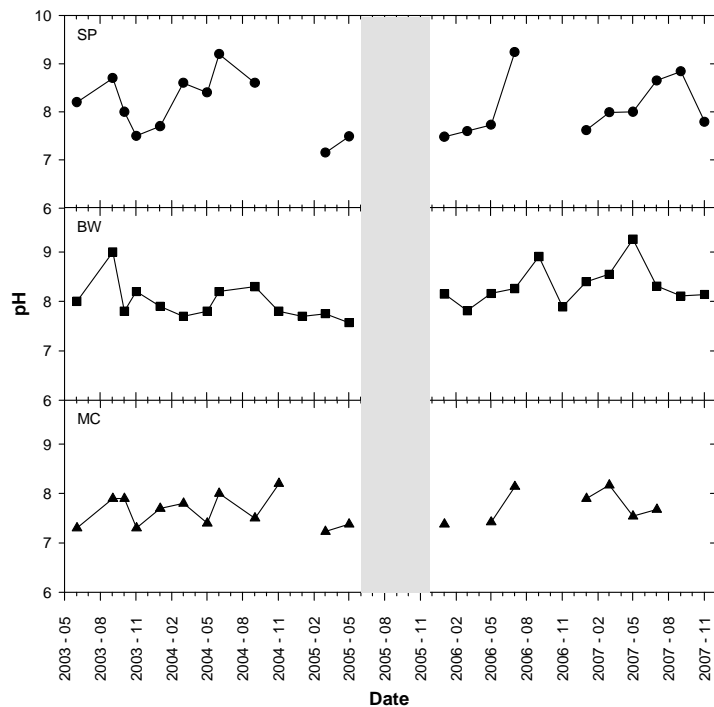


Figure 5.6: pH at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

The flow pattern (**Fig. 5.7**) at the three sites followed the same general pattern as those of the rainfall and the monthly discharge, with increases in flow during the summer “rainy” months, which are reflected by the turbidity (**Fig. 5.8**) pattern.

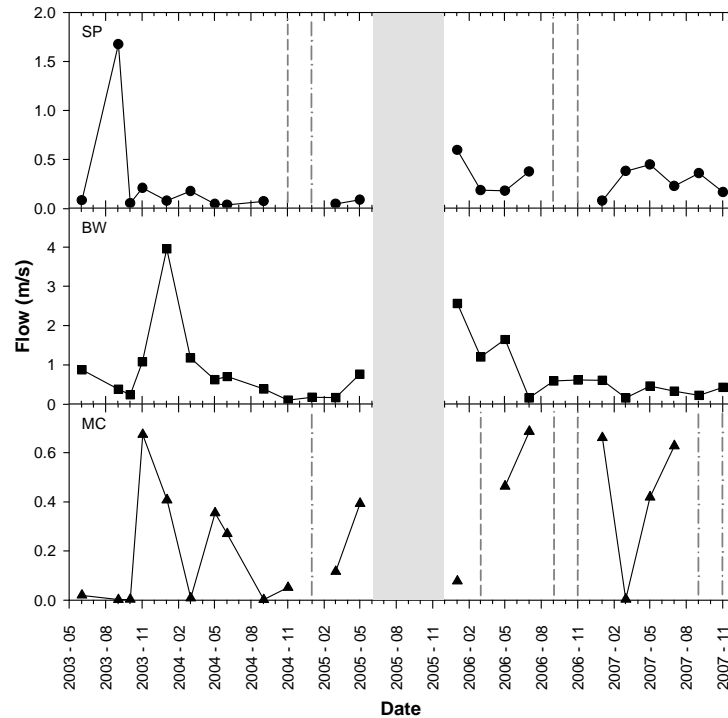


Figure 5.7: Flow (m/s) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period, the dashes (_ _ _) are sampling days with very high flow or floods, and dots and dashes (_ . _ . _) the very low flow or dry sampling days.

The phytoplanktonic chlorophyll-a (chl-a, **Fig. 5.9**) concentration at all the sites did not have a definite seasonal pattern, but were influenced by the nutrients and TDS concentrations. The chl-a concentration has a partial influence on the oxygen concentrations, as some of their fluxes correspond.

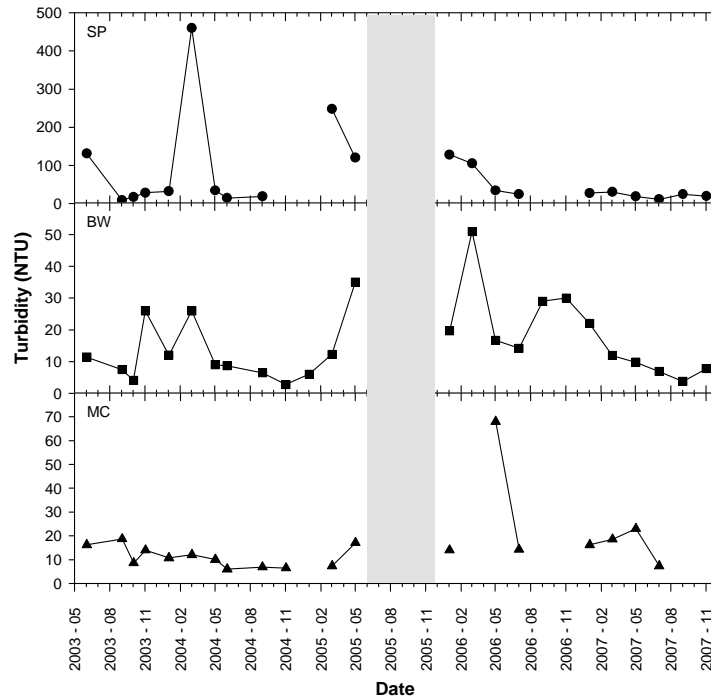


Figure 5.8: Turbidity (NTU) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

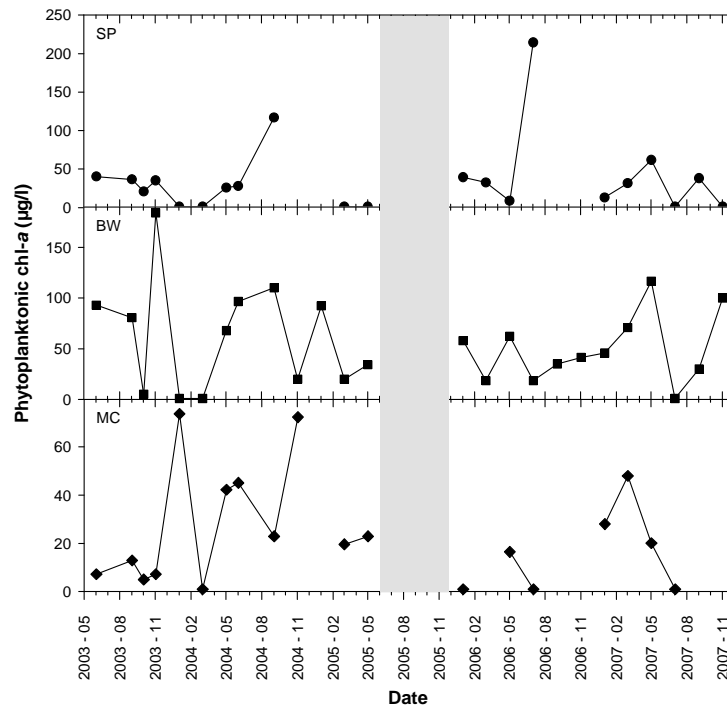


Figure 5.9: Phytoplanktonic chlorophyll-a (µg/l) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

The periphytic chlorophyll-*a* (pchl-*a*; **Fig. 5.10**) concentrations at all the sites have the same general pattern, although it was on different scales. The pchl-*a* concentration was the highest during the spring seasons and lower during summer to winter.

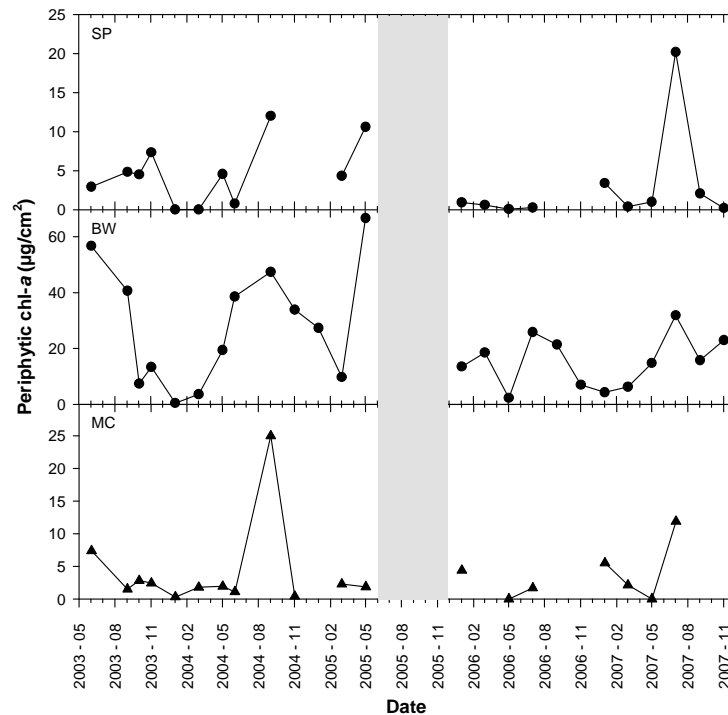


Figure 5.10: Periphytic chlorophyll-*a* ($\mu\text{g}/\text{cm}^2$) at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

5.2.2 NUTRIENTS (DIP & DIN) AND TOTAL DISSOLVED SALTS (TDS)

The DIP concentrations (**Fig. 5.11**) at sites SP and BW showed higher concentrations during the summers than during the winters for the 2003–2005 period and lower concentrations during the summers than during the winters for the 2006–2007 periods. The DIP concentration at MC showed an order of less magnitude, but had an inverse pattern to those of the other two sites during the different periods.

The seasonal pattern of the DIN concentration (**Fig. 5.12**) was the most distinct at BW, where it was high during the winter and low during the summer. The pattern at MC was less distinct, but did concur with that of BW. The DIN concentration pattern at SP was a little different. From 2003–2005 it was high during the summer, but during 2006–2007 it was high during the winter, as was the case at the other sites.

Sites SP and BW had clear seasonal patterns with regard to the TDS concentration (**Fig. 5.13**). For the period 2003–2005, the concentrations were higher during the summer and for the period 2006–2007, the inverse was true. The TDS concentration at MC displayed the same pattern, even if it was not as clearly defined.

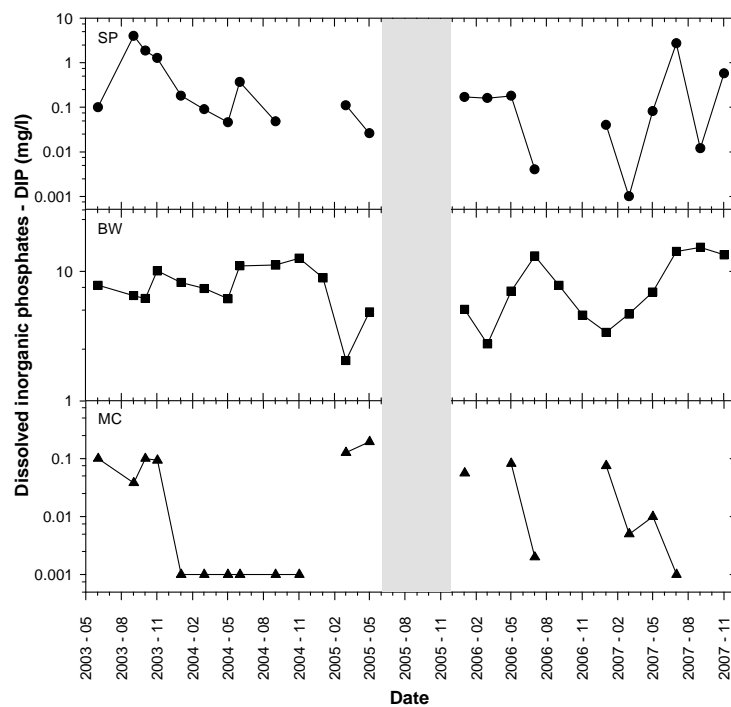


Figure 5.11: Dissolved inorganic phosphorus (DIP – mg/l) at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

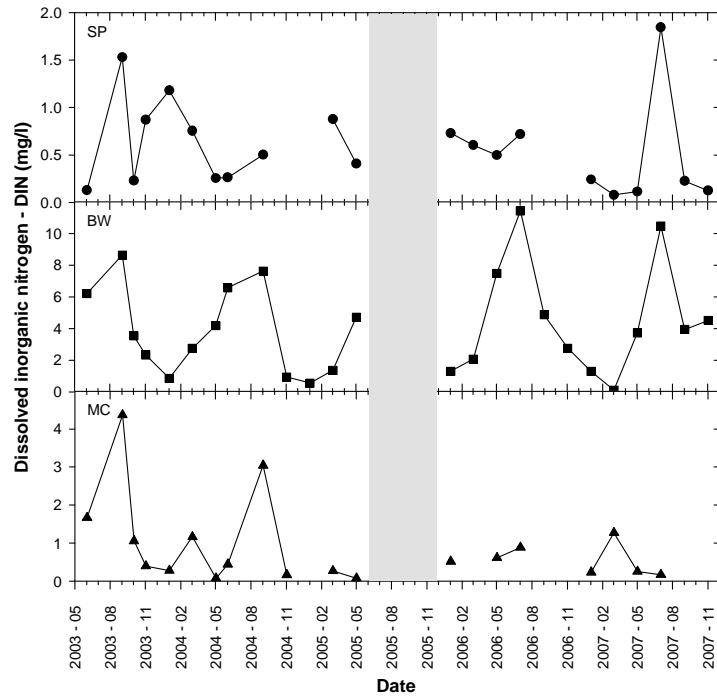


Figure 5.12: Dissolved inorganic nitrogen (DIN – mg/l) at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

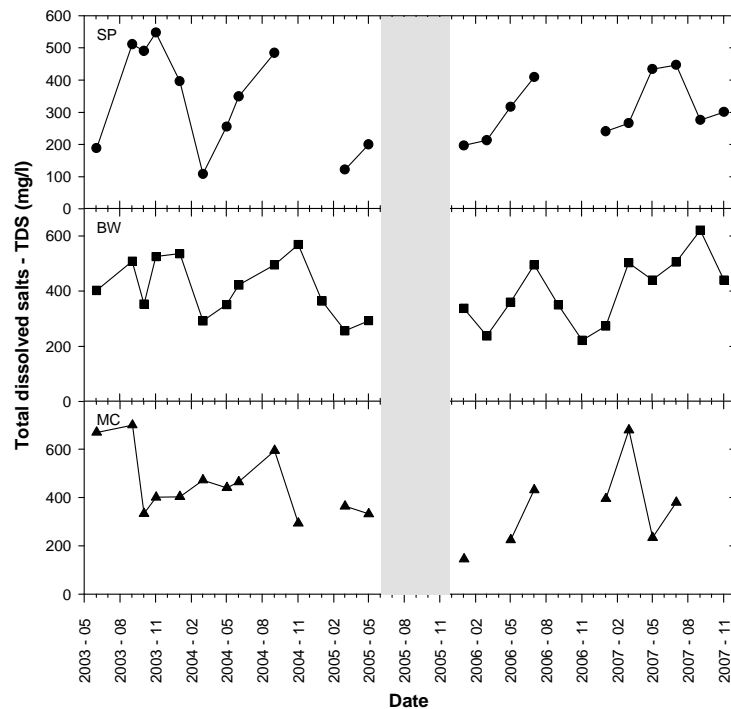


Figure 5.13: Total dissolved salts (TDS – mg/l) at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

5.2.3 BIOLOGICAL

The periphytic algal concentration (**Fig. 5.14**) and the periphytic algal biovolume (**Fig. 5.15**) displayed the same pattern, although the intensity of the peaks differs. The periphyton concentration and biovolume patterns also compared well with those of the pchl-a concentration. Seasonal patterns were also found in the periphytic algal assemblages (**Fig. 5.16 & 5.17**). The Bacillariophyta were mostly dominant during spring, and the Chlorophyta and the Cyanophyta showed an increase during the summer. Although the Euglenophytes displayed low concentrations throughout the study period, it did dominate the periphytic algal biovolume at times.

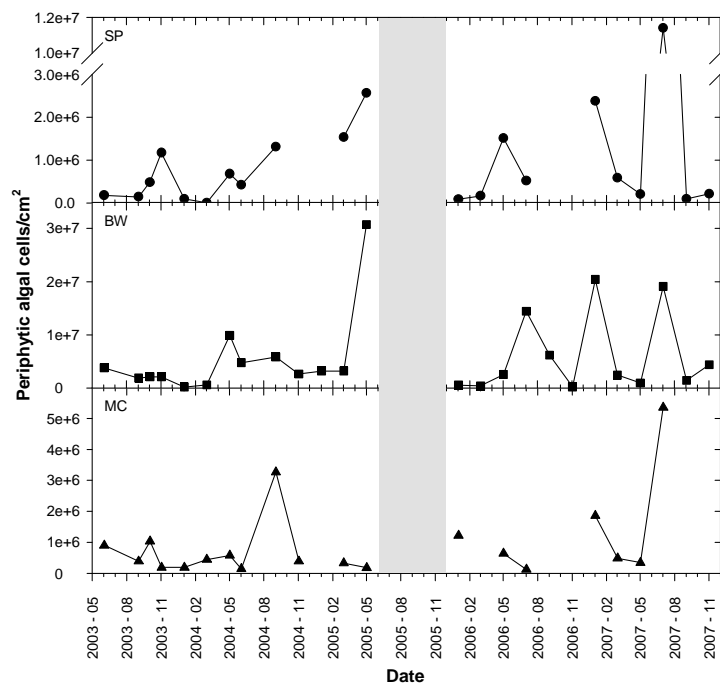


Figure 5.14: Total periphytic algal (cells/cm²) concentration at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

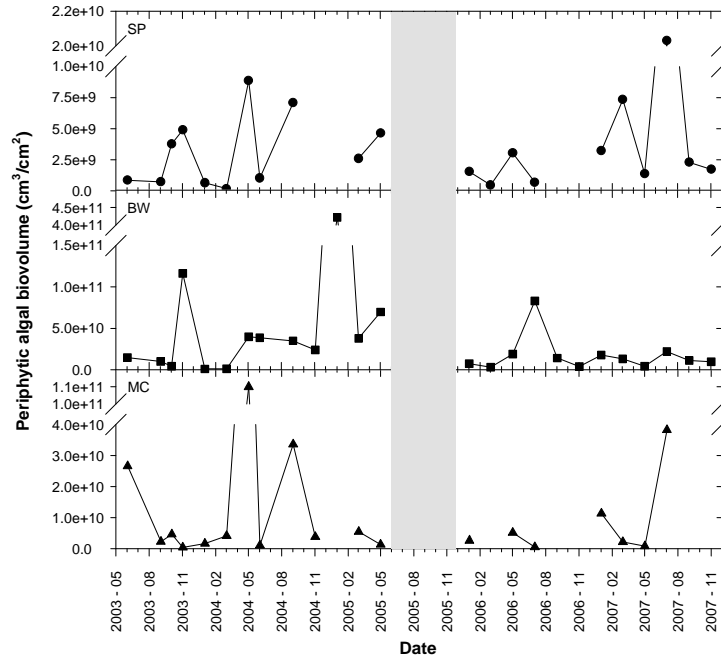


Figure 5.15: Total periphytic algal ($\mu\text{m}^3/\text{cm}^2$) biovolume at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

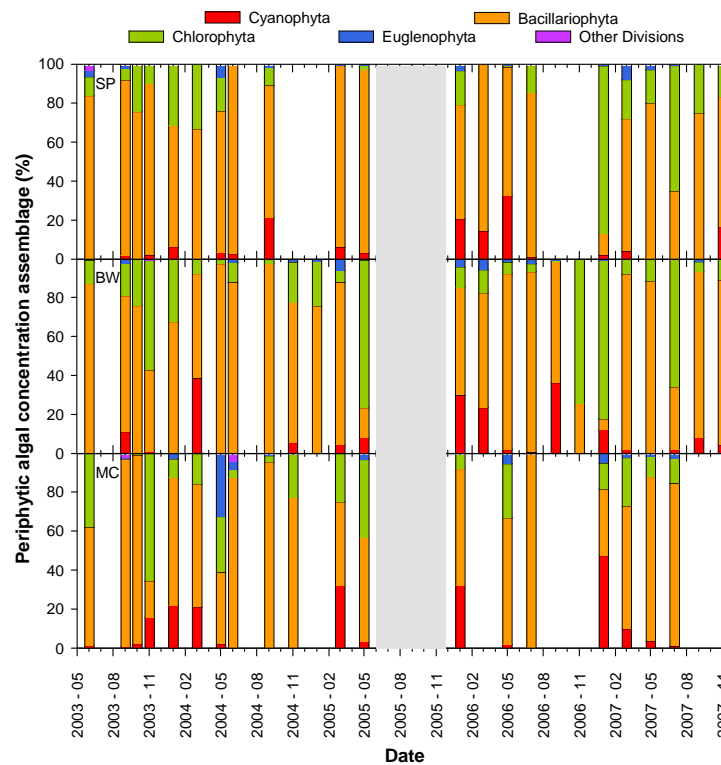


Figure 5.16: Stacked vertical bars displaying the periphytic algal division assemblage as cell concentrations for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

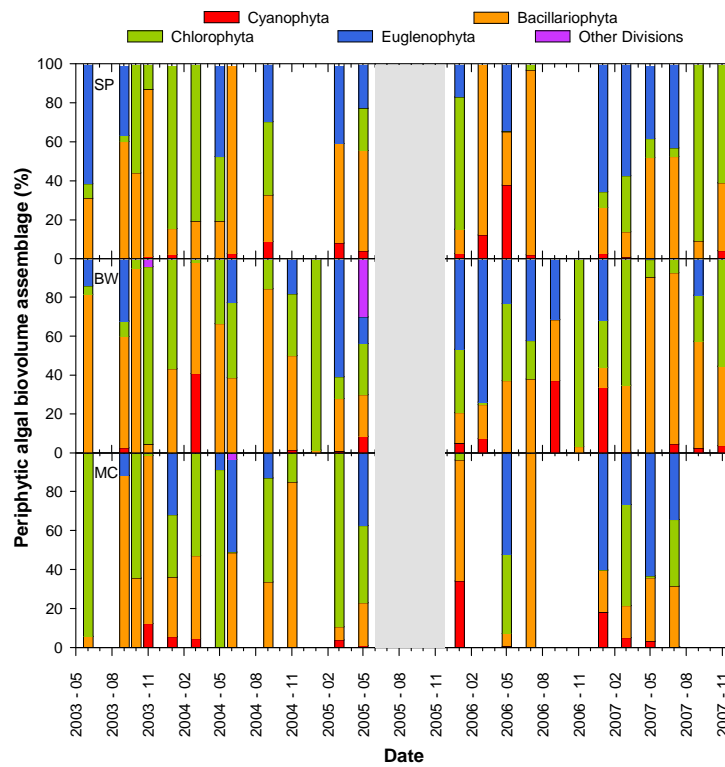


Figure 5.17: Stacked vertical bars displaying the periphytic algal division assemblage as cell biovolume for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

Both the periphyton concentration and biovolume displayed inverse patterns to those of the total SASS5 (**Fig. 5.18**) and total ASPT (**Fig. 5.19**) scores, as well as to the stones in current (SIC) SASS5 (**Fig. 5.20**) and SIC ASPT (**Fig. 5.21**) scores.

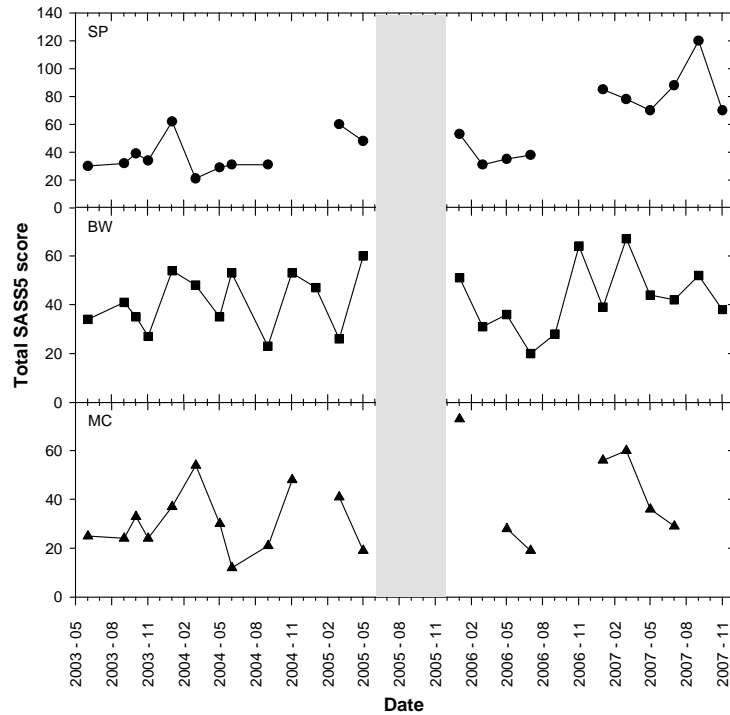


Figure 5.18: Total SASS5 scores at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

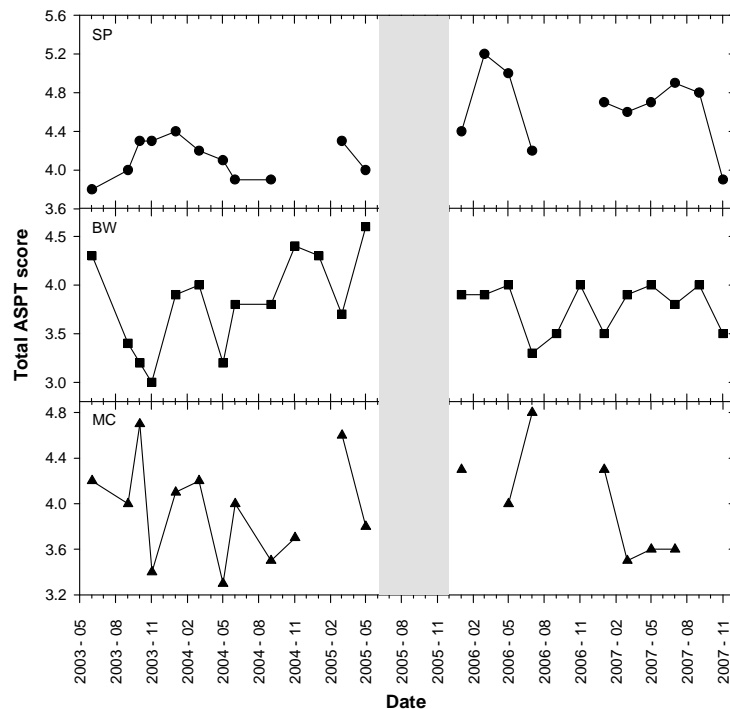


Figure 5.19: Total ASPT scores at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

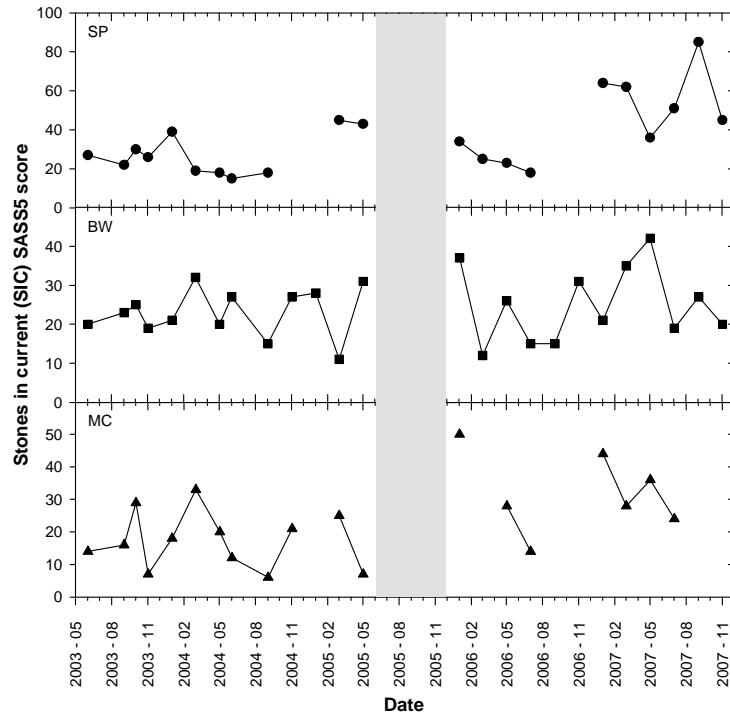


Figure 5.20: Stones in current (SIC) SASS5 scores at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

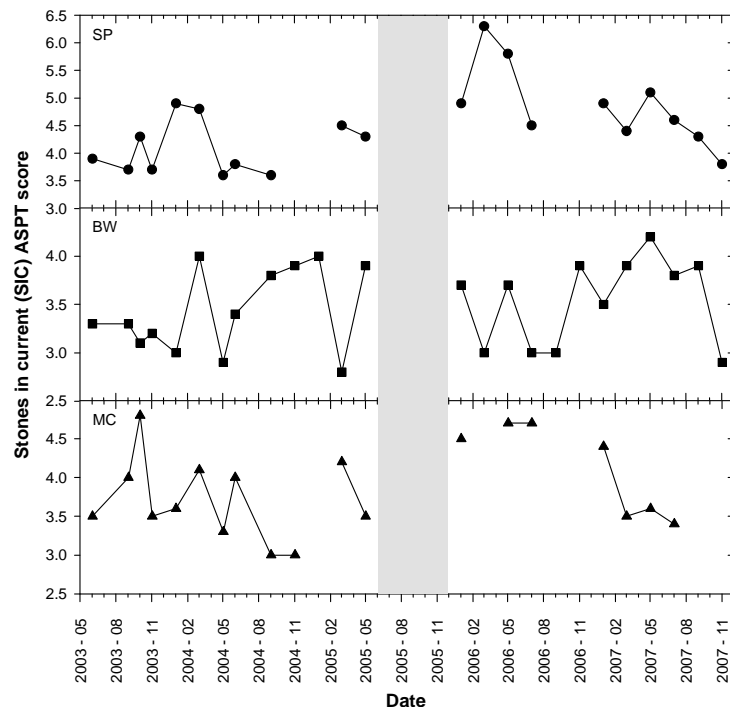


Figure 5.21: Stones in current ASPT scores at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The grey area was the six-month non-sampling period.

5.2.4 MULTIVARIATE RELATIONSHIPS

The combined data of the sites for the summer (SP, BW and MC; $n = 18$; **Fig. 5.22a**) showed no linear correlation or significance ($r^2 = 6.59 \times 10^{-3}$; $p = >0.1$) between the total periphytic algal concentration and biovolume, but during the winter ($n = 24$) there was an average correlation with statistical significance ($r^2 = 0.262$; $p = <0.02$). The pchl-*a* concentration and biovolume displayed an average correlation ($r^2 = 0.284$; $p = <0.02$; **Fig. 5.22b**), but with statistical significance during the summer, and a weak, insignificant correlation during the winter ($r^2 = 0.171$; $p = <0.1$). No significant linear correlation ($r^2 = 0.014$; $p = >0.1$; **Fig. 5.22c**) was detected between the periphytic algal concentration and the pchl-*a* concentration during the summer, while a strong, statistically highly significant correlation was detected during the winter ($r^2 = 0.595$; $p = <0.001$).

The Bacillariophyta was the dominant division found with the combined periphytic data with the periphytic algal concentration during both the summer and winter periods, followed by the Chlorophyta with the periphytic algal concentration during both the summer and the winter (**Table 5.1**). The Chlorophyta displayed the highest percentage periphytic algal biovolume during the summer. The Cyanophyta and the Euglenophyta were not so significant. The mean and median percentages between the division's biovolumes are closer than the concentration. The overall median Bacillariophyta and Euglenophyta increased from the summer to the winter season, while the other divisions decreased.

The total pchl-*a* concentration had a strong linear and a statistically highly significant correlation with the Bacillariophyta concentration ($r^2 = 0.648$; $p = <0.001$) during the summer. The Bacillariophyta biovolume had strong and moderate, statistically highly significant correlations ($r^2 = 0.733$; $p = <0.001$ and $r^2 = 0.418$; $p = <0.01$) with the total pchl-*a* concentration during the summer and winter seasons, respectively. The total pchl-*a* concentration displayed moderate, statistically significant to statistically highly significant correlations with the Chlorophyta algal concentration ($r^2 = 0.460$; $p = <0.001$), the Cyanophyta algal

concentration ($r^2 = 0.375$; $p = <0.01$) and biovolume ($r^2 = 0.376$; $p = <0.01$) during the winter season.

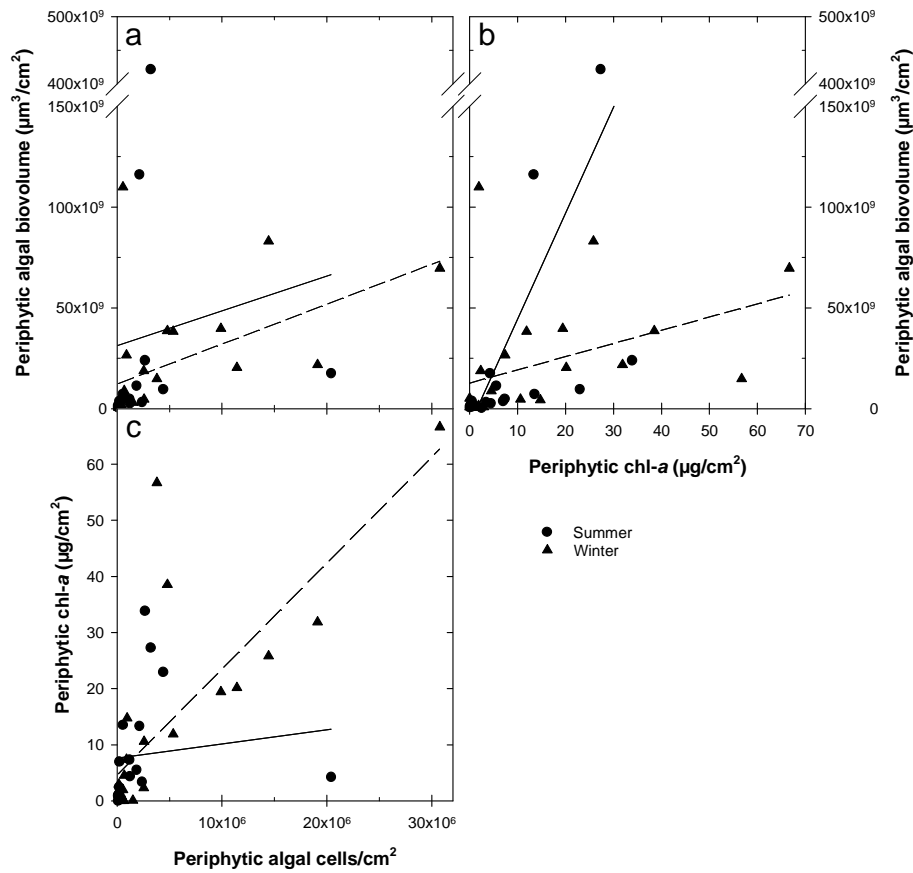


Figure 5.22: The linear relationships between the combined periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) for the summer (—) and winter (- - -) seasons.

Table 5.1: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found with the combined data for all sites during the summer and winter seasons, percentage of the periphytic algal concentration and biovolume.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Summer (n = 18)	Min	0.00	0.00	5.26	1.48	8.00	0.37	0.00	0.00
	Max	47.37	34.15	88.00	86.24	85.71	98.44	5.26	65.63
	Mean	12.10	7.03	53.92	34.61	32.73	43.02	1.19	15.10
	Median	5.89	2.70	61.25	27.09	21.73	32.25	0.00	0.00
	SD	13.73	10.84	25.21	28.75	26.98	34.26	1.71	22.58
Winter (n = 24)	Min	0.00	0.00	15.24	0.37	0.00	0.00	0.00	0.00
	Max	32.56	37.82	100.00	100.00	75.56	94.34	32.58	63.30
	Mean	2.90	2.77	73.92	47.15	19.78	23.31	3.07	25.37
	Median	1.21	0.29	84.07	38.20	11.51	14.64	1.29	23.04
	SD	6.59	7.75	23.64	30.76	21.91	26.11	6.56	21.43

The physical and chemical factors had very weak, insignificant correlations with periphytic algal concentration (**Table 5.2; Fig. D.13**) during the winter season. The DIP and DIN concentrations had moderate to strong, statistically significant correlations. The physical and chemical factors (**Table 5.2; Fig. D.14**) showed no clear or significant correlations with the periphytic biovolume range for the summer or winter seasons. The data (**Table 5.2; Fig. D.15**) showed that the physical and chemical factors displayed negligibly insignificant correlations with the pchl-a concentration. The DIP concentration displayed very strong (statistically highly significant) correlations to the pchl-a concentration during the summer season. During the winter season, the DIP and DIN concentrations displayed moderate to strong, statistically significant correlations to the pchl-a concentrations.

Table 5.2: The coefficient of determination (r^2) and probability (p) values for the combined data at all sites, for the summer and winter seasons, between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration.

