

# **Ecosystem health and water quality of the Mooi River and associated impoundments using diatoms and macroinvertebrates as bioindicators**

**ELC Pelser**

**21698759**

Dissertation submitted in fulfillment of the requirements for the degree  
*Magister Scientiae* in **Environmental Sciences** at the Potchefstroom  
Campus of the North-West University

Supervisor: Dr JC Taylor  
Co-supervisor: Prof S Barnard

November 2015

# ACKNOWLEDGEMENTS

First I thank my heavenly Father, for love, opportunity, healthy mind and patience to complete this dissertation. All Glory to the one and only God.

A very special thank you to my supervisor Dr. J.C. Taylor. It was a privilege and an honour to be mentored by one of the best in the field. Thank you for all the advice, knowledge, the long hours of corrections, and the passion in which you perceive the field. You are and will always be an inspiration and motivation to me.

Professor Sandra, thank you for all the advice, comments and inputs. I admire and respect you and your knowledge in the field and in life, and it has been an honour to learn different ways of doing things, and perceiving information taught me a lot.

To my family, friends and the love of my life Soné, I give thanks. The constant motivation and inspiration was invaluable and kept me going. Your love and support never failed me, and I will forever be grateful. To my father Johan a special thank you, you are the man I one day wish to grow into, I can only hope to be half the man you raised me to be. My moeder, thank you, from childhood you were always the one to comfort, motivate and support me no matter what. I love each and every one of you more than I could ever describe.

Thank you to Lucia, Adrian and Marinus for the help with the fieldwork, and lab analysing of the water samples.

Thank you Done vd Westhuizen for the GIS work done for the area map.

A last and special thank you to Lucia and the MidVaal Water Company for the free water quality analysis, it will never be forgotten, and you were a big part in the completion of this project.

# ABSTRACT

Water is the most important element on earth for sustaining the life of all living organisms. Fresh water is needed for human life and throughout history concentrated human populations were found in close proximity to a fresh water source. Urbanization, industrialization, mining and over population have negative effects on water quality. Clean potable water has become a limiting resource worldwide and particularly in South Africa due to developing communities and informal settlements forming around rivers, mining, heavy industry, agriculture and poorly managed sanitation. These impacts are problematic for both the human population and for the aquatic organisms which are dependent on this resource as a habitat. The monitoring and management of freshwater is thus critical to this resource.

In order to manage resources impacts need to be accurately identified. In the case of aquatic ecosystems constant monitoring will allow for the prevention or early detection of any threats to the integrity of the resource.

The river system chosen for the present study was the Mooi River. It is the source for potable water to various communities in the area including the city of Potchefstroom. The Mooi River originates near Koster and flows south to its confluence with the Vaal River south of Potchefstroom. The water quality of the Mooi River is impacted by mining pollution from Wonderfontein spruit (a tributary of the Mooi River), urban influences from Potchefstroom, agricultural activities and informal communities situated in the catchment area.

In this study the measured water quality variables, diatom analysis and macroinvertebrate analysis were used in combination to monitor the ecosystem health of the Mooi River for the calendar year of 2014 in order to identify problem areas in the catchment and the time of year that the influence of these impacts were greatest. All of the above mentioned biomonitoring tools showed a gradual decline in ecosystem health from the origin of the Mooi River flowing downstream toward the Vaal River.

This decline in ecosystem health, throughout the Mooi River, could be ascribed to the influence of the Wonderfontein spruit and also the impact of Potchefstroom and its surrounding (sub) urban area and industries. The alteration in the physical and chemical regime in the river was clearly reflected by changes within the habitat integrity and community structure of the aquatic biota.

In addition, low rainfall in the winter period had a slight impact on the ecosystem health, as pollutants become more concentrated.

It can be concluded that the methods used in the study were applied successfully to identify the main detrimental influences on the water quality of the Mooi River, and that the different bioindicators used in the study were sufficient to determine the health of the Mooi River ecosystem.

**Key Words: Biomonitoring; Aquatic Health; Diatom; Macroinvertebrates; Aquatic Ecosystems; Water quality.**

## Opsomming

Water is een van die belangrikste elemente op aarde wat die onderhouding van alle lewende organismes betref. Menslike oorlewing, en lewe oor die algemeen, is afhanklik van water, reeds vanaf die vroegste tye word menslike populasie in gekonsentreerde groepe, aangetref by nabygeleë waterbronne. Verstedeliking, industrialisering, mynbou en oorbevolking het 'n negatiewe uitwerking op die nodige waterbronne se kwaliteit. Die beskikbaarheid van drinkbare water word wêreldwyd, en spesifiek in Suid-Afrika, al hoe meer beperk as gevolg van ontwikkelende gemeenskappe en informele nedersettings wat naby ons riviere- en alternatiewe waterbronne, tot stand kom en sodoende hierdie waterbronne besoedel met 'n "slagorde" van verskeie besoedelingsmiddele uit alle oorde. Om hierdie varswaterbronne behoorlik te bestuur is dit dus van kardinale belang om die bronne te onderhou en bewaar.

Voor hierdie waterbronne egter behoorlik bestuur kan word, is dit belangrik om die probleemareas te identifiseer. Op die manier kan oorsprong van die probleem bestuur word; voorkoming is egter beter as genesing. Hierdie potensiele probleme, asook die bron-spessifieke besoedeling kan identifiseer word deur konstante monitering van waterbronne. Die studie word gedoen in die Mooirivier, wat hoofsaaklik Potchefstroom, en omliggende areas van drinkbare water voorsien. Hierdie rivier se oorsprong is naby Koster en vloei suid, in die rigting van die Vaal Rivier (suid van Potchefstroom). Die waterkwaliteit van die Mooirivier word hoofsaaklik beïnvloed deur verskeie bronne soos die mynbesoedeling uit die Wonderfonteinspruit, verstedeliking van Potchefstroom, landboukundige aktiviteite in die area en informelenedersettings wat in die "voerarea" ontwikkel.

In die studie word 'n kombinasie van water-veranderlikes, diatomiese analise en makro-invertebratiese analise gebruik om die ekosisteem gesondheid van die Mooi Rivier konstant te monitor, probleemareas in die opvangsgebied te identifiseer, en te bepaal in watter tyd van die jaar die impak van hierdie probleemareas die grootste was. Al die bogenoemde biomonitoring indikators het die gewenste resultate gekry en die gelydelike afname in die ekosisteem gesondheid van die Mooirivier (wat stroomaf vloei na die Vaalrivier) kon duidelik aangedui word deur die verskeie metodes wat in die studie gebruik is.

Die resultate het 'n afname in die ekosisteem gesondheid, regdeur die Mooi Rivier aangedui: Die grootste impak het plaasgevind by die samevloeiing van die Wonderfonteinspruit en weer stroomaf van Potchefstroom, met die verandering in die deklinasie in habitat integriteit en die gemeenskapsstruktuur van die water biota. Die lae reënvalsifer in die winter het ook 'n klein

impak op die ekosisteem gesondheid soos besoedelingstowwe meer gekonsentreerd en beskikbaar raak. Daar kan afgelei word dat die metodes wat in die studie gebruik is, suksesvol aangewend was om die hoof probleemareas wat die water kwaliteit van die Mooi Rivier beïnvloed te identifiseer, asook dat die verskeie bio-aanwysers wat in die studie gebruik was, voldoende was om die ekosisteem gesondheid van die Mooi Rivier te bepaal.

**Sleutel woorde: Biomonitoring; Akwatiese Gesondheid; Diatom; Macroinvertebrate; Akwatiese Ecosisteme; Water Kwaliteit.**

# TABLE OF CONTENTS

<b>ACKNOWLEDGEMENTS.....</b>	<b>I</b>
<b>ABSTRACT .....</b>	<b>II</b>
<b>OPSOMMING .....</b>	<b>IV</b>
<b>CHAPTER 1 GENERAL INTRODUCTION.....</b>	<b>1</b>
<b>1.1 Water scarcity and need for Biomonitoring in South Africa .....</b>	<b>1</b>
<b>1.2 Factors influencing water quality of the Mooi River .....</b>	<b>4</b>
1.2.1 Description of the Mooi River catchment .....	4
1.2.2 Mining related pollution .....	6
1.2.3 Agriculture.....	7
1.2.4 Urban runoff via Potchefstroom and wastewater .....	8
<b>1.3 Water quality changes.....</b>	<b>9</b>
1.3.1 Nutrients .....	9
1.3.2 Temperature.....	10
1.3.3 pH .....	10
1.3.4 Electrical Conductivity and Total Dissolved Solids.....	11
<b>1.4 The use of aquatic organisms as bioindicators .....</b>	<b>12</b>
1.4.1 Diatoms.....	12
1.4.2 Diatoms in biomonitoring.....	13
1.4.3 Diatom studies in South Africa .....	14
<b>1.5 Macroinvertebrates .....</b>	<b>14</b>

1.5.1	Macroinvertebrates as bioindicators .....	15
1.5.2	History of macroinvertebrate studies in South Africa .....	16
<b>1.6</b>	<b>Research outcomes .....</b>	<b>16</b>
1.6.1	Research question .....	16
1.6.2	Hypotheses.....	16
1.6.3	Aims.....	17
1.6.4	Objectives .....	17
<b>1.7</b>	<b>Dissertation outline.....</b>	<b>17</b>
<b>CHAPTER 2 MATERIALS AND METHODS .....</b>		<b>19</b>
<b>2.1</b>	<b>Study Sites .....</b>	<b>19</b>
2.1.1	Bovenste Oog (BVO) .....	20
2.1.2	Klerkskraal Dam (KKD) .....	20
2.1.3	Upstream of Wonderfonteinspruit (BWS).....	21
2.1.4	Downstream of Wonderfonteinspruit (OWS).....	22
2.1.5	Boskop Dam (BKD) .....	23
2.1.6	Mooi River at the bridge to Carletonville (PDM) .....	24
2.1.7	Trompie Kitsgras (RWP).....	25
2.1.8	Elbrixon Bridge (EBR) .....	26
<b>2.2</b>	<b>Site attributes. ....</b>	<b>27</b>
<b>2.3</b>	<b>Water Quality .....</b>	<b>28</b>
2.3.1	Field Measurements.....	28
2.3.2	Laboratory analysis.....	28

<b>2.4</b>	<b>Diatoms .....</b>	<b>28</b>
2.4.1	Diatom Field Collection .....	28
2.4.2	Diatom preparation .....	29
2.4.3	Diatom enumeration .....	30
2.4.4	Diatom identification .....	30
2.4.5	Diatom index calculations .....	30
<b>2.5</b>	<b>Macroinvertebrates .....</b>	<b>31</b>
2.5.1	Sampling sites.....	31
2.5.2	Sampling method .....	32
2.5.3	Identification.....	32
2.5.4	Relevant references and score attribution .....	32
<b>2.6</b>	<b>Statistical analysis.....</b>	<b>33</b>
 <b>CHAPTER 3 WATER QUALITY - MEASURED PHYSICAL-CHEMICAL</b>		
	<b>PARAMETERS .....</b>	<b>34</b>
<b>3.1</b>	<b>Water quality .....</b>	<b>34</b>
<b>3.2</b>	<b>Results and discussion .....</b>	<b>34</b>
3.2.1	Dissolved Calcium and Magnesium .....	34
3.2.2	Nutrients .....	38
3.2.3	Sulphate .....	42
3.2.4	Temperature.....	45
3.2.5	pH .....	46
3.2.6	Electrical conductivity .....	48

3.2.7	Turbidity .....	50
3.2.8	Spatial and Temporal variation in Water quality in the Mooi River .....	51
3.2.8.1	Spatial variation of water quality variables .....	52
3.2.8.2	Temporal variation of water quality .....	55
<b>CHAPTER 4 DIATOMS .....</b>		<b>58</b>
<b>4.1</b>	<b>Introduction .....</b>	<b>58</b>
<b>4.2</b>	<b>Results and discussion .....</b>	<b>58</b>
4.2.1	Diatom species list.....	59
4.2.2	Diatoms and water quality .....	66
4.2.3	Diatom indices .....	68
4.2.3.1	Pollution Tolerant Valves (%PTV) .....	69
4.2.3.2	Generic Diatom Index (GDI).....	71
4.2.3.3	Biological Diatom Index (BDI) .....	74
4.2.3.4	Species Pollution sensitivity Index (SPI) .....	76
4.2.4	Spatial and Temporal variation of diatom assemblages .....	80
4.2.4.1	Spatial variation of diatoms in the Mooi River .....	80
4.2.4.2	Temporal variation of diatoms in the Mooi River .....	85
<b>CHAPTER 5 MACROINVERTABRATES .....</b>		<b>86</b>
<b>5.1</b>	<b>Introduction .....</b>	<b>86</b>
<b>5.2</b>	<b>Results and discussion .....</b>	<b>86</b>
5.2.1	Macroinvertebrate taxa per site .....	87
5.2.2	Macroinvertebrates and water quality .....	88

5.2.3	Macroinvertebrate indices .....	91
5.2.4	Spatial and temporal variation in macroinvertebrate assemblages.....	94
5.2.4.1	Spatial variation of macroinvertebrates in the Mooi River .....	94
5.2.4.2	Temporal variation of macroinvertebrates in the Mooi River .....	97
<b>CHAPTER 6 CONCLUSION AND RECOMMENDATIONS.....</b>		<b>99</b>
<b>6.1</b>	<b>Conclusions.....</b>	<b>99</b>
<b>6.2</b>	<b>Recommendations.....</b>	<b>101</b>
<b>CHAPTER 7 BIBLIOGRAPHY.....</b>		<b>103</b>
<b>ANNEXURES.....</b>		<b>121</b>
<b>Appendix A</b>	<b>.....</b>	<b>121</b>
<b>Appendix B</b>	<b>.....</b>	<b>123</b>
<b>Appendix C</b>	<b>.....</b>	<b>131</b>
<b>Appendix D</b>	<b>.....</b>	<b>138</b>

## LIST OF TABLES

Table 2-1: A summary of the sites, relevance in terms of impact detection, coordinates and altitude.....	27
Table 2-2: Interpretation of the various indices used in this study .....	31
Table 2-3: A representation of the ecological classes representing the SASS5 scores .....	33
Table 3-1: Summary of canonical correspondence analysis (CCA) for both physical and chemical parameters.....	53
Table 4-1 Diatom species encountered in the Mooi River at each site in 2014. ....	59
Table 4-2: Summary of canonical correspondence analysis (CCA) for Diatom and the physical and chemical water quality parameters of the Mooi River in 2014.....	67
Table 5-1: Distribution of macroinvertebrate taxa per site during the 2014 sampling season.....	87
Table 5-2: Summary of canonical correspondence analysis (CCA) for macroinvertebrates and the physical and chemical water quality parameters of the Mooi River in 2014.....	89

## LIST OF FIGURES

Figure 2-1: Map of the Mooi River system, indicating the sites chosen for the present study. The study sites are (Upstream to downstream) Bovenste Oog (BVO), Klerkskraal Dam (KKD), Upstream of Wonderfonteinspruit (BWS), Downstream of Wonderfonteinspruit (OWS), Boskop Dam (BKD), Downstream of Potchefstroom Dam (PDM), Trompie Kitsgras (RWP) and Elbrixon Bridge (EBR). .....	19
Figure 2-2 Left, the aquatic vegetation, stones and sand substrate is shown and right, the stones in a current just downstream of the figure on the left. ....	20
Figure 2-3: The two photographs show the sampling site below the dam, rocks and vegetation are available for sampling. ....	21
Figure 2-4: Left illustrates the shallow water upstream of the sampling site and right the shallow water downstream of the sampling site. ....	21
Figure 2-5: Both photographs illustrate the deep flowing water of the site, heavily encroached upon by trees and bushes. ....	22
Figure 2-6: Sampling site at Boskop Dam situated within the Boskop Dam Nature reserve. ....	23
Figure 2-7: Left is upstream of the site and right is downstream of the site. Note the different sampling biotopes.....	24
Figure 2-8: Left is upstream of the site and right is downstream of the site. Note the different sampling biotopes.....	25
Figure 2-9: Left and right show the deep flowing waters and marginal vegetation of EBR. ....	26
Figure 3-1: A box-and-whisker plot illustrating the change in mean dissolved calcium concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	35
Figure 3-2: A box-and-whisker plot illustrating the change in mean dissolved magnesium concentration in mg/L between sites from upstream (1- BVO) to	

downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	37
Figure 3-3: A box-and-whisker plot illustrating the change in mean NO <sub>3</sub> and NO <sub>4</sub> (Nitrate) concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	39
Figure 3-4: A box-and-whisker plot illustrating the change in mean phosphate concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	41
Figure 3-5: A box-and-whisker plot illustrating the change in mean sulphate concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	43
Figure 3-6 A box-and-whisker plot illustrating the change in mean temperature in °C between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	45
Figure 3-7: A box-and-whisker plot illustrating the change in mean pH between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation. ....	47
Figure 3-8: A box-and-whisker plot illustrating the change in mean Electrical Conductivity (EC) in mS/m between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	48
Figure 3-9: A box-and-whisker plot illustrating the change in mean Turbidity in NTU between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	50
Figure 3-10: A Canonical correspondence analysis scatter biplot, showing the spatial variation between measured water quality variables and sites (January 2014 – December 2014).....	52

Figure 3-11: A Canonical correspondence analysis scatter biplot showing the seasonal temporal variation of the water quality variables at each site.....	56
Figure 4-1: Canonical correspondence analysis scatter biplot illustrating dominant diatom species (weight range of more than 15%) and their relation to water quality variables measured in the Mooi River for the time period 2014. ....	66
Figure 4-2: A Box and whisker plot illustrating the change in mean %PTV between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	69
Figure 4-3: A Box and whisker plot illustrating the change in mean GDI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	71
Figure 4-4: A Box and whisker plot illustrating the change in mean BDI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	75
Figure 4-5: A Box and whisker plot illustrating the change in mean SPI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.....	77
Figure 4-6: Detrended correspondence analysis scatterplot represents the spatial variation of dominant diatom species recorded from each site.....	81
Figure 4-7: Detrended correspondence analysis scatterplot represents the temporal variation of dominant diatom species represented at each site.....	85
Figure 5-1: Canonical correspondence analysis scatterplot illustrating macroinvertebrate taxa currency of occurring in and their relation to differences in water quality. ....	89
Figure 5-2: A Box and whisker plot illustrating the change in mean SASS5 score between sites from upstream (1- BVO) to Potchefstroom (6- PDM) and downstream towards (7- RWP) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation. ....	91

Figure 5-3: A Box and whisker plot illustrating the change in mean ASPT score between sites from upstream (1- BVO) to Potchefstroom (6- PDM) and downstream towards (7- RWP) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation. ....	92
Figure 5-4: Detrended correspondence analysis scatterplot represents the spatial variation of the macroinvertebrate taxa at each site.....	95
Figure 5-5: Detrended correspondence analysis scatterplot representing the temporal variation of macroinvertebrate taxa found at each site. ....	97

# CHAPTER 1 GENERAL INTRODUCTION

## 1.1 Water scarcity and need for Biomonitoring in South Africa

Water is the most important resource for sustaining life. It is indispensable for social, economic, industrial and agricultural activities. Water is a resource that is over utilised and the impact is borne by aquatic ecosystems (Nilsson *et al.*, 2007). The degradation of water and aquatic ecosystems is caused by the unsustainable use of water by the above mentioned industries and activities, which in turn deposit nutrients (nitrogen and phosphorus) of anthropogenic origin and other substances such as salts into catchments (Xue *et al.*, 2009). The increase of nutrients in aquatic ecosystems has widespread consequences on ecosystem deterioration such as a loss of biodiversity and an increase in the establishment and proliferation of invasive species (Green and Galatowitsch, 2002).

Freshwater is an invaluable source of environmental goods and services such as drinking water, food (fish), recreational activities and tourism attractions, which are essential to sustain human societies (Nilsson *et al.*, 2007). Population growth increase also escalates the demand for natural resources and infrastructure. This places pressure on social and economic industries to increase use and production rate of these resources, and this influences the quality and diversity of ecosystems (Ohl *et al.*, 2007). This makes a large contribution to the impacts on aquatic ecosystems, the constant urbanization of land, channelizing of streams all contribute to the degradation of ecosystems causing a loss of biodiversity and habitat degradation, thus freshwater ecosystems have become endangered (Meyer *et al.*, 2005; Vinson and Hawkins, 1998; LaBonte *et al.*, 2001; Dudgeon *et al.*, 2006).

South Africa is a semi-arid country with an average rainfall of 497 mm per year, considerably less than the global average of 860mm (Mantel *et al.*, 2010), the highest rain fall in South Africa is in Matiwa in Limpopo with an annual rain fall of 2004mm per year and the driest is Alexander Bay in the Northern Cape with 46mm (South African Weather Service, 2015). More than 80% of our rivers are currently threatened, because of the overuse of natural resources (Nel *et al.*, 2004). In South Africa, society utilises water as if it is an inexhaustible resource, and this has resulted in the degradation of our rivers, dams and wetlands. The effects can be seen in the environmental impacts caused by pollution and degradation in water quality (Dudgeon, 2005). The growing

water demand increases effluent returns into the ecosystem, this alters and reduces the natural state of the rivers and associated impoundments by adding chemicals to the ecosystem. The changes in chemical composition of the water influences the biota of an aquatic ecosystem and its surroundings.

The Mooi River originates north of Potchefstroom and due east of Koster in the North-West Province in the Boons area, it flows south to join the Vaal River just south-west of Potchefstroom. The Mooi River catchment includes problematic areas (in terms of pollution) such as the far West-Rand of Gauteng, where the Wonderfonteinspruit originates, contributing pollutants associated with mining activities (Venter *et al.*, 2013). The Mooi River tributaries include Wonderfonteinspruit and Loopspruit. Several impoundments are situated along the Mooi River catchment including the Klerkskraal, Klipdrift (Loopspruit catchment), Boskop and Potchefstroom dams (Barnard *et al.*, 2013). The Mooi River catchment receives an array of contaminants from a wide variety of point and non-point sources. For example the Wonderfonteinspruit has multiple abandoned tailing dams, agricultural activities along the Mooi River and its tributaries and urban pollution associated with Potchefstroom and the activities therein. There are also various active gold mines in the area that contribute to heavy metal pollution and Acid Mine Drainage (AMD) (Annandale and Nealer, 2011; Barnard *et al.*, 2013; Winde, 2010a).

The Mooi River catchment area is underlain by dolomite and three of these dolomite compartments (Bank, Oberholzer and Venterspost) located in the Wonderfonteinspruit catchment, are used by gold mines and are dewatered (Winde, 2010a). The water of the Mooi River and its tributaries is used by local municipalities that include Potchefstroom, Fochville and Carletonville. Developing communities such as Kagiso, utilise water from the Rietvlei outside Krugersdorp, which in turn contributes to the Wonderfonteinspruit and causes pollution to the Mooi River. Several large industries located in the town of Potchefstroom and surrounding areas as well as farmers abstract water from the Mooi River (Van Aardt and Erdman, 2004). The Mooi River and its catchments are utilised by a great variety of stakeholders (mining, farming and production industries to name a few), hence there is a need to study the effects these activities have on the aquatic ecosystem, in order to determine the influences of the various uses.

Water resources must be preserved and monitored (Rotter *et al.*, 2011). One method to assess aquatic ecosystems is through bio-monitoring. A lack of resources makes it impossible to analyse all physical and chemical constituents at once, and thus we use indicators. The use of

biota to evaluate the condition of an aquatic ecosystems is relatively simple and rapid. Aquatic biota are continuously exposed to the pollutants in the water, and they will thus reflect the effect of the pollution, in the area (De la Rey *et al.*, 2004). The overall condition of an aquatic ecosystem is determined by the interaction of all its physical, chemical and biological components. The biological responses of the ecosystem are then used to monitor change in the specific environment (Roux, 1999). In addition indicators are used to give us information about the state of environmental quality not obtainable in other ways, including synergistic and antagonistic effects of pollutants.

## 1.2 Factors influencing water quality of the Mooi River

### 1.2.1 Description of the Mooi River catchment

The Mooi River is situated in the North-West Province of South Africa, and is one of the tributaries to the Vaal River. The Mooi River is in the Highveld Ecoregion, classified as Ecoregion 11 in the Level 1 Ecoregion classification (Klynhans *et al.*, 2005). The elevation of the Ecoregion is 1400 to 1800m above sea level, and means that the landscape is navigated by meandering rivers, such as the Mooi and Vaal rivers. According to Klynhans *et al.* (2005) the rainfall ranges between 400 and 900 mm in the summer months but not evenly distributed throughout the region and has an average evaporation potential of 1650mm (van der Walt *et al.*, 2002). Average temperatures range from a maximum of 21-24°C and a minimum of 2-6°C. The region is dominated by grasslands, and is susceptible to frost, fires and heavy grazing (historically by wild animals, now by cattle and sheep). The region is however heavily degraded due to the expansion of communities, overgrazing of the grasslands, planted wattle and eucalyptus, agriculture (growing crops and irrigation) and mining of gold and coal (Low and Rebelo, 1998; Cowling *et al.*, 1997; Mallett, 1999). The Mooi River catchment has a total area of 1800km<sup>2</sup>.

The Mooi River originates near Koster (a small town consisting predominantly of farmers in the area), in the Boons area, and flows south towards the confluence with the Vaal River between Orkney and Potchefstroom. The Mooi River catchment is underlain with dolomite, and this changes the chemical properties of the river, causing higher pH and electrical conductivity levels and high calcium and magnesium concentrations (Henderson-Sellers, 1991) as the Mooi River flows downstream. As the Mooi River flows south it encounters several sources of pollution. The first is agricultural and occurs between the Bovenste Oog (where the Mooi River originates) and the first impoundment located in the Mooi River (Klerkskraal Dam) (Venter *et al.*, 2013). In the North-West Province of South Africa, agricultural irrigation consumes 62% of surface water, this is the largest single water use in the country (Schreiner and Van Koppen, 2002). The amount of water use leads to runoff and ground water pollution by herbicides and pesticides, top soil and the nutrients used in excess. The effects of these pollutants are discussed in section 1.2.3. Between Klerkskraal Dam and the Vaal River diamond diggings are common and contribute to the pollution and alteration of the Mooi River floodplains and vegetation throughout (Currie, 2001; van der Walt *et al.*, 2002).

The second impoundment found in the Mooi River is the Boskop Dam, and upstream of Boskop Dam the river is impacted upon by pollutants associated with the tributaries of the Mooi River. First and the most influential is the Wonderfonteinspruit. The Wonderfonteinspruit originates in the far West-Rand of Gauteng between Krugersdorp and Randfontein, around abandoned goldmines, and their residue deposits (Riedel, 2003). Some of the richest goldmines are located in the Wonderfonteinspruit catchment area, having produced approximately 18000 tons of gold up to 1994 (van der Walt *et al.*, 2002). Several dams (Donaldson Dam, Harry's Dam and Andries Coetzee Dam) are located in the Wonderfonteinspruit and receive water pumped from dolomitic compartments. Along the Wonderfonteinspruit there are several communities utilising the water of the spruit, one of these is the Westonaria community which deposits sewage effluent back into the Wonderfonteinspruit. The upper Wonderfonteinspruit is also contaminated by the developing community of Kagiso, depositing organic sludge of uranium, from the nearby tailings into the Rietvlei wetland, providing water to the Wonderfonteinspruit. The Wonderfonteinspruit flows towards the Mooi River and reaches the Venterspost dolomitic compartment where the stream is diverted into a 1m diameter pipe 32km long to prevent water flowing back into three dewatered dolomite compartments (Venterspost, Bank and Oberholzer) and then enters used irrigation canals and its original streambed in the Boskop-Turffontein Compartment (van der Walt *et al.*, 2002; Winde, 2010a). In the lower Wonderfonteinspruit large scale mining is the main land use, leading to a lower water table, and the formation of sink holes. Winde (2010b) claims the Wonderfonteinspruit dries up in the dry months. It however replenishes the dolomitic karst aquifer Boskop-Turffontein Compartment, which feeds Boskop Dam, is thus contributes to the source of Potchefstroom's drinking water. The mines in the Wonderfonteinspruit area use lime to treat the effluents, and together with the dolomitic geology, there is little concern for Acid Mine Drainage (AMD). AMD may still however prove to be of concern as tailings dams having pH levels as low as 1.7 (Wittmann and Förstner, 1977). Another mining related contaminant that is of concern is Salt Mine Drainage (SMD) (Labuschagne, 2007), the effects of these two mining related contaminants will be presented in section 1.2.2.

Between the Wonderfonteinspruit and Boskop Dam the Gerhard Minnebron Oog joins the Mooi River, with dolomitic geology. Peat is mined on the farm and high salt concentrations enter the Mooi River. The Boskop Dam Nature Reserve, is a popular destination for the residents of nearby communities, with fishing and camping activities being frequent over the weekends and holidays.

The Mooi River flows from Boskop Dam toward Potchefstroom Dam with agricultural activity taking place all along the river. The town of Potchefstroom has a population of 250 000, sustaining a university, several large industries such as Nestlé, South African Breweries depot, an abattoir and several fertilizing manufacturers. A phospho-gypsum heap is also located outside Potchefstroom, contributing to the pollution of the Mooi River. The Wasgoedspruit is a canalized tributary of the Mooi River, containing industrial effluent from the above mentioned industries, along with urban- and storm water runoff, flowing into the Mooi River without prior treatment. Trompie Kitsgras is a producer of several types of grass and is situated on the banks of the Mooi River. The waste water treatment plant of Potchefstroom is situated at the southern town edge, releasing treated sewage back into the Mooi River. Heavy rainfall in the summer months may cause overflows at the waste water treatment plant and raw untreated or semi-treated sewage may flow into the Mooi River.

The Loopspruit is another large tributary of the Mooi River and joins the Mooi River downstream of Potchefstroom, it is utilised by farmers for the irrigation of their crops, and grazing animals. Two gold mines are situated in the Loopspruit as well as the informal settlement Kukosi, situated between the two goldmines and Klipdrift Dam. These three pollution sources cause elevated nutrient levels in the Loopspruit and the effects will migrate towards the Mooi River (van der Walt *et al.*, 2002).

After the Loopspruit, the Mooi River flows downstream towards the Vaal River, with diamond diggings and heavy agricultural activity (irrigation) along the Mooi River having an effect on the water quality.

### **1.2.2 Mining related pollution**

The Wonderfonteinspruit joins the Mooi River just north of the Boskop Dam. Ongoing, large-scale mining in the Randfontein area, and the resulting mine effluents discharged into Wonderfonteinspruit (Coetzee *et al.*, 2006), are a cause of concern when assessing the water quality of the Mooi River. A description of the Wonderfonteinspruit and the structure regarding layout and pollution sources can be found in section 1.2.1. Mining leads to physical and chemical changes in water quality (Ashton *et al.*, 2001).

The chemical constituents contained within mine effluent vary depending on the nature of the type of mining. The contaminants may have synergistic effects on an ecosystem that are difficult to quantify (Pulles *et al.*, 1996). Effects include, nutrient enrichment, pH fluctuations and

dissolved oxygen reduction. Mining not only impacts the local environment but it pollutes on a regional scale as the effluent may be widely distributed through drainage systems. A serious problem that occurs with mining related pollution is acid mine drainage (AMD). AMD acidifies a water body and increases bioavailability of heavy metal contaminants and thus has a negative effect on all biota in the area (Newete *et al.*, 2014). In a study done by Wittmann and Förstner, (1977) tailing dams in the West-Rand had pH levels of 1.7, a low acidic pH of 1.7 will have an effect on the pH of the Mooi River although it is buffered by the dolomitic geology. Another contaminant found in the Wonderfonteinspruit is Salt Mine Drainage (SMD), this occurs from the dewatering of dolomitic compartments, the treatment of mining effluent with lime and Peat mining at Gerhard Minnebron. The salts, such as high sulphates, magnesium and calcium cause an increase in the electrical conductivity. The effects of electrical conductivity are discussed in section 1.2.1.

As already mentioned, the geology of the Mooi River catchment is underlain with dolomite and the Wonderfonteinspruit contains three dewatered dolomite compartments (section 1.2.1), the three dams in the Wonderfonteinspruit catchment (section 1.2.1) are also heavily contaminated with raw dolomite dewatering effluent, having an alkaline effect on the water downstream.

Heavy metal pollution such as Uranium, from the Wonderfonteinspruit (Winde, 2010a&b) may have a negative effect on diatoms and macroinvertebrates, as the metal sensitive species are eliminated and deformed (diatoms) (Hirst *et al.*, 2002; Medley and Clements, 1998).

### **1.2.3 Agriculture**

The North-West Province of South Africa is well known for its farming, particularly maize production. Pesticides, herbicides and fertilizers are often used in the production of crops and this coupled with extensive irrigation, allows these products to be introduced into the aquatic ecosystem.

Agriculture is particularly associated with non-point source pollution, the levels of which escalate when the agricultural activities are poorly managed (Bermudez-Couso *et al.*, 2007). These activities include the preparation of the fields for planting, and the accompanying soil disruption, erosion, removal of vegetation and a loss of biodiversity. The loss of vegetation has a natural tendency to cause erosion, and thus increases the turbidity and sediment load within a river (Dallas and Day, 2004). These same practices also contribute to the salinisation of rivers (Williams, 2001; Williams, 1987), increase in nutrient (phosphorus and nitrogen) concentrations

(Smil, 2009) and cause eutrophication which has an immediate effect on aquatic biota (Chambers *et al.*, 2003).

Pesticides are used in the North-West Province in the months of March/April and August/October when crops are planted. Two of the highly utilised pesticides are deltamethrin and cypermethrin (Ansara-Ross *et al.*, 2008) and the two most used herbicides are glyphosate and 2,4-Dichlorophenoxyacetic acid. Pesticides and herbicides enter the river via groundwater and runoff, and have an acute and chronic effect on biota (Helfrich *et al.*, 2009). Herbicides, such as glyphosate have a long half-life and kills all plants that are not inoculated against it. Toxins may affect aquatic biota causing weight loss, reproductive abnormalities, loss of awareness, and the ability of organisms to tolerate temperature variations (Ward *et al.*, 2002).

#### **1.2.4 Urban runoff via Potchefstroom and wastewater**

The rapid growth of urban, and informal areas in South Africa is one of the biggest ecological problems South Africa is currently facing, often informal settlements have poor infrastructure and sanitation (Wimberley and Coleman, 1992). Urbanization in and around water bodies such as the Mooi River alters stream flow and degrades habitat, in an around the water body (Leopold, 1968; Finkenbine *et al.*, 2000).

Potchefstroom has a large industrial area, and is surrounded by several informal and semi-formal communities such as Ikageng and Promosa (Van Aardt and Erdman, 2004). Potchefstroom is an urban ecosystem that is influenced by human activities, ecological processes and the interactions between them (Grimm *et al.*, 2000).

Effluents associated with urban runoff include suspended solids, chemicals derived from a variety of sources (industrial and commercial) and human waste effluents (Epstein, 2002) as well as storm water. Although management principles are in place concerning urban runoff, the Waste Water Treatment Plant (WWTP) cannot control everything, and it is possible, and often the case, that these effluents enter an aquatic ecosystem untreated.

An increase in conductivity and elevated levels of nutrients is commonly associated with urban runoff (Walker and Pan, 2006). Further effects are sedimentation, eutrophication, thermal pollution, a decrease in dissolved oxygen, microbial contamination, salinisation, trace metal contamination, pesticide and hydrocarbon contamination (UNEP GEMS, 2008).

## 1.3 Water quality changes

Changes in water quality/chemistry in aquatic ecosystems alter the compounds in the system and can have significant effects on the bioavailability of said compounds (Carere *et al.*, 2011). In addition, the rate of flow, seasonal changes and the contribution of biological activities change the chemical and physical composition of surface water (Augustyn *et al.*, 2012). The constant monitoring and management of water is important, and measurement of certain parameters gives us a snapshot of the water quality at a specific time. A number of water quality variables were selected for this study. The most important of these, in relation to the growth and reproduction of aquatic biota are nutrients, pH, temperature, electrical conductivity (EC) (Saunders *et al.*, 2009; Freeland *et al.*, 1999). These variables will be discussed below.

### 1.3.1 Nutrients

Human activities such as agriculture, industry and waste water treatment plants have an effect on the amount of nutrients available in an aquatic ecosystem (Braid and Ong, 2000; Li *et al.*, 2009).

The phosphate and nitrogen in water are collectively referred to as nutrients (Bouamra *et al.*, 2012). Phosphate and nitrogen are important nutrients in an aquatic ecosystem as they are essential in metabolic processes that produce proteins and phospholipids during DNA and RNA synthesis (Conley *et al.*, 2009).

Nutrient enrichment of aquatic ecosystems has been linked to cyanobacterial (blue-green algal) blooms that are harmful to the environment (Paerl *et al.*, 2001). Algae in general bloom in the presence of excessive nutrients in a water body as a result of eutrophication, and this affects the biological integrity of an aquatic ecosystem (Paerl *et al.*, 2011). Eutrophication has several important effects on aquatic biota.

Dissolved oxygen was not measured in this particular study but the effects that nutrients have on dissolved oxygen is worth mentioning. A decline in dissolved oxygen is one of the main effects eutrophication has on an aquatic ecosystem (Deegan and Buchsbaum, 2005). High nutrient loads can cause anoxic conditions as the biomass produced decomposes, this results in the death of aquatic biota and in the long term alters the community structure of aquatic biota (Deegan and Buchsbaum, 2005; Castro *et al.*, 2003).

An increase in turbidity of surface water has been associated with eutrophication (Doan *et al.*, 2015). Turbidity increase for a prolonged time period has effects on biota and community

structure, the organisms that are mostly affected are the primary producers and primary consumers (Dallas and Day, 2004).

### **1.3.2 Temperature**

The average surface temperature of the earth has increased by 0.6°C over the last 100 years (Houghton *et al.*, 2001). Temperature variations depend on the characteristics of a river such as the source of water, groundwater contribution, flow rate, volume of water and inflow from tributaries (Dallas, 2009).

Temperature increase in an aquatic ecosystem influences change in water chemistry reducing oxygen solubility (Carere *et al.*, 2011). Warmer temperatures trigger a longer than normal stratification period in dams, mixing in the water column is delayed thus bringing cold, low oxygen level and nutrient-rich water to the surface in late summer and autumn (Coats *et al.*, 2006). Increased temperature also influences the chemical composition of substances such as cyanide - its toxicity increases with temperature (Dallas and Day, 2004).

Both the distribution, and physiology of aquatic biota are dependent on water temperature (Hughes, 2000). Higher temperatures increase the metabolic rate of organisms, and thus they require more nutrition and oxygen demand increases (Zang *et al.*, 2015) as chemical reactions increase this may lead to the increased intake of toxins (Carere *et al.*, 2011). Most aquatic biota do not have the ability to regulate their own body temperature, colder temperatures cause decreases metabolic rate and constrains growth (Falkowski and Raven, 1997).

### **1.3.3 pH**

The Mooi River catchment is underlain by dolomite (Van Aardt and Erdman, 2004) which together with mining effluents in the Wonderfontein spruit (Coetzee *et al.*, 2006) influence the pH levels of the Mooi River (section 1.2.2).

The stoichiometry, kinetics and equilibrium of chemical reactions in an aquatic ecosystem are dependent on pH (Yang *et al.*, 2014; Flores-Alsina *et al.*, 2015) which means that pH influences the way molecules and elements move through membranes and the solubility of heavy metals.

The calculation of pH in a water body is based on the basic and acidic compounds dissolved in a water body and its temperature (Kim and Jeong, 2016). Changes in pH, whether the water body is either acidic or alkaline, affect the bioavailability of compounds such as cyanide, nutrients, trace metals and biocides (Dallas and Day, 2004).

Eukaryote organisms have several organelles that function separately of each other to form one functioning unit (Gabaldon and Pittis, 2015). Each organelle has a functional pH range and variations in pH different to the organisms niche has an adverse reaction to cells by influencing the properties of protein based films in the cell wall, and thus is able to cause dysfunctions in the cells and in turn affects the organism in a negative way (Yu *et al.*, 2015; Masamba *et al.*, 2016).

These changes on cellular level affect certain individual species more than others, potentially causing the loss at a less tolerant species, and thus loss of one species may have significant effect on community structure and ecological function (Wang *et al.*, 2016).

#### **1.3.4 Electrical Conductivity and Total Dissolved Solids**

The electrical conductivity (EC) of a water body is based on the ability of the water to conduct an electrical current (Hem, 1989). EC is widely used in water monitoring and usually has a direct relationship with Total Dissolved Solids (TDS) (Marandi *et al.*, 2013). TDS measurement indicates the amount of dissolved salts in the water, and correlates directly with the EC (DWAf, 1996). EC measurements are a useful tool in detecting waste water pollution, and provide us with indistinct information for contaminant discharges (de Sousa *et al.*, 2014). The bioavailability of metals are impacted by various physical and chemical changes in an aquatic ecosystem, one of these key factors is salinity (Blewett and Wood, 2015).

## 1.4 The use of aquatic organisms as bioindicators

The overall condition of an aquatic ecosystem is determined by the interaction of all its physical, chemical and biological components. The biological responses of the ecosystem are then used to monitor change in the specific environment (Roux, 1999). Biota live within the aquatic system, and are exposed to all of the above mentioned components, toxins and pollutants accumulate within the organism and thus provide us with a long and short term indication of environmental conditions (Taylor *et al.*, 2005 ).

Macroinvertebrates and diatoms are good ecological indicators (see discussion below) and it was found that macroinvertebrates give a good indication of catchment disturbance, while diatoms relate better to changes in water quality (Sonneman *et al.*, 2001). In this study diatoms and macroinvertebrates were used as indicator organisms, the use which will briefly be explained.

### 1.4.1 Diatoms

Diatoms are unicellular protists, are characterised by possessing silica walls (the frustule), and are one of the most common groups of algae found in water bodies (Moustafa *et al.*, 2009). The group is responsible for approximately one fifth of the primary production in the world (Nelson *et al.*, 1995; Round *et al.*, 1990). The structures of diatoms and their functions have been studied for over a hundred years (Tanaka *et al.*, 2015). Their robust and highly patterned silica structure makes them resistant to breaking during sample collection and preparation which aids in species identification (Taylor *et al.*, 2009). Most diatoms are autotrophic and contain one or several chloroplasts and they photosynthesise by using chlorophyll *a*, *c* and fucoxanthin, and are brown to yellow-brown in appearance (Janse van Vuuren *et al.*, 2006).

Diatoms are primary producers that utilise inorganic nutrients for growth and reproduction, and as such thus they can provide useful information concerning the health at the base of an aquatic ecosystem (McCormick and Cairns, 1994). Diatoms respond directly to changes in nutrient and pollution changes in the environment, and have proven useful to indicate specific problems in water quality such as, heavy metals, sewage (organic) effluents and eutrophication (Kriel, 2008; Taylor *et al.*, 2005).

Diatoms colonize new habitats rapidly and can be found in great abundance in a variety of aquatic ecosystems (Round *et al.*, 1990). Diatoms are highly abundant in most ecosystems with many thousands of species already described. Each diatom species has its own unique

requirements for survival and thus the changes in water quality in turn cause a change in the growth and abundance of diatoms. The growth of some species is inhibited while others flourish and this in turn transforms the community structure and this change can be used to indicate the state of the ecosystem (Cholnoky, 1960). Diatoms that are attached to substrate use the nutrients in the aquatic ecosystem as an energy source and thus they respond directly to fluctuations in water chemistry (Kriel, 2008).

### **1.4.2 Diatoms in biomonitoring**

Assessing biological integrity of aquatic ecosystems is an essential part of maintaining human health. That said, humans and their activities alter the environment of aquatic ecosystem and thus alter the biotic interactions between different trophic levels (Karr and Chu, 1999). Alterations in the aquatic ecosystem cause the biota to respond in a predictable manner (Odum, 1985) and this allows us to use biota as biomonitoring tools.

Diatoms serve as reliable biological indicators and community structure respond to changes in pH, salinity, nutrients and organic enrichment in the water (Koekemoer and Taylor, 2010). Diatoms are one of the best groups of organisms that can be found in any aquatic system to reflect on the changes in the water dynamics associated with human activities (Kelly, 2002). Diatom community structure varies in species composition, due to the wide range of tolerances within the diatom community. Water chemistry and habitat requirements play a large role in the diatoms found in said community structure (Bere. 2014; Harding *et al.*, 2005; Round *et al.*, 1990).

Diatom community structures are responsive to various environmental stressors such as temperature, current velocity, grazing and water chemistry, and differences also occur due to temporal and spatial variation (Pan *et al.*, 1996; Round, 1991). In relation to water quality diatoms respond to eutrophication because they are affected by nutrient abundance and light transmission (Tilman *et al.*, 1982).

There are however, problematic aspects of using diatoms and they are:

- The preparation time from raw material to microscope slide takes relatively longer than field based techniques.
- Inexperience may cause difficulty in identification.
- Counting frustules may take a long time due to inexperience.
- Diatoms cannot usually be found where there is no water.

