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# A spatial and temporal analysis of the changes in alien macrophyte communities and a baseline assessment of the macroinvertebrates associated with Eurasian water-milfoil, *Myriophyllum spicatum* L. (Haloragaceae) in the Vaal River.

Thesis

Submitted in fulfilment of the requirements for the degree of Master of Science

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# ABSTRACT

The majority of South Africa's fresh water (lotic and lentic), is eutrophic and this has resulted in water hyacinth, *Eichhornia crassipes* (C.Mart.) Solms. (Pontederiaceae) becoming South Africa's most damaging aquatic macrophyte. Recently however, concerns have also been voiced over the presence of highly invasive submerged macrophyte species, such as Eurasian water-milfoil, *Myriophyllum spicatum* L. (Haloragaceae) in the Vaal River. Interaction studies between floating and submerged macrophytes have shown that floating macrophyte dominance restricts light penetration into the water column shading out submerged macrophytes while submerged macrophyte dominance reduces nutrient availability in the water column limiting floating macrophyte growth. This cycle ensures that these species cannot coexist in the same habitat for extended periods of time. The aims of this thesis were to:

1. Investigate changes in the historical and current macrophyte dominance in the Vaal River
2. Determine whether these changes could be attributed to stochastic events, such as floods and herbicide control measures.
3. The physio-chemical conditions of the water column, and whether pressure from herbivory by macroinvertebrates had possibly influenced Eurasian water-milfoil's ability to dominate.

Spatial and temporal analysis of satellite imagery revealed that water hyacinth and submerged macrophyte species dominated different regions of the study area over different periods of time from 2006 to 2010. This was significantly correlated with nitrate concentrations of the water column. One of the lower Vaal River Water Management Areas (WMA) had changed from a water hyacinth dominated state in 2006 to an alternative submerged macrophyte dominated stable state in 2008. It was concluded that this change could be attributed to: a stochastic flooding event in 2006; perturbation from integrated control measures implemented

against water hyacinth; and low nitrate concentrations of the WMA. The lack of any substantial macroinvertebrate herbivory pressure or control measures implemented against Eurasian water-milfoil, compared to similar surveys conducted in the U.S.A. and its native range in Eurasia was shown to contribute to its dominance. Future successful integrated control programmes, including biological control against Eurasian water-milfoil, could provide the perturbation required to restore the ecosystem. However, without the reduction in nitrate concentration levels, water hyacinth will remain the dominant stable state of the rest of the Vaal River.

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# ABBREVIATIONS AND ACRONYMS

DWA – Department of Water Affairs

GIS – Geographic Information System

GPS - Global Positioning System

IUCN – International Union for Conservation of Nature

MMU – Minimum Mapping Unit

NRCS - Natural Resources Conservation Science

ORP – Oxygen reduction Potential

RQS – Resource Quality Services

SANSA – South African National Space Agency

SPOT - Systeme Pour l'Observation de la Terre

TDS – Total Dissolved Solids

WfW – Working for Water

WMA –Water Management Area

USA – United States of America

USDA – United States Department of Agriculture

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# CHAPTER 1 – General Introduction

## 1.1 *Introduction*

The probability of an alien plant becoming invasive is generally governed by the 'Rule of Tens' in invasion biology. Williamson *et al.* (1986) introduced this concept when they noted that exotic species introduced into England quickly naturalise, but remain restricted to highly disturbed areas. Mathematical modelling conducted by Williamson *et al.* (1986) showed that on average, 10% of all introduced species escape and are able to transform into coloniser species, a further 10% of those become naturalised, and lastly 10% of those naturalised species start to dominate existing ecosystems, becoming pests (Williamson & Fitter 1996). Debate over the clear definitions of invasion biology was settled by Richardson *et al.* (2000), who define an introduced species as a species that has established a population outside its indigenous geographical range. Coloniser species are defined as introduced species that are able to form a self-sustaining population of individuals, and naturalised species are those that have the ability to create new separate coloniser populations and become integrated into the indigenous communities. The International Union for Conservation of Nature (IUCN) defines an alien species as:

*"An alien organism is a species, subspecies, or lower taxon introduced outside its normal past or present distribution; includes any part, gametes, seeds, eggs, or propagules of such species that might survive and subsequently reproduce."*
McNeely (2001)

The IUCN emphasises that one of the most serious threats to ecosystem biodiversity is invasion by alien species (Heywood 1989). Globalisation has resulted in alien species inhabiting every ecosystem in the world, even Antarctica (Frenot *et al.* 2005), and is primarily responsible for creating pathways that alien species use as vectors to enter new habitats

(Jones 1998). McNeely (2001) termed globalisation the supreme form of dispersal for any alien species. Globalisation resulted from the dropping of trade barriers which opened distant markets to exotic goods. The sheer volume of goods has not been effectively controlled via government quarantine procedures, and thus this has become the main port of entry for alien species (McNeely 2001). Pimentel (2001) calculated that approximately 480,000 alien species across taxa have become introduced species globally, while significant ecosystem impacts caused by alien invasive plant species have occurred only within the past 40 years (Richardson 2001). A study by Stohlgren *et al.* (2011) showed that of the 120 most widely distributed alien vascular plant species, the Republic of South Africa ranked 5$^{th}$ (having 22.5 % of the 120 species) amongst the 13 bioregions globally which have significant survey data.

Sax *et al* (2002) and Davis (2003) regard the spread of invasive alien plants as the major cause of biotic homogenisation, which is the process whereby community species composition becomes increasingly similar (McKinney & Lockwood 1999). Globally, the rapid influx of alien plants has resulted in increasing similarity of plant community species compositions (McKinney & Lockwood 1999), with unique rare species undergoing constant extirpation (in most regions), and invasion rates of alien plants exceeding the extirpation rates, effectively increasing species richness via homogenisation (Sax *et al.* 2002, Stohlgren *et al.* 2008). Studies of the consequences of homogenisation however have shown that it has had no impact on the beta diversity of plant communities (Olden & Poff 2003, Cassey *et al.* 2006) but it has generally led to local species community composition becoming less distinct (McKinney & Lockwood 2001, Olden & Poff 2003, Pino *et al.* 2009, Winter *et al.* 2009).

One of the constraints on studies that investigate homogenisation of the biota is the scale at which the studies were conducted. These studies are commonly conducted at either a continental, intercontinental or global scale, thereby lacking definitive detail (numerous examples of which are mentioned in Stohlgren *et al.* 2011) (Olden & Rooney 2006, Pino *et al.* 2009). The other constraint is that homogenization has always been measured using classical similarity indices based largely on species numbers (Olden & Rooney 2006, Pino *et al.* 2009). A more comprehensive and complete analysis of homogenisation can be obtained by using species frequencies and abundances (Stohlgren *et al.* 2011). These measurements provide insight into community species dominance rather than just composition, and therefore provide a better analysis of the impact that invasive alien plants are having on the ecosystem (McKinney & Lockwood 2005, McKinney & La Sorte 2007, Lambdon *et al.* 2008).

Following an alien plant's introduction, its ability to become established or naturalised is directly proportional to the number of propagules initially introduced, as well as the number of subsequent introductions from genetically different sources (Richardson 2001). These actions directly increase the possibility of a genotype best suited for its new biotic and abiotic conditions, being introduced. Only once the population is able to reproduce in its new environment does it become naturalised (Richardson 2001).

The last phase of a successful alien plant infestation is the ability of the plant to spread and disperse. This can be achieved via passive vectors such as wind or water, or alternatively, via biotic vectors, such as animals. Once dispersed, additional vegetative dispersal methods often assist the newly established plants to become naturalised, thereby cementing their presence in the ecosystem (Richardson 1997). Rates of spread vary considerably, based on species and the presence of conditions favourable for establishment and naturalisation.

### 1.1.1 Global aquatic plant invasions

It is estimated that between 25 and 30% of all known aquatic plant species are endemic (Sculthorpe 1967, Cook 1974), as a result of the physical barriers that aquatic macrophytes have to overcome to be introduced into a new region (Ashton & Mitchell 1989). In contrast, several hardier aquatic macrophyte families have managed to reach far off regions and become cosmopolitan (Ashton & Mitchell 1989), while on introduction into new ecosystems, several aquatic macrophyte species have shown tendencies to explode in terms of population growth (Sculthorpe 1967, Mitchell 1974), becoming completely dominant (Arthington & Mitchell 1986, Ashton *et al.* 1986).

The most invasive free-floating aquatic macrophytes have been described as the worst invasive weeds of the world (Pieterse & Murphy 1993). These highly successful aquatic plant invaders have some, if not all, of the following traits: an ability to reproduce rapidly, an ability to reproduce vegetatively (often not even sexually), and the ability to enter new habitats via vegetative propagules transported either via human or faunal activities (Ashton & Mitchell 1989). Besides these traits, floating plants, such as water hyacinth, *Eichhornia crassipes* (C.Mart.) Solms. (Pontederiaceae) and Kariba weed, *Salvinia molesta* D.Mitch. (Salviniaceae) also often gain the competitive edge over indigenous species through their ability to photosynthesise via the whole plant, and by occupying large areas of water, shading out submerged competitors (Khanna *et al.* 2011). Their tolerance of a wide range of abiotic conditions and independence of substrate conditions are also contributing factors for their high success rates as alien invasive weeds (Chambers *et al.* 2008). When space becomes limiting, free-floating aquatic macrophytes, such as water hyacinth, are also able to amass organic matter and create what is known as a secondary swamp, which promotes the growth of emergent vegetation thus floating plants are often referred to as primary colonisers of aquatic ecosystems (Mitchell 1974, Ashton *et al.* 1986).

In comparison to free floating plants, submerged and emergent aquatic plants are more reliant on a stable hydrological regime and also require specific abiotic conditions for survival (Bunn & Arthington 2002). In particular, submerged plants require suitable substrate and are negatively affected by turbid water, algae and phytoplankton blooms (Ashton *et al.* 1986). However, given all of these limitations to establishment, should a submerged plant be introduced into suitable habitat, they are often able to rapidly colonise all of the available habitat (Scheffer *et al.* 2003). The most successful submerged aquatic weeds such as Eurasian water-milfoil, *Myriophyllum spicatum* L. (Haloragaceae), Brazilian waterweed, *Egeria densa* Planch. (Hydrocharitaceae), Canadian waterweed, *Elodea canadensis* Mitch. (Hydrocharitaceae), hydrilla, *Hydrilla verticillata* (L.f.) Royle (Hydrocharitaceae), and curly waterweed *Lagarosiphon major* Ridl. Moss ex Wager (Hydrocharitaceae), remain buoyant by means of their intercellular lacunal systems, lack any form of structural rigidity, are able to enter dormancy to avoid unfavourable conditions (Sastroutomo 1981), regenerate from small fragments and produce large quantities of vegetative propagules (Sculthorpe 1967, Nichols & Shaw 1986).

### 1.1.2 South African aquatic invasive alien plants

South Africa has a history of aquatic alien plant infestations, mainly due to eutrophication of rivers and change in natural flow regime which has resulted from the construction of large impoundments (de Villiers & Thiart 2007). The five dominant exotic aquatic weeds found in South Africa are water hyacinth; *Myriophyllum aquaticum* (Vell.) Verdcourt (Haloragaceae) (parrot's feather); *Salvinia molesta* (Kariba weed); *Azolla filiculoides* Lam. (Azollaceae) (red water fern) and *Pistia stratiotes* L. (Araceae) (water lettuce) (Figure 1-1) (Guillarmod 1979, Richardson & van Wilgen 2004).

Over the past two decades, effective control measures implemented against these five species, in particular biological control, have resulted in increases in other aquatic alien plants (Coetzee *et al.* 2011a). According to these writers, submerged aquatics such as Brazilian waterweed, Eurasian water-milfoil and hydrilla are already present in South African rivers and have recently increased in cover, while cabomba, *Cabomba caroliniana* A.Gray (Cabombaceae) and Canadian waterweed pose a serious threat, due to the nature of South African aquatic systems, even though they are not yet present in South Africa. Other hydrophytes such as watercress, *Nasturtium officinale* R.Br. (Brassicaceae), have established, but are not considered as damaging as the aforementioned alien aquatics (Henderson 2001). Coetzee & Hill (2012) have however stated that in South Africa, the invasive alien aquatic plants respond to ecosystem disturbances, and the success of their invasion is often linked to eutrophication of South African water bodies.

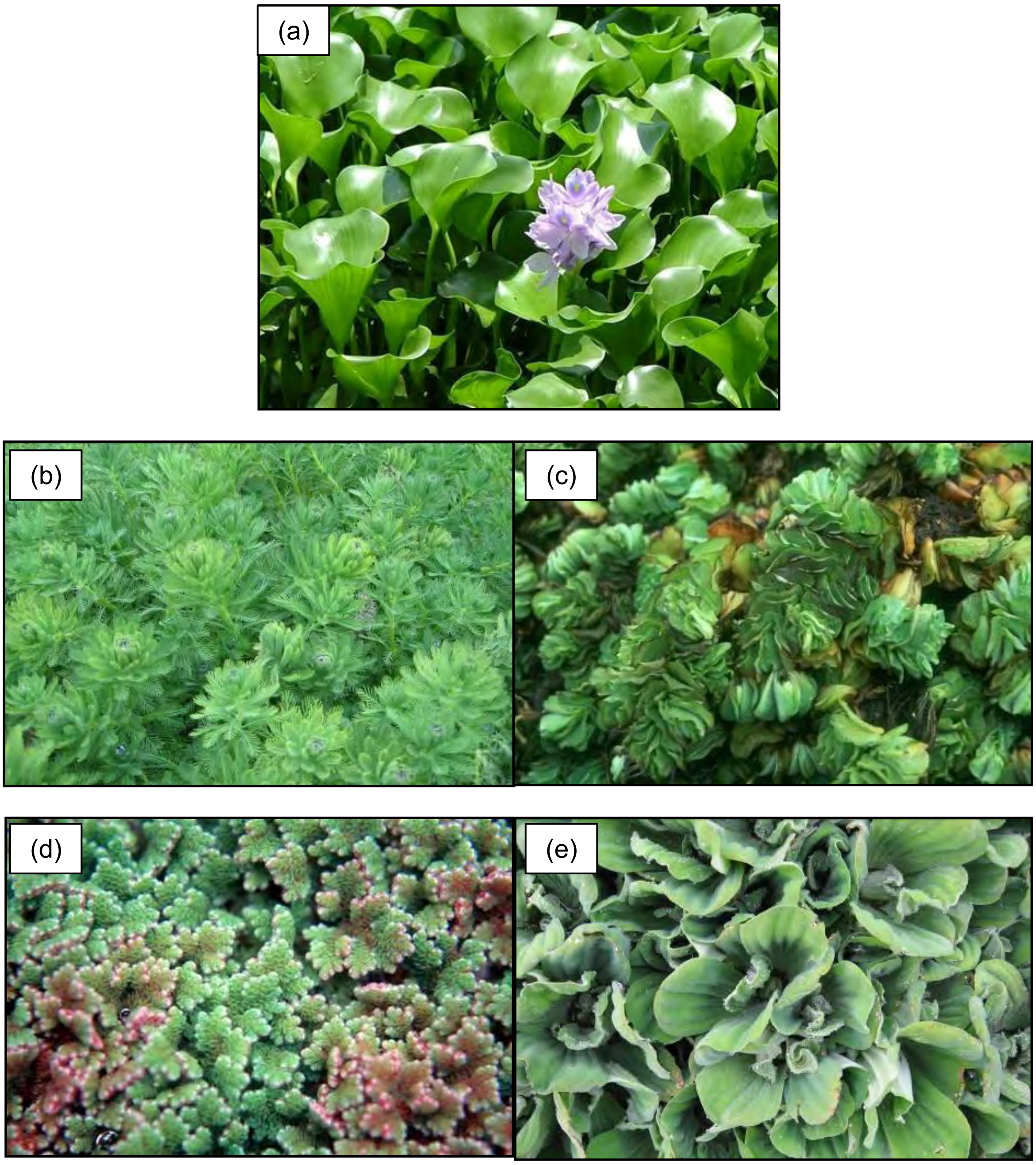


**Figure 1-1:** The five dominant floating invasive aquatic macrophytes in South Africa, (a) *Eichhornia crassipes*, (b) *Myriophyllum aquaticum,* (c) *Salvinia molesta,* (d) *Azolla filiculoides* and (e) *Pistia stratiotes* (Photos courtesy of Dr. Julie Coetzee).