	Algal conc. (cells/cm ²)		Biovolume (µm ³ /cm ²)		Pchl-a (µg/cm ²)		
	r^2	p	r^2	p	r^2	p	
Summer (n = 18)	Temperature	0.114	>0.1	(-) 0.018	>0.1	(-) 0.062	>0.1
	Diss. oxygen	2.14x10 ⁻³	>0.1	0.050	>0.1	0.086	>0.1
	Redox	(-) 0.291	<0.02	(-) 1.39x10 ⁻³	>0.1	(-) 0.071	>0.1
	pH	0.261	<0.05	1.97x10 ⁻⁴	>0.1	0.035	>0.1
	Turbidity	(-) 9.92x10 ⁻³	>0.1	(-) 0.027	>0.1	(-) 0.098	>0.1
	Flow	(-) 7.56x10 ⁻³	>0.1	(-) 0.012	>0.1	(-) 0.011	>0.1
	DIP	0.025	>0.1	0.144	>0.1	0.680	<0.001
	DIN	0.029	>0.1	(-) 0.83x10 ⁻³	>0.1	0.165	<0.1
	TDS	(-) 0.011	>0.1	9.28x10 ⁻³	>0.1	0.167	<0.1
Winter (n = 24)	Temperature	(-) 0.027	>0.1	0.026	>0.1	(-) 0.025	>0.1
	Diss. oxygen	0.081	>0.1	0.59x10 ⁻³	>0.1	0.181	<0.05
	Redox	0.043	>0.1	0.096	>0.1	0.013	>0.1
	pH	(-) 2.76x10 ⁻³	>0.1	(-) 0.075	>0.1	(-) 0.78x10 ⁻³	>0.1
	Turbidity	(-) 0.018	>0.1	(-) 0.063	>0.1	(-) 0.026	>0.1
	Flow	0.022	>0.1	0.011	>0.1	0.084	>0.1
	DIP	0.331	<0.01	0.134	<0.1	0.420	<0.01
	DIN	0.363	<0.01	0.160	<0.1	0.388	<0.01
TDS	0.011	>0.1	0.079	>0.1	0.016	>0.1	

(-) = indicates a negative correlation.

5.3 DISCUSSION

The seasonal patterns found in the atmospheric temperature (**Fig. 5.1**) and rainfall data (**Fig. 5.2**) comply with those of the climatic region (Tyson, 1986). The seasonal pattern of the discharge (**Fig. 5.3**) was a result of the rainfall patterns.

5.3.1 PHYSICAL AND CHEMICAL

The seasonal pattern found in the water temperature (**Fig. 5.4**) was a reflection of the atmospheric temperature. This unwavering pattern shows that no external sources influence the natural seasonal pattern of the water temperature of high in summer and low in winter (Macan, 1958). The solubility of O₂ in water is influenced by water temperature (Giller & Malmqvist, 1998; Allan & Castillo, 2007); thus, as O₂ dissolves more easily in cold water, the seasonal pattern (**Fig. 5.5**) was inverse to that of the water temperature – higher in winter and lower in summer. Oxygen saturation can be as much as 50% less during the spring and summer, as temperatures increase (Horne & Goldman, 1994). At SP, BW and MC the oxygen concentration was respectively 60%, 40% and 30% less during the summer than the winter, while the percentage saturation was respectively 45%, 20% and 10% less during the summer than the winter.

The seasonal pattern of pH at BW and SP, and weak seasonality at MC (**Fig. 5.6**) could partly be a result of the soils surrounding the sites. MC and BW have a higher base structure than that at SP (AGIS, n.d.). The productivity of periphyton and phytoplankton also has an influence on the pH through photosynthesis, where CO₂ are used and O₂ produced. The higher median of periphyton and phytoplankton algal components during the winter at SP and BW probably assisted with the seasonality of the pH (and diss. oxygen). At MC, the periphyton and phytoplankton algal components (except periphyton algal biovolume) were higher during the summer and probably had some influence on the seasonality of the pH, even though it was weak.

The flow (**Fig. 5.7**) at the sites had a seasonal pattern that corresponded to the rainfall and the discharge data, namely high in summer and low in winter. This seasonal pattern influenced that of the turbidity, which showed a correlation to the seasonal pattern of the flow. The faster the river flow, the higher the turbidity or sediment loads (Ryan, 1991). As the flow increases in summer, the suspended particles are suspended and transported and increases the turbidity (Closs *et al.* 2004). With rain, an increase of particles get washed into a river, and together with the extra erosion from the banks the turbidity increases further (Wetzel, 2001; Jakob *et al.*, 2003). Elevated turbidity in the summer can also be part of the reason the temperature is higher in summer, due to absorbance and scattering of light/heat (Paaijmans *et al.*, 2008). However, too much turbidity can also reflect heat (Ryan, 1991).

Phytoplanktonic algae in rivers mostly have their origins from slow-flowing parts of the river and pools, and do not react in the same way in flowing water as they would in standing water. Seasonality has a bigger influence on the growth of phytoplankton in standing water than in flowing water (Wetzel, 2001; Hart, 2006) and thus on the chlorophyll-*a* concentration (**Fig. 5.9**). Periphyton, on the other hand, is influenced by the factors of flowing water and is reflected in the pchl-*a* concentration (**Fig. 5.10**). Thus, the pchl-*a* concentration shows an inverse pattern to those of the atmospheric and water temperatures, rainfall and flow. The same seasonal pattern corresponds with that of oxygen as well as a combination of the nutrient patterns. The above-mentioned has an impact on either the photosynthesis rate or the productivity, or both (Ryan, 1991).

5.3.2 NUTRIENTS (DIP & DIN) AND TOTAL DISSOLVED SALTS (TDS)

The DIP concentrations corresponded with the periphyton and phytoplankton growth patterns. The DIP concentrations were usually higher at SP and BW during the winter and at MC during summer (**Fig. 5.11**). This might have caused the decrease in the DIP concentration in the mentioned season. During 2006–2007, the rainfall was higher and diluted some of the phosphates in the water,

while washing phosphates into the system during the winter, especially at SP and BW.

The higher flow during the summer months diluted the DIN concentrations in the river (Taylor *et al.*, 2007b) and the high chl-*a* and pchl-*a* concentrations extracted further nitrogen from the water (**Fig. 5.12**). During spring and summer, the increased uptake of nutrients by periphyton can be the cause of lower nutrients (Matheson *et al.*, 2012).

The higher TDS concentrations in the summer during 2003–2005 were a combined result of lack of rainfall and high evaporation during the sampling days (**Fig. 5.13**). During 2006–2007, the summer periods were wetter and the rainfall, runoff and discharge had a diluting effect on the concentration of the TDS (DWAF, 1996a).

5.3.3 BIOLOGICAL

The similar pattern in the periphytic algal concentration and the biovolume is a result of the same algal assemblage that is supporting them (**Fig. 5.14 & 5.15**). These patterns were driven by a combination of flow, turbidity and DIP concentrations. The influence of the different factor changes from season to season (Hillebrand & Sommer, 2000), and is reflected in the variability of the periphyton concentrations and biovolumes. Bacillariophyta usually dominates the algal community in spring, while Chlorophyta and Cyanophyta prefer the warmer late summers and even sometimes autumn (Biggs, 1996; Vis *et al.*, 1998; Flynn *et al.*, 2002). The Euglenophyta cells are big; therefore, a slight increase in concentration can cause a large increase in the biovolume (**Fig. 5.16 & 5.17**).

The periphytic algal concentration and biovolume are higher in winter with the lower flow; therefore, no algae are washed away, causing the filamentous algae biovolume to increase and even overgrow the habitat of the macroinvertebrates (**Fig. 5.18, 5.19, 5.20 & 5.21**).

5.3.4 MULTIVARIATE RELATIONSHIPS

The correlations and probabilities between the total periphytic algal biovolume ($\mu\text{m}^3/\text{cm}^2$) and those of the total periphytic algal concentrations (cells/cm^2) and periphytic chlorophyll-*a* concentrations (pchl-*a*; $\mu\text{g}/\text{cm}^2$) showed no relation of significance during the summer, but did so during the winter (**Fig. 5.22**). This is most probably a result of flow. The higher flow events (and rainfall) during the summer tend to dislodge periphyton from their substrate (Biggs & Thomsen, 1995; Mosisch & Bunn, 1997) and break filamentous algae (Biggs *et al.*, 1998a). The periphytic algal assemblages at all three sites consisted of more genera in winter than in summer. The average (statistically significant) and weak (insignificant) correlations between total periphytic biovolume and pchl-*a* concentrations during the summer and winter, respectively, and the non-existing (insignificant) and strong (statistically highly significant) correlations between total periphytic algal and pchl-*a* concentrations during the summer and winter, respectively, were a result of the division of algae present during a specific season. The temperature influences the composition of the periphyton community as more green algae occur during the summer months (Biggs, 1996; Vis *et al.*, 1998; Flynn *et al.*, 2002).

The combined data of all the sites showed dominance by Bacillariophyta during both the summer and winter seasons for the periphytic algal concentration and biovolume (winter) followed by the Chlorophyta (**Table 5.1**). The increase of Chlorophyta and Cyanophyta concentrations and biovolumes during the summer correspond to the increase in temperature and their affinity to warmer water (Biggs, 1996; Vis *et al.*, 1998; Flynn *et al.*, 2002), in spite of the higher flow. As Bacillariophyta is the dominant division, it is reflected in the strong correlation and high statistical significance of the concentration and biovolume with the pchl-*a* concentration.

The weak (insignificant) correlations between the physical and chemical factors with the periphytic algal concentration and biovolume, as well as pchl-*a* concentration (**Table 5.2; Fig. D.13, D.14 & D.15**) could be that so many factors

interact simultaneously with the algal components that their influence and effects are masked (Figueroa-Nieves *et al.*, 2006). In spite of all the interactions with the algal components, the nutrients had the best correlations with them. Nutrient availability is essential to the growth of periphyton in lotic ecosystems (Von Schiller *et al.*, 2007) as it is in all systems. The DIN concentrations correlated best with the periphytic algal concentrations and the DIP concentrations with the biovolume.

CHAPTER 6

IMPORTANCE OF HYDROLOGY

6.1 INTRODUCTION

The change in climatic factors may alter the transport of matter through runoff; thus, the occurrence of extreme events (droughts and floods) alters the quality of water from the norm (Delpla *et al.*, 2011). Changes in climate can also result in prolonged dry or wet periods, which can last several years (Tyson, 1986). Different flow regimes support different periphytic algal assemblages (Asaeda & Hong Son, 2000).

The sampling period of June 2003 to November 2007 was divided into two separate periods: June 2003 to May 2005 (dry) and January 2006 to November 2007 (wet). **Table 6.1** shows the number of samples in each period. This was done according to site and flow observation. The terms “dry” and “wet” are used to distinguish between the periods, as the first period was drier than the latter.

Table 6.1: Number of samples in each period (2003–2005 & 2006–2007).

Sites	Number of samples	
	Dry period (2003–2005)	Wet period (2006–2007)
Sannaspos	11	10
Bishop's Weir	13	12
Modder above Confluence	12	7

6.2 RESULTS

The monthly rainfall data (SAWS, n.d.(c); **Fig. 6.1**) shows that the mean and median of 2006–2007 (wet) were higher (even if slightly) than that of 2003–2005

(dry). The rainfall data for Bloemfontein (station no. 0261516B0) and Kimberley (station no. 0290468A9) showed roughly the same pattern (**Fig. 6.1**) with less rain in the winter months and more during the summer months than expected, as the sampling sites fall within the summer rainfall area of South Africa. However, the duration at the Bloemfontein site was longer and the results indicate that the Kimberley site is slightly drier than the Bloemfontein site.

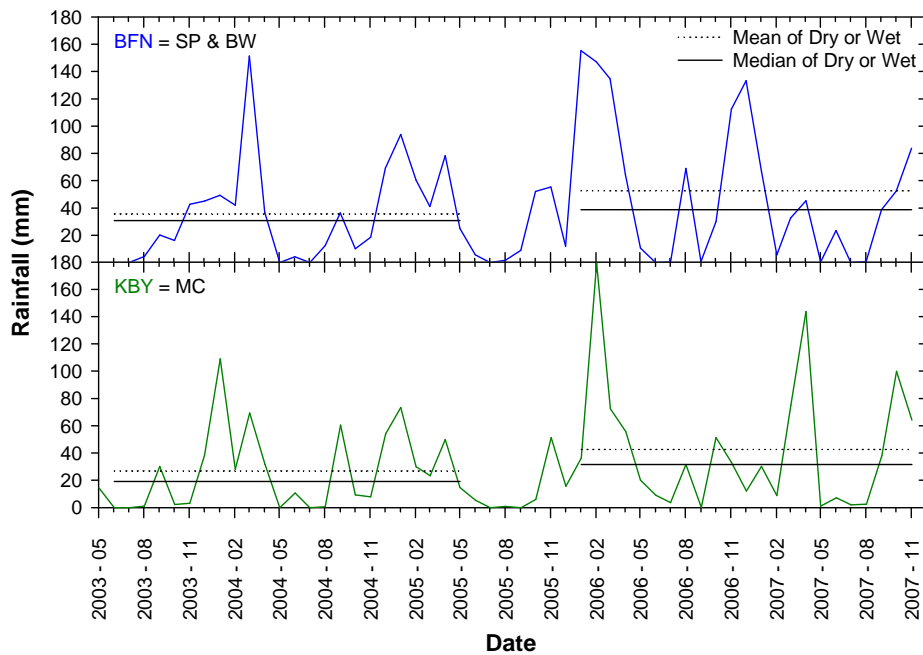


Figure 6.1: Rainfall data of Bloemfontein (BFN) and Kimberley (KBY) over the period from May 2003 to November 2007. The monthly mean (···) and median (—) of each are also showed (SAWS, n.d.(c)).

The monthly median and mean discharges measured at the gauging stations of the three sites were all higher during the wet period, except the median of SP (**Table 6.2**). As expected, the total discharge was higher during 2006–2007 than during 2003–2005. At Sannaspos (SP), the difference in total discharge was $61.3 \times 10^6 \text{ m}^3/\text{s}$; at Bishop's Weir (BW) it was $30.4 \times 10^6 \text{ m}^3/\text{s}$; and at Modder above Confluence (MC) the difference in total discharge was $287.9 \times 10^6 \text{ m}^3/\text{s}$.

Table 6.2: The monthly median and mean, and total discharge ($10^6 \text{ m}^3/\text{s}$) at the three sites (DWA, n.d.).

Sites	Gauging Station no	Year	Monthly median	Monthly mean	Total for period
Sannaspos	C5H003	2003–2005	3.0	2.2	53.9
		2006–2007	2.8	5.0	115.2
Bishop's Weir	C5H054	2003–2005	1.3	1.5	35.7
		2006–2007	1.8	2.9	66.1
Modder above Confluence	C5H035	2003–2005	0.4	0.4	10.1
		2006–2007	1.0	13.0	298.0

The median atmospheric temperatures over the whole study period and the medians of the minimums and maximums of the specific sampling dates (**Table 6.3**) showed that the atmospheric temperatures for Bloemfontein and Kimberley during the drier period (2003–2005) were higher than those of the wetter period (2006–2007). There was a slight difference in the maximum temperatures but a considerable difference in the minimum temperatures (SAWS, n.d.(b)).

Table 6.3: The median atmospheric temperatures ($^{\circ}\text{C}$) of the study sites (SAWS, n.d.(b)).

Weather station no.	Median over sampling period (maximum)		Median of sampling dates			
			Minimum		Maximum	
	2003–2005	2006–2007	2003–2005	2006–2007	2003–2005	2006–2007
0261516B0 (Bloemfontein)	24.95	24.7	10.10	4.05	23.40	23.25
0290468A9 (Kimberley)	26.80	26.60	11.55	8.35	26.00	25.10

Additional processed data tables are in **Appendix D.3**. Tables and Figures in **Appendix D** are numbered as, e.g. **Fig. D.16**.

6.2.1 PHYSICAL AND CHEMICAL

The median and mean water temperature (**Fig. 6.2a**) at SP, BW and MC were higher during the wet period than during the dry period, while the median and mean dissolved oxygen concentration (O_2 ; **Fig. 6.2b**) were lower during the wet period. The median and mean pH (**Fig. 6.2c**) at SP were lower during the wet period, while at both BW and MC the pH of the wet period were higher.

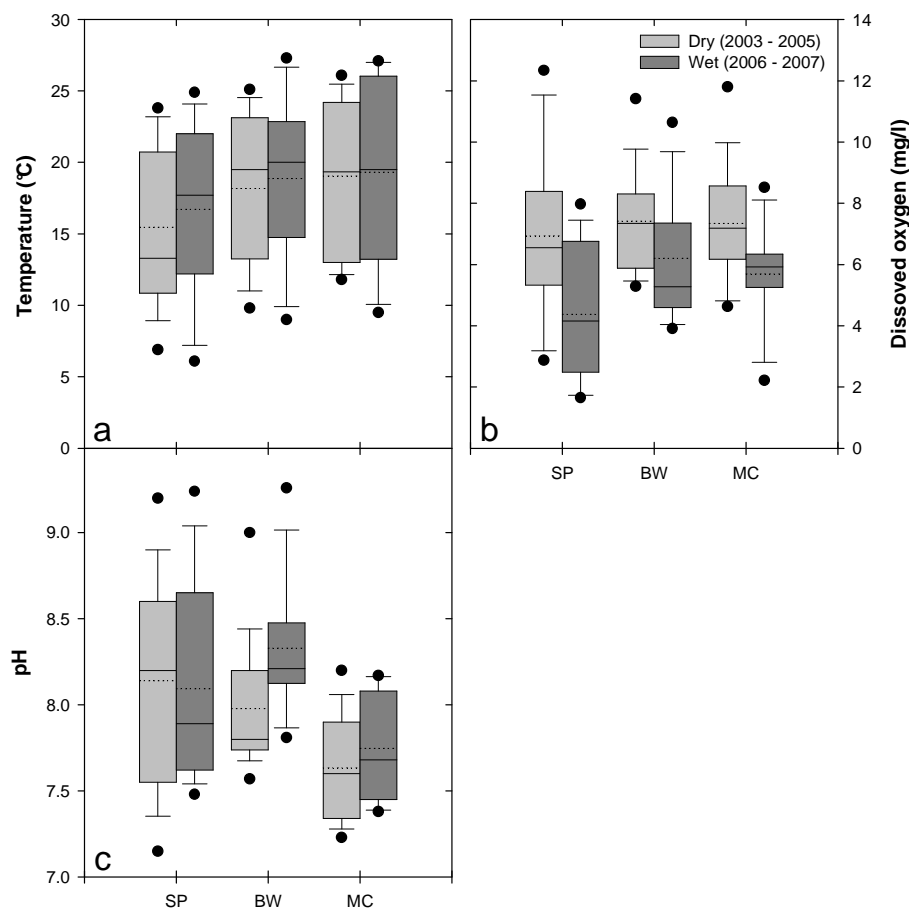


Figure 6.2: Box plots displaying the mean (···) and median (—) of (a) temperature, (b) dissolved oxygen and (c) pH for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

The turbidity (NTU; **Fig. 6.3a**) median and mean at SP were higher in the dry period, whereas those of BW and MC were higher in the wet period. The mean and median average daily discharge (**Fig. 6.3b**) measured at the gauging stations (**Table 6.2**) on the sampling days, were lower during the dry period for

all the sites, except for the mean at SP. The flow median and mean (measured at sampling point) (**Fig. 6.3c**) were lower during the dry period at SP and MC, but higher at BW.

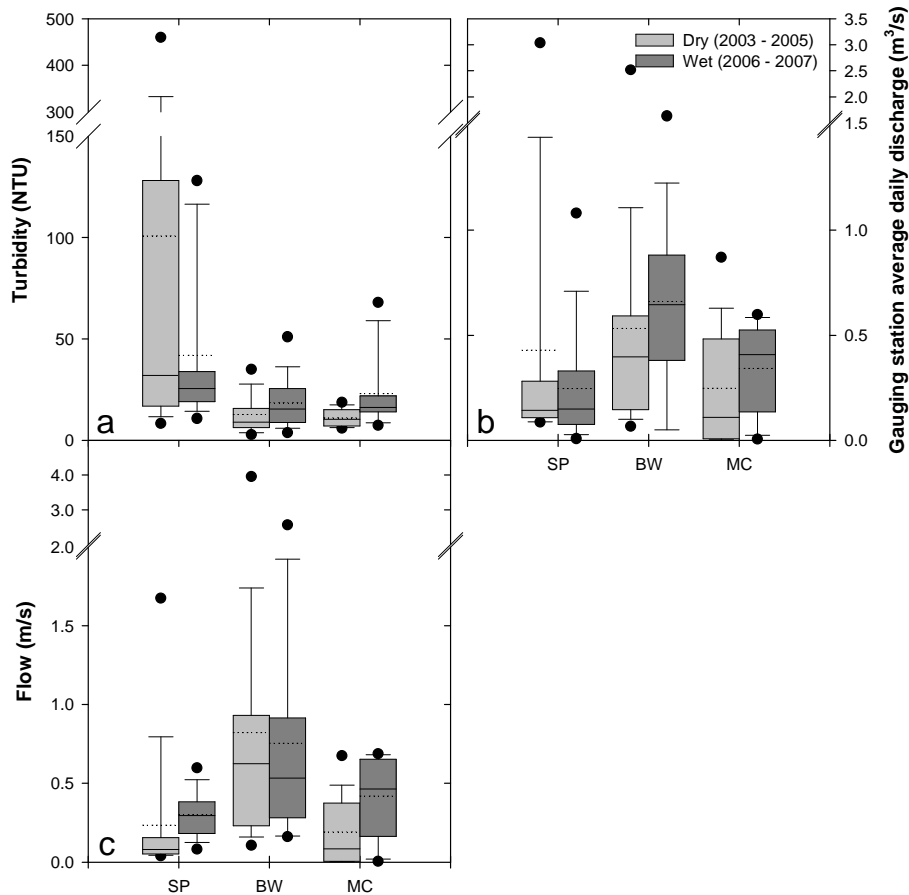


Figure 6.3: Box plots displaying the mean (····) and median (—) of (a) turbidity, (b) average daily discharge and (c) flow for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

The mean and median of the phytoplanktonic chlorophyll-*a* concentration (chl-*a*; **Fig. 6.4a**) at SP were higher during the wet period than the drier period, while the phytoplanktonic chlorophyll-*a* concentrations at BW and MC were lower during the wet period than during the drier period. The periphytic chlorophyll-*a* concentration (pchl-*a*; **Fig. 6.4b**) median and mean at SP and BW were lower during the wet period, while at MC the mean was slightly lower during the wet period, while the median was slightly higher.

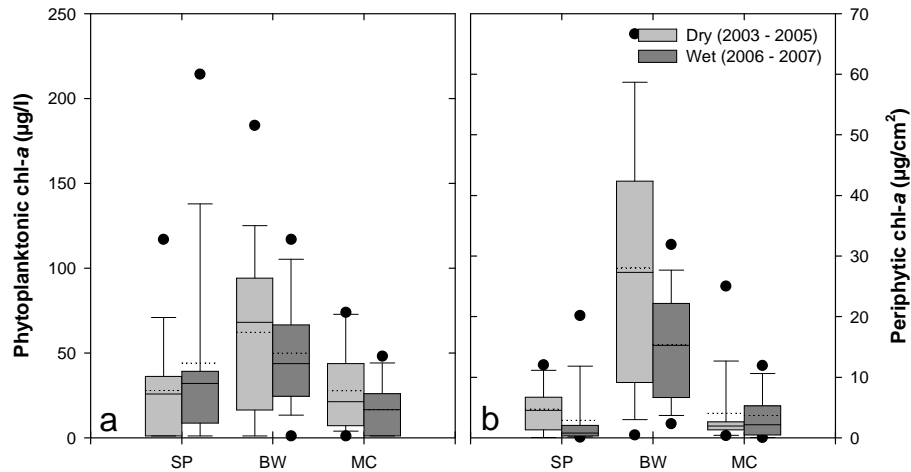


Figure 6.4: Box plots displaying the mean (····) and median (—) of (a) phytoplanktonic and (b) periphytic chlorophyll-*a* for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

6.2.2 NUTRIENTS (DIP & DIN) AND TOTAL DISSOLVED SALTS (TDS)

At SP, the median of the dissolved inorganic phosphorus (DIP) concentration for the dry period was slightly lower than that of the wet period, while the median at BW and MC were lower during the wet period. The median of the dissolved inorganic nitrogen (DIN) concentration showed the inverse of the DIP pattern (**Fig. 6.5a & b**). The median total dissolved salt (TDS) concentrations at all the sites were lower during the wet period (**Fig. 6.5c**) due to the dilution effect of increased flow in the river.

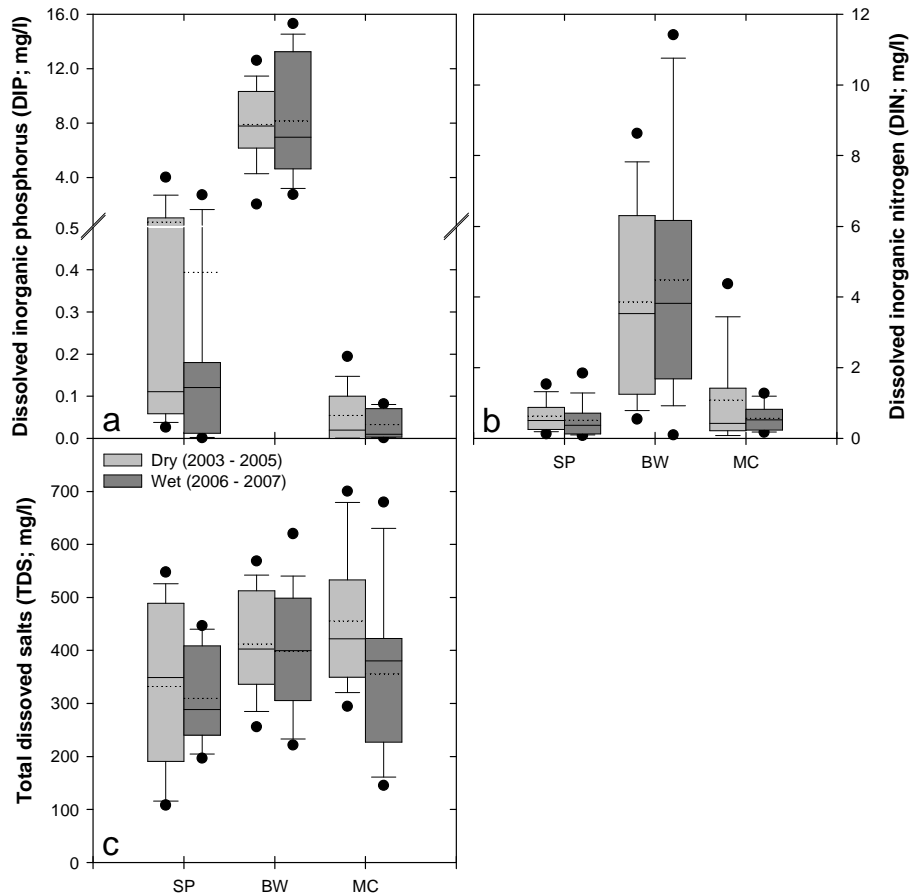


Figure 6.5: Box plots displaying the mean (···) and median (—) of (a) dissolved inorganic phosphorus (note y-axis break), and (b) -nitrogen, and (c) total dissolved salts for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

6.2.3 BIOLOGICAL

The SASS5 stone in current (SASS5 SIC) scores as well as the ASPT (SIC) were higher during the wet period (**Fig. 6.6**). The total SASS5 and ASPT scores shows the same pattern with an exception at BW (**Table 6.4**).

SP and BW showed a higher median of the periphytic algal concentration in the dry period than in the wet period, and at MC the periphytic algal concentration was higher during the wet period. The median periphytic algal biovolumes were lower during the wet period at all the sites (**Fig. 6.7**).

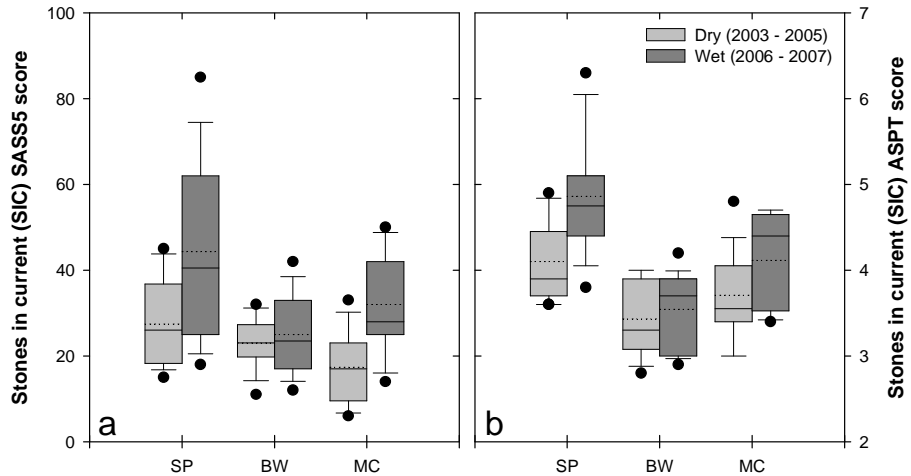


Figure 6.6: Box plots displaying the mean (.....) and median (—) of (a) SASS5 score and (b) ASPT score for stones in current for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

Table 6.4: The monthly median and mean of SASS5 and ASPT scores (totals).

Sites	Year	SASS5		ASPT	
		Monthly median	Monthly mean	Monthly median	Monthly mean
Sannaspos	2003–2005	32.00	37.91	4.14	4.10
	2006–2007	70.00	66.80	4.69	4.64
Bishop's Weir	2003–2005	41.00	41.23	3.81	3.81
	2006–2007	40.50	42.67	3.90	3.78
Modder above Confluence	2003–2005	27.50	30.67	4.00	3.95
	2006–2007	36.00	43.00	4.00	4.02

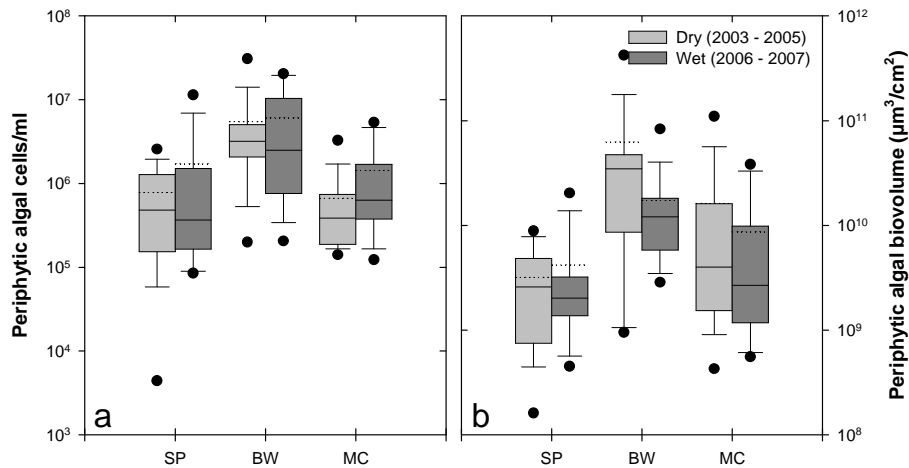


Figure 6.7: Box plots displaying the mean (···) and median (—) of (a) periphytic algal concentration and (b) biovolume (note log on y-axes) for the study periods at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC).

With the individual divisions (**Fig. 6.8 & 6.9**), the median of the Cyanophyta periphytic algal concentration and biovolume both increased from the dry to the wet period, while those of the Bacillariophyta decreased (except the periphytic algal concentration at MC). The median of the Chlorophyta periphytic algal concentration increased at SP and MC, but decreased at BW from the dry to the wet period. The periphytic algal biovolume increased at SP and decreased at BW and MC. The median of the periphytic algal concentration and biovolume of the Euglenophyta increased both from the dry to the wet period.

Fig. 6.10 shows the shift in algal division assemblage (percentage) in the concentration and biovolume at the sites. When displayed as concentration, the Bacillariophyta dominated in numbers (except during the wetter period at SP; **Fig. 6.10a**). The biovolume graph (**Fig. 6.10b**) shows the percentage of “3-dimensional space” each division occupied on the substrate/rock.

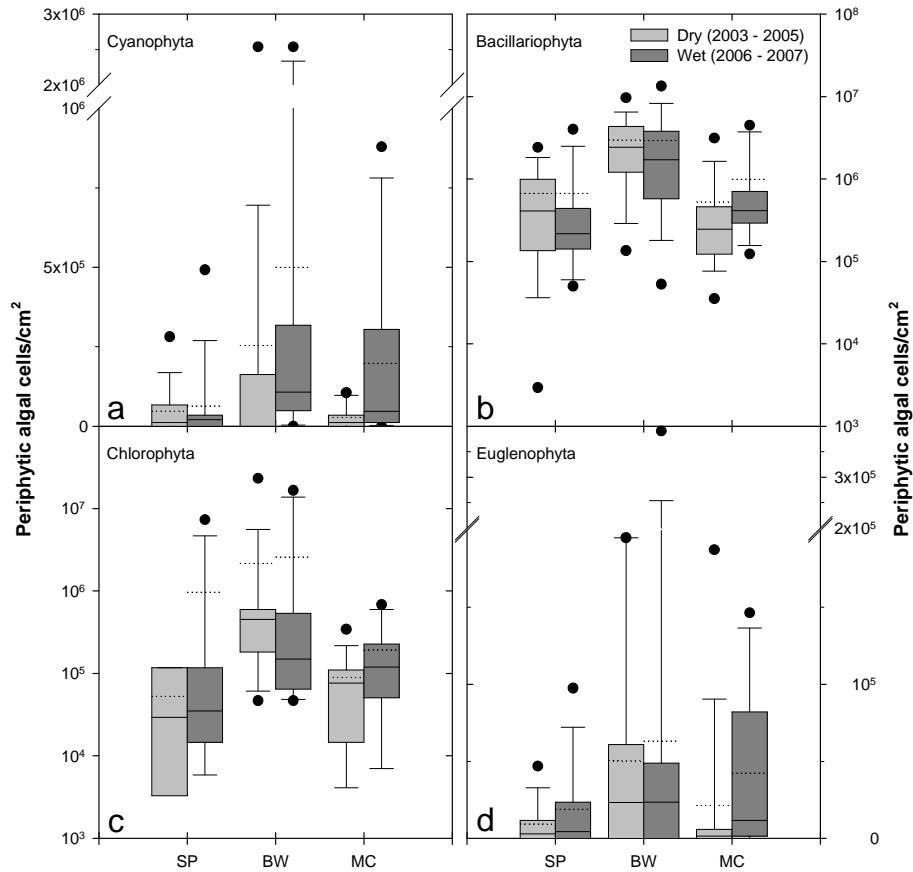


Figure 6.8: Box plots displaying the mean (····) and median (—) of (a) Cyanophyta (note y-axis break), (b) Bacillariophyta (note log on y-axis), (c) Chlorophyta (note log on y-axis) and (d) Euglenophyta (note y-axis break) cell concentration for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

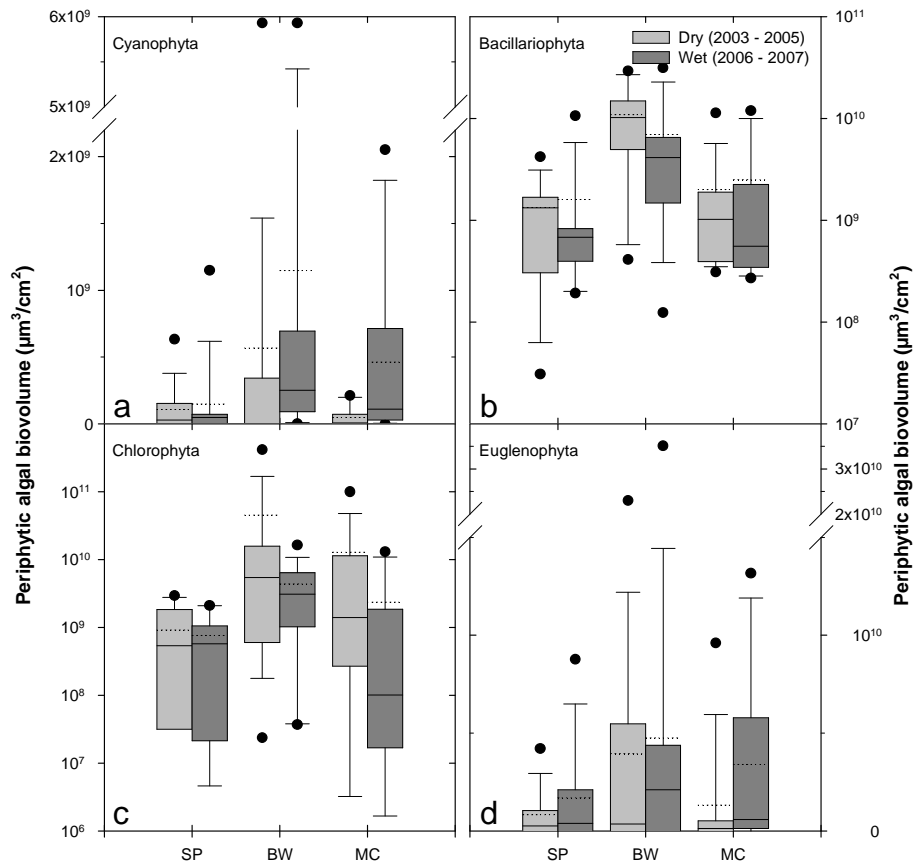


Figure 6.9: Box plots displaying the mean (····) and median (—) of (a) Cyanophyta (note y-axis break), (b) Bacillariophyta (note log on y-axis), (c) Chlorophyta (note log on y-axis) and (d) Euglenophyta (note y-axis break) cell biovolume for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

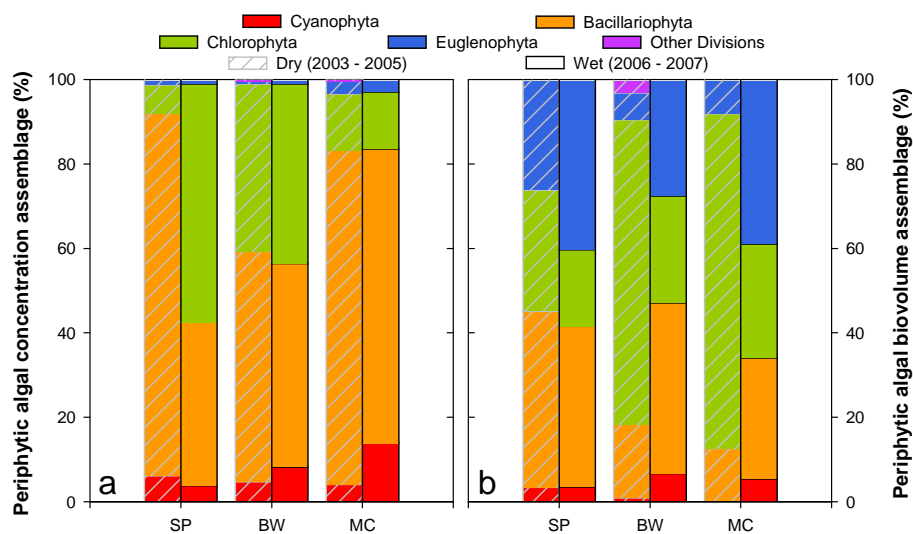


Figure 6.10: Stacked vertical bars displaying the periphytic algal division assemblage as (a) cell concentrations and (b) cell biovolume for the study periods at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC).