### 1.4.3 Diatom studies in South Africa

Diatoms were first recognised as water quality indicators in South Africa by Dr. B.J Chohnoky. After Chohnoky in (1968) other scientists started diatom research in South Africa (Archibald, 1972; Schoeman, 1976). In more recent times the possible use of diatom indices in South African in freshwater aquatic ecosystems has been investigated extensively (Bate *et al.*, 2002; de la Rey *et al.*, 2004; Harding *et al.*, 2005; Taylor *et al.*, 2005; Taylor *et al.*, 2007a). Diatoms have been used in The River Health Program in South Africa as an indicator of water quality (RHP, 2005).

## 1.5 Macroinvertebrates

Macroinvertebrates are a diverse group of animals that occur in aquatic ecosystems which include worms, crustaceans, mollusks, mites and other insects. Macroinvertebrates have no backbone, and come in an array of sizes that can be retained in a net with a mesh size of between 0.2 and 0.5mm. Macroinvertebrates inhabit different habitats within a stream and represent almost every taxonomical group occurring in freshwater habitats. They live on, under and in different habitats, substrates in and around an aquatic ecosystem (Hynes, 1970; Winterbourn, 1999). Macroinvertebrates are an important link in the aquatic ecosystem and feed on periphyton, break down organic matter, cycle nutrients and they are prey to larger predators. Macroinvertebrates have limited mobility, and have a wide range of sensitivities to water quality changes (Jimoh *et al.*, 2011; Uwem *et al.*, 2011; Abel, 1989). Macroinvertebrates (as with diatoms) have their own unique environmental requirements and a change in water quality variables and in the extent of pollution will have a positive or negative influence on taxon depending on the sensitivity of said organism (Dallas and Day, 1993). Importantly macroinvertebrates are limited in their range of movement and thus are confined to their current habitat (Barbour *et al.*, 1999).

Aquatic macroinvertebrates have multiple life stages depending on taxa, macroinvertebrates could be (USEPA, 2002b):

- Multivoltine and bivoltine, having life cycles of half a year or less, usually seasonal and include midges, blackflies and mayflies.
- Univoltine, producing one generation a year, with a life expectancy of one year, and include, caddisflies, mayflies and stoneflies
- Semivoltine, with generations and lifecycles being more than one year and include, mussels, crabs and a dragonflies.

### 1.5.1 Macroinvertebrates as bioindicators

Macroinvertebrates are used widely as bioindicators because of their sensitivity to changes within aquatic ecosystems (Rosenberg and Resh, 1993). Macroinvertebrates are a critical part of aquatic ecosystems and perform roles that are critical to maintain functionality in an ecosystem (O’Keeffe and Dickens, 2000). Macroinvertebrates can indicate specific environmental issues, as they are limited on movement and thus are confined to their current habitat (Barbour *et al.*, 1999). Macroinvertebrates are prey to several organisms, involved in the processing of organic matter and they are a big part of the biodiversity that is supported by the aquatic ecosystem (Snaddon, 2009). Metal pollution via mines, urban runoff and industries has a particular influence on orders such as Tricoptera, Ephemeroptera and Placoptera, these sensitive orders presence are lost within a community in the presence of metal pollution (Beasley and Kneale, 2003). Another advantage to using macroinvertebrates are that stream composition, climate and altitude have an effect on the distribution and community structure of macroinvertebrates (Grab, 2014). Macroinvertebrates are easily visible, easy to identify and they have rapid life cycles and thus the community structure adapts to changes in an aquatic ecosystem (Dickens and Graham, 2002).

The limitations regarding the use of macroinvertebrates as ecological indicators include the following (De la Rey *et al.*, 2004)

- Distribution of macroinvertebrates is affected by an array of factors not just water quality.
- Natural distribution of species may occur regardless of water quality.
- Species are sometimes restricted by disruption barriers or obstacles to migration and are thus not found in a region and not because of water quality.
- Seasons and flow rate influence species composition.
- Water level may be too deep and it can be difficult and dangerous to sample.
- Community structure is influenced by flow disturbance created by dams, bridges etc.

Macroinvertebrates are often used in studies to provide information on the health of the ecosystem (Wolmerans *et al.*, 2014; de la Ray *et al.*, 2004; Mantel *et al.*, 2010). These organisms were chosen in this study to aid in determining the health of the Mooi River ecosystem, and as a biomonitoring tool to assess environmental change throughout 2014.

## **1.5.2 History of macroinvertebrate studies in South Africa**

Macroinvertebrates can be used as a bioassessment tool for the purpose of water quality control and to determine the ecological status of a river system (Hart and Fuller, 1974; Dickens & Graham 2002). Change can therefore easily be noted in changing populations. In 1998 an index was developed for a quick and cost effective means to assess water quality using macroinvertebrates, the system was called SASS (South African Scoring System) (Chutter, 1994; Chutter 1998). SASS is based on macroinvertebrates and their sensitivity towards pollutants, where each taxon is allocated a sensitivity score, and the presence or absence of certain taxa in river systems (Gordan *et al.*, 2015).

In 2002 Dickens and Graham released SASS Version 5 where sampling methods and identification techniques were standardised. SASS5 is a quick easy bio-monitoring method to determine the ecological status of a river system (Wolmerans *et al.*, 2014). It is an ideal method to use when determining the health of rivers with low to medium flow, but not ideal for wetlands and estuaries (Dickens and Graham, 2002). SASS5 is ideal for impact studies but can't indicate a pollution source so incorporation of chemical studies is advisable when dealing with pollutants (Gordan *et al.*, 2015).

## **1.6 Research outcomes**

### **1.6.1 Research question**

How will the natural and anthropogenic influences on the Mooi River affect the health of the aquatic ecosystem as the river progresses downstream towards the Vaal River?

### **1.6.2 Hypotheses**

- Degradation of the aquatic ecosystem health will occur gradually as the Mooi River progresses downstream towards the Vaal River.
- The influence that Potchefstroom exerts on the quality of the Mooi River will be greater than the pollution associated with and emanating from the Wonderfonteinspruit.
- Impoundments will have a positive effect on aquatic ecosystem health, as indicated by recovery and the change in the community composition of biotic indicators.

### **1.6.3 Aims**

- First, to determine the aquatic ecosystem health of the Mooi River
- Second, to assess organism community structure change in relation to the water quality variables.
- Third to assess the organism community structure change in relation to temporal and spatial variation.
- Lastly to compare the use of diatom and macroinvertebrate assemblage data for indicating ecosystem health.

### **1.6.4 Objectives**

- Collecting data on several water variables, diatom- and macroinvertebrate community structures from eight pre-selected sites in the Mooi River every month throughout the 2014 calendar year.
- Comparing the community structure data of the above mentioned biota with literature available on the effects pollutants have on an aquatic ecosystem, and the biotic response to pollutants.
- To use the data collected, and integrating the components to assess the health of the Mooi River aquatic ecosystem.

## **1.7 Dissertation outline**

The chapters that follow were selected as individual parts and contain information relating to the subject of the chapter. In the end a chapter is dedicated to conclusions where all the chapters are seen as one working unit. Each chapter contains an introduction, materials and methods, results and discussion and a conclusion of the chapter.

Chapter 2 encompasses site descriptions and the reasons for site selection. The site selection section in the material and methods of each chapter will refer back to this chapter unless stated otherwise. The rest of Chapter 2 contains the material and methods used in the study.

Chapter 3 focuses on the measured water quality variables and the change thereof throughout the period of the study and between study sites. The water quality results will be used as supplementary information for the chapters that follow.

Chapter 4 focuses on the diatoms, the various diatom-based indices, community structure and the influences that impact them.

Chapter 5 is used to discuss macroinvertebrates, various macroinvertebrate indices and the community structure and the influences that impact them.

Chapter 6 is a summary of all the chapters, a unified overview of aquatic ecosystem health within the Mooi River system accompanied by general conclusions and recommendations.

Chapter 7 is the reference list, containing all the references used in the study.

Appendices follow the last chapter and include detailed figures, tables and interesting findings in the study that will receive later attention and research not included in this dissertation.

# CHAPTER 2 MATERIALS AND METHODS

## 2.1 Study Sites

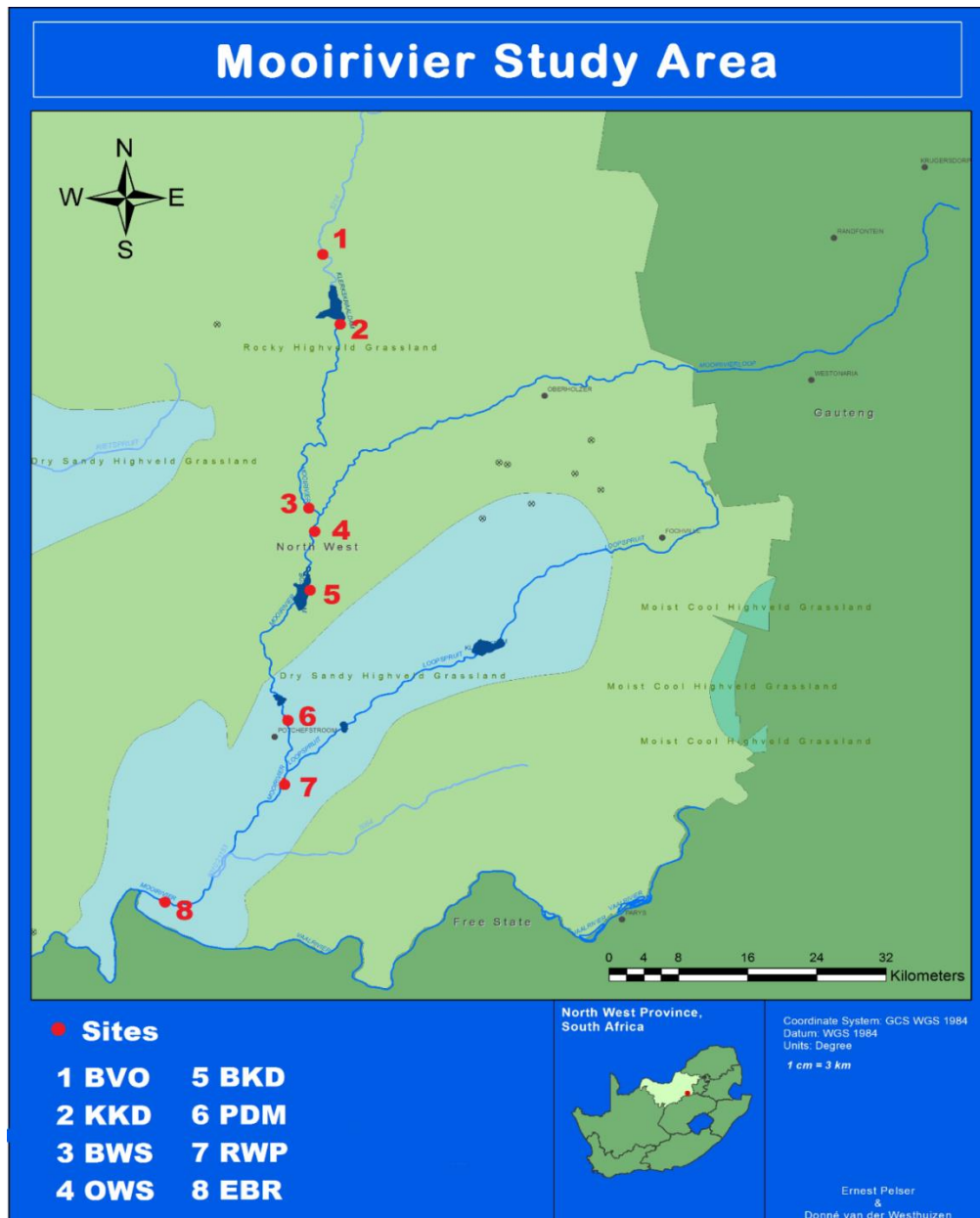


Figure 2-1: Map of the Mooi River system, indicating the sites chosen for the present study. The study sites are (upstream to downstream) Bovenste Oog (BVO), Klerkskraal Dam (KKD), Upstream of Wonderfonteinspruit (BWS), Downstream of Wonderfonteinspruit (OWS), Boskop Dam (BKD), Downstream of Potchefstroom Dam (PDM), Trompie Kitsgras (RWP) and Elbrixon Bridge (EBR).

The Mooi River has several influences throughout the catchment, eight sites were selected from the origin of the Mooi River to the Elbrixon Bridge, just before the Mooi River joins the Vaal River, this was in order to determine the effects of the different pollutants on the ecosystem.

### **2.1.1 Bovenste Oog (BVO)**

Bovenste oog (BVO) is a natural spring, located north of Klerkskraal Dam, it is the origin of the Mooi River, and is used as a reference point throughout the study.

BVO has shallow flowing waters, and a deep pool, the water is extremely clear. Aquatic vegetation and marginal vegetation are plentiful with an abundance of rocks and cobbles. The pool area of BVO has a sandy sediments with a muddy substrate.

Factors that may influence the water quality of BVO are grazing cattle which could influence the bacterial abundance and cause elevated nutrients.



**Figure 2-2 Left, the aquatic vegetation, stones and sand substrate are shown and right, the stones in a current just downstream of the figure on the left.**

### **2.1.2 Klerkskraal Dam (KKD)**

Klerkskraal Dam (KKD) is the first of three impoundments in the Mooi River. KKD is located just downstream of BVO and has no known sources of pollutants, and is used as a reference for the impoundments.

Sampling took place just below the dam wall, where there was plenty of marginal vegetation and rocks.

The only activity in the dam is recreational with the occasional fishing off of boats.



**Figure 2-3: The two photographs show the sampling site below the dam wall, rocks and vegetation are available for sampling.**

### **2.1.3 Upstream of Wonderfonteinspruit (BWS)**

Above Wonderfonteinspruit (BWS) is as the name suggests, it is the site just upstream of where Wonderfonteinspruit joins the Mooi River. The site was chosen in order to assess the influence of Wonderfonteinspruit on the Mooi River.

BWS is not unimpacted; several farms are located between KKD and BWS. Crops and cattle are the main potential sources of pollution.

Sampling at the site was difficult, the water was generally only about 20cm deep, with marginal vegetation, no stones present, and the sediment was made up of clay and sludge.



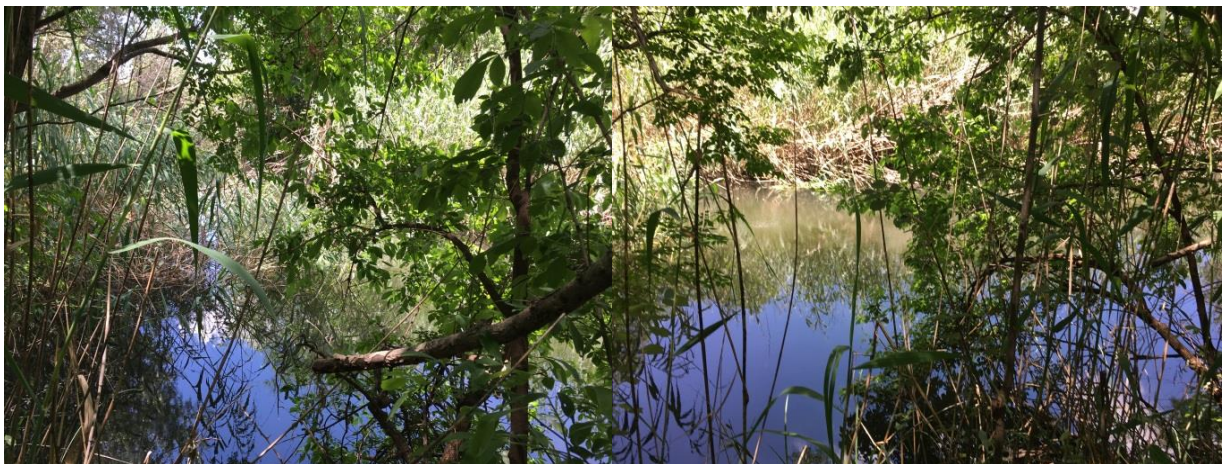
**Figure 2-4: Left illustrates the shallow water upstream of the sampling site and right the shallow water downstream of the sampling site.**

#### **2.1.4 Downstream of Wonderfonteinspruit (OWS)**

Downstream of Wonderfonteinspruit (OWS) is located downstream of the point where the Wonderfonteinspruit joins the Mooi River. OWS is situated on a farm that plants crops and has quite a number of cattle grazing on the farm.

This site was chosen to detect any influence of Wonderfonteinspruit and the potential mining impact on the aquatic ecosystem.

The sampling site is encroached on by trees and bushes, and is deep (approximately 1.8m) and slow flowing, with thick marginal vegetation, and sludge- clay sediment.



**Figure 2-5: Both photographs illustrate the deep flowing water of the site, heavily encroached upon by trees and bushes.**

### **2.1.5 Boskop Dam (BKD)**

Boskop Dam (BKD) is the second impoundment in the Mooi River system, and is a source of drinking water for the town of Potchefstroom.

The sampling site is in the dam itself with marginal vegetation, few trees and a stony substrate.



**Figure 2-6: Sampling site at Boskop Dam situated within the Boskop Dam Nature reserve.**

### **2.1.6 Mooi River at the bridge to Carletonville (PDM)**

This site is located approximately 3km below Potchefstroom Dam, and was chosen to determine the effect of impacts between BKD and PDM, at this point it is largely free from the influence of the town of Potchefstroom.

The sampling site had a combination of habitats that ranged from rocky areas instream, and out of stream to deep pools consisting of soft clay and sand. Marginal vegetation was present as well as several large willow trees partially shading the site.



**Figure 2-7: Left is upstream of the site and right is downstream of the site. Note the different sampling biotopes.**

### **2.1.7 Trompie Kitsgras (RWP)**

The site Trompie Kitsgras (RWP) is located just downstream of the waste water treatment plant (WWTP) and was allocated to determine the cumulative effect of Potchefstroom and the town's waste water treatment plant (WWTP) on the health of the ecosystem. The site was chosen to potentially detect the effect of urban runoff, agriculture (the Loopspruit joins the Mooi River just downstream of Potchefstroom and above RWP) and WWTP on the ecosystem.

The sampling site has several habitats that range from rocky areas in stream, and out of stream, and pools consisting of soft clay, sludge and sand. Marginal vegetation and aquatic vegetation are present.



**Figure 2-8: Left is upstream of the site and right is downstream of the site. Note the different sampling biotopes.**

### **2.1.8 Elbrixon Bridge (EBR)**

Elbrixon bridge (EBR) is located just upstream of where the Mooi River meets the Vaal River. Downstream of the bridge there is little to no pollution flowing towards the Vaal River. EBR is influenced by agricultural pollution, large irrigated lands and livestock farming.

The sampling site was deep (approximately 1.9m) and slow flowing with little to no rocks, and covered by marginal vegetation. The sediment was composed of mud and sand, the site is also used for water abstraction, the reason for which is unknown.



**Figure 2-9: Left and right show the deep flowing waters and marginal vegetation of EBR.**

## 2.2 Site attributes.

The following is a summary of the sites, potential impacts and geographical location.

**Table 2-1: A summary of the sites, relevance in terms of impact detection, coordinates and altitude.**

Site	Relevance	Possible Pollution source	Coordinates and Elevation
BVO	Reference site	Agriculture	26°11'53" S 27°9'53" E 1480m asl
KKD	Impoundment reference	Recreational	26°15'10" S 27°9'35" E 1470m asl
BWS	Upstream of Wonderfonteinspruit	Agriculture	26°27'19" S 27°7'38" E 1400m asl
OWS	Downstream of Wonderfonteinspruit	Agriculture Mining	26°30'9" S 27°7'32" E 1440m asl
BKD	Drinking water to Potchefstroom	Agriculture Mining	26°32'33" S 27°7'2" E 1350m asl
PDM	Upstream of Potchefstroom	Agriculture Recreational	26°41'04" S 27°06'01.8" E 1335m asl
RWP	Downstream of Potchefstroom	Urban Runoff Recreational Waste water treatment Loopspruit	26°45'51" S 27°5'29" E 1320m asl
EBR	Last site and final view of all factors	Agriculture	26°52'2" S 27°1'30" E 1310m asl

## **2.3 Water Quality**

### **2.3.1 Field Measurements**

Physical and chemical water quality variables were measured *in situ* before biological samples were taken at each site on the last Wednesday of every month in the 2014 calendar year. Electrical conductivity (EC), pH, total dissolved solids (TDS) and temperature measurements were taken using a HANNA multimeter (Model HI 9813-6) which was cleaned and calibrated before each sampling trip.

### **2.3.2 Laboratory analysis**

One liter samples were taken at each site every month, by filling sterilised bottles with surface water from each site, which were then immediately placed in a cool box. The samples were then taken to the Midvaal Water Company, where alkalinity, turbidity, dissolved calcium, dissolved magnesium, nitrite and nitrate, orthophosphate, sulphate, dissolved uranium and chlorophyll *a* were measured using standard accredited methodology. The laboratory at Midvaal Water Company is SANAS accredited (T0132).

## **2.4 Diatoms**

The methodology for diatom sampling and laboratory processing was that according to Taylor *et al.* 2005.

### **2.4.1 Diatom Field Collection**

Diatoms were sampled on the last Wednesday of each month during the year 2014. Diatoms were sampled from cobbles, boulders and vegetation depending on substrate availability at the site. Between 5 and 10 pieces of substrate showing signs of diatom growth were selected. The surface of the selected substrate was scraped with a toothbrush and rinsed, using river water, into a collecting tray. The collected sample was transferred to a container and ethanol was added (20% by volume) to preserve the sample.

## 2.4.2 Diatom preparation

Diatoms were cleaned using the hot hydrochloric acid (HCl) and potassium permanganate (KMnO<sub>4</sub>) method (Hasle, 1978).

Diatom samples were re-suspended and poured into a test tube through a coarse filter to keep debris (e.g. plant remains) out of the samples. The same amount of KMnO<sub>4</sub> was added to each sample, and left to stand for 24 hours. After 24 hours the colour changes from a purple colour to a brown colour indicating that the KMnO<sub>4</sub> had reacted with the organic material in the test tube. In a fume cabinet 10ml, 32% HCl was added to the samples and boiled in a beaker filled with water on a hotplate until the samples were clear. Hydrogen peroxide was added to all of the samples to confirm that no organic matter was left in the samples.

The samples were left to cool. After cooling sample contents were transferred to 13ml centrifuge tubes and placed in a centrifuge. The samples were centrifuged for 4 cycles of 10 minutes each at a speed of 2500 rpm, after every cycle the supernatant was discarded and 10ml distilled water was added. After the last cycle the supernatant was discarded and the diatoms re-suspended in a smaller volume of distilled water in order to provide a dilution suitable for slide preparation. The volume of water differs with every sample, depending on the concentration of diatom cells within the sample. One drop of 10% ammonium chloride (NH<sub>4</sub>Cl) was added to each sample to neutralise electrostatic charges which causes the diatoms to aggregate (McBride, 1988). The samples were then mixed with a vortex mixer, approximately 1ml of the clean dilute diatom suspension was placed on a clean 18mm round coverslips, and left to air dry. The rest of the suspension was poured into a glass vial with 99% ethanol to preserve the cleaned sample for future use. Slides and material are accessioned into the SAIAB National Diatom Collection currently housed at the North-West University Potchefstroom.

After the cover-slips had dried they were placed on a hotplate for a short period until the residual NH<sub>4</sub>Cl had sublimated. The hotplate and cover-slips were allowed to cool, and microscope slides were prepared for each sample. When the hotplate was at the desired temperature, the cover-slip was placed on the hotplate and 1-2 drops of Pleurax (mountant with high refractive properties – R.I. 1.73) was placed on the cover-slip. The microscope slide was lowered onto the cover-slip until the cover-slip was attached to the microscope slide. The slide was then inverted and placed on the hotplate. The Pleurax was allowed to boil slightly and the solvent evaporated. The slide was heated in this manner until the desired result was attained. The slide was then removed and left to cool.

### **2.4.3 Diatom enumeration**

The prepared microscope slides were observed with a Nikon 80i compound light microscope with a 100x oil immersion objective (1.4 N.A.) and differential interference contrast. The diatom frustules were enumerated using standard methods to a total of approximately 400 frustules.

### **2.4.4 Diatom identification**

Identification was done using mainly Taylor *et al.* (2007b). After initial identification of cells, these were confirmed by Dr J. C Taylor.

### **2.4.5 Diatom index calculations**

The data that were acquired after identification was sorted and formatted. The data were then entered into OMNIDIA (Lecointe *et al.*, 1993). OMNIDIA is software that aids with the interpretation of data and can be used to calculate various diatom indices scores.

It was decided that four diatom indices would be used:

- Percentage Pollutant Tolerant Valves (%PTV) (Kelly and Whitton, 1995) indicates the percentage of diatoms present in the community that is tolerant to polluted conditions; it also indicates the degree of organic pollution vs. eutrophication.
- Generic Diatom Index (GDI) (Coste and Ayphassorho, 1991), a relatively simple index that calculates its final score based on diatom tolerances to pollution at genus level.
- Biological Diatom Index (BDI) (Lenoir and Coste, 1996) can be used to accurately indicate on water quality, as it uses 14 water quality variables associated with diatom sensitivity, however it uses far less species than the SPI.
- Specific Pollution sensitivity Index (SPI) (CEMAGREF, 1982), contains more species in its database than any other index.

The maximum score of the above mentioned indices is 20, indicating a high ecosystem quality the exception is %PTV and is calculated out of 100. The table below indicates the pollution levels associated with the various index scores.

**Table 2-2: Interpretation of the various indices used in this study**

Index Score (20)	Ecosystem quality	Trophic level
>17	High Quality	Oligotrophic
15-17	Good Quality	Oligo-Mesotrophic
12-15	Moderate Quality	Mesotrophic
9-15	Poor Quality	Meso-eutrophic
<9	Bad Quality	Eutrophic

## 2.5 Macroinvertebrates

### 2.5.1 Sampling sites

For the purpose of the study, only 3 of the 8 sites were selected for sampling. The reason is that the SASS5 method was used which has various restrictions in terms of site selection. The following sites were not used during the study for SASS; motivations are included:

- Klerkskraal Dam- This is an impoundment and according to SASS5 requirements the method cannot be applied at impoundments.
- Above Wonderfonteinspruit- The water is very shallow, only ankle depth, there are no stones present in the area and the habitat is insufficient for a sample to give consistently accurate results.
- Below Wonderfonteinspruit- The water depth is dangerously high (shoulder depth), there were no stones in the area, and the sludge was deep.
- Boskop Dam- This is an impoundment and according to SASS5 requirements the method cannot be applied at impoundments.
- Elbrixon- The water depth is dangerously high (shoulder depth), there were no stones in the area, and it was decided that insufficient habitat was available.

Sites that were used had sufficient available habitat, was easily assessable and held no personal risks.

### **2.5.2 Sampling method**

Macroinvertebrates were sampled at all three sites on the last Wednesday of every month in the 2014 calendar year. Macroinvertebrates were sampled according to the SASS5 methods using a standardized 1000 µm mesh with a 30cm x 30cm frame SASS net at three different biotopes (Dickens and Graham, 2002). The biotopes included the following:

- Gravel, sand and mud (GSM),
- Vegetation (VEG) and
- Stones in current (SIC)
- Stones out of current (SOC)

The stones biotope was sampled for 120 seconds by vigorously kicking and disturbing the water to dislodge macroinvertebrates while the net was held downstream to collect the macroinvertebrates.

A stretch of 2m<sup>2</sup> was used to sample vegetation, sampling in the vegetation was done by sweeping the net vigorously through the roots and leaves of the plant, this was done with both marginal and submerged vegetation.

A sample time of 60 seconds was used to sample the gravel sand and mud biotope, sampling was done by kicking up dirt and sweeping over the dirt with the SASS net.

### **2.5.3 Identification**

Before sampling began, visual observation and identification of water surface invertebrates were made.

After sampling each biotope, the samples were tipped into a SASS tray. The macroinvertebrates were identified to genus level. Identification time was 15 minutes for each biotope or until no new taxa were identified for 5 minutes. The relative abundance rating was noted and the SASS score calculated.

### **2.5.4 Relevant reference sites and score attribution**

The Mooi River is located in Ecoregion 11 in the lower Highveld ecoregion according to the Ecoregion Level 1 classification (Kleynhans *et al.*, 2005). Each level 1 Ecoregion has been studied and relative SASS5 scores were calculated to be used as a reference site, the actual

SASS5 score was then be converted to a percentage in relation to the reference site (Dallas, 2007).

The maximum SASS5 and ASPT scores for the Lower Highveld Ecoregion are 144 and 6.4 respectively.

**Table 2-3: A representation of the ecological classes representing the SASS5 scores**

<b>Biological Category</b>	<b>Band/Ecological Category</b>	<b>Ecological Category Name</b>	<b>Description</b>	<b>Colour</b>
A		Natural	Unmodified natural	Blue
B		Good	Largely natural with few modifications	Green
C		Fair	Moderately modified	Yellow
D		Poor	Largely modified	Red
E		Seriously modified	Seriously modified	Purple
F		Critically modified	Critically or extremely	Black

## 2.6 Statistical analysis

The statistical analysis between the different datasets was conducted by means of Statistica version 12, software. The Kolmogorov-Smirnov and Lilliefors test for normality were used to determine whether the datasets were distributed parametrically. The distribution of the variables in the datasets was not parametric. The Kruskal-Wallis ANOVA (for non-parametric data), which can be used to compare multiple independent samples, was used to compare differences between the sites. A standard two-tailed Pearson correlation was used to determine significant differences ( $P < 0.05$ ) between water quality variables at the different sites, and multivariate statistical analysis was done using CANOCO 4.5 (Ter Braak and Šmilauer, 2002). Canonical correspondence analysis was done to illustrate spatial and temporal variation of water quality parameters measured as well as the diatom species and macroinvertebrate taxa. Detrended correspondence analysis was also used to illustrate spatial and temporal variation of diatom species as well as macroinvertebrate taxa.

# **CHAPTER 3 WATER QUALITY - MEASURED PHYSICO-CHEMICAL PARAMETERS**

## **3.1 Water quality**

The following chapter contains the results and discussion of the physical and chemical variables collected at each sampling occasion. The results are presented per water quality variable to illustrate the differences between sites further the results are presented as an average per site throughout the 12 months of sampling. A full table with individual results can be found in Appendix B Table A-1 and will be referred to during the discussion. Thereafter temporal and spatial results are presented to reflect the differences in water quality between sites and seasonal changes.

## **3.2 Results and discussion**

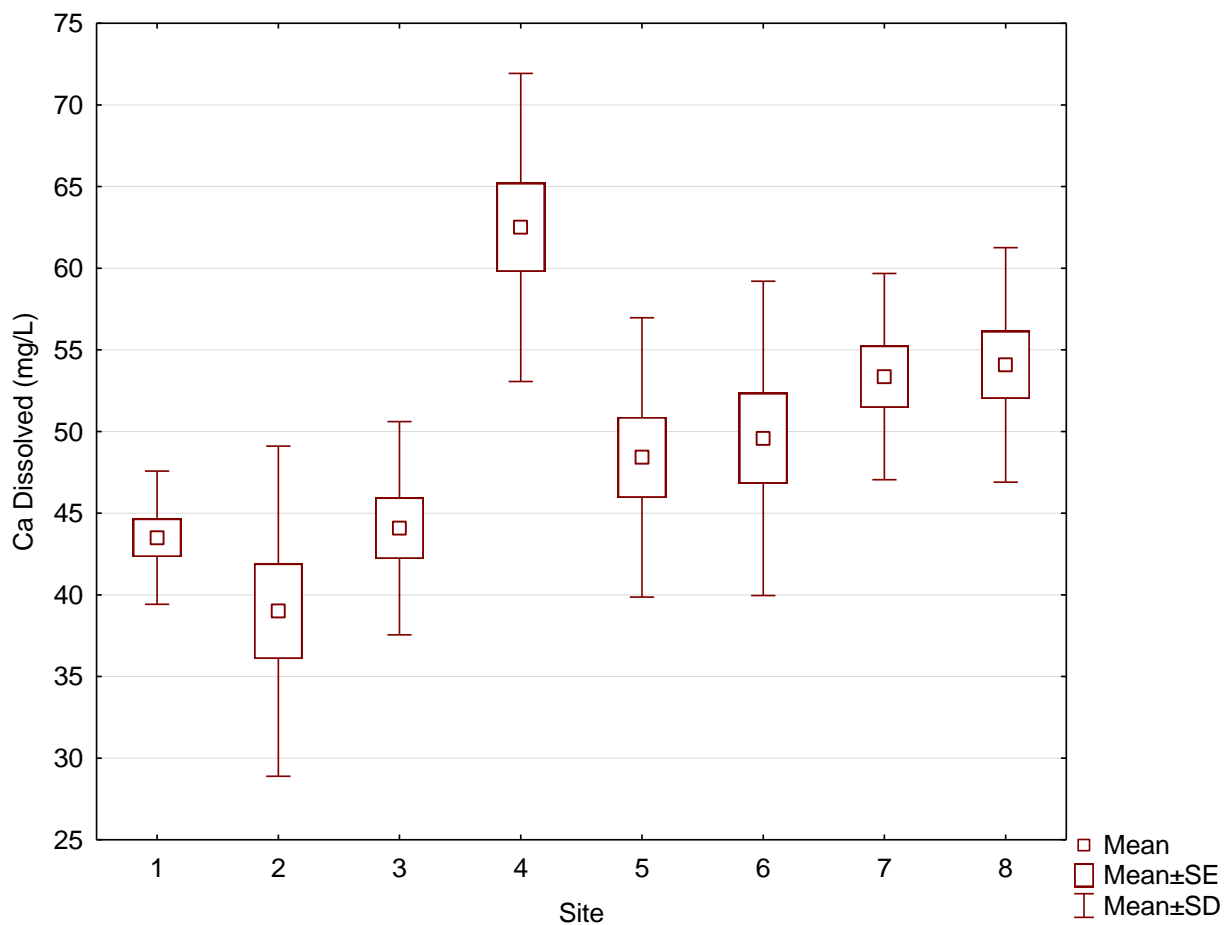
Sample sites and their abbreviations are as follows:

1. Bovenste Oog (BVO)
2. Klerkskraal Dam (KKD),
3. Upstream of Wonderfonteinspruit (BWS),
4. Downstream of Wonderfonteinspruit (OWS),
5. Boskop Dam (BKD),
6. Downstream of Potchefstroom Dam (PDM),
7. Trompie Kitsgras (RWP)
8. Elbrixon Bridge (EBR)

### **3.2.1 Dissolved calcium and magnesium**

Soluble and insoluble materials can be found in natural waters and can be organic or inorganic of origin (Mestre *et al.*, 2015). The above mentioned materials contribute to the hardness of water

and the main ions are calcium ( $\text{Ca}^{2+}$ ) and magnesium ( $\text{Mg}^{2+}$ ), concentrations of which are naturally dependent on the geological characteristics of a region (Troise *et al.*, 1990). The dolomitic characteristics of the Mooi River will have a contribution to increasing salt concentrations (section 1.2.1). An increase in Ca and Mg is usually associated with an increase of pH (Maybeck, 2003). The presence of these elements in water often also correlates closely to the land use within the catchment area (Wons *et al.*, 2012). The average concentrations of dissolved Ca in mg/L at each site is illustrated in Figure 3-1, site 1 is BVO and 8 is EBR, thus the figure illustrates the changes in dissolved Ca from upstream to downstream.



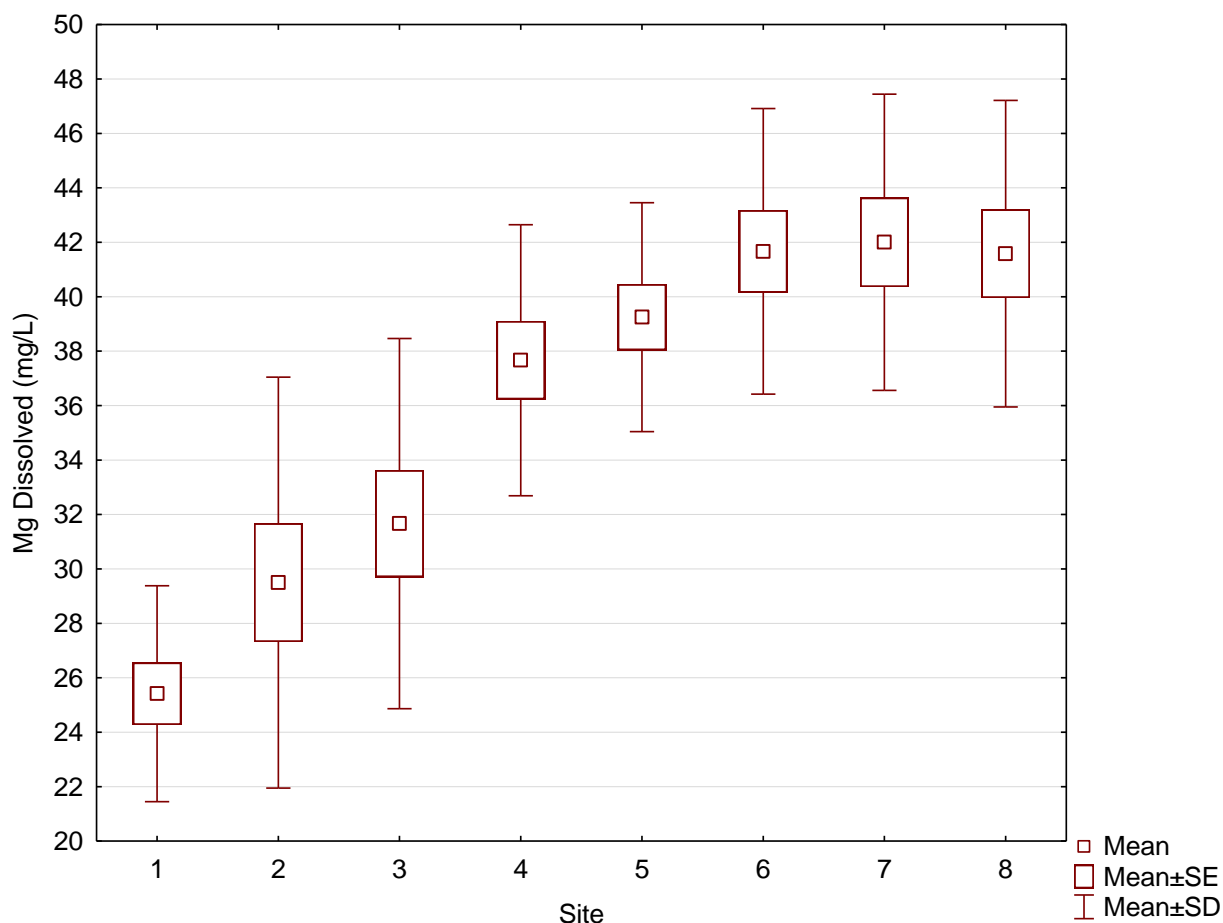
**Figure 3-1: A box-and-whisker plot illustrating the change in mean dissolved calcium concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

Figure 3-1 illustrates the difference in Ca concentrations between the different sites, when referring to Appendix B Table A-2 it is seen that OWS differs significantly ( $p < 0.05$ ) from the sites upstream of the confluence of the Wonderfontein spruit. Furthermore the only other significant differences are between KKD (Site with the lowest Ca concentrations) and RWP and EBR which are located downstream close to the confluence with the Vaal River.

The site with the lowest dissolved Ca concentrations is KKD, this may be due to the dilution of ions occurring naturally in an impoundment such as KKD with the maximum and minimum concentrations measured at KKD being 64mg/L in May 2014 and 26mg/L in November 2014 respectively (Appendix B Table A-1). This effect is again seen clearly as the Ca concentrations drop by from an average of 63mg/L to 48mg/L from OWS to BKD for the same months mentioned above, according to Potasznik and Szymczyk (2015) this can be explained as Ca bioaccumulates in the sediment at the bottom of a lake or impoundment. The Ca concentrations are the highest at OWS, with the maximum and minimum concentrations measured at OWS being 78mg/L in January and 47mg/L in May this is related to the influence of mining (see section 1.2.1) and the underlying dolomite geology of the Wonderfonteinspruit (Van Aardt and Erdman, 2004; Annandale and Nealer, 2011; Coetzee *et al.*, 2006; Ashton *et al.*, 2001). Noting that the maximum Ca concentration at KKD is in May and the minimum concentration Ca at OWS is in May gives a result opposite to the trend of the averages shown in Figure 3-1, this may be due to lower agricultural activity (crops), and thus less agricultural runoff. The next sudden rise in dissolved Ca is at RWP, this can be expected as the sampling site shows the cumulative effects Potchefstroom and its industries, the WWTP and Loopspruit have on the water quality, although the increase is evident in the figure it does not significantly differ from the concentrations found at PDM. There are however significant differences between the first 3 sites and the last 5 sites according to p-values calculated from a Kruskal-Wallis test Appendix B Table A-2.

The average dissolved Mg in mg/L is illustrated in Figure 3-2, site 1 is BVO and 8 is EBR, the figure illustrates the changes in dissolved Mg from upstream to downstream.

Dissolved Mg is essential for enzyme production and tissue contraction and is thus an essential element. Mg is also an essential component for chlorophyll production (Orzepowski and Pulikowski, 2008). Usually found in high concentrations, it is unlikely to act as a restrictive nutrient or contaminant (Dallas and Day, 2004).



**Figure 3-2: A box-and-whisker plot illustrating the change in mean dissolved magnesium concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

Figure 3-2 illustrates the gradual but constant rise of dissolved Mg as the Mooi River progresses downstream, significant differences are seen between BVO and all the sites downstream of OWS. KKD differs significantly from sites from PDM and downstream, while BWS differs significantly from RWP (Appendix B table A-3).

A constant rise in dissolved Mg concentration is observed with the lowest average concentration of dissolved Mg occurring at BVO. This may be due to the fact that BVO is not influenced by mining activities or irrigation and only cattle farming. When observing chlorophyll *a* concentrations in Appendix B Figure A-2 it is seen that there is almost no chlorophyll *a* present in the water at BVO, this may also be due to the water at BVO originating at the spring and flows downstream immediately. The maximum and minimum dissolved Mg at BVO were 30 mg/L in April and 16 mg/L in May. The high Mg concentrations can be seen throughout the month of May (Appendix B Table A-1) and all the increases are downstream from the first impoundment KKD. This may be an indication of stratification occurring and the bioaccumulated

Mg in the sediment of an impoundment and cold deeper waters mixing with the cooling surface water and releasing nutrients and ions (Coats *et al.*, 2006). The effects of this can be seen as the presence of Chlorophyll *a* (which indicates algal blooms) is at its highest throughout the river (all sites averaged per month) in July. It must be said that Mg is a contributing factor but not a direct cause for algal growth. In contrast the site with both the lowest and the highest dissolved Mg concentrations was RWP with the maximum Mg concentration of 49 mg/L in October and a minimum of 32 mg/L in September. The reason for dissolved Mg increasing from its minimum concentration to its maximum concentration in one month can be explained by the start of the rainy season, with the first rain shower falling in October 2014 leading to pollutants and salts as part of urban runoff from the Potchefstroom area to flowing into the aquatic ecosystem. Another explanation could be that the farmers plant maize during early October and use the water from the Loopspruit for irrigation. The sudden increase in dissolved Mg at OWS, from an average of 31.667mg/L at BWS to 39.25mg/L at OWS could be as a result of mining activities and the dolomite geology of the Wonderfonteinspruit.

Dissolved Mg concentrations also rise steadily from upstream to downstream and when turbidity increases are compared to this trend a similar pattern is noted. The increasing Mg concentrations is due to the dolomite geology (Williams *et al.*, 2007) of the Mooi River, as the river flows downstream, Mg accumulates and thus increased concentrations. Turbidity also increases with overland return flow from agricultural lands and pastures, this return flow may also deposit Mg in the river system.

