

**A BIOPHYSICAL FRAMEWORK FOR THE
SUSTAINABLE MANAGEMENT OF WETLANDS
IN THE LIMPOPO PROVINCE WITH
NYLSVLEY AS A REFERENCE MODEL**

**Report to the
Water Research Commission**

by

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WRC REPORT NO.: 1258/1/06

ISBN NO: 1-77005-462-6

APRIL 2006

This Report is obtainable from:

Water Research Commission
Private Bag X03
Gezina
0031
Pretoria, South Africa

The Report emanates from a Water Research Commission project
K5/1258: titled:
A BIOPHYSICAL FRAMEWORK FOR THE SUSTAINABLE MANAGEMENT
OF WETLANDS IN THE NORTHERN (LIMPOPO) PROVINCE WITH
NYLSVLEY AS A REFERENCE MODEL

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EXECUTIVE SUMMARY

Water in South Africa is a critically important natural resource. The Department of Water Affairs and Forestry list it as one of the most limiting resources in South Africa. South Africa is a semi-arid to arid region, receiving only an average of 500 mm of rain per annum, which is only 60% of the world average. Sixty five percent of the country receives less than 500 mm per annum, with 21% receiving less than 200 mm per annum. These conditions are further associated with highly variable climatic processes such as long periods of drought and floods, resulting in impacts on water resources that are difficult to manage. Jay O’Keeffe mentioned that the country’s rivers are often short and high seasonal flow regimes occur during the rainy season.

The ecological value of wetlands is demonstrated by their physical and geological presence, shape and vegetation it supports. Wetlands control stream flow, attenuate the force of floods, store water, slowly releasing it over time and remove certain pollutants. The value of wetlands was previously not appreciated and the international trend was to drain wetlands. In 1996, Dr. Geoff Cowan (DEAT) estimated that more than half of the wetlands in South Africa had already been destroyed or otherwise lost. Where wetlands are destroyed in the catchment of a river, the likelihood of flooding can increase in the lower reaches, resulting in serious damage. The recent floods in the Limpopo Province, Mpumalanga and Mozambique are clear evidence of this fact and a large proportion of the damage can be attributed to poor wetland management. The lack of wetlands could not buffer or minimise the impacts of the floods.

A possible reason for the lack of awareness of the importance of wetlands may be the fact that research on vleis and sponge areas in the past focused mainly on specific agricultural activities and related matters in catchment areas. This resulted in the fact that rivers and associated wetlands were merely considered as sources of freshwater to be utilised at will.

Although there is an increase in public awareness of the environmental importance of water and the importance of conservation of the natural environment when developing water resources (Heath and Claassen, 1999), much must still be done to make the larger community aware of this aspect. This is especially important when one refers to wetland conservation. In semi-arid areas, it is critical to find a balance between water resource development and the maintenance of the natural environment (DWAF, 1995).

At a workshop in 2000 on Wetlands in the Limpopo Province, a needs analysis identified the following five main areas for research: 1) a need to compile baseline information of wetland research

in the Limpopo Province; 2) preparation and utilisation of information and knowledge in education and awareness programmes; 3) development of scientific information to assist in the management approach of these sensitive habitats; 4) development of expertise on vlei's and sponge areas in the province (centre of excellence) and 5) to identify development potentials in the area.

Nylsvley as a RAMSAR site is under threat. Recent studies have shown that the water quality is poor. During the last ten seasons, water bird numbers have shown a decline of 80-90% in Nylsvley, indicating deterioration in the wetland. It was also mentioned that the frog numbers have declined, as only a few calling frogs were heard in February, after the good rains in the Limpopo Province (Wetlands Workshop, 2000). A study in this area will assist the Limpopo Province Department of Finance and Economic Development (Environmental Affairs) to compile a management plan for the reserve. The Nylsvley project is proposed in an attempt to obtain a holistic perspective of the current state of the vlei area and to identify impacts that could lead to further degradation and also identify areas where restoration could be affective.

A further important consideration for focusing in the area is that the Waterberg is registered as a Biosphere Reserve, making it an important international and national conservation area. The project will address various issues such as pollution from various quarters that have impacted on the resources and the effects of the increasing human population on the water quality in the area.

Introduction:

The project was initiated after a two-day workshop in 2000 hosted by Dr Steve Mitchell of the Water Research Commission near Nylsvley Nature Reserve. The aim of the workshop was to gather all role players to identify needs as far as biomonitoring of wetlands are concerned. After the consultation with the community, researchers and conservation agencies, a specialist workshop was held to compile the proposal for the project.

Objectives:

- 1 To develop draft water quality guidelines in vlei areas for key variables/parameters.
- 2 To develop potential biomonitoring indices specific to vlei areas.
- 3 To compile a draft sustainable management programme, focusing on the biophysical aspects, for Nylsvley.
- 4 To propose a strategic management plan for sustainable utilization of wetlands in the Waterberg region.

A further important aspect of the project was the building of capacity. The lack of scientists specializing in wetlands is still problem is South Africa. A result from the larger project to address

the problem is the completion of a PhD and two MSc studies. Various other students were involved in the project as research assistants, although the projects they completed as part of their studies focused on other areas.

Summary of the different chapters:

Chapter 1 is a summary of the different sites used during the study. A total of 18 sites were used, 10 for water and sediment sampling and 8 for the whole spectrum of water, sediment and biota.

Chapter 2 is a comprehensive summary of the water quality study. A range of parameters was evaluated and compared to an earlier study that focused on the Nylsvley Reserve area only. In the latter study, sites in the upper catchment of some of the important tributaries were included. The idea was to use these sites to get some baseline water quality data to really be able to quantify the pollution of the water in the Nyl River system and the associated floodplain.

Chapter 3 gives an overview of the potential pollutants in the sediments. This study was included to give some overview of the “pollution history” of the study area.

Chapter 4 is focusing on pesticides as potential pollutants. During the initial phase of the project, it was assumed that metals formed the most important pollutants in the system. As the project developed, we have decided to investigate the possible impact of pesticides on the biota of the system.

Chapter 5 is a summary of the work done on the macro-invertebrates of the whole system. The idea was to test the current indices used in the River Health Programme (RHP) to test its applicability to the Nylsvley area as a monitoring tool. The methodology and interpretation of the results were tested to evaluate the potential of SASS5 as a monitoring tool for Nylsvley and similar wetlands.

Chapter 6 is included to give some view on the potential of frogs in particular as a monitoring agent for wetlands. The idea is to develop this work further into an index that can form part of a biomonitoring programme.

Chapter 7 focused on the plants as monitoring organisms. The feeling was that the current Riparian Vegetation Index (RVI) used in the RHP is not practical and a more specific index must be developed. This is an effort to get a working document on the table.

Chapter 8 is evaluating the Fish Assemblage Integrity Index (FAII) as a tool to monitor Nylsvley and similar floodplain areas.

Chapter 9 is a proposed method to use the various indices from this project to get an overall picture of the health of wetlands.

Chapter 10 an example of what a management plan must consist of and we used one of the main issues identified to illustrate the use of the framework.

Recommendations:

This is a summary of the recommendations from the various sections in the report. Each author has highlighted the most important aspects. These recommendations form the basis for the management framework we have compiled for the management of Nylsvley in particular and the whole floodplain and the catchment areas contributing to this sensitive system. Towards the end of the project a flight in micro-light airplanes was undertaken to get a overall view of the study area in particular and a general impression of the area. It must be emphasized that this gave a huge insight into the system, its functioning and problems not picked up during normal surveys and traveling in the area. It is therefore recommended that an aerial survey must form part of the initial surveys or planning of any catchment based research programmes. The value of this form of evaluation of the study area is still not fully appreciated and can help to plan better, understand the system and pick up on hidden problems which will help to interpret results more effectively.

One important aspect that was mentioned throughout the study and especially during the compilation of the final report was the importance to develop a monitoring protocol and an early warning system. Managers and landowners need a simplified system to give them enough time to respond to changes in the sensitive wetland system and the development of Thresholds of Possible Concern (TPC's) will be an important strategy to address this issue. Although this study didn't produce all the TPC's one will need to monitor a system such as the Nyl floodplain and its tributaries, the foundations had been laid and the data and management plan will definitely give some pointers to all concerned.

Water:

The results from the water analysis give a comprehensive summary of the water quality in the Nyl River System. Of the many variables and parameters analysed the bacteriological study indicated most cause for concern. Total coliform and faecal coliform counts increased along the course of the river and into the wetland. The faecal coliform counts increased dramatically from the point where the Modimolle Sewage Treatment Works releases its effluent into the Klein Nyl River. This increase in faecal coliform content can have deleterious effects on the system as well as the people who rely on the water in the system. There are three possible causes for the increase in faecal coliform content along the course of the Nyl River namely:

1. The Modimolle Sewage Treatment Works is unable to cope with the volume of effluent that passes through it. This is visible in figure 2.39, which clearly shows a pipe leading into the field along the banks of the Klein Nyl River. This could lead to raw sewage leaching into the river and the subsequent contamination thereof.

2. The second possible cause for the increase in faecal coliform content in the river is run off from the surrounding informal settlements such as Phagameng. Insufficient municipal services could have an effect in the quantity of bacteria entering the system during the rainy season.
3. The third possible cause for the increased faecal coliform content in the system can be attributed to increased run off from agricultural land. The surrounding areas contain a large number of live stock farmlands including cattle, pigs, chickens, game and crocodiles. Run off during the rainy season could cause an increase of faecal coliform bacteria in the system.

Although there is little one can do about the increase of livestock farming in the surrounding areas it is possible to address the other two points of contamination. The Modimolle Sewage Treatment Works needs to be upgraded to cope with the volumes of sewage received and possibly to make provision for any further expansion of Modimolle. The incorporation of the informal settlements into the sewage network would also decrease the amount off of bacteria entering the system. Although parts of the settlements may have been incorporated, personal observation indicated that the pipelines are in a poor state and blockages in the system cause spills of raw sewage. Maintenance of these systems is also vitally important.

The floodplain helps in the purification of the water in the river system and aids in decreasing coliform counts. Continual contamination of the system however eventually exceeds the systems ability to cope with the bacterial levels and process them, leading to health risk problems further downstream. Increased levels of coliform bacteria in the wetland could also lead to the contamination of ground water supplies.

Toxicity tests done on the water in the system indicated that the water is suitable for sustaining aquatic life. The acute static screening toxicity tests on *Poecillia reticulata* and *Daphnia pulex* indicated little mortality and thus indicate that the water is of a fair quality.

The ICP-MS analysis of the whole water samples indicated that the water has got heavy metal concentrations above the TWQR set out by the Department of Water Affairs and Forestry. These metals however occur naturally in the system with the metal levels in the water remaining relatively constant from the source of the Klein and Groot Nyl Rivers to the furthest sampling site at Moorddrift. The water parameters such as the conductivity, oxygen content and pH also indicated little cause for concern. These factors together with the metal concentrations show that the water is of a fair quality. The determination of the present ecological state of the system indicates that although the nutrient levels in the system are on the increase, they pose little cause for concern, as the levels of toxic ammonia are still low. All these chemical constituents would indicate that the system is relatively un-impacted.

Regular monitoring of metal content and nutrient levels are not necessary and once a year should provide enough information to determine if the system is deteriorating. It is however recommended that a regular bacterial (Coliform) monitoring program be initiated. Bacterial contamination as mentioned earlier poses the biggest threat to the system.

Sediment:

The results clearly indicate the metals are not a problem in the system. Figures 3.10 (A-D) indicate a stacked graph of the percentage makeup of the different fractions for the different metals. The graphs indicate that the majority of the metals are partitioned into the 3rd, 4th and 5th fractions. Indications are that the metals generally are not very bio available. This would indicate that metals from the sediments pose little to no potential threat to the organisms in the system. This would also imply that most of the metals in the system are from a natural source.

The results also indicate that all metal concentrations fell within the lower end of the Sediment Quality Guideline Range. These will thus have little or no effect on the organisms in the system. Zinc was the only metal that had concentrations greater than the guideline values. This is however little cause for concern as they were recorded in the residual or inert fraction (Fraction 5). This would imply that they are natural concentrations in the sediment.

It may thus be concluded that the bioavailability of metals may be related to chemical form in sediment structure and not total concentration.

Regular monitoring of sediment metal content is not necessary and once a year should provide enough information to determine if the system is deteriorating.

Pesticides:

These results indicate that POP's are present in the system but the results are not conclusive, as data from sediments may not be representative of biota concentrations and cannot give information on contamination patterns in the upper levels of the food chain (Binelli and Provini, 2003). Further analysis on tissues from organisms in the system is recommended to ascertain the true extent of the pesticide contamination in the Nyl River System.

Macroinvertebrates:

Water quality may be assessed by examining the species present, the species richness or diversity (i.e. the number of species recorded at each site), abundance (number of individuals) and community structure (the relationship between organisms at different levels in the food chain). Sites such as Oilifantspruit with high species diversity can normally be considered healthy, although the ostrich

feedlot higher up in the system is a major source of organic contamination. Severe organic pollution such as at the Jasper site causes depletion of oxygen in the water and the majority of non-air breathing macro-invertebrates are largely eliminated except for species such as oligochaetes (worms) and chironomids (midges), which are tolerant to low oxygen levels. In less severe organic pollution sites such as Abba and Donkerpoort Dam the macro-invertebrate diversity has been reduced, and there is an abundance of the more tolerant organisms.

Human activities within the Nyl catchment or within the Groot and Klein Nyl Rivers itself can significantly alter the characteristics of the associated invertebrate communities. Changes in the sediment load, clearance of riparian vegetation, invasion of alien vegetation, modification of river structure and hydrological regime, and increases in nutrient and effluent input will all have an impact on the macro-invertebrate community structure. Suspended solids reduce the light penetration and therefore limit photosynthesis and affect the invertebrate composition. Sediments deposited on the river-bed can smother bottom-dwelling communities and alter the biotope suitability. Riparian vegetation supplies the majority of food in the form of organic material (leaves, bark etc.). The removal of this energy source will not only directly affect invertebrates that depend on this food source, but will also increase the amount of light penetration in areas previously shaded by overhanging vegetation. Loss of shade results in an increased algal production which smothers certain biotopes such as stones-in-current and limits macro-invertebrate communities (increase in grazers). Rises in nutrient levels from the catchment run-off (feed lots, sewage farms, septic tanks) also increases the algal production. Increase solar radiation may also raise surface water temperatures affecting certain organisms. Barriers such as farm dams and weirs alter the natural flow regime, temperatures and water chemistry. They may obstruct invertebrate drift, or movement downstream, which may affect any possible recolonisation. Agricultural practices such as monoculture crop planting (ploughing and tilling soil) and overgrazing by livestock result in increased levels of erosion and siltation; resulting in increased levels of turbidity or sediment input into the river. Sewage and agricultural products (pesticides and herbicides) contain many components, including toxic substances, which are deleterious to certain macro-invertebrates.

It should be however stressed that SASS was designed as a rapid biomonitoring tool for the assessment of water quality in flowing rivers and only allow basic comparisons of invertebrate communities present at sites. The method is sensitive primarily to organic pollution. Its relationship to other types of pollution cannot be inferred without further development and testing of the method. SASS cannot be used to establish the precise nature or cause of the impairment. For more in-depth studies, more detailed investigate research is required. This involves detailed surveys of the invertebrates, with identification to specific level, and an understanding of their ecology. Although

flowing rivers with a depositing substrate (stones-in-current) support a characteristic rich macro-invertebrate fauna, such biotopes are not always available in wetland habitats. The lack of this important biotope may severely restrict the available benthic fauna. The marginal and emergent vegetation is the most important biotope in the majority of wetland sites. The availability of marginal vegetation will fluctuate seasonally, reducing in availability during periods of low flow or during dry seasons. It is therefore necessary to investigate alternative sampling techniques and biotopes independent of the availability of natural substrates.

The use of artificial substrates or colonisation samplers should be investigated particularly in the wetland sites. Artificial substrates may provide a valid alternative sampling method for macro-invertebrates and the possibility of standardising the sampling effort in wetland habitats. Artificial substrates have been used in sampling surface and hyporheic macro-invertebrates. The artificial substrates can be used whatever the depth, the nature of the stream/wetland bed and the flow rate. More organisms (and often more species) are usually collected than the classical methods (SASS5). A preliminary wetland assessment protocol using macro-invertebrates found in the marginal and aquatic vegetation biotope and the habitat quality and surrounding land usage can be found in Chapter 7.

Amphibians:

Amphibians are an important component of South Africa's exceptional biodiversity and are such worthy of both research and conservation effort. Amphibian populations are declining throughout the world. It has become clear that declines cannot be attributed to any single causative factor and those complex mechanisms involving abiotic and biotic interactions are responsible for this phenomenon. These declines have been attributed to a combination of factors, including climate change, chemical pollution, habitat loss and disease.

Evidence for a countrywide decline in frog populations in South Africa is lacking. Amphibian declines in southern Africa have been observed, but only at the local population level, and are usually confined to areas directly impacted upon by relevant threats. Among many threats faced by amphibians in southern Africa, the most frequently implicated is habitat destruction resulting from wetland drainage, afforestation, crop farming, invasive alien vegetation and urbanisation. Like other animals, amphibians fall prey to viruses and fungi, and to parasitic infections by protozoans and various helminths.

Most frogs are intimately associated with wetlands. Much of southern Africa is semi-arid with seasonal and localised standing water and relatively few permanent wetlands. A reduction in water quality may arise from direct and indirect contamination. This may include direct chemical

contamination, or secondary run-off of petroleum and rubber compounds from roads, agricultural pesticides and herbicides, acid precipitation from atmospheric pollution, and eutrophication from fertilizer run-off. Owing to widespread pesticide use, increasing numbers of non-target species are exposed to chemical contamination. Amphibian populations are particularly susceptible to such contamination and causal relationships between pesticide usage and amphibian declines are of increasing concern.

In South Africa 88 of the 105 (84%) described species use wetland habitats. In South Africa, 19 frog species are permanent residents of wetlands or surrounding areas, 60 use wetlands for breeding and feeding during the rainy season, and 17 don't use wetlands. Destruction of essential aquatic breeding habitats negatively impacts on certain frog species and may act synergistically with factors such as habitat deterioration and fragmentation resulting in population declines.

Certain tadpoles are permanent or long-term residents in water-bodies and can be monitored quickly and economically. Different pollutants affect different systems in tadpoles. This includes substances affecting red blood cell number, neuronal activity, cell division, fertilization and a disruption in the normal biochemical pathways that lead to growth.

Proposed amphibian index (AI)

The potential use of amphibians as biological indicators is still in its infancy in Southern Africa. More intensive long-term amphibian population studies as well as intensive autecological studies need to be conducted. There is an extensive lack of accurate knowledge on the specific habitat requirements (migratory, foraging and breeding) of the majority of frog species. The tolerance levels or sensitivity to pollution of the majority of Southern African frog and tadpoles species is unknown. The use of *Xenopus* tadpoles may not be an accurate reflection of the sensitivity of other frog species to water contamination, or the effects of pesticides. *Xenopus* is widely known as a highly tolerant species often occurring in highly contaminated water bodies such as sewage purification works. The possible use of *Afrana angolensis* (Common River Frog) tadpoles should be investigated for both laboratory analysis as well as captive caged tadpoles in field experiments. *Afrana* tadpoles are found throughout the majority of South Africa throughout both the summer and winter months.

Plants:

The type of monitoring system implemented depends on the objectives of the study. From a vegetation and plant species diversity point of view there are a number of potential monitoring techniques available that may focus at different spatial scales or on different elements of the natural system.

A multi-temporal and multi-spatial scale monitoring framework is proposed here that may include some of the following components:

- Broad-scale vegetation map with landcover analysis
- Red-list species as species to monitor
- Aerial photos to see change in reed-bed size over time as indicator of siltation/ nutrient retention – system is not flushed as often or as intensively as it should be.
- Long-term NDVI data compared to rainfall data – RUE & variability in vegetation production.

When looking at the vegetation monitoring sites for species change and composition, it is especially important to note the relative contribution to cover and richness by alien versus indigenous species. A larger number of sites duplicated according to similar positions in the catchment and environmental attributes can provide useful comparative data.

Recommended further studies:

It is very important that a comprehensive inventory of the wetlands of the catchment is undertaken that includes the following:

1. Mapping of the location and size of wetlands of different types using data from sources such as aerial photography, satellite imagery, hydrogeomorphological information, landform and climate data and soil data;
2. Characterizing species composition, diversity, structure and functional attributes of vegetation of mapped wetland units from detailed ground verification surveys;
3. Capturing the information in an electronic form in a GIS database with associated vegetation and species attributes.

A baseline wetland inventory of the catchment is essential to be able to protect wetlands and ensure that the wetlands work optimally for the catchment. It would also provide an essential information source and tool for the management of the catchment.

It is recommended that use is made of historical data, such as aerial photography, to detect changes in the vegetation of the catchment. One possibility is to analyse changes in reed-bed size over time as an indicator of siltation and/or nutrient retention due to the fact that the system is not flushed as often or as intensively as it should be as a result of the proliferation of dams along the stream. This could be achieved with the use of historical aerial photographs from different dates and processed using digital photogrammetric procedures. Nutrient and substrate retention dynamics is an important functional component of wetlands and should be better understood in the Nylsvley catchment.

Floodplain vegetation undergoes seasonal changes in species composition that are relatively predictable. Long-term, regular sampling of monitoring sites can produce data that provides predictable information on the status of the vegetation. If the species composition at some point spirals out of this predictable cycle and cannot be explained by known factors, then this may be reason for concern. It is, therefore, recommended that regular sampling of monitoring sites is undertaken that covers intra- and inter-annual changes in the vegetation of the floodplain. Simple multivariate analysis of this data (using, for example, ordination methods) on an ongoing basis can provide a technique for evaluating vegetation change.

Detailed population information on many of the threatened species is required to assess and monitor them effectively.

Fish:

- Berms and channelisation in the floodplain must be rehabilitated, as these structures have a major impact on water flow and fish migration.
- Lack of sufficient culverts under roads and railways must be addressed.

The few culverts currently in place again limit flow and cause water to be hold back above the roads and railway. A further impact is on the fish population. Large predators (various bird species and in particular *Clarias gariepinus*, the sharptooth catfish) use these “bottlenecks” to prey on the smaller fish species migrating to spawn. The catfish take up position in the culverts and feed extensively on the migrating fish. One can argue that more culverts will give more opportunity to catfish to set up an ambush, but it is important that the design of culverts must be addressed, in a similar fashion that the designs of fish ways receive attention. It will be important to add some small structures in the culverts which can act as “substrate” for the migrating fish, i.e. some structures which will protect the mall fish from predators.

- Modification of the habitat in general is problematic. A concerted effort must be undertaken to address the problem and part of the management plan must focus on habitat restoration and rehabilitation.
- From the research done it is evident that the methodology developed for the RHP can be applied in a system such as the Nyl floodplain. As sampling is generally conducted during the low flow period in rivers, sampling techniques as used in the FAII can be used in the floodplain. In addition to the electroshocker, baited traps and small seine nets can be used to sample fish. Sampling during the high flow period was difficult as discussed earlier.

CAPACITY BUILDING

An important aspect of this project was the capacity development. We had a wonderful mentor in Prof Johan van Vuren (University of Johannesburg, Department of Zoology).

- ❖ The team as a whole gained insight into Nylsvley, its diverse catchment and wetlands in general.
- ❖ We had good interaction with a variety of landowners and are convinced that they learned something about the resource at their disposal, its conservation and management.
- ❖ At the University of Limpopo and the University of Johannesburg a number of students benefited from the project:
 - Support Chavalala (University of Limpopo) – was a valuable team member and gained a lot of experience through his participation, although he did a Masters degree (An assessment of the flow velocity, depth and substrate preference of selected aquatic macro-invertebrates in the Levuvhu River) that was not part of this project. He was employed by the Department of Conservation, Environment and Land (Gauteng) as the Wetland Rehabilitation Officer and more recently as an Ecologist;
 - Clayton Cook (University of Limpopo) – was an important part of the team, especially assisting younger students in the group with their projects. He was busy with his Doctoral studies (Aspects of the ecological integrity of the Debengeni and Magoebaskloof River Systems (Limpopo Province) with special reference to flow sensitive Trichoptera larvae.);
 - Richard Greenfield (University of Johannesburg) – has completed a worthwhile PhD thesis on the project (An assessment protocol for water quality integrity and management of the Nyl River Wetland System) working on water quality and pesticide pollution.
 - Ramogale Sekwele (University of Limpopo) – Completed his Masters thesis (Metal bioaccumulation in fauna of the Tobiasspruit, Nyl floodplain; Limpopo Province) and is currently employed in Natal by the Institute of Natural Resources (University of KZN) after spending some time as an Aquatic Scientist in the Northern Cape Conservation Department.

- Makoma Shai (University of Limpopo) – who is currently employed in DWAF (Water Quality section) in Polokwane completed a Masters thesis (An assessment of ecological disturbances of the Nyl floodplain) as part of the project. She gained tremendously by working in this environment.
 - Liesl Moolman (University of Johannesburg) – also completed a Masters thesis during our work on the project (The use of selected freshwater gastropods as bio-monitors to assess water quality). Although the project was not funded from the WRC programme, this work on the Nyl floodplain has provided valuable information on the use of indigenous organisms as biomonitoring tools.
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- ❖ During the study, various third year and Honours (University of Limpopo and University of Johannesburg) students were exposed to the work of the post graduate students and this has stimulated a greater interest in water research – not easy to quantify, but more students are approaching the Department to do post graduate studies in freshwater ecology.
 - ❖ Various newspaper articles and radio interviews resulted from the project.
 - ❖ All the authors presented papers at national and international conferences and the students were exposed to many national conferences where they presented their work, first as posters during the initial phase of study and later as oral presentations.
 - ❖ We have a great working relationship with the “Friends of Nylsvley” and they contact me regularly for comments with regard to developments (property and golf courses) in the area. We are busy setting up a forum with the EIA section of Limpopo Conservation to discuss issues on a more regular basis.

In general, it is our feeling that the floodplain has gained recognition as a significant area that needs protection.

Dr. Wynand Vlok

April 2006

ACKNOWLEDGMENTS

- The Water Research Commission for the funding of the project.
- Dr Steve Mitchell for his support, guidance and patience – thank you Steve.
- All the members of the Steering Committee for their inputs.
- The farmers for their cooperation and permission to use their land.
- Prof Victor Wepener for his assistance and guidance in the study on water quality.
- Support Chavalala, Charles Sekewele and Caroline Shai.
- Prof Pieter Olivier for his assistance during the initial planning of the project.
- The project team, all who worked hard during the surveys. A special thanks to Richard and Clayton.
- The various institutions represented by the team: the University of Limpopo, the University of Johannesburg, the South African National Biodiversity Institute and the consultants.
- Prof Braam van Wyk for his inputs in the planning phase as well as the work with Janine, David and Tony.

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CHAPTER 1: HABITAT: DESCRIPTION OF SAMPLING SITES FOR WATER, SEDIMENT, FISH AND INVERTEBRATES**1.1 Introduction:**

During the study, a total of eighteen (18) sites (Figure 1.1) were used for the collection of samples. Eight (8) sites were used for the sampling on the fish and invertebrates. The sites covered the entire Nyl system; including sites in the upper catchment of the system. The reasoning was that we wanted to include some sites where the potential pollution of the water resources will be limited. For this reason, we selected sites near the origin of the Groot Nyl River, the Klein Nyl River and another important tributary of the Nyl River system, the Olifantspruit. Some of the sites are considered to be “riverine sites” (Donkerpoort, Abba, Groot Nyl, Jasper and Olifantspruit), whilst the remainder of the sites were selected in the floodplain habitat (Vley, Haakdoorn and Moorddrift). The aim was to determine the current fish and macro-invertebrate species present in the rivers and compare it to the historical fish surveys conducted on the system (Kleynhans, 1990). This will then be used as the basis of the current status (absence/presence) of fish species and compare that to the fish assemblages found in the floodplain sites.

At each of these sites a habitat description of the site was done. During the sampling all major disturbances or impacts were noted. In-stream physical parameters collected as part of the survey included; flow velocity, depth and width of the stream. Fixed point photographs were taken over the study period as reference to the current study.

The sites selected for the fish and macro-invertebrate surveys were: Donkerpoort and Abba (upper reaches of the Klein Nyl River), Groot Nyl (middle reaches of the Groot Nyl River), Olifantspruit (in the Olifantspruit, a tributary of the Nyl system above the Nylsvley Nature Reserve), Jasper (Nyl River - after the confluence of the Groot and the Klein Nyl, but before the confluence of the Olifantspruit), Nylsvley (in the nature reserve), Haakdoorn (downstream of the nature reserve and Mookgopong) and Moorddrift (just above Mokopane). Water and sediment samples were also collected at these sites.

The other ten (10) sites were used for the sampling of water and sediment and are: Klein Nyl Oog (origin of the Klein Nyl River) and Koh-I-Noor (an unnamed tributary of the Klein Nyl flowing into the Donkerpoort Dam from the north), Groot Nyl Oog (close to the origin of the Groot Nyl River), Sewerage Works (below the effluent pipes of the sewerage works in Modimolle), Hessie-se-Water (just upstream from the confluence of Hessie-se-Water with the Olifantspruit), Bad-se-Loop (a tributary feeding into the Nyl system downstream of the reserve), Tobias Oog (close to the origin of the Tobiasspruit on the farm Bergland), Tobias (just upstream of the confluence of the Tobiasspruit and the Nyl floodplain, but below the confluence of the Naboomspruit), Mine (just below the fluorspar mine north-east of Mookgopong on the Naboomspruit), Mosdene (below the private reserve Mosdene in the flood plain area, below the inflow of run-off and sewerage effluent from Mookgopong).

1.2 Description of the sample sites:

(a) Fish and invertebrate sample sites (water and sediment collected as well).

1.2.1 Donkerpoort site (S 24° 40,542 E 28° 20,019)

This site is in the Klein Nyl River is approximately 2 kilometres below the Donkerpoort Dam (S 24° 40,233 E 28° 19,338). The site is situated on the farm Donkerpoort (406 KR) at a low weir (approximately 1,5m height). A section of the weir (towards the left bank) was damaged and washed away during previous floods (Figure 1.2). This weir is just upstream from the road bridge when travelling on the gravel road to the dam. This gravel road turns off from the Vaalwater/Modimolle road.

Biotopes sampled for fish and macro-invertebrates included: pools with a sand, gravel and mud bottom; shallow fast and slow runs; a deep run (medium fast flow velocity); marginal vegetation and a small cascade where the weir has been washed away (shallow fast). The pool situated above the weir reaches a maximum depth of 1,5m and the bottom is composed mainly with large bedrock areas covered in silt, gravel and sand.

Below the weir, towards the left bank, fish and macro-invertebrates were collected from the cascade and rapids downstream. Due to the break in the weir, this cascade/rapid contained water for the duration of the surveys. Towards the end of the project, an alternative site was selected at the road bridge (S 24° 40,631 E 28° 20,158); due to the fact that access to the farm became more difficult. It was not always possible to contact the owner to get the keys to the locks on the gate.

The vegetation on the banks is dominated by several large clumps of *Eucalyptus* trees with limited forb undergrowth and grasses and sedges on the riparian banks. The majority of *Eucalyptus* has been mechanically eradicated (ring-barked by “Working for Water”) and the potential for rehabilitation looks promising.

1.2.2 Abba site: (S 24° 40,800 E 28° 16,746)

This site is situated in the upper reaches of the Klein Nyl River, about 3 kilometres upstream of the Donkerpoort Dam, near the Abba Game Lodge (farm Vaalkop 405 KR). During the initial surveys, sampling was done at the road bridge (Figure 1.3). The habitat of the site consists of a deep slow pool/channel, approximately 3 meters wide, but the water pushes into the reeds and sedges (5 – 7 meters wide, depending on flow volumes).

The substrate consists mainly of mud and a few small stones (10 – 20 cm in diameter) with a depth of more than a meter in the centre of the channel. Dense marginal vegetation (reeds) covered the whole width of the broader channel. The pool was later silted up (Figure 1.4).

An alternative site was selected downstream from the original site during the 2002 sampling season. This site (S24° 40,114 E 28° 18,204) was about a kilometre from the first site. The biotopes of the site were restricted to a slow deep run and marginal fringing vegetation. The site proved more reflective of the fish communities present and more specimens were collected. This may be due to the fact that it was possible to wade for longer distances during sampling (not possible at the first site).



Figure 1.2: Donkerpoort site in the Klein Nyl River, downstream from the Donkerpoort Dam.



Figure 1.3: The Abba site near the game lodge (in the background) showing the site with dense vegetation (dominated by typha).



Figure 1.4: The deep pool at the initial Abba site pool silted up after the 2003 rainy season.



Figure 1.5: The Groot Nyl site, showing the narrow channel with the muddy/sandy substrate.

1.2.3 Groot Nyl (S 24° 45,645 E 28° 20,757)

This site is situated on the farm Modderpoort (454 KR), on the Groot Nyl River, just upstream from the road bridge on the Bela-Bela/Modimolle road. The narrow channel (2 – 4.5 meter wide) is dominated mainly by a sandy bottom. The biotopes sampled included marginal vegetation, a pool and the run (fast and slow) with occasional shallow fast run.

The banks are dominated with *Eucalyptus* trees (some ring-barked) and some sedges and grass. The water depth varied between 0.19 and 0.60 meter and a constant moderate flow.

1.2.4 Olifantspruit (S 24° 39,191 E 28° 28,596)

Originally this site was considered as one of the reference sites, as it was believed that the site contained relatively few impacts from the surrounding catchment area. The site is on the farm Rietspruit (412 KR) on the gravel road turn-off Groenkloof – on the Modimolle/Mookgopong road. At the second river crossing, the site is approximately 100 meters downstream from the bridge. Two alternative sections were sampled. Initially during the first two surveys (2001), the area around the bridge was sampled. Due to massive reed invasion sampling became ineffective. An alternate site was selected downstream from the bridge.

The biotopes at the road bridge site include a deep pool (substrate mostly mud and gravel) upstream of the bridge (formed by the debris clogging the culverts of the bridge) and a fast shallow cascade downstream of the bridge (Figure 1.6 and 1.7). Fringing marginal vegetation was sampled around the pool and along the margins of the cascade.

The second site consisted of an artificial deep pool above a rapid. The landowner had previously constructed low water bridge at the top of the rapid, damming the water upstream of the structure for a cattle drinking point (Figure 1.9). The resultant change in hydrological flow created a deep pool with dense reeds on the banks. The rapid consist of a few shallow pools below the river crossing and various shallow, fast flowing channels cascading into a small pool. Riparian vegetation comprised limited pockets of natural vegetation (*Acacia karroo*) as well as exotic tree species, *Melia azedarach* (Syringas). Reeds, grass and a few sedges form the bulk of the marginal bank vegetation. The bottom substrate consist of bedrock with bolder and cobbles. Course gravel was also present in the shallow pools. Limited backwaters (depending on flow depth) formed near the right hand bank and the bottom substrate was dominated by sand and mud.

After the completion of the initial project, another project was implemented on the Olifantspruit (MSc project). During the initial visitation for sample site selection; a huge ostrich farm, right on the banks of the river was observed. The operation raise between 4 000 and 4 500 birds per season in a feedlot system. All the faeces and the run-off from the feedlot end up in the Olifantspruit. During a recent site inspection (October 2004), it was discovered that the current landowners has cleared all the vegetation on the rivers banks around the road bridge site (Figure 1.8). Massive siltation and sediment deposits were evident downstream from the impacted site.



Figure 1.6: The Olifantspruit site, showing the dense vegetation downstream of the bridge.



Figure 1.7: The deep pool formed by the bridge.



Figure 1.8: The reed bed after clearing in 2004.



Figure 1.9: The second site selected a 100 m downstream from the road bridge site.

1.2.5 Jasper (S 24° 42,552 E 28° 28,794)

This site is situated on the Nyl River (downstream of the confluence of the Groot and Klein Nyl rivers) at the road bridge on the farm Doordraai (415 KR). The site is reached from the Modimolle/Mookgopong road at the turn off to Jasper railway siding. This site was constantly impacted from effluent from Modimolle (run-off), the sewerage works at Modimolle and effluent and run-off from the informal settlement around Phagameng.

Above the road bridge, the site is dominated by a shallow drift (pebbles, cobbles and sand) and pool (a muddy substrate, 8 – 10 meters wide) and further upstream the river narrows to form a deep run (cobbles) and pools (sand and mud) (Figure 1.10). Downstream of the bridge, a narrow rapid (2 – 4 meters wide) is followed by a deeper run near the left bank and a shallower rapid on the right-hand bank. Pools and rapids occur further downstream. Marginal vegetation consisted of reeds, grasses and sedges with small patches of reeds growing on isolated small “islands” in the river channel (Figure 1.11).

1.2.6 Nylsvley (S 24° 38,940 E 28° 41,419)

The sampling was conducted at the small weir (Figure 1.12 and 1.13) in the nature reserve (Nylsvley 560 KR), but later sampling site shifted to the Jacana hide (S 24° 38,864 E 28° 41,757) (Figure 1.14) and the bird-hides (Dragonfly, Dabchick and Crake) at Vogelfontein (S 24 36 894 E 28 41 520) Figure 1.1 and 1.16). This was done to get a more representative fish and macro-invertebrate sample in the reserve.

The site at the weir consisted mostly of a small stream, 1 – 3 meters wide. The weir (stone and soil) is partially destroyed from previous flooding activities. During moderate flow rates, water is retained above the weir, forming a pool. At the point where the weir was damaged, a stone foundation is still present retaining water even at low flows. The substrate consists of mud and fine gravel with rye grass the dominant vegetation (biotypes sampled: pool, marginal vegetation and the run - deep or shallow, depending on flow rates and volumes).

At the Kingfisher hide, the biotopes sampled included a deep pool that formed during moderate to high flows, or the run during low flows, and the marginal vegetation. The site at Vogelfontein is situated in the reserve (south of the road) and consists of a large pool, dammed behind the artificial berms created to retain water and walk-ways to the bird-hides. The bottom substrate consists mainly of mud. Fringing marginal vegetation and some floating macrophytes were present. At the hide outside the reserve (Dragonfly) (north of the road) a smaller impoundment was sampled. During the low flow surveys, it consisted of a shallow pool with a muddy substrate and dense aquatic macrophytes (reeds and water-lilies). The small inflow stream was also sampled during the study. Macro-invertebrates were not collected at any of these sites.

1.2.7 Haakdoorn (S 24° 25,804 E 28° 54,711)

This sampling site (Figure 1.17) is situated on the farm De Hoop (334 KR) and consists of an artificial impoundment formed by the earthen-wall constructed in the river channel. The water depth varied between 0.5 and 2.5 meter, depending on flow velocity. The bottom substrate consists of mud and the water is covered with a variety of macrophytes. The biotopes sampled included the pool (muddy substrate) and vegetation.



Figure 1.10: The Jasper site, upstream of the road bridge, showing the shallow pool area.



Figure 1.11: Downstream of the bridge, the run and shallow rapid.



Figure 1.12: The pool formed behind the old weir at the site in the NNR.



Figure 1.13: A view of the weir during the low flow season, with the broken section indicated by the arrow.



Figure 1.14: The sampling site used at the Jacana Bird Hide in the NNR.



Figure 1.15: A view of the shallow impoundment at Vogelfontein (Dragonfly hide).



Figure 1.16: The shallow pool formed by the berms at the Dabchick and Crake hides.



Figure 1.17: The impoundment at Haakdoorn – a spill over was constructed in the two-meter high earthen wall, which retains a large quantity of the flood water.

1.2.8 Moorddrift (S 24° 15,175 E 28° 58,521)

This site is situated on the farm Moorddrift (289 KR) and is dominated by a large impoundment (earthen-wall – Figure 1.18)) constructed upstream from the Nyl pans. Sampling was conducted on the shoreline in the shallow water between the macrophytes (reeds). Biotopes sampled for macro-invertebrates included the muddy bottom and vegetation.



Figure 1.18: The impoundment at the Moorddrift sampling site.

Description of the sample sites: (b) Water and sediment sampling sites:

1.2.9 Klein Nyl Oog (S 24° 42,967 E 28° 14,542)

This site (De Nyl Zyn Oog – 423 KR) is situated in one of the many springs and sponge areas feeding the Klein Nyl River (Figure 1.19 and 1.20). Water and sediment samples were collected from small artificial impoundment constructed below the sponge area.

The site is situated on the gravel road past the Abba Game Lodge, just before the cross road to Modimolle/Bela-Bela. The farm is located to the right of the road opposite the residential area.

1.2.10 Groot Nyl Oog (S 24° 46,959 E 28° 15,290)

The site is situated on the farm Groot Nyls Oog (447 KR) near the origin of the Groot Nyl River (Figure 1.21 and 1.22). The site was selected opposite bridge due to access problems. Current owner of the property does not reside on the farm. Water samples were collected upstream of the road bridge. The site contained water throughout the sampling period.

1.2.11 Sewerage Works (S 24° 42,335 E 28° 25,747)

This site was selected just below the effluent pipes (some illegal) of the Modimolle Sewerage Works (Figure 1.23) on the farm Nylstroom (419 KR) on the Klein Nyl River. Access to the site was gained through the property of the sewerage works.

This site was sampled to determine the effectiveness of the sewerage works as well as an indicator of the presence of bacterial pollution in the Klein Nyl River. Another major impact occurring at this site was the storm water run-off from Modimolle Township into the river.



Figure 1.19: The small impoundment below the spring where water was sampled.



Figure 1.20: The small seep formed after rain in the area of the spring, feeding into the impoundment (Figure 1.19).



Figure 1.21: The grassland area showing the seeps and springs giving rise to the Groot Nyl River.



Figure 1.22: The sampling site upstream of the road bridge.



Figure 1.23: The site near the sewerage works in Modimolle, just downstream of the various effluent pipes.



Figure 1.24: The site, Koh-I-Noor, selected in one of the tributaries of the Klein Nyl River.

1.2.12 Koh-I-Noor (S 24° 38,812 E 28° 17,813)

This site is on the game farm, Koh-I-Noor (Figure 1.24), on the road between Vaalwater and Modimolle (Nooitgedacht 404 KR). One must follow an indistinctive vehicle track towards the mountainous areas. The site was situated on the tributary of the Klein Nyl River in the pool below the waterfall. This site was selected as a reference site of water quality in the head waters of the Nyl River system. The tributary flows into the Donkerpoort Dam.

1.2.13 Hessie-se-Water (S 24° 33,948 E 28° 28,408)

This site (on the border of the farms Rietspruit and Olifantspoort 414) was near the confluence of Hessie-se-Water and the Olifantspruit (Figure 1.25) and was selected to indicate the impacts of various activities in the catchment area on water quality and as a comparative site for Olifantspruit. The sampling was conducted downstream of the road bridge crossing Hessie-se-Water on the Modimolle/Mookgoong road.

1.2.14 Tobiasoog (S 24° 28,995 E 28° 35,933)

This site is situated on the farm Rietfontein (513). Currently, the portion is known as Bergland and the site was approximately 2.5 km downstream from the origin (spring) of the Tobiasspruit (Figure 1.26). This site was selected as a reference site to evaluate the impacts of surrounding mining activities on the Naboomspruit.

1.2.15 Mine (S 24° 28,342 E 28° 40,894)

The site is on the road between the hot springs and a holiday resort and Mookgoong township. The tar road crosses the Naboomspruit, just downstream on the fluorspar mine. The site was selected to determine the impact of run-off and possible leaching from the mining activities. The site (Buffelsfontein 347 KR) was at the road bridge, downstream from the mine and the weir on the southern site of the road (Figure 1.27).

1.2.16 Tobias (S 24° 27,504 E 28° 45,080)

The site in the Tobiasspruit was situated on the farm Tobias Zyn Loop (339 KR) on the Mookgoong/Mokopane road (Figure 1.28). The site was selected below the confluence of the Naboomspruit and the Tobiasspruit to determine the cumulative effects of the two rivers water quality. The site was located upstream from the confluence with the Nyl floodplain.

1.2.17 Bad-se-Loop (S 24° 34,448 E 28° 38,687)

This site was situated on the Bad-se-Loop road between Modimolle and Mookgoong (Figure 1.29). The site is located on the farm Vischat (520 KR) at the road bridge.

This site was selected as one of the pristine sites with limited impacts currently influencing the water quality and the aim was to determine its impact or value on the floodplain.

1.2.18 Mosdene (S 24° 33,950 E 28° 45,850)

Situated on the farm Du Toits Kraal (532 KR) is the Mosdene Private Reserve. This site is located downstream from the Nylsvley Nature Reserve on the road between Roedtan and Mookgoong (Figure 1.30). The storm water runoff and sewerage effluent from the sewerage works impact on this site. A further impact to the water quality is the potential sewerage runoff from the informal settlement to the east of Mookgoong.



Figure 1.25: The site, Hessie-se-Water, during the low flow season.

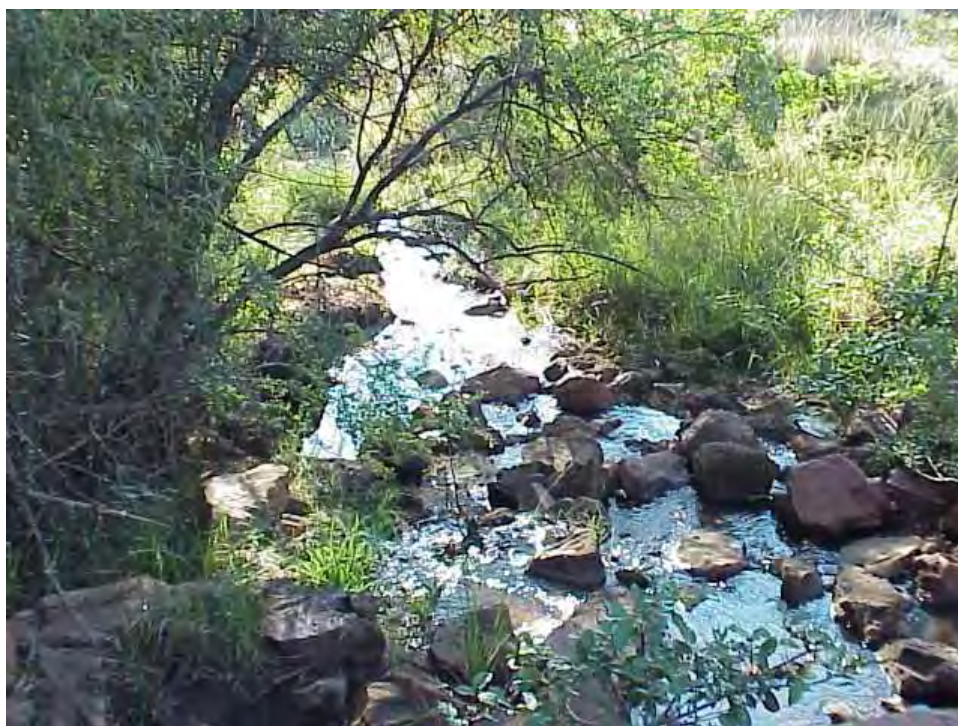


Figure 1.26: The site a short distance from the springs forming the Tobiaspruit.



Figure 1.27: The site below the Mine (S 24° 28,342 E 28° 40,894



Figure 1.28: The Tobias site, showing the river crossing underneath the old road between Mookgopong and Mokopane.



Figure 1.29: The Bad-se-Loop site, showing the river downstream of the road bridge.



Figure 1.30: The site at the private reserve, Mosdene, showing the small pools formed in the floodplain.

CHAPTER 2: WATER

2.1 Introduction:

The analysis of the water in the wetland is vitally important as it forms the life's blood of the system. It provides habitat for most of the organisms involved in the system, both terrestrial and aquatic. Many of the invertebrates in the systems depend on the hydrosphere for all or part of their life cycle. The chemical characteristics of the water indicate the level of contamination in the system. This is important as it provides vital information on the suitability of the systems water for both animal and human consumption and the provision of a suitable living medium for aquatic organisms.

The water in the system was analysed for the following chemical and physical parameters: Water parameters, inorganic constituents, organic constituents, toxicity, bacteria concentrations and metal concentrations. These analyses form a suite of tests that are carried out on the water to provide a holistic indication of the levels of contamination and the suitability of the water for the many users involved.

2.2 Materials and methods:

2.2.1 Field Work:

Field sampling trips were conducted between March 2001 and March 2003. These trips took place on a quarterly basis and incorporated the sampling of water, sediment and aquatic invertebrates.

Water samples were collected in 500ml plastic bottles. Two bottles were collected for different analyses. The water in one bottle was not treated and used for further analysis of macro variables and organic constituents. The water in the second bottle was treated with 5 ml nitric acid to preserve the metal content in the water. Once the water samples were collected they were placed on ice until they could be frozen. Once frozen the water samples were returned to the laboratory for further analysis. A further 2l of water was taken in plastic sample bottles for toxicity testing back in the University of Johannesburg (Kingsway Campus) Aquarium facility. These samples were also frozen till the testing could take place in the University of Johannesburg (Kingsway Campus) environmental control facilities in the aquarium. The water parameters were also recorded. These were taken using a variety of Eutech instruments. The water parameters recorded were O₂ (concentration and saturation), pH, Total dissolved solids (TDS), conductivity and temperature. The oxygen concentrations and saturations were determined using the Eutech Cyberscan DO 300 oxygen meter, the pH was determined using the Eutech Cyberscan pH 300 meter and the conductivity and TDS were determined using the Eutech Cyberscan con 410 conductivity and TDS Combination meter. All three meters contained built in thermometers. The meters were calibrated according to the instruction manuals with standards provided by Selectech. The pH meter underwent a 3 point calibration using the 4, 7 and 10 pH standards. The conductivity meter was calibrated to 1413 µs standard and the TDS calibration standard was the 300 ppm standard. The oxygen meter was calibrated according to altitude and air saturation (van Vuren *et al.*, 1994).

2.2.2 Laboratory work:

In the laboratory the different samples were analysed for the required information. Prior to analysis all glassware and plastic containers were washed according to the protocol prescribed by Giesy and Wiener in 1977.

The pre-washed apparatus was placed in a phosphate free soap bath for 24 hours containing a 2% concentration of Contrad soap (Merck). It was then removed, rinsed with distilled water and placed in a 2% HCl acid (Merck) bath for 24 hours. The glassware and plastic

containers were then rinsed with distilled water and placed on drying racks and allowed to air dry. The clean apparatus was then placed on racks for storage until needed.

2.2.2.1 Metal Analysis:

Water samples were analysed at Waterlab in Pretoria for metal content, nutrients and inorganic constituents and bacteria content. The acidified water samples were analysed for metals using Inductively Coupled Plasma Mass Spectrophotometry (ICP-MS). These samples under went a scan and were analysed for 70 elements.

2.2.2.2 Nutrients and macro variables:

The non-acidified water samples collect were defrosted and then analysed for nutrients using the standard methods prescribed by the American Public Health Association (APHA, 1998). The following macro variable levels were determined: Chloride, Sulphates, Ortho-phosphates, Nitrites, Nitrates and Fluorides. Table 2.1 indicates the macro variable and the APHA methods used in analysis.

Table 2.1: Table of APHA methods used for the determination of water macro variables.

Macro Variable	APHA Method
Chlorides	4500-Cl B. Argentometric Method
Sulphates	4500-SO ₄ ²⁻ E. Turbidimetric Method
Flourides	4500-F ⁻ C. Ion-Selective Electrode Method
Ortho-phosphates	4500-P D. Stannous Chloride Method
Nitrates	4500-NO ₃ ⁻ H. Automated Hygrazine Reduction Method
Nitrites	4500-NO ₂ ⁻ B. Colometric Method

2.2.2.3 Bacteria:

Prior to sampling the bottles were sterilised. The clean glass bottles were sterilised in an autoclave with a 1mℓ/100mℓ sodium thiosulphate solution. Heterotrophic plate counts, faecal coliform and total coliform counts were then carried out on the samples according to the methods used at Waterlab (APHA, 1998). The different counts were done by culturing the bacteria present on the appropriate agar based medium in an incubator and then the developed colonies were counted and expressed as standard plate counts per mℓ.

2.2.2.4 Toxicity testing:

The use of toxicity testing is important in the assessment of water throughout a system. The tests indicate the response of living material to the total effect of actual and potential disruptions in water, and thus complement the chemical analysis in monitoring harmful chemicals in water (Blaise *et al.*, 1988). The chemical analysis of water indicates what constituents/contaminants the water contains of and if they pose a potential problem in the water to aquatic organisms. It does not however give an indication to the cumulative effects of the constituents/ contaminants to organisms inhabiting the water. The use of toxicity testing can give an indication of these cumulative effects on the organisms placed in the water (Slabbert *et al.*, 1998). It was thus decided to subject the water to acute toxicity testing using both the water flea, *Daphnia pulex*, and the guppy, *Poecillia reticulata*, as test organisms.

Toxicity tests were carried out in an Environmental room at the University of Johannesburg (Kingsway Campus) aquarium. These tests were carried out according to the guideline set out by the Institute for Water Quality Studies (IWQS).

i) *Daphnia* culturing:

The *Daphnia pulex* was cultured in a separate environmental room to the one in which the toxicity testing took place. The culture was kept at a temperature of approximately 20 °C. A photoperiod of 16 hours daylight was also maintained. *Daphnia* cultures were kept in 3 litre glass beakers in a reconstituted medium to promote culture growth. The culture was refreshed every 3 weeks to keep daphnia densities in the media down to promote reproduction.

The *Daphnia* media was made up according to method 3001 001 of the Acute Toxicity Assessment Methods Manual (Truter, 1994). The medium used was moderately hard, reconstituted water. It was made up by placing 1ℓ of stock reagent into 19 ℓ of double distilled water and the medium was then aerated for 24 hours before use. New medium was made up weekly. The following stock reagent was used: NaHCO₃, CaSO₄.2H₂O, MgSO₄.7H₂O and KCl.

The stock reagent was made up as follows: 2.59g of NaHCO₃, 1.2g of CaSO₄.2H₂O, 0.08g of KCl and 2.46g of MgSO₄.7H₂O was dissolved in 1ℓ of deionised water in a volumetric flask.

2500 ml aerated medium was then added to a 3000ml glass beaker and 5 ml of food was added before the transfer of *Daphnia* took place. The feeding of the *Daphnia* culture is very important and fresh food was made up weekly. The *Daphnia* food is a suspension of trout pellets, alfalfa and brewers yeast. Food was made up as follows: 6.3g of trout pellets, 2.6g of dried yeast and 0.5g of alfalfa was added to 1ℓ of warm deionised water. The food was left to stand for approximately 15 minutes to allow the pellets to soften and then placed on a magnetic stirrer for about 2 hours. This allowed the particles to go into suspension. The food was then placed in the fridge over night and the following morning the top 500ml was poured off into a 500ml volumetric flask leaving solid particles behind in the preparation beaker. The food was kept in the fridge. *Daphnia* were fed three times a week on Mondays, Tuesdays and Fridays. The food was left to reach room temperature before it was introduced into the cultures. 5 ml of food was pipetted into each culture beaker.

ii) Guppy culture.

Guppies were bought from a fish farm near Polokwane and held in the aquarium for less than a week before being used. The guppies used in the tests were between 1 and 2 weeks old.

iii) Test Media

The tests were carried out according to the guidelines set out by the IWQS for both guppies and *Daphnia* (IWQS, 1998). Both diluents used in the tests were moderately hard reconstituted water, where the *Daphnia* culture medium was used for the *Daphnia* tests and guppy medium was used for the guppy tests. The preparation of the *Daphnia* medium preparation has been explained earlier in this chapter.

iv) Guppy medium:

The preparation of the guppy medium was done according to method 3001 002 in the Acute Toxicity test methods manual (IWQS, 1998). Four stock solution for the preparation of reconstitutes moderately hard water were prepared using milli-Q water.

The four stock solutions were a sodium hydrocarbonate solution, a magnesium sulphate solution, a calcium carbonate solution and a potassium chloride solution. Each stock solution was prepared in 1ℓ volumetric flasks and contained:

11.76g of CaCl₂.2H₂O diluted to 1ℓ.

4.93g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ diluted to 1ℓ.

2.59g of NaHCO_3 diluted to 1ℓ.

0.23g of KCl diluted to 1ℓ.

250 ml of each of these four stock solutions was then added to a container containing 19ℓ of de ionised water and aerated over night to allow stabilisation of the medium and for the medium to attain the required room temperature of 21 degrees.

v) Toxicity testing:

Acute screening toxicity tests were performed on all the water samples collected at the different localities. These tests were carried out every three months for a period of a year as part of a monitoring programme for the water. For the *Daphnia* the 48hour acute static screening tests were used and for the guppy the 96 hour acute static screening tests were performed. Rand (1995) describes a screening test as a short test usually in the early stages of a programme to establish the potential of an effluent or chemical to elicit an adverse effect. All tests were carried out in duplicate for confirmation of mortality rates (Truter, 1994).

The effluent or stream water tested was defrosted and left to reach room temperature before testing took place. *Daphnia* tests were carried out in 40 ml glass beakers and guppy tests in 600ml glass beakers. Tests were conducted at both the 100 percent and 50 percent dilution levels. All dilutions were made with reconstituted water. Positive controls were run for both tests to monitor the medium toxicity. As these were acute tests the feeding of organisms was not necessary during the test period.

Daphnia pulex test

In the case of the *Daphnia* tests 40 ml of stream water was used in the 100 percent test and for the 50 percent test 20 ml stream water and 20 ml of medium were added to 40 ml glass beakers. The water variables were then read using the Eutech range of water quality meters before the *Daphnia pulex* were added. 5 individuals were added to each beaker, individuals were approximately a day old. The water quality was monitored every day and mortalities noted. The mortalities after the completion of the tests were then analysed using the Spearman Karber computer program and the lethality concentration 50 (LC50) was determined (Truter, 1994).

Guppy test

In the case of the guppy test 400ml of stream water was used in the 100 percent test. For the 50 percent test 200ml of stream water was added to 200ml of guppy medium. Water quality parameters were then read using the Eutech range of water parameter meters and the test organisms were then added. Five guppies (week old) were placed in each beaker and their mortalities were recorded. The mortalities were also analysed using the Spearman Karber computer program to determine the LC 50.

2.3 Results and discussion:

Historical data:

Reference conditions for the Nyl River system were determined using the method of Bath *et al.* (1999)¹. This method was developed for the determination of reference conditions for water quality variables to account for 1) the influence of point and diffuse sources, 2) the lack of consistent spatial and temporal data, and 3) natural changes in quality along the length of a river (Bath *et al.*, 1999)¹.

Historical data was obtained from the Department of Water Affairs and Forestry (DWAF⁹). The sites chosen were taken from available data with the sites covering the main river and the more important tributaries. The data obtained dated from 1972 to September 1999. Data from gauging stations at 7 localities was used to obtain a representative reference condition for the Nyl River system or desired state. The seven localities used are listed in table 2.2 with gauging station numbers.

Table 2.2: List of gauging stations to determine reference conditions and number of samples used.

Station Name	Station number	Total Sample size	Sample size period one	Sample size period two
Klein Nyl at Modimolle	A6H006Q01	178	48	130
Donkerpoort Dam	A6r003Q01	63	31	32
Hessie-se-Water	A6H019Q01	113	51	62
Olifantspruit	A6H012Q01	221	76	145
Nylsvley at Deelkraal	A6H002Q01	47	9	38
Bad-se-Loop	A6H010Q01	214	50	164
Tobiasspruit	A6H023Q01	25	19	6

From the figures obtained (Figures 2.1-2.21) two time periods were determined. These time periods were determined visually from the graphs judging from the visible onset of the increase in concentration/level. These periods ran from February 1972 to January 1984 and from February 1984 to September 1999.

These reference conditions cannot be used in a full reserve determination as they are calculated for a single period and not monthly as one would conduct for a full reserve determination. They are however suitable to act as a guideline for comparisons of sampled data and to denote changes in the overall status of the system.

The trend lines obtained indicate that the water quality of the system has deteriorated over the last 20 years. This is true for most of the variables at most of the sites. There are however a few exceptions. These polynomial trend lines are indicated in figures 2.1 to 2.21. The positive trend indicates an increase in the level/concentration of the specific variable, where a negative trend indicates a decrease in the variable level/concentration. These trend lines are however not regression lines and thus no formulae can be determined. The temporal changes can be caused by a number of factors.

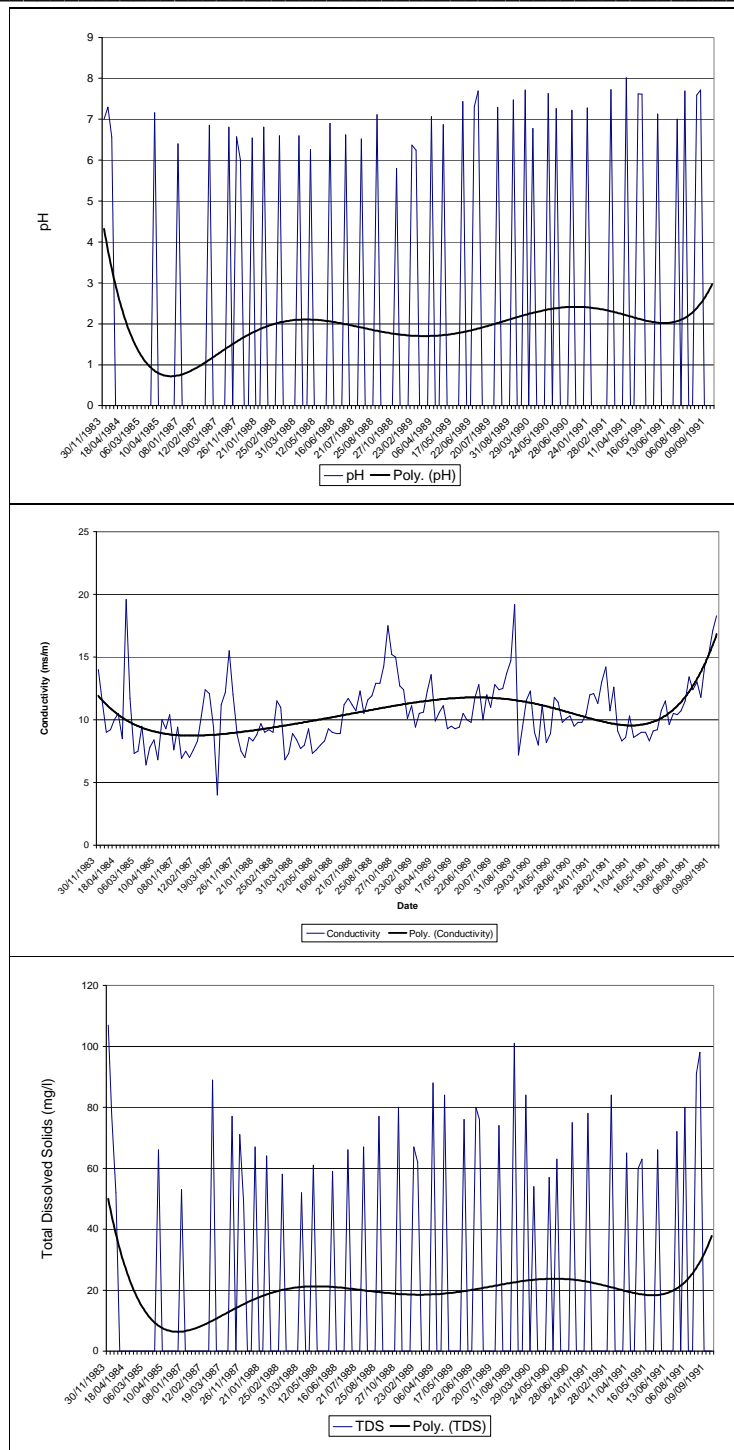


Figure 2.1: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Klein Nyl River at Modimolle (KNN).

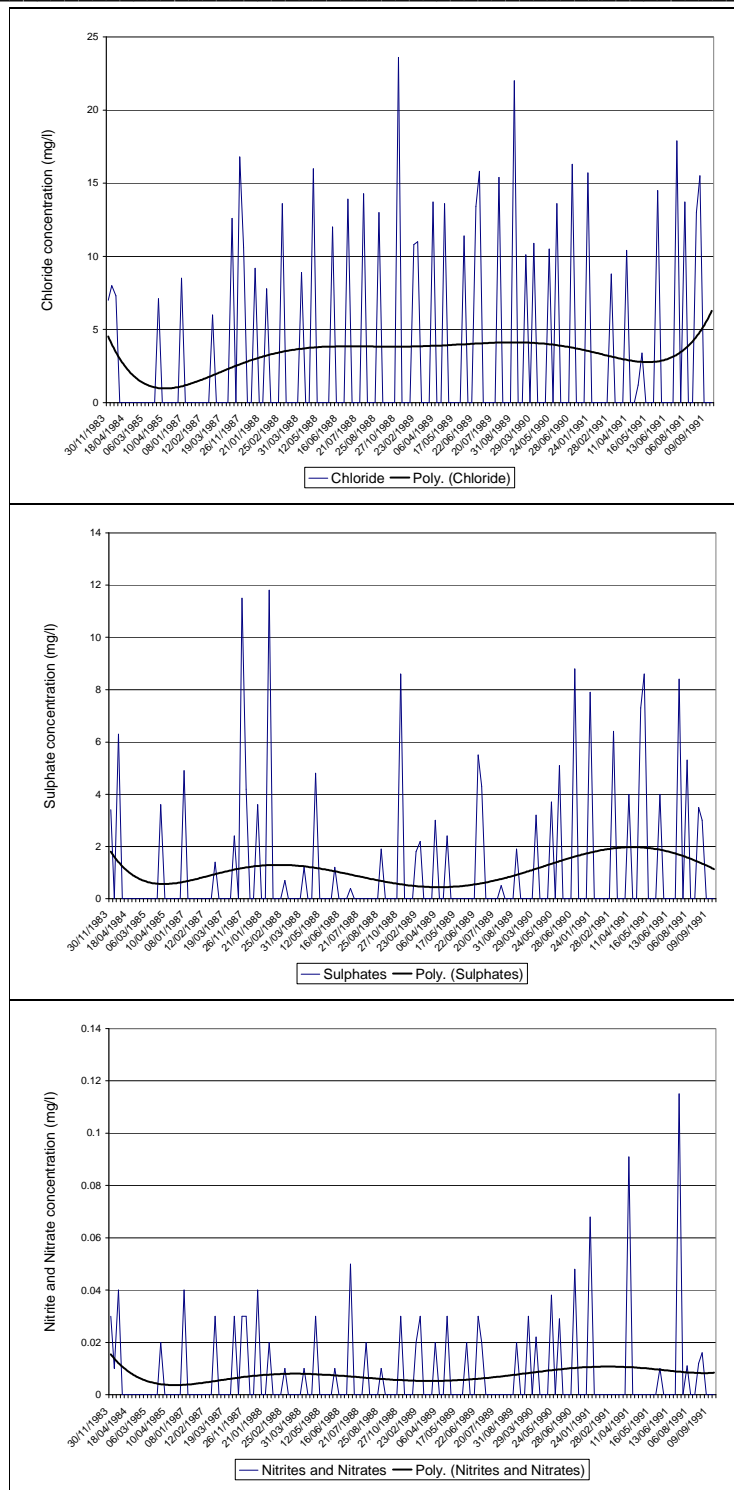


Figure 2.2: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Klein Nyl River at Modimolle (KNN).

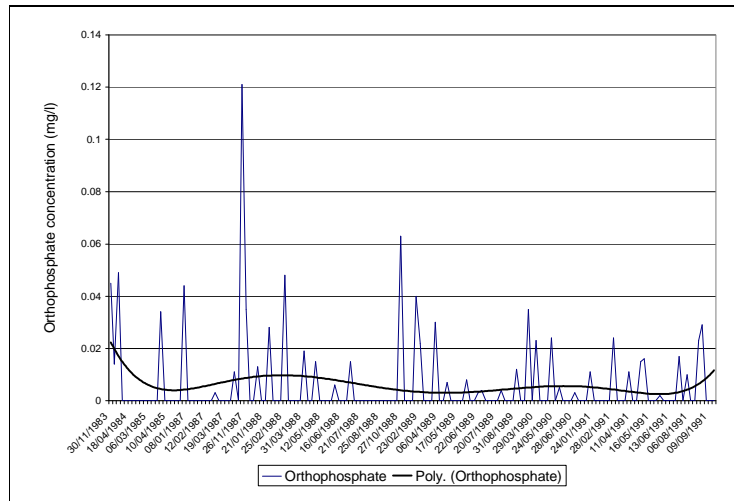


Figure 2.3: Graphic representation of historical data on a temporal scale. Orthophosphate at Klein Nyl River at Modimolle (KNN).

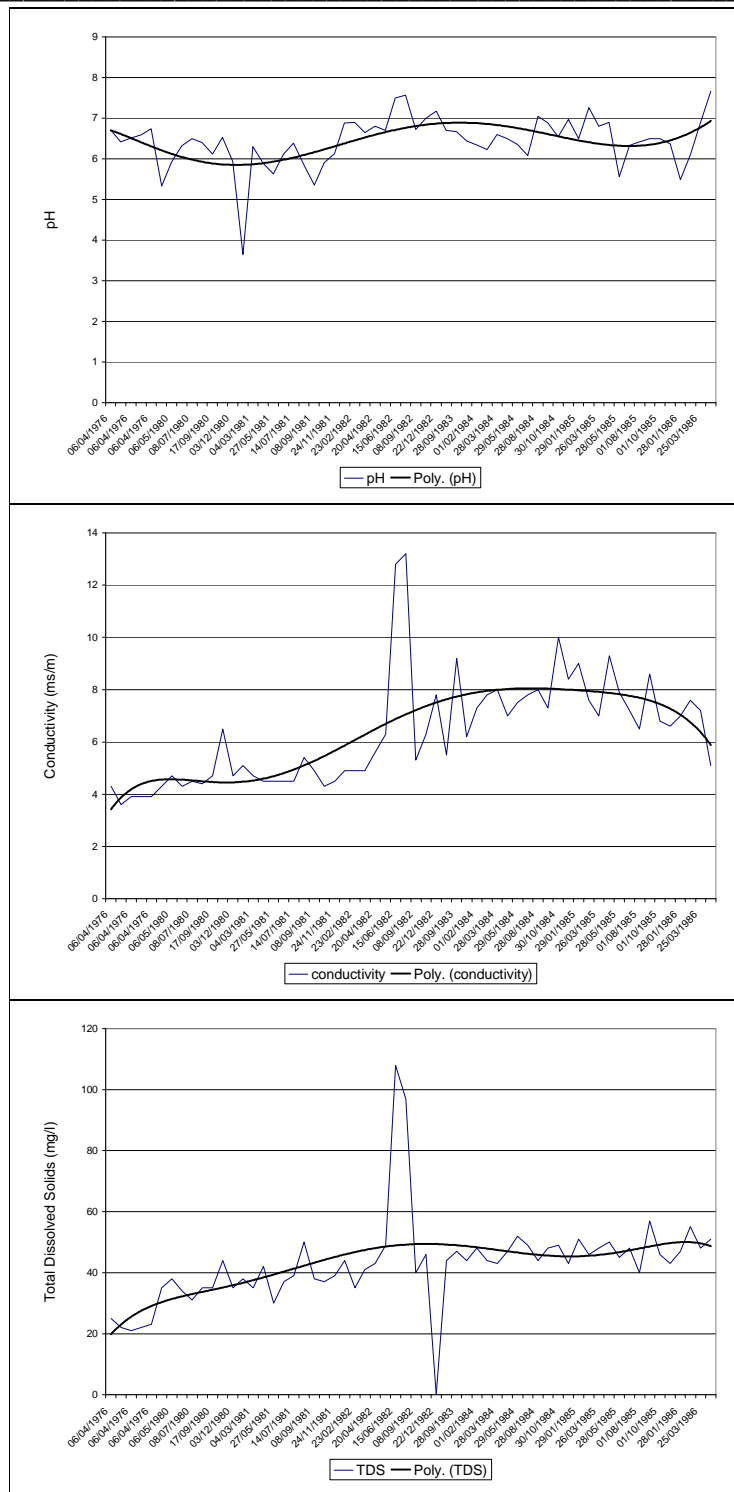


Figure 2.4: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Donkerpoort Dam (DPD).

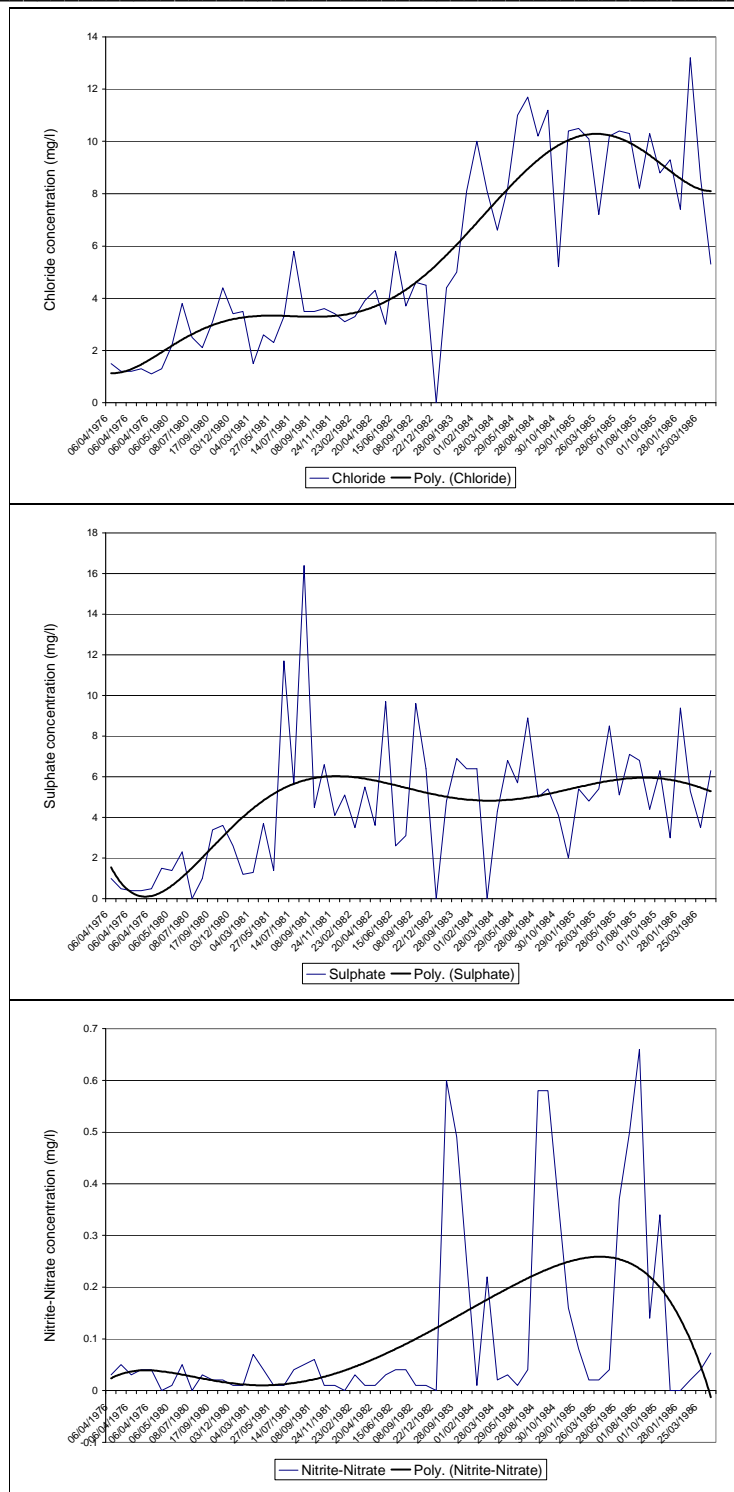


Figure 2.5: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Donkerpoort Dam (DPD).

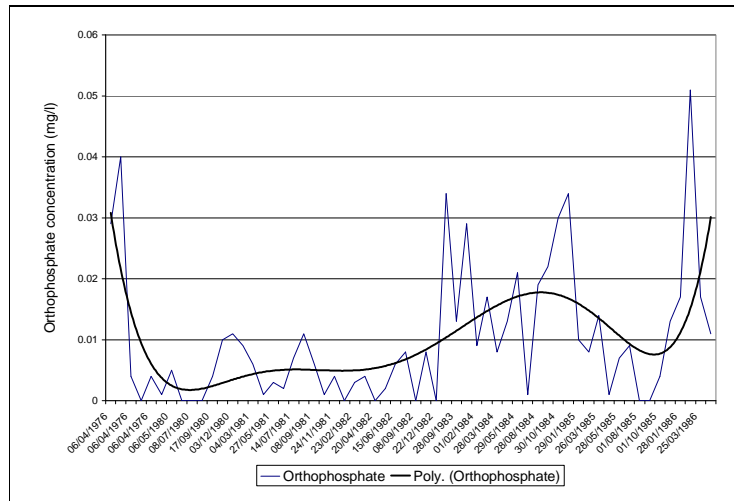


Figure 2.6: Graphic representation of historical data on a temporal scale. Orthophosphate at Donkerpoort Dam (DPD).

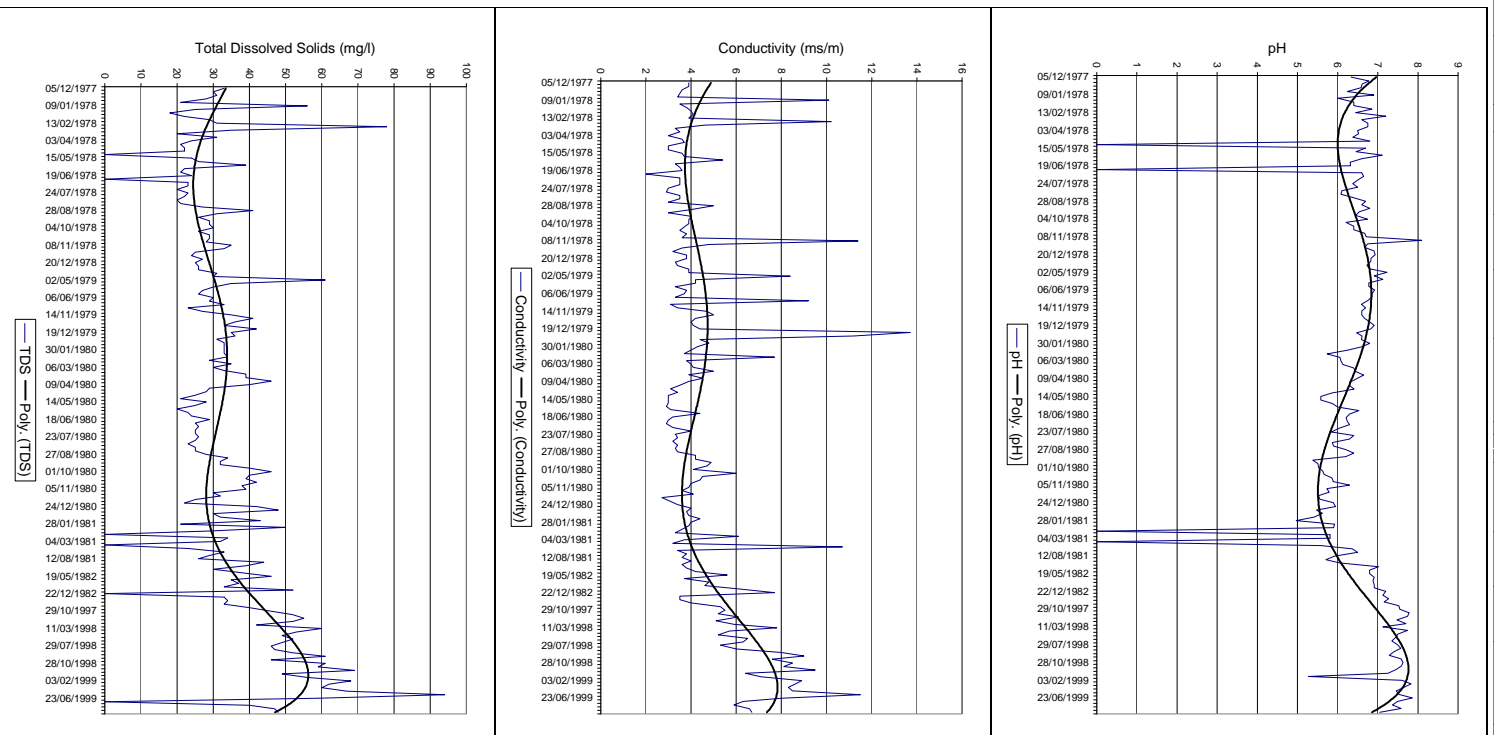


Figure 2.7: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Hesse-se-Water (HSW).

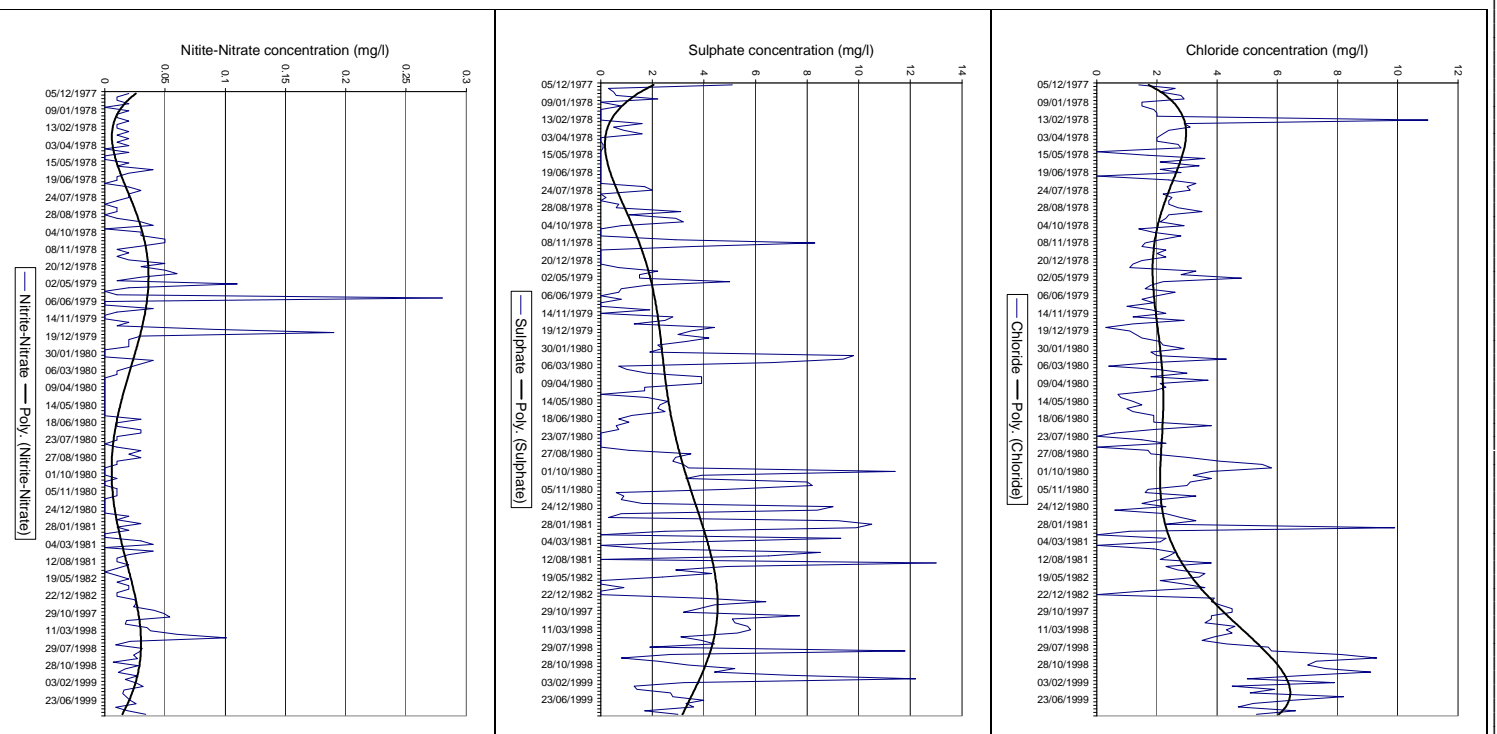


Figure 2.8: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Hesse-se-Water (HSW).

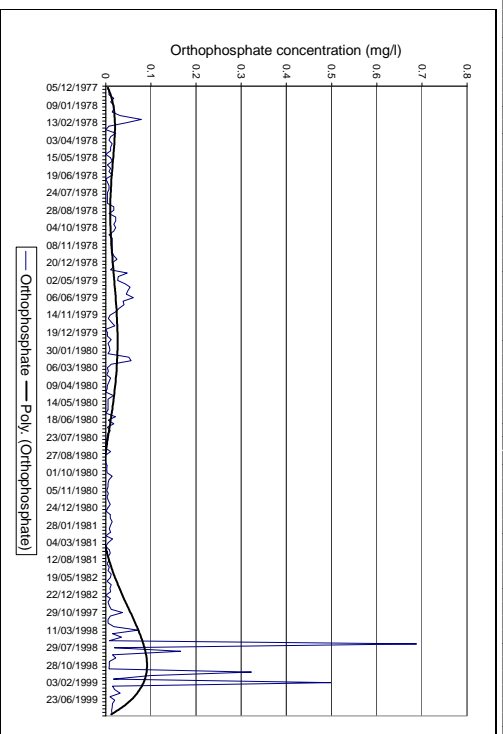


Figure 2.9: Graphic representation of historical data on a temporal scale. Orthophosphate at Hesse-se-Water (HSW).