Table 6.5 shows that 62 genera (from seven divisions) were found at all three sites during the total study period, of which 32 occurred throughout the study period, while 21 genera were only found during the drier (2003–2005) period and nine genera during the wet (2006–2007) period. The Cryptophyta, Dinophyta and Chrysophyta are mostly planktonic algae (Dodds, 2002) and their genera (one each) were found only during the dry period. **Table 6.6** shows the genera that were found either during the drier or wetter period at the three sites. Half of these periphytic genera were either filamentous or colonial. Out of 11 colonial genera, four each were found only during the dry or wet periods, while two filamentous genera were only present during the dry period and five during the wet period, of a total of 11 filamentous genera.

Table 6.5: The number of periphyton genera that only occurred during the total study period and during either 2003–2005 (dry) or 2006–2007 (wet) period at the three study sites.

Divisions	Nr of periphyton genera			Total over full period
	Only dry	Only wet	Dry and wet	
Cyanophyta	4	3	1	8
Bacillariophyta	1	1	17	19
Chlorophyta	13	5	10	28
Cryptophyta*	1	0	0	1
Dinophyta*	1	0	0	1
Euglenophyta	0	0	4	4
Chrysophyta*	1	0	0	1
Total	21	9	32	62

* = mostly planktonic (Dodds, 2002)

Table 6.6: Periphyton genera that only occurred during the 2003–2005 (dry) or 2006–2007 (wet) periods at all three study sites.

Genera	Only during dry	Only during wet	Genera	Only during dry	Only during wet
<i>Anabaena</i> (F)	√		<i>Crucigenia</i> (C)		√
<i>Merismopedia</i> (C)	√		<i>Mesotaenium</i>		√
<i>Microcystis</i> (sc)		√	<i>Micractinium</i> (C)		√
<i>Nostoc</i> (F)	√		<i>Mougoetia</i> (F)	√	
<i>Oscillatoria</i> (F)		√	<i>Oocystis</i> (C)	√	
<i>Rivularia</i>	√		<i>Oocystis</i> (sc)	√	
<i>Phormidium</i> (F)		√	<i>Pandorina</i> (C)	√	
<i>Amphora</i>		√	<i>Sphaerocystis</i> (C)	√	
<i>Skeltonema</i>	√		<i>Spirogyra</i> (F)	√	
<i>Actinastrum</i> (C)		√	<i>Staurastrum</i>	√	
<i>Ankistrodesmus</i>	√		<i>Tetrastrum</i> (C)		√
<i>Carteria</i>	√		<i>Ulothrix</i> (F)	√	
<i>Chlorococcum</i>	√		<i>Cryptomonas</i>	√	
<i>Chodatella</i>	√		<i>Peridinium</i>	√	
<i>Cosmarium</i>	√		<i>Chrysococcus</i>	√	

C = colonial; F = filamentous; sc = a single cell of a colony. Red = Cyanophyta; orange = Bacillariophyta; green = Chlorophyta; light pink = Cryptophyta; purple = Dinophyta; yellow = Chrysophyta.

6.2.4 MULTIVARIATE RELATIONSHIPS

Additional processed data tables are in **Appendix D.3**.

6.2.4.1 Sannaspos

During the dry period, Sannaspos (SP; **Fig. 6.11**) (n = 11) had an average linear correlation (insignificant) between the total periphytic algal concentrations

(cells/cm²) and biovolume ($\mu\text{m}^3/\text{cm}^2$) ($r^2 = 0.269$; $p = <0.1$). The periphytic chlorophyll-*a* (pchl-*a*; $\mu\text{g}/\text{cm}^2$) concentrations and biovolume ($r^2 = 0.469$; $p = <0.02$), as well as the periphytic algal concentration and pchl-*a* concentration ($r^2 = 0.607$; $p = <0.01$) had moderate and strong, statistically significant linear correlations, respectively. For the wet period ($n = 10$), the linear correlations were all very strong and statistically highly significant: the total periphytic algal concentrations and biovolume ($r^2 = 0.893$; $p = <0.001$), pchl-*a* concentration and biovolume ($r^2 = 0.873$; $p = <0.001$), and periphytic algal concentration and the pchl-*a* concentration ($r^2 = 0.963$; $p = <0.001$).

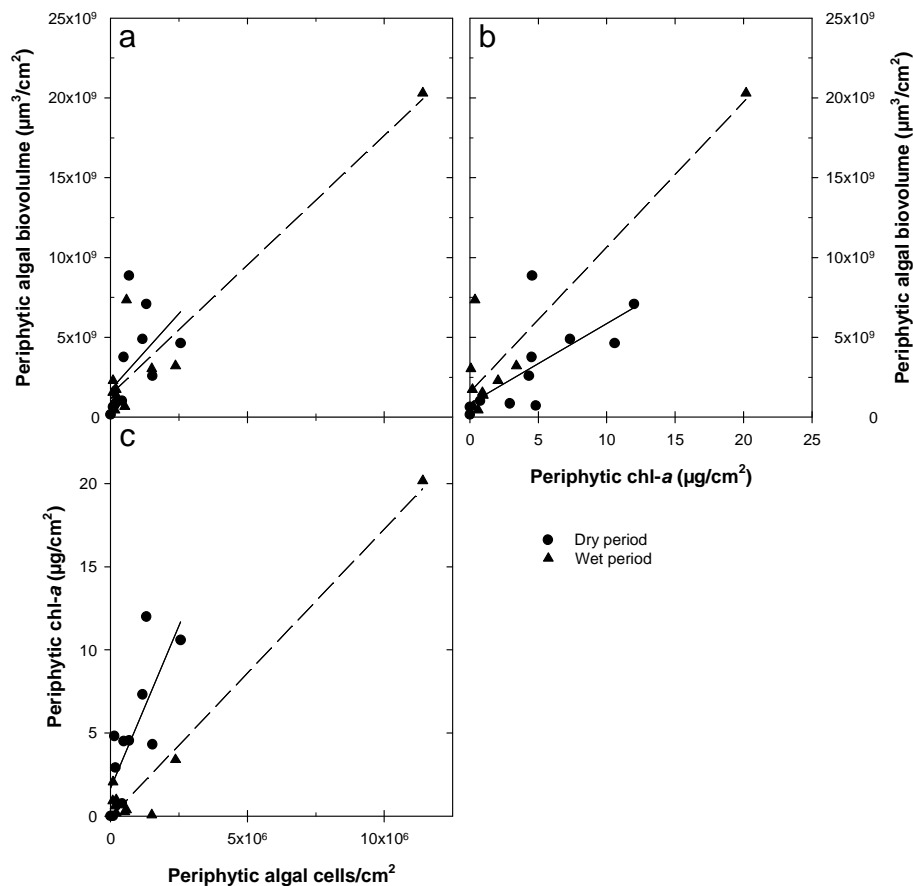


Figure 6.11: The linear relationships between periphyton periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-*a* concentration and algal biovolume, and (c) algal concentration and chlorophyll-*a* concentration at Sannaspos (SP) for the dry (—) and wet (- -) periods.

During both the dry and wet periods at SP, the dominant division was Bacillariophyta, followed by the Chlorophyta, Euglenophyta and Cyanophyta (**Table 6.7**). The mean and median percentages of the division's biovolume are much closer together than that of the concentration. The mean and median percentage of Bacillariophyta algal concentration and biovolume decreased from the dry period to the wet period, causing the other divisions' percentages to increase.

During the dry period the total pchl-*a* concentration had strong, statistically significant correlations with the periphytic algal concentration of the Cyanophyta ($r^2 = 0.550$; $p = <0.01$) and Bacillariophyta ($r^2 = 0.496$; $p = <0.02$), and strong and moderate (statistically significance) correlations with the periphytic algal biovolume of the Cyanophyta ($r^2 = 0.551$; $p = <0.01$) and Bacillariophyta ($r^2 = 0.413$; $p = <0.05$), respectively. During the wet period the total pchl-*a* concentration had very strong and highly statistically significant correlations with the periphytic algal concentration of the Chlorophyta ($r^2 = 0.973$; $p = <0.001$), Bacillariophyta ($r^2 = 0.8937$; $p = <0.001$) and Euglenophyta ($r^2 = 0.770$; $p = <0.001$), and with the periphytic algal biovolume of the Bacillariophyta ($r^2 = 0.962$; $p = <0.001$) and Euglenophyta ($r^2 = 0.770$; $p = <0.001$).

Table 6.7: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Sannaspos for dry and wet periods, percentage of the periphytic algal concentration and biovolume.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Dry (n = 11)	Min	0.00	0.00	62.50	13.29	0.00	0.00	0.00	0.00
	Max	21.43	8.94	97.22	97.31	33.33	84.56	6.90	61.51
	Mean	4.30	2.56	81.01	45.02	13.02	30.67	1.38	21.75
	Median	2.78	1.12	83.87	44.08	9.68	21.77	0.46	22.75
	SD	6.09	3.21	12.40	27.78	11.96	31.13	2.13	22.95
Wet (n = 10)	Min	0.00	0.00	11.33	9.14	0.00	0.00	0.00	0.00
	Max	32.56	37.82	85.71	94.82	85.71	90.86	8.00	65.63
	Mean	9.13	6.23	63.04	40.74	26.14	27.39	1.69	25.65
	Median	2.99	2.34	67.33	31.02	17.22	8.80	0.82	25.89
	SD	11.34	11.68	23.39	30.69	27.38	33.48	2.53	25.53

During the dry period at SP there was no physical, chemical or nutrient factor that had a meaningful, significant correlation with the periphytic algal concentration, biovolume or pchl-a concentration (**Table 6.8; Fig. D.16a & b, D.17a & b, D.18a & b**). During the wet period at SP the DIP and DIN concentrations were the only of the factors that had very strong ($r = >0.7$; $r^2 = >0.490$) linear correlations and were highly statistically significant ($p = <0.001$) with the periphytic algal concentration, biovolume and pchl-a concentration (**Table 6.8; Fig. D.16c & d, D.17c & d, D.18c & d**).

Table 6.8: The coefficient of determination (r^2) and probability (p) values at Sannaspos, between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration for the dry and wet periods.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)		
	r^2	p	r^2	p	r^2	p	
Dry (n = 11)	Temperature	(-) 0.029	>0.1	(-) 0.036	>0.1	(-) 0.104	>0.1
	Diss. oxygen	(-) 0.069	>0.1	(-) 3.77×10^{-3}	>0.1	0.064	>0.1
	Redox	(-) 4.12×10^{-3}	>0.1	0.501×10^3	>0.1	(-) 0.032	>0.1
	pH	(-) 0.280	<0.1	(-) 0.016	>0.1	(-) 0.060	>0.1
	Turbidity	(-) 2.42×10^{-3}	>0.1	(-) 0.135	>0.1	(-) 0.104	>0.1
	Flow	(-) 0.073	>0.1	(-) 0.087	>0.1	0.18×10^{-3}	>0.1
	DIP	(-) 0.084	>0.1	(-) 0.050	>0.1	0.36×10^{-3}	>0.1
	DIN	(-) 0.033	>0.1	(-) 0.145	>0.1	(-) 0.017	>0.1
	TDS	(-) 0.018	>0.1	0.037	>0.1	0.087	>0.1
	Wet (n = 10)	Temperature	(-) 0.282	<0.1	(-) 0.205	>0.1	(-) 0.254
Diss. oxygen		0.268	>0.1	0.238	>0.1	0.285	<0.1
Redox		(-) 0.104	>0.1	(-) 0.119	>0.1	(-) 0.135	>0.1
pH		0.071	>0.1	0.070	>0.1	0.096	>0.1
Turbidity		(-) 0.108	>0.1	(-) 0.123	>0.1	(-) 0.083	>0.1
Flow		(-) 0.083	>0.1	(-) 0.026	>0.1	(-) 0.039	>0.1
DIP		<i>0.898</i>	<i><0.001</i>	<i>0.825</i>	<i><0.001</i>	<i>0.908</i>	<i><0.001</i>
DIN		<i>0.723</i>	<i><0.001</i>	<i>0.560</i>	<i><0.01</i>	<i>0.719</i>	<i><0.001</i>
TDS	0.251	>0.1	0.198	>0.1	0.232	>0.1	

(-) = indicates a negative correlation.

6.2.4.2 Bishop's Weir

During the dry period (n = 13; **Fig. 6.12**) at BW the periphytic algal concentrations and biovolume ($r^2 = 1.01 \times 10^{-3}$; $p = >0.1$), pchl-a concentrations and biovolume ($r^2 = 1.87 \times 10^{-3}$; $p = >0.1$) had no linear or significant correlation between them; while the periphytic algal concentration and the pchl-a

concentrations ($r^2 = 0.380$; $p = <0.02$) had a moderate, statistically significant correlation. During the wet period ($n = 12$) the periphytic algal concentrations and biovolume ($r^2 = 0.283$; $p = <0.1$) had an average, insignificant correlation, while pchl-a concentrations and biovolume ($r^2 = 0.131$; $p = >0.1$), and the periphytic algal concentration and the pchl-a concentrations ($r^2 = 0.111$; $p = >0.1$) had weak, insignificant correlations.

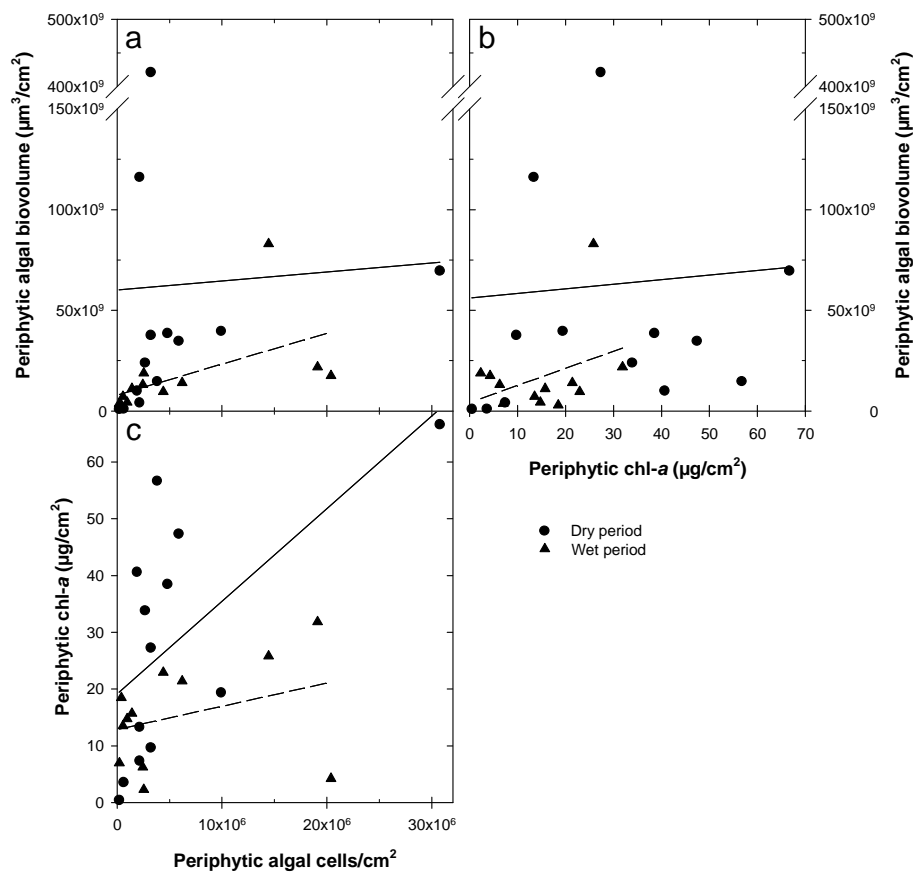


Figure 6.12: The linear relationships between periphyton periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration at Bishop's Weir (BW) for the dry (—) and wet (- - -) periods.

During the dry and wet periods, the Bacillariophyta was the dominant division, followed by the Chlorophyta (**Table 6.9**). The means and medians of both periods were closer with the percentages of the algal biovolume than with the percentage of algal concentration. The percentage algal concentrations and

biovolume of the Bacillariophyta decreased from the dry period to the wet period, while the Chlorophyta, Cyanophyta and Euglenophyta increased.

Pchl-a concentrations had the best correlations (average; statistically significant to insignificant, respectively) with the periphytic algal concentration of Chlorophyta ($r^2 = 0.302$; $p = <0.05$) and Cyanophyta ($r^2 = 0.276$; $p = <0.1$) during the dry period; and a moderate, statistically significant correlation with Bacillariophyta ($r^2 = 0.363$; $p = <0.05$) during the wet period. The best correlations (average; insignificant to statistically significant) between pchl-a concentration and periphytic algal biovolume were with Cyanophyta ($r^2 = 0.276$; $p = <0.1$) and Bacillariophyta ($r^2 = 0.267$; $p = <0.1$) during the dry period and Bacillariophyta ($r^2 = 0.346$; $p = <0.05$) during the wet period.

Table 6.9: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Bishop's Weir for dry and wet periods, percentage of the periphytic algal concentration and biovolume.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Dry (n = 13)	Min	0.00	0.00	15.24	1.48	2.50	2.19	0.00	0.00
	Max	38.46	40.85	97.50	94.85	75.56	98.44	6.06	60.91
	Mean	5.32	4.19	71.118	48.16	22.27	32.52	1.17	12.48
	Median	0.00	0.00	75.61	48.49	16.67	26.19	0.61	0.09
	SD	10.64	11.27	23.10	29.50	21.64	31.98	1.74	18.19
Wet (n = 12)	Min	0.00	0.00	5.26	3.33	0.79	0.28	0.00	0.00
	Max	36.22	37.18	92.57	89.99	81.34	96.68	5.88	73.91
	Mean	10.17	8.00	64.14	38.31	24.14	31.24	1.56	22.45
	Median	3.37	3.13	73.32	35.37	10.87	23.93	0.87	21.17
	SD	12.68	13.06	29.68	27.67	30.34	28.99	1.89	23.93

The periphytic algal concentration's best correlation was with the physical factors, the redox potential (moderate, statistically significant) and turbidity (average, statistically significant), during the dry period (**Table 6.10**; **Fig. D.19**). The periphytic algal biovolume (**Table 6.10**; **Fig. D.20**) had no significant

correlation during the dry period, except with the redox potential (average, statistically significant) and the DIN concentrations (strong, statistically significant) during the wet period. The pchl-a concentration (**Table 6.10; Fig. D.21**) had strong and moderate, statistically significant correlations with the temperature and the DIN correlation, respectively, during the dry period, and moderate, statistically significant correlations with temperature, DIP and DIN concentrations during the wet period.

Table 6.10: The coefficient of determination (r^2) and probability (p) values at Bishop's Weir between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration for the dry and wet periods.

	Algal conc. (cells/cm ²)		Biovolume (µm ³ /cm ²)		Pchl-a (µg/cm ²)		
	r^2	p	r^2	p	r^2	p	
Dry (n = 13)	Temperature	(-) 0.100	>0.1	0.068	>0.1	(-) 0.490	<0.01
	Diss. oxygen	(-) 0.13x10 ⁻³	>0.1	(-) 0.151	>0.1	0.155	>0.1
	Redox	0.382	<0.02	5.71x10 ⁻³	>0.1	0.082	>0.1
	pH	(-) 0.101	>0.1	(-) 0.05	>0.1	0.046	>0.1
	Turbidity	(-) 0.321	<0.05	(-) 2.97x10 ⁻³	>0.1	0.011	>0.1
	Flow	(-) 0.019	>0.1	(-) 0.054	>0.1	(-) 0.132	>0.1
	DIP	(-) 0.091	>0.1	0.016	>0.1	0.024	>0.1
	DIN	0.037	>0.1	(-) 0.136	>0.1	0.383	<0.02
	TDS	(-) 0.145	>0.1	(-) 0.014	>0.1	2.89x10 ⁻³	>0.1
Wet (n = 12)	Temperature	(-) 0.084	>0.1	(-) 0.287	<0.1	(-) 0.372	<0.05
	Diss. oxygen	0.164	>0.1	0.069	>0.1	0.215	>0.1
	Redox	0.011	>0.1	0.389	<0.05	0.030	>0.1
	pH	8.58x10 ⁻³	>0.1	(-) 0.32x10 ⁻³	>0.1	2.68x10 ⁻³	>0.1
	Turbidity	(-) 0.0459	>0.1	(-) 0.049	>0.1	(-) 0.024	>0.1
	Flow	0.124	>0.1	0.086	>0.1	0.093	>0.1
	DIP	0.072	>0.1	0.176	>0.1	0.451	<0.02
	DIN	0.234	>0.1	0.506	<0.01	0.373	<0.05
TDS	0.013	>0.1	0.108	>0.1	0.155	>0.1	

(-) = indicates a negative correlation.

6.2.4.3 Modder above Confluence

At Modder above Confluence during the dry period ($n = 12$; **Fig. 6.13**), the periphytic algal concentration and biovolume ($r^2 = 0.069$; $p = >0.1$), and pchl-*a* concentration and biovolume ($r^2 = 0.045$; $p = >0.1$) had no significant linear correlation between them, while the periphytic algal concentration and the pchl-*a* concentration ($r^2 = 0.936$; $p = <0.001$) had a very strong, statistically highly significant correlation. During the wet period ($n = 7$) the periphytic algal concentrations and biovolume ($r^2 = 0.974$; $p = <0.001$), pchl-*a* concentration and biovolume ($r^2 = 0.842$; $p = <0.001$) and the periphytic algal concentration and the pchl-*a* concentration ($r^2 = 0.915$; $p = <0.001$) all had very strong, highly statistically significant correlations.

At MC, the Bacillariophyta was the dominant division of the periphytic algae, followed by the Chlorophyta (**Table 6.11**) during the dry period and the wet period. The percentage division periphytic algal biovolume means and medians were, as with the other sites, also closer than the percentage concentration. The percentage Bacillariophyta increased from the dry to the wet period, with the other division percentages decreasing over the same period.

The Bacillariophyta had a very strong and statistically highly significant correlation with the pchl-*a* concentration with both the periphytic algal concentration ($r^2 = 0.906$; $p = <0.001$) and –biovolume ($r^2 = 0.845$; $p = <0.001$) during the dry period. During the wet period the pchl-*a* concentration had strong correlations with both the periphytic algal concentration and biovolume of Bacillariophyta ($r^2 = 0.825$; $p = <0.01$ and $r^2 = 0.886$; $p = <0.001$), Chlorophyta ($r^2 = 0.804$; $p = <0.01$ and $r^2 = 0.669$; $p = <0.02$) and Euglenophyta ($r^2 = 0.717$; $p = <0.01$ and $r^2 = 0.764$; $p = <0.01$), ranging from statistically significance to highly statistically significance.

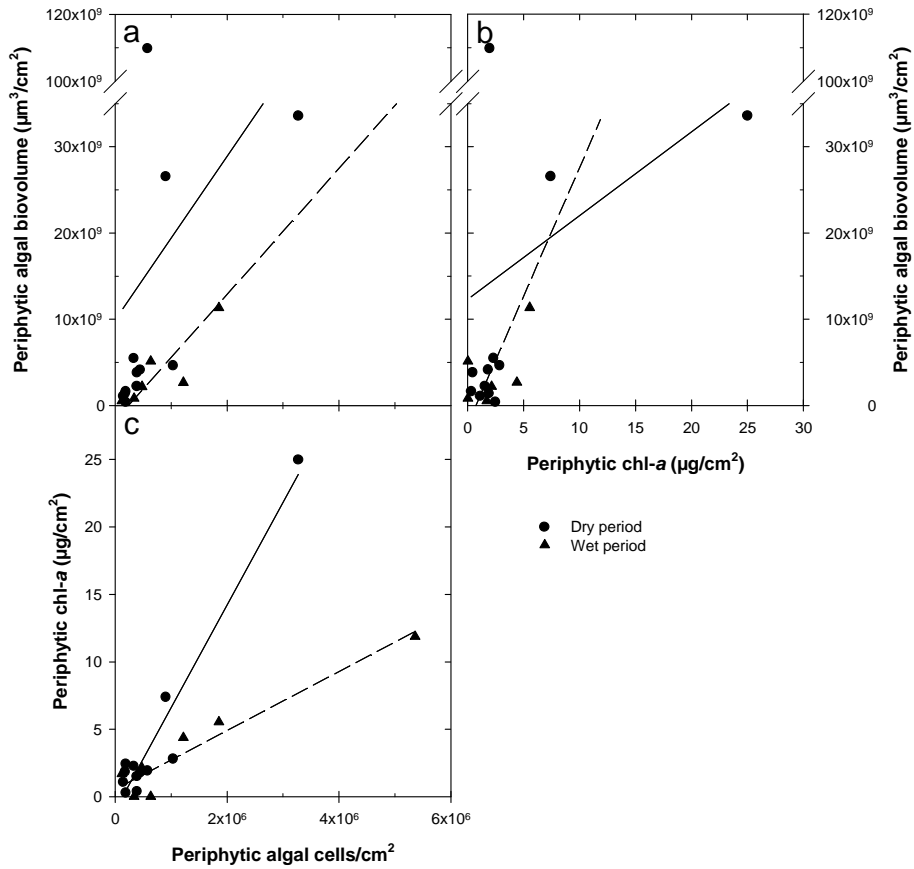


Figure 6.13: The linear relationships between periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration at Modder above Confluence (MC) for the dry (—) and wet (- -) periods.

Table 6.11: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at Modder above Confluence for dry and wet periods, percentage of the periphytic algal concentration and biovolume.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Dry (n = 12)	Min	0.00	0.00	18.76	0.37	0.00	0.00	0.00	0.00
	Max	32.14	12.20	96.60	88.23	65.62	94.34	32.58	47.53
	Mean	8.37	2.28	66.19	40.39	21.12	44.51	3.79	12.52
	Median	2.15	0.12	64.39	34.52	19.26	46.32	0.38	4.37
	SD	11.21	3.74	25.45	31.46	19.83	35.68	9.20	16.99
Wet (n = 7)	Min	0.00	0.00	34.21	6.61	0.00	0.00	0.00	0.00
	Max	47.37	34.15	100.00	100.00	27.69	51.98	5.54	63.30
	Mean	13.61	8.76	70.05	38.58	13.81	18.85	2.53	33.81
	Median	3.45	3.29	64.93	31.08	12.75	3.79	2.43	34.41
	SD	18.62	12.86	21.40	32.21	9.57	22.54	2.24	26.59

With the physical, chemical and nutrient factors' correlation with the periphytic algal concentration, the DIN:DIP ratio had a strong (statistically significant) correlation during the dry period, and the DIN concentrations (**Table 6.12; Fig. D.22**) had an average (insignificant) correlation with the periphytic algal concentration during the dry and the wet periods. There were no significant correlations with the periphytic algal biovolume during the dry period, while during the wet period there was a moderate correlation with the oxygen and an average correlation with the DIN concentration, both insignificant (**Table 6.12; Fig. D.23**). The best correlation with pchl-a concentration was with the DIN:DIP ratio (strong, statistically significant) during the dry period. With the DIN concentration (**Table 6.12; Fig. D.24**), the correlation was weak and insignificant during the dry and wet periods. Only the DIN:DIP ratios had any statistical significance to the pchl-a concentration.

Table 6.12: The coefficient of determination (r^2) and probability (p) values at Modder above Confluence between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-a concentration for the dry and wet periods.

	Algal conc. (cells/cm ²)		Biovolume ($\mu\text{m}^3/\text{cm}^2$)		Pchl-a ($\mu\text{g}/\text{cm}^2$)		
	r^2	p	r^2	p	r^2	p	
Dry (n = 12)	Temperature	(-) 0.122	>0.1	(-) 0.031	>0.1	(-) 0.155	>0.1
	Diss. oxygen	0.032	>0.1	(-) 0.035	>0.1	3.88×10^{-3}	>0.1
	Redox	0.112	>0.1	(-) 5.20×10^{-3}	>0.1	0.175	>0.1
	pH	(-) 0.015	>0.1	(-) 0.108	>0.1	(-) 0.069	>0.1
	Turbidity	(-) 0.079	>0.1	(-) 0.011	>0.1	(-) 0.039	>0.1
	Flow	(-) 0.193	>0.1	3.89×10^{-3}	>0.1	(-) 0.103	>0.1
	DIP	(-) 0.045	>0.1	(-) 0.077	>0.1	(-) 0.019	>0.1
	DIN	0.251	<0.1	(-) 5.08×10^{-3}	>0.1	0.239	<0.1
	TDS	0.142	>0.1	0.025	>0.1	0.195	>0.1
	DIN:DIP	0.732	<0.01	0.011	>0.1	0.752	<0.01
Wet (n = 7)	Temperature	(-) 0.154	>0.1	(-) 0.246	>0.1	(-) 0.063	>0.1
	Diss. oxygen	0.279	>0.1	0.422	<0.1	0.149	>0.1
	Redox	(-) 0.33×10^{-3}	>0.1	(-) 5.94×10^{-3}	>0.1	(-) 0.015	>0.1
	pH	(-) 0.023	>0.1	(-) 8.98×10^{-3}	>0.1	2.12×10^{-6}	>0.1
	Turbidity	(-) 0.136	>0.1	(-) 0.086	>0.1	(-) 0.312	>0.1
	Flow	0.106	>0.1	0.166	>0.1	0.072	>0.1
	DIP	(-) 0.022	>0.1	(-) 0.032	>0.1	(-) 0.031	>0.1
	DIN	(-) 0.313	>0.1	(-) 0.277	>0.1	(-) 0.213	>0.1
	TDS	(-) 0.04×10^{-3}	>0.1	3.02×10^{-3}	>0.1	8.25×10^{-3}	>0.1
	DIN:DIP	(-) 0.019	>0.1	(-) 5.74×10^{-3}	>0.1	(-) 0.76×10^{-3}	>0.1

(-) = indicates a negative correlation.

6.2.4.4 Combined sites

It was found that the combined data of all the sites during the dry period (SP, BW and MC; n = 36; **Fig. 6.14a**) showed no linear correlation or significance ($r^2=$

0.035; $p = >0.1$) between the total periphytic algal concentrations and biovolume. However, during the wet period ($n = 29$), there was a moderate correlation with statistically high significance ($r^2 = 0.393$; $p = <0.001$). The pchl-a concentrations and biovolume had no correlation or significance ($r^2 = 0.073$; $p = >0.1$; **Fig. 6.14b**) during the dry period, and during the wet period an average, statistically highly significant correlation ($r^2 = 0.312$; $p = <0.001$). A strong linear correlation with highly statistically significance ($r^2 = 0.498$; $p = <0.001$; **Fig. 6.14c**) was found during the dry period between the periphytic algal concentration and the pchl-a concentration and a moderate to strong correlation (statistically significant) during the wet period ($r^2 = 0.351$; $p = <0.01$).

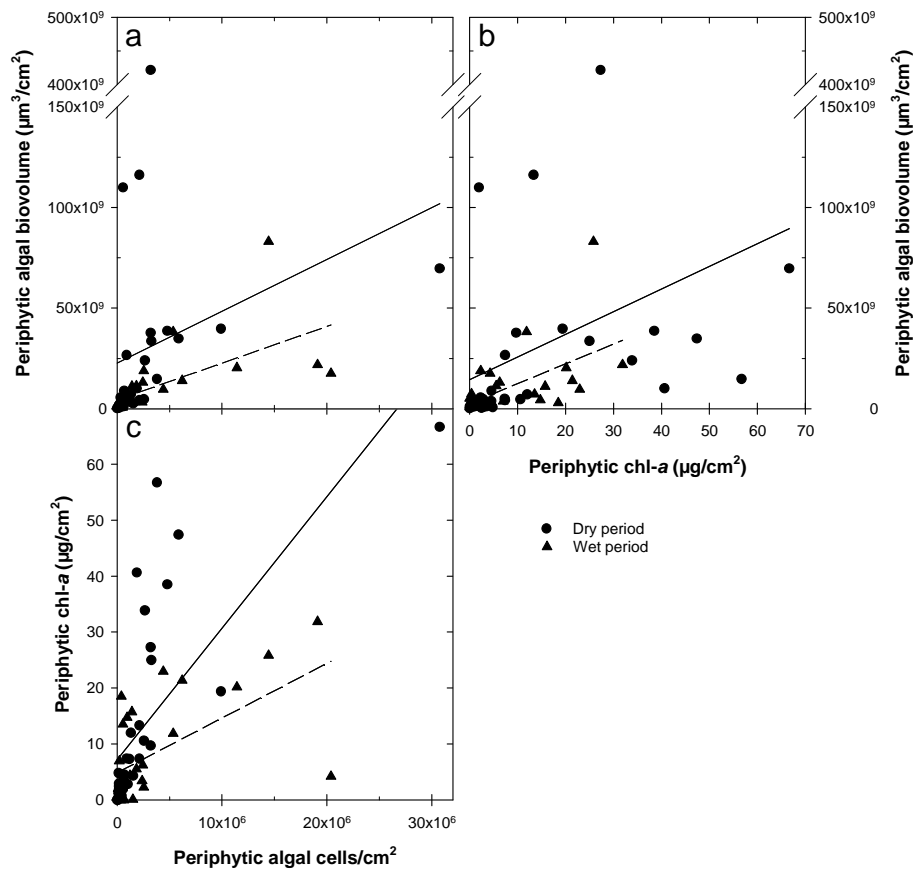


Figure 6.14: The linear relationships between the combined periphyton (a) algal concentration and algal biovolume, (b) chlorophyll-a concentration and algal biovolume, and (c) algal concentration and chlorophyll-a concentration of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) for the dry (—) and wet (- - -) periods.

The Bacillariophyta was the dominant division found with the combined periphytic data, followed by the Chlorophyta with both the periphytic algal concentration and biovolume (**Table 6.13**) during the dry and wet periods. The Cyanophyta and the Euglenophyta took the third and fourth places. The mean and median percentages between the division's biovolumes are closer than the concentration. Overall, the Bacillariophyta decreased from the dry period to the wet period, while the other divisions increased.

The total pchl-a concentration had moderate to strong linear correlations with the Bacillariophyta concentration ($r^2 = 0.435$; $p = <0.001$ and $r^2 = 0.502$; $p = <0.001$) and -biovolume ($r^2 = 0.545$; $p = <0.001$ and $r^2 = 0.496$; $p = <0.001$) during the dry and wet periods, respectively, with highly statistically significance. The total pchl-a concentration had an average correlation with the Chlorophyta algal concentration ($r^2 = 0.305$; $p = <0.001$), and Cyanophyta algal concentration ($r^2 = 0.282$; $p = <0.01$) and biovolume ($r^2 = 0.277$; $p = <0.01$) during the dry period, with statistically highly to statistically significance.