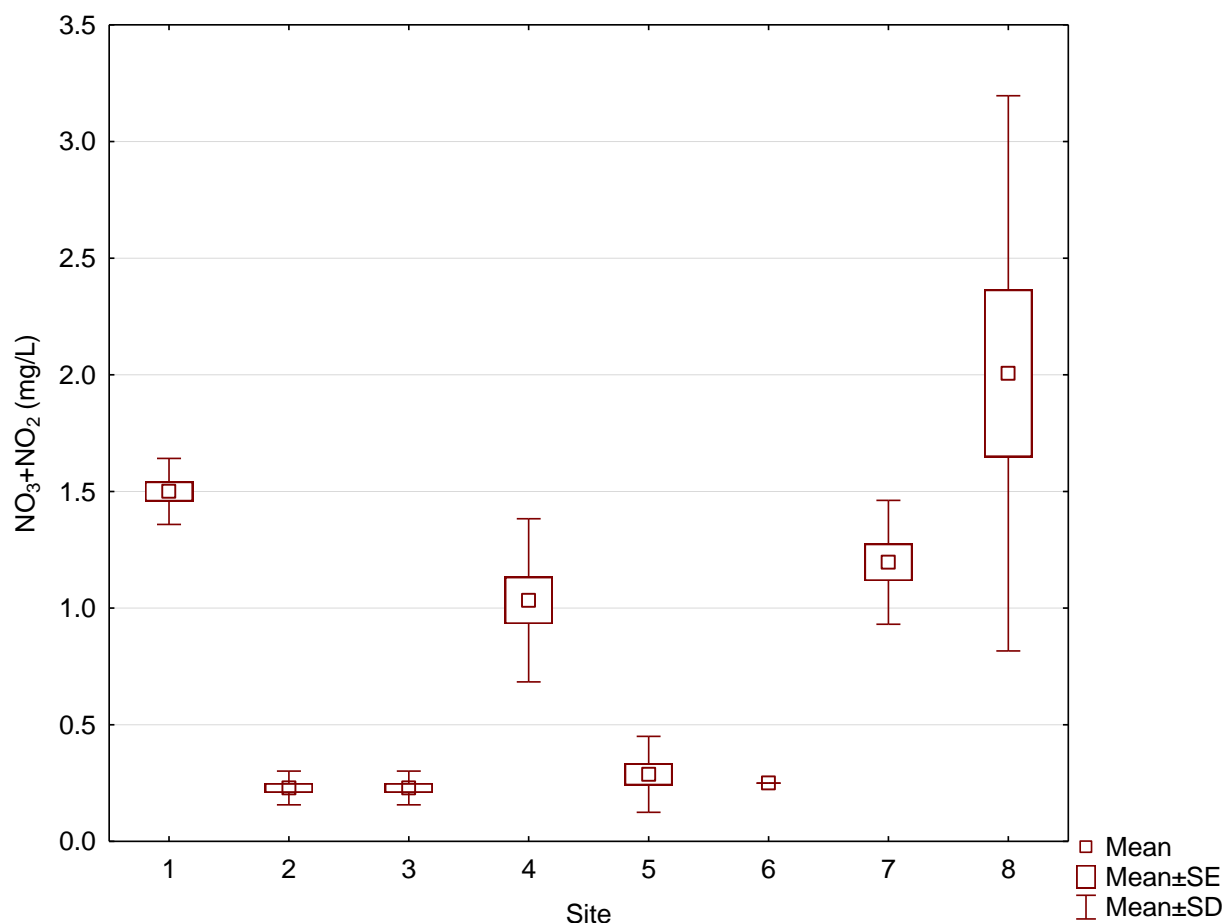
To summarise, dissolved Ca and Mg concentrations in the water increases from BVO to EBR, and the major increases can be seen at two sites OWS influenced by mining pollution from Wonderfonteinspruit, and at RWP where the effects of urban runoff and the WWTP can be observed. Dissolved Ca and Mg both correlate well with the increase in pH, which will be discussed in section 3.2.4.

### **3.2.2 Nutrients**

One of the largest problems that aquatic ecosystems face is eutrophication through the increase and availability of nutrients. One of the best examples of eutrophication in South Africa is Hartebeespoort Dam, which was once described as an oligotrophic and oxygen rich water body (Hutchinson *et al.*, 1932; Chloko, 1958). A considerable amount of sewage and industrial effluents from the catchment flowed into the impoundment for many years and introduced a high concentration of nutrients, and thus the dam became (hyper) eutrophic with highly visible, toxic

algal blooms being one of the end results (Steyn *et al.*, 1975; Paerl *et al.*, 2001). The root cause of such phenomena can be ascribed to compounds collectively referred to as nutrients, the most important of which are phosphate and nitrate (Bouamra *et al.*, 2012).

The average  $\text{NO}_3$  and  $\text{NO}_2$  (will be referred to as nitrate) concentration in mg/L at the different sites are illustrated in Figure 3-3. Site 1 is BVO and 8 is EBR, thus the figure illustrates the changes in  $\text{NO}_3$  and  $\text{NO}_2$  from upstream to downstream.



**Figure 3-3: A box-and-whisker plot illustrating the change in mean Nitrate ( $\text{NO}_3$  and  $\text{NO}_2$ ) concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

It can be noted in Figure 3-3 that BVO differs significantly from all the sites except OWS, RWP and EBR. Seen in Appendix B Table A-4 these are the three sites that had significant differences from one another, in terms of nitrate ( $p < 0.05$ ). The guidelines set out by DWAF in 1996 stated that nitrate concentrations between 2.5-10mg/L are considered as eutrophic conditions.

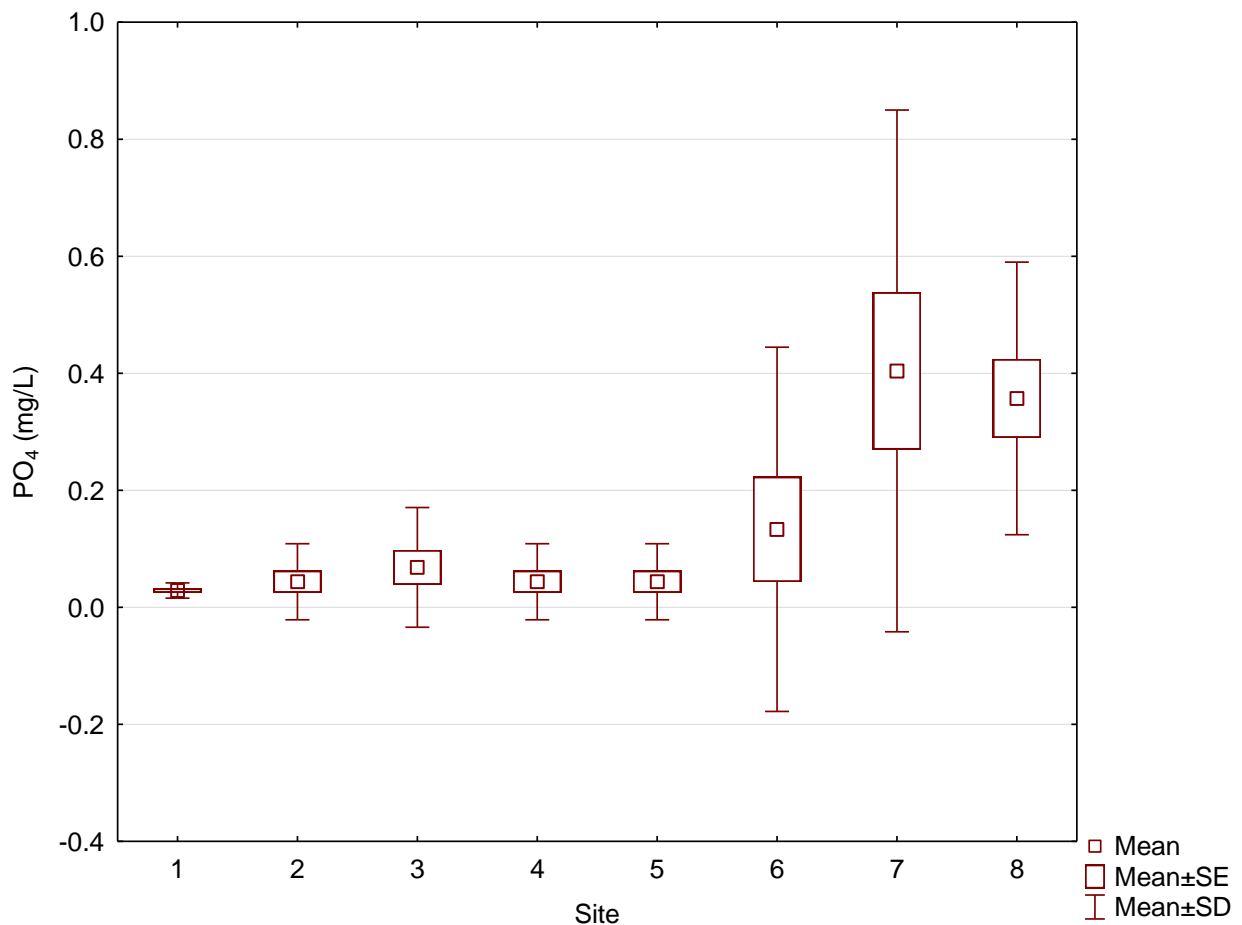
Nitrate concentrations were high at BVO with the maximum concentration and minimum concentration recorded as 1.7mg/L in July and 1.2mg/L in February respectively. According to

Dallas and Day (2004), nitrite and nitrate enter water systems via agricultural runoff, fertilizers, aquaculture and sewage spills, BVO is impacted by agriculture as cattle graze nearby and use the dolomitic eye as a drinking water source and the faeces of the cattle contaminate the water. The farmer destroys the aquatic vegetation and removes it from the water and places the dead plant material on the bank where it rots, and thus provides an additional source of organic material – nitrate included (Ladd *et al.*, 1981). The available nitrite is oxidized to nitrate by nitrifying bacteria in oxygenated systems (DWAF, 1996), and this could in part explain the high concentrations of nitrate present at BVO. A higher nitrate concentration is again seen at OWS and this is possibly associated with mining and agricultural pollution, however the nitrate concentrations at OWS are constant and the difference between the maximum and minimum values is 0.5 mg/L indicating a steady input into the system. What is surprising are the low nitrate values at PDM. It was expected to show the cumulative effect of the agricultural pollution occurring between BKD and PDM, a significant number of irrigated lands are situated along the Mooi River between these two sites. Nitrate concentrations were elevated and variable at RWB and this is expected due to the WWTP, urban runoff, industrial effluents and agricultural pollution from the Loopspruit. After RWP the last site EBR had the highest nitrate concentrations with an average of 2.1 mg/L with the maximum concentration of 3.9 mg/L recorded in November and the minimum concentration of 0.5 mg/L recorded in October. As mentioned in the discussion of dissolved Mg, maize is planted in October and is under irrigation during November and December, which explains the high nitrate concentrations in the water as coupled with the onset of the summer rains. Fertilizer, pesticides and herbicides used after planting form part of the runoff. This is contributed to by the piggery occurring on the river bank upstream from EBR. The turbidity at EBR is very high and the statement could be made that, algae do not have sufficient light to absorb and use the available nitrate. The main concern is that nitrate in the Mooi River at EBR was elevated, reaching 3.9 mg/L, which is similar to concentrations reported in the Hartbeespoort Dam (4.1 mg/L on 17/04/1972). This will not directly affect the Mooi River, but just downstream the Mooi River flows into the Vaal River, which is one of the most economically valuable rivers in South Africa.

The average PO<sub>4</sub> concentration in mg/L is illustrated in Figure 3-4, site 1 is BVO and 8 is EBR, illustrating the changes in PO<sub>4</sub> from upstream to downstream.

It should be noted that phosphorus, along with nitrate, is a limiting growth factor in photosynthetic living organisms and the optimum concentration for cell growth is described as

the Redfield ratio (N:P). The optimum ratio is 7:1, meaning for every 1 mg available phosphorus in the environment, 7mg available nitrate is needed for optimum cell growth (Kelly, 2001).



**Figure 3-4: A box-and-whisker plot illustrating the change in mean phosphate concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

From Figure 3-4 it can be observed that the phosphate concentrations are initially relatively low, with a rise in phosphate concentrations seen at PDM. RWP differs significantly from all the sites except with EBR, and EBR differs significantly with all the sites except from RWP (Appendix B table A-5).

Low phosphate concentrations in a water quality sample do not necessarily mean that the phosphate concentrations are limiting; the value simply reflects available phosphate concentrations in the water, and not the phosphate that has already been used up during cell metabolism (Mantyla *et al.*, 2008; Psarra *et al.*, 2005; Hartshorn *et al.*, 2016). Phosphate concentrations can be related to the chlorophyll *a* (Appendix B Figure A-2) present in the water

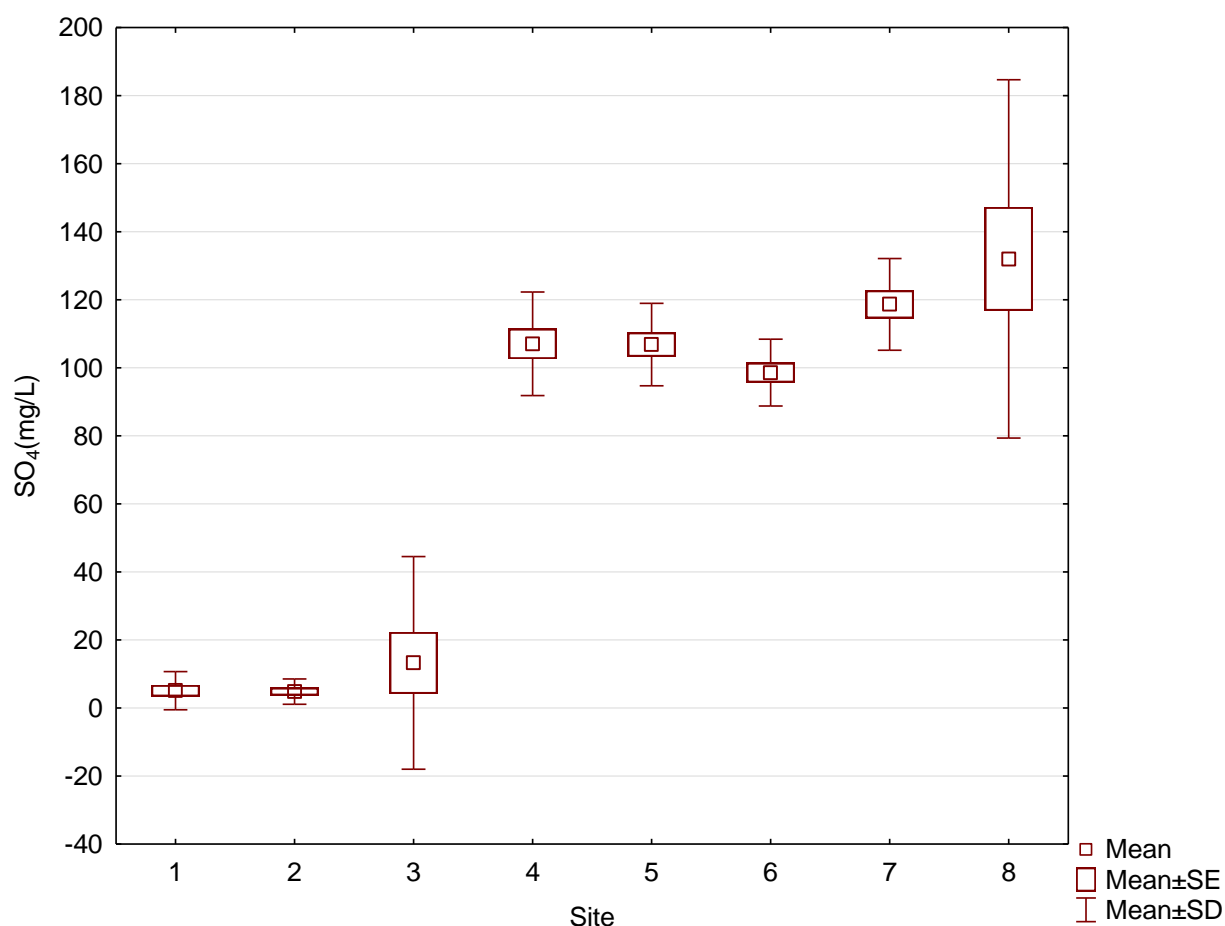
as well as the turbidity of the water that may influence light penetration, which plants need to photosynthesise and use the nutrients. At BVO the phosphate concentrations were less than 0.3 mg/L throughout the year, the chlorophyll *a* was also very low, and turbidity was low, so it can be assumed that the true phosphate concentrations were also very low, and this explains the higher available nitrate concentrations at BVO if we look at the Redfield ratio as nitrate cannot be effectively used in the relative absence of phosphate. Low phosphate concentrations at KKD and low turbidity but with a higher chlorophyll *a* concentration (average of 27.7 µg/L) make it clear that the phosphate were used by the cells of the algae. The next three sites OWS, BWS and BKD have low phosphate concentrations, and only BKD has slightly elevated chlorophyll *a* concentrations. This is explained by Kelly (2001) where he states that reservoirs/dams are recognised as traps for nutrients, and he specifically mentions phosphate. Reservoirs/dams reduce the delivery of these nutrients, thus very low phosphate concentrations. High turbidity levels are observed from PDM to EBR coupled with low chlorophyll *a* concentrations and may explain lower phosphate concentrations.

RWP differs significantly from all the sites except with EBR. A rise in phosphate concentrations is seen at PDM coupled with heavy agricultural activity between BKD and PDM this can be expected. The highest phosphate concentrations are seen at RWP, due to the effects of Potchefstroom and activities in and around the town, agricultural runoff from Loopspruit and effluents from the WWTP. The average phosphate concentration at RWP is 0.4 mg/L with the maximum value of 1.7 mg/L in December and the minimum value of 0.2 mg/L in September. The low concentrations in September can be due to the lack in rain and thus little to no urban runoff, and the high concentrations in December can be attributed to agricultural activities and heavy rainfall.

### 3.2.3 Sulphate

The average  $\text{SO}_4$  in mg/L is illustrated in Figure 3-5, site 1 is BVO and 8 is EBR, illustrating the changes in  $\text{SO}_4$  from upstream to downstream.

Sulphur occurs naturally in water as sulphate ( $\text{SO}_4^{2-}$ ), and is reduced to sulfhydryl (-SH) groups that are used in protein synthesis. Although  $\text{SO}_4^{2-}$  is not considered toxic at high concentrations for aquatic biota, but may lead to the formation of sulphuric acid, that could lower pH levels (Dallas and Day, 2004). Sulphates have different effects on trace metals and, for example, reduce the toxicity of copper (DWAF, 1996).



**Figure 3-5: A box-and-whisker plot illustrating the change in mean sulphate concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

From Figure 3-5 it can be observed that sulphate concentrations rise dramatically at OWS and concentrations stay elevated downstream of OWS. Significant differences are seen between BVO and OWS and downstream of OWS, where KKD and BWS show significant differences from OWS downstream except for the site PDM, which only differs significantly from BVO (Appendix B Table A-6).

The elevated sulphate concentrations from OWS may be explained by the influence of the Wonderfonteinspruit and the salts generated and released in effluent by gold mining activities. The sulphate concentrations rise from an average of 13.5mg/L at BWS to 107.1mg/L at OWS, this illustrates that Wonderfonteinspruit has a large contribution to sulphate concentrations in the Mooi River. Sulphate concentrations do not decrease after OWS and it may be assumed that the agricultural impacts ensure that sulphate concentrations remain elevated. At RWP the sulphate concentrations rise again and this is due to the effects of various runoffs from Potchefstroom, the WWTP and the effects of the agricultural and mining activities in the Loopspruit (section 1.2.1).

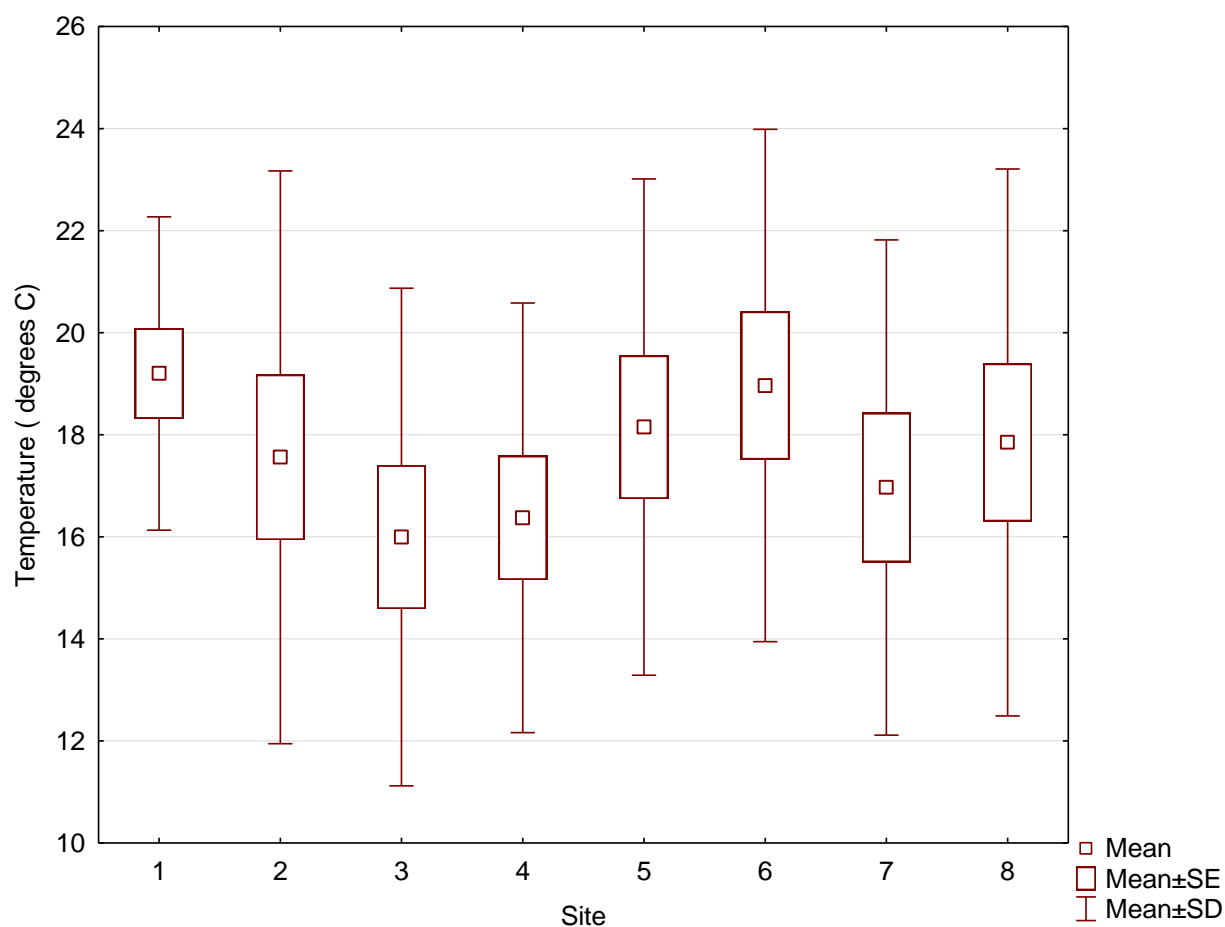
The highest sulphate concentrations of all the sites can be observed at EBR, with the difference between the maximum and minimum values are 288mg/L in February and 93mg/L in March. It can be assumed the differences in sulphate are influenced mostly by mining activities as indicated by Figure 3-5 as well as agricultural activities, as the site with the highest sulphate values is at EBR and the main source of pollution is agricultural in nature.

To summarise, it can clearly be observed from the data presented that nutrient concentrations increase significantly at OWS, that the site is heavily impacted by mining related pollution and that RWP is impacted by industrial, urban and agricultural pollution, therefore it may be concluded that activities near to or upstream of site RWP have the greatest influence on nutrient concentration in the Mooi River catchment area.

### 3.2.4 Temperature

Water temperature is one of the most important water quality variables as it affects the solubility of nutrients, toxins and oxygen (section 1.3.2). The effects on aquatic biota are also significant and the alteration of metabolic pathways and oxygen consumption may lead to drastic changes in community structure and the chemical and physical properties of a water body.

The variation in temperature will be discussed referring to Figure 3-6, and the possible effects that the temperature has on the water quality variables that were analysed in this study. The environmental impacts that may cause a rise in temperature will also be discussed.



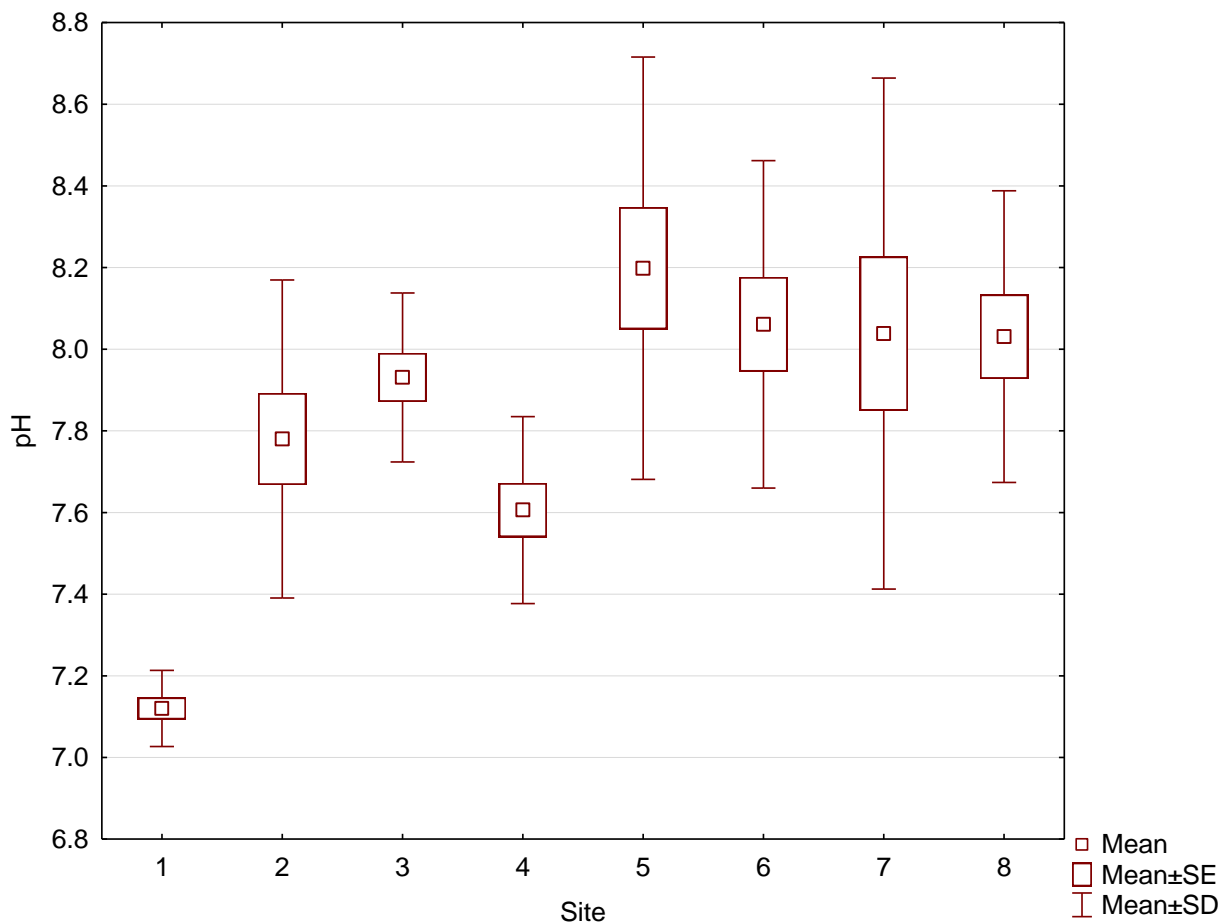
**Figure 3-6 A box-and-whisker plot illustrating the change in mean temperature in °C between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

Figure 3-6 illustrates the average temperatures per site for 2014. There are however, no significant differences between any of the sites as can be seen in Appendix B Table A-8.

It is interesting to note the high average temperature at BVO of 19.2 °C, the minimum temperature at this site was 9.6°C recorded in May. The constant high temperature of BVO is attributed to the fact that BVO is a natural spring, and there is constant upwelling of relatively warm water. The constant temperature makes BVO the ideal location for aquatic life as the oxygen consumption and metabolic reactions remain constant. It was also observed that there is an increase in temperature between BWS and OWS, here it is again clearly seen how the Wonderfonteinspruit influences the characteristics of the Mooi River. The elevation in temperature could be a result of warmer mine effluents being deposited into Wonderfonteinspruit and the warmer water mixing with the cooler Mooi River water causing a slight elevation in water temperature. The Wonderfonteinspruit also has a lower volume than the Mooi River and thus its ambient temperature will also be higher. The elevation in temperature at PDM may be a result of warmer surface water released from Potchefstroom Dam from the sluices (top release) in turn increasing stream temperatures for several kilometers downstream.

### 3.2.5 pH

pH is defined as the negative  $\log_{10}$  value of the hydrogen ion concentration activity in a water body (Dodds, 2002). Along with temperature it is one of the most important water quality variables and as mentioned in section 1.3.1, the stoichiometry, kinetics and equilibrium of chemical reactions in an aquatic ecosystem are dependent on pH (Yang *et al.*, 2014; Flores-Alsina *et al.*, 2015). The calculation of pH in a water body is based on the basic and acidic compounds dissolved in a water body and its temperature (Kim and Jeong, 2016). Changes in pH, whether the water body is either acidic or alkaline, affects the bioavailability of compounds such as cyanide, nutrients, trace metals and biocides (Dallas and Day, 2004).



**Figure 3-7: A box-and-whisker plot illustrating the change in mean pH between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

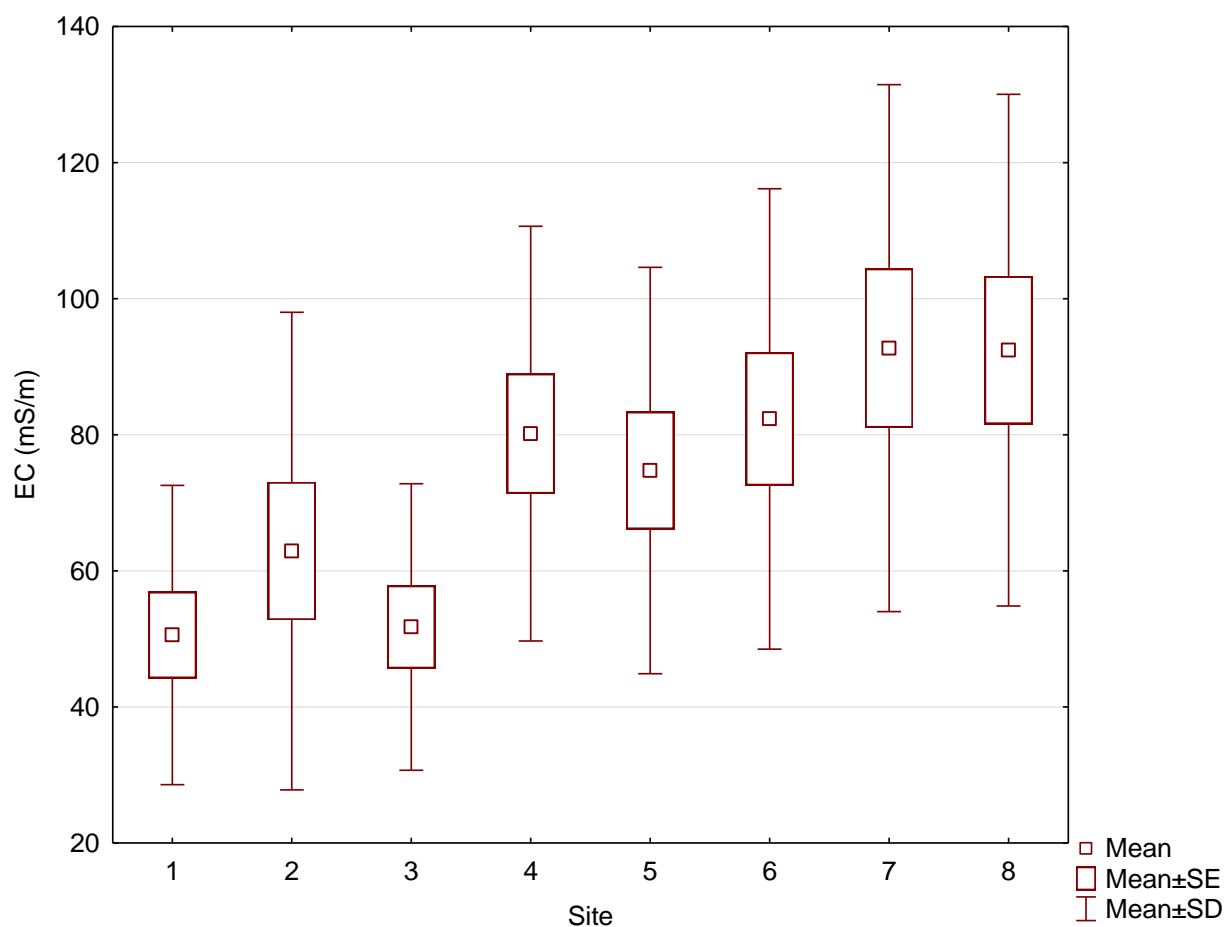
As seen in Figure 3-7, the Mooi River is an alkaline water system. There are only significant differences in pH between BVO and the rest of the sites excluding OWS (Appendix B Table A-9).

The lowest pH values are observed at BVO with an average of 7.12 with the maximum pH readings values of 7.2 in May, June, July, August and November and the minimum of 6.95 in January. BVO has a circumneutral (close to pH 7) pH and without fluctuations which is ideal for the growth and reproduction of aquatic organisms when combined with consistent temperatures (Figure 3-6). A drop in pH can be seen at OWS, and could be a result of AMD from Wonderfontein spruit (Pulles *et al.*, 1996; Dallas and Day, 2004). The pH values exceed 8 from BKD downstream to EBR and this is of concern as for example the balance between  $\text{NH}_4^+$  and  $\text{NH}_3$  may shift towards  $\text{NH}_3$  at pH levels above 8, and this can be a very toxic substance (Dallas and Day, 2004). pH values were especially high in June with values of 8.12, 9.7, 9.24 and 9.73 at

OWS, BKD, PDM and RWP respectively, pH levels this high could have a negative effect on the community structure of organisms, and this will be discussed in the following chapters.

### 3.2.6 Electrical conductivity

The amount of material that is dissolved within a water body in general is seen as a major descriptor of water quality (Dallas and Day, 2004). There are three forms in which dissolved material can be observed in water, Total Dissolved Solids (TDS) which can be found in Appendix B Figure A-3, salinity and Electrical Conductivity (EC) (Dodds, 2002) that will be discussed using Figure 3-8 with reference to the above mentioned variables.



**Figure 3-8: A box-and-whisker plot illustrating the change in mean Electrical Conductivity (EC) in mS/m between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

Figure 3-8 clearly illustrates an increase in average EC from BVO downstream to EBR. Significant differences in EC are observed between BVO, OWS, RWP and EBR. KKD and BWS only shows significant differences between KKD and RWP, EBR, and between BWS and RWP,

EBR (Appendix B Table A-10). Thus, it can be inferred that the EC at OWS, RWP, and EBR have an influence on the Mooi River. For this study EC was measured in mS/m. The results obtained from the EC measurements correlates well with the measured salts (dissolved Ca and dissolved Mg) discussed in Chapter 3.3.1 and with sulphates discussed in Chapter 3.3.2 and illustrated in Figure 3-5. The EC correlates almost perfectly with TDS (as would be expected) as seen in Appendix B Figure A-3. In 2004 data EC collected by Taylor in the Vaal River correlated 100% with TDS values. With TDS and EC correlating along with the dissolved salts it is safe to make the deduction that with the increase in EC, salinity (although not measured) will also increase, as EC, TDS and salinity usually correlate (Dodds, 2002). Thus from this point only EC will be discussed.

There is a clear increase in EC between BVO and KKD, and a decrease to BWS, there are however no significant differences between these sites. A high EC can be seen at OWS, and this can be related to the mining activities in the Wonderfontein spruit, mining effluents have significant effects on EC, TDS and salinity (Dallas and Day, 2004; Pulles *et al.*, 1996). The average EC at OWS is 80.2 mS/m and is an increase from BWS which has an average EC of 51.75. The maximum EC at OWS is 175 mS/m in March (Appendix B Table A-1) and this could be a consequence of late rainfall, rising water levels, and thus comprising the dried salt and nutrient-rich crust on the previously dried river bank. The EC of all the sites in the Mooi River was highly elevated in March 2014, this could be because of very low rainfall in April 2014 (25mm-50mm) and then heavy rainfall in March 2014 (100mm-200mm) (South African Weather Service, 2015b) causing run off from various sources.

The highest EC values observed were at RWP. The average EC at RWP is 92.73 mS/m with the maximum and minimum value occurring at 209 mS/m in March 2014 and 72 mS/m in December. Elevated EC can be attributed to urban runoff, industrial pollution and the effluents of the WWTP and agricultural pollution associated with the Loopspruit, this agrees with the literature regarding flows and contaminants expected from urban areas (Epstein, 2002; Walker and Pan, 2006; Williams, 2001; Williams, 1987).

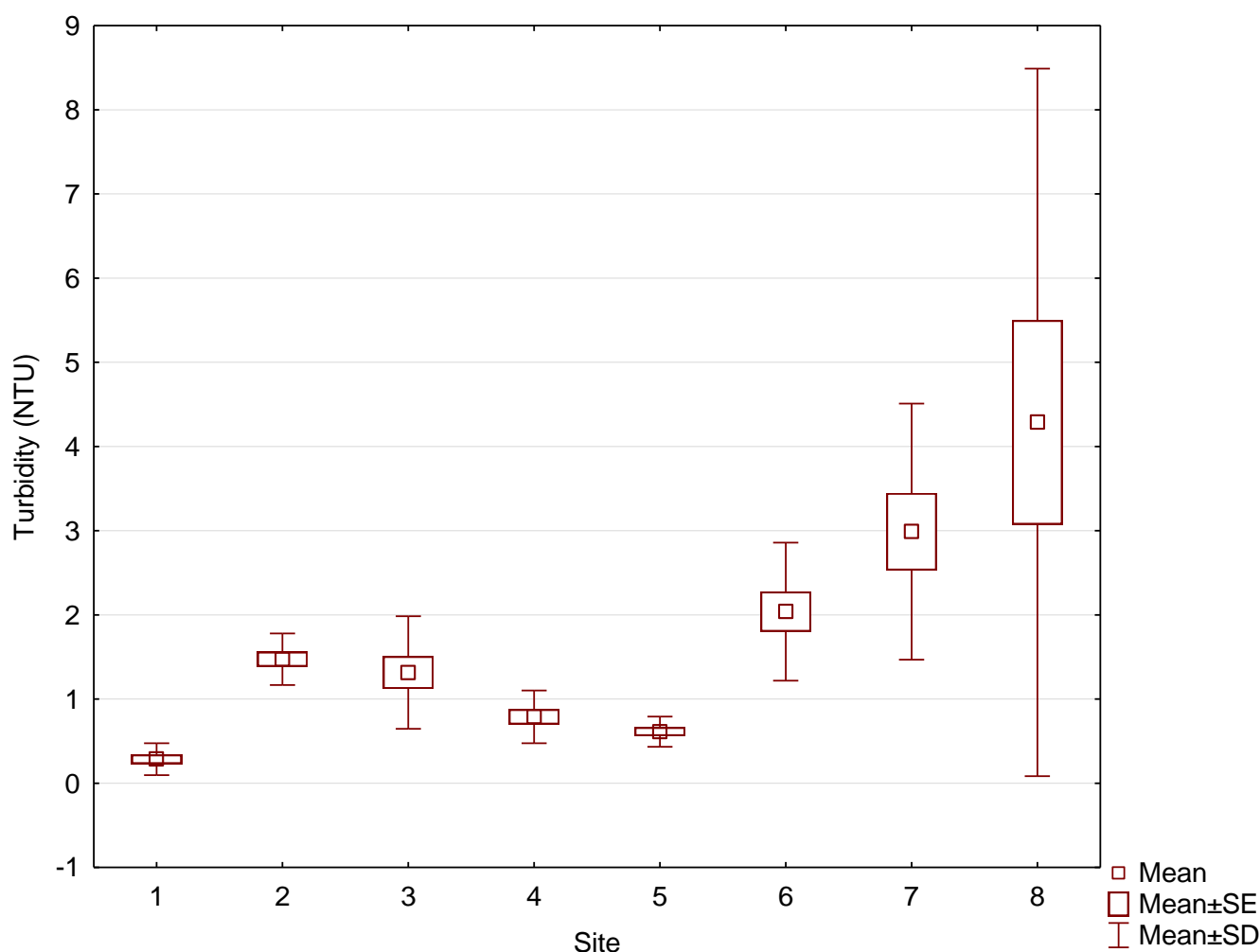
The second highest EC values are at EBR and indicates that the agricultural activities allowed the continued high EC values, the EC values at this site are important as they are potentially problematic in terms of impacting the water quality and biological integrity of the Vaal River.

To summarise, the effects of the different pollution impacts can clearly be seen in the results, RWP being the site that shows the highest contributor to pollution considering EC.

### 3.2.7 Turbidity

Turbidity (NTU) gives a good indication of the degree of light refraction in water. Turbidity affects primary production as autotrophic organisms at this trophic level use light for photosynthesis (Dallas and Day, 2004). As observed in the previous discussion of nutrients, measurable phosphate has a correlation to chlorophyll *a* concentrations, and chlorophyll *a* is dependent on light penetration and thus turbidity.

The average NTU is illustrated in Figure 3-9, site 1 is BVO and 8 is EBR, thus it illustrates the changes in NTU from upstream to downstream. The NTU values were divided by 10, so 1 unit in Figure 3-9 is 10 NTU.



**Figure 3-9: A box-and-whisker plot illustrating the change in mean Turbidity in NTU between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

In Figure 3-9 it can be observed that turbidity rises from BVO to KKD and BWS which shows a significant difference in turbidity. BVO also differs significantly from PDM, RWP and EBR. The other significant differences are observed between OWS and RWP and EBR. The other significant differences are observed at BKD which differs from PDM, RWP and EBR (Appendix B Table A-12).

BVO is the site with the lowest turbidity and has an average of 2.9NTU. This can be due to the low flow rate at BVO and due to the fact that the sediment is mostly sand particles with minimal mud and clay present minimal inflow which could carry such particles. An increase in turbidity is seen at KKD, with the average being 14.7 NTU this may this can be due to low flow rate at BVO and due to the sandy sediment. be due to recreational activities in the dam, and the fact that the sediment is disturbed by the channels leading water from KKD to BKD. Turbidity values are higher at PDM, and can be explained by the high flow rate of the water and the clay/mud composition of the sediment.

Very high turbidity was observed at RWP with an average of 29.8NTU, at this site it can be observed that the turbidity levels have an influence on primary production as inferred by the low chlorophyll *a* concentrations (Appendix B Figure A-2) and high available phosphates (Figure 3-4). The high turbidity results from pollution and urban runoff from the Potchefstroom area, the high flow rate of the water and the muddy composition of the sediment will also be a contributing factor. The site with the highest turbidity is EBR, possibly due to the high agricultural activity in the area, the trucks disturbing the soil and marginal vegetation to pump water out of the river during water abstraction and the sediment which consists of a mixture of mud and clay. The effects of high turbidity with an average of 42.9NTU can be seen in the low chlorophyll *a* concentrations and high phosphates (Figure 3-4).

To summarise, turbidity is a very useful supplementary variable as it can easily alter the physical and chemical attributes of water and impact upon the biota.

### **3.2.8 Spatial and Temporal variation in Water quality in the Mooi River**

Canonical correspondence analysis (CCA) biplots are presented in the following sections to illustrate changes in water quality variables in relation to space and time.

### 3.2.8.1 Spatial variation of water quality variables

Figure 3-10, a CCA biplot, illustrates the measured water quality variables used, and how the various variables relate to the study sites. The discussion of Figure 3-10 will be per site.

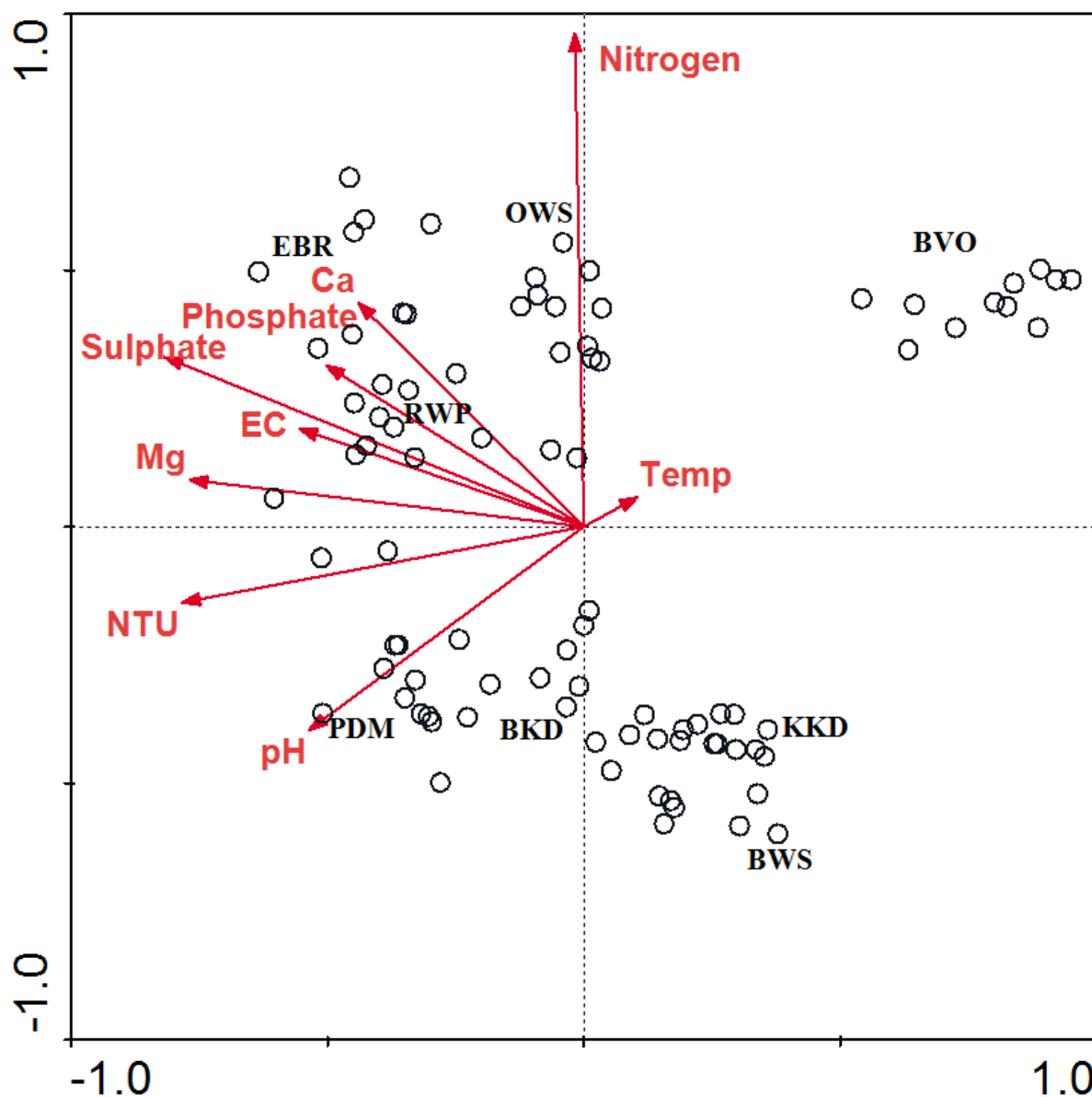


Figure 3-10: A canonical correspondence analysis scatter biplot, showing the spatial variation between measured water quality variables and sites (January 2014 – December 2014).