## 1.2 *South African aquatic systems*

Water quality in South Africa is rapidly declining as a result of the increase in pollution due to its rapidly expanding economy (Oberholster & Ashton 2008). Sources of pollution include urbanisation, industry, mining, agriculture, afforestation and power generation (Ashton *et al.* 2008), which all have various detrimental effects on water quality. In addition to the threats posed to water quality, by 2005, almost all of the country's available freshwater resources had been allocated (Oberholster & Ashton 2008) and by 2030 it is expected that South Africa's freshwater resources will be fully allocated and unable to meet the demands of this growing economy (National Committee on climate change 1998). In response to this high demand, South Africa has created 497 large reservoirs (capacity >1 00 000m$^3$) and 150 000 smaller ones, highlighting the importance of South African water resources (Basson *et al.* 1997).

South Africa, a water-scarce country, has also modified the flow regimes of the majority of its rivers, such as those on the Vaal River (Figure 1-2), via the construction of numerous dams, reservoirs and interbasin transfers, since the Molteno Dam built in 1881 (de Villiers & Thiart 2007). This has changed the seasonal patterns of the flow regimes and by reducing water velocity nutrients sink out of the water column (de Villiers & Thiart 2007). Besides additional pollution inputs, water quality in South African river systems is further deteriorating as a result of poor sewage treatment and the overloading of treatment plants (Oberholster & Ashton 2008). The most important macronutrient levels that result in eutrophication are nitrates, nitrites and phosphates (de Villiers & Thiart 2007). This high nutrient input alters the balance of the aquatic ecosystem, particularly the balance between plant species, giving some species a distinct competitive edge (Coetzee *et al.* 2005). A change in floral composition often has a knock-on effect for the rest of the aquatic community as it provides shelter, food and breeding habitats for the faunal communities. Thus, when rivers become eutrophic, the aquatic ecosystem as a whole is changed (de Villiers & Thiart 2007). The climax of eutrophication events in aquatic ecosystems occurs when there is an excessive build-up of organic matter which begins to decompose and deplete oxygen levels, leading to mass fish and invertebrate deaths (de Villiers & Thiart 2007).

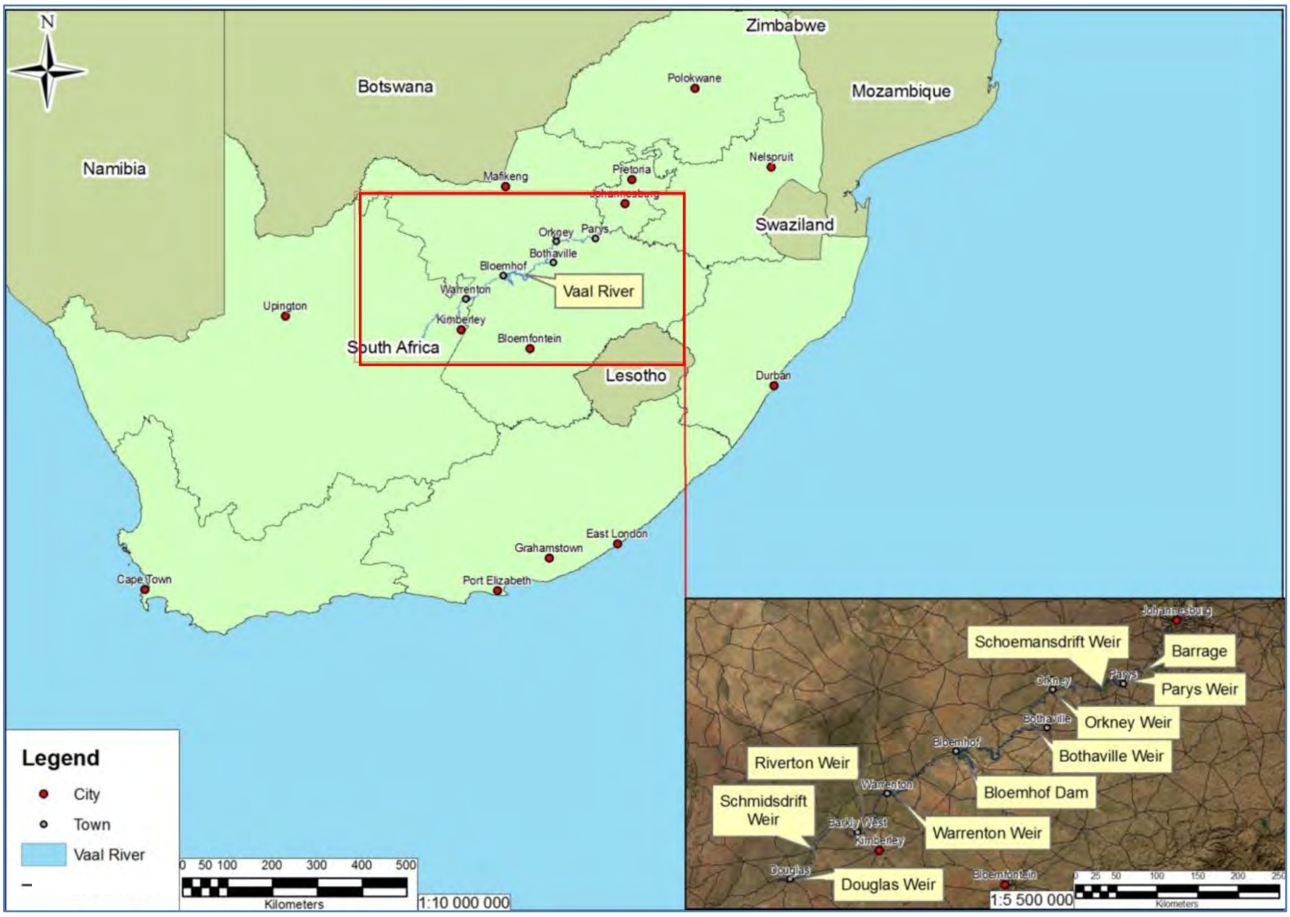


**Figure 1-2:** A map illustrating the location of the Vaal River in South Africa, with an insert depicting significant impoundments along the river.

## 1.3 *Water hyacinth,* Eichhornia crassipes

Eutrophication in South Africa has allowed water hyacinth to dominate systems throughout the country (Coetzee & Hill 2012). Water hyacinth was first discovered in South Africa in 1908 (Stent 1913) and has since spread countrywide (Guillarmod 1979). Its attractive flowers led to its introduction as an ornamental aquatic plant for garden ponds and water features (Ashton *et al.* 1979). Water hyacinth is a free floating water weed which reproduces both by seed and by vegetative budding (Henderson & Cilliers 2002). By means of outcompeting native flora and replicating itself every 11-18 days (Cilliers 1991), water hyacinth remains South Africa's most dominant aquatic weed (Coetzee *et al.* 2011b). It forms dense mats of individuals that completely cover the water surface and prevents sunlight from entering the aquatic community.

### 1.3.1 Taxonomy and origin

The taxonomy of water hyacinth has not undergone any recent changes (Table 1-1).

**Table 1-1:** Taxonomy of water hyacinth (Gopal 1987)

| | | |
|---|---|---|
| **Kingdom** | Plantae | -Plants |
| **Subkingdom** | Tracheobionta | -Vascular plants |
| **Division** | Magnoliophyta | -Angiosperms or flowering plants |
| **Class** | Liliopsida | -Monocots |
| **Subclass** | Commelinidae (Lilidae) | |
| **Order** | Pontederiales (Philydrales) | |
| **Family** | Pontederiaceae | |
| **Genus** | *Eichhornia* | |
| **Species** | *Eichhornia crassipes* | -Water hyacinth |

The genus *Eichhornia* consists of six species and is widely distributed across the continent of South America, with only one species endemic to Northern Africa (Table 1-2) (Gopal 1987). There are no indigenous *Eichhornia* species found in South Africa.

**Table 1-2:** The indigenous geographical locations of the six *Eichhornia* species.

| **Species** | **Geographical location** |
|---|---|
| *Eichhornia azurea* Kunth | (Jamaica and South American) |
| *Eichhornia crassipes* (Mart.) Solms-Laub. | (South America) |
| *Eichhornia diversifolia* (Vahl) Urban | (Brazil, Surinam, Cuba, Haiti, Puerto Rico, Santo Domingo) |
| *Eichhornia paniculata* (Spreng) Solms | (northern Argentina , Brazil) |
| *Eichhornia paradoxa* (Solms) | (Endemic to Brazil and possibly Caracus Venezuela) |
| *Eichhornia natans* (Beauv.) Solms | (Senegal, Sudan, Nigeria and Mali) |

### 1.3.2 Morphology and reproduction

Water hyacinth is a perennial aquatic free floating or anchored plant that changes in response to its abiotic environment (Henderson & Cilliers 2002). Depending on the abiotic conditions it is exposed to, its roots can constitute as much as 50% of a single plant's biomass (Gopal 1987). To increase root surface area, the root structure is made up of fine feathery adventitious rhizomes. Petioles of water hyacinth plants vary in size from an average of 100-200mm to 1m in height when growing under favourable conditions in dense mats (Penfound & Earle 1948). The leaves are shiny dark green in colour, arranged in rosettes with a distinct erect swollen bladder-like petiole (Penfound & Earle 1948). Its attractive pale violet or blue flowers measure up to 50mm in diameter, with the upper petal having a yellow central patch (Penfound & Earle 1948). The resultant capsules can contain from 40 – 300 fine seeds (Cronk & Fenessy 2001) that can remain dormant in the sediment (Obeid & Tag El Seed 1976) for a period of greater than 5 years (Edwards & Musil 1975, Thompson *et al.* 1998, Gunnarsson & Petersen 2007) until dormancy is broken via a cycle of wetting, drying and re-wetting (Baskin *et al.* 2003). Despite its effective seed production, water hyacinth primarily spreads via the production of daughter plants through the formation of stolons (Penfound & Earle 1948). The reinfestation of South African water-bodies, following effective control measures, is highly likely due to the large seed banks present in South African water-bodies (Albano Pérez *et al.* 2011).

### 1.3.3 Abiotic conditions

One of the primary reasons that water hyacinth is such a successful invasive weed is its ability to grow under a variety of abiotic conditions. It can grow in both highly acidic and alkaline waters, which can vary from clean to highly polluted, shallow ponds to deep waters and can survive in fairly fast flowing waters (Gopal 1987). It does not however thrive in rough waters or aquatic systems which have salinity levels exceeding 15% of that of sea water (Penfound & Earle 1948).

The plant's ability to modify its morphology is key to this success. When exposed to clean nutrient-depleted waters, the plant responds by increasing its root biomass to absorb the optimum amount of nutrients and, decreases its above-water biomass (Pieterse 1978, Xie 2004). The converse is also true, when water hyacinth is exposed to highly eutrophic waters, with high concentrations of nutrients and organic matter, the plant has a low root biomass and rather invests heavily in above-water biomass (Pieterse 1978, Xie 2004).

The ability of water hyacinth to remediate polluted water has been extensively studied in infested river and marshland systems (Gossett & Norris 1971, Mitsch 1977). The plant has the ability to capture and store N and P in excess of what it requires for growth (Reddy & Tucker 1983, Reddy & Reddy 1987, Alves *et al.* 2003, Fox 2009). Even though this is useful for N and P removal from systems, it varies in effectiveness between scenarios and water hyacinth alone cannot remediate 100% of the N or P pollution from solution (Fox 2009).

### 1.3.4 Impact

The highly invasive nature of water hyacinth and great rate of reproduction (Cilliers 1991) results in it disrupting any activity associated with water body utilisation (Coetzee & Hill 2008). Modifications to natural South African aquatic systems, coupled with water nutrient enrichment, have resulted in many of these systems becoming highly susceptible to water hyacinth infestation (MacDougall & Turkington 2005). As water is a valuable resource in South Africa the economic and ecological impacts associated with infestations of water hyacinth can be very significant. A number of additional negative impacts have been associated with water hyacinth infestations, summarised in Table 1-3.

**Table 1-3:** A modified summary of the impacts of dense water hyacinth on its environment (Jones 2009).

| **Water hyacinth infestation impacts** | **Impact** | **Reference** |
|---|---|---|
| Increased water utilisation for plant physiological processes. | Increased water loss through evapotransporation compared to open surface water loss rates. | Cilliers 1991 |
| Decreased light penetration into the water column. | Oxygen levels decrease and carbon dioxide levels increase, having negative effects on aquatic fauna and flora. | Timmer & Weldon 1966, Howard & Harley 1998, Gratwicke & Marshall 2001 |
| Water flow rates can be reduced by 40 to 95%. | Increases severity of flooding events and negative effects on the general ecology of the system. | Gopal 1987 |
| Changes water quality. | Lowers water temperature, pH, bicarbonate, alkalinity and dissolved oxygen content, while concurrently increasing free carbon dioxide content and nutrient levels. | Ultsch 1973, McVea & Boyd 1975 |
| Physical damage to ecosystem. | Dense mats can damage riparian vegetation in strong winds. | Gowanloch 1944 |
| Knock on effect to surrounding ecosystems. | All these above mentioned impacts directly modify the ecosystem, which indirectly has a negative effect on the surrounding resident biota. | Crooks 2002 |

Water hyacinth in South Africa is most damaging at high altitudes (above 1500m), where it thrives in nutrient enriched waters (Hill & Olckers 2001).The cold winters of these altitudes, in South Africa, do have a negative effect on the plant (Byrne *et al.* 2010), however despite the climate, the aforementioned conditions have attributed to its dominance of the Vaal River. The weed was first observed as a problem plant in the Vaal River in 1972 and since then it has spread a distance of 300km (Henderson & Cilliers 2002). During a survey conducted in April 1983 and March 1984 (Bruwer *et al.* 1985), three macrophytes species occurred along the Vaal River, included which only two aquatic macrophytes were noted to be alien invasives namely: water hyacinth and the submerged macrophyte, Eurasian water-milfoil.

### 1.3.5 Control Methods

In South Africa, the Department of Water Affairs (DWA) has a mandate to protect and secure the Republic's water quality and quantity. To assist in fulfilling their mandate, the DWA formed the Working for Water (WfW) programme in 1995 with the vision of improving water quality and quantity by controlling alien plant infestations, such as water hyacinth (Turpie *et al.* 2008). In the recent past, various control measures have been implemented against water hyacinth infestations in South Africa. There are four broad categories of control: mechanical, herbicidal, biological and integrated (Cilliers *et al.* 1996).

#### 1.3.5.1 Mechanical control

Traditionally, mechanical control has always been the first option of water hyacinth control, however this method is ineffective against infestations larger than a hectare due to the plant's rapid growth rate and the fact that it constitutes 90% water (Penfound & Earle 1948). Usually mechanical removal is labour intensive, where workers use rakes and pitchforks to remove the water hyacinth from the water (Hill & Coetzee 2008). Mechanical harvesters are also used for larger infestations and have had some success in the past in Port Bell and the Owen Falls Dam on the Ugandan side of Lake Victoria, where the port, hydroelectric power generation and water intake pipes have been kept free of water hyacinth (Mailu 2001).

In South Africa the first machine bought for water hyacinth removal was the Watermaster Classic III, bought in February 2010, by Ekurhuleni Metropolitan Municipality in Gauteng Province, to assist in the removal of water hyacinth from Benoni Lakes (Figure 1-3). This machine cost R7 Million and although it has been effective in clearing infestations, the long-term running costs are yet unknown (http://www.looklocal.co.za/looklocal/content/en/benoni-news). Hill (2003) suggests that although mechanical control provides immediate relief from the infestation, the cost benefit analysis of mechanical control are outcompeted by integrated control measures.

**Figure 1-3**: Watermaster Classic III machine bought by Ekurhuleni Metropolitan Municipality (http://www.looklocal.co.za/looklocal/content/en/benoni/)

Apart from manual removal using pitchforks and rakes, another form of mechanical control used in South Africa is the spanning of cables across rivers (Cilliers *et al.* 1996). This prevents small mats of the plants from floating downstream and consolidates the smaller mats together into a singular large mat. The cables can be raised or lowered, thereby allowing for effective aerial spraying, or they can be utilised to split large infestations into smaller clumps to make spraying the weed more manageable (Cilliers *et al.* 1996). This method of mechanical control has been successfully implemented across the Vaal River (as part of an integrated control programme), where the water hyacinth closest to the bridge has undergone herbicide treatment from the bridge with a high pressure sprayer (Figure 1-4).