Table 6.13: The minimum, maximum, mean, median and standard deviation (SD) of the major algal divisions found at the combined sites for dry and wet periods, percentage of the periphytic algal concentration and biovolume.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		conc.	biovol.	conc.	biovol.	conc.	biovol.	conc.	biovol.
Dry (n = 36)	Min	0.00	0.00	15.24	0.37	0.00	0.00	0.00	0.00
	Max	38.46	40.85	97.50	97.31	75.56	98.44	32.58	61.51
	Mean	6.02	3.05	72.49	44.61	19.06	35.95	2.11	15.32
	Median	2.04	0.36	75.61	42.81	13.86	31.95	0.53	4.42
	SD	9.58	7.18	21.62	29.00	18.49	32.65	5.51	19.32
Wet (n = 29)	Min	0.00	0.00	5.26	3.32	0.00	0.00	0.00	0.00
	Max	47.37	37.82	100.00	100.00	85.71	96.68	8.00	73.91
	Mean	10.64	7.57	65.19	39.21	22.34	26.92	1.84	26.29
	Median	3.45	2.66	66.66	32.41	12.75	19.66	0.96	26.57
	SD	13.49	12.14	25.06	28.76	25.43	28.70	2.17	24.63

The physical, chemical and nutrient factors had very weak (mostly insignificant) correlations with periphytic algal concentration (**Table 6.14; Fig. D.25**) during the dry period. During the wet period, the DIP and DIN concentrations had average correlations, with statistical significance. There were no correlations or significance with the periphytic algal biovolume (**Table 6.14; Fig. D.26**) during the dry period, while having an average (statistically significant) and moderate (statistically highly significant) correlation with DIP and DIN concentrations, respectively, for the wet period. Data (**Table 6.14; Fig. D.27**) showed that the physical and chemical factors had negligible and insignificant correlations with the pchl-*a* concentration, although the DIN concentration had strong, statistically highly significant correlations and DIP concentrations had moderate to strong correlations, statistically highly significant, to the pchl-*a* concentration during the dry and wet periods.

Table 6.14: The coefficient of determination (r^2) and probability (p) values of the combined sites between the physical, chemical and nutrient factors and the periphytic algal concentration, biovolume and chlorophyll-*a* concentration for the dry and wet periods.

	Algal conc. (cells/cm ²)		Biovolume (µm ³ /cm ²)		Pchl- <i>a</i> (µg/cm ²)		
	r^2	p	r^2	p	r^2	p	
Dry (n = 36)	Temperature	(-) 0.025	>0.1	0.024	>0.1	(-) 0.097	<0.1
	Diss. oxygen	0.23x10 ⁻³	>0.1	(-) 0.026	>0.1	0.039	>0.1
	Redox	0.083	<0.1	(-) 1.93x10 ⁻³	>0.1	(-) 5.5x10 ⁻⁶	>0.1
	pH	(-) 0.013	>0.1	(-) 0.013	>0.1	0.010	>0.1
	Turbidity	(-) 4.83x10 ⁻³	>0.1	(-) 0.015	>0.1	(-) 0.032	>0.1
	Flow	4.4x10 ³	>0.1	(-) 0.80x10 ⁻³	>0.1	2.45x10 ⁻³	>0.1
	DIP	0.078	<0.1	0.125	<0.05	0.404	<0.001
	DIN	0.167	<0.02	(-) 8.82x10 ⁻⁵	>0.1	0.565	<0.001
	TDS	(-) 0.013	>0.1	0.47x10 ⁻³	>0.1	0.015	>0.1
Wet (n = 29)	Temperature	(-) 0.064	>0.1	(-) 0.136	<0.1	(-) 0.091	>0.1
	Diss. oxygen	0.205	<0.02	0.169	<0.05	0.271	<0.01
	Redox	(-) 7.39x10 ⁻³	>0.1	0.069	>0.1	(-) 0.017	>0.1
	pH	0.062	>0.1	0.014	>0.1	0.108	<0.1
	Turbidity	(-) 0.073	>0.1	(-) 0.075	>0.1	(-) 0.119	<0.1
	Flow	(-) 0.011	>0.1	(-) 1.19x10 ⁻³	>0.1	8.80x10 ⁻³	>0.1
	DIP	0.235	<0.02	0.243	<0.01	0.647	<0.001
	DIN	0.344	<0.01	0.444	<0.001	0.545	<0.001
	TDS	0.053	>0.1	0.100	>0.1	0.168	<0.05

(-) = indicates a negative correlation.

6.3 DISCUSSION

Even though both periods, dry and wet, show the same seasonal pattern (**Fig. 6.1**), the difference between the two periods was that during the dry period the winter rainfall (when present) was less than that of the wet period. The same was seen for the summer rainfall in the dry period. The start of the

spring/summer rainfall was slow, as well as lower than that of the wet period. This influenced the mean and median of each period, as seen in **Fig. 6.1**, and contributed to the separation of the two periods.

SP is downstream from the Rustfontein Dam, which is opened periodically to supply water to the downstream drinking water purification plant. Thus, the mean monthly discharge values included the high volume flowing through, whereas the median sees the data only as number of highs and lows. The huge fluctuation between the flooding from the dam and dryness of the site at other times influenced the median during the dry period, so that it was slightly higher than that of the wet period (**Table 6.2**). All the other median and mean monthly discharge volumes were higher during the wet period, which corresponded with the rainfall and total discharge data of the sites.

The median atmospheric maximum temperatures over the study period and minimum and maximum temperatures on specific sampling days (**Table 6.3**) corresponded with the above-mentioned data. As 2003–2005 was drier than 2006–2007 (median: BFN 20.9% and KBY 39.5% less rainfall; mean: BFN 31.9% and KBY 36.7% less rainfall), it was natural that the atmospheric temperatures during the dry period were warmer (even if slightly).

During the summer rainfall season the maximum atmospheric temperatures did not reach such a high during the wet period (cloud cover reduced solar radiation) (Rutherford *et al.*, 1997). The Bloemfontein summer medians and maximums were 14.6 °C and 22.2 °C for the dry period, respectively, and 13.4 °C and 19.8 °C, for the wet period, respectively. The Kimberley summer medians and maximums were 32.7 °C and 40.5 °C for the dry period, respectively, and 32.3 °C and 39.7 °C, for the wet period, respectively.

6.3.1 PHYSICAL AND CHEMICAL

The water temperature (**Fig. 6.2a**) at all the sites showed an increase from the dry to the wet period, even though the atmospheric temperatures showed a

slight decrease over the same period. Various early studies had already shown that river water tends to be warmer after rain, especially in summer or if the sun shine afterwards (Smith & Lavis, 1975; Ward, 1985; Wiley & Seelbach, 1997).

As the solubility of diss. oxygen concentration in water depends on the water temperature of a water body (Giller & Malmqvist, 1998; Allan & Castillo, 2007) and on air pressure (SAWS, n.d.(a); Davies & Day, 1998; Wetzel, 2001), it was not unusual to find that the oxygen of the wet period was lower than that of the dry period. When the flow and turbidity are lower, as found in drier periods (**Fig. 6.3a, b & c**), more algal growth occurs, leading to increased photosynthesis, which increases the oxygen concentrations in the water (Reynolds, 1992; Walling & Webb, 1992; Wetzel & Ward, 1992; Horne & Goldman, 1994; Giller & Malmqvist, 1998; Wetzel, 2001; Dallas & Day, 2004), as found during the dry period.

The pH decrease from the dry to the wet period at SP was expected, in conjunction with the oxygen concentration decrease. This is most probably as a result of respiration by organisms decomposing extra organic matter that washed down the river with the increased flow. Oxygen is used and converted to CO₂, and with that the pH decreases (Allan & Castillo, 2007). The increase in pH at sites BW and MC did not follow the expected pattern, as the pH increased from the drier to the wetter period. This could be a direct result from the upstream soil types at these sites. The soils at BW and MC do have more lime in the soil as well as higher base (more alkaline) structure than the soils around SP (AGIS, n.d.). When the climate is wetter due to more rain, the water draining through the soil became more alkaline; so also the water in the river.

The mean and median turbidity of BW and MC were higher during the wet period as the daily discharge at these sites was higher during the same period (**Fig. 6.3a & b**). With higher volumes of water in a river, more particles get suspended and transported, and lead to increased turbidity (Closs *et al.*, 2004). The decrease of turbidity at SP from the dry to the wet period can be a result of the vast fluctuation during the dry period from very low flow (dry) to flooding, when the sluices of Rustfontein Dam were opened through regulated water releases.

These vast fluctuations could have caused more erosion from the streambed and banks than what would normally occur with lesser fluctuations. This would further increase the suspended sediments in the water column in addition to re-suspension of the sediments (Wetzel, 2001).

The flow, measured at each site, increased at SP and MC from the dry to wet periods as expected, since the discharge also increased from one sampling period to the next (Horne & Goldman, 1994; **Fig. 6.3b & c**). The slight decrease in flow during the wet period, from the dry period at BW, could be a direct result of the flow meter and its measurement. At BW the rocks and the stones in current sample area are, with few examples of cobble, pebble and gravel, mostly boulders. As the flow and discharge increase over these boulders, it may cause “flow pockets” where flow is less than what is expected, or from multiple directions that interfere with the flow meter’s propeller, thus giving a measurement of less than what it should be.

Flow is one of the limiting factors for phytoplankton (Allan & Castillo, 2007) and results in lower phytoplanktonic chl-a concentration at BW and MC during the wetter period (**Fig. 6.4a**). At SP the high turbidity during the drier period also played a role in limiting phytoplankton growth through light limitation (Reynolds, 1992; Walling & Webb, 1992; Giller & Malmqvist, 1998). This could have a bigger effect on the phytoplanktonic chl-a concentration during the drier period than the flow and discharge during the wetter period. The lower pchl-a concentration during the wetter period (**Fig. 6.4b**) was mainly influenced by the higher flow and discharge that occurred. High flow dislodges periphytic algae from their habitat. It also prevents filamentous algae to grow long filaments, as transported sediments damage the algae (Dallas & Day, 2004) and prevent sufficient light penetration.

6.3.2 NUTRIENTS (DIP & DIN) AND TOTAL DISSOLVED SALTS (TDS)

The median DIP concentration (**Fig. 6.5a**) at BW and MC was (slightly) lower during the wet period than during the dry period. As phosphorous has no gas

phase; it must be obtained by dissolving a source or a direct input from a source (Horne & Goldman, 1994; Dodds *et al.*, 2002; Allan & Castillo, 2007). With increase in flow and discharge the DIP concentration usually also increase (Webb & Walling, 1992), but as phosphorus are prone to be adsorbed to particles and sediment (Webb & Walling, 1992) the higher turbidity during the wetter period could be the reason the DIP concentration were lower. At SP the median DIP concentration was slightly higher during the wetter period, and this could be attributed to the higher flow, as well as the lower turbidity during the same period.

The lower DIN concentration (**Fig. 6.5b**) at SP and higher DIN concentration at BW and MC during the wetter period (higher flow/discharge), could be a result of the ration between nitrate and ammonium in the water, as ammonium (like phosphate) is retained by soil and sediment, and nitrate not (Webb & Walling, 1992; Horne & Goldman, 1994). There are also larger irrigation areas along the rivers near BW and MC, from where DIN could have come via fertiliser return flows (Allan & Castillo, 2007).

As the TDS concentration (**Fig. 6.5c**) decreased form the dry to the wet period, it can be concluded that a dilution effect is observed with the higher runoff and discharge (Webb & Walling, 1992) during the wet period. The effect of increased salinity along the longitudinal gradient is observed in the Modder River as the TDS increase from SP to MC (downstream site) for both the periods (Gordon *et al.*, 2004).

6.3.3 BIOLOGICAL

The increase in the SASS5 (SIC) and ASPT (SIC) scores at the sites (**Fig. 6.6a & b**) could not be attributed to specific influences, but it could be as a result of a combination of influences from all the factors, as it seemed that the wet period had slightly better water quality, due to the dilution effect. This is also reflected in the SASS5 and ASPT (total) scores (**Table 6.4**).

According to ranges adapted from Thirion (2003), the SASS5 and ASPT scores are poor to very poor, with the exception of a few fair to good scores (mostly during the wet period). However, Dallas (2007a) mentioned that with low SASS5 scores the ASPT scores are unreliable, as the occurrence of a sensitive taxon can deceptively increase the ASPT score. At SP and MC, one or more families were found during the wet period that scored SASS5 8 or higher, which did not occur at the sites during the dry period.

Even though many of the physical and chemical factors have direct influences on the growth and distribution of periphytic algae, it seems that flow had the bigger influence at the study sites. The overall periphytic algal concentration and biovolume (**Fig. 6.7a & b**) decreased at all the sites (except the periphytic algal concentration at MC) from the dry to the wet period. This is most probably a result from flow increase and higher occurrences of high flow conditions.

The direct influence of flow is that as it increased from the dry to wet period, it dislodges algae and leads to a decrease in the concentration and biovolume (Wetzel, 2001; Allan & Castillo, 2007, Lampert & Sommer, 2007). On the indirect influences, higher flow increases the turbidity (Closs *et al.*, 2004), which decreases the light availability, causing less photosynthesis and growth in periphytic algae (Reynolds, 1992; Walling & Webb, 1992, Wetzel & Ward, 1992; Horne & Goldman, 1994; Giller & Malmqvist, 1998, Wetzel, 2001; Dallas & Day, 2004).

Grazing of macroinvertebrates on periphyton also decreases the concentrations, yet not always the biovolume (Lampert & Sommer, 2007). The increase in the SASS5 (SIC) scores may not necessary indicate an increase in macroinvertebrates at these sites, but if so, it could also be a reason for the decrease in concentration and biovolume of the periphytic algae.

From **Fig. 6.8** and **6.10a** it is found that the Cyanophyta and Euglenophyta (**Fig. 6.8a & d**) are the minority in the periphytic algal assemblages of the three sites and did not influence the periphytic algal concentration decrease at SP and BW during the wet period (**Fig. 6.7a**). What was evident was the influence of the

Bacillariophyta and Chlorophyta (**Fig. 6.8b & c**) concentrations, either individually or together, on the periphytic algal concentration. Bacillariophyta and Chlorophyta are two of the commonest divisions found in periphyton (Horne & Goldman, 1994; Allan & Castillo, 2007).

At SP the Bacillariophyta decreased from the dry to the wet period (**Fig. 6.8 & Ja**), while the Chlorophyta increased, but the Bacillariophyta was by far the majority during both periods. The decrease in the Bacillariophyta leads to the decrease in total periphyton concentration. At BW, both the Bacillariophyta and Chlorophyta concentrations attributed towards the decrease in the periphyton concentration. The Bacillariophyta and Chlorophyta concentrations at MC increased over the two study periods thus the reason for the increase in periphytic alga concentration at BW.

The decrease in the periphytic algal biovolume (**Fig. 6.7b**) was also mostly the result of the decrease in biovolume of Bacillariophyta or Chlorophyta, or both (**Fig. 6.9b & c**), as the Cyanophyta and Euglenophyta (**Fig. 6.9a & d**) increased slightly at all the sites. The Euglenophyta is big cells and the slight increase in concentration has a bigger impact on the biovolume composition (%; **Fig. 6.10b**), especially if the Bacillariophyta and Chlorophyta biovolume decrease.

It is evident in **Table 6.5** that flow had an influence on the periphytic algal genera, as it is seen that a third of the total amount of genera that occurred over the total study period occurred only during the dry period, and about a sixth during the wet period. This supports the known fact that periphyton diversity in a river with high velocity is low (Wetzel, 2001). That left only half of the genera to be present during both periods.

It is also seen in **Fig. 6.10b** how the percentage shift in periphytic algal biovolume took place. It is clear that the Chlorophyta lost the most biovolume, as is reflected in **Table 6.6**, where it shows that most of the genera (specifically the Chlorophyta) that were lost during the shift from dry to wet were either filamentous or colonial. Chlorophyta and Cyanophyta found in rivers are mostly

filamentous, where Bacillariophyta is most often unicellular pennates with some filamentous centric ones.

Filamentous algae are normally counted as one, but comprise large biovolumes. With the increase in flow/discharge from the dry to the wet period, some planktonic genera were also lost as they were swept away in the current from between the periphytic algae, as the turbulence also removes periphyton that are loosely attached to their substrate (Wetzel, 2001)

6.3.4 MULTIVARIATE RELATIONSHIPS

6.3.4.1 Sannaspos

The difference in the linear regression correlations and statistical significance between the total periphytic algal concentrations, the biovolume and pchl-*a* concentrations at Sannaspos during the dry and the wet periods were that those of the wet period were all very strong and statistically highly significant (**Fig. 6.11**). This could be due to the fact that, during the wet period many of the filamentous and colonial periphytic algae that were present during the dry period were absent. Studies have found that filamentous Chlorophyta prefers lower flow <0.09 m/s (Horner *et al.*, 1990). This was also reflected in the median values of total periphytic algal concentrations, the biovolume and pchl-*a* concentrations for both periods.

As seen in **Chapter 4**, the Bacillariophyta was the dominant division during both the dry and wet periods, followed by the Chlorophyta (**Table 6.7**). The decrease in the Bacillariophyta periphytic algal concentration and biovolume could probably be ascribed to the increase in flow during the wet period. As flow increases slightly at SP, loosely adhered Bacillariophyta can be dislodged and thus opens space for Chlorophyta and Cyanophyta to colonise (Biggs & Close, 1989). Sites downstream of dams usually have a higher proportion of filamentous algae (Chester & Norris, 2006). This can be observed in the mean and median (0.425 and 0.445, respectively) Chlorophyta percentage of

periphytic algal concentration:biovolume (c:bv) ratios. The increase in flow was probably the driver behind the decrease in the Chlorophyta biovolume.

Even though there were no meaningfully significant correlations between the physical and chemical factors and periphytic algal concentration, biovolume and pchl-*a* concentration during both the dry and wet periods, some factors' correlations increased during the wet period with all, or at least two, of the algal components (**Table 6.8; Fig. D.16, D.17 & D.18**). The strong (statistically highly significant) correlations of the algal components with the nutrients (DIP and DIN) could also be ascribed to the increase in flow, as it eliminated the diffusion limitation problem in the algal mat (Rier & Stevenson, 2006).

6.3.4.2 Bishop's Weir

At Bishop's Weir, the inversion of the correlations between the periphytic algal components from the dry to the wet period could be the result of the huge increase in the percentage Euglenophyta biovolume with its slight increase in concentration against the overall decrease in total periphytic algal concentration, biovolume and pchl-*a* concentration from the dry to the wet period (**Fig. 6.12**).

The slight decrease of the percentage Bacillariophyta and Chlorophyta concentrations and biovolumes from the dry to the wet period results in a composition shift to an increase in the Euglenophyta and Cyanophyta (**Table 6.9**). The loss in the Bacillariophyta and Chlorophyta percentages of the composition could not be ascribed to the flow, as the flow decreased from the dry period to the wet period. The shift could probably be as a result of the slight median increase of temperature over the period or the decrease in the water clarity over the same period.

The moderate to strong (statistically significant) negative correlation of temperature with the pchl-*a* concentrations during both the dry and the wet periods (**Table 6.10; Fig. D.19, D.20 & D.21**) goes against the literature, where it is mentioned that an increase in water temperature promotes the increase in

algal activities (Wetzel, 2001), but as the site has little riparian shade over the river, the increase in temperature could also be an indication of an increase of solar radiation (light intensity) at the site (Ward, 1985; Webb *et al.*, 2008). The higher solar radiation with the relative low turbidity could result in photo-inhibition, as the chloroplast is damaged (or bleached) and thus a decrease in the pchl-*a* concentration occur (Quesada *et al.*, 1995; Roos & Vincent, 1998).

The statistically significant correlations of the periphytic algal components with the nutrient (DIP and DIN) concentrations increase from the dry period to the wet period. This could be due the slightly lower flow, as the flow decreases to 0.53 m/s (median) during the wet period. As discussed in **Section 6.3.1**, during higher flow, the boulders may cause “flow pockets” with lower flow. Nutrients were always in abundance (hypertrophic state) at this site.

6.3.4.3 Modder above Confluence

The linear correlations and significance at Modder above Confluence, between the periphytic algal concentrations, biovolume and pchl-*a* concentrations (**Fig. 6.13**) were very strong and highly statistically significant during the wet period as, with the increase in flow, the periphytic algal biovolume decreased (Bowes *et al.*, 2012). The periphytic algal concentration and pchl-*a* concentration, however, increased as the flow increased. Flow up to 0.5 m/s can still stimulate periphytic growth and thus also an increase of pchl-*a* concentration (Horner *et al.*, 1990). At lower flow, nutrients can become limited in the algal mat, and flow higher than 0.25 m/s annihilate this effect (Rier & Stevenson, 2006). The fact that the periphytic algal and pchl-*a* concentrations had very strong correlations and significances during both dry and wet periods indicated that the pchl-*a* concentration is an ideal indicator for calculating the living biomass at MC, and can be used as trophic level indicator (Wellnitz & Poff, 2006; Kilroy *et al.*, 2013).

The Bacillariophyta's (dominant during both the periods) percentage of assemblage remained the same, but the percentage biovolume decreased slightly (**Table 6.11**). This, together with the decrease in the Chlorophyta's

percentages of concentration and biovolume, indicates that the five-fold increase in the median flow might have been the reason for the periphytic algal concentration increase and the decrease of the biovolume. The higher flow most probably removed loose attached and damaged algae, and filamentous/colonial algae, making space for smaller ones to attach and colonise (Biggs & Close, 1989; Biggs *et al.*, 1998b; Flynn *et al.*, 2013). Filamentous green algae prefer flow <0.09 m/s (Horner *et al.*, 1990). With the increase of flow at MC, many of the filamentous/colonial algae were absent during the wet period. Removal of the filamentous algae also eradicated the self-shading effect (Nijboer & Verdonschot, 2004; Rier *et al.*, 2006). The slight increase in the Euglenophyta percentage concentrations caused a huge increase in the percentage biovolume of the Euglenophyta, and so helped with the increase in the total pchl-a concentration (big cells).

The weak correlations and insignificance between the physical, chemical and the nutrient factors with the periphytic algal components (**Table 6.12**; **Fig. D.22, D.23 & D.24**) could be that, at various moments, any one of the factors was in a limiting position (Horner *et al.*, 1990), as it takes only one factor to limit growth at any particular time (Liebig's law of the Minimum; Hill & Fanta, 2008)

6.3.4.4 Combined sites

The correlations and significance between the total periphytic algal concentrations, biovolume and pchl-a concentrations of the combined data of the three sites (SP, BW & MC; **Fig. 6.14**) were mostly influenced by the site with the stronger correlations. The overall better correlations during the wet period were still portrayed in most of the data. For the combined data, the pchl-a concentration and biovolume, and periphytic algal and pchl-a concentrations' correlations were the best.

Since the Bacillariophyta was dominant at all the sites (**Table 6.13**), it was found to be dominant with the combined data, followed by the Chlorophyta. The general trends from the separate sites flow through to the combination of data

where Bacillariophyta percentages were less during the wet period for both the concentration and biovolume. The Chlorophyta percentage concentration was a little less during the wet period, whereas the percentage biovolume was much less. The wet period was more favourable towards the Euglenophyta and Cyanophyta, where both the divisions' percentage concentration and biovolume increased. The reason for this overall algal assemblage shift could be that the change in flow from the dry to the wet period had an effect in the availability of nutrients and light to the periphytic algal mat attached to the substrata (Biggs & Close, 1989; Nijboer & Verdonschot, 2004; Rier & Stevenson, 2006; Rier *et al.*, 2006).

With the correlations between the physical, chemical and nutrient variables with the periphytic algal components (**Table 6.14; Fig. D.25, D.26 & D.27**), the same pattern was observed with the combined data. As found in other studies (Justus *et al.*, 2010), it was mostly the nutrients (DIP and DIN) that had the best correlations with the periphytic algal components, and with better correlations and higher significance during the wet period than during the dry period.

It was found that during the dry and the wet periods, the pchl-a concentration at each of the sites gave the best indication of the trophic status of the river reach upstream from it. The shift in periphytic algal composition through absence or presence of filamentous/colonial algae from one period to the other could be important in estimating the flow regime. If a site does not have filamentous/colonial algae present, it could indicate fast-flowing water or regular floods (natural or regulated) at the site.

CHAPTER 7

APPRAISAL OF BIOMONITORING INDICATOR METHODOLOGIES

7.1 INTRODUCTION

By carrying out biological monitoring, a direct measure of the ecological integrity is provided, using the response of the biota to the environmental change (Lavoie *et al.*, 2008; Dallas, 2013). Algal indices have already been proven efficient with the monitoring of water quality, as did benthic macroinvertebrates (SASS) and fish indices (Justus *et al.*, 2010; Kireta *et al.*, 2012).

It was also found that algal indices had better correlations with the nutrient index than macroinvertebrates and fish had, because of their short generation time (De la Rey *et al.*, 2008; Kireta *et al.*, 2012; Schneider *et al.*, 2012). Studies have found that macroinvertebrate indices have stronger relationships with change in the habitat and function of the aquatic environment (De la Rey *et al.*, 2008). The FRAI index is more linked to the catchment or river reach health and integrity of a river, as well as the habitat availability (Kleynhans, 2007).

This chapter looks at various biomonitoring indices, the basics of their methodologies and the results found at Sannaspos, Bishop's Weir and Modder above Confluence.

7.2 RESULTS

Additional processed data tables in **Appendix D.4. Table 7.1** show the n-values for the probabilities (p) at the various sites and study periods (excl. fish), the fish were sampled as a once off each year.

Table 7.1: The n-values for the probabilities (p) at the study sites, for the various periods of study.

	n-values				
	Total study period	Dry period	Wet period	Summer	Winter
Sannaspos	21	11	10	NA	NA
Bishop's Weir	25	13	12	NA	NA
Modder above Confluence	19	12	7	NA	NA
Combined Sites	65	36	29	18	24

7.2.1 FISH

The FAIL (old index) and FRAI indices essentially use the same information, but their procedure differs. The FAIL index was “*developed for application in the broad synoptic assessment required for the RHP and does not have a particularly strong cause-and-effect basis*”. FRAI “*provide a habitat-based cause-and-effect underpinning to interpret the deviation of the fish assemblage from the reference condition*” (Kleynhans, 2007).

Thus, FRAI can be defined as an assessment index measuring “*the environmental intolerances and preferences of the reference fish assemblage and the response of the constituent species of the assemblage to particular groups of environmental determinants or drivers*” (Kleynhans, 2007).

The FRAI method was developed on perennial rivers and it is regarded as such a method (Avenant, 2010). Avenant (2000; 2010) found in two different studies that the hydrological phases in regulated and non-perennial rivers are very important, especially if the rivers have been modified by dams and weirs. The opening of dam sluices sends large amounts of water down a river at one time and can cause damage to fish habitat, while the damming of a river by a weir removes natural flow habitat and increases pool habitat.

Avenant (2000) and Seaman *et al.* (2007) further mention that the fish species in the Modder River are all generalists, and require no specialist niches. If this is combined with the absence of sensitive fish species from the FRAI list, it is inevitable to calculate a low score for the said river reach.

The Modder River is a highly regulated river with various dams (reservoirs) from which water are regularly released and weirs that dam the main stream flow. FAII and FRAI results from 2003 to 2007 (**Table 7.2**) found the reaches in which SP, BW and MC fall to be highly modified (Seaman *et al.*, 2003; 2004; 2005; 2007). The hydrological phase effect is also seen in the data, as the scores increased from 2003 to 2007 as the hydrology cycle moved from dry tot wet.

Table 7.2: The FAII and FRAI scores of the river reaches in which Sannaspos, Bishop's Weir and Modder above Confluence fall over the study period* (adapted from Seaman *et al.*, 2003; 2004; 2005; 2007).

Modder River Reach	Sites	2003	2004	2005	2007	
		FAII	FAII	FAII	FRAI	FRAI
Upper-Middle	SP	D – 50.1%	E – 38.2%	E – 36.9%	C – 64.7%	C – 62.8%
Lower-Middle	BW	D – 49.3%	E – 25.6%	E – 26.0%	C – 61.2%	D/E – 40.9%
Lower	MC	D – 49.8%	E – 31.0%	D – 41.6%	D – 56.7%	D – 49.1%

(C = moderately modified; D = largely modified; E = seriously modified)

* = no sampling was done during 2006.

7.2.2 BENTHIC MACROINVERTEBRATES

The taxa (families) used in the SASS5 method are scored on a sensitivity scale from one (1) to 15, and can be subdivided into three categories according to their tolerance to pollution. Scale one to five (1-5) for taxa with a high tolerance to pollution, scale six to ten (6–10) for those with a moderate tolerance to pollution and the scale 11 to 15 for the taxa with a very low tolerance to pollution (Gerber & Gabriel, 2002). Therefore, if taxa with scores that range from 11 to 15 are

found in waters, it can be assumed that the water is unpolluted or relatively unpolluted, but taxa with a score from one to five may occur in any water.

The SASS5 scores are calculated by adding the sensitivity scores of taxa found in any of the biotopes sampled and the ASPT scores by dividing the SASS5 score by the number of taxa found in the sample (Dickens & Graham, 2002). Depending on how the sites are structured, one biotope could dominate the SASS5 score. The absence of a biotope also has an influence on the SASS5 and ASPT scores (Dallas, 2007b).

From **Tables 7.3, 7.4** and **7.5** it is seen that the SASS5 and ASPT scores had moderate to strong linear correlations with statistically high significance (but a few) between their respective total and SIC scores. The correlations between the SASS5 and ASPT total scores, and SASS5 and ASPT SIC scores were mostly weak (variety of significance). BW had strong, statistically highly significant correlations between the SASS5 and ASPT SIC for the whole study period (**Table 7.3**) and during the wet period (**Table 7.5**), while the combined data showed moderate and strong correlations (statistically highly significant) between the SASS5 and ASPT total scores, and SASS5 and ASPT SIC scores during the summer (**Table 7.4**).

Table 7.3: The coefficient of determination (r^2) and probability (p) values at the separate sites and combined sites, between the SASS5 (total and SIC) and ASPT (total and SIC) scores, and total (SASS5 and ASPT) and SIC (SASS5 and ASPT) scores.

Sites	#Total & SIC				§SASS5 & ASPT			
	SASS5		ASPT		Total		SIC	
	r^2	p	r^2	p	r^2	p	r^2	p
SP	0.896	<0.001	0.659	<0.001	0.196	<0.05	8.19×10^{-3}	>0.1
BW	0.558	<0.001	0.412	<0.001	0.324	<0.01	0.559	<0.001
MC	0.684	<0.001	0.648	<0.001	0.013	>0.1	0.267	>0.1
*Comb.	0.794	<0.001	0.669	<0.001	0.178	<0.01	0.191	<0.01

* = combined sites; # = correlation between the total and SIC scores of SASS5 and ASPT, respectively; § = correlation between the SASS5 and ASPT scores.

Table 7.4: The coefficient of determination (r^2) and probability (p) values for the combined sites, between the SASS5 (total and SIC) and ASPT (total and SIC) scores, and total (SASS5 and ASPT) and SIC (SASS5 and ASPT) scores, for summer and winter seasons.

Season	#Total & SIC				\$SASS5 & ASPT			
	SASS5		ASPT		Total		SIC	
	r^2	p	r^2	p	r^2	p	r^2	p
Summer	0.806	<0.001	0.605	<0.001	0.450	<0.001	0.587	<0.001
Winter	0.639	<0.001	0.580	<0.001	0.197	<0.05	0.119	>0.1

= correlation between the total and SIC scores of SASS5 and ASPT, respectively; \$ = correlation between the SASS5 and ASPT scores.

Table 7.5: The coefficient of determination (r^2) and probability (p) values at the separate sites and combined sites, between the SASS5 (total and SIC) and ASPT (total and SIC) scores, and total (SASS5 and ASPT) and SIC (SASS5 and ASPT) scores for dry and wet periods.

Sites	#Total & SIC				\$SASS5 & ASPT				
	SASS5		ASPT		Total		SIC		
	r^2	p	r^2	p	r^2	p	r^2	p	
Dry	SP	0.790	<0.001	0.367	<0.05	0.279	<0.1	0.344	<0.05
	BW	0.652	<0.001	0.442	<0.01	0.363	<0.05	0.435	<0.01
	MC	0.732	<0.001	0.671	<0.001	0.093	>0.1	0.346	<0.05
	*Comb.	0.347	<0.001	0.497	<0.001	0.137	>0.1	0.325	<0.001
Wet	SP	0.901	<0.001	0.585	<0.01	1.59x10 ⁻³	>0.1	0.270	<0.1
	BW	0.525	<0.001	0.567	<0.01	0.427	<0.02	0.683	<0.001
	MC	0.667	<0.02	0.751	<0.01	(-) 0.012	>0.1	6.93x10 ⁻³	>0.1
	*Comb.	0.809	<0.001	0.790	<0.001	0.163	<0.05	0.078	>0.1

(-) = indicates a negative correlation; * = combined sites; # = correlation between the total and SIC scores of SASS5 and ASPT, respectively; \$ = correlation between the SASS5 and ASPT scores.

The percentage representation of biotopes in the total SASS5 scores was dominated by the stones-in-current (SIC) (**Table D.10**), with a median and mean representation of over 50% at all the sites during all periods (dry/wet) and seasons (summer/winter), except at MC during summer. Of all three sites, BW had the lowest percentage representation of SASS5 SIC scores in the total SASS5 score (except at MC during summer). At BW and MC, these percentage representations increased from summer to winter and decreased at SP and BW from the dry period to the wet.

Most correlations between the SASS5 and ASPT scores with the physical, chemical and nutrient factors were weak with a few exceptions. At SP there were moderate to strong correlations between the SASS5 and ASPT (total and SIC) scores with the dissolved oxygen and pH (both neg.) during the dry period, as well as between the ASPT (total and SIC) scores and the temperature. At BW no noticeable correlations between SASS5 and ASPT (total and SIC) scores and any of the physical, chemical and nutrient factors were found. At MC there were moderate to strong correlations between the SASS5 (total and SIC) scores and the temperature (stronger during the wet period), and it had strong correlations with the dissolved oxygen (neg.) during the wet period. The combined summer data for all the sites showed that there were moderate to strong correlations between the SASS5 and ASPT (total and SIC) scores and dissolved oxygen (neg.).

There were little to no correlation between the SASS5 and ASPT scores and those of the periphytic algal components. The best correlations (moderate) were between the ASPT total score and the total periphytic algal concentrations and biovolumes at BW during the wet period.

Macroinvertebrate taxa with sensitivity scores of 11 to 15 were not found at any of the three sites during the study period (**Figure C.10; Table D.11**). Taxa with a score from six to ten were scarce and were mostly present at sites during the wet period (**Table D.13**). Taxa with scores from one to five occurred at all the sites throughout the study period. Four taxa (Ecnomidae, 8; Hydraenidae, 8; Planorbinae, 3; Sphaeriidae, 3) at SP were found to be not present during either

summer or winter (**Table D.12**), but in autumn and spring, and only during the wet period. These four taxa were not found at the BW and MC during the study period. Various taxa were found to be present at only one or two of the three sites.

SP was the site that had the highest number of taxa (20) present at one sampling over the study period, but the median number of taxa present at any of the sites was six and seven (**Table 7.6**). The highest maximum number of taxa found at all the study sites was during the wet period. Samples with low numbers of taxa were found throughout the dry and wet periods. The minimum and maximum occurrences of numbers of taxa occur at the same sampling times at the three sites. Higher numbers of taxa (median and mean) were found during the wet period and during summer.

The median SASS5 scores at SP were either fair (dry and summer) or poor. At BW, the medians all indicated poor scores, while at MC the medians were poor (dry and winter) or very poor SASS5 scores (**Table 7.7**).

Table 7.6: The minimum, maximum, mean, median and standard deviation (SD) of the number of macroinvertebrate taxa found at the study sites for the various periods of study.

		No. of taxa present during various period				
		Total study period	Dry period	Wet period	Summer	Winter
Sannaspos	Min	4.0	4.0	4.0	7.0	4.0
	Max	20.0	10.0	20.0	13.0	11.0
	Mean	8.0	6.6	9.6	9.4	6.5
	Median	7.0	7.0	9.0	8.0	6.0
	SD	4.2	2.1	5.3	2.9	2.8
Bishop's Weir	Min	4.0	4.0	4.0	6.0	5.0
	Max	10.0	8.0	10.0	10.0	10.0
	Mean	6.8	6.7	6.9	7.3	7.0
	Median	7.0	7.0	7.0	7.0	7.0
	SD	1.7	1.4	2.0	1.3	1.7
Modder above Confluence	Min	2.0	2.0	3.0	2.0	2.0
	Max	11.0	8.0	11.0	11.0	10.0
	Mean	5.8	4.6	7.9	7.0	5.1
	Median	6.0	4.5	8.0	7.0	5.0
	SD	2.8	2.1	2.8	3.7	2.6
Combined Sites	Min	2.0	2.0	3.0	2.0	2.0
	Max	20.0	10.0	20.0	13.0	11.0
	Mean	6.9	6.0	8.1	7.8	6.2
	Median	7.0	6.0	7.0	7.0	6.0
	SD	3.1	2.1	3.7	2.6	2.4

7.2.3 PHYTOPLANKTON

Phytoplankton is suspended and free-floating in the water column of lakes and rivers (mostly in the pelagic zone). In rivers, they are swept downstream by the longitudinal current (Wetzel, 2001). Algae are a very important component of the aquatic ecosystem, as they trap almost all the energy that is used by ecosystems (Felip & Catalan, 2000). Thus, they reflect the health and trophic status of a river through their biomass (chl-a), diversity and density (Huszar *et al.*, 1998; Guo *et al.*, 2010).

Phytoplankton are included (and settled) as part of biomonitoring programs, either through their chl-a concentration, biovolume or diversity, or all combined. In South Africa, they were included in the RHP as part of the NBPAE. Chl-a (a green photosynthetic pigment) is the dominant pigment in algal cells, but differs from genera to genera, even from species to species. Even though it fluctuates according to the light intensity, it remains a valuable indicator of the trophic status of an aquatic system (Felip & Catalan, 2000; Gregor & Maršálek, 2004; Hart, 2006).