**Table 3-1: Summary of canonical correspondence analysis (CCA) for both physical and chemical parameters.**

Axes	1	2	3	4	Total
Eigenvalues	0.688	0.509	0.372	0.185	4.915
Species-environment correlations	0.960	0.920	0.887	0.639	
Cumulative percentage variance of species data	14.0	24.4	31.9	35.7	
Cumulative percentage variance of	33.2	57.8	75.7	84.6	
Sum of all eigenvalues					4.915
Sum of all canonical eigenvalues					2.071

From Figure 3-10 it can be observed that the first site BVO, associates with higher temperatures, this was discussed in section 3.2.3 as there little was fluctuation in this variable and an average temperature of 19.2 °C throughout 2014. The temperature at the other sites was lower during the cold winter months. The constant temperature is attributed due to the fact that BVO is a natural spring and the temperature of the upwelling water remains rather constant independent of season. BVO is also associated with high nitrate concentrations, at an average of 1.5 mg/L the second highest nitrate concentration of all the sites. The high nitrate concentrations could be a result of cattle using BVO as a water source, as well as the dead plant material and macrophytes the owner of the land removes and discards next to the water.

The next site were KKD and BWS, the two sites are discussed together because of their close grouping in Figure 3-10. The two sites are not associated with elevated levels of water quality variables, the two sites are seldom exposed to any high levels of variables but are associated with higher pH values. BWS is the site that is the least influenced by any of the water quality variables, and based on Figure 3-10 BWS could be considered as the least impacted or polluted site.

Downstream of BWS is OWS - the site that was of most concern in the beginning of the study in terms of potential impact. OWS is the site located downstream of the confluence of Wonderfonteinspruit and the Mooi River. The Wonderfonteinspruit is heavily impacted by mining activities in the far West-rand and is partly underlain by dolomite. OWS directly corresponds with high nitrate, phosphate and sulphate concentrations as well as dissolved calcium and magnesium, elevated temperatures, and elevated EC. This confirms that the Wonderfonteinspruit has a large influence on the water quality of the Mooi River. The elevated

levels of the various measured variables observed at OWS will have drastic effects on biological integrity of the aquatic ecosystem and community structure of the biota in the Mooi River.

BKD is the next site after OWS and it was observed that this site had elevated pH, dissolved magnesium and sulphate. From the CCA it can be seen that the BKD associate negatively with nitrogen and OWS the impoundment acts to dilute the elevated water quality variables that was measured OWS. BKD is still relatively impacted by pollution when comparing BKD to BVO, KKD and BWS.

PDM is located downstream of Potchefstroom Dam and is impacted by agricultural activity along the Mooi River between BKD and PDM. PDM correlates with elevated pH levels, high turbidity, elevated dissolved magnesium and elevated phosphate and sulphate concentrations. The association with turbidity can be due to higher phosphate concentrations and the high agricultural activity upstream of PDM. Habitat integrity and community structure will potentially be affected by the elevated pollution levels when compared to the upstream sites.

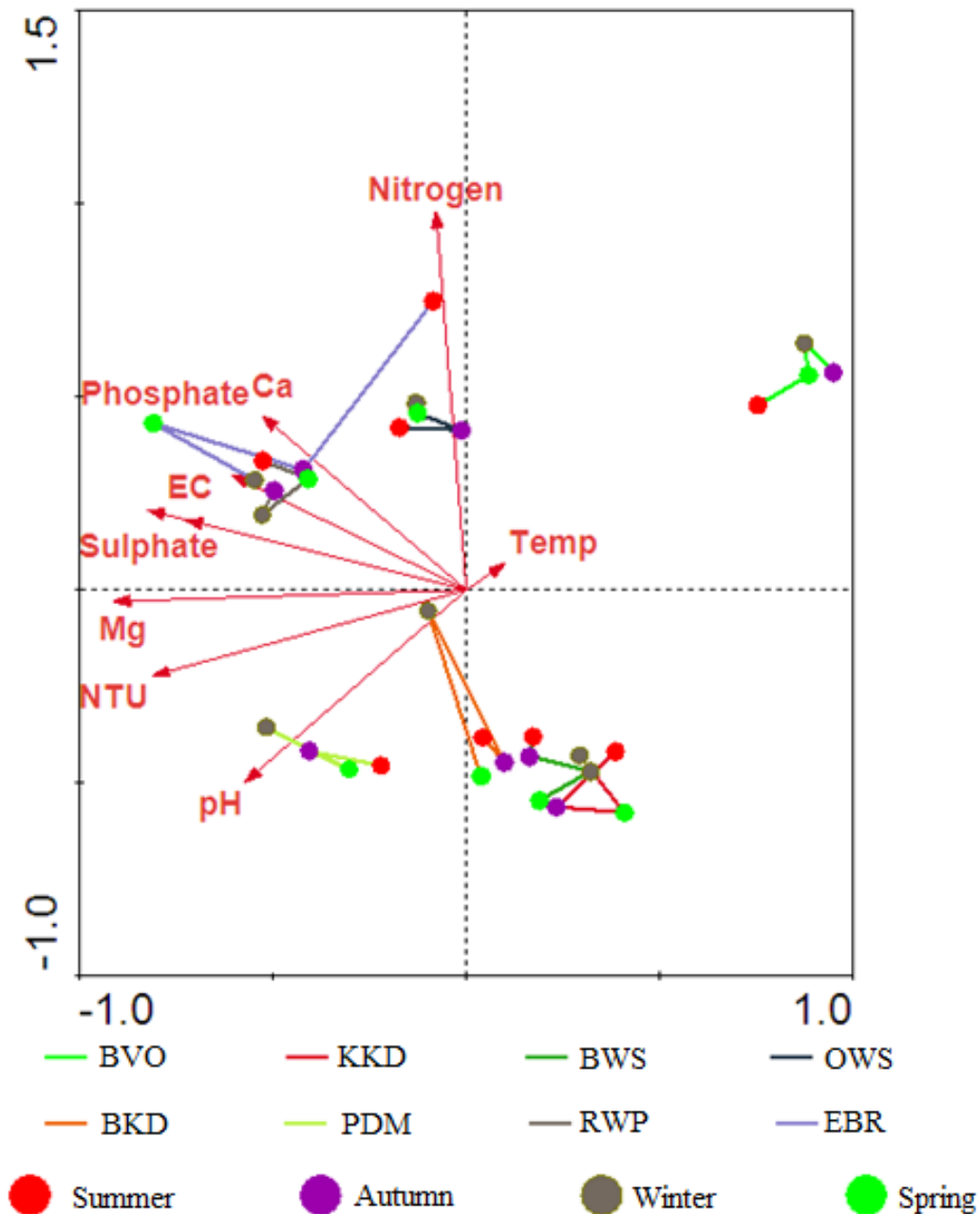
The effects that the industrial effluents, urban runoff and agricultural activities from Loopspruit and Potchefstroom's WWTP effluents have on the aquatic ecosystem and water quality as can be observed and easily recognised at site RWP. RWP is associated with elevated concentrations of nutrients, salts, turbidity and EC. Elevated levels of these variables indicate that RWP is polluted and that Potchefstroom has a significant influence on the water quality of the Mooi River. The potential for the bioavailability of toxic trace metals and other elements is exponentially increased at RWP due to higher average temperatures from Wasgoedspruit. Habitat integrity and community structure of aquatic biota will be heavily affected by the pollutants and water chemistry composition that occurs at RWP.

The last site is EBR and although there is often potential for recovery of water quality through natural assimilation this cannot be seen to be demonstrated at EBR as there is slight to no improvement in water quality when compared to RWP. EBR correlates significantly with high turbidity, phosphate and nitrate concentrations, high concentrations of dissolved salts and high EC levels. The possible reason for the ecosystem not recovering is that high agricultural activity that impacts on the Mooi River in the form of irrigation, abstraction, piggery and cattle farming leading to the destruction of aquatic and marginal vegetation allowing high levels of pollutants to enter the with diffuse overland flow river with no natural 'filters' to absorb and use the nutrients.

In conclusion the effects of different land uses produce different pollutants that enter the Mooi River. Whether it is intentional disposal of effluents, runoff caused by rain or irrigation, or livestock grazing on the land, all have noticeable effects on the water quality. The largest contributor of pollution is the town of Potchefstroom and the pollutants associated with urbanisation and light industry. The second most influential is Wonderfonteinspruit, with the effects of the mining industry. A clear, steady and significant trend of decreasing water quality from upstream to downstream is clearly apparent from the discussion above.

### **3.2.8.2 Temporal variation of water quality**

Figure 3-11 illustrates the CCA biplot containing information on the water quality variables measured, and how the various variables correlate between the study sites and the influence of seasonal change. Figure 3-11 will be discussed in terms of seasonality in relation to water quality.



**Figure 3-11: A Canonical correspondence analysis scatter biplot showing the seasonal temporal variation of the water quality variables at each site.**

In Figure 3-11 the temporal variation of water quality across the study sites is illustrated. The temporal grouping was done by averaging the water quality results seasonally, and displaying them on a CCA plot. The seasonal changes indicate that sites have slightly better water quality in the summer months.

The only sites where changes can be seen are at BKD and EBR. BKD associates with elevated levels of all the measured water quality variables during the winter months, and this can be

attributed to the lower water levels occurring in the dry season, causing a higher concentration of pollutants in BKD.

EBR shows an improvement in water quality in the summer, and correlates strongly with elevated levels of nitrate, this may be due to the urban runoff, due to rainfall, higher through flow and possible spills at the waste water treatment plant, causing higher levels of nitrate.

The other six sites show little seasonal variation.

# CHAPTER 4 DIATOMS

## 4.1 Introduction

The following chapter contains the results and discussion of the diatom data collected at every sampling opportunity. The results will be presented as species lists per site, correlation analyses between the various diatom indices and the measured water quality variables. The results are presented as an average per site throughout the 12 months of sampling. A full table with all results can be found in Appendix C and will be referred to during the discussion. Thereafter the results of temporal and spatial analyses are presented to show the differences in diatom community structure between sites as well as the effect of seasonal changes.

## 4.2 Results and discussion

The sites and their abbreviations are as follows:

1. Bovenste Oog (BVO)
2. Klerkskraal Dam (KKD)
3. Upstream of Wonderfonteinspruit (BWS)
4. Downstream of Wonderfonteinspruit (OWS)
5. Boskop Dam (BKD)
6. Downstream of Potchefstroom Dam (PDM)
7. Trompie Kitsgras (RWP)
8. Elbrixon Bridge (EBR)

### 4.2.1 Diatom species list

Table 4-1 lists the diatom species observed at each of the sites throughout 2014. Dominant species (10% and above) are marked in bold.

**Table 4-1 Diatom species encountered in the Mooi River at each site in 2014.**

Species	Abbreviation
<b>Bovenste Oog (BVO)</b>	
<i>Adlafia bryophila</i> (Petersen) Moser, Lange-Bertalot & Metzeltin	ABRY
<i>Amphora copulata</i> (Kützing) Schoeman & Archibald	ACOP
<i>Achnanthes eutrophila</i> Lange-Bertalot	AEUT
<i>Amphora inariensis</i> Krammer	AINA
<i>Amphora</i> sp. C.G. Ehrenberg	AMPH
<i>Brachysira neoexilis</i> Lange-Bertalot	BNEO
<i>Caloneis</i> sp. Cleve	CALO
<b><i>Cymbella kappii</i> (Cholnoky) Cholnoky</b>	<b>CKPP</b>
<b><i>Craticula molestiformis</i> (Hustedt) Lange-Bertalot</b>	<b>CMLF</b>
<i>Cyclotella ocellata</i> Pantocsek	COCE
<b><i>Cocconeis placentula</i> Ehrenberg</b>	<b>CPLA</b>
<i>Denticula</i> sp. F.T. Kützing	DENT
<b><i>Denticula kuetzingii</i> Grunow</b>	<b>DKUE</b>
<i>Diploneis oblongella</i> (Naegeli) Cleve-Euler	DOBL
<i>Denticula subtilis</i> Grunow	DSUB
<i>Epithemia adnata</i> (Kützing) Brébisson	EADN
<b><i>Encyonopsis subminuta</i> Krammer &amp; Reichardt</b>	<b>ESUM</b>
<i>Eunotia</i> sp. C.G. Ehrenberg	EUNO
<b><i>Fragilaria ulna</i> (Nitzschia) Lange-Bertalot</b>	<b>FULN</b>
<i>Geissleria decussis</i> (Oestrup) Lange-Bertalot & Metzeltin	GDEC
<i>Geissleria</i> sp. Lange-Bertalot & Metzeltin	GEIS
<i>Gomphonema</i> sp.1 C.G. Ehrenberg	GOMP
<i>Gomphonema parvulum</i> (Kützing) Kützing	GPAR
<i>Luticola kotschy</i> (Grunow)	LKOT
<i>Navicula cryptocephala</i> Kützing	NCRY
<i>Navicula radiosa</i> Kützing	NRAD
<b><i>Pseudostaurosira brevistriata</i> (Grunow) Williams &amp; Round</b>	<b>PBTG</b>
<i>Pinnularia platycephala</i> (Ehrenberg) Cleve	PPLA
<i>Psammothidium rossii</i> (Hustedt) Bukhtiyarova & Round	PROS
<i>Staurosira construens</i> Ehrenberg	SCON
<i>Surirella gracilis</i> Grunow	SGRA
<i>Stauroneis anceps</i> Ehrenberg	STAN
<i>Staurosira</i> sp. (C.G. Ehrenberg) D.M. Williams & F.E. Round	STRS

<b>Klerkskraal Dam (KKD)</b>	
<i>Aulacoseira ambigua</i> (Grunow) Simonsen	AAMB
<i>Achnanthes</i> sp. J.B.M. Bory de St. Vincent	ACHN
<i>Amphora copulata</i> (Kützing) Schoeman & Archibald	ACOP
<i>Amphipleura pellucida</i> Kützing	APEL
<i>Brachysira neoexilis</i> Lange-Bertalot	BNEO
<i>Cymbella cymbiformis</i> Agardh	CCYM
<i>Cymbella kolbei</i> Hustedt	CKOL
<b><i>Cymbella kappii</i> (Cholnoky) Cholnoky</b>	<b>CKPP</b>
<i>Cocconeis</i> sp. C.G Ehrenberg	COCS
<b><i>Denticula kuetzingii</i> Grunow</b>	<b>DKUE</b>
<i>Discostella stelligera</i> (Cleve & Grunow) Houk & Klee	DSTE
<i>Encyonopsis cesatii</i> (Rabenhorst) Krammer	ECES
<i>Eunotia subarcuatooides</i> Alles, Nörpel & Lange-Bertalot	ESUB
<i>Eunotia</i> sp. C.G. Ehrenberg	EUNO
<i>Fragilaria crotonensis</i> Kitton	FCRO
<i>Fallacia insociabilis</i> (Krasske) D.G. Mann	FINS
<i>Fragilaria nanana</i> Lange-Bertalot	FNAN
<b><i>Fragilaria ulna</i> (Nitzsch.) Lange-Bertalot</b>	<b>FULN</b>
<i>Gomphonema insigne</i> Gregory	GINS
<i>Gomphonema italicum</i> Kützing	GITA
<i>Gomphonema ventricosum</i> Gregory	GVEN
<i>Mastogloia elliptica</i> (C.A. Agardh) Cleve	MELL
<b><i>Navicula</i> sp. J.B.M. Bory de St. Vincent</b>	<b>NAVI</b>
<i>Nitzschia fonticola</i> Grunow	NFON
<i>Nitzschia</i> sp. A.H. Hassall	NITZ
<i>Navicula radiosa</i> Kützing	NRAD
<b><i>Navicula zanoni</i> Hustedt</b>	<b>NZAN</b>
<i>Pinnularia borealis</i> Ehrenberg	PBOR
<b><i>Pseudostaurosira brevistriata</i> (Grunow) Williams &amp; Round</b>	<b>PBTG</b>
<i>Rhopalodia gibba</i> (Ehrenberg) O.Müller	RGIB
<b><i>Staurosira construens</i> Ehrenberg</b>	<b>SCON</b>
<b><i>Staurosirella pinnata</i> (Ehrenberg) Williams &amp; Round</b>	<b>SPIN</b>
<i>Stauroneis</i> sp. C.G. Ehrenberg	STAU
<b>Upstream of Wonderfonteinspruit (BWS)</b>	
<i>Aulacoseira ambigua</i> (Grunow) Simonsen	AAMB
<b><i>Achnanthes</i> sp. J.B.M. Bory de St. Vincent</b>	<b>ACHN</b>
<i>Amphora copulata</i> (Kützing) Schoeman & Archibald	ACOP
<i>Amphora pediculus</i> (Kützing) Grunow	APED
<i>Amphipleura pellucida</i> Kützing	APEL
<i>Cymbella kappii</i> (Cholnoky) Cholnoky	CKPP

<i>Cocconeis pediculus</i> Ehrenberg	CPED
<b><i>Cocconeis placentula</i> Ehrenberg</b>	<b>CPLA</b>
<i>Cymbella similis</i> Krasske	CSIM
<i>Cymbella tumida</i> (Brébisson) Van Heurck	CTUM
<i>Discostella stelligera</i> (Cleve & Grunow) Houk & Klee	DSTE
<i>Encyonopsis leei</i> Krammer	ENLE
<i>Eunotia subarcuatooides</i> Alles, Nörpel & Lange-Bertalot	ESUB
<i>Fragilaria ulna</i> (Nitzsch) Lange-Bertalot	FULN
<i>Gomphonema acuminatum</i> Ehrenberg	GACU
<b><i>Gomphonema minutum</i> (Agardh) Agardh</b>	<b>GMIN</b>
<i>Gomphonema</i> sp. C.G. Ehrenberg	GOMP
<i>Gomphonema parvulum</i> (Kützing) Kützing	GPAR
<i>Gyrosigma scalproides</i> (Rabenhorst) Cleve	GSCA
<i>Hantzschia amphioxys</i> (Ehrenberg) Grunow	HAMP
<b><i>Navicula</i> sp. J.B.M. Bory de St. Vincent</b>	<b>NAVI</b>
<i>Navicula capitata</i> Ehrenberg (Hippodonta)	NCAP
<i>Nitzschia dissipata</i> (Kützing) Grunow	NDIS
<b><i>Navicula tridentula</i> Krasske</b>	<b>NTRI</b>
<i>Pseudostaurosira brevistriata</i> (Grunow) Williams & Round	PBTG
<b><i>Rhopalodia gibba</i> (Ehrenberg) O.Müller</b>	<b>RGIB</b>
<i>Rhopalodia</i> sp. O Müller	RHOP
<b><i>Staurosira construens</i> Ehrenberg</b>	<b>SCON</b>
<b><i>Staurosirella pinnata</i> (Ehrenberg) Williams &amp; Round</b>	<b>SPIN</b>
<i>Stauroneis smithii</i> Grunow	SSMI
<i>Tryblionella apiculata</i> Gregory	TAPI
<b>Downstream of Wonderfonteinspruit (OWS)</b>	
<i>Amphora ovalis</i> (Kützing) Kützing	AOVA
<b><i>Craticula ambigua</i> (Ehrenberg) Mann</b>	<b>CAMB</b>
<i>Cymbella cuspidata</i> Kützing	CCUS
<b><i>Cocconeis placentula</i> Ehrenberg</b>	<b>CPLA</b>
<i>Cymbella similis</i> Krasske	CSIM
<i>Cymbella tumida</i> (Brébisson) Van Heurck	CTUM
<b><i>Diatoma vulgare</i> Bory</b>	<b>DVUL</b>
<i>Eunotia bilunaris</i> (Ehrenberg) Mills	EBIL
<i>Encyonopsis cesatii</i> (Rabenhorst) Krammer	ECES
<i>Eunotia subarcuatooides</i> Alles, Nörpel & Lange-Bertalot	ESUB
<i>Fragilaria ulna</i> (Nitzsch) Lange-Bertalot	FULN
<i>Frustulia vulgare</i> (Thwaites) De Toni	FVUL
<i>Gomphonema acuminatum</i> Ehrenberg	GACU
<i>Gomphonema affine</i> Kützing	GAFF
<i>Gomphonema italicum</i> Kützing	GITA
<b><i>Gomphonema parvulum</i> var. <i>subellipticum</i> Cleve</b>	<b>GPSE</b>

<b><i>Gomphonema pumilum</i> (Grunow) Reichardt &amp; Lange-Bertalot</b>	<b>GPUM</b>
<i>Gyrosigma rautenbachiae</i> Cholnoky	GRAU
<i>Hippodonta capitata</i> (Ehrenberg) Lange-Bert.Metzeltin & Witkowski	HCAP
<i>Nitzschia amphibia</i> Grunow	NAMP
<i>Navicula capitatoradiata</i> Germain	NCPR
<i>Navicula cryptocephala</i> Kützing	NCRY
<i>Nitzschia heufleriana</i> Grunow	NHEU
<b><i>Nitzschia</i> sp. A.H. Hassall</b>	<b>NITZ</b>
<i>Nitzschia sinuata</i> (Thwaites) Grunow	NSIN
<i>Navicula viridula</i> (Kützing) Ehrenberg	NVIR
<i>Placoneis</i> sp. C. Mereschowsky	PLAC
<i>Planothidium rostratum</i> (Oestrup) Lange-Bertalot	PRST
<b><i>Rhoicosphenia abbreviata</i> (C.Agardh) Lange-Bertalot</b>	<b>RABB</b>
<i>Rhopalodia gibba</i> (Ehrenberg) O.Müller	RGIB
<i>Stauroneis</i> sp. C.G. Ehrenberg	STAU
<i>Surirella</i> sp. P. J.F. Turpin	SURI
<i>Tabularia fasciculata</i> (Agardh)Williams & Round	TFAS
<i>Thalassiosira weissflogii</i> (Grunow) Fryxell & Hasle	TWEI
<b>Boskop Dam (BKD)</b>	
<i>Adlafia bryophila</i> (Petersen) Moser, Lange-Bertalot & Metzeltin	ABRY
<i>Amphora copulata</i> (Kützing) Schoeman & Archibald	ACOP
<i>Achnanthes exilis</i> Kützing	AEXI
<i>Asterionella gracillima</i> (Hantzsch) Heiberg	AGRA
<i>Brachysira neoexilis</i> Lange-Bertalot	BNEO
<i>Craticula ambigua</i> (Ehrenberg) Mann	CAMB
<i>Cymbopleura naviculiformis</i> (Auerswald) Krammer	CBNA
<i>Caloneis clevei</i> var. <i>attenuata</i> Manguin	CCLA
<i>Cymbella cymbiformis</i> Agardh	CCYM
<i>Cymbella kappii</i> (Cholnoky) Cholnoky	CKPP
<b><i>Cyclotella ocellata</i> Pantocsek</b>	<b>COCE</b>
<i>Cocconeis placentula</i> Ehrenberg	CPLA
<i>Denticula kuetzingii</i> Grunow	DKUE
<i>Denticula subtilis</i> Grunow	DSUB
<i>Diatoma vulgare</i> Bory	DVUL
<b><i>Encyonopsis minuta</i> Krammer &amp; Reichardt</b>	<b>ECPM</b>
<i>Encyonopsis</i> sp. Krammer	ENCP
<b># <i>Eunotia</i> sp.C.G. Ehrenberg</b>	<b>EUNO</b>
<i>Fragilaria capucina</i> Desmazieres	FCAP
<i>Fragilaria crotonensis</i> Kitton	FCRO
<b><i>Fragilaria ulna</i> (Nitzsch) Lange-Bertalot</b>	<b>FULN</b>
<b># <i>Gomphonema</i> sp.2 C.G. Ehrenberg</b>	<b>GOMP</b>

<i>Mastogloia elliptica</i> (C.A. Agardh) Cleve	MELL
<i>Nitzschia dissipata</i> (Kützing) Grunow abnormal form	NDTG
<i>Nitzschia</i> A.H. Hassall	NITZ
<i>Navicula libonensis</i> Schoeman	NLIB
<i>Navicula subrhynchocephala</i> Hustedt	NSRH
<i>Navicula zanoni</i> Hustedt	NZAN
<b><i>Pseudostaurosira brevistriata</i> (Grunow) Williams &amp; Round</b>	<b>PBTG</b>
<b><i>Rhopalodia gibba</i> (Ehrenberg) O.Muller</b>	<b>RGIB</b>
<b><i>Staurosira construens</i> Ehrenberg</b>	<b>SCON</b>
<b><i>Staurosirella pinnata</i> (Ehrenberg) Williams &amp; Round</b>	<b>SPIN</b>
<i>Sellaphora pupula</i> (Kützing) Mereschkowsky	SPUP
<i>Tabularia fasciculata</i> (Agardh) Williams et Round	TFAS
<b>Downstream of Potchefstroom Dam (PDM)</b>	
<b><i>Achnanthes exilis</i> Kützing</b>	<b>AEXI</b>
<i>Asterionella gracillima</i> (Hantzsch.) Heiberg	AGRA
<b><i>Amphora pediculus</i> (Kützing) Grunow</b>	<b>APED</b>
<i>Craticula ambigua</i> (Ehrenberg) Mann	CAMB
<i>Cocconeis placentula</i> Ehrenberg	CPLA
<i>Cyclotella meneghiniana</i> Kützing	CMEN
<i>Cyclotella ocellata</i> Pantocsek	COCE
<b><i>Cocconeis pediculus</i> Ehrenberg</b>	<b>CPED</b>
<b><i>Diatoma vulgare</i> Bory</b>	<b>DVUL</b>
<i>Encyonema caespitosum</i> Kützing	ECAE
<i>Encyonopsis minuta</i> Krammer & Reichardt	ECPM
<i>Fragilaria ulna</i> (Nitzsch) Lange-Bertalot	FULN
<i>Gomphonema acuminatum</i> Ehrenberg	GACU
<i>Gomphonema auritum</i> A.Braun ex Kützing	GAUR
<b><i>Gomphonema parvulum</i> (Kützing) Kützing</b>	<b>GPAR</b>
<i>Gomphonema pumilum</i> (Grunow) Reichardt & Lange-Bertalot	GPUM
<i>Gyrosigma rautenbachiae</i> Cholnoky	GRAU
<i>Melosira varians</i> Agardh	MVAR
<i>Navicula capitatoradiata</i> Germain abnormal form	NCPG
<i>Navicula cryptotenella</i> Lange-Bertalot	NCTE
<b><i>Nitzschia dissipata</i> (Kützing) Grunow</b>	<b>NDIS</b>
<i>Nitzschia littoralis</i> Grunow	NLIT
<i>Navicula schroeteri</i> var. <i>escambia</i> Patrick	NSES
<i>Nitzschia sinuata</i> (Thwaites) Grunow	NSIN
<i>Navicula tripunctata</i> var. <i>arctica</i> Patrick & Freese	NTPA
<b><i>Pseudostaurosira brevistriata</i> (Grunow) Williams &amp; Round</b>	<b>PBTG</b>
<i>Rhoicosphenia abbreviata</i> (Agardh) Lange-Bertalot	RABB
<i>Surirella angusta</i> Kützing	SANG

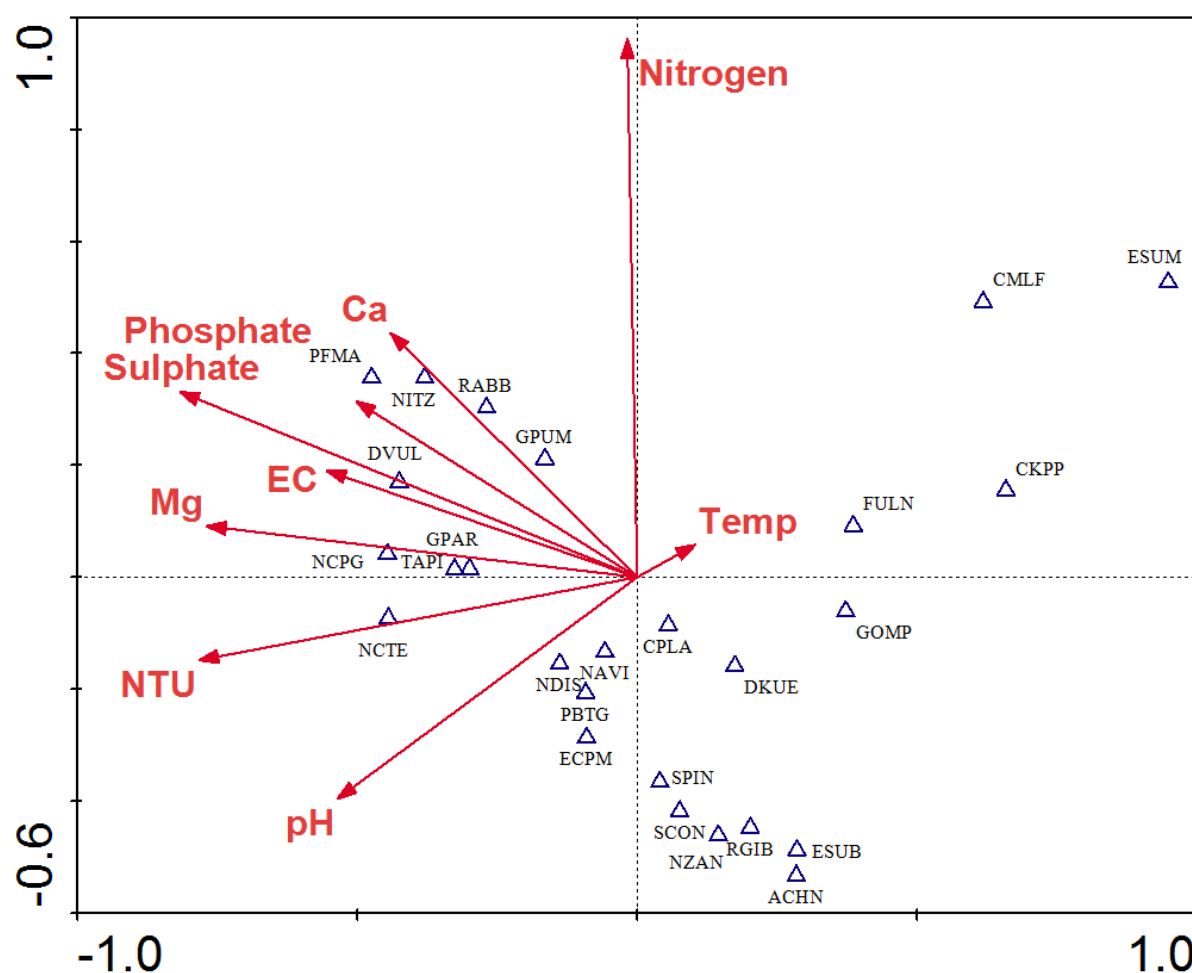
<i>Staurosira construens</i> Ehrenberg	SCON
<i>Surirella ovalis</i> Brebisson	SOVI
<b><i>Staurosirella pinnata</i> (Ehrenberg) Williams &amp; Round</b>	<b>SPIN</b>
<b><i>Sellaphora pupula</i> (Kützing) Mereschkowsky</b>	<b>SPUP</b>
<i>Tryblionella apiculata</i> Gregory	TAPI
<i>Tabularia fasciculata</i> (Agardh) Williams & Round	TFAS
<i>Tryblionella levidensis</i> W. Smith	TLEV
<b>Trompie Kitsgrass (RWP)</b>	
<b><i>Amphora pediculus</i> (Kützing) Grunow</b>	<b>APED</b>
<i>Caloneis</i> sp. Cleve	CALO
<i>Cyclotella meneghiniana</i> Kützing	CMEN
<i>Craticula molestiformis</i> (Hustedt) Lange-Bertalot	CMLF
<b><i>Cocconeis placentula</i> Ehrenberg</b>	<b>CPLA</b>
<i>Craticula cuspidata</i> var. <i>media</i> (Meister) Aboal	CRCM
<i>Cymatopleura solea</i> (Brebisson) W.Smith	CSOL
<i>Cymbella tumida</i> var. <i>borealis</i> (Grunow) Cleve	CTBO
<i>Diadesmis confervacea</i> Kützing	DCOF
<b><i>Diatoma vulgare</i> Bory</b>	<b>DVUL</b>
<i>Encyonema caespitosum</i> Kützing	ECAE
<i>Fragilaria biceps</i> (Kützing) Lange-Bertalot	FBCP
<b><i>Gomphonema gracile</i> Ehrenberg</b>	<b>GGRA</b>
<b><i>Gomphonema parvulum</i> (Kützing) Kützing</b>	<b>GPAR</b>
<i>Gyrosigma acuminatum</i> (Kützing) Rabenhorst	GYAC
<i>Melosira varians</i> Agardh	MVAR
<b><i>Navicula</i> sp. J.B.M. Bory de St. Vincent</b>	<b>NAVI</b>
<b><i>Navicula capitatoradiata</i> Germain abnormal form</b>	<b>NCPG</b>
<b><i>Navicula cryptotenella</i> Lange-Bertalot</b>	<b>NCTE</b>
<i>Nitzschia dissipata</i> (Kützing) Grunow	NDIS
<i>Navicula erifuga</i> Lange-Bertalot	NERI
<b><i>Nitzschia</i> A.H. Hassall</b>	<b>NITZ</b>
<i>Navicula tripunctata</i> (O.F.Müller) Bory	NTPT
<i>Navicula veneta</i> Kützing	NVEN
<i>Pseudostaurosira brevistriata</i> (Grunow) Williams & Round	PBTG
<i>Planothidium frequentissimum</i> var. <i>magnum</i> (Straub) Lange-Bertalot	PFMA
<i>Rhoicosphenia abbreviata</i> (C.Agardh) Lange-Bertalot	RABB
<i>Surirella angusta</i> Kützing	SANG
<i>Staurosira construens</i> Ehrenberg	SCON
<i>Surirella ovalis</i> Brébisson	SOVI
<i>Staurosirella pinnata</i> (Ehrenberg) Williams & Round	SPIN
<b><i>Sellaphora pupula</i> (Kützing) Mereschkowsky</b>	<b>SPUP</b>
<i>Sellaphora seminulum</i> (Grunow) D.G. Mann	SSEM

<b><i>Tryblionella apiculata</i> Gregory</b>	<b>TAPI</b>
<b>Elbrixon bridge (EBR)</b>	
<b><i>Amphora pediculus</i> (Kützing) Grunow</b>	<b>APED</b>
<i>Caloneis</i> sp. Cleve	CALO
<i>Cyclotella meneghiniana</i> Kützing	CMEN
<i>Craticula molestiformis</i> (Hustedt) Lange-Bertalot	CMLF
<b><i>Cocconeis placentula</i> Ehrenberg</b>	<b>CPLA</b>
<i>Craticula cuspidata</i> var. <i>media</i> (Meister) Aboal	CRCM
<i>Cymatopleura solea</i> (Brébisson) W.Smith	CSOL
<i>Cymbella tumida</i> (Brébisson) Van Heurck var. <i>borealis</i> (Grunow) Cleve	CTBO
<i>Diadesmis confervacea</i> Kützing	DCOF
<b><i>Diatoma vulgare</i> Bory</b>	<b>DVUL</b>
<i>Encyonema caespitosum</i> Kützing	ECAE
<i>Fragilaria biceps</i> (Kützing) Lange-Bertalot	FBCP
<b><i>Gomphonema gracile</i> Ehrenberg</b>	<b>GGRA</b>
<b><i>Gomphonema parvulum</i> (Kützing) Kützing</b>	<b>GPAR</b>
<i>Gyrosigma acuminatum</i> (Kützing) Rabenhorst	GYAC
<i>Melosira varians</i> Agardh	MVAR
<b><i>Navicula</i> sp. J.B.M. Bory de St. Vincent</b>	<b>NAVI</b>
<b><i>Navicula capitatoradiata</i> Germain</b>	<b>NCPG</b>
<b><i>Navicula cryptotenella</i> Lange-Bertalot</b>	<b>NCTE</b>
<i>Nitzschia dissipata</i> (Kützing) Grunow	NDIS
<i>Navicula erifuga</i> Lange-Bertalot	NERI
<b><i>Nitzschia</i> sp. A.H. Hassall</b>	<b>NITZ</b>
<i>Navicula tripunctata</i> (O.F.Müller) Bory	NTPT
<i>Navicula veneta</i> Kützing	NVEN
<i>Pseudostaurosira brevistriata</i> (Grunow) Williams & Round	PBTG
<i>Planothidium frequentissimum</i> var. <i>magnum</i> (Straub) Lange-Bertalot	PFMA
<i>Rhoicosphenia abbreviata</i> (C.Agardh) Lange-Bertalot	RABB
<i>Surirella angusta</i> Kützing	SANG
<i>Staurosira construens</i> Ehrenberg	SCON
<b><i>Surirella ovalis</i> Brébisson</b>	<b>SOVI</b>
<i>Staurosirella pinnata</i> (Ehrenberg) Williams & Round	SPIN
<i>Sellaphora pupula</i> (Kützing) Mereschkowsky	SPUP
<i>Sellaphora seminulum</i> (Grunow) D.G. Mann	SSEM
<b><i>Tryblionella apiculata</i> Gregory</b>	<b>TAPI</b>

# - Possible new species (Appendix B Figures A-4 and -5)

## 4.2.2 Diatoms and water quality

Diatom community structures consist of different species, each one unique and displaying different characteristics and responses to change in environmental changes (Azim *et al.*, 2005; Pan *et al.*, 1996; Potapova and Charles 2003). Thus the relationship between water quality and diatom community structure is important as different diatoms have different ecological requirements to thrive in their particular habitat. Some diatoms are more sensitive to changes and conditions in their specific habitat than other more tolerant species and for this reason are powerful indicators of ecological change. The canonical correspondence analysis scatterplot - Figure 4-1, allows us to observe the different diatom species assemblages in relation to a variety of environmental factors. The results displayed are those obtained from all the sites in the study area throughout 2014.



**Figure 4-1: Canonical correspondence analysis scatter biplot illustrating dominant diatom species (weight range of more than 15%) and their relation to water quality variables measured in the Mooi River for the time period 2014.**

**Table 4-2: Summary of canonical correspondence analysis (CCA) for Diatom and the physical and chemical water quality parameters of the Mooi River in 2014.**

Axes	1	2	3	4	Total
Eigenvalues	0.688	0.509	0.372	0.185	4.915
Species-environment correlations	0.960	0.920	0.887	0.639	
Cumulative percentage variance of species data	14.0	24.4	31.9	35.7	
Cumulative percentage variance of species-	33.2	57.8	75.7	84.6	
Sum of all eigenvalues					4.915
Sum of all canonical eigenvalues					2.071

In the previous chapter, trends in water quality were observed and it was clear that the sites with better water quality were BVO, KKD, BWS and BKD. Sites impacted by pollution were OWS, PDM, RWP and EBR. The effects of pollution could clearly be seen as the Mooi River flowed downstream towards the Vaal River. Similarly in Figure 4-1 the changes in diatom community structure and composition can be clearly seen, in relation to the changes in water quality.

When studying Figure 4-1 it is clear that *Cymbella kappii*, *Fragilaria ulna*, *Encyonopsis subminuta* and *Craticula molestiformis* group together in the top right quadrant of the CCA, these species associate with elevated levels of nitrate and elevated temperatures. The grouping of these organisms are slightly different to the ecology and tolerances described by Van Dam in 1994 and Taylor *et al.* in 2007b, this could be explained by the fact that they occur at BVO. In the discussion in section 3.2.7.1, BVO is described as a site with constant temperatures (averages), and high nitrate levels. The fact that the temperature is constant at ~20 °C and that the above mentioned species show a strong positive relationship to temperature may explain the slightly unexpected results.

The next group of diatom species to be discussed is located in the bottom right quadrant of the CCA scatterplot. The taxa *Achnanthes* sp., *Denticula kuetzingii*, *Navicula zanonii*, *Staurosira construens*, *Rhopalodia gibba* and *Staurosirella pinnata*, do not associate with the elevated water variables, and it can be deduced, that the species mentioned above indicate moderate to good water quality. These species usually occur in good quality water that is alkaline, the exception are *Rhopalodia gibba* and *Staurosirella pinnata* that favours moderate to high electrolyte content (Taylor et al., 2007c; Van Dam 1994). These species are dominant in KKD and BWS, which corresponds to the conclusions made in the previous chapter.

Species that are grouped together in the middle of the bottom half of the CCA scatterplot are *Cocconeis placentula*, *Encyonopsis minuta*, *Navicula* sp., *Nitzschia dissipata* and *Pseudostaurosira brevistriata*, these species associates with elevated pH, turbidity, EC, sulphate

and dissolved Mg and dissolved Ca. These species can be found in waters which are alkaline, calcareous, meso- to eutrophic with moderate to high electrolyte content (Taylor et al., 2007c; Van Dam 1994). These species are dominant in OWS and BKD, which supports the conclusions regarding quality made in the previous chapter.

As pollution increased species composition changed, although some species that indicate moderate to good water quality such as *Staurosira construens* are still present, the abundance is much reduced. The last species in the grouping in Figure 4-1 in the left half are *Diatoma vulgaris*, *Gomphonema parvulum*, *Gomphonema pumilum*, *Nitzschia* sp., *Navicula capitatoradiata*, *Navicula cryptotenella*, *Rhoicosphenia abbreviata* and *Tryblionella apiculata*. These species associate strongly with elevated to high concentration of the measured water quality variables. These species are collectively indicative of heavily polluted waters and the specific ecological requirements of some will be discussed below.

*Diatoma vulgaris* is a commonly occurring diatom and is closely associated with elevated to high levels of pollution - especially inorganic nutrients (Taylor et al., 2007c). Once a year, around spring time the diatom assemblages in the Mooi River become completely dominated by *Diatoma vulgaris*, this has happened yearly for the past several years (J.C. Taylor pers comm).

*Gomphonema parvulum* is described in many studies as a species that is tolerant to high levels of pollution, is resistant to trace metal pollution, low levels of oxygen, high levels of organic pollution and is dominant at sites where treated sewage effluent is present (Bere, 2014; Van Dam et al., 1994; Fukushima et al., 1994; Bere and Tundisi, 2012; Duong et al., 2010; Gold et al., 2003; Lobo et al., 2002).

It is clearly visible from Table 4-2 and Figure 4-1, how the diatom community structure changes, with relation to the change in the water quality, as the Mooi River flows downstream.

### **4.2.3 Diatom indices**

The following section contains results concerning the diatom indices that were used in the study. The information is used to evaluate the water quality integrity and aquatic ecosystem health of the Mooi River.