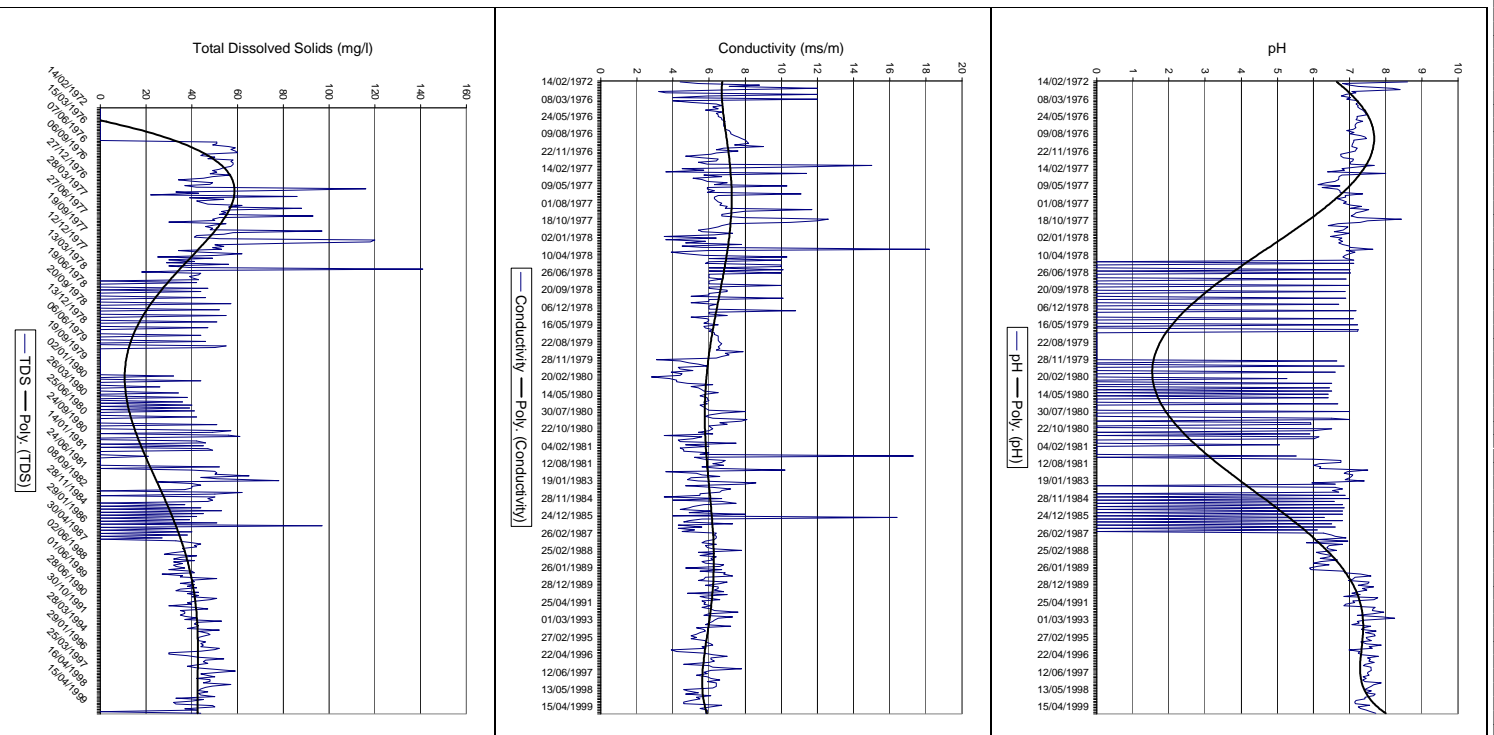


Figure 2.10: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Olfantanspruit.

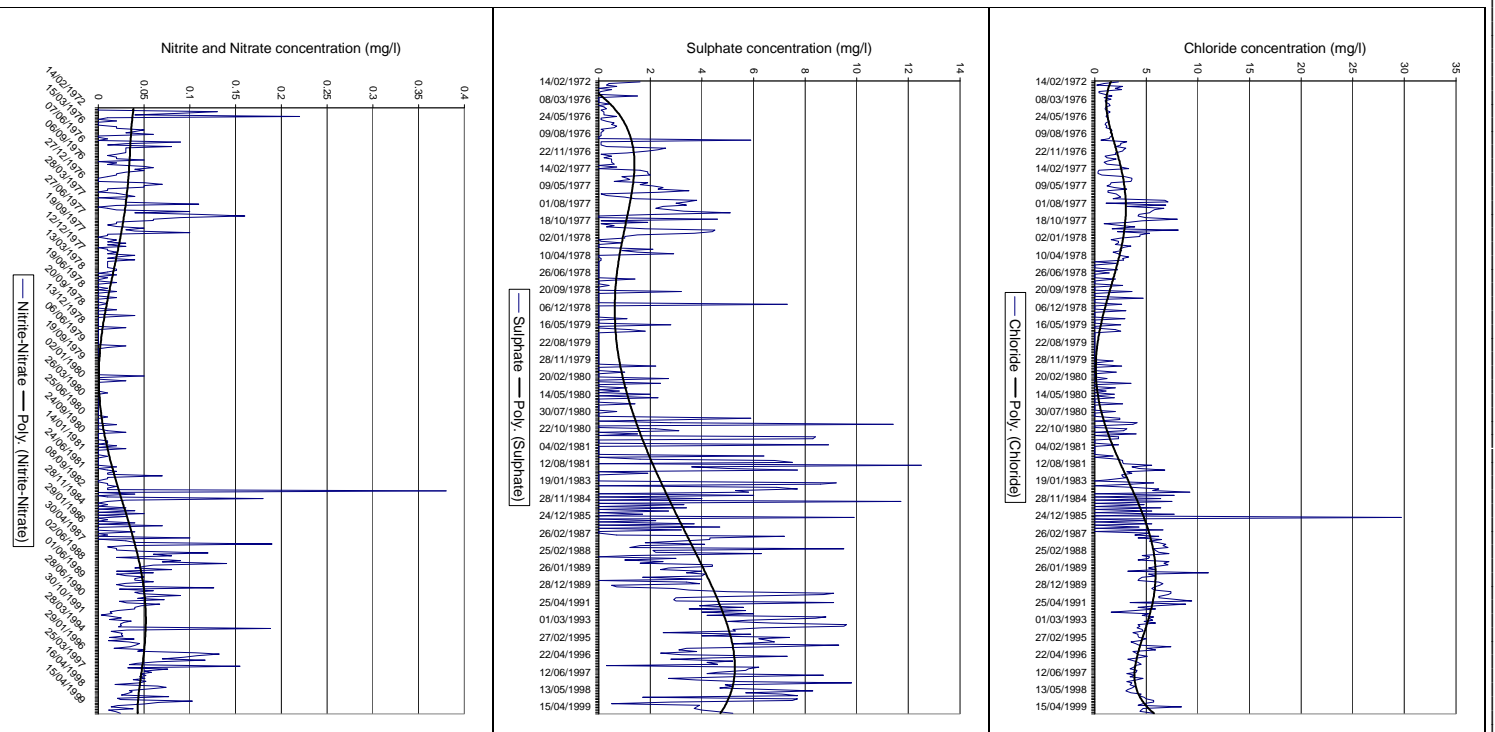


Figure 2.11: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Olifantspruit.

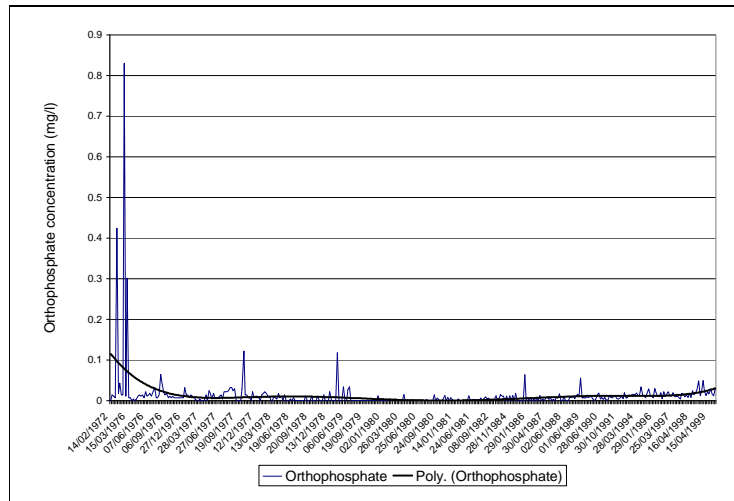


Figure 2.12: Graphic representation of historical data on a temporal scale. Orthophosphate at Olifantspruit.

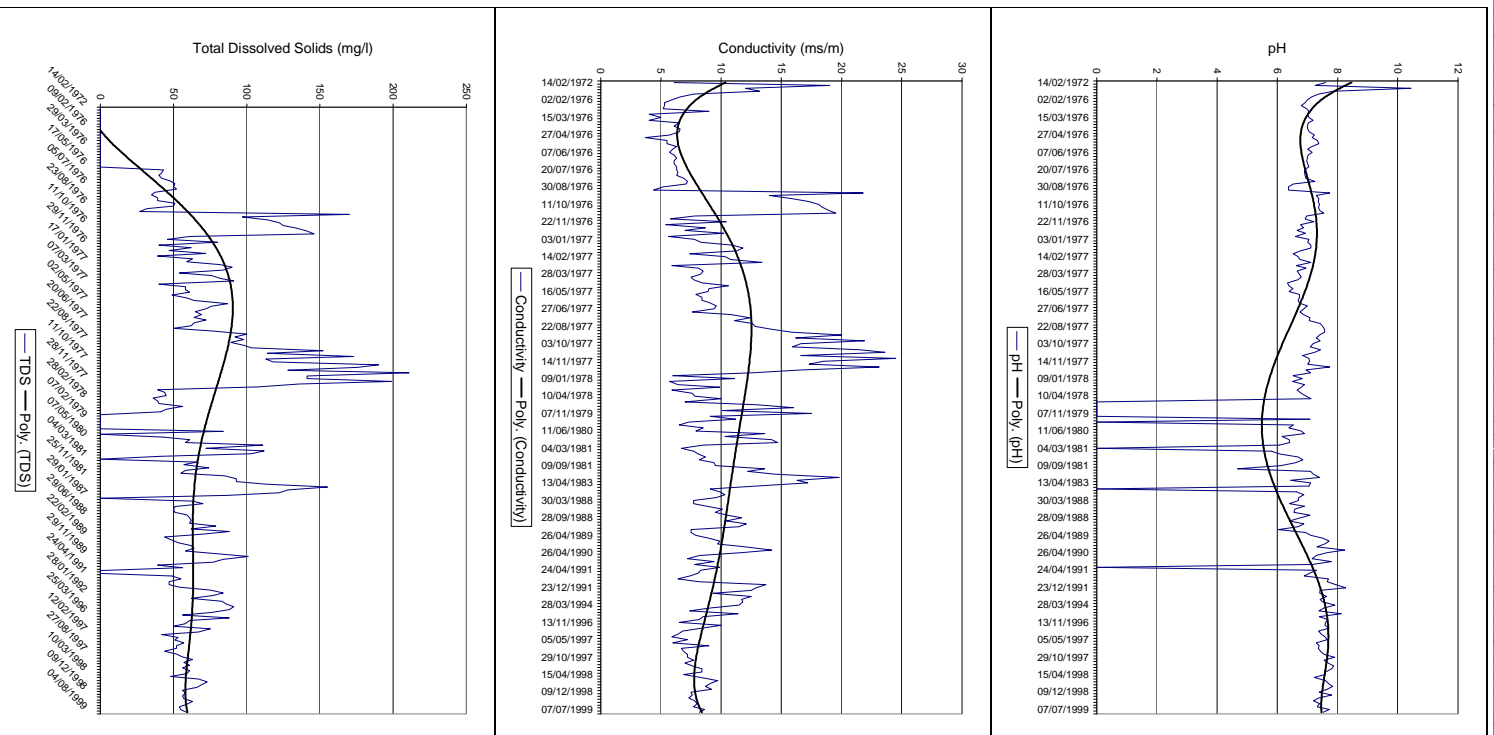


Figure 2.13: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Nylsvley at Deelkraal.

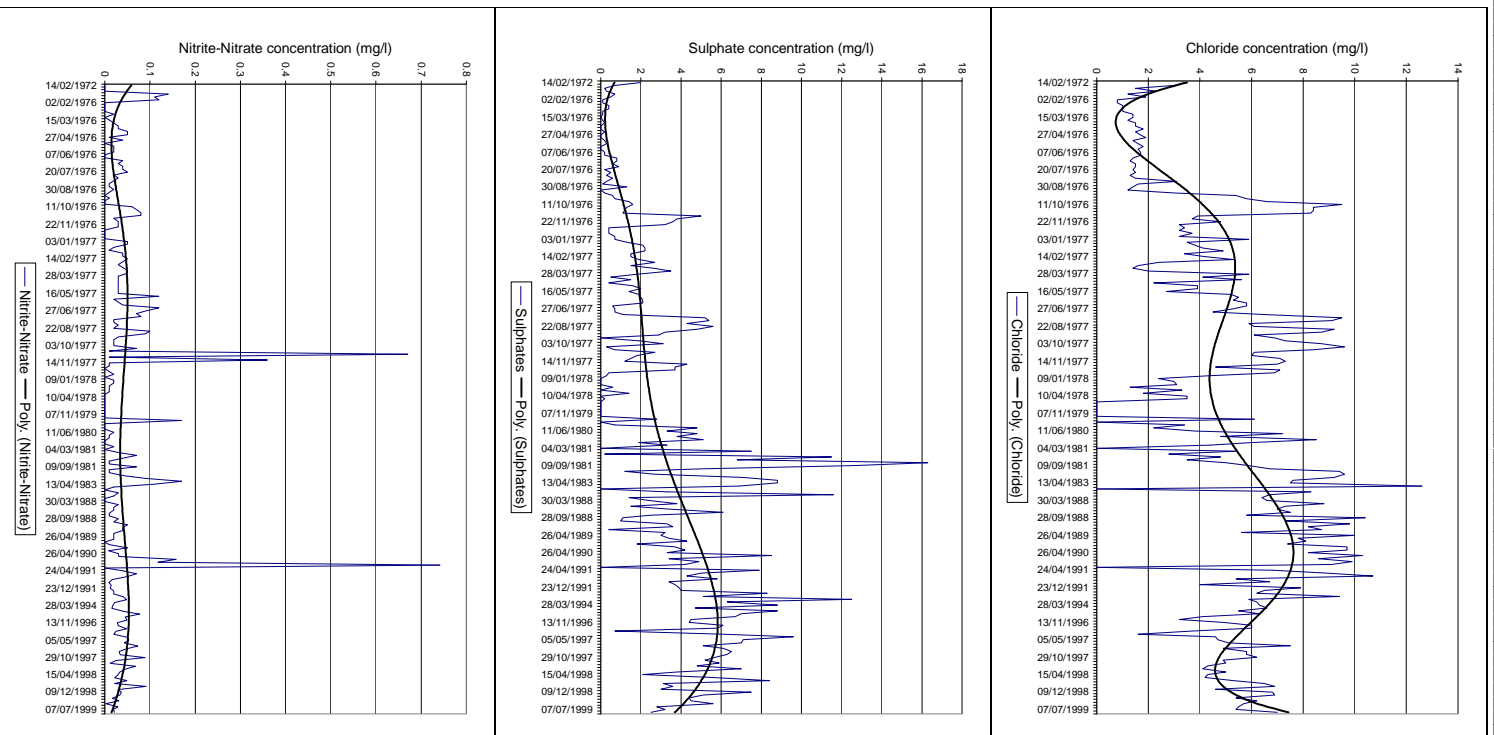


Figure 2.14: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Nylsvley and Deelkraal.

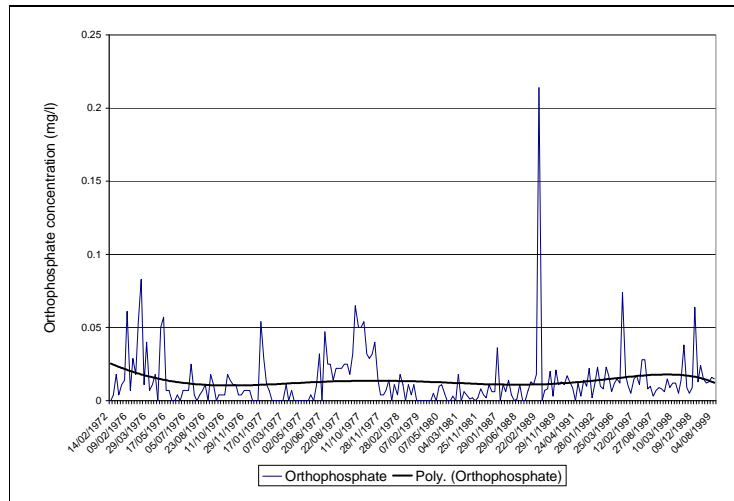


Figure 2.15: Graphic representation of historical data on a temporal scale. Orthophosphate at Nylsvley at Deelkraal.

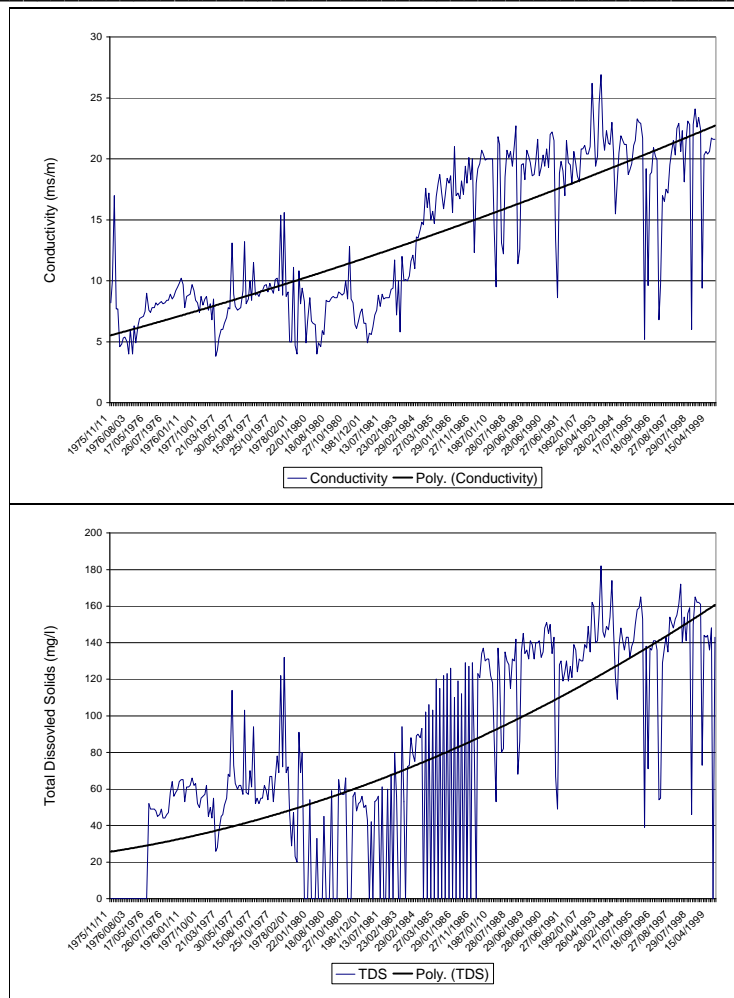


Figure 2.16: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Bad-se-Loop.

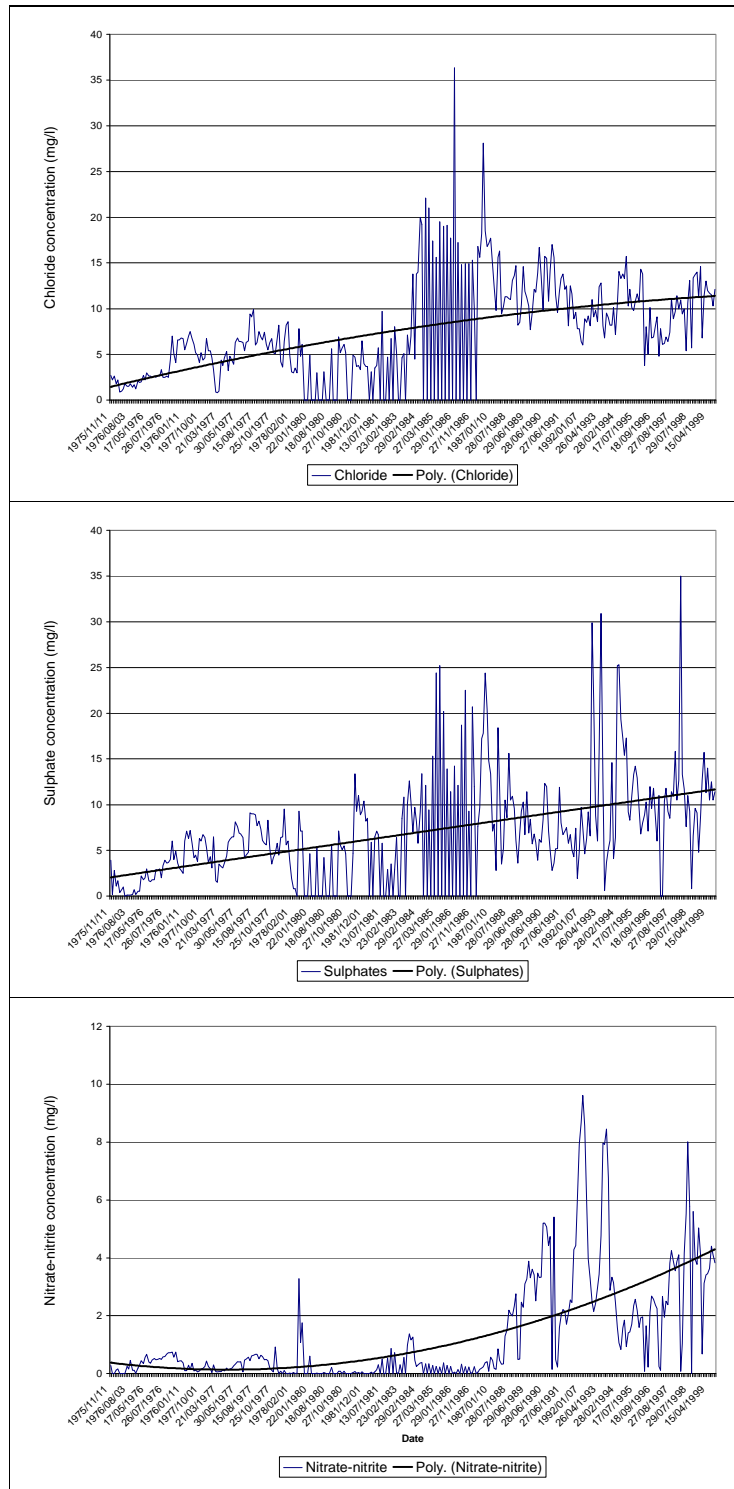


Figure 2.17: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Bad-se-Loop.

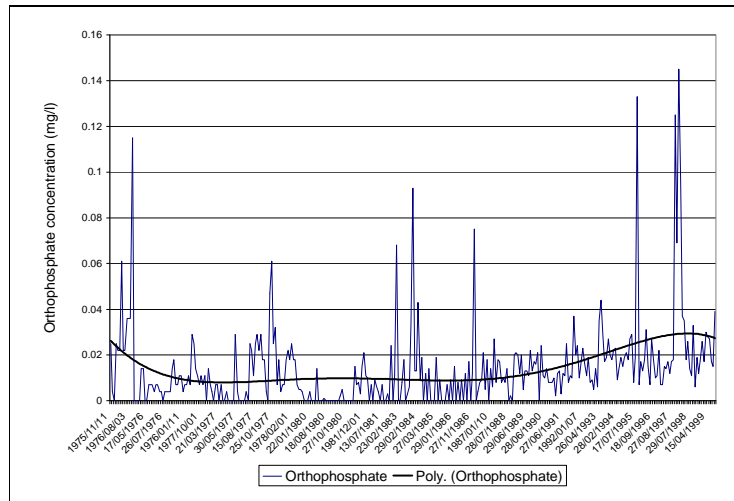


Figure 2.18: Graphic representation of historical data on a temporal scale. Orthophosphate at Bad-se-Loop.

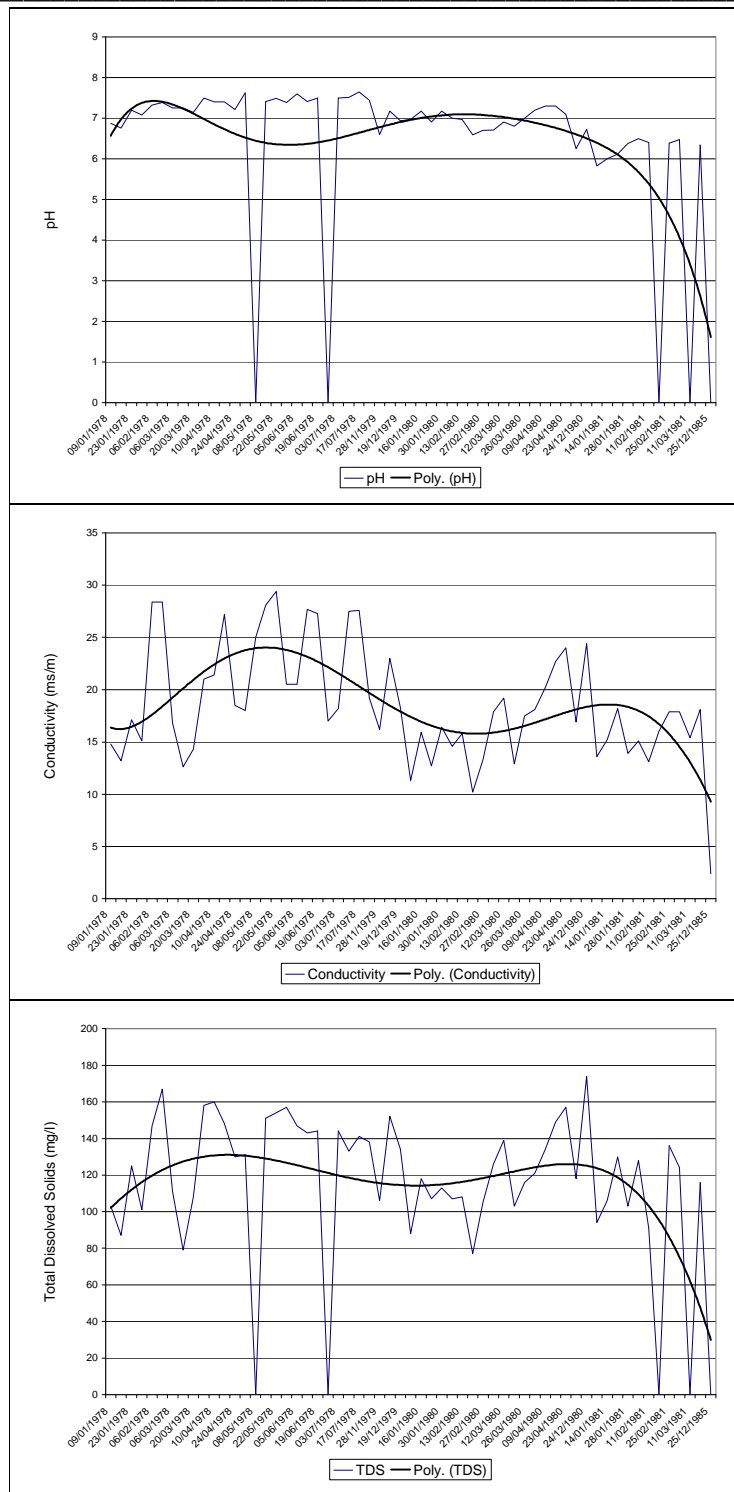


Figure 2.19: Graphic representation of historical data on a temporal scale. pH, Conductivity and TDS at Toboasspruit.

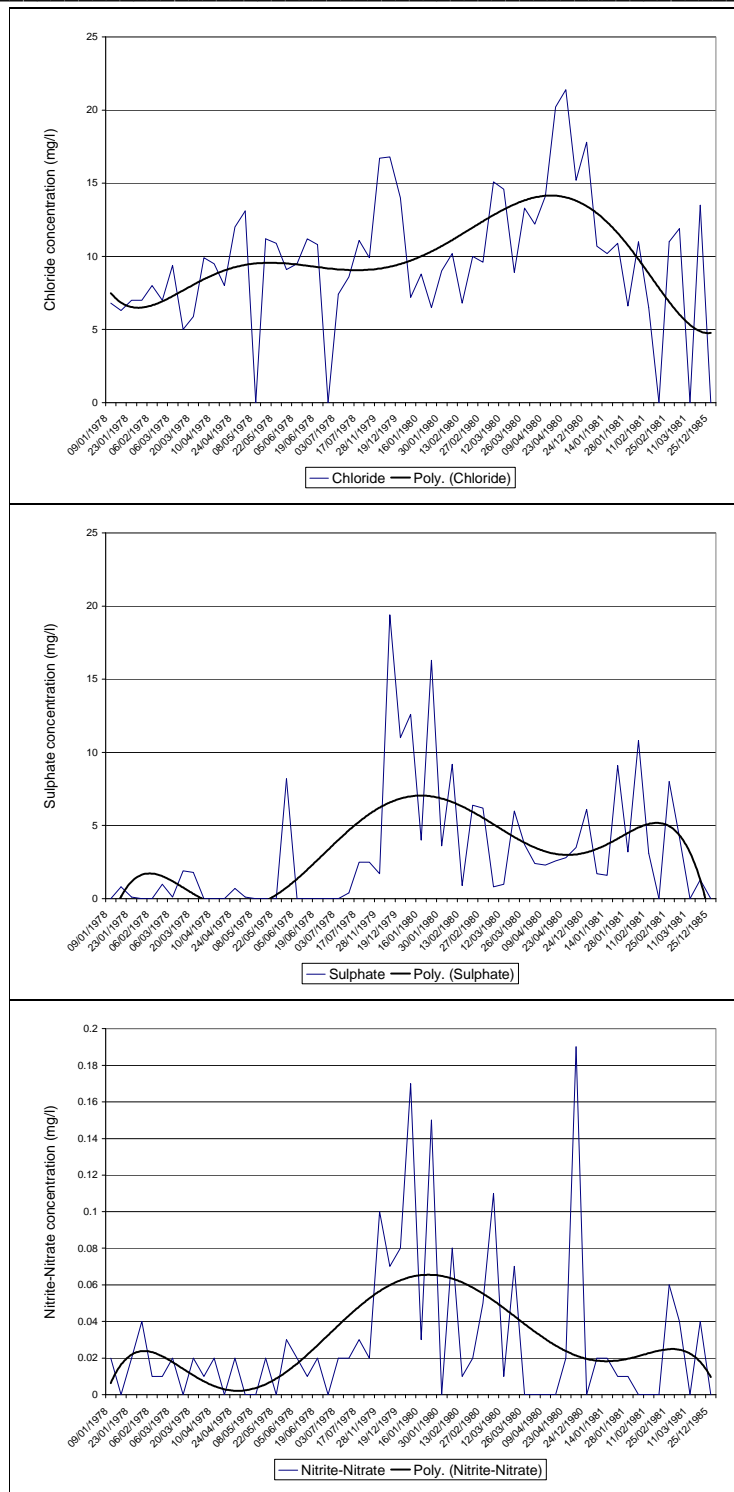


Figure 2.20: Graphic representation of historical data on a temporal scale. Chloride, Sulphate and Nitrite-Nitrate at Toboasspruit.

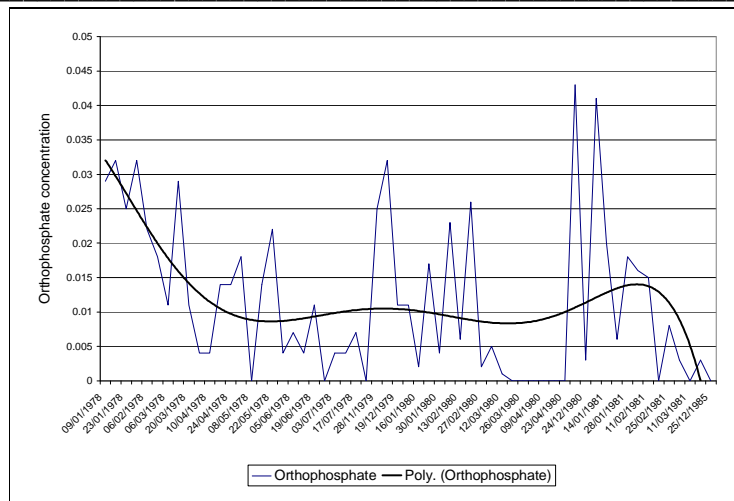


Figure 2.21: Graphic representation of historical data on a temporal scale. Orthophosphate at Tobiasspruit.

Various activities take place around these sites and increased farming and mining activities over the last 20 years may have lead to these changes occurring. The variables that indicated negative trends or decreases in concentrations are the conductivity and total dissolved solids at the source of the Klein Nyl River and in the Olifantspruit.

The reference conditions for the sites were then calculated using the medians of the data for the first time period. Three percentile values were determined for each site with the 50th percentile giving the median concentration/levels for the system and the 25th and 75th percentiles indicating a suitable operating range. Table 2.3 shows these values and thus the guideline ranges for the water in the system. It is important to remember that these values are just a guideline for water quality in the system in the period from 1972 to 1984 and are thus not pristine conditions for the system but any deviations from these ranges denote deterioration in the systems integrity. In the case of Nylsvley at Deelkraal the data set was small so the first time period was taken from October 1983 to January 1988.

The results in table 2.3 indicate that at most of the sites the medians for period two show increased levels/concentrations than those of period one. These indicate an overall deterioration of the water quality in the system over the past 20 years. The 25th and 75th percentiles provide a guideline range from which one can compare present conditions. These indicate conditions close to the natural state of the river system. Table 2.4 indicates the preferred water quality ranges from the Nyl River system from data provided by gauging weirs. The conductivity and TDS at the source of the Klein Nyl Oog indicates a different trend and thus the data from period two has been used instead of the values from period one. The same trend is present at Olifantspruit and at the site Nylsvley at Deelkraal.

Table 2.3: Water quality ranges for historical sites indicating two time periods and temporal changes in variables.

	pH	Ec	Tds	Cl	SO4	NO2-NO3	PO4
Klein Nyl at Modimolle							
25th p period 1	6.53	71.75	49.00	3.08	0.68	0.02	0.00
median period 1	6.90	87.00	64.00	4.40	1.85	0.03	0.01
75th p period 1	7.10	137.50	103.00	6.63	4.80	0.05	0.02
25th p period 2	7.33	75	51	5.8	3.9	0.03	0.01
median period 2	7.54	82.3	58	7.5	4.8	0.04	0.01
75th p period 2	7.67	99.69	70	10	6.5	0.05	0.02
Donkerpoort Dam							
25th p period 1	6.03	45.00	35.00		2.60	0.01	0.00
median period 1	6.44	49.00	39.00		3.70	0.03	0.01
75th p period 1	6.72	59.00	44.00		6.40	0.05	0.01
25th p period 2	6.44	55.15	43.25		5.00	0.03	0.01
median period 2	6.75	72.00	46.50		5.35	0.08	0.01
75th p period 2	6.99	79.50	48.75		6.68	0.24	0.02
Hessie-se-Water							
25th p period 1	6.14	34.25	24.00	2.00	1.20	0.01	0.01
median period 1	6.60	39.00	30.00	2.30	2.30	0.02	0.01
75th p period 1	6.80	42.00	35.00	2.90	4.45	0.02	0.01
25th p period 2	7.22	41.05	38.35	5.03	4.00	0.04	0.01
median period 2	7.45	52.75	49.00	10.00	4.52	0.04	0.02
75th p period 2	7.59	63.08	52.00	10.00	5.80	0.04	0.03
Olifantspruit							
25th p period 1	6.52	56.00	42.00	1.80	0.70	0.01	0.01
median period 1	6.89	64.00	49.00	2.60	1.90	0.02	0.01
75th p period 1	7.11	72.25	55.00	3.60	5.15	0.03	0.01
25th p period 2	6.87	56.40	36.00	4.30	3.40	0.03	0.01
median period 2	7.41	60.10	41.00	5.70	4.40	0.04	0.01
75th p period 2	7.56	64.00	45.00	8.25	6.00	0.07	0.02
Nylsvley							
25th p period 1	6.79	101.50	64.50	7.15	4.13	0.02	0.03
median period 1	7.00	113.00	77.00	7.30	4.85	0.03	0.05
75th p period 1	7.15	126.50	92.00	7.65	5.58	0.04	0.05
25th p period 2	6.78	93.25	61.25	9.80	1.90	0.02	0.01
median period 2	7.09	114.50	67.00	11.50	3.60	0.03	0.02
75th p period 2	7.57	125.50	80.00	14.00	5.00	0.04	0.04

Table 2.3: Water quality ranges for historical sites indicating two time periods and temporal changes in variables. (Continued)

	pH	Ec	Tds	Cl	SO4	NO2-NO3	PO4
Bad-se-Loop							
25th p period 1	6.42	72.25	53.25	3.10	3.10	0.11	0.01
median period 1	6.67	86.00	58.00	5.10	5.60	0.35	0.01
75th p period 1	6.97	99.25	66.75	6.60	7.30	0.56	0.02
25th p period 1	7.32	170.75	121.25	9.80	7.18	0.55	0.01
median period 2	7.66	199.75	135.00	12.00	10.15	2.47	0.02
75th p period 1	7.92	216.00	143.00	15.53	12.67	4.12	0.02
Tobias							
25th p period 1	6.55	149.50	106.50	8.75	1.70	0.02	0.00
median period 1	6.94	179.00	124.00	11.90	2.95	0.04	0.01
75th p period 1	7.29	182.50	139.00	14.55	8.43	0.10	0.02
25th p period 2	7.00	112.78	123.00	15.00	5.00	0.20	0.20
median period 2	7.07	272.50	160.00	25.87	5.00	0.20	0.20
75th p period 2	7.17	318.00	170.75	29.00	5.00	0.20	0.20

Table 2.4: Water Quality Ranges for water in the Nyl River System.

	pH	Ec	Tds	Cl	SO4	NO2-NO3	PO4
Klein Nyl at Modimolle							
Lower Limit	7.33	75	51	5.8	3.9	0.03	0.01
Upper Limit	7.67	99.69	70	10	6.5	0.05	0.02
Donkerpoort Dam							
Lower Limit	6.03	45.00	35.00		2.60	0.01	0.00
Upper Limit	6.72	59.00	44.00		6.40	0.05	0.01
Hessie-se-Water							
Lower Limit	7.22	41.05	38.35	5.03	4.00	0.04	0.01
Upper Limit	7.59	63.08	52.00	10.00	5.80	0.04	0.03
Olifantspruit							
Lower Limit	6.87	56.40	36.00	4.30	3.40	0.03	0.01
Upper Limit	7.56	64.00	45.00	8.25	6.00	0.07	0.02
Nylsvley							
Lower Limit	6.79	93.25	61.25	7.15	4.13	0.02	0.03
Upper Limit	7.15	125.5	80.00	7.65	5.58	0.04	0.05
Bad-se-Loop							
Lower Limit	7.32	170.75	121.25	9.80	7.18	0.55	0.01
Upper Limit	7.92	216.00	143.00	15.53	12.67	4.12	0.02
Tobias							
Lower Limit	6.55	149.50	106.50	8.75	1.70	0.02	0.00
Upper Limit	7.29	182.50	139.00	14.55	8.43	0.10	0.02

2.3.1 Water parameters:

pH:

The pH is a measure of the hydrogen ion activity in water. As the concentration of H^+ ions increases in water the pH value decreases making the water more acidic (DWAF⁷, 1996). pH is regulated by the carbonate-bicarbonate cycle consisting of $H_2CO_3 \leftrightarrow CO_2 \leftrightarrow CO_3^{2-} \leftrightarrow HCO_3^-$ (Polling, 1999). pH fluctuations may occur either naturally or by anthropogenic activities.

Natural fluctuations can either occur diurnally or seasonally. Diurnal fluctuations are caused by the changes in the carbonate- bicarbonate levels due to the actions of photosynthesis in productive systems. Extreme rates of photosynthesis can result in high pH values in still standing eutrophied waters. Seasonal variations are caused by the hydrological cycle (DWAF⁷, 1996).

Artificial changes brought on by anthropogenic activities take place in 3 ways. (1) Low pH point source from industries, (2) mine drainage, which is almost always acidic, and (3) acidic precipitation brought on by the presence of air bourn pollution. During the burning processes of coal and combustion engines SO^2 and NO^2 are released into the atmosphere. These gasses bind with the water in the atmosphere to form a sulphuric or nitric acid compound which leads to acid rain (DWAF⁷, 1996). The acid rain interferes with the nutrient availability.

The pH plays an important role in physio-chemical and biological processes in water (Train, 1979). Depending on the number of free H^+ ions, cations and anions are released or bonded to other ions in the water making them more or less toxic to aquatic organisms. An example of this is the influence pH can have on the reproductive rates of fish. This was indicated by Tucker and Robinson (1990) in Channel catfish. They noted that disturbances in respiration, osmoregulation and blood pH/acid-base balance result from exposure to low water pH levels.

This caused reduced growth, reproduction and disease resistance. "Acid rain" can cause situations that will negatively influence egg production and hatching time of fish eggs (Mount, 1973; Nelson, 1982). Egg production can also be negatively influenced for example some species of fish show signs of embryonic deformities (Lee and Germing, 1980; Peterson *et al.*, 1976).

The pH of the water can also have a role in the release of toxic substances from sediments (DWAF⁷, 1996), which is important in wetland systems as they act as sediment traps.

The pH of the water in the system (table 2.5) for the most part falls within the target water quality range (TWQR) set out by the Department of Water Affairs and Forestry.

Less than 25 percent of the readings showed values less than the lower limit set out by DWAF of 6.5 pH units. Less than five percent of the readings fell outside the upper limit of 9 pH units. The TWQR of 6.5-9 is set out for Aquaculture (DWAF⁶, 1996). A maximum value of 9.52 was measured at the site located at the Moorddrift Dairy during sampling in April 2001. The minimum value of 5.52 pH units was recorded at the source of the Klein Nyl River (Klein Nyl Oog) during March 2002. The increased pH levels at the Haakdoorn and Moorddrift site can be attributed to the eutrophication of the water. Both sites are dammed areas situated at the lower end of the Nyl River Floodplain and would thus provided good points for nutrient deposition and eutrophication. This increase could be an effect of commercial cattle farming.

Table 2.5: pH values of sampled water in the Nyl River System.

Locality	pH						
	Apr-01	Aug-01	Nov-01	Mar-02	Jul-02	Nov-02	Mar-03
Koh-I-Noor	5.89	7.76	5.7	N/A	7.14	N/A	N/A
Klein Nyl Oog	5.91	8.39	6.62	5.52	7.33	6.5	N/A
Abba	6.21	7.56	6.55	6.64	6.43	6.57	6.57
Donkerpoort	6.11	7.51	6.8	6.68	6.45	N/A	6.85
Groot Nyl Oog	6.06	7.36	6.45	5.61	5.88	7.1	N/A
Groot Nyl	6.41	7.07	6.66	6.34	6.72	6.8	N/A
Sewerage Works	7.2	7.66	N/A	7.14	7.34	7.31	N/A
Jasper	6.29	8.81	6.67	6.86	7.16	7.39	6.75
Hessie-se-Water	6.33	7.02	6.94	6.61	6.43	7.35	N/A
Olifantspruit	6.3	7.31	6.79	6.58	6.64	7.08	6.28
Nylsvley	7.04	7.31	6.78	6.92	7.56	6.8	7.62
Bad-se-Loop	6.2	7.5	7.05	7.56	7.31	7.4	N/A
Tobiasoog	6.45	7.35	7.02	6.9	7.44	7.2	N/A
Mine	6.72	7.13	7	7.18	7.01	7.29	N/A
Tobias	6.87	N/W	6.73	7.69	N/W	N/A	N/A
Mosdene	6.72	6.92	6.75	6.38	N/W	N/A	N/A
Haakdoorn	6.9	8.47	6.6	8.26	7.29	N/A	N/W
Moorddrif	9.56	9.5	8.94	7.97	7.52	N/A	7.59

N/A: not available, N/W: no water. Light grey cells indicate tributaries

Oxygen:

Dissolved oxygen concentrations are probably the most important abiotic factors for aquatic organisms (Davies and Day, 1998). Dissolved oxygen enters the aquatic environment by one of two methods. The first is the dissolving of atmospheric O₂ into the water via surface interface interactions. The second is via the photosynthetic interactions of aquatic plants (DWAFF⁷, 1996). Concentrations fluctuate diurnally due to the metabolic processes of photosynthesis and respiration (Davies and Day, 1998). Oxygen concentration levels are usually lowest just before dawn and increase during the day, peaking in the afternoon. The amount of oxygen that can dissolve in a water body is dependant on certain factors such as temperature, aeration rates from the atmosphere, air pressure and salinity (Davies and Day, 1998). Oxygen concentrations in a water body can be influenced by the following factors: (1) organism respiration rates, (2) the presence of organic matter and (3) the resuspension of anoxic sediments either via dredging or natural flooding (DWAFF⁷, 1996).

In wetland systems the presence of oxidizable organic matter (detritus) and the rates of photosynthesis and respiration are important factors in the oxygen concentrations in water. These factors can have an influence on the oxygen levels and thus affect the presence of high life in the system. Organic matter will settle out in the system due to the slow movement of the water in the system. This is part of the normal functioning of wetlands as previously discussed in chapter one.

Table 2.6: Dissolved Oxygen concentrations during the study period.

Locality	Dissolve Oxygen (mg/l)						
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul	02-Nov	03-Mar
Koh-I-Noor	8.1	7.78	8.67	N/A	8.64	N/A	N/A
Klein Nyl Oog	5.4	8.48	6.96	4.04	8	5.23	N/A
Abba	7.3	6.53	6.96	7	5.44	0.13	0.13
DPD	7.6	8.38	8.84	8.13	8.45	N/A	6.36
Groot Nyl Oog	6.2	7.31	7.22	8.68	8.26	7.34	N/A
Groot Nyl	7.9	9.03	8.39	9.2	6.89	6.54	N/A
Sewerage	7.5	8.3	N/A	8.07	5.18	N/A	N/A
Jasper	7.9	10.75	5.95	8.35	7.75	4.96	2.04
Hessie-se-Water	6	8.85	6.71	7.16	8.74	1.5	N/A
Olifantspruit	6.7	6.93	8.75	8.97	5.68	4.64	6.86
Nylsvley	6.7	6.11	5.41	2.23	7.11	1.15	3.3
Bad-se-Loop	6.4	8.42	8.37	8.82	8.03	7.1	N/A
Tobiasoog	7.2	7.55	8.42	7.74	6.89	5.69	N/A
Mine	4.7	2.1	7.95	6.71	4	3.24	N/A
Tobias	5.96	N/W	8.14	8.15	N/W	N/A	N/A
Mosdene	0.4	3.6	1.48	0	N/W	N/A	N/A
Haakdoorn	6	9.75	0.33	7.65	3.4	N/A	N/W
Moorddrift	10.1	7.36	5.9	8.74	8.63	N/A	8.84

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Tables 2.6 and 2.7 indicate results obtained from the in-situ O₂ determinations. The results varied greatly. The maximum O₂ concentration was recorded during the August 2001 survey at Jasper. The site had an O₂ concentration of 10.75 mg/l. The minimum value recorded was at Mosdene in March 2002.

Figure 2.22 indicates the mean oxygen concentration at the different sites along the course of the Klein Nyl River, past the confluence of the Groot and Klein Nyl Rivers, and along the course of the Nyl River. The graph clearly indicates that the oxygen levels decrease dramatically as the water flows through the wetland. This can be attributed to the increased levels of organic matter and the break down thereof. A more detailed explanation will be discussed later on in this chapter. The oxygen saturation (Table 2.7) indicated slightly different results with the maximum value being 139% at Moorddrift in April 2001. The minimum value was once again found at Mosdene in March 2002. The values at Mosdene in March 2002 can be attributed in part to an oily layer that was found floating on the surface of the sampling site. This layer inhibited the transfer of oxygen from the atmosphere into the water. The oxygen levels fluctuated greatly and thus it makes it difficult to list all the sites that fell outside the TWQR for aquatic ecosystems. The TWQR as prescribed by the Department of Water Affairs and Forestry state that oxygen saturation levels below 60% are sublethal to aquatic organisms and below 40% are lethal. The sampling site located at Mosdene consistently provided oxygen saturation readings below the lethal TWQR of 40%, with many of the other sites having sub lethal concentrations of oxygen.

Table 2.7: Oxygen Saturation during the study period.

Locality	Dissolved O ₂ (% Saturation)						
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul	02-Nov	03-Mar
Koh-I-Noor	99	70.4	90.8	N/A	79.4	N/A	N/A
Klein Nyl Oog	69	102	77.1	50	75.4	67.3	N/A
Abba	91	73	75.5	87	68.4	1.8	1.8
Donkerpoort	96	83	95.6	100.2	85.8	N/A	83.5
Groot Nyl Oog	82	78.9	80.2	108.1	75.6	96.6	N/A
Groot Nyl	104	92.2	90.5	109.4	72.1	80.4	N/A
Sewerage Works	96	79.2	N/A	100.5	N/A	59.3	N/A
Jasper	98	113	63.8	104	71.8	55.6	3.75
Hessie-se-Water	79	78.1	70.6	88	83.1	20	N/A
Olifantspruit	91	71.2	93.5	108.6	66.8	52.8	82
Nylsvley	74	55.7	62.6	27.6	89.4	14.1	45.3
Bad-se-Loop	81	87	92.3	107.9	81.9	90.5	N/A
TobiasoogO	87	75.6	89	95.8	75.9	74.6	N/A
Mine	61	21.4	86.1	82.9	36.1	41	N/A
Tobias	71	N/W	90	96.4	N/W	N/A	N/A
Mosdene	6	31.7	16.7	0	N/W	N/A	N/A
Haakdoorn	77	101.7	3.5	117.7	32.8	N/A	N/W
Moorddrift	139	82.1	69.2	102.6	90	N/A	117.8

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

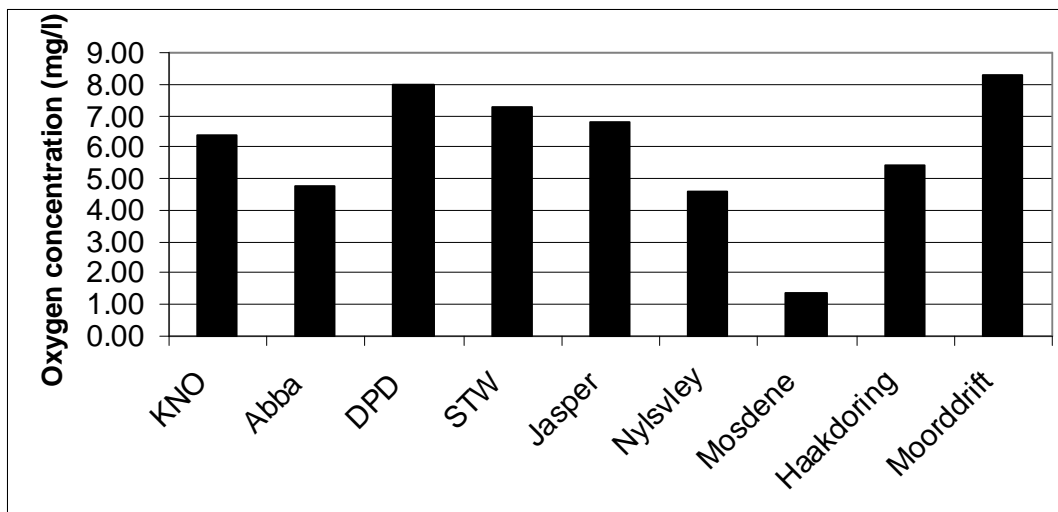


Figure 2.22: Graph indicating mean oxygen concentration level fluctuations in the Klein Nyl River as it flows from the source through the Nyl River Floodplain.

Conductivity (EC) and Total Dissolved Solids (TDS):

Electrical conductivity is the measure of a body of water's ability to conduct an electrical current (DWAF⁷, 1996). It is closely related to the waters. Total Dissolved Solids (TDS) (DWAF⁷, 1996). Conductivity is usually the parameter of choice to measure as it is easier to measure than TDS. TDS can be determined by using the conductivity in an equation for TDS determination. This calculation is done by multiplying the conductivity in ms/m by a factor of 6.5. This gives a TDS value in mg/l (Kempster and van Vliet, 1991).

A water bodies electrical conductivity is a result of the presence of ions in the form of carbonates, bicarbonates, sulphates, nitrates, sodium, potassium, calcium and magnesium. Organic compounds dissolve in water and thus do not affect the EC of the water (Davies and Day, 1998). Natural waters contain varying quantities of TDS and their conductivities thus also vary. These solids enter the water column via a number of routes namely: (1) the mechanical weathering of rocks, (2) evaporation and (3) rainfall (Davies and Day, 1998)

TDS concentrations can increase due to anthropogenic activities. These can be either from direct input of effluents from industrial processes, or from surface runoff from urban, industrial or cultivated areas. Increased TDS concentrations affect metabolic processes of organisms and the fate and impact of other chemical constituents in the aquatic environment (DWAF⁷, 1996).

Changes in TDS concentration can affect organisms on the following levels: (1) effects on, and adaptations of individual species, (2) effects on community structure, and (3) effects on microbial and ecological processes such as metabolic rates and nutrient cycling (Dallas *et al.*, 1998). Little is known about the tolerance levels of aquatic organisms to changes in TDS concentrations but the following generalizations can be made. Rate of change and duration are more important than absolute TDS changes, especially in systems where organisms are not adapted to fluctuations in TDS. Juvenile stages appear more sensitive to changes in TDS concentration.

Synergistic and antagonistic effect usually appear as secondary effects of TDS concentration changes (metals may become more toxic), and organisms adapted to low salinities are generally more sensitive to changes in TDS concentrations (Dallas *et al.*, 1998).

The results in table 2.8 indicate that the conductivities at the sites range between 8.17 $\mu\text{s}/\text{cm}$ and 685 $\mu\text{s}/\text{cm}$. Figure 2.23 indicates that at points where the conductivity readings decrease a dilution of the main stream occurs and where increases occur the tributaries are adding to the contamination. The results also indicate that the Sewerage Works has an effect on the increase of the conductivity of the Nyl River. The Tobiasspruit also has a negative effect on the water quality of the Nyl River. The highest conductivity reading (685 $\mu\text{s}/\text{cm}$) was recorded at the Sewerage Treatment Works in November 2002. The EC reading is higher than the 635 $\mu\text{s}/\text{cm}$ recorded by Polling 1999 in the Selati River. Polling (1999) reported that the conductivity of 635 $\mu\text{s}/\text{cm}$ was almost acceptable and the conductivity reading at the Sewerage Works can thus be deemed to be unacceptable. This however is an isolated incident with the median conductivity for the system being 91.4 $\mu\text{s}/\text{cm}$, which is acceptable. The conductivity values obtained in the lower reaches of the study area do not conform to the TWQR guideline values specified by the DWAF. They do differ from each other by the specified 15% given for aquatic ecosystems (DWAF⁷, 1996).

Table 2.8: EC values for the sampling sites for the period April 2001 to March.

Locality	Conductivity ($\mu\text{s}/\text{cm}$)						
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul	02-Nov	03-Mar
Koh-I-Noor	19.3	26.1	18.1	N/A	49.9	N/A	N/A
KLei Nyl Oog	8.17	47.2	24.6	14.48	46.6	17.3	N/A
Abba	45.5	44.2	62.2	48.9	67.8	73.2	73.2
Donkerpoort	48.5	47.1	53.5	55.7	91.4	N/A	134.7
Groot Nyl Oog	16.99	16.2	16.57	25.8	40.6	26.1	N/A
Groot Nyl	35.2	36.8	39.6	30.3	72.5	54.1	N/A
Sewerage Works	233	206	N/A	150.4	287	685	N/A
Jasper	132.3	151.9	151.5	165.7	229	503	163
Hessie-se-Water	60.8	61	73.5	63.9	51.4	141.3	N/A
Olifantspruit	53.6	52.4	29.9	57.1	86.2	60.3	42
Nylsvley	98.1	120	59	156.3	190.1	358	130.5
Bad-se-Loop	167	236	47.7	162.8	242	238	N/A
Tobiasoog	92.6	102.2	51.2	80	128.4	118.5	N/A
Mine	306	346	70.7	239	322	35.8	N/A
Tobias	375	N/W	137	292	N/W	N/A	N/A
Mosdene	94.9	152.2	121	224	N/W	N/A	N/A
Haakdoorn	325	361	196.7	215	468	N/A	N/W
Moorddrift	179.5	218	232	166	163.7	N/A	311

N/A: not available, N/W: no water. Light grey cells indicate tributaries

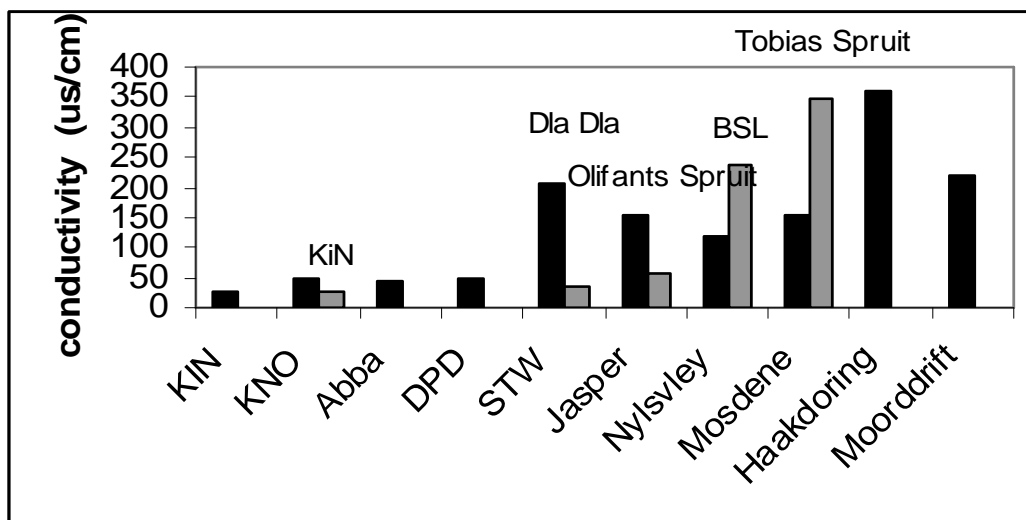


Figure 2.23: Graph indicating the increase in conductivity through the Klein Nyl River during August 2001. The light grey bars indicate the points where water from the tributaries mix with the water from Nyl River. Levels given for the tributaries are average conductivity readings from the different sites in the tributaries.

Table 2.9 gives the total dissolved solid values for the system. These values correspond to the conductivity levels, showing the same increase or decrease in trends. The TWQR for TDS state that the levels should not change by more than 15% of normal conditions for that time of year (DWAF⁷, 1996). These levels do increase by more than 15% if one compares the levels obtained during the same time periods for 2001 and 2002. One must take into account the difference in stream flow due to the poor rainy season experienced during 2002, which leads to decreased water available during the periods of low flow. The maximum concentration recorded of 342 mg/l was recorded at the Sewerage Works during November 2002. The minimum concentration of total dissolved solids (7.13 mg/l) was recorded at the source of the Klein Nyl River in March 2002. The 2001- 2002 rainy season was very good and the dilution factor of rainwater can be taken into account for the low concentrations. Similarly the poor 2002-2003 rainy season can be a contributing factor to the increased TDS concentrations recorded in the November 2002 sampling period. According to the DWAF water quality guidelines rainwater has a TDS concentration of around 1mg/l (DWAF⁷, 1996).

Table 2.9: TDS values for the sampling sites for the period April 2001 to March 2003.

Locality	Total Dissolved Solids (mg/l)						
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul	02-Nov	03-Mar
Koh-I-Noor	10.2	13	N/A	N/A	24.2	N/A	N/A
Klein Nyl Oog	15.84	24.1	12.4	7.18	23.3	8.5	N/A
Abba	23.7	22.2	31.4	24.7	40.8	36.6	36.6
Donkerpoort	25.1	23.2	26.8	26.7	45.8	N/A	67.2
Groot Nyl Oog	8.93	8.05	8.25	13.1	19.9	16.2	N/A
Groot Nyl	18.5	18.9	19.8	14.8	38.4	27.1	N/A
Sewerage Works	118	104	N/A	75.5	152	342	N/A
Jasper	69.6	76.1	74.9	83.3	116	251	81.6
Hessie-se-Water	30.1	30.6	36.7	32.3	51.7	70.2	N/A
Olifantspruit	27.8	26.3	15	28.1	44	29.7	21.1
Nylsvley	51	59.9	29.5	73.7	97.5	178	65.3
Bad-se-Loop	83	118	23.9	82.4	117	120	N/A
Tobiasoog	48.3	50.1	25.8	41.5	59.6	59.1	N/A
Mine	159	174	35.5	111	161	178	N/A
Tobias	190	N/W	69.5	144	N/W	N/A	N/A
Mosdene	49	74.8	60.6	112	N/W	N/A	N/A
Haakdoorn	170	183	96.8	109	235	N/A	N/W
Moorddrift	93.3	109	116	82.8	81.8	N/A	157

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Temperature:

The water temperature (Table 2.10) in the system ranged between 8.3 °C at Koh-I-Noor during the winter month of July 2002 and 31.3 °C at Moorddrift during the summer month of March 2003. These drastic temperature changes can be attributed to the season but the high water temperature can also be attributed to the lack of water circulation in water

inundated with a high prevalence of aquatic vegetation. The mean water temperature during the winter months is 13.1 °C and during summer 22.6 °C.

2.3.2 Inorganic constituents:

Total Alkalinity and water hardness:

Total alkalinity is the measure of all bases dissolved in a water body (Polling, 1999). It is directly involved in the buffering capacity of a water body and is closely related to water hardness. Water hardness is determined by the CaCO₃ concentrations in the water (DWA⁶, 1996). Hardness plays a role in the toxicity of certain metals. Carbonates and bicarbonates have a dampening effect on the toxicity of certain metals (Train, 1979). As a general rule the toxicity of metals decreases as the hardness or CaCO₃ concentrations increases (Bell, 1976). A total alkalinity range of between 20 and 100 mg/ℓ is optimal for primary and secondary production in aquatic ecosystems (Stickney, 1979). Water can be classified into four main classes according to its CaCO₃ concentrations. Table 2.11 indicates these classification classes and the CaCO₃ concentration ranges that they are split into.

Table 2.10: Water temperatures measured during the study period.

Locality	Temperature (°C)						
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul	02-Nov	03-Mar
Koh-I-Noor	16.4	14.7	18	N/A	8.3	N/A	N/A
Klein Nyl Oog	19.9	19.6	21	28.7	12.7	26.6	N/A
Abba	17.8	15.7	19.9	25.7	14.3	27.4	27.4
Donkerpoort	18.9	15.2	21.8	26.5	15	N/A	27
Groot Nyl Oog	19.8	19	19.1	23.9	10.9	26.5	N/A
Groot Nyl	21.5	14.5	19.6	23.9	8.8	25.2	N/A
Sewerage Works	18.4	11.7	N/A	27.3	13.2	22	N/A
Jasper	17	14.7	20.7	23.9	5.9	21.1	27
Hessie-se-Water	19.7	10.1	19.5	24.8	6.2	21.7	N/A
Olifantspruit	20.4	13.3	19.5	25	12	21.7	23.7
Nylsvley	20.7	10.9	22.9	25.9	11.9	26.1	28.3
Bad-se-Loop	17.4	17.2	19.7	26.6	12.5	27	N/A
Tobiasoog	16.1	12.1	18.6	26.6	12.9	29	N/A
Mine	19	16.4	19	27.3	10.8	25.2	N/A
Tobias	20.2	N/W	20.3	23.9	N/W	N/A	N/A
Mosdene	17	8	21.5	21.6	N/W	N/A	N/A
Haakdoorn	22.6	15.8	20.9	25.8	16.8	N/A	N/W
Moorddrift	22.3	14.9	24.6	23.5	14.7	N/A	31.3

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Table 2.11: Water classification according to CaCO₃ concentrations in mg/ℓ.

Classification	CaCO ₃ concentration range (mg/ℓ)
Soft water	< 60
Medium water	60-119
Hard water	120-179
Very hard water	>180

Table 2.12: Measured CaCO₃ concentrations in mg/ℓ.

Locality	Alkalinity mg/l CaCO ₃				
	1-Apr	1-Aug	1-Nov	2-Mar	2-Jul
Koh-I-Noor	2.6	5	N/A	20	15
Klein Nyl Oog	1.4	5	15	20	<5
Abba	15.2	10	15	30	20
Donkertpoort	13	25	25	25	40
Groot Nyl Oog	0.6	<5	10	60	20
Groot Nyl	10	10	20	15	15
Sewerage Works	60.8	65	55	35	70
Jasper	46.8	35	85	40	<5
Hessie-se-Water	25.4	20	35	35	30
Olifantspruit	18.6	15	25	20	<5
Nylsvley	36.2	25	65	30	35
Bad-se-Loop	56	25	45	15	50
Tobiasoog	30.4	30	35	25	40
Mine	99.2	105	85	20	120
Tobias	124	N/W	105	70	N/W
Mosdene	56.4	55	60	35	N/W
Haakdoorn	122.4	140	100	85	225
Moorddrift	91.8	90	85	95	65

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

The results indicate (Table 2.12) that for the most part the water is soft in nature with the system having a median value of 35 mg/ℓ. A maximum concentration of 225 mg/ℓ was recorded at Haakdoorn in July 2002 and the minimum concentration of 0.6 mg/ℓ recorded at source of the Groot Nyl River in April 2002. The average concentrations of CaCO₃ for the 2001 and 2002 sampling periods were both 45 mg/ℓ with the standard deviations for 2001 being 37 mg/ℓ and for 2002 42 mg/ℓ. The soft nature of the water can change the speciation of the metals in the water and thus, lead to the increased toxicity of the metal found in the system.

Chlorides:

Chlorides are considered one of the major inorganic anions in water after carbonates and sulphates (Hutchinson, 1975). Although there is no TWQR available for chlorides, Ayers and Wescott (1985) considered chloride concentrations of 60 mg/l safe for agricultural use and values up to 100mg/l safe for livestock, game animals and industrial use. Aucamp and Viviers (1990) considered 250 mg/l to be the maximum limit safe for potable water sources.

Chlorides are a product of the binding action of chlorine to most elements. Chlorine is used widely as a bleaching agent in industry and for water purification. Chlorides are relatively non-toxic to organisms with an effective excretion mechanism. To organisms without an effective excretion mechanism for example Plants, chlorides can prove to be toxic (Kempster *et al.*, 1980). Chlorine may enter the aquatic environment via irrigation runoff, industrial processes and sewage effluent (DWAF⁶, 1996). Chlorides interact with metals to enhance and accelerate the oxidation processes.

Table 2.13: Recorded chloride levels at the sampling sites during the study periods.

Locality	Cl- concentration mg/l				
	1-Apr	1-Aug	1-Nov	2-Mar	2-Jul
Koh-I-Noor	3.33	<5	N/A	<5	10
Klein Nyl Oog	N/A	<5	<5	<5	6
Abba	N/A	20	<5	<5	12
Donkerpoort	N/A	<5	<5	<5	11
Groot Nyl Oog	2.9	<5	5	<5	10
Groot Nyl	N/A	16	<5	<5	7
Sewerage Works	25.63	17	20	<5	37
Jasper	13.07	20	15	9	613
Hessie-se-Water	4.01	<5	<5	6	11
Olifantspruit	N/A	14	<5	10	608
Nylsvley	10.15	16	11	<5	34
Bad-se-Loop	11.61	15	11	<5	26
Tobiasoog	5.62	6	6	<5	14
Mine	25.87	29	15	<5	29
Tobias	25.15	N/W	16	9	N/W
Mosdene	5.24	31	11	6	N/W
Haakdoorn	18.29	<5	<5	6	41
Moorddrift	8.01	9	<5	12	12

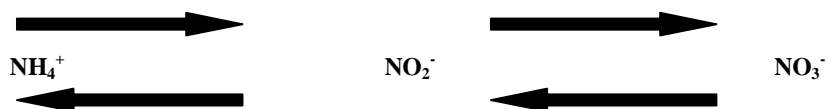
N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Nitrates and Nitrites:

Nitrates and nitrites are two inorganic constituents that are interlinked. Nitrites are the intermediate in the nitrogen cycle where ammonia is broken down by the process of nitrification to form nitrites and then once again broken down to form nitrates. This is done by the action of two bacteria (*Nitromonas* spp. and *Nitrobacter* spp.) under aerobic conditions. Under anaerobic conditions nitrates are oxidised to form nitrites by a number of

species of common facultative anaerobic bacteria, which use nitrite as an exogenous terminal electron acceptor during the oxidation of organic compounds. The basic equation for these reactions is as follows.

Nitromonas spp. and *Nitrobacter* spp.



Common facultative anaerobic bacteria

The processes of nitrification, denitrification and active uptake of nitrates by algae and higher plants is regulated by temperature and pH. Some of the bacteria used in these processes are affected by cold temperatures. High levels of nitrates impact on the eutrophication of a water body (DWAF⁶, 1996).

Ammonia and thus nitrates and nitrites are indicative of contamination by organic industrial effluents, runoff from farming activities and by far the biggest providers of ammonia into a system are sewage effluent and intensive animal culturing e.g. dairy farming (Davies and Day, 1998). High levels of nitrites are indicative of organic contamination and therefore lead to the contamination of fish flesh by metals and bacteria such as *E. coli*. Nitrites are more toxic than nitrates to aquatic organisms and their rate of uptake is influenced by temperature, pH and Cl⁻ concentrations (DWAF⁶, 1996). Nitrates are usually more stable than nitrites and are more abundant in aquatic ecosystems (DWAF⁷, 1996).

Table 2.14: Nitrate concentrations at the different sampling sites.

Locality	Nitrates (mg NO ₃ -N/l)				
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul
Koh-I-Noor	N/A	<0.2	N/W	<0.2	<0.2
Klein Nyl Oog	N/A	<0.2	<0.2	<0.2	0.3
Abba	0.08	<0.2	<0.2	<0.2	<0.2
Donkerpoort	N/A	<0.2	<0.2	<0.2	<0.2
Groot Nyl Oog	N/A	<0.2	<0.2	<0.2	<0.2
Groot Nyl	1.47	0.7	0.2	<0.2	0.5
Sewerage Works	1.32	<0.2	0.2	<0.2	3.3
Jasper	N/A	<0.2	0.2	<0.2	<0.2
Hessie-se-Water	N/A	<0.2	<0.2	<0.2	<0.2
Olifantspruit	0.08	<0.2	<0.2	0.5	<0.2
Nylsvley	N/A	<0.2	0.2	<0.2	<0.2
Bad-se-Loop	14.53	8	2.7	<0.2	6.8
Tobiasoog	N/A	<0.2	0.2	<0.2	<0.2
Mine	N/A	<0.2	<0.2	0.2	0.2
Tobias	N/A	N/W	<0.2	0.2	N/W
Mosdene	N/A	<0.2	<0.2	<0.2	N/W
Haakdoorn	N/A	<0.2	<0.2	<0.2	<0.2
Moorddrift	N/A	<0.2	0.2	<0.2	<0.2

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Table 2.14 indicates the nitrate concentrations recorded at the different localities during the study period. The site at Bad-se-Loop had the highest nitrate concentration recorded of 14.53 mg NO₃-N/ℓ during April 2001. Bad-se-Loop also consistently showed the highest nitrate concentrations through out the study period. The average nitrate concentration for the system (Excluding the increased results from the Bad-se-Loop) was 0.56 mg NO₃-N/ℓ although most of the sites indicated results of <0.2 mg NO₃-N/ℓ. The sites at the sewage treatment works and at Groot Nyl were the other sites that indicated results of more than 0.2 mg NO₃-N/ℓ. The Department of Water Affairs and Forestry Guidelines for aquaculture indicates that a nitrate concentration of less than 5 mg NO₃-N/ℓ signifies that the water is unimpacted. This hold true for most of the sites with the site at Bad-se-Loop being the only site that shows signs of any impact (DWAF⁶, 1996).

The nitrite concentrations in the system (Table 2.15) were all very low. No reading of above 0.1 mg NO₂-N/ℓ was recorded at any of the sites along the course of the study area.

The TWQR for nitrite in aquaculture lies between 0.06 and 0.25 mg NO₂-N/ℓ. This range is considered safe for many warm water fish species (DWAF⁶, 1996). A comparison of the recorded results and the TWQR would indicate that nitrites are not impacting on the organisms in the system.

Table 2.15: Nitrite concentrations at the sampling sites.

Locality	Nitrite (mg NO ₂ -N/ℓ)				
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul
Koh-I-Noor	N/A	<0.1	<0.1	<0.1	<0.1
Klein Nyl Oog	N/A	<0.1	<0.1	<0.1	<0.1
Abba	N/A	<0.1	<0.1	<0.1	<0.1
Donkerpoort	N/A	<0.1	<0.1	<0.1	<0.1
Groot Nyl Oog	N/A	<0.1	<0.1	<0.1	<0.1
Groot Nyl	N/A	<0.1	<0.1	<0.1	<0.1
Sewerage Works	N/A	<0.1	0.3	<0.1	<0.1
Jasper	N/A	<0.1	<0.1	<0.1	<0.1
Hessie-se-Water	N/A	<0.1	<0.1	<0.1	<0.1
Olifantspruit	N/A	<0.1	<0.1	<0.1	<0.1
Nylsvley	N/A	<0.1	<0.1	<0.1	<0.1
Bad-se-Loop	N/A	<0.1	<0.1	<0.1	<0.1
Tobiasoog	N/A	<0.1	<0.1	<0.1	<0.1
Mine	N/A	<0.1	<0.1	<0.1	<0.1
Tobias	N/A	N/A	N/W	<0.1	N/W
Mosdene	N/A	<0.1	<0.1	<0.1	N/W
Haakdoorn	N/A	<0.1	<0.1	<0.1	<0.1
Moorddrift	N/A	<0.1	<0.1	<0.1	<0.1

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Sulphates:

Sulphates are the oxy anions of sulphur in the +IV oxidation state (DWAF¹, 1996). They are non toxic to humans and animals but can become so at extremely high concentrations. They are slightly less toxic to plants than chlorides (Kempster *et al.*, 1980).

The sulphate levels in the system (Table 2.16) are typical of surface water of 5 mg/l (DWA⁸, 1996). Most of the readings are less than 5 mg/l. The majority of the readings recorded during all sampling months other than April 2001 are <5 mg/l due to minimum detection limits on the apparatus used during analysis. The readings could however be somewhat closer to those found in April 2001. A maximum value of 520 mg/l was recorded at Olifantspruit in July 2002 with a minimum value recorded at Haakdoorn (0.19 mg/l) during April 2001. Most of the sulphate levels recorded are within the TWQR of between 0 and 200 mg/l set out for domestic consumption. No TWQR is available for aquatic ecosystems and so the TWQR for domestic use has been used as suitable for the organisms. Jasper and Olifantspruit were the only two sites that had readings that fell outside these guideline values with readings of 363 and 520 mg/l respectively.

These readings were recorded in July 2002. These values however are below the median guideline value prescribed by Kempster *et al.* in 1980 of 1400 mg/l.

Table 2.16: Sulphate levels in the Nyl River System.

Locality	Sulphates mg/l				
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul
Koh-I-Noor	0.33	<5	<5	<5	<5
Klein Nyl Oog	N/A	<5	<5	<5	18
Abba	0.31	<5	<5	<5	<5
Donkerpoort	0.62	<5	<5	<5	<5
Groot Nyl Oog	N/A	<5	<5	<5	<5
Groot Nyl	N/A	<5	<5	<5	<5
Sewerage Works	10.76	<5	<5	<5	17
Jasper	2.43	<5	<5	<5	363
Hessie-se-Water	0.25	<5	<5	<5	<5
Olifantspruit	0.55	<5	<5	<5	520
Nylsvley	0.44	<5	<5	<5	<5
Bad-se-Loop	8.93	<5	<5	<5	9
Tobiasoog	1.4	<5	<5	5	<5
Mine	6.43	<5	<5	<5	<5
Tobias	3.03	N/A	<5	6	N/W
Mosdene	N/A	<5	5	<5	N/W
Haakdoorn	0.19	<5	<5	5	<5
Moorddrift	N/A	<5	<5	13	<5

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

Phosphates:

Phosphates can be either inorganic or organic in nature. They are measured as orthophosphates, total dissolved phosphorus or total inorganic phosphate. Orthophosphate is the only form of phosphorus that is immediately available for aquatic biota. It can be transformed into an available form via natural processes (DWA⁷, 1996). Phosphates are essential to a wide variety of aquatic organisms. In animals they play a vital role in the building of nucleic acids and the storage and use of energy in cells. Plants readily utilise it in unimpacted waters by converting it into cell structures in the process of photosynthesis.

Phosphates enter the water in one of two ways, either naturally or by anthropogenic activities.

Naturally they enter the water body by the weathering of rocks and the decomposition of organic matter. Anthropogenic activities that release phosphates into the water include domestic and industrial effluents (point source), atmospheric precipitation, urban run off and drainage from agricultural activities (non- point source) (Dallas and Day, 1993).

Table 2.17 indicates the phosphate levels determined in the Nyl River System. The maximum value recorded (4540 $\mu\text{g}/\ell$) was recorded at the Sewerage Works. This would indicate that the Sewerage Works has extremely high phosphate values and the system would thus be hypertrophic in nature. Only one other reading higher than 200 $\mu\text{g}/\ell$ was recorded at the Sewerage Works.

The rest of the readings were less than 200 $\mu\text{g}/\ell$ which would indicate that the entire system is eutrophic, and susceptible to algal blooms and increased aquatic macrophyte growth.

Table 2.17: Orthophosphate levels in the Nyl River system.

Locality	Orthophosphates mg/l				
	01-Apr	01-Aug	01-Nov	02-Mar	02-Jul
Koh-I-Noor	N/A	<0.2	<0.2	<0.2	<0.2
Klein Nyl Oog	N/A	<0.2	<0.2	<0.2	<0.2
Abba	N/A	<0.2	<0.2	<0.2	<0.2
Donkerpoort	N/A	<0.2	<0.2	<0.2	<0.2
Groot Nyl Oog	N/A	<0.2	<0.2	<0.2	<0.2
Groot Nyl	N/A	<0.2	<0.2	<0.2	<0.2
Sewerage Works	4.54	<0.2	0.4	<0.2	2.2
Jasper	N/A	<0.2	<0.2	<0.2	<0.2
Hessie-se-Water	N/A	<0.2	<0.2	<0.2	<0.2
Olifantspruit	N/A	<0.2	<0.2	<0.2	<0.2
Nylsvley	N/A	<0.2	<0.2	<0.2	<0.2
Bad-se-Loop	N/A	<0.2	<0.2	<0.2	<0.2
Tobiasoog	N/A	<0.2	<0.2	<0.2	<0.2
Mine	N/A	<0.2	<0.2	<0.2	<0.2
Tobias	N/A	N/W	<0.2	<0.2	N/W
Mosdene	N/A	<0.2	<0.2	<0.2	N/W
Haakdoorn	N/A	<0.2	<0.2	<0.2	<0.2
Moorddrift	N/A	<0.2	<0.2	<0.2	<0.2

N/A: not available, N/W: no water. Light grey cells indicate tributaries.

The TWQR for phosphates is listed in table 2.18. Different ranges have been set out for different levels of nutrient loads in a water body.

Table 2.18: List of TWQR's (DWA⁷, 1996).

Average Summer Inorganic Phosphorus Concentration (ug/l)	Effects
<5	Oligotrophic conditions; usually moderate levels of specie diversity; low productivity with rapid nutrient cycling; no nuisance growth of aquatic plants or blue green algae
5 – 25	Mesotrophic conditions; usually high levels of species diversity; usually productive systems; nuisance growth of aquatic plants and blue green algal blooms; algal blooms usually non toxic
25- 250	Eutrophic conditions; usually low levels of specie diversity; usually highly productive systems, with nuisance growth of aquatic plants and blooms of blue green algae; algal blooms may include species which are toxic to man, livestock and wildlife.
>250	Hypertrophic conditions; usually very low levels of specie diversity; usually very highly productive system; nuisance growth of aquatic plants and blooms of blue green algae, often including species which are toxic to man, livestock and wildlife.

2.3.3 Metals and metalloids:

All living organisms need a certain amount of trace elements for effective and proper metabolic functioning (Galvin, 1996). The functions of these elements can be varied and include: (1) the role played in physiological processes, (2) their requirement for respiration and gonadal development and (3) their role as an integral part of protein and enzymatic systems (Heath, 1987). Natural waters contain these elements at low concentrations and an increase in the concentrations can lead to an increase in the accumulation by the aquatic organisms (Nussey *et al.*, 1999). Metal levels in water can be either toxic or non toxic to aquatic organisms, depending on the metal, concentration or whether the element is an essential element or not. The Department of Water Affairs and Forestry has set out a list of guideline values with which to compare metal levels in a water body and to assess if they are toxic or not. These guideline values form a target water quality range (TWQR). If the levels fall below the TWQR organisms can become susceptible to disease. If the concentrations are exceed the upper limit of the TWQR then the elements become toxic to the aquatic organisms (du Preez *et al.*, 1998). The presence of heavy metals in water becomes harmful when the concentrations present rise above that of the background concentrations found in water and sediment.

Metals enter the natural surface water systems either naturally or via anthropogenic activities. Natural processes include chemical geological weathering and decomposition of biotic matter. Metal concentrations can also be increased by anthropogenic activities such as industrial pollution, agriculture and mining. Industrial pollution is usually point source in nature where as agriculture and mining activities act as diffuse sources of metal contamination (Heath and Claasen, 1999). Mining and industrial effluent are usually the major sources of increased metal concentrations in rivers. There are two factors that contribute to the damaging effect of metals as environmental pollutants namely (1) the inadequacy of biological degradation of inert metals and (2) the trend of metals to accumulate and largely remain in the aquatic environment (Robinson and Avenant-Oldewage, 1997).

Metals entering fresh water systems could undergo various changes before temporary or final stability is reached. In aqueous solutions metal ions can be complexed with water (Hydrated) or associated with organic or inorganic matter through the process of adsorbtion, chemical combination or complex formation (Förstner and Muller, 1973). The ambient water quality determines the actions of these processes, for example a low pH causes some

metals (e.g. Al) to become more toxic (Klein *et al*, 1975). The specie of the metal occurring in the water plays an important role in the bioavailability and toxicity of that metal (Wade *et al*, 1995). The formation of complexes greatly reduces the toxicity of the free metal ion. The final toxicity of the metal is further influenced by the interactions between the pollutant, the developmental stage of the aquatic organism and the interspecies variations in susceptibility to metals (Hellawell, 1986; Ellis, 1989).

From this list of the analysed elements (Table 2.19) comparisons were made to the TWQR for aquatic ecosystems. Due to the complex nature of the interactions of metals in water and organisms TWQR's for most of the elements are not available at this time. Those elements that exceeded the existing TWQR's will be discussed in this chapter.

Table 2.20 lists the metals for which TWQR's that exist and the specified guideline values. Ranges are taken from two different sources and where discrepancies occur the lowest value has been used for comparison.

Table 2.19: Table of the elements analysed via ICP-MS.

Element symbol						
Li	Cr	Rb	I	Ga	Pr	Yb
B	Mn	Sr	W	Ge	Nd	Lu
Na	Fe	Zr	Tl	Y	Sm	Hf
Mg	Co	Mo	Pb	Ru	Eu	Ta
Al	Ni	Ba	Hg	Rh	Gd	Re
Si	Cu	Pd	Bi	Nb	Tb	Os
K	Zn	Ag	U	Te	Dy	Ir
Ca	As	Cd	Be	Cs	Ho	Pt
Ti	Se	Sn	P	La	Er	Au
V	Br	Sb	Sc	Ce	Tm	Th

Table 2.20: Guideline values for metal content in aquatic ecosystems.

Element	DWAF ⁷ 1996 (Aquatic Ecosystems)	Canadian Water Quality Guidelines
Al	≤10ppb	0.1 ppm
As	≤10	0.05
Cd	≤0.25 med water	0.8
Cr	≤7	0.002
Cu	≤0.3 med hard water	0.002
Pb	≤0.5	0.001
Mn	180	
Hg	≤0.04	0.001
Se	≤2	0.001
Zn	≤2	0.03

Ten of the 70 elements analysed indicated values above the guideline values set out by the Department of Water Affairs and Forestry. These elements are listed in table 2.20, as well as the summary statistics. It is however important to note that these are total metal concentrations and not the dissolved concentration. It also does not differentiate between the more toxic species of the metals such as hexavalent chromium.

Aluminium:

Aluminium is a metal that is strongly dependent on pH. In the soluble state it forms the highly toxic hexahydrate form. Developmental stage and organism species determines the toxicity of aluminium. It can have the following possible deleterious effects on organisms, (1) interference with osmotic and ionic balance, (2) respiratory defects as a result of coagulation of mucous on the gills, (3) interference with the functioning of calcium regulating proteins, and (4) calcium metabolism in the brain and other organs (DWAF⁷, 1996). Aluminium toxicity increases as the water pH decreases (Savory and Wills, 1991). Possible sources of aluminium are industrial effluents and aluminium is used as a flocculant in the purification of drinking water (Kempster *et al.*, 1980).