Phytoplankton species has a rapid reproduction rate and short life cycles. It is directly affected by changes in physical and chemical factors, e.g. nutrient concentration, meteorological and hydrological conditions (Seaman *et al.*, 2001; Lin *et al.*, 2008). The sampling of phytoplankton is easy, inexpensive, rapid to quantify and creates minimal disturbance to the aquatic environment. Relatively standard methods exist for the analyses and evaluation of the chl-a concentration and algal communities (Felip & Catalan, 2000; Seaman *et al.*, 2001).

An assessment of the chl-a concentrations and phytoplanktonic algal concentrations during seasons and dry/wet periods revealed the following results. The chl-a concentration at SP had an average, statistically significant correlation with the pH ($r^2 = 0.255$; $p = <0.02$), and strong, statistically highly significant correlations with the DIN:DIP ratio ($r^2 = 0.586$; $p = <0.001$) and the total phytoplanktonic algal concentrations ($r^2 = 0.696$; $p = <0.001$). The total

phytoplanktonic algal concentrations also had strong, statistically highly significant correlations with the DIN:DIP ratio ($r^2 = 0.776$; $p = <0.001$).

At BW the strongest correlation was close to an average correlation between the chl-*a* concentration and the total phytoplanktonic algal concentration ($r^2 = 0.234$; $p = >0.1$) but was insignificant. The correlations at MC were average (statistically significant) between the total phytoplanktonic algal and diss. oxygen concentrations ($r^2 = 0.234$; $p = <0.01$). Over the total study period, the total combined data of all the sites showed an average correlation between the chl-*a* and phytoplanktonic algal concentrations ($r^2 = 0.323$; $p = <0.001$), but statistically highly significant.

With the combined summer data, the chl-*a* concentration had very weak to average correlations with pH ($r^2 = 0.273$; $p = <0.05$), DIP ($r^2 = 0.242$; $p = <0.05$) and DIN ($r^2 = 0.235$; $p = <0.05$) concentrations, and the total phytoplanktonic algal concentrations ($r^2 = 0.294$; $p = <0.02$), and moderate correlations with the Chlorophyta concentration ($r^2 = 0.382$; $p = <0.01$). The total phytoplanktonic algal concentrations had a moderate correlation with the TDS concentrations ($r^2 = 0.372$; $p = <0.01$). These were all statistically significant correlations. The combined winter data showed strong (statistically highly significant) correlations between the chl-*a* concentration and the Bacillariophyta ($r^2 = 0.564$; $p = <0.001$), and the total phytoplanktonic algal concentrations ($r^2 = 0.611$; $p = <0.001$).

During the dry period at SP, the chl-*a* concentration had a moderate correlation with the diss. oxygen ($r^2 = 0.455$; $p = <0.02$), while the phytoplanktonic algal concentrations had strong and moderate correlations with the DIP ($r^2 = 0.501$; $p = <0.02$) and TDS concentrations ($r^2 = 0.422$; $p = <0.05$), respectively. During the wet period at SP the chl-*a* concentration had moderate or strong correlations with pH ($r^2 = 0.400$; $p = <0.05$), DIN:DIP concentration ($r^2 = 0.774$; $p = <0.001$), Bacillariophyta ($r^2 = 0.907$; $p = <0.001$), Chlorophyta ($r^2 = 0.376$; $p = <0.05$) concentrations, and total phytoplanktonic algal concentrations ($r^2 = 0.908$; $p = <0.001$). The phytoplanktonic algal concentrations had moderate and strong correlations to the pH ($r^2 = 0.453$; $p = <0.05$) and DIN:DIP concentration ($r^2 =$

0.805; $p = <0.001$), respectively. The mentioned correlations were either statistically significant or statistically highly significant.

During the dry period at BW, all correlations with the aquatic factors were weak with no significant statistical probabilities, while during the wet period the chl-a concentration displayed average or strong correlations with the Bacillariophyta ($r^2 = 0.310$; $p = <0.05$), Chlorophyta ($r^2 = 0.355$; $p = <0.05$) and the phytoplanktonic algal concentrations ($r^2 = 0.500$; $p = <0.01$), all statistically significant.

During the dry period at MC the phytoplanktonic algal concentrations had an average correlation with the diss. oxygen ($r^2 = 0.349$; $p = <0.05$) and during the wet period it had a strong correlation with the TDS concentration ($r^2 = 0.558$; $p = <0.05$), both statistically significant. The chl-a concentration had strong, statistically significant correlations with the Cyanophyta ($r^2 = 0.684$; $p = <0.02$) and Chlorophyta ($r^2 = 0.544$; $p = <0.05$) concentrations during the wet period.

The combined data of the three sites showed that during the dry period the chl-a concentration had only weak to no significant correlation with all the aquatic factors. On the contrary, during the wet period the chl-a concentration had average to strong, statistically highly significant correlations with the pH ($r^2 = 0.374$; $p = <0.001$), Bacillariophyta ($r^2 = 0.576$; $p = <0.001$), Chlorophyta ($r^2 = 0.356$; $p = <0.001$), and the phytoplanktonic algal concentrations ($r^2 = 0.596$; $p = <0.001$).

The Chlorophyta was found to be the dominant division, followed by the Bacillariophyta (**Table D.14**). These two divisions had inverse patterns during the various study periods (wet and dry, and seasonally). The Chlorophyta decreased at BW and MC from the dry to the wet period, while the Bacillariophyta increased. SP displayed an inverse pattern. From the summer to winter seasons, the Chlorophyta decreased, while the Bacillariophyta increased (except at MC).

The median chl-*a* concentration was higher at BW and MC during the dry period (**Table D.15**), but higher at SP during the wet period. Regarding the seasons the median chl-*a* concentration was higher during the winter season at SP and BW, while at MC it was higher during summer. The phytoplanktonic algal concentrations followed the same pattern, except at SP, where the phytoplanktonic algal concentrations were higher during the summer.

The median phytoplanktonic chl-*a* concentration ranges for SP was mostly in eutrophic ranges, except during the summer when it was mesotrophic. For BW the medians were all in the eutrophic ranges, while at MC the medians were all mesotrophic except for the summer when it was eutrophic.

For all the sites, the total phytoplanktonic algal assemblage had 68 different algal genera from seven divisions (**Tables D.16, D.17 & D.18**). Of these algal genera, 44 were on Palmer's list (Palmer, 1969) list of 60 most organic pollutant tolerant algal genera. Of the top 25 genera on Palmer's list, only three genera were not present at the three sampling sites.

Over the total study period, there were differences in the phytoplanktonic algal assemblages between the three sites, with BW the highest with 61 genera. At SP, 36 out of a total 52 genera were found on Palmer's list (Palmer, 1969) during the study period, at BW 43 out of 61 genera; and at MC 38 out of 52 genera.

7.2.4 PERIPHYTON

Periphyton can be divided into two major groups: macroalgae, which includes filamentous strands, and microalgae, which are mostly single cells such as Bacillariophyta and other soft algae (Arnwine & Sparks, 2003). Periphyton can grow on any surface if the required energy (light and nutrients) is abundant (Vermaat, 2005). It forms the basis of many food webs, which include fish and macroinvertebrates (Biggs & Kilroy, 2000).

Bacillariophyta have been used all over the world, especially Europe, as indicators for some time (Lavoie *et al.*, 2008). In South Africa, comprehensive studies of Bacillariophyta were undertaken by Cholnoky after 1952. A large number of papers have been produced, mainly by Cholnoky, Archibald and Schoeman, and many of the papers were published internationally. In the 1990s, Bate looked at the ecological aspects of Bacillariophyta assemblages and applying the European Bacillariophyta index to South African conditions. During this period, Harding initiated further diatom studies, which produced protocol sets. After 2000, the applications of numerical indices were tested on South African rivers, which was later adapted and improved by Taylor and Harding (Bate *et al.*, 2002; Taylor *et al.*, 2007a).

Bacillariophyta indices are simply based on identifying Bacillariophyta species and applying an equation that takes its tolerance to water quality conditions and species abundance in a sample into account (Taylor *et al.*, 2007a). While Bacillariophyta indices are very site specific on water quality, it can be time consuming and costly, and it is a specialised discipline. Identification of periphyton as a group and only to genera does not need such a high degree of specialisation, while it is easy to sample and analyse pchl-a.

The periphyton (epilithic) in this study was sampled from rocks submerged (stones-in-current) in the river. The samples were identified to genus level; total concentrations and biovolumes were calculated, as were the periphytic chlorophyll-a concentrations. This data were looked at over the whole study period (**Chapter 4**), seasonal periods (summer and winter; **Chapter 5**) and greater hydrological periods (dry and wet years; **Chapter 6**) (also see **Appendix D.1 – D.3**).

When investigating the sites' data for the whole study period, it was found that at SP there were strong correlations between all the periphytic algal components (concentration, biovolume and pchl-a), while at BW and MC, both only had moderate correlations between the periphytic algal concentration and the pchl-a concentration. It was also noticed that SP had less filamentous and colonial

algal genera as part of the total periphytic algal assemblage than MC and BW. The latter had the most filamentous and colonial genera.

At all three sites, the Bacillariophyta was the dominant division and the Chlorophyta the second-most dominant division. The difference in the percentage of periphytic algal concentration was bigger between the Bacillariophyta and the other divisions than the difference between the percentages of periphytic algal biovolume.

At SP it was found that the DIN concentration had moderate correlations with both the periphytic algal concentration and the pchl-*a* concentration, while at BW and MC there were no correlation between the nutrients (DIP and DIN) and the periphytic algal components. At MC there was a strong correlation between the DIN:DIP ratio and the pchl-*a* concentration.

The seasonal data showed that the temperature, dissolved oxygen, turbidity and flow were clearly seasonal and most likely influenced each other's pattern. The pchl-*a* concentration had an inverse seasonal pattern to that of the flow and turbidity patterns; yet, it had the same seasonal pattern as the nutrients. The periphytic algal concentrations and biovolume seasonal pattern seemed to be influenced by a combination of the flow, turbidity and DIP concentrations.

The combined seasonal data of the sites showed that there were correlations between the pchl-*a* and DIP concentrations during both summer (strong) and winter (moderate), but only between the pchl-*a* and DIN concentrations during winter (moderate).

During the respective dry and wet periods, SP displayed strong correlations between all the periphytic algal components, the correlations were stronger during the wet period than the dry period. BW had only moderate correlations between the pchl-*a* and periphytic algal concentrations during the dry period. At MC, the pchl-*a* and periphytic algal concentrations displayed strong correlations during the dry period. During the wet period, there were strong correlations between all the periphytic algal components at MC.

The algal divisions were dominated by the Bacillariophyta and Chlorophyta during both the dry and wet periods, but while their concentration and biovolume decreased from the dry to the wet period, there was an increase in both the concentrations and biovolumes of the Cyanophyta and Euglenophyta. The filamentous and colonial algae (Chlorophyta and Cyanophyta) were mostly absent during the wet period or present in very low concentrations.

The DIN and DIP concentrations at SP both had strong correlations with all the periphytic algal components during the wet period. At BW, there was a moderate correlation between the DIN and pchl-a concentrations during the dry period. During the wet period, moderate and strong correlations exist between the DIN and pchl-a concentrations and the DIN concentrations and periphytic algal biovolume, respectively. MC had strong correlations between the DIN:DIP ratio and all the periphytic algal components.

The median pchl-a concentrations of SP were found to be either oligotrophic or mesotrophic (dry), BW either eutrophic (wet and summer) or hypertrophic, while MC had medians between oligotrophic and mesotrophic (wet and summer) (**Table 7.7**). According to Palmer's list of pollutant tolerant genera for phytoplankton, the periphyton genera (**Tables D.3, D.6 & D.9**) found at the three sites had only one that was not included in the top 25 genera of the list (Palmer, 1969).

7.3 DISCUSSION

7.3.1 FISH

Hydrological phases and flow regime are important in the working concept behind FRAI. The irregular and regulated flow in non-perennial and regulated rivers, respectively, causes major changes in fish habitat and the outcome of the calculated scores (**Table 7.2**). The scores can thus also be seen as a scale of

river modification and not necessarily of just the quality and health of water in a river reach, catchment or system.

7.3.2 BENTHIC MACROINVERTEBRATES

The results showed that the biotope that contributes most to the total SASS5 and ASPT scores were the SIC biotope. Strong correlations and statistically high significance (**Tables 7.3, 7.4 & 7.5**) were found throughout the dry and wet periods, as well as the different seasons of the study period. It was also found that more than 50% of the total SASS5 score comprise the SIC score (**Table D.10**). At BW, the higher flow could be the reason the SIC score comprise less of the total SASS5 score.

No profound correlation was found between the nutrient concentrations and any of the SASS5 and ASPT scores. It was found that temperature, oxygen and pH did play a role in the macroinvertebrate community as seasons or hydrological phases changed. This was also seen in the number of taxa present (**Table 7.6**) during the various seasons and hydrological phases, as well as the presence and absence of these taxa at a site (**Tables D.11, D.12 & D.13**).

Thirion (2007) lists four components effecting aquatic organisms as flow regime, physical habitat structure, water quality (e.g. temperature and dissolved oxygen), and energy inputs from a catchment (e.g. nutrients and organic matter).

This all corresponds to studies that found that macroinvertebrate indices (incl. SASS) relate more to the change in their habitat and aquatic environment function than directly to nutrient changes. The SASS5 score found the sites to be primarily poor to very poor.

7.3.3 PHYTOPLANKTON

The combined data for each site (and total) showed a weak to strong correlation between chl-*a* concentration and the total phytoplanktonic algal concentrations. The combined data for the summer and winter seasons indicated that, although weak, the nutrients did have a bigger influence on the chl-*a* concentration during the summer. During the summer the chl-*a* concentration correlated better with the Chlorophyta and during the winter with the Bacillariophyta (**Table D.14**). Bacillariophyta more likely occur during the winter and Cyanophyta during the summer (Horne & Goldman, 1994). This gave the chl-*a* concentration moderate to strong correlations with the total phytoplanktonic algal concentrations for summer and winter, respectively.

It was also found that during the wet seasons the correlations between the chl-*a* concentration and phytoplanktonic algal concentrations were better, also with the individual algal divisions, especially Bacillariophyta and Chlorophyta (**Table D.14**). The higher chl-*a* and phytoplanktonic algal concentrations during the dry period and winter (lower flow periods) are an indication that hydrology plays a role in the occurrence and growth of phytoplankton (**Table D.15**) (Lin *et al.*, 2008).

Although the nutrients have no visible correlation with the chl-*a* concentration, the median chl-*a* concentrations indicated a mesotrophic to eutrophic range for the sites (**Table 7.7**). This corresponded with the algal genera that are, according to Palmer's list (Palmer, 1969), mostly all pollutant tolerant (**Tables D.16, D.17 & D.18**). Most of the algal genera that were absent during the wet period were on Palmer's list. With the wet period, also having lower DIP concentrations than during the dry period further indicates that the nutrients have influence over the algal composition and assemblage (Lin *et al.*, 2008). BW, with the highest nutrient concentrations, had the highest number of genera present, illustrating the relationship between the nutrients and species richness (Guo *et al.*, 2010).

7.3.4 PERIPHYTON

From the results, it was found that the correlations between the various algal components are driven by the physical and chemical nature of each site, which affects the periphytic algal composition and what percentage of the algae is filamentous or colonial. The correlation between the pchl-a and periphytic algal concentrations was found to be the strongest at all the sites through both the seasonal and hydrological periods of the study periods.

No significant correlations were found between the physical (e.g. temperature, turbidity) and chemical (e.g. dissolved oxygen, pH) factors at any of the sites during any period. However, moderate to strong correlations were found between the nutrient (DIP and DIN) concentrations and some periphytic algal components.

The best correlation between the nutrient concentrations and periphytic algal components was found when there was enough flow, as found during the wet period. This eliminates the diffusion-limiting problem caused by thick mats of periphytic algae on substrates (Rier & Stevenson, 2006). During periods of high flow and sites with high flow (e.g. BW), the nutrients correlate best with the pchl-a concentration. The higher flow removed most of the filamentous and colonial periphytic algae, causing a loss of concentration and biovolume. Flow increase can also cause a shift in the periphytic algal composition (Biggs & Close, 1989).

The periphytic algal component that had the best correlations with the nutrients was the pchl-a concentration. However, it was also found that at MC only weak relationships exist between the nutrients and the pchl-a concentration. The strong correlation between the DIN:DIP ratio and the algal components during dry periods may indicate that the algal growth is frequently limited by one nutrient.

The best results linking periphytic algae to water quality and the changing of water quality over time was found with the pchl-a concentration. Chlorophyll-a concentration measures living algal biomass, whereas algal concentrations and

biovolume do not distinguish between the dead and living algal matter (Wellnitz & Poff, 2006; Kilroy *et al.*, 2013). However, the composition indicated that pollution did occur, as pollution tolerant genera were present. Sampling during very low and high flow should also be avoided to achieve optimal results.

The DIP, DIN and TDS concentrations, together with both the periphytic and phytoplanktonic chl-*a* concentrations and benthic macroinvertebrates, indicated that the three sites fell into moderate to very poor water quality ranges (**Table 7.7**), while the fish indices perceived the sites as moderately to seriously modified.

Although the mean and median of the nutrients (DIP and DIN concentrations) remained at the same trophic levels at the three sites from the dry to the wet period, the SASS scores showed a decrease, which suggests that the water quality was poorer during the wet condition or just a result of flow increase. However, the pchl-*a* concentrations indicated an improvement in quality and phytoplanktonic chl-*a* concentrations suggested no change. As both the macroinvertebrates and the periphyton decreased in numbers with the increase in flow, they result in a different indication towards the water quality trend; the combination of the two data sets will complement each other.

The increase or decrease of the means and medians of the nutrient concentrations between seasons at the three sites were corroborated by the SASS scores and both the chl-*a* concentrations. However, the pchl-*a* concentration did suggest a slightly better water quality at the sites, especially at MC, which had the lowest DIP concentrations of the three sites. The more polluted the sites were, the closer did the pchl-*a* concentrations reflect the trophic ranges of that of the nutrients.

CHAPTER 8

FINAL DISCUSSION & CONCLUSIONS

8.1 FINAL DISCUSSION

The main aim of this study was to determine the feasibility of periphyton as biomonitoring tool. This was achieved through the establishment of significant correlations between the physical, chemical and biological factors, the influences on the periphytic algae community, and during various seasons and hydrological cycles in regulated rivers. It was determine further which of the periphytic algal components (cell concentration, biovolume or pchl-a concentration) are the most accurate indicator of water quality and to establish possible water quality indicators among the periphyton community.

It was expected that the temperature and flow would have a better correlation with the periphytic algal concentration, biovolume or pchl-a concentrations. Temperature's daily and seasonal variations are important factors that determine the presence and distribution of aquatic organisms (Vannote *et al.*, 1980), as well as the concentrations and composition of periphyton (Williams & Boorman, 2012). With the regulation of rivers come changes in flow (diversion, impoundment and abstraction) that affect the temperature (Webb *et al.*, 2008).

River regulation also causes large changes in the flow regime and the effects of flow on the periphyton community. With the increase of flow in a river, the scouring effect on the periphytic algae from their substrates (Bowes *et al.*, 2012) also increases. As flow increased slightly in slow-flowing water, it dislodged loosely attached (or dead) Bacillariophyta and Cyanophyta from their substrates, leaving the algal community to be dominated by Chlorophyta and some Cyanophyta (Biggs & Close, 1989), causing a shift in the algal assemblage. As the flow increased to above 0.5 m/s, the scouring and dislodging decreased the periphytic algal numbers and biovolume (Horner *et al.*, 1990).

Sannaspos (SP) and Modder above Confluence (MC) had low median flows (0.18 and 0.27 m/s, respectively), while Bishop's Weir (BW) had higher medium flow (0.6 m/s). Thus, the different flow velocities at the various sites had different effects on their periphytic community. Further, in low-velocity streams the nutrients in the periphytic mat can become limited, but in flow higher than 0.25 m/s it overcomes the diffusion limitation (Rier & Stevenson, 2006) without excess drag, decreasing the algal mat. Therefore, at SP and MC a nutrient limiting effect could have occurred at times.

Temperature and flow can be the primary driving factors behind water clarity, nutrients, substrate availability and grazing (Horner *et al.*, 1990). With all these secondary factors working with and/or against one another, it may seem that temperature and flow have no correlation with some of the periphyton aspects like concentration and biovolume.

Seasonal patterns were found in all the physical and chemical factors. Some factors had similar patterns (e.g. flow and turbidity), while others had inverse patterns (e.g. temperature and diss. oxygen). Even though the Bacillariophyta was the dominant division during both the summer and winter seasons, the Chlorophyta and Cyanophyta increased during the summer in both concentration and biovolume.

This increase of the Chlorophyta and Cyanophyta corresponds with their affinity to warmer water (Biggs, 1996; Vis *et al.*, 1998; Flynn *et al.*, 2002), like their phytoplankton counterparts. During the summer, the Chlorophyta had the highest percentage biovolume of all the divisions, but the Bacillariophyta had the highest cell concentration, especially during the winter. The highest periphytic algal components (with one or two exceptions) were found during the winter.

In the same way that extreme climatic events (e.g. droughts and floods) can alter water quality (Delpla *et al.*, 2011), so can prolonged periods of dry and wet periods, through a river's flow regime, support different periphytic algal assemblages (Asaeda & Hong Son, 2000). So it was found that 2003–2005 was

a dry period compared with 2006–2007 with regard to the medians and means of the rainfall (SAWS, n.d.(c)) and total discharge (DWA, n.d.; in spite of river regulation) over the two periods.

As with the seasonality of the physical and chemical factors, it was found that some of the factors showed the same patterns with increases or decreases from the dry to the wet period, while other variables showed an inverse pattern (e.g. temperature and diss. oxygen). With this mentioned, not all the patterns were as expected, for example, the water temperature was higher at the sites during the wet period.

Although the flow did not show any correlation with the periphytic algal components, it was observed that during the wet period the periphytic algal concentrations were lower at all the sites, as well as the biovolumes and pchl-*a* concentrations at Sannaspos (SP) and Bishop's Weir (BW). This corresponded with the higher flows and discharges of the wet period, through scouring, which dislodged periphytic algal cells from rocks and broke filamentous algal strands (Horner *et al.*, 1990). More filamentous and colonial algae were present during the dry period, resulting in higher biovolume and pchl-*a* concentration.

Finally, it was the nutrients (DIP and DIN) that had the more significant effect on the periphyton, in particular the pchl-*a* concentration, and most probably the limiting factor at these sites, as found with various studies (Marcarelli & Wurtsbough, 2007; Hill & Fanta, 2008). With some exceptions, the periphytic algal biovolume had the weakest correlations with the physical and chemical factors, and nutrients at all the sites. This could be attributed to the regulation of the rivers, as frequent flooding and flow regulation have a huge impact on the biovolume of the periphyton, especially with regard to filamentous algae (Lavoie *et al.*, 2006), making the periphytic algal biovolume not a usable periphytic metric.

From further data analysis it was found that the various biomonitoring indices had different spatial scales and measurements of the aquatic ecosystem (**Table 8.1**), as mentioned in the literature (Griffith *et al.*, 2005; Yagow *et al.*, 2006;

Kleynhans, 2007; De la Rey *et al.*, 2008; Justus *et al.*, 2010). Each of the indicators is influenced by different factors; even periphyton and phytoplankton are affected differently by natural environmental variables (Kireta *et al.*, 2012). Flow (with other physical factors) is an overall, direct, system influence, whereas nutrients (food source) influence the algae directly.

Table 8.1: The spatial scales and measurements of each biomonitoring indicator.

Indicator	Spatial scale	Measurement	Major strengths
Fish (FRAI)	Catchment or river reach (site non-specific index)	Health and integrity of a river, and the habitat availability	The ability to integrate the long-term effect of the environment through a life cycle over multiple years
Macroinvertebrates (SASS5)	Ecosystem and habitat (site specific index)	Change in the habitat and function of the aquatic environment	Abundance in most streams and a life cycle from weeks to a year
Phytoplankton	Upstream from sampling site (site non-specific index)	Change in water quality, physical and chemical factors	Rapid response to environmental changes through rapid reproduction rates and short lives
Periphyton	Upstream and at sampling site (site specific index – more local)		

8.2 CONCLUSION

This study examined the periphyton (diatom and soft, non-diatom periphytic algae) with the aim of using them as indicators of water quality. In this study, it was found that:

- ◆ Of the three periphytic algal components investigated, the algal concentration and chlorophyll-a had strong correlations with each other

- ◆ for each of the sites and the combined data,
- ◆ during the different seasons, and
- ◆ during different hydrological conditions.
- ◆ Of the physical and chemical conditions, the nutrients had the best statistical significant correlations with the periphytic algal components
 - ◆ for each of the sites and the combined data,
 - ◆ during the different seasons, and
 - ◆ during different hydrological conditions.
- ◆ The strongest correlations were found between the nutrients and the periphytic chlorophyll-*a* concentrations.
- ◆ Bacillariophyta was the dominant division present in the periphyton assemblage, followed by the Chlorophyta, but
 - ◆ during the summer the percentage Chlorophyta increased in concentration and biovolume, and
 - ◆ during lower flow conditions the percentage Chlorophyta increased in concentration and biovolume.
- ◆ The change in seasons and hydrological regime not only changes the division composition of periphyton species, but also the algal divisions' composition.
- ◆ It is clear from this study of the three periphytic metrics investigated that the periphytic chlorophyll-*a* concentration is the best periphytic measure to use in regulated rivers.

Furthermore, if the periphytic chlorophyll-*a* concentration is combined with the number of taxa present in the periphytic assemblage, especially if they are noted to be pollution-tolerant or not, as in Palmer's Index (Palmer, 1969), it strengthens the result and outcome, making periphyton a contributor in the field of water quality indicators. There are Cyanophyta that produces toxins, which can affect the other benthic organisms negatively; this knowledge could be important in the accurate interpretation of the benthic macroinvertebrate data.

The biomonitoring indices used at the three study sites, Sannaspos, Bishop's Weir and Modder above Confluence, confirm the following. The combined data for each site indicate that the fish indices perceived the reaches as moderately to

seriously modified. The SASS5 scores (macroinvertebrate index) indicated that the sites have poor water quality. The phytoplanktonic and periphytic chl-*a* concentration, respectively, indicated Sannaspos as eutrophic and mesotrophic (poor and fair water quality), Bishop's Weir as eutrophic and eu-hypertrophic (poor and very poor water quality) and Modder above Confluence as mesotrophic and oligo-mesotrophic (fair water quality).

According to the nutrients, Bishop's Weir was the site with the poorest water quality of the three sites and Modder above Confluence the best water quality. The periphytic algal genera at the three sites did indicate pollution, as most of the genera found are pollutant tolerant. The presence of the Cyanophyta (especially *Oscillatoria*) could have had a negative impact on the fish and macroinvertebrates grazing on the periphyton.

While it was accepted that all biotic interactions respond directly to or are influenced by community structure and function, a single index would not always give the most complete description of a biotic community, particularly in regulated rivers. Consequently, it was found that that by using periphyton in conjunction with other biomonitoring indices (fish, benthic macroinvertebrates, phytoplankton and nutrients) it would give a more comprehensive understanding of the biotic interactions, the trophic level and health of an aquatic ecosystem.

Therefore, it is recommended that a suite of biomonitoring indices be used in conjunction with each other, including, if possible, periphyton as well.

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APPENDIX A

STUDY SITES

The 1:50 000 Topographic Map Sheets of the study sites also show captured parts for each study site, as indicated in **Chapter 3.2: Study sites**. Coordinates are given as Lat/Lon hddd°mm'ss.s" (WGS84).

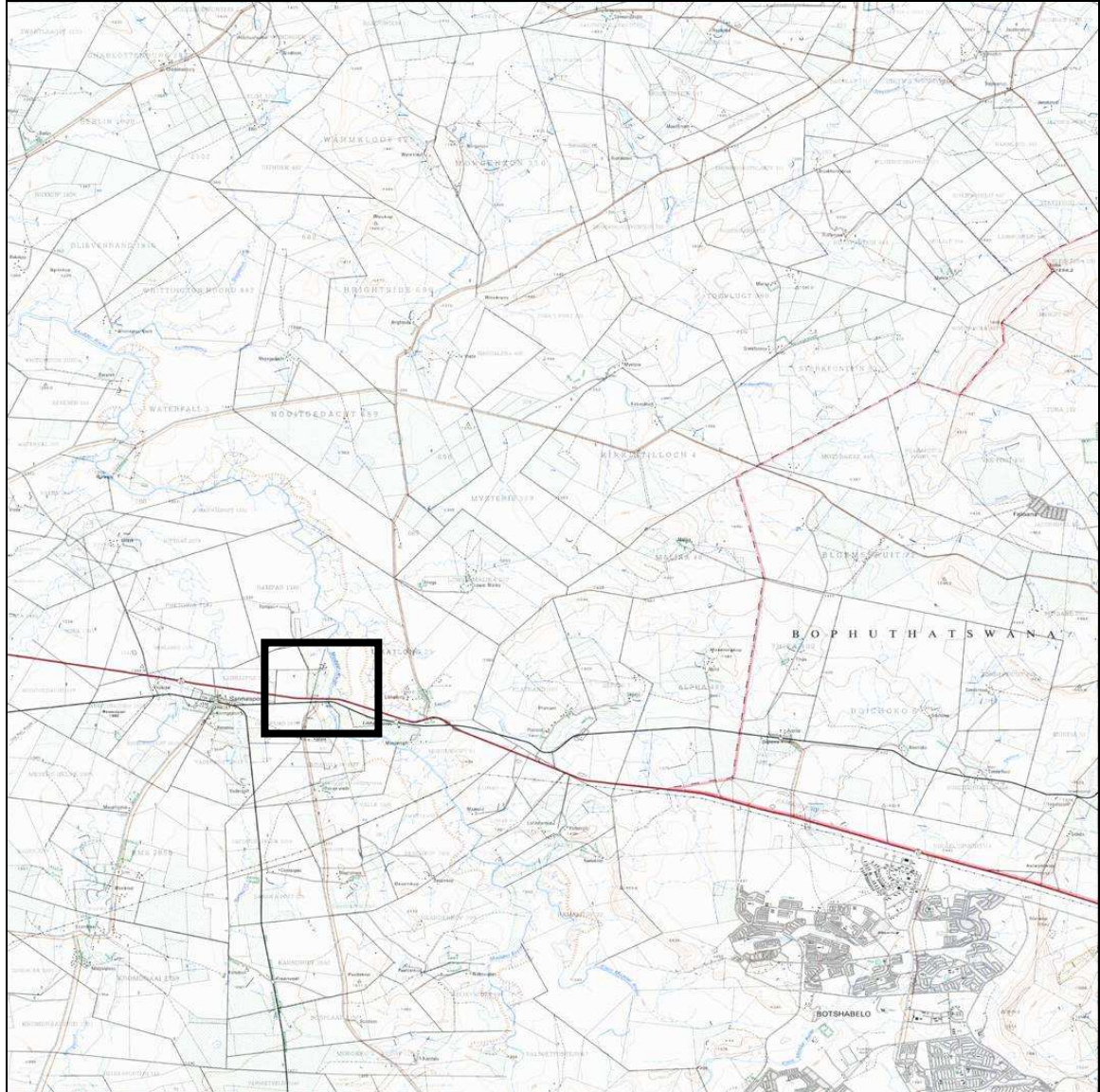
A.1 MODDER RIVER**A.1.1 SANNASPOS (-29°09'40.0"S, 26°34'19.0"E)**

Figure A.1: 1:50 000 Topographic Map Sheet no: 2926BA Sannaspos (CD:SM, 2002).
(Scale of map: 15 minutes on latitudinal and longitudinal axis).

A.1.2 MODDER ABOVE CONFLUENCE (-29°01'42.0"S, 24°38'18.0"E)

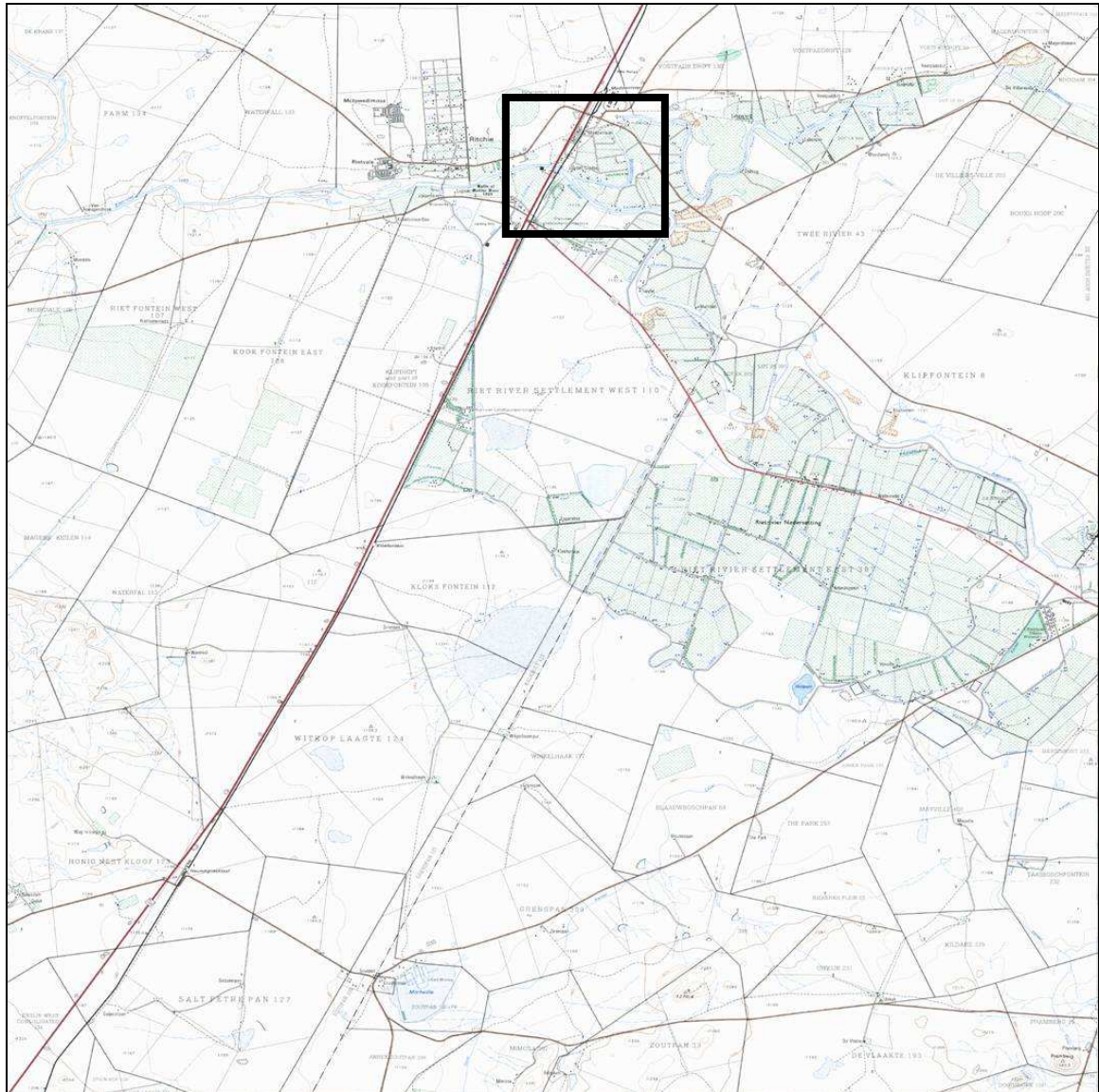


Figure A.2: 1:50 000 Topographic Map Sheet no: 2924BA Modderrivier (CD:SM, 2002).

(Scale of map: 15 minutes on latitudinal and longitudinal axis).