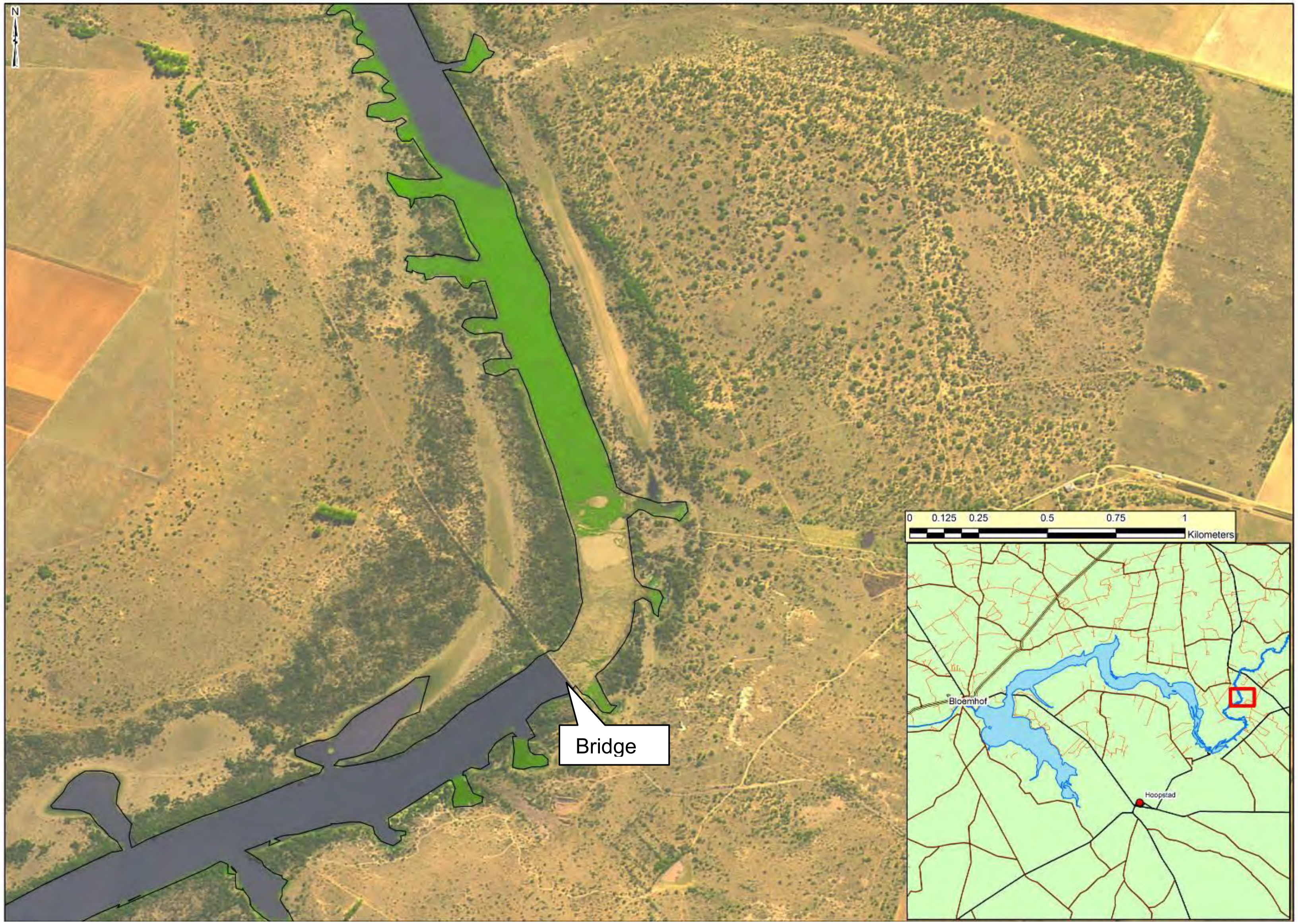


**Figure 1-4**: A 2009 SPOT V satellite image showing a 2km long infestation of water hyacinth that has resulted from backing up against a cable spanned across a bridge located at the top of Bloemhof Dam (insert depicting exact location of the bridge). The brown regions of the infestation, next to the bridge, have undergone herbicide treatment.

#### 1.3.5.2 Herbicide control

The first record of water hyacinth control in South Africa dates from the early 1970s, by the Department of Water Affairs when it was controlled on Hartebeespoort Dam via aerial application of the herbicide Clarosan 500FW (Ashton *et al.* 1979). Several studies summarised in Ueckermann and Hill (2001) showed that the best herbicide for treatment of water hyacinth is glyphosate-based, despite the detrimental effects of glyphosate on aquatic fauna and riverine flora (Relyea 2005a, 2005b, 2005c, Relyea *et al.* 2005).

Ueckermann & Hill (2001) however showed that should glyphosate be used responsibly at the correct concentrations, it is non-toxic to both biocontrol agents and indigenous invertebrates. Currently WfW uses a 2–4% lethal dose solution of glyphosate, recommended by Ueckermann and Hill (2001), in an effort to maintain populations of biological control agents. Only a percentage of the water hyacinth population is treated on the infestations of the Vaal River (Byrne *et al.* 2010), which costs R1194/ha on average, including follow-up spraying (D. Sharp, WfW, Pers. comm. 2011).

Despite its safe and responsible use, the extent of nontarget effects of glyphosate on neighbouring vegetation is directly linked to the vegetation species composition and population maturity. Seedlings have been shown to be the most at risk regarding herbicidal drift at concentrations lethal to water hyacinth, while mature plants are much better equipped to withstand infrequent applications of herbicidal drift (Mars *et al.* 1993). Mars *et al.* (1993) suggest that in the light of this evidence, a minimum of 6m no-spray buffer zones be implemented alongside mature non-sensitive vegetation stands, and 20m no-spray buffer zone be used to minimise impacts on sensitive vegetation which comprises a high seedling count. On the Vaal River, 50 – 60% of the central portions of all plugs are sprayed, minimising herbicide spray drift (D. Sharp, WfW, Pers. comm. 2011).

The primary disadvantage of using herbicidal control is that following a costly herbicidal treatment, water hyacinth rapidly regenerates, either via daughter plant production of surviving individuals or via seed germination from increased light entering the aquatic environment (Charudattan *et al.* 1996), and additional treatments are therefore required. Additional disadvantages are as a result of the efficiency of the chemicals. Herbicidal half-life has had to be reduced following concerns that in rural areas, communities often use untreated water for domestic consumption, while the massive die-off and decomposition of water hyacinth mats following herbicidal treatment can affect the aquatic environment and result in an increase of anoxic conditions (Ueckermann & Hill 2001). In comparison to the negative effects that a water hyacinth mat poses, these effects are negligible (Ueckermann & Hill 2001).

#### 1.3.5.3 Biological control

Biological control is defined by Eilenberg *et al.* (2001) as a method of control whereby living organisms are used to reduce the population of a specific pest organism, either via directly decreasing its density or impairing the pest organism's ability to cause damage to its surrounding environment. Biological control strategies include classical biological control, inoculation biological control, inundation biological control and conservation biological control (Eilenberg *et al.* 2001), of which classical biological control is the most commonly used for landscape level effectiveness (Van Driesche *et al.* 2010). If the classical biological control option is effective, it will result in the desired ecological modification over extensive areas, for a fraction of the cost and effort required by any other control methods implemented across the same extent (Van Driesche *et al.* 2010).

Biological control is considered the most effective form of water hyacinth control and has a high benefit to cost ratio (Harley 1990). The first biological control agent introduced into South Africa against water hyacinth was *Neochetina eichhorniae* Warner (Coleoptera:

Curculionidae), initially onto the Vaal River in 1977. This failed to establish until a second introduction was made in 1986 (Cilliers 1991, Julien & Griffiths 1998). Additional agents released onto the Vaal River in 1990 include another weevil, *Neochetina bruchi* Hustache (Coleoptera: Curculionidae) and the moth *Niphograpta albiguttalis* (Warren) (=*Sameodes albiguttalis* (Warren)) (Lepidoptera: Pyralidae) (Cilliers 1991). Initially, the effectiveness of the agents was limited by herbicidal treatment, which when used at concentrations above 4% killed the agents, either by direct toxicity, or by the destruction of their food source, and flooding damaging or displacing the plants (Center 1994). As a result, Cilliers (1991) proposed that "biological control reserves" be established, where no herbicidal treatment of the weed would take place, which would have a beneficial effect on agent populations.

Currently four of the six arthropod biological control agents released for the control of water hyacinth in South Africa have established on the Vaal River. The leaf feeding mite *Orthogalumna terebrantis* Wallwork (Oribatida: Galumnidae), has not established (M. Hill, Rhodes University, Pers. comm. 2011) and the grasshopper *Cornops aquaticum* (Brüner) (Orthoptera: Acrididae) has only been recently released in two locations, Misty Hills, Muldersdrift (Gauteng) and Dudley Pringle Dam at Tongaat Sugar Estate (KwaZulu-Natal) not in the Vaal River (Coetzee *et al.*, 2011b). Three of these agents, the weevils *N. bruchi*, *N. eichhorniae,* and the moth *N. albiguttalis,* have been described by Julien & Griffiths (1998) as the most effective biocontrol agents against water hyacinth in the world, with *C. aquaticum* being classed by Coetzee *et al.* (2009a) as South Africa's most promising agent.

#### 1.3.5.4 Integrated control

Integrated control should be the preferred option for water hyacinth control (Law 2007). An integrated approach has resulted in a decrease of water hyacinth cover on the Vaal River. The strategy of integrated control on the Vaal River involves using cables to manipulate the infestation sizes, with the central 60% of the clumps undergoing spraying with a lethal glyphosate solution (2-4%), while the remaining portion of the plants is left as a nursery for biological control agents (Byrne *et al.* 2010). Any smaller infestations undergo spraying from boats and knapsacks with lethal doses of herbicide (Cilliers *et al.* 1996). The integrated approach has resulted in a reduction in water hyacinth populations over the past five years. The removal of water hyacinth as the dominant aquatic weed has opened a niche in the ecosystem. Unfortunately there has been a concurrent observed increase in the density and percentage surface area covered by an alien submerged aquatic weed, Eurasian water-milfoil, from when it was first observed in 1885 near Barkly West on the Vaal River (28°32’45”S 24°30’50”E) (SAPIA Database, ARC – PPRI).

## 1.4 *Eurasian water-milfoil,* Myriophyllum spicatum

*Myriophyllum spicatum* L. (Haloragaceae), Eurasian water-milfoil was first discovered by Linnaeus in 1753 and is indigenous native to Eurasia and northern Africa (Couch & Nelson 1985). Following which it was recorded to have invaded North America in 1814 (Bergquist & Bergquist 1970). It has also been introduced into Australia, India and South Africa (Holm *et al.* 1979, Couch & Nelson 1985). Eurasian water-milfoil was first recorded in South Africa in 1885, near Barkly West on the Vaal River (28°32’45”S 24°30’50”E) ((SAPIA Database, ARC – PPRI). While it is classed as a Category 1 weed by the Conservation of Agricultural Resources Act (CARA) of 1983*, it was not regarded as a problem weed until 2005 (Coetzee *et al.* 2011a). It was recorded to have established at a few other locations in South Africa but the only other confirmed record is in Lake Sibaya in northern KwaZulu Natal (27°24’37”S; 32°42’20”E) (Coetzee *et al.* 2011a).

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### 1.4.1 Taxonomy and origin

Since Linnaeus’s description, the taxonomy of the genus has undergone several revisions. The latest taxonomy of Eurasian water-milfoil is shown in Table 1-4. There are 16

*CARA is the legislation used in South Africa to govern how serious an invader, particular plant species, can be classed, and thus be used to limit trade and govern how species are treated.

*Myriophyllum* species worldwide but no indigenous species of *Myriophyllum* occur in South Africa; only the invasive Parrots feather (*M. aquaticum*) and Eurasian water-milfoil are present (Coetzee *et al.* 2011a). Parrots feather is particular abundant in the Eastern and Western Cape, North-West and Mpumalanga provinces

**Table 1-4**: Taxonomy of *Myriophyllum spicatum* (Cock *et al.* 2008a).

| | | |
|---|---|---|
| **Kingdom** | Plantae | -Plants |
| **Subkingdom** | Tracheobionta | -Vascular plants |
| **Division** | Magnoliophyta | -Angiosperms or flowering plants |
| **Class** | Magnoliopsida | -Dicots |
| **Subclass** | Rosidae | |
| **Order** | Haloragales | |
| **Family** | Haloragaceae | |
| **Genus** | *Myriophyllum* L. | -Water milfoil |
| **Species** | *Myriophyllum spicatum* L. | -Eurasian water-milfoil |

Eurasian water-milfoil is a submerged macrophyte native to Eurasia and North Africa (Meusel & Jager 1978). It has however been introduced and found to be invasive in a number of countries including 43 states in the U.S.A., Australia and South Africa. Its invasiveness can be attributed to its effective modes of reproduction and morphology.

### 1.4.2 Morphology and reproduction

Eurasian water-milfoil plants root in the sediment and grow rapidly upwards to form a dense canopy at the surface (Figure 1-5). They typically grow in water 1- 4m in depth, however in ideal conditions, growth in depths of up to 10m has been recorded, with densities exceeding 300 plants/$m^2$ (Aiken *et al.* 1979). The stems vary considerably in appearance, from pink to yellow and usually accommodate 12 – 21 leaflet pairs, which lose structural integrity and go limp when removed from water (Johnson & Blossey 1997). The plant's roots are adventitious persisting along the stem in the lower buried portions and becoming visible prior to auto-fragmentation along the upper parts of the stem (Smith & Barko 1990). Unlike other submerged aquatics, Eurasian water-milfoil is essentially an “evergreen plant” and does not form turions or any other form of specialised overwintering structures to help it survive the extreme cold conditions in its native range (Smith & Barko 1990). Instead the plant stores carbohydrates throughout its roots and shoots. In spring, the plants only start to grow when water temperatures reach 15$^{o}$C (Titus & Adams 1979, Perkins & Systsma 1987).

The plant typically flowers in early summer in its native range. Only those that have reached the surface flower (Figure 1-5) (Johnson & Blossey 1997). The resulting inflorescence has separate male (located on the upper half of the spike) and female flowers (located on the lower portion of the spike) (Squire & Hawes 2004). Flowers are primarily pollinated anemophily and the resulting seeds are dispersed by waterfowl or water currents (Grace & Wetzel 1978). Each fruit separates into four separate nutlets which are in total 2-3mm long and subglobose in shape (Squire & Hawes 2004). Seeds require scarification to germinate, often resulting in germination in their second spring after seeding with an 85% success rate (Pattern 1955, Guppy 1897). The resultant seedling is very delicate and can be easily damaged or destroyed (Pattern 1955), resulting in vegetative reproduction being the primary form of population expansion (Grace & Wetzel 1978). Eurasian water-milfoil has also been shown to hybridize with *Myriophyllum sibiricum* Kom. (Haloragaceae) (a native North American species) (Moody & Les 2002).

### 1.4.3 Abiotic conditions

The high rates of reproduction and morphological adaptations of Eurasian water-milfoil make it a highly adaptable plant which can grow under a wide variety of conditions. Once established however, many of the conditions listed in Table 1-5 are on feed-back loop systems which can be influenced by varying degrees depending on the size of Eurasian water-milfoil population. For example, when dense stands of submerged vegetation become established, they can reduce current velocity and allow heavier particles to settle out of the water column, thereby increasing water clarity, which has a positive effect on the rate at which the plant grows and allows the plant to grow in deeper waters (Scheffer *et al.* 1994). Besides carbon dioxide, Eurasian water-milfoil has the ability to absorb carbon in the form of bicarbonate from the water column (Grace & Wetzel 1978). The removal of carbon in the form of carbon dioxide increases the pH of the water column creating available bicarbonate and alkaline waters in turn assist the plant to remove additional bicarbonate from the water column (Grace & Wetzel 1978). In colder climates, the winter die-back of the plant also directly increases the N and P levels in the water column (Nichols 1991). These feedback loops, where the population is able to manipulate environmental variables to enhance its growth rate, are one of the primary reasons why Eurasian water-milfoil is such a successful alien invasive weed (Grace & Wetzel 1978, Smith & Barko 1990).

**Figure 1-5:** A dense stand of flowering Eurasian water-milfoil bed in the Vaal River taken in 2010, Rooipoort Nature Reserve (24°10'23.986" E; 28°32'59.651" S).