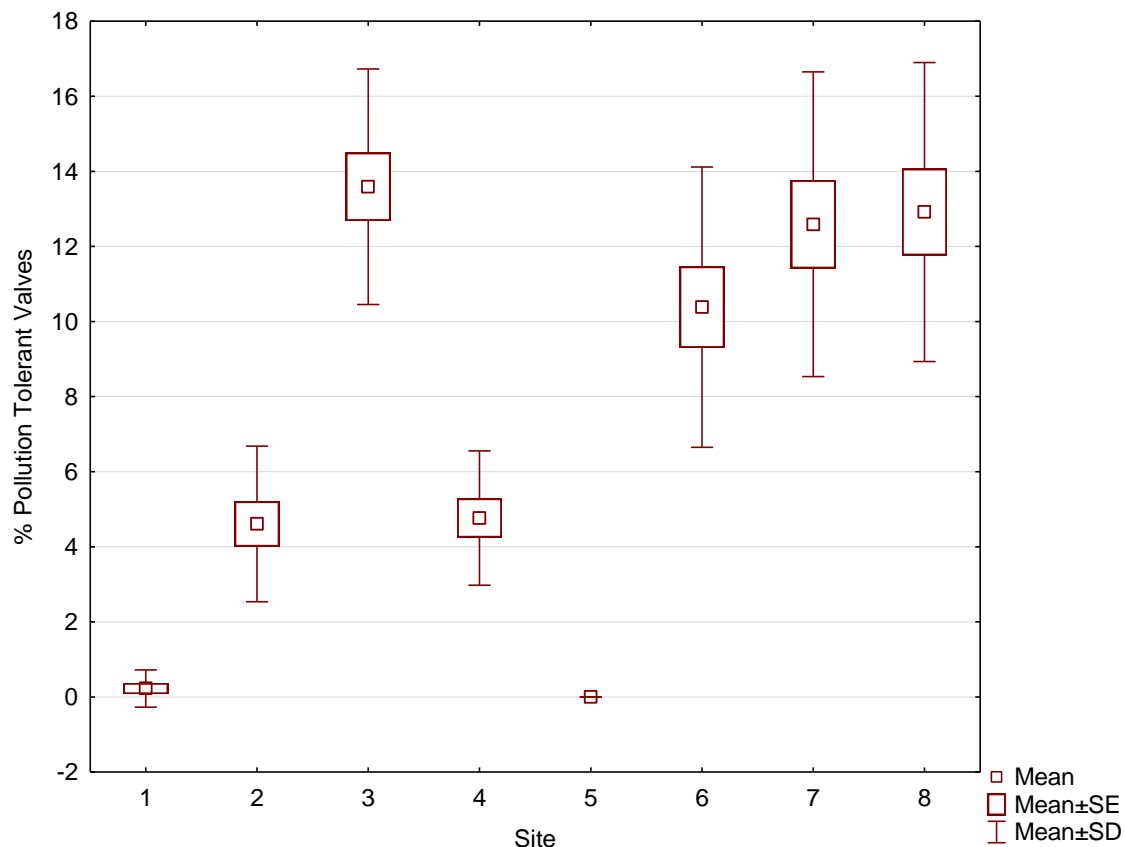
Appendix C Table A-13 contains Pearson correlations between water quality variables and the indices used, to determine the effectiveness of the indices. The marked values indicate significant differences with a  $p < 0.05$ . Appendix C Table A-14 contains Pearson correlations

between the indices used and positive correlations with significant differences occur between GDI, SPI and BDI. The % Percentage Pollutant Tolerant Valves has a significant negative correlation with GDI, SPI and BDI.

#### 4.2.3.1 Pollution Tolerant Valves (%PTV)

The Percentage Pollutant Tolerant Valves (%PTV) (Kelly and Whitton, 1995) indicates the percentage of diatoms present in the community that are tolerant to polluted conditions and can be used as a descriptor to discriminate between organic pollution and eutrophication.

Referring to Appendix C Table A-13, the %PTV correlates significantly with dissolved Mg and phosphate concentrations and pH, EC, TDS and turbidity.



**Figure 4-2: A Box and whisker plot illustrating the change in mean %PTV between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

Figure 4-2 indicates the %PTV of each site throughout 2014. The graph is a box and whiskers plot based on values calculated with the non-parametric Kruskal-Wallis test. BVO differs significantly from and BWS, PDM, RWP and EBR. BWS differs significantly from BVO, KKD,

OWS and BKD. BWS has the best measured water quality (Chapter 3) but has the highest average %PTV of all the sites, with an average of 13.9% which is a rather low score. All sites score below 20%, a score of above 20% would indicate the presence of organic waste in the river system, as none of the scores are higher than this there is no evidence for any sustained influx of organic material into the Mooi River system. However this is not to say that there is no nutrient present just that when it is present it is in an inorganic form.

As seen in Chapter 3, the typical upwards curve of the figures from PDM can be seen in Figure 4-2 as well, and this confirms that nutrient pollution does increase as the Mooi River flows downstream.

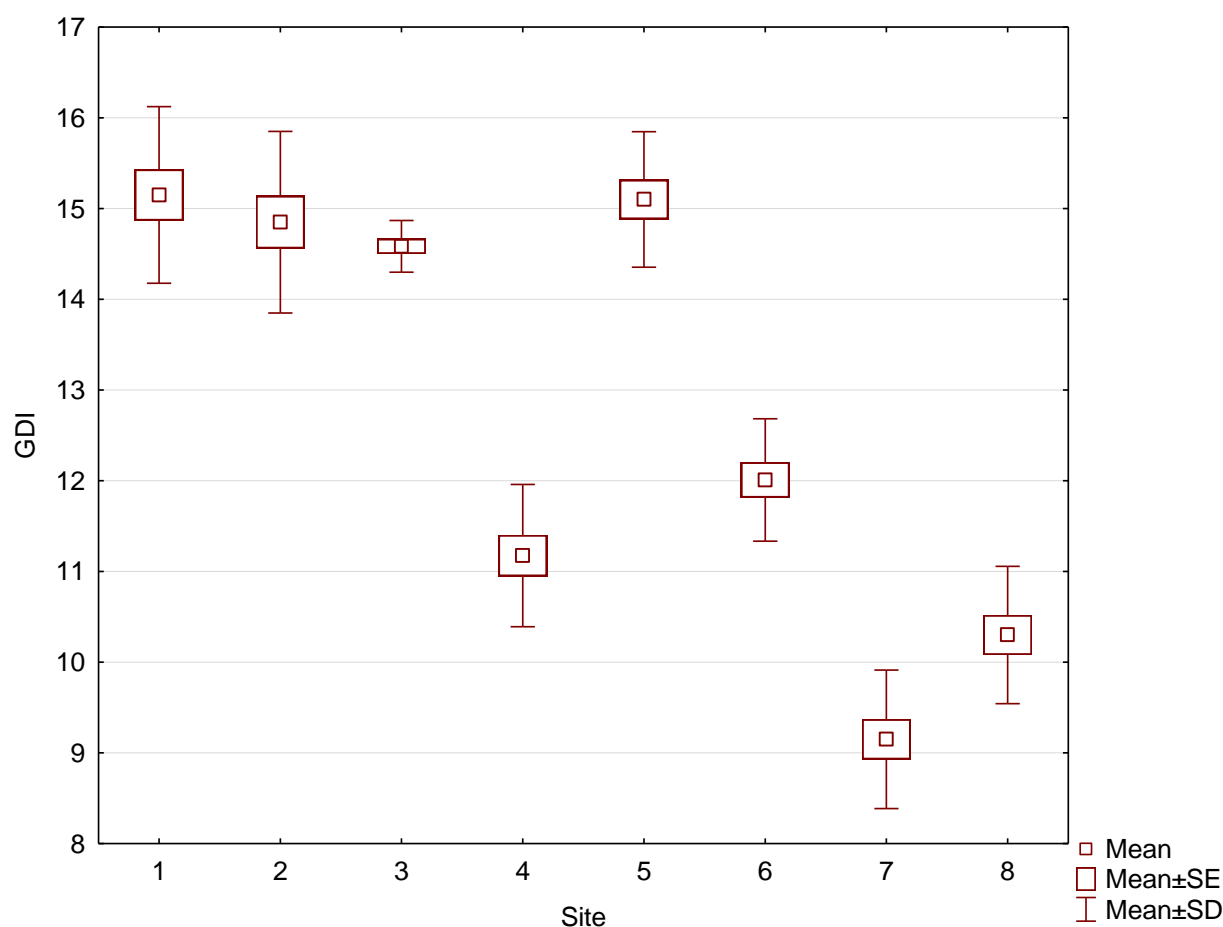
From Table A-12 Appendix C it can be seen that there is a slight increase in the %PTV in the months of March and October, and this indicates increased pollution levels during the months mentioned. Heavy rainfall experienced in late February (Appendix A: Figure A-1), would increase the runoff of pollutants and influence the change in community structure in the month to follow. The increase in %PTV in October can also be due to rainfall and the preparation of soil for the planting of crops.

The %PTV at the sites are not high, and only twice do the values reach over 20%, once in October at BWS with the %PTV at 20.8%, and once at EBR in March with the %PTV at 21.8%. Although the values are generally not high this index does illustrate the trend of increased pollution in the Mooi River from BVO flowing downstream towards EBR.

### 4.2.3.2 Generic Diatom Index (GDI)

The Generic Diatom Index (GDI) (Coste and Ayphassorho, 1991) is an index based on genus level identifications and with tolerance values ascribed at the generic rank. A total of 48 genera were identified in this study during 2014.

Referring to Appendix C Table A-13, Pearson correlation shows the GDI has a significant ( $p$  value  $< 0.05$ ) negative correlations to Ca, Mg, nitrate, phosphate, sulphate and EC, TDS and turbidity.



**Figure 4-3: A Box and whisker plot illustrating the change in mean GDI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

In Figure 4-3 the GDI scores are presented as an average per site throughout 2014. The GDI scores per site per sampling occasion may be found in Appendix C Table A-16. Significance of difference between the sites was calculated using the non-parametric Kruskal-Wallis test. The GDI of BVO, KKD and BKD differ significantly from OWS, PDM, RWP and EBR. BWS

differs significantly from RWP and EBR. In Figure 4-3 it can be observed that higher index scores are at BVO, KKD and BWS, with a gradual decline from BVO to BWS. A sudden drop in GDI occurs at OWS, and scores recover as the river flows to BKD. Low scores were obtained at PDM, and after Potchefstroom at RWP the GDI is at its lowest, and increases again slightly at EBR indicating some natural recovery of the Mooi River.

The average GDI score at BVO is 15.15 and ranging between 13 and 17, which indicates good water quality, with oligo- to mesotrophic water. The highest score of 17 in January and falls within the same water quality range, and the lowest scores are 13 in November and 14.3 in December, these scores fall below the average and indicate moderate water quality with the water in a mesotrophic state. The lower values may be a result of rainfall (Appendix A Figure A-1) and the increased pollutants associated with the agricultural runoff and grazing cattle.

The average GDI score at KKD is 14.85 and ranging between 13 and 17, which indicates good water quality, with oligo- to mesotrophic water. The GDI scores at KKD are constantly over 15 in January, February, March, April, May and October which indicates good water quality. The scores are the highest is 16.6 in March, although runoffs in February influenced %PTV in March. The reason KKD is in the moderate water category is scores in the winter were low, with the lowest score of 13.2 in June reducing the average to below 15. The opposite happened in the colder months, mixing of cold deep water with surface water leads to increased nutrient availability, this along with a decrease in water levels, due to little to no rainfall in the winter period, increases the nutrient levels of KKD and the score GDI directly indicates this response.

Downstream of KKD is BWS, and according to Chapter 3, the site with the best water quality. The average GDI of BWS is 14.58, which falls into the moderate water quality category with the water in a mesotrophic state. Scores at BWS are relatively constant with scores decreasing in the colder months. The highest score is in April, at 15.1 which indicates good water quality. This trend is similar to the slightly increased GDI scores at both BVO and KKD for the same period. The lowest GDI scores are in November and December with scores of 14.2 and 14 respectively. Although still in the moderate water quality range it is slightly lower than the average, and again follows the same trend as BVO and KKD for the same period. This could be a result of increased water temperature and the organisms response to acclimation. The time period is also associated with field preparation and planting of crops, and increased rainfall that causes runoff of pollutants accumulated on the land during the dry season.

OWS was shown in Chapter 3 to be impacted by the mining activity in Wonderfonteinspruit, has an average GDI of 11.18 and falls into the poor water quality class; the Mooi River is classified as meso- to eutrophic. The GDI is relatively constant, there are however lower scores in July and August of 10.8 and 10 respectively, and can be attributed to the increase in concentration of pollutants due to the lower water levels during the dry season. An increase in scores occur with the start of the spring, but falls again to 10.6 and 9.8 in November and December respectively and is probably due to the same reasons as already discussed.

BKD is the next site and has an average GDI of 15.1 which indicates good water quality which is oligo- to mesotrophic. Low scores in the winter months of 13.6 and 14.5 in June and July follow the trend at KKD. The scores are lower again in December with a score of 14.2 and following the same trend for this month as the other sites already discussed.

Lower GDI scores can be noted at Figure 4-3 at PDM with an average of 12.01 which indicates moderate water quality in a mesotrophic state. The index scores are not constant and also do not follow the same trend in score fluctuations as the upstream sites. There is agricultural activity from BKD and PDM and the results of this are seen in a lower GDI scores than those from BKD, the reason that there are no seasonal fluctuation patterns may be due to the fact that PDM is downstream of Potchefstroom dam, and the flow of the river is regulated from Potchefstroom dam and therefore PDM has abnormal fluctuations in GDI scores.

RWP has an average GDI of 9.15 which indicates poor water in a meso- to eutrophic state. This was expected as RWP is influenced by various pollutant sources that include industrial and waste water effluents and urban and agricultural runoff. The GDI scores do not have the same seasonal fluctuations as the upstream sites. Higher GDI scores were obtained in the dry season in June, July and August with GDI scores of 9.7, 9.4 and 10.5 respectively. In general higher rainfall introduces more water to an aquatic ecosystem, this dilutes pollutants, and the water quality appears higher. At RWP the situation is opposite. However, if we consider when rainfall is low, there is less urban runoff, the flow of the Loopspruit is lower and thus flushes less pollutants into the Mooi River. The WWTP thus did not experience flushing events associated with high rainfall. With heavy rains WWTP may have overflows and may lead to semi-treated sewage entering the Mooi River.

The last site is EBR, and has an average GDI of 10.3 which indicates poor water quality in a meso- to eutrophic state. The GDI scores are slightly improved from RWP but still indicate poor water quality. The GDI show a slight improvement in water quality. EBR is heavily impacted by

agricultural activities, and the lowest GDI is observed in May at 9.1, this can be attributed to the harvesting of crops. GDI scores fall in May due to the influence of waste water from Potchefstroom. There is also a lower GDI score in December, and can be attributed to late field preparations in November.

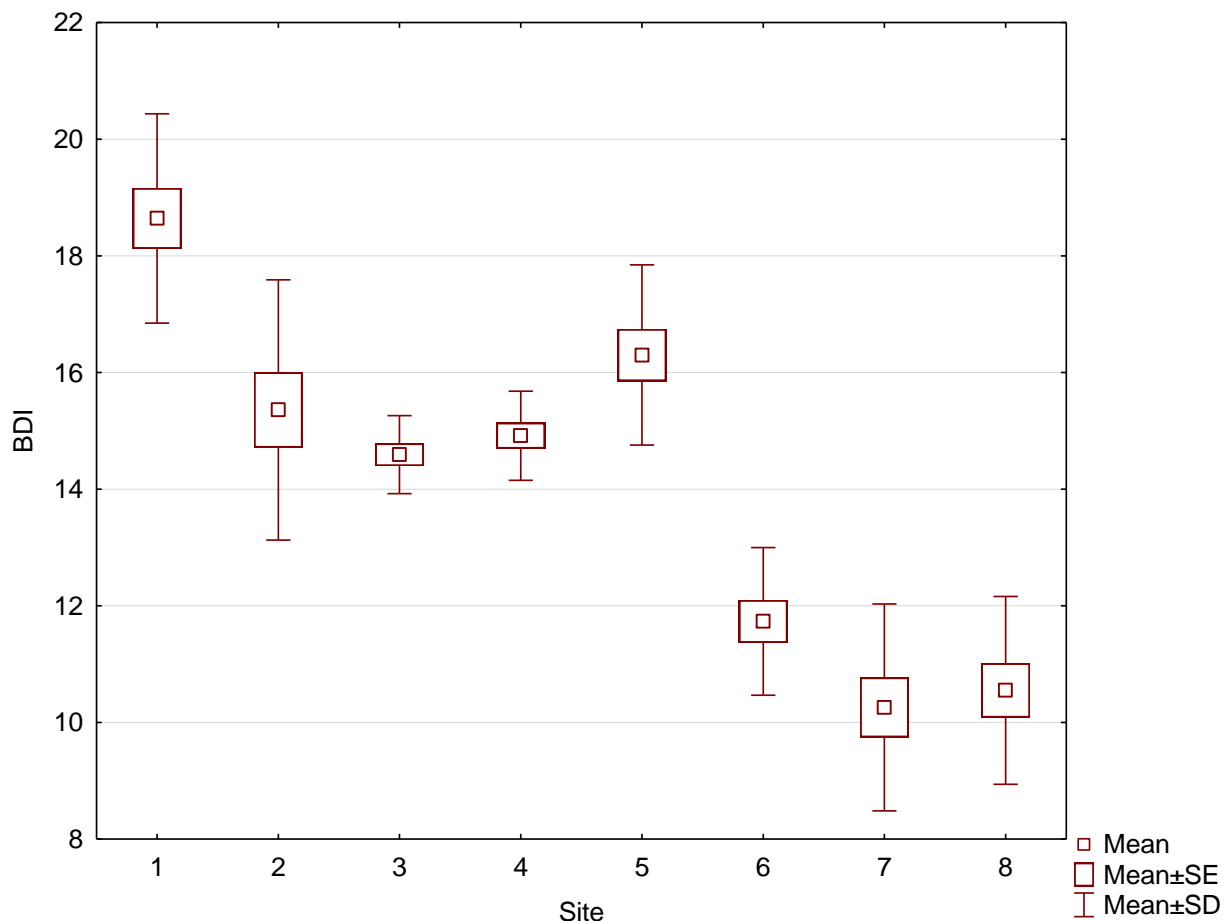
To summarise, the GDI clearly indicates and increase in water quality impacts from upstream to downstream as demonstrated by the measured water quality variables. It is clear that pollution increases as the Mooi River flows from BVO to EBR, and the different pollutant sources have different effects on the GDI scores. It is also clear that the most impacted site is RWP, and is expected, due to the cumulative effect of the various pollution sources.

It should however be pointed out that the GDI is based on identifications at a genus level, only the tolerances and sensitivities of the genera are taken into account and this may yield only a coarse indication of the actual state of the river. Water quality indication based on genera scores can be useful, especially as it requires less skills in identification, however specific species within a genus might have very different sensitivity values than the rest of the genus making indices which use species more accurate.

#### **4.2.3.3 Biological Diatom Index (BDI)**

Biological Diatom Index (BDI) (Lenoir and Coste, 1996) uses 14 water quality variables associated with diatom sensitivity. The BDI does not use as many species as the SPI and groups some species that are difficult to separate and identify under the light microscope together.

From Appendix C Table A-13, Pearson correlation shows significant ( $p$  value  $< 0.05$ ) negative correlations between the SPI scores Ca, Mg, phosphate and sulphate concentrations as well as pH, EC, TDS and turbidity.



**Figure 4-4: A Box and whisker plot illustrating the change in mean BDI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

In Figure 4-5 the BDI scores are presented as an average per site throughout 2014. The BDI scores per site per sampling occasion may be found in Appendix C Table A-16. Significance of difference between the sites was calculated using the non-parametric Kruskal-Wallis test. The BDI of BVO and KKD differ significantly from PDM, RWP and EBR. BWS and OWS differ significantly from RWP and EBR. BKD differ significantly from PDM, RWP and EBR. In Figure 4-5 it is observed that higher index scores are noted at BVO, KKD, BWS, OWS and BKD with a gradual decrease from BVO to BWS. An increase in BDI scores is observed at OWS and BKD. Low scores were calculated for PDM, and after Potchefstroom at RWP the BPI is at its lowest, and increases again at EBR indicating some natural recovery of the Mooi River.

As mentioned above the BDI does not have as many species available on which the calculation of the index score is based. Figure 4-5 that the BDI calculates score according to water quality, and OWS has an average score of 14.59, indicating moderate water quality. OWS has an average score of 14.91, indicating that this site, heavily influenced by mining pollution, seems to have

better water quality than BWS. Another site that does not agree with the general trend found in this study is BKD, with an average BDI of 16 would then appear to be of better quality than KKD which has an average score of 15.36. The explanation could be, at KKD only 59.9% of species are included in the calculation and at BKD 75.55% of species are included. At OWS and BWS 74.29% and 77.17% respectively are included in the calculation. The BDI score being higher at OWS than at BWS can be explained by *Nitzschia* sp., not being included in the calculation at OWS, as already mentioned, *Nitzschia* is a genus associated with high levels of pollution. These inclusion rates of species into the index calculation are rather low when comparing to the SPI or BDI which generally has inclusion rates ranging from 90-100%.

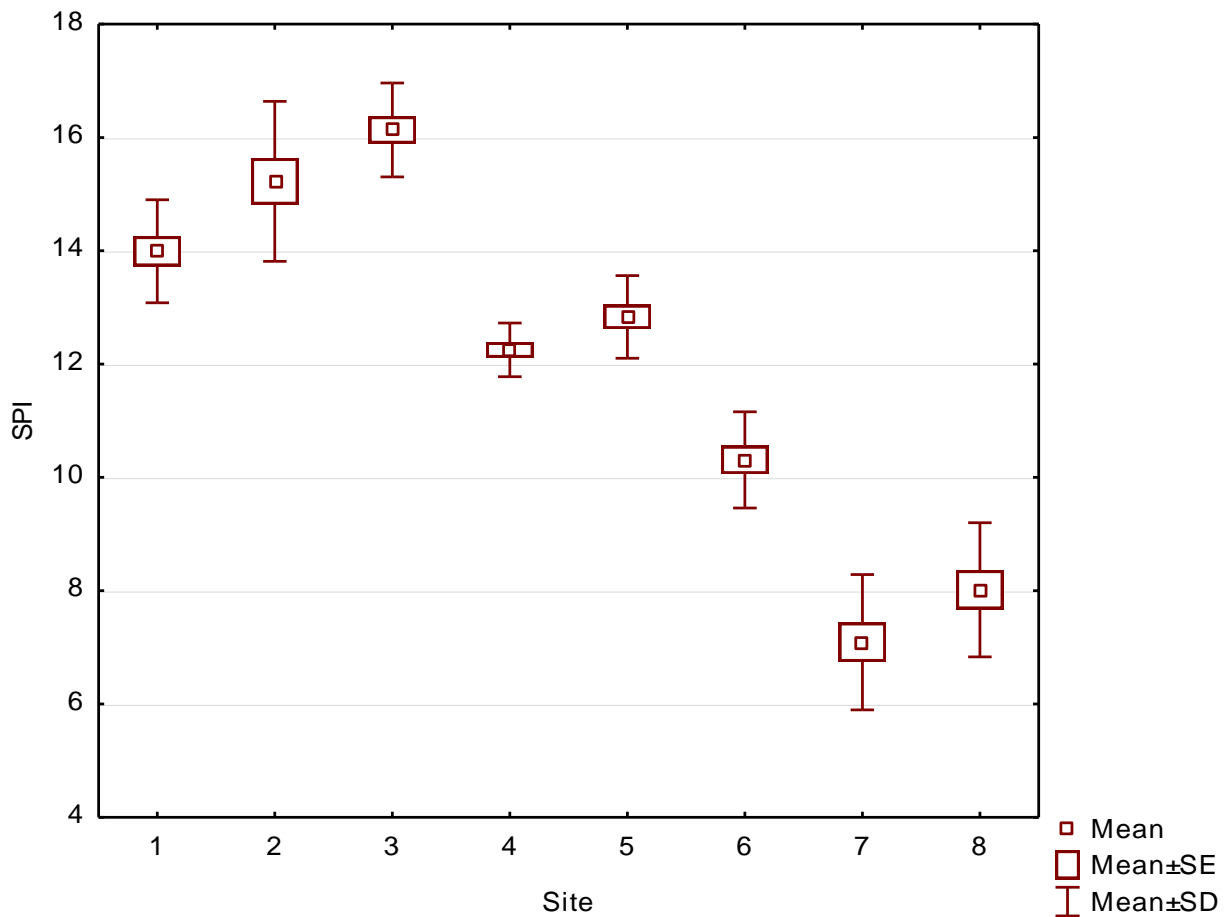
This trend of differing index scores can also be noted at PDM, RWP and EBR.

To summarise, the BDI did not reflect trends in water quality as accurately as the GDI and SPI, but still indicated an overall decline in aquatic ecosystem health in the Mooi River from BVO to EBR, with RWP as the most polluted site and slight recovery from RWP to EBR. The BDI also shows that Potchefstroom has a greater influence on pollution than Wonderfontein spruit. High scores after Wonderfontein spruit can be attributed to non-inclusion of species in the calculation of the index score.

#### **4.2.3.4 Species Pollution sensitivity Index (SPI)**

The Specific Pollution sensitivity Index (SPI) (CEMAGREF, 1982), is the index containing the most species in its database and incorporates species into five discrete sensitivity groups.

From Appendix C Table A-13, Pearson correlation shows significant ( $p$  value  $< 0.05$ ) negative correlations between the SPI scores and dissolved Ca, Mg, nitrogen, phosphate and sulphate concentrations and elevated pH, EC, TDS and turbidity. This shows that the SPI gives an accurate reflection of measured water quality variables.



**Figure 4-5: A Box and whisker plot illustrating the change in mean SPI score between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

In Figure 4-4 the SPI scores are presented as an average per site throughout 2014. The SPI scores per site per sampling occasion may be found in Appendix C Table A-16. Significance of difference between the sites was calculated using the non-parametric Kruskal-Wallis test. The SPI of BVO and KKD differ significantly from PDM, RWP and EBR. BWS differs significantly from OWS, PDM RWP and EBR. OWS differs significantly from BWS which has a higher SPI and RWP which has a lower SPI. BKD differs significantly from RWP and EBR. In Figure 4-4 it can be observed that higher index scores occur at BVO, KKD and BWS, with a gradual increase from BVO to BWS. A sudden drop in SPI is observed at OWS, and slightly improves as the river flows to BKD. Low scores are seen at PDM, and after Potchefstroom at RWP the SPI is at its lowest, and increases again slightly at EBR indicating slight natural recovery of the Mooi River.

The average SPI score at BVO is 13.99 and ranging between 13 and 17, which indicates good water quality, with oligo- to mesotrophic water. BVO is the first site and has an SPI average

score of 13.99, and falls into moderate water quality category with water in a mesotrophic state. The SPI at BVO moves out of its index range twice, with SPI scores of 15.4 and 15.3 in January and September respectively, and falls into the 15 to 17 which indicates good water quality, with oligo- to mesotrophic water. A drop in SPI is seen in the winter months of June July and August, and follows the same trend as the measured water quality variables - elevated EC, turbidity and nitrate concentrations. The elevated concentrations of water quality variables and lower SPI scores are possibly a result of little to no rainfall (Appendix A; Figure A-1) and a drop in the water level, causing more concentrated nutrients at BVO. A lower SPI is seen in November, and this can be ascribed to the reasons discussed in section 4.3.3.2.

KKD is the first impoundment and downstream of BVO and has an SPI average score of 15.23 and falls into the good water quality, with oligo- to mesotrophic water. The highest SPI score is 17.1 in January and indicates high water quality and oligotrophic conditions. The SPI scores are lower in the colder winter months, this also follows the trend in measured water quality variables in particular elevated sulphate levels. Chlorophyll *a* concentrations are exceptionally high in July at 202µg/L - an increase from 2.8µg/L in June. This was also accompanied by higher temperature - increasing from 6.11°C in June to 11.7°C in July. The SPI score is also at its lowest in July at 12.4 and falls into the water quality class with the water in a mesotrophic state. Water quality results indicated an algal bloom in this time period, which can be caused by an increase in nutrient concentrations due to lower water levels and possible stratification. Water levels decreased due to lower rainfall and pollutants became more concentrated. With rainfall in October comes an increase in the SPI.

BWS had the highest SPI average score of 16.13 and falls into good water quality, with oligo- to mesotrophic water. The highest SPI score is 17.3 in April, with another score over 17 in February indicating high water quality and oligotrophic conditions. The highest score in April could be due to high rainfall in March with minimal agricultural activity in that time (time before harvesting). This is confirmed by water quality at BWS in April, with low EC levels and nutrient concentrations. The lowest SPI score is in July of 14.5, and is possibly due to the combination of agricultural activity and low water levels. A lower SPI score of 15 was calculated for November 15 and follows the same trend at the BD, and may be due to the preparation of fields for planting of maize.

At OWS we see the influence of mining pollution in Wonderfontein spruit, Figure 4-4 shows lower SPI scores compared to the upstream site. OWS has an average SPI score of 12.25 and places OWS in the category moderate water quality with the water in a mesotrophic state. The

SPI scores are relatively constant with no major fluctuations. The SPI score is lower in June and July, and may be due to lower water levels, and slightly elevated nutrients. The water quality of OWS is also relatively constant throughout 2014, and it is reflected in the SPI score. The BDI score is also constant at OWS (Section 4.2.3.3).

The second impoundment in the study is BKD and has an SPI average score of 12.83 and falls in category moderate water quality in a mesotrophic state. The highest SPI score is in May at 14.1 and it falls to 11.9 in June, elevated sulphate levels can be seen in June but no other water quality fluctuations were observed. The scores in the winter months are lower and follow the same trend with the BDI and GDI scores of KKD and the BDI of BKD, and can be due to colder, deep nutrient rich water mixing with cooling surface temperature water which allows for resuspension of nutrient and for it to become available to the algae. Water levels decreased due to lower rainfall with a concomitant increase in the concentration of a pollutants.

PDM is influenced by agricultural activity, and Figure 4-4 shows lower SPI scores for PDM with an average of 10.31 and falls in the poor water classification and in a meso- to eutrophic state. The index scores are not constant at this site and also do not follow the same trend as the SPI score fluctuations of the upstream sites. There is agricultural activity from BKD and PDM and the results of these activities are seen in a lower SPI average score than that at BKD. The reason there are no fluctuation patterns may be due to the fact that PDM is downstream of Potchefstroom Dam, and the flow of the river is regulated by Potchefstroom Dam.

RWP is the site with the highest cumulative pollution as seen in Chapter 3 and has an average SPI of 7.1 and falls in bad water quality class in a eutrophic state. The lowest SPI score was recorded in February (5.3). This was expected as RWP is influenced by various pollutant sources that include industrial and waste water effluents and urban and agricultural runoff. The SPI score is 6 in January and constantly increases as the rainfall decreases, reaching the highest value of 8.6 in August. Warmer temperatures in September with low rainfall cause an increase in nutrient concentration due to lower water levels. And in October the SPI is again lower because of heavy rainfall, causing runoff of accumulated pollutants into the Mooi River, and possible overflows at the WWTP, causing SPI scores to decrease.

The last site is EBR, and has an SPI average score of 8.03, and falls in the bad water quality category indicating that the Mooi River is in a eutrophic state at EBR. The SPI is a slight improvement from RWP but still indicates bad water quality. As with the GDI, the SPI follows the same trend as the water quality data, which indicates a very slight improvement. EBR is

heavily impacted by agricultural activities, and the lowest SPI was observed in March, and correlates with very high EC levels in the whole Mooi River for that time. There is also a lower SPI score in December, and can be attributed to late agricultural field preparations in November.

To summarise, the SPI indicate an increase in water quality from BVO with its high nitrogen concentrations to BWS with very good water quality. The SPI correlates significantly with the water quality data collected and SPI also confirms that Potchefstroom has the biggest influence on water quality, and the effects are seen until the last site at EBR.

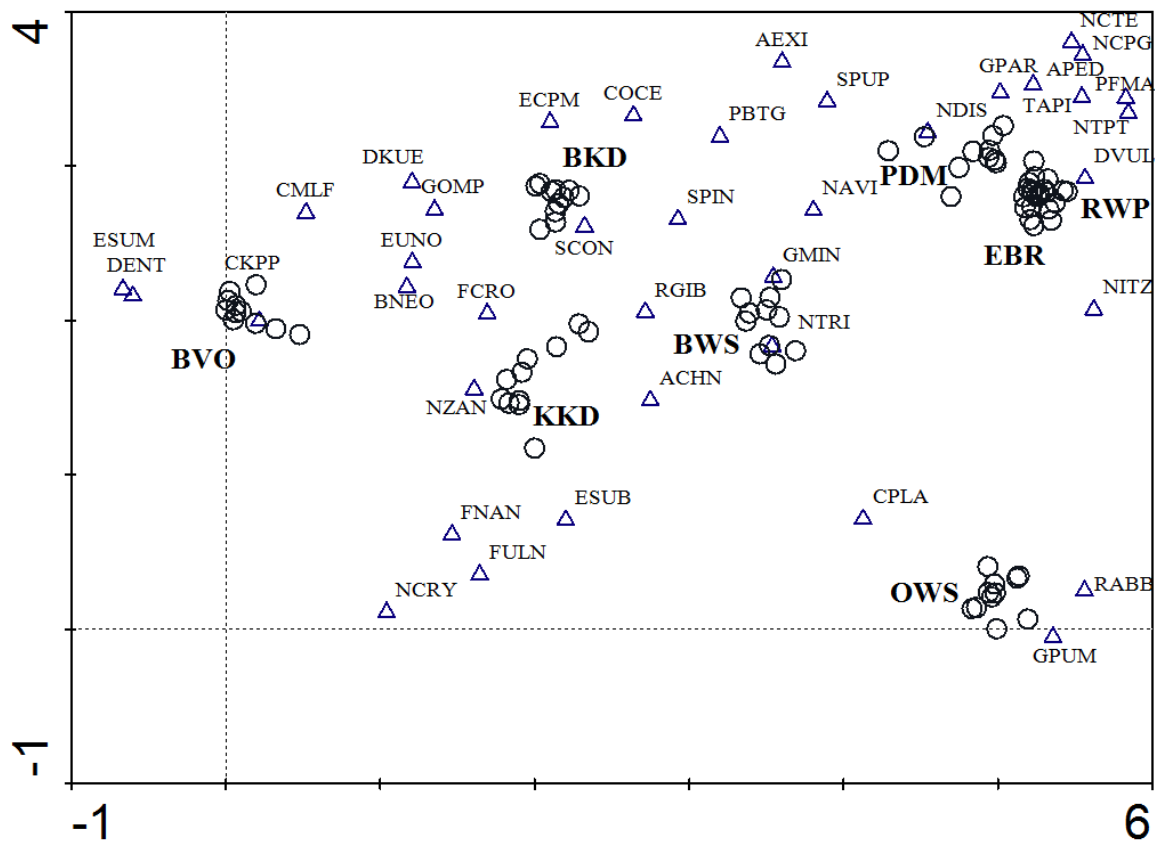
The species specific calculations of the SPI shows more accurate results than GDI or BDI, but the possibility of errors is increased due to the minor differences between certain species making species identification difficult, sometimes the consequences of incorrect identification can lead to significant errors in interpretation of water quality.

#### **4.2.4 Spatial and temporal variation of diatom assemblages**

In this section changes in diatom community structure and assemblage, related to differences between sites (spatial) and in time (temporal) will be presented. Temporal changes are shown calculated per site, with averages of species abundances found for each season. The discussion will refer back to the spatial and temporal variations in water quality as supplementary information (section 3.2.8.1).

##### **4.2.4.1 Spatial variation of diatoms in the Mooi River**

Figure 4-1 shows how diatom community structure changes in relation to the changes in water quality within the Mooi River in 2014. Compare this figure to Figure 3-10 which illustrates the how each site is associated with different water quality variables. Figure 4-6 is used to illustrate how the different sites in the Mooi River are associated with the different diatom species. Figure 4-6 will thus illustrate the changes in diatom community structure per site



**Figure 4-6: Detrended correspondence analysis scatterplot represents the spatial variation of dominant diatom species recorded from each site.**

Figure 4-6 shows the difference in diatom community in order to demonstrate which diatom species were found at each site, as well to show the diatom species that are distributed between sites. This provides an interpretation tool to illustrate changes in community structure as pollution increases from upstream to downstream.

The first site discussed will be BVO as we move from an upstream to downstream direction. BVO is relatively unimpacted by pollution and the species that occur at BVO are either intolerant towards any pollutants, or to slight elevation in pollutant levels. The species that are predominantly found at BVO are *Cymbella kappii*, *Encyonopsis subminuta* and an unidentified *Denticula* sp. These species are usually found in cleaner water, but can tolerate slightly elevated levels of pollution (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). Species also occurring as BVO are *Craticula molestiformis*, *Denticula kuetzingii*, *Gomphonema* sp.1 and *Eunotia* sp. species that also occur at BKD and *Brachysira neoexilis*, *Fragilaria crotonensis* and *Fragilaria ulna* that

share the species with KKD these species also occur at these other sites, and are not dominant at either of the sites. *Cymbella kappii* occurs at KKD as well, but never in large numbers, but are extremely dominant at BVO and that is why it is seen in strong correlation with BVO.

KKD is the first impoundment and the impoundment with the best water quality in the Mooi River catchment. The species that are found regularly at the site KKD are *Navicula zaroni*, *Fragilaria crotonensis* and *Eunotia subarcuatoidea*. These diatoms prefer clean habitats with low electrolyte content and a water body in an oligotrophic state (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). *Eunotia subarcuatoidea*, *Fragilaria nanana* and *Navicula cryptocephala* also occur at KKD but in low numbers, and do not occur at other sites or are found in very low numbers. *Staurosira construens*, *Rhopalodia gibba*, *Achnanthes* sp. and *Fragilaria crotonensis*, are present at KKD, BKD and BWS, and are neither indicators of good water quality nor are they indicators of bad water quality (indifferent) with the exception of *Staurosira construens* which prefer but is not limited to good water quality (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). They are common enough in these sites to have an influence on the diatom indices already discussed.

BWS is the site with the best water quality confirmed by the SPI scores and the water quality results found in Chapter 3. The species that are found mainly at BWS are *Gomphonema minutum*, *Navicula tridentula*, *Rhopalodia gibba* and *Staurosirella pinnata*, these species occur in oligo- mesotrophic waters, with preference to moderate electrolyte content, but not in waters that are more than moderately polluted (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). The diatom species distributed between both KKD and BKD have already been discussed above. The other species occurring at BWS are *Pseudostaurosira brevistriata* and *Cocconeis placentula*, but their abundances were very low.

OWS is impacted by mining activities. The species that are predominantly found at OWS are *Cocconeis placentula*, *Gomphonema pumilum* and *Rhoicosphenia abbreviata*. *Cocconeis placentula* occurs in meso- to eutrophic standing water and is found in large abundances on plants. *Gomphonema pumilum* and *Rhoicosphenia abbreviata* both occur in meso- to eutrophic water and are able to tolerate in high to critical levels of pollution (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). *Nitzschia* sp. and *Diatoma vulgare* are also associated with OWS and both are known as indicators of poor water quality, *Nitzschia* sp. and *Diatoma vulgare* ordinate at some distance from OWS as they are the two dominant species at two sites, and were also found at the impacted sites EBR and RWP.

BKD shows an improvement in water quality as is reflected by the similar community structure at both BWS and KKD. The dominant species at BKD are *Staurosira construens*, *Encyonopsis minuta* and *Cyclotella ocellata*, are found in clean water, with elevated electrolyte levels. *Cyclotella ocellata*, is found in meso-eutrophic water and prefer high pH values (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994) matching the chemical profile of the site. BKD has many diatom species in common with PDM. Other species that are associated and are slightly dominant in both of these sites are *Pseudostaurosira brevistriata*, *Achnanthes exilis*, *Sellaphora pupula*, an unknown *Navicula* sp. and *Staurosirella pinnata*, these species occur in rather unpolluted water with high electrolyte content, and prefer meso- to eutrophic conditions with *Sellaphora pupula* able to withstand strongly polluted water (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). There is a possibility that a new or unusual *Eunotia* species was encountered, the shape and occurrence are unique, as *Eunotia* prefers acidic water but the species occurred regularly in the high pH waters of BKD. A new *Gomphonema* species may also have been discovered at BKD, the diatom is similar to *Gomphonema rautenbachiae*, but with the internal and external openings of the proximal raphe fissures strongly offset – a rather unique character. Images of these species can be seen in Appendix C Figure A-4&5.

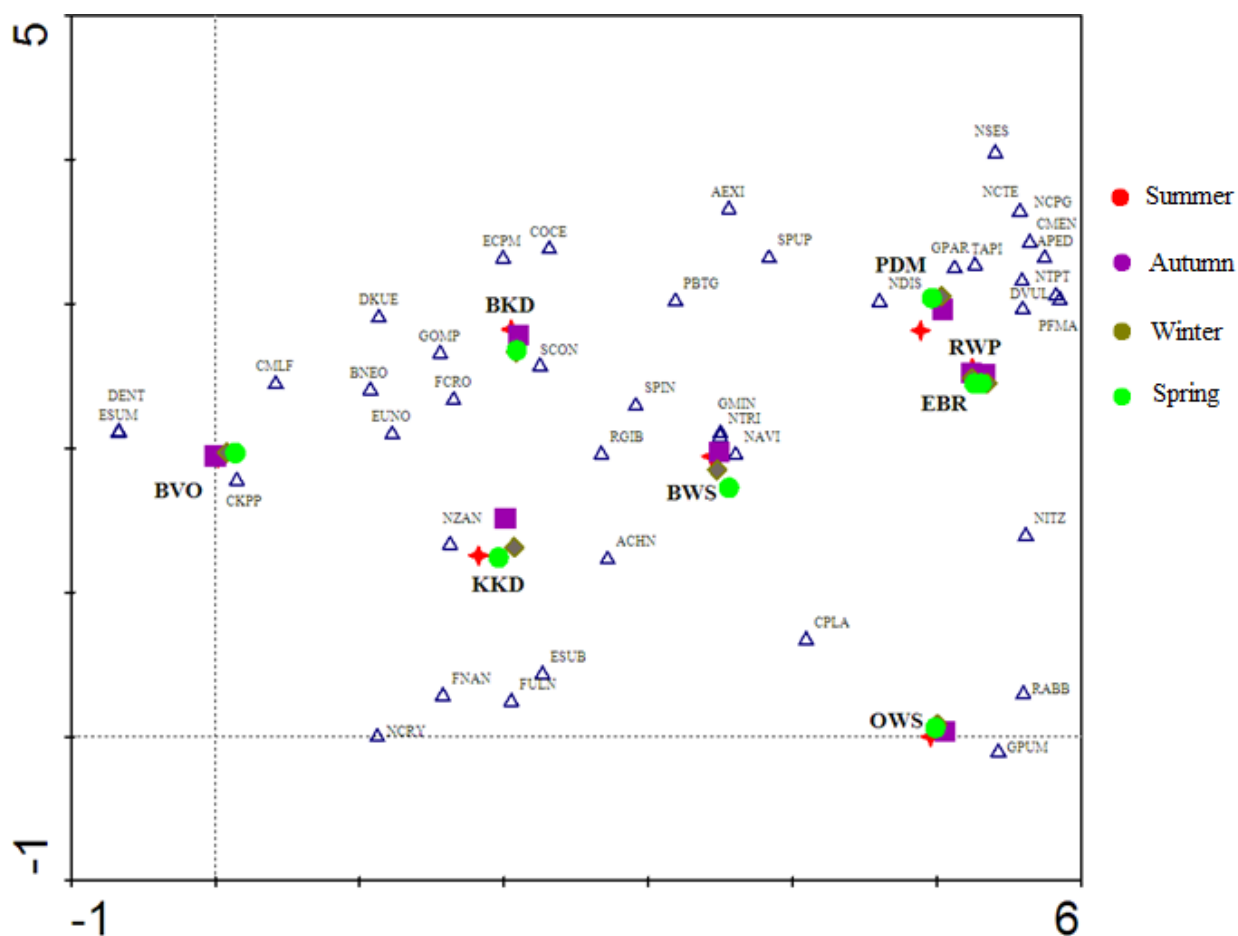
PDM is the next site, downstream of Potchefstroom Dam, and is affected by agricultural pollution upstream of the dam. As mentioned above PDM and BKD a notable shift in community structure commences, most of the diatom species found at PDM were not present at the sites BVO, KKD and BWS despite the relatively short geographical distance separating these sites. The site PDM is dominated by *Sellaphora pupula*, *Navicula* sp., *Nitzschia dissipata*, *Tryblionella apiculata*, *Amphora pediculus*, and *Gomphonema parvulum*, these species are tolerant of critical levels of pollution preferring eutrophic waters with high electrolyte content (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). PDM is closely associated with RWP, but the species PDM and BKD have in common are the only those species present at PDM that are not associated with critical levels of pollution. The diatom indices confirm with the spatial variation in species found at PDM, as well as the increase in water quality impacts as discussed in Chapter 3.

Sites RWP and EBR will be discussed together as the two sites share the same species, water quality and diatom index scores. The community structure at RWP and EBR is completely dissimilar to the species composition of BVO. RWP and EBR are dominated by *Amphora pediculus*, *Nitzschia* sp., *Diatoma vulgare*, *Navicula tripunctata*, *Navicula cryptotenella*, *Navicula capitatoradiata* and *Tryblionella apiculata*, all found in waters with critical levels of

pollution, with high electrolyte content high nutrient content (eutrophic) (Taylor *et al.*, 2007c; Van Dam *et al.*, 1994). These two sites do not correspond with any of the sites having good quality water, and they only share a few species with PDM although there are already changes in community structure from PDM to RWP. The pollution that Potchefstroom contributes to the Mooi River can clearly be seen at RWP, and as with the other results, RWP is again the site that is influenced the most within the system.