Aluminium concentrations ranged between 2089 µg/l at the source of the Tobiasspruit (Tobiasoog) during November 2001 to a lowest recorded value of 18 µg/l, at the source of the Groot Nyl River (Groot Nyl Oog), in July 2002. Various readings of 0 µg/l were recorded during July 2002 with July 2002 have significantly lower readings than the other sampling months. November 2001 had the highest recorded aluminium concentrations. Although aluminium concentrations were high they are significantly lower than aluminium levels recorded in the upper catchment of the Olifants River during the autumn of 1994. A comparison of the levels (2089 µg/l as apposed to 43 680 µg/l) indicates that the levels are almost one twentieth of those found in the upper catchment of the Olifants River (van Vuren *et al.*, 1999). Average aluminium levels recorded during the low flow period between August and November 2001 (913.58 µg/l) were also found to be lower then the average low flow level (8840 µg/l) found in unfiltered water by Greenfield (2001) during a previous study on Nyl River. Figures 2.24 A-F indicate that all the aluminium concentrations recorded were above the TWQR of 10 µg/l for aquatic ecosystems.

Chromium:

Chromium is a relatively scarce metal that can occur in several states. Chromium VI is the most toxic state. It is the highly oxidized state of chromium and is highly soluble at all pH ranges. Chromium II and III are the more reduced states of the chromium ion and are seen to be less toxic. In aquatic environments chromous compounds tend to be oxidized to chromic forms, whilst chromium VI can be reduced to form chromium III by heat and the presence of organic matter and reducing agents. Chromium exists in natural waters in three oxidation states. These are however difficult to distinguish due to inter-conversion reactions. The presence of oxydisable organic matter and iron (II) salts encourages reduction to lower and less toxic oxidation states (Chromium III) (DWAF⁷, 1996). Water hardness has a significant effect on the toxicity. Chromium (III) becomes more toxic in soft water (CCME, 1992). The toxicity of fish and invertebrates to chromium is comparable in soft water. Phytoplankton have been shown to be more sensitive to chromium than are fish (U.S. EPA 1985e). According to the Canadian Water Quality Guidelines (CCME, 1992) the total chromium concentration in water should not exceed 200 µg/l.

Table 2.21: Table of summary statistics for metal concentrations in water ($\mu\text{g}/\ell$).

		Aug-01	Nov-01	Mar-02	Jul-02
Aluminium	Average	779.83	1047.33	228.50	16.17
	Stdev	± 253.19	± 552.73	± 119.17	± 28.81
	25th percentile	681.25	639.00	171.00	0.00
	75th percentile	945.25	1343.75	257.75	19.50
	Maximum	1144.00	2089.00	598.00	97.00
	Minimum	0.00	313.00	0.00	0.00
Chromium	Average	403.72	42.56	41.78	0.00
	Stdev	± 1119.29	± 12.46	± 10.64	± 0.00
	25th percentile	0.00	33.25	43.00	0.00
	75th percentile	11.75	47.75	45.75	0.00
	Maximum	3679.00	71.00	48.00	0.00
	Minimum	0.00	26.00	0.00	0.00
Manganese	Average	243.83	111.11	407.11	63.67
	Stdev	± 682.97	± 91.93	± 1164.78	± 98.58
	25th percentile	5.25	49.50	52.75	11.25
	75th percentile	25.00	136.00	185.75	57.50
	Maximum	2882.00	343.00	5047.00	348.00
	Minimum	0.00	29.00	0.00	0.00
Iron	Average	1578.94	3053.56	2815.17	376.17
	Stdev	± 3709.14	± 1988.45	± 4425.11	± 548.40
	25th percentile	0.00	1780.25	1061.75	120.00
	75th percentile	216.25	3758.25	2540.75	389.00
	Maximum	13310.00	9306.00	19901.00	2321.00
	Minimum	0.00	852.00	0.00	0.00
Copper	Average	21.06	53.67	3.17	1.17
	Stdev	± 42.28	± 168.79	± 1.50	± 1.34
	25th percentile	4.25	7.50	2.25	0.00
	75th percentile	9.00	18.75	3.75	1.75
	Maximum	151.00	729.00	7.00	4.00
	Minimum	0.00	4.00	0.00	0.00
Zinc	Average	520.50	438.33	111.94	1.28
	Stdev	± 496.10	± 264.28	± 28.50	± 5.42
	25th percentile	114.25	189.50	112.25	0.00
	75th percentile	1037.00	692.25	121.75	0.00
	Maximum	1350.00	871.00	130.00	23.00
	Minimum	0.00	132.00	0.00	0.00

Arsenic	Average	25.22	8.17	1.22	0.78
	Stdev	± 23.50	± 6.65	± 1.06	± 1.35
	25th percentile	11.25	3.25	0.25	0.00
	75th percentile	25.00	10.75	2.00	1.50
	Maximum	79.00	25.00	4.00	4.00
	Minimum	0.00	1.00	0.00	0.00
Selenium	Average	3.94	2.00	6.17	2.39
	Stdev	± 2.44	± 1.41	± 2.57	± 2.00
	25th percentile	3.00	1.00	5.00	1.00
	75th percentile	4.75	3.00	7.00	3.00
	Maximum	10.00	5.00	11.00	7.00
	Minimum	0.00	0.00	0.00	0.00
Cadmium	Average	21.33	0.00	0.00	0.00
	Stdev	± 5.39	± 0.00	± 0.00	± 0.00
	25th percentile	22.00	0.00	0.00	0.00
	75th percentile	23.00	0.00	0.00	0.00
	Maximum	24.00	0.00	0.00	0.00
	Minimum	0.00	0.00	0.00	0.00
Lead	Average	38.11	72.06	7.22	2.06
	Stdev	± 19.75	± 32.40	± 2.05	± 0.87
	25th percentile	26.75	47.50	7.00	2.00
	75th percentile	48.25	82.50	8.00	2.75
	Maximum	89.00	175.00	10.00	3.00
	Minimum	0.00	43.00	0.00	0.00

From the data indicated in table 2.21 it can be seen that chromium levels ranged between 3679 µg/l, at Nylsvley in August 2001, to 26 µg/l, at Donkerpoort in November 2001. Various sites throughout the study period however did have zero readings. Although August 2001 had the highest recorded chromium levels it was only at seven of the 18 sampled localities. Cr levels were however recorded at all eighteen localities during the November 2001 and March 2002 surveys. These levels were however all below the TWQR, for aquatic ecosystems, for both chromium (VI) (70 µg/l) and (III) (120 µg/l). Chromium levels only seemed to be a problem (3675 µg/l) during August 2001. This alarmingly high level was however a once off reading and thus no cause for concern. These results are clearly illustrated in figures 2.25 A-F.

Manganese:

Manganese is an essential element in animals and plants (Health and Welfare Canada, 1980). Manganese is a functional component in nitrate assimilation and an essential catalyst of numerous enzyme systems in animals, plants and bacteria (DWAF⁷, 1996). In vertebrates manganese deficiencies can lead to skeletal deformities and a reduction in reproduction processes. High concentrations are toxic and can lead to disturbances in various pathways

such as the central nervous system caused by the inhibition of dopamine formation (DWAF⁷, 1996).

External sources of manganese into the environments include (1) the steel industry and the production of dry cell batteries, (2) the fertilizer industry, (3) the chemical industry and (4) acid mine drainage. Dissolved manganese concentrations are influenced by redox potential, dissolved oxygen, pH and organic matter. In surface waters divalent manganese (Mn^{2+}) is rapidly oxidized to insoluble manganese dioxide (MnO_2), which settles out of the water column (DWAF⁷, 1996).

In well oxygenated waters manganese levels are lower than in waters with low dissolved oxygen concentrations. This is because most soluble manganese compounds are rapidly oxidised and precipitate out (Galvin, 1996). As the pH in the system decreases the toxicity of manganese increases, as the manganese becomes more prevalent in its ionic state (Wang, 1987). In natural fresh water the manganese concentration rarely exceeds concentrations of 1000 $\mu g/l$ (Hellowell, 1986).

Manganese levels ranged between 5047 $\mu g/l$, at Mosdene in March 2002, and 3 $\mu g/l$, at Olifantspruit in August 2001. For the most part manganese levels were higher during November 2001 and March 2002 (high flow period) than in August 2001 and July 2002 (low flow period). This would indicate that manganese levels are higher during the rainy season and that the increased water flow released manganese that has precipitated out during periods of low to no flow. The average manganese concentrations for the different sampling months (Table 2.21) would indicate this not to be the case, but high concentrations at Mosdene (2882 $\mu g/l$) and at Mine (639 $\mu g/l$) have skewed the August 2001 average concentrations. The same applies for the Mosdene reading (5047 $\mu g/l$) during November 2001. Zero readings obtained at Tobias, Mosdene and Koh-I-Noor have been calculated in the averages, but they are due to the sites being dry at the time of sampling and thus add an extra bias. Figures 2.27 (A-F) illustrate the localities that indicated readings above the TWQR (180 $\mu g/l$). These readings show no clear trend as to increases and decreases in manganese concentrations, although Mosdene, Jasper, Nylsvley, Abba, Klein Nyl Oog and Mine indicate the sites with high levels of manganese compared to the TWQR for aquatic ecosystems.

Iron:

On the basis of limited toxicity and bioavailability iron is classified as a non-critical element. Two common oxidation states are found in water namely: divalent ferrous iron (Fe^{2+}) and trivalent ferric iron (Fe^{3+}) (DWAF⁷, 1996). Iron is present in natural waters in varying quantities depending on the geological makeup of the specific region (Train, 1979). Iron originates in natural waters via rock dissolution and via the anthropogenic activities of steel production and other industrial waste waters (Galvin, 1996). The main source of iron entering the study area through human activities is via effluent from sewage treatment works (DWAF⁷, 1996). Iron is readily oxidized and at high concentrations may lead to oxygen depletion in the water (Dallas and Day, 1993). In natural fresh waters total iron is found at levels between 0.5-50 mg/l (WHO, 1993). In surface waters iron is generally in the trivalent form ranging between 0.1 and 0.3 mg/l , this is due to the precipitation of $Fe(OH)_3$ at pH 7.5 and lower. Divalent salts start to precipitate out at pH 6 and lower (Galvin, 1996). In organic rich waters, such as waters in wetlands, divalent iron and the organic matter form stable Fe^{2+} -organic matter complexes, which cause serious problems in the subsequent treatment of these waters (Galvin and Mellado, 1993).

Iron is an essential element and is required in respiratory enzymes of all organisms. It makes up the basic component of the haeme containing respiratory pigment (haemoglobin), and is present in cytochromes and several redox enzymes. (DWAF⁷, 1996; Galvin, 1996).

Gills, liver and kidneys are the main accumulation points for iron (Nussey *et al.*, 1999). One of the main toxic actions of iron is that it precipitates on the gill surface causing increased mucous production and suffocation (Muniz and Leivestad, 1980).

Iron concentrations ranged between 19901 µg/l, at Mosdene in March 2002, and 110 µg/l, at Jasper in July 2002. Various values of 0 µg/l were recorded but this is mainly in August 2001. The averages in table 2.21 for iron indicate that iron levels are higher during November 2001 (3053.4 µg/l) and March 2002 (2815.2 µg/l), High flow periods, than in August 2001 (1578.9 µg/l) and July 2002 (376.2 µg/l), low flow periods. The maximum concentration of total iron recorded (19.901 mg/l) falls within the range indicated by Galvin (1996), of between 0.5 and 50 mg/l, for nature fresh water.

Figures 2.27 (A-F) illustrate the high levels of iron in the water and the non-uniform nature of the peaks obtained in the water sampled.

Copper:

Copper is a common environmental metal and is found in the Cu⁺, Cu²⁺ and Cu³⁺ oxidation states. It is an essential metal in cellular metabolism, but is also potentially highly toxic to fish (Grosell *et al.*, 1997). The toxicity of copper in water is largely attributed to monovalent (Cu⁺) and CuOH⁺, which is present in small quantities in fresh water. Divalent (Cu²⁺) ions are rarely found in the loose form in water as they bind rapidly to inorganic and organic substances and can be adsorbed to particulate matter (Robinson and Avenant-Oldewage, 1997).

Copper dissociates in acidic conditions to the divalent form and thus the toxicity of copper increases as the pH decreases (Benedetti *et al.*, 1989). In alkaline conditions it tends to precipitate out of water (Grosell *et al.*, 1997). Toxicity of copper increases in water when (1) there is a reduction in water hardness, (2) there is a decrease in dissolved oxygen concentrations, and (3) it acts synergistically with other elements found in the water column (DWAF⁷, 1996; Benedetti *et al.*, 1989; EIFAC, 1978).

The toxicity of copper is greater in the presence of zinc than it is as a single toxicant (Scheinberg, 1991). Copper toxicity decreases with an increase in alkalinity. The presence of certain compounds, such as sodium nitrate, sodium nitrite and calcium, can also increase the toxicity of copper to fish (DWAF⁷, 1996).

Copper enters the environment through the oxidation of sediment, but heavy increases are due to anthropogenic activities. The main routes of copper into the environment with respect to this study are: (1) sewage treatment effluent, (2) aquatic algacides, (3) the runoff from fungicides and pesticides from agricultural land use, and (4) the manufacturing and use of fertilizers (Kotze *et al.*, 1999; Nussey *et al.*, 1999). Copper compounds such as copper sulphate are effective in the treatment of water to eliminate algae and micro-organisms such as *E. coli*. This action is made possible by the copper obstructing the micro-organisms membrane wall preventing oxygen uptake. Moderate levels of copper in the water can have negative effects on fish (Galvin, 1996).

Copper is an essential element that aids in bone formation, maintenance of myelin and the synthesis of haemoglobin (Nussey *et al.*, 1999). Its accumulation has a specific pattern of uptake namely liver > gills > skin and muscle (Kotze *et al.*, 1999). Copper accumulation takes place across the gills and through the digestion of food and sediment. In the food chain, tolerant plants and invertebrates may accumulate copper posing a risk to organisms higher up in the food chain. About half the ingested copper is excreted in the faeces (Scheinberg, 1991). In comparable ecosystems water plants accumulates up to three times more copper than terrestrial plants.

The copper concentrations ranged between 729 µg/l, at Jasper during November 2001, and 0 µg/l, at both the sources of the Groot (Groot Nyl Oog) and Klein (Klein Nyl Oog) Nyl Rivers during July 2002. Table 2.21 indicates that August 2001 and November 2001 have

higher copper concentrations than March and July 2002. This would indicate that the copper concentrations are not season related but rather temporally. Figures 2.28 (A-F) illustrates that all recorded concentrations of copper were above the TWQR of 0.3 µg/l. This would indicate that the system naturally has a high copper content.

Zinc:

Zinc is an essential micronutrient for all organisms because it forms the active site for various metalloenzymes (DWAF⁷, 1996). It is also essential for life (Sola and Duran, 1994). Zinc occurs in water in two oxidation states namely: as the metal and as a divalent zinc ion (Zn²⁺). It is the divalent form in water that is toxic to fish and aquatic organisms (DWAF⁷, 1996). Zinc in waters is usually low being detected as inorganic, ionic or colloidal compounds. In this way the mean values of zinc in surface waters are usually lower than 10 µg/l (Galvin, 1996). Zinc chloride and zinc sulphates in water react with dissolved carbon dioxide yielding hydroxides and carbonates which are adsorbed onto sediments and muds of river beds or lakes (Galvin, 1996).

Zinc has an antagonistic and toxic effect in the uptake of cadmium. It helps in the synthesis of metallothionines, which bind to cadmium ions to detoxify them (Kargin and Çoşun, 1999). According to Kargin and Çoşun (1999) Zn²⁺ ions may inhibit the uptake of cadmium by the gills but increases the movement of cadmium to the internal organs. The antagonistic effect of zinc arises due to the competition between Zn and Cd for protein binding sites.

Gills are usually the first organs to be affected by metal pollution due to their direct contact with the water (Hoogstrand *et al.*, 1994). Zinc causes death to the fish as it destroys the gill tissue. Acute increases in water borne Zn²⁺ concentrations impair branchial Calcium influx. This causes hypocalcaemia by inhibiting Ca²⁺ transporting ATPase in baso-lateral membranes of chloride cells. Zinc is essential for maintaining structure and functioning of cell membranes. It binds to the plasma and internal membranes to stabilize them (Viarengo, 1988). It also plays a role in biological functions such as enzyme activity, nucleic acid metabolism, protein synthesis and hormone activity (Ohnesorge and Wilhelm, 1991).

Zinc shows the greatest bioconcentration factor in skin and bone although the liver, gills and kidney also accumulate it to a considerable extent (Heath, 1987). The LC₅₀ value for zinc is higher in warm water than in cold water but fish are more sensitive to zinc at cold temperatures. At increased temperatures there is an increased concentration of zinc in the gills (Rattner and Heath, 1995). Water hardness also plays a role in Zn toxicity. As the water hardness increases the Zn uptake rate across the gills decreases thus reducing zinc caused lethality in fish. Toxicity also increases as the quantity of dissolved oxygen decreases (Rattner and Heath, 1995). Copper increases Zn toxicity in soft water and prolonged exposure can cause liver necrosis (DWAF⁷, 1996).

Zinc concentrations ranged between 1350 µg/l, at Mosdene in August 2002, and 23 µg/l at Koh-I-Noor in July 2003. Average zinc concentrations were higher during August 2002 and November 2002 than those recorded in March 2003 and July 2003. This would indicate that the good rainy season experienced at the end of 2002 had a flushing effect on the system. All zinc concentrations recorded were above the TWQR of 2 µg/l. Table 2.21 indicates the summary statistics for zinc in the water of the Nyl River system. The statistics for July 2003 indicate average zinc concentrations below the TWQR, but these statistics are skewed by all sites except one (Koh-I-Noor) having zinc concentrations of 0 µg/l. The reasons for this are unclear except for the sites of Tobias and Mosdene where the site had no water. Figures 2.29 (A-F) illustrate the spatial and temporal fluctuations in zinc concentrations and their deviation from the TWQR.

Arsenic:

Arsenic is a metalloid element (Rodriguez *et al.*, 2003). It is an analyte of high concern to the scientific community due to its toxic properties (Pizzaro *et al.*, 2003). Arsenic is widely distributed as a trace constituent in rocks and soils, natural waters and organisms. It can be mobilized mainly by weathering and microbial activities (Garcia-Sanchez and Alvarez-Ayuso, 2003). Arsenic may enter the natural environment via either point source or diffuse sources (Carbonell *et al.*, 1998). Anthropogenic activities that can lead to an increase in arsenic levels include (1) agricultural use of pesticides and fertilizers, (2) mining wastes, (3) industrial processes and (4) mineral debris (Rodriguez *et al.*, 2003; Garcia-Sanchez and Alvarez-Ayuso, 2003).

Arsenic concentrations found in natural waters range from less than 0.5 µg/l to more than 5000 µg/l (Huang *et al.*, 2003). Arsenic has effects on the functioning of the central nervous system, genotoxicity and cell disruption (Rodriguez *et al.*, 2003). Arsenic is also carcinogenic, mutagenic and teratogenic (Newman and McIntosh, 1991). Arsenic, although toxic, can also play a role in the conversion of methionine to its metabolites taurine, labile methyl and possibly the polyamines (Rodriguez *et al.*, 2003). Arsenical compounds may also be used to treat trypanosomiasis. In fish arsenical compounds can have effects on fish embryos causing skeletal malformations at concentrations as low as 250 µg/l (Newman and McIntosh, 1991).

Arsenic concentrations ranged between 79 µg/l, at Mosdene in August 2002, and 0 µg/l at various localities. The average arsenic concentrations (Table 2.21) indicate that August 2002 had the highest average concentration of 25.22 µg/l. This average concentration is higher than the TWQR of 10 µg/l for aquatic ecosystems. The other localities all had average concentrations below the TWQR. Results indicate that August 2002 and November 2002 had significantly higher average arsenic concentrations than March 2003 and July 2003. This could be attributed to the flushing effect that the good rainy season had on the March and July sampling period. Arsenic concentrations approximately five times greater than the TWQR were experienced at four localities during August 2002. The sites Koh-I-Noor and Klein Nyl Oog are situated at the source of the Klein Nyl River and these high concentrations could be attributed to natural arsenic levels. The sites situated at the Modimolle Sewage Treatment Works and Mosdene may be seen as point sources of arsenic pollution. Figures 2.30 (A-F) illustrate the spatial and temporal arsenic levels throughout the study area and their deviations from the TWQR.

Cadmium:

Cadmium is a non-essential volatile trace element. In natural waters it occurs primarily as a divalent ion (Cd^{2+}), cadmium chloride and cadmium carbonate. Cadmium enters the environment via a number of anthropogenic activities. The ones most prevalent to this system are via (1) Sewage sludge, (2) Fertilizers and (3) Pesticides (DWAF⁷, 1996).

Cadmium toxicity increases as temperature increases due to the suppression of calcium ion by the cadmium ions. Cadmium suppresses ventilation in catfish but causes elevation in gill ventilation in most other fish species. Cadmium precipitates in hard waters (Rattner and Heath, 1995). As pH decreases the toxicity of cadmium increases, due to the fact that cadmium is highly soluble in acidified waters (DWAF⁷, 1996).

Cadmium accumulation takes place in the kidneys, liver, gastro-intestinal tract and gills. It does not accumulate in muscle tissue. It can be excreted via the faeces of organisms (Spry and Wiener, 1991). The toxic effects of cadmium are worsened by the fact that it has a long half-life. This means that it stays in the tissues for a long time after accumulation thus making it available for longer periods of time to prospective predators (WHO, 1992).

Cadmium causes decreased growth rates and can have negative effects on embryonic development (Newman and McIntosh, 1991). Cadmium has various physiological effects,

and can cause anaemia due to the destruction of red blood cells. It also causes a disturbance in renal functioning by obstructing the renal tubes. It can cause hypocalcaemia, which is the destruction of skeletal bones (Larsson *et al.*, 1994).

Another physiological effect of cadmium is the interruption of ionic balance by altering the permeability of cell membranes. It affects the passive movement and active transport processes by inhibiting Na and K ATPases. This toxicant also displaces the beneficial metals from their active binding sites of enzymes and binds to the deactivating site on the molecule (Viarengo, 1985).

Fish can survive and accumulate high levels of cadmium in the liver. The cadmium is immobilized to a non-toxic form by the formation of Cd-thionien complex (Carpene *et al.*, 1987). Bioavailability of cadmium to benthic organisms is limited by its strong adsorption to environmental components such as sediment and organic matter (Sanders *et al.*, 1999).

Cadmium concentrations during August 2002 ranged between 24 µg/l, at Abba and HSW, and 21 µg/l, at Groot Nyl Oog and Koh-I-Noor. The site located at Tobias had a zero reading as it was dry. Zero readings were also obtained at all the sites during the other sampling months (November 2002, March 2003 and July 2003). Reasons for this are unclear. All the readings obtained for August 2002 were above the TWQR (0.25 µg/l) for aquatic ecosystems. This however is no cause for concern as the cadmium levels are very similar throughout the system with the average for the system being 21.3 µg/l ± 2.6 µg/l and the sources of the Groot and Klein Nyl Rivers having concentrations of 21 µg/l and 22 µg/l respectively. Figures 2.31 (A-F) illustrate the uniformity of cadmium concentrations recorded during August 2002, and the lack of readings for the other sampling months.

Lead:

Lead is a ubiquitous non-essential trace element (Ewers and Schlipkoter, 1991). It exists in several oxidation states namely, Pb, Pb⁺, Pb²⁺ and Pb⁴⁺. Divalent lead (Pb²⁺) is the most toxic form and is bio-accumulated by aquatic organisms. It enters the environment via a number of different ways namely: (1) Combustion of fossil fuels, (2) Industrial and municipal waste, (3) Precipitation fall out and (4) Street runoff (DWAF⁷, 1996; Manahan, 1993).

Lead pollution is localized near points of discharge due to its low solubility.

The bioavailability of lead (Pb²⁺) increases as pH decreases. An increase in water hardness decreases the toxicity of lead. Lead toxicity increases with decreased dissolved oxygen in the water. The toxicity of lead is also dependent on the life stage of the organism and the presence of organic material (Hellowell, 1986). In South African surface waters lead is usually particulate in form and results in a decreased bioavailability to fish (Seymore *et al.*, 1995). Uptake of lead usually takes place across the gills (Coetzee, 1996). Animal exposure experiments have shown that lead is a renal and vascular poison (Browning, 1961). Lead poisoning in the wild have been said to be the cause of central nervous disorders, motor abnormalities and blindness (Ewers and Schlipkoter, 1991). Low levels of lead affect fish by causing the formation of a thin film of mucous over the gills and subsequently the entire body. Death of the fish is then caused by suffocation. Lead does not appear to bio magnify through the food web (DWAF⁷, 1996).

Lead concentrations ranged between 175 µg/l, at the Sewerage Works in November 2002, and 2 µg/l at various sites during July 2003. August and November 2002 indicate average lead concentrations higher than those recorded during March and July 2003. This can be attributed to the good rainy season experienced and the resultant increased street runoff. Lead levels in 2002 were significantly higher than those recorded in 2003. All lead concentrations recorded were above the TWQR (0.5 µg/l) for aquatic ecosystems. The increased levels experienced (Figure 2.32) in November can be attributed to the washing effect of run off from the roads around the urban areas as well as the old and new N1 north.

Although runoff increased the lead levels in the water the sources of the Groot and Klein Nyl Rivers as well as the tributaries indicate relatively high natural lead concentrations.

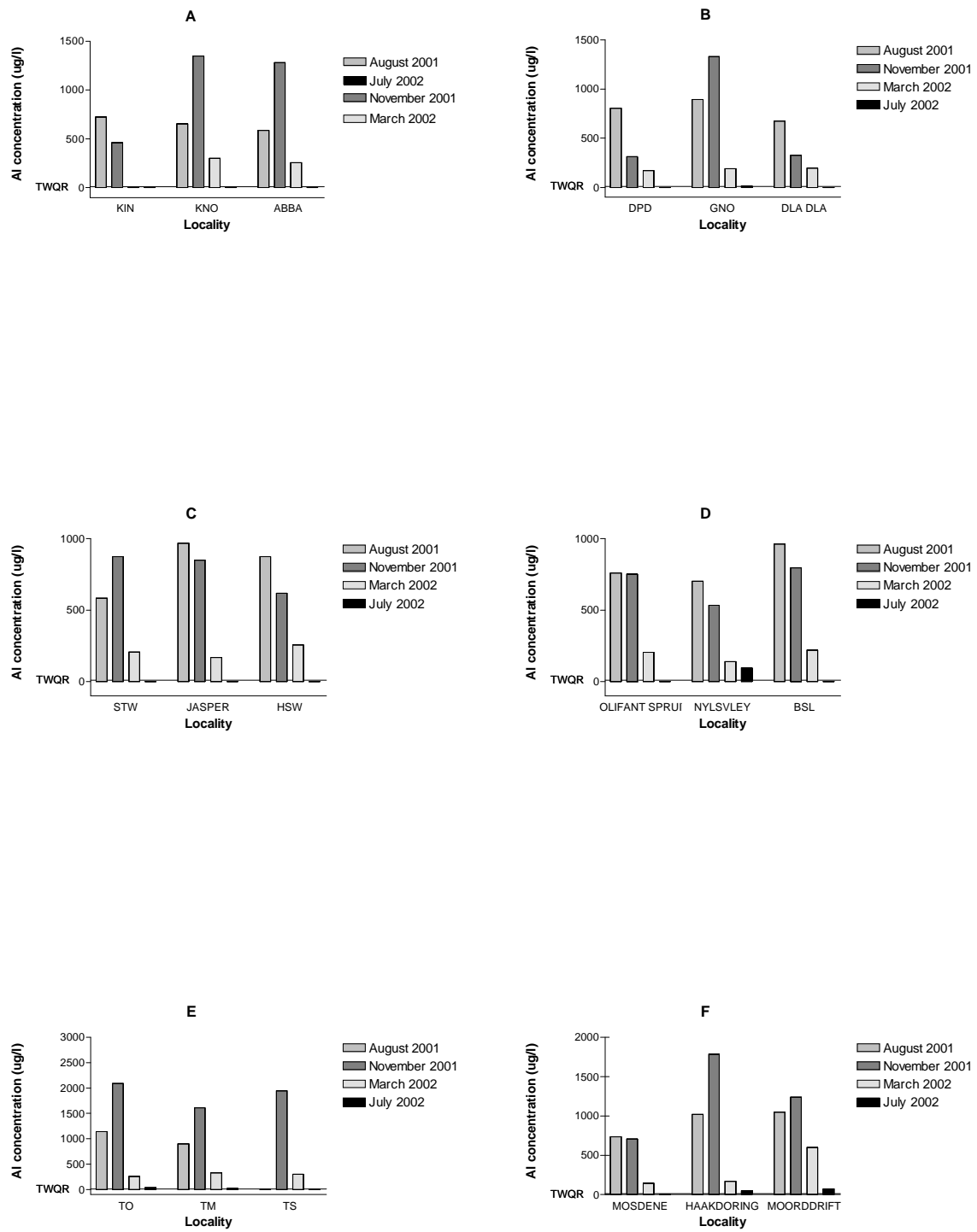
Selenium:

Selenium is a metalloid element similar to sulphur. It occurs in many forms but the tetravalent form is the most common form found. Small quantities are essential to animals and bacteria to help in the functioning of certain enzymatic systems (DWAF⁷, 1996). Selenium has not been recognised as an essential element for plants (Cao *et al.*, 2001). Selenium occurs naturally in various rock types. It may enter the water body by mechanical & chemical weathering of the rocks and by the deposition of organic compounds from decaying plant matter (DWAF⁷, 1996). In natural waters selenium usually occurs in nanogram quantities. The organic form of selenium is more toxic than the inorganic form (Newman and McIntosh, 1991). Selenium is an important element to ecotoxicologists due to its protective effects it has on the toxicity of certain heavy metals such as mercury and cadmium (Bhattacharya *et al.*, 2003).

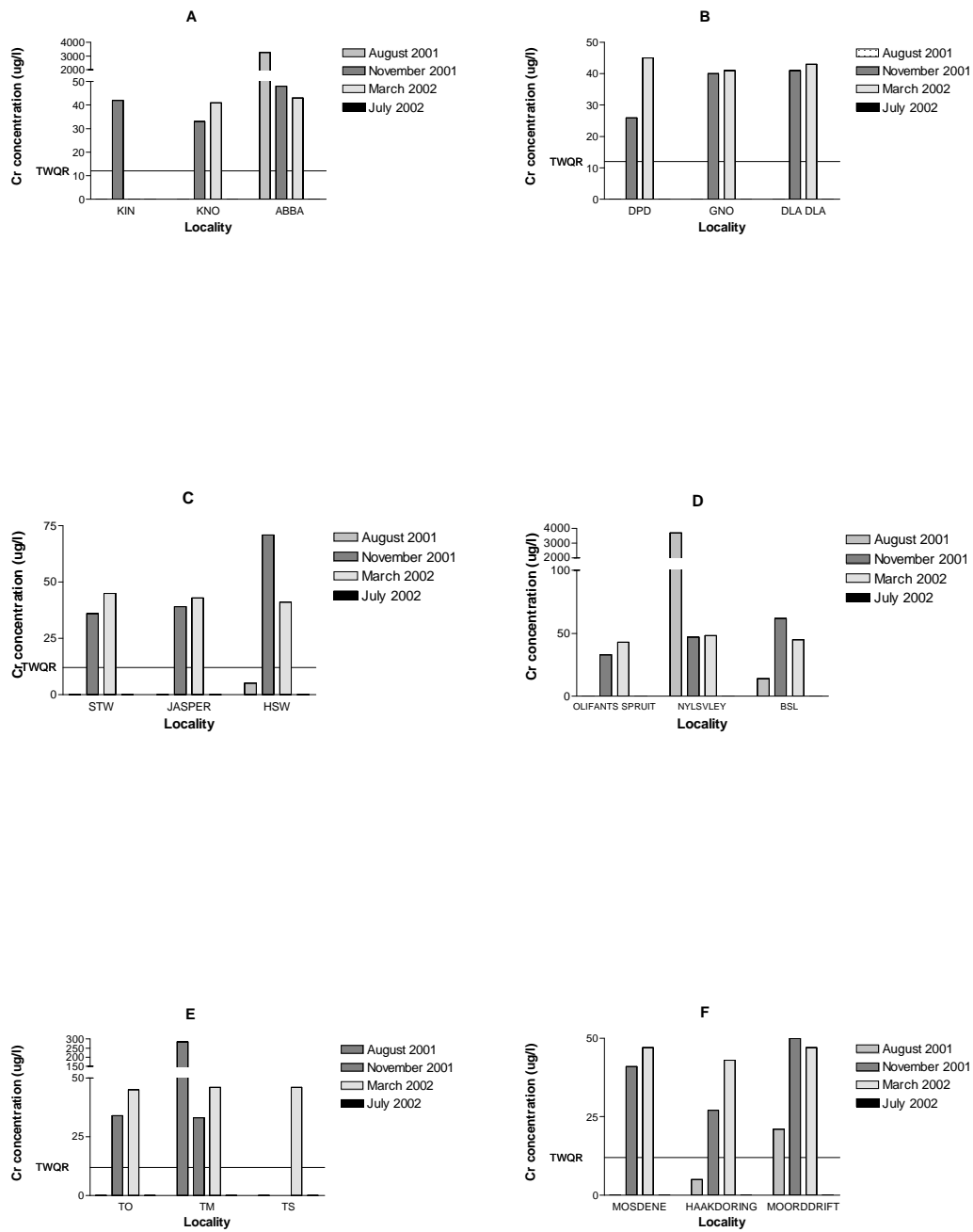
Selenium levels can also enter the environment via anthropogenic activities. These activities include industrial activities and agricultural activities (Liu *et al.*, 1987). Industrial activities that lead to increased selenium levels in the environment include (1) the paint industry, (2) the food processing industry, (3) the steel industry, (4) the pesticide manufacturing industry and (5) the combustion of fossil fuels during the smelting process and the operation of coal fired power plants (DWAF⁷, 1996, Bhattacharya *et al.*, 2003).

Selenium is a very toxic element to cells when it occurs over critical levels (Cao *et al.*, 2001). Its toxicity to fish is directly related to water temperature. pH decreases in the water column decrease the toxicity of selenium. This is because it becomes less soluble as the pH decreases and hence pH has little effect on selenium toxicity (DWAF⁷, 1996). Toxic effects of selenium include changes in feeding behaviour, skeletal anomalies in off spring, decreases growth rates and eventually death (DWAF⁷, 1996, Newman and McIntosh, 1991). Selenium however at the right levels can increase an animal's resistance to some diseases and cancers (Cao *et al.*, 2001).

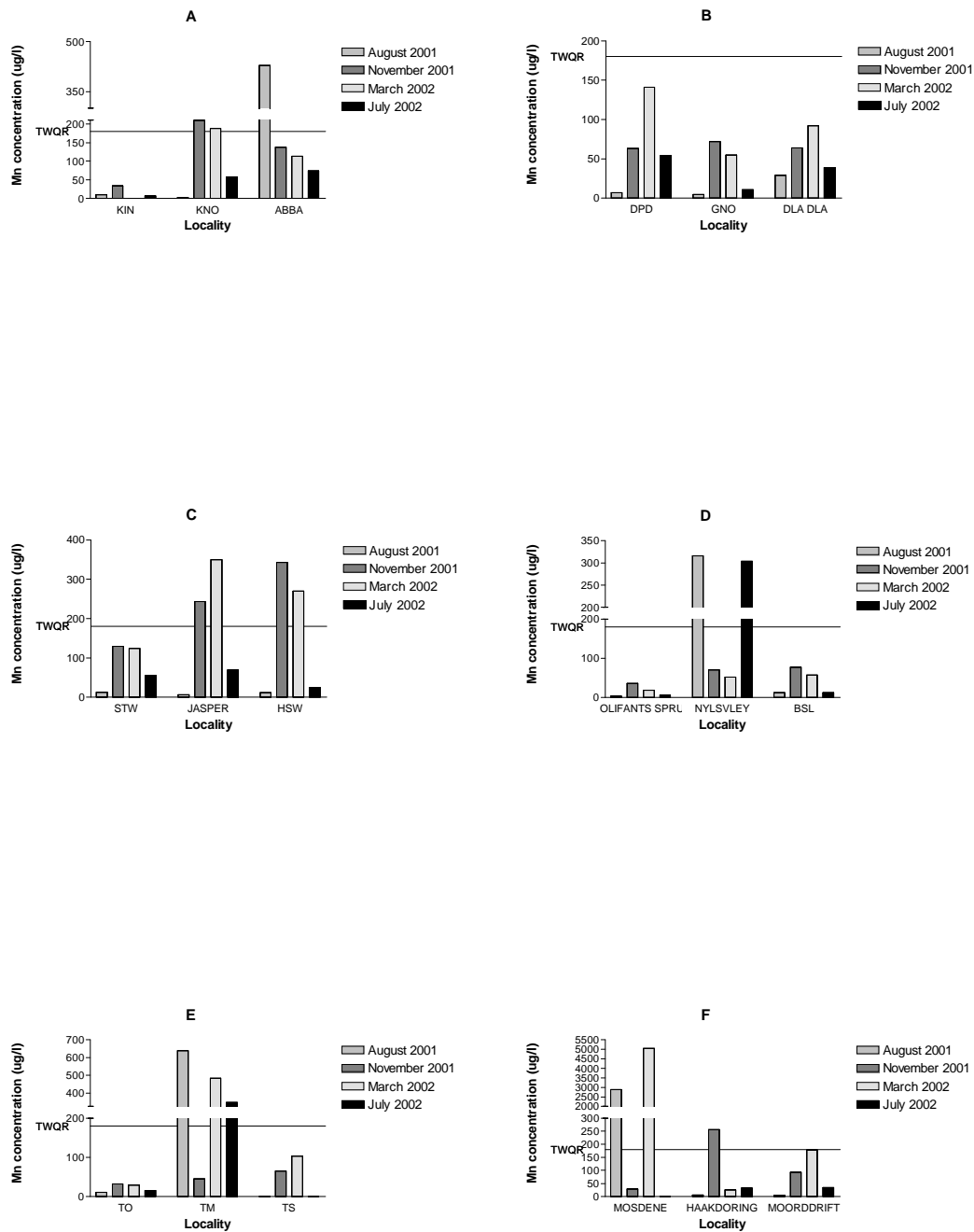
Selenium concentrations ranged between 11 µg/l, at Bad-se-Loop in March 2003, and 0 µg/l, at various sites throughout the system. The summary statistics (Table 2.21) indicate that November 2002 had the lowest average selenium levels (2 µg/l) and that March 2003 (6.17 µg/l) had the highest. During the August 2002 and March 2003 sampling months most of the sites had selenium concentrations above the TWQR value of 2 µg/l. Mosdene, DPD and Abba showed selenium concentrations below the TWQR during August 2002 and no site indicated (Figure 2.33) values below the TWQR for March 2003. Average concentrations equal to or slightly higher were recorded during November and July (2 µg/l and 2.3 µg/l respectively). This would indicate that selenium levels during these months were acceptable. The higher levels experienced during August and March could be attributed to a few factors. These factors could be (1) increased use of pesticides in agricultural activities, (2) increased combustion of fossil fuels in informal settlements during winter months and (3) increased weathering of the surrounding geology.



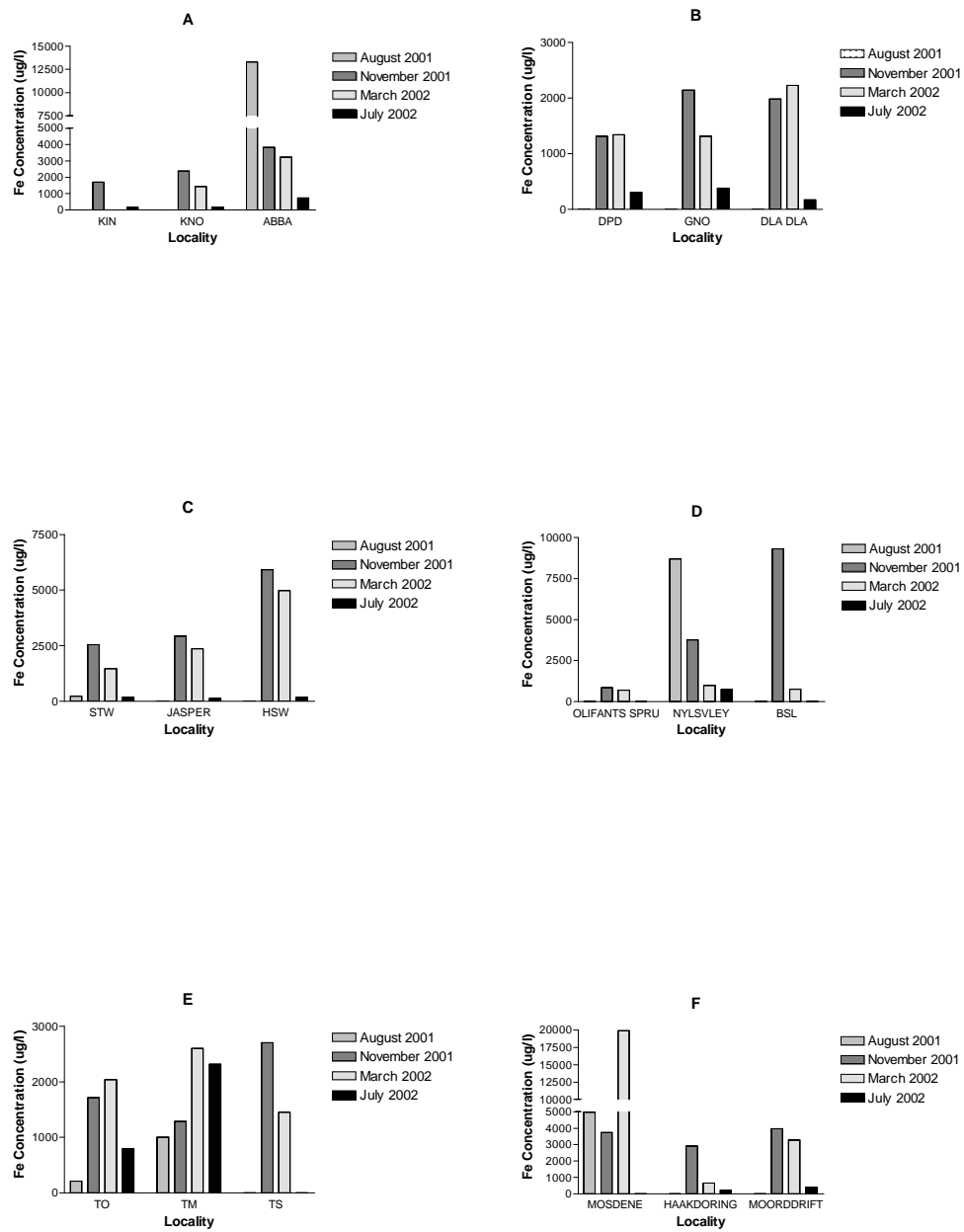
Figures 2.24 (A-F): Spatial and temporal representations of aluminium concentrations recorded in the water.



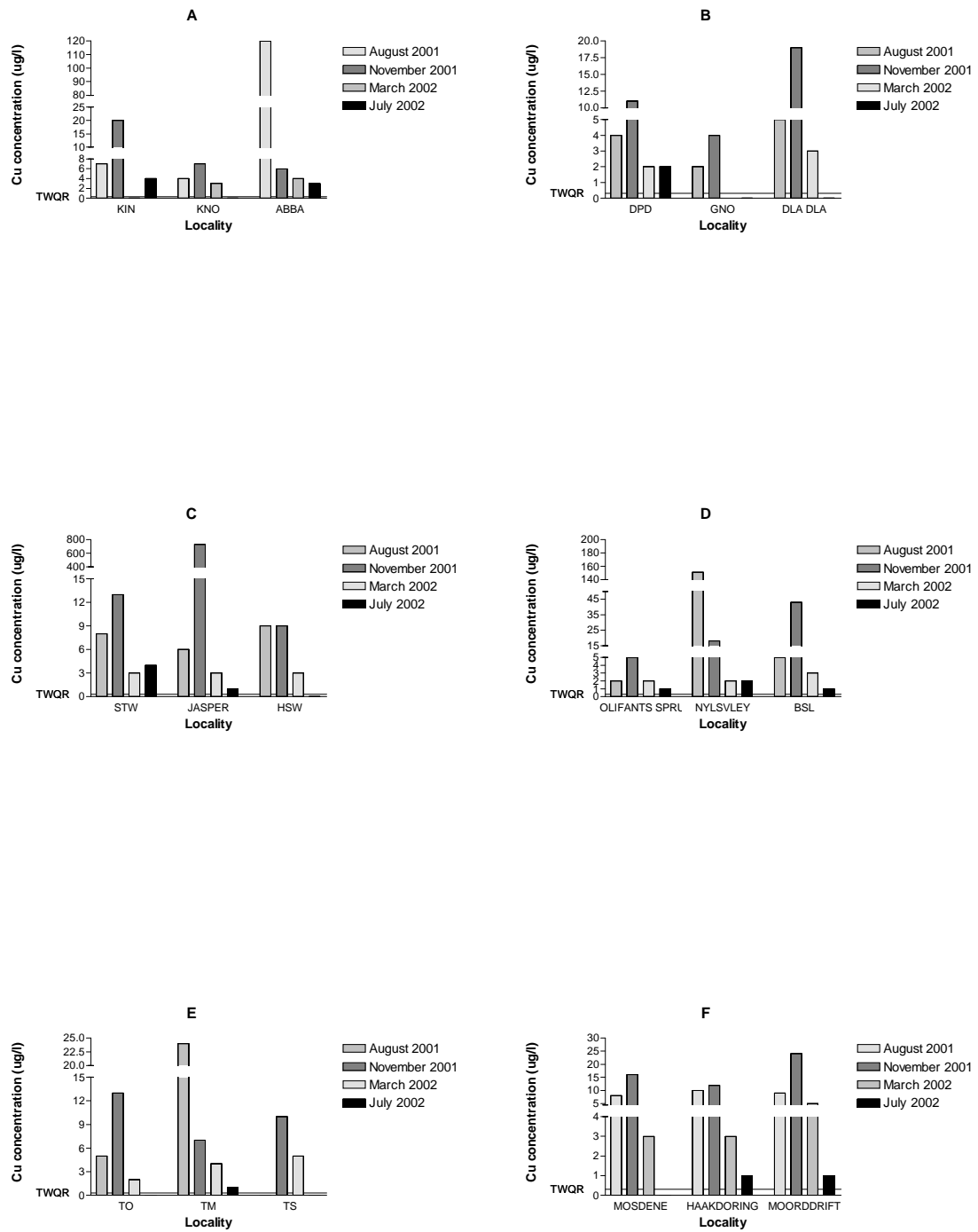
Figures 2.25 (A-F): Spatial and temporal representations of total chromium concentrations recorded in the water.



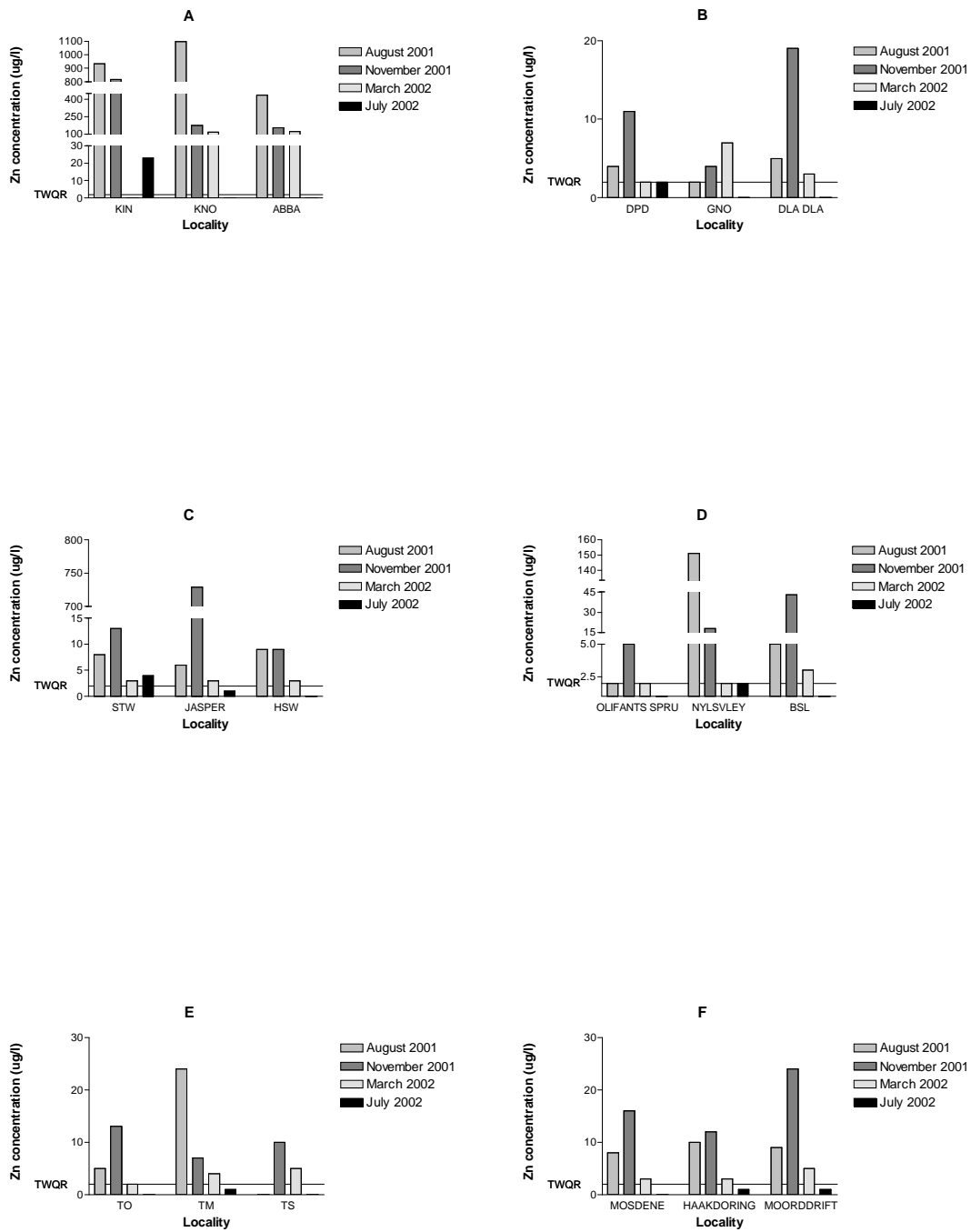
Figures 2.26 (A-F): Spatial and temporal representations of manganese concentrations recorded in the water.



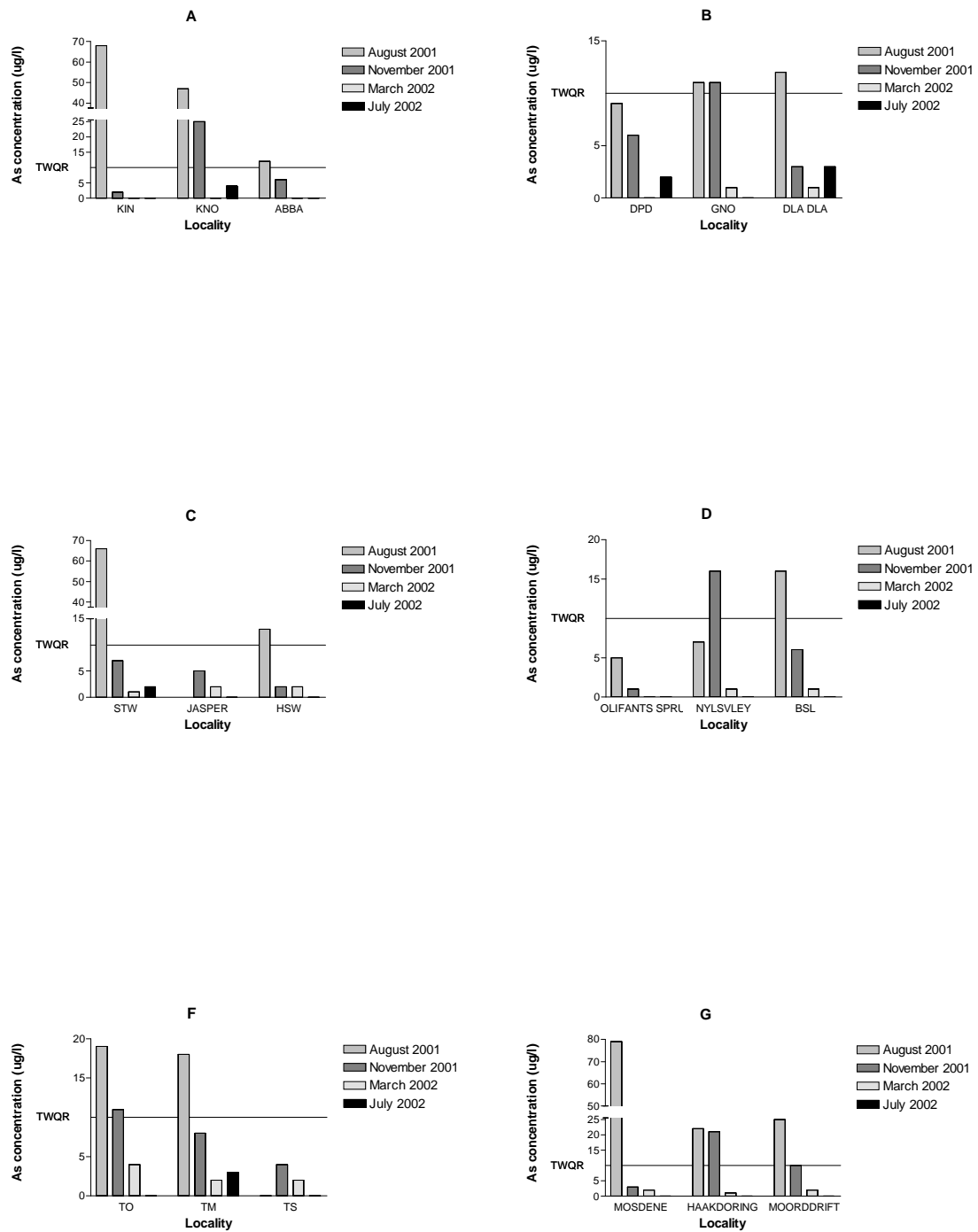
Figures 2.27 (A-F): Spatial and temporal representations of iron concentrations recorded in the water.



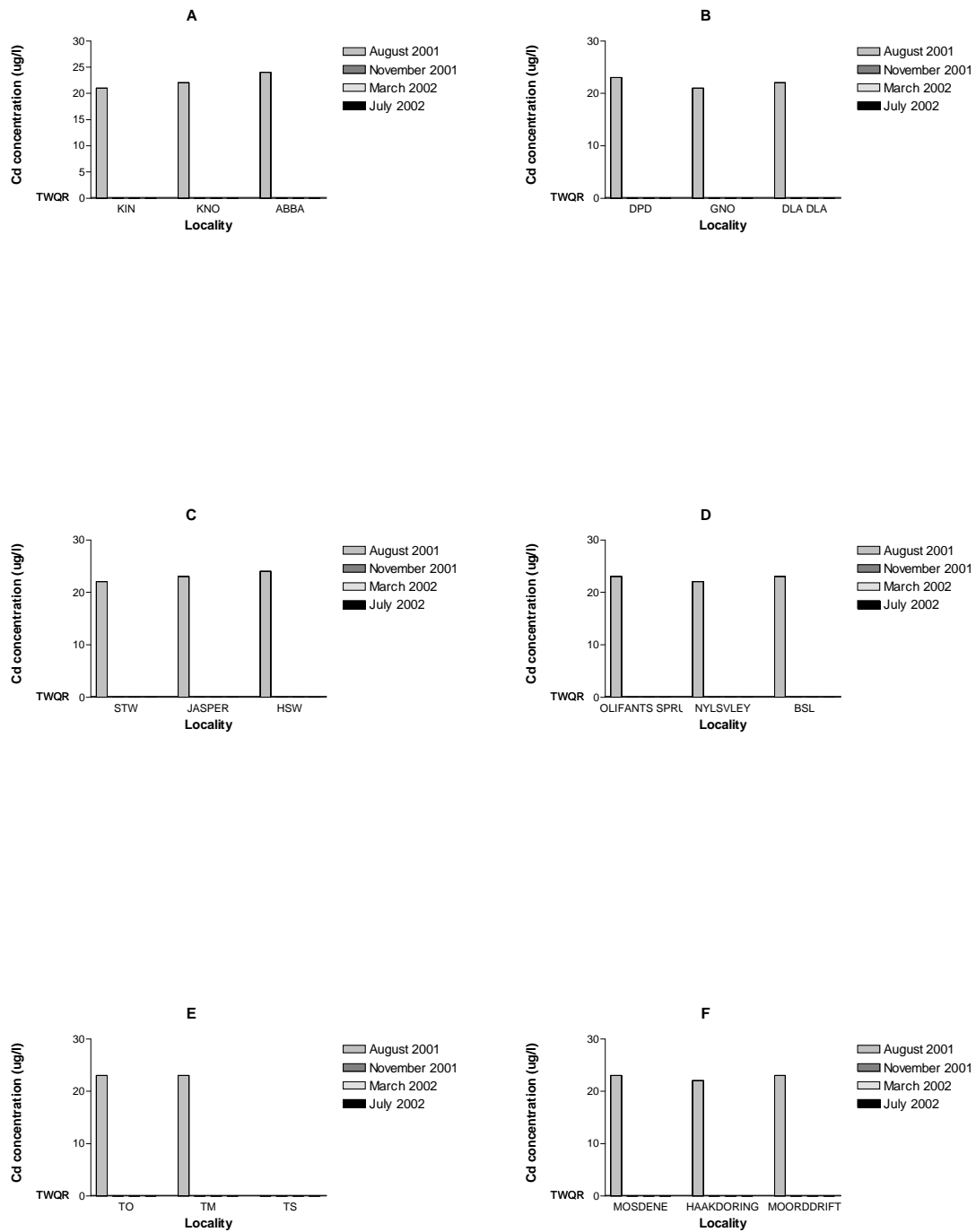
Figures 2.28 (A-F): Spatial and temporal representations of copper concentrations recorded in the water.



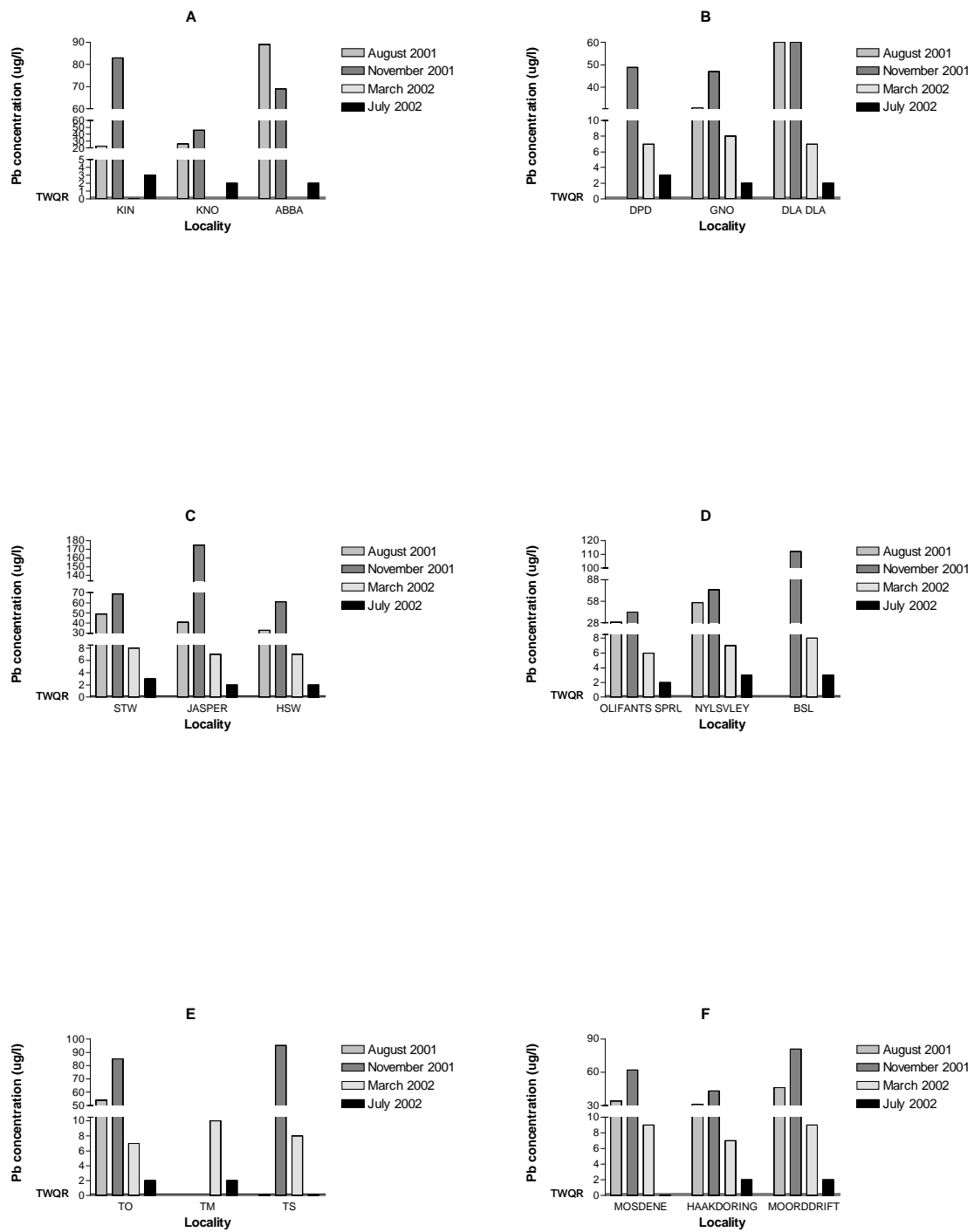
Figures 2.29 (A-F): Spatial and temporal representations of zinc concentrations recorded in the water.



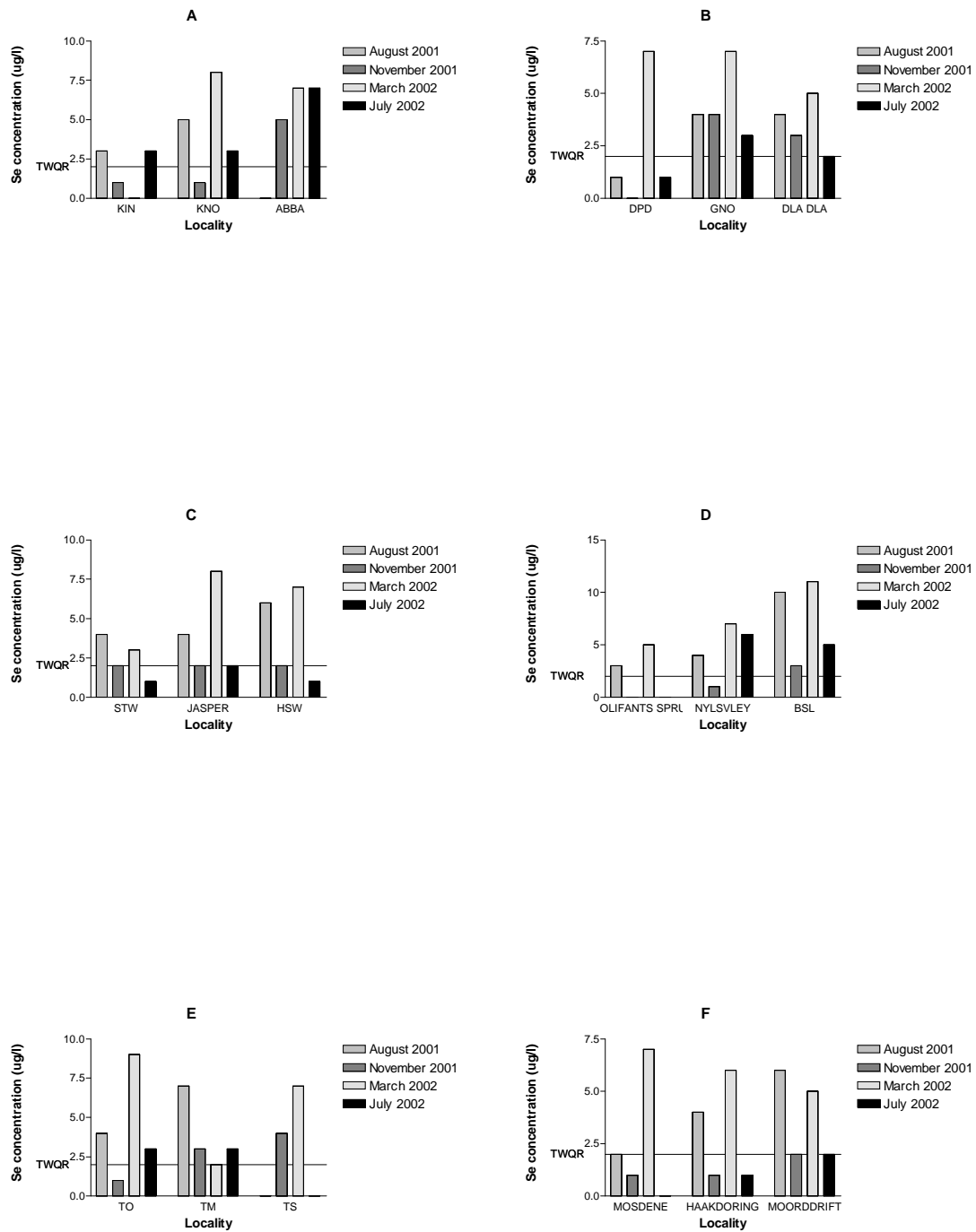
Figures 2.30 (A-F): Spatial and temporal representations of arsenic concentrations recorded in the water.



Figures 2.31 (A-F): Spatial and temporal representations of cadmium concentrations recorded in the water.



Figures 2.32 (A-F): Spatial and temporal representations of lead concentrations recorded in the water.



Figures 2.33 (A-F): Spatial and temporal representations of selenium concentrations recorded in the water.

2.3.4 Bacteriology:

Bacteria:

One of the functions of a wetland is water filtration. Suspended organic matter and bacteria settle out in a wetland due to the action of gravity on the particles in the slow moving water. Bacteria and pathogens enter the system via leaking sewer lines, sewage effluents and direct input from developing urban and rural areas (Vega *et al.*, 2003), as well as from farm animals.

Bacterial contamination is of great concern as the bacteria and pathogens can contaminate ground water supplies. This is of importance as ground water supplies are the main source of potable water supplies for the future (Vrhovsek *et al.*, 1996). Rodgers *et al.* (2003) stated that an important but often overlooked aspect of bacterial contamination of water courses is faecal contamination from organic matter via agricultural run off. Faecal coliforms are abundant in the faeces of warm-blooded animals and their presence indicates the potential risk to humans of faecal contamination, bacteria and viral agents. Bacterial water quality in rivers and wetlands is thus essential in planning policy decisions and providing an indication of the health risk posed by faecal contamination (Wilkinson *et al.*, 1995).

A study by Mvungi *et al.* (2003) on the impact of home industries on water quality concluded that improper waste management practices, sewage overflows and pollutant runoff increased the contaminant and bacterial content of tributaries draining urban areas in Harare. This fact is evident in the results obtained in this study.

The results indicate that the Modimolle Sewage Treatment Works (Sewerage Works) is having an effect on the bacterial content of the system. From figures 2.34 (A-C) it is evident that bacterial content of the water increases once the water flows past the Sewerage Works. Another source of bacteria in the system can be attributed to agricultural run off from the many cattle farms in the area, as well as run off from the informal settlement of Phagameng.

Total coliforms:

Total coliform counts indicate a dramatic increase in number of colonies per 100mℓ (N/100mℓ) between Donkerpoort and the sewage treatment works (Figure 2.34 A). This trend is most visible during the April 2001 sampling period, with counts increasing from 35 N/100mℓ to 77000 N/mℓ. The levels then slowly decrease as the water moves through the wetland to the site at Mosdene where the bacterial levels rise sharply once gain. This would indicate that there is a point source of bacterial contamination entering the system upstream from the site. This trend holds true for four of the five sampling periods with March 2002 indicating a different trend. The total coliform decrease experienced between Sewerage Works and Nylsvley during the sampling months of April 2001, August 2001, November 2001 and July 2002 can be attributed to one or more of the following factors: (1) dilution factor due to an influx of water from the tributaries Hessie-se-Water and the Olifantspruit, (2) bacterial breakdown/die off due to changes in water chemistry, (3) bacterial die off due to water temperature changes, and (4) the settling action exerted on bacterial particles bound to particulate organic matter and sediments as the water moves slowly through the wetland system (An *et al.*, 2002, Zaccone *et al.*, 2002, Maul and Cooper, 2000). The increases in total coliform concentrations can be attributed to (1) point sources of contamination e.g. Sewerage Works and increased run off from cattle farms in the area, (2) increased agricultural run off from precipitation and (3) resuspension of sediments bound to microbes during periods of increased flow (Interlandi and Crockett, 2003, Jagalsa, 1997, An *et al.*, 2002). Fisher *et al.*, (2000) concluded in their study of the Oconee River in Georgia that total coliform number increased with rainfall events but decreased more rapidly in grazed watersheds.

Table 2.22: Table of the effects of total coliforms on human health (DWAF¹, 1996).

Total coliforms (counts/100ml)	Effects
TWQR (0-5)	Negligible risk of microbial infection
5-100	Indicative of inadequate treatment, post contamination or after growth in the water distribution system. Risk of infectious disease transmission with continuous exposure and a slight risk with occasional exposure.
>100	Indicative of poor treatment, post treatment contamination or definite after growth in the water distribution system. Significant and increasing risk of infectious disease transmission.

Table 2.22 indicates the guideline ranges set out by the Department of Water Affairs and Forestry for drinking water or domestic use. The results indicate that the total coliform count falls within the lower two classes and that the risk for microbial infection if water from both Mosdene and the Sewerage Works are ingested is significant and increasing. The other three sites indicate a high risk of infection if water is ingested.

Faecal coliforms:

Faecal coliform counts followed similar trends to those of the total coliforms. This is logical as they form part of the total coliform count. Sources of faecal contamination to the system could be attributed to (1) run off from developed and developing urban areas, (2) agricultural runoff from both cattle and chicken farms as well as from wild areas such as game reserves, and (3) sewage effluents. High numbers of coliform bacteria from wild areas could account for variation in coliform levels when water quality is otherwise good (Fisher *et al.*, 2000).

The decreases illustrated in Figure 2.34 B can be attributed to the binding of the microbes to particulate organic matter and sediment particles, and the subsequent settling of the bacteria into the sediment layers of the wetland (Fisher *et al.*, 2000, An *et al.*, 2002). The faecal coliforms however indicated different trends during August 2001, November 2001 and March 2002 with peaks in the trend line occurring at Jasper and not at the Sewerage Works as is the case with the April sampling period. Studies by Maul and Cooper (2000) and by Newman *et al.* (2000) have shown that standing water and wetlands decrease faecal coliform counts. Faecal coliform counts were all above the recommended TWQR for drinking water (DWAF¹, 1996) of 0 counts/ 100mℓ. A maximum faecal coliform count was recorded at the Sewerage Works in April 2001 of 37500 counts/100mℓ. Faecal coliform guideline values for aquatic ecosystems are not available.

Table 2.23 indicates the Department of Water Affairs and Forestry guidelines for domestic use of water. These ranges indicate the different effects that water contaminated with faecal coliforms could have on humans. The water at all of the sites falls outside of the TWQR of 0 counts per 100 mℓ range and pose varying risks to human health from slight risks to significant and increasing risks. Jasper is the site that indicated the highest faecal contamination.

Table 2.23: Table of effects of faecal coliforms to humans (DWAF¹, 1996).

Faecal coliforms (counts/100ml)	Effects
TWQR 0	Negligible risk of microbial infection
0-10	Slight risk of microbial infection with continuous exposure; negligible risk with occasional exposure
10-20	Risk of infectious disease transmission with continuous exposure; slight risk with occasional exposure.
>20	Significant and increasing risk of infectious disease transmission. As faecal coliform levels increase, the amount of water ingested required causing infection decreases.

Heterotrophic Plate Counts:

Heterotrophic bacteria are recognised as an important component of the planktonic community contributing significantly to the regulation of the flux of organic matter. They are also a critical link in the microbial loop, which starts with the production of dissolved organic matter and ends with the oxidation to CO₂ (Zaccone *et al.*, 2002). Heterotrophic plate counts are used to test the general microbial quality of the water. They do not represent the total bacterial population present. They do however indicate the efficacy of the water treatment process and indicate if the treatment process is adequate or not (DWAF¹, 1996). Heterotrophic plate counts of between 100-1000 counts per ml indicate that the water has undergone inadequate treatment, post treatment contamination or after growth in the water. There is a slight risk of microbial infection (DWAF¹, 1996). The primary role of heterotrophic bacteria is classified as the decomposition and mineralization of dissolved and particulate organic nitrogen. Significant heterotrophic utilization of dissolved inorganic nitrogen would have profound effects on the fluxes of nitrogen and carbon in the water column (Allen *et al.*, 2002). Heterotrophic bacteria include species from many bacterial genera such as the nitrifying bacteria *Nitrosomonas*, *Nitrobacter*, and the denitrifying bacteria (Sakai *et al.*, 1997).

Heterotrophic plate counts indicated a different trend (Figure 2.34 C) to that of the coliform bacteria. All sampling periods indicate the steady increase in bacteria numbers from DPD to Jasper and then the trend lines form a plateau. Heterotrophic plate counts also exceeded the TWQR of 0-100 counts/ml except at the DPD site. All the plate counts fell between the ranges of 100-1000 counts/ml. As mentioned earlier in this section, this would indicate that the water entering the system, as effluent is not being treated properly. This would also indicate that the water in the system would pose a slight risk to human health. The plateau that is visible in the trend line could indicate a normal trend in wetlands as heterotrophic bacteria play a vital role in the filtration/purification of water. The bacteria help in the reduction of nutrient loads in the water column by oxidising toxic ammonia to nitrites, nitrates and finally nitrogen which is released into the atmosphere (Kupchella and Hyland, 1993).

Table 2.24: Table of effects of heterotrophic bacteria to human health (DWAF¹, 1996).

Heterotrophic plate count (Counts per ml)	Effects
TWQR (0-100)	Negligible risk of microbial infection
100-1000	Indicative of inadequate treatment, post contamination or after growth in the water distribution system. Slight risk of microbial infection
>1000	Indicative of poor treatment, post treatment contamination or definite after growth in the water distribution system. Increased risk of infectious disease transmission.

Table 2.24 indicates the effects of differing heterotrophic bacteria ranges to human health. The water in the system falls within the 100-1000 counts per ml range and thus poses a slight risk of microbial infection to humans.

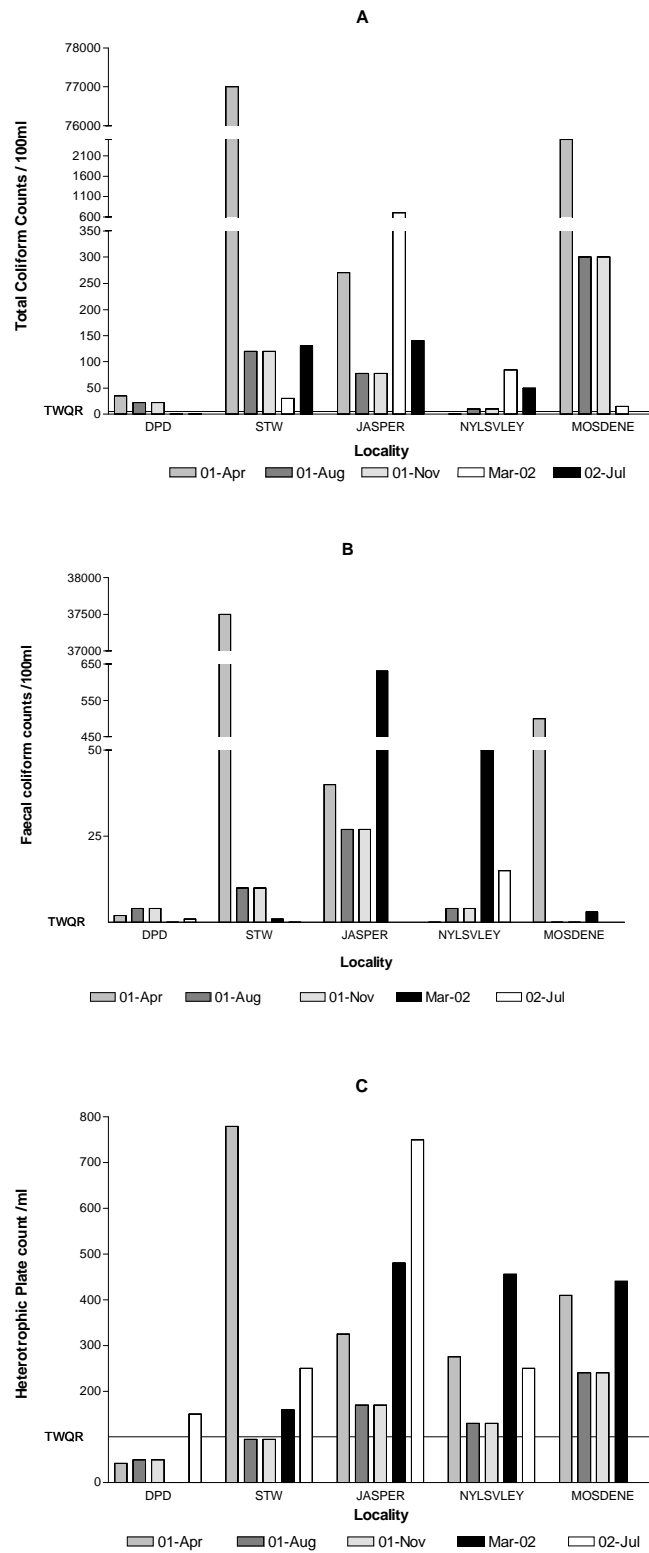
Due to the nature of the system and the functions of wetlands it could be said that high heterotrophic bacterial content in the water is not a major problem to the system. With the purification process/function of wetlands one would expect to have a relatively high heterotrophic plate count due to the presence of the bacteria needed to complete the different nutrient cycles, and organic matter break down. A high count however could indicate the contamination of the system with nutrients from agricultural activities. The concerning bacterial counts in the system are the high faecal and total coliform counts. These indicate that the system is definitely being contaminated by sewage effluent and agricultural run off from the different farms either game farms or cattle farms.

2.3.5 Toxicity testing:

Pollution of the environment means the contamination of soil, air and water. As water is essential for life, its pollution will not only endanger aquatic life, but also terrestrial organisms, including man (Kfir, 1981). Living material responds to the total effect of actual and potential disruptions in the water and therefore, the use of biological toxicity testing has become an important approach to complement chemical analysis to monitor and control harmful chemicals in water (Blaise *et al.*, 1988). Bioassays are used to determine the potential of substances to cause environmental harm, and are sometimes able to elicit responses at concentrations below chemical detection limits (Muller and Palmer, 2002). Bioassays have both advantages and disadvantages and these are listed in table 2.25.

An objective of the standard toxicity test is to assess whether the measured treatment of one end-point is significantly different from the measured end-point in another treatment, usually a control (Denton and Norberg-King, 1996). The aims of toxicity testing are therefore the ability of the test to detect either acute or chronic toxicity of an effluent (Chapman *et al.*, 1996).

The results discussed in this section are reported as LC50 values of the whole effluent tested. The results were obtained by calculating the LC50 with the Spearman Karber computer program. During the tests the mortalities and various parameters were recorded. During the tests the pH, conductivity, TDS, temperature and oxygen levels were monitored.



Figures 2.34 (A-C): Spatial and temporal representation of bacterial counts in the Nyl River System.

Table 2.25: Table of advantages and disadvantages to toxicity testing (Muller and Palmer, 2002).

Advantages	Disadvantages
A holistic approach, integrating effects of all stressors (especially useful for investigating whole effluent).	Tests do not indicate which stressors are causing the observed effects.
Tests can be simple and cost effective	Tests can be too simple and result in environmentally unsound answers
The effect on the selected organism(s) is (are) observed at the exposure(s) tested	Not all organisms, exposures and end-points can be tested
Tests are normally carried out under controlled laboratory conditions	Field conditions are different from laboratory conditions and extrapolating results from laboratory to field is associated with its own set of complications

For the tests to be validated the test conditions were monitored according to the International Standards Organisation (ISO), Institute for Water Quality Studies (IWQS) and the Organisation for Economic Change and Development (OECD) guidelines for static testing. These guidelines require the test conditions to conform to the following regulations.

Constant conditions must be maintained as far as possible throughout the test. Dissolved oxygen concentrations should not dip below the 60% saturation level throughout the duration of the test. The mortality of control organism should not exceed 10%. The proportion of control organisms showing abnormal behaviour should not exceed 10% of the total number of organisms in the control medium. Organisms should be healthy with mortalities in holding tanks kept down to a minimum (ISO, 1996, OECD, 1992, IWQS, 1998, Truter E. 1994)

From the results obtained the test conditions conformed to the required test condition specified by the OECD and ISO. Zero mortality was experienced under control conditions. The Temperature of the test conditions did not differ by more the 0.5 degrees during the tests and the oxygen concentrations remained more or less above the 80% saturation level. Table 2.26 indicates the LC50 values that were obtained during both the *Daphnia pulex* and *Poecilia reticulata* tests. The results will be discussed according to the different sampling months.

The *Daphnia* test for August 2001 produced no LC50 values during the tests. This is simply due to the lack of mortality experienced with not enough *Daphnia* dying to allow Spearman Karber to calculate the LC50 value. The guppy test produced LC50 values ranging between 73.49 percent, at the Mine site, and 100 percent at Mosdene and Bad-se-Loop. These results however are not statistically significant as they are not 95 percent reliable. Moorddrift, Sewerage Works, Koh-I-Noor and Nylsvley were the other sites that produced LC50 values but once again the values were not reliable.

From the samples from November 2001, March 2002 and July 2002 no LC50 values could be calculated due to lack of mortalities in both the *Daphnia* and guppy tests. The only site during November 2001 where an LC50 value was calculable was at Mine. The LC50 value was 100 percent, but it was not statistically reliable. The only site where an LC50 value was calculable was Moorddrift during July 2002. The LC50 value was calculated at 84.9 percent.

The results from the whole battery of toxicity tests would indicate that the water is relatively non toxic to aquatic organisms and that during the sampling periods the water posed no potential threat to maintaining aquatic life.

Table 2.26: Table of LC 50 values obtained during the study.

	<i>Daphnia (Daphnia pulex)</i>		<i>Guppy (Poecillia reticulata)</i>		
Locality	LC50	Trim	Locality	LC 50	Trim
Aug-01					
			Moorddrift	77.11	Not 95% reliable
			Sewerage Works	74.3	Not 95% reliable
			Mosdene	100	Not 95% reliable
All sites		Not available trim too large	Mine	73.49	Not 95% reliable
			Bad-se-Loop	100	Not 95% reliable
			Koh-I-Noor	82.03	Not 95% reliable
			Nylsvley	82.03	Not 95% reliable
			Rest of the sites		Not available trim too large
Nov-01					
Mine	100	Not 95% reliable	All sites		Not available trim too large
Rest of sites.		Not available trim too large			
Mar-02					
All sites		Not available trim too large	All sites		Not available trim to large
Jul-02					
All sites		Not available trim too large	Moorddrift	84.9	
			Rest of sites		Not available trim too large

2.3.6 Integrated water quality:

For the integrated water quality localities were compared to other localities with respect to variables analysed during the study except for the bacteria and toxicity test results. The data was analysed using PRIMER 5 statistics program. The data underwent a principle component analysis (PCA) with the results being plotted on graphs. The PCA's were conducted for each of the four sampling periods. Data was normalised to give a better comparison between the different localities.

August 2001

Eigen values calculated indicate that 46.8 percent of the variation occurred with the first two of the components calculated. Principle component one indicates that chromium, manganese, iron, copper, chlorides and oxygen saturation and concentration are causing the variation in water qualities. Principle component two indicates that aluminium, selenium, alkalinity, conductivity and TDS are causing the variability in the water.

This would indicate that the above mentioned variables play a vital role in the systems health, and that they are important stressors in the system to be monitored. Figure 2.35

illustrates the two components plotted on a graph. The X axis depicts the principle component one and the Y axis principle component two. The sites grouped close together indicate water with similar qualities. In figure 2.35 the sites grouped together are all situated before the Sewerage Works at Modimolle and would thus indicate that the Sewerage Works is having an effect on the water downstream.

November 2001

The calculated Eigen values indicate that 41.6 percent of the variation in the system occurs in the first two principle components. Principle component one indicates that the variation in the water is caused by the water parameters and the alkalinity. In component two the metals chromium, iron, zinc, arsenic, and lead are primarily responsible for the variation. Alkalinity also is one of the causes of variation. Figure 2.36 indicates the graphical representation of the similarities between the water on a spatial scale. No definitive grouping can be determined from the graph of the principle components. Groot Nyl Oog and Donkerpoort are the only sites that show any similarity in the water body.

March 2002

The Eigen values calculated indicate that 43.9 percent of the variation in the water occurs in the first two principle components. In principle component one the aluminium, chromium, lead, chloride, sulphate, pH, conductivity, and TDS are responsible for the spatial variation in the water. In principle component two manganese, iron, zinc, chloride and oxygen saturation and concentration can account for the variation in the water. Figure 2.37 indicates the spatial graphical representation of the first two principle components. From the figure a group emerges of localities with similar water qualities. This can be due to a possible increase in water volume or stability in the water due to less water entering the system via precipitation.