### 1.4.4 Impact

As a result of its dense canopy-forming properties, Eurasian water-milfoil can have direct financial costs, including damage to irrigation and pump equipment, and reducing recreational access to the water body (Johnson & Blossey 1997). Johnson and Blossey

(1997) also showed that Eurasian water-milfoil's dominance of ecosystems is assisted by the plant's superior competitive ability and tolerance of a wide variety of growing conditions (Table 1-5). Eurasian water-milfoil's growth rate is often also faster than native macrophyte species, resulting in dense canopies forming earlier in spring, before native species have reached their maximum growth rate. These dense canopies shade out competing species, altering community species composition and thus having severe negative effects on biodiversity. According to Keast (1984), a Eurasian water-milfoil dominated habitat contains significantly fewer macroinvertebrates (including benthic invertebrates), than native macrophyte habitat and this has an indirect impact on the abundance and spawning of native fish species (Aiken *et al.* 1979, Johnson & Blossey 1997).

**Table 1-5**: Environmental variables affecting Eurasian water-milfoil growth rates (Smith & Barko 1990)

| **Environmental Variable** | ***Myriophyllum spicatum* response** |
|---|---|
| **Light conditions** | Turbid waters restricts the plant to shallow depths, clearer waters allows for growth at deeper depths. |
| **Temperature** | Plant growth is inhibited at higher water temperatures (>30$^o$C) and optimum growth rates are achieved between 15-25$^o$C, growth is severely hampered below 15$^o$C. |
| **Carbon** | Plants prefer to take up inorganic carbon in the form of $H_2CO_3$, for this to occur, alkaline pH levels are needed, the more acidic the conditions, the less rigorous the growth rate. |
| **Nutrient levels** | Roots are responsible for major nutrient uptake (N and P), while cations and bicarbonate are absorbed directly from the water column. Plant populations perform best in eutrophic systems. |
| **Sediment** | Growth rates are highest while growing as intermediate sized populations in fine textured sediments. |
| **Flow rates** | Water currents assist in the spread of the plant, however it does not fare well in high energy locations |
| **Ice scour** | This limits plant growth in shallow areas. |
| **Desiccation** | Water level fluctuation is an effective control measure to limit shallow water growth as the plants are highly affected by desiccation and freezing during winter. |

### 1.4.5 Control Methods

The high costs of a Eurasian water-milfoil infestation, both financially and ecologically, have made it a target for a number of control measures. According to the United States Department of Agriculture (USDA) and Natural Resources Conservation Science (NRCS) (2010), the aggressive manner by which Eurasian water-milfoil invades habitats, has led to the implementation of a number of different management options, all of which are currently used throughout various states of the U.S.A., currently there are no control measures being implemented against South African Eurasian water-milfoil populations.

#### 1.4.5.1 Prevention or eradication

The primary control option in the U.S.A. is prevention (Parkinson *et al.* 2010). The vegetative manner in which Eurasian water-milfoil spreads means that a single small strand of two nodes (approximately 2.5cm long) has the ability to grow and establish a new population (Riis *et al.* 2009). Regular, meticulous inspections of recreational equipment (Figure 1-6) are conducted at recreational sites before and after the equipment enters or leaves the water. Removal of any strands is then done on site, mostly via a wash bay (Prather *et al.* 2007).

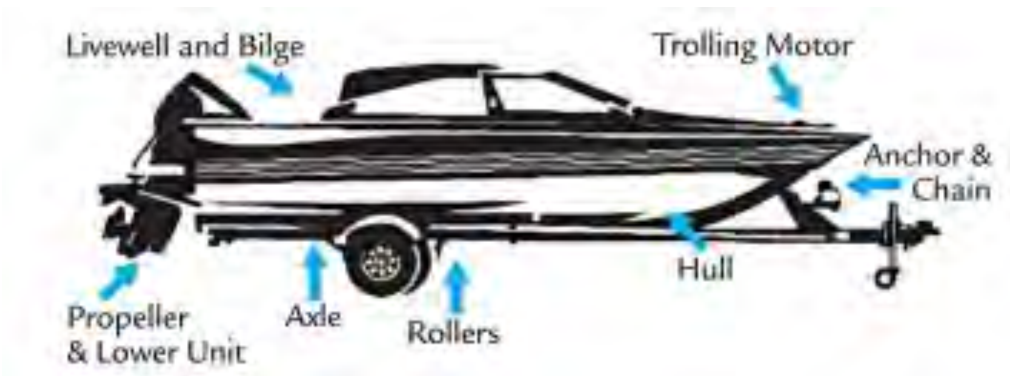



**Figure 1-6:** Arrows indicate the locations of inspection points on recreational boats, where Eurasian water-milfoil strands are often found (Parkinson *et al.* 2010)

The high reproduction potential of Eurasian water-milfoil, however, means that new infestations will inevitably become established and the use of early detection of new infestations plays an essential role in limiting the spread of this macrophyte (Parkinson *et al.* 2010). The identification of water-bodies at risk and regular surveying of these waters for new infestations allows for the early detection of future infestations, and can result in the eradication of the macrophyte (if the population is small and isolated enough), via the use of herbicides or mechanical control options (Parkinson *et al.* 2010).

#### 1.4.5.2 Mechanical control

Eurasian water-milfoil undergoes routine mechanical control in many states of the U.S.A. For small populations, the most commonly method is manual removal via rake drags, often in conjunction with fragment barriers (Gettys *et al.* 2009). This is best implemented just prior to attainment of peak seasonal biomass (Madsen 2005) and although labour intensive and limited to shallow waters it is highly effective against small isolated individuals and plants (Boylen *et al.* 1996). In some states, deeper small populations are removed by divers equipped with suction pipes to extract the entire plant (Boylen *et al.* 1996).Divers are also employed to fasten benthic barriers to lake bottoms. These barriers limit Eurasian water-milfoil growth in deeper waters and have been particularly useful near boat ramps or areas that are frequently disturbed (Laitala 2007). Constant maintenance of these structures is however required, as sediment accumulation just 4cm deep allows for plant establishment (Laitala 2007).

For larger infestations, the development of mechanical floating harvesters was first initiated in the 1950s by a Wisconsin company, which serviced hundred of lakes in the Upper Midwest, specifically aimed at harvesting Eurasian water-milfoil and curly leaf pondweed (Gettys *et al.* 2009). Since the 1950s, mechanical harvesters have become highly sophisticated, manoeuvrable machines, able to operate in water depths of 0.38 - 0.46m

(Gettys *et al.* 2009). Currently mechanical harvesters are used where extensive infestations are located, especially in the Northeast and Midwest of the U.S.A. (Madsen 2005). These machines cut off the growing upper portions of the plant (1.5m deep and 2.4m wide), and this in turn severely hampers its growth rate (Sheldon & O'Bryan 1996). Not all harvesters can collect the abscised portion of the plant and often this process assists the spread of Eurasian water-milfoil (Madsen 2000).

In addition to mechanical removal of the upper portions of the plants, rotavating operations damage and destroy the root crowns, but the floating plant debris also assists in the spread of Eurasian water-milfoil. Dredging of the sediment deepens the water column and is effective against small beds of Eurasian water-milfoil, but removes all available habitat for other macrophyte species and is more costly than any other form of mechanical control (Gettys *et al.* 2009).

Overall, according to the AERF (2009), advantages of mechanical control revolve around the fact that it is site specific, it results in little removal of nutrients from the system (1-3% which is especially important in oligotrophic systems), and that the water body can immediately be utilised following implementation. The disadvantages are that it is relatively costly, that it does not differentiate between plant species (except for diver or manual removal methods) and certain methods physically damage other species such as fish and crayfish (AERF 2009). Mechanical operations also primarily remove the upper portions of the Eurasian water-milfoil plant where the majority of phytophagous insects reside and harvesting decimates the population levels of these insects resulting in a faster re-growth of the Eurasian water-milfoil populations (Newman & Inglis 2009).

### 1.4.5.3 Herbicide control

Eurasian water-milfoil control in the U.S.A. also involves herbicide treatment, although the legislation governing use of herbicides varies on a state basis (Newman, University of Minnesota, Pers. comm. 2011). The fastest acting of the herbicides used are contact heterocyclic cationic herbicide Diquat and the dicarboxylic acid herbicide, Endothall. According to Parkinson *et al.* (2010), these herbicides are used in fast flowing systems and have the ecological advantages of been fast acting, with only a short contact period, required and they have a short half-life. Unfortunately these herbicides do not translocate into the roots of Eurasian water-milfoil and thus re-growth is likely (Vassios 2010). Following application of these herbicides, it is not safe to use the water for recreational activity for one day. Human, livestock consumption and irrigation activities of water treated with Endothall can only resume after 7-25 days, or when the concentration levels drop below 0.5ppb (Petty 2005). When Diquat is applied to the water column, human consumption can resume after 3 days or when the concentration levels drop below 0.05ppb, while livestock consumption can resume a day after treatment and irrigation activities can resume after 3 days (Petty 2005).

Triclopyr and 2,4-D are better suited for use against Eurasian water-milfoil beds in slower-flowing systems (Parkinson *et al.* 2010). They require an intermediate contact time with the macrophyte, but they have no effect on the majority of the native U.S.A. aquatic macrophyte species, excluding other native U.S.A. *Myriophyllum* species (Vassios 2010). Triclopyr does not restrict recreational use of the water body following application, there is no restriction on livestock consumption and human consumption can resume after concentration levels have dropped to 0.4ppb (this varies in time). Irrigation activities, however, are severely restricted and cannot resume until the concentration levels drop below 1.0ppb or alternatively after 120 days (Petty 2005). The restrictions following 2,4-D application are much less severe, recreational activities can resume a day after treatment, while livestock and human consumption as well as irrigational activities can resume 21 days after treatment, or when concentration levels drop below 0.03ppb (Petty 2005). Despite these disadvantages, both Triclopyr and 2,4-D are the most commonly used herbicides against Eurasian water-milfoil, both for their selectivity and cost effectiveness (Parkinson *et al.* 2010).

In Eurasian water-milfoil-infested lake systems of the U.S.A., Fluridone is the herbicide of choice (Parkinson *et al.* 2010). It requires a very long contact period (60 days) and is broad spectrum (Vassios 2010). The advantages of using Fluridone are linked to the water use. It has no restrictions regarding recreational activities, and human water consumption can resume when levels drop below 150ppb, while use for livestock consumption is not restricted and irrigation is even possible with limited restrictions (based on the water body and crop type) (Petty 2005). Although Fluridone is a broad spectrum herbicide, it does not affect all plants to the same degree, but is highly effective against Eurasian water-milfoil (Gettys *et al.* 2009). Fluridone treatment is recommended to be applied only to lake systems with a diverse submerged macrophyte species composition (R. Newman, University of Minnesota, Pers. comm. 2011). This is done to eliminate the possibility of the simultaneous complete destruction of the entire submerged macrophyte flora. The removal of all submerged macrophytes from a lake system changes the abiotic environment, often destabilising the system and creating unfavourable conditions for subsequent submerged aquatic plant growth (R. Newman, University of Minnesota, Pers. comm. 2011).

#### 1.4.5.4 Biological control

The severity of the Eurasian water-milfoil invasion in the U.S.A. for the past 30 years has necessitated the development of an effective biological control programme (Johnson & Blossey 1997). Despite all the historical efforts to combat the invasion using other methods of control, only recently have declines in its densities been observed. This is mainly attributed to feeding damage caused by an introduced midge, *Cricotopus myriophylli* Oliver (Diptera: Chironomidae) (MacRae *et al.* 1990), the introduced pyralid moth, *Acentria* *ephemerella* Denis and Schiffermüller (syn.: *Acentria nivea* Olivier) (Lepidoptera: Crambidae) (both feed on the apical tip meristematic tissue (MacRae *et al.* 1990, Johnson & Blossey 1997), and the native weevil, *Euhrychiopsis lecontei* Dietz (Coleoptera: Curculionidae) (adults feed on the leaves, stem and meristem and larvae feed on meristem and vascular tissue) (Cock *et al.* 2008a). The most damaging and effective agent used against Eurasian water-milfoil is the milfoil weevil (Cock *et al.* 2008a), currently undergoing host specificity testing in quarantine at Rhodes University, Grahamstown, South Africa.

#### 1.4.5.5 Integrated control

Integrated control measures are not fully recognised and used throughout the U.S.A. against Eurasian water-milfoil. Control measures differ between states and coordination between biological, mechanical and herbicidal treatments is not implemented on a national scale (Newman, University of Minnesota, Pers. comm. 2011). The majority of integrated control measures happen only via coincidence, for example particular lakes have restrictions placed on the percentage of a lake that can be mechanically harvested. This is done to maintain a portion of the habitat for the fish population, but as a consequence of which, milfoil weevil populations are also protected. The protection of the milfoil weevil populations was not considered when the management plan was drawn up, but this is effectively a form of integrated control (Newman, University of Minnesota, Pers. comm. 2011). This lack of a nationwide consensus, coordination and strategy to deal with Eurasian water-milfoil infestations is probably the reason it still thrives in regions of the U.S.A. today (Homans & Newman 2011).

In South Africa, unlike the U.S.A., Working for Water (WfW), a division of the Department of Water Affairs (DWA), has the mandate of controlling alien plant infestations at a nationwide scale. Eurasian water-milfoil however, is currently not under any form of control. Without control, this highly problematic plant is envisaged to have a greater impact on the agricultural, environmental and recreational sectors of South African river systems (specifically those whose characteristics fit in with Table 1-5) due to the absence of severe winter conditions that cause massive die-back in the U.S.A. river systems.

## 1.5 *Study Aims*

Alien invasive weeds are known to be one of the main driving forces behind biodiversity losses in ecosystems, often resulting in the entire collapse of populations in communities, either directly or indirectly (Didham *et al.* 2005). In shallow water aquatic ecosystems worldwide, macrophytes feature prominently (Coops *et al.* 2002, Janauer 2006, Feldmann & Nõges 2007). The presence of indigenous macrophytes in aquatic ecosystems is threatened by the increase in alien weed infestations in these habitats, and the negative impacts of these weeds cannot be ignored.

Chapter 2 addresses the **primary** aim of this thesis, which is a spatial and temporal analysis of Eurasian water-milfoil and water hyacinth: the dominant alien macrophytes in the Vaal River. This study correlated abiotic conditions, both historical and present day, to macrophyte cover and determined where along the Vaal River stable populations of these macrophytes exist and why. WfW also has an integrated management plan for the alien macrophytes of the Vaal River. Their spray records were correlated with the historical data. For effective monitoring of future control measures which will be implemented against Eurasian water-milfoil, the conditions in which milfoil thrives were quantified, and its interactions with water hyacinth understood. This will allow for future accurate assessment of the effectiveness of the level of control, by means of monitoring how the above-mentioned variables change in response to levels of control.

To successfully adopt an integrated management strategy, the **secondary** aim of this dissertation was a baseline faunal survey, chapter 3. This survey determined what macroinvertebrates are associated with, and using, Eurasian water-milfoil, forming the basis for future research, especially regarding the naturalisation of Eurasian water-milfoil. The purpose of this survey was to identify what species of macroinvertebrates were associated with Eurasian water-milfoil and to establish whether the abundance and species richness of these macroinvertebrates were compromising Eurasian water-milfoil's competitive ability to dominate habitat. Similar studies were conducted on other invasive alien species in South Africa, including those by Hill (1998) on red water fern and by Schutz (2007) and Baars *et al.* (2010) on the indigenous coarse oxygen weed, *Lagarosiphon major* Ridley (Hydrocharitaceae) and Schutz (2007) on fine oxygen-weed, *Lagarosiphon muscoides* Harvey (Hydrocharitaceae).

Chapter 4 is a general discussion chapter, placing the findings of this research in a broader context. It interprets the results from Chapter 2 and 3 in light of the ecological theories of succession management and alternative stable states. In addition to this, Chapter 4 provides recommendations for future research studies on the Vaal River and discusses future control measures regarding any Eurasian water-milfoil infestations in South Africa.

# CHAPTER 2– Spatial Analysis of Dominant Macrophytes

## 2.1 *Chapter overview*

This chapter focuses on spatial and temporal changes in macrophyte dominance in the Vaal River, and the abiotic conditions that drive these changes. Satellite imagery was used to analyse changes in dominant cover of the macrophytes, differentiating between the floating water hyacinth, *Eichhornia crassipes* (C. Mart.) Solms. (Pontederiaceae) and submerged macrophytes, predominately Eurasian water-milfoil, *Myriophyllum spicatum* L. (Haloragaceae). The established macrophyte communities were mapped by a visual classification method using a temporal range of SPOT 5 satellite imagery in ArcView 9.3.1. Geographic Information Systems (GIS). In addition to establishing where and when the communities were growing or shrinking, the analysis also determined where and when communities changed in terms of macrophyte dominance. Historical water physio-chemistry data and herbicide spraying records were also obtained from Working for Water (WfW), and their relationships with changes in macrophyte cover were investigated. The aim of this chapter was to map the historical and current extent of the water hyacinth and Eurasian water-milfoil invasion to understand the mechanisms behind the recent changes in dominance.