The deterioration of the aquatic ecosystem health is quite apparent when observing the spatial variation of the diatom species. The complete change in community structure is so severe that *Encyonopsis subminuta*, a species found at BVO, has a negative correlation greater than 6 with *Planothidium frequentissimum* a species found at RWP and EBR.

#### 4.2.4.2 Temporal variation of diatoms in the Mooi River



**Figure 4-7: Detrended correspondence analysis scatterplot represents the temporal variation of dominant diatom species represented at each site.**

Figure 4-7 represents the temporal variation found in the diatom community structure of the Mooi River system. The temporal grouping was done by averaging the diatom counts per site seasonally, and displaying them on a DCA plot. Comparing Figure 4-7 with Figure 4-6, there are no significant changes in community structure with regard to seasons.

# **CHAPTER 5 MACROINVERTEBRATES**

## **5.1 Introduction**

The following chapter contains the results and discussion of the macroinvertebrate data collected at every sampling opportunity. The results will be presented as species composition at the sites, correlations between the SASS5 index and water quality variables and index values will also be presented to illustrate the differences in ecosystem health between sites. A table with individual results per site per sampling occasion can be found in Appendix D and will be referred to during the discussion. Thereafter temporal and spatial will be illustrated to reflect differences in macroinvertebrate community structure between sites as well as seasonal changes in a one year period.

## **5.2 Results and discussion**

The sites and their abbreviations are as follows:

1. Bovenste Oog (BVO)
6. Downstream of Potchefstroom Dam (PDM),
7. Trompie Kitsgras (RWP)

## 5.2.1 Macroinvertebrate taxa per site

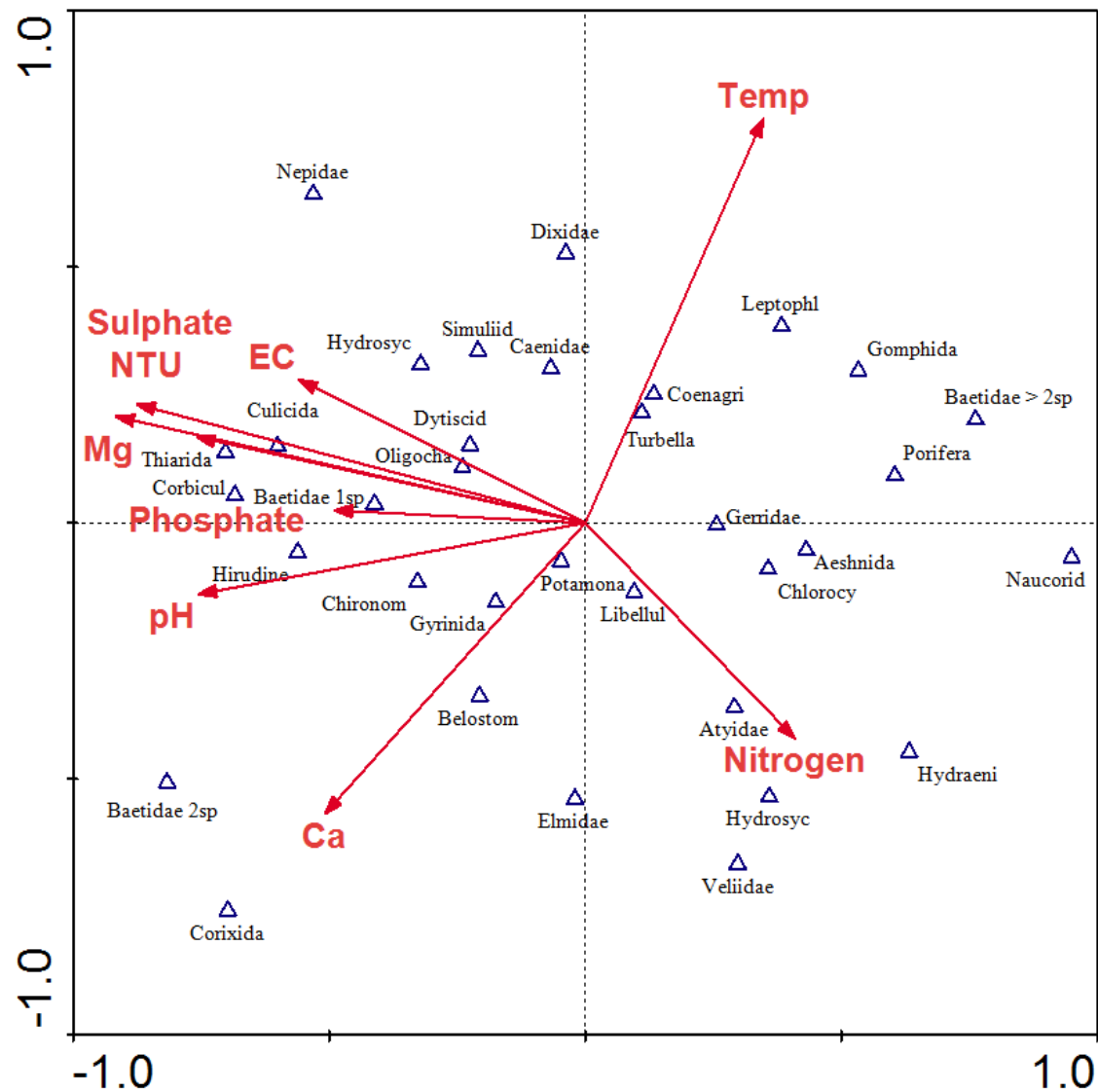
The taxa found at each site are presented in Table 5-2, this table will be referred to throughout the following discussion.

**Table 5-1: Distribution of macroinvertebrate taxa per site during the 2014 sampling season.**

BVO	PDM	RWP
Aeshnidae	Aeshnidae	Aeshnidae
Amphipoda	Atyidae	Atyidae
Atyidae	Baetidae >2	Baetidae 1
Baetidae >2	Baetidae 2	Baetidae 2
Baetidae 2	Belostomatidae	Belostomatidae
Belostomatidae	Caenidae	Caenidae
Blepharoceridae	Chironomidae	Chironomidae
Caenidae	Chlorocyphidae	Chlorocyphidae
Chironomidae	Coenagrionidae	Coenagrionidae
Chlorocyphidae	Corbiculidae	Corbiculidae
Coenagrionidae	Corixidae	Corixidae
Corixidae	Culicidae	Culicidae
Culicidae	Dixidae	Dixidae
Dixidae	Dytiscidae	Dytiscidae
Dytiscidae	Elmidae	Elmidae
Ecnomidae	Gerridae	Gerridae
Elmidae	Gomphidae	Gomphidae
Gerridae	Gyrinidae	Gyrinidae
Gomphidae	Hirudinea	Hirudinea
Gyrinidae	Hydraenidae	Hydraenidae
Heptageniidae	Hydrosychidae	Hydrosychidae
Hirudinea	Hydrosychidae	Hydrosychidae
Hydraenidae	Leptophlebiidae	Libellulidae
Hydrosychidae	Lestidae	Lymnaeidae
Hydrosychidae	Libellulidae	Naucoridae
Hydrosychidae	Limnichidae	Nepidae
Leptophlebiidae	Lymnaeidae	Oligochaeta
Libellulidae	Nepidae	Potamonautidae
Limnichidae	Notonectidae	Psychodidae
Lymnaeidae	Oligochaeta	Simuliidae
Naucoridae	Philopotamidae	Turbellaria
Notonectidae	Porifera	Veliidae
Oligochaeta	Potamonautidae	
Philopotamidae	Psychodidae	
Porifera	Simuliidae	
Potamonautidae	Thiaridae	
Psychodidae	Turbellaria	
Simuliidae	Veliidae	
Turbellaria		
Veliidae		

### **5.2.2 Macroinvertebrates and water quality**

Knowledge of the relationship between water quality and macroinvertebrate community structure is a valuable source of information for inferring reigning environmental conditions. Different macroinvertebrate taxa have different ecological requirements which need to be met in order to thrive in their particular habitat. Some macroinvertebrates are therefore more sensitive than others to changes in conditions in their specific habitat. It is the knowledge of these sensitivities which allows us to draw inferences on the state of the environment based on macroinvertebrate community structure. This is illustrated in the canonical correspondence analysis scatterplot (Figure 5-1) in which the macroinvertebrate taxa show specific preferences to specific environmental factors. The results represented in the CCA are those obtained from the three sites suitable for SASS for the period 2014.



**Figure 5-1: Canonical correspondence analysis scatterplot illustrating macroinvertebrate taxa occurrence in relation to differences in water quality.**

**Table 5-2: Summary of canonical correspondence analysis (CCA) for macroinvertebrates and the physical and chemical water quality parameters of the Mooi River in 2014.**

Axes	1	2	3	4	Total
Eigenvalues	0.251	0.130	0.081	0.063	1.520
Species-environment correlations	0.952	0.901	0.807	0.803	
Cumulative percentage variance of species data	16.5	25.1	30.4	34.6	
Cumulative percentage variance of species-	32.5	49.4	59.9	68.1	
Sum of all eigenvalues					1.520
Sum of all canonical eigenvalues					0.771

In Chapter 3, changes in water quality were observed and it was clear that the sites with better water quality were BVO, KKD, BWS and BKD. The sites impacted by pollution were OWS, PDM, RWP and EBR. The effects of pollution could clearly be seen as the Mooi River flowed downstream towards the Vaal River. In Figure 5-1 the changes in macroinvertebrate community structure and composition can be seen clearly in relation to the change in water quality due to various pollutants.

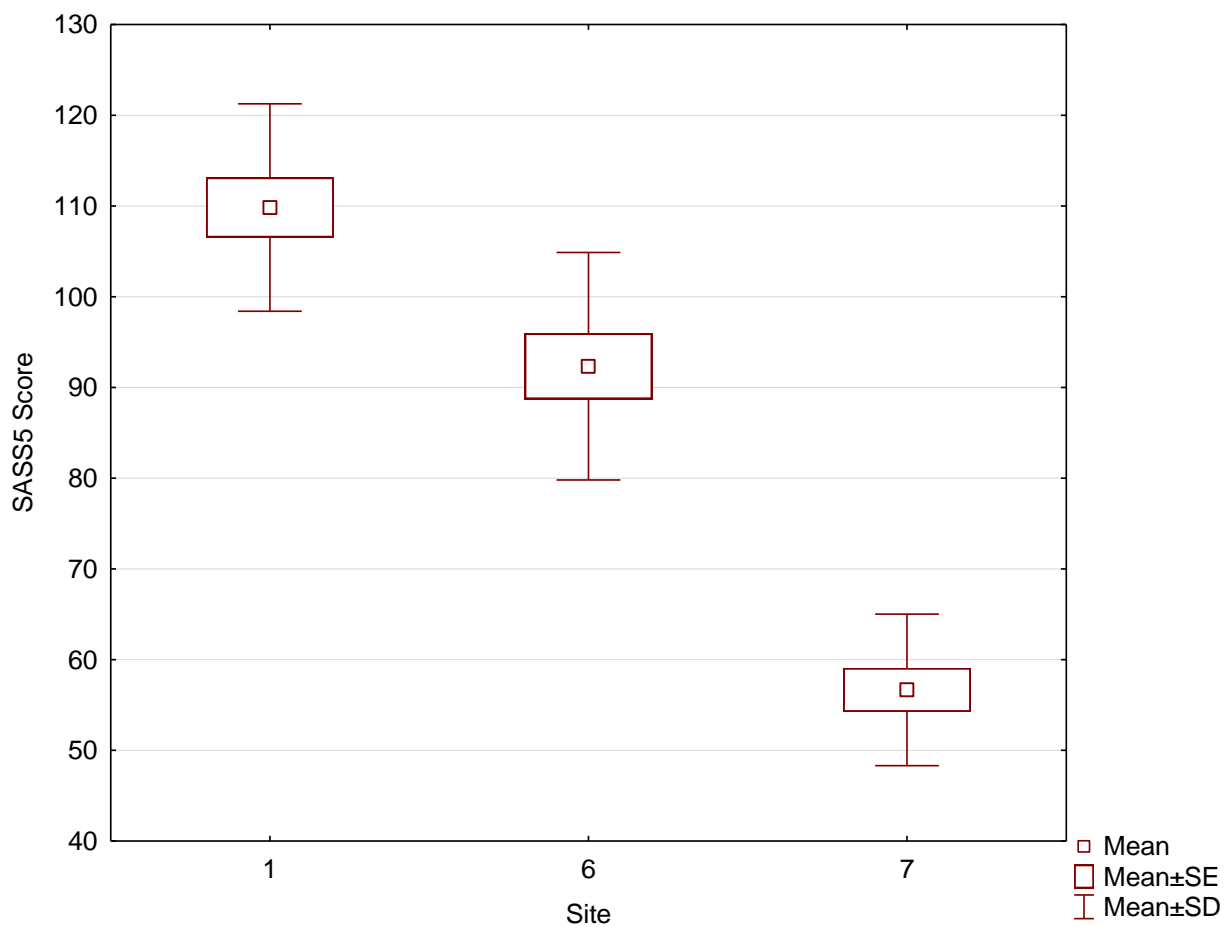
When studying Figure 5-1 it can be observed that Aeshnidae, Leptophlebiidae, Baetidae >2, Gomphidae, Naucoridae, Chlorocyphidae and Porifera are grouped together on the right of the CCA, these taxa are associated with elevated levels and nitrogen and elevated temperatures. The groupings of these organisms, are slightly different to the established sensitivity scores as most of the above mentioned taxa have high sensitivity scores (Dickens and Graham, 2002; Gerber and Gabriel, 2002). However these taxa all occur abundantly at BVO a site with constant temperatures (ground water fed), and high nitrogen levels but with no other form of impact.

Taxa that are only partially or slightly influenced by elevated levels of pollution are found in the center of the CCA, they are not specifically grouped together as the taxa in the negative quadrant prefer slightly elevated levels of nitrogen, dissolved calcium, pH and phosphates, whereas the organisms in the positive quadrant are found in water with slightly elevated levels of dissolved magnesium, sulphate, turbidity and phosphates. These taxa are Atyidae, Belostomatidae, Caenidae, Coenagrionidae, Dixidae, Hydrosychidae, Libellulidae, Nepidae Potamonautidae, Simuliidae, Turbellaria and Veliidae. The grouping of these organisms agrees somewhat with established sensitivity scores and most of the above mentioned taxa have average sensitivity scores at around 7 (Dickens and Graham, 2002; Gerber and Gabriel, 2002).

As pollution increased, the occurrence of taxa found at the above mentioned sites became less dominant and community structure shifted although some taxa that indicate good water quality such as Baetidae 2sp, Dixidae and Elmidae were still present but in lower numbers. The last taxa grouped together in Figure 5-1 on the left half are Baetidae 1sp, Corbiculidae, Culicidae, Chironomidae, Gyrinidae, Hirudinea, Nepidae, Oligochaeta and Coenagrionidae. The occurrence of these taxa were strongly associated with elevated to high levels of the water quality variables measured in the Mooi River for 2014. The sensitivity scores of these taxa are also low with an average of 5 (Dickens and Graham, 2002; Gerber and Gabriel, 2002)

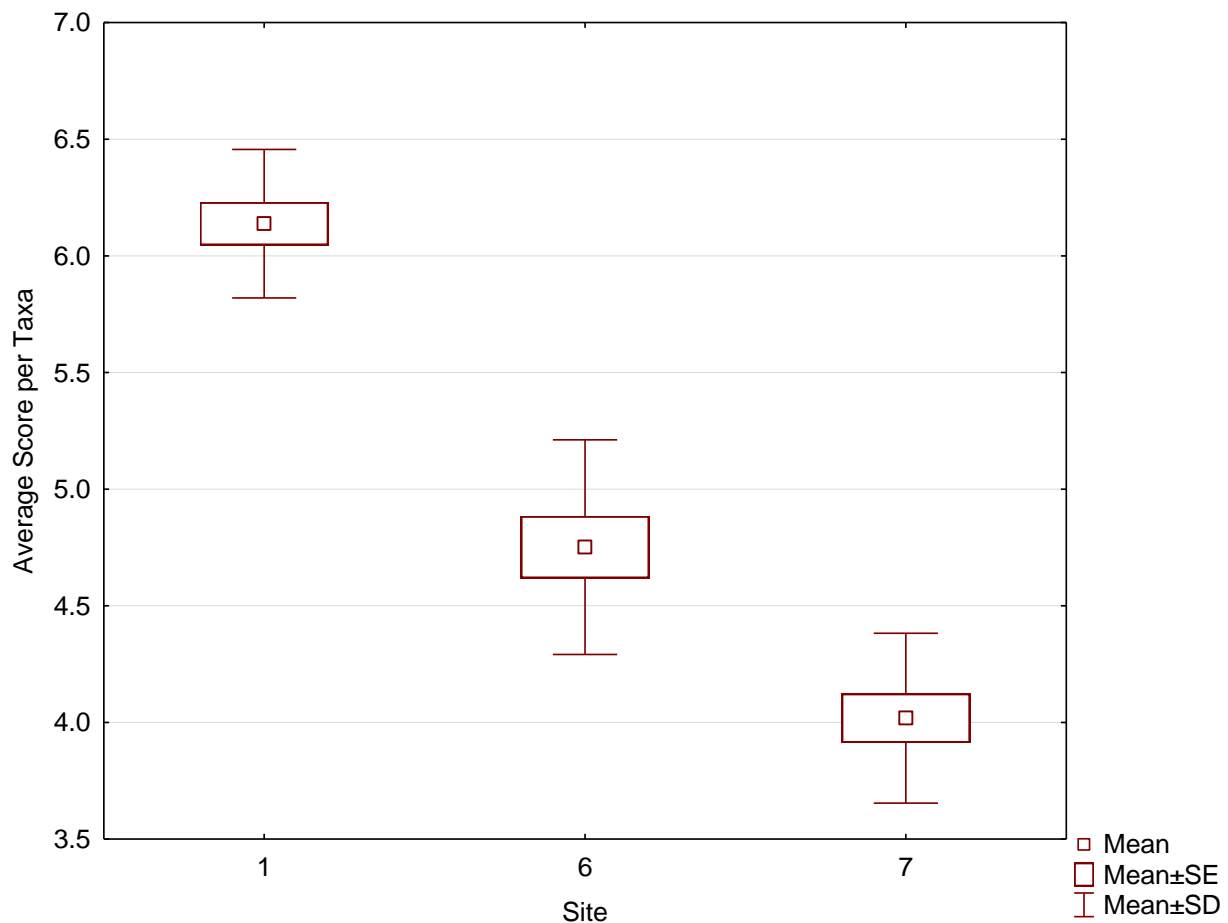
### 5.2.3 Macroinvertebrate indices

The South African Scoring System version 5 (SASS5) and Average Score Per Taxa (ASPT) were the indices chosen to determine the aquatic ecosystem health of the Mooi River. SASS5 and ASPT scores will be discussed together. SASS5 is calculated based on the total combined sensitivity values of each taxa and the ASPT scores are calculated based on the SASS5 score divided by the number of taxa found. Figure 5-2 and 5-3 illustrate SASS5 scores and ASPT respectively. ASPT



**Figure 5-2: A box-and-whisker plot illustrating the change in mean SASS5 score between sites from upstream (1- BVO) to Potchefstroom (6- PDM) and downstream towards (7- RWP) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

From Figure 5-2 it is clear that the SASS5 scores decrease from BVO to PDM and also from PDM to RWP a drastic decrease is seen. This agrees with the diatom-based SPI scores from section 4.2.3.3. When significant differences are determined (Appendix D Table A-21) it can be seen that only RWP show significant differences from BVO and PDM.



**Figure 5-3: A Box and whisker plot illustrating the change in mean ASPT scores between sites from upstream (1- BVO) to Potchefstroom (6- PDM) and downstream towards (7- RWP) (January 2014 – December 2014). SE = Standard Error; SD = Standard Deviation.**

From Figure 5-3 it can be observed that the ASPT scores decline from upstream to downstream, with the biggest drop in score from BVO to PDM, then from PDM to RWP a decrease in scores is again noted. This follows the same trend as the SPI scores discussed in section 4.2.3.3. When significant differences are determined (Appendix D Table A-22), it can be seen that only BVO showed a significant difference to PDM and RWP.

Both the above figures indicate BVO to be the site with the highest ecological integrity. BVO has an average SASS5 score of 109.83 and an average ASPT of 6.14 and the average number of taxa at BVO was 17.91. This places the site BVO in the B/A ecological category based on both SASS5 and ASPT scores. Table 2-3 classifies an A as natural unmodified ecological status, and a B as a good, natural ecological condition with slight modification. The lowest SASS5 score at BVO was 92 in July and the second lowest in August, giving the BVO for these months an ecological classification of B. The ASPT for the same period was 5.75 and 5.81 respectively and it still gave an ecological integrity score of A. This slight shift can be explained by an increase in

nutrients, EC and turbidity at BVO for these months. Seasonal change could also have had an effect, in particular causing lower water levels and increasing the concentration of pollutants. The highest SASS5 score is in March at 131, and the ASPT for the same period is 5.95, this is due to the number of taxa decreasing to 22 from 17. Hence the SASS classification at BVO places it in a category A - a higher ecological category. The ASPT score is lower but is still in the A category, but it is lower than the average, the increase in taxa in March had lower sensitivity scores than those usually found at BVO.

The community at PDM was impacted cumulatively by all of the pollution occurring from BVO to PDM, including the mining and agricultural pollution. PDM has an average SASS5 score of 92.33 which places it in the B category for ecological integrity, indicating a good, natural ecological condition with slight modification. The average ASPT is 4.75 and places it in the C category which indicates fair ecological integrity that is moderately modified. There were a large number of taxa found at PDM with moderate sensitivities, the high SASS5 score can be attributed to the higher diversity and the lower ASPT average to the sensitivity of these taxa. The highest SASS5 score is 106 in February, and the ASPT for the same period is 5.3, which puts PDM in a B category for both scores. As seen in section 4.2.3.3 the diatom SPI did not fluctuate over the seasons at PDM, and this was attributed to the influence of Potchefstroom Dam. The macroinvertebrates however show a response in the form of seasonal change with lower scores in the winter with a lowest SASS5 score of 58 in July, and an ASPT for the same period of 3.87, The SASS score gave a D classification and the ASPT an E, which suggests that pollution occurred in the month of July to classify PDM as seriously modified to critically modified. This may have been a result of low flow at this period limiting available habitat for invertebrates at the site.

The last site studied was at RWP and the community structure was affected by the pollution associated with urban runoff, the WWTP and agricultural pollution from Loopspruit. The average SASS5 and ASPT scores were 56.67 and 4.02 respectively, and placed RWP in the ecological category of D and E respectively which indicate largely to seriously modified habitat and/or pollution. This same result was obtained in section 4.3.3.3 using the SPI which indicated very bad water quality. The highest SASS5 and ASPT at RWP were 73 and 4.3, this indicates better ecological integrity of a class C and D respectively in December. Again this does not correspond with the diatom index scores which indicate a decline in water quality, and not an improvement. This could be as a result of higher nutrient availability, and hence more food for the macroinvertebrates, attracting and allowing slightly more sensitive taxa to flourish because of

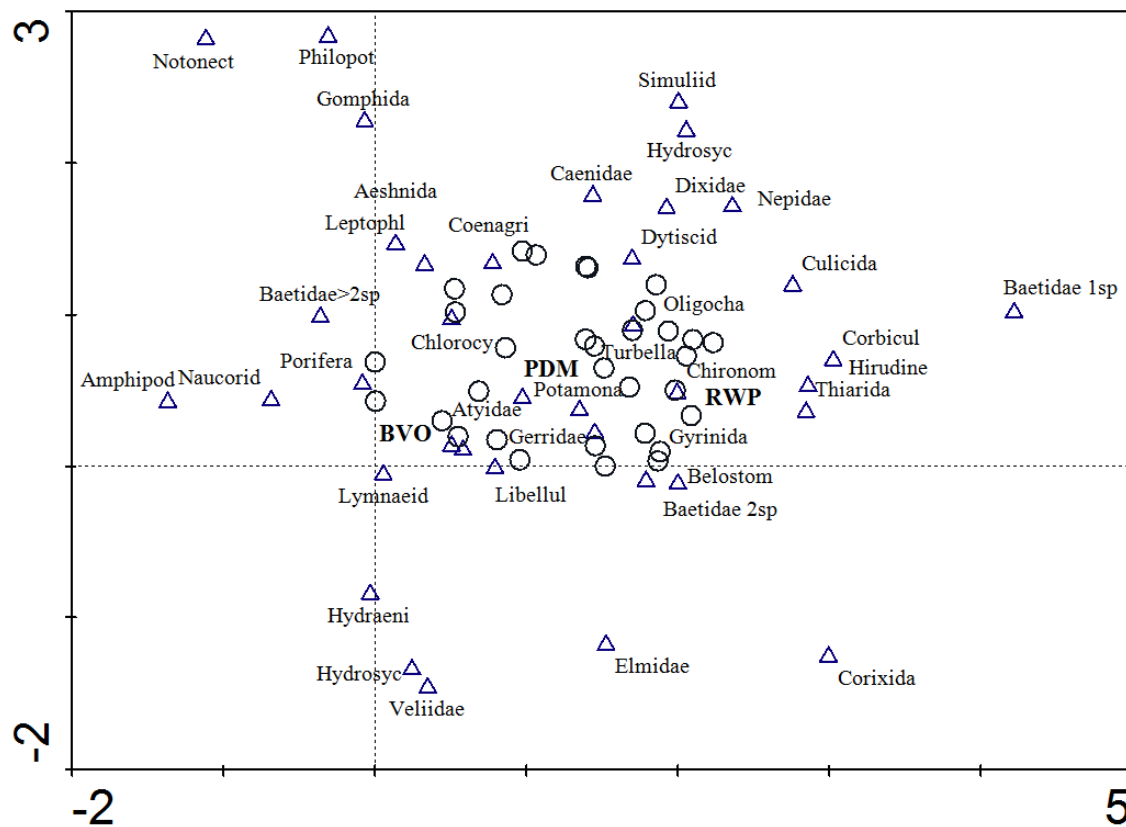
food availability in the start of the summer. The lowest SASS5 and ASPT can be observed in November only a month before the best score in December. The scores fall in the E/F category, and indicate seriously to critically modified conditions, this can be due to higher rainfall and associated increased urban runoff, potential WWTP spills and the preparation of fields for the planting of maize in the Loopspruit catchment.

## **5.2.4 Spatial and temporal variation in macroinvertebrate assemblages**

The results in this section are based on the changes in macroinvertebrate community structure and assemblages between sites (spatial) and in time (temporal). Temporal changes were also calculated per site, with averages of species abundances found in each season. The discussion will be refer back to the spatial and temporal variations in water quality (Chapter 3) as supplementary information.

### **5.2.4.1 Spatial variation of macroinvertebrates in the Mooi River**

Figure 5-4 will be used as the basis for a discussion of how the macroinvertebrate community structures change in relation to the changes in water quality in the Mooi River 2014. Figure 5-4 illustrates how each site is associated with different water quality variables. The detrended correspondence analysis in Figure 5-4 illustrates associations between sites and the various macroinvertebrate taxa. Figure 5-4 is thus presented to illustrate the changes in macroinvertebrate community structure per site.



**Figure 5-4: Detrended correspondence analysis scatterplot represents the spatial variation of the macroinvertebrate taxa at each site.**

Figure 5-4 illustrates distribution of macroinvertebrate taxa in order to reflect the community structure at each site, as well as the taxa that are have a shared distribution between sites. This is to illustrate how the community structure changes in response to increases in pollution.

Spatial variation of macroinvertebrates is presented as frequency of occurrence. The taxa close to the 3 and minus 2 horizontal axes have low frequency of occurrence and the taxa occurring close to the central axis have a high frequency of occurrence.

At BVO various taxa are found but only a few are restricted to this site only. Aeshnidae, Amphipoda, Leptophlebiidae, Baetidae >2, Gomphidae, Naucoridae, Notonectidae, Chlorocyphidae and Porifera are taxa that have a high sensitivity rating, with the exception of Porifera (Dickens and Graham, 2002; Gerber and Gabriel, 2002). These species form part of an aquatic ecosystem that is in a good condition and this is supported by the results from the water quality and diatom analyses. BVO shares many taxa with PDM and RWP, which include Atyidae, Belostomatidae, Caenidae, Coenagrionidae, Dixidae, Hydrosychidae, Libellulidae,

Nepidae, Simuliidae, Turbellaria and Veliidae, these are taxa with moderate sensitivity scores (Dickens and Graham, 2002; Gerber and Gabriel, 2002).

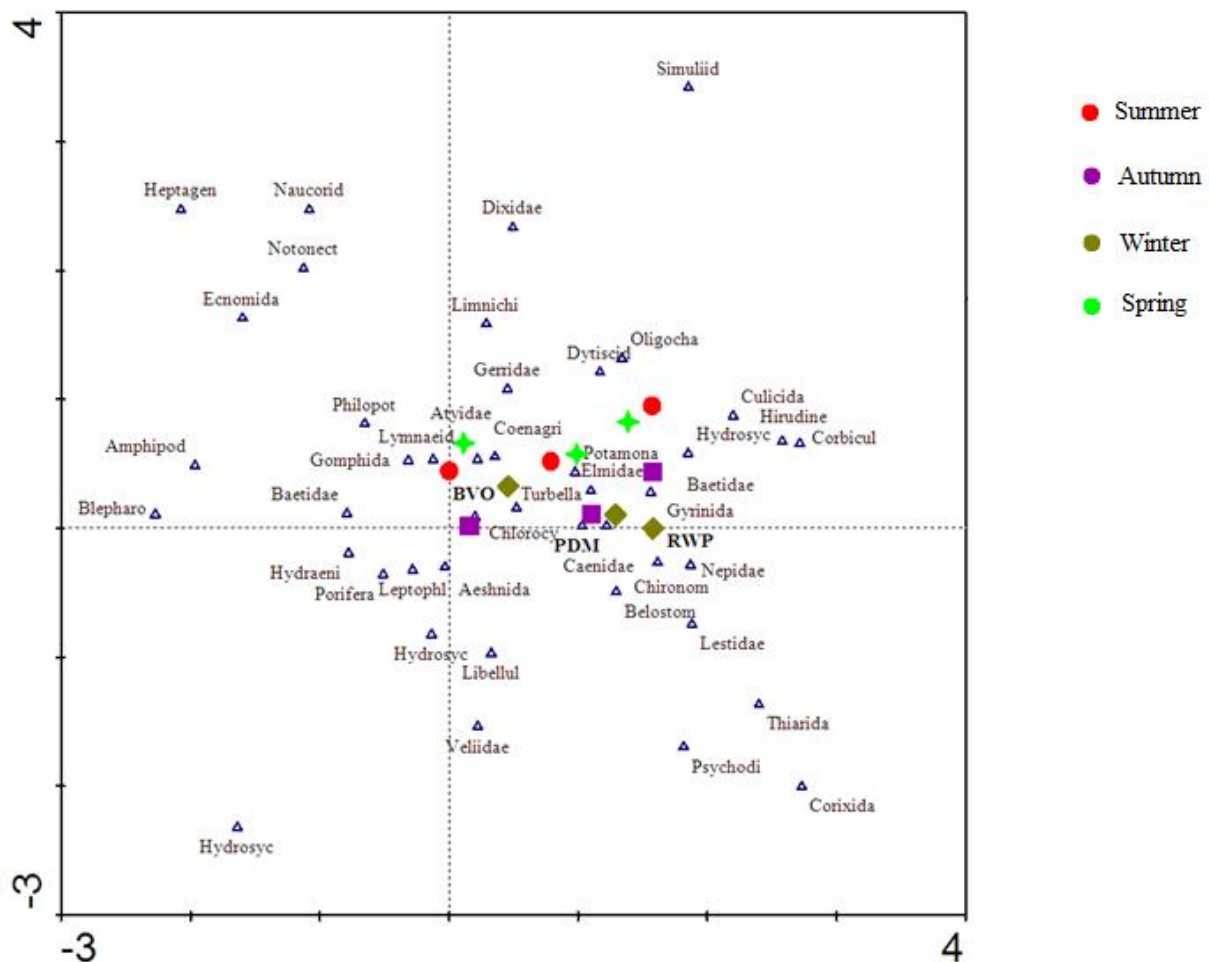
PDM is the site between BVO and RWP, and as expected species found at PDM also occur at BVO as it is not severely polluted. PDM also shares taxa with RWP as well as PDM is polluted and not in a pristine condition. The taxa found at PDM are Atyidae, Belostomatidae, Caenidae, Coenagrionidae, Dixidae, Hydrosychidae, Libellulidae, Nepidae, Potamonautidae, Simuliidae, Turbellaria, Veliidae, Baetidae 1sp and 2sp, Corbiculidae, Culicidae, Chironomidae, Gyrinidae, Hirudinea, Nepidae and Oligochaeta. These taxa range between high and low sensitivity (Dickens and Graham, 2002; Gerber and Gabriel, 2002), and as with the spatial variation in diatoms, a shift in community structure can be observed at PDM.

The cumulative effect of pollutants from Potchefstroom is apparent at RWP, with the occasional sensitive taxa occurring but with most being tolerant to pollution and having relatively low sensitivity scores (Dickens and Graham, 2002; Gerber and Gabriel, 2002). The taxa occurring at the highest frequency are Baetidae 1sp, Corbiculidae, Culicidae, Chironomidae, Gyrinidae, Hirudinea, Nepidae, Oligochaeta and Coenagrionidae. These taxa are as already mentioned are tolerant to pollution, and are the same taxa that in Figure 5-1 occur in water with high levels impact.

Changes in macroinvertebrate community structure are constant from BVO towards RWP. The taxa that occur most often at BVO are Amphipoda, and differ with a negative correlation of about -5 from the taxa at RWP at which only one species of Baetidae is present. The Amphipoda have a sensitivity rating of 13, and 1 species Baetidae has a sensitivity of 4 (Dickens and Graham, 2002; Gerber and Gabriel, 2002).

### 5.2.4.2 Temporal variation of macroinvertebrates in the Mooi River

The temporal variation of macroinvertebrate community structure can be observed in Figure 5-5.



**Figure 5-5: Detrended correspondence analysis scatterplot representing the temporal variation of macroinvertebrate taxa found at each site.**

Figure 5-5 illustrates the temporal variation of macroinvertebrate taxa from each of the studied sites. It becomes apparent from the graph that seasonal changes had an influence on the composition of the macroinvertebrate fauna. Tolerant taxa with low sensitivity scores are located on the right of the graph in the positive quadrants and the sensitive taxa on the left in the negative quadrants. For all three sites, the taxa composition shifts to tolerant, less sensitive taxa in the winter and autumn. This shift corresponds with the changes in SASS5 and ASPT scores which indicate worsening water quality in the dry months, when water levels are low.

The taxa located farthest from the neutral axis are, Corixidae, Ecnomidae, Heptageniidae, Hydrosychidae >2, Naucoridae, Notonectidae, Psychodidae, Simuliidae and Thiaridae. These taxa those that are most affected by seasonal change, they also differ from the taxa found in the

spatial variations non correlating taxa. These taxa are all multivoltine, except Hydrosychidae >2 which is univoltine (USEPA, 2002b).

The temporal variation shows a clear a shift in community structure, however this is expected, as unlike the diatoms, the macroinvertebrates have different life stages depending on taxa. Multivoltine taxa may not occur in the colder months and then a shift in community structure is expected as the seasons change. When one taxon is absent, for breeding or hibernation, another taxon takes its place, until the eggs of the absent taxa are hatched and a change in community structure is seen again (USEPA, 2002b).

# CHAPTER 6 CONCLUSION AND RECOMMENDATIONS

## 6.1 Conclusions

The preceding chapters, gave a description and discussion of the results of physical and chemical water quality variables (Chapter 3) diatom analysis in relation to water quality, indices and community structures (Chapter 4), and macroinvertebrates in relation to water quality, indices and community structures (Chapter 5) of the Mooi River for the 2014 calendar year. Conclusions from each chapter can be drawn and are presented in the paragraphs below, followed by a general conclusion describing the aquatic ecosystem health of the Mooi River.

Chapter 3 highlighted the change in water quality throughout the Mooi River, from its origin and flowing downstream towards the Vaal River. Changes in physical and chemical variables can clearly be observed in the individual figures in Chapter 3. A summary of the variation in water quality variables is presented in Figure 3-10, where data were plotted and classified by site.

The Wonderfontein spruit is the first major source of pollution in the Mooi River and the impact of the mining in the West Rand area is clearly seen in the elevated levels of pollution, after the Wonderfontein spruit joins the Mooi River. The impact of agricultural activity is clearly evident at PDM with higher concentrations of water quality variables measured than at Boskop Dam.

Potchefstroom had the largest contribution of pollution to the Mooi River with various sources of pollution entering the Mooi River. Very high concentrations of measured water quality variables, especially phosphates, nitrate, EC and sulphates, clearly indicate the severity of pollution and impact.

Chapter 4 was the first of two chapters using biotic aquatic ecosystem health in the Mooi River. Diatom species associations to changes in water quality were elucidated using canonical correspondence analysis and showed clear groups and the preferences of the taxa to either good or bad water quality. The preferences matched those described in national and international literature. The diatom indices that were used were the %PTV, GDI, SPI and BDI. All indices, with the exception of the BDI, indicated a decrease in water quality after the Wonderfontein spruit and the indices correlated significantly with water changes in water quality variables. The diatom indices showed significant differences between sites and their related

water quality. Potchefstroom was again identified as the chief source of pollution and had the most impact on ecosystem health.

In terms of spatial variation a clear shift in diatom community structure was observed. Gradual changes were seen in community composition with species favoring good water quality occurring at Bovenste oog, Klerkskraal Dam, upstream of Wonderfonteinspruit and Boskop Dam while species capable of tolerating elevated pollution levels occurred at downstream of the Wonderfonteinspruit tributary, downstream of Potchefstroom Dam, Trompie Kitsgras and Elbririxon. A complete change in community structure between Bovenste oog and Elnrixon was observed, indicating a deterioration in aquatic ecosystem health. There were few temporal changes, the water quality was slightly worse in the winter months but the community structure shifted only slightly in terms of dominant species. Seasonal change does thus not have a significant influence on diatom community structure in this heavily impacted system. Seasonal shifts may be more readily observed in less impacted waters.

Chapter 5 presented the macroinvertebrate data collected in the Mooi River for 2014. The results of this analysis corresponded well to the sensitivity scores assigned to each taxon on the SASS5 score sheet. With the more sensitive taxa residing in the sites with low levels of pollution and the tolerant taxa with low sensitivity scores located at the sites with elevated water quality variables. The indices used were SASS5 and Average Score per Taxa (ASPT), and they were compared to the reference site as described by Dallas (2007). The index scores showed a clear decline in habitat integrity from Bovenste oog to Potchefstroom Dam and lastly to Trompie Kitsgras as the Mooi River flows downstream towards the Vaal River. The macroinvertebrate indices used showed significant differences between the sites and water quality variables. Correlation analysis showed that macroinvertebrate index scores were significantly and inversely correlated to water quality variables. Potchefstroom was the source of pollution that was associated with the highest levels of impact.

The spatial and temporal variations between sites were discussed in the last sub-section of Chapter 5. The taxa with the highest sensitivity scores were located at Bovenste oog. Taxa with moderate sensitivity scores resided at all three sites, and the taxa with low sensitivity scores and high tolerances were located at Potchefstroom Dam and Trompie Kitsgras, with the latter containing the most tolerant taxa. Macroinvertebrates showed changes in community structure in relation to seasonal changes, with multivoltine taxa occurring less in the winter months, and causing community shifts at Bovenste oog and Potchefstroom Dam towards low sensitivity, tolerant taxa that are uni- or semivoltine.

In conclusion, it is clear that water quality and biotic integrity decreases gradually as the Mooi River flows downstream towards the Vaal River. The first noticeable influence is seen at downstream of the Wonderfonteinspruit just after the confluence of the highly polluted Wonderfonteinspruit with the Mooi River. Recovery is observed at Boskop Dam with the impoundment having a clear influence on the reduction of the concentration of pollutants, and thus improving the water quality. The agricultural activity between Boskop Dam and Potchefstroom is clearly evident when assessing all of the results obtained from the study. Potchefstroom and its surrounding activities, has an undeniable and significant influence on the aquatic ecosystem health of the Mooi River, and demonstration of deterioration of biotic integrity greater than Wonderfonteinspruit was shown. The agricultural activity and utilization of the Mooi River from Potchefstroom to Elbrixon allowed little to no improvement in the aquatic ecosystem health, and thus the Mooi River would have a significant deleterious effect on the Vaal River.

To answer the research question posed in Chapter 1: How will the natural and anthropogenic influences on the Mooi River affect the health of the aquatic ecosystem as the river progresses towards the Vaal River? The natural and anthropogenic exerted in the catchment influence the Mooi River extensively. The general aquatic ecosystem health of the Mooi River can still be described as healthy near to the source but as it flows downstream the aquatic ecosystem health decreases to the point at Trompie Kitsgras where it could be described as poor, and critically modified by a variety of pollution sources.

## **6.2 Recommendations**

It is clear that Potchefstroom influences the aquatic ecosystem health negatively, even though Potchefstroom is a relatively small town, with few sources of pollution. The poor quality of the Mooi River at Elbrixon is of concern, as the Mooi River would in turn contribute to the water quality of the Vaal River.

It would be recommended that the present study be expanded to take more water quality variables into account and with the addition of other biotic indicators such as the fish response and health indices to indicate long term change. It would also be useful to conduct bacteriological studies (to reflect the impact on human health) and to include algal studies of the whole periphyton as this will provide a more complete overview of the ecosystem health of the Mooi River. It would also be advisable to extend the study into the tributaries of the Mooi River, to determine the exact point and extent of pollution. An extended study could be also be

conducted in the Vaal River up- and downstream of the confluence with the Mooi River, to assess the influence of the Mooi River on a larger aquatic ecosystem, and to evaluate the extent of the effects.

This study has shown the effectiveness of the use of aquatic biota as indicators of water quality and habitat integrity. The methodologies discussed provide time integrated results and if applied regularly could be used for the identification of the precise sources of pollution and this in turn could help to inform managers in order mitigate the effects of pollutants entering the river systems in South Africa and thus improving water quality for both human use and for the biota dependent on these ecosystems.