A.2 RENOSTER SPRUIT

A.2.1 BISHOP'S WEIR (-28°58'00.0"S, 26°19'09.0"E)

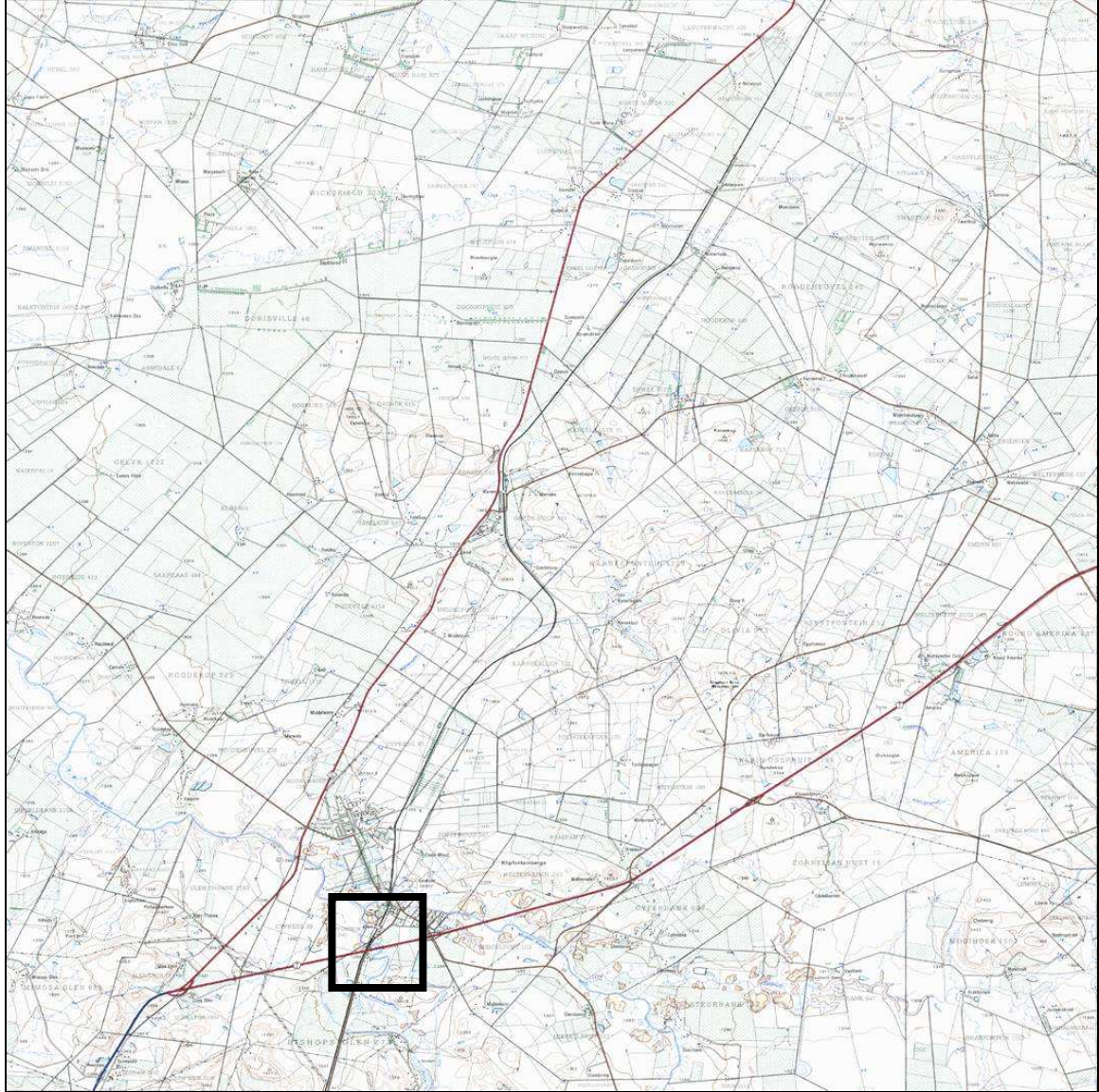


Figure A.3: 1:50 000 Topographic Map Sheet no: 2826CD Glen (CD:SM, 2002). (Scale of map: 15 minutes on latitudinal and longitudinal axis).

APPENDIX B

STUDY SITE PHOTOGRAPHS

Photographs of study sites are shown here. They include normal flow conditions, low flow and/or dry conditions, high flow and/or flood conditions, as well as photos of algae on rocks and rock that were sampled throughout the entire study period, as indicated in **Chapter 3.2: Study sites**.

All photographs were taken by either the author/researcher or Marie Watson.

B.1 MODDER RIVER

B.1.1 SANNASPOS



Figure B.1: Sannaspos during “normal” flow conditions.



Figure B.2: Sannaspos during dry/no flow conditions.



Figure B.3: Sannaspos during higher flow conditions.



Figure B.4: Sannaspos during flood conditions (water release from Rustfontein Dam).

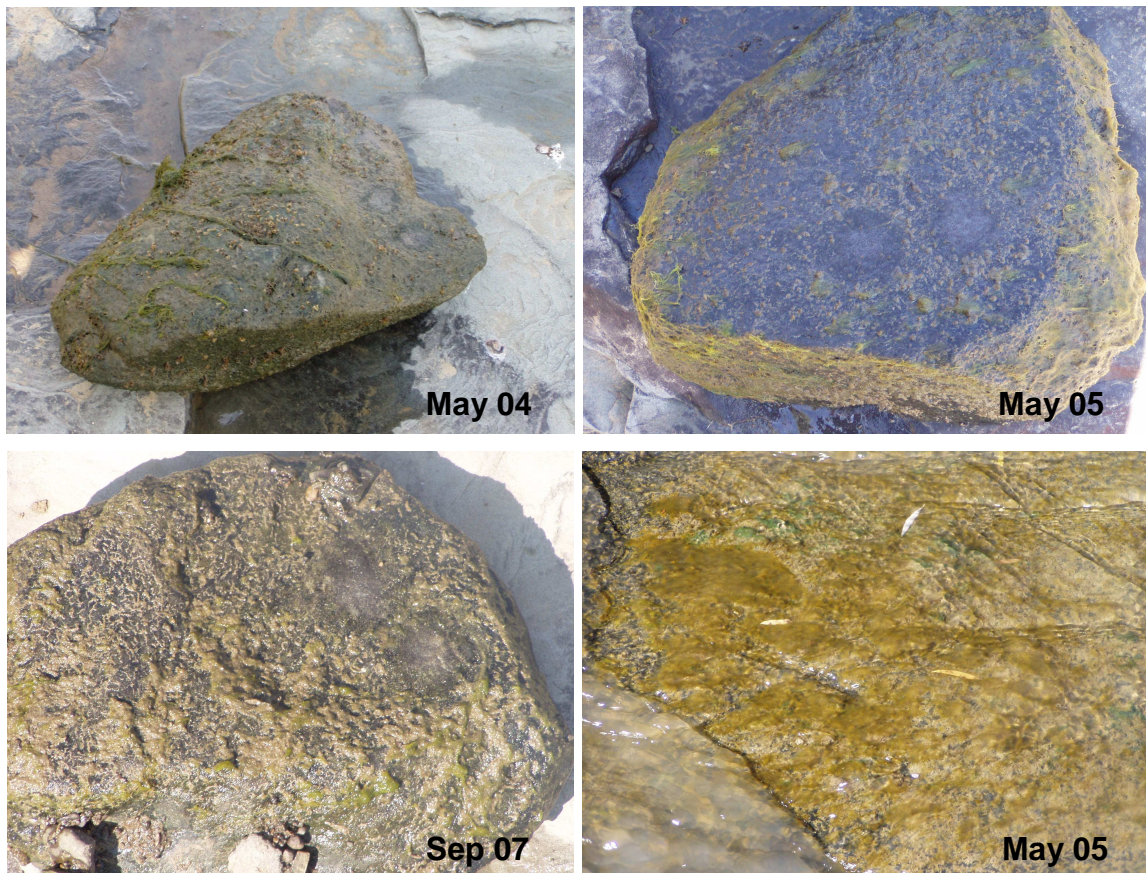


Figure B.5: Rocks and bedrock covered with periphyton at Sannaspos.



Figure B.6: Bedrock covered with filamentous algae at Sannaspos.

B.1.2 MODDER ABOVE CONFLUENCE



Figure B.7: Modder above Confluence during “normal” flow conditions.



Figure B.8: Modder above Confluence during low flow conditions.



Figure B.9: Modder above Confluence during dry/no flow conditions.



Figure B.10: Modder above Confluence during high flow conditions.

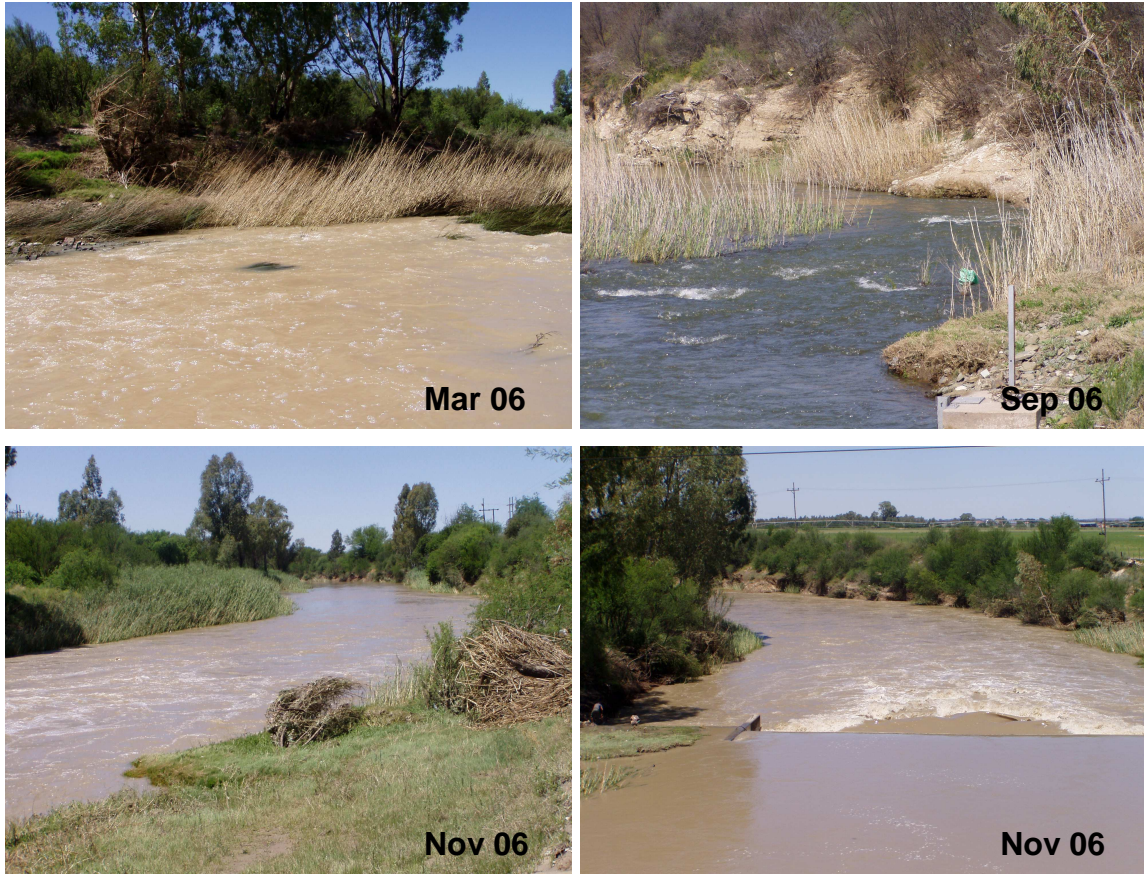


Figure B.11: Modder above Confluence during flood conditions.



Figure B.12: Rocks covered with periphyton at Modder above Confluence.

B.2 RENOSTER SPRUIT

B.2.1 BISHOP'S WEIR



Figure B.13: Bishop's Weir during "normal" flow conditions.



Figure B.14: Bishop's Weir during low flow conditions.



Figure B.15: Bishop's Weir during high flow conditions.



Figure B.16: Rocks covered with periphyton at Bishop's Weir.

APPENDIX C

MATERIALS & METHODS

Photographs of some equipment used for the measurement of biological characteristics for this study as indicated in **Chapter 3.3: Materials & Methods**.

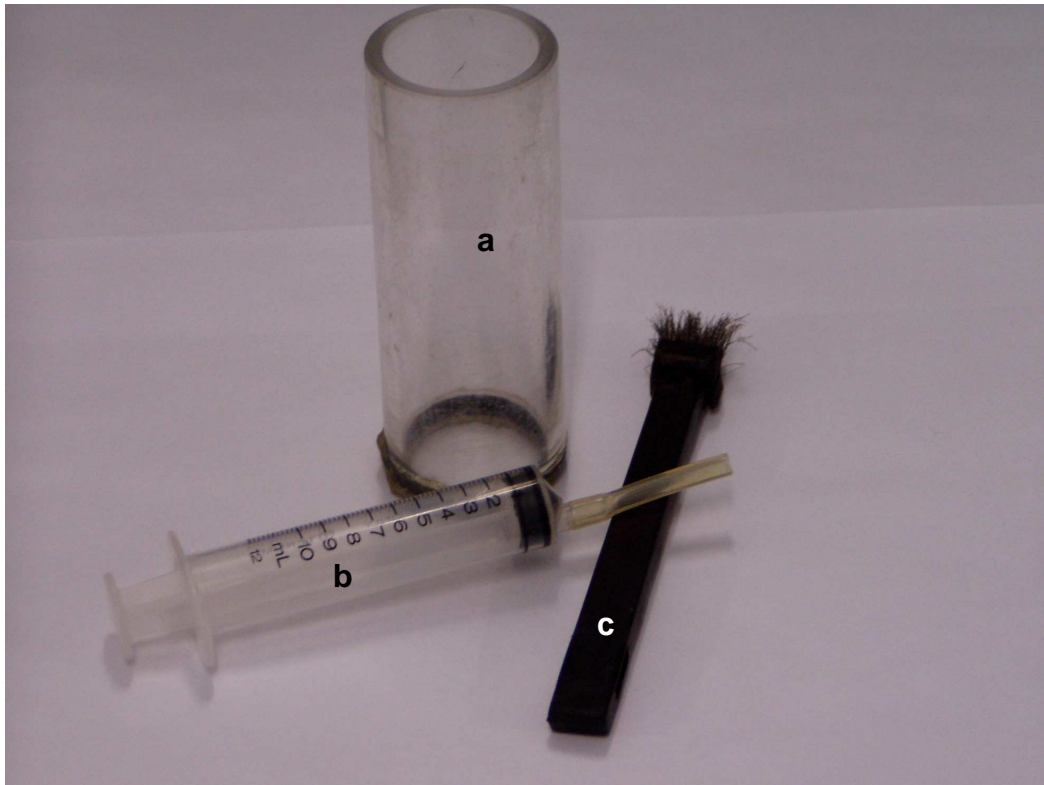


Figure C.1: Periphyton sampling equipment – (a) Perspex tube with O-ring (diameter of 3.09 cm), (b) syringe with rubber tube and (c) stainless steel bristle brush.

Diameter of chamber: 16,450 μm
 Surface of chamber bottom: 212,610,000 μm^2

Dimensions of grid (L and W) in μm :

Objective	1	2	3
10.0x	870	87	17.4
16.0x	540	54	10.8
40.0x	220	22	4.4

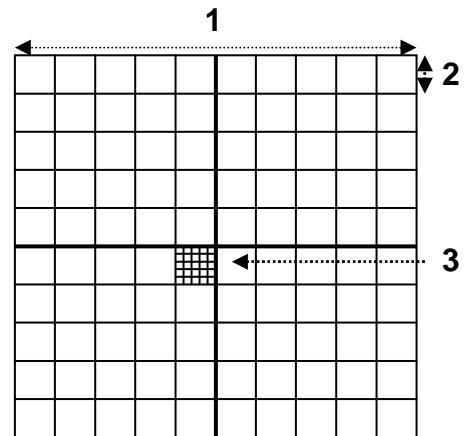


Figure C.2: Inverted Zeiss Light Microscope sedimentation chamber and grid dimensions.

Version date: Sep 2005

SASS Version 5 Score Sheet


Date (dd-mm-yr): Site Code: Collector/Sampler: River: Level 1 Ecoregion: Quaternary Catchment:	Grid reference (dd mm ss.s) Lat: S Datum (WGS84/Cape): Altitude (m): Zonation: Routine or Project? (circle one) Project Name: Flow: Clarity (cm): Turbidity: Colour:	Biotopes Sampled (tick & rate) Rating (1 - 5) Time (min)	
Temp (°C): pH: DO (mg/L): Cond (mS/m): Riparian Disturbance: Instream Disturbance:	Taxon QV S Veg GSM TOT PORIFERA (Sponge) COELENTERATA (Cnidaria) TURBELLARIA (Flatworms) ANNELIDA Hirudinea (Earthworms) Hirudinea (Leeches) CRUSTACEA Amphipoda (Scuds) Potamonautidae* (Crabs) Aysidae (Freshwater Shrimps) Palaemonidae (Freshwater Prawns) HYDRACARINA (Mites) PLECOPTERA (Stoneflies) Notonemouridae Perlidae EPHEMEROPTERA (Mayflies) Baetidae 1 sp Baetidae 2 sp Baetidae > 2 sp Caenidae (Squaregills/Cairflies) Ephemeridae Heptageniidae (Flatheaded mayflies) Leptophlebiidae (Pronghills) Oligoneuridae (Brushlegged mayflies) Poliancyidae (Pale Burrowers) Prosoptomalidae (Water specks) Telepsopidae SMC (Spry Crawlers) Tricorythidae (Stout Crawlers) ODONATA (Dragonflies & Damselflies) Golepterygidae ST,T (Demoselles) Chlorocyphidae (Jewels) Synlestidae (Chlorostidae) (Syphs) Coenagrionidae (Sprites and blues) Lesidae (Emerald Damselflies/Spreadwings) Platycentridae (Stream Damselflies) Protoneridae (Threshwings) Aeshnidae (Hawkers & Emperors) Corduliidae (Cruisers) Gomphidae (Clubtails) Libellulidae (Darters/Skimmers) LEPIDOPTERA (Aquatic Caterpillars/Moths) Crambidae (Pyralidae)	Taxon QV S Veg GSM TOT DIPTERA (Flies) Athericidae (Shipe flies) Belostomatidae (Giant water bugs) Corixidae* (Water boatmen) Gerridae* (Pond skaters/Water striders) Hydrometridae* (Water measurers) Naucloretidae* (Creeping water bugs) Nepidae* (Water scorpions) Notonemidae* (Backswimmers) Pleidae* (Pygmy backswimmers) Veliidae/M. velidae* (Ripple bugs) MEGALOPTERA (Fishflies, Dobsonflies & Alderflies) Corydalidae (Fishflies & Dobsonflies) Sialidae (Alderflies) TRICHOPTERA (Caddisflies) Dipseuropsidae Ecnomidae Hydroptilidae 1 sp Hydroptilidae 2 sp Hydroptilidae > 2 sp Philopotamidae Polycentropodidae Psychomyiidae/Xiphocentronidae Caseid caddis: Barbatrochontidae SWC Calamoceratidae ST Glossosomatidae SWC Hydroptilidae Hydroscaphidae SWC Lepidostomatidae Leptoceridae Petrohrinidae SWC Pisulidae Sericosomatidae SWC COLEOPTERA (Beetles) Dytiscidae/Notidae* (Diving beetles) Elmidae (Dytiscidae* (Rifle beetles) Gyrinidae* (Whirligig beetles) Halpidae (Crawling water beetles) Helocidae (Marsh beetles) Hydraenidae (Minute moss beetles) Hydrophilidae (Water scavenger beetles) Limnichidae (Marsh-Loving Beetles) Psephenidae (Water Pennies)	Taxon QV S Veg GSM TOT Gastropoda (Snails) Anoploleidae (Limpets) Burinidae* Hydrobiidae* Lymnaeidae* (Pond snails) Physidae* (Pouch snails) Planorbinae* (Olb snails) Thiardidae* (=Melanidae) Viviparidae* ST PELECYPODA (Bivalves) Corbiculidae (Clams) Sphaeriidae (Pill clams) Unionidae (Pearly mussels) SASS Score No. of Taxa Other biota: Comments/Observations:

Figure C.3: South African Scoring System version 5 (SASS5) scoring sheet.

Procedure: Kick SIC & bedrock for 2 mins. max. 5 mins. Kick SOOC & bedrock for 1 min. Sweep marginal vegetation (IC & COC) for 2m total and aquatic veg 1m². Stir & sweep gravel, sand, mud for 1 min total. Hand picking & visual observation for 1 min - record in biotope where found (by circling estimated abundance on score sheet). Score for 15 mins biotope but stop if no new taxa seen after 5 mins. Estimate abundances: 1 = 1, A = 2-10, B = 10-100, C = 100-1000, D = >1000. S = Stone, rock & solid objects; Veg = Vegetation; GSM = Gravel, sand, mud. * = airbreathers. Rate each biotope sampled, every poor (de limited diversity), 5=highly suitable (i.e. wide diversity). Rate colour: transparent, tea brown, light brown, dark brown, light green, dark green, yellow, red, grey, milky white, black. Rate flows: Zero, trickle, low, medium, high, flood.

APPENDIX D

RESULTS

Tables showing additional processed data for **Chapters 4–7** not mentioned in the respective chapters' result sections.

D.1 ADDITIONAL PROCESSED DATA FOR CHAPTER 4

Table D.1: Additional minimum, maximum, mean, median and standard deviation of the physical, chemical and standard deviation of the physical, chemical and algal data over the study period for each site and the three sites combined.

	Temperature (°C)	Diss. oxygen (mg/l)	Redox (mV)	pH	Turbidity (NTU)	Flow (m/s)	DIP (mg/l)	DIN (mg/l)	TDS (mg/l)	DIN:DIP	Pchl-a (µg/cm ²)	Periphytic algal	
												Conc.	Biovol.
Sannaspos	Min	1.65	-117.20	7.15	8.30	0.04	0.001	0.079	108.12	0.12	0.013	4,401.0	161,356,526.5
	Max	24.90	12.34	-11.60	9.24	460.00	1.68	4.010	1,844	547.45	169.11	20.177	11,404,348.0
	Mean	16.06	5.71	-58.32	8.12	72.86	0.27	0.575	0.580	321.36	16.38	3.853	1,225,999.5
	Median	16.00	5.53	-54.60	8.00	28.00	0.18	0.111	0.499	300.65	4.29	2.064	480,338.0
	SD	6.00	2.89	29.72	0.60	107.05	0.36	1.056	0.479	131.59	38.88	5.077	2,450,915.1
Bishop's Weir	Min	9.00	3.91	-130.40	7.57	2.80	0.11	2.050	221.55	0.02	0.460	199,148.0	950,634,732.5
	Max	27.30	11.41	-7.05	9.26	51.00	3.95	15.300	620.10	1.33	66.640	30,749,458.0	421,346,390,442.9
	Mean	18.51	6.83	-68.16	8.15	15.60	0.79	8.039	405.91	0.54	21.972	5,751,277.4	40,804,933,755.6
	Median	19.50	6.86	-65.90	8.14	11.90	0.60	7.370	402.50	0.60	18.495	2,647,146.0	14,724,358,301.8
	SD	5.65	2.00	29.28	0.43	11.70	0.86	3.705	3.122	111.25	0.33	17.494	7,585,916.6
Modder above Confluence	Min	9.50	2.22	-84.10	7.23	6.00	0.00	0.001	145.19	0.42	0.010	123,022.0	426,502,686.4
	Max	27.10	11.80	-17.80	8.20	68.00	0.69	0.194	700.21	3044.70	24.990	5,358,879.0	109,848,334,934.7
	Mean	19.15	6.73	-47.43	7.68	15.57	0.28	0.047	419.12	334.09	3.929	948,985.7	13,477,557,046.1
	Median	19.50	6.37	-49.80	7.68	14.00	0.27	0.010	401.26	82.40	1.950	445,207.0	3,838,543,797.5
	SD	5.95	2.07	18.87	0.32	13.60	0.26	0.057	154.39	716.06	5.861	1,310,923.8	26,011,665,247.7
Combined Sites	Min	6.10	1.65	-130.40	7.15	2.80	0.00	0.001	108.12	0.02	0.010	4,401.0	161,356,526.5
	Max	27.30	12.34	-7.05	9.26	460.00	3.95	15.300	700.21	3044.70	66.640	30,749,458.0	421,346,390,442.9
	Mean	17.90	6.44	-58.92	8.00	34.09	0.47	3.291	382.45	103.16	10.844	2,885,517.7	20,820,478,160.4
	Median	19.30	6.37	-55.20	7.90	16.20	0.33	0.182	380.29	1.33	4.390	901,462.0	4,632,350,840.2
	SD	5.91	2.36	27.75	0.50	66.43	0.63	4.455	136.37	408.78	14.529	5,400,494.3	55,809,772,158.2

Table D.2: The mean and median periphytic algal concentration:biovolume ratio over the study period for each site and the three sites combined.

		Periphytic algal divisions (%)			
		Cyanophyta	Bacillariophyta	Chlorophyta	Euglenophyta
SP	Mean	1.532	1.686	0.662	0.065
	Median	1.376	2.153	1.276	0.034
BW	Mean	1.270	1.560	0.726	0.079
	Median	2.283	1.958	0.458	0.047
MC	Mean	2.205	1.702	0.526	0.163
	Median	4.261	2.003	0.385	0.128
3*	Mean	1.594	1.641	0.643	0.098
	Median	2.499	2.046	0.526	0.056

* = Combined sites

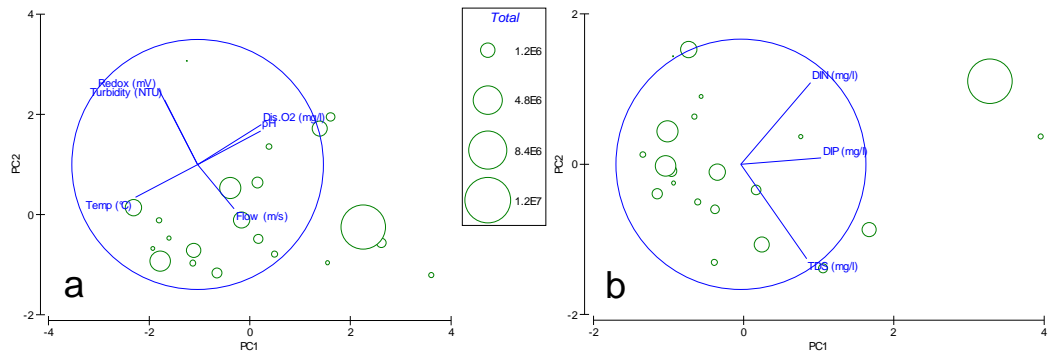


Figure D.1: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Sannaspos (SP) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal concentration (cells/cm²).

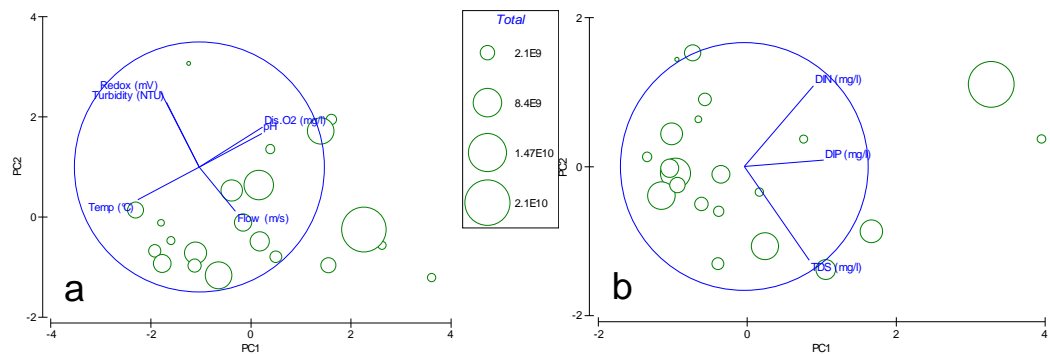


Figure D.2: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Sannaspos (SP) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal biovolume (cm³/cm²).

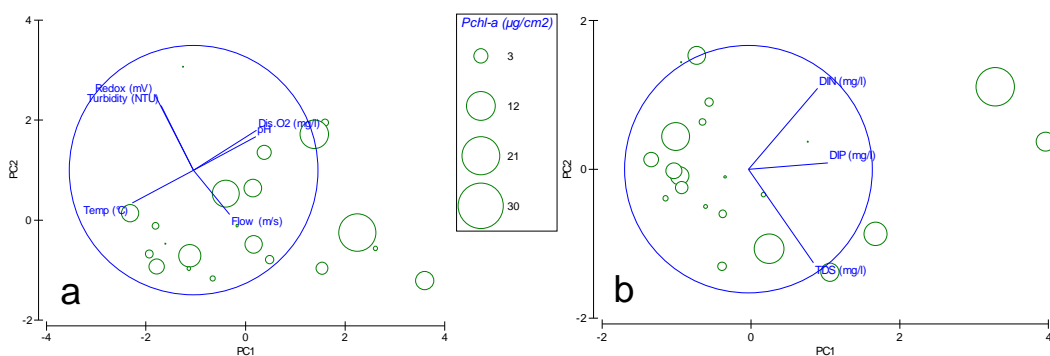


Figure D.3: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Sannaspos (SP) over the period from May 2003 to November 2007. The bubble overlay is the periphytic chlorophyll-a concentration (µg/cm²).

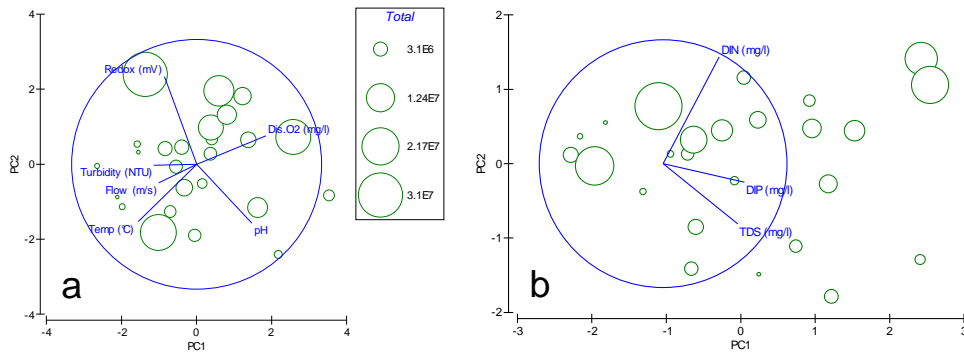


Figure D.4: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Bishop's Weir (BW) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal concentration (cells/cm²).

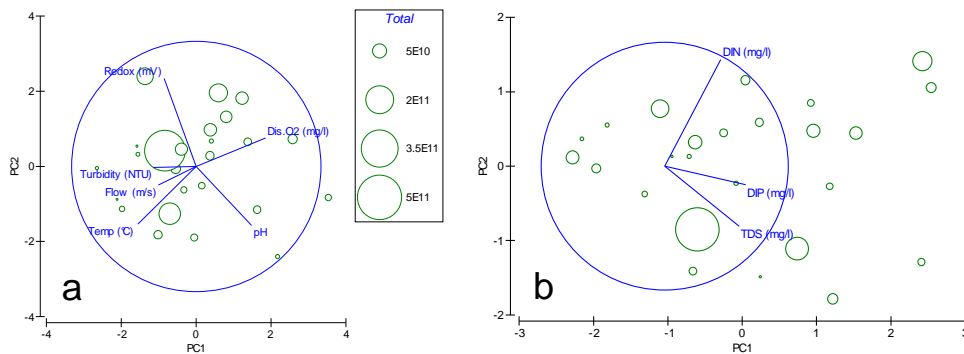


Figure D.5: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Bishop's Weir (BW) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal biovolume (cm³/cm²).

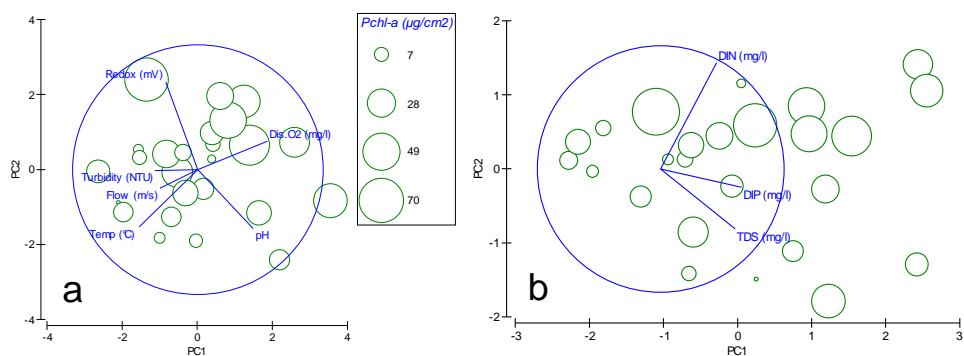


Figure D.6: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Bishop's Weir (BW) over the period from May 2003 to November 2007. The bubble overlay is the periphytic chlorophyll-a concentration (µg/cm²).

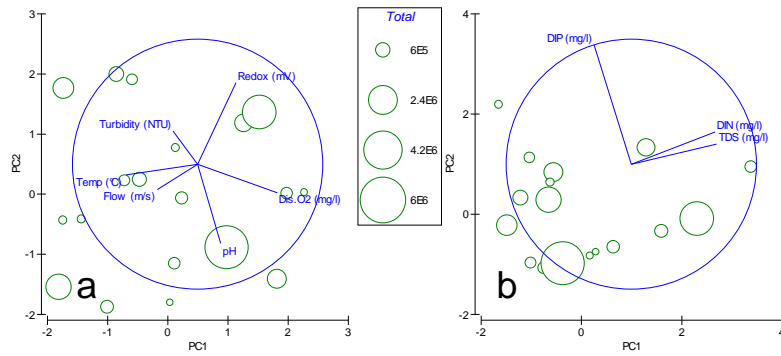


Figure D.7: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal concentration (cells/cm²).

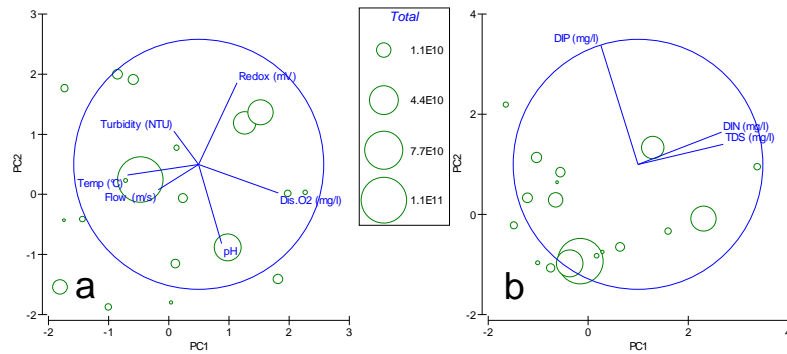


Figure D.8: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal biovolume (cm³/cm²).

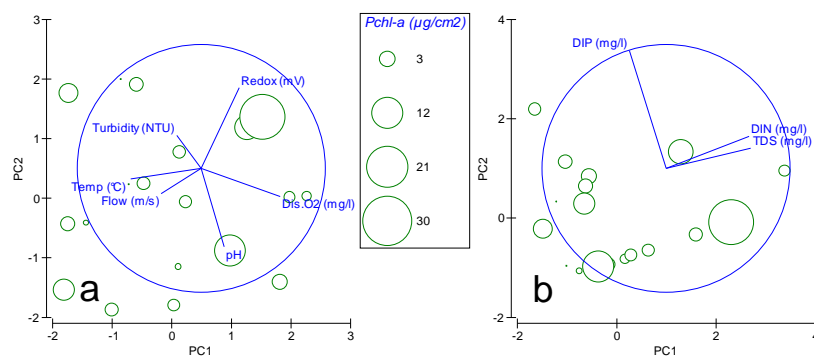


Figure D.9: The PCA graphs: (a) physical and chemical and (b) nutrient factors at Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the periphytic chlorophyll-a concentration (µg/cm²).

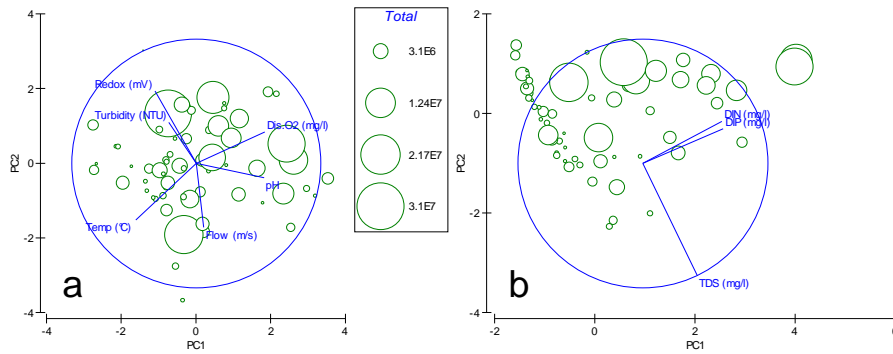


Figure D.10: The combined PCA graphs: (a) physical and chemical and (b) nutrient factors of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal concentration (cells/cm^2).

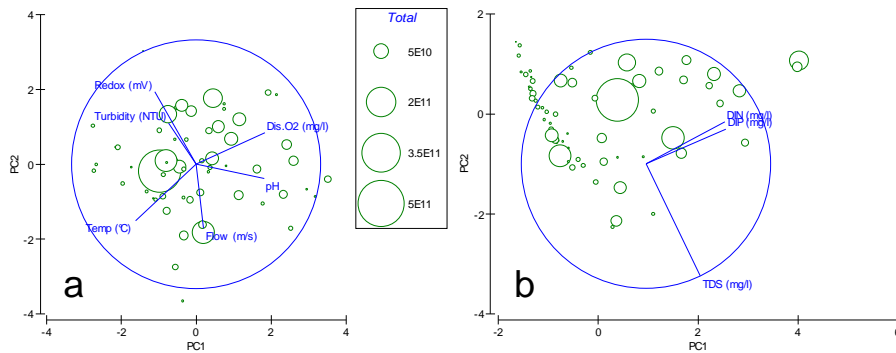


Figure D.11: The combined PCA graphs: (a) physical and chemical and (b) nutrient factors of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the total periphytic algal biovolume (cm^3/cm^2).

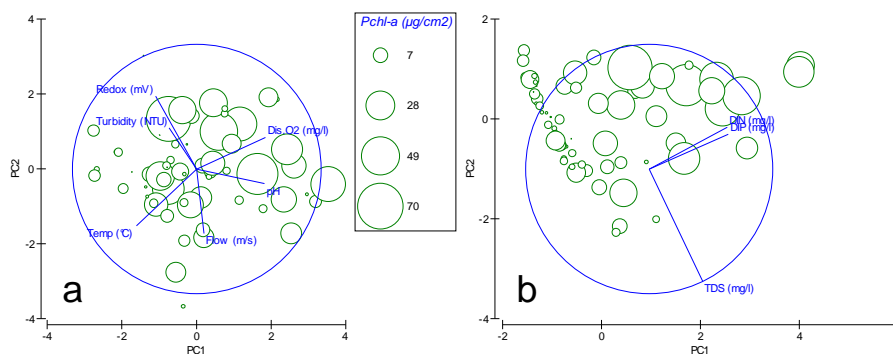


Figure D.12: The combined PCA graphs: (a) physical and chemical and (b) nutrient factors of Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC) over the period from May 2003 to November 2007. The bubble overlay is the periphytic chlorophyll-a concentration ($\mu\text{g}/\text{cm}^2$).