## 2.2 *Introduction*

The Vaal River is one of only two large rivers flowing westwards from the Drakensburg Mountains (Van Vuuren & Pieterse 2005). As a result of its size and location, it supports 37% of South Africa's economic activity (Basson *et al.* 1997). The stretch of the Vaal River between the Vaal Barrage (26° 45' 54" S; 27° 41' 02" E) and its confluence with the Orange River below Douglas Weir (29° 04' 15" S; 23° 38' 09" E) is 793 km long and covers approximately 31 438.29ha of water area. This area is supplemented by a catchment of 194

000 $km^2$, making it the major potable water resource in the north western provinces of South Africa (Bruwer *et al.* 1985). The river's gentle slope between the Vaal Barrage and Douglas Weir (with an average drop of 0.58m/km), has resulted in its forming a series of large pools separated by small rapids (Bruwer *et al.* 1985). The Vaal River is also the primary route of disposal for various industrial, agricultural and domestic effluent (Cloot & Roux 1997) and the elevated nutrient levels (especially nitrates and phosphates), together with slow-moving water, leads to a system that provides an ideal habitat for aquatic macrophyte growth over a vast area, even though it is often subject to large stochastic events such as floods (Cloot & Roux 1997).

The Vaal River climate has a mean minimum temperature of $0^{o}C$ during peak winter months (June/July), resulting in 104 days of frosting a year (Schulze *et al.* 1997). In the absence of severe winters compared to the Northern Hemisphere, the most influential stochastic events taking place along the system are flooding and droughts (Chambers *et al.* 1991, Biggs 1996, French & Chambers 1996), however these are offset by the high number of impoundments found on the Vaal River (Braune & Rogers 1987) (Figure 1-2). Flooding events submerge macrophyte beds below levels where photosynthesis can take place and often scour the system, removing submerged aquatic plant beds and displacing floating macrophytes from the water, where they die (Bunn & Arthington 2002). Flooding also oxidises the sediment. This, combined with decaying vegetation matter and runoff from agricultural activities in the catchment, influences water chemistry and nutrient levels of the waters, which in turn promotes macrophyte growth (Bailey-Serres & Voesenek 2008).

### 2.2.1 Dominant macrophytes of the Vaal River

The abiotic conditions present in the Vaal River, as discussed in Chapter 1, have presented alien aquatic macrophytes with the perfect platform for invasion. Parrots feather, *Myriophyllum aquaticum* (Vell.) Verdcourt (Haloragaceae); red water fern, *Azolla filiculoides*

Lam. (Azollaceae) and water hyacinth are floating macrophytes that occur in the Vaal River (with water hyacinth being the most problematic) (Coetzee *et al.* 2011b). The integrated control measures introduced against the floating weeds, and high variability of stochastic events such as flooding, have allowed submerged plants, particularly the invasive Eurasian water-milfoil, to proliferate in the system.

Water hyacinth was first observed as a problem plant in the Vaal River in the 1980s and since then it has spread a distance of 300km (Henderson & Cilliers 2002). Traditionally, water hyacinth has dominated much of the system due to its rapid rate of reproduction and ability to shade out submerged macrophytes. The reliance of floating plants on high nutrient concentrations is a direct result of their growth form. Floating plants are able to extract nutrients only from their roots, while submerged macrophytes are often able to extract nutrients from both sediment and water column, making them better competitors in nutrient-depleted waters (Scheffer *et al.* 2003). However, an integrated management plan implemented by Working for Water (WfW), making use of biological control as well as herbicidal treatment, has resulted in a recent noticeable decline in water hyacinth and a concurrent increase in Eurasian water-milfoil (as introduced in Chapter 1).

Eurasian water-milfoil is a highly problematic plant that is expected to have a greater impact on the agricultural, environmental and recreational sectors of South African river systems than in North America, due to the absence of severe winter conditions that cause massive die-back in the U.S.A. river systems. Since its unknown mode of introduction in 1885, near Barkly West on the Vaal River (28°32’45” S; 24°30’50” E) (SAPIA ARC PPRI Database), Eurasian water-milfoil has remained at low levels of cover until it recently became a problem plant from 2007 (D.Sharpe, WfW, Pers. comm. 2011).

Submerged aquatic macrophytes such as Eurasian water-milfoil, and floating aquatic macrophytes such as water hyacinth are not able to coexist in the same area for extended periods of time (Scheffer *et al.* 1993). Floating macrophytes shade out submerged macrophytes, or conversely, submerged macrophytes limit floating plant dominance through access to nutrients from the water column. Besides integrated control measures introduced by WfW against water hyacinth, the abiotic environment and competition between the plants could also have contributed to the observed change in dominance. According to Ruiz Téllez *et al.* (2008), water hyacinth prefers pH levels between 6 and 8 as well as high levels of phosphates (62mg/L) and nitrates (21mg/L) in the water column and thus growth rates are restricted at low water nutrient levels. At suitable pH, the more nutrients available to water hyacinth, the better it performs. Similarly, Eurasian water-milfoil enjoys eutrophic waters (high nitrate and phosphorous levels), but can also survive in oligotrophic systems, as a result of its ability to abstract nutrients from sediments (Barko 1983). Ali & Soltan (2006) showed how calcium levels were singled out as the most important sediment nutrient regarding the success of Eurasian water-milfoil populations in Egypt. Smith *et al.* (2002) showed how significant increases in auto-fragmentation rates were recorded when total soil nutrient nitrogen levels were greater than 0.44 mg/g. Sediment nutrient uptake rate is however dependent on the sediment particle size composition, as evidenced by Wang *et al.* (2009) who showed that Eurasian water-milfoil grows better in sandy sediments than in clay or silt sediments. It is evident that sediment characteristics play an essential role in the health and competitive ability in the aquatic ecosystems.

With linear ecosystems such as rivers, there are various influences that consistently affect the quality of the aquatic abiotic environment. Water quality differs over the entire extent of the Vaal River (Figure 2-1) (with dams such as Bloemhof, Grootdraai and the Vaal Dam acting as buffers increasing water quality downstream of them) and therefore, to determine where macrophytes proliferate, spatial and temporal analysis of the distribution and dominance of macrophytes is required to better manage the system and prepare for species responses to physio-chemical changes in water quality. The various regions of the Vaal River require specific techniques to manage the system as the plants are exposed to localised conditions, and therefore respond differently. Hence analysis on both a spatial and temporal scale is required to fully understand the system.

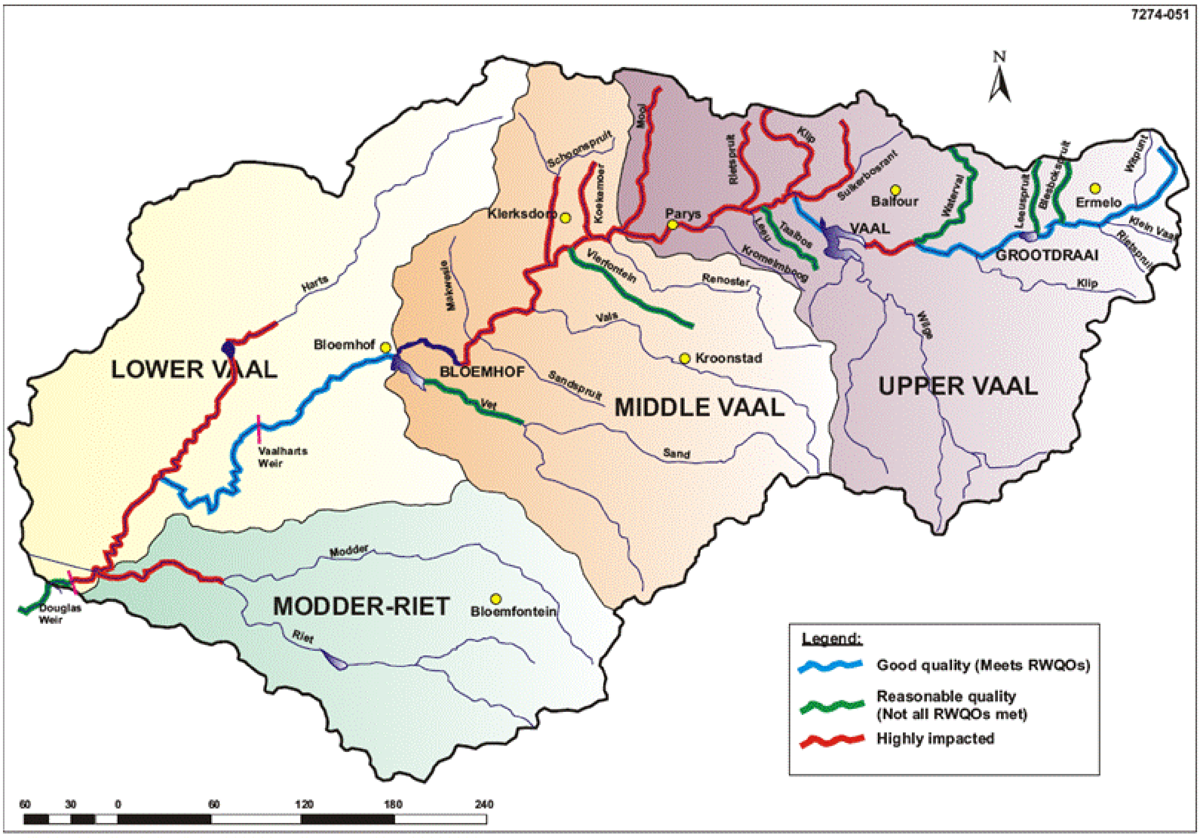



**Figure 2-1:** A map illustrating the compliance of different regions in the Vaal River to Resource Water Quality Objectives (RWQOs) set out by the Department of Water Affairs (Moodley *et al.* 2006).

## 2.2.2 Spatial and temporal analysis

The two primary surveying methods utilised to monitor spatial and temporal changes in aquatic ecosystems are ground-based sampling or field surveys, and Geographic Information Systems (GIS) remote sensing (Shuman & Ambrose 2003). GIS have long been used to analyse land-use trends. The development of one of the first GIS, the Canada Geographic Information System in the 1960s, was driven by the need for policies over the use of land (Longley *et al.* 2001). GIS has since been defined as computer systems having the ability not only to display geospatial data, but to effectively scrutinize, challenge, process and digitally save such data (Chang 2006). GIS analysis allows the user to accurately assess the history of an ecosystem and is primarily used for habitat and species mapping, biodiversity determination, land change detection and conservation area monitoring (Ozesmi & Bauer 2002, Shuman & Ambrose 2003, Turner *et al.* 2003, Cohen & Goward 2004, Schmid *et al.* 2005, Baker *et al.* 2006, 2007).

Ground or field surveys can provide highly accurate spatial and taxonomic data, but are limited in terms of timeframe (providing only a snapshot of the current situation), sampling time (it takes large quantities of time to complete), scale (limited timeframes result in smaller areas sampled), and in addition to all of these factors, the actual sampling event generally disturbs the ecosystem which is being sampled, with the potential of skewing results (Phinn *et al.* 1996). It is commonplace to combine the advantages of both methods (remote sensing and field surveying) to obtain an effective strategic sampling method. This method has inaccuracies in terms of relatively low resolution of data and cost implications, but when combined with ground surveys, the data become highly accurate and applicable (Shuman & Ambrose 2003). This is particularly true in the case where large volumes of spatial data are mapped and are then informed and corrected for by field work, where the field worker can accurately delineate what was mapped by visually ground-truthing the data. These techniques are increasingly being used in biological sciences as an efficient, accurate and robust tool to illustrate large changes in aquatic ecosystems over time (Davranche *et al.* 2010). Examples of such studies that mapped Eurasian water-milfoil include those conducted by Boylen *et al.* (2006) in North America, Davranche *et al.* (2010) in France and Yuan & Zhang (2008) in China. To date no such studies involving Eurasian water-milfoil have been conducted in South Africa. The accuracy of the GIS remote sensing result is however highly dependent on the classification techniques selected, which is customised depending on the user's scale and accuracy requirements.

### 2.2.3 Image classification techniques

GIS have various classification techniques that can be used to analyse imagery, and are based on a set of user-defined parameters. Depending on the chosen classification technique, different results could be obtained from the same image and thus the skill of the "classifier" is paramount to the outcome. A general flowchart regarding a range of acceptable classification procedures is presented in Figure 2-2.

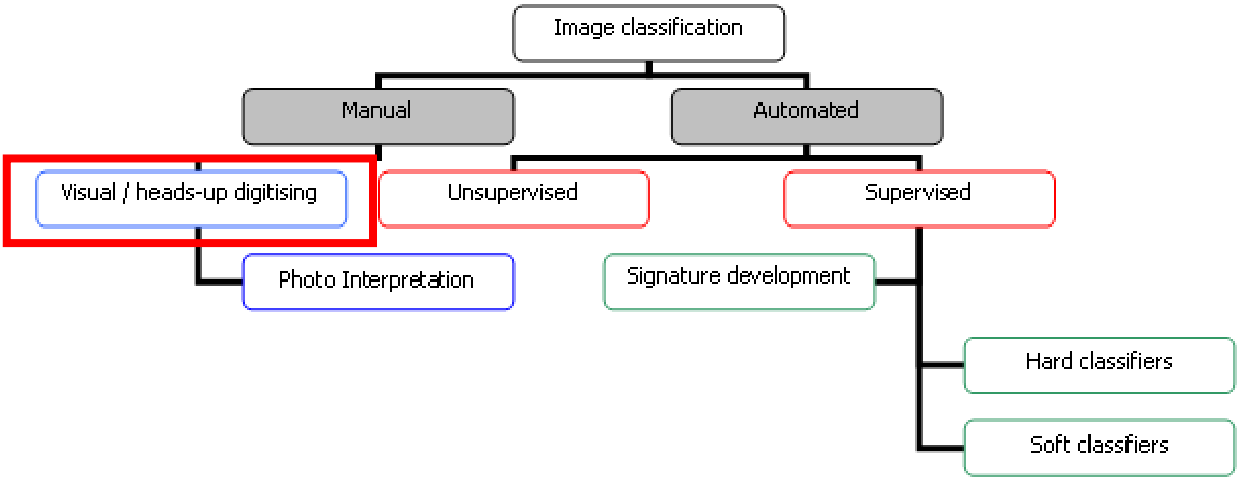



**Figure 2-2:** A flow diagram of different image classification procedures. The red box indicates the technique selected in this study, Visual/heads-up digitising (Eastman 1999, Pouncey *et al.* 1999)

There are several different remote sensing classification techniques used in biological sciences; the only manual method is Visual Interpretation, while automated methods include Unsupervised Classification or Clustering and Supervised Classification (Ozesmi & Bauer 2002). The primary advantage of using an automated remote sensing technique is that once the system is correctly configured, the user can repeat the technique on several images, in relatively quick succession and use hard or soft classifiers to achieve the most accurate representation (Cobbing 2006). Hard classifiers confine each pixel to one definitive class, the most commonly used hard classifier is that of the Maximum Likelihood classifier (Eastman, 1999). Soft classifiers break down the likelyhood of a pixel being confined to a specific class and assign the pixel a range of values so that the user can analyse what the probability is for each pixel has to belong to a specific class (Eastman, 1999).

Automated remote sensing techniques can be quicker, and each replicate has the same degree of error, resulting in consistent results, allowing for less bias, but then may not be as accurate as visual classification techniques. Manual visual classification techniques are better suited for smaller scale studies and studies that require fine detail classification, especially when analysing aquatic vegetation habitats which lack distinctive differences in colour and texture (Cobbing 2006). Both methods have distinct limitations and advantages related to the scale of the study and the detail required from the exercise in the form of the Minimum Mapping Unit (MMU) requirement.

MMU can be defined as “the smallest size aerial entity to be mapped as a discrete entity” (Lillesand *et al.* 1994). Based on the MMU requirements, the user will select the most appropriate image depending on the availability, cost and resolution. The importance of low MMU values was illustrated by Seher and Tueller (1973), Shima *et al.* (1976), Howland (1980), Lehmann and Lachavanne (1997), who all concluded that the lowest MMU values and smallest pixel resolution was the most suitable for detailed aquatic vegetation mapping.