## CHAPTER 7 BIBLIOGRAPHY

- Abel P.D. 1989. *Water Pollution Biology*. Ellis Horwood, Chichester. United Kingdom. 232pp.
- Annandale E. and Nealer E. 2011. Exploring aspects of the water history of the Potchefstroom region and the local management of it. *New Contree*. Vol. 62: 111–124
- Ansara-Ross T.M., Wepener V., Van den Brink P.J. and Ross M.J. 2008. Probabilistic risk assessment of the environmental impacts of pesticides in the Crocodile (West) Marico catchment, North West Province. *Water SA*. Vol. 34 No. 5: 637- 644
- Archibald R.E.M. 1972. Diversity in some South African diatom associations and its relation to water quality. *Water Research*. Vol. 6: 1229-1238
- Ashton P.J., Love D., Mahachi H. and Dirks P.H.G.M. 2001. An Overview of the Impact of Mining and Mineral Processing Operations on Water Resources and Water Quality in the Zambezi, Limpopo and Olifants Catchments in Southern Africa. *Contract Report to the Mining, Minerals and Sustainable Development Southern Africa*. Report No. ENV-P-C 2001-042. Xvi: 336
- Augustyn Ł., Kaniuczak J and Stanek-Tarkowska J. 2012. Selected physicochemical and chemical properties of surface water of the Wisłoka used as potable water. *Inżynieria Ekologiczna*. Vol. 26: 7-19
- Azim M. E., Verdegem M. C. J., Van Dam A. A. and Bederidge M. C.M. 2005. *Periphyton Ecology, Exploitation and Management*. CABI Publishing. Cambridge: 352pp.
- Barbour M.T., Gerritsen J., Snyder B.D. and Stribling J.B. 1999. Rapid bioassessment protocols for use in streams and wadeable rivers: Periphyton, benthic macroinvertebrates and fish. *Environmental Protection Agency*. 841-B-99-002
- Barnard S., Venter A. and Van Ginkel C.E. 2013. Overview of the influences of mining-related pollution on the water quality of the Mooi River system's reservoirs, using basic statistical analyses and self organised mapping. *Water SA*. Vol. 39 No. 5: 655-662

- Bate G.C. Adams J.B. and Van der Molen J.S. 2002. Diatoms as indicators of water quality in South African river systems. *Water research commission, Pretoria*. WRC Report No. 814/1/02
- Beasley G. and Kneale P.E. 2003. Investigating the influence of heavy metals on macroinvertebrate assemblages using Partial Canonical Correspondence Analysis (pCCA). *Hydrology and Earth System Sciences*. Vol. 7 No.2: 221–233
- Bere T. 2014. Ecological preferences of benthic diatoms in a tropical river system in São Carlos-SP, Brazil. *Tropical Ecology*. Vol. 55: 47-61.
- Bere T., Chia M.A. Tundisi J.G. 2012. Effects of Cr III and Pb on the bioaccumulation and toxicity of Cd in tropical periphyton communities: implications of pulsed metal exposures. *Environmental Pollution*. Vol. 163: 184-191
- Bermudez-Couso A., Arias-Esteves M., Novoa-Munoz J.C., Lopez Periago E., Soto-Gonzalez B. and Simal-Gandara J. 2007. Seasonal distributions of fungicides in soils and sediments of a small river basin partially devoted to vineyards. *Water Research*. Vol. 31: 4515-4525
- Blewett T.A. and Wood C.M. 2015. Low salinity enhances NI-mediated oxidative stress and sub-lethal toxicity to the green shore crab (*Carcinusmaenas*). *Ecotoxicology and Environmental Safety*. Vol. 122: 159-170
- Bouamra F., Drouiche N., Ahmed D.S. and Lounici H. 2012. Treatment of water loaded with Orthophosphate by electrocoagulation. *Procedia Engineering*. Vol. 33: 155-162
- Braid W. and Ong S.K. 2000. Decomposition of nitrite under various pH and aeration conditions. *Water Air and Soil Pollution*. Vol. 118: 13-26
- Carere M., Miniero R. and Cicero M.R. 2011. Potential effects of climate change on chemical quality of aquatic biota. *Trends in Analytical Chemistry*. Vol. 30 No. 8: 1214-1221
- Castro M.S., Driscoll C.T., Jorden T.E., Reay W.G. and Boynton W.R. 2003. Sources of nitrogen to estuaries in the United States. *Estuaries*. Vol. 26 No. 3: 803-814

- Cemagref B. 1982. *Etude des méthodes biologiques d'appréciation quantitative de la qualité des eaux.* [A study on the biological methods of qualitative assessment of water quality. A report of the Water Quality Division Lyon-Outflow Rhône River section catchment] Rapport Division Qualite des Eaux Lyon-A.F. Bassin Rhône-Méditerranée-Corse, Pierre-Bénite, 218 pp. [in French]
- Chambers P.A., Guy E., Roberts E., Charlton M.N., Kent R., Gannon C., Grove G., Foster N., de Kimpe C. and Giddings. 2003. Nutrients- nitrogen and phosphorus. In threats to sources of drinking water and aquatic ecosystem health in Canada. *NWRI Scientific Assessment Report*. Series No. 1
- Cholnoky B.J. 1958. Hydrobiologische untersuchungen in Transvaal 3. *Hydrobiologia*. Vol 11: 205-266
- Cholnoky B.J. 1960. Beiträge zur Kenntniss der Ökologie der diatomeen in dem Swartkops-Bache nahe Port Elizabeth (Südost-Kaapland). *Hydrobiologia*. Vol. 16: 229-287
- Chutter F.M. 1994. The rapid biological assessment of streams and river water quality by means of macroinvertebrate communities in South Africa. In: Uys M.C. Editor. *Classification of rivers and environmental health indicators*. (pp. 217-234). *Water Research Commission*. WRC Report No. TT 63/94
- Chutter F.M. 1998. Research on the Rapid Biological Assessment of Water Quality Impacts in Streams and Rivers. *Water Research Commission*. Pretoria. WRC Report No 422/ 1/98
- Coats R., Perez-Losada J., Schladow G., Richards R. and Goldman C. 2006. The warming of lake Tahoe. *Climate Change*. Vol. 76: 121-148
- Coetzee H., Winde F., Wade P. 2006. An assessment of sources, pathways, mechanisms and risks of current and potential future pollution of water and sediments in gold Mining areas of the Wonderfontein spruit catchment (Gauteng/North West Province, South Africa). *Water Research Commission*. Pretoria. WRC Report No. 1214/1/06
- Conley D.J., Paerl H.W., Howarth R.W., Boesch D.F., Seitzinger S.P., Havens K.E., Lancelot C. and Likens G.E. 2009. Controlling eutrophication: nitrogen and phosphorus. *Science*. Vol. 323: 1014-1015

- Coste M. and Ayphassorho H. 1991. Etude de la qualité des eaux du bassin Artois Picardie à l'aide des communautés de diatomées benthiques (Application des indices diatomiques), Rapport Cemagref Bordeaux – Agence de l'Eau Artois Picardie. 227 pp. [in French]
- Cowling, R.M., Richardson D.M. and Pierce S.M. 1997. Vegetation of South Africa. Cambridge: Cambridge University Press.
- Currie S.L. 2001. *The implementation of an environmental decision-making support system: the Mooi River Catchment as a case study*. PhD thesis, Potchefstroom University for Christian Higher Education. 152 pp.
- Dallas H. 2009. The effect of water temperature on aquatic organisms: A review of knowledge and methods for assessing biotic responses to temperature. *Water Research Commission*. Pretoria. WRC Project No. K8/690
- Dallas H. 2007. River Health Programme: South African Scoring System (SASS) data interpretation guidelines. A report by the Freshwater Consulting Group? Freshwater Research Unit. *University of Capetown and Department of Water Affairs and Forestry*. Cape Town. 85pp.
- Dallas H. F. and Day J.A. 2004. The effect of water quality variables on aquatic ecosystems. *Water Research Commission*. Pretoria. WRC Report No. TT 224/04
- Dallas H. F. and Day J.A. 1993. The effect of water quality variables on riverine ecosystems: A review. *Water Research Commission*. Pretoria. WRC Report No. TT 61/93
- Deegan L.A. and Buchsbaum R.N. 2005. The effect of habitat loss and degradation on fisheries. In: Buchsbaum R., Pederson J. and Robinson W.E. Editors: *The decline on fisheries resources in New England: evaluating the impact of overfishing, contamination, and habitat degradation*. Cambridge. *MIT Sea Grant College Program*. Publication No. MITSG 05-5.: 67-96
- Department of Water Affairs and Forestry (DWAF). 1996. The Department of Water Affairs and Forestry. South African Water Quality Guidelines: Volume 7-Aquatic ecosystems.

- de Sousa D.N.R., Mozeto A.A., Carneiro R.L. and Fadini P.S. 2014. Electrical conductivity and emerging contaminant as markers of surface freshwater contamination by wastewater. *Science of the Total Environment*. Vol. 484: 19–26
- Dickens C.W.S. and Graham M. 2002. The South African Scoring System (SASS) Version 5 rapid bio-assessment method for rivers. *African Journal of Aquatic Science*. Vol. 27: 1-10
- de la Ray P.A., Taylor J.C., Laas A., van Rensburg L. and Vosloo A. 2004. Determining the possible application value of diatoms as indicators of general water quality: A comparison with SASS 5. *Water SA*. Vol. 30 No. 3: 325-332
- Doan P.T.K., Némery J., Schmid M. and Gratiot N. 2015. Eutrophication of turbid tropical reservoirs: Scenarios of evolution of the reservoir of Cointzio, Mexico. *Ecological Informatics*. Vol. 29: 192-205
- Dodds W.K. 2002. Freshwater Ecology. Concepts and Environmental Applications. *Academic Press*. USA. 821pp.
- Dudgeon D. 2005. River management for conservation of fish biodiversity in Monsoonal Asia. *Ecology and Society*. Vol. 10(2):15
- Dudgeon D., Arthington A.H., Gessner M.O., Kawabata Z., Knowler D.J., Leveque C., Naiman R.J., Prieur-Richard A., Soto D., Stiassny M.L.J. and Sullivan C.A. 2006. Freshwater biodiversity: importance threats, status and conservation challenges. *Biology Review*. Vol. 81: 163-182
- Duong T., Coste M., Feurtet-Mazel A., Dang D., Gold C., Park Y. and Boudou A. 2006. Impact of urban pollution from the Hanoi area on benthic diatom communities collected from the Red, Nhue and Tolich Rivers (Vietnam). *Hydrobiologia*. Vol. 563: 201-216
- Epstein E. 2002. Land applications of sewage sludge and biosolids. Boca Raton (FL): *CRC Press*: 216 pp.
- Falkowski P.G. and Raven A.J. 1997. Aquatic photosynthesis, *Blackwell science*. Massachusetts. USA: 375pp.

- Finkenbine J. F., Atwater J. W. and Mavinic D. S. 2000. Stream health after urbanization. *Journal of the American Water Resources Association*. Vol. 36: 1149-1160
- Flores-Alsina X., Christian Kazadi Mbamba C.M., Solon K., Vrecko D., Tait S., Batstone D.J., Jeppsson U. and Gernaey K.V. 2015. A plant-wide aqueous phase chemistry module describing pH variations and ion speciation/pairing in wastewater treatment process models. *Water Research*. Vol. 85: 255-265
- Freeland J. A., Richardson J.L. and Foss L.A. 1999. Soil indicators of agricultural impacts on Northern Prairie wetlands: Cottonwood Lake research area, North Dakota, USA. *Wetlands*. Vol. 19 No. 1: 56-64
- Fukushima S., Koichi Y and Fukushima H. 1994. Effects of self-purification on periphytic algal communities. *Ver Int Verein Theor Angewandte Limnology*. Vol. 25: 1966- 1970
- Gabaldon T. and Pittis A.A. 2015. Origin and evolution of metabolic sub-cellular compartmentalization in eukaryotes. *Biochimie*. Vol. xxx (Article in Press): 1-7
- Gerber A. and Gabriel M.J.M. 2002. Aquatic Invertebrates of South African Rivers- Version 1. *Department of Water affairs and Forestry*. Pretoria. 150pp.
- Gold, C., Feurte-Mazel A., Coste M. and Boudou A. 2003. Effects of cadmium stress on periphytic diatom communities in indoor artificial streams. *Freshwater Biology*. Vol. 48: 316-328
- Gordan A.K., Griffin N.J. and Palmer C.G. 2015. The relationship between concurrently measured SASS (South African Scoring System) and turbidity data archived in the South African River Health Programme's Rivers Database. *Water SA*. Vol. 41 No. 1: 21-26
- Grab S. 2014. Spatio-temporal attributes of water temperature and macroinvertebrate assemblages in the headwaters of the Bushmans River, southern Drakensberg. *Water SA*. Vol. 40 No. 1: 19-26

- Green E.K. and Galatowitsch S.M. 2002. Effects of *Phalaris arundinacea* and nitrate- N addition on the establishment of wetland plant communities. *Journal Applied Ecology*. Vol. 39: 134-144
- Grimm N. B., Grove J.M., Pickett S.T.A and Redman C.L. 2000. Integrated approaches to long-term studies of urban ecological systems. *BioScience*. Vol. 50: 571–584.
- Harding W.R., Archibald C.G.M. and Taylor J.C. 2005. The relevance of diatoms for water quality assessments in South Africa: A position paper. *Water SA*. Vol 30 No. 1: 41-46
- Hartshorn N., Marimon Z., Xuan Z., Cormie J., Chang N. and Wanielista M. 2016. Complex interactions among nutrients, chlorophyll-a, and microcystins in three stormwater wet detention basins with floating treatment wetlands. *Chemosphere*. Vol. 144: 408-419
- Hasle G.R. 1978. Some specific preparations: diatoms. In: *Phytoplankton Manual*. Sournia A. Editor. *United Nations Educational, Scientific and Cultural Organisation (UNESCO)*. Paris.
- Helfrich L.A., Weigmann D.L., Hipkins P. and Stinson E.R. 2009. Pesticides and Aquatic Animals: A Guide to Reducing Impacts on Aquatic Systems. *College of Agriculture and Life Sciences, Virginia Polytechnic Institute and State University*. Publication: 420-013
- Hem J.D., 1989. The Study and Interpretation of the Chemical Characteristics of Natural Water, third ed. *United States Geology. Survey Water-Supply*. Paper: 2254
- Henderson-Sellers B. 1991. *Water quality modelling, Decision support techniques for lakes and reservoirs*. Volume 4. CRC Press. United States. Boston.
- Hirst H., Juttner I. and Ormerod S.J. 2002. Comparing the responses of diatoms and macroinvertebrates to metals in upland streams of Wales and Maizewall. *Freshwater Biology*. Vol. 47: 1752–1765
- Houghton J., Ding Y., Griggs D., Noguer M., Van Der Linden P., Dai X., Maskell K. and Johnson C. 2001. Climate change 2001: the scientific basis. *Cambridge University Press*, Cambridge.
- Hughes L. 2000. Biological consequences of global warming: is the signal already apparent? *Trends in Ecology and Evolution*. Vol. 15: 56-61.

- Hutchinson G.E., Pickford G.E. and Schuurman J.F.M. 1932. A contribution to the hydrobiology of pans and other inland waters of South Africa. *Hydrobiologia*. Vol. 24: 1-154
- Hynes H.B.N. 1970. *The Ecology of Running Waters*. Liverpool University Press. England. 555pp.
- Inhaber H. 1976. Environmental Indices. John Wiley & Sons, New York, New York.
- Janse van Vuuren S., Taylor J.C., Gerber A. and van Ginkel C. 2006. Easy identification of the most common Freshwater Algae. *North-West University and Department of Water Affairs and Forestry*. Potchefstroom. 35-85
- Jimoh A.A., Clarke E.O., Olusegun W.O. and Haleemah B.A. 2011. Food and feeding habits of the African river prawn (*Macrobrachium vollenhovenii*, Herklots, 1857) in Epe Lagoon, Southwest Nigeria. *International Journal of Fish Aquaculture*. Vol. 3 No. 1:10-15
- Karr J.R. and Chu E.W. 1999. Restoring life in running waters: better biological monitoring. *Island Press*, Washington, D.C. pp. 220
- Kelly M.G. and Whitton B.A. 1995. The trophic Diatom Index: a new index for monitoring eutrophication in rivers. *Journal of Applied Phycology*. Vol 7: 433-444
- Kelly M.G. 2002. Role of benthic diatoms in the implementation of the Urban Wastewater Treatment Directive in the River Wear, North-East England. *Journal of Applied Phycology*. Vol. 14: 9-18
- Kelly V.J. 2001. Influence of reservoirs on solute transport: a regional-scale approach. *Hydrological Processes*. Vol.15: 1227-1249
- Kim T.H. and Jeong J.H. 2016. Evaluation method of iodine re-evolution from an in-containment water pool after a loss of coolant accident, Part I: pH estimation of a solution with various chemicals. *Annals of Nuclear Energy*. Vol. 87: 324–330

- Kleynhans C.J., Thirion C. and Moolman J. 2005 A level 1 River Ecoregion classification System for South Africa. Lesotho and Swaziland. *Department of Water Affairs and Forestry*. South Africa. Report No. N/0000/00/REQ0104
- Koekemoer S. and Taylor J.C. 2010. Rapid riverine assessment method for level 3 ecological water resource monitoring. Supporting Document for the Rapid Habitat Assessment Method (RHAM) Manual. *Department of Water Affairs and Forestry*. South Africa. Report No. RDM/NAT/00/CON/0707
- Kriel G.P. 2008. Urban water quality monitored, waste water pollution management. *Urban Green File*. Vol. 13: 30-33
- Kruskal, W. H. and Wallis W. A. 1952. Use of ranks in one-criterion variance analysis. *Journal American Statistic Association*. Vol. 47: 583-621.
- LaBonte J.R., Scott D.W., McIver J.D. and Hayes J.L. 2001. Threatened, endangered and sensitive insects in Eastern Oregon and Washington forests and adjacent lands. *Northwest Science*. Vol.75: 185-198
- Labuschagne P. 2007. *Update plume delineation and liability estimation for Vaal River*. Groundwater Consulting Service (PTY) LTD. Report to AngloGold Ashanti.
- Ladd J.N., Oades J.M. and Amato M. 1981. Microbial biomass formed from  $^{14}\text{C}$ ,  $^{15}\text{N}$ -labelled plant material decomposing in soils in the field. *Soil Biology and Biochemistry*. Vol. 13 No. 2: 119-126
- Lenoir A. and Coste M. 1996. Development of a practical diatom index of overall water quality applicable to the French National Water Board network. In Whitton B.A. and Rott E. Editors. *Use of Algae for Monitoring River II*. Universität Innsbruck: 29-45
- Leopold L.D. 1968. Hydrology for urban planning? a guidebook on the hydrologic effects of urban land use. *USGS Circular*. 554pp.

- Li Y.F., Wan W.F., Song J., Wu Y., Xu Y. and Zhang M. 2009. Classification of groundwater contamination in Yuxi river valley, Shaanxi Province, China. *Bulletin of Environmental Contamination and Toxicology*. Vol.82: 234-238
- Lobo E. A., Callegaro V. L., Hermany G., Bes D., Wetzel C. E. and Oliveira M.A. 2004. Use of epilithic diatoms as bioindicator from lotic systems in southern Brazil, with special emphasis on eutrophication. *Acta Limnologia Brasiliense*. Vol. 16: 25-40
- Low A.B. and Rebelo A.B. 1998. *Vegetation of South Africa, Lesotho and Swaziland*. Department of Environmental Affairs and Tourism, Pretoria. 2nd ed.
- Mallett V. 1999. After Hours- Journey to the center of the earth. Business Day. July 16.
- Mantel S.K., Muller N.W.J. and Hughes D.A. 2010. Ecological impacts of small dams on South African rivers Part 2: Biotic response – abundance and composition of macroinvertebrate communities. *Water SA*. Vol. 36 No. 3: 361-370
- Mantyla A.W., Bograd S.J. and Venric E.L. 2008. Patterns and controls of chlorophyll-*a* and primary productivity cycles in the Southern California Bight. *Journal of Marine Systems*. Vol. 73: 48-60
- Marandi A., Polikarpus M and Jöeleht A. 2013. A new approach for describing the relationship between electrical conductivity and major anion concentration in natural waters. *Applied Geochemistry*. Vol. 38: 103-109
- Masamba K., Li Y., Hategekimana J., Ma J. and Zhong F. 2016. Effect of drying temperature and pH alteration on mechanical and water barrier properties of transglutaminase cross linked zein-oleic acid composite films. *Food Science and Technology*. Vol. 65: 518-531
- Maybeck M. 2003 Global occurrence of major elements in rivers. In: Drever J.I. Editor. *Surface and ground water, weathering and soils*. Elsevier-Pergamon. 207–24 pp.
- McBride. 1988. Preparing random distribution of diatom valves on microscope slides. *Limnological Oceanography*. Vol. 33: 1627-1629

- McCormick P.V. and Cairns J. 1994. Algae indicators of environmental change. *Journal of Applied Phycology*. Vol. 6: 509-526
- Medley C.N. and Clements W.H. 1999. Responses of diatom communities to heavy metals in streams: The influence of longitudinal variation. *Ecology Applications*. Vol. 8:631–644
- Meyer J.L., Paul M.J. and Taulbee W.K. 2005. Stream ecosystem functions in urbanizing landscapes. *Journal of the North American Benthological Society*. Vol. 24: 602-612
- Mestre A.S., Nabico A., Figueiredo P.L., Pinto M.L., Santos M.S.C.S. and Fonseca I.M. 2015. Enhanced clofibric acid removal by activated carbons: water hardness as a key parameter, *Chemical Engineering Journal*. Vol .XXX: XX-XX
- Moustafa A., Beszteri B., Maier U.G., Bowler C., Valentin K. and Bhattacharya D. 2009. Genomic footprints of a cryptic plastidendosymbiosis in diatoms. *Science*. Vol. 324:1724-1726
- Nelson D.M., Treguer P., Brzezinski M.A., Leynaert A. and Queguiner B. 1995. Production and dissolution of biogenic silica in the ocean-revised global estimates, comparison with regional data and relationship to biogenic sedimentation. *Global Biogeochemistry Cycle*. Vol. 9: 359-372
- Nel J., Maree G., Roux D., Moolman J., Kleynhans N., Silberbauer M. and Driver A. 2004. South African National Spatial Biodiversity Assessment 2004: Technical 40 Report. Vol. 2: River Component. *Council for Scientific and Industrial Research*. CSIR Report No. ENV-S-I-2004-063
- Newete S.W., Erasmus B.F.N., Weiersbye I.L. and Byrne M.J. 2014. The effect of water pollution on biological control of water hyacinth. *Biological Control*. Vol. 79: 101–109
- Nilsson C., Jansson R., Malmqvist R. and Naima R.J. 2007. Restoring riverine landscapes: the challenge of identifying priorities, reference states and techniques. *Ecology and Society*. Vol. 12 No. 1:16
- O’Keeffe J. and Dickens C. 2000. Aquatic invertebrates. In King J.M., Tharme R.E. and deVilliers M.S. Editors. *Environmental Flow Assessments for Rivers: Manual for the Building Block Methodology*. *Water Research Commission*. WRC Report No. 576/1/98. pp. 231-244

- Odum E.P. 1985. Trends expected in stressed ecosystems. *Bioscience*. Vol. 35 No.7: 419-422.
- Ohl C., Krauze K. and Grünbühel C. 2007. Towards an understanding of long-term ecosystem dynamics by merging socio-economic and environmental research Criteria for long-term socio-ecological research sites selection. *Ecological Economics*. Vol. 63: 383-391
- Pan, Y., Stevenson R.J., Hill B.H., Herlihy A.T. and Collins G.B. 1996. Using diatoms as indicators of ecological conditions in lotic systems: a regional assessment. *Journal of North American Benthological Society*. Vol. 15: 481-495
- Paerl H.W., Fulton R.S., Moisander P.H. and Dyble J. 2001. Harmful freshwater algal blooms, with an emphasis on cyanobacteria. *The Scientific World Journal*. Vol 1: 76-113
- Paerl H.W., Xu H., McCarthy M.J., Zhu G.W., Qin B.Q., Li Y.P. and Gardner W.S. 2011. Controlling harmful cyanobacterial blooms in a hyper-eutrophic lake (Lake Taihu, China): the need for a dual nutrient (N & P) management strategy. *Water Research*. Vol. 45: 1973-1983
- Potapova, M. G. and D. F. Charles. 2003. Distribution of benthic diatoms in U.S. rivers in relation to conductivity and ionic composition. *Freshwater Biology*. Vol: 48: 1311-1328
- Potasznik A. and Szymczyk S. 2015. Magnesium and Calcium concentrations in the surface water and bottom deposits of a river-lake system. *Journal of Elementology*. Vol. 20 No. 3: 677-692
- Psarra S., Zohary T., Krom M.D., Mantoura R.F.C., Polychronaki T., Stambler N., Tanaka T., Tselepides A. and Thingstad T.F. 2005. Phytoplankton response to a Lagrangian phosphate addition in the Levantine Sea (Eastern Mediterranean). *Deep-Sea Research*. Part II. Vol. 52: 2944-2960
- Pulles W., Howie D., Otto D. and Easton J. 1996. A Manual on Mine Water Treatment and Management Practices in South Africa. *Water Research Commission*. Pretoria. WRC Report No.527/1/96

- Riedel K. 2003. *The establishment of a catchment management plan for the Mooi River (Kromdraai) catchment, North West Province, South Africa*. M.Sc. dissertation, University of Pretoria, Pretoria.
- River Health Programme (RHP). 2005. State of rivers Report: Monitoring and managing the ecological state of rivers in the Crocodile (West) Marico Water Management Area. *Department of Environmental affairs and Tourism (DEAT)*. Pretoria. South Africa
- Rosenberg D.M. and Resh V.H. 1993. *Freshwater biomonitoring and benthic macroinvertebrates*. Chapman and Hall. USA. pp. 488
- Rotter S., Sans-pichè F., Streck G. and Schummit-Jans G. 2011. Active bio-monitoring of contamination in aquatic systems-An in situ translocation experiment applying the Pitch concept. *Aquatic topology*. Vol. 101 No.1: 228-236
- Round F.E. 1991. Diatoms in river water-monitoring studies. *Journal of Applied Phycology*. Vol. 3: 129-145
- Round F.E., Crawford R.M., Mann D.G. 1990. The Diatoms. Biology and Morphology of the Genera. *Cambridge University Press*. Cambridge. pp 747
- Roux D.J. 1999. Design of a national programme for monitoring and assessing the health of aquatic ecosystems, with specific reference to the South African river health programme. *Environmental science forum*. Vol. 96: 13-32
- Saunders K.M., Hodgson D.A. and McMinn A. 2009. Quantitative relationships between benthic diatom assemblages and water chemistry in Macquarie Island lakes and their potential for reconstructing past environmental changes. *Antarctic Science*. Vol. 21 No.1: 35–49
- Schoeman F.R. 1976. Diatom indicator groups in the assessment of water quality in the Jukskei-Crocodile river system (Transvaal, Republic of South Africa). *Journal of the Limnological Society of Southern Africa* .Vol. 2 No. 1: 21-24

- Schreiner B. and Van Koppen B. 2002. Catchment management agencies for poverty eradication in South Africa. *Physical Chemistry of the Earth*. Vol. 27: 969-976
- Smil V. 1999. Nitrogen in crop production. *Global Biogeochemistry*. Vol. 13: 647-662.
- Snaddon K. 2009. Spatial and temporal changes in the riverine macroinvertebrate community composition in the Berg River, and the expected response following the development of the Berg River Dam. *Transactions of the Royal Society of South Africa*. Vol. 64 No.2: 119-141
- Sonneman, J. A., Walsh C.J., Breen P.F. and Sharpe A.K. 2001. Effects of urbanization on streams of the Melbourne region, Victoria, Australia. II. Benthic diatom communities. *Freshwater Biology*. Vol. 46: 553–565
- South African Weather Service. 2015a. What are the temperature, rainfall and wind extremes in SA? <http://www.weathersa.co.za/learning/climate-questions/39-what-are-the-temperature-rainfall-and-wind-extremes-in-sa>. Date of access: 26 July 2015
- South African Weather Service. 2015b. Historical Rain Maps. <http://www.weathersa.co.za/climate/historical-rain-maps>. Date of access: 16 February 2015
- Steyn D.J., Toerien D.F. and Visser J.H. 1975. Eutrophication levels of some South African Impoundments. Part 2. Hartebeespoort Dam. *Water SA*. Vol. 1 No. 3: 93-101
- Tanaka A., de Martino A., Amato A., Montsant A., Mathieu B., Rostaing P., Tirichine L. and Bowler C. 2015. Ultrastructure and Membrane Traffic during Cell Division in the Marine Pennate Diatom *Phaeodactylum trimaizeutum*. *Protist*. Vol. 166:506-521
- Taylor J.C. 2004. The application of diatom-based pollution indices in the Vaal catchment. Unpublished M.Sc. Dissertation. North-West University, Potchefstroom Campus. Potchefstroom. 121pp.
- Taylor J.C., de la Ray A. and van Rensburg L. 2005. Recommendations for the collection, preparation and enumeration of diatoms from riverine habitats for water quality monitoring in South Africa. *African Journal of Aquatic Science*. Vol. 30 No.1: 65-75

- Taylor J.C., Janse van Vuuren M.S. and Pieterse A.J.H. 2007a. The application and testing of diatom based indices in the Vaal and Wilge Rivers, South Africa. *Water SA*. Vol 33:-60
- Taylor J.C., Harding W.R. and Archibald C.G.M. 2007b. An illustrated guide to some common Diatom species from South Africa. *Water Research Commission*. Pretoria. WRC Report No.TT282/07
- Taylor J.C., Prygiel J., Vosloo A., de la Ray P.A. and van Rensburg L. 2007c. Can diatom-based pollution indices be used for biomonitoring in South Africa? A case study of the Crocodile West and Marico water management area. *Hydrobiologia*. Vol 592: 455-464
- Taylor J.C., Pope N. and van Rensburg L. 2009. Community participation in river monitoring using diatoms: A case study from the Buffelspoort Valley Conservancy. *African Journal of Aquatic Science*. Vol. 32 No. 2: 173-182
- ter Braak C.J.F. 1986. Canonical Correspondence Analysis: a new eigenvector technique for multivariate direct gradient analysis. *Ecology*. Vol. 67 No. 5: 1167-1179
- Tilman D., Kilham S.S. and Kilham P. 1982. Phytoplankton community ecology: The role of limiting nutrients. *Annual Review of Ecological Systems*. Vol. 13: 349-372
- Troise F.L., Todd D.K. and Leeden F.V.D. 1990. The water encyclopedia. 2nd edition. *Lewis Publishers*. United States of America. 824 pp.
- United Nations Environment Programme Global Environment Monitoring System Water Programme Office. 2008. Water Quality for Ecosystem and Human Health, 2nd Edition. *United Nations Environment Programme Global Environment Monitoring System (GEMS)/Water Programme*. Burlington. Ontario. L7R 4A6 CANADA. ISBN 92-95039-51-7: 120 pp.
- United States Environmental Protection Agency (USEPA). 2002a. Summary of biological assessment programs and biocriteria development for States, Tribes, Territories and interstate commissions: Streams and wadeable rivers. *United States Environmental Protection Agency*. EPA: 822-R-02-048

- United States Environmental Protection Agency (USEPA). 2002b. Methods for evaluating wetland condition: Developing an invertebrate index of biological integrity for wetlands. Office of Water. *United States Environmental Protection Agency*. EPA: 822-R-02-019
- Uwem G.U., Albert P.E. and George E. 2011. Food and feeding habits of *Ophiocephalus obscura* (African snakehead) in the Cross River estuary, Cross River State, Nigeria. *International Journal of Fish Aquaculture*. Vol. 3 No. 13: 231-238
- Van Aardt W.J. and Erdmann R. 2004. Heavy metals (Cd, Pb, Cu, Zn) in mudfish and sediments from three hard-water dams of the Mooi River catchment, South Africa. *Water SA*. Vol. 30 No. 2: 211- 218
- Van Dam H., Mertens A. and Sinkeldam J. 1994. A coded checklist and ecological indicator values of freshwater diatoms from the Netherlands. *Netherlands Journal of Aquatic Ecology*. Vol. 28 No. 1: 117-133
- van der Walt L.J., Winde F. and Nell B. 2002. Integrated catchment management: The Mooi River (North West Province, South Africa) as a case study. *Cuadernos de Investigación Geográfica*. Vol. 28: 109-126
- Venter A., Barnard S., Dickinson M.A., Janse van Vuuren S., Levanets A. and Taylor J.C. 2013. Planktonic algae and cyanoprokaryotes as indicators of ecosystem quality in the Mooi River system in the North-West Province, South Africa. *Water SA*. Vol. 39 No. 5: 707-720
- Vinson M.R. and Hawkins C.P. 1998. Biodiversity of stream insects: variation at local, basin and regional scales. *Annual Review of Entomology*. Vol. 43: 193-271
- Walker C.E. and Pan Y. 2006. Using diatom assemblages to assess urban stream conditions. *Hydrobiologia*. Vol. 561: 179-789
- Wang Z., Meador J.P. and Leung K.M.Y. 2016. Metal toxicity to freshwater organisms as a function of pH: A meta-analysis. *Chemosphere*. Vol. 144: 1544-1552

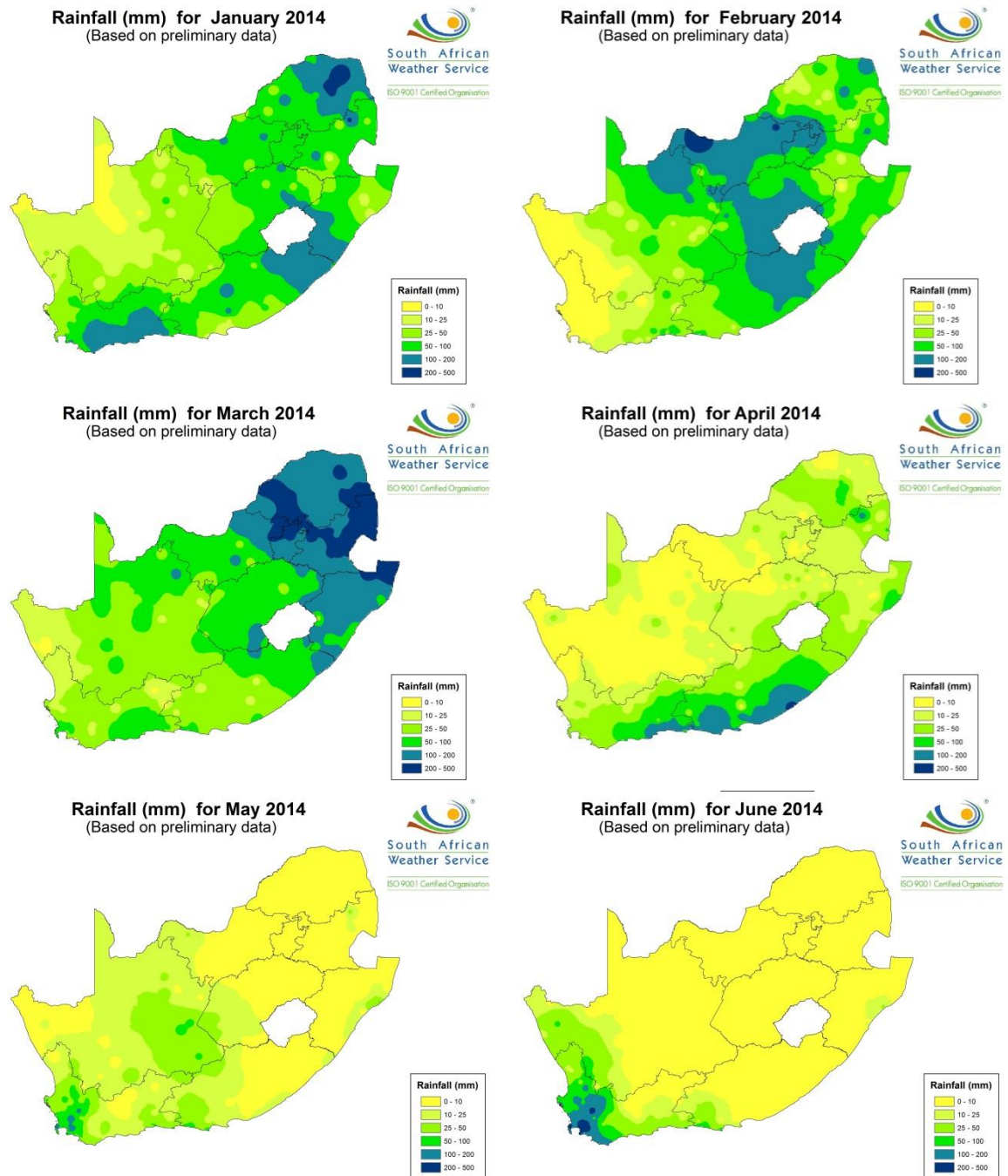
- Ward S., Arthington A.H. and Pusey B.J. 2002. The effects of a chronic application of chlorpyrifos on macroinvertebrate fauna in an outdoor artificial stream system: Species responses. *Ecotoxicology and environmental safety*. Vol. 30 No. 1: 2-23
- Williams E.L., Szramek J.J., Ku C.W.T. and Walter L.M. 2007. The carbonate system geochemistry of shallow groundwater–surface water systems in temperate glaciated watersheds (Michigan, USA): Significance of open-system dolomite weathering. *Geological Society of America*. Vol 119 No. 5/6: 515-528
- Williams W.D. 1987. Salinisation of rivers and streams: an important environmental hazard. *Ambio*. Vol. 16: 180-185
- Williams W.D. 2001. Anthropogenic salinisation of inland waters. *Hydrobiologia*. Vol. 466: 329-337
- Winde F. 2010a. Uranium pollution of the Wonderfonteinspruit, 1997-2008 Part 1: Uranium toxicity, regional background and mining-related sources of uranium pollution. *Water SA*. Vol. 36 No. 3: 239-256
- Winde F. 2010b. Uranium pollution of the Wonderfonteinspruit, 1997-2008 Part 2: Uranium in water, concentrations, loads and associated risks. *Water SA*. Vol. 36 No. 3: 257-278
- Wimberley F.R. and Coleman T.J. 1992. The effect of different urban development types on storm-water runoff quality: A comparison between two Johannesburg catchments. *Water SA*. Vol. 19 No. 4: 325-330
- Winterbourn MJ, Rounick J.S. and Cowie B. 1981. Are New Zealand stream ecosystems really different? *New Zealand Journal of Marine and Freshwater Research*. Vol. 15: 321-328
- Wittmann G.T.W. and Förstner U. 1977. Heavy metal enrichment in mine drainage: III. The Klerksdorp, West Wits and Evander Goldfields. *South African Journal of Science*. Vol. 73: 53-57
- Wolmarans C.T., Kemp M., de Kock K.N., Roets W., van Rensburg L. and Quinn L. 2014. A semi-quantitative survey of macroinvertebrates at selected sites to evaluate the ecosystem health of the Olifants River. *Water SA*. Vol. 40 No. 2: 245-254

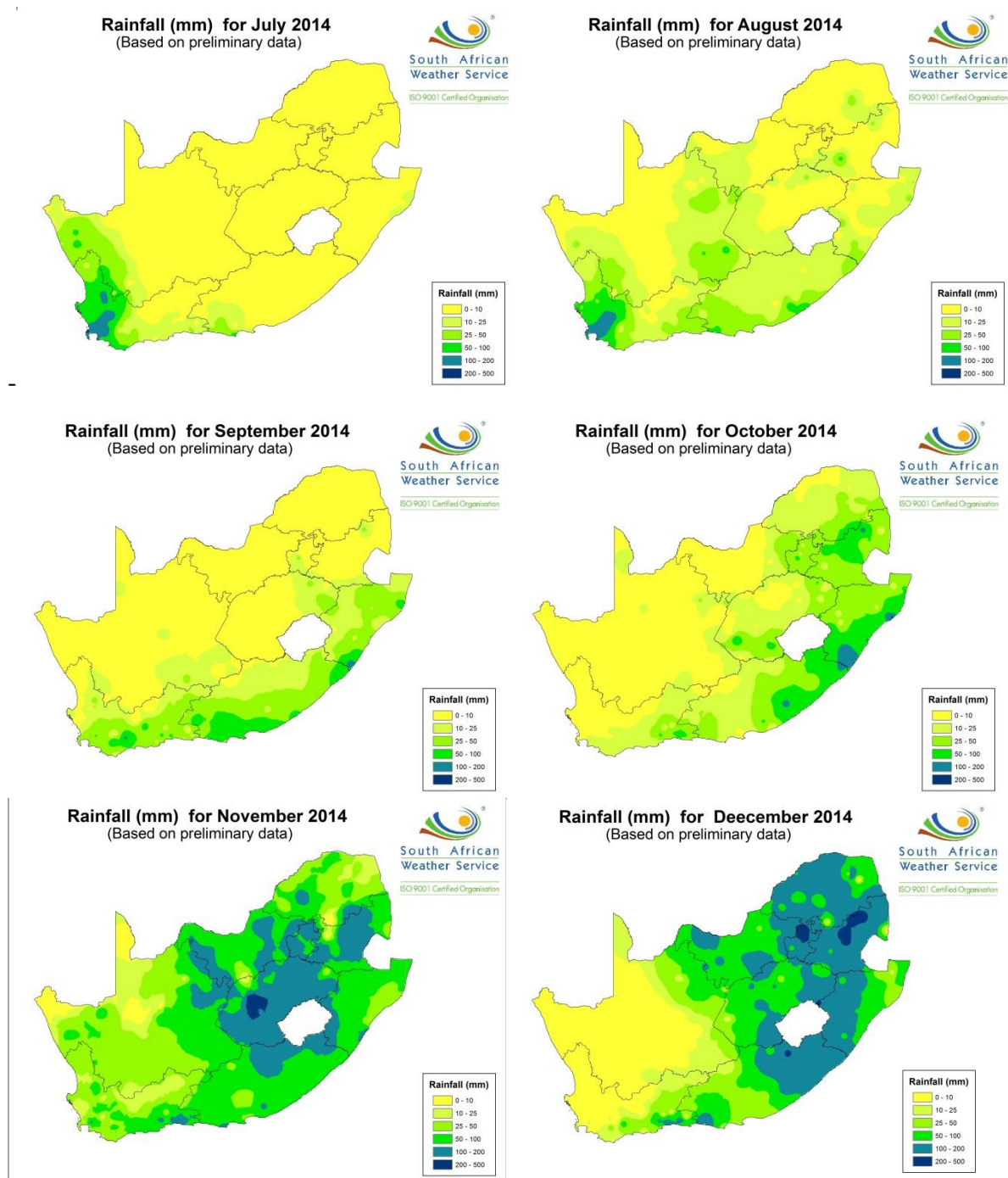
- Xue D., Botte J., De Baets B., Accor F., Nestler A., Taylor P., Van Cleemput O., Berglund M. and Boeckx P. 2009. Present limitations and future prospects of stable isotope methods for nitrate source identification in surface and groundwater. *Water Research*. Vol. 43: 1159-1170
- Yang B., Patsavas M.C., Byrne R.H. and Ma J. 2014. Seawater pH measurements in the field: A DIY photometer with 0.01 unit pH accuracy. *Marine Chemistry*. Vol 160: 75–81
- Yu H., Sun M., Zhang K., Zhu H., Liu Z., Zhang Y., Zhao J., Wu L., Zhang Z and Wang S. 2015. A reversible near-infrared pH probes for optical measurements of pH in complete water system and living cells. *Sensors and Actuators B*. Vol. 219: 294–300
- Zang Y., Wu Z., Liu M., He J., Shi K., Zhou Y., Wang M. and Liu X. 2015. Dissolved oxygen stratification and response to thermal structure and long-term climate change in a large and deep subtropical reservoir (Lake Qiandaohu, China). *Water Research*. Vol. 75: 249-258

# ANNEXURES

## Appendix A

### Rain fall maps of 2014





**Figure A- 1: The figures above represents the rainfall of every month for the 2014 calendar year (South African Weather Service, 2015b)**

## Appendix B

### Water quality variables

**Table A- 1: Table containing the raw water quality data collected in the Mooi River for the 2014 calendar year**

Date	Site	Dis Ca	Dis Mg	NO <sub>3</sub> & NO <sub>2</sub>	PO <sub>4</sub>	SO <sub>4</sub>	Chlor <i>a</i>	Temp	pH	EC ( mS/m)	TDS	Turbidity (NTU)
JAN	BVO	49	28		0.025	2.5	4.9	19.7	6.95	48.9	317	0.32
FEB	BVO	46	27	1.2	0.025	13	0.4	19.6	7	48	310	0.1
MRT	BVO	47	28	1.3	0.025	20	1.6	19.6	7.19	120	781	0.2
APR	BVO	46	30	1.5	0.025	2.5	4.9	20.4	7.1	43	312	0.3
MAY	BVO	40	16	1.5	0.025	2.5	3.8	9.6	7.2	43	319	0.2
JUN	BVO	43	24	1.5	0.025	2.5	2.5	20.3	7.2	45	325	0.3
JUL	BVO	47	28	1.7	0.025	2.5	5.8	19.2	7.2	46	332	0.45
AUG	BVO	40	23	1.6	0.025	2.5	3.6	20.1	7.2	42	305	0.28
SEP	BVO	46	27	1.6	0.025	2.5	2.8	20.4	7.1	43	312	0.19
OCT	BVO	43	29	1.5	0.025	5.5	7.5	20.1	7	42	311	0.1
NOV	BVO	35	22	1.5	0.025	2.5	0.5	21.3	7.2	43	313	0.8
DEC	BVO	40	23	1.6	0.07	2.5	0.6	20.1	7.1	43	313	0.19
JAN	KKD	30	34		0.025	2.5	81	21.3	7.01	42	275	1.42
FEB	KKD	36	24	0.25	0.025	15	7.8	22.7	7.41	38	245	0.82
MRT	KKD	35	22	0.25	0.025	7.1	1.9	21.4	7.49	99	642	1.67
APR	KKD	40	27	0.25	0.025	<5	3.8	21.1	7.5	98	286	1.86
MAY	KKD	64	50	0.25	0.025	2.5	5.2	10	8.2	152	385	1.46
JUN	KKD	42	26	0.25	0.025	7	2.8	6.11	7.45	48	343	1.92
JUL	KKD	47	32	0.25	0.025	2.5	202	11.7	7.9	46	334	1.27
AUG	KKD	43	25	0.25	0.025	5.4	16	16.1	8	54	390	1.64
SEP	KKD	42	31	0.25	0.025	2.5	1.2	16.9	8	51	383	1.41
OCT	KKD	34	33	0.25	0.025	5.9	5.7	18.1	8.1	46	291	1.31
NOV	KKD	26	25	0.25	0.025	2.5	3	23.5	8.1	40	287	1.21
DEC	KKD	29	25	0.25	0.25	2.5	1.8	21.8	8.2	41	307	1.69
JAN	BWS	45	44		0.025	112	40	22	7.63	52	335	1.12
FEB	BWS	42	28	0.25	0.025	13	17	21.1	7.54	44	287	1.52
MRT	BWS	44	28	0.25	0.025	5.3	1.6	19.1	7.8	118	767	0.86
APR	BWS	43	31	0.25	0.025	<5	2.7	16.4	7.8	49	358	0.88
MAY	BWS	58	46	0.25	0.025	2.5	1.7	10.1	8.1	44	336	0.98
JUN	BWS	43	27	0.25	0.025	5.9	0.6	9.96	7.9	43	335	1.04
JUL	BWS	49	33	0.25	0.025	2.5	0.5	9.4	8.1	44	318	0.68
AUG	BWS	41	26	0.25	0.025	2.5	1.6	11	8.1	45	344	0.85
SEP	BWS	48	32	0.25	0.025	2.5	6	14	8.2	44	332	1.1