D.2 ADDITIONAL PROCESSED DATA FOR CHAPTER 5

Table D.4: Additional minimum, maximum, median and standard deviation of the physical, chemical and standard deviation of the physical, chemical and algal data for the summer and winter seasons for each site and the three sites combined.

	Temperature (°C)	Diss. oxygen (mg/l)	Redox (mV)	pH	Turbid- ity (NTU)	Flow (m/s)	DIP (mg/l)	DIN (mg/l)	TDS (mg/l)	DIN:DIP	Pchl-a (µg/cm ²)	Periphytic algal		
												Conc.	Biovol.	
Summer	Min	19.40	1.65	-62.50	7.48	19.10	0.08	0.040	0.12	196.63	0.22	0.013	84,963.0	634,502,756.6
	Max	24.90	5.26	-36.80	7.79	128.00	0.60	1.280	1.18	547.45	6.48	7.320	2,378,005.0	4,890,973,961.3
	Mean	22.84	2.92	-50.26	7.62	46.82	0.23	0.449	0.63	336.20	3.55	2.369	787,818.2	2,400,326,832.7
	Median	23.30	2.48	-54.60	7.62	28.00	0.17	0.182	0.73	300.65	4.29	0.920	210,922.0	1,725,145,991.7
	SD	2.07	1.48	10.42	0.13	45.62	0.21	0.505	0.44	139.75	2.95	3.081	998,057.6	1,671,207,694.4
	Min	20.70	3.91	-93.60	7.70	2.80	0.11	3.380	0.55	221.55	0.06	0.460	199,148.0	950,634,732.5
	Max	27.30	6.75	-44.50	8.40	30.00	3.95	13.401	4.50	568.65	0.60	33.860	20,402,129.0	421,346,390,442.9
	Mean	23.69	5.17	-70.29	8.02	15.79	1.19	8.280	1.81	408.13	0.26	15.336	4,216,358.0	75,081,643,803.1
	Median	23.85	5.21	-67.30	8.02	15.85	0.62	8.540	1.29	401.80	0.24	13.435	2,389,614.5	13,609,015,230.6
	SD	2.05	0.85	17.63	0.24	10.02	1.36	3.711	1.32	129.02	0.18	11.746	6,713,657.0	144,899,562,945.7
	Min	23.80	2.22	-71.40	7.30	6.50	0.05	0.001	0.17	145.19	3.15	0.310	187,437.0	426,502,686.4
	Max	27.10	6.37	-27.80	8.20	16.20	0.67	0.093	0.52	403.35	280.00	5.541	1,854,777.0	11,344,190,447.0
	Mean	25.76	5.09	-54.88	7.70	12.28	0.37	0.045	0.32	327.88	93.17	2.622	767,305.6	3,986,434,518.0
	Median	26.10	5.59	-55.20	7.70	14.00	0.41	0.056	0.28	395.32	9.33	2.450	388,616.0	2,671,659,264.4
	SD	1.30	1.72	17.80	0.37	3.78	0.30	0.042	0.14	111.95	126.19	2.338	742,489.4	4,301,544,956.4
Min	19.40	1.65	-93.60	7.30	2.80	0.05	0.001	0.12	145.19	0.06	0.013	84,963.0	426,502,686.4	
Max	27.30	6.75	-27.80	8.40	128.00	3.95	13.401	4.50	568.65	280.00	33.860	20,402,129.0	421,346,390,442.9	
Mean	24.03	4.52	-60.44	7.82	23.43	0.70	3.817	1.07	365.86	26.98	8.203	2,305,915.7	35,143,719,843.3	
Median	23.85	5.01	-58.85	7.80	17.65	0.42	0.926	0.79	379.58	0.64	4.316	861,058.0	3,781,954,801.6	
SD	2.12	1.60	17.70	0.31	27.56	1.00	4.757	1.12	126.18	74.40	10.171	4,692,045.8	100,009,176,498.0	
Min	6.10	4.90	-117.20	7.49	10.70	0.04	0.004	0.11	187.93	0.68	0.080	180,411.0	678,078,198.2	
Max	13.50	8.46	-16.20	9.24	131.00	0.45	2.720	1.84	446.63	169.11	20.177	11,404,348.0	20,302,617,660.2	
Mean	10.35	6.61	-61.85	8.36	48.23	0.19	0.441	0.53	324.73	24.67	5.042	2,186,057.4	5,093,767,677.8	
Median	11.25	6.31	-52.16	8.30	29.00	0.14	0.090	0.34	332.74	2.09	1.957	600,400.5	2,205,262,908.3	
SD	2.92	1.45	34.33	0.64	48.52	0.16	0.928	0.57	102.56	58.58	7.038	3,811,245.6	6,738,739,591.7	
Min	9.00	6.86	-130.40	7.57	6.90	0.16	4.840	3.71	292.49	0.54	2.290	966,039.0	4,307,160,456.2	
Max	16.60	10.64	-7.05	9.26	35.00	1.65	14.200	11.42	505.87	1.07	66.640	30,749,458.0	83,010,616,328.3	
Mean	12.54	8.14	-62.36	8.20	13.96	0.70	8.874	6.84	408.55	0.78	32.002	10,737,989.9	36,330,102,145.9	
Median	12.30	7.73	-63.95	8.18	10.60	0.67	7.400	6.39	412.20	0.77	28.851	7,345,935.0	30,240,996,470.5	
SD	2.82	1.35	38.69	0.50	9.07	0.45	3.443	2.84	72.97	0.18	21.479	10,199,966.9	27,554,779,770.3	
Min	9.50	5.92	-66.70	7.30	6.00	0.02	0.001	0.08	224.60	0.42	0.010	123,022.0	555,182,304.2	
Max	19.50	9.19	-17.80	8.14	68.00	0.69	0.194	1.67	669.77	551.35	11.884	5,358,879.0	109,848,334,934.7	
Mean	14.56	7.13	-45.26	7.61	20.26	0.40	0.049	0.53	397.29	162.76	3.245	1,031,169.1	22,967,574,415.6	
Median	14.15	6.74	-46.13	7.48	15.25	0.41	0.006	0.35	406.12	54.37	1.795	457,493.5	3,277,220,387.2	
SD	3.59	1.29	15.59	0.31	20.07	0.21	0.071	0.54	142.82	216.31	4.196	1,770,335.7	37,886,320,437.6	
Min	6.10	4.90	-130.40	7.30	6.00	0.02	0.001	0.08	187.93	0.42	0.010	123,022.0	555,182,304.2	
Max	19.50	10.64	-7.05	9.26	131.00	1.65	14.200	11.42	669.77	551.35	66.640	30,749,458.0	109,848,334,934.7	
Mean	12.48	7.29	-56.49	8.06	27.48	0.43	3.121	2.63	376.86	62.74	13.429	4,651,738.8	21,463,814,746.4	
Median	12.25	7.04	-52.85	8.00	15.25	0.39	0.140	0.67	391.39	1.36	3.735	933,750.5	7,001,341,238.5	
SD	3.47	1.46	30.89	0.58	33.10	0.36	4.601	3.45	111.67	143.54	18.476	7,523,116.7	29,199,700,945.6	
Min	9.00	6.86	-130.40	7.57	6.90	0.16	4.840	3.71	292.49	0.54	2.290	966,039.0	4,307,160,456.2	
Max	16.60	10.64	-7.05	9.26	35.00	1.65	14.200	11.42	505.87	1.07	66.640	30,749,458.0	83,010,616,328.3	
Mean	12.54	8.14	-62.36	8.20	13.96	0.70	8.874	6.84	408.55	0.78	32.002	10,737,989.9	36,330,102,145.9	
Median	12.30	7.73	-63.95	8.18	10.60	0.67	7.400	6.39	412.20	0.77	28.851	7,345,935.0	30,240,996,470.5	
SD	2.82	1.35	38.69	0.50	9.07	0.45	3.443	2.84	72.97	0.18	21.479	10,199,966.9	27,554,779,770.3	
Min	9.50	5.92	-66.70	7.30	6.00	0.02	0.001	0.08	224.60	0.42	0.010	123,022.0	555,182,304.2	
Max	19.50	9.19	-17.80	8.14	68.00	0.69	0.194	1.67	669.77	551.35	11.884	5,358,879.0	109,848,334,934.7	
Mean	14.56	7.13	-45.26	7.61	20.26	0.40	0.049	0.53	397.29	162.76	3.245	1,031,169.1	22,967,574,415.6	
Median	14.15	6.74	-46.13	7.48	15.25	0.41	0.006	0.35	406.12	54.37	1.795	457,493.5	3,277,220,387.2	
SD	3.59	1.29	15.59	0.31	20.07	0.21	0.071	0.54	142.82	216.31	4.196	1,770,335.7	37,886,320,437.6	
Min	6.10	4.90	-130.40	7.30	6.00	0.02	0.001	0.08	187.93	0.42	0.010	123,022.0	555,182,304.2	
Max	19.50	10.64	-7.05	9.26	131.00	1.65	14.200	11.42	669.77	551.35	66.640	30,749,458.0	109,848,334,934.7	
Mean	12.48	7.29	-56.49	8.06	27.48	0.43	3.121	2.63	376.86	62.74	13.429	4,651,738.8	21,463,814,746.4	
Median	12.25	7.04	-52.85	8.00	15.25	0.39	0.140	0.67	391.39	1.36	3.735	933,750.5	7,001,341,238.5	
SD	3.47	1.46	30.89	0.58	33.10	0.36	4.601	3.45	111.67	143.54	18.476	7,523,116.7	29,199,700,945.6	

Table D.5: The mean and median periphytic algal concentration:biovolume ratio for the summer and winter seasons for each site and the three sites combined.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		sum	win	sum	win	sum	win	sum	win
C*	Mean	1.720	1.049	1.558	1.568	0.761	0.849	0.079	0.121
	Median	2.183	4.216	2.261	2.201	0.674	0.786	0.000	0.056

* = Combined sites; sum = summer; win = winter

Table D.6: Periphyton genera found at the study three sites during the summer and winter seasons for 2003–2007.

Sites	Periphyton genera	Sampling dates											
		2003/06	2004/05	2004/06	2005/05	2006/05	2006/07	2007/05	2007/07	2003/11	2004/01	2007/01	2007/11
Sannaspos	Anabaena (F)	-	-	-	-	-	-	-	-	-	-	-	-
	Calothrix (F)	-	-	-	-	-	-	-	-	-	-	-	-
	Merismopedia (C)	-	-	-	-	-	-	-	-	-	-	-	-
	Microcystis (sc)	-	-	-	-	-	-	-	-	-	-	-	-
	Nostoc (F)	-	-	-	-	-	-	-	-	-	-	-	-
	Oscillatoria (F)	-	-	-	-	-	-	-	-	-	-	-	-
	Phormidium (F)	-	-	-	-	-	-	-	-	-	-	-	-
	Rivularia	-	-	-	-	-	-	-	-	-	-	-	-
	Achnanthes	-	-	-	-	-	-	-	-	-	-	-	-
	Amphora	-	-	-	-	-	-	-	-	-	-	-	-
	Cocconeis	-	-	-	-	-	-	-	-	-	-	-	-
	Cyclotella	-	-	-	-	-	-	-	-	-	-	-	-
Gymbella	-	-	-	-	-	-	-	-	-	-	-	-	
Diatoma	-	-	-	-	-	-	-	-	-	-	-	-	
Gomphonema	-	-	-	-	-	-	-	-	-	-	-	-	
Gyrosigma	-	-	-	-	-	-	-	-	-	-	-	-	
Melosira (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Navicula	-	-	-	-	-	-	-	-	-	-	-	-	
Nitzschia	-	-	-	-	-	-	-	-	-	-	-	-	
Pennate (other)	-	-	-	-	-	-	-	-	-	-	-	-	
Pinnularia	-	-	-	-	-	-	-	-	-	-	-	-	
Skeletonema	-	-	-	-	-	-	-	-	-	-	-	-	
Stauroneis	-	-	-	-	-	-	-	-	-	-	-	-	
Centric (other)	-	-	-	-	-	-	-	-	-	-	-	-	
Stephanodiscus	-	-	-	-	-	-	-	-	-	-	-	-	
Surirella	-	-	-	-	-	-	-	-	-	-	-	-	
Synedra	-	-	-	-	-	-	-	-	-	-	-	-	
Actinastrium (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Ankistrodesmus	-	-	-	-	-	-	-	-	-	-	-	-	
Carteria	-	-	-	-	-	-	-	-	-	-	-	-	
Chlamydomonas	-	-	-	-	-	-	-	-	-	-	-	-	
Chlorella	-	-	-	-	-	-	-	-	-	-	-	-	
Chlorococcum	-	-	-	-	-	-	-	-	-	-	-	-	
Chodatella	-	-	-	-	-	-	-	-	-	-	-	-	
Cladophora (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Closterium	-	-	-	-	-	-	-	-	-	-	-	-	
Coelastrum (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Cosmarium	-	-	-	-	-	-	-	-	-	-	-	-	
Crucigenia (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Mesostaelium	-	-	-	-	-	-	-	-	-	-	-	-	
Micractinium (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Monoraphidium	-	-	-	-	-	-	-	-	-	-	-	-	
Mougeotia (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Oocystis (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Oocystis (sc)	-	-	-	-	-	-	-	-	-	-	-	-	
Pandorina (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Pediastrum (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Scenedesmus (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Schroederia	-	-	-	-	-	-	-	-	-	-	-	-	
Sphaerocystis (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Sprogyra (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Staurastrum	-	-	-	-	-	-	-	-	-	-	-	-	
Stigeoclonium (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Tetrastrum (C)	-	-	-	-	-	-	-	-	-	-	-	-	
Ulothrix (F)	-	-	-	-	-	-	-	-	-	-	-	-	
Cryptomonas	-	-	-	-	-	-	-	-	-	-	-	-	
Peridinium	-	-	-	-	-	-	-	-	-	-	-	-	
Euglena	-	-	-	-	-	-	-	-	-	-	-	-	
Lepocyclis	-	-	-	-	-	-	-	-	-	-	-	-	
Phacus	-	-	-	-	-	-	-	-	-	-	-	-	
Trachelomonas	-	-	-	-	-	-	-	-	-	-	-	-	
Chrysococcus	-	-	-	-	-	-	-	-	-	-	-	-	

Legend – Table 1st row: red = Cyanophyta; orange = Bacillariophyta; green = Chlorophyta; light pink = Cryptophyta; purple = Dinophyta; blue = Euglenophyta; yellow = Chrysophyta. – Table body: light green = genera that did not occur at each site; tan = genera that did not occur during either the summer or winter seasons.

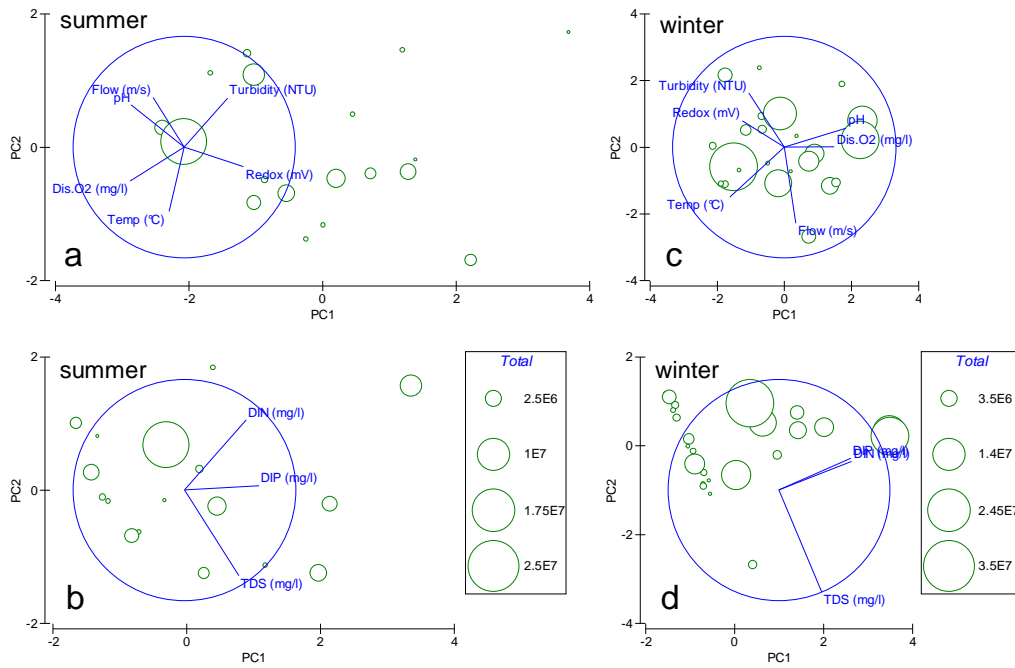


Figure D.13: The PCA graphs: summer period (a) physical and chemical and (b) nutrient, and winter season (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic algal concentration (cells/cm²).

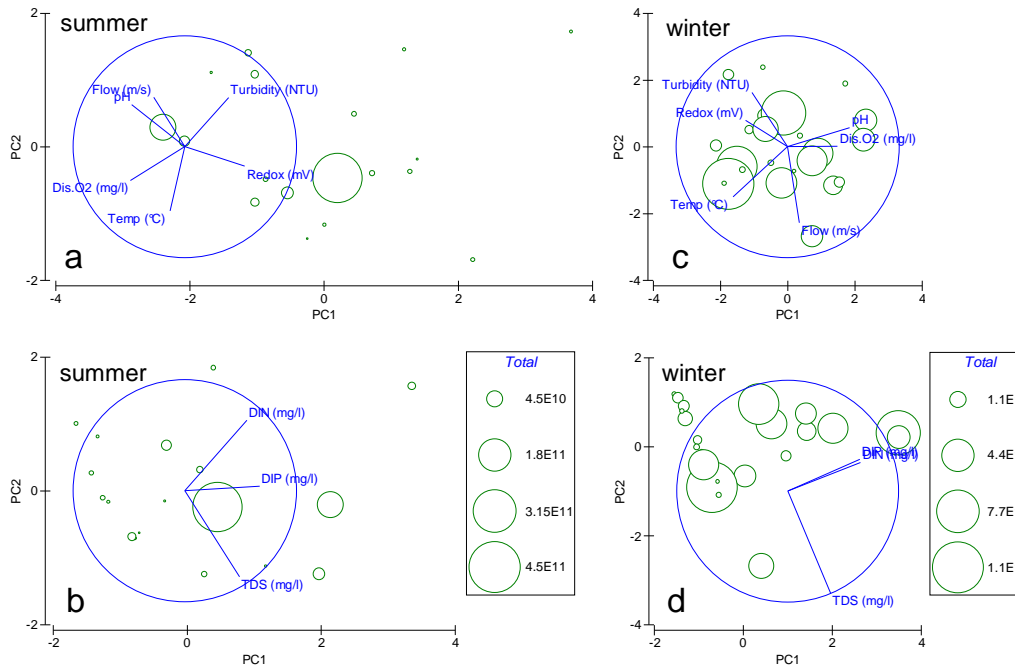


Figure D.14: The PCA graphs: summer season (a) physical and chemical and (b) nutrient, and winter season (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop’s Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic algal biovolume (cm³/cm²).

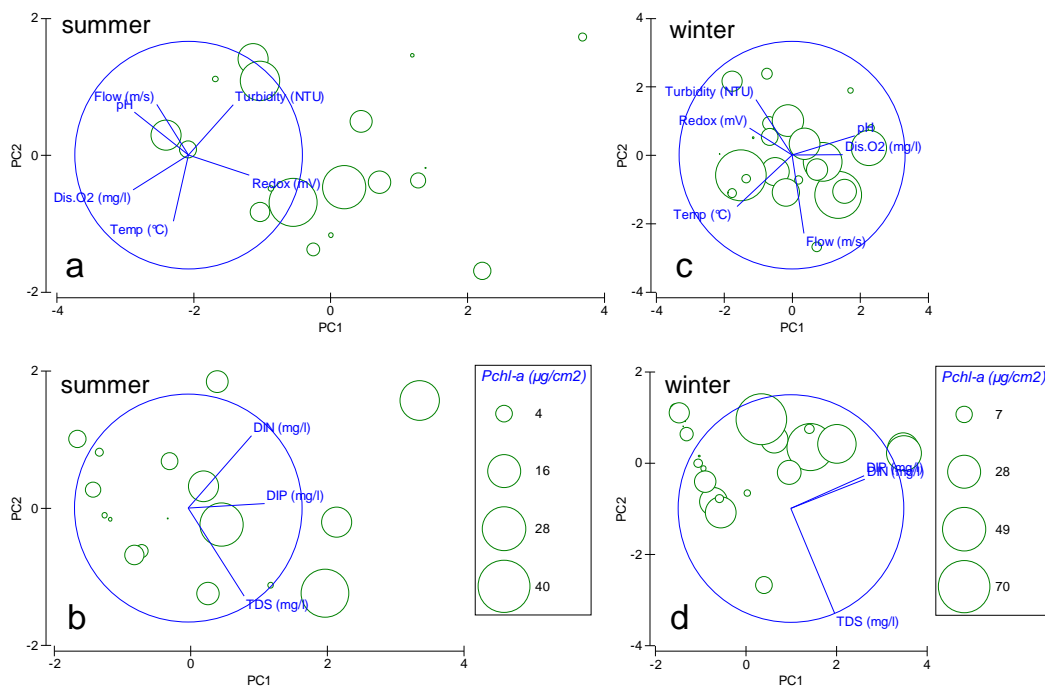


Figure D15: The PCA graphs: summer season (a) physical and chemical and (b) nutrient, and winter season (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic chlorophyll-a concentration ($\mu\text{g}/\text{cm}^2$).

D.3 ADDITIONAL PROCESSED DATA FOR CHAPTER 6

Table D.7: Additional minimum, maximum, mean, median and standard deviation of the physical, chemical and algal data for the dry and wet periods for each site and the three sites combined.

	Temperature (°C)	Diss. oxygen (mg/l)	Redox (mV)	pH	Turbidity (NTU)	Flow (m/s)	DIP (mg/l)	DIN (mg/l)	TDS (mg/l)	DIN:DIP	Pchl-a (µg/cm ²)	Periphytic algal	
												Conc.	Biovol.
Dry	Min	2.87	-91.00	7.15	8.30	0.04	0.026	0.13	108.12	0.12	0.013	4,401.0	161,356,526.5
	Max	23.80	12.34	-11.60	9.20	460.00	1.68	4.010	1.53	547.45	15.77	12.000	2,565,456.0
	Mean	15.46	6.93	-45.70	8.14	100.92	0.23	0.739	0.64	331.86	5.24	4.711	780,657.5
	Median	13.30	6.55	-42.40	8.20	32.00	0.08	0.111	0.50	348.80	5.59	4.510	480,338.0
	SD	5.75	2.92	24.73	0.63	140.17	0.48	1.241	0.45	164.19	5.12	3.971	791,134.5
	Min	9.80	5.29	-114.80	7.57	2.80	0.11	2.050	0.55	256.14	0.06	0.460	199,148.0
	Max	25.10	11.41	-22.90	9.00	35.00	3.95	12.600	8.63	568.65	1.33	66.640	30,749,458.0
	Mean	18.17	7.42	-60.41	7.98	12.87	0.82	7.905	3.86	412.70	0.55	28.072	5,469,781.6
	Median	19.50	7.34	-58.30	7.80	9.00	0.63	7.790	3.53	402.50	0.60	27.300	3,201,899.0
	SD	5.60	1.75	25.80	0.38	9.85	1.00	2.891	2.73	104.54	0.38	21.214	7,994,706.0
	Min	11.80	4.63	-70.20	7.23	6.00	0.00	0.001	0.08	294.28	0.42	0.310	140,621.0
	Max	26.10	11.80	-17.80	8.20	18.70	0.67	0.194	4.37	700.21	3044.70	24.990	3,270,249.0
Mean	19.04	7.35	-42.28	7.63	11.19	0.19	0.055	1.09	455.99	444.84	4.079	668,094.7	
Median	19.35	7.19	-40.98	7.60	10.40	0.08	0.019	0.42	421.83	99.00	1.910	385,890.5	
SD	5.58	2.00	16.73	0.32	4.42	0.22	0.066	1.35	133.32	883.36	6.831	867,355.8	
Min	6.90	2.87	-114.80	7.15	2.80	0.00	0.001	0.08	108.12	0.06	0.013	4,401.0	
Max	26.10	12.34	-11.60	9.20	460.00	3.95	12.600	8.63	700.21	3044.70	66.640	30,749,458.0	
Mean	17.63	7.25	-49.87	7.91	39.21	0.43	3.099	1.95	402.43	150.08	12.936	2,436,431.4	
Median	17.80	7.00	-50.11	7.80	12.20	0.18	0.188	0.91	401.88	1.32	4.530	790,477.0	
SD	5.68	2.19	23.61	0.49	85.89	0.72	4.100	2.31	140.08	538.46	17.513	5,261,220.9	
Min	6.10	1.65	-117.20	7.48	10.70	0.08	0.001	0.08	196.63	0.22	0.080	84,963.0	
Max	24.90	7.97	-35.40	9.24	128.00	0.60	2.720	1.84	446.63	169.11	20.177	11,404,348.0	
Mean	16.71	4.37	-72.21	8.09	41.99	0.30	0.394	0.52	309.81	28.63	2.909	1,715,875.7	
Median	17.70	4.16	-67.15	7.89	25.50	0.30	0.120	0.37	288.19	4.03	0.766	366,115.5	
SD	6.52	2.31	29.56	0.60	40.17	0.16	0.835	0.53	90.79	54.88	6.154	3,484,877.6	
Min	9.00	3.91	-130.40	7.81	3.80	0.16	2.750	0.10	221.55	0.02	2.290	205,028.0	
Max	27.30	10.64	-7.05	9.26	51.00	2.56	15.300	11.42	620.10	1.07	31.870	20,402,129.0	
Mean	18.88	6.20	-76.55	8.33	18.57	0.75	8.183	4.49	398.55	0.54	15.365	6,056,231.2	
Median	20.00	5.28	-81.20	8.21	15.45	0.53	6.960	3.82	399.66	0.57	15.247	2,489,303.0	
SD	5.93	2.13	31.56	0.41	13.21	0.72	4.560	3.60	122.35	0.30	9.231	7,458,839.5	
Min	9.50	2.22	-84.10	7.38	7.40	0.00	0.001	0.17	145.19	3.15	0.010	123,022.0	
Max	27.10	8.52	-27.80	8.17	68.00	0.69	0.082	1.28	679.80	551.35	11.884	5,358,879.0	
Mean	19.33	5.69	-56.27	7.75	23.07	0.42	0.033	0.57	355.92	144.22	3.671	1,430,513.1	
Median	19.50	5.92	-58.50	7.68	16.20	0.46	0.010	0.52	380.29	26.35	2.140	634,627.0	
SD	7.01	1.87	20.27	0.33	20.37	0.28	0.037	0.40	177.61	202.60	4.171	1,829,760.3	
Min	6.10	1.65	-130.40	7.38	3.80	0.00	0.001	0.08	145.19	0.02	0.010	84,963.0	
Max	27.30	10.64	-7.05	9.26	128.00	2.56	15.300	11.42	679.80	551.35	31.870	20,402,129.0	
Mean	18.24	5.44	-70.16	8.11	27.73	0.52	3.530	2.17	379.66	44.91	8.247	3,443,004.2	
Median	19.40	5.23	-69.60	8.11	19.10	0.42	0.180	0.73	349.74	1.41	4.242	966,039.0	
SD	6.27	2.22	28.77	0.51	28.11	0.52	4.924	3.02	129.71	114.75	9.301	5,610,871.7	

Table D.8: The mean and median periphytic algal concentration:biovolume ratio for the dry and wet periods for each site and the three sites combined.

		Periphytic algal divisions (%)							
		Cyanophyta		Bacillariophyta		Chlorophyta		Euglenophyta	
		dry	wet	dry	wet	dry	wet	dry	wet
SP	Mean	1.681	1.465	1.799	1.547	0.425	0.954	0.063	0.066
	Median	2.481	1.275	1.902	2.170	0.445	1.957	0.020	0.032
BW	Mean	1.000	1.271	1.269	1.674	1.477	0.773	0.041	0.069
	Median	1.000	1.078	0.000	2.073	1.559	0.454	0.000	0.041
MC	Mean	3.666	1.553	1.639	1.816	0.474	0.732	0.303	0.075
	Median	17.785	1.049	1.866	2.089	0.416	3.360	0.087	0.071
3*	Mean	1.972	1.405	1.625	1.662	0.530	0.830	0.138	0.070
	Median	5.592	1.295	1.766	2.057	0.434	0.649	0.121	0.036

* = Combined sites

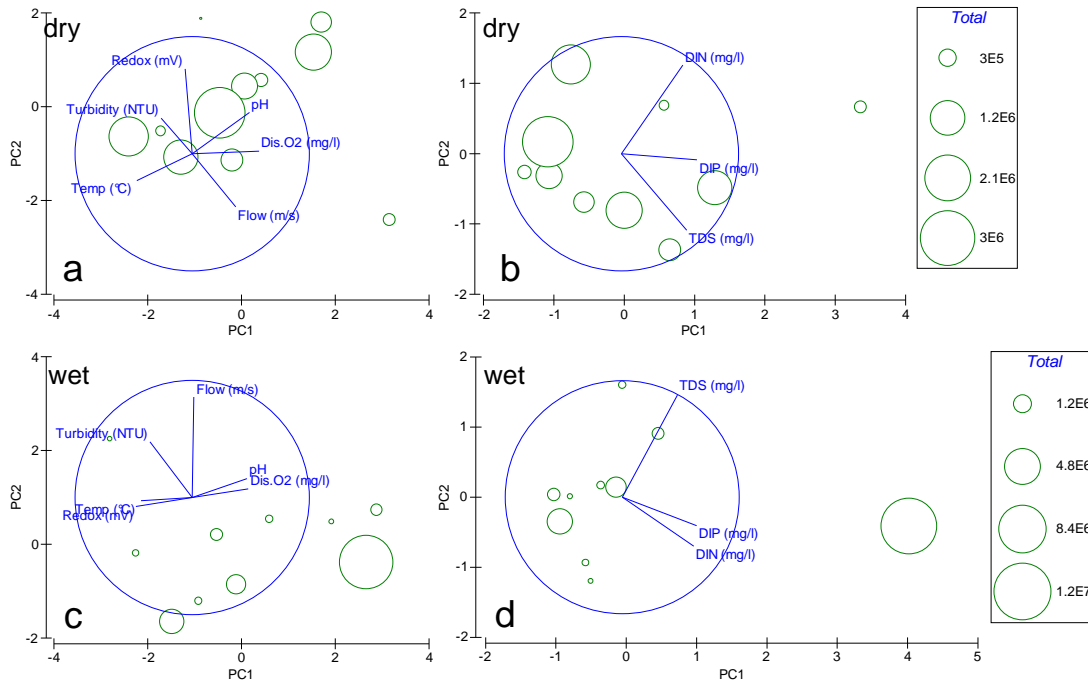


Figure D.16: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP). The bubble overlay is the periphytic algal concentration (cells/cm²).

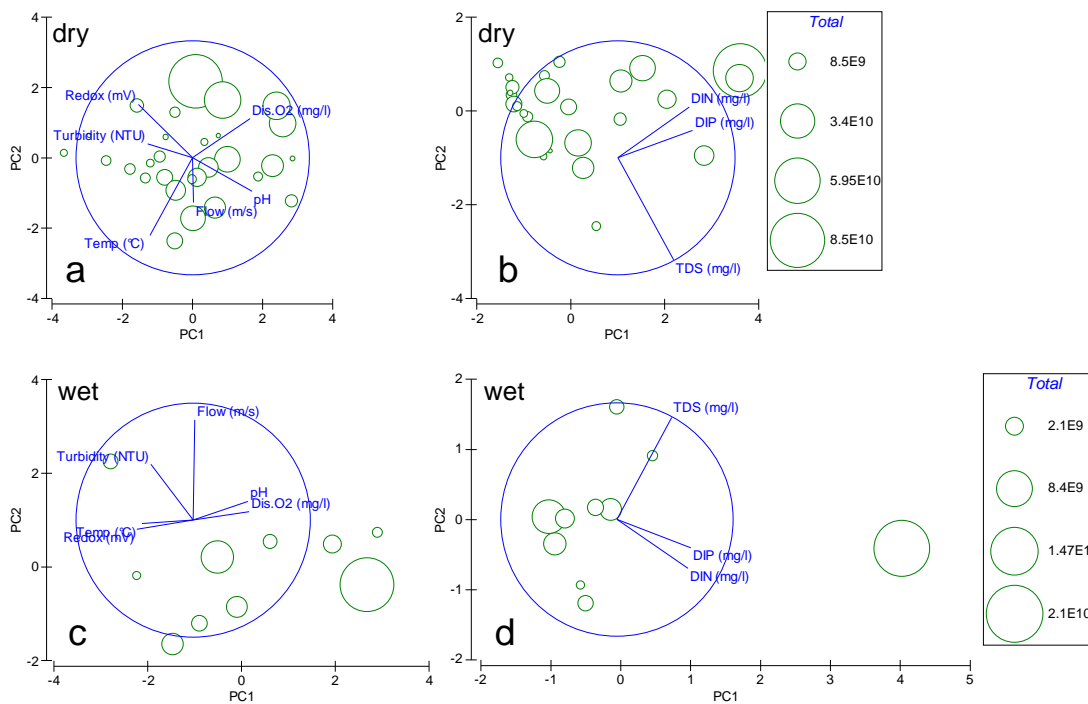


Figure D.17: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP). The bubble overlay is the periphytic algal biovolume (cm³/cm²).

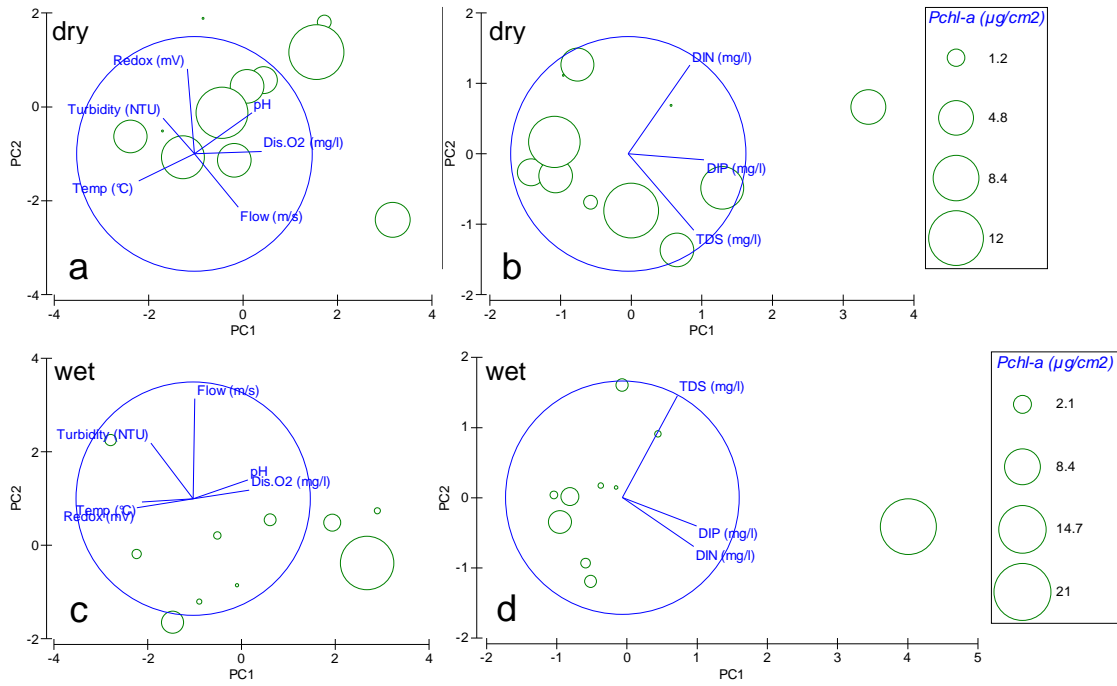


Figure D.18: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP). The bubble overlay is the periphytic chlorophyll-a concentration (µg/cm²).