### 2.2.4 South African image availability and suitability

The oldest images available for remote sensing of the Vaal River from the South African National Space Agency (SANSA) are National Oceanic and Atmospheric Administration Advanced Very High Resolution Radiometer (NOAA-AVHRR) and Landsat MSS Landsat 5 TM / Landsat 7 ETM images. Although these images for the Vaal River have coverage dating back to as early as 1984 (Table 2-1), they are only suitable for studies that can tolerate high MMU and thus were not suitable for mapping aquatic macrophytes in this study.

**Table 2-1:** A table illustrating the suitability of images available from the South African National Space Agency (SANSA)

| Minimum Mapping Unit (ha)* | Theoretical pixel size (m) | Equivalent satellite/sensor ** | Year available from | Size of Scene | Theoretical number of scenes required per year for study site | Cost of scene | Suitability |
|---|---|---|---|---|---|---|---|
| 1000 | 1000 X 1000 | NOAA-AVHRR | 1984 | 2400km X 6400km | 1 | Free | Low |
| 50 | 224 X 224 | TERRA-MODIS | 2003 | 10km X 10km | 600 | Free | Low |
| 30 | 175 X 175 | SAC-C | 2008 | 90km X 1150km | 2 | Free | Low |
| 5 | 71 X 71 | Landsat MSS | 1972 | 170km X 185km | 3 | R 3 000 | Low |
| 1 | 32 X 32 | Landsat 5 TM / Landsat 7 ETM | 1972 | 170km X 185km | 3 | R 3 000 | Low |
| 0.4 | 20 X 20 | CBERS | 2008 | 130km X 130km | 7 | Free | Low |
| 0.25 | 16 X 16 | Landsat 7 Pan | 1999 | 170km X 185km | 3 | R 3 000 | Low |
| 0.1 | 10 X 10 | SPOT 2 | 1994 | 60km X 60km | 10 | RSA university student 10 free per year after which (R1000/scene) | Low |
| 0.0025 | 5 X 5 | SPOT 4 | 1999 | 60km X 60km | 10 | RSA university student 10 free per year after which (R1000/scene) | Low |
| 0.006 | 2.5 X 2.5 | SPOT 5 | 2006 | 60km X 60km | 10 | Free for RSA Government Departmental Projects | Highly suitable |
| 0.0006 | 0.75 X 0.75 | Aerial Image CD:NGI | 1980 | 5km X 5km | 120 | R 21/dvd, 21c/image | Suitable |

*This is only a theoretical MMU, which is the equivalent to one pixel, in practice MMUs consist of a minimum of three pixels (Chang 2006)

** All images can be obtained from SANSA except the aerial images which are available from the Chief Directorate of National Geospatial Information (CD NGI) at the Department of Rural Development and Land Affairs.

The largest database of historical high resolution GIS imagery is the National set of black and white aerial imagery, held by CD NGI. The advantage of using aerial images is that they have the highest spatial resolution available and smallest pixel resolution (Table 2-1), and can give an accurate historical account of the study area. The limitations of using this form of imagery stem mainly from the fact that these images are black and white. This makes it particularly difficult to identify submerged macrophytes or differentiate between water hyacinth and sediment. The Northern Cape is also the most sparsely populated region in South Africa and thus the motivation to have the area flown for aerial images is minimal as the Department of Rural Development and Land Affairs concentrates its funds on flying populated regions of the country and therefore there is also only partial coverage for the study area (Figure 1-2). Similar limitations were encountered by Adam *et al.* (2009), who concluded that aerial photography was not feasible on a regional scale and that satellite imagery such as Systeme Pour l'Observation de la Terre (SPOT) imagery was better suited for mapping aquatic vegetation communities as a result of its spatial coverage. The primary reason for this is the recent improvements in SPOT imagery pixel resolution and availability.

The SPOT series of satellites was first launched by the French Space Agency, Centre National d'Etudes Spatiales (CNES), in 1986 with SPOT I. Since then SPOT II, III, IV and V have been launched (Chang 2006). The SPOT V satellite was launched in 2002 and the first images were received by SANSA in late 2006. The SPOT V satellite carries two types of sensor and the images are broken down into four separate spectral bands (Figure 2-3). The first sensor captures multi-spectral images with a 10m X 10m resolution, while the second sensor captures panchromatic images (PAN), with a 2.5m x 2.5m resolution (Chang 2006). Each image has a swathe of 60km x 60km and the entire research study area (Figure 1-2) is covered by 10 images.

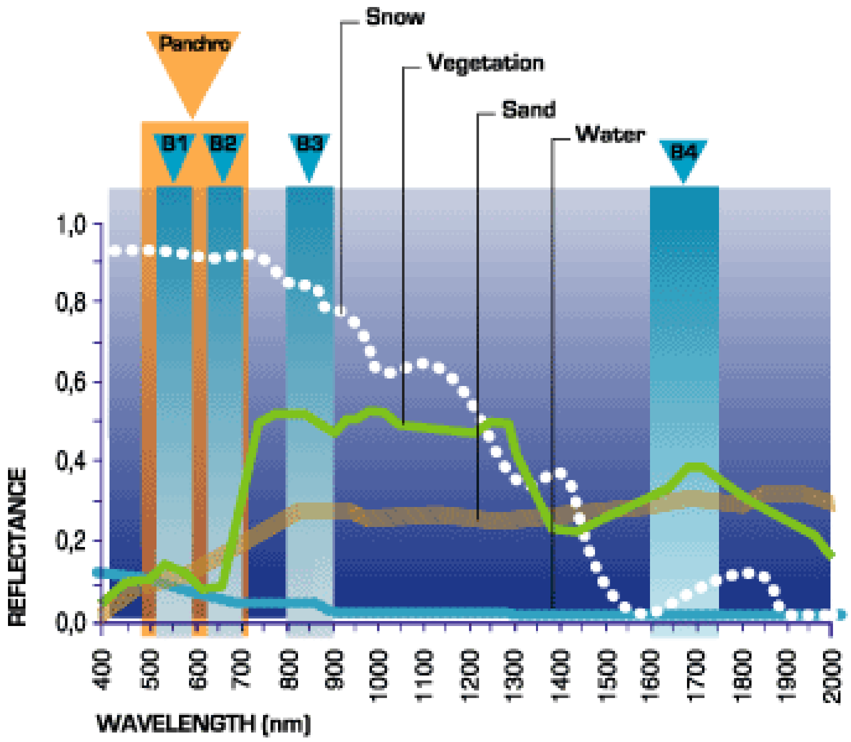



**Figure 2-3:** A breakdown of the reflectance bands which the SPOT V sensor collects (www.spotimage.com).

SANSA takes the separate image band data from each sensor and merges and processes each image band together for a combined pan-enhanced image with a resolution of 2.5m x 2.5m (Chang 2006). These are known as pan-enhanced images and although they cannot be used for any automated multispectral image classification (due to the merging of the various spectral bands), they are suitable for manual visual mapping purposes, which were used in this study. The accuracy of using SPOT V imagery to map submerged macrophytes is directly influenced by the proportion of the plants that float on the surface of the water as well as the density of the plant beds and the water clarity and salinity (Davranche *et al.* 2010). Despite these limitations, several authors have managed to successfully use satellite imagery to map macrophytes. Boylen *et al.* (2006) for example, effectively mapped the extent of Eurasian water-milfoil populations in North America, while Khanna *et al.* (2011) mapped the changes in water hyacinth and aquatic macrophytes over time in Florida. Similar studies in South Africa have coarsely determined the extent of freshwater macrophytes at a large scale, for the National Wetlands Inventory and also used remote sensing as a tool to delineate macrophytes (Darwall *et al.* 2009). The physical extent of water hyacinth versus that of a submerged aquatic plant over a vast extent of river has never been mapped before in South Africa, but given the success of using GIS and its ability to monitor changes in ecosystems over time (Ozesmi & Bauer 2002), it was selected as the most appropriate method for this study.

Ecosystems are dynamic and change continuously, and satellite photographs are snapshots in time. The aim of this chapter was to analyse the historical and current extent of the dominant macrophyte infestations in the Vaal River, as well as to comprehend mechanisms behind their observed dominance. Visual classification of SPOT V satellite imagery was used to monitor changes in the macrophyte populations since 2006 and to correlate of these data with historical abiotic data.

Over such a protracted time period of analysis weather conditions to which the Vaal ecosystem is exposed change. The frequency of stochastic events, such as flooding or droughts, have long term impacts on the macrophytes ability to proliferate and thus these factors were considered and also recorded. A field survey was then conducted to collect data on macrophyte plant population densities, growth forms (water hyacinth populations only), presence of biological control agents (water hyacinth populations only), physio-chemical properties (of both water and sediment) and to determine precise spatial locations which was also used to determine the accuracy of the remote sensing analysis.

## 2.3 *Materials and Methods*

### 2.3.1 Study Site

The study area spanned from the Vaal River Barrage to the confluence of the Vaal and the Orange Rivers (Figure 1-2). This area is divided into three large Water Management Areas (WMA) by Working for Water (WfW), which consist of 24 smaller sub-areas (Figure 2-4). Each WMA (8, 9 and 10) is managed separately by a different WfW manager who is responsible for the application of herbicides on alien weeds within his/her region.

### 2.3.2 Significant threats to macrophyte populations

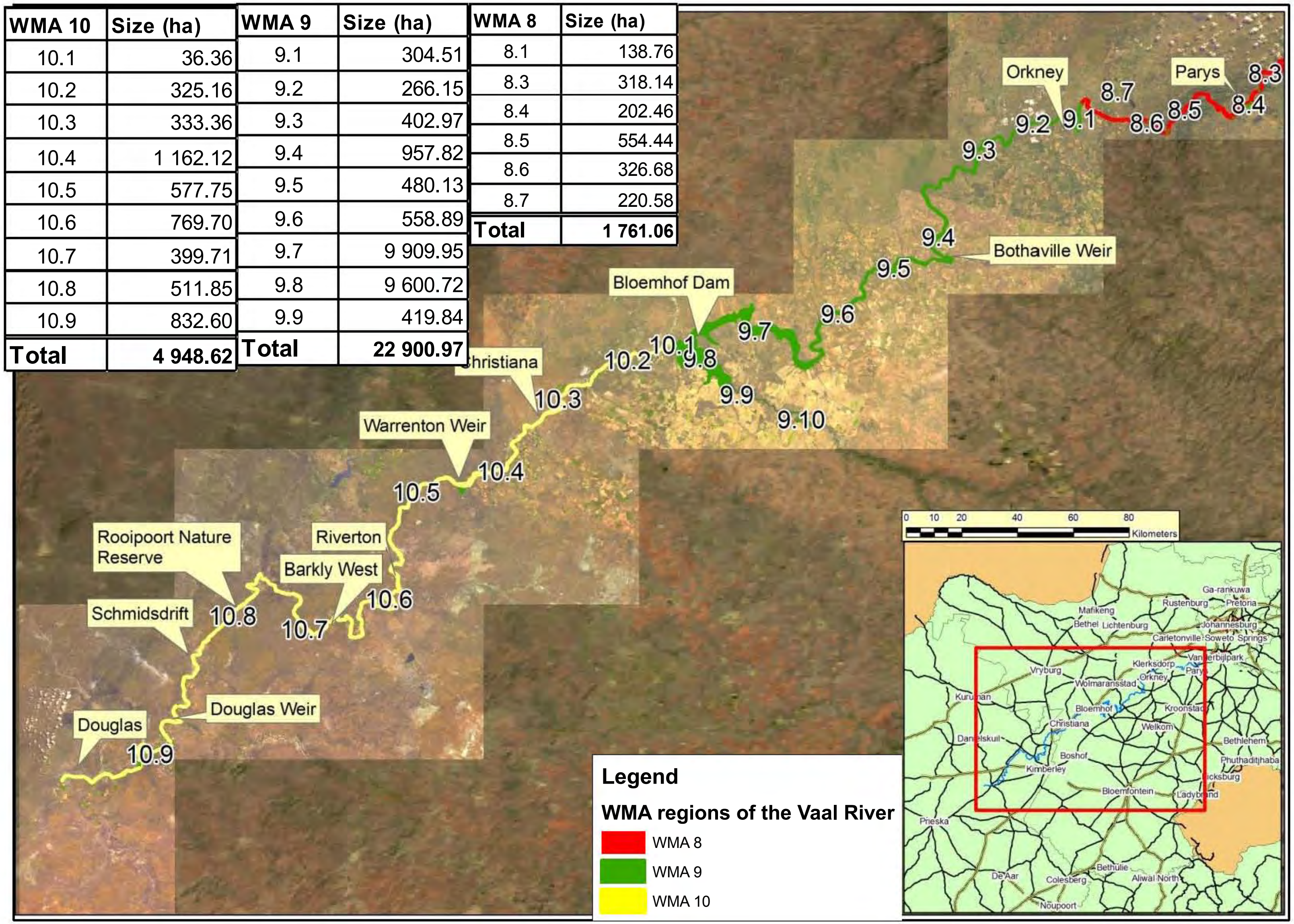
Natural stochastic events (such as weather extremes) have the ability to completely rest a system in terms of macrophyte dominance. This is achieved by means of decimating populations of macrophytes thus resetting niches in the ecosystem (Scheffer *et al.* 2001) Information on stochastic events in the form of the frequency of floods and droughts was obtained from DWA RQS in July 2011, for the entire Vaal River. Another significant threat to the alien macrophytes population levels is the integrated control measures used by WfW. No integrated control measures are yet in place against Eurasian water-milfoil, yet water hyacinth populations are subjected to integrated control measures in the form of herbicide application and biological control agents. Herbicide application records were obtained by personal communication with the three WfW WMA managers of the study area, but historical records of biological control agent population levels were not kept. All these data were then compared to the results of the spatial analysis to determine how changes in spatial cover of the macrophytes were affected by stochastic events. Every region was also mapped in its entirety, using remote sensing imagery, and the majority of the study area was accessed and the spatial data was visually ground-truthed during the field survey.

### 2.3.3 Spatial Analysis

Prior to spatial analysis, it was important to determine when major stochastic events such as floods occurred to determine how these events may have influenced results. Flood data were obtained from DWA Resource Quality Service division (RQS) for the Vaal River and were statistically correlated with the spatial analyses results to determine what effect these events had on the macrophyte populations of the Vaal River from 2006 until 2010.

ArcMap v9.3.1 (ESRI, Redlands, California) was used to analyse SPOT V imagery. Suspected patches of submerged macrophytes and water hyacinth were mapped using a combination of different visual classification techniques from SPOT 5 satellite imagery in ArcView GIS. All raster datasets were resampled using the cubic convolution technique to improve the visual effect. This technique averages the nearest 16 pixels to determine a new pixel value (ESRI, Redlands, California), which produces a smoother image while maintaining edge detail (Repaka *et al.* 2004). Cubic convolution was selected by authors such as Fuller *et al.* (2002), who preferred it over other resampling techniques (such as nearest neighbour), and although it marginally changes the original pixel value, the increase in edge detail makes it easier to visually map vegetation communities (Lillesand *et al.* 1994, De Wit & Clevers 2004). Delineation of the polygon edges of the water hyacinth mats was compiled using the true colour SPOT V image (Figure 2-5). Submerged macrophytes were more difficult to delineate and the raster SPOT V symbology properties of the images were manipulated to produce a false colour SPOT V image (Figure 2-6).

False colour images assisted in highlighting the extent of submerged macrophyte beds (Figure 2-6), not visible on the true colour image (Figure 2-5). To compile the false colour image in ArcMap v9.3.1, only the blue (Band 2) and red bands (Band 3) of the image were displayed (Figure 2-6).




| WMA 10 | Size (ha) |
|---|---|
| 10.1 | 36.36 |
| 10.2 | 325.16 |
| 10.3 | 333.36 |
| 10.4 | 1 162.12 |
| 10.5 | 577.75 |
| 10.6 | 769.70 |
| 10.7 | 399.71 |
| 10.8 | 511.85 |
| 10.9 | 832.60 |
| **Total** | **4 948.62** |

| WMA 9 | Size (ha) |
|---|---|
| 9.1 | 304.51 |
| 9.2 | 266.15 |
| 9.3 | 402.97 |
| 9.4 | 957.82 |
| 9.5 | 480.13 |
| 9.6 | 558.89 |
| 9.7 | 9 909.95 |
| 9.8 | 9 600.72 |
| 9.9 | 419.84 |
| **Total** | **22 900.97** |

| WMA 8 | Size (ha) |
|---|---|
| 8.1 | 138.76 |
| 8.3 | 318.14 |
| 8.4 | 202.46 |
| 8.5 | 554.44 |
| 8.6 | 326.68 |
| 8.7 | 220.58 |
| **Total** | **1 761.06** |

**Figure 2-4:** The study area, showing the location and sizes of the WfW Management Areas (WMA) the extent of these segments of the river were defined by WfW for management and reference purposes, extending from the Vaal River Barrage (26° 45' 54"S; 027° 41' 02" E) to the confluence at Douglas (29° 04' 15" S; 23° 38' 09" E).