July 2002

The calculated Eigen values indicate that 40.7 percent of the variation in the water occurs within the first two components of the principle component analysis. The first component indicates that the water parameters and alkalinity account for the variation in the water. In principle component two the metals aluminium, manganese, selenium and lead as well as the chlorides and sulphates account for variation in the water. Figure 2.38 indicates the grouping of the sites with similar water qualities.

A comparison of the graphs (Figures 2.35-2.38) indicates that site groupings occur during three of the four sampling months (August 2001, March 2002 and July 2002) with many of the sites in these groups corresponding. The fourth sampling period (November 2001) is the period that differs. This can be attributed to a change in water stability due to an influx of water from runoff and the associated influx of contaminants. The precipitation causing the influx could also be changing the water chemistry with the associated dilution effect of the addition of large amounts of fresh water to the system. The sites excluded from the grouping are Bad-se-Loop, Haakdoorn, Jasper, Hessie-se-Water, Tobias and Moorddrift.

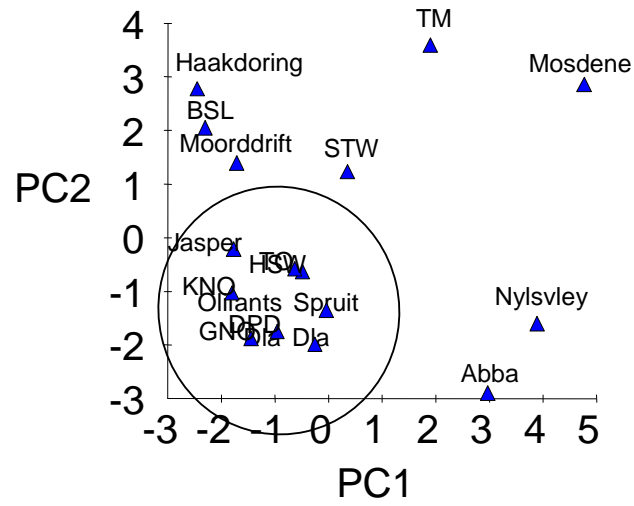


Figure 2.35: Principle component plot of spatial similarities of water in the system for August 2001.

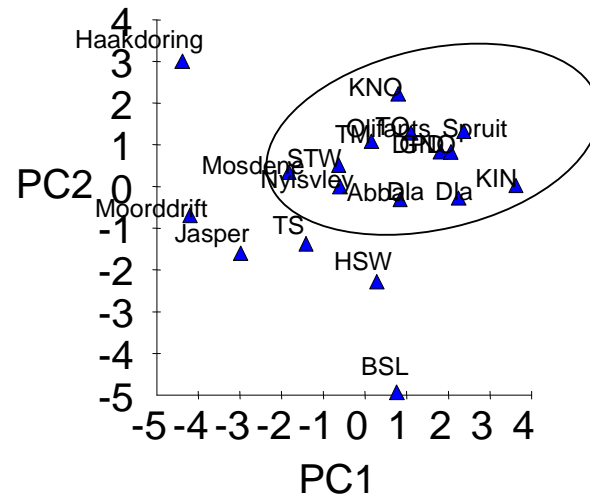


Figure 2.36: Principle component plot of spatial similarities of water in the system for November 2001.

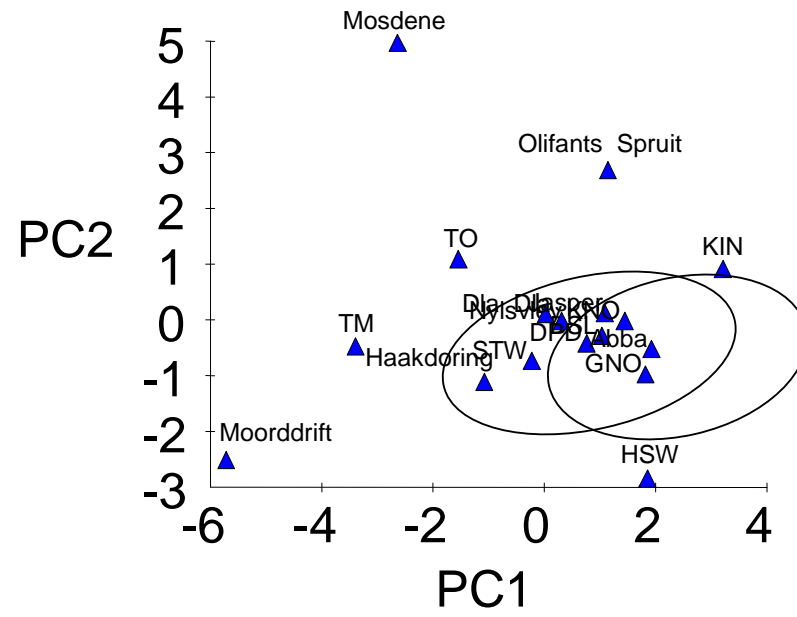


Figure 2.37: Principle component plot of spatial similarities of water in the system for March 2002.

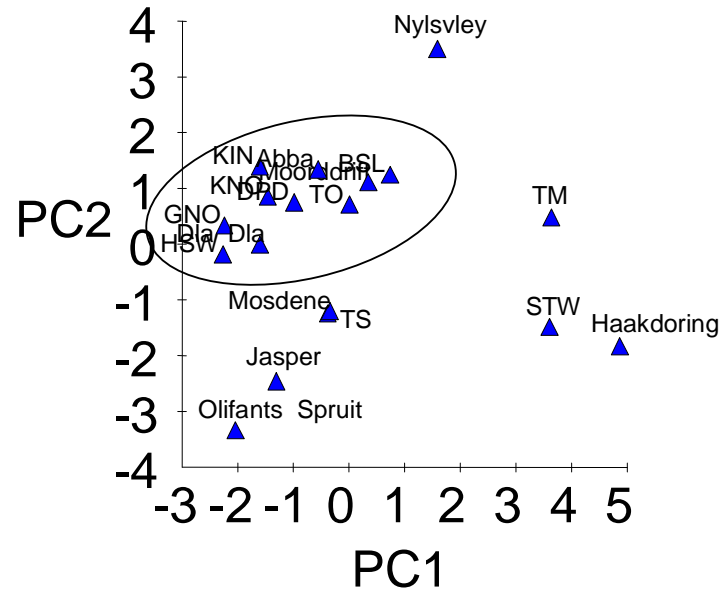


Figure 2.38: Principle component plot of spatial similarities of water in the system for July 2002.

2.3.7 Present ecological state of the system:

Data from the Gauging stations used to determine the reference condition was obtained from the Department of Water Affairs and Forestry and used to determine the present ecological state of the system. This data was analyzed using the method set out by Bath *et al.* (1999)² for the determination of the present ecological status of the system. This method is used in the resource directed measures for a reserve determination. A full reserve determination was however not conducted, but results obtained may be used in a reserve determination at a later stage. These results represent the changes that have occurred in the system over the last 20 years. Changes determined include the water parameters pH, TDS and conductivity as well as nutrients (N:P ratios and ammonia concentrations). The results were then compared to the reference conditions determined in the beginning of this chapter.

2.3.7.1 Nutrients

Ammonia

Ammonia is the toxic nitrogen based compound in an aquatic environment. It is present in air soil and water and in large amounts in decomposing organic matter. Ammonia is a common pollutant in water and contributed greatly to eutrophication in a system (DWAF⁷, 1996). External sources of un-ionised ammonia include (1) fish farm effluents, (2) sewage discharges, (3) atmospheric deposition of ammonia from the combustion of coal and (4) the biological degradation of manure. Increases in temperature and pH increase the proportion of toxic un-ionized ammonia in solution and hence increase its toxicity to aquatic organisms (DWAF⁷, 1996).

The ammonia levels at the gauging stations were determined using the water temperature, ammonium levels and table 2.27. The ammonium concentration was multiplied by the factor give in table 2.27, for the corresponding water temperature and pH, and divided by 100 to give the proportion of un-ionized ammonia in the water. This concentration was then compared to table 2.28 and assigned a letter depicting the class range that the water fell in. Table 2.28 gives the different class ratings, ammonia ranges and impact status of a water body.

Table 2.27: Relationship between water temperature, pH and un-ionized ammonia (Bath *et al.*, 1999)².

pH	Water Temperature (°C)				
	10	15	20	25	30
6.5	0.06	0.09	0.12	0.18	0.25
7	0.18	0.27	0.39	0.56	0.79
7.5	0.58	0.85	1.2	1.7	2.4
8	1.8	2.6	3.8	5.3	7.3
8.5	5.5	7.9	11	15	20

Table 2.28: Nutrient assessment using the un-ionized ammonia concentration to assign assessment classes for rivers (Bath *et al.*, 1999)².

General classes	Assessment classes	Ammonia (un-ionized) concentration (expressed as mg-N/l as NH ₃)
Un-impacted	A	<0.007
	B	<0.015
Moderately impacted	C	<0.030
	D	<0.070
Highly impacted system	E	<0.100
	F	>0.100

Nitrogen:Phosphorus (N:P) Ratio

The N:P ratio is the ratio of Total Inorganic Nitrogen (TIN) to Soluble Phosphates (SP). This ratio is determined by calculating the median TIN for the system and the median SP concentrations. These concentrations are then compared to each other to provide a ratio. This ratio is then used along with the SP concentration and Table 2.29 to assign nutrient status classes.

Table 2.29: Assessment of nutrient status based on N:P ratio using only orthophosphate data (Bath *et al.*, 1999)².

		Total inorganic Nitrogen to Soluble Phosphate Ratio			
		<10:1	>10:1 & <20:1	>20:1 & <30:1	>30:1
Orthophosphate concentration (expressed in mg-P/l)	<0.01	C	B	A	A
	<0.05	D	C	B	A
	<0.07	E/F	D	C	B
	<0.1	F	E/F	D	C
	>0.1	F	F	E/F	D/E

Table 2.30 Resultant classes assigned to sites analyzed.

	Historical state	Present state	Present state
	N:P	N:P	Un-ionised Ammonium.
KNO	C	F	A
DPD	C		
HSW	C	F	A
Olifantspruit	D	F	A
Nylsvley	D		
Bad-se-Loop	A	F	A
Tobiasspruit	D		

The class descriptions are listed in table 2.33.

The result indicated in table 2.30 show that although the system was moderately impacted twenty years ago the nutrient levels have increased to a highly impacted state.

The present day ammonia levels would however indicate that although the system has been impacted by water nitrification the toxic levels are as un-impacted and the water is thus not harmful to the aquatic organisms.

System variables

For the purpose of this study the following system variables were calculated TDS and pH. For TDS the sites were classified using the following method. The median reference condition (RC) was calculated using the data collected earlier on in this chapter (Historical Data). The median TDS value was also calculated for the present study from March 2001 to March 2003. These values were then plugged into the following equation:

$$\% \text{ difference [TDS]} = ((\text{TDS}_{\text{median}} - \text{TDS}_{\text{median RC}}) / \text{TDS}_{\text{median RC}}) * 100$$

Table 2.31 was then used with the percentage difference to assign a class rating to each site. These were then compared to the reference condition to depict the ecological status of the system.

Table 2.31: System variable assessment for total dissolved solids (TDS) (Bath *et al.*, 1999)².

Assessment class	TDS
	The median TDS concentration should not differ from the upstream Reference Condition (RC) by greater than:
A	15 percent
B	20 percent
C	30 percent
D	40 percent
E and F	> 40 percent

Reference conditions were calculated for pH previously in this chapter (Historical Data). These conditions were then compared to the present pH conditions to assign assessment class ratings. For pH the percentage variation was calculated using the following formula:

$$\% \text{ pH} = ((\text{pH}_{\text{median}} - \text{pH}_{\text{median RC}}) / \text{pH}_{\text{median RC}}) * 100$$

Assessment classes were then assigned using table 2. 32.

Table 2.34 indicates the results of the present ecological state of the system in comparison to reference conditions. The reference conditions were assigned an assessment class rating A as they are the considered to be pristine or un-impacted conditions. Table 2.34 indicates the state of the system according to the class ratings.

From the results indicated in table 2.34 it can be concluded that the pH of the system has not changed over the last twenty years. The only site indicating moderate impact is Bad-se-Loop. TDS assessment class ratings indicate that the system is being impacted and the salt levels in the system are increasing. These increases are taking place at Olifantspruit, Tobiasspruit and in the Bad-se-Loop. The TDS values at Nylsvley are un-impacted which would indicate the system health and effective functioning of the wetland.

Table 2.32: Present ecological state for the assessment of pH in Rivers (Bath *et al.*, 1999)².

Assessment class	pH The median pH value should not differ from the upstream reference condition (RC) by greater than:
A	< ± 5 percent
B	± 7 percent
C	± 10 percent
D	± 12 percent
E	> ± 12 percent
F	

Table 2.33: Class Assessment Descriptions.

Class assessment	Description
A	Un-impacted/ pristine
B	Few modifications
C	Moderately modified
D	Largely modified
E	Greatly modified
F	

Table 2.34: Present ecological state of the sites in the Nyl River system.

	Reference condition (R/C)	pH	TDS
Klein Nyl Oog	A	A	A
Donkerpoort	A	A	A
Hessie-se-Water	A	A	B
Olifantspruit	A	A	E
Nylsvley	A	A	A
Bad-se-Loop	A	C	F
Tobiasspruit	A	A	F

2.4 Conclusions and recommendations:

The results from the water analysis give a comprehensive summary of the water quality in the Nyl River System. Of the many variables and parameters analysed the bacteriological study indicated most cause for concern. Total coliform and faecal coliform counts increased along the course of the river and into the wetland. The faecal coliform counts increased dramatically from the point where the Modimolle Sewage Treatment Works releases its effluent into the Klein Nyl River. This increase in faecal coliform content can have deleterious effects on the system as well as the people who rely on the water in the system.

There are three possible causes for the increase in faecal coliform content along the course of the Nyl River namely:

1. The Modimolle Sewage Treatment works is unable to cope with the volume of effluent that passes through it. This is visible in figure 2.39, which clearly shows a pipe leading into the field along the banks of the Klein Nyl River. This could lead to raw sewage leaching into the river and the subsequent contamination thereof.



Figure 2.39: Effluent discharge pipe leading onto the banks of the Klein Nyl River (November 2002).

2. The second possible cause for the increase in faecal coliform content in the river is run off from the surrounding informal settlements such as Phagameng. Insufficient municipal services could have an effect in the quantity of bacteria entering the system during the rainy season.
3. The third possible cause for the increased faecal coliform content in the system can be attributed to increased run off from agricultural land. The surrounding areas contain a large number of live stock farmlands including cattle, pigs, chickens, game and crocodiles. Run off during the rainy season could cause an increase of faecal coliform bacteria in the system.

Although there is little one can do about the increase of livestock farming in the surrounding areas it is possible to address the other two points of contamination. The Modimolle Sewage Treatment works needs to be upgraded to cope with the volumes of sewage received and possibly to make provision for any further expansion of Modimolle. The incorporation of the informal settlements into the sewage network would also decrease the run off of bacteria entering the system. Although parts of the settlements may have been incorporated already personal observation indicated that the pipelines are in a poor state and blockages in the system cause spills of raw sewage. Maintenance of these systems is also vitally important.

The floodplain helps in the purification of the water in the river system and aids in decreasing coliform counts. Continual contamination of the system however eventually exceeds the systems ability to cope with the bacterial levels and process them, leading to health risk problems further down stream. Increased levels of coliform bacteria in the wetland could also lead to the contamination of ground water supplies.

Toxicity tests done on the water in the system indicate that the water is suitable for sustaining aquatic life. The acute static screening toxicity tests on *Poecillia reticulata* and *Daphnia pulex* indicated few mortalities and thus indicate that the water is of a fair quality.

The ICP-MS analysis of the whole water samples indicated that the water has got heavy metal concentrations above the TWQR set out by the Department of Water Affairs and Forestry. These metals however occur naturally in the system with the metal levels in the water remaining relatively constant from the source of the Klein and Groot Nyl Rivers to the furthest sampling site at Moorddrift. The water parameters such as the conductivity, oxygen content and pH also indicated little cause for concern. These factors together with the metal concentrations show that the water is of a fair quality. The determination of the present ecological state of the system indicates that although the nutrient levels in the system are on the increase, they pose little cause for concern, as the levels of toxic ammonia are still low. All these chemical constituents would indicate that the system is relatively un-impacted.

Regular monitoring of metal content and nutrient levels are not necessary and once a year should provide enough information to determine if the system is deteriorating. It is however recommended that a regular bacterial (Coliform) monitoring program be initiated. Bacterial contamination as mentioned earlier poses the biggest threat to the system.

2.5 References:

- ALLEN, A.E., HOWARD-JONES, M.H., BOOTH, M.G., FRISCHER, M.E., VERITY, P.G., BRONK, D.A. and SANDERSON, M.P. (2002). *Journal of Marine Systems* 38: 93-108.
- AN, Y., KAMPBELL, D.H. and BREIDENBACH, G.P. (2002). *Escherichia coli* and total coliforms in water and sediment at lake marinas. *Environmental Pollution* 120: 771-778.
- APHA 1998, Standard Methods for the Examination of Water and Wastewater, 20th Edition. United Book Press, Baltimore, Maryland. pp 4-67 – 4-179.
- AUCAMP, P.J. and VIVIER, F.S. (1990). Proposed Water Quality Criteria in South Africa. *Technology SA* (June): 21 – 30.
- AYERS, R.S. and WESTCOTT, D.W. (1985). Water Quality for Agriculture. FAO Irrigation and Drainage Paper No. 29. FAO Rome. pp 19.
- BATH, A., JOOSTE, S., HOHLS, B., MACKAY H., ASHTON, P. and PALMER, C. (1999)¹. Part one of appendix x: determination of reference conditions for water quality variables. Unpublished Report, Institute for Water Quality Studies, Pretoria.
- BATH, A., JOOSTE, S., HOHLS, B., MACKAY, H., ASHTON, P. and PALMER, C. (1999)². Part two of appendix x: determination of present ecological statuses: water quality . Unpublished Report, Institute for Water Quality Studies, Pretoria.
- BELL, A.V. (1976). Waste control at base metal mines. *Environmental Science and Technology* 10 (2): 130 – 135.
- BENEDETTI, I., ALBANO, A.G., and MOLA, L. (1989). Histomorphological changes in some organs of the brown bullhead, *Ictalurus nebulosus* LeSueur, following short- and long-term exposure to copper. *Journal of Fish Biology* 34: 273-280.
- BHATTACHARYA, B., SARKAR, S.K. and DAS, R. (2003). Seasonal variations and inherent variability of selenium in marine biota of a tropical wetland ecosystem: implications for bioindicator species. *Ecological Indicators* 2: 367-375.
- BLAISE, C., SERGY, G., WELLS, P., BERMINGHAM, N and COILLIE, R.V. (1988). Biological testing- development, application, and trends in Canadian environmental protection laboratories. *Toxicity assessment* 3: 385-406.
- BROWNING, E. (1961). Toxicity of industrial metals. Butterworth, London. pp. 325
- CANADIAN COUNCIL OF THE MINISTERS OF THE ENVIRONMENT (CCME), (1992). Canadian water quality guidelines. Ottawa, Ontario, Canada.
- CAO, Z.H., WANG, X.C., YAO, D.H., ZHANG, X.L. and WONG, M.H. (2001). Selenium geochemistry of paddy soils in the Yangtze River Delta. *Environment International* 26: 335-339.
- CARBONELL, A.A., AARABI, M.A., DELAUNE, R.D., GAMBRELL, R.P. and PATRICK jr, W.H. (1998). Arsenic in wetland vegetation: Availability, phytotoxicity, uptake and effects on plant growth and nutrition. *The Science of the Total Environment* 217: 189-199.
- CARPENE, E., CORTESI, P., TACCONI, S., CATTONI, O., ISANI, G. and SERRAZANETTI, G. P. (1987) Cd-metallothionein and metal enzyme interactions in the goldfish, *Carassius auratus*. *Comparative biochemistry and Physiology* 86c 2: 267-272.

- CHAPMAN, G.A., ANDERSON, B.S., BAILER, A.J., BAIRD, R.B., BERGER, R., BURTON, D.T., DENTON, D.L., GOODFELLOW, W.L. jr, HEBER, M.A., MCDONALD, L.L., NORBERG-KING, T.J. and RUFFIER, P.J. (1996). Methods and appropriate endpoints: discussion synopsis. In: whole effluent toxicity testing: an evaluation of methods and prediction of receiving impacts. GROTHE, D.R., DICKENSON, K.L. and REED-JUDKINS, D.K. (Eds), SETAC, Pensacola. pp 51-78.
- COETZEE, L. (1996). Bioaccumulation of metals in selected fish species and the effects of pH on aluminium toxicity in a cichlid *Oreochromis mossambicus*. MSc thesis, R.A.U.
- DALLAS, H.F. and DAY, J.A. (1993). The effects of water quality variables on riverine ecosystems: a review. Water Research Commission Report Number TT 61/93.
- DALLAS, H.F., DAY, J.A, MUSIBONO, D.E. and DAY, E.C. (1998). Water Quality for Aquatic Ecosystems: Tools for Evaluating Regional Guidelines. Water Research Commission Report Number 626/1/98. pp 240.
- DAVIES, B. and DAY, J. (1998). Vanishing Waters. University of Cape Town Press. Rondebosch, Cape Town. pp 487.
- DENTON, D.L. and NORBERG-KING, T.J. (1996). Whole effluent toxicity statistics: a regulatory perspective. In: whole effluent toxicity testing: an evaluation of methods and prediction of receiving impacts. GROTHE, D.R., DICKENSON, K.L. and REED-JUDKINS, D.K. (Eds), SETAC, Pensacola. pp 83-102.
- DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF¹). (1996). South African Water Quality Guideline volume 1: Domestic water use: pp 197.
- DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF⁶). (1996). South African Water Quality Guideline volume 6: Agricultural water use: aquaculture. pp 193.
- DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF⁷). (1996). South African Water Quality Guideline volume 7: Aquatic ecosystems. pp 159.
- DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF⁹). (1999). Water Quality on Disk V 1.0: Water Quality from the Hydrological Information System (HIS).
- EIFAC, (1978). Report on copper and freshwater fish. *Water Research* 12: 277-280.
- EWERS, U. and SCHLIPKOTER, H.W. (1991). Lead In: E. MERIAN [ed.]. Metals and their compounds in the environment, Occurrence, analysis and biological relevance. pp. 1438
- FISHER, D.S., STEINER, J.L., ENDALE, D.M., STUEDEMANN, J.A., SCHOMBERG, H.H., FRANZLUEBBERS, A.J. and WILKINSON, S.R. (2000). The relationship of land use practices to surface water quality in the upper Oconee Watershed of Georgia. *Forest Ecology and Management* 128: 39-48.
- GALVIN, R.M. (1996). Occurrence of metals in waters: An overview. *Water SAI* 22 (1): 7-18.
- GALVIN, R.M. and MELLADO, J.M. (1993). A note on the use of chlorine dioxide vs. chlorine for potable water treatment. *Water SA* 19 (3): 231-234.
- GARCIA-SANCHEZ, A. and ALVAREZ-AYUSO, E. (2003). Arsenic in soils and water and its relation to geology and mining activities (Salamanca Province, Spain). *Journal of Geological Exploration* 80: 69-79.

- GIESY, J.P. and WIENER, J.G. (1977). Frequency distribution of trace metal concentrations in five freshwater fishes. *Transactions of the American fisheries society* 106: 393-403.
- GREENFIELD, R.(2001). Bioaccumulation of selected metals in water, sediment and selected tissues of *Oreochromis mossambicus* and *Clarias gariepinus* in the Nyl River and Nylsvley. Unpublished M.Sc thesis, University of the North, Sovenga, South Africa. pp 200
- GROSELL, M.H., HOGSTRAND, C. and WOOD, C.M. (1997). Copper uptake and turnover in both copper acclimated and non-acclimated rainbow trout (*Oncorhynchus mykiss*). *Aquatic Toxicology* 38. pp. 257-276.
- HEALTH AND WELFARE CANADA, (1980). Guidelines for Canadian drinking water quality 1978. supporting documentation. Supply and services Canada, Hull.
- HEATH, A.G. (1987). Water pollution and fish physiology. C.R.C Press, inc., Boca Raton, Florida pp. 245.
- HELLAWELL, J.M. (1986). Biological indicators of freshwater pollution and environmental management. Elsevier Applied Science Publishers Ltd., London pp. 546.
- HOGSTRAND, C., WILSON, R.W., POLGAR, D. and WOOD, C.M. (1994). Effects of zinc on the kinetics of branchial calcium uptake in freshwater rainbow trout during adaptation to waterborne zinc. *Journal of Experimental Biology* 186: 55-73.
- HUTCHINSON, G.E. (1975). A treatise on limnology, Volume III: Limnological Botany. Wiley and Sons Inc, New York. pp 660.
- INTERLANDI, S.J. and CROCKETT, C.S. (2003). Recent water quality in the Schuylkill River, Pennsylvania, USA: a preliminary assessment of the relative influence of climate, river discharge and suburban development. *Water Research* 37: 1737-1748.
- INTERNATIONAL STANDARD ORGANISATION (ISO), 1996. Water quality- Determination of the acute lethal toxicity of substances to a freshwater fish [Brachydanio rerio Hamilton-Buchanan (Teleostei, Cyprinidae)] Part 1: Static method, Second edition. ISO 7346-1:1996(E). pp 11.
- IWQS 1998. Methods Manual: Method 3001 002 acute toxicity assessment using *Poecillia reticulata*. pp 14
- KARGIN, F. and ÇOŞUN, N.Y. (1999). Metal interactions during accumulation and elimination of zinc and cadmium in tissues of the freshwater fish *Tilapia nilotica*. *Bulletin of Environmental Contamination and Toxicology* 63: 511-519
- KEMPSTER, P.L. and VAN VLIET, H.R. (1991). Water quality fitness for use curves for domestic water. Draft internal report, Hydrobiological Research Institute, Department of Water Affairs and Forestry, Pretoria, South Africa.
- KEMPSTER, P.L., HATTINGH, W.A.J. and VAN VLIET, H.R. (1980). Summarized water quality criteria. Department of Water Affairs and Environmental Conservation, Hydrological Research Institute. Technical Report No TR 108. pp 45.
- KFIR, R. (1981). The detection and assay of potential carcinogens and toxicants in water by tissue culture techniques. Thesis for the degree of Doctor of Science in the Faculty of Science, University of Pretoria.

- KOTZE, P., DU PREEZ, H.H. AND VAN VUREN, J.H.J. (1999). Bioaccumulation of copper and zinc in *Oreochromis mossambicus* and *Clarias gariepinus*, from the Olifants River, Mpumalanga, South Africa. *Water S.A* 25(1): 99-110
- KUPCHELLA, C.E. and HYLAND, M.C. (1993). Environmental Science: Living within the System of Nature 3rd ed. Prentice Hall International Ltd. London. Pp 579.
- LARSSON, A., BENATSSON, B.E. and SVANBERG, O. (1976). Some haematological and biochemical effects of cadmium on fish. In: LOCKWOOD, A.P.M. [ed.] Effects of pollutants on aquatic organisms. Cambridge University Press, London pp. 193.
- LEE, R.M. and GERMING, S.D. (1980). Survival and reproductive performance of the desert pupfish, *Cyprinidon nevadensis* (Eigenmann and Eigenmann) in acid water. *Journal of Fish Biology* 17: 507
- LIU, D.L., YANG, Y.P., HU, M.H., HARRISON, P.J. and PRICE, N.M. (1987). Selenium content of marine food chain organisms from the coast of China. *Marine Environmental Research* 22: 151-165.
- MANAHAN, S.E. (1993). Fundamentals of environmental chemistry. Lewis Publishers, Boca Raton. Pp. 417-437.
- MAUL, J.D. and COOPER, C.M. (2000). Water quality of seasonally flooded agricultural fields in Mississippi, USA. *Agriculture, Ecosystems and Environment* 81: 171-178.
- MOUNT, D.I. (1973). Chronic effects of low Ph on fathead minnow survival, growth and reproduction. *Water Research* 7: 987 - 993
- MULLER, W.J. and PALMER, C.G. (2002). The use of *Daphnia spp.* and indigenous river invertebrates in whole effluent toxicity testing in the Vaal catchment. WRC Report No 815/1/02. pp 57.
- MUNIZ, I.P. and LEIVESTAD, H. (1980). Toxic effects of aluminium on the brown trout, *Salmo trutta*. Pp 320-321. In: D.DRABLOS & A. TOLLEN [ed.] Ecological impact of acid precipitation. SNSF Project, Norway.
- MVUNGI, A., HRANOVA, R.K., LOVE, D. (2003). Impact of home industries on the water quality in a tributary of the Marimba River, Harare: implications for urban management. *Physics and Chemistry of the Earth* 28: 1131-1137.
- NELSON, J.A. (1982). Physiological observations on developing rainbow trout, *Salmo gairdneri* (Richardson), exposed to low pH and varied calcium iron concentrations. *Journal of Fish Biology* 20: 359
- NEWMAN, J.M., CLAUSEN, J.C. and NEAFSEY, J.A. (2000). Seasonal performance of a wetland constructed to process dairy milkhouse wastewater in Connecticut. *Ecological Engineering* 14: 181-198.
- NEWMAN, M.C. and MCINTOSH, A.W. (1991). Metal Ecotoxicology: Concepts and Applications. Lewis Publishing, Michigan. pp 399.
- NUSSEY, G., VAN VUREN, J.H.J. and DU PREEZ, H.H. (1999). Bioaccumulation of Al, Cu, Fe and Zn in the tissues of the moggel from Witbank Dam, upper Olifants River catchment (Mpumalanga). *South African Journal of Wildlife Research* 29(4): 129-144.

- OHNESORGE, F.K. and WILHELM, M. (1991). Zinc In: E. MERIAN [ed.]. Metals and their compounds in the environment: Occurrence, analysis and biological relevance. pp. 1438.
- ORGANISATION FOR ECONOMIC CHANGE AND DEVELOPMENT (OECD), (1992). Guideline for testing of chemicals: fish acute toxicity test 203. pp 9.
- PETERSON, R.H., DAYE, P.G., LACROIX, G.L. and GARSIDE, E.T. (1976). Reproduction in fish experiencing acid and metal stress. In: JOHNSON, R.E. (ed), Acid Rain and Fisheries. American Fisheries Society, Bethesda, Md. pp 177.
- PIZZARO, I., GOMEZ, M., CAMARA, C. and PALACIOS, M.A. (2003). Arsenic speciation in environmental and biological samples Extraction and stability studies. *Analytica Chimica Acta* 495: 85-98.
- POLLING, L. (1999). Ecological aspects of the Ga-Selati River System, Northern Province, Republic of South Africa. Unpublished Phd. Thesis, University of the North, Pietersburg, South Africa. pp 312.
- RAND, G.M., WELLS P.G. and McCARTY L.S. (1995). Introduction to aquatic toxicology. In: G.M. Rand (ed.) Fundamentals of aquatic toxicology: effects, environmental fate and risk assessment. Taylor and Francis, United States. pp 3-66.
- RATTNER, B.A. and HEATH, A.G. (1995). Environmental factors affecting contaminant toxicity in aquatic and terrestrial vertebrates. In D.J. HOFFMAN, B.A. RATTNER, G.A. BURTON. Jr. and J.CAIRNS Jr. [ed.] Handbook of ecotoxicology. Lewis Publishers. Boca Raton.
- RATTNER, B.A. and HEATH, A.G. (1995). Environmental factors affecting contaminant toxicity in aquatic and terrestrial vertebrates. In D.J. HOFFMAN, B.A. RATTNER, G.A. BURTON. Jr. and J.CAIRNS Jr. [ed.] Handbook of ecotoxicology. Lewis Publishers. Boca Raton.
- ROBINSON, J. and AVENANT- OLDEWAGE, A. (1997). Chromium, copper, iron and manganese bioaccumulation in some organs and tissues of *Oreochromis mossambicus* from the lower Olifants River, inside the Kruger National Park. *Water S.A.* 23(4): 387-404.
- RODGERS, P., SOULSBY, C., HUNTER, C. and PETRY, J. (2003). Spatial and temporal quality of a lowland agricultural stream in northeast Scotland. *The Science of the Total Environment* 314-316: 289-302.
- RODRIGUEZ, V.M., JIMENEZ-CAPDEVILLE, M.E., and GIORDANO, M. (2003). The effects of arsenic exposure on the nervous system. *Toxicology letters* 145: 1-18.
- SAKAI, K., NAKAMURA, K., WAKAYAMA, M. and MORIGUCHI, M. (1997). Change in nitrite conversion direction from oxidation to reduction in heterotrophic bacteria depending on the aeration conditions. *Journal of Fermentation and Bioengineering* 84 (1): 47-52.
- SANDERS, M.J., DU PREEZ, H.H. and VAN VUREN J.H.J. (1999). Monitoring Cadmium and zinc contamination in fresh water systems with the use of the fresh water crab, *Potamanautius warrenii*. *Water S.A.* 25(1): 91-98.
- SAVORY, J. and WILLS, M.R. (1991). Aluminium In: E. MERIAN (ed.). Metals and their components in the environment: Occurrence, analysis and biological relevance. pp 1438.
- SCHEINBERG, I.H. (1991). Copper In: E. MERIAN [ed.]. Metals and their compounds in the environment: Occurrence, analysis and biological relevance. pp. 1438.

- SEYMORE, T., DU PREEZ, H.H. and VAN VUREN, J.H.J. (1995). Manganese, lead and strontium bioaccumulation in the tissues of the yellow fish, *Barbus marequensis* from the lower Olifants River, Eastern Transvaal. *Water S.A.* 21 (2): 159-171.
- SLABBERT, J.L., OOSTEHUIZEN, J., VENTER, E.A., HILL, E., DU PREEZ, M. and PRETORIUS, P.J. (1998). Development of guidelines for toxicity bioassaying of drinking and environmental waters in South Africa. Water Research Commission Report Number 358/1/98. Water Research Commission, Pretoria. pp 101
- SOLA, M. and DURAN, M. (1994). El zinc y los enzimas: Importancia y estudio mediante modelos. *Quimica e Industria* 42 (7): Pp 24-28 In: GALVIN, R.M. (1996). Occurrence of metals in waters: An overview. *Water SA* 22 (1): 7-18.
- SPRY, D. and WIENER, J.G. (1991). Metal bioavailability and toxicity to fish in low alkalinity lakes: A critical review. *Environmental pollution* 71(2-4): 243-304.
- STICKNEY, R.R. (1979). Principles of warm water aquaculture. Interscience Publication, John Wiley and Sons Inc. New York, USA.
- TRAIN, R.E. (1979). Quality criteria for water. US Environmental Protection Agency, Washington DC. Castle House Publications. pp 256.
- TRUTER, E. 1994. Methods for estimating chronic toxicity of a chemical or water sample to the Cladoceran *Daphnia pulex*. IWQS Report number N0000/00/OEQ/1394.
- TUCKER, C.S.; ROBINSON, E.H. 1990. Channel catfish farming handbook. Van Nostrand Reinhold, New York: 454pp
- VAN VUREN, J.H.J., DU PREEZ, H.H. and DEACON, A.R. (1994). Effects of pollutants on the physiology of fish in the Olifants River (Eastern Transvaal). Water Research Commission Report Number 350/1/94. Water Research Commission, Pretoria. pp 214.
- VAN VUREN, J.H.J., DU PREEZ, H.H., WEPENER, V., ADENDORFF, A., BARNHOORN, I.E.J., COETZEE, L., KOTZE, P. and NUSSEY, G. (1999). Lethal and Sublethal Effects of Metals on the Physiology of Fish: An Experimental Approach with Monitoring Support. Water Research Commission Report Number 608/1/99.
- VEGA, E., LESIKAR, B. and PILLAI, S.D. (2003). Transport and survival of bacterial and viral tracers through submerged-flow constructed wetland and sand filter system. *Bioresource Technology* 89: 49-56.
- VIARENGO, A. (1985) Biochemical effects of trace metals. *Marine Pollution Bulletin* 16(4): 153-158.
- VIARENGO, A. (1985) Biochemical effects of trace metals. *Marine Pollution Bulletin* 16(4): 153-158.
- VRHOVSEK, D., KUKANJA, V. and BULC, T. (1996). Constructed wetland (CW) for industrial waste water treatment. *Water Research* 30 (10): 2287-2292.
- WANG, W. (1987). Factors affecting metal toxicity to (and accumulation by) aquatic organisms- Overview. *Environment International* 13: 437-457
- WHO (World Health Organization) (1992). Cadmium-environmental aspects. World Health Organization, Geneva pp. 156.

-
- WILKINSON, J., JENKINS, A., WYER, M. and KAY, D. (1995). Modelling faecal coliform dynamics in streams and rivers. *Water Research* 29 (3): 847-855.
- WORLD HEALTH ORGANIZATION (WHO) (1993). Guideline for drinking water quality (2nd edn.) Vol 1. geneve (Switzerland).
- ZACCONE, R., CARUSO, G. and CALI, C. (2002). Heterotrophic bacteria in the northern Adriatic Sea: seasonal changes and ectoenzyme profile. *Marine Environmental Research* 54: 1-19.

CHAPTER 3: SEDIMENT:**3.1 Introduction:**

Metals have a high toxicity and worldwide distribution in the aquatic environment. They are also known to accumulate in sediments (Klavins *et al.*, 1998). Data concerning environmental effects of chemicals clearly indicates the accelerated and negative effects of the dispersal of metals and metalloids in the environment by anthropogenic activities, and the changes made to global chemical cycles (Mester *et al.*, 1998). The study of sediments in wetlands is important as wetlands act as natural filters for water in a system and thus act as a sink for contaminated suspended particles in the water column. Sediments also provide an indication of potential contamination on a temporal scale. The analysis of water indicates the contamination status at present where as sediment can provide information on the systems contamination history. Wetlands can also lead to increased contaminant levels in a water body during periods of increased water flow by remobilizing the settled particles and thus the resuspension of the contaminants in the water. These sediments are transported downstream and affect the ecosystems of the river downstream as well as flooded wetlands (Ulbrich *et al.*, 1997). The mobilization of sediments by water flow allows contaminants to penetrate deep into wetlands by flood waters (Ulbrich *et al.*, 1997). It is thus important to assess the sediment quality throughout the system.

Contaminants bind to the sediment particles. These contaminants can be either metallic in nature or originate from chemical compounds released into the system via a number of anthropogenic activities. This Chapter deals with the metal and metalloid contamination of sediments with sediment contamination by pesticides discussed in the next Chapter (Chapter 4).

Metals in a system can originate from either natural or anthropogenic sources. These metals may be present in several geochemical phases that act as reservoirs or sinks of trace metals in the environment (Li *et al.*, 1995). These phases include the broad categories: exchangeable, specifically adsorbed, carbonate, Fe-Mn oxides, organic matter and mineral lattice (Li *et al.*, 1995) it is thus recognised that the quantification of the chemical forms of metals in the sediment is essential for estimating the mobility and bioavailability of metals in the environment (Leschber *et al.*, 1985, Li *et al.*, 1995). Van Ryssen *et al.* (1999) noted in an article on the mobilization potential of trace metals in aquatic sediments that in anoxic sediments the exchangeable and carbonate fractions are negligible for all elements except Mn, where as 50-90% of metals are bound to the residual fraction (Van Ryssen *et al.*, 1999)

It is generally recognised that information about the physico-chemical forms of elements is necessary to understand their environmental behaviour such as mobility and bioavailability (Tack and Verloo, 1995). To this end the sediment samples collected from the 18 localities throughout the system were subjected to a five point sequential extraction. The sequential extraction of metals from solid media is a common tool used in the analysis of environmental geochemistry (Sutherland and Tack, 2003). This process uses different reducing and oxidising agents to remove each fraction of the bound metals in the sample to allow for the evaluation of the total metal content available for uptake by organisms or bioavailability.

3.2 Materials and methods:

The sediment samples were taken from the upper 5 cm of the substrate and placed in 350 ml plastic honey jars. The samples were then placed on ice till freezing could take place. Once frozen the samples were returned to the lab for further analysis.

3.2.1 Sequential extraction:

In the laboratory the sediment samples underwent a process of sequential extraction. It was decided to use a five fraction extraction, which identifies the non residual metal concentrations among the three basic operationally- defined host fractions (Ngiam and

Lim, 2000). The process followed was a modified from the process set out by Tessier *et al.* in 1979. The process involves subjecting the sediment samples to chemicals of decreasing pH and increasing oxidising strength, to remove the operationally defined host fractions corresponding to the exchangeable, carbonate, reducible and organic/sulphide phases (Ngiam and Lim, 2000). Sediment samples were dried in an oven at 60 degrees to remove any moisture from them. Approximately 1g of dry sample was placed in a 50 ml nalgene polyethylene centrifuge tube before it underwent extraction. Figure 3.1 gives a brief outline of the process followed during the extraction process. The five fractions removed were:

- 1: The exchangeable fraction,
- 2: The fraction bound to carbonates,
- 3: The fraction bound to iron and manganese oxides,
- 4: The fraction bound to organic matter,
- 5: The residual or inert fraction.

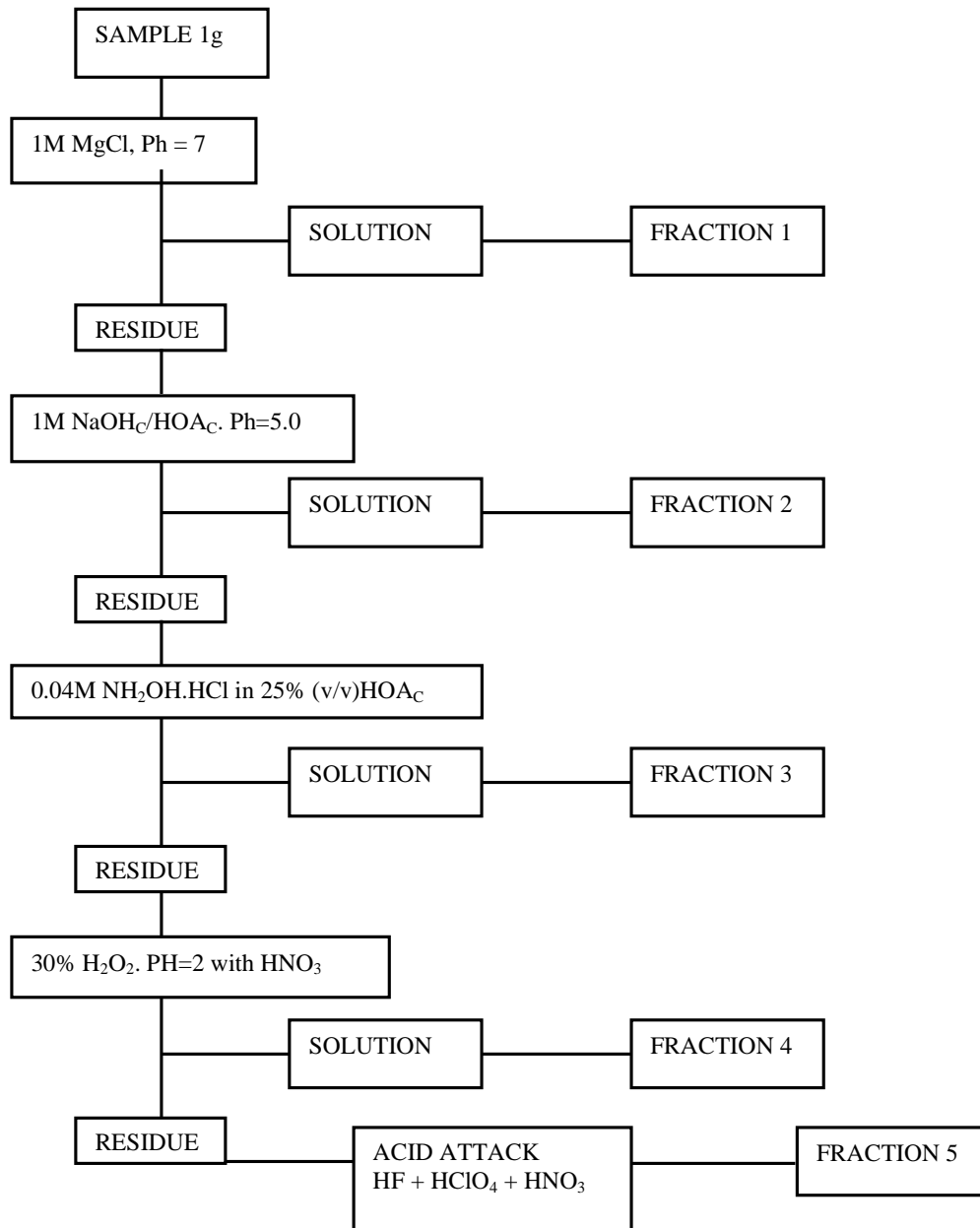


Figure 3.1: Flow diagram for Tessier sequential extraction (Coetzee, 1992)

3.2.2. Extraction process:

Once in the centrifuge tube the 1g of sediment was subjected to different digestive processes. For first fraction was subjected to a 1M Magnesium chloride solution. This was made up by dissolving 23.793g of magnesium chloride (Merck) in 250 ml of deionised water. The pH was then adjusted to 7 using NaOH. 8ml was then added to each sample and left at room temperature for one hour. The samples were then centrifuged at 3000 r/min for 30 minutes before the extraction phase was removed and placed in amber glass bottles. The extraction was then diluted to 50 ml.

The residue then underwent further extraction with 8ml of a 1M sodium acetate/acetic acid buffer to pH5 for five hour at room temperature. The sodium acetate/acetic acid buffer was made up by dissolving 20.5g of sodium acetate (BDH Lab Reagents) in 250 ml deionised water, buffered to pH5 with acetic acid (Associate Chemical Enterprises). The samples were then centrifuged at 3000 r/min for 30 minutes and the extraction was removed and placed in amber glass bottles and made up to a volume of 50ml.

The residue then underwent the 3rd extraction under mild reducing conditions. 20ml of a 0.4M hydroxyl amine hydrochloride in 25 % (v/v) acetic acid was added to the residue and incubated in a warm bath for 6 hours at 96 ± 3 degrees. The hydroxyl amine hydrochloride solution was prepared by dissolving 6.949g of hydroxyl ammonium chloride (Saarchem) in 25 % (v/v) acetic acid to a volume of 250ml. The reduced samples were then centrifuged at 3000 r/min for 30 minutes and the extract was placed in amber glass bottles and made up to a volume of 50 ml.

The residue underwent the 4th reduction. The reducing agent in this fraction was a nitric acid/ hydrogen peroxide solution. 3ml of a 0.02M nitric acid (Saarchem) and 5ml 30% (v/v) hydrogen peroxide (Saarchem) were added to each sample. The mixture was then heated in a warm bath at $85^{\circ}\text{C} \pm 2^{\circ}\text{C}$ for 3 hours. Once cool 5ml of a 3.2 M ammonium acetate was added to the samples and left to stand for an hour before the samples were centrifuged at 3000 r/min for 30 min. the extract was then removed and placed in amber glass bottles and made up to a volume of 50 ml. 3.2M ammonium acetate was made up by dissolving 26.6656g of ammonium acetate in 100ml 20% (v/v) nitric acid.

The 5th fraction was then extracted from the residue. This was achieved by digesting the residue with a 5:1 mixture of hydrofluoric acid (Merck) and perchloric acid (Saarchem). The samples were placed in a warm bath at $96 \pm 3^{\circ}\text{C}$ for 3 hours and then left to cool. They were centrifuged at 3000r/min for 30 minutes and the extract removed and placed in plastic falcon tubes. The extract was made up to a volume of 50 ml.

3.2.3 ICP and statistical analysis:

After the sequential extraction process the samples were subjected to an ICP-MS analysis and the results were then statistically analysed using the SPSS statistical analysis software. From the scan performed metals that corresponded to those found to be potential problems from the water scan performed earlier were chosen for discussion. Table 3.1 indicates the metals to be discussed and the detection limits of the ICP-MS. Table 3.1 also indicates the sediment guideline levels used to determine if the concentrations reported are toxic to aquatic life.

Sediment Guideline levels give a range determined by the Effect Range-Low (ERL) and the Effect Range Median (ERM) (EPA, 1999).

Table 3.1: Table of potential problem metals, ICP-MS detection limits and SQG levels in mg/kg.

Metal	ICP-MS Detection limit (mg/kg)	SQG (mg/kg)
Aluminium	0.000112	
Chromium	0.000008	81-370
Copper	0.000062	34-270
Cadmium	0.000014	1.2-9.6
Zinc	0.000028	150-410
Manganese	0.000008	
Lead	0.000022	46.7-218
Arsenic	0.000002	8.2-70

3.3 Results and discussions:

The results in this Chapter will be discussed on a metal for metal basis. The results reported cover a comparison of the different fractions of the sequential extraction between the different sampling months as well as the fractions within the sampling months. Maximum concentrations will also be reported.

Aluminium

The increased bioavailability of aluminium in sediments comes about by the remobilization of sediment particles by increased water flow and agitation in conjunction with decrease in pH. This increased bioavailability can have various physiological effects on the organisms in the system. These negative physiological effects have been discussed in Chapter 2 and will thus not be discussed in this Chapter.

Figures 3.2 A-D indicate the various aluminium concentrations observed at each locality during the different sampling periods for each fraction. The concentrations recorded indicate that the majority of the aluminium in the sediment is partitioned into the 4th and 5th fractions. This would indicate that the aluminium in the sediment is not very bio available and will only be release in the presence of a strong reducing agent or if the pH in the system were to decrease.

Figure 3.2 A indicates the aluminium concentrations during the August 2001 sampling period. The localities situated at Nylsvley, Haakdoorn and Moorddrift had the highest concentrations of aluminium with concentrations of 25938.12 mg/kg, 17787.95 mg/kg and 24757.47 mg/kg respectively.

No significant differences were found between the mean aluminium concentrations in fraction 4. Significant differences ($P < 0.05$) were however noted between the following fractions:

- Fraction 1 and fraction 3
- Fraction 1 and fraction 5
- Fraction 2 and fraction 3
- And Fraction 2 and fraction 5.

A comparison between fraction 1 and 2 indicated no significant difference.

Figure 3.2 B indicates the aluminium concentrations during the November 2001 sampling period. The figure indicates that fraction 5 once again had the highest concentrations of aluminium with the highest concentrations being found at Nylsvley, Haakdoorn, the sewage treatment works (STW) and Tobias (T.Station).

Nylsvley

Sediment

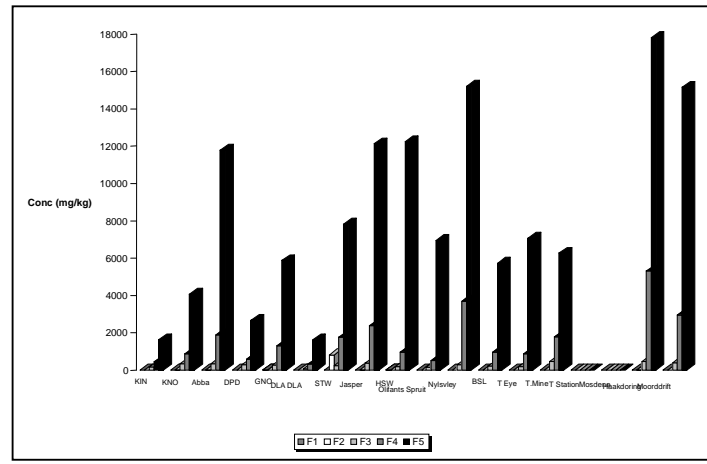
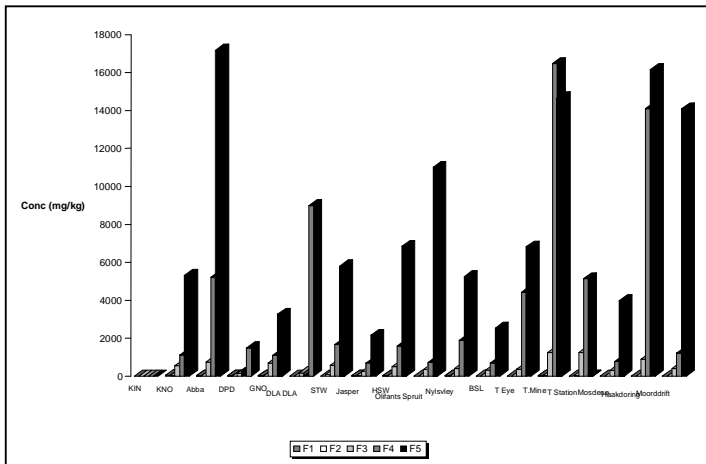
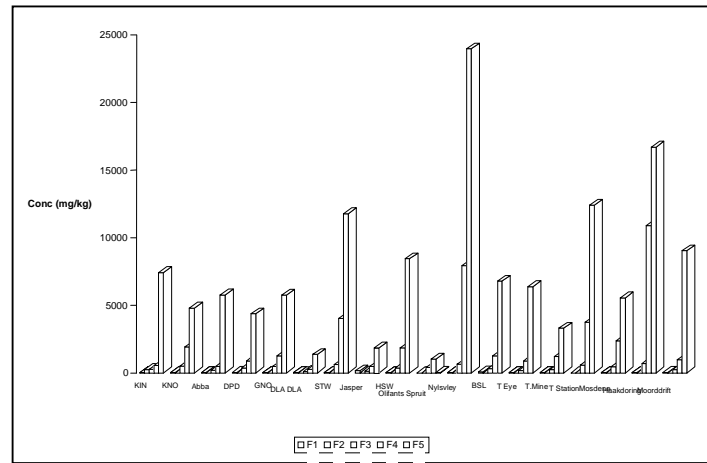
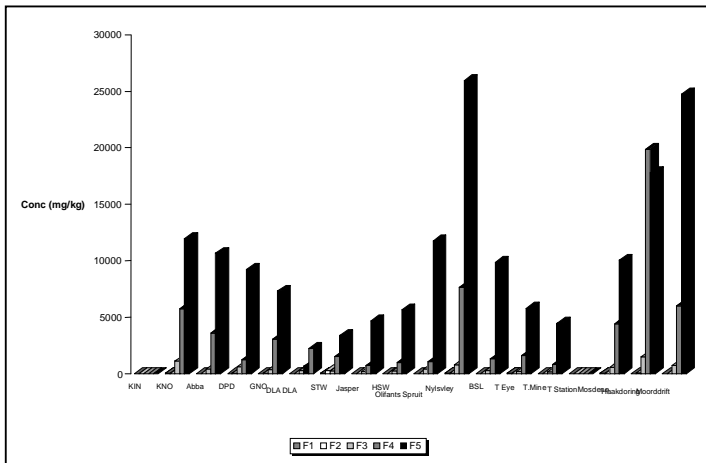


Figure 3.2: Aluminium concentrations (mg/kg) in the different fractions during the different sampling months.
 A: August 2001, B: November 2001, C: March 2002 and D: July 2002.

Maximum aluminium concentrations of 23951.68 mg/kg, 16692.37 mg/kg, 11751.84 mg/kg and 12390.49 mg/kg were recorded respectively. The statistical analysis of the November 2001 sampling period indicates that all mean fraction concentrations were significantly different with a few exceptions. The difference between fraction 1 and 2 and fraction 3 and 4 were not significantly different with p values being greater than 0.05.

Figure 3.2 C indicates the aluminium concentrations recorded during the March 2002 sampling period. Abba, Mine (T.Mine), Haakdoorn and Moorddrift had the highest aluminium concentrations with concentrations of 17182.31 mg/kg, 14677.73 mg/kg, 16162.49 mg/kg and 14090.91 mg/kg respectively. Significant differences ($P < 0.05$) between fraction means in the system were recorded between the following fractions:

- Fraction 1 and fraction 3
- Fraction 2 and fraction 3
- Fraction 1 and fraction 5
- Fraction 2 and fraction 5

Figure 3.2 D indicates the aluminium concentrations for the July 2002 sampling period. Maximum concentrations were found in fraction 5 and were found at Haakdoorn, Moorddrift and Nylsvley. These sites had aluminium concentrations of 17822.93 mg/kg, 15160.46 mg/kg and 15225.94 mg/kg respectively. Significant differences were recorded between the following fractions:

- Fraction 1 and fraction 3
- Fraction 1 and fraction 4
- Fraction 1 and fraction 5
- Fraction 2 and fraction 3
- Fraction 2 and fraction 4
- Fraction 2 and fraction 5
- Fraction 3 and fraction 4
- Fraction 3 and fraction 5

No significant differences were found between the mean aluminium concentrations for fractions 1, 2, 4 and 5 when the fractions for each month were compared to each other. The comparison between the fraction 3 concentrations for March and July were however significantly different.

The results indicate that the majority of the aluminium is concentrated in the 5th or inert fraction. This would indicate that the majority of the aluminium extracted from the sediment is from a lattice or detrital origin and can be taken as from a natural source (Jain, 2004).

Chromium:

Chromium is a relatively scarce metal that occurs in several states. The most toxic of these states is the chromium VI or hexavalent state. The physiological effects of chromium on aquatic organisms vary. These physiological effects have been discussed in Chapter 2 and will thus not be discussed again.

Fytianos and Lourantou observed in their study of sediment from Lake Volvi and Koronia in Northern Greece, that chromium is primarily distributed in the reducible (Fe/Mn oxide), residual and oxidizable fractions. They found that metals bound to these different fractions have different potentials for remobilization and for uptake by biota (Fytianos and Lourantou, 2004).

Figure 3.3 indicates the chromium concentrations found in the samples collected during the different sampling months. The graphs illustrate during the different months the chromium is released in the different fractions. Chromium is predominantly remobilized in the oxidizable (F4) and inert fractions (F5). During the August 2001, November 2001 and March 2002 sampling period the observed partitioning of chromium concentrations is primarily in the 4th and 5th fractions. During the July sampling the majority of the chromium is found in the 5th fraction.

Figure 3.3 A indicates chromium concentrations throughout the system during the August 2001 sampling period. Fraction 4 had the highest chromium concentration. The maximum concentration recorded was 244.5129 mg/kg at Mosdene. Bad-se-Loop, Nylsvley, Haakdoorn and Moorddrift also had high concentrations of chromium in fraction 4. A comparison of mean chromium concentrations showed significant differences between fractions 1&3, 1&5, 2&3 and 2&5 with P values less than 0.05.

Figure 3.3 B indicates the chromium concentrations recorded during the November 2001 sampling period. The results indicate that the chromium concentrations are highest in fractions 4 and 5. The highest chromium concentrations were recorded at Nylsvley, Tobias, Mosdene and Haakdoorn. Concentrations were 34.82428 mg/kg, 37.47542 mg/kg, 50.98731 mg/kg and 41.76947 mg/kg respectively. Significant differences ($P < 0.05$) in mean chromium concentrations were recorded between fractions 1&2, 1&3, 1&4, 1&5, 2&3, 2&4 and 2&5.

Figure 3.3 C indicates chromium concentrations recorded during the March 2002 sampling period. The results indicate that the majority of the chromium is partitioned into the 3rd, 4th and 5th fractions. The maximum concentrations recorded were at Haakdoorn (F4) of 58.32133 mg/kg, Abba (F4) of 34.44767 mg/kg and at Mosdene (F5) 45.16986 mg/kg.

Figure 3.3 D indicates chromium concentrations recorded during the July 2002 sampling period. The results indicate that the majority of the chromium is partitioned in the 5th fraction. The maximum concentration observed was observed at Haakdoorn (78.3164 mg/kg). Abba, Sewerage Works, Jasper, Hessie-se-Water, Olifantspruit and Nylsvley also indicated elevated chromium levels compared to the other sites. Significant differences ($P < 0.05$) in mean chromium concentrations were observed between the following fractions 1&3, 1&4, 1&5, 2&3, 2&4 and 3&5.

In the comparison between the different fractions and the months sampled, fraction 4 and 5 indicated no significant differences ($P > 0.05$) between August 2001, November 2001, March 2002 and July 2002. Fraction 3 indicated a significant difference in mean chromium concentration between March 2002 and July 2002. Fraction 1 indicated significant differences between August 2001 & July 2002 but no significant differences between August 2001 and March 2002 and July 2002, and November 2001 and March 2002 and July 2002, but not between August 2001 and November 2001. Fraction 2 indicated significant differences between August 2001 and November 2001, August 2001 and March 2002, November 2001 and March 2002 and November 2001 and July 2002.

The highest chromium concentration recorded all fell within the Sediment Quality Guideline Range of 81 – 370 mg/kg (EPA, 1999). The majority of the chromium concentrations were below the lower limit of 81 mg/kg or ERL (effect range low) (EPA, 1999).

Nylsvley

Sediment

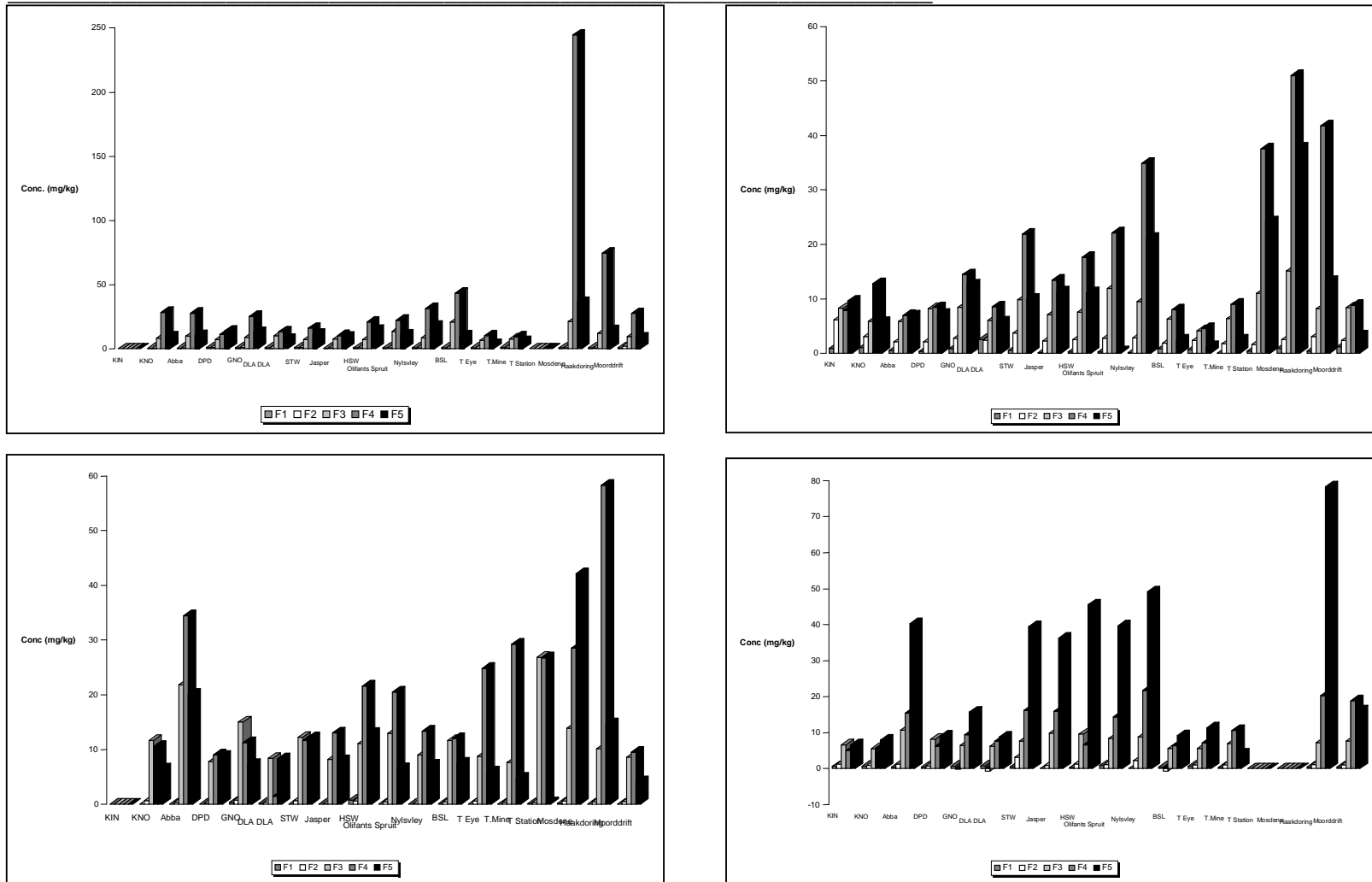


Figure 3.3: Chromium concentrations (mg/kg) in the different fractions during the different sampling months.
 A: August 2001, B: November 2001, C: March 2002 and D: July 2002.

Manganese

Manganese is an essential element (Health and Welfare Canada, 1980). It is a functional component in nitrate assimilation and is used as a catalyst in many enzymatic systems in both plants and animals (DWAF⁷, 1996). Manganese is readily oxidizable and settles out of the water column as MnO₂ (DWAF⁷, 1996). For an overview of the physiological effects of manganese on aquatic organisms refer to Chapter 2.

Figure 3.4 (A-D) indicates the manganese concentrations recorded during the sampling period in the system. The results indicate that the third fraction had the highest concentrations on manganese during the sampling period.

Figure 3.4 A indicates the manganese concentrations recorded during the August 2001 sampling period. One notable spike is visible on the graph at Mosdene. The 3rd and 4th fractions spike with manganese concentrations of 1742.529 mg/kg and 791.1142 mg/kg respectively. No significant differences in mean manganese concentrations were found between the different fractions for August 2001.

Figure 3.4 B indicates manganese concentrations recorded during the November 2001 sampling period. Notable spike in the third fraction occur at Groot Nyl Oog and Tobias with concentrations of 1112.466 mg/kg and 1904.166 mg/kg respectively. No significant differences were found between the fractions for the November 2001 sampling period.

Figure 3.4 C indicates the manganese concentrations recorded during the March 2002 sampling period. Fraction 3 indicated the greatest partition of manganese. Abba had the highest manganese concentration with all five fractions indicating increased levels of manganese. T. Mine and Jasper showed higher concentrations of manganese in fraction one (105.3879 mg/kg and 135.5336 mg/kg) and HSW recorded high concentrations in fraction one and two (130.9786 mg/kg and 141.4849 mg/kg). The increased concentrations in fraction one and two indicate manganese that is readily available and that could cause a potential threat to the system. No significant differences in mean manganese concentration were found between the different fractions for March 2002.

Figure 3.4 D indicates manganese concentrations recorded during the July 2002 sampling period. Fraction 3 indicated the highest concentrations with Jasper, Bad-se-Loop and Haakdoorn having concentrations of 2237.896 mg/kg, 1518.569 mg/kg and 1316.884 mg/kg respectively. No significant differences were recorded in mean manganese concentration between fractions for July 2002.

No significant differences were also recorded between corresponding fractions of each sampling month.

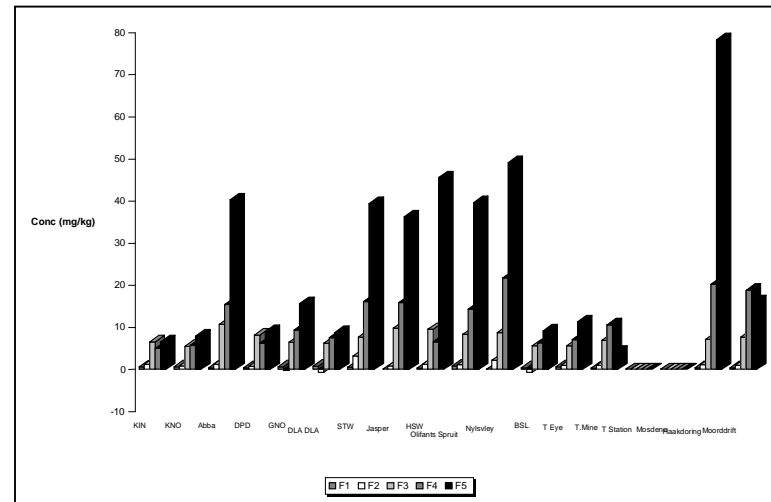
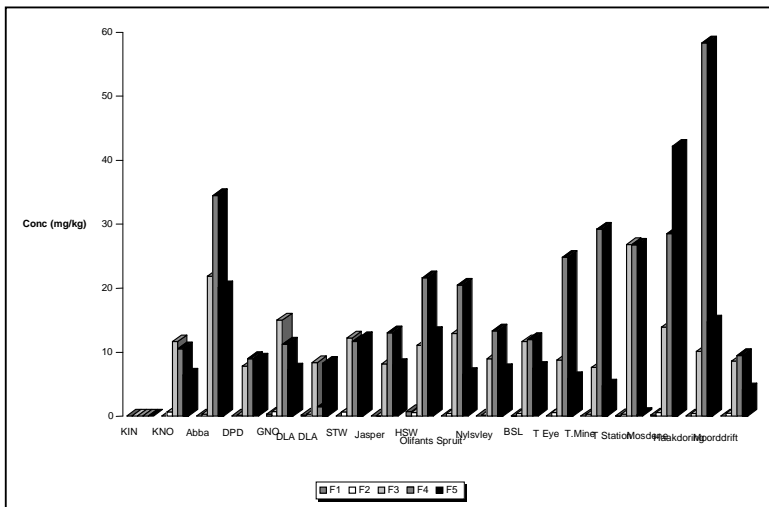
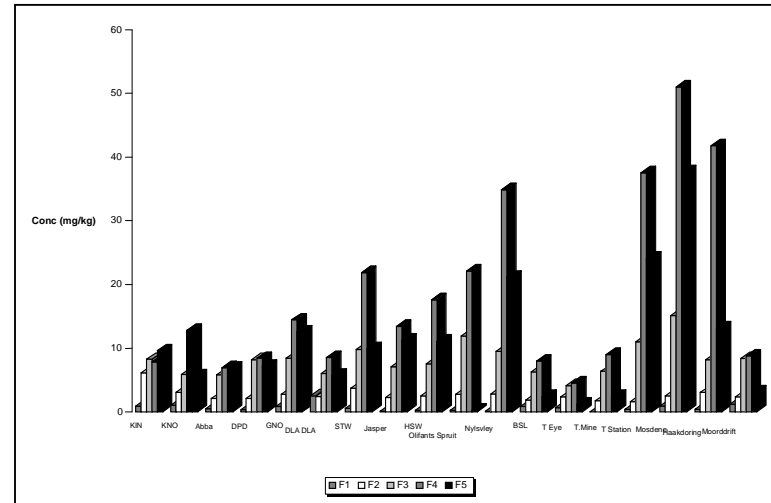
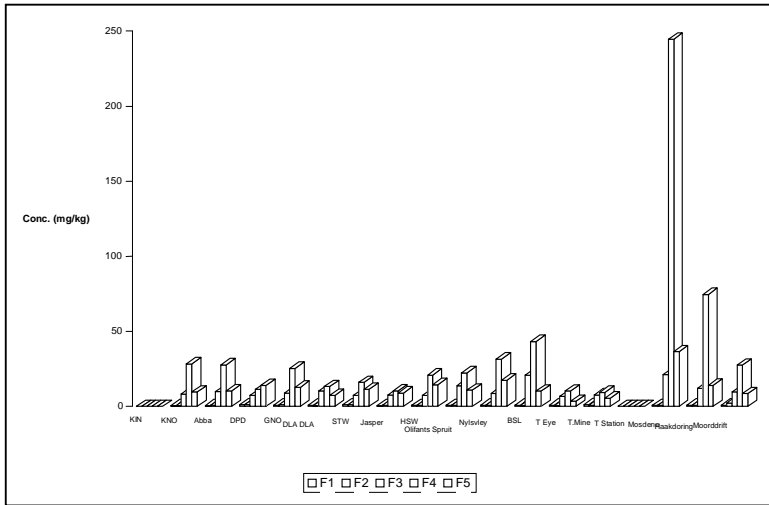
Zinc

Zinc is an essential micronutrient for all organisms. It forms the active site for various metalloenzymes (DWAF⁷, 1996). The physiological effects of zinc on organisms have been discussed in the previous Chapter, so for an overview of the physiological effects refer to Chapter 2.

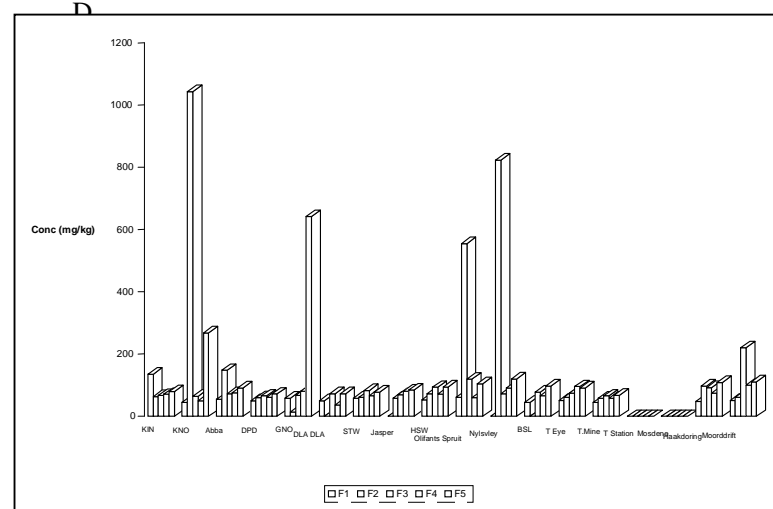
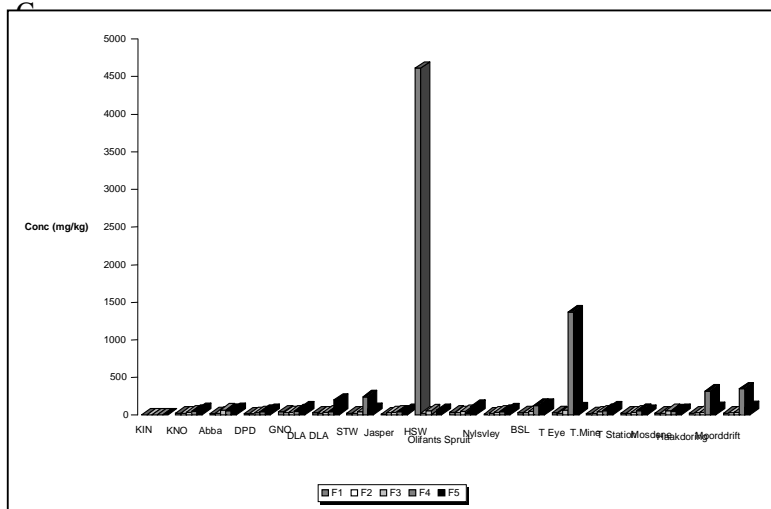
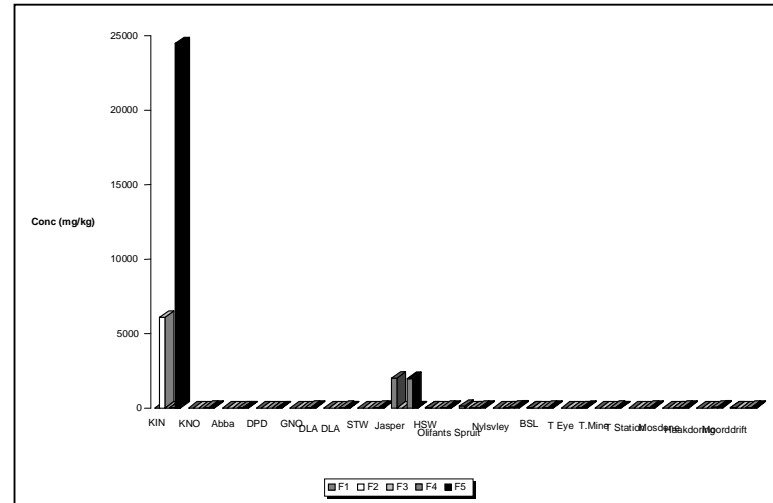
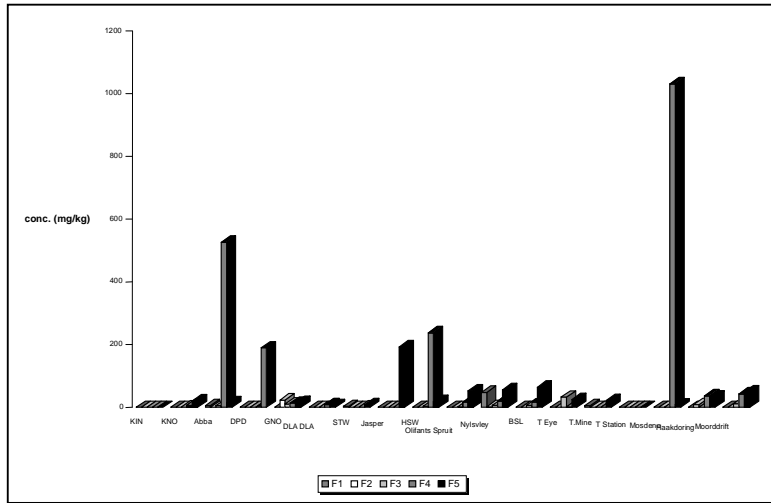
Figure 3.5 illustrates the zinc concentrations in the system during the sampling period. Figure 3.5 A indicates zinc concentrations recorded during the August 2001 sampling period. The results indicate that the majority of the zinc is partitioned into the 4th and 5th fractions.

Nylsvley

Sediment



Figures 3.4 (A-D). Manganese concentrations (mg/kg) in the different fractions during the different sampling months. A: August 2001, B: November 2001, C: March 2002 and D: July 2002.



Figures 3.5 (A-D): Zinc concentrations (mg/kg) in the different fractions during the different sampling months. A: August 2001, B: November 2001, C: March 2002 and D: July 2002.

Mosdene exhibited the highest concentration of zinc (1031.88 mg/kg). Fraction 4 at Abba and Mosdene exhibited zinc concentrations higher than the Sediment Quality Guideline (SQG) for aquatic ecosystems (EPA, 1999) of 150-410 mg/kg. No significant differences were found between the mean zinc concentrations for the different fractions during August 2001.

Figure 3.5 B indicates the zinc concentrations recorded in the system during the November 2001 sampling period. The highest zinc concentrations recorded were in the residual or inert fraction (Fraction 5) at Koh-I-Noor (24461.8 mg/kg). Fraction 2 Koh-I-Noor (6090.918 mg/kg), fraction 1 at Jasper (2000.789 mg/kg) and fraction 4 at Jasper (1959.353 mg/kg) were the other fractions that exhibited Zn concentrations higher than the SQG range. No significant differences were recorded for the mean zinc concentrations for the different fractions.

Figure 3.5 C indicates the zinc concentrations recorded throughout the system for the March 2002 sampling period. Fraction 3 at Hessie-se Water and fraction 4 at T. Eye exhibited the highest zinc concentrations of 4608.765 mg/kg and 1371.047 mg/kg respectively. All the other zinc concentrations recorded were below the SQG. Significant differences ($P < 0.05$) were found between fractions 2&3, 2&5 and 3&5.

Figure 3.5 D indicates the zinc concentrations recorded during the July 2002 sampling period. All sites and fractions indicate increased zinc concentrations for each of the five fractions. The highest concentrations were recorded at Klein Nyl Oog (fraction 2, 1043.751 mg/kg), Nylsvley (fraction 2, 824.1389 mg/kg), Groot Nyl Oog (fraction 5, 642.0671 mg/kg) and Olifantspruit (fraction 2, 554.6227 mg/kg). These four sites had zinc concentrations greater than the SQG. Three of these sites indicated these high concentrations in the readily available fraction 2 (ions bound to carbonates). No significant differences were found between the mean zinc concentrations in the different fractions.

Fraction 4 and 5 indicated no significant differences in the mean zinc concentrations during the different sampling months. In fraction 3 significant differences ($p < 0.05$) were found between August 2001 & July 2002, August 2001 & March 2002, November 2001 & July 2002 and March 2002 & July 2002. Fraction 2 indicated significant differences between August 2001 & March 2002 and fraction 1 between August 2001 & July 2002.

Copper:

Copper is a common environmental metal. It is essential in cellular metabolism but at high concentrations it can be highly toxic to fish (Grosell *et al*, 1997). Copper is generally remobilized with acid-base ion exchange and oxidation mechanism (Gomez Ariza *et al*, 2000).

Figures 3.6 (A-D) illustrates the copper concentrations observed in the system during the sampling period. Copper concentrations recorded all generally fell below or at the lower end of the SQG range (34-270mg/kg) with one exception. An excessively high copper concentration (786.533 mg/kg) was recorded at the source of the Klein Nyl River (Klein Nyl Oog - KNO) during July 2002 (Figure 5.5 D). This level was found in the second fraction which would indicate that copper is readily available. The site is however situated on a farm about 50 m from the source of the Klein Nyl River so it can be assumed the levels come from natural sources.

Figure 3.6 A illustrates the copper concentrations recorded during August 2001. The results indicate that the majority of the copper is found in fractions 3, 4 and 5. The maximum concentration recorded was in fraction 4 at Mosdene (64.3368 mg/kg). Mosdene also recorded the highest concentrations in fraction 3 (30.9564 mg/kg) and fraction 5 (26.5269 mg/kg). Significant differences were found in copper concentrations between fractions 1&5 and 2&5.

Figure 3.6 B indicates the copper concentrations in the system for November 2001 sampling period. Fractions 3, 4 and 5 exhibited the highest concentrations with Tobias having the highest concentration of 36.86751 mg/kg. Significant differences were found between fractions 1, 2 & 3 and fraction 5 as well as between fraction 2&4.

Figure 3.6 C indicates the copper concentrations recorded during the March 2002 sampling period. The majority of the copper is partitioned into fractions 3, 4, and 5. A maximum concentration was recorded at Bad-se-Loop (BSL) in fraction 4 (62.1038 mg/kg). Significant differences were recorded between fractions 1&5, 2&3 and 2&5.

Figure 3.6 D indicates the copper concentrations recorded during the July 2002 sampling period. As mentioned earlier Klein Nyl Oog (KNO) had a spike with copper levels higher than all the other sites. No significant differences were found in mean copper concentrations between the fractions in the system.

A comparison between the corresponding fractions from each sampling month indicated that fraction 3 was the only fraction that exhibited significant differences. Significant differences in mean copper concentration were between November 2001&July 2002 and March 2002 and July 2002.

Arsenic:

Arsenic is a highly toxic metalloid element (Rodrigues *et al.*, 2003, Pizzaro *et al.*, 2003). It is widely distributed as a trace element in rocks and soils and is mainly mobilized by microbial activities (Garcia-Sanchez and Alvarez-Ayuso, 2003). The physiological effects of arsenic on aquatic organisms have been discussed in the previous Chapter (Chapter 2) and will thus not be referred to in this Chapter.

Figures 3.7 (A-D) indicates the arsenic concentrations in the system during the sampling period. The graphs indicate that most of the arsenic is concentrated in the residual or inert fraction (fraction 5).

Figure 3.7 A indicates arsenic concentrations in the system during the August 2001 sampling period. The highest concentrations were recorded at Jasper (57.0031 mg/kg) and Tobiasoog (58.25977 mg/kg). The results indicate that arsenic concentrations from the different fractions fell within the SQG of between 8.2 and 70 mg/kg. Significant differences in mean arsenic concentrations were found between fractions 1, 2, 3 and fraction 5.

Figure 3.7 B indicates arsenic concentrations recorded in the system during the November 2001 sampling period. The graphs indicate that the majority of the arsenic is partitioned into the fifth fraction. The results indicate that arsenic concentrations ranged between 0.181436 mg/kg (Olifantspruit) and 8.835832 mg/kg (Tobiasoog). This range falls within or below the ERL of the SQG range. Fractions 1, 2, 3 and 4 indicated a significant difference in mean arsenic concentration from fraction 5.

Figure 3.7 C indicates arsenic concentrations during the March 2002 sampling period. The results indicate that the majority of the arsenic is once again partitioned in the residual fraction. The maximum concentration observed however was observed in the fourth fraction at Tobiasoog (12.75781 mg/kg). All concentrations occurred toward or below the lower end of the SQG range of between 8.2 and 70 mg/kg. Significant differences were found in mean arsenic concentrations between fractions 1, 2 3 and fraction 5 respectively.

Figure 3.7 D indicates the arsenic concentrations recorded during the July 2002 sampling period. The majority of the arsenic is partitioned in the fifth fraction. A spike on the graph at Tobiasoog can be observed with a maximum concentration of 28.77561 mg/kg. No significant differences were observed for July 2002 between the fractions.

Fraction 1 and 5 were the only fractions to show significant differences between sampling months. Fraction 1 indicated a difference in mean arsenic concentration for August 2001 and July 2002. Fraction 5 indicated a significant difference in mean arsenic concentration between November 2001 and March 2002.