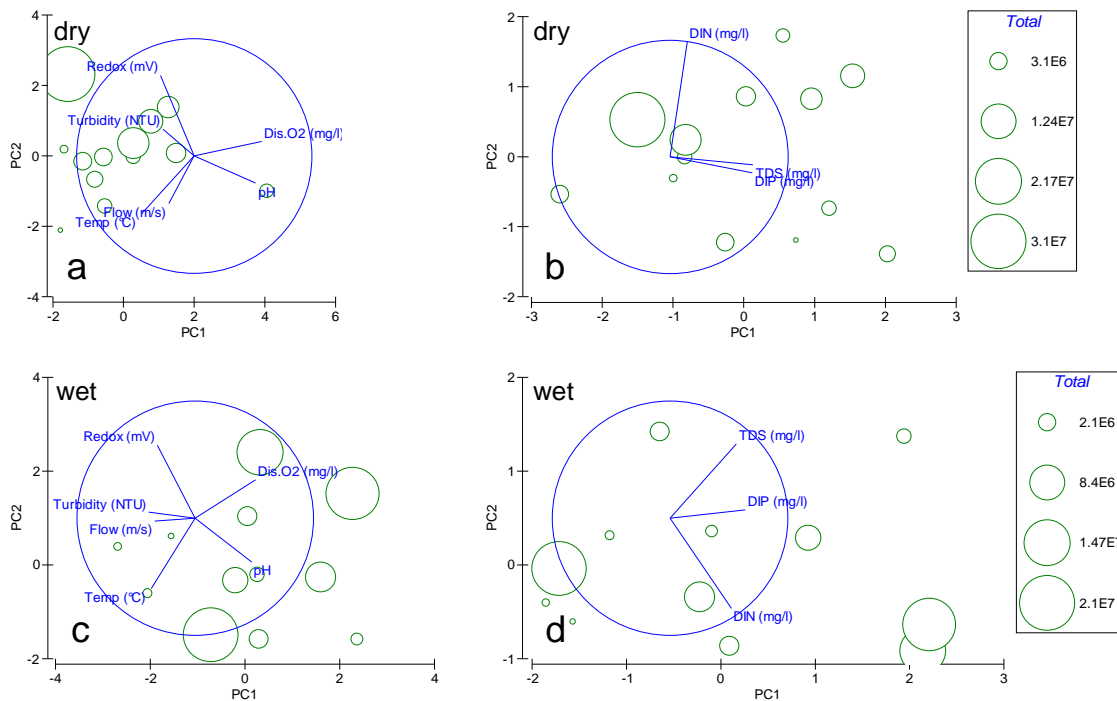


Figure D.19: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Bishop's Weir (BW). The bubble overlay is the periphytic algal concentration (cells/cm²).

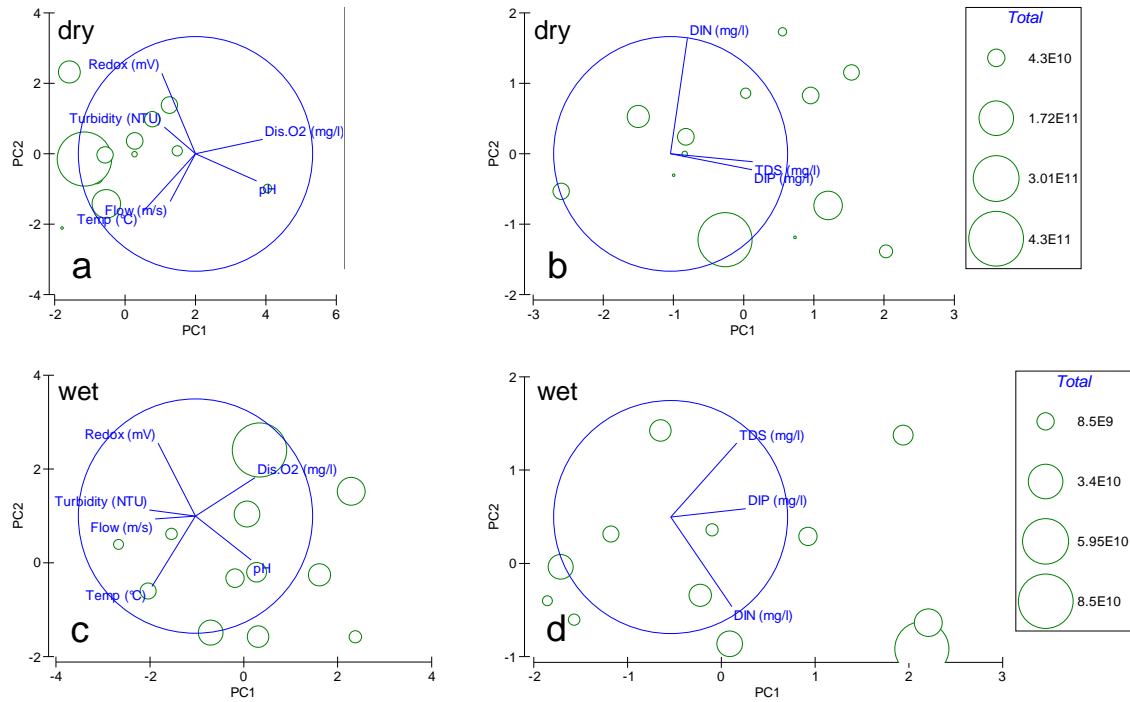


Figure D.20: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Bishop's Weir (BW). The bubble overlay is the periphytic algal biovolume (cm³/cm²).

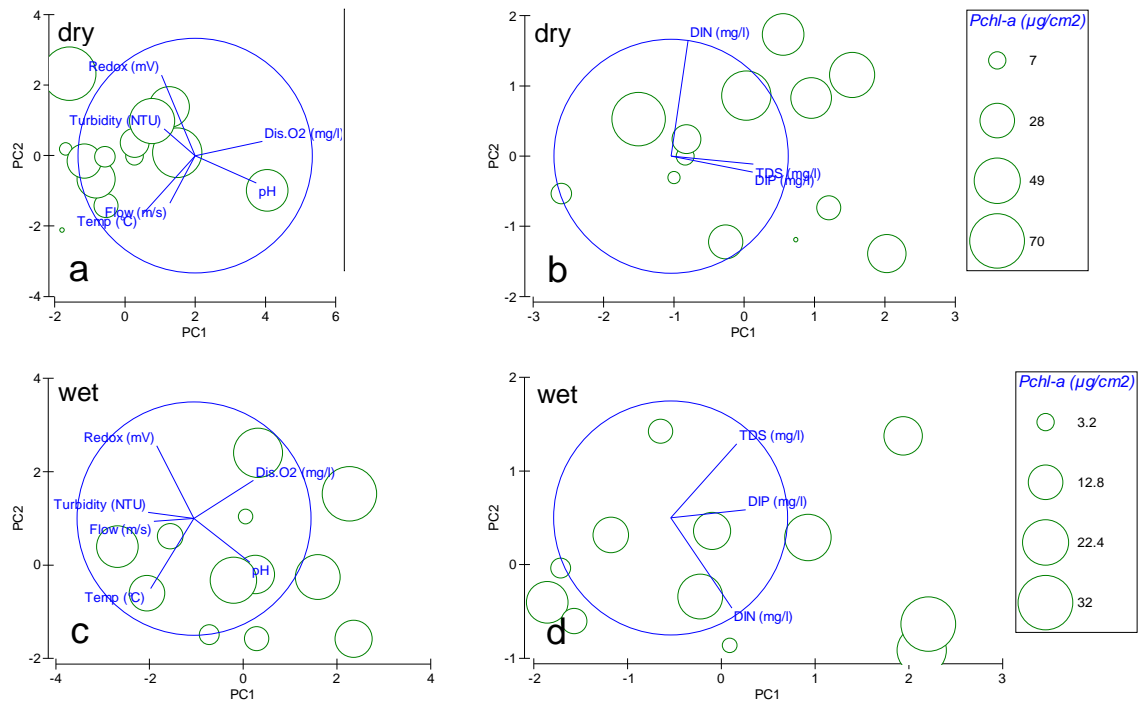


Figure D.21: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Bishop's Weir (BW). The bubble overlay is the periphytic chlorophyll-a concentration (µg/cm²).

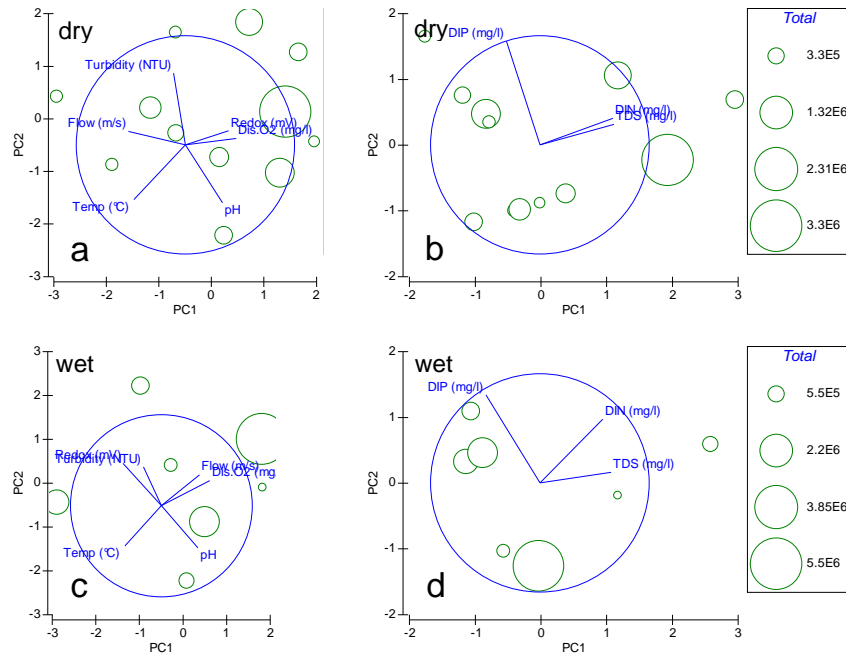


Figure D.22: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Modder above Confluence (MC). The bubble overlay is the periphytic algal concentration (cells/cm²).

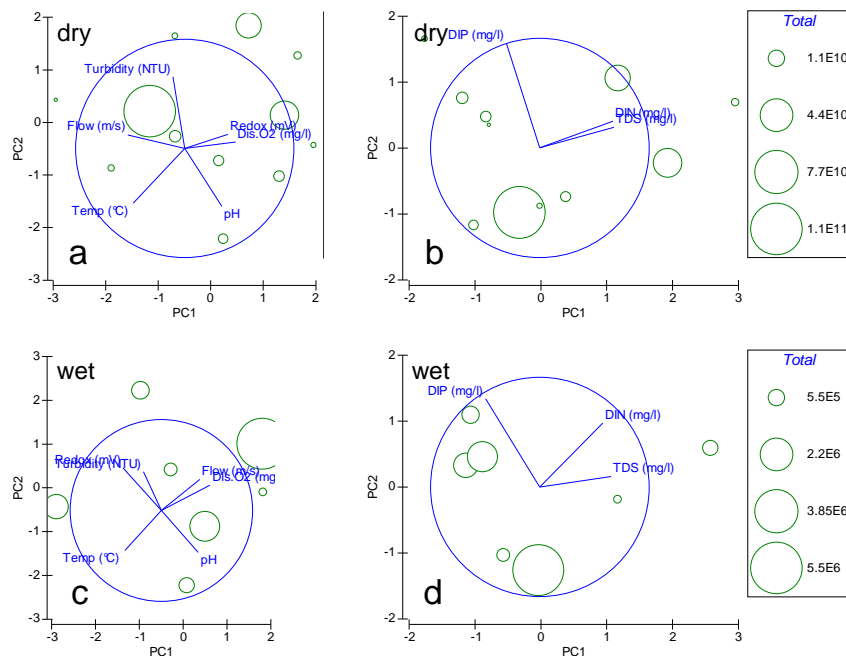


Figure D.23: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Modder above Confluence (MC). The bubble overlay is the periphytic algal biovolume (cm³/cm²).

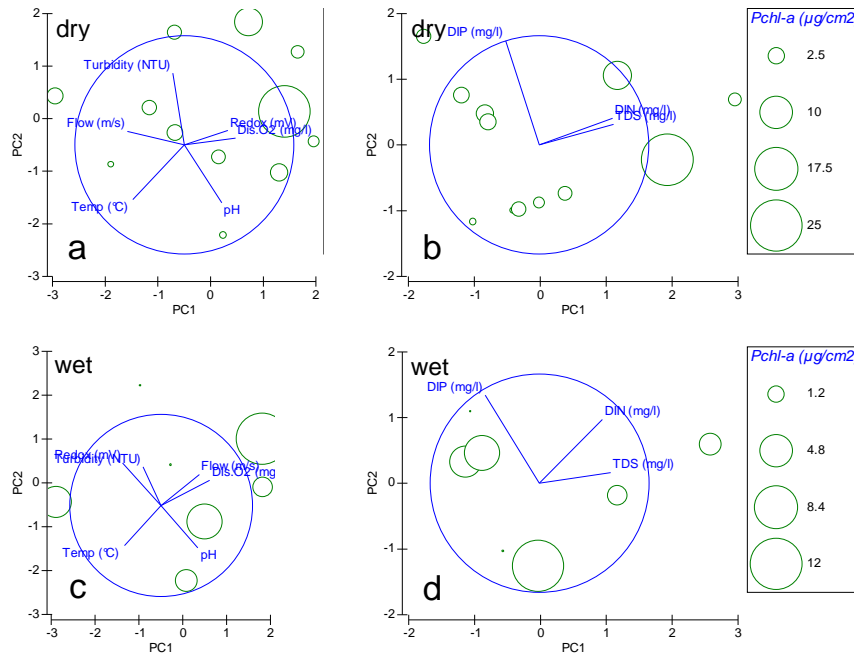


Figure D.24: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Modder above Confluence (MC). The bubble overlay is the periphytic chlorophyll-a concentration ($\mu\text{g}/\text{cm}^2$).

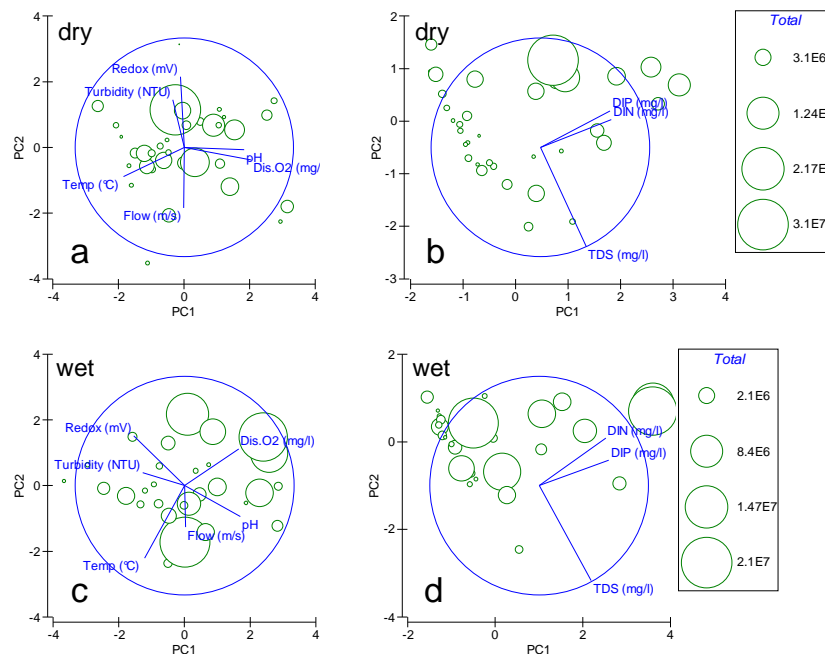


Figure D.25: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic algal concentration (cells/ cm^2).

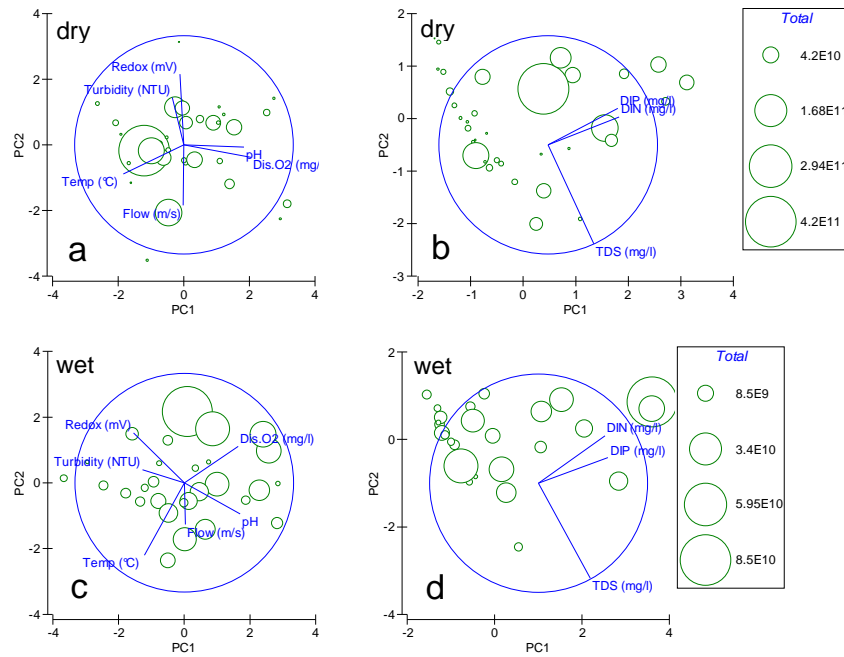


Figure D.26: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic algal biovolume (cm^3/cm^2).

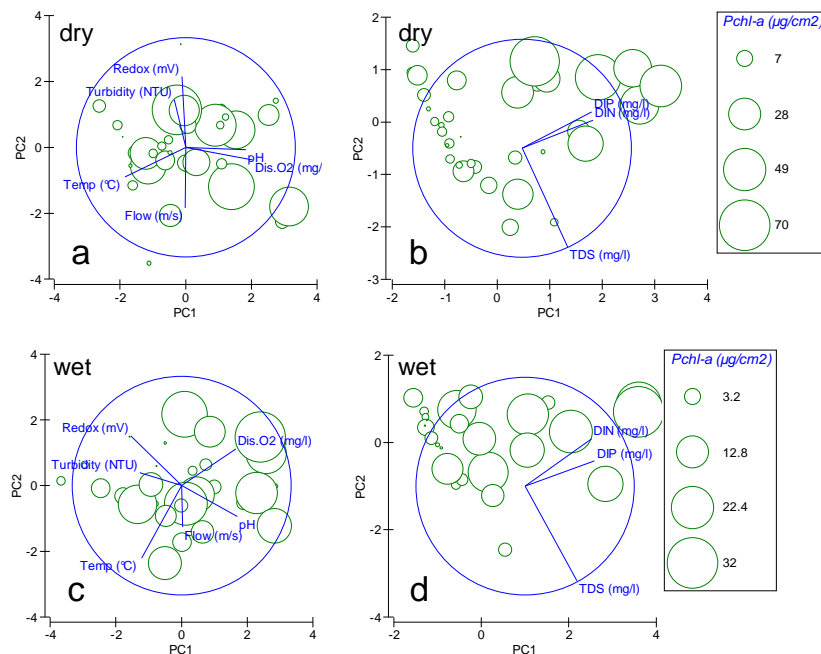


Figure D.27: The PCA graphs: wet period (a) physical and chemical and (b) nutrient, and dry period (c) physical and chemical and (d) nutrient factors at Sannaspos (SP), Bishop's Weir (BW) and Modder above Confluence (MC). The bubble overlay is the periphytic chlorophyll-a concentration ($\mu\text{g}/\text{cm}^2$).

D.4 ADDITIONAL PROCESSED DATA FOR CHAPTER 7

Table D.10: Additional minimum, maximum, mean, median and standard deviation of the SASS and ASPT data over (a) the study period, (b) the summer and winter seasons and (c) the dry and wet periods for each site and the three sites combined.

	SASS5				ASPT			
	Total	SIC	%*	Total	Total	SIC	%*	Total
(a)	Min	21.0	15.0	47.4	3.8	3.6		
	Max	120.0	85.0	90.5	5.2	6.3		
	Mean	51.7	35.5	69.3	4.4	4.4		
	Median	39.0	30.0	68.8	4.3	4.4		
	SD	25.7	18.3	12.9	0.4	0.7		
Sannaspos	Min	20.0	11.0	38.7	3.0	2.8		
	Max	67.0	42.0	95.5	4.6	4.2		
	Mean	41.9	24.0	58.1	3.8	3.5		
	Median	41.0	23.0	53.8	3.9	3.5		
	SD	12.8	7.9	13.0	0.4	0.4		
Bishop's Weir	Min	12.0	6.0	28.6	3.3	3.0		
	Max	73.0	50.0	100.0	4.8	4.8		
	Mean	35.2	22.7	65.1	4.0	3.9		
	Median	30.0	21.0	66.7	4.0	3.6		
	SD	16.3	12.2	22.8	0.4	0.6		
Confidence above Modder	Min	12.0	6.0	28.6	3.3	3.0		
	Max	73.0	50.0	100.0	4.8	4.8		
	Mean	35.2	22.7	65.1	4.0	3.9		
	Median	30.0	21.0	66.7	4.0	3.6		
	SD	16.3	12.2	22.8	0.4	0.6		
Combined Sites	Min	12.0	6.0	28.6	3.0	2.8		
	Max	120.0	85.0	100.0	5.2	6.3		
	Mean	43.1	27.3	63.8	4.0	3.9		
	Median	38.0	25.0	62.9	4.0	3.8		
	SD	19.6	14.2	16.9	0.5	0.7		
(b)								
Sannaspos	Min	34.0	26.0	62.9	3.9	3.7		
	Max	85.0	64.0	76.5	4.7	4.9		
	Mean	60.8	41.6	68.6	4.3	4.4		
	Median	62.0	39.0	64.3	4.4	4.9		
	SD	19.0	14.3	6.7	0.3	0.6		
Bishop's Weir	Min	27.0	19.0	38.9	3.0	2.9		
	Max	64.0	37.0	72.5	4.4	4.0		
	Mean	46.6	25.5	55.9	3.8	3.5		
	Median	49.0	24.0	53.2	3.9	3.6		
	SD	11.6	6.4	11.2	0.5	0.4		
Confidence above Modder	Min	24.0	7.0	29.2	3.4	3.0		
	Max	73.0	50.0	78.6	4.3	4.5		
	Mean	47.6	28.0	53.7	4.0	3.8		
	Median	48.0	21.0	48.6	4.1	3.6		
	SD	18.6	18.2	19.8	0.4	0.6		
Combined Sites	Min	24.0	7.0	29.2	3.0	2.9		
	Max	85.0	64.0	78.6	4.7	4.9		
	Mean	50.8	30.7	58.8	4.0	3.8		
	Median	52.0	27.5	61.2	4.1	3.7		
	SD	16.2	13.9	13.9	0.4	0.7		
(c)								
Sannaspos	Min	21.0	15.0	48.4	3.8	3.6		
	Max	62.0	45.0	90.5	4.4	4.9		
	Mean	37.9	27.5	72.6	4.1	4.1		
	Median	32.0	26.0	75.0	4.1	3.9		
	SD	13.2	10.6	14.0	0.2	0.5		
Bishop's Weir	Min	23.0	11.0	38.9	3.0	2.8		
	Max	60.0	32.0	71.4	4.6	4.0		
	Mean	41.2	23.0	56.9	3.8	3.4		
	Median	41.0	23.0	57.1	3.8	3.3		
	SD	12.1	6.1	10.0	0.5	0.4		
Confidence above Modder	Min	12.0	6.0	28.6	3.3	3.0		
	Max	54.0	33.0	100.0	4.7	4.8		
	Mean	30.7	17.3	57.2	4.0	3.7		
	Median	27.5	17.0	58.5	4.0	3.6		
	SD	12.4	8.8	21.9	0.4	0.5		
Combined Sites	Min	12.0	6.0	28.6	3.0	2.8		
	Max	62.0	45.0	100.0	4.7	4.9		
	Mean	36.7	22.5	61.8	3.9	3.7		
	Median	34.0	21.0	61.0	4.0	3.7		
	SD	13.0	9.3	17.1	0.4	0.5		
(d)								
Sannaspos	Min	31.0	18.0	47.4	3.9	3.8		
	Max	120.0	85.0	80.6	5.2	6.3		
	Mean	66.8	44.3	65.7	4.6	4.8		
	Median	70.0	40.5	65.0	4.7	4.7		
	SD	28.0	21.3	11.2	0.4	0.7		
Bishop's Weir	Min	20.0	12.0	38.7	3.3	2.9		
	Max	67.0	42.0	95.5	4.0	4.2		
	Mean	42.7	25.0	59.3	3.8	3.5		
	Median	40.5	23.5	53.1	3.9	3.7		
	SD	14.0	9.6	16.1	0.3	0.5		
Confidence above Modder	Min	19.0	14.0	46.7	3.5	3.4		
	Max	73.0	50.0	100.0	4.8	4.7		
	Mean	43.0	32.0	78.6	4.0	4.1		
	Median	36.0	28.0	78.6	4.0	4.4		
	SD	20.0	12.3	18.6	0.5	0.6		
Combined Sites	Min	19.0	12.0	38.7	3.3	2.9		
	Max	120.0	85.0	100.0	5.2	6.3		
	Mean	51.1	33.3	66.2	4.1	4.1		
	Median	44.0	28.0	65.7	4.0	3.9		
	SD	23.5	17.0	16.6	0.5	0.8		

* = SASS5 SIC as a percentage of the total SASS5 score

Table D.11: Macroinvertebrate families found at the three study sites during the period 2003–2007.

Sites	Macro-invertebrates present/absent	Sampling dates	Macroinvertebrate Families																																					
			PORIFERA	TURBELLARIA	Oligochaeta	Hirudinae	Potamonautidae	Atyidae	HYDRACARINA	Baetidae	Caenidae	Tricorythidae	Coenagrionidae	Aeshnidae	Gomphidae	Belostomatidae	Corixidae	Gerridae	Naucoridae	Notonectidae	Velidae	Ecnomidae	Hydropsychidae	Dytiscidae	Gyrinidae	Hydraenidae	Hydrophilidae	Ceratopogonidae	Chironomidae	Culicidae	Muscidae	Simuliidae	Tabanidae	Ancylidae	Lymnaeidae	Physidae	Planorbinae	Sphaeriidae		
Sannaspos		2003/06																																						
		2003/09				P																																		
		2003/10																																						
		2003/11																																						
		2004/01																																						
		2004/03																																						
		2004/05																																						
		2004/06																																						
		2004/09																																						
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		2007/01																																						
		2007/03																																						
		2007/05																																						
		2007/07																																						
		2007/09																																						
	2007/11																																							
Bishop's Weir		2003/06																																						
		2003/09																																						
		2003/10																																						
		2003/11																																						
		2004/01																																						
		2004/03																																						
		2004/05																																						
		2004/06																																						
		2004/09																																						
		2004/11																																						
		2005/01																																						
		2005/03																																						
		2005/05																																						
		2006/01																																						
		2006/03																																						
		2006/05																																						
		2006/07																																						
		2006/09																																						
		2006/11																																						
		2007/01																																						
	2007/03																																							
	2007/05																																							
	2007/07																																							
	2007/09																																							
	2007/11																																							
Modder above Confluence		2003/06																																						
		2003/09																																						
		2003/10																																						
		2003/11																																						
		2004/01																																						
		2004/03																																						
		2004/05																																						
		2004/06																																						
		2004/09																																						
		2004/11																																						
		2005/03																																						
		2005/05																																						
		2006/01																																						
		2006/05																																						
		2006/07																																						
		2007/01																																						
	2007/03																																							
	2007/05																																							
	2007/07																																							

Legend – Table 1st row: the different colours indicate the families belonging to the same order.
 Table body: light green = genera that did not occur at each site.

Table D.14: Additional minimum, maximum, mean, median and standard deviation of the phytoplanktonic algal divisions' data over (a) the study period, (b) the summer and winter seasons and (c) the dry and wet periods for each site and the three sites combined.

	Phytoplanktonic algal divisions (%)				
	Cyano- phyta	Bacillario- phyta	Chloro- phyta	Eugleno- phyta	
(a)	Min	0.00	0.00	3.61	0.00
	Max	12.10	95.59	80.69	60.00
	Mean	2.66	25.69	44.45	23.81
	Median	0.00	22.82	44.45	24.92
	St. Dev.	4.11	23.54	21.68	19.75
	Min	0.00	5.48	15.94	0.00
	Max	63.61	50.00	88.68	27.62
	Mean	7.45	24.31	50.98	8.41
	Median	3.98	25.08	52.57	6.90
	St. Dev.	13.53	12.66	19.51	7.50
(b)	Min	0.00	4.80	21.43	0.00
	Max	57.12	58.79	85.81	46.82
	Mean	4.77	26.10	50.49	11.53
	Median	0.00	20.78	41.80	9.57
	St. Dev.	12.96	16.47	22.65	10.95
	Min	0.00	0.00	3.61	0.00
	Max	63.61	95.59	88.68	60.00
	Mean	5.12	25.28	48.73	14.30
	Median	0.92	23.71	48.72	9.53
	St. Dev.	11.20	17.61	21.04	14.91
(c)	Min	0.00	0.00	7.04	0.00
	Max	12.10	58.11	80.69	60.00
	Mean	3.39	27.63	39.61	25.66
	Median	1.95	23.97	30.01	23.32
	St. Dev.	4.26	19.48	22.40	23.09
	Min	0.00	5.48	30.15	0.00
	Max	63.61	39.99	88.68	18.37
	Mean	11.46	21.57	55.62	7.00
	Median	4.02	20.69	55.73	6.90
	St. Dev.	17.74	11.95	17.97	6.44
Summer	Min	0.00	4.80	31.61	0.00
	Max	57.12	48.02	85.81	21.04
	Mean	6.07	21.76	56.59	7.98
	Median	0.00	20.54	51.54	7.25
	St. Dev.	16.26	13.59	21.88	7.21
	Min	0.00	0.00	7.04	0.00
	Max	63.61	58.11	88.68	60.00
	Mean	7.20	23.49	51.05	13.03
	Median	2.18	20.78	46.17	9.26
	St. Dev.	14.42	14.94	21.54	15.98
Winter	Min	0.00	2.39	3.61	0.00
	Max	11.13	95.59	77.18	44.45
	Mean	1.85	23.56	49.78	21.78
	Median	0.00	15.59	49.35	25.56
	St. Dev.	4.00	28.29	20.67	16.30
	Min	0.00	6.40	15.94	0.00
	Max	12.50	50.00	76.68	27.62
	Mean	3.10	27.27	45.94	9.93
	Median	1.03	25.51	50.32	7.37
	St. Dev.	4.09	13.24	20.61	8.53
Dry	Min	0.00	11.84	21.43	5.91
	Max	7.19	58.79	70.55	46.82
	Mean	2.54	33.53	40.04	17.61
	Median	0.00	37.56	29.45	12.52
	St. Dev.	3.25	19.32	21.43	13.99
	Min	0.00	2.39	3.61	0.00
	Max	12.50	95.59	77.18	46.82
	Mean	2.53	27.50	45.84	15.87
	Median	0.00	25.01	48.72	12.52
	St. Dev.	3.78	20.51	20.41	13.58
Wet	Min	0.00	0.00	0.00	0.00
	Max	11.13	95.59	63.76	55.55
	Mean	2.88	35.15	32.54	26.08
	Median	0.00	33.70	35.99	28.84
	St. Dev.	4.47	30.91	21.38	22.86
	Min	0.00	5.48	15.94	0.00
	Max	12.50	50.00	88.68	22.90
	Mean	4.21	22.98	46.15	9.50
	Median	1.69	17.81	38.29	8.73
	St. Dev.	4.98	17.28	28.78	7.89
Summer	Min	0.00	0.00	3.61	0.00
	Max	11.13	95.59	63.76	55.55
	Mean	2.88	35.15	32.54	26.08
	Median	0.00	33.70	35.99	28.84
	St. Dev.	4.47	30.91	21.38	22.86
	Min	0.00	5.48	15.94	0.00
	Max	12.50	50.00	88.68	22.90
	Mean	4.21	22.98	46.15	9.50
	Median	1.69	17.81	38.29	8.73
	St. Dev.	4.98	17.28	28.78	7.89
Winter	Min	0.00	0.00	3.61	0.00
	Max	11.13	95.59	63.76	55.55
	Mean	2.88	35.15	32.54	26.08
	Median	0.00	33.70	35.99	28.84
	St. Dev.	4.47	30.91	21.38	22.86
	Min	0.00	5.48	15.94	0.00
	Max	12.50	50.00	88.68	22.90
	Mean	4.21	22.98	46.15	9.50
	Median	1.69	17.81	38.29	8.73
	St. Dev.	4.98	17.28	28.78	7.89

Table D.15: Additional minimum, maximum, mean, median and standard deviation of the phytoplankton components data over (a) the study period, (b) the summer and winter seasons and (c) the dry and wet periods for each site and the three sites combined.

	Phytoplankton				Phytoplankton				Phytoplankton					
	No. of genera	Chl-a (µg/l)	Conc. (cells/ml)		No. of genera	Chl-a (µg/l)	Conc. (cells/ml)		No. of genera	Chl-a (µg/l)	Conc. (cells/ml)			
(a)	Min	4.0	1.00	884.0	(b)	Min	4.0	1.00	2419.0	(c)	Min	7.0	1.00	884.0
	Max	20.0	214.23	202662.0		Max	20.0	116.79	22555.0		Max	20.0	39.17	22555.0
	Mean	10.6	36.51	15862.1		Mean	10.5	27.90	8305.4		Mean	12.0	17.84	7356.8
	Median	11.0	27.94	4399.0		Median	10.0	25.79	4544.0		Median	11.0	12.90	3671.0
	St. Dev.	4.0	49.70	43153.3		St. Dev.	4.9	33.35	6841.6		St. Dev.	5.6	18.34	8698.3
(a)	Min	8.0	1.00	592.0	(b)	Min	12.0	1.00	1843.0	(c)	Min	12.0	1.00	1843.0
	Max	22.0	1402.41	37562.0		Max	20.0	184.14	37562.0		Max	21.0	184.14	22556.0
	Mean	16.0	107.69	7264.7		Mean	16.9	62.10	10722.0		Mean	16.6	67.92	7616.1
	Median	16.0	45.86	4619.0		Median	18.0	68.07	6597.0		Median	16.5	20.35	3597.0
	St. Dev.	3.7	273.23	8118.6		St. Dev.	2.3	54.54	10004.3		St. Dev.	3.3	57.58	7084.9
(a)	Min	4.0	1.00	591.0	(b)	Min	4.0	1.00	954.0	(c)	Min	10.0	1.00	1253.0
	Max	19.0	73.80	19481.0		Max	18.0	73.80	19481.0		Max	15.0	73.80	7476.0
	Mean	11.7	22.97	4879.7		Mean	12.0	27.61	6294.8		Mean	12.6	36.47	4380.4
	Median	11.0	16.48	3082.0		Median	12.5	20.78	3875.0		Median	13.0	28.01	3861.0
	St. Dev.	3.9	23.13	5241.5		St. Dev.	4.0	25.32	6050.5		St. Dev.	2.1	34.89	2884.6
(a)	Min	4.0	1.00	591.0	(b)	Min	4.0	1.00	954.0	(c)	Min	7.0	1.00	884.0
	Max	22.0	1402.41	202662.0		Max	20.0	184.14	37562.0		Max	21.0	184.14	22556.0
	Mean	13.0	59.93	9345.2		Mean	13.3	40.15	8507.9		Mean	14.2	45.27	6645.3
	Median	13.0	28.01	4104.0		Median	14.5	24.36	4984.5		Median	14.5	37.14	4058.5
	St. Dev.	4.5	174.34	25217.6		St. Dev.	4.6	42.65	7918.8		St. Dev.	4.2	47.08	6521.5
(a)	Min	4.0	1.00	591.0	(b)	Min	7.0	1.00	884.0	(c)	Min	6.0	1.00	2059.0
	Max	15.0	214.23	202662.0		Max	15.0	214.23	202662.0		Max	15.0	214.23	202662.0
	Mean	10.6	45.97	24174.5		Mean	10.6	45.97	24174.5		Mean	10.3	49.94	29282.8
	Median	11.0	32.01	3741.5		Median	11.0	32.01	3741.5		Median	11.0	26.87	4324.0
	St. Dev.	2.8	63.74	62766.7		St. Dev.	2.8	63.74	62766.7		St. Dev.	3.3	71.32	70081.1
(a)	Min	8.0	1.00	592.0	(b)	Min	8.0	1.00	592.0	(c)	Min	8.0	1.00	592.0
	Max	22.0	1402.41	8798.0		Max	22.0	1402.41	8798.0		Max	20.0	1402.41	37562.0
	Mean	14.9	157.08	3519.3		Mean	14.9	157.08	3519.3		Mean	14.5	222.09	8895.0
	Median	14.0	43.71	3491.0		Median	14.0	43.71	3491.0		Median	15.0	65.21	4075.5
	St. Dev.	4.6	393.09	2279.0		St. Dev.	4.6	393.09	2279.0		St. Dev.	4.5	478.13	12266.3
(a)	Min	7.0	1.00	591.0	(b)	Min	7.0	1.00	591.0	(c)	Min	7.0	1.00	591.0
	Max	19.0	48.01	6235.0		Max	19.0	48.01	6235.0		Max	17.0	45.14	17433.0
	Mean	11.3	15.02	2453.7		Mean	11.3	15.02	2453.7		Mean	10.6	18.21	4353.0
	Median	11.0	9.65	1253.0		Median	11.0	9.65	1253.0		Median	11.0	13.07	2936.0
	St. Dev.	4.1	17.69	2105.5		St. Dev.	4.1	17.69	2105.5		St. Dev.	3.5	17.39	5461.4
(a)	Min	7.0	1.00	591.0	(b)	Min	7.0	1.00	591.0	(c)	Min	6.0	1.00	591.0
	Max	22.0	1402.41	202662.0		Max	22.0	1402.41	202662.0		Max	20.0	1402.41	202662.0
	Mean	12.6	84.47	10384.6		Mean	12.6	84.47	10384.6		Mean	11.8	96.74	14176.9
	Median	12.0	31.53	3454.0		Median	12.0	31.53	3454.0		Median	11.0	26.87	3926.5
	St. Dev.	4.3	257.04	37056.0		St. Dev.	4.3	257.04	37056.0		St. Dev.	4.1	282.11	40893.4