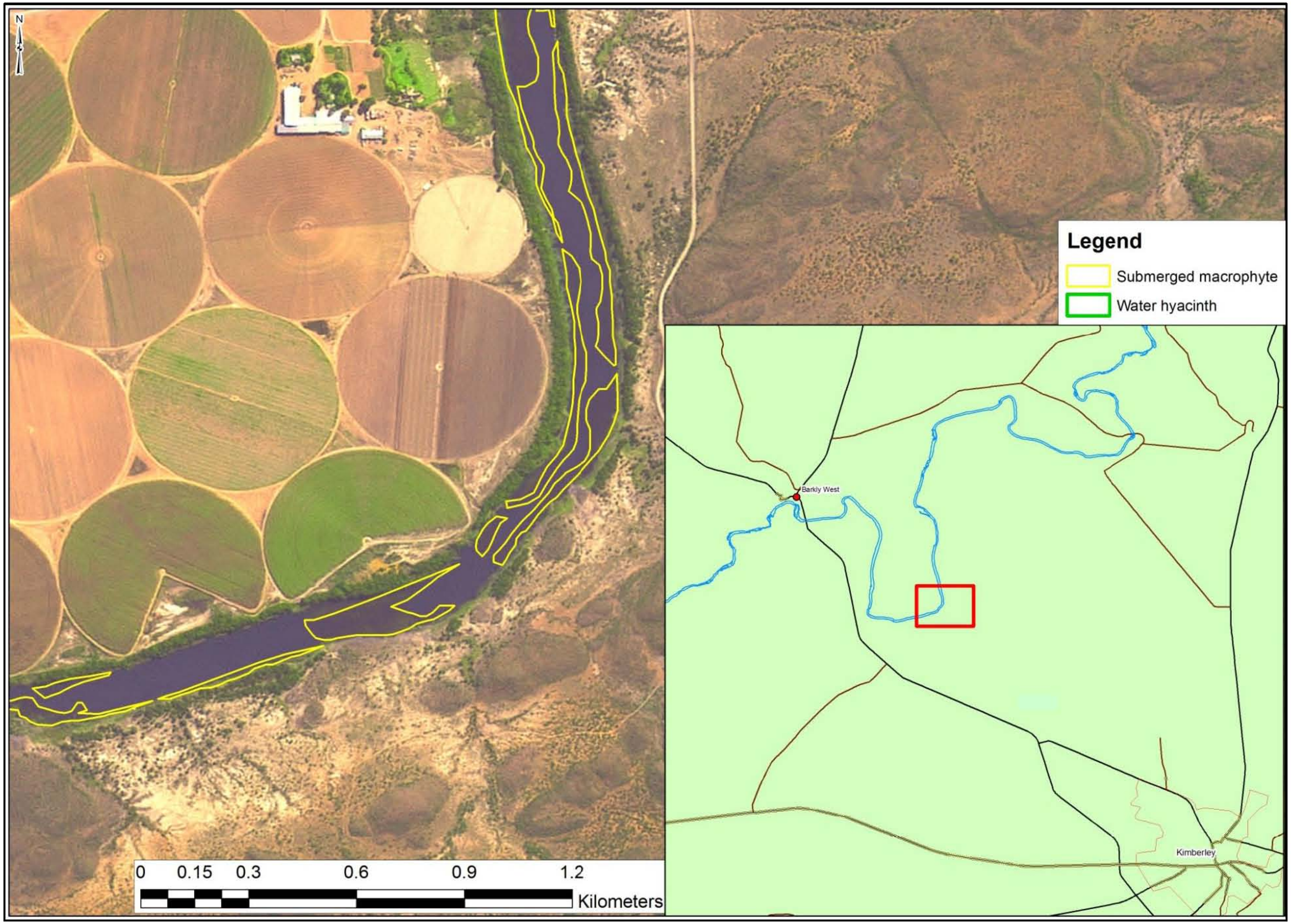



**Figure 2-5:** A true colour SPOT V image (South East of Barkly West on the Vaal River) without any band prioritisation, using default colours. Submerged aquatic macrophytes are outlined in yellow. The insert illustrates the location of the site relative to the nearest city, Kimberley.

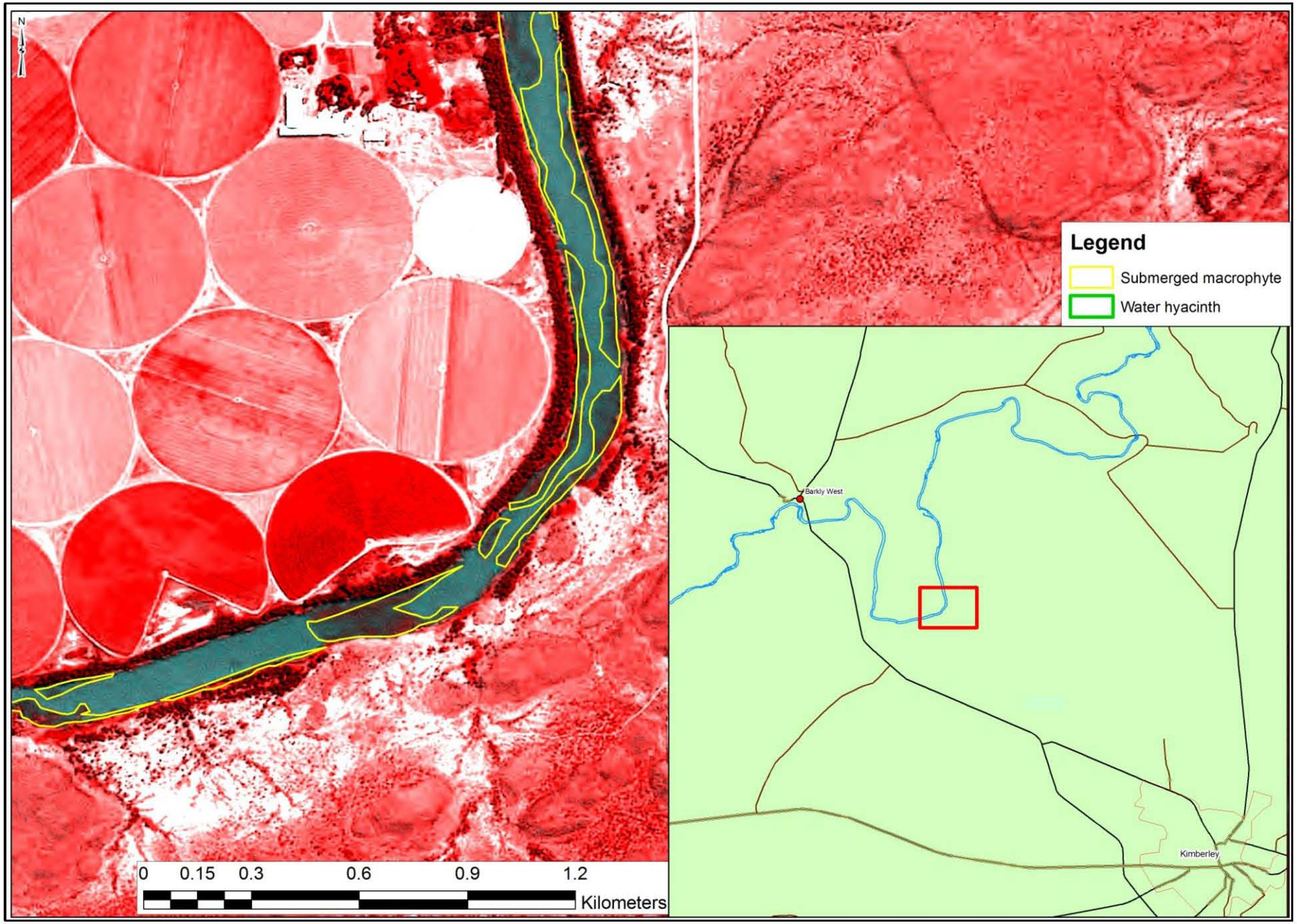



**Figure 2-6:** A false colour SPOT V image (South East of Barkly West on the Vaal River) with red band prioritisation, submerged aquatic macrophytes are outlined in yellow. The insert illustrates the location of the site relative to the nearest city, Kimberley.

The red band of the image is closest to the vegetation reflectance value (Figure 2-3), and by prioritising this band the vegetation had its highest reflectance properties, this resulted in deeper submerged macrophyte beds of lower densities becoming more visible for delineation purposes. The blue band of the image allowed for high water reflectance of the Vaal River, facilitating the accurate delineation of submerged macrophyte beds by illustrating the actual extent of the Vaal River even if there were large trees shading portions of the river (Figure 2-6).

Brightness and contrast settings of the image were also fine-tuned (generally a contrast setting of 70% and a brightness setting of 5% was used) to highlight the submerged macrophytes present in the river (Figure 2-6). The setting change resulted in the SPOT V RGB false colour composite image, which better illustrated the edges of the submerged macrophyte beds and assisted in mapping the macrophytes (Figure 2-6). The resultant layer was compared to the true colour image and modified if any additional beds were missed, before proceeding to the next image. This technique was primarily responsible for assisting in delineating the true extent of submerged macrophyte beds that were not visible in the true colour image. Submerged macrophyte beds that were not dense enough to float on the water surface were difficult to delineate using just the true colour image and the false colour image was invaluable in highlighting these beds which may have otherwise gone unmapped. This technique was applied across the entire study area for every year from 2006 until 2010.

### 2.3.4 Ground-truthing of spatial data

The primary aims of the ground-truthing field trip were to determine the accuracy of the original visually-mapped spatial data, and secondly to collect information regarding the abiotic conditions of the river. The field trip took place (in October 2010), after the spatial data were captured in the form of vector shapefile, and all suspected macrophyte units were digitised (Figure 2-5). The maps used during the field trip to locate suspected macrophyte patches that were remotely delineated, consisted of the polygons delineated and displayed against 2009 SPOT V imagery, as 2010 imagery was not available until August 2011. Field maps and datasheets were then printed and the digitised data were verified in-field for species, density, and growth-form in the case of water hyacinth. Field observations were used to update the previously digitised map data. Abiotic physio-chemical measurements, water and sediment samples, were also collected and used in later analysis (described in detail in 2.3.5). Statistica v10 was used to conduct all the statistical analyses, scatterplot data and correlations, while Microsoft Office 2007 was used to compile graphs.

Distribution and abundance of water hyacinth and submerged macrophytes on the Vaal River was determined during a field survey in October 2010 using a boat to navigate down (bordering) one bank and then back up the opposite bank of the Vaal River returning to the launch site. Due to the size of the study area (Figure 1-2), a rapid assessment technique was developed. Wherever Eurasian water-milfoil or water hyacinth was observed from the boat, a Global Positioning System (GPS) co-ordinate (point) was taken with a Garmin e-Trex GPS, and the width of each population from the river bank was estimated. In addition to these data, Eurasian water-milfoil beds were also assigned a variety of density classes (Figure 2-8).

Kenow *et al.* (2006) found a linear relationship between plant biomass and density of the Eurasian water-milfoil beds. Based on this relationship, a density score was assigned to submerged macrophyte beds based on the frequency and abundance of the submerged macrophytes (Figure 2-8). Determination of Eurasian water-milfoil biomass was undertaken within all the density classes; dense (5), abundant (4), frequent (3), occasional (2) and rare (1), and replicated at 19 different random locations throughout the system, at an average of one different location per day in the field. Data were converted back to macrophyte dry mass via the methods developed by Kenow *et al.* (2006) used in North America for submerged macrophyte sampling (Yin *et al.* 2000).

The procedure began with the random placement of a boat inside the extent of a Eurasian water-milfoil bed, following which the bed was scored based on the observed percentage cover of the bed (Figure 2-8). A double-sided rake used for collecting had a 3m long handle with a 36cm wide head, with 14 5cm long teeth on each side, similar in design to the rake used by Yin *et al.* (2000). The rake was then dragged for 2m along the river bed in six different directions (Figure 2-7) and rotated 180$^{o}$ before being pulled to the surface and scored according to the percentage of the rake teeth that were covered (1 = <20%, 2 = 20 - 40%, 3 = 40 – 60%, 4 = 60 – 80%, 5 = 80-100%). The dry biomass of the submerged macrophyte patches was determined from an average of the six rake drag scores at each site using the following formula:

$$\mathrm{Exp}^{(\ln(gDM.m\text{-}2))}-1 = -0.070+(3.343*rake)+(-0.836*rake^{2})+(0.081*rake^{3})$$

From the formula, $gDM.m^{-2}$ represents grams of dry mass of the Eurasian water-milfoil per square meter, and rake is an average rake score. These results were used to calculate the total biomass of Eurasian water-milfoil of the Vaal River, by multiplying $gDM.m^{-2}$ by the area covered for each density score (Figure 2-7). The statistical analysis was performed on the field survey data and density of the observed Eurasian water-milfoil beds. Pearson's correlation coefficient tests determined the degree of correlation.

In the U.S.A., this field sampling method was found to be effective in determining the presence and composition of submerged aquatic vegetation beds of the Mississippi River. The Eurasian water-milfoil had a significant correlation of 0.725 to the harvested quadrat of biomass sampled (Kenow *et al.* 2006). The method is also used by the United States Geological Survey Long Term Resource Monitoring Programme throughout the U.S.A. However, due to the size of this study a randomised sampling procedure was preferred over the grid system of analysis used by Yin *et al.* (2000), in an attempt to reduce the number of sites investigated in a given area.

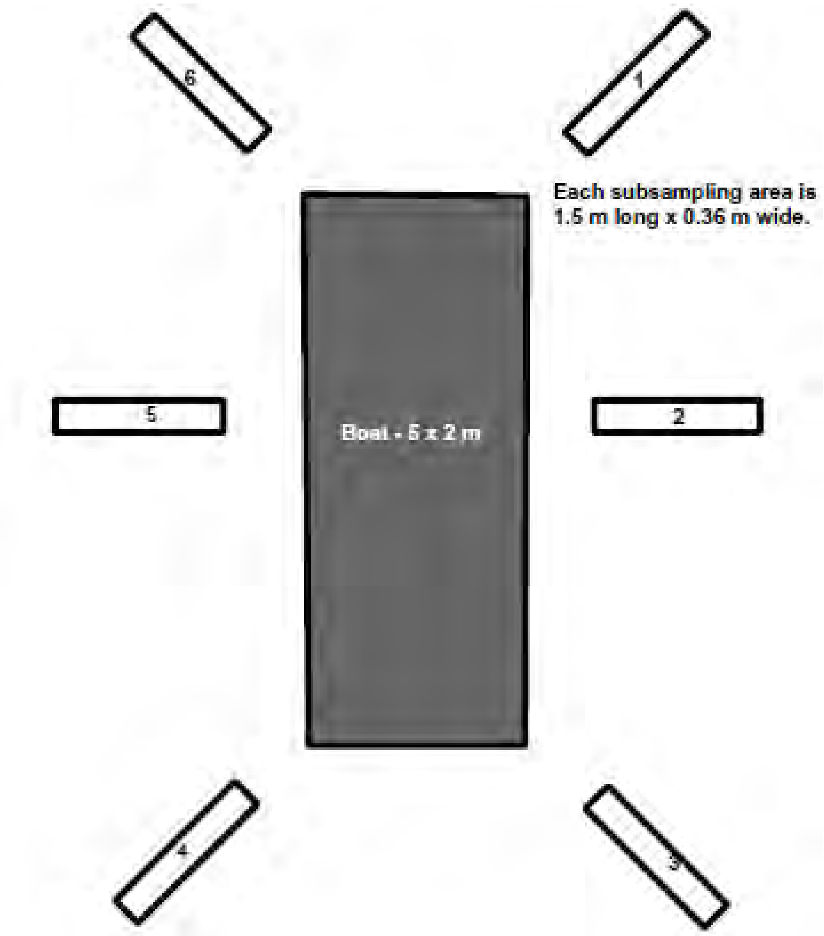



**Figure 2-7:** The various directions that each rake was dragged to determine the density of Eurasian water-milfoil beds (Yin *et al.* 2000)

Water hyacinth populations were analysed to determine the extent of their cover and state of dominance. The following plant and agent parameters were recorded at each site: number of ramets, flowers, leaves, maximum root length, length of the petiole of the second leaf, length of the longest petiole, and presence or evidence of biological control agent feeding (Center 1981). The arrangement of water hyacinth leaves is in the form of a spiral around the crown of the plant, with the newest leaf unfurled closest to the crown/centre of the plant followed by leaf two which is the next closest leaf (Center 1981).