Cadmium:

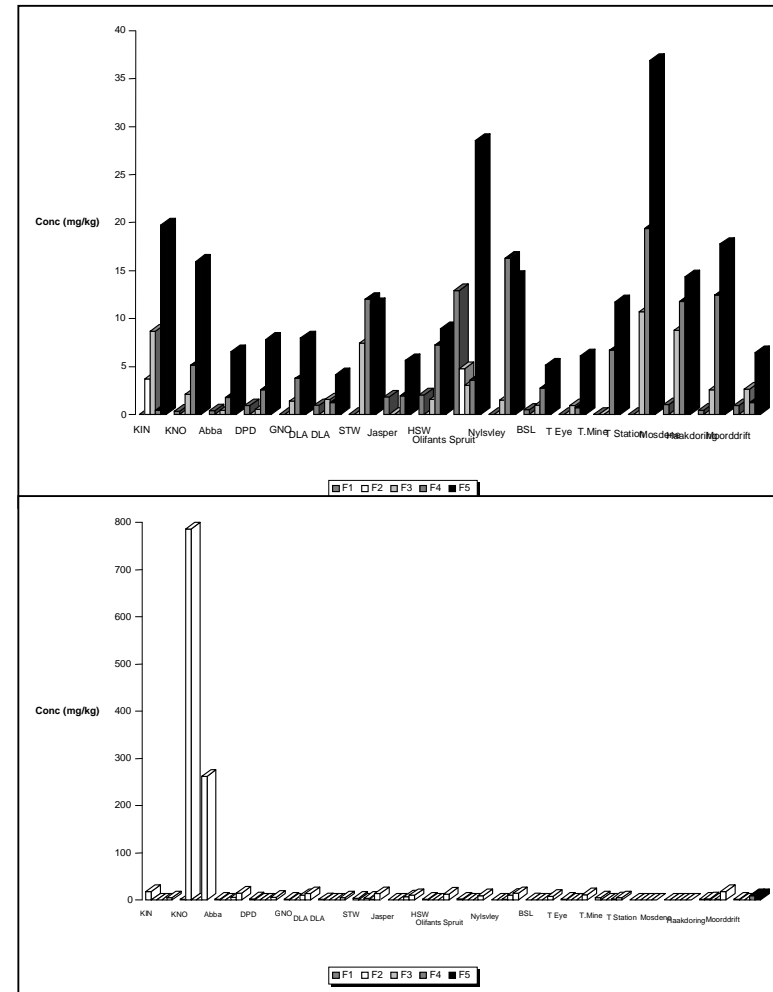
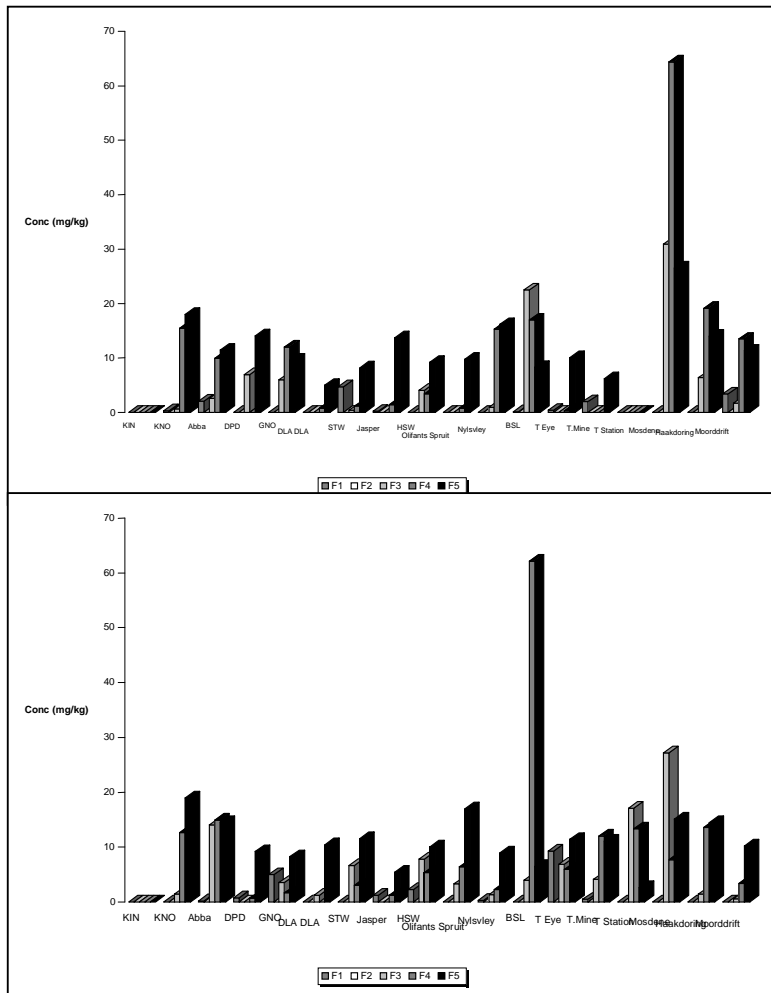
Cadmium is a non essential trace element that enters the environment via anthropogenic activities such as sewage sludge, fertilizers and pesticides (DWA⁷, 1996). Cadmium adsorbs strongly to sediments and organic matter (Sanders *et al*, 1999). The physiological effects such as decreased growth rates and negative effects on embryonic development (Newman and Mc Intosh, 1991) have been discussed in Chapter 2 and will thus not be discussed here. Figures 3.8 (A-D) indicate the cadmium concentrations recorded during the sampling period.

Figure 3.8 A indicates the cadmium concentrations determined for the system during August 2001. The maximum concentration recorded was in fraction 5 of 0.373014 mg/kg. Cadmium concentrations ranged between 0.000014 mg/kg and 0.373014 mg/kg. Significant differences ($P < 0.05$) were only noted in mean cadmium concentrations between fractions for fraction 3 and 4.

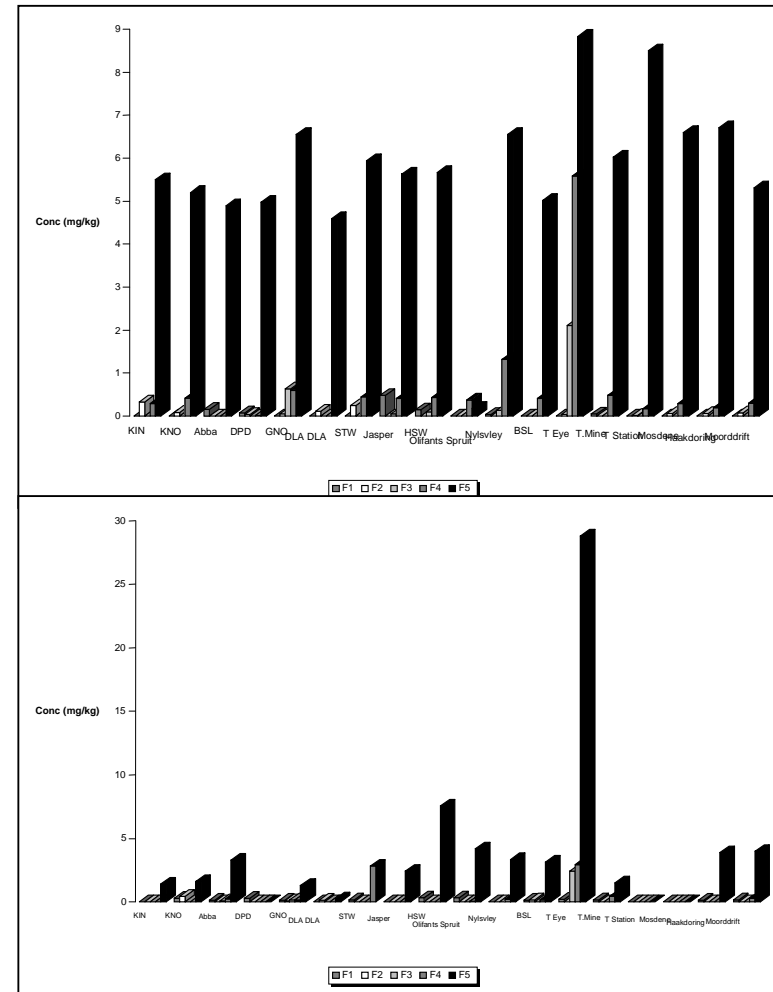
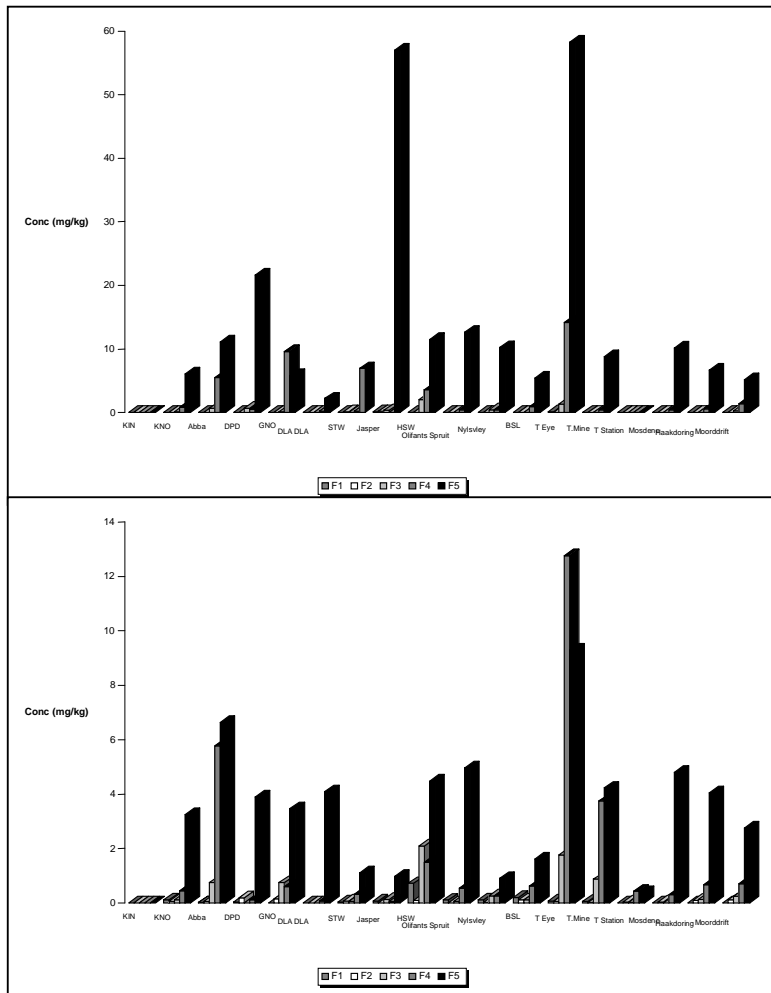
Figure 3.8 B indicates the cadmium concentrations recorded during the November 2001 sampling period. The maximum concentration recorded was at Olifantspruit in fraction 1 (1.032342 mg/kg). This would indicate that the cadmium is readily available. Significant differences were recorded between fractions 2&4 and 3&4.

Figures 3.8 C and D indicate cadmium concentrations recorded during the March 2002 and July 2002 sampling periods. 2 distinct peaks can be seen on the graphs. In figure 3.7 C the maximum concentration recorded, was recorded in fraction 3 at Tobiasoog (2.514241 mg/kg). In figure 3.7 D the maximum concentration of 0.422359 mg/kg was recorded for fraction 4 at Mine. No significant differences were recorded in mean cadmium concentrations between fractions.

The maximum cadmium concentration of 2.514241 mg/kg recorded during sampling fell within the SQG and thus cadmium poses little potential threat to the organisms in the system. No significant differences were recorded between corresponding fractions for each month with the exception of fraction 4. Significant differences were recorded between November 2001& and March 2002 and November 2001 and July 2002.



Figures 3.6 (A-D): Copper concentrations (mg/kg) in the different fractions during the different sampling months.
 A: August 2001, B: November 2001, C: March 2002 and D: July 2002.



Figures 3.7 (A-D): Arsenic concentrations (mg/kg) in the different fractions during the different sampling months.
 A: August 2001, B: November 2001, C: March 2002 and D: July 2002.

Lead:

Lead is a non essential trace element (Ewers and Schlipkoter, 1991). The toxicity of lead is dependent on life stage of the organism and the presence of organic material (Hellawell, 1986). A variety of physico-chemical factors lead to increased toxicity. Physiological effects caused by lead toxicity have been discussed in Chapter 2 and will thus not be discussed.

Figures 3.9 (A-D) indicates lead concentrations determined in the system during the sampling period. The results indicate that the majority of the lead is partitioned in fractions 3, 4 and 5.

Figure 3.9 A indicates lead concentrations recorded during the August 2001 sampling period. The majority of the lead is partitioned in the fifth fraction. Maximum lead concentrations were recorded at Mine (1226.425 mg/kg) and Olifantspruit (1118.408 mg/kg). Significant differences were found in mean lead concentrations of lead between fractions 1&5, 2&5, 3&5 and 4&5.

Figure 3.9 B indicates lead concentrations recorded during the November 2001 sampling period. The lead is partitioned primarily in fractions 3, 4 and 5. The maximum concentration was recorded at Nylsvley in fraction 3 (37,89936 mg/kg). Significant differences were recorded between fractions 1&2, 1&3, 1&4, 1&5, 2&3, 2&4 and 2&5.

Figure 3.9 C indicates lead concentrations recorded during the March 2002 sampling period. The majority of the lead is partitioned in fractions 3, 4 and 5. The maximum lead concentrations were recorded at Bad-se-Loop, Mine, Mosdene and Haakdoorn. The maximum values were all in fraction 3 and were 36.2352 mg/kg, 38.08004 mg/kg, 30.92636 mg/kg and 23.71075 mg/kg respectively. Significant differences were recorded between fractions 1&3, 1&4, 1&5, 2&3, 2&4 and 2&5.

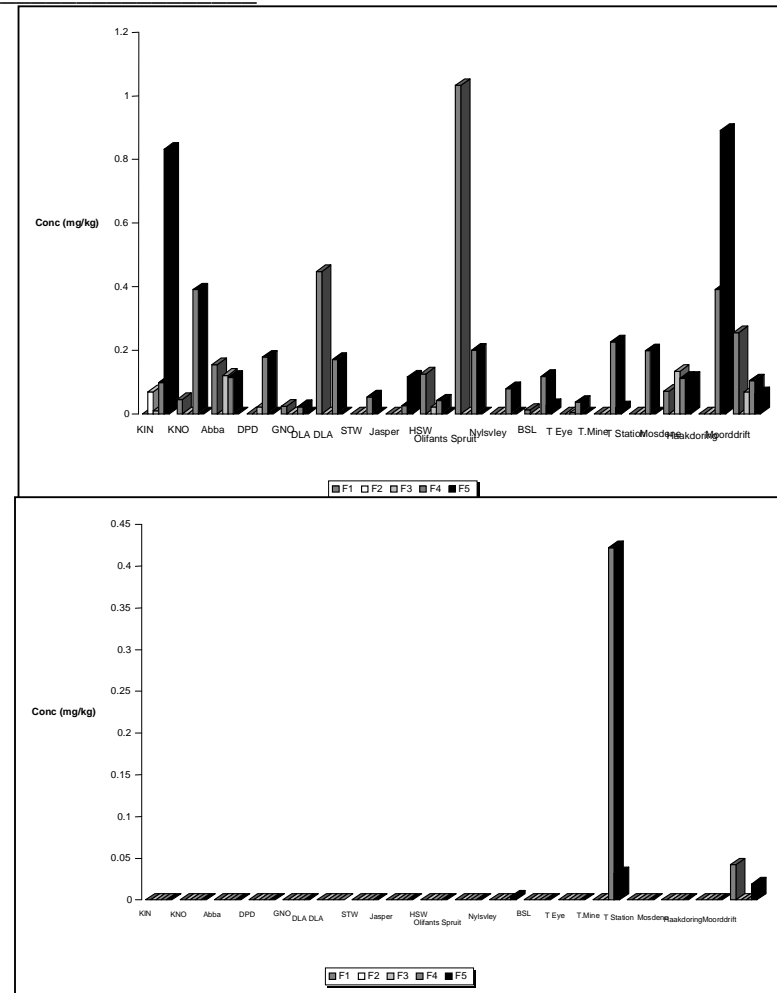
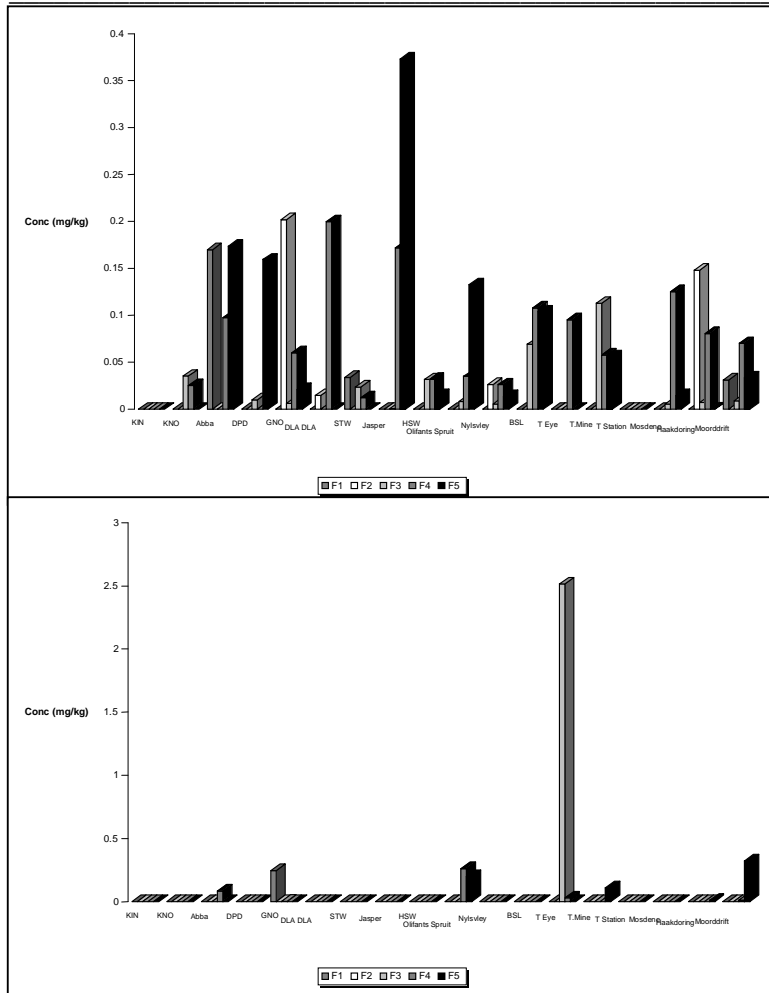
Figure 3.9 D indicates the lead concentrations recorded for the system during July 2002. The majority of the lead is partitioned in fraction 3. Maximum concentrations were recorded at Bad-se-Loop (44,19074 mg/kg) and Klein Nyl Oog (Fraction 2, 26,05897 mg/kg). Significant differences were recorded between fractions 1&3, 1&4 and 1&5.

No significant differences were noted in mean lead concentrations between corresponding fractions for fraction 2 and 3 from August 2001 to July 2002. In fraction 1 significant difference's were recorded between August 2001 & March 2001, August 2001 & July 2002, November 2001 & March 2001 and November 2001 & July 2002. In fraction 5 significant differences were recorded between August 2001 & November 2001, March 2002 & July 2002, November 2001 & March 2002 and November 2001 & July 2002

All lead concentrations fell within the SQG range except for fraction 5 in August 2001. This is little cause for concern as fraction 5 is the residual/inert fraction and the lead would thus be from natural sources.

Nylsvley

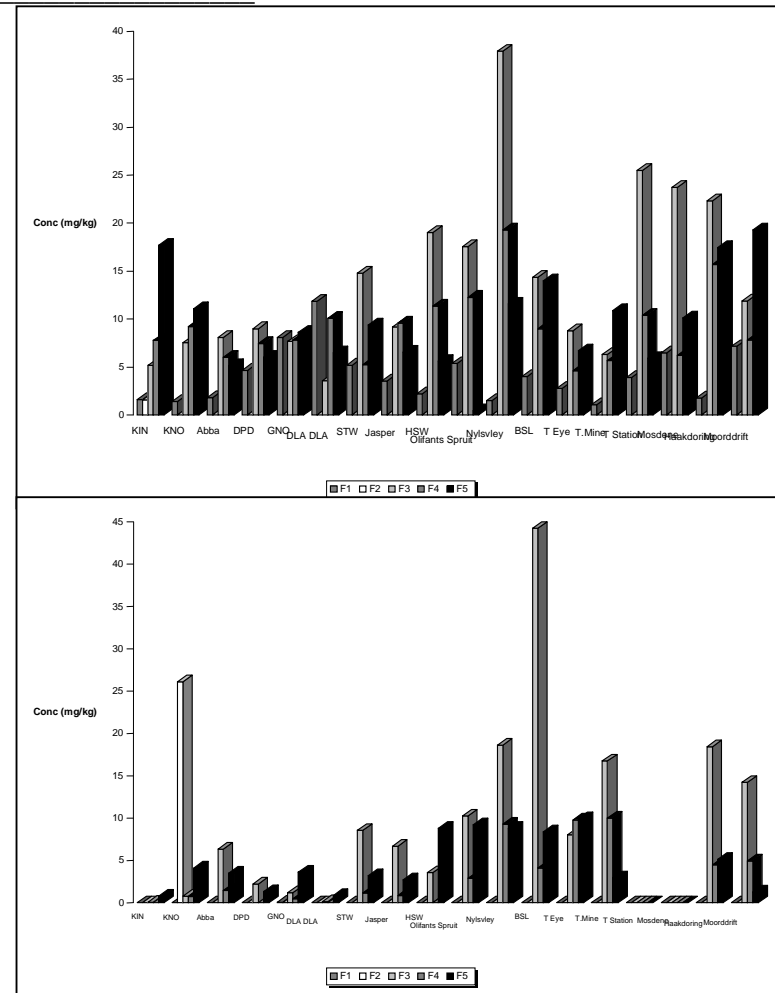
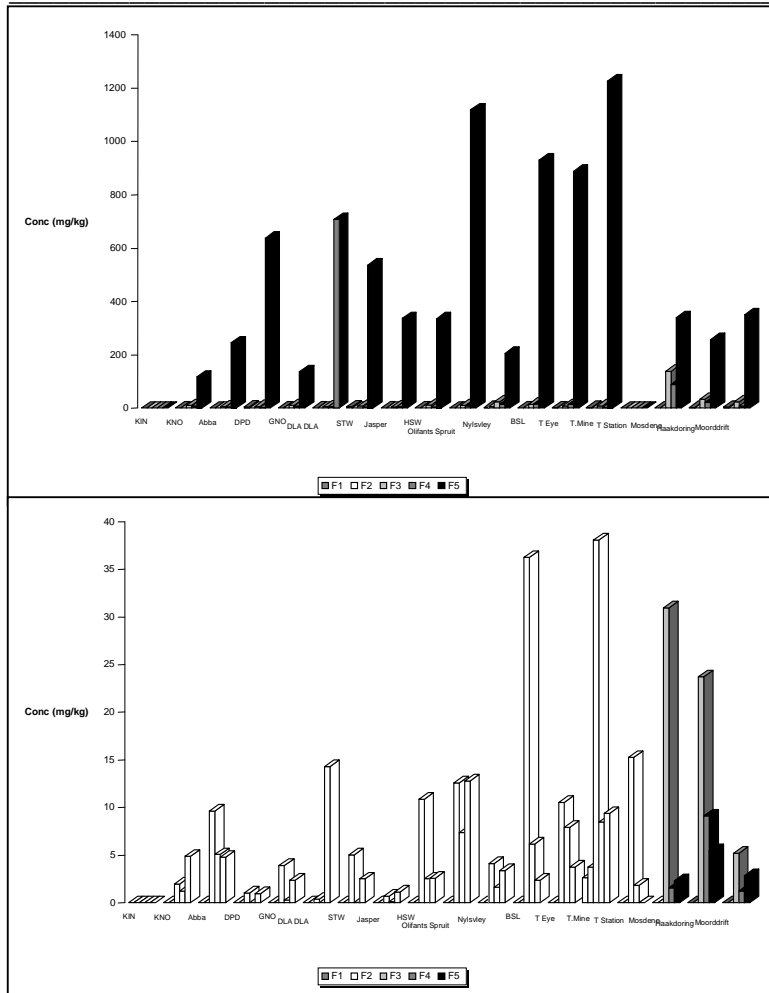
Sediment



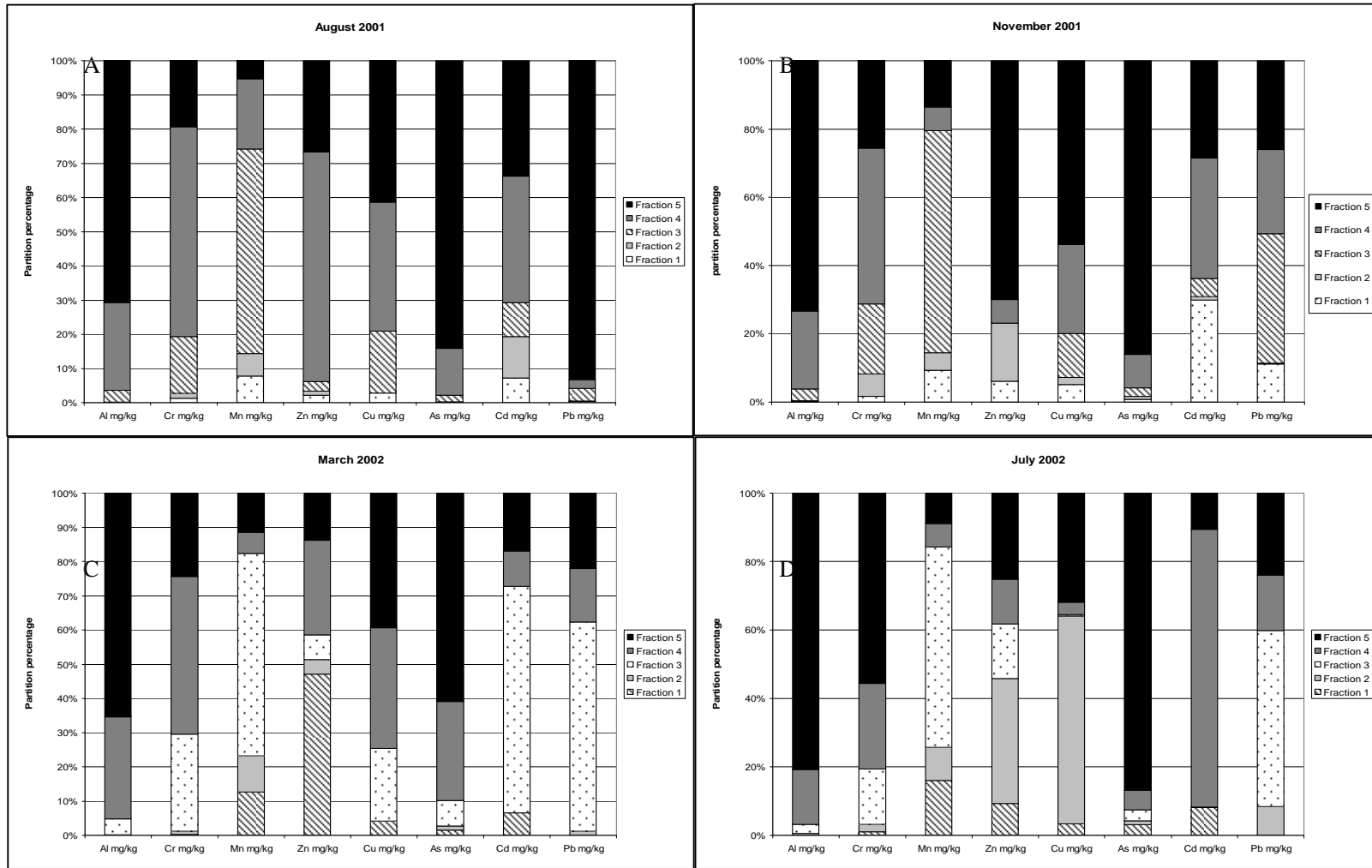
Figures 3.8 (A-D): Cadmium concentrations (mg/kg) in the different fractions during the different sampling months. A: August 2001, B: November 2001, C: March 2002 and D: July 2002.

Nylsvley

Sediment



Figures 3.9 (A-D): Lead concentrations (mg/kg) in the different fractions during the different sampling months.
 A: August 2001, B: November 2001, C: March 2002 and D: July 2002.



Figures 3.10 (A-D). Fraction percentages of mean metal concentrations.

3.4 Conclusions and recommendations:

The results clearly indicate the metals are not a problem in the system. Figures 3.10 (A-D) indicate a stacked graph of the percentage makeup of the different fractions for the different metals. The graphs indicate that the majority of the metals are partitioned into the 3rd, 4th and 5th fractions. Indications are that the metals generally are not very bio available. This would indicate that metals from the sediments pose little to no potential threat to the organisms in the system. This would also imply that most of the metals in the system are from a natural source.

The results also indicate that all metal concentrations fell within the lower end of the Sediment Quality Guideline Range. These will thus have little or no effect on the organisms in the system. Zinc was the only metal that had concentrations greater than the guideline values. This is however little cause for concern as they were recorded in the residual or inert fraction (Fraction 5). This would imply that they are natural concentrations in the sediment.

It may thus be concluded that the bioavailability of metals may be related to chemical form in sediment structure and not total concentration (Tüzen, 2003).

Regular monitoring of sediment metal content is not necessary and once a year should provide enough information to determine if the system is deteriorating.

3.5 References:

- COETZEE, P. (1993). Determination and speciation of heavy metals in sediments of the Hartbeespoort Dam by sequential chemical extraction. *Water SA* 19 (4): 291-300.
- DEPARTMENT OF WATER AFFAIRS AND FORESTRY (DWAF7). (1996). South African Water Quality Guideline volume 7: Aquatic ecosystems. pp 159.
- ENVIRONMENTAL PROTECTION AGENCY (EPA). (1999). Sediment Quality Guidelines developed for the national status and trends program. Report number 6/12/99. <http://www.epa.gov/waterscience/cs/pubs.htm> (May 2004)
- EWERS, U. and SCHLIPKOTER, H.W. (1991). Lead In: E. MERIAN [ed.]. Metals and their compounds in the environment, Occurrence, analysis and biological relevance. pp. 1438
- FYTIANOS, K. and LOURANTOU, A. (2004). Speciation of elements in sediment samples collected at lakes Volvi and Koronia, N. Greece. *Environment International* 30: 11-17.
- GARCIA-SANCHEZ, A. and ALVAREZ-AYUSO, E. (2003). Arsenic in soils and water and its relation to geology and mining activities (Salamanca Province, Spain). *Journal of Geological Exploration* 80: 69-79.
- GOMEZ ARIZA, J.L., GIRÁLDEZ, I., SÁNCHEZ-RODAS, D. and MORALES, E. (2000). Comparison of the feasibility of three extraction procedures for trace metal partitioning in sediments from south west Spain. *The Science of the Total Environment* 246: 271-283.
- GROSELL, M.H., HOGSTRAND, C. and WOOD, C.M. (1997). Copper uptake and turnover in both copper acclimated and non-acclimated rainbow trout (*Oncorhynchus mykiss*). *Aquatic Toxicology* 38: 257-276.
- HEALTH AND WELFARE CANADA, (1980). Guidelines for Canadian drinking water quality 1978. Supporting documentation. Supply and services Canada, Hull.
- HELLAWELL, J.M. (1986). Biological indicators of freshwater pollution and environmental management. Elsevier Applied Science Publishers Ltd., London pp. 546.
- JAIN, C.K. (2004). Metal fractionation study on bed sediments of River Yamuna, India. *Water Research* 38: pp 569-578.
- KLAVINS, M., RODINOV, V. and VERESKUNS, G. (1998). Metal and organochlorine compounds in fish from Latvian lakes. *Bulletin of Environmental Contamination and Toxicology* 60: 538-545.
- LESCHBER, R., DAVIS, R.D. and L'HERMITE, P. (1985). Chemical methods for assessing Bio-available metals in sludge and soils. Elsevier, London, 96 pp.
- LI, X., COLES, B.J., RAMSEY, M.H. and THORNTON, I. (1995). Sequential extraction of soils for multielement analysis by ICP-AES. *Chemical Geology* 124: 109-123.
- MESTER, Z., CREMISINI, C., GHIARA, E. and MORABITO, R. (1998). Comparison of two sequential extraction procedures for metal fractionation in sediment samples. *Analytica Chimica Acta* 359: 133-142.
- NEWMAN, M.C. and MCINTOSH, A.W. (1991). Metal Ecotoxicology: Concepts and Applications. Lewis Publishing, Michigan. pp 399.

- NGIAM, L. and LIM, P. (2001). Speciation patterns of heavy metals in tropical estuarine anoxic and oxidized sediments by different sequential extraction schemes. *The Science of the Total Environment* 275 (1-3): 53-61.
- PIZZARO, I., GOMEZ, M., CAMARA, C. and PALACIOS, M.A. (2003). Arsenic speciation in environmental and biological samples Extraction and stability studies. *Analytica Chimica Acta* 495: 85-98.
- RODRIGUEZ, V.M., JIMENEZ-CAPDEVILLE, M.E., and GIORDANO, M. (2003). The effects of arsenic exposure on the nervous system. *Toxicology letters* 145: 1-18.
- SANDERS, M.J., DU PREEZ, H.H. and VAN VUREN J.H.J. (1999). Monitoring Cadmium and zinc contamination in fresh water systems with the use of the fresh water crab, *Potamanautius warrenii*. *Water S.A.* 25(1): 91-98.
- SUTHERLAND, R.A. and TACK, F.M.G. (2003). Fractionation of Cu, Pb and Zn in certified reference soils SRM 2710 and SRM 2711 using the optimized BCR sequential extraction procedure. *Advances in Environmental Research* 8: 37-50.
- TACK, F.M.G. and VERLOO, M.G. (1995). Chemical speciation and fractionation in soil and sediment heavy metal analysis: a review. *International Journal of Environmental Analytical Chemistry* 59: 225-238.
- TESSIER, A. CAMPBELL, P.G.C. and BISON, M. (1979). Sequential extraction procedure for speciation of particulate trace metals. *Analytical Chemistry* 51: 844-851.
- TÜZEN, M. (2003). Determination of trace metals in the River Yesilirmak sediments in Tokat, Turkey using sequential extraction procedure. *Microchemical Journal* 74: 105-110.
- ULBRICH, K., MARSULA, R., JELTSCH, F., HOFMANN, H. and WISSEL, C. (1997). Modelling the ecological impact of contaminated river sediments on wetlands. *Ecological Modelling* 94: 221-230.
- VAN RYSSSEN, R., LEERMAAKERS, M. and BAEYENS, W. (1999). The mobilisation potential of trace metals in aquatic sediments as a tool for sediment quality classification. *Environmental Science and Policy* 2: 75-86.

CHAPTER 4: PESTICIDES

4.1 Introduction:

An increasing population growth over the last century or so has led to the increasing need for food production. This increase has been met by the increased use of pesticides to control problem plants, insects and diseases. Hundreds of these chemicals are introduced into commerce every year and may eventually end up in the environment (Fernandez-Alba *et al.*, 2002). Many of these pesticides are harmful to the biospheres they land up in. It was decided to analyse the sediment in the system to determine what toxins are in the system, regions of input and if possible to quantify their levels. Schultz (2001) concluded in a study of rainfall-induced pesticide input from orchards of the Lourens River that runoff played a valuable role in the deposition of pesticides into the system. The decision was made to analyse sediment samples to indicate what pesticides are present in the system, with an eye on further analysis of potential problem toxins in fish tissues of organisms found in the system.

4.1.1 Polychlorinated biphenyls:

Persistent organic pollutants or POPs are chemicals that may persist for long periods of time in the environment (Suchan *et al.*, 2004). POPs include polychlorinated biphenyls (PCBs), polychlorinated dibenzo p-dioxins (PCDDs), dibenzo furans (PCDFs), polycyclic aromatic hydrocarbons (PAHs) and organochlorine pesticides (OCPs). POPs have been cause for concern since the 60's (Nowell *et al.*, 1999). Due to their toxicity, persistence, tendency to accumulate in biota and their adverse effects on wildlife, the use of the majority of organochlorine pesticides was banned in the USA (Sapozhnikova *et al.*, 2004).

PCBs are categorized as members of the group of ubiquitous, persistent, lipophilic, bio accumulative, highly toxic micro contaminants in the global environment (Falandysz *et al.*, 2004). Due to their lipophilic nature they are persistent and are chemically prone to long range transport through the atmosphere and along the course of river systems (Suchan *et al.*, 2004). Their lipophilic nature also allows them to accumulate in ecosystems.

PCBs are primarily industrial in origin but due to their physical and chemical properties they mimic organochlorine pesticides (Sapozhnikova *et al.*, 2004). PCBs have a long half-life and their presence in the environment is cause for concern relating to human health and toxicity in animals (Gallant *et al.*, 2000). PCBs are a group of halogenated aromatic compounds, which consist of a large group of 209 congeners (Storelli *et al.*, 2004, Kimbrough, 1995)

In the aquatic environment fish are exposed to hydrophobic organic compounds (HOCs) both via direct contact with water and via the food uptake routes (Burreau *et al.*, 2004). Uptake from water takes place over the gill membranes and in food over the membranes in the gastro-intestinal tract.

PCBs have various adverse physiological effects on aquatic organisms, which include development, reproduction and behaviour (Oliver, 1985 and Ferraro *et al.*, 1991). They also have a carcinogenic and mutagenic effect on organisms (Suchan *et al.*, 2004). As PCBs bio-magnify in the food chain the contamination of a system by PCBs can have negative effects on humans as well. Exposure to PCBs can lead to non-Hodgkin lymphoma, serious intellectual impairment in newborns, lymphatic/haematological malignancies and breast cancer in humans (Zuccato *et al.*, 1999).

4.1.2 Pyrethroids:

Synthetic pyrethroids are amongst the most potent and effective insecticides available, and account for more than 30% of the worlds market in insecticides (Philip and Rajasree, 1996). Synthetic pyrethroids such as Cypermethrin are being used increasingly due to their low toxicity in mammals, non-persistence and efficiency (Moore and Waring, 2001). Their efficiency and non-persistence lends them to be a good pesticide to use although they a

highly toxic to fish (Coats and O'Donnel-Jeffery, 1979). They are used extensively in agriculture; in house holds and low doses are used in aquaculture as an effective insecticide on the ectoparasite *Argulus* spp. (Das & Mukherjee, 2003; Adhikari *et al.*, 2004).

Cypermethrin is a neurotoxin and is highly toxic to fish, aquatic and terrestrial invertebrates and some beneficial arthropods such as shrimps and lobsters (Polat *et al.*, 2002). The toxicity is due to the organisms in ability to degrade and metabolise pyrethroids (David *et al.*, 2004). They are also known to induce alterations in carbohydrate metabolism (Philip *et al.*, 1995). The hypersensitivity of fish to this group of toxins is partially due to differences in specie specific pyrethroid metabolism and increased sensitivity of the piscine nervous system (Moore and Waring, 2001). The main route of uptake of pyrethroids in fish is via the gills (Adhikari *et al.*, 2004). Pyrethroids like cypermethrin have also a negative effect on the reproduction rates of some fish species. Studies by Moore and Waring (2001) indicate that the pyrethroids inhibit olfactory detection of the male reproductive priming pheromone from female Atlantic salmon. The pheromone is considered to be involved in the synchronisation of spawning between the sexes and thus negatively effects the spawning and reproduction of salmon.

4.2 Materials and methods:

Sediment samples from five localities along the system were analysed for the presence of five main groups of compounds. These localities were analysed under both high and low flow conditions during 2002. Table 4.1 indicates the five main groups analysed and the derivatives in these groups. All in all 55 derivatives were scanned.

The sediment samples were analysed at the SANAS accredited laboratory, Testing and Conformity Services (Pty) Ltd, an affiliate of the SABS. The single determination was carried out using test method AP0 27 A: Method for the determination of OC and OP pesticides, Triazines, Pyrethroids, PCB's and Carbamates in sediment.

4.3 Results and discussion:

The role of pesticides in the Nyl River system is not yet known. Sediment samples were analysed for a qualitative analysis at Testing and Conformity Services (Pty) Ltd, an affiliate of the SABS, and a the SANAS accredited laboratory. The results obtained indicate that pesticides pose no potential threat to the system. Of the 51 potential contaminants in the system results were obtained for four of them. These derivatives (PCB 118, 101 and 52 and the pyrethroid, cypermethrin) were all below 12 µg/kg, which are concentrations too low to quantify accurately.

4.4 Conclusions and recommendations:

These results indicate that POPs are present in the system but the results are not conclusive, as data from sediments may not be representative of biota concentrations and cannot give information on contamination patterns in the upper levels of the food chain (Binelli and Provini, 2003). Further analysis on tissues from organisms in the system is recommended to ascertain the true extent of the pesticide contamination in the Nyl River System.

Table 4.1: Pesticide derivatives analysed in sediments to determine pesticide concentrations in sediment.

Carbamates	Organochlorine pesticides	Organophosphorus pesticides & Triazines	Pyrethroids	PCB's
Aldicarb	Alpha-BHC	Dichlorvos	Lambda-Cyhalothrin	2,4-Dichlorobiphenyl (PCB-8)
Carbofuran	Beta-BHC	Phosdrin	Permethrin	2,3,3-Trichlorobiphenyl (PCB-20)
Carbaryl	Gamma-BHC (Lidane)	Diazinon	Cypermethrin	2,4,4-trichlorobiphenyl (PCB-28)
Methomyl	Delta-BHC	Chlorpyrifos-methyl	Deltamethrin	2,2',5,5'-tetrachlorobiphenyl (PCB-52)
Oxamyl	Heptachlor	Chlorpyrifos-ethyl	Esfenvalerate	2,2',4,5,5'-pentachlorobiphenyl (PCB-101)
3-Hydroxycarbofuran	Aldrin	Malathion		2,3',4,4',5-pentachlorobiphenyl (118)
Aldicarbocofuran	Epoxide	Bromophos-methyl		2,2',3,4,4',5' hexachlorobiphenyl (PCB-138)
Aldicarb sulfoxide	Endosulfan-I	Parathion		2,2',4,4',5,5'-hexachlorobiphenyl (PCB-153)
Methiocarb	4,4-DDE	Carbophenothion		2,2',3',4,4',5,5'-heptachlorobiphenyl (PCB-180)
Propoxur	Dieldrin	Atrazine		
	Endrin	Simazine		
	Endosulfan-II	Terbutylazine		
	DDD	Clorothalonil		
	Endrin aldehyde			
	4,4-DDT			
	Endosulfan sulphate			
	Endrin ketone			
	metoxychlor			

4.5 References:

- ADHIKARI, S., SARKAR, B., CHATTERJEE, A., MAHAPATRA, C.T. and AYYAPPAN, S. (2004). Effects of cypermethrin and carbofuran on certain haematological parameters and prediction of their recovery in a freshwater teleost, *Labeo rohita* (Hamilton). *Ecotoxicology and Environmental Safety* 58: 220-226.
- BINELLI, A. and PROVINI, A. (2003). The PCB pollution of Lake Iseo (N.Italy) and the role of biomagnification in the pelagic food web. *Chemosphere* 53: 143-151.
- BUREAU, S. ZEBUHR, Y. BROMAN, D. and ISHAQ, R. (2004). Biomagnification of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) studied in pike (*Esox lucius*), perch (*Perca fluviatilis*) and roach (*Rutilus rutilus*) from the Baltic Sea. *Chemosphere* 55: 1043-1052.
- COATS, J.R. and O'DONNELL-JEFFERY, N.L. (1979). Toxicity of four synthetic pyrethroid insecticides to rainbow trout. *Bulletin of Environmental Contamination and Toxicology* 23: 250.
- DAS, B.K. and MUKHERJEE, S.C. (2003). Toxicity of cypermethrin in *Labeo rohita* fingerlings: biochemical, enzymatic and haematological consequences. *Comparative Biochemistry and Physiology part c* 134: 109-121.
- DAVID, M., MUSHIGERI, S.B., SHIVAKUMAR, R. and PHILIP, G.H. (2004). Response of *Cyprinus carpio* (Linn) to sublethal concentration of cypermethrin: alterations in protein metabolic profiles. *Chemosphere* 56: 347-352.
- FALANDYSZ, J., WYRZYKOWSKA, B., WARZOCHA, J., BARSKA, I., GARBACIK-WESOŁOWSKA, A. and SZEFER, P. (2004). Organochlorine pesticides and PCBs in perch *Perca fluviatilis* from the Odra/Oder River Estuary, Baltic Sea. *Food Chemistry* 87: 17-23.
- FERRARO, S.P., LEE II, H., SMITH, L.M., OZRETICH, R.J. and SPECHT, D.T. (1991). Accumulation factors for eleven polychlorinated biphenyl congeners. *Bulletin of Environmental Contamination and Toxicology* 46: 276-283.
- GALLANT, T.L., SINGH, A. and CHU, I. (2000). PCB 118 induces ultrastructural alterations in the rat liver. *Toxicology* 145: 127-134.
- KIMBROUGH, R.D. (1995). Polychlorinated biphenyls (PCBs) and human health: an update. *CRC Critical Revision of Toxicology* 25: 133-163.
- MOORE, A. and WARING, C.P. (2001). The effects of a synthetic pyrethroid pesticide on some aspects of reproduction in Atlantic salmon (*Salmo salar* L.). *Aquatic Toxicology* 52: 1-12.
- NOWELL, L.H., CAPEL, P.D. and DILEANIS, P.D. (1999). Pesticides in stream sediment and aquatic biota: distribution, trends, and governing factors. Lewis Publishers, Boca Raton, FL.
- OLIVER, B.G. and NIIMI, A.J. (1985). Bioconcentration factors of some halogenated organics for the rainbow trout: limitations in their use for prediction of environmental residues. *Environmental Science Technology* 19: 842-849.
- PHILIP, G.H. and RAJASREE, B.H. (1996). Action of cypermethrin on tissue transamination during nitrogen metabolism in *Cyprinus carpio*. *Ecotoxicology and Environmental Safety* 34: 174-179.

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- PHILIP, G.H., REDDY, P.M. and SRIDEVI, G. (1995). Cypermethrin-induced *in vivo* alterations in the carbohydrate metabolism of freshwater fish, *Labeo rohita*. *Ecotoxicology and Environmental Safety* 38: 173-178.
- POLAT, H., ERKOÇ, F.Ü., VIRAN, R. and KOÇAK, O. (2002). Investigation of acute toxicity of beta-cypermethrin on guppies *Peocilia reticulata*. *Chemosphere* 49: 39-44.
- SAPOZHNIKOVA, Y., BAWARDI, O. and SCHLENK, D. (2004). Pesticides and PCBs in sediments and fish from the Salton Sea, California, USA. *Chemosphere* 55: 797-809.
- SCHULTZ, R. (2001). Rainfall-induced sediment and pesticide input from orchards into the Lourens River, Western Cape, South Africa: importance of a single event. *Water Research* 35(8): 1869-1876.
- STORELLI, M.M., STORELLI, A., D'ADDABBO, R., BARONE, G. and MARCOTRIGIANO, G.O. (2004). Polychlorinated biphenyl residues in deep-sea fish from Mediterranean Sea. *Environment International* 30: 343-349.
- SUCHAN, P., PULKRABOVÁ, J., HAJŠLOVÁ, J. and KOCOUREK, V. (2004). Pressurized liquid extraction in determination of polychlorinated biphenyls and organochlorine pesticides in fish samples. *Analytica Chimica Acta* xxx (2004). pp xxx-xxx. Article in press.
- ZUCCATO, E., CALVARESE, S., MARIANI, G., MANGIAPAN, S., GRASSO, P., GUZZI, A. and FANELLI, R. (1999). Level sources and toxicity of polychlorinated biphenyls in the Italian diet. *Chemosphere* 38 (12): 2753-2765.

CHAPTER 5: MACROINVERTEBRATES

5.1 Introduction:

A recent worldwide development is the introduction of in-stream biological effects or response monitoring in water resources management. This type of response monitoring, commonly referred to as biomonitoring, is increasingly being recognised as an important component in the overall monitoring and assessment of water resources. The use of biological field assessments of, for example fish or macro invertebrate communities, provides an integrated and sensitive measurement of environmental problems and represents progress in the assessment of ecological impacts, and hence in the management of water resources (Karr and Chu, 1997).

Several local advances in applied aquatic science provide a basis for integrating *in situ* biological assessments into the country's surface water monitoring and assessment strategy. These advances include the development and standardisation of rapid bio-assessment techniques and the delineation of homogenous ecological regions, which provide a spatial framework for selecting reference and monitoring sites within the biomonitoring context.

The South African Scoring System (SASS) is a rapid biological assessment method which was developed to evaluate the impact of changes in water quality using aquatic macroinvertebrates as indicator organisms. The technique has been developed, tested and refined over several years in South Africa and is in its 5th version. In SASS5, the macroinvertebrate families are scored according to their sensitivity to deterioration in water quality. The scores range from 0 to 15, with highly pollution-tolerant species scoring low and intolerant/highly sensitive species scoring high. The three index values derived from SASS5 are the total score (sum of scores for the taxa present), the number of taxa present in the sample, and the average score per taxon (ASPT = total score/no. of taxa). SASS5 and ASPT scores have been shown to relate directly to water quality, and are particularly sensitive to organic pollution. The SASS5 score is considerably influenced by the number of biotopes from which the organisms are collected (Chutter, 1995). The most pollution tolerant taxa occur in almost all biotopes, so the scores for severely polluted sites tend not to be influenced by biotope diversity. Conversely, where water quality is "natural", SASS5 total scores tend to be extremely sensitive to biotope diversity (Chutter, 1995).

Since resident aquatic benthic macro invertebrate communities integrate and reflect the effects of chemical and physical impacts, occurring over extended periods of time, they are regarded as good indicators of overall ecological integrity. A major problem when using benthic macro invertebrate communities as indicators of water quality in rivers is the inherent natural differences in community structure caused by factors other than water quality. Methods suitable in shallow waters are not feasible in deeper waters where alternative methods have to be used. Although several techniques exist for sampling benthic communities in streams, little or no guidance is available on sampling benthic communities in wetland habitats.

The major aims of this project are:

- To assess the diversity of macro invertebrate communities in the entire Nyl system and to describe distribution and abundance, and to show how these fluctuate in different sections of the system during seasonal changes from 2001-2003.
- To describe the differences between the macroinvertebrates of different biotopes.
- Attempt to modify current SASS 5 sampling techniques and scoring system developed for lotic river systems and adapt it for lentic wetland habitats as well as to investigate alternative sampling techniques.

The macro invertebrate fauna is the most diverse and abundant group of aquatic animals in a river and wetland system. The types and densities of invertebrates found at any point along the system are a reflection of water quality, habitat availability and flow regime of that

reach, and could be described as an integrated reflection of the condition of the river/wetland. For this reason the invertebrate fauna is most often used to provide an index of conditions, and particularly water-quality. Invertebrates are also a key community in the ecological functioning of a river/wetland – breaking down organic detritus in association with the micro flora to recycle nutrients, filtering material out of the water column, grazing algae and fungi from the river bed, turning over the sediments, and serving as important food source for other species such as fish and frogs.

5.2 Materials and methods:

The sampling method selected was a modification on the method used in the South African Scoring system version 5 (SASS5). The sampling method should be simple and not require the services of highly trained personnel. Due to the nature of the habitat available in most wetlands, it was decided to sample the aquatic and marginal vegetation component in the system. This habitat is important in the system and provides the necessary refuge and food sources for the macro invertebrates to colonise. In the SASS5 protocol three habitats are sampled namely stones, vegetation and sand/gravel/mud. Due to the lack of biotope variation in wetlands, it was decided that the vegetation biotope was the most suitable biotope to sample, as it contains a larger diversity of organisms. Aquatic macro-invertebrates were sampled using a 30 cm by 30 cm 1000 micron nylon mesh net. Approximately five meters of marginal vegetation, submerged vegetation or a mixture of both was sampled, by sweeping the net through the vegetation to brush and macro-invertebrates into the net. The invertebrates were then transferred into a photographic tray or similar sort of vessel for on site identification (Adapted from Dickens and Graham, 2002). The organisms were identified and recorded on a score sheet. Identification of organisms was aided using Aquatic invertebrates of South African Rivers: Illustrations (Gerber and Gabriel, 2002^b). Figure 9.2 illustrates the score sheet used during the data collection phase of the protocol.

The sampler will need the following equipment which is relatively inexpensive and easily obtainable: Waders*, Sampling Net, A3 photo tray or similar container, sample bottles*, forceps*, preservation solution*, data sheets and Aquatic Macro-invertebrate identification field guide. The items marked with an asterisk are optional. Samples can be identified either in the laboratory or in the field.

Sample sites (8) were selected along the entire course of the Nyl River and the Nyl floodplain. Three different biotopes/habitats were sampled depending on their availability namely: stones-in-current (riffles)/bedrock runs, marginal vegetation and sediments (mud, sand and gravel). Benthic macro invertebrates were collected from these selected biotopes (habitats). In stones-in-current (SIC); the net was placed directly on the riverbed and the area immediately upstream disturbed by kicking the surrounding stones and dislodging the benthic invertebrates. Marginal and fringing vegetation (MV) were sampled by sweeping the net back and forth through the biotope. Sediments were sampled by stirring the bottom sediments and sweeping the net through the disturbed area, collecting dislodged organisms. The SASS 5 scores obtained from the surveys are presented in the Tables 5.1-5.6.

5.3 Results:

Table 5.1: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the August 2001 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of Taxa	Combined SASS5 Score	Combined ASPT
Abba	MV	22	124	5.6	-	-	-
Donkerpoort	S	10	66	6.6			
	MV	14	76	5.4	22	112	5.1
	GSM	9	49	5.4			
Dladla	MV	8	47	5.9			
	GSM	10	45	4.5	17	81	4.8
Jasper	MV	12	52	4.3			
	GSM	4	14	3.5	14	61	4.4
Olifanstspruit	S	14	86	6.1			
	MV	14	83	5.9	22	130	5.9
	GSM	7	36	5.1			
Nylsvley	MV	11	50	4.5			
	GSM	3	7	2.3	13	64	4.9
Haakdoring	MV	17	82	4.8	-	-	-
Moorddrift	MV	8	39	4.9	-	-	-

SIC= Stones-in-current biotope, MV= Marginal vegetation

GSM= Sediments, (gravel, sand and mud), ASPT= Average Score per Taxon

Table 5.2: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the November 2001 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of Taxa	Combined SASS5 Score	Combined ASPT
Abba	MV	22	124	5.6	-	-	-
Donkerpoort	MV	82	12	6.8			
	GSM	3	13	4.3	13	68	5.2
Dladla	MV	13	73	5.6			
	GSM	4	19	4.8	15	84	5.6
Jasper	MV	12	52	4.3			
	GSM	4	14	3.5	14	61	4.4
Olifanstspruit	MV	15	87	5.8			
	GSM	9	53	5.9	20	102	5.1
Nylsvley	MV	17	70	4.1	-	-	-
Haakdoring	MV	6	25	4.2			
	GSM	4	12	3.0	7	26	3.7
Moorddrift	MV	12	55	4.6			
	GSM	10	38	3.8	17	74	4.4

Table 5.3: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the April 2002 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of Taxa	Combined SASS5 Score	Combined ASPT
Abba	MV	15	65	4.3			
	GSM	1	1	1	66	16	4.1
Donkerpoort	S	10	57	5.7			
	MV	15	84	5.6	105	19	5.5
Dladla	MV	10	60	6.0			
	GSM	7	33	4.7	14	77	5.3
Jasper	MV	13	67	5.2			
	GSM	7	33	4.7	16	78	4.9
Olifanstspruit	MV	13	82	6.3			
	GSM	10	58	5.8	18	98	5.4
Nylsvley	MV	9	42	4.7			
	GSM	3	14	4.7	11	52	4.7
Haakdoring	MV	73	16	4.6	-	-	-
Moorddrift	MV	16	78	4.9	-	-	-

SIC= Stones-in-current biotope, MV= Marginal vegetation

GSM= Sediments, (gravel, sand and mud), ASPT= Average Score per Taxon

Table 5.4: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the July 2002 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of Taxa	Combined SASS5 Score	Combined ASPT
Abba	MV	11	61	5.6			
Donkerpoort	S	10	57	5.7			
	MV	15	84	5.6	105	19	5.5
Dladla	MV	8	37	4.6			
	GSM	7	38	5.4	11	58	5.3
Jasper	MV	5	20	4.0			
	GSM	7	29	4.1	10	46	4.6
Olifanstspruit	S	13	83	6.4			
	MV	10	54	5.4	22	134	6.1
Nylsvley	MV	12	51	4.3			
	GSM	3	6	2.0	12	57	4.8
Haakdoring	MV	15	15	4.3	-	-	-
Moorddrift	MV	14	66	4.7	-	-	-

SIC= Stones-in-current biotope, MV= Marginal vegetation

GSM= Sediments, (gravel, sand and mud), ASPT= Average Score per Taxon

Table 5.5: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the November 2002 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of Taxa	Combined SASS5 Score	Combined ASPT
Abba	S	6	24	4.0	-	-	-
	MV	14	56	4.0	15	57	3.8
Donkerpoort	S	7	30	4.3			
	MV	11	48	4.4	14	59	4.2
Dladla	MV	8	38	4.8			
	GSM	6	21	4.4	12	53	4.4
Jasper	MV	11	45	4.1	-	-	-
Olifanstspruit	S	10	50	5.0			
	MV	11	63	5.7	24	145	6.0
	GSM	14	96	6.9			
Nylsvley	MV	10	48	4.8	-	-	-
Haakdoring	MV	13	57	4.4	-	-	-
Moorddrift	S	9	35	3.9			
	MV	8	25	3.1	11	39	3.5
	GSM	12	45	3.8			

Table 5.6: SASS5 Scores, number of taxa sampled and Average Score Per Taxon (ASPT) for the April 2003 survey.

Sample Site	Biotope Sampled	Number of Taxa	SASS5 Score	ASPT	Combined Total No of taxa	Combined SASS5 Score	Combined ASPT
Abba	MV	15	75	5.0	-	-	-
Donkerpoort	S	5	22	4.4			
	MV	11	54	4.9	15	73	4.9
	GSM	1	8	8.0			
Dladla	MV	15	90	6.0	-	-	-
Jasper	S	6	21	3.5			
	MV	9	37	4.1	13	55	4.2
	GSM	4	15	3.8			
Olifanstspruit	S	13	82	6.3			
	MV	8	46	5.8	18	115	6.4
Nylsvley	MV	16	72	4.5	-	-	-
Haakdoring	MV	17	82	4.8	-	-	-
Moorddrift	MV	11	39	3.9			

SIC= Stones-in-current biotope, **MV**= Marginal vegetation

GSM= Sediments, (gravel, sand and mud), **ASPT**= Average Score per Taxon

Table 5.7: Categories used to classify SASS4 and ASPT values (Thirion *et al.*, 1995).

Total Score	ASPT	Integrity Class	Conditions
>140	>7	A	Excellent
100-140	5-7	B	Good
60-100	3-5	C	Fair
30-60	2-3	D	Poor
<30	<2	E	Very Poor

Table 5.8: Macroinvertebrate taxa collected from the stones-in-current (SIC) biotope.

Taxa	Family	Average Sample Abundance
Planarians	Turbellaria	A
Oligochaeta	Annelida	A
Atyidae	Crustacea	B
Potamonautidae	Crustacea	A
Hydracarina	Hydracarina	B
Beatidae > 2sp	Ephemoptera	C
Caenidae	Ephemoptera	B
Heptagenidae	Ephemoptera	B
Leptophlebiidae	Ephemoptera	B
Chlorocyphidae	Odonata	B
Gomphidae	Odonata	B
Aeshnidae	Odonata	B
Libellulidae	Odonata	B
Cordulidae	Odonata	C
Corixidae	Hemiptera	C
Gerridae	Hemiptera	C
Vellidae	Hemiptera	B
Hydrosychidae 2sp	Trichoptera	B
Philopotamatidae	Trichoptera	B
Dytiscidae	Coleoptera	B
Gyrinidae	Coleoptera	B
Ceratopogonidae	Diptera	B
Chironomidae	Diptera	C
Culicidae	Diptera	B
Tabanidae	Diptera	B
Simuliidae	Diptera	B
Planorbidae	Molluscs	B

Table 5.9: Macroinvertebrate taxa collected from the marginal vegetation (MV) biotope.

Taxa	Family	Average Sample Abundance
Planarians	Turbellaria	A
Oligochaeta	Annelida	A
Hirudinea	Annelida	A
Atyidae	Crustacea	B
Potamonautidae	Crustacea	A
Hydracarina	Hydracarina	B
Beatidae > 2sp	Ephemoptera	C
Caenidae	Ephemoptera	B
Leptophlebiidae	Ephemoptera	B
Chlorolestidae	Ephemoptera	B
Coenagriidae	Odonata	B
Gomphidae	Odonata	B
Aeshnidae	Odonata	B
Libellulidae	Odonata	B
Cordulidae	Odonata	B
Notonectidae	Hemiptera	B
Corixidae	Hemiptera	C
Gerridae	Hemiptera	C
Naucoridae	Hemiptera	B
Nepidae	Hemiptera	A
Belostomatidae	Hemiptera	B
Vellidae	Hemiptera	B
Leptoceridae	Trichoptera	B
Dytiscidae	Coleoptera	B
Elmidae	Coleoptera	B
Gyrinidae	Coleoptera	B
Haliplidae	Coleoptera	A
Hydrophilidae	Coleoptera	B
Ceratopogonidae	Diptera	B
Chironomidae	Diptera	C
Culicidae	Diptera	B
Tipulidae	Diptera	B
Tabanidae	Diptera	B
Simuliidae	Diptera	B
Lymnaeidae	Molluscs	B
Planorbidae	Molluscs	B
Physidae	Mollusca	B

Table 5.10: Macroinvertebrate taxa collected from the soft-sediment biotope (Gravel, Sand and Mud).

Taxa	Family	Average Sample Abundance
Oligochaeta	Annelida	A
Hirudinea	Annelida	A
Potamonautidae	Crustacea	A
Hydracarina	Hydracarina	B
Beatidae > 2sp	Ephemoptera	B
Caenidae	Ephemoptera	B
Leptophlebiidae	Ephemoptera	A
Chlorocyphidae	Odonata	A
Gomphidae	Odonata	A
Aeshnidae	Odonata	A
Libellulidae	Odonata	A
Cordulidae	Odonata	A
Corixidae	Hemiptera	B
Gerridae	Hemiptera	B
Vellidae	Hemiptera	B
Hydropsychidae	Trichoptera	A
Hydroptilidae	Trichoptera	A
Philopotamatidae	Trichoptera	A
Dytiscidae	Coleoptera	A
Gyrinidae	Coleoptera	A
Ceratopogonidae	Diptera	B
Chironomidae	Diptera	B
Culicidae	Diptera	B
Tipulidae	Diptera	A
Tabanidae	Diptera	A
Athericade	Diptera	A
Physidae	Mollusca	A

5.4 Discussion, conclusions and recommendations:

Water quality may be assessed by examining the species present, the species richness or diversity (i.e. the number of species recorded at each site), abundance (number of individuals) and community structure (the relationship between organisms at different levels in the food chain). Sites such as Oilifantspruit with high species diversity can normally be considered healthy, although the ostrich feedlot higher up in the system is a major source of organic contamination. Severe organic pollution such as at the Jasper site causes depletion of oxygen in the water and the majority of non-air breathing macroinvertebrates are largely eliminated except for species such as oligochaetes (worms) and chironomids (midges), which are tolerant to low oxygen levels. In less severe organic pollution sites such as Abba and Donkerpoort Dam the macroinvertebrate diversity has been reduced with the abundance of the more tolerant organisms.

Human activities within the Nyl catchment or within the Groot and Klein Nyl Rivers itself can significantly alter the characteristics of the associated invertebrate communities. Changes in the sediment load, clearance of riparian vegetation, invasion of alien vegetation, modification of river structure and hydrological regime, and increases in nutrient and

effluent input will all have an impact on the macroinvertebrate community structure. Suspended solids reduce the light penetration and therefore limit photosynthesis and affect the invertebrate composition. Sediments deposited on the river-bed can smother bottom-dwelling communities and alter the biotope suitability. Riparian vegetation supplies the majority of food in the form of organic material (leaves, bark etc.). The removal of this energy source will not only directly affect invertebrates that depend on this food source, but will also increase the amount of light penetration in areas previously shaded by overhanging vegetation. Loss of shade results in an increased algal production which smothers certain biotopes such as stones-in-current and limits macroinvertebrate communities (increase in grazers). Rises in nutrient levels from the catchment run-off (feed lots, sewage farms, septic tanks) also increases the algal production. Increase solar radiation may also raise surface water temperatures affecting certain organisms. Barriers such as farm dams and weirs alter the natural flow regime, temperatures and water chemistry. They may obstruct invertebrate drift, or movement downstream, which may affect any possible recolonisation. Agricultural practices such as monoculture crop planting (ploughing and tilling soil) and overgrazing by livestock result in increased levels of erosion and siltation; resulting in increased levels of turbidity or sediment input into the river. Sewage and agricultural products (pesticides and herbicides) contain many components, including toxic substances, which are deleterious to certain macroinvertebrates.

It should be however stressed that SASS was designed as a rapid biomonitoring tool for the assessment of water quality in flowing rivers and only allow basic comparisons of invertebrate communities present at sites. The method is sensitive primarily to organic pollution (Chutter 1994). Its relationship to other types of pollution cannot be inferred without further development and testing of the method. SASS cannot be used to establish the precise nature or cause of the impairment. For more in-depth studies, more detailed investigate research is required. This involves detailed surveys of the invertebrates, with identification to specific level, and an understanding of their ecology. Although flowing rivers with a depositing substrate (stones-in-current) support a characteristic rich macroinvertebrate fauna, such biotopes are not always available in wetland habitats. The lack of this important biotope may severely restrict the available benthic fauna. The marginal and emergent vegetation is the most important biotope in the majority of wetland sites. The availability of marginal vegetation will fluctuate seasonally, reducing in availability during periods of low flow or during dry seasons. It is therefore necessary to investigate alternative sampling techniques and biotopes independent of the availability of natural substrates.

The use of artificial substrates or colonisation samplers should be investigated particularly in the wetland sites. Artificial substrates may provide a valid alternative sampling method for macroinvertebrates and the possibility of standardising the sampling effort in wetland habitats. Artificial substrates have been used in sampling surface and hyporheic macroinvertebrates. The artificial substrates can be used whatever the depth, the nature of the stream/wetland bed and the flow rate. More organisms (and often more species) are usually collected than the classical methods (SASS5). A preliminary wetland assessment protocol using macroinvertebrates found in the marginal and aquatic vegetation biotope and the habitat quality and surrounding land usage can be found in Chapter 9.

5.4 References:

- CHUTTER, F.M. 1995. *Research on the rapid assessment of water quality impacts on streams and rivers*. Final report to Division of Water Technology, CSIR. Pretoria.
- DICKENS, D.W.S. AND GRAHAM, P.M. 2002. The South African Scoring System (SASS) version 5 Rapid Bioassessment Method for Rivers. *African Journal of Aquatic Science* 27: 1-10.
- GERBER, A. AND GABRIEL, M.J.M. 2002 ^a. *Aquatic Invertebrates of South African Rivers: Field Guide (1st ed.)*. Institute for Water Quality Studies. Department of Water Affairs and Forestry, Pretoria. pp 150.
- GERBER, A. AND GABRIEL, M.J.M. 2002 ^b. *Aquatic Invertebrates of South African Rivers: Illustrations (1st ed.)*. Institute for Water Quality Studies. Department of Water Affairs and Forestry, Pretoria.
- KARR, J.R. AND CHU, E.W. 1997. *Biological Monitoring and Assessment: Using Multimetric Indexes Effectively*. EPA 235-R97-001. University of Washington, Seattle.

CHAPTER 6: AMPHIBIAN ASPECT**6.1 Introduction:**

Conservation efforts to protect the planet's vertebrate diversity have been disproportionate for the various groups and have tended to favour mammals and birds. The so-called 'lower vertebrates' such as fish, amphibians and reptiles; generally have a lower public appeal and are typically neglected in conservation programmes, yet these groups are of fundamental importance at an ecosystem level. In terms of species richness, amphibians outnumber mammals with more than 4700 living species currently recognised and with an expected total exceeding 5000 (Glaw and Kohler, 1998). Ironically, at a time when taxonomists are unravelling and describing this richness at an unprecedented rate, alarming reports of amphibian population declines and species extinctions are being recorded around the world. Amphibians are proportionally the most threatened group of vertebrates (Branch 1988).

With the world's human population more than doubling during the second half of the 20th century to reach six billion in October 1999 (Brown *et al.*, 1999), a concurrent increase in the rate of habitat loss and species extinction has become the greatest conservation concern. Biologists and conservation authorities realise that strategies geared towards reducing the risk and rate of extinction need to be implemented to ensure viable ecosystem functioning in the long term. These strategies can be at a global as well as a regional or national level, and include:

- Habitat preservation
- Intensified legislation and regulation
- Additional field research
- Investigations into the ecological roles of key/ flagship species
- Development of improved biomonitoring techniques especially concerning frogs and tadpoles as indicators of ecological status of wetlands
- Specific managed populations or even captive breeding programmes for potential restocking of wild populations

Amphibians are an important component of South Africa's exceptional biodiversity (Siegfried, 1989) and are such worthy of both research and conservation effort. Amphibian populations are declining throughout the world (Wyman, 1990; Wake, 1991; Kiesecker *et al.*, 2001; Blaustein and Kiesecker, 2002). It has become clear that declines cannot be attributed to any single causative factor and those complex mechanisms involving abiotic and biotic interactions are responsible for this phenomenon (Blaustein and Kiesecker, 2002). These declines have been attributed to a combination of factors, including climate change, chemical pollution, habitat loss and disease (Blaustein *et al.*, 2003).

Evidence for a countrywide decline in frog populations in South Africa is lacking (Channing and Van Dijk, 1995). Amphibian declines in southern Africa have been observed, but only at the local population level, and are usually confined to areas directly impacted upon by relevant threats. Among many threats faced by amphibians in southern Africa, the most frequently implicated is habitat destruction resulting from wetland drainage, afforestation, crop farming, invasive alien vegetation and urbanisation (Harrison *et al.*, 2001). Like other animals, amphibians fall prey to viruses and fungi, and to parasitic infections by protozoans and various helminths.

Three of the most commonly implicated diseases of amphibians associated with recent population declines are chytridiomycosis, saprolegniosis and *Ranavirus* infections (Cunningham *et al.*, 1996; Berger *et al.*, 1998). Chytridiomycosis is caused by a single-

celled fungus, *Batrachochytrium dendrobatidis*, and has the strongest association with amphibian population declines (Daszak *et al.*, 2003). The emergence of an infectious disease such as chytridiomycosis is usually linked to several factors and may be triggered off by environmental change. The chytrid fungus has been identified in a number of South African Frogs, particularly *Xenopus laevis* (Speare, 2000; Weldon, 2002).

Most frogs are intimately associated with wetlands. Much of southern Africa is semi-arid with seasonal and localised standing water and relatively few permanent wetlands. With a burgeoning human population and its consequent demands on limited water resources, more than one-third of South Africa's wetlands have been destroyed (Breen and Begg, 1989). Those that remain are increasingly threatened by water abstraction and pollution (Begg, 1990). A reduction in water quality may arise from direct and indirect contamination. This may include direct chemical contamination, or secondary run-off of petroleum and rubber compounds from roads, agricultural pesticides and herbicides, acid precipitation from atmospheric pollution, and eutrophication from fertilizer run-off. Owing to widespread pesticide use, increasing numbers of non-target species are exposed to chemical contamination. Amphibian populations are particularly susceptible to such contamination and causal relationships between pesticide usage and amphibian declines are of increasing concern (Boone *et al.*, 2001; Sparling *et al.*, 2001).

The extent and impacts of pollution on South African frogs have not been addressed, even though pollution and pesticides pose serious threats. Langton (2002) noted high levels of cadmium and copper in frog carcasses in the United Kingdom, and the copper (as copper thalocyanide) was linked to the use of slug pellets in suburban gardens. Channing (1998) discussed the toxicity of many pesticides to tadpoles inhabiting streams and rivers, but gave no examples of deaths resulting from their use in the wild. Herbicide and endocrine-disrupting contaminants have also been showed to cause amphibian abnormalities and mortalities (Pickford and Morris, 1999; Saka, 1999; Bettaso *et al.*, 2002). Frog mortalities and deformities can arise secondarily via synergistic links between trematode infections and pesticide exposures (Kiesecker, 2002).

Wetland habitats are biologically diverse and productive areas that support numerous species of both aquatic and terrestrial organisms and are critical habitats. Frogs appear to be particularly vulnerable to population losses. Most South African frogs are terrestrial, with an aquatic larval stage associated with a freshwater system. The majority of frogs use wetlands for breeding, and many are found in or near bodies of water outside the breeding season. In South Africa 88 of the 105 (84%) described species use wetland habitats. In South Africa, 19 frog species are permanent residents of wetlands or surrounding areas, 60 use wetlands for breeding and feeding during the rainy season, and 17 don't use wetlands (Channing and Van Dijk, 1995). Destruction of essential aquatic breeding habitats negatively impacts on certain frog species and may act synergistically with factors such as habitat deterioration and fragmentation resulting in population declines.

Most frogs have a biphasic life cycle, where eggs laid in water develop into tadpoles and these live in the water until they metamorphose into juvenile frogs living on the land. This fact, coupled with being covered by a semi-permeable skin makes frogs particularly vulnerable to pollutants and other environmental stresses. Consequently frogs are useful environmental bio-monitors (bio-indicators) and may act as an early warning system for the quality of the environment. Frogs are recognised as important indicators of the health of aquatic systems (Boyer and Grue, 1995; Bunn, 1995).

Toxicants can affect frog populations by:

- Affecting disease susceptibility

-
- Retarding growth
 - Affecting escape behaviour
 - Affecting reproductive development
 - Directly increasing mortality (Carey and Bryant, 1995).

A comparative toxicity data-base for amphibians has been suggested as a means of assessing toxicity risks (Linder *et al.*, 1990).

One of the most difficult problems in conservation biology is the lack of baseline data against which to measure population changes. The result has been that much of the information concerning amphibian declines is anecdotal; this is especially pertinent for South Africa. In some instances, especially in the case of disappearances, the anecdotal information is useful, but all ecologists are aware that populations undergo fluctuations in size under ordinary circumstances. Accordingly, a decline over a period of two or three years might be more than offset by a single year of successful recruitment. Those concerned with biodiversity, in general, have long recognised the need to establish long term studies, using standardised methods for natural populations. There is a growing interest in species and groups that might serve as indicators of the state of health of the environment. Many different taxa could be useful indicators, and amphibians have received considerable attention because of a combination of biological attributes:

- Permeable skin that acts in respiration and osmoregulation
- Biphasic life cycle with aquatic and terrestrial phases
- Feeding shifts in many species from herbivorous diets as larvae to carnivorous diets as adults
- Exposed developmental biology offering ease of investigation
- Amphibians are abundant and functionally important in most freshwater and terrestrial habitats in tropical, subtropical and temperate regions.

6.2 Tadpoles:

Tadpoles are the ephemeral, feeding, non-reproductive larvae in the life cycle of anuran amphibians. In many ways, tadpoles are the only vertebrate analogues to the larvae of aquatic macroinvertebrates e.g. certain insect orders (Trichoptera; Odonata; Diptera and others). The complex life cycle of frogs, allows the larval and adult stages to occupy entirely different ecological settings. This is a situation unique among tetrapods (Altig and McDiarmid, 1999).

Tadpoles regulate their metabolic state by physiological and behavioural means. Among anuran larvae, important biochemical -and physiological processes known to change as a function of the abiotic environment are: differentiation rate; growth rate; body size at metamorphosis; mechanisms of gas exchange and metabolic rate. The ionic composition of the plasma of tadpoles is markedly hyper-osmotic and hyper-ionic relative to the surrounding environment. These osmotic and ionic gradients are the driving force for the continual osmotic uptake of water and diffusive leak of plasma ions. Water may be taken up across the gills, skin or gut. As with freshwater fishes, tadpoles remain in positive ionic balance, by excreting copious quantities of dilute urine and by actively transporting ions across external epithelia. These two physiologically mediated processes are subject to the effects of temperature, salinity and pH (Altig and McDiarmid, 1999).

Of all the physical parameters in the aquatic environment, temperature is perhaps the most dramatic in its effect on the physiology, ecology, and behaviour of anuran larvae.

Environmental temperature affects the time taken to metamorphoses, differentiation and growth rates, body size at metamorphoses, mechanisms of gas exchange, and rates of energy metabolism. The thermal environment of all anuran larvae is variable in space and time, often extremely so in shallow water exposed to intense isolation. Anuran larvae have generally accommodated to variation in the thermal environment by:

- Possessing the physiological ability to function over a wide range in body temperature,
- Behaviourally regulating body temperature,
- Physiologically adjusting to changes in environmental temperature (i.e. acclimatization)

Tadpoles regulate body temperature behaviourally by moving or changing posture among available thermal microenvironments. Behaviourally mediated temperature regulation allows for maximization or control of differentiation and growth rates (Bradford and Freda, 1999).

Competition, environmental temperature, and other aspects of environmental quality such as DO and pH may influence growth rates. The influence of temperature on differentiation and growth rates probably is the most dramatic effect of environmental temperature on the ecological physiology of anuran larvae. This influence determines larval body size and length of the larval period, which in turn are directly related to survival and reproductive success. Typically, an increase in temperature result in faster rates of development and growth, a shorter time to metamorphosis and smaller body size at metamorphosis. Competition could be of several types, including intra- and inter-specific competition, competition with other vertebrates and members of other phyla e.g. macroinvertebrates. The mechanism of competition could be exploitation or interference. Interference competition may be behavioural or allelopathic, which could be caused by chemicals, parasites, or pathogens. Rates of development may respond directly to environmental influences such as dissolved oxygen (DO), pH, and temperature. Survival is also dependent on biotic and abiotic environmental factors – the composition of the assemblage of potential predators and the probability of environmental changes (Altig and McDiarmid, 1999).

6.3 Habitat selection:

Tadpoles have little control over the general habitat type (lotic or lentic, permanent or temporary) they occupy. This is determined by the choice of spawning sites by adults, which determines the range of species with which tadpoles may potentially interact. The timing of frog reproduction determines the temporal distribution of tadpoles within sites. This affects the interactions each cohort of tadpoles experience with other cohorts of the same species, with other species of anurans, and with other vertebrates and invertebrates. Reproductive site choice by frogs, and thus possible tadpole community composition, is influenced by a complex interaction between environmental quality, environmental uncertainty, life history, predation risk, and resource availability.

The general type of reproductive habitat chosen has a strong influence on the entire developmental strategy followed by many species. Most anuran larvae inhabit temporary habitats that range from small pools to larger pans situated in lower lying areas or depressions. Unpredictable temporal and spatial distributions and cyclic patterns of nutrient availability are common features of these habitats. Others develop in more complex permanent aquatic habitats as temporary invaders in established communities such as rivers, streams and dams. Numerous physical (e.g. distance from shore, oxygen concentration, substrate qualities, water depth and flow rate, site duration, and temperature) and biological (e.g. presence and distribution of vegetation, other tadpoles, other organisms including predators, and the phenology of all organisms) factors influence the spatial and temporal distribution of tadpoles among microhabitats.

Tadpoles are thus likely to encounter a variety of intra- and interspecific competitors, other types of herbivores, and predators. Predation begins at the egg stage and continues through metamorphosis. The presence of potential predators has been suggested as a limiting factor in habitat use by many anurans (Bradford, 1989; Kats *et al.*, 1988; Sexton and Phillips, 1986; Woodward, 1983). Tadpoles are vulnerable to a wide range of invertebrate and vertebrate predators. Their vulnerability can be influenced by tadpole behaviour, body size, coloration, genotype, habitat preferences, and palatability. The influence of body size on tadpole vulnerability to predation has received considerable attention. In general, mechanical limitations should cause most predators to prefer prey within a limited range of body sizes. This is less true for some predators, such as odonata nymphs, than for gape limited predators like fish. Shifts in microhabitat use are one method some tadpoles may use to decrease predation rates (Morin, 1986).

6.4 Tadpoles and pollutants:

Certain tadpoles are permanent or long-term residents in water-bodies and can be monitored quickly and economically. Different pollutants affect different systems in tadpoles. This includes substances affecting red blood cell number, neuronal activity, cell division, fertilization and a disruption in the normal biochemical pathways that lead to growth. Caged tadpoles of *Rana temporaria* have been used to monitor pesticide run-off or spray drift. They show mortality, deformities, reduced rates of metamorphosis and reduced growth (Cooke, 1981). Tadpoles and eggs have also been transplanted to ponds as an experimental monitoring procedure (Freda and McDonald, 1993). Tadpoles have been used to assay for metal pollution, as they accumulate metals (Sparling and Lowe, 1996). The FETAX (Frog Embryo Teratogenesis Assay- *Xenopus*) has been positively evaluated as a short-term test for developmental effects (Sabourin and Faulk, 1987). Biomonitoring is a widely used component of ecosystem management. Tadpoles are present in the majority of wetland habitats during the rainy season when run-off and spray drift can be expected to contaminate the waterbodies. Tadpoles could provide a low cost alternative to expensive water chemical analyses especially for routine surveys and long-term monitoring projects.

Southern African tadpoles are still not completely described or known. The first comprehensive synthesis was by Van Dijk (1966), who provided characters, keys and illustrations to many tadpoles. Other description of African tadpoles includes the works of Van Dijk (1971), Channing (1998), Channing (2001) and Lambiris (1987, 1989).

6.5 Study area:

The veld type of Nylsvley (24°29' S; 28°42' E) falls within the vegetation unit classified as Mixed Bushveld (Veld Type 18, Low and Rebelo, 1998, (A18) Acocks, 1988) forming part of the Savanna Biome. This bushveld represents a great variety of plant communities, with many variations and transitions. The elevation of the area varies between 1 080m and 1 140 m above sea level with the seasonal Nyl river forming a floodplain from the South West to the North East. On account of the low drainage elevation from the one end of the Reserve to the other (7m), extensive flooding results during the rainy season. Prominent hills are Maroelakop (1 140m) in the south and Stemmerskop (1 090m) further to the northwest. The southern elevations are mainly underlain by sandstone and conglomerate bands of the Waterberg system, while the northern elevations are underlain by felsites of the Bushveld Igneous Complex. Several soil series occur associated with differences in geology and topography. The higher lying areas have relatively sandy soils and carry mostly a broad-leaved deciduous savanna as opposed to the lower lying areas (lowlands), where the soils are largely calcareous clay with microphyllous deciduous thorn savanna. Both areas exhibit a marked seasonality with regard to the grass layer. Termitaria are a feature of the northern elevations of the felsites, as well as along the edge of the Nyl floodplain. The area experiences a moderately low summer rainfall with a dry winter period. The climate of the

area is semi-arid with distinct seasons; a hot, wet season from October to April, followed by a cool, dry season from May to September. The mean annual rainfall is 630 mm and the mean annual temperature is 18,6° C.

The summer months are characterised by hot days and warm nights whilst during winter the daytime temperatures are moderate and cold at night. Ground frosts are experienced nightly in the lower lying areas of the Reserve but are relatively mild. The reserve was previously utilised for cattle ranching and was conserved as far as possible. Small areas of the lowlands were previously ploughed as well as certain disturbed areas (kraal sites) around previous settlements (Jacobsen, 1982).

6.6 Methods:

The survey entailed intensive monitoring of selected localities from the beginning of February 2001 until the end of March 2003. Four survey methods were used during this amphibian survey to obtain the species inventory (Table 6.1). These included visual encounter surveys (VES) of the terrestrial and aquatic habitats, road surveys for live and road-killed animals, anuran call surveys at potential breeding habitats and dip-netting for tadpoles in various aquatic habitats. This included nocturnal and diurnal surveys conducted after a sufficient rainfall period. Nocturnal surveys were conducted of selected habitats to determine the number of adult male frogs calling. Daytime surveys were conducted on known and potential breeding sites searching for evidence of any previous breeding activities (eggs, tadpoles).

6.7 Results:

Table 6.1: List of frog species recorded at Nylsvley Nature Reserve during the base-line amphibian survey between 2001-2004.

Common Name	Scientific Name	Status and Distribution	Habitat
Giant Bullfrog	<i>Pyxicephalus adspersus</i>	Limited records of adults and juveniles at Nylsvley.	Temporary shallow depressions and floodplain areas of the vlei. Also requires undisturbed open veld for foraging and sandy soils for burrowing.
Tremolo Sand Frog	<i>Tomopterna cryptotis</i>	Abundant throughout Nylsvley	Temporary rain pools, pans and floodplain areas of Nylsvley.
Natal Sand Frog	<i>Tomopterna natalensis</i>	Limited records during survey at Nylsvley.	Temporary and semi-permanent seepage and mud pools.
Knocking Sand Frog	<i>Tomopterna krugerensis</i>	Limited records during survey.	Seasonal mud pools, small ponds and floodplain areas of Nylsvley.
Common Caco	<i>Cacosternum boettgeri</i>	Limited records around Vogelfontein.	Temporary marshes, ditches and grass inundated to a depth of about 2cm.
Bubbling Kassina	<i>Kassina senegalensis</i>	Scattered records, especially around Vogelfontein.	Semi-permanent vleis, pans and shallows around dams.
Red Toad	<i>Schismaderma carens</i>	Limited numbers of adults were observed. No tadpoles recorded.	Semi-permanent dams & ponds with water depth of more than one metre.
Olive Toad	<i>Bufo garmani</i>	Most common toad species at Nylsvley.	Permanent or semi-permanent bodies of water, natural or man-made in open wooded savannah.
Guttural Toad	<i>Bufo gutturalis</i>	Common throughout Nylsvley.	Permanent and semi-permanent ponds and backwaters in open habitat.