OCT	BWS	46	33	0.25	0.32	5.3	3.5	17.6	8	43	351	2.04
NOV	BWS	30	25	0.25	0.025	2.5	1.9	20.8	8.1	48	344	3.02
DEC	BWS	40	27	0.25	0.25	2.5	2.6	20.5	7.9	47	340	1.7
JAN	OWS	78	44		0.025	121	7.2	21.5	7.56	88	568	0.96
FEB	OWS	64	37	0.8	0.025	93	3.2	19.9	7.56	73	471	0.66
MRT	OWS	63	36	1.3	0.025	77	2.9	18.9	7.44	175	1142	0.66
APR	OWS	64	43	1.2	0.025	102	0.6	17.9	7.8	69	325	0.62
MAY	OWS	47	31	1.1	0.025	99.5	1.9	12.1	7.7	69	523	0.61
JUN	OWS	61	35	1.3	0.025	110	1.9	9.28	8.12	68	524	0.56
JUL	OWS	67	41	1.1	0.025	119	1.9	12.1	7.69	64	512	0.72
AUG	OWS	56	33	1.1	0.025	112	10	12.4	7.7	74	535	0.64
SEP	OWS	69	41	1.2	0.025	121	3.8	13.8	7.5	66	511	0.74
OCT	OWS	75	45	1.1	0.025	128	2.4	17.2	7.4	72	535	0.68
NOV	OWS	48	31	1.1	0.025	89	8.2	20.5	7.6	74	526	0.91
DEC	OWS	58	35	1.1	0.25	113	6.1	20.9	7.2	70	512	1.71
JAN	BKD	48	46		0.025	95	67	23.9	7.96	74	479	1.01
FEB	BKD	43	38	0.25	0.025	102	19	24	7.9	63	408	0.57
MRT	BKD	47	38	0.25	0.025	91	2.5	22.5	7.82	169	1100	0.58
APR	BKD	49	41	0.25	0.025	91	9.8	18.1	8.4	64	464	0.59
MAY	BKD	51	33	0.25	0.025	93	2.8	14.9	8	65	510	0.53
JUN	BKD	52	35	0.25	0.025	109	2	10.7	9.7	67	511	0.54
JUL	BKD	61	42	0.25	0.025	121	3.9	11.6	8.1	69	498	0.96
AUG	BKD	51	34	0.6	0.025	112	2	14.1	7.9	66	514	0.58
SEP	BKD	60	41	0.6	0.025	113	2.1	15.1	8.1	63	490	0.52
OCT	BKD	51	44	0.25	0.025	120	0.95	18	7.8	65	502	0.44
NOV	BKD	38	43	0.25	0.025	124	1.8	21.4	8.4	63	455	0.47
DEC	BKD	30	36	0.25	0.25	111	0.5	23.5	8.3	69	508	0.57
JAN	PDM	50	47		1.1	95	95	23.7	7.99	76	495	1.76
FEB	PDM	46	39	0.25	0.025	104	30	24.6	8.12	65	424	1.95
MRT	PDM	47	39	0.25	0.025	92	4.5	23.9	7.98	188	1222	2.02
APR	PDM	51	45	0.25	0.025	86	3.1	19.4	8.1	68	481	2.12
MAY	PDM	64	50	0.25	0.025	87	6.8	15.4	7.9	76	542	1.98
JUN	PDM	52	37	0.25	0.025	102	1.9	10.98	9.24	73	533	0.94
JUL	PDM	60	45	0.25	0.025	115	2.3	12.8	8.1	62	510	1.67
AUG	PDM	52	36	0.25	0.025	85	8.5	14.9	8.1	70	507	4.29
SEP	PDM	61	44	0.25	0.025	104	18	15	7.9	81	556	1.97
OCT	PDM	43	47	0.25	0.025	106	11	18.9	7.9	77	539	1.57
NOV	PDM	39	34	0.25	0.025	110	7.5	23.3	7.6	82	596	1.53
DEC	PDM	30	37	0.25	0.25	97	5	24.7	7.8	70	503	2.67
JAN	RWP											
FEB	RWP	51	40	1.3	0.36	105	10	22.3	7.5	79	479	2.4
MRT	RWP	52	42	1.1	0.2	97	8.8	20.5	7.59	209	1359	3.11

APR	RWP	56	48	1.2	0.34	107	2	19.1	7.8	86	622	2.98
MAY	RWP	59	44	1.2	0.26	105	4.4	13.3	8.1	83	581	3.16
JUN	RWP	57	39	1.2	0.025	131	5.7	9.33	9.73	82	589	1.14
JUL	RWP	63	46	0.78	0.45	139	2.6	11.4	8.1	81	584	2.59
AUG	RWP	56	40	1.6	0.28	126	6.5	13.6	8.1	82	592	4.85
SEP	RWP	45	32	1	0.17	129	1.7	14.2	8.1	82	574	2.23
OCT	RWP	58	49	1.1	0.25	118	3.1	17.4	8.2	81	571	2.03
NOV	RWP	43	35	0.98	0.41	129	11	22	7.8	83	544	1.82
DEC	RWP	47	47	1.7	1.7	119	3.1	23.5	7.4	72	536	6.57
JAN	EBR	54	39		0.025	104	8.7	24.2	7.62	91	588	3.33
FEB	EBR	52	40	2.6	0.46	288	4.2	22.9	7.64	79	519	3.36
MRT	EBR	53	43	2.8	0.36	93	4.6	21.2	7.51	211.2	1370	2.49
APR	EBR	55	47	0.5	0.21	101	1.4	20.9	8.1	76	548	2.52
MAY	EBR	50	30	2.1	0.2	98	9.6	12.4	8.2	80	605	2.66
JUN	EBR	55	40	1.9	0.07	121	10	11.8	8.3	81	608	1.85
JUL	EBR	64	48	2.9	0.33	134	7	10.6	8.4	83	597	2.42
AUG	EBR	55	39	1	0.29	121	5.5	13.8	8.4	79	611	2.1
SEP	EBR	66	48	0.7	0.48	118	2.4	12.7	8.3	81	602	4.7
OCT	EBR	59	49	0.47	0.34	124	2.9	16.7	8.3	82	667	3.33
NOV	EBR	47	37	3.9	0.79	164	5.2	24.6	8.1	80	579	5.49
DEC	EBR	39	39	3.2	0.73	118	2.8	22.4	7.5	86	635	17.2

**Table A- 2: Indicates the significant differences in dissolved calcium between sites, using the Kruskal-Wallis test**

Dissolved Ca	Multiple Comparisons z' values						
	Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:29.458	2 R:20.667	3 R:32.250	4 R:79.417	5 R:48.167	6 R:50.500	7 R:61.136
1		0.781160	0.248046	4.438914	1.662279	1.869601	2.752798
2	0.781160		1.029206	5.220074	2.443439	2.650761	3.516788
3	0.248046	1.029206		4.190868	1.414233	1.621555	2.510205
4	4.438914	5.220074	4.190868		2.776635	2.569313	1.588545
5	1.662279	2.443439	1.414233	2.776635		0.207322	1.127058
6	1.869601	2.650761	1.621555	2.569313	0.207322		0.924292
7	2.752798	3.516788	2.510205	1.588545	1.127058	0.924292	
8	3.024681	3.805842	2.776635	1.414233	1.362402	1.155080	0.205398

**Table A- 3: Indicates the significant differences in dissolved magnesium between sites, using the Kruskal-Wallis test**

Dissolved Mg	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:13.625	2 R:24.750	3 R:32.292	4 R:53.208	5 R:58.917	6 R:67.208	7 R:68.682
1		0.988482	1.658577	3.517071	4.024270	4.761004	4.784399
2	0.988482		0.670095	2.528589	3.035788	3.772522	3.817644
3	1.658577	0.670095		1.858495	2.365693	3.102427	3.162278
4	3.517071	2.528589	1.858495		0.507199	1.243933	1.344635
5	4.024270	3.035788	2.365693	0.507199		0.736734	0.848585
6	4.761004	3.772522	3.102427	1.243933	0.736734		0.128045
7	4.784399	3.817644	3.162278	1.344635	0.848585	0.128045	
8	4.746195	3.757713	3.087619	1.229124	0.721925	0.014809	0.142528

**Table A- 4: Indicates the significant differences in nitrate between sites, using the Kruskal-Wallis test**

NO <sub>3</sub> +NO <sub>2</sub>	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:77.636	2 R:23.583	3 R:23.583	4 R:58.208	5 R:27.583	6 R:25.500	7 R:65.773
1		4.849515	4.849515	1.743039	4.490644	4.579070	1.041968
2	4.849515		0.000000	3.176294	0.366936	0.171959	3.785137
3	4.849515	0.000000		3.176294	0.366936	0.171959	3.785137
4	1.743039	3.176294	3.176294		2.809357	2.934517	0.678660
5	4.490644	0.366936	0.366936	2.809357		0.186912	3.426266
6	4.579070	0.171959	0.171959	2.934517	0.186912		3.537102
7	1.041968	3.785137	3.785137	0.678660	3.426266	3.537102	
8	0.235541	4.608909	4.608909	1.502432	4.250038	4.343530	0.806427

**Table A- 5: Indicates the significant differences in phosphate between sites, using the Kruskal-Wallis test**

PO <sub>4</sub>	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:36.333	2 R:37.000	3 R:41.042	4 R:37.000	5 R:37.000	6 R:42.042	7 R:77.318
1		0.059235	0.418346	0.059235	0.059235	0.507199	3.561554
2	0.059235		0.359112	0.000000	0.000000	0.447964	3.503622
3	0.418346	0.359112		0.359112	0.359112	0.088852	3.152404
4	0.059235	0.000000	0.359112		0.000000	0.447964	3.503622
5	0.059235	0.000000	0.359112	0.000000		0.447964	3.503622
6	0.507199	0.447964	0.088852	0.447964	0.447964		3.065504
7	3.561554	3.503622	3.152404	3.503622	3.503622	3.065504	
8	3.765118	3.705883	3.346771	3.705883	3.705883	3.257919	0.120803

**Table A- 6: Indicates the significant differences in sulphate between sites, using the Kruskal-Wallis test**

SO4	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:16.125	2 R:19.833	3 R:22.292	4 R:63.625	5 R:62.375	6 R:52.125	7 R:76.000
1		0.329494	0.547923	4.220486	4.109420	3.198684	5.203095
2	0.329494		0.218429	3.890992	3.779926	2.869190	4.880844
3	0.547923	0.218429		3.672563	3.561498	2.650761	4.667216
4	4.220486	3.890992	3.672563		0.111065	1.021802	1.075379
5	4.109420	3.779926	3.561498	0.111065		0.910736	1.184003
6	3.198684	2.869190	2.650761	1.021802	0.910736		2.074721
7	5.203095	4.880844	4.667216	1.075379	1.184003	2.074721	
8	5.138627	4.809132	4.590704	0.918141	1.029206	1.939943	0.177419

**Table A- 7: Indicates the significant differences in chlorophyll-a between sites, using the Kruskal-Wallis test**

Chl a	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:36.792	2 R:53.958	3 R:35.625	4 R:44.042	5 R:40.542	6 R:65.042	7 R:53.727
1		1.525298	0.103661	0.644179	0.333196	2.510078	1.471692
2	1.525298		1.628959	0.881119	1.192102	0.984780	0.020079
3	0.103661	1.628959		0.747840	0.436857	2.613739	1.573075
4	0.644179	0.881119	0.747840		0.310983	1.865899	0.841672
5	0.333196	1.192102	0.436857	0.310983		2.176882	1.145820
6	2.510078	0.984780	2.613739	1.865899	2.176882		0.983213
7	1.471692	0.020079	1.573075	0.841672	1.145820	0.983213	
8	1.595640	0.070341	1.699301	0.951460	1.262444	0.914439	0.088874

**Table A- 8: Indicates the significant differences in temperature between sites, using the Kruskal-Wallis test**

Temp	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:53.042	2 R:49.833	3 R:38.042	4 R:39.083	5 R:52.000	6 R:56.417	7 R:44.182
1		0.285068	1.332785	1.240230	0.092555	0.299877	0.769915
2	0.285068		1.047717	0.955163	0.192513	0.584945	0.491113
3	1.332785	1.047717		0.092555	1.240230	1.632662	0.533575
4	1.240230	0.955163	0.092555		1.147676	1.540107	0.443055
5	0.092555	0.192513	1.240230	1.147676		0.392431	0.679394
6	0.299877	0.584945	1.632662	1.540107	0.392431		1.063200
7	0.769915	0.491113	0.533575	0.443055	0.679394	1.063200	
8	0.174002	0.111065	1.158782	1.066228	0.081448	0.473879	0.599737

**Table A- 9: Indicates the significant differences in pH between sites, using the Kruskal-Wallis test**

pH	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:7.4583	2 R:45.917	3 R:54.708	4 R:31.708	5 R:66.417	6 R:59.125	7 R:54.727
1		3.417113	4.198273	2.154669	5.238585	4.590704	4.107637
2	3.417113		0.781160	1.262444	1.821473	1.173591	0.765635
3	4.198273	0.781160		2.043604	1.040313	0.392431	0.001646
4	2.154669	1.262444	2.043604		3.083916	2.436035	2.000330
5	5.238585	1.821473	1.040313	3.083916		0.647882	1.015800
6	4.590704	1.173591	0.392431	2.436035	0.647882		0.382159
7	4.107637	0.765635	0.001646	2.000330	1.015800	0.382159	
8	5.068285	1.651172	0.870012	2.913616	0.170300	0.477581	0.849243

**Table A- 10: Indicates the significant differences in EC between sites, using the Kruskal-Wallis test**

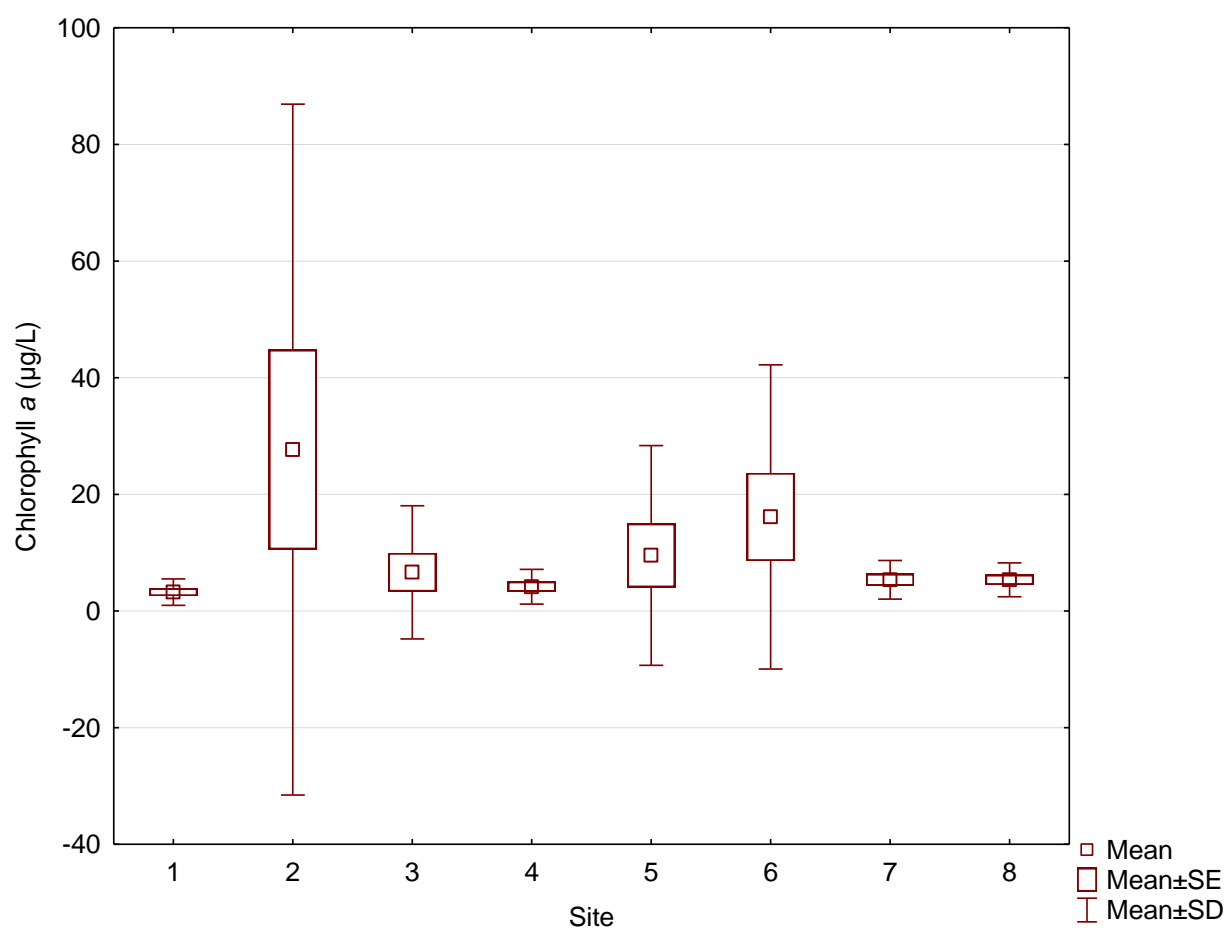
EC	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:20.042	2 R:33.417	3 R:24.542	4 R:55.792	5 R:45.417	6 R:58.167	7 R:75.000
1		1.188400	0.399835	3.176471	2.254628	3.387495	4.775840
2	1.188400		0.788564	1.988071	1.066228	2.199095	3.613562
3	0.399835	0.788564		2.776635	1.854792	2.987660	4.384794
4	3.176471	1.988071	2.776635		0.921843	0.211024	1.669191
5	2.254628	1.066228	1.854792	0.921843		1.132867	2.570771
6	3.387495	2.199095	2.987660	0.211024	1.132867		1.462805
7	4.775840	3.613562	4.384794	1.669191	2.570771	1.462805	
8	4.783217	3.594817	4.383382	1.606746	2.528589	1.395722	0.097762

**Table A- 11: Indicates the significant differences in TDS between sites, using the Kruskal-Wallis test**

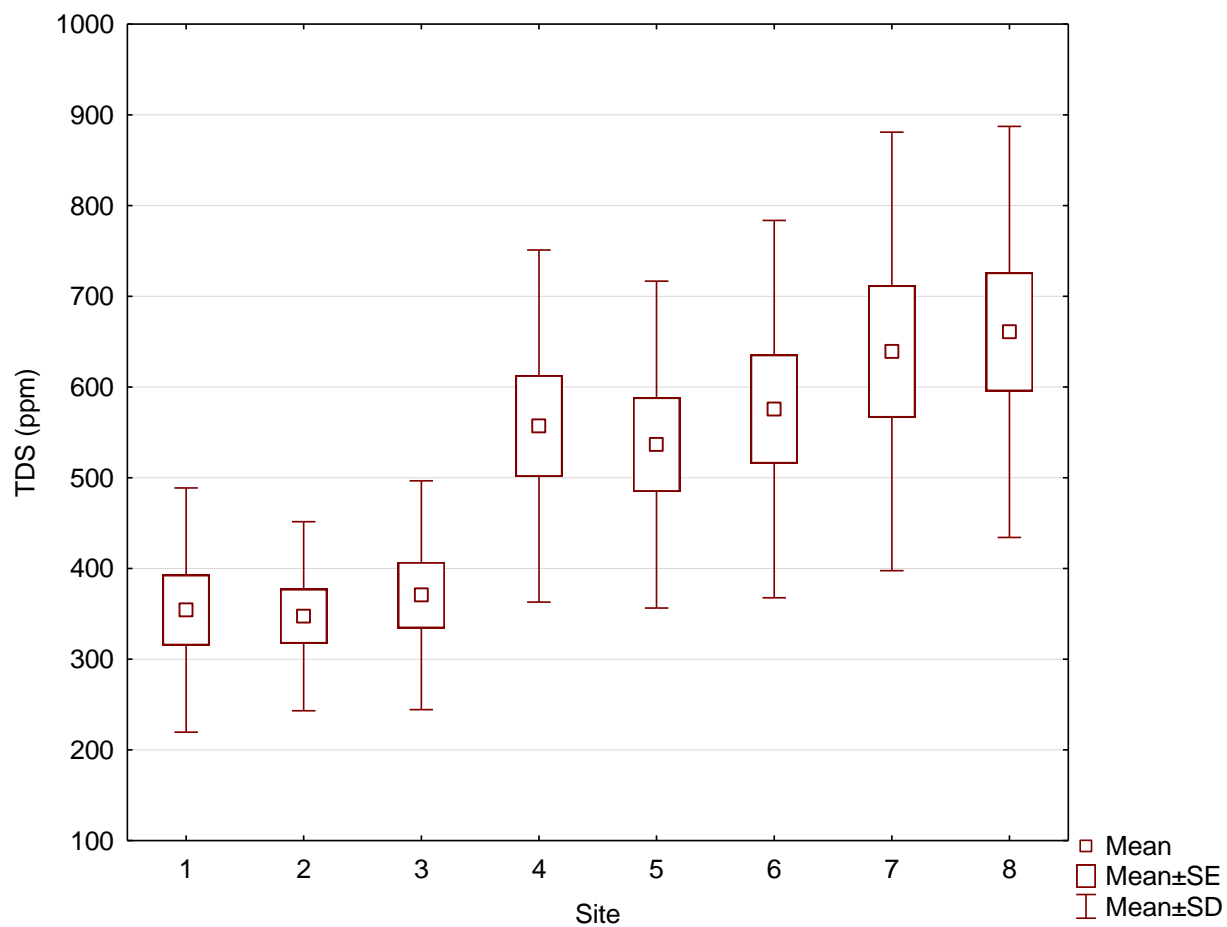
TDS	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:19.750	2 R:21.625	3 R:28.833	4 R:56.917	5 R:48.625	6 R:58.375	7 R:72.500
1		0.166598	0.807075	3.302345	2.565611	3.431921	4.583938
2	0.166598		0.640477	3.135747	2.399013	3.265323	4.421002
3	0.807075	0.640477		2.495270	1.758536	2.624846	3.794603
4	3.302345	3.135747	2.495270		0.736734	0.129576	1.354181
5	2.565611	2.399013	1.758536	0.736734		0.866310	2.074721
6	3.431921	3.265323	2.624846	0.129576	0.866310		1.227453
7	4.583938	4.421002	3.794603	1.354181	2.074721	1.227453	
8	5.301522	5.134924	4.494447	1.999177	2.735911	1.869601	0.601053

**Table A- 12: Indicates the significant differences in turbidity between sites, using the Kruskal-Wallis test**

NTU	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate $p < 0.05$						
	1 R:8.2500	2 R:52.792	3 R:47.958	4 R:30.750	5 R:20.875	6 R:65.542	7 R:77.364
1		3.957631	3.528178	1.999177	1.121761	5.090498	6.005926
2	3.957631		0.429453	1.958453	2.835870	1.132867	2.135287
3	3.528178	0.429453		1.529001	2.406417	1.562320	2.555300
4	1.999177	1.958453	1.529001		0.877417	3.091321	4.050692
5	1.121761	2.835870	2.406417	0.877417		3.968737	4.908823
6	5.090498	1.132867	1.562320	3.091321	3.968737		1.027321
7	6.005926	2.135287	2.555300	4.050692	4.908823	1.027321	
8	6.634307	2.676676	3.106129	4.635130	5.512547	1.543809	0.482554



**Figure A- 2: A Box and whisker plot illustrating the change in mean Chlorophyll-*a* concentration in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014) SE = Standard Error; SD = Standard Deviation**



**Figure A- 3: A Box and whisker plot illustrating the change in mean TDS levels in mg/L between sites from upstream (1- BVO) to downstream (8- EBR) (January 2014 – December 2014) SE = Standard Error; SD = Standard Deviation**

# Appendix C

## Supplementary diatom data

**Table A- 13: Displaying the diatom indices scores from all the samples collected in the 2014 calendar year organised by site**

Date	Site	%PTV	GDI	SPI	BDI
JAN	BVO	0	17	15.4	19.3
FEB	BVO	0	15.8	13.6	19.6
MRT	BVO	0	15.4	13.8	19.7
APR	BVO	0	15.6	14.8	20
MAY	BVO	0	15.2	14.3	19.2
JUN	BVO	0.2	14.7	14.1	18.9
JUL	BVO	0	15.3	13.2	16.7
AUG	BVO	0	14.6	13.6	18.2
SEP	BVO	0	15.8	15.3	19.8
OCT	BVO	1.5	15.1	13.7	19.6
NOV	BVO	1	13	12.1	13.7
DEC	BVO	0	14.3	14	19
JAN	KKD	5.3	15.6	17.1	17.8
FEB	KKD	2.8	15.7	15.9	17
MRT	KKD	2.5	16.6	16.1	17.2
APR	KKD	1.8	15.8	16.4	17.8
MAY	KKD	4	15.2	14.5	14.4
JUN	KKD	8	13.2	13.4	12.6
JUL	KKD	2.5	13.9	12.4	11.1
AUG	KKD	7.3	13.8	15.9	15.6
SEP	KKD	5.8	14.3	14.1	12.5
OCT	KKD	4	15.3	15	15.9
NOV	KKD	7	14.4	15.2	16.5
DEC	KKD	4.3	14.4	16.7	15.9
JAN	BWS	12.7	14.9	16.3	15.9
FEB	BWS	16	14.6	17.2	14.3
MRT	BWS	13.3	14.6	16.7	15.3
APR	BWS	12	15.1	17.3	14.7
MAY	BWS	11	14.5	16.3	14.4
JUN	BWS	10	14.6	15.6	14
JUL	BWS	10	14.5	14.5	13.4
AUG	BWS	11.3	14.7	15.8	14.6
SEP	BWS	15	14.7	16.1	14.8

OCT	BWS	20.8	14.6	16.1	14.7
NOV	BWS	15.5	14.2	15	13.9
DEC	BWS	15.5	14	16.7	15.1
JAN	OWS	6.3	11.2	12.2	14.6
FEB	OWS	3.3	11.5	11.9	14.6
MRT	OWS	3.8	12.1	12.5	15.6
APR	OWS	2.8	11.9	12.3	16.2
MAY	OWS	4.5	11.3	12.6	14.1
JUN	OWS	3	12.2	12.9	15.1
JUL	OWS	4	10.8	11.8	15.4
AUG	OWS	6.3	10	11.9	14.3
SEP	OWS	3.3	11.8	12.5	15.9
OCT	OWS	5.3	10.9	12.6	13.8
NOV	OWS	8.8	10.6	12.6	15.2
DEC	OWS	5.8	9.8	11.2	14.2
JAN	BKD	0	16	12.8	17.5
FEB	BKD	0	16.2	12.4	17.4
MRT	BKD	0	15.3	13.3	17.2
APR	BKD	0	14.8	12.1	16.3
MAY	BKD	0	15.7	14.1	18.5
JUN	BKD	0	13.6	11.9	14.8
JUL	BKD	0	14.5	11.8	13.6
AUG	BKD	0	15	13.6	17
SEP	BKD	0	15.5	13.3	14.5
OCT	BKD	0	15.1	13.3	17.2
NOV	BKD	0	15.3	13.1	17.1
DEC	BKD	0	14.2	12.3	14.5
JAN	PDM	14.5	11.3	11	12.3
FEB	PDM	10	11.9	10.8	11.2
MRT	PDM	13.3	11	9.6	10.2
APR	PDM	12.5	11.8	9.4	9.9
MAY	PDM	11.3	12.7	9.9	10.8
JUN	PDM	12.5	11.4	9.2	10.6
JUL	PDM	6	12.6	11.6	13.4
AUG	PDM	7.3	13.2	10.2	12.4
SEP	PDM	8.3	12.5	11.7	14.1
OCT	PDM	16.8	11.3	9.4	11.9
NOV	PDM	7.8	12.2	10.5	12.3
DEC	PDM	4.3	12.2	10.4	11.7
JAN	RWP	11.8	8.1	6	8.9
FEB	RWP	12.8	8	5.3	8.1

MRT	RWP	18.8	9.7	6.1	7.8
APR	RWP	18.5	9.1	5.7	8.3
MAY	RWP	17.8	8.4	6.6	8.9
JUN	RWP	9	9.7	6.8	10.2
JUL	RWP	6.3	9.4	7.3	11.4
AUG	RWP	12.3	10.5	8.6	12.6
SEP	RWP	7.5	9.7	8.5	11.7
OCT	RWP	12.5	9.7	8.4	11.1
NOV	RWP	11.5	8.8	7.3	11.6
DEC	RWP	12.3	8.7	8.5	12.5
JAN	EBR	13.3	9.9	6.9	8.6
FEB	EBR	10.3	9.7	6.7	8.6
MRT	EBR	21.8	9.8	6.3	8.5
APR	EBR	15	10.2	6.9	8.9
MAY	EBR	15.3	9.1	7.2	9.6
JUN	EBR	10.8	11	8.1	10.7
JUL	EBR	12.3	11.6	9.5	11.7
AUG	EBR	12.8	10.6	8.9	12.8
SEP	EBR	4.5	11.3	9.5	11.4
OCT	EBR	12.3	10.2	8.7	12.1
NOV	EBR	14.3	10.7	9.4	11.6
DEC	EBR	12.3	9.5	8.1	12.1

**Table A- 14: Indicates the significant differences in water quality variables and diatom indices, using the Pearson correlation test**

Marked correlations are significant at $p < 0.05$				
Variable	%PTV	GDI	SPI	BDI
Ca Dissolved	0.074734	-0.436335	-0.394244	-0.285481
Mg Dissolved	0.288954	-0.479	-0.574061	-0.518919
Nitrate & Nitrate	0.135294	-0.447022	-0.455463	-0.171219
Orthophosphate	0.364233	-0.523059	-0.465093	-0.380309
Sulphate	0.155607	-0.65882	-0.73089	-0.514835
Chlorophyll a	-0.070461	0.113427	0.067467	-0.037713
Temp	-0.008318	0.062336	0.015962	0.130601
pH	0.218749	-0.18421	-0.260762	-0.413069
EC (mS/m)	0.252652	-0.345342	-0.429442	-0.360439
TDS	0.307163	-0.446236	-0.525971	-0.426395
NTU	0.4148	-0.456847	-0.426939	-0.435364

**Table A- 15: Indicates the significant differences between diatom indices, using the Pearson correlation test**

Variable	Marked correlations are significant at $p < .05000$			
	%PTV	GDI	SPI	BDI
%PTV	1.000000	-0.557492	-0.388776	-0.714841
GDI	-0.557492	1.000000	0.853679	0.783805
SPI	-0.388776	0.853679	1.000000	0.771496
BDI	-0.714841	0.783805	0.771496	1.000000

**Table A- 16: Indicates the significant differences in %PTV between sites, using the Kruskal-Wallis test**

%PTV	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate $p < 0.05$						
	1 R:14.000	2 R:37.708	3 R:76.583	4 R:38.667	5 R:11.000	6 R:64.208	7 R:71.958
1		2.084711	5.503050	2.168978	0.263795	4.414897	5.096367
2	2.084711		3.418339	0.084268	2.348505	2.330186	3.011656
3	5.503050	3.418339		3.334072	5.766845	1.088153	0.406683
4	2.168978	0.084268	3.334072		2.432773	2.245919	2.927388
5	0.263795	2.348505	5.766845	2.432773		4.678692	5.360161
6	4.414897	2.330186	1.088153	2.245919	4.678692		0.681470
7	5.096367	3.011656	0.406683	2.927388	5.360161	0.681470	
8	5.264902	3.180191	0.238148	3.095924	5.528697	0.850005	0.168535

**Table A- 17: Indicates the significant differences in GDI between sites, using the Kruskal-Wallis test**

GDI	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate $p < 0.05$						
	1 R:77.167	2 R:71.000	3 R:65.542	4 R:30.292	5 R:76.167	6 R:39.583	7 R:8.2083
1		0.542245	1.022204	4.121792	0.087932	3.304761	6.063614
2	0.542245		0.479960	3.579547	0.454313	2.762516	5.521369
3	1.022204	0.479960		3.099587	0.934273	2.282557	5.041409
4	4.121792	3.579547	3.099587		4.033860	0.817031	1.941822
5	0.087932	0.454313	0.934273	4.033860		3.216829	5.975682
6	3.304761	2.762516	2.282557	0.817031	3.216829		2.758853
7	6.063614	5.521369	5.041409	1.941822	5.975682	2.758853	
8	5.023090	4.480846	4.000886	0.901298	4.935159	1.718329	1.040523

**Table A- 18: Indicates the significant differences in SPI between sites, using the Kruskal-Wallis test**

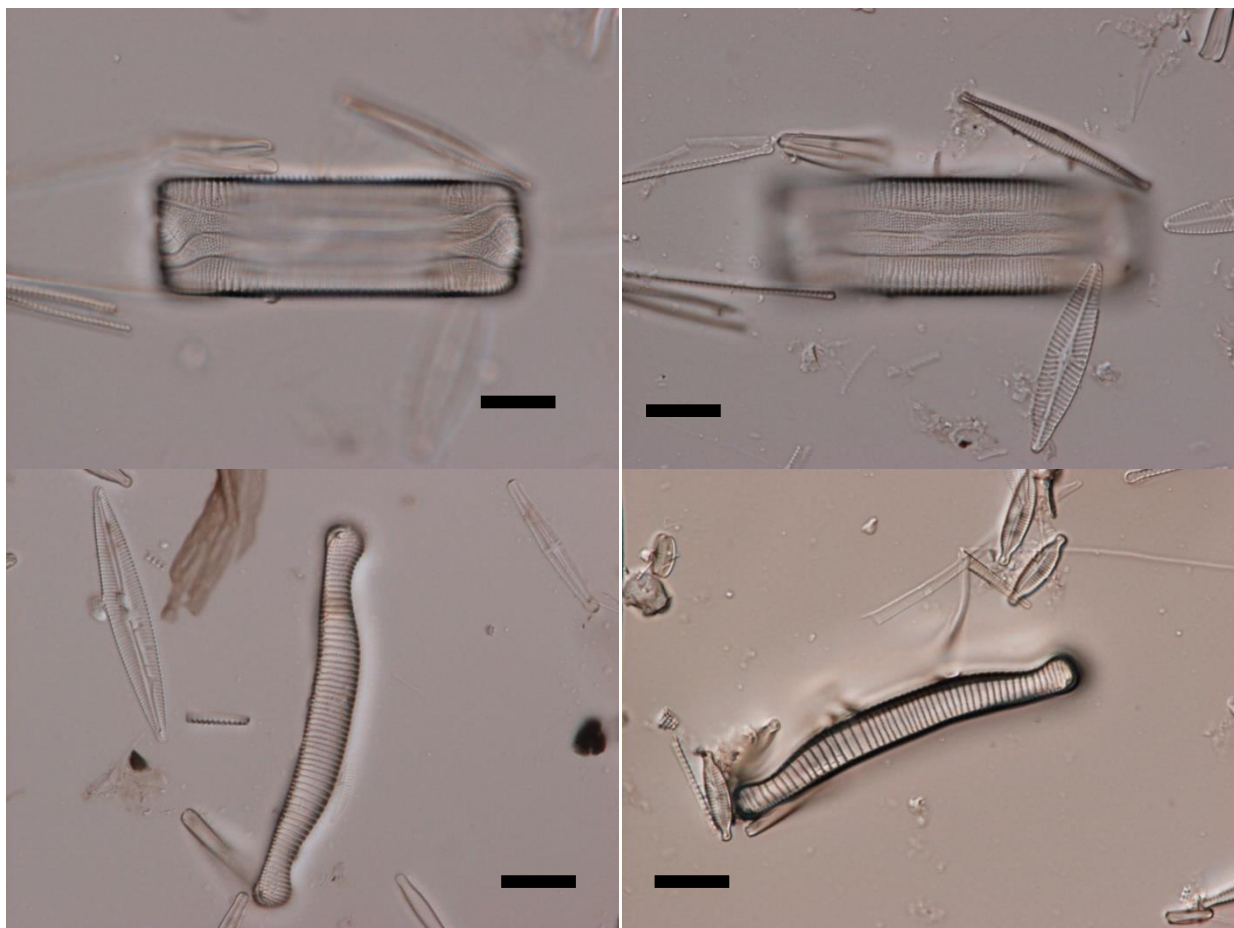
SPI	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:67.208	2 R:78.125	3 R:86.417	4 R:46.917	5 R:53.667	6 R:30.000	7 R:9.9167
1		0.959920	1.689019	1.784278	1.190740	3.271787	5.037745
2	0.959920		0.729099	2.744197	2.150659	4.231706	5.997665
3	1.689019	0.729099		3.473297	2.879759	4.960805	6.726764
4	1.784278	2.744197	3.473297		0.593538	1.487509	3.253468
5	1.190740	2.150659	2.879759	0.593538		2.081047	3.847006
6	3.271787	4.231706	4.960805	1.487509	2.081047		1.765959
7	5.037745	5.997665	6.726764	3.253468	3.847006	1.765959	
8	4.524811	5.484731	6.213830	2.740534	3.334072	1.253025	0.512934

**Table A- 19: Indicates the significant differences in BDI between sites, using the Kruskal-Wallis test**

BDI	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05						
	1 R:85.833	2 R:61.833	3 R:54.083	4 R:57.542	5 R:70.333	6 R:25.125	7 R:15.750
1		2.110357	2.791827	2.487730	1.362939	5.338178	6.162537
2	2.110357		0.681470	0.377373	0.747418	3.227821	4.052179
3	2.791827	0.681470		0.304097	1.428888	2.546351	3.370710
4	2.487730	0.377373	0.304097		1.124791	2.850448	3.674806
5	1.362939	0.747418	1.428888	1.124791		3.975239	4.799598
6	5.338178	3.227821	2.546351	2.850448	3.975239		0.824358
7	6.162537	4.052179	3.370710	3.674806	4.799598	0.824358	
8	6.008656	3.898299	3.216829	3.520926	4.645717	0.670478	0.153880



**Figure A- 4: Illustrations of a possible new *Gomphonema* species found in Boskop Dam. Scale bar = 10µm**



**Figure A- 5: Illustrations of a possible new *Eunotia* species found in Boskop Dam. Scale bar = 10μm**

## Appendix D

Supplementary macroinvertebrate data

**Table A- 20: Displaying the macroinvertebrate index scores from all the samples collected in the 2014 calendar year organised by date, and biological integrity**

Date	Site	SASS5	No.	ASPT	Category
Jan	BVO	112	17	6.588235	B/A
Jan	PDM	101	19	5.315789	B
Jan	RWP	51	12	4.25	D/C
Feb	BVO	113	17	6.647059	B/A
Feb	PDM	106	20	5.3	B
Feb	RWP	52	13	4	C/E
Mrt	BVO	131	22	5.954545	A
Mrt	PDM	95	18	5.277778	B
Mrt	RWP	54	13	4.153846	D/E
Apr	BVO	106	17	6.235294	B/A
Apr	PDM	98	21	4.666667	B/C
Apr	RWP	64	14	4.571429	C/D
May	BVO	98	16	6.125	B/A
May	PDM	92	20	4.6	B/C
May	RWP	62	14	4.428571	D
Jun	BVO	110	18	6.111111	B/A
Jun	PDM	92	21	4.380952	B/D
Jun	RWP	59	16	3.6875	D/E
Jul	BVO	92	16	5.75	B/A
Jul	PDM	58	15	3.866667	C/E
Jul	RWP	55	16	3.4375	C/E
Aug	BVO	93	16	5.8125	B/A
Aug	PDM	90	21	4.285714	B/D
Aug	RWP	50	14	3.571429	E
Sept	BVO	115	19	6.052632	B/A
Sept	PDM	104	22	4.727273	B/C
Sept	RWP	66	18	3.666667	C/E
Oct	BVO	123	20	6.15	A
Oct	PDM	84	17	4.941176	B
Oct	RWP	51	12	4.25	D
Nov	BVO	114	20	5.7	B/A
Nov	PDM	90	20	4.5	B/D
Nov	RWP	43	11	3.909091	E
Dec	BVO	111	17	6.529412	B/A
Dec	PDM	98	19	5.157895	B
Dec	RWP	73	17	4.294118	C/D

**Table A- 21: Indicates the significant differences in water quality variables and macroinvertebrate indices, using the Pearson correlation test**

Variable	Marked correlations are significant at p <0 .05	
	SASS5 Score	ASPT
Ca Dissolved	-0.488845	-0.574581
Mg Dissolved	-0.587224	-0.721432
Nitrate & Nitrate	0.044805	0.366340
Orthophosphate	-0.403198	-0.353883
Sulphate	-0.761691	-0.921698
Chlorophyll a	-0.020552	-0.134981
Temp	0.307540	0.375360
pH	-0.497225	-0.706510
EC (mS/m)	-0.333657	-0.382767
TDS	-0.365568	-0.430060
NTU	-0.577572	-0.683274

**Table A- 22: Indicates the significant differences in SASS5 score between sites, using the Kruskal-Wallis test**

SASS5	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05		
	1 R:29.042	6 R:19.542	7 R:6.9167
1		2.208705	5.143958
6	2.208705		2.935253
7	5.143958	2.935253	

**Table A- 23: Indicates the significant differences in ASPT between sites, using the Kruskal-Wallis test**

ASPT	Multiple Comparisons z' values Kruskal-Wallis test: Marked values indicate p<0.05		
	1 R:30.500	6 R:17.333	7 R:7.6667
1		3.061188	5.308642
6	3.061188		2.247454
7	5.308642	2.247454	



NORTH-WEST UNIVERSITY  
YUNIBESITHI YA BOKONE-BOPHIRIMA  
NOORDWES-UNIVERSITEIT  
POTCHEFSTROOM CAMPUS

Higher Degree Administration

## AMENDMENTS FORM

In accordance with the Statute of the North-West University, the undersigned declares that the candidate mentioned below has made the changes to the satisfaction of the supervisor/promoter as indicated by him/her.

Candidate: Ernest Pelser

University number: 2 1 6 9 8 7 5 9

Qualification: MSc Environmental Science

Registered Title of the mini-dissertation/dissertation/thesis (exactly as approved and registered):

Ecosystem health and water quality of the Mooi River and Associated impoundments using diatoms and macroinvertebrate as bioindicators

Student Signature

1 1 - 0 5 - 2 0 1 6

Date

### For completion by the supervisor/promoter:

Have the changes indicated by the supervisor/promoter been made?

☒ Yes ☐ No

Are the final copies (printed and electronic) in order and according to the specifications?

☒ Yes ☐ No

Signature of Sup

11-05-2016

Date

# FAKULTEIT NATUURWETENSKAPPE

## Uitslag van die Magistereksamen

Verhandeling	x	Skripsie		Merk toepaslike blokkie
Kandidaat:	Mnr ELC Pelser			Universiteitsnommer: 21698759
Graad:	MSc			Kurrikulumnommer: N830P
Beskrywende naam van kurrikulum soos in die jaarboek:		Omgewingswetenskappe		
Fokusarea / Navorsingseenheid: Omgewingswetenskap- en bestuur				
Titel: Ecosystem health and water quality of the Mooi River and associated impoundments using diatoms and macroinvertebrate as bioindicators				

**A** Die ondergetekendes gee as die betrokke skool- en navorsingsdirekteure, aan die Fakulteitsbestuur kennis dat die uitslag van die Magistereksamen van bogenoemde kandidaat soos volg is:

- ☐ Die kandidaat slaag en die verhandeling/skripsie word onvoorwaardelik aanvaar. Die geringe taal-, tik- en/of tegniese foute wat voorkom, word vir regstelling aan die kandidaat onder toesig van sy/haar studieleier oorgelaat.
- ☒ Die verhandeling/skripsie slaag, maar daar is te veel tik-, taal- en/of tegniese foute en/of geringe wetenskaplik-inhoudelike foute vir opsie 1; daarom moes 'n gekorrigeerde eksemplaar eers aan die navorsingsdirekteur vir finale goedkeuring voorgelê word. **Die gekorrigeerde eksemplaar is deur ons nagegaan en die verhandeling is finaal aanvaar en die kandidaat slaag.**
- ☐ Die verhandeling/skripsie slaag nie in sy huidige vorm nie, maar word na die kandidaat terugverwys vir eenmalige omvattende hersiening en/of uitbreiding en herindiening vir hereksaminering. 'n Aanbeveling oor die eksaminatore vir hierdie tweede eksaminering sal aan die Fakulteitsbestuur gemaak word.
- ☐ Die verhandeling/skripsie word nie aanvaar nie en die kandidaat druipt.

B PUNTEBEREKENING VIR MAGISTERGRAAD		PUNT	UITSLAG
Gemiddelde punt aan verhandeling/skripsie toegeken:		67%	Druip
Gemiddelde punt behaal in mondelinge/skriftelike eksamen(s):			Slaag
Finale punt bereken volgens die verhouding:			Slaag met lof
Kredietpunt totaal vraestelle : Kredietpunte verhandeling/skripsie = ..... : .....			

Navorsingsdirekteur

Skooldirekteur

Datum

24/02/2016