Common Name	Scientific Name	Status and Distribution	Habitat
*Raucous Toad	<i>Bufo rangeri</i>	Not recorded during this survey. Historic records.	Permanent or semi-permanent bodies of water, natural or man-made.
Common Platanna	<i>Xenopus laevis</i>	Common throughout Nylsvley.	Permanent or semi-permanent bodies of water, natural or man-made.
Common River Frog	<i>Afrana angolensis</i>	Common throughout the Nyl system.	Present in Groot and Klein Nyl systems as well as permanent standing water and floodplain areas of Nylsvley.
Banded Rubber Frog	<i>Phrynomantis bifasciatus</i>	Limited records during survey period.	Shallow ponds or inundated grass in savanna and Acacia veld.
Snoring Puddle Frog	<i>Phrynobatrachus natalensis</i>	Rare throughout Nylsvley	Pools or marshy area associated with the vlei
*Striped Stream Frog	<i>Strongylopus fasciatus</i>	Not recorded during survey. Historic records.	Shallow pools (seasonal), inundated grassland areas of the vlei and dams.
Plain Grass Frog	<i>Ptychadena anchietae</i>	Rare, mainly recorded from Vogelfontein but scattered records throughout Nylsvley.	Shallow pools (seasonal), inundated grassland areas of the vlei and dams.
Bushveld Rain Frog	<i>Breviceps adpersus</i>	Abundant throughout Nylsvley	Eggs deposited in a subterranean chamber about 30cm below surface.

* Limited historic records (Jacobsen, 1982).

6.8 Discussion:

During the survey period (2001-2004) a total of fifteen (15) frog species were recorded for the Nylsvley Nature Reserve. Historic surveys conducted by Jacobsen between 1975 and 1977 recorded a total of 17 frog species (Jacobsen, 1983). Two frog species were not recorded during this survey being the Raucous Toad (*Bufo rangeri*) and Striped Grass Frog (*Strongylopus fasciatus*). The range of the Raucous Toad appears to be contracting in the north and east of South Africa, in Limpopo, Mpumalanga, Gauteng and coastal Kwazulu-Natal provinces (new atlas data). The apparent range contraction seems to complement the range expansion of the Guttural Toad (*Bufo gutturalis*) and it's possible that these are linked, that is, *B. gutturalis* displaces *B. rangeri* and/or habitat modification affects these species differently. *B. rangeri* hybridises with *B. gutturalis* at scattered sites and several authors have suggested that hybridisation is a historically recent phenomenon and that the creation of artificial breeding sites, such as farm dams, has broken down natural separation based on breeding habitat (Carruthers, 2001). *B. gutturalis* readily colonizes farm dams and reach high abundance, so it is possible that modern peri-urban and agricultural development has extended the potential distribution of this species at the expense of *B. rangeri*. However, the two species also co-occur and hybridise in natural situations, such as around slow flowing streams or stream-side pools (Cunningham, 2004).

The absence of the Striped-Stream Frog (*Strongylopus fasciatus*) is possibly due to the fact that they breed mainly in winter months, which were not sampled.

Several species recorded during the survey were only rarely encountered in the Nylsvley Nature Reserve. These included the Giant Bullfrog (*Pyxicephalus adpersus*), Plain Grass Frog (*Ptychadaena anchietae*), Banded Rubber Frog (*Phrynomantis bifasciatus*), Natal Sand Frog (*Tomopterna natalensis*), Knocking Sand Frog (*Tomopterna krugerensis*) and Red Toads (*Schismaderma careens*).

Jacobsen (1983) found that two species, *Bufo garmani* and *Kassina senegalensis*, made up 86,4% of species captured. If one includes the terrestrial breeder, *Breviceps adpersus* the three species make up 95% of all amphibians captured. During the survey period the most abundant frog species were *Bufo garmani*, *Bufo gutturalis* and *Tomopterna cryptotis*. *Kassina senegalensis* was present in low numbers (<50) throughout the survey period.

Several factors may be influencing the amphibian populations in the Nyl system and Nylsvley Nature Reserve and these are listed below.

6.9 Habitat destruction, fragmentation and degradation:

Habitat loss, in all its many forms is still the most pervasive threat facing amphibian populations in South Africa. Habitat loss in the Nyl system most commonly results from agricultural development, alien invasive plants, localised urban developments and associated infrastructure such as roads. The destruction and alteration (alien vegetation) of the majority of riparian vegetation along the Nyl floodplain system directly affect the frog species utilising this habitat, such as the Common River Frog (*Afrana angolensis*). The removal of natural vegetation and the invasion of alien vegetation result in increased erosion and consequent siltation of the system.

Agricultural developments include the construction of farm dams and the overgrazing and trampling of livestock. Farms dams although providing additional habitat to certain species such as the Common Plattana (*Xenopus laevis*), tend to reduce or completely eliminate the downstream flow of certain streams and rivers, reducing the suitability of riparian species. Overgrazing and trampling of sensitive wetland vegetation results in increased erosion and consequent increase siltation in water catchments. Domestic stock levels increase and larger herds overgraze the vegetation around natural water sources, leading to siltation and increased evaporation, which is detrimental for developing larvae. Agricultural practices such as the planting of monoculture crops destroy large areas of natural vegetation and the natural soil structures and result in increased levels of erosion and siltation.

Major road networks result in severe habitat fragmentation and increased road mortalities of migrating amphibians. Roads adjacent to wetland areas may cause unsuitable rates of mortality, particularly in the case of toads and Giant Bullfrogs. Populations can be decimated during migration to and dispersal from the breeding sites and this may lead to local extinction. Habitat loss and fragmentation can lead to genetic depletion in isolated populations and this may be reflected in lower larval fitness (Hitchings and Beebee, 1997).

The encroachment of invasive alien vegetation reduces the available groundwater levels and increases the risk of wildfires. The increased burden of woody material cause extremely hot fires, which are damaging to amphibian populations. Fire has a considerable impact on the vegetation as well as the fauna. Fires in the savannah usually occur during the winter months when the majority of amphibian species are dormant (hibernating) in holes and in logs. Frogs inhabiting logs lying on the ground are burned. Others using vegetation as refuge areas are also incinerated. The increased use of water by alien vegetation affects the duration of seasonal water bodies which have a major influence on the success rate of developing larvae. Desiccation of larvae is one of the major factors influencing explosive breeders' reproductive success. Seasonal ponds should ideally contain water for at least 30 days for larvae to successfully develop and metamorphose. Alien vegetation also directly destroys certain breeding sites by invading the habitat such as Poplars (*Populus deltoides*) and reeds.

A reduction in water quality of the Nyl floodplain system arises from direct and indirect contamination. This includes direct contamination, or secondary run-off petroleum and

rubber compounds from roads (especially N1), agricultural pesticides and herbicides and eutrophication from sewerage farm, septic tanks, feeding lots as well as fertilizer run-off.

Seasonal fluctuations in amphibian population sizes occur depending on certain environmental parameters such as amount of rainfall, temperature and humidity. Precipitation strongly influences amphibian activity, distribution and dispersion patterns, reproductive cycles, and rates of growth and development. Many species remain underground or in aboveground retreats except during wet periods. Therefore, the best time to survey an area is often during the wet season or following rain.

Rainfall at Nylsvley is extremely variable and concentrated between October and March. Rain falls most frequently in the form of heavy diurnal thunderstorms of relatively short duration. Considerable fluctuations in rainfall occur from year to year. Rainfall is important for amphibians in that it initiates activity and reproduction. Most of the amphibians at Nylsvley are fossorial, avoiding desiccation during the dry winter months. Certain species such as the Sand Frogs (*Tomopterna sp.*) and Giant Bullfrogs (*Pyxicephalus adspersus*) dig their own burrows whilst species such as the Bubbling Kassina (*Kassina senegalensis*) use the burrows of other animals, natural crevices or under rotting logs. Levels of amphibian activity are influenced by the intensity and duration of rainfall as well as temperature and humidity. Heavy summer downpours result in adult migrations to suitable breeding habitat and the initiation of breeding events. Following some form of courtship, adults of oviparous species deposit eggs in or near the water. Large numbers of eggs are deposited to ensure survival of certain tadpoles. The eggs and tadpoles are adapted to these ephemeral habitats and develop rapidly. The tadpoles are major consumers in aquatic environments. After a period of growth, the tadpoles undergo metamorphosis and move back to the terrestrial environment where they feed and continue to grow. When mature, they return to their specific aquatic environment or habitat to breed, completing the biphasic, complex life cycle.

Limited rainfall results in dehydration or desiccation of eggs and larvae and is thus, therefore, one of the major factors determining population sizes of amphibians. Larval mortality is extremely high in seasonal rainfall areas. Temperature and rainfall combine to influence the humidity of the environment. Temperature can be a limiting factor for amphibians; especially eggs and larvae, which are, exposed to extremely high water temperatures (>35°C) during the midday heat. Several Giant Bullfrog tadpoles were observed in a shallow pool with water temperatures exceeding 39°C. Extreme temperatures are a major limiting factor for denaturing amphibian eggs and desiccating tadpoles.

6.10 Proposed sampling techniques for amphibians:

Questions concerning amphibian biodiversity basically fall into two categories: (1) those related to habitats, sites, or areas and (2) those concerned with species assemblages. The primary goal of habitat or area-based questions is to inventory the species that occur in habitats or areas at a specific site. Species-based studies focus on one or more populations over an extended period of time. Several techniques are available for generating species lists or information on species richness for a site. For the most part, field techniques are methods of general collecting, as historically practiced by herpetologists. Typically, they involve searching for and collecting amphibians in all possible microhabitats both during the day and at night. Below a few general collecting techniques are discussed. These are adapted from Heyer *et al.*, 1994.

6.10.1 Surveys at breeding sites:

Many amphibians are most conspicuous at breeding ponds. Therefore, surveys conducted at breeding sites are especially effective. Sampling at the breeding site involves counting the

animals are some predetermined fashion. Generally, adults are counted along visual or aural transects. Data from surveys at breeding sites can be used to estimate species richness at one or several sites. Across-site comparisons are useful for identifying areas most suitable for development or preservation, studying the effects of pollution from point sources and determining the presence of predators. The techniques can also be used to monitor changes in population levels of species, to detect changes in species assemblages through time, or to carry out detailed autecological studies.

6.10.2 Target organisms and habitat:

The techniques described below can be adapted for the study of any amphibian that breeds in communal aggregations in temporary or permanent ponds, vleis, streams or rivers. Breeding site surveys can focus on adults or larvae. Adults are usually more conspicuous and easier to sample and identify than larvae. However, larvae are typically present at the breeding site for longer periods than adults. Sampling both adults and larvae is the best approach. Monitoring adults at a breeding site is easiest when breeding is concentrated in a narrow, well-defined period, but it can also be implemented when the breeding period is extended. In arid areas most breeding is rain-dependent, and developmental times of larvae are often short; surveys must be undertaken during suitable weather conditions, whenever they occur (Wells, 1977).

For short, infrequent surveys, larval sampling yields more complete species lists than adult surveys do. However, if there is any doubt as to larval identification the larvae should be reared through metamorphosis. Larval densities can be strongly influenced by local factors (e.g. climate and co-occurring predators) and can vary greatly over short periods (Woodward and Mitchell, 1991). Larval densities are not good predictors of adult population size. Breeding site studies are most thorough in small, shallow bodies of water that are free of emergent vegetation and that can be surveyed by observers in a relatively short period. The use of the Giant Bullfrog (*Pyxicephalus adspersus*) as a grassland and savannah “flagship species” should be further investigated.

6.10.3 Data treatment and interpretation:

The following information should be recorded by the observer include:

- Surface-water and deep-water temperatures.
- Presence or absence of calling sites (bushes, trees, floating, fringing or emergent vegetation).
- Reproductive activity of adult frogs (Calling, oviposition, etc.)
- Developmental stages of larvae.

The data from breeding-site surveys can be used to produce a list of frog species encountered, or they can be combined with other information to form a basis for detailed ecological and population analyses. Species lists can be compared across sites, although one should be cautious to attribute too much to species absences if only a few breeding sites are examined during a single season survey. Breeding-site surveys can be used to estimate effective population size and operational sex ratio (OSR), two parameters that are important for conservation work (Falconer, 1989). For these purposes, surveys must be made over an extended period because breeding populations vary widely from night to night at a single pond (Ryan, 1985).

6.11 Short-term sampling (sts):

Short collecting visits to a single site cannot give much insight into the total number of species present. However, using time-constrained collecting techniques, rates of species accumulation in different habitats or sites can be compared if amphibian populations are

similar. The first step in time-constrained, short-term sampling is to identify and define the major habitat types at the study site. All habitats should be sampled during the first few days of the sampling period. Information derived from this broad scale sampling can be used to plan how to distribute subsequent sampling among habitats. Many factors influence the efficiency of short-term surveys, and they must be recognized and controlled if comparisons are to be made among different sites and habitats. Some of these variables are:

- Total time spent on the survey
- Number and experience of fieldworkers
- Topography
- Area of the site
- Weather conditions and climate
- Season, date, and time of day
- Time required sampling each major habitat.

6.12 Assumptions:

The results obtained from short-term sampling are highly dependent on collecting and the environmental variables. Some of these variables include weather (both prior and during sampling), collector's experience, and level of sampling effort in each habitat, diversity of collecting techniques used, and phenology of the amphibian species. This is especially important when results from similar habitats are compared. Any effects of these variables must be recognised and controlled. Time constrained searches must standardise collecting effort within the selected habitat types.

6.13 Audio strip transects (AST):

In the vast majority of frog species, males in reproductive condition use distinctive species-specific calls to advertise their position to potential mates and rivals (Wells 1977). The audio strip transect (AST) technique exploits this species-specific behaviour. All calling frogs along a selected transect are counted. The width of the transect varies according to the detection distance of each species' advertisement call. The counts are then used to estimate or determine:

- Relative abundance of calling males
- Relative abundance of all adults
- Species composition
- Breeding habitat or microhabitat use
- Breeding phenology of species.

Counting calling male frogs or aggregations of calling males (choruses) along strip transects can be the most effective way to:

- Inventory species composition
- Provide a first approximation of relative abundance of breeding frogs
- Determine breeding habitat use
- Map distributions of most frog species throughout a large area.

Several factors may affect the accuracy of assessments of abundance and habitat occupancy based on audio strip transects (AST). For example, if counts of a species are not made during its peak-breeding period, when the maximum number of males is calling, differences between surveys may simply reflect differences in stage of breeding cycle. This is especially pertinent for certain frog species such as the Giant Bullfrogs

(*Pyxicephalus adspersus*), which may not emerge for several years to breed due to unfavourable environmental parameters such as insufficient rainfall. Observers must be fully knowledgeable of the species-specific frog calls, must have full hearing ability and be experienced in this sampling technique. Observers should ideally make audio recordings of all the species present in the choruses for historic records as well as confirmation of species identification.

6.13.1 Limitations:

The audio strip transect method has several limitations including:

- Number of calling males cannot be determined aurally for chorusing species or in situations of high call overlap such as *Cacosternum boettgeri* and *Tomopterna cryptotis*.
- Some reproductively active males of certain species don't call and others such as the Giant Bullfrog have a diurnal reproductive strategy.
- Explosively breeding frog species are acoustically evident for extreme short periods and are probably not sampled adequately. Certain male and female Giant Bullfrogs are only present for a few hours during their short-duration reproductive event.
- Absolute population size cannot be estimated, because male and female survivorships may not be related and operational sex ratios (males to females) may differ greatly.

6.14 Quantitative sampling of tadpoles:

Most tadpoles occur in aquatic habitats including lentic waters (streams and rivers) and lotic waters (ponds and dams). Tadpoles are often found in large concentrations at breeding sites over longer periods. As a result, sampling tadpoles rather than the adults may be a more efficient method for inventorying species at a site, even though eggs and larvae of many species are poorly known. In addition the collection of tadpole voucher specimens is easier and has less impact on the population than collecting adults.

There are various methods for sampling and identification of amphibian larvae (tadpoles) from water bodies. These methods include seining, dipnetting, trapping and enclosure sampling. These techniques provide a fast, relatively thorough, qualitative or quantitative sample with minimal personnel, material and time. In addition, the techniques generally do not harm the animals so can be used to monitor rare or endangered species. The two primary goals of these procedures are to assess the species richness of larvae in a body of water and determine larval population size.

Amphibian larvae occur in three basic habitat types: small bodies of water, ponds/dams and streams/rivers. Several points must be considered when sampling tadpoles. First, most tadpoles are medium to strong swimmers that can out swim a slow-moving net. Second, tadpoles commonly escape by hiding in the bottom substrate, making it essential to keep a seine or dipnet on the bottom when sampling. Third, vegetation and/or irregular bottom surfaces make any sampling difficult. Seine and frigid-frame samplers prove ineffective in bodies of water with abundant vegetation. Fourth, many tadpoles are microhabitat specialists, so all the biotopes should be sampled.

6.14.1 Equipment:

At one extreme, a small aquarium net (about 10cm wide) with a bendable frame is useful in capturing tadpoles from small, shallow pools. A slightly larger net serves well for general collecting in both rivers and ponds. Wire-mesh sieves (kitchen strainers) with a handle work well in densely vegetated areas or rocky areas. Net size and mesh size determine passage

rates through the water; some experience will be needed to find the net optimal for the body of water and tadpoles to be sampled. Fine mesh nets capture all larvae but are easily clogged with filamentous algae and debris. They are also cumbersome and move through the water relatively slowly and tear easily. Large mesh nets are easier to use but may miss small individuals. The use of several mesh size nets is recommended to ensure all sizes of tadpoles are caught. There are no definitive rules about the number of sweeps needed to sample a habitat adequately. It is not uncommon to cover almost all of the surface area in small ponds or pools whereas only a fraction of larger bodies of water are sampled, such as shorelines of dams and pans. Twenty to fifty sweeps can be made in an hour depending on how much vegetation and detritus must be removed from the net and how many larvae need to be identified. A reasonable procedure is to survey each habitat for an equal period or with an equal number of sweeps. Making more sweeps in the larger habitats increases the chance of encountering rare species. Increasing the number of sweeps also increases the chance of capturing highly habitat-specific species. Tadpoles are fragile often with delicate tails, which are easily damaged in nets. Trauma can be minimised by keeping tadpoles, cool, un-crowded and in the net for as little time as possible. Tadpoles should not be picked up with your hands from the net but collected in a glass beaker and placed into a larger bucket. Consideration should also be taken for the water body sampled. Major disturbances, such as massive sediment up-swelling by sweeping a net in small ponds may result in the death of all associated animals.

6.15 Proposed amphibian index (AI)

Amphibian communities inhabit a variety of different habitats. Certain species are completely terrestrial whilst others inhabit wetlands continuously, and hence possess a broad-based ability to integrate the effects of chemical, biological and physical attributes over extended periods of time. Structural and functional attributes of these amphibian communities could reflect and be used to monitor the cumulative effects of multiple stressors at the selected site. The proposed amphibian index should be based on categorisation of frog species according to an intolerance rating which reflects the species habitat specialisation or preference, breeding and larval requirements and sensitivity to habitat deterioration. Results of species observed can be compared with available historical lists or what species should have occurred in the absence of human impacts.

The potential use of amphibians as biological indicators is still in its infancy in Southern Africa. More intensive long-term amphibian population studies as well as intensive autecological studies need to be conducted. There is an extensive lack of accurate knowledge on the specific habitat requirements (migratory, foraging and breeding) of the majority of frog species. The tolerance levels or sensitivity to pollution of the majority of Southern African frog and tadpoles species is unknown. The use of *Xenopus* tadpoles may not be an accurate reflection of the sensitivity of other frog species to water contamination, or the effects of pesticides. *Xenopus* is widely known as a highly tolerant species often occurring in highly contaminated water bodies such as sewage purification works. Eutrophic waters seem to produce the highest densities. The possible use of *Afrana angolensis* (Common River Frog) tadpoles should be investigated for both laboratory analysis as well as captive caged tadpoles in field experiments. *Afrana* tadpoles are found throughout the majority of South Africa (entire Nyl system) throughout both the summer and winter months.

Long-term experiments using caged tadpoles in selected sites in specific drainage systems must still be conducted. A comparative toxicity data-base for South African amphibians needs to be compiled as a means of assessing the potential toxicity risks. More detailed amphibian surveys are required on specific wetlands and which specific habitats are utilised by which species in these areas. The development of an amphibian index should take into consideration the following aspects:

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- Presence or absence of frog and tadpoles species at a selected site.
 - Total number of observed frog species or species richness for the selected site versus what species should ideally occur in absence of human impacts.
 - Total number of frog species breeding at the site (evidence of amplexus, oviposition or tadpoles)
 - Presence of intolerant (sensitive) species: Sensitive species are usually the first to disappear following environmental disturbances.
 - The proportion of species tolerant of poor water quality conditions in the sample population. These species usually dominate or increase in abundance in degraded habitats.
 - The presence of abnormalities or malformations in adults and larvae (tadpoles).

6.16 Limitations:

There are certain prerequisites inherent to the use of amphibians as biological indicators and may limit its application in South Africa.

- Limited knowledge on the autecology of the majority of frog species. Including specific habitat requirements and species tolerance to environmental degradation.
- Relatively extensive training and experience are required for the sampling and identification of frogs and larvae (tadpoles).
- In certain parts of South Africa amphibian species diversity is naturally low.
- Amphibian species assemblages must be used in conjunction with a habitat assessment of the selected site.

6.17 References:

- ACOCKS, J.P.H. 1988. *Veld Types of South Africa. Memoirs of the Botanical Survey of South Africa*. 57: 1-146.
- BEGG, G.W. 1990. *Policy Proposals for the wetlands of Natal and Kwazul*. Natal Town and Regional Planning Commission. Report 75. Pietermaritzburg.
- BERGER, L., SPEARE, R., DSAZAK, P., GREEN, D.E., CUNNINGHAM, A.A., GOGGIN, C.L., SLOCOMBE, R., RAGAN, M.A., HYATT, A.D., MACDONALD, K.R., HINES, H.B., LIPS, K.R., MARANTELLI, G., AND PARKES, H. 1998. Chytridiomycosis causes amphibian mortality associated with population declines in the rain forests of Australia and Central America. *Proceedings of the National Academy of sciences of the United States of America* 95: 9031-9036.
- BETTASO, J.B.; WELSH, H.H., AND PALMER, B.D. 2002. Northern red-legged frogs and endocrine disrupting compounds. *Froglog* 52:1-2.
- BLAUSTEIN, A.R.; AND WAKE, D.B. 1995. The puzzle of declining amphibian populations. *Scientific American* 1995(April):56-61.
- BLAUSTEIN, A.R.; AND WAKE, D.B. 1990. Declining amphibian populations. A global phenomenon? *Trends in Ecology and Evolution* 5: 203-204.
- BLAUSTEIN, A.R., AND KIESECKER, J.M. 2002. Complexity in conservation: lessons from the global decline of amphibian populations. *Ecology Letters* 5:597-608.
- BLAUSTEIN, A.R., ROMANSIC, J.M.; KIESECKER, J.M., AND HATCH, A.C. 2003. Ultraviolet radiation, toxic chemicals and amphibian population declines. *Diversity and Distributions* 9: 123-140.
- BOONE, M.D., BRIDGES, C.M., AND MILLS, N.E. 2001. A hierarchical approach in studying the effects of an insecticide on amphibian communities. *Froglog* 48: 4-5.
- BOYER, R & GRUE, C.E. 1995. The need for water quality criteria for frogs. *Environmental Health Perspectives*. 103(4): 352-357.
- BRADFORD, D.F. 1989. Allopatric distribution of native frogs and introduced fishes in high Sierra Nevada lakes of California: Implications of the negative effect of fish introductions. *Copeia* 1989: 775-778.
- BRANCH, W.R. (ed) 1988. South Africa Red Data Book: Reptiles and Amphibians. *S. Afr. Nat. Sci. Prog. Rpt.* 151, IV, 109pp.
- BREEN, C.M., AND BEGG, G.W. 1989. Conservation status of southern African wetlands. Pp. 254-263 in Huntley, B.J., (ed). *Biotic diversity in Southern Africa: Concepts and Conservation*. Oxford University Press, Cape Town.
- BROWN, L.R., GARDNER, G. AND HALWEIL, B. 1999. *Beyond Malthus: Nineteen dimensions of the population challenge*. Washington, DC: Worldwatch Institute.
- BUNN, S.E. 1995. Biological monitoring of water quality in Australia: Workshop summary and future decisions. *Australian Journal of Ecology*. 20(1): 220-227.

- CAREY, C. AND BRYANT, C.J. 1995. Possible interrelations among environmental toxicants, amphibian development and decline of amphibian populations. *Environmental Health Perspectives*. 103(4): 13-17.
- CARRUTHERS, V.C. 2001. *Frogs and Frogging in Southern Africa*. Struik, Cape Town.
- CHANNING, A. 1998. *Tadpoles as Bio-indicators of Stream Quality: A Baseline Study*. WRC Report 718/1/98.
- CHANNING, A. 2001. *Amphibians of Central and Southern Africa*. Cornell University Press, Ithaca.
- CHANNING, A. & VAN DIJK, D.E. 1995. Amphibia: In : Cowan, G.I. (ed). *Wetlands of South Africa*. Department of Environmental Affairs and Tourism, Pretoria.
- COOKE, A.S. 1981. Tadpoles as indicators of harmful levels of pollution in the field. *Environ.Poll.* (A) 25: 123-133.
- COWAN, G.I. (ed). 1995. *Wetlands of South Africa*. Department of Environmental Affairs and Tourism, Pretoria.
- CUNNINGHAM, M. 2004. *Bufo rangeri* species account in: Minter, L.R., Burger, M., Harrison, J.A., Braak, H.H., Bishop, P.J., and Kloepfer, D. (eds). *Atlas and Red data Book of the Frogs of South Africa, Lesotho and Swaziland*. SI/MAB Series #9. Smithsonian Institution, Washington, DC.
- CUNNINGHAM, A.A., LANGTON, T.E.S., BENNET, P.M., LWEIN, J.F., DRURY, S.E.N., DASZAK, P., CUNNINGHAM, A.A, AND HYATT, A.D. 2003. Infectious disease and amphibian population declines. *Diversity and Distributions* 9:141-150.
- FALCONER, D.S. 1989. *Introduction to Quantitative Genetics*. 3rd ed. Longman-Wiley, New York.
- FREDA, J., AND MCDONALD, D.G. 1993. Toxicity of amphibian breeding ponds in the Sudbury region. *Can. J. Fish Aquat. Sci.* 50: 1497-1503.
- Gough, R.E. and Macgregor, S.K. 1996. Pathological and microbiological findings from incidents of unusual mortality of the common frog (*Rana temporaria*). *Philosophical Transactions of the Royal Society of London: Biological Science* 351: 1529-1557.
- GLAW, F. AND KOHLER, J. 1998. *Amphibian species diversity exceeds that of mammals*. *Herp Review* 29:11-14.
- HARRISON, J.A., BURGER, M., MINTER, L.R., DEVILLIERS, A.L., BAARD, E.H.W., SCOTT, E., BISHOP, P.J. AND ELLIS, S. (eds) 2001. *Conservation assessment and management plan for Southern African Frogs*. Final Report. IUCN/SSC Conservation Breeding Specialist Group.
- HEYER, W.R., DONNELLY, M.A., MCDIARMID, R.W., HAYEK, L-A.C., AND FOSTER, M.S. (eds) 1994. *Measuring and monitoring biological diversity. Standard methods for amphibians*. Biological Diversity Handbook Series. Washington, D.C.: Smithsonian Institution Press.
- HITCHINGS, S.P., AND BEEBEE, T.J.C. 1997. Genetic substructuring as a result of barriers to gene flow in urban *Rana temporaria* (common frog) populations: implications for biodiversity conservation. *Heredity* 79: 117-127.

- JACOBSEN, N.H.G. 1982. *The ecology of the reptiles and amphibians in the Burkea Africana Eragrostris pallens savannas of the Nylsvley Nature Reserve*. Unpublished MSc. thesis, University of Pretoria, Pretoria.
- KATS, L.B., PETRANKA, J.W., AND SHI, A. 1988. Anti-predator defences and persistence of amphibian larvae with fishes. *Ecology* 69:1865-1870.
- KIESECKER, J.M. 2002. Synergism between trematode infection and pesticide exposure: A link to amphibian limb deformities in nature? *Proceedings of the National Academy of Sciences of the United States of America* 99:990-994.
- KIESECKER, J.M., BLAUSTEIN, A.R., AND BELDEN, L.K. 2001. Complex causes of amphibian population declines. *Nature* 410:681-684.
- LAMBIRIS, A.J.L. 1987. Description of the tadpole of *Strongylopus hymenopus* (Boulenger, 1920) (Amphibia:Anura:Ranidae) and a key to describe southern African tadpoles of the genus. *Ann.Natal.Mus.* 28(2):455-462.
- LAMBIRIS, A.J.L. 1989. A review of the amphibians in Natal. *Lammergeyer* 39:1-210.
- LANGTON, T.E.S. 2002. On the trail of a serial killer. *BBC Wildlife*. February 2002:57-60.
- LINDER, G., BARBITTA, J. AND KWAISER, T. 1990. Short-term amphibian toxicity tests and paraquat toxicity assessment. In: Landis, W.G. & van der Schalie, W.M. (eds) ASTM STP 1096. *Aquat.Tox.Risk Assess.* 13: 189-198.
- LOW, A.B. AND REBELO, A.G.1998. *Vegetation of South Africa, Lesotho and Swaziland*. DEAT., Pretoria.
- MCDIARMID, R.W. & ALTIG, R. 1999. *Tadpoles: The biology of anuran larvae*. pp 7-24. University of Chicago Press, Chicago.
- MINTER, L.R., BURGER, M., HARRISON, J.A., BRAAK, H.H., BISHOP, P.J., AND KLOEPFER, D. (eds). *Atlas and Red data Book of the Frogs of South Africa, Lesotho and Swaziland*. SI/MAB Series #9. Smithsonian Institution, Washington, DC.
- MORIN, P.J. 1986. Interactions between interspecific competition and predation in an amphibian predator-prey system. *Ecology* 67:713-720.
- PICKFORD, D.B., AND MORRIS, I.D. 1999. Effects of endocrine-disrupting contaminants on amphibian oogenesis: methoxychlor inhibits progesterone-induced maturation of *Xenopus laevis* oocytes in vitro. *Environmental Health Perspective* 107(4): 285-292.
- RYAN, M.J. 1985. *TheTungara Frog: A Study in Sexual Selection and Communication*. University of Chicago Press, Chicago.
- SABOURIN, T.D. & FAULK, R.T. 1987. Comparative evaluation of a short-term test for developmental effects using frog embryos. In: Machlachlan, J.A.; Pratt, R.M., & Marken, L. (eds). *Developmental Toxicology: Mechanisms and Risk*. Banbury Report # 26. Cold Spring Harbour Laboratory NY. pp 203-215.
- SAKA, M. 1999. Acute toxicity tests on amphibian larvae using thiobencarb, a component of the rice paddy herbicides. *Herpetology Journal* 9(2): 73-81.

-
- SEXTON, O.J., AND PHILLIPS, C. 1986. A qualitative study of fish-amphibian interactions in 3 Missouri ponds. *Trans.Missouri.Acad.Sci.* 20:25-35.
- SIEGFIED, W.R. 1989. *Preservation of species in southern African nature reserves*. In: Huntley, B.J. (Ed). *Biotic Diversity in Southern Africa*, 186-201. Cape Town: Oxford University Press.
- SPARLING, D.W., AND LOWE, T.P., 1996. Metal concentrations of tadpoles in experimental ponds. *Environmental Pollution* 91(2):149-159.
- SPARLING, D.W., FELLERS, G.M., AND MCCONNELL, L.L. 2001. Pesticides and amphibian population declines in California, USA. *Environmental Toxicology and Chemistry* 20: 1591-1595.
- SPEARE, R. 2000. Global distribution of chytridiomycosis in amphibians.
<http://www.jcu.edu.au/school/phtm/PHTM/frogs/chyglog.htm>.
- VAN DIJK, D.E. 1966. Systematic and field keys to the families, genera and described species of southern African Anuran tadpoles. *Ann. Natal Mus.* 18(2): 231-286.
- VAN DIJK, D.E. 1971. A further contribution to the systematics of southern African Anuran tadpoles- The genus *Bufo*. *Ann. Natal Mus.* 21: 71-76.
- WAKE, D.B. 1991. *Declining amphibian populations*. *Science* 253: 860.
- WELDON, C. 2002. Chytridiomycosis survey in South Africa. *Froglog* 51:1-2.
- WELLS, K.D. 1977. The social behaviour of anuran amphibians. *Animal Behaviour* 25:666-693.
- WOODWARD, B.D. 1983. Predator-prey interactions and breeding-pond use of temporary-pond species in a desert anuran community. *Ecology* 64:1549-1555.
- WOODWARD, B.D., AND MITCHELL, S.L. 1991. The community ecology of desert anurans. Pp 223-248. In Pollis, G. (Ed). *The Ecology of Desert Communities*. University of Arizona Press, Tucson, Arizona.
- WYMAN, R.L. 1990. *What's happening to amphibians?* *Conservation Biology* 4:350-352.

CHAPTER 7: PLANTS

7.1 Introduction:

7.1.1 Terms of reference:

This chapter reports on the vegetation and plant species of the Nylsvley catchment. The aim of this chapter is to develop a framework for the sustainable management of wetlands in the Limpopo Province with specific focus on the Waterberg and Nylsvley region. Specifically, the objective, as set in the original proposal, was to develop potential biomonitoring indices specific to the wetland areas.

Since the health of the wetlands is integrally linked to the management of the entire catchment and may be reflected in the health of the terrestrial vegetation and sensitive plant species, it was also necessary to:

- review current knowledge on vegetation patterns in the catchment,
- evaluate land-use impacts on the vegetation of the catchment,
- collect baseline vegetation information from monitoring sites, and
- compile a list of threatened plant species from the catchment.

The study examines the entire floodplain, although it focuses on the immediate floodplain and riparian vegetation of the catchment.

7.1.2 Broad vegetation patterns:

The Nyl River Catchment is found approximately 100 km north of Pretoria in the Savanna Biome (Rutherford and Westfall, 1994). The upper reaches of the river start south and south-west of Modimolle (Nylstroom) and run north-eastwards from there, through the Nylsvley Nature Reserve towards Mokopane (Potgietersrus). At its widest the floodplain only just exceeds 30 km wide. The floodplain is surrounded by bushveld savanna that consists primarily of broad-leaved species.

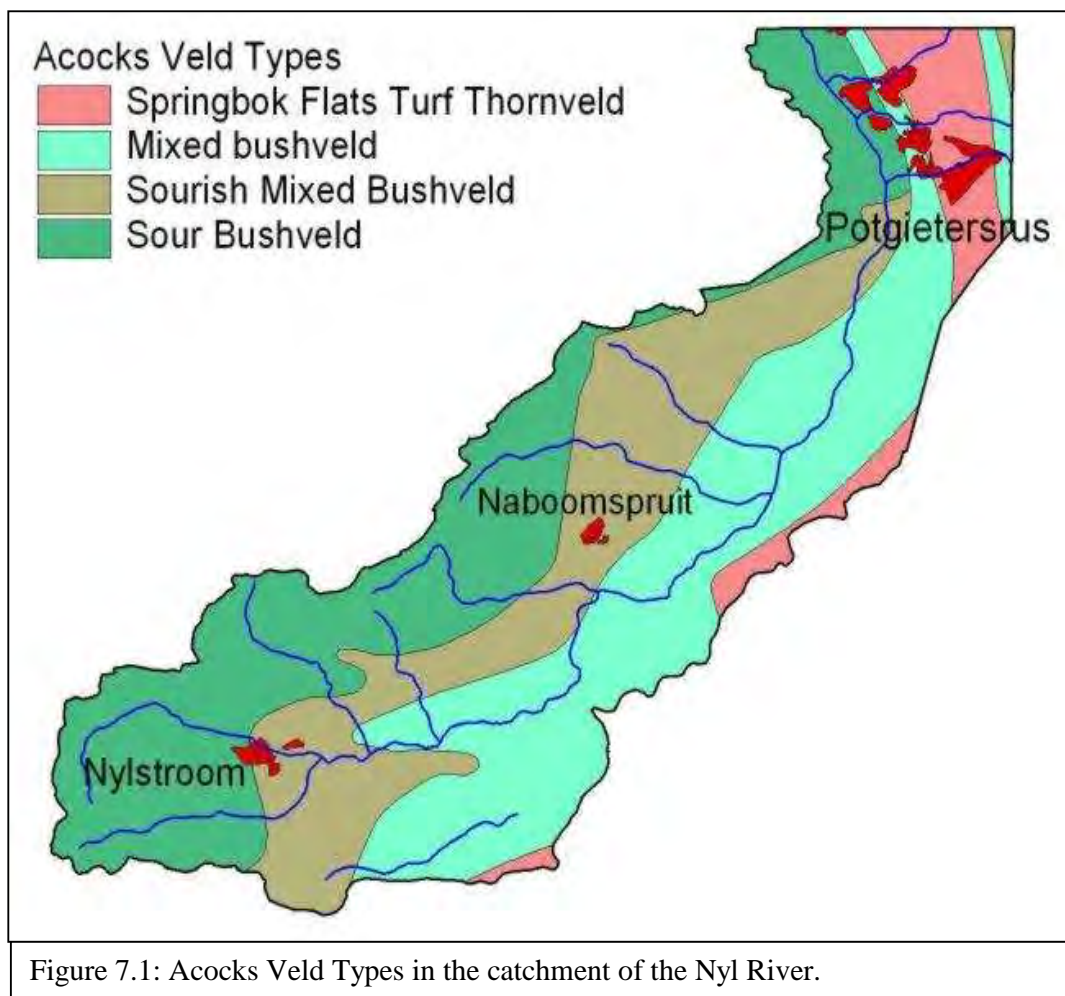
The Nyl floodplain contains the largest ephemeral floodplain vlei in southern Africa and, when inundated, covers an area in of 8 000 – 16 000 hectares (Duthie and Tarboton, 1991). The floodplain is well-known for its exceptional avifaunal diversity, but the system also plays an important role for frogs and fish and is the only recorded locality for wild rice (*Oryza longistaminata*) and the grass *Elytrophorus globularis* in South Africa. The dynamics of the floodplain vegetation are determined by rainfall and flooding cycles that vary from dry and lifeless to a sea of highly productive grasses and sedges that provide the habitat for the diversity of birds and animals found here.

The elevation and topography changes from the upper to middle and lower reaches and, consequently, the surrounding vegetation also changes (Figure 7.1). The upper reaches are hilly and relatively steeply sloping and occur within Sour Bushveld (Acocks, 1988), which is the vegetation of the bushveld mountains, such as the Waterberg (Figure 7.1). Sour Bushveld is an open savanna of *Faurea saligna* trees occurring within tall, tufted sour grasses on sandy soils of quartzite, sandstone and shale origin (Acocks, 1988).

A wide variety of other woody species also occur within Sour Bushveld. The next part of the catchment, which is still near to the upper reaches with moderately hilly topography, passes through Sourish Mixed Bushveld (Acocks, 1988), a vegetation type transitional between the sour mountain vegetation and the sweet plains vegetation (Figure 7.1). Sourish Mixed Bushveld is an open savanna dominated by the tree *Acacia caffra* and the grasses *Cymbopogon plurinodis*, *Themeda triandra*, *Elionurus muticus* and *Hyparrhenia* species (Acocks, 1988). Below the Sourish Mixed Bushveld is Mixed Bushveld. Most of the flat, wide part of the Nyl River, constituting the middle to lower reaches, occurs in Mixed Bushveld (Acocks, 1988) (Figure 7.1). On the basis of a study by Irvine (1941 in Acocks 1988), Acocks recognises two main variations of Mixed Bushveld, of which the study area falls into Mixed *Terminalia-Dichapetalum* Veld.

This is a dense, fairly tall vegetation type dominated by *Terminalia sericea*, *Ochna pulchra* and *Burkea africana* (Acocks, 1988). A small amount of Springbok Flats Turf Thornveld enters the southern and south-eastern boundary of the catchment, mostly around Makopane (Potgietersrus) (Figure 7.1). It is an open thornveld, the principal trees being *Acacia karroo*, *A. nilotica* and *Ziziphus mucronata* that occur on turf soils of the hot, flat plains of the catchment.

7.2 Materials and Methods:



7.2.1 Broad vegetation patterns:

A desk top study was undertaken to provide background information on general vegetation patterns in the catchment of the Nyl River (from a national perspective) and broad vegetation patterns in the Nylsvley area. This section of the report forms the background and introduction to the remainder of the study. A vegetation map for the catchment and surrounding areas is available in draft form from the VegMap Project (Mucina and Rutherford, in press). Units for this vegetation map are based on surrogate environmental information, including topographic, geological and landtype data, as well as floristic data, where available, and expert knowledge. A landtype is an area with relatively uniform soil, topographic and climatic characteristics (Land Type Survey Staff, 1984). Most of those parts of the Nyl River that flow through marshy areas occur in the Ca land type (Ca90). This constitutes the broad floodplain of the catchment. Surrounding this is vegetation occurring on various land types that can be divided into mountainous areas with shallow soils and undulating slopes with deeper soils.

7.2.2 Flora:

The collection of plant specimens in the Nyl catchment have been undertaken for various purposes in the past, but most intensively in the Nylsvley Nature Reserve. Checklists of species from the Nylsvley Nature Reserve have been supplemented with specimen data obtained from the National Herbarium as well as from localised studies done in the catchment, where available. This provides a plant species checklist for the catchment that provides an indication of the floristic diversity in the study area. This checklist is given in Appendix 1.

7.2.3 Field survey of monitoring sites:

It is important to describe how the health of the wetlands of the Nylsvley catchment changes over time. This is achieved by undertaking monitoring, which involves collecting information at regular intervals that measures the extent of variation from an acceptable standard or condition and/or change in condition over time. Depending on what aspects are being monitored, the monitoring process can be carried out at different levels of intensity and frequency. It is important, however, that a baseline inventory is in place and that the extent, scale and likely causes of problems have already been identified. Only then can appropriate variables and methods be chosen to provide information for monitoring. The vegetation at eight monitoring sites was surveyed in order to provide baseline vegetation data for the study. The monitoring sites were positioned to include variation in the different reaches of the catchment, but were located as close as possible to sites for monitoring of other physical and biological components of the overall study. At each site the following information on vegetation was collected:

- vegetation physiognomy / structure;
- floristic composition;
- species diversity;
- presence of invasive species.

7.2.4 Threatened plant species evaluation:

Information on the presence and habitat preferences of threatened plant species were compiled. Each of the species was evaluated in terms of habitat requirements and availability, threats and the contribution that the presence of these species may make to determining vegetation health in the Nylsvley area. A total of 29 threatened plant species (Hilton-Taylor, 1996) have been historically recorded in the area surrounding Nylsvley.

7.2.5 Land-use impacts on vegetation:

Wetlands may be under pressure from a number of damaging factors that could lead to the loss of benefits derived from them. It is important to understand these impacts and threats in order to make decisions about important biological variables to monitor to be able to detect changes in the health of ecosystems. Impacts that may be having an effect on vegetation of the Nylsvley catchment include cultivation, grazing by domestic animals, inappropriate burning practice, invasion by alien plants, the proliferation of bridges and dams and direct pollution. Some of these pressures can be detected with the use of land cover data and can, therefore, be monitored as future updated land cover information becomes available. Land cover data were used to provide an example of how impacts on indigenous vegetation in the Nylsvley River catchment may be detected. Satellite-derived land cover data (Fairbanks, *et al.*, 2000) overlaid on vegetation patterns to determine the extent of transformation in the catchment that could be attributed to various land-uses. Information on the number of road-crossings and farm dams were determined using 1:50 000 topographic maps of the area.

7.2.6 Biomonitoring techniques:

A brief literature survey was undertaken to determine the type of monitoring techniques used in wetlands, primarily in South Africa. Included in this component was an evaluation of the Riparian Vegetation Index (Kemper, 1999) as a technique for biomonitoring.

7.3 Results:

7.3.1 Broad vegetation patterns:

The vegetation of the Nyl catchment has not been studied at a uniform level and only small areas are well-understood. The vegetation of the Nylsvley Nature Reserve has been the focus of a number of studies, including a detailed survey by Coetzee *et al.* (1976). Although limited to the extent of the Reserve, the results of this study provide information that can be extrapolated to similar areas in the catchment, including most of those parts of the river that flow through wide marshy areas. The local vegetation patterns along this part of the river are dependant on soil properties and local elevation. Broad-leaved savanna occurs on sandy soils of sandstone and felsite origin, whereas microphyllous and mixed savanna is found on clay and vertic clay of alluvial or basaltic origin. Grasslands occur on bottom-slopes where drainage is poor (adjacent to the vleis areas) or where a hard, impenetrable layer underlies the soil. The grasslands adjacent to the vleis are dominated by *Tristachya rehmannii* and *Setaria perennis*, accompanied by *Trachypogon spicatus*, *Elionurus muticus*, *Digitaria erianthe* and *Eragrostis gummiflua*.

7.3.2 Flora of Nylsvley catchment:

A total of 950 species were found in the floodplain of the Nyl River and its immediate surroundings from previous collections done in the region. A checklist of these species is given in Appendix 1. A detailed vegetation map of the Nyl catchment from the VegMap Project (Mucina and Rutherford, in press) is given in Figure 7.2.

Most of the Nyl floodplain, including much of the Nylsvley Nature Reserve, is classified as Subtropical Freshwater Wetlands (Figure 7.2). Surrounding these are Springbokvlakte Thornveld, mostly to the southeast, and Central Sandy Bushveld, mostly to the north-west. With elevation on the northern side of the floodplain, the vegetation becomes Waterberg Mountain Bushveld and then, at the summit, Waterberg-Magaliesberg Summit Sourveld.

7.3.2.1 Field survey of monitoring sites:

The vegetation of the different sites varied according to its position in the catchment (Table 7.1). The upper reaches of the catchment tended to be more wooded than the lower reaches. The upper reaches have steep banks, shallow soils, rocky substrates and water is fast-moving. The lower reaches have fluctuating water levels, sloping banks, deeper soils with few rocks and generally slow-moving, deeper water. Certain morphological types of plant are more suitable for the different habitats of these different reaches.

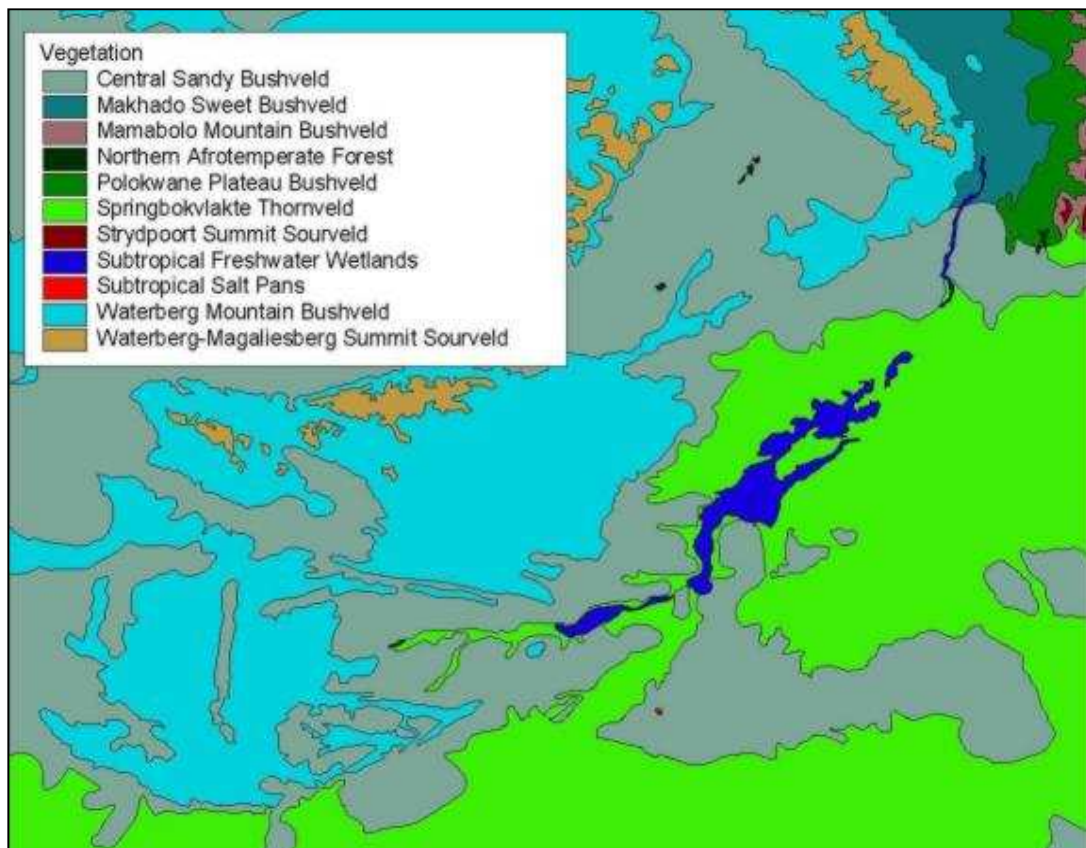


Figure 7.2: Vegetation communities of the broad area surrounding the Nyl River catchment.

Although there are too few sites to make statistically meaningful inferences, it is interesting to compare data from sites in similar positions in the catchment. Those sites with a higher proportion of alien species have lower relative species richness and are generally in poorer condition. At a course level, it can be deduced that lower cover and richness of indigenous species is a sign of degradation and that monitoring of these attributes can provide a useful assessment of catchment health.

7.3.2.2 Threatened plant species evaluation:

Information on the presence and habitat preferences of threatened plant species were compiled. Each of the species was evaluated in terms of habitat requirements and availability, threats and the contribution that the presence of these species may make to determining vegetation health in the Nylsvley area. A total of 29 threatened plant species have been historically recorded in the area surrounding Nylsvley (Table 7.2). Eight of these may be directly associated with wetland habitats, the remainder occurring in terrestrial habitats in the catchment.

Threatened species can provide a useful indicator of ecosystem health since:

1. They are vulnerable to many impacts that lead to habitat degradation;
2. Many of the threats that affect habitats have an impact on threatened species (e.g. Victor and Dold, 2003). Assessing threats to these species therefore provides an indication of the threats to habitat.

The first step to monitoring the threatened species is to collect baseline information on the population status in the catchment area. Due to the rarity of many of these species, detailed information on populations is usually lacking. The following is a discussion of species that need to be located, assessed and monitored.

The purpose of monitoring for each species is different (as discussed below), but in general, all can be used as indicator species for general ecosystem health, as well as to ensure their persistence in the area:

Barleria bolusii is a relatively poorly known species that needs information collected in order to assess its status.

Brachiaria subulifolia occurs in damp areas. It would therefore be affected by any alteration of drainage and, being rare and sensitive, could provide a good indication of ecosystem disturbance.

Ceropegia stentiae and *Ceropegia turricula* both require monitoring since they are very rare species that need monitoring to ensure that they will survive or to provide an early indication if they are declining so that steps can be taken to prevent further decline.

Cucumis humifructus is undergoing a decline because of local declining populations of the Aardvark, on which it is symbiotically dependent. Monitoring of this species can, therefore, provide information on the status of animal populations in the catchment.

Cullen holubii and *Elephantorrhiza obliqua* var. *glabra* are both species that need to be located and monitored, so that a better assessment of their Red List status can be gained.

Elytrophorus globularis has only been found four times at Nylsvlei. It only germinates as pans dry out, therefore if the flooding regime changes over time it would be severely affected. It would therefore be an ideal species to monitor as an indicator species, to provide information on the flooding regime of the floodplain.

Encephalartos eugene-maraisii subsp. *eugene-marais* is threatened by cycad collectors, and needs to be protected and monitored to ensure its survival. It is, however, a species where the threat is specific to the species and does not necessarily provide information on habitat degradation.

Eucomis autumnnalis subsp. *clavata* is a widespread but heavily harvested species. It is not as sensitive to ecosystem disturbance as other threatened species, but monitoring will provide a good indication of the level of harvesting that is happening in the area and would be a useful indication of what level of harvesting is or is not sustainable.

Hermbstaedia capitata, *Justicia minima*, *Peristrophe transvaalensis* and *Portulaca trianthemoides* are all restricted species that need proper Red List assessment and monitoring.

Oryza longistaminata has a wide distribution throughout Africa. In South Africa, however, *O. longistaminata* is found only on the floodplains of Nylsvley wetland system, where it is a keystone species in the ecology of the wetland system. Dominance by this species is checked by the disturbance of warthogs that forage on the floodplain during the dry season and create patches where other species have an opportunity to become established. At Nylsvley, *O. longistaminata* only spreads vegetatively and it is thought that the entire population is a single genetic clone, effectively a single individual. This makes it extremely vulnerable to disturbance since it does not have the genetic variability at this site to accommodate environmental shifts. This characteristic may be present throughout the whole geographic range of the species but no research has been undertaken to investigate this.

The following methods are proposed for monitoring of threatened species:

1. Locate all subpopulations and map their distributions using GIS software.
2. Count population numbers in the flowering season. In the case of the *Encephalartos*, counting should be undertaken regularly, at any time of the year.
3. At each subpopulation, record the threats that are currently acting to cause a decline or any potential threats that could cause a decline in the future. It is important to distinguish between acting and potential threats.
4. Note all alien invasive plants that occur within 200m of the subpopulation.
5. Note any signs of harvesting or removal of plants including other plants comprising the community in which the threatened plant grows.

At least, these surveys should be conducted annually, and the Threatened Species Programme of the South African National Biodiversity Institute should be consulted for feedback and updated Red List status.

7.3.2.3 Land-use impacts on vegetation:

Impacts may be separated into those acting directly on the vegetation of the floodplain or riparian zone and those acting on the vegetation of the entire catchment, including mostly terrestrial vegetation. The major existing and potential impacts on the vegetation of the Nyl catchment, as identified from literature sources (e.g. Duthie and Tarboton, 1991), are:

- the proliferation of farm dams in the catchment and proposed dams of the Olifantspruit River that together affect the supply of runoff water into the floodplain;
- Cultivation that removes vegetation and causes nutrient-rich chemicals and fertilizers to runoff into the Nyl River as well as soil erosion that silts up channels;
- Proposed afforestation – 23 000 hectares of the Nyl floodplain catchment has been identified as having afforestation potential.

Other local impacts are caused by factors such as road crossings that divert the normal flow of water in the catchment into concentrated channels, rather than diffuse overland flow. This alters the local nature of floodplain vegetation. From Table 7.3 it can be seen that a large number of bridges impede or redirect water-flow in the floodplain and that numerous dams affect the hydrological regime of wetlands in downstream positions. It can, therefore, be expected that the distribution of floodplain vegetation may have been affected by these constructions and that the dynamics of floodplain vegetation may be being affected by the change in flooding conditions imposed by damming. It can also be expected that species sensitive to changes in the hydrological regime, as well as to pollutants and siltation, may be being affected by the impacts identified here.

Table 7.1: Summary of information obtained from monitoring sites.

For full species list at each site, refer to Appendix 2.

Site number	Co-ordinates	Description	Height / structure	Species per 100 m ²
Site 1	S: 24.67791 E: 28.33559	Dense tall reeds, shrubs and herbaceous vegetation	Trees up to 18 m tall, shrubs up to 2 m tall, reeds up to 2 m tall, grasses, sedges and forbs 10 – 140 cm tall	24
Site 2	S: 24.67077 E: 28.30352	Invaded streambank vegetation	Shrubs up to 5 m tall, reeds up to 3 m tall, grasses, sedges and forbs 30 – 150 cm tall	21
Site 3	S: 24.76125 E: 28.35009	Riparian woodland with reeds	Trees up to 20 m tall, shrubs up to 4 m tall, reeds up to 4 m tall, grasses, sedges and forbs 15 – 120 cm tall	27
Site 4	S: 24.65389 E: 28.47612	Riparian woodland	Trees up to 10 m tall, shrubs up to 3 m tall, reeds 3 m tall, grasses, sedges and forbs 30 – 150 cm tall	45
Site 5	S: 24.71006 E: 28.47916	Riparian woodland	Trees up to 10 m tall, shrubs up to 2 m tall, reeds and sedges up to 3 m tall, grasses and forbs 10 – 50 cm tall	25
Site 6	S: 24.61681 E: 28.69188	Seasonally inundated grassland	Sedges, grasses and forbs predominantly 30 cm tall, but varying from 5 – 100 cm	6
Site 7	S: 24.44234 E: 28.90590	Seasonally inundated grassland	Sedges, grasses and forbs predominantly 10 cm tall, but varying from 5 – 120 cm	15
Site 8	S: 24.27468 E: 28.97655	Tall reeds	Reeds predominantly 2 m tall, but varying from 1.2 – 2.5 m, interspersed with grasses, sedges and forbs with a height of approximately 60 cm., varying from 20 – 120 cm	10

Table 7.2: Threatened plant species that have been historically recorded in the Nyl River catchment. Unless otherwise stated, all assessments according to IUCN 3.1 are global assessments.

Taxon	Pre-1994 status	IUCN 3.1	Habitat	Notes
<i>Barleria bolusii</i> Oberm.	K	DD	Deep sandy soil. Flowers from December to February.	Only known from Naboomspruit area and attempts to relocate it have failed
<i>Brachiaria subulifolia</i> (Mez) Clayton	Nt	NT	Damp, seepage areas	
<i>Ceropegia stentiae</i> E.A.Bruce	R	LC – Rare	Basaltic loam. Flowering December to February.	Rare plant in the Waterberg (Naboomspruit)
<i>Ceropegia turricula</i> E.A.Bruce	K	NT	Unknown. Flowering December to February.	Naboomspruit area
<i>Commelina bella</i> Oberm.	K	NE	Grassland on turf soil. Flowering November to January.	Confined to Springbok Flats of NW Transvaal - threatened by agriculture
<i>Cucumis humifructus</i> Stent	K	EN B1ab(i,ii,iii,iv) + 2ab (i,ii,iii,iv)	Woodland and grassland on deep sand. Flowering January and April.	Waterberg
<i>Cullen holubii</i> (Burt Davy) C.H.Stirt.	E	*VU	Black turf flats amongst grass. Flowering January to April.	Recorded from Nylsvlei Nature Reserve. Rare and declining.
<i>Cyphostemma anatomicum</i> (C.A.Sm.) Wild & R.B.Drumm.	K	NE	Grassland, woodland, streambanks. Flowering in January.	Pietersberg district.
<i>Elephantorrhiza obliqua</i> Burt Davy var. <i>glabra</i> E.Phillips	K	NE	Grassland among rocks. Flowering October to November.	Appears to be restricted to the Limpopo province and possibly North West and Mpumalanga, where it occurs in grassland.
<i>Elytrophorus globularis</i> Hack.	K (National) Nt (Global)	VU D2 (National) LC (Global)	Vleis, pans, shallow water or damp places	Occurs throughout tropical Africa, but, in South Africa, is only found at Nylsvlei.

Nylsvley

Plants

Taxon	Pre-1994 status	IUCN 3.1	Habitat	Notes
<i>Encephalartos eugene-maraisii</i> I. Verd. subsp. <i>eugene-maraisii</i>	V	VU D1	Mountainous areas.	Bokpoort & Purekrans in the Waterberg.
<i>Erythrophysa transvaalensis</i> I. Verd.	R	LC	Rocky slopes. October to February.	Shrub to small tree. Waterberg
<i>Eucomis autumnalis</i> (Mill.) Chitt. subsp. <i>clavata</i> (Baker) Reyneke	K	NT	Open grassland and marshes. Flowering November to April.	Geophyte. Common and widespread, but used medicinally
<i>Hermbsstaedtia capitata</i> Schinz	I	Could be extinct?	Unknown. Flowering in January.	Decumbent to upright herb.
<i>Justicia minima</i> A. Meeuse	R	NE	Crevices in rocks and on shallow soil. Flowering October to January and in April.	Shrublet. Nylsvley Nature Reserve and other areas
<i>Lophacme digitata</i> Stapf	K	LC	Open highveld sourveld. Flowering February to April.	Slender, rhizomatous, tufted grass.
<i>Loudetia pedicellata</i> (Stent) Chippind	K	LC	Burkea-Terminalia veld. Flowering December to April.	Tufted grass.
<i>Mosdenia leptostachys</i> (Ficalho & Hiern) Clayton	K	LC	Bushveld, usually on sandy soil. Flowering January to April.	Rhizomatous grass.
<i>Nuxia gracilis</i> Engl.		LC	Along watercourses in low, dry areas. Flowering September to December and in March.	Bushy shrub.
<i>Oryza longistaminata</i> A. Chev. & Roehr.		VU D2 (National) LC (global)	Floodplains of the Nylsvlei wetland system.	
<i>Otholobium polyphyllum</i> (Eckl. & Zeyh.) C.H. Stirt.	R	LC	No information.	Common, appears to occur throughout the country; unlikely to occur in the Nylsvley region
<i>Ozoroa albicans</i> R. & A. Fern	K	LC	Woodland. Flowering January to March.	Shrub. Pilgrims Rest area & north central Tvl.

Taxon	Pre-1994 status	IUCN 3.1	Habitat	Notes
<i>Panicum volutans</i> J.G.Anderson	K	LC	Mainly in black turf soil in cultivated and disturbed areas and areas of high moisture. Flowering January to March.	Annual grass.
<i>Parapodium costatum</i> E.Mey.	K	LC	Grassland. Flowering October to January.	Herb. Widespread in northern parts of South Africa.
<i>Peristrophe transvaalensis</i> (C.B.Clarke) K.Balkwill	K	NE	Hillsides and undergrowth. Flowering January to June.	Perennial herb.
<i>Portulaca trianthemoides</i> Bremek.	V	NE	Dry places. Flowering in April.	Succulent herb.
<i>Rhus wilmsii</i> Diels	K	LC	Mountainsides. Flowering January to April.	Shrublet.
<i>Rhynchosia nitens</i> Benth.	K	LC	Bushveld, grassy slopes. Flowering August to May.	Shrublet.
<i>Triaspis glaucophylla</i> Engl.	R	LC	Wooded grassland, woodland, bushveld. Flowering October to April.	Scandent shrub. Northern provinces of S.A. in woodland.
<i>Tristachya biseriata</i> Stapf	K	LC	Shallow stony soils on hillsides and rocky outcrops. Flowering October to March.	Tufted grass. Widely distributed in northern provinces of S.A.

Table 7.3: In-stream constructions that influence water-flow in the Nyl River and its major tributaries.

Construction	Klein Nylrivier	Groot Nylrivier	Nylrivier
Bridges	38	29	35
Dams	27	15	24

An example of how land cover data can be used to detect indigenous vegetation removal is given in Table 7.4 and Figure 7.3.

From this data it can be seen that the factor responsible for the most direct removal of natural vegetation in the entire catchment is cultivation. Cultivation is not restricted to the terrestrial environment and has contributed to a 10% direct loss of floodplain vegetation, as represented by the vegetation type “Subtropical Freshwater Wetlands” (Table 7.4). Cultivation affects erosion and runoff in the catchment that may lead to increased siltation in floodplain areas as well as contributing to nutrient enrichment of the water. Field experience in the widely separated Eastern Cape and Gauteng Provinces indicates that satellite-derived land cover data under-estimates cultivated areas since it is unable to distinguish between natural vegetation and secondary vegetation on old lands. The true amount of cultivated land may be much greater.

Urbanisation has transformed only a small amount of vegetation (Table 7.4), but the urban areas may contribute to pollutants in the river. The same applies to mines and quarries that may have a disproportionate impact on the catchment relative to their overall size. Effects from these sources act as point pollution sources which should be detectable downstream from the source. Urban areas are also often located on rivers, streams or drainage lines.

The amount of transformation is not evenly spread. The two vegetation types that border directly on the Nyl wetlands are Springbokvlakte Thornveld and Central Sandy Bushveld. These are transformed by 72% and 38% respectively (Table 7.5), some of the most transformed vegetation in the broad area including the Nyl catchment.

Table 7.4: Land cover information for the broad area that includes the Nyl River catchment as well as for Subtropical Freshwater Wetlands that represents the lower wide floodplain.

Land cover	Proportion of total catchment and surroundings (%)	Proportion of Subtropical Freshwater Wetlands (%)
Natural vegetation		
1. Forest / woodland	37.9	80.0
2. Thicket / bushland	24.8	10.0
3. Grassland	1.4	0.0
4. Wetlands/waterbodies	0.2	0.0
Cultivation	33.2	10.0
Urban	0.9	0.0
Degraded	1.6	0.0

Table 7.5: Amount of transformation per vegetation type from land cover information for the broad area that includes the Nyl River catchment.

Vegetation type	Transformed (%)	Natural (%)
Springbokvlakte Thornveld	71.9	28.1
Makhado Sweet Bushveld	70.6	29.4
Central Sandy Bushveld	37.8	62.2
Polokwane Plateau Bushveld	27.3	72.7
Subtropical Freshwater Wetlands	10.0	90.0
Waterberg Mountain Bushveld	6.2	93.8
Mamabolo Mountain Bushveld	0.0	100.0
Waterberg-Magaliesberg Summit Sourveld	0.0	100.0
Strydpoort Summit Sourveld	0.0	100.0

It is unknown from landcover data what the relative grazing impact in the catchment is or the appropriateness of timing and frequency of burning. These factors can lead to changes in species composition and loss of vegetation cover that could promote soil erosion or loss of sensitive species. Detecting fire and grazing effects on vegetation requires detailed fieldwork supplemented by data on fire frequency and stocking rates. This requires a large project on its own.

7.3.2.4 Biomonitoring techniques:

There are various techniques that have been used for monitoring wetland vegetation, based variously on species, functional groups, habitat condition/integrity, etc. Many of these methods are designed to arrive at a score that gives an indication of the quality of the habitat at a site and are comparative in nature, i.e. they are useful in terms of comparing sites to one another or the same site at different times.

In North America a technique called Floristic Quality Assessment is used to determine the relative quality of wetlands in terms of their ecological condition on the basis of the species composition (Mushet *et al.*, 2002). Species are assigned a score on the basis of their fidelity to wetland systems and their general geographical distribution and individual species scores are accumulated to arrive at a site score. The technique provides good comparative data, but relies on good knowledge of the constituent species and on botanists that are familiar with the region's flora.

A number of wetland species can be used as indicators of system health due either to their ecological properties (Marnewick pers. comm.) or their fidelity to individual habitats (Mushet *et al.*, 2002). However, experience in semi-arid systems in southern Africa indicates that these are only reliable in relatively static systems. In the highly dynamic Nylsvley system these species are more likely to give an indication of the dynamic state of the system than ecosystem health. The Nylsvley floodplain system cycles naturally from one dynamic state to another depending on the season and hydrological regime (Marnewick, PhD) and the indicator species lose resolving power within this extreme variability. Dynamic indicators are, therefore, more important than static indicators.

The monitoring technique of Kemper (1998) is site specific and concentrates on trees and therefore requires some improvement to be applicable to a system such as at Nylsvlei. The biomonitoring technique of Kemper is limited in its potential application because:

1. It considers a limited number of species, thus ignoring many species occurring in the Nylsvley catchment;
2. It considers only some habitats in the riparian zone, which are not widely distributed at Nylsvley.

The methodology described by Kleynhans (1995) for determining Habitat Integrity could be used. It may be necessary to modify the approach slightly due to the lack of distinct riparian zones in areas of the floodplain and the complex riparian zone that occurs in others.

A summary of the approach is provided as follows:

- According to the standard method, the river would be divided into appropriate resource units.
- The riparian zone would be videod (helicopter survey)
- Using ArcView, the riparian zone would be mapped at an appropriate scale and buffered to an appropriate distance to allow for the capture of dominant land cover along the banks;
- Land cover within the buffer zone would be mapped using the land cover data provided by the GIS specialist on the project team;
- Land use impacts on the riparian zone as well as the level of impact would then be described. This process enables some quantification of an otherwise relatively subjective approach. Specialist judgment would also be applied in this part of the assessment;
- Air photo-mosaics and orthophotograph analysis would also be used to supplement the

video footage analysis.

The standard method for determining Habitat Integrity could then be applied as follows:

- The river would be divided into segments based on the desktop analysis, video footage and air photo analysis. In addition to this, sections of the river would be divided into zones of homogenous character;
- Methodologies as described by the Department of Water Affairs and Forestry (1999b) would be closely followed. Riparian components would be rated using largely qualitative procedures. For each segment of the shoreline, the general status of the riparian zone would be recorded, impacts noted and riparian cover status and instream habitat availability assessed.

The assessment of the severity of impacts would be based on the standard descriptive classes and a five point rating system for scoring within each class. Experience and specialist judgment would be required to weight the scores. The estimated impacts of all criteria would be summed, expressed as a percentage, and subtracted from 100 to arrive at a provisional assessment of habitat integrity for the shoreline and riparian components respectively. Modifications of the scoring system may be required due to the unusual application of the method for the riparian zone along the Nyl and where cumulative impacts occur. Total scores would be used to assign Habitat Integrity.

7.4 Discussion

Healthy wetland vegetation has a number of attributes that may be used for the purposes of monitoring ecosystem health. It is tolerant of inter- and intra-seasonal fluctuations – such fluctuations lead to the typical zonation found in many wetland systems.

It also has the ability to survive stochastic catastrophic events, e.g. floods. Plants have morphological attributes, such as strong root systems, that promote physical persistence. Healthy ecosystems maintain populations and species adapted to that environment. Therefore diversity and composition are stable within cyclical variation and rare species are most likely to be affected by unusual modifications. This maintenance of genetic variability permits natural successional processes after small-scale natural disturbances to occur since propagules are readily available. Healthy ecosystems resist invasion by alien organisms.

The information provided in this report is discussed in terms of the broad objectives of the overall study, specifically with respect to developing biomonitoring indices specific to the vleis area. Recommendations include further evaluation of the status of threatened species in the area and possible long-term monitoring of indicator species to evaluate the health of the natural environment.

The survey of the vegetation at the monitoring sites provides useful baseline data on vegetation patterns at the monitoring sites that can be compared to other monitored indices, i.e. fauna, water quality, etc. However, the vegetation monitoring sites were placed to coincide with the water quality and faunal monitoring sites, which was not necessarily suitable for vegetation monitoring.

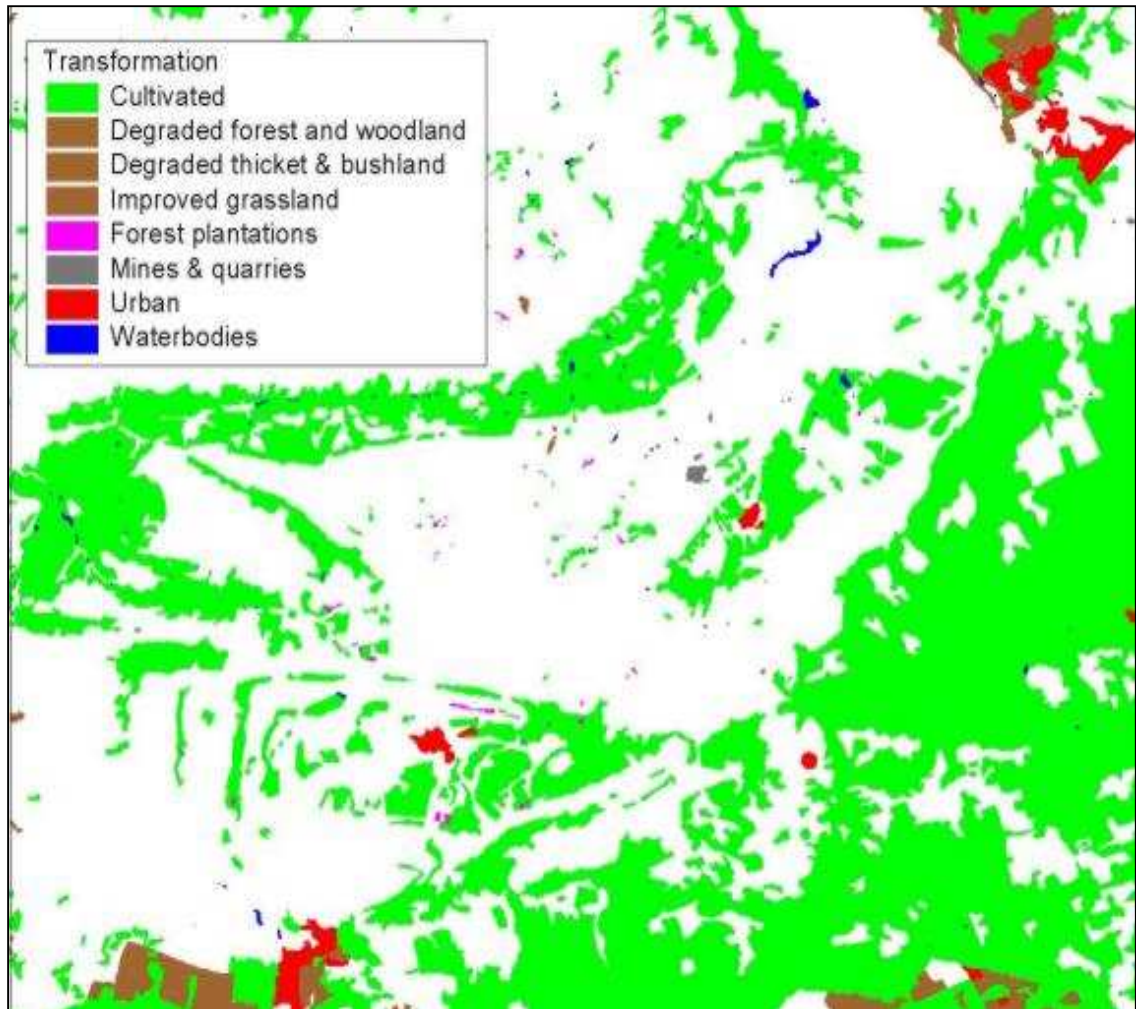


Figure 7.3: Land cover of the Nyl River catchment and surrounding areas.

The number of monitoring sites and the placement of these is not sufficient to provide an overall baseline description of the vegetation of the Nyl River catchment. Future repeat surveys can detect changes in species composition, cover, structure and species diversity in monitoring sites, if these are located in places that serves the objectives of a vegetation monitoring programme. Potential problems in undertaking repeat surveys of vegetation monitoring sites are that the exact same site needs to be surveyed, otherwise compositional turnover in space will be recorded rather than temporal turnover. The sites therefore have to be adequately marked and/or laid out.

7.5 Recommendations:

7.5.1 Guidelines for biomonitoring:

The type of monitoring system implemented depends on the objectives of the study. From a vegetation and plant species diversity point of view there are a number of potential monitoring techniques available that may focus at different spatial scales or on different elements of the natural system.

A multi-temporal and multi-spatial scale monitoring framework is proposed here that may include some of the following components:

- Broad-scale vegetation map with landcover analysis to detect overall impacts on the entire catchment
- Red-list species as species to monitor, e.g. *Elytrophoris globularis*, since these are sensitive to local changes in ecological conditions.
- Aerial photos to see change in reed-bed size over time as indicator of siltation/nutrient retention – the system may not be flushed as often or as intensively as it should be and this may provide some indication of the effect of this condition.
- Using long-term satellite NDVI data (Normalized Difference Vegetation Index) compared to rainfall data to give an indication of rain use efficiency and variability in vegetation production
- Vegetation monitoring sites for species change & composition. One important factor is the relative contribution to cover and richness by alien versus indigenous species. A larger number of sites duplicated according to similar positions in the catchment and environmental attributes is necessary to provide useful comparative data. This depends on a more detailed knowledge of vegetation patterns on the floodplain so that sites can be optimally located.

7.6 Recommended further studies:

It is very important that a comprehensive inventory of the wetlands of the catchment is undertaken that includes the following:

1. Delimitation and mapping of the location and size of wetlands of different types using data from sources such as aerial photography (primarily), satellite imagery, hydrogeomorphological information, landform and climate data, and soil data;
2. Characterizing species composition, diversity, structure and functional attributes of vegetation of mapped wetland units from detailed ground verification surveys;
3. Capturing the information in an electronic form in a GIS database with associated vegetation and species attributes.

A baseline wetland inventory of the catchment is essential to be able to protect wetlands and ensure that the wetlands work optimally for the catchment. It would provide an essential information source and tool for the management of the catchment. The optimal layout of monitoring sites is dependant on a good knowledge of wetland patterns in the catchment.

It is recommended that use is made of historical data, such as aerial photography, to detect changes in the vegetation of the catchment. One possibility is to analyse changes in reed-bed size over time as an indicator of siltation and/or nutrient retention due to the fact that the system is not flushed as often or as intensively as it should be as a result of the proliferation of dams along the stream. This could be achieved with the use of historical aerial photographs from different

dates and processed using a digital photogrammetric procedures.

Nutrient and substrate retention dynamics is an important functional component of wetlands and should be better understood in the Nylsvley catchment.

Floodplain vegetation undergoes seasonal changes in species composition that are relatively predictable. Long-term, regular sampling of specific sites can produce data that provides predictable information on the status of the vegetation. If the species composition at some point spirals out of this predictable cycle and cannot be explained by known factors, then this may be reason for concern. It is, therefore, recommended that regular sampling of monitoring sites is undertaken that covers intra- and inter-annual changes in the vegetation of the floodplain. Simple multivariate analysis of this data (using, for example, ordination methods) on an ongoing basis can provide a technique for evaluating vegetation change. However, it is essential that the monitoring sites are placed more meaningfully with respect to vegetation patterns rather than according to the needs of water quality and faunal monitoring objectives.

Detailed population information on many of the threatened species is required to assess and monitor them effectively.

7.7 References:

- ACOCKS, J.P.H. 1988. Veld types of South Africa (3rd edn.). Memoirs of the Botanical Survey of South Africa No 28. Government printer, Pretoria.
- COETZEE B.J., VAN DER MEULEN, F., ZWANZIGER, S., GONSALVES, P. AND WEISSER, P.J. 1976. A phytosociological classification of the Nylsvly Nature Reserve. South African National Scientific Programmes Report no. 20. pp.1 – 31.
- Department of Water Affairs and Forestry. 1999. Resource Directed Measures for Protection of River Water Resources. Appendix R7 (CJ Kleynhans, Assessment of Ecological Importance and Sensitivity), River Ecosystems Version 1.0, Pretoria.
- DUTHIE, A. AND TARBOTON, W. 1991. Nylsvlei. A conservation imperative. *African Wildlife* 45: 398–401.
- FAIRBANKS, D.H.K., THOMPSON, M.W., VINK, D.E., NEWBY, T.S., VAN DEN BERG, H.M. & EVERARD, D.A. 2000. The South African land-cover characteristics database: a synopsis of the landscape. *S. Afr. J. Sci.* 96: 69–82.
- GERMISHUIZEN, G. & MEYER, N.L. (eds) 2003. Plants of southern Africa: an annotated checklist. *Strelitzia* 14. National Botanical Institute, Pretoria.
- HILTON-TAYLOR, C. 1996. Red Data List of Southern African Plants. *Strelitzia* 4.
- IUCN (2000) IUCN Red List Categories. Prepared by the IUCN—Species Survival Commission, Gland, Switzerland.
- KEMPER, N.P. 1998. Riparian Vegetation Index Development. Progress report to the Water Research Commission for Project K5/850.
- KLEYNHANS, C.J. 1999. A procedure for the determination of the ecological reserve for the purposes of the national water balance model for South African Rivers. Institute for Water Quality Studies. Department of Water Affairs and Forestry, Pretoria.
- Land Type Survey Staff, 1984. Land types of the maps 2628 East Rand and 2630 Mbabane. *Mem. agric. nat. Resour. S. Afr.* No. 5.
- MUCINA, L AND RUTHERFORD, M.C. (editors) in press. Vegetation map of South Africa, Lesotho and Swaziland: an illustrated guide. *Strelitzia* xx, National Botanical Institute, Pretoria.
- MUSHET, D.M., EULISS, N.H. & SHAFFER, T.L. 2002. Floristic quality assessment of one natural and three restored wetland complexes in North Dakota, USA. *Wetlands* 22: 126 – 138.
- RUTHERFORD, M.C. & WESTFALL, R.H. (1994). *Biomes of southern Africa: an objective categorization*. National Botanical Institute: Pretoria.
- VICTOR, J.E. & DOLD, A.P. 2003. Threatened plants of the Albany Centre of Floristic Endemism, South Africa. *S.Afr.J.Sci.* 99: 437–446.

APPENDIX 1:

Checklist of plant species recorded at the study area during the current study, as well as various local checklists and National Herbarium collection records.

Species marked with an asterisk are naturalized exotics. Species taxonomy is according to Germishuizen and Meyer, (2003).

PTERIDOPHYTA**ADIANTACEAE**

- Cheilanthes deltoidea Kunze
- Cheilanthes hirta Sw.
- Cheilanthes hirta Sw. var. brevopilosa W. & N. Jacobsen
- Pellaea calomelanos (Sw.) Link var. calomelanos
- Pellaea dura (Willd.) Hook. var. dura

APONOGETONACEAE

- Marsilea villifolia* Brem. & Oberm. ex Alston & Schelpe
- Marsilea macrocarpa C. Presl
- Marsilea nubica A. Braun var. gymnocarpa (Lepr. ex A. Braun) Launert

SELAGINELLACEAE

- Selaginella dregei (C. Presl) Hieron.

GYMNOSPERMAE**PODOCARPACEAE**

- Podocarpus latifolius (Thunb.) R. Br. ex Mirb.

ZAMIACEAE

- Encephalartos eugene-maraisii I. Verd.

MONOCOTYLEDONAE**ALLIACEAE**

- Tulbaghia leucantha Baker

AMARYLLIDACEAE

- Ammocharis coranica (Ker Gawl.) Herb.
- Crinum buphanoides Welw. ex Baker
- Crinum lugardiae N. E. Br.
- Crinum macowanii Baker
- Crinum paludosum I. Verd.

Pancratium tenuifolium Hochst. ex A.Rich.

Scadoxus puniceus (L.) Friis & Nordal

MONOCOTYLEDONAE

ANTHERICACEAE

Chlorophytum angulicaule (Baker) Kativu

Chlorophytum cooperi (Baker) Nordal

Chlorophytum galpinii (Baker) Kativu var. *galpinii*

Chlorophytum recurvifolium (Baker) C.Archer & Kativu

Chlorophytum trichophlebium (Baker) Nordal

APONOGETONACEAE

Aponogeton junceus Lehm.

Aponogeton rehmannii Oliv.

ARACEAE

Stylochaeton natalensis Schott

ASPARAGACEAE

Asparagus angusticladus (Jessop) J.-P.Lebrun & Stork

Asparagus buchananii Baker

Asparagus cooperi Baker

Asparagus exuvialis Burch. forma *ecklonii* (Baker) Fellingham & N.L.Mey.

Asparagus exuvialis Burch. forma *exuvialis*

Asparagus flavicaulis (Oberm.) Fellingham & N.L.Mey. subsp. *setulosus* (Oberm.)
Fellingham & N.L.Mey.

Asparagus nodulosus (Oberm.) J.-P.Lebrun & Stork

Asparagus suaveolens Burch.

Asparagus virgatus Baker

ASPHODOLACEAE

Aloe aculeata Pole-Evans

Aloe arborescens Mill.

Aloe dolomitica Groenew.

Aloe greatheadii Schönland var. *greatheadii*

Aloe marlothii A.Berger subsp. *marlothii*

Aloe pretoriensis Pole-Evans

Aloe zebrina Baker

Bulbine angustifolia Poelln.

Bulbine capitata Poelln.

Bulbine frutescens (L.) Willd.
Bulbine narcissifolia Salm-Dyck
Kniphofia ensifolia Baker subsp. *ensifolia*
Trachyandra laxa (N.E.Br.) Oberm. var. *laxa*
Trachyandra saltii (Baker) Oberm. var. *saltii*

COLCHICACEAE

Ornithogalum flexuosum (Thunb.) U. & D. Müll.-Doblies
Ornithogalum prasinum Lindl.
Ornithogalum tenuifolium F. Delaroché subsp. *tenuifolium*

COMMELINACEAE

Commelina africana L. var. *krebsiana* (Kunth) C.B. Clarke
Commelina africana L. var. *lancispatha* C.B. Clarke
Commelina bella Oberm.
Commelina eckloniana Kunth
Commelina livingstonii C.B. Clarke
Commelina modesta Oberm.
Commelina subulata Roth
Cyanotis speciosa (L.f.) Hassk.
Floscopa glomerata (Willd. ex Schult. & Schult.f.) Hassk.

CORNACEAE

Curtisia dentata (Burm.f.) C.A. Sm.

CYPERACEAE

Ascolepis capensis (Kunth) Ridl.
Bulbostylis burchellii (Ficalho & Hiern) C.B. Clarke
Bulbostylis hispidula (Vahl) R.W. Haines
Carex cognata Kunth
Courtoisina cyperoides (Roxb.) Soják
Cyperus compressus L.
Cyperus denudatus L.f. var. *denudatus*
Cyperus difformis L.
Cyperus digitatus Roxb. subsp. *auricomus* (Sieber ex Spreng.) Kük.
Cyperus distans L.f.
Cyperus esculentus L. var. *esculentus*

CYPERACEAE

Cyperus fastigiatus Rottb.
Cyperus fulgens C.B. Clarke var. *fulgens*

Cyperus indecorus Kunth var. *decurvatus* (C.B.Clarke) Kük.
Cyperus iria L.
Cyperus longus L. var. *tenuiflorus* (Rottb.) Boeck.
Cyperus margaritaceus Vahl var. *margaritaceus*
Cyperus obtusiflorus Vahl var. *obtusiflorus*
Cyperus procerus Rottb.
Cyperus pseudovestitus (C.B.Clarke) Kük.
Cyperus sexangularis Nees
Cyperus sphaerospermus Schrad.
Eleocharis acutangula (Roxb.) Schult.
Eleocharis atropurpurea (Retz.) C.Presl
Eleocharis limosa (Schrad.) Schult.
Eleocharis variegata (Poir.) C.Presl
Fuirena pubescens (Poir.) Kunth var. *pubescens*
Fuirena stricta Steud. var. *stricta*
Kyllinga alata Nees
Kyllinga alba Nees
Kyllinga erecta Schumach. var. *erecta*
Lipocarpha chinensis (Osbeck) Kern
Lipocarpha rehmannii (Ridl.) Goetgh.
Mariscus dregeanus Kunth
Mariscus uitenhagensis Steud.
Pycreus chrysanthus (Boeck.) C.B.Clarke
Pycreus flavescens (L.) P.Beauv. ex Rchb.
Pycreus macranthus (Boeck.) C.B.Clarke
Pycreus pelophilus (Ridl.) C.B.Clarke
Pycreus pumilus (L.) Nees
Pycreus rehmannianus C.B.Clarke
Schoenoplectus corymbosus (Roth ex Roem. & Schult.) J.Raynal
Schoenoplectus muricinux (C.B.Clarke) J.Raynal
Scirpoides burkei (C.B.Clarke) Goetgh., Muasya & D.A.Simpson

DRACAENACEAE

Sansevieria aethiopica Thunb.

ERIOCAULACEAE

Syngonanthus wahlbergii (Körn.) Ruhland var. *wahlbergii*

ERIOSPERMACEAE

- Eriospermum cooperi Baker var. cooperi
- Eriospermum flagelliforme (Baker) J.C.Manning
- Eriospermum mackenii (Hook.f.) Baker subsp. galpinii (Schinz) P.L.Perry
- Eriospermum sp.

HYACINTHACEAE

- Dipcadi marlothii Engl.
- Dipcadi rigidifolium Baker
- Dipcadi viride (L.) Moench
- Drimia depressa (Baker) Jessop
- Drimia sanguinea (Schinz) Jessop
- Drimiopsis burkei Baker subsp. burkei

HYACINTHACEAE

- Eucomis autumnalis (Mill.) Chitt. subsp. clavata (Baker) Reyneke
- Ledebouria cooperi (Hook.f.) Jessop
- Ledebouria graminifolia (Baker) Jessop
- Ledebouria marginata (Baker) Jessop
- Ledebouria revoluta (L.f.) Jessop
- Ledebouria undulata (Jacq.) Jessop
- Schizocarphus nervosus (Burch.) Van der Merwe

HYDROCHARITACEAE

- Lagarosiphon muscoides Harv.
- Lagarosiphon verticillifolius Oberm.
- Ottelia ulvifolia (Planch.) Walp.

HYPOXIDACEAE

- Hypoxis filiformis Baker
- Hypoxis hemerocallidea Fisch. & Avé-Lall.
- Hypoxis iridifolia Baker
- Hypoxis rigidula Baker var. rigidula

IRIDACEAE

- Babiana bainesii Baker
- Dierama mossii (N.E.Br.) Hilliard
- Freesia grandiflora (Baker) Klatt
- Gladiolus antholyzoides Baker
- Gladiolus dalenii Van Geel subsp. dalenii
- Gladiolus elliotii Baker

Gladiolus oatesii Rolfe
Gladiolus papilio Hook.f.
Gladiolus pardalinus Goldblatt & J.C.Manning
Gladiolus permeabilis D.Delaroche subsp. *edulis* (Burch. ex Ker Gawl.) Oberm.
Gladiolus rehmannii Baker
Lapeirousia sandersonii Baker
Moraea natalensis Baker
Moraea pallida (Baker) Goldblatt

JUNCACEAE

Juncus effusus L.

NAJADACEAE

Najas horrida A.Braun

ORCHIDACEAE

Disa woodii Schltr.
Eulophia clitellifera (Rchb.f.) Bolus
Eulophia hereroensis Schltr.
Eulophia hians Spreng. var. *hians*
Eulophia hians Spreng. var. *nutans* (Sond.) S.Thomas
Eulophia ovalis Lindl. var. *bainesii* (Rolfe) P.J.Cribb & la Croix
Eulophia schweinfurthii Kraenzl.
Habenaria epipactidea Rchb.f.
Satyrium trinerve Lindl.

POACEAE

Acroceras macrum Stapf
Agrostis continuata Stapf
Agrostis lachnantha Nees var. *lachnantha*
Alloteropsis semialata (R.Br.) Hitchc. subsp. *eckloniana* (Nees) Gibbs Russ.
Androcymbium melanthioides Willd. subsp. *melanthioides*
Andropogon appendiculatus Nees
Andropogon chinensis (Nees) Merr.
Andropogon eucomus Nees
Andropogon huillensis Rendle
Andropogon schirensis A.Rich.
Anthephora pubescens Nees
Aristida adscensionis L.
Aristida aequiglumis Hack.

Aristida bipartita (Nees) Trin. & Rupr.
Aristida canescens Henrard subsp. *canescens*
Aristida congesta Roem. & Schult. subsp. *barbicollis* (Trin. & Rupr.) De Winter
Aristida congesta Roem. & Schult. subsp. *congesta*
Aristida diffusa Trin. subsp. *burkei* (Stapf) Melderis
Aristida junciformis Trin. & Rupr. subsp. *junciformis*
Aristida meridionalis Henrard
Aristida mollissima Pilg. subsp. *argentea* (Schweick.) Melderis
Aristida pilgeri Henrard
Aristida rhiniochloa Hochst.
Aristida scabrivalvis Hack. subsp. *scabrivalvis*
Aristida spectabilis Hack.
Aristida stipitata Hack. subsp. *graciliflora* (Pilg.) Melderis
Aristida stipitata Hack. subsp. *stipitata*
Bewsia biflora (Hack.) Gooss.
Bothriochloa bladhii (Retz.) S.T.Blake
Bothriochloa insculpta (Hochst. ex A.Rich.) A.Camus
Brachiaria bovonei (Chiov.) Robyns
Brachiaria brizantha (A.Rich.) Stapf
Brachiaria deflexa (Schumach.) C.E.Hubb. ex Robyns
Brachiaria eruciformis (Sm.) Griseb.
Brachiaria nigropedata (Ficalho & Hiern) Stapf
Brachiaria serrata (Thunb.) Stapf
Brachiaria subulifolia (Mez) Clayton
Brachiaria xantholeuca (Schinz) Stapf
Cenchrus ciliaris L.
Cenchrus incertus M.A.Curtis
Chloris gayana Kunth
Chloris pycnothrix Trin.
Chloris virgata Sw.
Cymbopogon caesius (Hook. & Arn.) Stapf
Cymbopogon nardus (L.) Rendle
Cymbopogon pospischilii (K.Schum.) C.E.Hubb.
Cynodon dactylon (L.) Pers.
Dactyloctenium aegyptium (L.) Willd.
Dactyloctenium giganteum Fisher & Schweick.

Diandrochloa namaquensis (Nees) De Winter
Dichanthium annulatum (Forssk.) Stapf var. *papillosum* (A.Rich.) de Wet & Harlan
Digitaria argyrograpta (Nees) Stapf
Digitaria debilis (Desf.) Willd.
Digitaria diagonalis (Nees) Stapf var. *diagonalis*
Digitaria eriantha Steud.
Digitaria eylesii C.E.Hubb.
Digitaria longiflora (Retz.) Pers.
Digitaria natalensis Stent
Diheteropogon amplectens (Nees) Clayton var. *amplectens*
Dinebra retroflexa (Vahl) Panz. var. *condensata* S.M.Phillips
Echinochloa colona (L.) Link
Echinochloa haploclada (Stapf) Stapf
Echinochloa holubii (Stapf) Stapf
Echinochloa jubata Stapf
Echinochloa stagnina (Retz.) P.Beauv.
Echinochloa ugandensis Snowden & C.E.Hubb.
Eleusine coracana (L.) Gaertn. subsp. *africana* (Kenn.-O'Byrne) Hilu & de Wet
Elionurus muticus (Spreng.) Kunth
Elytrophorus globularis Hack.
Enneapogon cenchroides (Licht. ex Roem. & Schult.) C.E.Hubb.
Enneapogon pretoriensis Stent
Enneapogon scoparius Stapf
Eragrostis amabilis (L.) Hook. & Arn.
Eragrostis aspera (Jacq.) Nees
Eragrostis barbinodis Hack.
Eragrostis barrelieri Daveau
Eragrostis biflora Hack. ex Schinz
Eragrostis capensis (Thunb.) Trin.
Eragrostis chloromelas Steud.
Eragrostis cilianensis (All.) Vignolo ex Janch.
Eragrostis curvula (Schrud.) Nees
Eragrostis echinochloidea Stapf
Eragrostis glandulosipedata De Winter
Eragrostis gummiflua Nees
Eragrostis heteromera Stapf

Eragrostis inamoena K.Schum.
Eragrostis lappula Nees
Eragrostis lehmanniana Nees var. *lehmanniana*
Eragrostis mexicana (Hornem.) Link subsp. *virescens* (J.Presl.) S.D.Koch & Sánchez Vega
Eragrostis micrantha Hack.
Eragrostis nindensis Ficalho & Hiern
Eragrostis obtusa Munro ex Ficalho & Hiern
Eragrostis pallens Hack.
Eragrostis patentipilosa Hack.
Eragrostis plana Nees
Eragrostis planiculmis Nees
Eragrostis racemosa (Thunb.) Steud.
Eragrostis rigidior Pilg.
Eragrostis rotifer Rendle
Eragrostis superba Peyr.
Eragrostis tef (Zuccagni) Trotter
Eragrostis trichophora Coss. & Durieu
Eragrostis viscosa (Retz.) Trin.
Eriochloa fatmensis (Hochst. & Steud.) Clayton
Eulalia aurea (Bory) Kunth
Eustachys paspaloides (Vahl) Lanza & Mattei
Fingerhuthia africana Lehm.
Helictotrichon turgidulum (Stapf) Schweick.
Heteropogon contortus (L.) Roem. & Schult.
Hyparrhenia anamesa Clayton
Hyparrhenia filipendula (Hochst.) Stapf var. *filipendula*
Hyparrhenia filipendula (Hochst.) Stapf var. *pilosa* (Hochst.) Stapf
Hyparrhenia hirta (L.) Stapf
Hyparrhenia poecilotricha (Hack.) Stapf
Hyperthelia dissoluta (Nees ex Steud.) Clayton
Ischaemum afrum (J.F.Gmel.) Dandy
Leersia hexandra Sw.
Leptochloa fusca (L.) Kunth
Loudetia flavida (Stapf) C.E.Hubb.
Loudetia pedicellata (Stent) Chippind.
Loudetia simplex (Nees) C.E.Hubb.

Melinis nerviglumis (Franch.) Zizka
Melinis repens (Willd.) Zizka subsp. *grandiflora* (Hochst.) Zizka
Melinis repens (Willd.) Zizka subsp. *repens*
Microchloa caffra Nees
Miscanthus junceus (Stapf) Pilg.
Mosdenia leptostachys (Ficalho & Hiern) Clayton
Oropetium capense Stapf
Oryza longistaminata A.Chev. & Roehr.
Panicum coloratum L. var. *coloratum*
Panicum deustum Thunb.
Panicum dregeanum Nees
Panicum maximum Jacq.
Panicum natalense Hochst.
Panicum schinzii Hack.
Panicum subalbidum Kunth
Panicum volutans J.G.Anderson
Paspalidium geminatum (Forssk.) Stapf
Paspalum dilatatum Poir.
Paspalum distichum L.
Paspalum scrobiculatum L.
Perotis patens Gand.
Phragmites australis (Cav.) Steud.
Phragmites mauritianus Kunth
Pogonarthria squarrosa (Roem. & Schult.) Pilg.
Sacciolepis typhura (Stapf) Stapf
Schizachyrium jeffreysii (Hack.) Stapf
Schizachyrium sanguineum (Retz.) Alston
Schmidtia pappophoroides Steud.
Sehima galpinii Stent
Setaria incrassata (Hochst.) Hack.
Setaria lindenbergiana (Nees) Stapf
Setaria nigrirostris (Nees) T.Durand & Schinz
Setaria pumila (Poir.) Roem. & Schult.
Setaria sphacelata (Schumach.) Stapf & C.E.Hubb. ex M.B.Moss var. *torta* (Stapf) Clayton
Setaria verticillata (L.) P.Beauv.
Sorghastrum friesii (Pilg.) Pilg.

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- Sorghum versicolor* Andersson
Sporobolus conrathii Chiov.
Sporobolus fimbriatus (Trin.) Nees
Sporobolus fimbriatus (Trin.) Nees var. *latifolius* Stent
Sporobolus ioclados (Trin.) Nees
Sporobolus natalensis (Steud.) T.Durand & Schinz
Sporobolus nitens Stent
Sporobolus panicoides A.Rich.
Sporobolus pyramidalis P.Beauv.
Sporobolus stapfianus Gand.
Stiburus alopecuroides (Hack.) Stapf
Stipagrostis uniplumis (Licht.) De Winter var. *neesii* (Trin. & Rupr.) De Winter
Stipagrostis uniplumis (Licht.) De Winter var. *uniplumis*
Themeda triandra Forssk.
Trachypogon spicatus (L.f.) Kuntze
Tragus berteronianus Schult.
Tricholaena monachne (Trin.) Stapf & C.E.Hubb.
Trichoneura grandiglumis (Nees) Ekman
Tripogon minimus (A.Rich.) Steud.
Triraphis andropogonoides (Steud.) E.Phillips
Triraphis schinzii Hack.
Tristachya biseriata Stapf
Urelytrum agropyroides (Hack.) Hack.
Urochloa brachyura (Hack.) Stapf
Urochloa mosambicensis (Hack.) Dandy
Urochloa oligotricha (Fig. & De Not.) Henrard
Urochloa panicoides P.Beauv.
- PONTEDERIACEAE
Heteranthera callifolia Rchb. ex Kunth
- POTAMOGETONACEAE
Potamogeton thunbergii Cham. & Schldtl.
- VELLOZIACEAE
Xerophyta humilis (Baker) T.Durand & Schinz
Xerophyta retinervis Baker
- XYRIDACEAE
Xyris congensis Büttner

 DICOTYLEDONAE

ACANTHACEAE

- Barleria bremekampii Oberm.
- Barleria crossandriiformis C.B.Clarke
- Barleria macrostegia Nees
- Blepharis leendertziae Oberm.
- Blepharis maderaspatensis (L.) Roth
- Blepharis subvolubilis C.B.Clarke
- Chaetacanthus costatus Nees
- Crabbea ovalifolia Ficalho & Hiern
- Crossandra greenstockii S.Moore
- Dyschoriste transvaalensis C.B.Clarke
- Isoglossa grantii C.B.Clarke
- Justicia betonica L.
- Justicia flava (Vahl) Vahl
- Justicia minima A.Meeuse
- Justicia protracta (Nees) T.Anderson subsp. protracta
- Peristrophe decorticans K.Balkwill
- Thunbergia atriplicifolia E.Mey. ex Nees
- Thunbergia neglecta Sond.

APIACEAE

- Heteromorpha arborescens (Spreng.) Cham. & Schldl. var. abyssinica (Hochst. ex A.Rich.)
H.Wolff

AIZOACEAE

- Gisekia africana (Lour.) Kuntze var. africana
- Gisekia pharnacioides L. var. pharnacioides
- Hypertelis salsoloides (Burch.) Adamson var. salsoloides
- Limeum fenestratum (Fenzl) Heimerl var. fenestratum
- Limeum sulcatum (Klotzsch) Hutch. var. sulcatum
- Limeum viscosum (J.Gay) Fenzl subsp. viscosum var. glomeratum (Eckl. & Zeyh.) Friedrich
- Limeum viscosum (J.Gay) Fenzl subsp. viscosum var. kraussii Friedrich
- Mollugo cerviana (L.) Ser. ex DC. var. cerviana
- Psammotropha mucronata (Thunb.) Fenzl var. mucronata

AMARANTHACEAE

- *Achyranthes aspera L. var. aspera
- Aerva leucura Moq.

Alternanthera pungens Kunth
Gomphrena celosioides Mart.
Hermbstaedtia odorata (Burch.) T.Cooke var. *aurantiaca* (Suess.) C.C.Towns.
Kyphocarpa angustifolia (Moq.) Lopr.
Kyphocarpa cruciata (Schinz) Schinz

ANACARDIACEAE

Lanea discolor (Sond.) Engl.
Lanea edulis (Sond.) Engl. var. *edulis*
Lanea gossweileri Exell & Mendonça subsp. *tomentella* (R.& A.Fern.) J.B.Gillett
Ozoroa albicans R.& A.Fern.
Ozoroa paniculosa (Sond.) R.& A.Fern. var. *paniculosa*
Protorhus longifolia (Bernh.) Engl.
Rhus dentata Thunb.
Rhus engleri Britten
Rhus keetii Schönland
Rhus lancea L.f.
Rhus leptodictya Diels
Rhus magalismontana Sond. subsp. *magalismontana*
Rhus pentheri Zahlbr.
Rhus pyroides Burch. var. *pyroides*
Rhus rigida Mill. var. *dentata* (Engl.) Moffett
Rhus wilmsii Diels
Schinus terebinthifolius Raddi
Sclerocarya birrea (A.Rich.) Hochst. subsp. *caffra* (Sond.) Kokwaro

ANNONACEAE

Annona senegalensis Pers. subsp. *senegalensis*
Hexalobus monopetalus (A.Rich.) Engl. & Diels var. *monopetalus*

APOCYNACEAE

Ancylobotrys capensis (Oliv.) Pichon
Carissa bispinosa (L.) Desf. ex Brenan
 **Catharanthus roseus* (L.) G.Don
Diplorhynchus condylocarpon (Müll.Arg.) Pichon

ASCLEPIADACEAE

Asclepias aurea (Schltr.) Schltr.
Asclepias brevipes (Schltr.) Schltr.
Asclepias densiflora N.E.Br.

Asclepias gibba (E.Mey.) Schltr. var. *media* N.E.Br.
Brachystelma hirtellum Weim.
Ceropegia decidua E.A.Bruce subsp. *decidua*
Cynanchum gerrardii (Harv.) Liede
Fockea edulis (Thunb.) K.Schum.
Gomphocarpus fruticosus (L.) Aiton f. subsp. *fruticosus*
Sarcostemma viminale (L.) R.Br. subsp. *viminale*
Secamone filiformis (L.f.) J.H.Ross

ARALIACEAE

Cussonia natalensis Sond.
Cussonia transvaalensis Reyneke

ASTERACEAE

Artemisia afra Jacq. ex Willd.
Athrix elata Sond.
Callilepis leptophylla Harv.
Conyza aegyptiaca (L.) Aiton
Conyza bonariensis (L.) Cronquist
Cotula anthemoides L.
Denekia capensis Thunb.
Dicoma gerrardii Harv. ex F.C.Wilson
Dicoma macrocephala DC.
Dicoma prostrata Schweick.
Felicia muricata (Thunb.) Nees subsp. *muricata*
Gazania krebsiana Less. subsp. *serrulata* (DC.) Roessler
Geigeria burkei Harv. subsp. *burkei* var. *burkei*
Geigeria burkei Harv. subsp. *burkei* var. *zeyheri* (Harv.) Merxm.
Geigeria elongata Alston
Gnaphalium filagopsis Hilliard & B.L.Burt
Gnaphalium polycaulon Pers.
Haplocarpha scaposa Harv.
Helichrysum argyrosphaerum DC.
Helichrysum caespititium (DC.) Harv.
Helichrysum callicomum Harv.
Helichrysum cerastioides DC. var. *cerastioides*
Helichrysum dasymallum Hilliard
Helichrysum difficile Hilliard

Helichrysum kraussii Sch.Bip.
Helichrysum mixtum (Kuntze) Moeser var. *mixtum*
Helichrysum mundtii Harv.
Helichrysum pilosellum (L.f.) Less.
Helichrysum setosum Harv.
Helichrysum stenopterum DC.
Kleinia stapeliiformis (E.Phillips) Stapf
Laggera crispata (Vahl) Hepper & J.R.I.Wood
Laggera decurrens (Vahl) Hepper & J.R.I.Wood
Litogyne gariepina (DC.) Anderb.
Lopholaena coriifolia (Sond.) E.Phillips & C.A.Sm.
Nidorella hottentotica DC.
Nidorella resedifolia DC. subsp. *resedifolia*
Nolletia rarifolia (Turcz.) Steetz
Pegolettia tenuifolia Bolus
Pseudognaphalium luteo-album (L.) Hilliard & B.L.Burt
Psiadia punctulata (DC.) Oliv. & Hiern ex Vatke
Pulicaria scabra (Thunb.) Druce
Senecio apiifolius (DC.) Benth. & Hook.f. ex O.Hoffm.
Senecio barbertonicus Klatt
Senecio coronatus (Thunb.) Harv.
Senecio glanduloso-pilosus Volkens & Muschl.
Senecio inornatus DC.
Senecio radicans (L.f.) Sch.Bip.
Stoebe vulgaris Levyns
Tarchonanthus camphoratus L.
Tripteris aghillana DC. var. *aghillana*
Vernonia oligocephala (DC.) Sch.Bip. ex Walp.
Vernonia poskeana Vatke & Hildebr. subsp. *botswanica* G.V.Pope
Vernonia staehelinoides Harv.
Xanthium strumarium L.

BORAGINACEAE

Ehretia amoena Klotzsch
Ehretia obtusifolia Hochst. ex A.DC.
Ehretia rigida (Thunb.) Druce subsp. *rigida*
Heliotropium ovalifolium Forssk.

Heliotropium steudneri Vatke

Trichodesma angustifolium Harv. subsp. *angustifolium*

Trichodesma physaloides (Fenzl) A.DC.

BRASSICACEAE

Erucastrum austroafricanum Al-Shehbaz & S.I. Warwick

BURSERACEAE

Commiphora neglecta I. Verd.

Commiphora schimperi (O.Berg) Engl.

CAMPANULACEAE

Wahlenbergia androsacea A.DC.

Wahlenbergia banksiana A.DC.

Wahlenbergia undulata (L.f.) A.DC.

CAPPARACEAE

Boscia albitrunca (Burch.) Gilg & Gilg-Ben.

Boscia foetida Schinz subsp. *rehmanniana* (Pestal.) Toelken

Cadaba aphylla (Thunb.) Wild

Cleome gynandra L.

Cleome hirta (Klotzsch) Oliv.

Cleome macrophylla (Klotzsch) Briq.

Cleome maculata (Sond.) Szyszyl.

Cleome monophylla L.

Cleome rubella Burch.

Maerua cafra (DC.) Pax

CARYOPHYLLACEAE

Polycarpea corymbosa (L.) Lam. var. *corymbosa*

Silene gallica L.

Silene pilosellifolia Cham. & Schtdl.

CELASTRACEAE

Elaeodendron transvaalense (Burt Davy) R.H. Archer

Gymnosporia buxifolia (L.) Szyszyl.

Gymnosporia polyacantha (Sond.) Szyszyl. subsp. *vaccinifolia* (P. Conrath) M. Jordaan

Gymnosporia tenuispina (Sond.) Szyszyl.

Maytenus undata (Thunb.) Blakelock

Pterocelastrus echinatus N.E. Br.

Salacia rehmannii Schinz

CERATOPHYLLACEAE

Ceratophyllum demersum L. var. *demersum*

CHENOPODIACEAE

Chenopodium cristatum F.Muell.

Lophiocarpus dinteri Engl.

CHRYSOBALANACEAE

Parinari capensis Harv. subsp. *capensis*

CLUSIACEAE

Hypericum lalandii Choisy

COMBRETACEAE

Combretum apiculatum Sond. subsp. *apiculatum*

Combretum erythrophyllum (Burch.) Sond.

Combretum hereroense Schinz

Combretum kraussii Hochst.

Combretum molle R.Br. ex G.Don

Combretum zeyheri Sond.

Terminalia brachystemma Welw. ex Hiern subsp. *brachystemma*

Terminalia sericea Burch. ex DC.

CONVOLVULACEAE

Cuscuta australis R.Br.

Evolvulus alsinoides (L.) L.

Falkia oblonga Bernh. ex C.Krauss

Ipomoea adenioides Schinz var. *adenioides*

Ipomoea bathycolpos Hallier f.

Ipomoea bolusiana Schinz

Ipomoea chloroneura Hallier f.

Ipomoea coptica (L.) Rot

Ipomoea coccinosperma Hochst. ex Choisy

Ipomoea crassipes Hook.

Ipomoea gracilisepala Rendle

Ipomoea magnusiana Schinz

Ipomoea obscura (L.) Ker Gawl. var. *obscura*

CONVOLVULACEAE

Ipomoea oenotherae (Vatke) Hallier f. var. *oenotherae*

Ipomoea sinensis (Desr.) Choisy subsp. *blepharosepala* (Hochst. ex A.Rich.) Verdc. ex A.Meeuse

Ipomoea transvaalensis A.Meeuse
Merremia palmata Hallier f.
Seddera capensis (E.Mey. ex Choisy) Hallier f.

CRASSULACEAE

Cotyledon barbeyi Schweinf. ex Baker
Cotyledon orbiculata L. var. *oblonga* (Haw.) DC.
Crassula lanceolata (Eckl. & Zeyh.) Endl. ex Walp. subsp. *transvaalensis* (Kuntze) Toelken
Crassula sarcocaulis Eckl. & Zeyh. subsp. *sarcocaulis*
Crassula setulosa Harv. var. *jenkinsii* Schönland
Crassula swaziensis Schönland subsp. *swaziensis* var. *swaziensis* forma *swaziensis*
Kalanchoe lanceolata (Forssk.) Pers.
Kalanchoe longiflora Schltr. ex J.M.Wood
Kalanchoe luciae Raym.-Hamet subsp. *luciae*
Kalanchoe paniculata Harv.
Kalanchoe rotundifolia (Haw.) Haw.

CUCURBITACEAE

Citrullus lanatus (Thunb.) Matsum. & Nakai
Coccinia adoensis (A.Rich.) Cogn.
Coccinia sessilifolia (Sond.) Cogn.
Coccinia variifolia A.Meeuse
Momordica balsamina L.
Momordica cardiospermoides Klotzsch
Momordica repens Bremek.
Trochomeria debilis (Sond.) Hook.f.

DICHAPETALACEAE

Dichapetalum cymosum (Hook.) Engl.

EBENACEAE

Diospyros lycioides Desf. subsp. *guerkei* (Kuntze) De Winter
Diospyros lycioides Desf. subsp. *lycioides*
Diospyros lycioides Desf. subsp. *sericea* (Bernh.) De Winter
Euclea crispa (Thunb.) Gürke subsp. *crispa*
Euclea linearis Zeyh. ex Hiern
Euclea natalensis A.DC. subsp. *angustifolia* F.White
Euclea undulata Thunb.

ELATINACEAE

Bergia capensis L.

Bergia decumbens Planch. ex Harv.

Bergia salaria Bremek. x *B. decumbens* Planch. ex Harv.

EUPHORBIACEAE

Acalypha angustata Sond.

Acalypha segetalis Müll.Arg.

Acalypha villicaulis Hochst.

Bridelia mollis Hutch.

Clutia monticola S.Moore var. *monticola*

Croton gratissimus Burch. var. *gratissimus*

Croton gratissimus Burch. var. *subgratissimus* (Prain) Burt Davy

Dalechampia capensis A.Spreng.

Euphorbia aeruginosa Schweick.

Euphorbia crotonoides Boiss. subsp. *crotonoides*

Euphorbia davyi N.E.Br.

Euphorbia griseola Pax subsp. *griseola*

Euphorbia inaequilatera Sond. var. *inaequilatera*

Euphorbia monteiri Hook.f. subsp. *ramosa* L.C.Leach

Euphorbia neopolycnemoides Pax & K.Hoffm.

Euphorbia prostrata Aiton

Euphorbia transvaalensis Schltr.

Euphorbia trichadenia Pax var. *trichadenia*

Flueggea virosa (Roxb. ex Willd.) Voigt subsp. *virosa*

Jatropha hirsuta Hochst. var. *hirsuta*

Jatropha hirsuta Hochst. var. *oblongifolia* Prain

Jatropha schlechteri Pax subsp. *schlechteri*

Jatropha zeyheri Sond.

Phyllanthus incurvus Thunb.

Phyllanthus maderaspatensis L.

Phyllanthus pentandrus Schumach. & Thonn.

Pseudolachnostylis maprouneifolia Pax var. *glabra* (Pax) Brenan

Pseudolachnostylis maprouneifolia Pax var. *maprouneifolia*

Pterococcus africanus (Sond.) Pax & K.Hoffm.

Spirostachys africana Sond.

Tragia incisifolia Prain

Tragia rupestris Sond.

FABACEAE

- Abrus precatorius* L. subsp. *africanus* Verdc.
Acacia ataxacantha DC.
Acacia burkei Benth.
Acacia caffra (Thunb.) Willd.
Acacia erioloba E.Mey.
Acacia erubescens Welw. ex Oliv.
Acacia galpinii Burt Davy
Acacia gerrardii Benth. subsp. *gerrardii* var. *gerrardii*
Acacia hebeclada DC. subsp. *hebeclada*
Acacia karroo Hayne
Acacia luederitzii Engl. var. *retinens* (Sim) J.H.Ross & Brenan
Acacia mellifera (Vahl) Benth. subsp. *detinens* (Burch.) Brenan
Acacia nilotica (L.) Willd. ex Delile subsp. *kraussiana* (Benth.) Brenan
Acacia robusta Burch. subsp. *robusta*
Acacia tenuispina I.Verd.
Acacia tortilis (Forssk.) Hayne subsp. *heteracantha* (Burch.) Brenan
Aeschynomene indica L.
Albizia tanganyicensis Baker f. subsp. *tanganyicensis*
Albizia versicolor Welw. ex Oliv.
Bauhinia petersiana Bolle subsp. *macrantha* (Oliv.) Brummitt & J.H.Ross
Bolusanthus speciosus (Bolus) Harms
Burkea africana Hook.
Calpurnia aurea (Aiton) Benth. subsp. *aurea*
Canavalia ensiformis (L.) DC.
Chamaecrista absus (L.) Irwin & Barneby
Chamaecrista biensis (Steyaert) Lock
Chamaecrista capensis (Thunb.) E.Mey. var. *capensis*
Chamaecrista mimosoides (L.) Greene
Chamaecrista stricta E.Mey.
Crotalaria barkae Schweinf. subsp. *barkae*
Crotalaria brachycarpa (Benth.) Burt Davy ex I.Verd.
Crotalaria globifera E.Mey.
Crotalaria laburnifolia L. subsp. *australis* (Baker f.) Polhill
Crotalaria lotoides Benth.
Crotalaria piscarpa Welw. ex Baker

Crotalaria podocarpa DC.
Crotalaria rhodesiae Baker f.
Crotalaria schinzii Baker f.
Crotalaria sphaerocarpa Perr. ex DC. subsp. *sphaerocarpa*
Cullen holubii (Burt Davy) C.H.Stirt.
Dichrostachys cinerea (L.) Wight & Arn. subsp. *africana* Brenan & Brummitt var. *africana*
Dichrostachys cinerea (L.) Wight & Arn. subsp. *africana* Brenan & Brummitt var. *setulosa* (Welw. ex Oliv.) Brenan & Brummi
Dichrostachys cinerea (L.) Wight & Arn. subsp. *nyassana* (Taub.) Brenan
Dolichos trilobus L. subsp. *transvaalicus* Verdc.
Elephantorrhiza burkei Benth.
Elephantorrhiza elephantina (Burch.) Skeels
Eriosema nutans Schinz
Eriosema pauciflorum Klotzsch var. *pauciflorum*
Eriosema psoraleoides (Lam.) G.Don
Erythrina x hennessyae Barneby & Krukoff
Hoffmannseggia burchellii (DC.) Benth. ex Oliv. subsp. *rubro-violacea* (Baker f.) Brummitt & J.H.Ross
Indigastrum costatum (Guill. & Perr.) Schrire subsp. *macrum* (E.Mey.) Schrire
Indigastrum parviflorum (B.Heyne ex Wight & Arn.) Schrire subsp. *parviflorum* var. *parviflorum*
Indigofera alternans DC. var. *alternans*
Indigofera arrecta Hochst. ex A.Rich.
Indigofera daleoides Benth. ex Harv. var. *daleoides*
Indigofera filipes Benth. ex Harv.
Indigofera heterotricha DC.
Indigofera hiliaris Eckl. & Zeyh. var. *hiliaris*
Indigofera melanadenia Benth. ex Harv.
Indigofera mollicoma N.E.Br.
Indigofera nebrowniana J.B.Gillett
Indigofera setosa N.E.Br.
Indigofera sordida Benth. ex Harv.
Lotononis bainesii Baker
Lotononis listii Polhill
Melilotus indica (L.) All.
Mundulea sericea (Willd.) A.Chev.
Neorautanenia amboensis Schinz

Otholobium polyphyllum (Eckl. & Zeyh.) C.H.Stirt.
Ooptera burchellii DC.
Peltophorum africanum Sond.
Psoralea rhizotoma C.H.Stirt.
Pterocarpus rotundifolius (Sond.) Druce subsp. *rotundifolius*
Ptychlobium plicatum (Oliv.) Harms subsp. *plicatum*
Rhynchosia albissima Gand.
Rhynchosia confusa Burt Davy
Rhynchosia densiflora (Roth) DC. subsp. *chrysadenia* (Taub.) Verdc.
Rhynchosia komatiensis Harms
Rhynchosia minima (L.) DC. var. *minima*
Rhynchosia minima (L.) DC. var. *prostrata* (Harv.) Meikle
Rhynchosia monophylla Schltr.
Rhynchosia nervosa Benth. & Harv. var. *nervosa*
Rhynchosia spectabilis Schinz
Rhynchosia totta (Thunb.) DC. var. *totta*
Senna italica Mill. subsp. *arachoides* (Burch.) Lock
Sesbania transvaalensis J.B.Gillett
Sphenostylis angustifolia Sond.
Stylosanthes fruticosa (Retz.) Alston
Tephrosia acaciifolia Baker
Tephrosia burchellii Burt Davy
Tephrosia capensis (Jacq.) Pers. var. *angustifolia* E.Mey.
Tephrosia elongata E.Mey. var. *elongata*
Tephrosia forbesii Baker subsp. *interior* Brummitt
Tephrosia linearis (Willd.) Pers.
Tephrosia longipes Meisn. subsp. *longipes* var. *longipes*
Tephrosia lupinifolia DC.
Tephrosia polystachya E.Mey. var. *latifolia* Harv.
Tephrosia polystachya E.Mey. var. *polystachya*
Tephrosia purpurea (L.) Pers. subsp. *leptostachya* (DC.) Brummitt var. *leptostachya*
Tephrosia purpurea (L.) Pers. subsp. *leptostachya* (DC.) Brummitt var. *pubescens* Baker
Tephrosia radicans Baker
Trifolium hybridum L. var. *hybridum*
Vigna unguiculata (L.) Walp. subsp. *unguiculata*
Vigna vexillata (L.) A.Rich. var. *vexillata*

Zornia glochidiata DC.

Zornia linearis E.Mey.

FLACOURTIACEAE

Flacourtia indica (Burm.f.) Merr.

Scolopia zeyheri (Nees) Harv.

GENTIANACEAE

Chironia purpurascens (E.Mey.) Benth. & Hook.f. subsp. *humilis* (Gilg) I.Verd.

GERANIACEAE

Monsonia angustifolia E.Mey. ex A.Rich.

Monsonia burkeana Planch. ex Harv.

Pelargonium dolomiticum R.Knuth

Pelargonium luridum (Andrews) Sweet

ICACINACEAE

Apodytes dimidiata E.Mey. ex Arn. subsp. *dimidiata*

ILLECEBRACEAE

Corrigiola litoralis L. subsp. *litoralis* var. *litoralis*

LAMIACEAE

Acrotome angustifolia G.Taylor

Aeollanthus buchnerianus Briq.

Becium angustifolium (Benth.) N.E.Br.

Becium obovatum (E.Mey. ex Benth.) N.E.Br. subsp. *obovatum* var. *obovatum*

Leucas capensis (Benth.) Engl.

Ocimum americanum L. var. *americanum*

Orthosiphon suffrutescens (Thonn.) J.K.Morton

Plectranthus hadiensis (Forssk.) Schweinf. ex Spreng. var. *hadiensis*

Plectranthus neochilus Schltr.

Pycnostachys reticulata (E.Mey.) Benth.

Salvia runcinata L.f.

Stachys natalensis Hochst. var. *natalensis*

Stachys spathulata Burch. ex Benth.

Tetradenia riparia (Hochst.) Codd

LENTIBULARIACEAE

Utricularia stellaris L.f.

LOBELIACEAE

Lobelia erinus L.

Monopsis decipiens (Sond.) Thulin

LOGANIACEAE

- Buddleja salviifolia (L.) Lam.
- Gomphostigma virgatum (L.f.) Baill.
- Strychnos cocculoides Baker
- Strychnos madagascariensis Poir.

LOGANIACEAE

- Strychnos pungens Soler.

LORANTHACEAE

- Agelanthus natalitius (Meisn.) Polhill & Wiens subsp. zeyheri (Harv.) Polhill & Wiens
- Erianthemum ngamicum (Sprague) Danser
- Tapinanthus quequensis (Weim.) Polhill & Wiens

MALPHIGIACEAE

- Sphedamnocarpus pruriens (A.Juss.) Szyszyl. subsp. galphimiifolius (A.Juss.) P.D.de Villiers & D.J.Botha
- Sphedamnocarpus pruriens (A.Juss.) Szyszyl. subsp. pruriens

MALVACEAE

- Cienfuegosia digitata Cav.
- Gossypium herbaceum L. subsp. africanum (Watt) Vollesen
- Hibiscus calyphyllus Cav.
- Hibiscus lunarifolius Willd.
- Hibiscus meyeri Harv. subsp. transvaalensis (Exell) Exell
- Hibiscus microcarpus Garcke
- Hibiscus nigricaulis Baker f.
- Hibiscus praeteritus R.A.Dyer
- Hibiscus schinzii Gürke
- Hibiscus trionum L.
- Malvastrum coromandelianum (L.) Garcke
- Sida alba L.
- Sida chrysantha Ulbr.
- Sida cordifolia L.
- Sida dregei Burt Davy
- Sida rhombifolia L. subsp. rhombifolia

MELASTOMATACEAE

- Antherotoma debilis (Sond.) Jacq.-Fél.

MELIACEAE

- Turraea obtusifolia Hochst.

MESEMBRYANTHEMACEAE

- Aptenia lancifolia* L. Bolus
- Delosperma davyi* N.E.Br.
- Delosperma herbeum* (N.E.Br.) N.E.Br.

MORACEAE

- Ficus abutilifolia* (Miq.) Miq.
- Ficus glumosa* Delile
- Ficus thonningii* Blume

MYRTACEAE

- Heteropyxis dehniae* Suess.
- Heteropyxis natalensis* Harv.

NYCTAGINACEAE

- Commicarpus pentandrus* (Burch.) Heimerl

NYMPHAEACEAE

- Nymphaea nouchali* Burm.f. var. *caerulea* (Savigny) Verdc.
- Nymphaea nouchali* Burm.f. var. *zanzibariensis* (Casp.) Verdc.
- Nymphoides indica* (L.) Kuntze subsp. *occidentalis* A.Raynal

OCHNACEAE

- Ochna inermis* (Forssk.) Schweinf.
- Ochna natalitia* (Meisn.) Walp.
- Ochna pulchra* Hook.f.

OLACACEAE

- Ximenia caffra* Sond. var. *caffra*

OLEACEAE

- Jasminum breviflorum* Harv. ex C.H.Wright
- Jasminum multipartitum* Hochst.
- Jasminum stenolobum* Rolfe
- Menodora africana* Hook.
- Olea capensis* L. subsp. *enervis* (Harv. ex C.H.Wright) I.Verd.

ONAGRACEAE

- Ludwigia adscendens* (L.) Hara subsp. *diffusa* (Forssk.) P.H.Raven
- Oenothera affinis* Cambess.

PASSIFLORACEAE

- Adenia digitata* (Harv.) Engl.
- Adenia glauca* Schinz

PEDALIACEAE

- Ceratotheca triloba* (Bernh.) Hook.f.
- Dicerocaryum senecioides* (Klotzsch) Abels
- Pterodiscus speciosus* Hook.
- Sesamum alatum* Thonn.

PERIPLOCACEAE

- Cryptolepis oblongifolia* (Meisn.) Schltr.
- Raphionacme hirsuta* (E.Mey.) R.A.Dyer
- Stomatostemma monteiroae* (Oliv.) N.E.Br.

PITTOSPORACEAE

- Pittosporum viridiflorum* Sims

PLANTAGINACEAE

- Plantago lanceolata* L.

PLUMBAGINACEAE

- Plumbago zeylanica* L.

POLYGALACEAE

- Polygala albida* Schinz subsp. *albida*
- Polygala hottentotta* C.Presl
- Polygala producta* N.E.Br.
- Securidaca longepedunculata* Fresen. var. *longepedunculata*

POLYGONACEAE

- Oxygonum dregeanum* Meisn. subsp. *canescens* (Sond.) Germish. var. *canescens*
- Oxygonum sinuatum* (Hochst. & Steud. ex Meisn.) Dammer
- Polygonum plebeium* R.Br.
- Persicaria decipiens* (R.Br.) K.L.Wilson
- Persicaria hystricula* (J.Schust.) Soják
- Persicaria limbata* (Meisn.) H.Hara
- Persicaria meisneriana* (Cham. & Schldl.) M.Gómez
- Persicaria senegalensis* (Meisn.) Soják forma *albotomentosa* (R.A.Graham) K.L.Wilson
- Persicaria senegalensis* (Meisn.) Soják forma *senegalensis*

PORTULACACEAE

- Portulaca kermesina* N.E.Br.
- Portulaca quadrifida* L.

PROTEACEAE

- Faurea saligna* Harv.
- Protea welwitschii* Engl. subsp. *welwitschii*
- Protea welwitschii* Engl. var. *glabrescens* (Beard) Beard

RANUNCULACEAE

- Clematis oweniae* Harv.
- Clematis villosa* DC. subsp. *villosa*
- Ranunculus multifidus* Forssk.

RHAMNACEAE

- Berchemia zeyheri* (Sond.) Grubov
- Ziziphus mucronata* Willd. subsp. *mucronata*

RUBIACEAE

- Agathisanthemum bojeri* Klotzsch subsp. *bojeri*
- Anthospermum rigidum* Eckl. & Zeyh. subsp. *rigidum*
- Canthium suberosum* Codd
- Fadogia homblei* De Wild.
- Gardenia volkensii* K.Schum. subsp. *volkensii* var. *volkensii*
- Kohautia caespitosa* Schnizl. subsp. *brachyloba* (Sond.) D.Mantell
- Kohautia cynanchica* DC.
- Kohautia virgata* (Willd.) Bremek.
- Oldenlandia capensis* L.f. var. *capensis*
- Oldenlandia cephalotes* (Hochst.) Kuntze
- Oldenlandia herbacea* (L.) Roxb. var. *flaccida* Bremek.
- Oldenlandia herbacea* (L.) Roxb. var. *herbacea*
- Pachystigma caffrum* (Sim) Robyns
- Pachystigma pygmaeum* (Schltr.) Robyns
- Pachystigma triflorum* Robyns
- Pavetta eylesii* S.Moore
- Pavetta gardeniifolia* A.Rich. var. *gardeniifolia*
- Pavetta gardeniifolia* A.Rich. var. *subtomentosa* K.Schum.
- Pavetta zeyheri* Sond.
- Pentanisia angustifolia* (Hochst.) Hochst.
- Psydrax livida* (Hiern) Bridson
- Pygmaeothamnus chamaedendrum* (Kuntze) Robyns var. *chamaedendrum*
- Pygmaeothamnus zeyheri* (Sond.) Robyns var. *zeyheri*
- Tapiphyllum parvifolium* (Sond.) Robyns

Tricalysia lanceolata (Sond.) Burtt Davy

Vangueria infausta Burch. subsp. *infausta*

RUTACEAE

Zanthoxylum capense (Thunb.) Harv.

SALICACEAE

Salix mucronata Thunb. subsp. *woodii* (Seemen) Immelman

SANTALACEAE

Osyris lanceolata Hochst. & Steud.

Thesium burkei A.W.Hill

Thesium costatum A.W.Hill var. *costatum*

Thesium gracilarioides A.W.Hill

SANTALACEAE

Thesium magalismontanum Sond.

Thesium procerum N.E.Br.

Thesium utile A.W.Hill

SAPINDACEAE

Cardiospermum corindum L.

Pappea capensis Eckl. & Zeyh.

SAPOTACEAE

Englerophytum magalismontanum (Sond.) T.D.Penn.

Mimusops zeyheri Sond.

SCROPHULARIACEAE

Alectra orobanchoides Benth.

Alectra vogelii Benth.

Buchnera sp.

Craterostigma plantagineum Hochst.

Cycnium tubulosum (L.f.) Engl. subsp. *tubulosum*

Diclis petiolaris Benth.

Freylinia tropica S.Moore

Jamesbrittenia aurantiaca (Burch.) Hilliard

Jamesbrittenia micrantha (Klotzsch) Hilliard

Jamesbrittenia montana (Diels) Hilliard

Manulea parviflora Benth. var. *parviflora*

Melanospermum foliosum (Benth.) Hilliard

Mimulus gracilis R.Br.

Nemesia fruticans (Thunb.) Benth.

-
- Striga asiatica* (L.) Kuntze
Striga bilabiata (Thunb.) Kuntze subsp. *bilabiata*
Striga elegans Benth.
Striga gesnerioides (Willd.) Vatke ex Engl.

SELAGINACEAE

- Selago lacunosa* Klotzsch
Selago welwitschii Rolfe var. *australis* Hilliard

SOLANACEAE

- Lycium cinereum* Thunb.
Solanum panduriforme E.Mey.

STERCULIACEAE

- Dombeya rotundifolia* (Hochst.) Planch. var. *rotundifolia*
Hermannia boraginiflora Hook.
Hermannia grisea Schinz

STERCULIACEAE

- Hermannia woodii* Schinz
Melhania acuminata Mast. var. *acuminata*
Melhania rehmannii Szyszyl.
Waltheria indica L.

THYMELIACEAE

- Gnidia kraussiana* Meisn. var. *kraussiana*
Gnidia sericocephala (Meisn.) Gilg ex Engl.

TILIACEAE

- Corchorus asplenifolius* Burch.
Corchorus confusus Wild
Corchorus kirkii N.E.Br.
Corchorus tridens L.
Grewia caffra Meisn.
Grewia flava DC.
Grewia flavescens Juss.
Grewia flavescens Juss. var. *olukondae* (Schinz) Wild
Grewia monticola Sond.
Grewia occidentalis L. var. *occidentalis*
Grewia retinervis Burret
Grewia subspathulata N.E.Br.
Triumfetta angolensis Sprague & Hutch.

Triumfetta sonderi Ficalho & Hiern

TURNERACEAE

Tricliceras longepedunculatum (Mast.) R.Fern. var. *longepedunculatum*

URTICACEAE

Pouzolzia mixta Solms

VAHLIACEAE

Vahlia capensis (L.f.) Thunb. subsp. *vulgaris* Bridson var. *linearis* E.Mey. ex Bridson

VERBENACEAE

Chascanum hederaceum (Sond.) Moldenke var. *hederaceum*

Clerodendrum glabrum E.Mey. var. *glabrum*

Clerodendrum ternatum Schinz

Duranta erecta L.

**Lantana camara* L.

Lippia javanica (Burm.f.) Spreng.

Vitex obovata E.Mey. subsp. *obovata*

Vitex pooara Corbishley

VERBENACEAE

Vitex rehmannii Gürke

VISCACEAE

Viscum combreticola Engl.

Viscum spragueanum Burt Davy

VITACEAE

Cyphostemma humile (N.E.Br.) Desc. ex Wild & R.B.Drumm. subsp. *dolichopus* (C.A.Sm.) Wild & R.B.Drumm.

Cyphostemma oleraceum (Bolus) J.J.M. van der Merwe

Cyphostemma puberulum (C.A.Sm.) Wild & R.B.Drumm.

Rhoicissus digitata (L.f.) Gilg & M.Brandt

Rhoicissus revoilii Planch.

Rhoicissus tridentata (L.f.) Wild & R.B.Drumm. subsp. *tridentata*

ZYGOPHYLLACEAE

Tribulus terrestris L.

APPENDIX 2

Detailed environmental and species composition information from each of the 8 monitoring sites sampled for the purposes of this study.

Site number 1:	
East of Nylstroom, south of Donkerpoort Dam adjacent to road crossing, disturbed vegetation dominated by Eucalyptus trees.	
Altitude (m) :	0000
Aspect (degrees):	0
Slope (degrees):	0
Cover total (%):	95
Cover open water (%):	10
Cover bare rock (%):	2
Height (highest) trees (m):	18
Height lowest trees (m):	14
Height (highest) shrubs (m):	2.0
Height lowest shrubs (m):	1.0
Aver. height (high) herbs (cm):	60
Aver. height lowest herbs (cm):	10
Maximum height herbs (cm):	140
Latitude (degr./min/sec):	24-40-40
Longitude (degr./min/sec):	28-20-08
<u>Species, growth form, % cover</u>	
Bidens pilosa-hl 1	Paspalum urvillei-hl 10
Chamaecrista mimosoide-hl 1	Persicaria lapathifoli-hl 2
Conyza albida-hl 3	Pseudognapha luteo-alb-hl 1
Eucalyptus camaldulens-t1 20	Psiadia punctulata-hl 2
Gomphocarpus fruticosu-s2 1	Schoenoplect corymbosu-hl 2
Hemarthria altissima-hl 5	Sesbania punicea-s2 1
Kyllinga alata-hl 1	Sida dregei-hl 1
Kyllinga species-hl 1	Solanum mauritianum-s2 1
Leersia hexandra-hl 5	Terminalia sericea-t3 1
Mariscus congestus-hl 1	Typha capensis-s2 20
Melinis nerviglumis-hl 2	Zinnia peruviana-hl 1
Nympha noucha v. caeru-hl 3	Ziziph mucron s. mucro-s2 1

Site number: 2	
East of Nylstroom, above Donkerpoort Dam adjacent to road.	
Altitude (m) :	0000
Aspect (degrees):	flat
Slope (degrees):	0
Cover total (%):	100
Cover open water (%):	0
Cover bare rock (%):	0
Height (highest) trees (m):	0
Height lowest trees (m):	0
Height (highest) shrubs (m):	5.0
Height lowest shrubs (m):	1.0
Aver. height (high) herbs (cm):	90
Aver. height lowest herbs (cm):	30
Maximum height herbs (cm):	150
Latitude (degr./min/sec):	24-40-15
Longitude (degr./min/sec):	28-18-13
<u>Species, growth form, % cover</u>	
Andropogon appendicula-hl 25	Persicaria lapathifoli-hl 1
Cirsium vulgare-hl 1	Phragmites australis-s2 5
Conyza albida-hl 1	Pseudognapha oligandru-hl 1
Cyperu escul v. escul-hl 1	Psiadia punctulata-hl 5
Cyperus sexangularis-hl 2	Ranunculus multifidus-hl 1
Diospy lycioi s. lycio-s2 1	Rhus pyroid v. pyroid-s1 10
Gunnera perpensa-hl 2	Sesbania punicea-s2 10
Helichrysum species-hl 1	Sida cordifolia-hl +
Kyllinga alata-hl 1	Typha capensis-s2 30
Leersia hexandra-hl 1	Verbena bonariensis-hl 5
Miscanthus junceus-hl 2	

Site number 3:			
South-west of Nylstroom at Modderspoort on road to Warmbad			
Altitude (m) :	0000		
Aspect (degrees):	0		
Slope (degrees):	flat		
Cover total (%):	95		
Cover open water (%):	5		
Cover bare rock (%):	0		
Height (highest) trees (m):	20		
Height lowest trees (m):	6		
Height (highest) shrubs (m):	4.0		
Height lowest shrubs (m):	2.0		
Aver. height (high) herbs (cm):	45		
Aver. height lowest herbs (cm):	15		
Maximum height herbs (cm):	120		
Latitude (degr./min/sec):	24-45-41		
Longitude (degr./min/sec):	28-21-00		
<u>Species, growth form, % cover</u>			
Acacia karroo-s1	1	Morus alba-t3	1
Celtis africana-t3	1	Oxalis species-hl	1
Cirsium vulgare-hl	1	Panicum maximum-hl	5
Combretum erythrophyll-t2	2	Paspalum dilatatum-hl	1
Conyza albida-hl	1	Persicaria lapathifoli-hl	1
Cyperu escule v. escul-hl	1	Phragmites australis-s2	30
Diospy lycioi s. lycio-s1	1	Psiadia punctulata-hl	2
Eucalyptus camaldulens-t1	30	Ranunculus multifidus-hl	1
Euphorbia heterophylla-hl	1	Schoenoplect corymbosu-hl	2
Kohautia cynanchica-hl	1	Setaria species-hl	1
Lantana rugosa-s2	1	Sida dregei-hl	1
Leersia hexandra-hl	2	Typha capensis-s2	2
Melia azedarach-t2	1	Verbena bonariensis-hl	1
Agave americana-s2	2		

Site number 4:	
East of Nylstroom on farm Grootfontein, upper reaches of catchment of Nyl River on Olifantspruit tributary, riparian vegetation on banks of running stream, relatively flat, but in landscape with steep topography.	
Altitude (m) :	1187
Aspect (degrees):	flat
Slope (degrees):	0
Cover total (%):	100
Cover open water (%):	0
Cover bare rock (%):	0
Height (highest) trees (m):	12
Height lowest trees (m):	0
Height (highest) shrubs (m):	4.0
Height lowest shrubs (m):	0.0
Aver. height (high) herbs (cm):	40
Aver. height lowest herbs (cm):	30
Maximum height herbs (cm):	150
Latitude (degr./min/sec):	24-39-14
Longitude (degr./min/sec):	28-28-34

<u>Species, growth form, % cover</u>			
Acacia karroo-s2	1	Melia azedarach-s1	1
Acacia mearnsii-t3	1	Melinis nerviglumis-hl	1
Asparagus aethiopicus-hl	1	Miscanthus junceus-s2	15
Bidens bipinnata-hl	1	Oxalis corniculata-hl	1
Buddleja salviifolia-s1	2	Panicum maximum-hl	3
Celtis africana-t3	1	Paspalum dilatatum-hl	1
Clematis brachiata-hl	1	Persicaria lapathifoli-hl	2
Commelina erecta-hl	1	Phragmites mauritianus-s2	15
Conyza canadensis-hl	1	Phyllanthus parvulus-hl	1
Conyza scabrida-hl	1	Rhus pyroid v. gracil-s1	20
Cynodon dactylon-hl	3	Rhynchosia species-hl	1
Cyperu escule v. escul-hl	1	Schkuhria pinnata-hl	1
Cyperus species-hl	1	Setari sphace v. torta-hl	2
Digitaria sanguinalis-hl	1	Sida dregei-hl	1
Diospy lycioi s. lycio-s1	2	Sporobolus africanus-hl	10
Dombey rotund v. rotun-t3	1	Tagetes minuta-hl	1
Eragrostis superba-hl	1	Terminalia sericea-t3	2
Euclea natale s. natal-s1	1	Teucrium trifidum-hl	1
Felici murica s. muric-hl	1	Themeda triandra-hl	1
Gomphrena celosioides-hl	1	Tithonia rotundifolia-hl	1
Ischaemum fasciculatum-hl	1	Verbena bonariensis-hl	1
Leersia hexandra-hl	1	Zanthoxylum capense-s2	1
Maytenus heterophylla-s1	1	Ziziph mucron s. mucro-t3	5
Salix mucronata-t3	1	Gomphocarpus fruticosa-hl	1
Xanthium strumarium-s2	1	Achyranthes species-hl	1
Schoenoplectus species-hl	1		

Site number 5: Jasper			
East of Nylstroom near to Jasper railway siding, upper reaches of Nylstroom river, riparian vegetation on banks of running stream with soft bottom, relatively flat, and in flat landscape, grassland on banks heavily grazed.			
Altitude (m) :	1134		
Aspect (degrees):			
Slope (degrees):	0		
Cover total (%):	100		
Cover open water (%):	0		
Cover bare rock (%):	0		
Height (highest) trees (m):	14		
Height lowest trees (m):	0		
Height (highest) shrubs (m):	2.0		
Height lowest shrubs (m):	0.0		
Aver. height (high) herbs (cm):	90		
Aver. height lowest herbs (cm):	10		
Maximum height herbs (cm):	150		
Latitude (degr./min/sec):	24-42-36		
Longitude (degr./min/sec):	28-28-45		
<u>Species, growth form, % cover</u>			
Acacia karroo-s1	2	Paspalum dilatatum-hl	5
Bidens bipinnata-hl	1	Paspalum distichum-hl	10
Bidens pilosa-hl	1	Persicaria lapathifoli-hl	10
Centella asiatica-hl	1	Phragmites mauritianus-hl	2
Commelina erecta-hl	1	Ranunculus multifidus-hl	1
Conyza canadensis-hl	1	Salix babylonica-t2	5
Cyperu escule v. escul-hl	1	Sesbania punicea-hl	2
Echinochloa holubii-hl	1	Sonchus species-hl	1
Gomphocarpus fruticosu-hl	1	Tagetes minuta-hl	1
Leersia hexandra-hl	5	Verbena bonariensis-hl	1
Panicum repens-hl	3	Potamogeton species-hl	1
Azolla species-hl	1	Senecio species-hl	1
Pseudognaphalium species	1	Combretum erythrophyllum	1
Rhus pyroides-s2	1	Rumex lanceolatus-hl	2
Aster squamatus-hl	1	Schoenoplectus species-hl	1
Cynodon dactylon-hl	1	Eragrostis curvula-hl	1
Sporobolus africanus-hl	1		

Site number 6: Vogelfontein			
North of Nylsvly Nature Reserve where vlei wetland exits the reserve, seasonally inundated hygrophilous grassland, flat, surface of vegetation with water-borne dead vegetation matter.			
Altitude (m) :	1099		
Aspect (degrees):			
Slope (degrees):	0		
Cover total (%):	100		
Cover open water (%):	0		
Cover bare rock (%):	0		
Height (highest) trees (m):	0		
Height lowest trees (m):	0		
Height (highest) shrubs (m):	0.0		
Height lowest shrubs (m):	0.0		
Aver. height (high) herbs (cm):	30		
Aver. height lowest herbs (cm):	5		
Maximum height herbs (cm):	100		
Latitude (degr./min/sec):	24-37-01		
Longitude (degr./min/sec):	28-41-31		
<u>Species, growth form, % cover</u>			
Commelina erecta	1	Malva species	1
Conyza canadensis	1	Panicu colora v. color-hl	1
Cynodon dactylon	1	Paspalum notatum-hl	1
Cyperus sexangularis	1	Persicaria lapathifoli-hl	3
Diplachne fusca	1	Scirpus species-hl	2
Echinochloa holubii	1	Senecio species	1
Leersia hexandra-hl	50	Centella asiatica-hl	1
Pseudognaphalium luteo-hl	1	Oryza longistaminata-hl	1
Carissa bispinosa-s2	1	Themeda triandra-hl	1
Schoenoplectus species-hl	1		

Site number 7: Haakdoring	
East of Naboomspruit on Haakdoring road, seasonally inundated hygrophilous grassland, flat, surface of vegetation with water-borne dead vegetation matter, grasses form raised hummocks, grazed by cattle.	
Altitude (m) :	1061
Aspect (degrees):	
Slope (degrees):	0
Cover total (%):	100
Cover open water (%):	0
Cover bare rock (%):	0
Height (highest) trees (m):	0
Height lowest trees (m):	0
Height (highest) shrubs (m):	0.0
Height lowest shrubs (m):	0.0
Aver. height (high) herbs (cm):	10
Aver. height lowest herbs (cm):	5
Maximum height herbs (cm):	120
Latitude (degr./min/sec):	24-26-32
Longitude (degr./min/sec):	28-54-21

<u>Species, growth form, % cover</u>			
Aristida adscensionis-hl	1	Hemarthria altissima-hl	2
Brachiaria dictyoneura	1	Ischaemum fasciculatum-hl	1
Centella asiatica	1	Panicum colora v. color-hl	1
Citrullus lanatus-hl	1	Paspalum distichum-hl	10
Conyza canadensis-hl	1	Persicaria lapathifoli	1
Crinum paludosum-hl	1	Phymaspermum parvifoli	1
Cyanotis speciosa-hl	1	Senecio pentactinus	1
Echinochloa holubii	1	Senecio species-hl	1
Eleocharis palustris-hl	5	Setaria incrassata-hl	5
Eragrostis inamoena-hl	1	Sonchus oleraceus-hl	1
Eragrostis species-hl	1	Tagetes minuta	1
Gomphocarpus fruticosu	1	Themeda triandra-hl	2
Helictotrich turgidulu-hl	1	Tithonia rotundifolia	1
Oryza longistaminata-hl	2	Schoenoplectus species-hl	1
Schistosteph crataeg-hl	1	Cynodon dactylon-hl	1
Acacia tortilis-s1	1	Phragmites australis-hl	1

Site number 8:	
South of Potgietersrus on side of main road at Moorddrift, tall reed wetland, disturbed - close to road and Eucalypt stand.	
Altitude (m) :	1057
Aspect (degrees):	
Slope (degrees):	0
Cover total (%):	100
Cover open water (%):	0
Cover bare rock (%):	0
Height (highest) trees (m):	0
Height lowest trees (m):	0
Height (highest) shrubs (m):	1.2
Height lowest shrubs (m):	2.5
Aver. height (high) herbs (cm):	60
Aver. height lowest herbs (cm):	20
Maximum height herbs (cm):	120
Latitude (degr./min/sec):	24-16-29
Longitude (degr./min/sec):	28-58-36

Species, growth form, % cover

Chloris gayana-hl	1	Phragmites mauritianus-s2	60
Cyperu escul v. escul-hl	1	Physalis angulata-hl	1
Datura stramonium-hl	1	Setaria verticillata-hl	1
Echinochloa holubii-hl	1	Verbena bonariensis-hl	2
Panicu colora v. color-hl	1		

8) FISH: MONITORING TECHNIQUES

8.1 Introduction:

As we know, the Department of Water Affairs and Forestry (DWAF) is the primary agency to manage the water resources in South Africa. They formulated a mission with respect to water quality stating that the Department must ensure the fitness of South Africa's surface water, groundwater and coastal marine resources, for water uses and for the protection of aquatic ecosystems on a sustainable basis (DWAF 1986). With this in mind, the "National Aquatic Ecosystem Biomonitoring Programme" was initiated in 1995. According to Hohls (1996) the importance of the aquatic ecosystem concept forms the basis for the programme, using biological diversity and biotic integrity as cornerstones.

- One of the indices developed for the riverine programme was the "Fish Assemblage Integrity Index" or FAII (Kleynhans, 1999). The purpose was to develop an index that was readily available and able to measure fish assemblage attributes that are responsive to human-induced environmental changes. The objectives of the FAII were:
 - To function in conjunction with other indices and to provide information to the public on the state of the nation's rivers on a regular basis.
 - To ensure it is usable within the limited available information, labour, expertise and financial resources.
 - To be structured in such a way that easy adaptation is possible when the available information on fish assemblages improves.
 - To provide information and answers within the context and framework of the legislation on South African water resources (e.g. the ecological reserve).
 - To be flexible for application in all ecoregions of the country.
 - To be developed in a hierarchical framework, as to make provision for different levels of monitoring intensity (Kleynhans, 1999).

In this chapter we will describe the fish distributed through the Nyl floodplain system and compare the historical surveys (Kleynhans, 1990) with the samples collected. As indicated earlier, the sample sites were in the Groot Nyl, the Klein Nyl and some of the more prominent tributaries. The riverine sites were included to get some indication of what species is potentially available and see if any species are absent if compared to the earlier surveys.

A further aim was to see if the sampling techniques used for the Fish Assemblage Integrity Index (FAII) used in the River Health Programme (RHP), is applicable to the palustrine wetlands. It is important to remember that the Nyl floodplain is only flooded for a very short period of time each year. When sampling is done during the "low flow" period, the floodplain has been reduced to a small stream with a few deeper pools. These pools are usually formed where farmers (commercial farms) and conservationists (Nylsvley Reserve) constructed berms and farm dams.

During the "high flow" period when the floodplain is flooded, sampling was very difficult. The main reasons being the following: 1) after a succession of wet seasons, the growth of the rye grass forms very dense stands and moving is just about impossible, 2) because of this very dense vegetation, sampling is virtually impossible using electroshocking, netting and traps, and 3) the conductivity of the water is so low that electroshocking is totally ineffective.

During the surveys listed by Kleynhans (1990), 22 species of which 2 are invasive alien species (Table 8.1) were recorded. Only one of the species is listed in Skelton (2001) as vulnerable, i.e. *Barbus brevipinnis*. None other are listed in the categories: near threatened, vulnerable, endangered or critically endangered.

8.2 Materials and methods:

During the seasonal surveys, sampling was done at the following sites: Abba, Donkerpoort, Groot Nyl, Jasper, Olifantspruit, Nylsvley, Haakdoorn and Moorddrift. During the initial surveys, sampling was done with a backpack type electroshocker (Smith-Root, Inc., Model 12-B POW Backpack Electrofisher Combo) and using hand-held nets to scoop the fish. The fish was placed in a bucket for later identification. Different biotopes at the different sites were sampled and the fish from each biotope were kept separate and the information captured accordingly. Sampling was done for a minimum of 30 minutes per site. In some instances, at the Groot Nyl site for instance, sampling times were shorter (between 15 and 20 minutes) because of the limited biotopes and the size of the stream.

During the rainy season of 2002, a boat-mounted shocker was used at the Nylsvley site. The reason was that moving in the floodplain area was difficult, mainly as a result of the dense growth of the rye grass (if compared to 2001). After the rainy season of 2001, the rye grass growth was not as dense as in 2002, partially because of the drier 2000 season. With the two wet seasons following on each other, the re-growth in 2002 was much more than 2001. Another reason was that the smaller backpack type electroshocker didn't have the same output as the much larger boat-mounted unit. Even when using the large unit, we had very little results, simply because of the very low conductivity of the water.

We also employed baited traps at all the floodplain sites (Nylsvley, Haakdoorn and Moorddrift) to test the effectiveness of this as a sampling technique. Little results were achieved in this way during the "high flow" period in the floodplain sites.

During "low flow" sampling, the backpack electroshocker gave satisfactorily results at all sites, both riverine and floodplain. At the Nylsvley site, water was reduced to a small stream, similar in flow to some of the other upstream sites. The diversity of biotopes was much lower and the substrate consisted mostly of a muddy bottom, marginal vegetation and some small cobbles. The cobbles were part of the small weir in the Nature Reserve that was washed away. At the Jacana Hide and the hide at Vogelfontein and the Crake and Dabchick hides (northern side on the Nylsvley Nature Reserve), sampling was done in the small impoundments formed as result of the low berms constructed to channel/divert water. These areas were the only places where water collected during the "low flow" periods. Again, the backpack type electroshocker was effective in the collection of fish specimens for identification.

At the sites at Haakdoorn and Moorddrift, the electroshocker and baited traps were used. The circular nets were made from 5 mm round bar and covered with shade cloth. A funnel shaped entrance was constructed to get the fish into the trap. A small "door" was used to place the bait into the trap and to remove the fish after the trap was left overnight at the sampling sites. A small float was attached to the traps to mark the trap and for retrieval of the trap from the deeper waters. Similar traps were employed by Weeks *et al.* (1996) in the Sabie-Sand River system. They used "valved minnow traps" with bread as bait and the traps were for at least three hours or where possible, overnight (Weeks *et al.*, 1996).

Although we were looking at a rapid assessment technique similar to those used in the RHP, we set the baited traps overnight. Brown bread and standard trout pellets were mixed with a little water and small balls of the bait were placed in the traps. These traps with the same bait were used in another project in the Blydepoort Dam with much success (Vlok and Engelbrecht, in preparation). Sampling (electroshocking) was then completed the following morning at these sites and the traps inspected. The fish collected were then identified and counted.

8.3 Results:

Table 8.1 gives a summary of the different surveys conducted in the Nyl River system in the period 1962 – 1990. Surveys were done in the Groot Nyl River, Klein Nyl River, Nyl River, Hessie-se-Water, Olifantspruit, Middelfonteinspruit, Bad-se-Loop, Naboomspruit and the Tobiasspruit. Surveys were conducted at infrequent intervals and not all sites were sampled in each year that surveys were conducted. In the Klein Nyl River for instance, the site on the

farm Donkerpoort were sampled 8 out of the 11 surveys, whilst a few sites only had 1 survey during the entire survey period (1962 – 1990). From Table 8.1, it is clear that the best data (most number of surveys conducted) were collected during the 1988 and 1989 surveys. Table 8.2 compare the sites surveyed during the recent study with the sites listed by Kleynhans (1990). In his document, Kleynhans (1990) refer to the farm names and farm numbers as indicated on the 1:50 000 maps. In Table 8.2, the site names as giving during the recent survey is correlated to the farm names and numbers and compared to the sites of the 1962 – 1990 surveys. Although the sampling may not be at the exact sites, the sites were compared to the farm names in the 1962 – 1990 surveys.

A total of 22 species (Table 8.3) were collected over the study period 1962 – 1990 (Kleynhans) and, including the introduced *Cyprinus carpio* and *Micropterus salmoides* and the vulnerable *Barbus brevipinnis* (Skelton, 2001) and *Clarias theodora*.

During the 1962 survey, only the Donkerpoort site was surveyed and only one species, *Labeo molybdinus*, was recorded. During the 1965 survey, two sites were sampled – Donkerpoort (Klein Nyl River) and Vischgat (Bed-se-Loop). Only four species were collected – *Barbus paludinosus*, *B. trimaculatus* (Vischgat), *B. unitaeniatus* and *Chetia flaviventris* (Donkerpoort). During the 1988 survey a total of 19 species (out of a possible 22 species) were collected. Both the exotic invasive species, *Cyprinus carpio* and *Micropterus salmoides*, were collected (Table 8.3).

In Table 8.5 the species caught at the floodplain sites are listed. From the historical data, a total of 12 species were collected and include *Barbus brevipinnis*, the only species off all species collected listed by Skelton (2001) as being vulnerable. The site on the farm Zandfontein (Kleynhans, 1990) is the first of the potential floodplain sites. From the map, it is not clear if this was a riverine or floodplain site. Two species, *Barbus unitaeniatus* and *Labeobarbus marequensis* were collected during the 1962 – 1990 surveys at this site and represent the records the furthest downstream in the system. If this site was excluded as a floodplain site, only 10 species would have been collected at the floodplain sites during the 1962 – 1990 surveys. The recent surveys will then correlate well with the historic surveys as far as species collected are concerned. During the recent surveys, five species were collected at Moorddrift, eight at Haakdoorn and seven at the various sites at the Nylsvley Nature Reserve (NRR). A “new record” for the Moorddrift and Haakdoorn sites is *Tilapia rendalli*, as it was not collected during the 1962 – 1990 surveys. A possible explanation for this can be the eagerness of farmers to improve their fishing stock. Tilapia is a popular angling species in the area and it is possible that the fish was distributed by local anglers. No evidence could be found to substantiate this theory however.

B. bifrenatus, which was collected 1979 and 1987 at Nylsvley, was not collected during the recent surveys. As it is listed by Skelton (2001) as vulnerable, this can be a matter for some concern. No specific explanation can be given for it not being present, other than habitat modification and predation. During surveys at Vogelfontein and the bird hides in the northern section of the NRR, large numbers of catfish (*C. gariepinus*) gathered in the few culverts in the roads crossing the floodplain. All migrating fish were forced into these limited migrating channels and thousands of small fish were caught in this manner. If a species were already present in low numbers, this can have an impact of the population. The structure of roads and the railway line across the floodplain, is a matter of concern and will be addressed in the management plan.

Table 8.1: Graphical presentation of fish samples over the study period (1962 – 1990) in the Nyl River and its tributaries (Kleynhans, 1990).

River	Site name (Farm name and number)	Presence (Year of sample)											
		62	65	68	78	79	83	84	87	88	89	90	
Klein Nyl	De Nyl Zyn Oog – 423					■	■				■	■	
	Vaalkop – 405											■	■
	Donkerpoort – 406	■	■	■		■		■		■	■	■	
	Rhenosterfontein – 407									■	■		
	Nylstroom – 419			■		■				■	■		
Groot Nyl	Groot Nylsoog – 447					■					■	■	
	Buffelspoort – 421					■				■	■		
	Modderpoort – 454					■				■	■		
	Rhenosterpoort – 455									■	■		
	Shangri-La – 459											■	■
	Grootvlei - 417										■		
Hessie-se-Water	Rietspruit - 412							■					■
Olifantspruit	Olifantspoort – 414					■					■	■	
	Rietspruit – 412										■	■	
	Groenfontein – 383							■			■		
	Buffelshoek – 384										■		■
Middelfonteinspruit	Middelfontein -564											■	
	Naauwpoort – 518											■	
Bad-se-Loop	Vischgat – 520		■								■	■	
Tobiasspruit	Buffelsfontein – 347												■
Nyl	Doorndraai – 415					■	■				■		
	Olifantspoort – 414					■							
	Zandfontein – 566			■		■				■	■		
	Deelkraal – 561									■	■		
	Nylsvley – 560									■	■		
	Vogelfontein – 527				■	■				■	■		
	Zyferkraal – 528									■			
	De Hoop – 334									■	■		
	Vaalkop – 325									■	■		
	Jaagbaan – 291									■	■		
	Moorddrift - 289					■				■			

Table 8.2: List of sampling sites – comparing the sites listed by Kleynhans (1990) and the sites for this study.

River	Site name (Farm name and number)	
	Kleynhans (1990).	Current study (2001 – 2003)
Klein Nyl	De Nyl Zyn Oog – 423 Vaalkop – 405 Donkerpoort – 406 Rhenosterfontein – 407 Nylstroom – 419	Klein Nyl Oog - De Nyl Zyn Oog – 423 Abba - Vaalkop – 405 Donkerpoort - Donkerpoort – 406 Sewerage - Nylstroom – 419 Koh-I-Noor – Nooitgedaght - 404
Groot Nyl	Groot Nylsoog – 447 Buffelspoort – 421 Modderpoort – 454 Rhenosterpoort – 455 Shangri-La – 459 Grootvlei – 417	Groot Nyl Oog - Groot Nylsoog – 447 Groot Nyl - Modderpoort – 454
Hessie-se-Water	Rietspruit – 412	Hessie-se-Water - Rietspruit - 412
Olifantspruit	Olifantspoort – 414 Rietspruit – 412 Groenfontein – 383 Buffelshoek – 384	Olifantspruit - Rietspruit – 412
Middelfonteinspruit	Middelfontein -564 Naauwpoort – 518	
Bad-se-Loop	Vischgat – 520	Bad-se-Loop - Vischgat – 520
Tobiasspruit	Buffelsfontein – 347	Tobiasoog - Rietfontein – 513 Tobias - Tobias Zyn Loop - 339
Naboomspruit		Mine - Buffelsfontein – 347
Nyl	Doorndraai – 415 Olifantspoort – 414 Zandfontein – 566 Deelkraal – 561 Nylsvley – 560 Vogelfontein – 527 Zyferkraal – 528 De Hoop – 334 Vaalkop – 325 Jaagbaan – 291 Moorddrift – 289	Jasper - Doorndraai – 415 Nylsvley - Nylsvley – 560 Mosdene – Du Toitskraal – 532 Haakdoorn - De Hoop – 334 Moorddrift - Moorddrift - 289

Table 8.3: List of fish species collected in the Nyl system during the survey period (1962 – 1990) compiled by Kleynhans (1990).

Species	Common names
<i>Amphilius uranoscopus</i>	Stargazer mountain catfish
<i>Aplocheilichthys johnstoni</i>	Johnston's topminnow
<i>Aplocheilichthys katangae</i>	Striped topminnow
<i>Barbus bifrenatus</i>	Hyphen barb
<i>Barbus brevipinnis</i> #	Shortfin barb
<i>Barbus paludinosus</i>	Staightfin barb
<i>Barbus trimaculatus</i>	Threespot barb
<i>Barbus unitaeniatus</i>	Longbeard barb
<i>Chetia flaviventris</i>	Canary kurper
<i>Clarias gariepinus</i>	Sharptooth catfish
<i>Clarias theodora</i>	Snake catfish
<i>Cyprinus carpio</i> *	Carp
<i>Labeobarbus marequensis</i>	Lowveld largescale yellowfish
<i>Labeo cylindricus</i>	Redeye labeo
<i>Labeo molybdinus</i>	Leaden labeo
<i>Marcusenius macrolepidotus</i>	Bulldog
<i>Mesobola brevianalis</i>	River sardine
<i>Micropterus salmoides</i> *	Largemouth bass
<i>Oreochromis mossambicus</i>	Mozambique tilapia
<i>Pseudocrenilabrus philander</i>	Southern mouthbrooder
<i>Tilapia rendalli</i>	Redbreast tilapia
<i>Tilapia sparrmanii</i>	Banded tilapia
TOTAL	22

= listed as vulnerable by Skelton (2001)

* = alien invasive species introduced into the Nyl River system.

Table 8.4: Fish collected at the riverine sites during this study, indicating the sites where they were found.

Species	Sites collected				
	Jasper	O/poort	G Nyl	D/poort	Abba
<i>Amphilius uranoscopus</i>		O/poort			
<i>Aplocheilichthys johnstoni</i>	Jasper		G Nyl	D/poort	Abba
<i>Aplocheilichthys katangae</i>					
<i>Barbus bifrenatus</i>	Jasper	O/poort	G Nyl	D/poort	Abba
<i>Barbus brevipinnis</i> #	Jasper		G Nyl	D/poort	
<i>Barbus paludinosus</i>	Jasper	O/poort	G Nyl		Abba
<i>Barbus trimaculatus</i>	Jasper	O/poort	G Nyl	D/poort	
<i>Barbus unitaeniatus</i>		O/poort			
<i>Chetia flaviventris</i>			G Nyl	D/poort	Abba
<i>Clarias gariepinus</i>	Jasper			D/poort	Abba
<i>Clarias theodora</i>					
<i>Cyprinus carpio</i> *					
<i>Labeobarbus marequensis</i>		O/poort	G Nyl	D/poort	Abba
<i>Labeo cylindricus</i>					
<i>Labeo molybdinus</i>		O/poort			
<i>Marcusenius macrolepidotus</i>			G Nyl	D/poort	
<i>Mesobola brevianalis</i>					
<i>Micropterus salmoides</i> *					
<i>Oreochromis mossambicus</i>				D/poort	
<i>Pseudocrenilabrus philander</i>	Jasper	O/poort	G Nyl	D/poort	Abba
<i>Tilapia rendalli</i>		O/poort	G Nyl	D/poort	
<i>Tilapia sparrmanii</i>	Jasper	O/poort	G Nyl	D/poort	Abba
22 species listed above the floodplain by Kleynhans	8	10	11	12	8

= listed as vulnerable by Skelton (2001)

* = alien invasive species introduced into the Nyl River system.

Table 8.5: Species collected at the floodplain sites.

Species			
<i>Aplocheilichthys johnstoni</i>			Vley
<i>Barbus bifrenatus</i>			
<i>Barbus brevipinnis</i> #			
<i>Barbus paludinosus</i>	M/drift	H/doorn	Vley
<i>Barbus trimaculatus</i>		H/doorn	Vley
<i>Barbus unitaeniatus</i>		H/doorn	
<i>Clarias gariepinus</i>		H/doorn	Vley
<i>Labeobarbus marequensis</i>			
<i>Marcusenius macrolepidotus</i>			Vley
<i>Oreochromis mossambicus</i>	M/drift	H/doorn	
<i>Pseudocrenilabrus philander</i>	M/drift	H/doorn	Vley
<i>Tilapia rendalli</i> (not listed by Kleynhans)	M/drift	H/doorn	
<i>Tilapia sparrmanii</i>	M/drift	H/doorn	Vley
12 species listed by Kleynhans at floodplain sites	5	8	7

= listed as vulnerable by Skelton (2001)

8.4 Discussion:

The aim of the project was to evaluate the sampling techniques used for the various indices developed for the River Health Programme (RHP) and test their applicability as biomonitoring techniques for wetlands and in particular the palustrine wetlands, such as the floodplain of the Nyl River system. As part of its 1994 discussion documents (NAEBP) the Department of Water Affairs and Forestry (DWAf) wanted to develop a whole suite of monitoring techniques for the diverse wetlands types in South Africa. As part of this proposal, the RHP was initiated and the biomonitoring indices developed to monitor the health of rivers.

The index using fish as a bio-indicator is known as the “Fish Assemblage Integrity Index” or FAII (Kleynhans, 1999). This index aims to measure the biological integrity of a river as based on the attributes of the fish assemblage’s native to a river. It is important to remember that all alien species to the river (introduced indigenous and exotic species) must not be included, as it was not integrated into the metrics of the index (Kleynhans, 1999). The presence and the distribution of these introduced species are however noted and one will use this information to interpret causes and declines of the indigenous fish community (Kleynhans, 1999). When this index was developed, three aspects of the fish assemblage were taken into account:

- The relative intolerance of the indigenous species expected to occur in each segment of the river.
- Abundance was however not included as a metric in the index, as it is difficult to obtain quantitative information.
- As it is difficult to use general health and well-being in the index, the percentage of fish with externally evident disease or other anomalies are used.

Parasite infestation is noted, but not included in the assessment, due to the lack of correlation between parasite burden and environmental quality (Kleynhans, 1999).

The various techniques used included electroshocking (backpack type and large boat mounted unit) and the use of baited traps. A small drop net was used once, but the dense plant growth made the use of this technique impractical. It was suggested that this drop net will ensure that all the fish in the vegetation can be trapped once the heavy metal frame was dropped over the study area. Transporting the net and moving it in the floodplain was very difficult and it was decided that this will not be an effective way to sample fish during high flow surveys.

The use of the baited traps was employed as an alternative technique to sample fish in the dense floodplain growth. The traps were set at various places at the floodplain sites and left overnight to ensure that fish active at sunset and dawn were collected. The traps were very effective to collect small fish in Lake Blydepoort (Blyde River, Mpumalanga Province, South Africa) during a study of the feeding ecology of the Smallmouth bass, *Micropterus dolomieu*. A wide variety of species were collected and the bait used was a mixture of brown bread and ground trout pellets mixed with the water from the impoundment. The species caught in the Blydepoort study included: *Micralestes acutidens*, *Clarias gariepinus*, *Pseudocrenilabrus philander*, *Oreochromis mossambicus*, *Tilapia sparmanii*, *Tilapia rendalli*, *Labeobarbus marequensis*, *L. polylepis*, *Barbus uniteaeniatus*, *B. euteania*, *B. trimaculatus*, *Labeo molybdinus* and *Micropterus dolomieu*. From this it is clear that the technique can be effective as a sampling technique, as virtually all species sampled during the study in the gut of *M. dolomieu* as prey was sampled in the traps. The only species not sampled in the traps which were collected in the gut of the smallmouth bass were *Marcusenius macrolepidotus*, *Petrocephalus catostoma* and *Chiloglanis pretoriae*.

According to Weeks *et al.* (1996), the valved minnow traps used in the Sabie-Sand study was effective to capture minnows, but not as effective to sample cichlids. The traps used in the Blydepoort study were effective for a wide range of species, including the different cichlids found in the impoundment.

During this study, very little success was achieved when the traps were used at the “vley” sites. Although the traps were set overnight, very few fish were caught. The fish sampled with this traps included: *Pseudocrenilabrus philander*, *Oreochromis mossambicus*, *Tilapia sparmanii* and *Tilapia rendalli*. Even when the shocker was used, low numbers of fish were collected from the floodplain sites, indicating relatively low numbers of fish in the system during low flow seasons.

The use of the backpack electroshocker was effective during low flow surveys. During surveys, seven species of fish were collected at the Nylsvley site and included *Marcusenius macrolepidotus*, *Barbus paludinosus*, *Pseudocrenilabrus philander* and *Clarias gariepinus*. Sampling at the floodplain sites during the low flow surveys was very similar to the riverine sites upstream of these sites. The main difference was that the diversity in biotopes was lower and consisted mostly of the river channel with a sandy or muddy substrate and marginal vegetation, mostly the rye grass and in some places small reed beds. The only floodplain site where some “stones in current” were encountered, were at the low weir in the Nylsvley Nature Reserve. The low structure was constructed using small rocks and cobbles to stabilise the earth wall. When the weir was damaged some time ago, the cobbles were left behind, giving some additional habitat in the otherwise sand/muddy environment.

A worrying factor is the modification of habitat associated with wetlands and the Nyl floodplain is no exception. A vast number of low weirs and berms have been constructed to contain some of the flow. A further modification is the numerous channels and canals that were dug over the years. The many roads and the railway add to the problem. During the high flow period of any flood, water is trapped behind the various structures, preventing a flushing of nutrients and sediment. This can result in the more dense growth of rye grass and reeds experienced by some of the farmers and that has led to more fires being employed to alleviate the “problems”. The cumulative effect is that habitat is changed which can have a negative impact on the structure of the fish community

In conclusion, we are of the opinion that the sampling techniques used for fish surveys developed for the “River Health Programme” (RHP) can be used in the floodplain areas similar to the Nylsvley floodplain. As is the protocol with the RHP, sampling must be done during the “low flow” season. Attempting to survey during the “high flow” season presented various problems. The very low conductivity, dense floodplain vegetation and the difficulty of moving in the area are the most prominent issues to mention.

Sampling can be done with the aid of an electroshocker, small seine nets and baited fish traps. The electroshocker was effective in the channel of the floodplain during low flow, as well as for sampling in the shallow pools formed on the floodplain. The baited traps are effective in the pool, especially between the vegetation in the floodplain and in the farms dams and shallow pools behind the earth berms.

8.5 Recommendations:

- Berms and canalisation in the floodplain must be rehabilitated, as these structures have a major impact on water flow and fish migration.
- Lack of sufficient culverts under roads and railways must be addressed. The few culverts currently in place again limit flow and cause water to be hold back above the roads and railway. A further impact is on the fish population. Large predators (various bird species and in particular *Clarias gariepinus*, the sharptooth catfish) use these “bottlenecks” to prey on the smaller fish species migrating to spawn. The catfish take up position in the culverts and feed extensively on the migrating fish. One can argue that more culverts will give more opportunity to catfish to set up an ambush, but it is important that the design of culverts must be addressed, in a similar fashion that the designs of fishways receive attention. It will be important to add some small structures in the culverts which can act as “substrate” for the migrating fish, i.e. some structures which will protect the mall fish from predators.
- Modification of the habitat in general is problematic. A conceded effort must be undertaken to address the problem and part of the management plan must focus on habitat restoration and rehabilitation.
- From the research done it is evident that the methodology developed for the RHP can be applied in a system such as the Nyl floodplain. As sampling is generally conducted during the low flow period in rivers, sampling techniques as used in the FAII can be used in the floodplain. In addition to the electroshocker, baited traps and small seine nets can be used to sample fish. Sampling during the high flow period was difficult as discussed earlier.

8.6 References:

- DWAF, 1986. *Management of the water resources of the Republic of South Africa*. Department of Water Affairs and Forestry, Pretoria, South Africa. 459 pp.
- HOHLS, D.R. 1996. National Biomonitoring Programme for Riverine Ecosystems: Framework document for the programme. NBP Report Series No 1. Institute for Water Quality Studies, Department of Water Affairs and Forestry, Pretoria, South Africa.
- KLEYNHANS, C.J. 1990. Grafiese voorstelling van die verspreiding van vissoorte in die Nylrivier en sytakke (Mogalakwenarivier, Limpoposisteam). Hoofdirektoraat Natuur- en Omgewingsbewing, Pretoria.
- KLEYNHANS, C.J. 1999. The development of a fish index to assess the biological integrity of South African rivers. *Water SA*. Vol. 25 No. 3. 265 – 278.
- SKELTON, P.H. 2001. A complete guide to the freshwater fishes of southern Africa. Struik Publishers. Cape Town. 395 pp.
- WEEKS, D.C., J.H. O'KEEFFE, A. FOURIE AND B.R. DAVIES. 1996. A pre-impoundment study of the Sabie-Sand River system, Mpumalanga with special reference to predicted impacts on the Kruger National Park. WRC Report No. 294/1/96.

CHAPTER 9: DEVELOPMENT OF A WETLAND ASSESSMENT PROTOCOL

Biological assessments evaluate the health of a water body by directly measuring the condition of one or more of its taxonomic assemblages (e.g. plants, macro-invertebrates, ect.) and supporting chemical and physical attributes. A major premise of bio-assessments is that biotic communities of plants and animals will reflect the health of the water body in which they live. Changes will occur in floral and faunal community structure, diversity, organism health and trophic structure after damage from anthropogenic activities has occurred (USEPA^a, 2002)

In wetlands minimal human activities will cause little effect on the biotic communities. These communities are usually very resilient and will recover quite quickly. At this point in the continuum the biological communities are said to have biological integrity. Biological integrity is said to be the ability to “support and maintain a balanced adaptive community of organisms having a species composition, diversity and functional organisation comparable to that of natural habitats within a region” (Karr and Dudley, 1981).

When the interaction of wetland plants and animals with their environment is disrupted many of the functions provided by the wetlands are lost or diminished (USEPA^a, 2002). The most direct and cost effective way to evaluate the integrity of a wetland is to directly measure the attributes of the floral and faunal communities that inhabit the wetland. Chemical endpoints are not suitable for evaluating wetlands as there are too many of them to monitor and many studies are limited to financial and staff resource constraints (USEPA^a, 2002).

Stream based bio-assessment programs have found that bio-assessment programs are less expensive than many chemical based assessment programs (Yoder and Rankin, 1995). There are many other factors that affect a system or which could be monitored to assess the biological integrity of a system. These factors are illustrated in Figure 9.1.

Bio-assessments can help prioritize where to follow up with additional monitoring, diagnosis of causes of degradation and assist in informed management decision making in the protection and rehabilitation of wetlands.

Bio-assessments are based on the premise that the biotic communities will reflect the health or integrity of the wetland system. Decreases in community structure, abundance and diversity are sure signs of damage to the system. Bio-assessment methods developed for streams and rivers can be and have been adapted for use in the monitoring of wetlands, lakes, estuaries and terrestrial systems (USEPA^b, 1998, Karr and Chu, 1999).

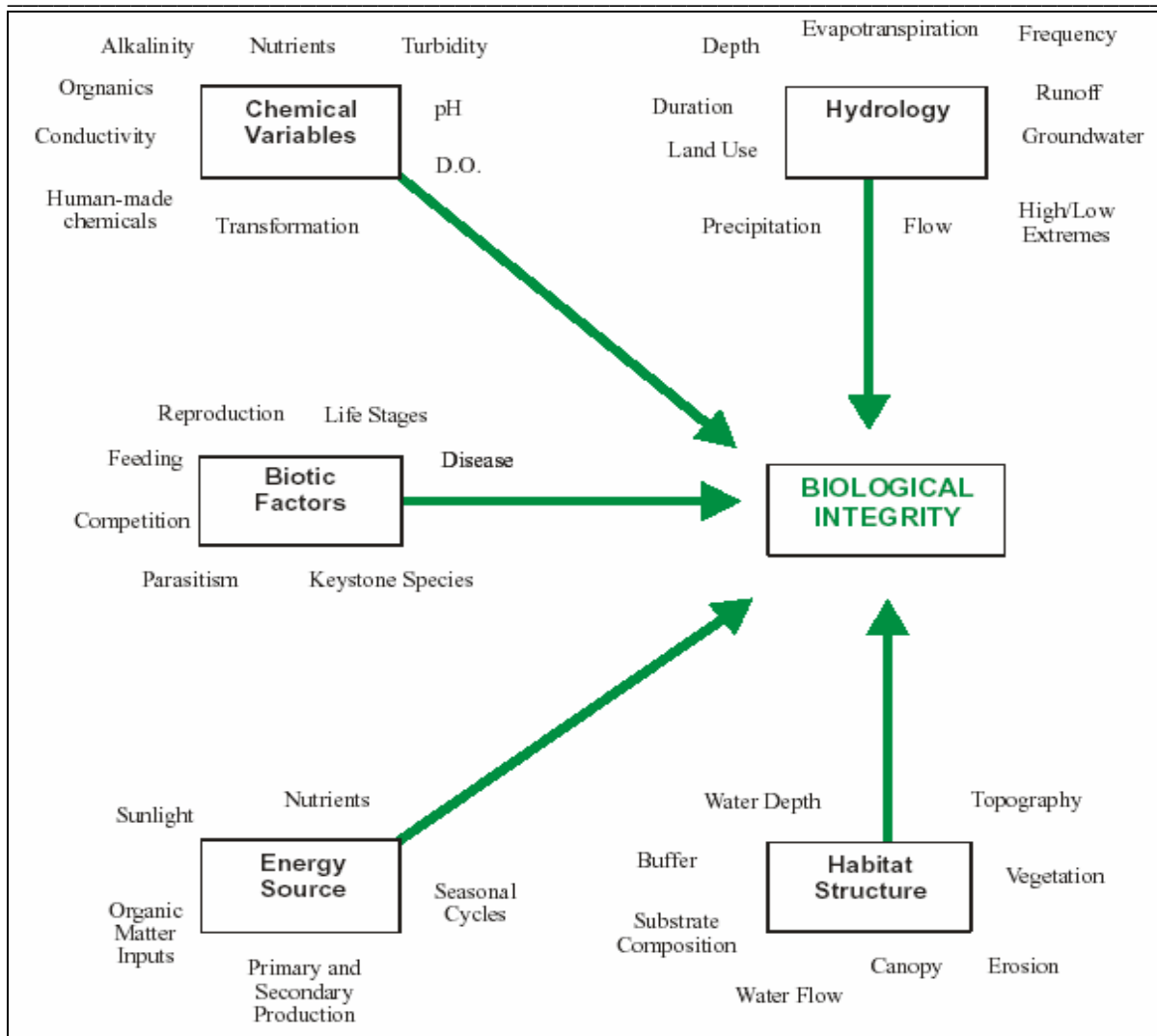


Figure 9.1: Ecosystem influences on biological integrity. (Adapted from Karr *et al.*, 1986).

The development of a wetland assessment protocol involves the following six stages of development:

1. Selection of the biological assemblage to monitor.
2. Classification of the wetland.
3. Selection of wetlands across a human disturbance gradient to monitor.
4. Sampling of chemical and physical characteristics to use in result validation.
5. Data analysis.
6. Result reporting.

The rest of this chapter will be discussed under these six headings with the aim of providing a framework for wetland assessment in the Limpopo Province.

9.1 Biological assemblages

A variety of biological assemblages can be used to assess the integrity of wetland systems. These assemblages include fish, plants, macro-invertebrates, algae and amphibians. Each of these assemblages has their own pit falls and merits. For the purpose of this assessment system macro-invertebrates were the assemblage of choice.

There are many reasons for the use of macro-invertebrates, the least of which is that they have successfully been used in the monitoring of river and stream integrity in South Africa. Table 9.1 indicates the advantages and disadvantages of using Macro-invertebrates as bio-indicators of system integrity.

Table 9.1: Table of advantages and disadvantages in using aquatic macro-invertebrates as bio-indicators of wetland integrity. (USEPA^c, 2002).

Advantages and disadvantages of using invertebrates for biological analysis of wetlands	
Advantages	Disadvantages
Invertebrates can be expected to respond to a wide array of stresses to wetlands, such as pollutants in water and bottom sediments, nutrient enrichment, increased turbidity, loss or simplification of vegetation, siltation, rearing of bait or game fish, input of storm water or wastewater runoff, introductions of exotic species, or alterations of the landscape around the wetland.	Because it is likely that multiple stressors are present, it may not be possible to pinpoint the precise cause of a negative change in the composition of invertebrates. However, data from major sources of human disturbance, e.g., water and sediment chemistry, the nearby wetland landscape features, sources of hydrologic alteration, and other disturbance factors can be assessed in relation to the invertebrate data to see which factors have the greatest effects.
Life cycles of weeks to months allow integrated responses to both chronic and episodic pollution, whereas algae recover rapidly from acute sources, and vertebrates and macrophytes may take longer to respond to chronic pollution.	Information on short-term, pulse impairments (using algae, zooplankton) or more long-term impairments (using macrophytes, vertebrates) or more landscape-level (using birds, amphibians) impairment may be desired.
Toxicological/laboratory based information is extensive. Invertebrates are used for a large variety of experimental approaches.	Toxicological response data may not be available for all invertebrates; data for some wetlands species are less extensive than for stream species.
There is an extensive history of analysis of aquatic invertebrates in biological monitoring approaches for streams.	Using invertebrates to assess the condition of wetlands is now under development in several States and organizations.
Invertebrates are used for testing bioaccumulation of contaminants to analyze effects of pollutants in food webs.	Tissue contaminant analyses are always costly. This is true for tissue analysis of any group of organisms: vertebrate, invertebrate, or plant.
Invertebrates are important in food webs of fish, salamanders, birds, waterfowl, and predatory invertebrates.	Aquatic invertebrates tend not to be valued by the public as much as fish, amphibians, turtles, or birds. However, citizens do respond to invertebrates.
Many invertebrates are ubiquitous in standing water habitats.	Invertebrate composition will differ in different wetland classes, as will other groups of organisms (plants, birds) that might be used to assess wetlands.

Advantages and disadvantages of using invertebrates for biological analysis of wetlands	
Advantages	Disadvantages
Many invertebrates are tightly linked to wetland conditions, completing their life cycles within the wetlands. They are exposed to site-specific conditions.	Some invertebrates migrate in from other water bodies; these taxa are not as tightly linked to the conditions in the specific wetland.
Many invertebrates depend on diverse wetland vegetation, some depend on particular types of vegetation for reproduction.	Loss of invertebrates may be a secondary effect from the loss of wetland vegetation, e.g., from herbicide treatments. Vegetation loss is an impairment.
Invertebrates have short and long life cycles and they integrate stresses to wetlands often within a 1-year time frame.	Many complete their life cycle within a year, they are not as "long-lived" as birds, amphibians or perennial vegetation.
Invertebrates can be easily sampled with standardized methods	Picking invertebrate samples is labor-intensive.
Invertebrates can be sampled once during the year, if the best index period is selected for optimal development of invertebrates.	Invertebrate composition of wetlands often varies within the seasons of the yearly cycle. Invertebrates mature at different times. This necessitates selecting an "index period" for sampling once, or alternatively, sampling more than once in the season.
Invertebrates can be identified using available taxonomic keys within labs of the entities doing the monitoring. Staff help develop biomonitoring programs.	Expertise is required to perform identifications of invertebrates. Some may choose to contract out some or all the identifications. There is a cost involved.
High numbers of taxa and individual counts permits the use of statistical ordination techniques that might be more difficult with just a few species, e.g. with amphibians.	Large numbers of taxa and individual counts make the sample processing more labor intensive than other groups. Adequate training and staff time are required. More lab time is needed than for some other groups of organisms.
Citizens can be trained to identify wetlands invertebrates and become interested and involved in wetlands assessment. Citizens are excited to see the richness of wetland invertebrates.	Citizen monitoring requires training to learn many invertebrates in a short time, a structured program, and a commitment by volunteers and local governments; citizens may tend to underrate high quality wetlands.

9.2 Wetland Classification

The classification of wetlands is of great importance in assessing wetlands and in being able to draw comparisons to other similar wetlands. The monitoring system must be validated in a number of different wetlands and it is important to compare similar system with differing levels of anthropogenic impacts. One of the ways to simplify the evaluation of wetlands is to classify the wetlands and only compare wetlands within the same class (USEPA^a, 2002)

There are many definitions of wetlands which are broad in nature and thus classification of the system a necessity. The overall goal of classification is to reduce the variability within classes caused by differences in natural condition related to factors such as geology, hydrology and

climate (USEPA^d, 2002). For the purposes of developing bio-assessment methods, the goal is to establish classes of wetlands that have similar biological communities and the respond to similar human disturbances (USEPA^a, 2002)

On a broad scale wetlands can be divided into the following categories. These categories are (1) swamps and marshes, (2) lakes, (3) estuaries, (4) marine, (5) riverine and (6) artificial wetlands. These classes can further be divided into permanent and seasonal classes with respect to duration of flow/ inundation (Frazier, 1999).

For the purpose of this protocol the system is being developed for ephemeral, riverine floodplains.

9.3 Wetland Selection

After wetland classification it is important to select sampling sites along a disturbance gradient. These sites are used to document how the biological assemblage responds to differing levels of human disturbance, or anthropogenic stressors (USEPA^e, 2002). The disturbance gradient used is not important, but rather the sites should be selected from minimally disturbed to severely disturbed. In selecting the sampling sites one should not become preoccupied with the disturbance gradient, as it is impossible to take every way that humans can disturb a wetland into account, but the gradient must be estimated sufficiently enough to allow for calibration of selected matrices (Karr and Chu, 1999). The expected disturbances should also be defined and described to allow for sufficient sampling at either end of the disturbance gradient (USEPA^a, 2002).

9.4 Sampling Method Selection

The sampling method selected was a modification on the method used in the South African Scoring system version 5 (SASS5). The sampling method should be simple and not require the services of highly trained personnel. Due to the nature of the habitat available in most wetlands, it was decided to sample the aquatic and marginal vegetation component in the system. This habitat is important in the system and provides the necessary refugia and food sources for the macro invertebrates to colonise. In the SASS5 protocol three habitats are sampled namely stones, vegetation and sand/gravel/mud. Due to the lack of biotope variation in wetlands, it was decided that the vegetation biotope was the most suitable biotope to sample, as it contains a larger diversity of organisms. Aquatic macro-invertebrates were sampled using a 30 cm by 30 cm 1000 micron nylon mesh net. Approximately five meters of marginal vegetation, submerged vegetation or a mixture of both was sampled, by sweeping the net through the vegetation to brush and macro-invertebrates into the net. The invertebrates were then transferred into a photographic tray or similar sort of vessel for on site identification. (Adapted from Dickens and Graham, 2002). The organisms were identified and recorded on a score sheet. Identification of organisms was aided using Aquatic invertebrates of South African Rivers: Illustrations (Gerber and Gabriel, 2002). Figure 9.2 illustrates the score sheet used during the data collection phase of the protocol. The sampler will need the following equipment which is relatively inexpensive and easily obtainable: Waders*, Sampling Net, A3 photo tray or similar container, sample bottles*, forceps*, preservation solution*, data sheets and Aquatic Macro-invertebrate identification field guide. The items marked with an asterisk are optional. Samples can be identified either in the laboratory or in the field.

SASS Version 5 Score Sheet				Taxon				Taxon				Taxon			
				S	Veg	GSM	Tot	S	Veg	GSM	Tot	S	Veg	GSM	Tot
Date: / / 200				PORFERA				HEMIPTERA				DIPTERA			
				COELENTERATA				Belostomatidae				Athericidae			
Collector:				TURBELLARIA				Corixidae				Blepharoceridae			
				ANNELIDA				Gerridae				Ceratopogonidae			
Grid Reference: WGS-84 Cape Date				Oligocheata				Hydrometridae				Chironomidae			
				Leeches				Naucoridae				Culicidae			
				CRUSTACEA				Nepidae				Dixidae			
S: " " , E: " " .				Amphipoda				Notonectidae				Empididae			
				Potamonautidae				Pleidae				Ephyridae			
Site Code:				Atyidae				Veliidae/M...veliidae				Muscidae			
River:				Palaemonidae				MEGALOPTERA				Psychodidae			
Site Description:				HYDRACARINA				Corydalidae				Simuliidae			
Weather Condition:				PLECOPTERA				Sialidae				Syrphidae*			
				Notonemouridae				TRICHOPTERA				Tabanidae			
Temp:..... pH:.....				Perlidae				Dipseudopsidae				Tipulidae			
DO:.....r Cond:.....ms/m				EPHEMEROPTERA				Ecnomidae				GASTROPODA			
Biotopes Sampled:				Baetidae 1sp				Hydropsychidae 1sp				Ancyliidae			
				Baetidae 2sp				Hydropsychidae 2sp				Bulinidae*			
SIC..... Time..... minutes				Baetidae >2 sp				Hydropsychidae >2sp				Hydrobiidae*			
SOOC..... Time..... minutes				Caenidae				Philopotamidae				Lymnaeidae*			
Average size of stones.....cm				Ephemeridae				Polycentropodidae				Physidae*			
Bedrock.....				Heptageniidae				Psychomyiidae/Xiphocen				Planorbidae*			
Aquatic veg'n..... Dom. Sp.....				Leptophlebiidae				Case Caddis:				Thiaridae*			
MvegIC..... Dom. Sp.....				Oligoneuridae				Barbarochthonidae SWC				Viviparidae* ST			
MvegOC..... Dom. Sp.....				Polymitarcyidae				Calamoceratidae ST				PELECYPODA			
Gravel.....				Prosopistomatidae				Glossosomatidae				Corbiculidae			
Sand.....				Teloganodidae SWC				Hydroptilidae				Sphaeriidae			
Mud.....				Tricorythidae				Hydrosalpingidae				Unionidae			
Hand picking/ visual observation.....				ODONATA				Lepidostomatidae				SASS Score			
Flow: Low/ Medium/ High				Calopterygidae				Leptoceridae				No. of Taxa			
Turbidity: Low/ Medium/ High				Chlorocyphidae				Petrothrincidae SWC				ASPT			
Riparian Land Use:				Chlorolestidae				Pisuliidae				Sample collection efforts exceeds method?.....			
				Coenagrionidae				Sericostomatidae SWC							
Disturbances in the river: eg. Sandwinning, cattle drinking point, floods etc.				Lestidae				COLEOPTERA				Other biota including juveniles:			
				Platycnemidae				Dytiscidae*							
				Protoneuridae				Elmidae/Dryopidae*				Comments:			
				Aeshnidae				Gyrinidae*							
				Corduliidae				Halipidae*							
Observations: eg. Smell and colour of water petroleum, dead fish, etc.				Gomphidae				Helodidae*							
				Libellulidae				Hydraenidae*							
				LEPIDOPTERA				Hydrophilidae*							
				Pyralidae				Limnichidae							
								Psephenidae							

Figure 9.2: SASS5 score sheet used for primary data collection (Dickens and Graham, 2002).

9.5 Data analysis and matrix determination.

The determination of a single matrix for the evaluation of wetland integrity is difficult due to the uniqueness of the floral and faunal assemblages as well as the ecological processes (Wissinger 1999). It is also unrealistic to expect to find a single index of wetland integrity for all wetlands, due to the wide variation in wetlands from a geographical, hydrological, wetland class and biological aspects (USEPA^c, 2002). According to Gemes and Helgen (1999) the monitoring of a number of assemblages increases the power of wetland bio-assessments. It is for this reason that a multi pronged approach was decided upon for the assessment of the Nyl River Floodplain. The three prongs of the index were Aquatic Macro-invertebrates, Habitat Quality Rating and a Land Usage Rating.

The scores obtained from the three prongs were then combined mathematically to provide a Wetland Biological Index Score or WBI.

9.6 Aquatic Macro-invertebrates.

The invertebrate families chosen to act as indicator species for the monitoring were chosen according to two criteria. The first criterion used was the determination of the families present in the system during sampling periods. Two high flow and two low flow periods were sampled using the SASS5 method of sampling and data collection (Dickens and Graham, 2002). The families identified in the vegetation biotope were then placed on a list and checked for compliance to the second criterion. The listed families were checked for habitat preference in the Aquatic Invertebrates of South Africa Field Guide (Gerber and Gabriel^a, 2002). Only species found on both lists were then used as indicator families for the WBI.

Table 9.2 indicates the invertebrate families chosen to act as indicator families in the Nyl River Floodplain System. The relative sensitivities obtained for and use in the South African Scoring System version 5 (SASS5) were used to obtain the invertebrate score.

9.7 Habitat Quality Rating

Habitat quality plays an important role in the monitoring of macro-invertebrates. Sites with a more diverse habitat will have a greater diversity of invertebrates, but this may not indicate the water quality in which they live (Pennak, 1978). It is thus important to try and negate the effects of habitat on the score obtained in the final WBI. For this reason a simple table of weighted questions was set up. Answers to five questions were assigned a value between 1 and 5 with the habitat rating score being calculated by adding the values together. Table 9.3 indicates the list of questions relating to habitat quality. Invertebrate community structures improve with an improvement in habitat quality. Weighted values were thus assigned to percentage bank cover, percentage aquatic vegetation and percentage fringing/ leafy vegetation

The weighted scores assigned fall within the following categories:

0-20%	1
21-40%	2
41-60%	3
61-80%	4
81-100%	5

Table 9.2: Invertebrate sampling list with weighted sensitivities according to the SASS5 score sheet (Adapted from Dickens and Graham, 2002).

Taxon	Sensitivity	Presence
Potamonautes	3	
Atyidae	8	
Hydrachnellae	8	
Baetidae	4	
Chlorocyphidae	10	
Chlorolestidae	8	
Coenagrionidae	4	
Lestidae	8	
Aeshnidae	8	
Belostomatidae	3	
Corixidae	3	
Gerridae	5	
Naucoridae	7	
Nepidae	3	
Notonectidae	3	
Pleidae	4	
Veliidae	5	
Leptoceridae	6	
Dytiscidae	5	
Gyrinidae	5	
Helodidae	12	
Chironomidae	2	
Culicidae	1	
Simuliidae	5	
Lymnaeidae	3	
Physidae	3	
Planorbidae	3	
Dixidae	13	
Leeches	3	
Hydrophilidae	5	
Lepistomatidae	10	
Pisuliidae	10	
Planerians	3	
Oligochaetes	1	
Elmidae	8	
	A	

These scores allow the sampler to rate the habitat with a high score denoting a good habitat quality and a low score denoting a poor habitat quality, or lack of habitat. The habitat quality rating is divided into five questions giving the habitat a score out of 25 in the end. The questions are simple but provide the relevant information as to habitat suitability for the presence of aquatic macro-invertebrates.

The five questions are:

1. Percentage right bank cover?
2. Percentage left bank cover?
3. Percentage submerged aquatic vegetation?
4. Percentage right bank cover fringing/Leafy?
5. Percentage left bank cover fringing/Leafy?

For the sampler to assess the habitat quality he/she should have an understanding of the relevant definitions.

Left or Right: the left or right hand side of the channel is determined according to the direction of flow of the river. This means that if you stand facing the same direction as to the river flow, the left bank will be on your left side and the right bank will be on your right side.

Percentage Bank Cover: defines the percentage of the bank that is covered by vegetation in comparison to bare ground.

Percentage Aquatic Vegetation: is defined as the amount of submerged or floating vegetation cover in comparison to open water and bare substrate.

Percentage Fringing Vegetation: this refers to the percentage of the bank cover that is herbaceous and leafy in nature hanging in or submerged in water. This plant cover provide better cover and more feed for the organisms than reeds, sedges and grasses do.

The habitat rating is very subjective but the broad nature of the differential percentage classes erases some of the subjectivity involved in the assessment process.

Table 9.3: Habitat Quality Rating Table.

Habitat Quality Rating					
	0-20%	21-40%	41-60%	61-80%	81-100%
% Left Bank Cover	1	2	3	4	5
% Right Bank Cover	1	2	3	4	5
% Aquatic Vegetation	1	2	3	4	5
% Left Bank Fringing Vegetation	1	2	3	4	5
% Right Bank Fringing Vegetation	1	2	3	4	5
				B	

9.8 Land Usage Rating:

Different anthropogenic land use activities can cause a decline in the macro-invertebrate community. These land use practises can be varied from mining activities, both agricultural and livestock farming activities and the construction of unnatural barriers (roads, impoundments, dams etc.). The land uses at the sites were assigned a weighted score between 0 and 3. Table 9.4 indicates the land use practises and the weighted score assigned to each practise.

Scores assigned to each activity were chosen randomly with the activity having a large impact scoring a zero and if the activity is not present the activity scores a three. The different activities were rated from sever to none. A high Land Use Rating score indicates a site with little observed impact and with a low rating score having a high level of anthropogenic impact. The maximum Land Use Rating obtainable for a site is a score of 15.

Table 9.4: Land Usage Rating Score Table.

Land Use Rating				
	Severe	Moderate	Minimal	None
Mining	0	1	2	3
Agriculture	0	1	2	3
Livestock	0	1	2	3
Urban	0	1	2	3
Other	0	1	2	3
			C	

9.9 Wetland Biological Index Score.

The three scores obtained were then placed in the following simple equation to establish the WBI.

$$WBI=A-B+C$$

Where:

A is the Invertebrate score

B is the Habitat Quality Rating

C is the Land Use Rating.

Due to the increases made on invertebrate scores by a high habitat rating it was decided to subtract the Habitat Quality Rating from the score to negate the habitats role in the invertebrate score. The land usage plays a role in the decline in invertebrate community structure so a high score means little impacts from land usage and thus plays a role in the integrity of the site. Figure 9.3 illustrates the score sheet developed for use in the wetland biological index.

9.10 Result Reporting:

The results obtained from the data sheets can then be reported with reference to weighted invertebrate scores, habitat quality and land use. The wetland integrity can also be reported with the WBI able to relate water quality of the system. The WBI does however not specify the cause of the disturbance in the invertebrate community structure but does allow management and monitors to make decisions on whether the system is being impacted or not.

The monitor should take into account that the seasons play a role in the invertebrate community structure and that WBI values may decrease during periods of low flow and extreme high flow. The system has as yet not been calibrated and compared to other wetland systems similar in nature so max values cannot be stated at this point in time.

9.11 Case study:

The Nyl River Floodplain was chosen as the wetland for this study. Six localities were chosen along the course of the Klein Nyl River with three sites inside the wetland itself and three sites in the river before the wetland.

As mentioned earlier in this chapter the Wetland Biological Index has been developed for this type of systems namely ephemeral riverine floodplains. The localities were situated along a pollution/impact gradient so as to provide the necessary information as to whether the WBI could indicate the effects of human impacts on the invertebrate community structure.

Invertebrates from the six localities were sampled using the specified protocol and the organisms were identified. The habitat was rated as well as the surrounding land uses. The results were then compared to the results obtained from the water analysis to determine if the WBI can depict if anthropogenic activities are having a negative effect on the water in the system.

Table 9.5 indicates the scores obtained at the different localities during the four sampling periods.

Date ___/___/20__

Site: _____

Sampler: _____

Taxon	Sensitivity	Presence	Taxon	Sensitivity	Presence
CRUSTACEA			COLEOPTERA		
Potaminautidae	3		Dytiscidae	5	
Atyidae	8		Gyrinidae	5	
Hydrachnellae	8		Elmidae	8	
EPHEMEROPTERA			Hydrophilidae	5	
Baetidae	4		Helodidae	12	
ODONATA			DIPTERA		
Chlorocyphidae	10		Chironomidae	2	
Chlorolestidae	8		Culicidae	1	
Coenagrionidae	4		Dixidae	13	
Lestidae	8		Simulidae	5	
Aeshnidae	8		GASTROPODA		
HEMIPTERA			Lymnaeidae	3	
Belostomatidae	3		Physidae	3	
Corixidae	3		Planorbidae	3	
Gerridae	5		ANNELIDA		
Naucoridae	7		Oligochaetes	1	
Nepidae	3		Leeches	3	
Notonectidae	3				
Pleidae	4		Planerians	3	
Veliidae	5				
CASED CADDIS:					
Leptoceridae	6			A	

Habitat Quality Rating (HQR)					
	0-20	21-40	41-60	61-80	81-100
% Left Bank Cover	1	2	3	4	5
% Right Bank Cover	1	2	3	4	5
% Aquatic Vegetation	1	2	3	4	5
% Left Bank Fringing	1	2	3	4	5
% Right Bank Fringing	1	2	3	4	5
				B	

Land Use Rating				
	Sever	Moderate	Minimal	None
Mining	0	1	2	3
Agriculture	0	1	2	3
Livestock	0	1	2	3
Urban	0	1	2	3
Other	0	1	2	3
			C	

WBI = A - B + C

A	B	C	WBI

Figure 9.3: WBI Data Sheet.

Table 9.5: Table of WBI scores calculated during study period.

Wetland Biological Integrity Index				
	August 2001	November 2001	March 2002	July 2002
Abba		88	43	53
DPD	33	62		58
Jasper		15	35	16
Nylsvley	37	42	29	42
Haakdoring	57	14	33	57
Moorddrift	37	43	54	52

The results were then compared to those obtained for the water analysis for the same period. The comparisons were made by super imposing the WBI scores onto Principle Component Analysis Plots of the water Quality at each locality. Figures 9.4 (A-D) illustrate the super imposed WBI scores on the PCR plots with the bubbles of similar size indicating water with similar WBI scores and water qualities.

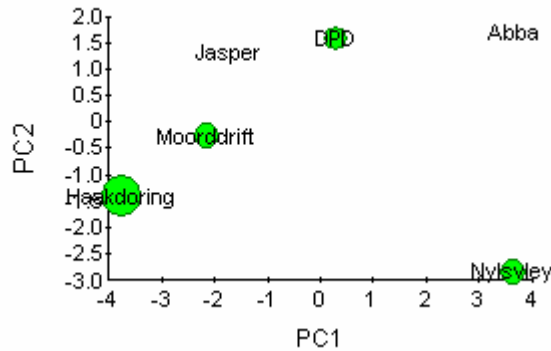


Figure 9.4 (A): Principle component analysis of Nylsvley water quality for August 2001 with the calculated WBI superimposed onto it.

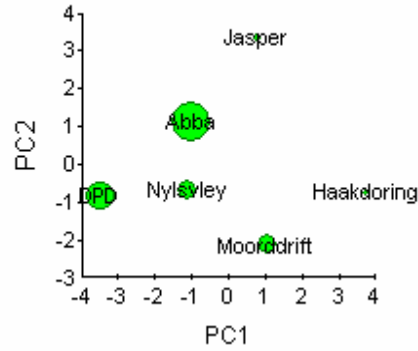


Figure 9.4 (B): Principle component analysis of Nylsvley water quality for November 2001 with the calculated WBI superimposed onto it.

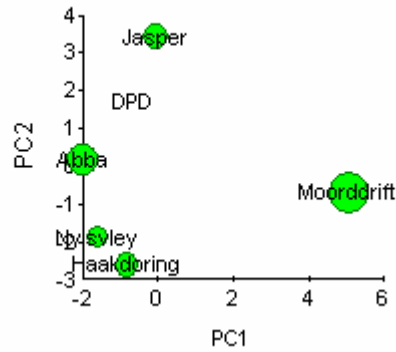


Figure 9.4 (C): Principle component analysis of Nylsvley water quality for March 2002 with the calculated WBI superimposed onto it.

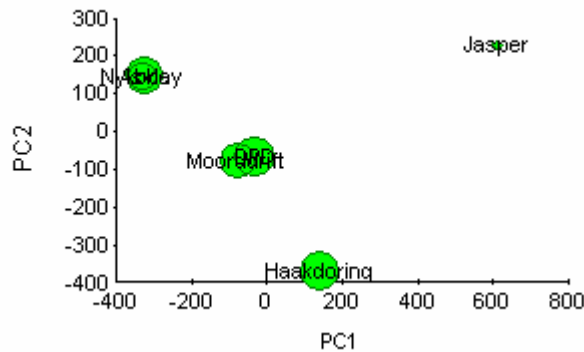


Figure 9.4 (D): Principle component analysis of Nylsvley water quality for July 2002 with the calculated WBI superimposed onto it.

The results discussed in chapter 2 indicate that for the most part the water in the Nyl River system is of a suitable quality. The water does however have bacterial contamination at the sites analysed. The results from the WBI indicate these trends that bacterial contamination is also affecting the aquatic invertebrate communities.

In figure 9.4 (A) the bubble superimposed onto the site at Haakdoring indicates that the WBI score at this site is high in relation to the other scores. The bubbles at Moorddrift, Nylsvley and DPD indicate that the invertebrate communities are similar. The large angles between Moorddrift, DPD and Nylsvley indicate that the water at these sites is not similar, but the variable/s causing dissimilarity in the water is not affecting the invertebrate communities adversely, in relation to one another. The larger bubble at Haakdoring indicating a greater WBI score and thus a healthier invertebrate community. This can be attributed to a lack of cattle in the area and the larger volume of water present during the dry conditions experienced throughout the system. No bubble values at Jasper and Abba are due to a lack of data so the WBI could not be applied at these sites.

In figure 9.4 (B) the bubble sizes at the sites indicate that the water quality at Abba and DPD are of a better quality and more suitable for sustaining Aquatic invertebrates. The increased water flow due to precipitation has led to a decrease in water quality due to increased runoff from both farming activities and farm lands. The small bubble values at both Jasper and Haakdoring indicate poor invertebrate community structure and would thus indicate a decrease in water quality. The bubble sizes at Nylsvley and Moorddrift indicate that the invertebrate communities at these sites are similar in nature and would indicate that the effects of the contamination at Jasper and Haakdoring are having less effect on the water quality, or that the water quality at these sites is improving. The poor invertebrate community structure can in all probability be as a result of bacterial contamination from coliform bacteria. This figure indicates clearly that the sites situated before Nylstroom/Modimolle have better invertebrate community structure and that Nylstroom/Modimolle is having an effect on the system. The acute angles occurring between the plotted sites of DPD Nylsvley and Abba indicate that these sites have similar water qualities.

In figure 9.4 (C) the invertebrate communities appear to be similar in nature. The acute angles between the water at five of the six sites indicate that the water is also very similar in nature. Moorddrift is the outlier with respect to water quality and invertebrate community structure indicating that the water leaving the floodplain is of a better quality than that of the water in the Klein Nyl River and entering the floodplain. This would indicate that the floodplain is functioning correctly by slowing down the water movement and removing possible contaminants from the water. March 2002 is at the end of the High flow period and the water is moving quicker than during dry months so possible contaminants are being spread throughout the system and causing the system to have similar water qualities. The size of the WBI bubbles as well as the WBI score indicates that the water invertebrate community structures are similar correlating to the similar water qualities.

In figure 9.4 (D) Jasper is the only site with a small bubble size, which would indicate that the invertebrate community at this site is under the influence of a stressor. Four of the other five sites namely: Haakdoring, Moorddrift, DPD and Abba have similar bubble sizes indicating similar invertebrate community structures. The Nylsvley Bubble size is slightly smaller than the other bubble sizes, which indicates the effects of the water from the contaminated Jasper site with a large degree of system recovery with respect to the water quality due to the increased WBI score.

The WBI indicates that throughout all sampling periods that increased levels of contamination cause the WBI scores to decrease. The clearest indication of this is the Sampling done during July 2002, with the WBI score indicating that the Jasper site is being contaminated, with sewage effluents, and this is having an effect on the invertebrate community structure.

The results indicated in the WBI indicate that the WBI scores show stressors to the system although more sampling and refinement is necessary before this index can be used as a tool to evaluate the integrity of ephemeral, floodplain wetland health. The index needs to be tested in systems with varying pollution gradients to test its sensitivity. Ephemeral wetland systems with greater pollution gradients need to be monitored using this tool to validate its efficacy.

The nature of the water quality in the Nyl River Floodplain is such that it has few contaminants thus making it not a very good system to calibrate an index of this nature. Calibration and refinements such as the addition or subtraction of invertebrate families can only take place once the Wetland Assessment Protocol has been tested on a number of systems and over a greater time frame.

9.12 References:

- DICKENS, D.W.S. and GRAHAM, P.M. (2002). The South African Scoring System (SASS) version 5 Rapid Bioassessment Method for Rivers. *African Journal of Aquatic Science* 27: 1-10.
- FRAZIER, S. (1999). Ramsar Sites Overview: A Synopsis of the World's Wetlands of International Importance. Wetlands International. pp 42.
- GEMES, M.C. and HELGEN, J.C. (1999). Indexes of Biotic Integrity (IBI) for Wetlands . Vegetation and invertebrates IBI's. Final Report to U.S.EPA. Assistance #CD 99515-01. Minnesota Pollution Control Agency, Environmental Outcomes Division.
- GERBER, A. and GABRIEL, M.J.M. ^a(2002). Aquatic Invertebrates of South African Rivers: Field Guide (1st ed.). Institute for Water Quality Studies. Department of Water Affairs and Forestry, Pretoria. pp 150.
- GERBER, A. and GABRIEL, M.J.M. ^b(2002). Aquatic Invertebrates of South African Rivers: Illustrations (1st ed.). Institute for Water Quality Studies. Department of Water Affairs and Forestry, Pretoria.
- KARR, J.R. and CHU, E.W. (1999). Restoring Life in Running Waters: Better Biological Monitoring. Washington, D.C.: Island Press. Pp 220.
- KARR, J.R. and DUDLEY, D.R. (1981). Ecological perspective on water quality goals. *Environmental Management* 5: 55-68.
- KARR, J.R., FAUSCH, K.D., ANGERMEIER, P.L., YANT, P.R. and SCHLOSSER, I.J. (1986). Assessment of Biological Integrity in Running Waters. A method and its rationale. Illinois Natural History Survey Special Publication 5.
- PENNAK, R.W. (1978). Freshwater invertebrates of the United States (2nd ed). John Wiley and Sons, New York. pp 803.
- UNITED STATES ENVIRONMENTAL PROTECTION AGENCY (USEPA^a) (2002). Methods for Evaluating Wetland Condition: # 1 introduction to Wetland Biological Assessment. Office of Water, Washington, D.C. EPA 822-R-02-014.pp 35.
- UNITED STATES ENVIRONMENTAL PROTECTION AGENCY (USEPA^b) (1998). Lake and Reservoir Bioassessment and Biocriteria: Technical Guidance Document. Office of Water, Washington D.C. EPA 841-B-98-007.
- UNITED STATES ENVIRONMENTAL PROTECTION AGENCY (USEPA^c) (2002).Methods for Evaluating Wetland Condition: #9 Developing an invertebrate Index of Biological Integrity for Wetlands. Office of Water, Washington, D.C. EPA 822-R-02-019.pp 50.
- UNITED STATES ENVIRONMENTAL PROTECTION AGENCY (USEPA^d) (2002).Methods for Evaluating Wetland Condition: #7 Wetland Classification. Office of Water, Washington, D.C. EPA 822-R-02-017.pp 35.
- UNITED STATES ENVIRONMENTAL PROTECTION AGENCY (USEPA^e) (2002).Methods for Evaluating Wetland Condition: #6 Developing Metrics and Indexes of Biological Integrity. Office of Water, Washington, D.C. EPA 822-R-02-016.pp 38.

- WISSINGER, S.A. (1999). Ecology of Wetland Invertebrates: Synthesis and Applications for Conservation and Management. In: BATZER, D.P., RADER, R.B., WISSINGER, S.A. (eds.). Invertebrates in Freshwater Wetlands of North America: Ecology and Management. John Wiley, New York. pp 1043-1086.
- YODER, C.O. and RANKIN, E.T. (1995). Biological criteria program development and implementation in Ohio. In: DAVIES, W.S., and SIMON, T.P., Eds. Biological Assessment and Criteria: Tools for Water Resource Planning and Decision Making. Boca Raton, FL: CRC Press. Pp 109-144.

CHAPTER 10: PROPOSED MANAGEMENT FRAMEWORK FOR THE NYL RIVER FLOODPLAIN

10.1 Introduction:

The chapter focuses on the main finding and recommendations during the study and is presented as a proposed framework to be used as a management plan for the larger Nyl floodplain that should include the whole catchment. The reason for this is that the system is not large, when compared to other systems in the area and any impact, how limited or insignificant to the observer, can have a huge negative result for the whole of the system. The spillage of chemicals into any of the rivers feeding water to the floodplain can serve as an example. Such an event can have a catastrophic result with regard to the biota of the system, including the birds, fish, macro-invertebrates and the frogs.

Two summaries of the impacts and issues were made. In table 10.1, Greenfield (2004) gave a summary of impacts that have an influence on the Nyl River floodplain. The recommendations from each section of the study was used to compile the “Matrix of issues” in table format (Table 10.2) which highlights the critical issues to be further investigated or considered as important aspects to guide managers, when a more comprehensive “Management Strategy for the Nyl floodplain and its catchment” is compiled. The strategy must have a holistic approach and must include all tributaries feeding water to the floodplain. In each of the smaller catchments, various activities, land uses and structures are compromising the integrity of the system. An example is the large number of impoundments in the Tobiasspruit which has reduced the contribution of water to the floodplain to virtually no water at all during an average rain season. On the floodplain itself, habitat modification has various negative impacts. Here one can list the number of roads and the railway line cutting across the sensitive habitat, as the most destructive.

One understands the need for this type of infrastructure development, but the design of the roads and railway line are really poor. Very few culverts are present, to ensure a free flow of water during the rainy season, resulting in a “backing-up” of water behind the roads. This leads to an increased build-up of nutrients in the system which eventually results in eutrophic conditions. If the system was not restricted, some flushing of the system would have been possible. It is a well known fact that the wetlands act as nutrient traps and this point is acknowledged. The problem in the Nyl floodplain is that the historic export of nutrient by herbivores in particular, is not as effective as it was when large migrating herds utilised the floodplain for short periods during the crossing of the Springbok Flats. Even fish contribute to this process as they were food to large numbers of migrating birds. The numbers of birds have dwindled over the last decades simply because the fish numbers are lower. One reason for this, is the habitat modification taking place due to the large number of dykes and berms channelling water away from the extensive floodplain areas and restricting it in many areas to a much narrower channel.

It is evident that an approach to the management of the floodplain will need to be well planned and holistic, to ensure that the habitat degradation is curbed, that all role players understand the impact of pollution on the system and that water recycling receives a high priority. Water abstraction must be addressed, as this will ensure that more water is available to sustain the fragile floodplain environment. If this does not happen, the whole system is threatened as biodiversity will be lost – this will include rare and endangered plants and plant communities, birds, frogs and the fish.

Water quality management must be improved as the high bacterial loads during certain periods, can lead to huge problems for people relying on the system for their basic water supply. The various Councils responsible for the treatment of sewerage must have this as a highest priority. From the results of this study it was evident that bacterial counts reached critical values on various occasions. Again, in a relatively small river system, the dilution effect is not sufficient to counter these kinds of breeches of protocol and attention must be paid to ensure that water effluent is of the highest quality at all times.

10.2 Why a management framework?

- We need to monitor the floodplain.
- There is a need to understand the functioning of the floodplain and the associated biota and interacting activities.
- Plans must be formulated (specific actions to be employed) to address issues of concern raised from this and other studies.
- It must be a dynamic process with regular feedback, evaluation, and modification of the “Management Framework” if needed.

The National Water Act (Act 36 of 1998) state that we are obliged to protect the water resources, firstly from pollution and secondly as part of the management strategy of natural resources. This fact is highlighted because the Nylsvley Nature Reserve (NRR) is a registered Ramsar site and requires further actions to protect this important wetland. From this study it became clear that the protection of the NRR is not sufficient, as the reserve is situated at the top end of the floodplain area. A larger, more inclusive protected area is needed to ensure that endangered plants, birds and fish are sufficiently protected. This can only be done, if the whole catchment receives more attention. For this reason it is suggested that the whole floodplain be included, from Modimolle to Mokopane, as part of a floodplain reserve. Further, it will be important to add the catchments of all the rivers and streams feeding water to the floodplain into this protected area. It can be a formal reserve, conservancy or landowners can form informal agreements to be part of the larger Nyl Floodplain Reserve.

To ensure its success, a formal plan is needed to guide managers, landowners, government agencies, municipal planners, developers and non-government agencies in their decision making processes. If the framework is to be a successful and effective document, one must develop criteria to guide all role players with respect to the following aspects:

- The importance of wetlands.
- Why monitoring?
- What will be monitored?
- Protocols for monitoring and databases to capture historic and new information.
- What will happen to this information?
- Who will manage it?
- “Thresholds of Probable Concern” (TPC’s) must be developed. Who will be responsible for this phase?
- What actions will be taken if the “red flags” (TPC’s) are raised?

All of these issues must be captured in the management plan for the Nyl Floodplain and its associated river catchments. A task team must be put together by the role players to facilitate this process and a programme for the successful implementation of a Management Plan must be formulated.

As proposed by Greenfield (2004), a four-phased programme is needed:

- Phase 1 - *The Planning Initiative*. This is the planning phase intended to build the management framework for identifying and solving problems and defining the steps in the decision-making processes.
- Phase II - *Characterisation and Problem Definition*. The goal of the system characterisation is to gather and summarise the existing knowledge concerning the state of the wetland, as well as the physical, chemical, and biological factors focusing on identifying existing and potential problems and exploring probable causes of such problems. Such cause-effect linkages between human activities and environmental changes provide the public and decision makers with the information necessary to develop priorities, set management strategies, and devise mitigating measures.

However, information gaps do exist and additional sampling and a subsequent monitoring programme can help to fill such information gaps.

- Phase III - *Development of a Comprehensive Conservation and Management Plan (CCMP)*. The CCMP is a major product of the Nyl Floodplain Programme and can be developed by a management conference to (1) summarise findings, (2) identify and prioritise problems, (3) determine environmental quality goals and objectives, (4) identify action plans and compliance schedules for pollution control and resource management and (5) ensure that designated uses of the wetland are protected.
- Phase IV - *CCMP Implementation*. All the interested parties should establish a committee to co-ordinate the implementation of the CCMP. The development of a monitoring programme to evaluate the effectiveness of actions specified in the CCMP is a required task of the management conference.

The update and adaptation of goals of the Management Plan is a critical component of this whole initiative. As more knowledge become available, each aspect and facet of the plan must be re-evaluated, to ensure that the plan is still effective. The phased layout of the proposed plan, indicates a sequential approach to the programme and in essence one phase must be completed before the next can be initiated. Coordination of a programme such as this will be critical and as Greenfield (2004) suggested, where possible, activities must overlap. If the one phase is in progress and clear activities or actions are identified, this can give rise to a simultaneous action and effective communication will be needed to guide the committee in this regard.

Greenfield (2004) proposes the following model (Fig 10.1) that can be used for this project. The process involves five steps:

- **Step 1. Develop monitoring objectives and performance criteria.** Once the clear objectives and corresponding performance criteria are developed for each component of the monitoring programme, the basis for the model is in place. The performance criteria are an indication of the changes or trends that the monitoring programme must be able to detect.
- **Step 2. Establish testable hypotheses and select statistical methods.** For each of the study objectives statistically testable hypotheses must be formulated. The advantage of this process ensures that the result of the monitoring programme will be unambiguous and that the objectives of the programme are met. A well defined hypothesis further guides the development of statistical strategies to determine sample locations and sampling times or frequencies and the determination of the statistical analysis or tests that will be used to analyse the data.
- **Step 3. Select analytical methods and alternative sampling designs.** The next important aspect of the programme is to develop detailed specifications for each monitoring variable (measurable endpoint) of the monitoring programme (e.g. sampling and laboratory procedures). As part of the design an alternative sampling design that specifies the number and location of stations must be devised.
- **Step 4. Evaluate expected monitoring programme performance.** The expected performance of the initial sampling design must be evaluated, as this is critical to determine the minimum difference that can be detected over time or between sampling localities. If this is not done, there is a risk of not collecting and analysing enough samples, to detect statistically significant temporal or spatial trends, or that an excessive number of samples with the associated high costs is analysed. The information generated should be constantly re-evaluated, as it will enable the project team to modify and improve the model or management plan.
- **Step 5. Design and implement data management plan.** In most cases, the constant updating of the management plan, and the development of a data management system, are neglected during the design and management of monitoring programmes. Greenfield (2004) rightly mentions that the data management system should be operational prior to

implementation of the monitoring programme. In this part of the programme, the data analysis methods and a timetable for analysing the data for assessing of the Comprehensive Conservation and Management Plan (CCMP) implementation progress results should be specified. As indicated in the model, the results of the performance assessment are used to refine programme objectives (loop back to the first step).

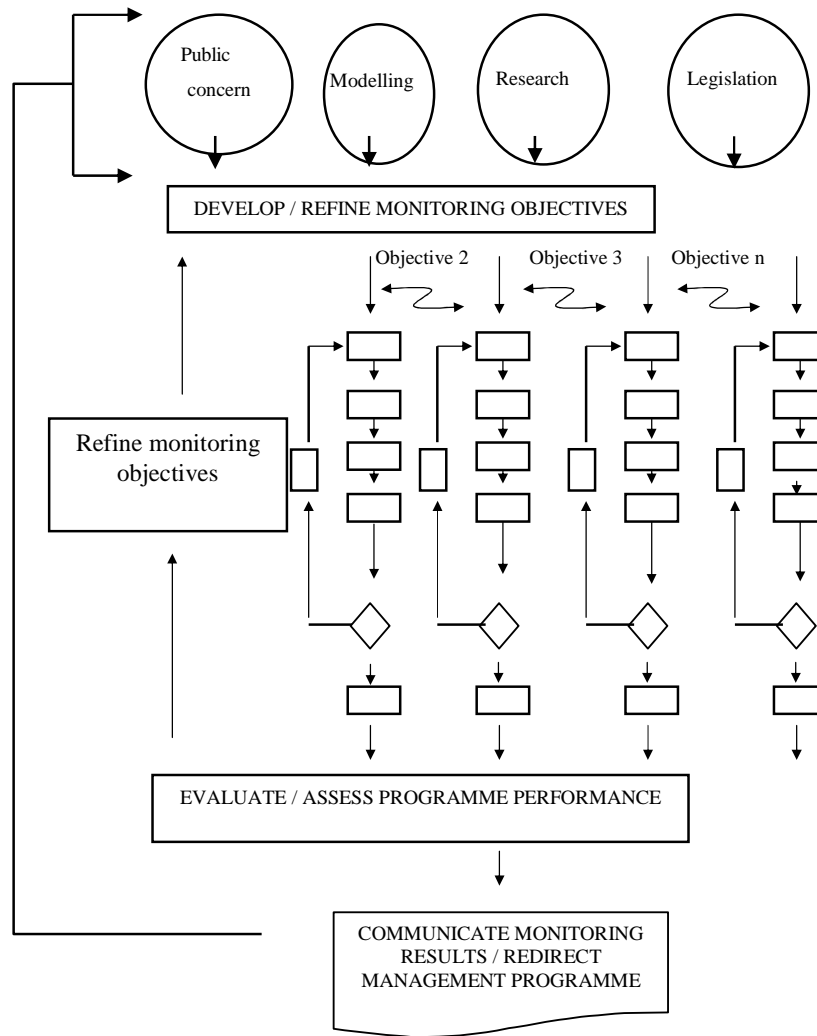


Fig 10.1: The conceptual framework to be used when designing a monitoring programme (Greenfield, 2004).

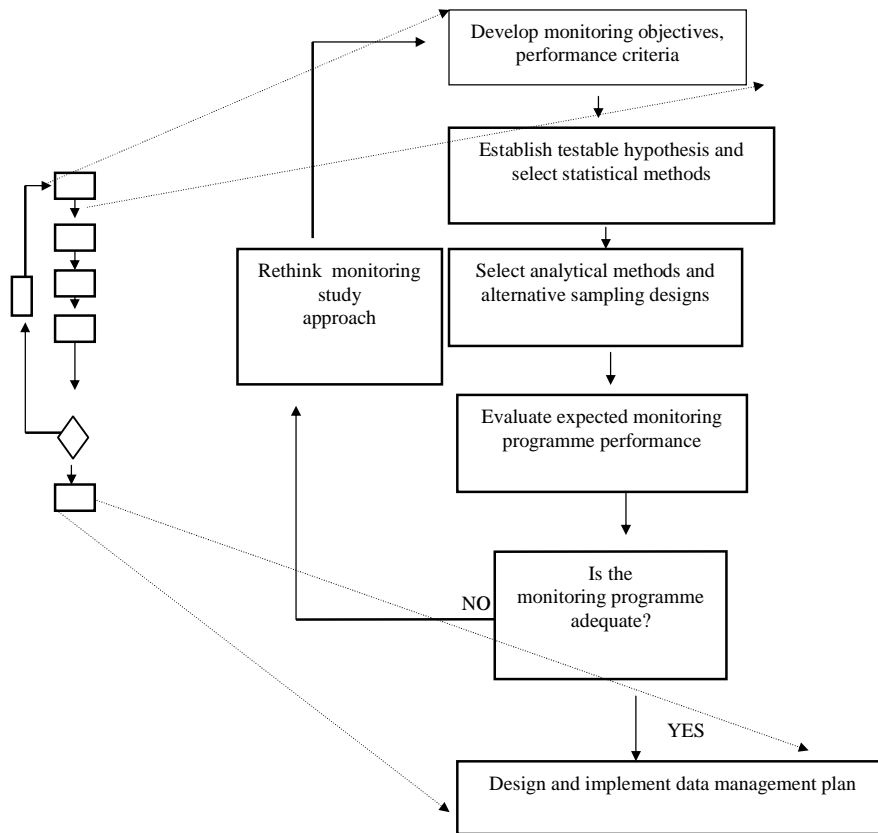


Figure 10.2: The suggested individual steps involved when designing each component of the Nyl River Catchment Management Plan (Greenfield, 2004).

In a large system such as the floodplain and its tributaries, it might be valuable to have more subunits into the framework model i.e. each catchment or tributary as a model, feeding information into the larger model. The reason for this, is that each subunit may have specific or unique issues or problems that need attention. These issues can be overlooked or lost if it forms part of the large floodplain system. As an example one can take the habitat modifications in the system. Although it is a problem in the whole system, habitat modifications in the Tobiasspruit is of particular importance. In this system, a large number of dams and weirs (between 12 and 15) have a huge impact on the Tobiasspruit in particular. Compared to the Bad-se-Loop where there are very few dams or weirs, the management strategy will be different. The issue will be addressed in the larger “Management Document”, but a specific strategy must be developed to address the problem in the Tobiasspruit.

In Table 10.2 we give a summary of the most important variables identified during the study and this can be used as the “objectives” for such a comprehensive document. Each of these issues must be analysed and transferred into the framework document as suggested by Greenfield (2004). The issues must be monitored and for each of these some “red flags” or Thresholds of Probable Concern (TPC’s) must be developed.

To illustrate the process, we will discuss one example from Table 10.1. Once the monitoring programme is ongoing, some of the objectives may show little variation and in a situation like this, the long-term impacts must be considered. A well designed management plan must

have the necessary “quality control” components incorporated, to ensure that the stable variables (those with little fluctuations) and resource wasting, is not part of the programme.

From his work, Greenfield (2004) compiled a table to highlight the impacts on the floodplain. Table 10.1 is an example of the impacts and the issues raised in the matrix, Table 10.2, must be used in conjunction with this summary. One of the most worrying factors currently affecting and threatening the floodplain is habitat modification. This includes the construction of weirs, dams and berms to divert water flow, roads and railways crossing the floodplain and utilisation of the floodplain for various agricultural activities.

In most of the rivers and streams feeding the floodplain, various structures were erected over the years, all extracting water destined for the floodplain. Most of these were constructed as weirs to draw water for household use, water for livestock and irrigation of crops. In many cases, the agricultural activities have either seized, or other sources such as groundwater are used, leaving the weirs just as water sources used for fishing.

Table 10.1: A summary of impacts that have an influence on the environment of the Nyl River Floodplain system.

Valued ecosystem components Sources of perturbation	Wetland	Phytoplankton	Zooplankton	Riparian vegetation	Marginal vegetation	Aquatic vegetation	Aquatic invertebrates	Pelagic fish	Demersal fish	Fish eggs and larvae	Aquatic birds	Human health	All
Flow regime disturbances													
Blooms\invasions													
Ecological interactions													
Wastewater outfalls													
Sewage effluent discharges													
Rivers\storm runoff													
Sport fishing													
Habitat loss\modification	3	1	1	2	2	1	3	3	3	2	4	4	3
Chemical spills													
All													

A key to illustrate the importance and level of understanding of the impacts as listed in Table 10.1.												
Potential importance				Understanding								
Controlling 1	Major 2	Moderate 3	Some 4	High	Moderate	Low						

10.3 Example of the implementation of the “Management Framework”.

When one develops a management plan for the Nyl floodplain, the first stage should involve the development of monitoring objectives to steer the management committee. In this instance when looking at the habitat modification and rehabilitation thereof, the following objectives were identified:

- a survey to determine the habitat modification of the floodplain and its tributaries,
- possible mitigating strategies to improve or rehabilitate the floodplain and its tributaries
- and future management strategies to minimise or prevent further habitat modification.

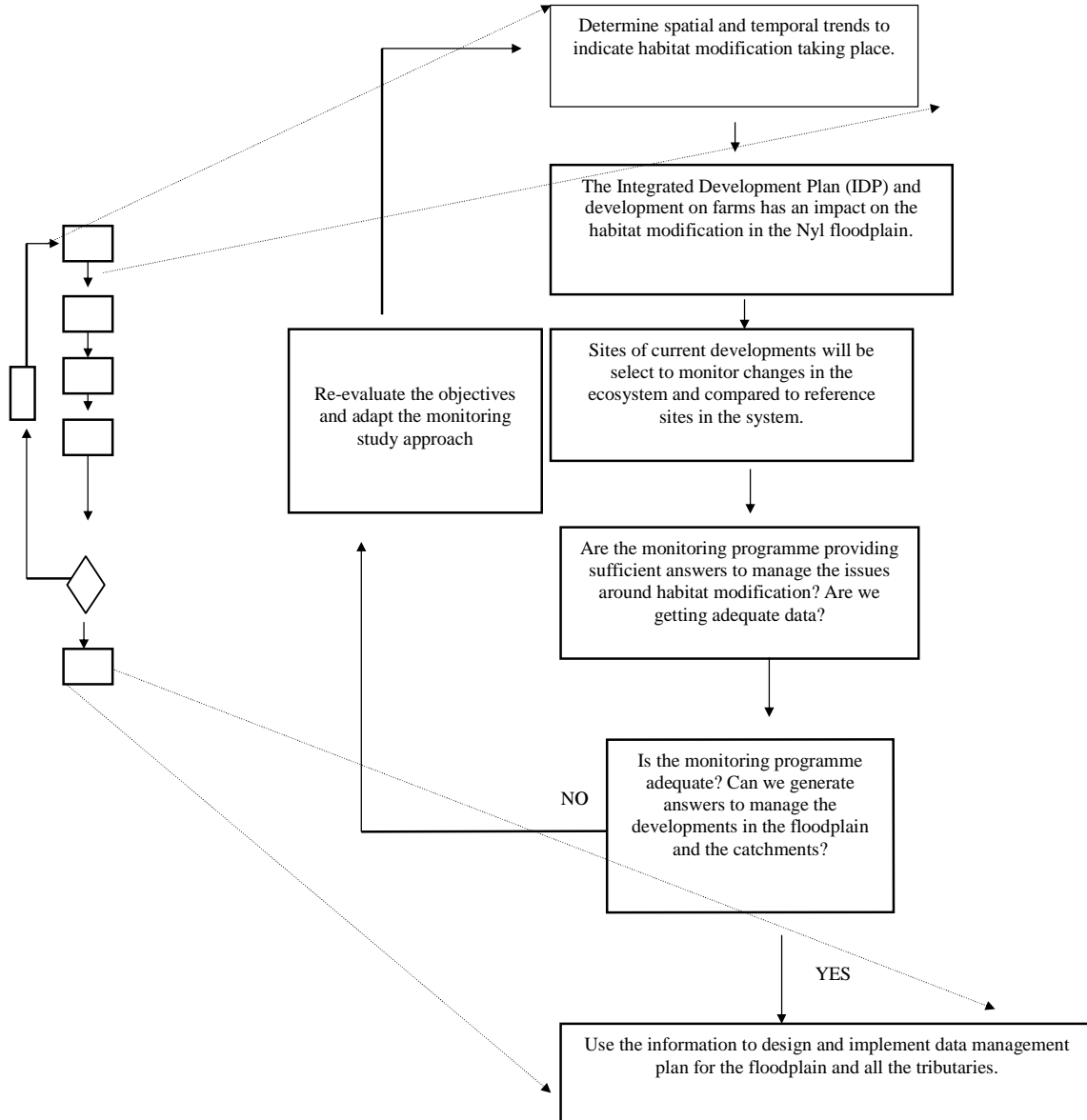


Figure 10.3: Incorporation of the identified habitat modification issues in the Nyl floodplain and its tributaries into the biomonitoring framework.

In figure 10.3 the issues to be incorporated into the management plan is refined. The first part of the strategy will be to do a detailed survey to determine all aspects related to habitat modification and the trends related to the impacts on the environment. Various expert opinions will determine changes effected over time and how this changed the system and related populations. Once these impacts have been identified and quantified, the rehabilitation options must be considered. This must form part of the strategy to when monitoring changes and improvements to the environment and related biota. The actions must form part of the larger Integrated Development Plan (IDP) for the region. With the current pressure by various development activities in the region, a well researched plan must be developed involving all role players.

The emphasis is on wealth creation - which it is an important aspect - yet, it can not be to the detriment of the sensitive environment and limited natural resources, such as water.

Once the broader strategy (IDP) is in place, the formal monitoring programme must be developed. Part of the process will be the development of indicators, referred to as the “Thresholds of Probable Concern” (TPC’s). For each TPC, an action or mitigating strategy must be developed to guide the managers to rectify threats to the system. The environmental indicators will act as red flags in the monitoring programme and once any of the TPC’s has been breached, the specific action or mitigating action will be employed. Regular reviews of the system, its functioning and rehabilitation will determine if the objectives of the programme are still relevant. If not, the plan or strategy must be revised, indicating that this “Management Plan” must be a dynamic process.

As more data is generated, the objectives and mitigating actions can be refined. The strategies for the larger floodplain and its tributaries will differ, as the impacts in each of the subsections are different. Yet, the overall strategy will be the management of a healthy floodplain for generations to follow.

In figure 10.4, the more specific impacts for the floodplain are listed. Here the focus is on the specific structures constructed over the years to divert or channel the water. A more detailed study of the weirs and dams will give detail on the way in which migration of fish is disturbed and how this can impact on the reproduction of the various species. Once the different impacts are listed and mapped (GIS), a strategy can be formulated to rehabilitate the system.

Again, a “multiple system approach” must be followed, as the impacts and mitigating strategies for the main stem of the Nyl River - and its tributaries, will be different. Once the rehabilitation programme is finalised, monitoring of water flow, flood frequencies, fish movement and numbers, plants, macro-invertebrates, frogs and birds, will be critical to determine the effectiveness of the rehabilitation programme. The information will be critical to guiding managers and landowners in the rehabilitation process, the successes and the improvement of the environment. The data generated will be used to refine the management strategy and the TPC’s, as this dynamic process will need to be updated on a regular basis.

The process can be further scaled down. If the need arises, a similar process can be developed for smaller subunits in the different catchments in the system. Depending on the complexity of any catchment, it might be necessary to develop a strategy for a specific farm or each farm in the area. This will mean that the landowner in conjunction with the forum can decide on a specific strategy to rehabilitate the habitat on his property, to implement a monitoring programme and to use the data generated to manage the specific farm. Again, management of the smaller unit will form part of the larger strategy (goals and objectives), but will be very specific for the conditions encountered on the property.

As seen in table 10.1 and table 10.2, the issues related to habitat modification are very complex. If one wants to tackle the issues as a single problem, failure will be guaranteed. As suggested earlier, it will be important to break the system into smaller components to ensure that each smaller tributary is managed as a separate unit. The overall goals of the larger floodplain will still be the driver to guide the rehabilitation of the system as a whole. In each of the smaller subunits, different issues will be drivers that must be addressed. In the Olifansspruit for instance, water quality and invasive plants infestations may rank as the top issues, whilst water abstraction and dams walls and weirs may be the critical drivers in the Tobiasspruit. The other issues will have an impact on the tributary, but may not be as important in that system (e.g. water quality in the upper reaches of the Tobiasspruit).

Once the issues for each tributary have been identified, the process of developing a management strategy and monitoring programme can be developed. Regular feedback will be critical to ensure that maximum effectivity will ensure the best outcomes for the system as a whole. To get the water flow regime restored, part of the process will involve detailed study to determine historical flow and flood regimes (reserve determination). All barriers, berms and weirs in the Nylsvley Nature Reserve must be documented and its functions must be determined. All structures not needed, must be rehabilitated. A long term plan must be formulated to upgrade roads and railways, crossing the floodplain.

It is important to upgrade the flow through these structures, as currently it restricts flow and migration routes for fish. It was suggested that the breeding sites for frogs around the floodplain must be rehabilitated too ensure that more food is available for migrating water birds.

In figure 10.3, we see a summary of what the components for a “management plan” should include. The issues must be incorporated in the IDP and this will drive further developments in the area. As part of the rehabilitation of the floodplain, it will be critical to ensure that the current structures are mapped. Each of these structures must be evaluated to determine what the impact of the structure is and what its functions are. If the structure is deemed to be “non functional”, it must be removed. This is the way in which we foresee the management plan will use the different components, to ensure that the system as a whole will benefit.

In figure 10.4, it is obvious that the issues listed for the Nylsvley Nature Reserve (NNR) are different for the larger floodplain area. The issues will be addressed for the specific subsystem and the results will feed into the larger management strategy.

Through this example, one can see that certain aspects are very complex. If this is compared with the example listed by Greenfield (2004) where he looked at the bacterial pollution on the floodplain, a more uniform approach is followed for the entire system. This illustrates the importance of the planning of the management of the Nyl floodplain and the rivers in its catchment.

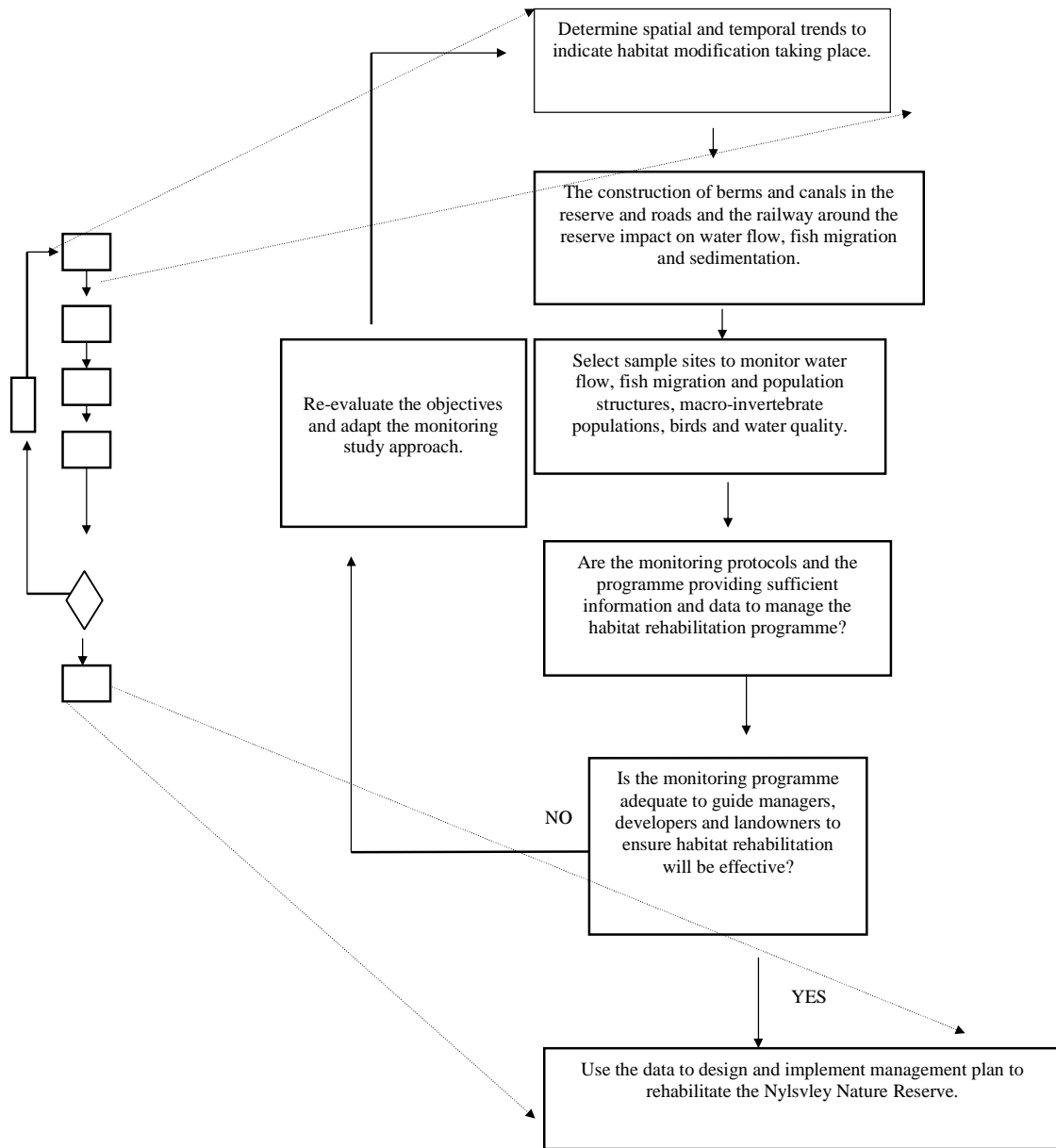


Figure 10.4: Impacts of habitat modification in the Nylsvley Nature Reserve.

Table 10.2: Matrix of management issues to be addressed in the detailed Management Plan.

	Description	Indicators	Means of verification	Mitigation
Objectives	The management of the Nyl floodplain to ensure its long term conservation.	Any further deterioration in water quality, numbers in biota and habitat degradation.	TPC’s in the monitoring programme must be determined.	Immediate action as part of the “Management Strategy”.
Specific objectives	<ul style="list-style-type: none"> • Conserve the Nylsvley Nature Reserve – maintain its Ramsar status. • Include the larger floodplain into a “conservation area”. • Conserve the rivers and its tributaries as part of the Nyl floodplain conservation area. • Ensure that effective education and awareness programmes are implemented to educate all role players. 	<ul style="list-style-type: none"> • Breach of TPC’s can lead to status being threatened and lowering in biodiversity in general. • The catchment as a whole must be the management unit. • A holistic approach will ensure a more coordinated management effort. • The lack of knowledge of the broader population in wetland conservation must be addressed. 	<ul style="list-style-type: none"> • TPC’s in the monitoring programme must be determined. Management of changes will be directed by management strategy. • See first bullet. • See first bullet. • Buy-in into management strategy and effort to lower pollution. 	<ul style="list-style-type: none"> • Compile a comprehensive management plan and strategy. • Get landowners to form conservancies. • Landowners buy-in – conservancies and management strategy. • Must form part of the management strategy as this will enhance the broad based effort in the conservation of the floodplain.

	Description	Indicators	Means of verification	Mitigation
Specific objectives (Continued)	<ul style="list-style-type: none"> Development of a long term monitoring programme focussing on water quality (including sediments), fish, macro-invertebrates and plants (bioaccumulation as part of the monitoring), frogs, birds and diatoms and regular evaluation of results, leading to an adaptive management strategy. 	<ul style="list-style-type: none"> Irregular or poor monitoring lead to useless data that can not be used to predict or highlight changes in trends and this will ensure that an adaptive management strategy can be implemented. 	<ul style="list-style-type: none"> See first bullet. 	<ul style="list-style-type: none"> Regular monitoring will give an early warning system linked to “Thresholds of Potential Concern” (TPC’s) and this will guide managers to specific actions. Regular monitoring will feed information into the management strategy for updates and changes in actions.
Water quality <ul style="list-style-type: none"> Monitoring Pesticides, solid waste and pollution Effluent monitoring Cooperation from polluters Recycling of water 	<p>Implement an effective and well managed water monitoring programme.</p> <ul style="list-style-type: none"> Identify potential sources of pollution. Monitor effluent from these sources. Get buy-in from “polluters” to minimise pollution. Investigate the use of recycled water and limit effluent into the Nyl system. 	<ul style="list-style-type: none"> Changes in trends and breach of TPC’s. Points were changes are most profound. TPC’s. Cooperation and adherence to permit conditions. Increase recycling should lower pressure on current resources. 	<ul style="list-style-type: none"> Regular monitoring and testing of water quality. Site visit to “ground truth” potential sources. Regular testing of water samples. Cooperation and self-regulation. Measuring of run-off and compare to previous data. 	<ul style="list-style-type: none"> Will give parameters to operate in and plans if TPC’s are exceeded. Will assist mangers to act early against polluters. TPC’s and early actions. Through education, pollution can be lowered. Need to improve this must be of national priority, will reduce pressure on limited resources.

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Education, awareness and training • Artificial damming and flow regime 	<ul style="list-style-type: none"> • Develop awareness/ education programme to sensitise all role players about potential pollution. • Increase in nutrients and pollutants in water and sediment. 	<ul style="list-style-type: none"> • An effective programme will alleviate pressure on water resources and lower pollution. • Water quality and sediment monitoring. 	<ul style="list-style-type: none"> • Participation and changes in attitudes of role players. • Use baseline data and information collected over the long term. 	<ul style="list-style-type: none"> • Understanding the issues will help role players to be active in protection of resource. • Increase baseline flow during high flow periods Promote flushing of the system
<p>Plants</p> <ul style="list-style-type: none"> • Monitoring • Red data and threatened species lists. • Comprehensive management plan for the catchment. • Education, awareness and training • Alien and invasive plants • Rehabilitation and riparian restoration 	<ul style="list-style-type: none"> • Develop effective monitoring index for floodplain plants. • Compile list of threatened species. • Compile strategy to conserve these plants. • Develop awareness/ education programme to sensitise all role players about plants, there role and its conservation. • Put effective alien/invasive plant eradication programme in place –awareness/education. • Develop programme to protect the riparian zone and 	<ul style="list-style-type: none"> • Changes in trends during regular monitoring. • Changes in the status of all species to be monitored. • Comprehensive strategy as part of the management plan. • Increased understanding of communities will be measure of success. • Part of the larger awareness/ education process. • Regular monitoring will 	<ul style="list-style-type: none"> • Guidelines and TPC’s as part of management strategy. • Regular verification of status of plants and/or populations. • TPC’s and management goals. • Feedback from groups and conservation authorities – are people changing? • Regular monitoring and comparison to historical maps of infestations. • Monitoring – are the riparian vegetation in 	<ul style="list-style-type: none"> • Get an early warning system in place with TPC’s as guidelines. • Will help to set TPC’s and management strategy. • Understanding of actions, activities and needs will enhance efforts on all levels. • Protection of indigenous plants, more water available and improvement of habitat in general. • Critical follow-up on alien vegetation clearance to protect habitat – limit erosion. • Will help in habitat

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Herbicides and pollution • Changes in flow regime • Fire • Erosion 	<p>rehabilitate areas where plants have been removed.</p> <ul style="list-style-type: none"> • Determine impact of herbicides and pollution on red data and threatened species. • Determine the changes in flow and the impact on plant communities and species. • Determine impacts of regular and uncontrolled fires on plant communities. • Impact of alien vegetation, clearing of alien plants, destruction of riparian vegetation and over grazing. 	<p>guide managers and landowners.</p> <ul style="list-style-type: none"> • Monitoring of populations. • Monitoring and mapping of changes. • Use indicator species and monitor changes in populations. • Monitoring and awareness. 	<p>good condition?</p> <ul style="list-style-type: none"> • Regular verification and comparison to historical and/or baseline data. • Rigorous monitoring programme. • Regular monitoring and education and awareness. • Regular follow-up in problem areas and awareness. 	<p>restoration and limit potential erosion.</p> <ul style="list-style-type: none"> • Education and training in use and application of herbicides. • Early action on TPC’s and other indicators – include education and awareness. • Introduce fire programme as part of the larger floodplain system – education, awareness and buy-in from all role players. • Management strategy to combat problems and support to assist all role players.
<p>Invertebrates</p> <ul style="list-style-type: none"> • Refine monitoring protocol • Education, awareness and training 	<ul style="list-style-type: none"> • Develop the “SASS for floodplains” further, as it must take the limited biotope diversity into account. • Develop awareness/ education programme to sensitise all role players about the importance 	<ul style="list-style-type: none"> • Regular monitoring and TPC’s. • Increased understanding of communities and increase in diversity will 	<ul style="list-style-type: none"> • Regular monitoring and evaluation of results will be indicators of success. • Are people aware of impacts on the biota and the system? 	<ul style="list-style-type: none"> • The TPC’s will act as warning system to managers. • A good understanding by all role players will enhance protection of the

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Water quality, pollution and species diversity • Habitat improvement • Exotic biota 	<p>of the macro-invertebrates, their role in the system and the impact of pollution and habitat destruction on them.</p> <ul style="list-style-type: none"> • Monitoring and increase knowledge base. • Changes in flow as a result from dams, berms and channels. • Introduction of exotic plants and animals can impact of the indigenous communities. 	<p>be measure of success.</p> <ul style="list-style-type: none"> • Changes in community structures. • Artificial barriers and lack of sufficient culverts. • Local extinctions and habitat modification. 	<ul style="list-style-type: none"> • Improve current knowledge base. • Improve understanding of flow dependence of invertebrates. • Monitoring and improvement knowledge base. 	<p>floodplain as a unit.</p> <ul style="list-style-type: none"> • Improve toxicity base and understanding for invertebrates – especially indigenous species. • Education & awareness – get role players to improve or remove structures and make rehabilitation programme part of the management strategy. • Educations and awareness to prevent new introductions and control programmes of existing problems.
<p>Fish</p> <ul style="list-style-type: none"> • Monitoring • Education, awareness and training 	<ul style="list-style-type: none"> • Implement a monitoring programme. • Develop awareness/ education programme to sensitise all role players about the importance of fish in the system, the 	<ul style="list-style-type: none"> • Regular monitoring will show changes in trends – TPC’s will dictate actions. • Changes in attitudes and more responsible actions will be indicative of understanding and caring 	<ul style="list-style-type: none"> • Compare to historic data and regular workshop on all aspects to guide managers. • Discussion with managers and conservation officials will indicate perception 	<ul style="list-style-type: none"> • TPC’s will be guidelines to actions. • Regular updates and information sharing will be critical – keep role players involved.

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Water quality and pollution • Artificial damming and flow regime • Introduction of exotic/alien biota 	<p>impact of habitat destruction and pollution on the communities.</p> <ul style="list-style-type: none"> • Changes in water quality impact on physiological processes in fish – especially reproduction and growth. • Changes in flow regime and habitat destruction. • Impact on habitat (alien plants) and communities by exotic fish (include parasites and diseases). 	<p>for the environment.</p> <ul style="list-style-type: none"> • Changes in reproductive success and health of populations. • Limited spawning and problems with predation in limited “migration” routes – barbel waiting for migrating fish in limited culverts. • High predation and poor recruitment and health problems. 	<p>and attitudes.</p> <ul style="list-style-type: none"> • Regular monitoring and effective protocols to evaluate. • Monitor improvements to problem areas. • Regular monitoring and expansion of knowledge and data base. 	<ul style="list-style-type: none"> • Education and awareness and stringent action against polluters. • Improve structures identified as problems, monitoring and education and awareness. • Education and awareness and control programmes. Prevention of new introductions is critical.
<p>Frogs</p> <ul style="list-style-type: none"> • Monitoring • Local farmers • Training 	<ul style="list-style-type: none"> • Implement a monitoring programme for frogs. • Involve farmers in the conservation of breeding sites away from the floodplain as an important habitat for the survival of frogs. • Through courses, expose the public to frogs – get them involved in the monitoring. 	<ul style="list-style-type: none"> • Well planned management plan and TPC’s. • Regular meetings and visits to monitor improvement of habitat and numbers of frogs. • Understanding by role players will be measure of success. 	<ul style="list-style-type: none"> • Compare new information to historic data. • Regular meetings and measure achievements against goals/objectives for programme. • Measure changes in attitudes. 	<ul style="list-style-type: none"> • TPC’s will guide measuring of successes. • Habitat improvement will be critical to get numbers up. • Regular feedback – meetings, papers, pamphlets.

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Population Status 	<ul style="list-style-type: none"> • Implement long-term monitoring programmes throughout S.A. 	<ul style="list-style-type: none"> • Monitor adult breeding populations and overall reproductive success). 	<ul style="list-style-type: none"> • Improve current limited base-line information on the status of selected amphibian species. 	<ul style="list-style-type: none"> • More employment opportunities for qualified herpetologists especially in government reserves.
<ul style="list-style-type: none"> • Pesticides and pollutants 	<ul style="list-style-type: none"> • Implement long-term monitoring programmes throughout S.A. 	<ul style="list-style-type: none"> • Monitor adult and tadpoles for abnormalities and deformities and overall reproductive success (offspring survivorship). 	<ul style="list-style-type: none"> • Improve current limited base-line information on the effects of pesticides and pollutants on selected amphibian species. 	<ul style="list-style-type: none"> • A comparative toxicity data-base for amphibians has been suggested as a means of assessing toxicity risks.
<ul style="list-style-type: none"> • Artificial Damming and Changes in flow Regime 	<ul style="list-style-type: none"> • Construction of dams and changes in flow regime due to culverts, weirs, bridges etc. 	<ul style="list-style-type: none"> • Monitor adult and larval reproductive success of selected species. 	<ul style="list-style-type: none"> • Improve current limited base-line information on the effects of habitat modification and flow modification on selected amphibian species. 	<ul style="list-style-type: none"> • Removal of unnecessary barriers such as at Vogelfontein as well as limiting future construction of dams along the system.
<ul style="list-style-type: none"> • Introduction of alien vegetation. 	<ul style="list-style-type: none"> • Reed invasion, invasion of alien invasive vegetation in riparian zones and open water bodies. 	<ul style="list-style-type: none"> • Monitor area of invasion and have eradication targets. 	<ul style="list-style-type: none"> • Constant monitoring of selected sites. Meetings with land owners and affected parties. 	<ul style="list-style-type: none"> • Removal of excessive reed beds or areas invaded by alien vegetation (Work for Wetlands project).
<ul style="list-style-type: none"> • Introduction of exotic animals. 	<ul style="list-style-type: none"> • Introduction of large mouth bass and carp. 	<ul style="list-style-type: none"> • Monitor population sizes of alien species. 	<ul style="list-style-type: none"> • Educational programme as well as long-term monitoring of selected sites. 	<ul style="list-style-type: none"> • Eradication programme including netting or intensive angling competitions on selected species.

	Description	Indicators	Means of verification	Mitigation
<ul style="list-style-type: none"> • Fire. 	<ul style="list-style-type: none"> • Uncontrolled frequent fires have a detrimental effect on herpetofauna. 	<ul style="list-style-type: none"> • Monitor amphibian and reptile numbers after fire destruction. 	<ul style="list-style-type: none"> • Educational awareness programme. 	<ul style="list-style-type: none"> • Implement a natural fire regime with burning of selected blocks.
<p>Habitat</p> <ul style="list-style-type: none"> • Intensive survey • Comprehensive data base • Management strategy for system 	<ul style="list-style-type: none"> • Do a survey of the floodplain and its tributaries to determine areas of modification. • This data must be mapped (GIS) and prioritised in a rehabilitation programme. • Compile a strategy to manage future development – ensure that the habitat will suffer minimal impacts. 	<ul style="list-style-type: none"> • Use historic information to determine changes. • Changes to current status will determine plans to be implemented. • All developments must form part of larger strategy for development in the area – local authorities must “buy-in”. 	<ul style="list-style-type: none"> • Regular updates of data/changes to environment. • Action plans and strategies, with TPC’s to measure failures and success. • TPC’s and goals/objectives of larger strategy for region. 	<ul style="list-style-type: none"> • Regular updates of changes. • Must include whole catchment to be effective. • Regular feedback to role players and monitoring. Plan must be revised on regular basis.

10.4 References:

GREENFIELD R 2004. An assessment protocol for water quality integrity and management of the Nyl River Wetland system. PhD, University of Johannesburg.