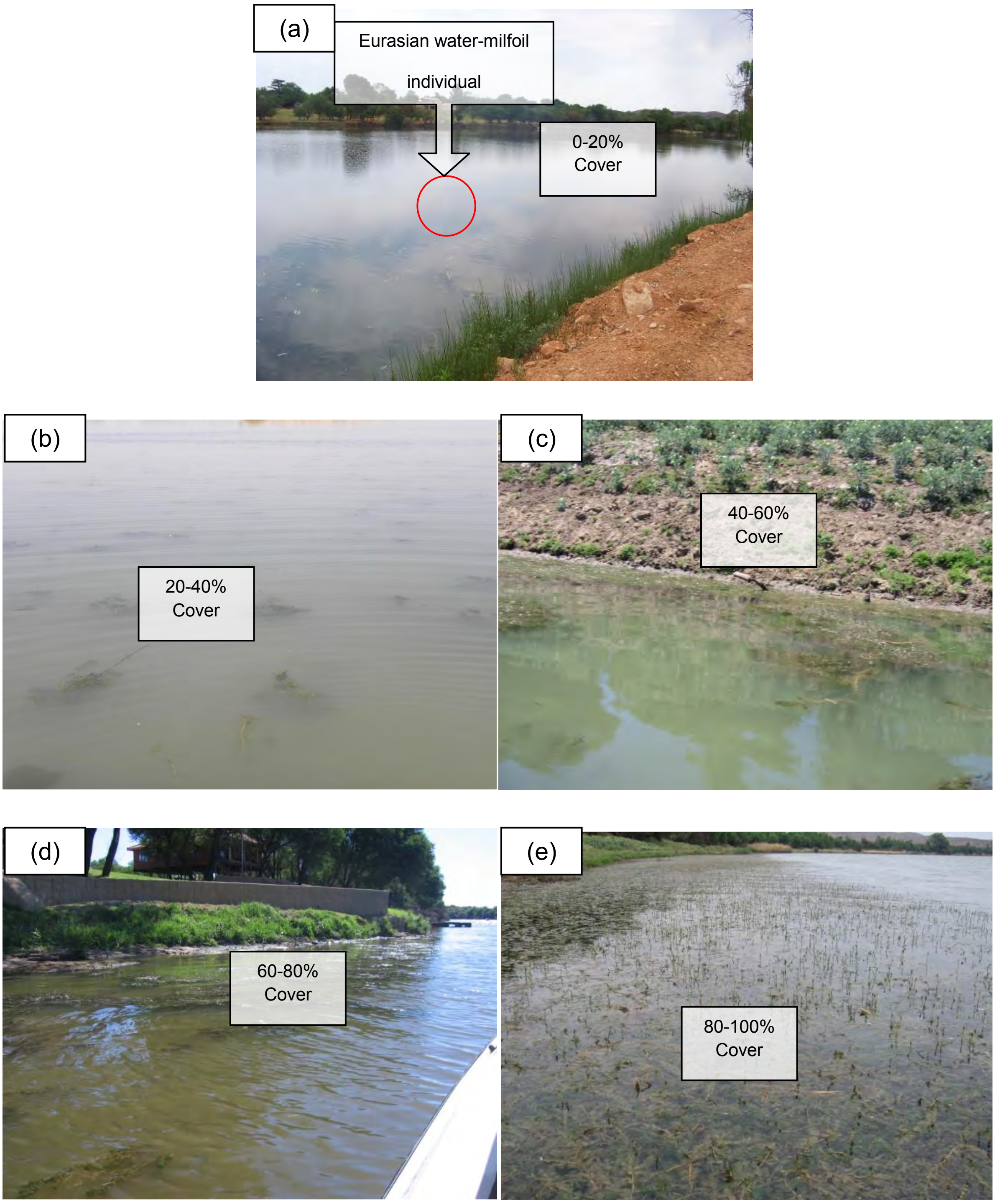



**Figure 2-8:** A selection of photos illustrating examples of Eurasian water-milfoil beds scored according to their density (a) rare bed with a single individual plant, (b) occasional bed, (c) frequent bed, (d) abundant bed and (e) dense bed.

A community of water hyacinth plants at the site was also classed by plant phenostage (growth form) according to Center *et al.* (1999): (1) incipient - few small plants, inflated petioles, (2) scattered - patches of small plants, open canopy, (3) coalescing: medium plants, large mat, closed canopy or (4) mature - mainly tall plants attenuated petioles, which gives an indication of the age of plants. Individually selected plants within a population were also classed according to growth form: (A) short - small healthy plants inflated petioles, (B) medium- medium height, healthy plants, petioles inflated to attenuating, (C) tall- tall healthy all attenuating petioles, or (D) impacted - small to moderate plants, tough spindly petioles, curled laminas. All these stages are directly linked to individual plant age and water quality (Center *et al.* 1999). If any of the characteristics changed within the extent of the macrophyte population, an additional GPS point was taken and changes in characteristics were noted. GPS points were assimilated and organised in ArcMap 9.3.1 (Figure 2-9).

### 2.3.5 Formation of macrophyte polygons and analysis of the accuracy of the spatial analysis

Attribute field data were captured and correlated with each GPS co-ordinate. Following data capture, a GPS point shapefile was generated (Figure 2-9 (a)). The GPS used (Garmin e-Trex), has a spatial inaccuracy of up to 15m in X;Y planes. Each point was snapped (snapping is a GIS term where a feature within a prescribed minimum distance is relocated to coincide exactly with the coordinates of another feature (ESRI, Redlands, California)) to its relevant river bank of the river bank polygon file (Figure 2-9 (b)), using the Hawths Analysis Tools 3.27 in ArcMap 9.3.1. This process limited GPS and field data capture inaccuracies by consistently having points located on river banks instead. It was also assumed that all beds of macrophytes extended up to the river bank.

The first set of points analysed were those where macrophyte beds observed during the field survey had been noted to have reoccurred in exactly the same position as previously mapped. These data formed the basis of the final macrophyte polygon shapefile. For any other extensive macrophyte beds, GPS points were snapped from the recorded position in the river to its relevant bank, and allowed for the usage of the river sample extraction tool from Hawths Analysis Toolset (Figure 2-9 (b)). The original Vaal River bank polygon shapefile was converted to a polyline (polyline is a form of GIS shapefile and consists of a continuous line consisting of one or more line segments (ESRI, Redlands, California)) feature within a pre shapefile and the river sample extraction tool (from the Hawths Analysis Toolset) was employed. The river sample extraction tool sequentially divided each bank polyline into segments from a designated start GPS to a designated end GPS point and the attributes of the points were aggregated and transferred to the polyline (Figure 2-9 (c)). Each polyline had to be given an average of the point score, e.g.: polyline between point A classed as rare (5 m wide), and B classed as frequent (3m wide), would be given the average score of occasional (4m wide). The transfer of point attributes to the polyline allowed for each polyline segment to be buffered (buffer is a GIS term where a polygon or polyline feature is created based on a user defined distance from another feature (ESRI, Redlands, California)), according to the newly calculated bed width which was the average width noted between the two points observed in the field (Figure 2-9 (d)).

In cases where the macrophyte community was small enough to be captured with one data point (e.g.: Eurasian water-milfoil population classed as rare, 5m in diameter), the GPS point was snapped to its relevant bank of the river bank polygon file, using the Hawths Analysis Tools 3.27 in ArcMap 9.3.1. buffers were then assigned to the river bank polygon shapefile at set distances of 0.5m, 1m, 2m, 3m, 4m, 5m, 10m and 20m, and the relevant points assigned to the relevant buffer distance, e.g.: if a GPS point indicated that the population size was 5m in diameter, it was snapped to the internal buffer of 5m. The GPS points were then buffered and attributes of the point shapefile were transferred across to the new buffer polygon areas. These areas were then unioned with the final macrophyte polygon shapefile, where the area, in hectares, was calculated using ArcMap 9.3.1.

The final analysis of the field survey data was the analysis of the accuracy of the spatial mapping exercise. The final macrophyte polygon layer was intersected (intersect is a GIS term where a new feature is created from the common area of two or more features of the same geometry type (ESRI, Redlands, California)), with the 2009 mapped macrophyte layer. This new layer, termed the 'confirmed-present' layer, shows only those areas that are common for both the 2009 layer and the final macrophyte layer. The intersect function from the ArcMap Toolbox 9.3.1. was then used to create a new polygon layer termed the confirmed-present polygon. The 'confirmed-present' polygon shapefile only maintained the regions which both the 2009 polygon and the final macrophyte polygon layer had in common and discarded any other polygon data which came from its parental shapefiles.

Similarly, the 'confirmed dead/MIA' was determined using the erase function from the ArcMap 9.3.1 Toolbox, and by overlaying the 2009 field map polygon and final map polygon shapefiles, the resulting shapefile represented polygons from the 2009 dataset that were within regions that were accessed during the field survey, but where no macrophytes were found at those locations.

To determine if the field trip had surveyed a large enough portion of the study area a 'potentially missed' polygon shapefile was created. Several reasons accounted for the inability to gain access to particular 2009 polygon patches. These include: river topography, where rapids and separate pools restricted access and  restricted launching access due to private land ownership or mining activities.

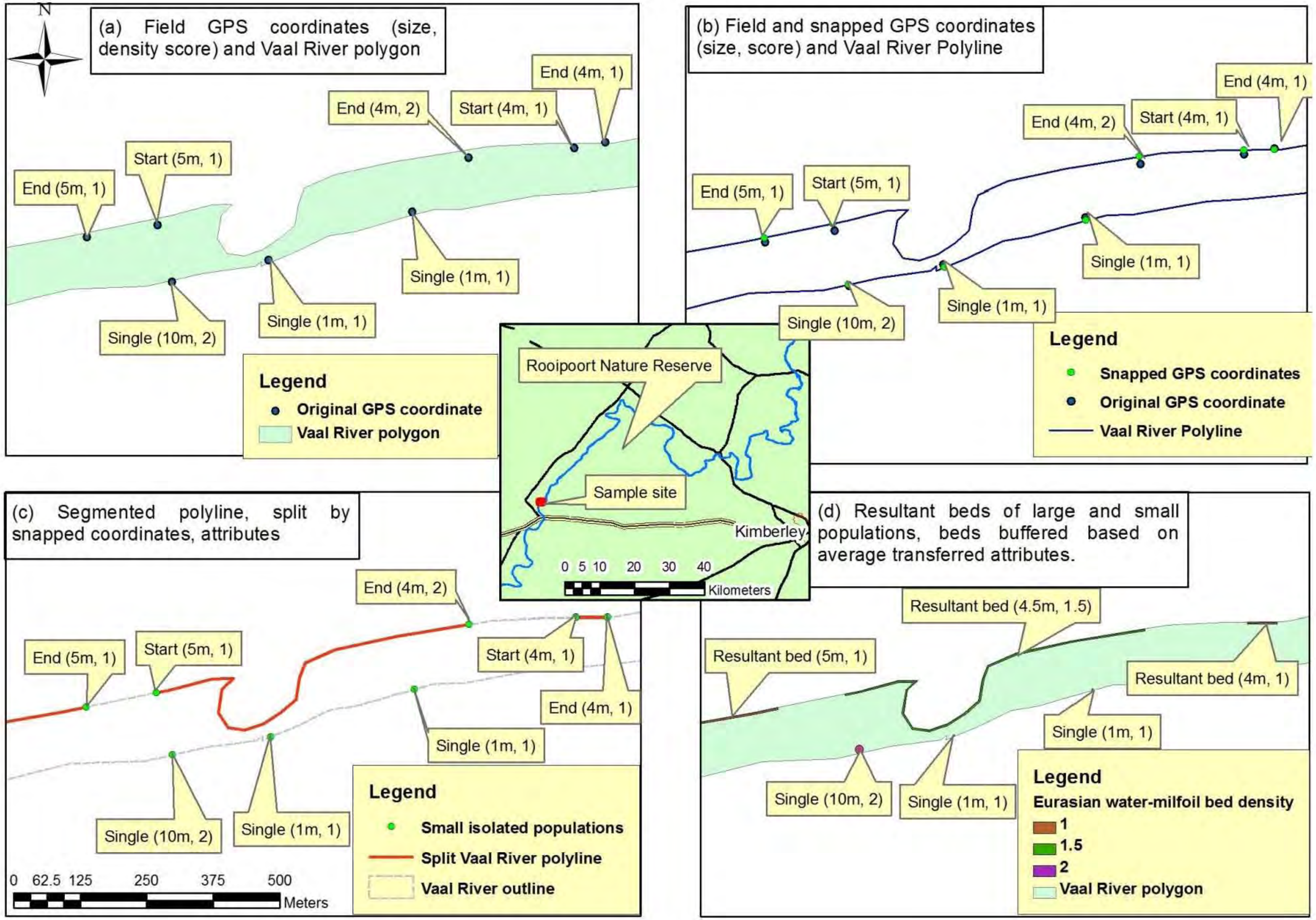



**Figure 2-9:** A selection of maps (a –d) illustrating the method used to compile the map of results from the field trip of Eurasian water-milfoil beds scored according to density.The insert illustrates the location of the site relative to the nearest city, Kimberley.

The ‘potentially missed’ polygon shapefile comprised polygons from the 2009 field maps dataset extents were determined using the erase function ArcMap 9.3.1. (Erase is a GIS term where a new polygon feature is created by the overlaying two source polygons features. the portions of the first source polygon falling outside the extent of the second source polygon are copied and used to create the new polygon feature (ESRI, Redlands, California)). The 2009 dataset polygon shapefile was overlaid on the field-surveyed layer of the Vaal River. The erase function removed all 2009 dataset polygons present within the field surveyed portions of the Vaal River polygon extent, and created a new polygon, with only the portions of the 2009 dataset that was inaccessible during the field survey.

#### 2.3.5.1 Data analysis: Determination of the dominant aquatic macrophytes

Changes in plant dominance over space and time can best be analysed using spatial analysis techniques (Yuan & Zhang 2008). Once all the GIS and fieldtrip data had been captured and corrected, the river was analysed in its entirety in terms of macrophyte dominance from the Barrage to the confluence, and all data were summarised to note overall trends of macrophyte dominance of the system. To best understand how integrated control measures implemented by WfW have impacted the distribution and abundance of dominant macrophytes of the system, the GIS data were then analysed per WMA (Figure 2-4), to determine where the macrophytes were located each year. Lastly, each WMA was divided into its respective region to note exactly which regions contained the most macrophytes and whether the extent and distribution of aquatic macrophytes had changed.

All aquatic macrophyte patches were classed as either submerged macrophyte or water hyacinth and each polygon was assigned a value according to Table 2-2, based on the year being analysed, e.g. all submerged macrophyte beds in 2006 were assigned an arbitrary numerical value to assist with the summation during the union function of the shapefiles. All resultant shapefiles were unioned in ArcGIS 9.3.1. This function combined all data to a single shapefile, which then allowed for all the values to be summed in a single column to determine the change in dominance of aquatic macrophytes. For the purposes of this study, 1 was used for submerged macrophytes and 2 was used for water hyacinth. The actual values per year did not influence the analysis, except that every subsequent year had its values increased from the previous year by a magnitude of 10, to prevent calculation confliction errors where confusion could have resulted. For example, if a water hyacinth bed occurred in the same location from 2006 until 2007, it received a value of 2 for each polygon. In 2008 submerged macrophytes dominated the region and the bed was given a value of 1. The summation would be 2+2+1 = 5, giving no additional information about when water hyacinth had dominated the patch, whereas if the Table 2-2 values had been assigned, the result would be 2+20+100 = 122, where $1^{2008}\,2^{2007}\,2^{2006}$ (superscripts are displayed simply for explanatory purposes), accurately indicate the history of succession. The change in magnitude between the years reduces conflict and keeps more complicated data intact. Numerical values, differing by factors of 10 per year, were preferred over alphabetical values as numerical values assisted in this regard.

**Table 2-2:** Values assigned to each mapped aquatic macrophyte patch.

| | Value | |
|---|---|---|
| Year | Submerged macrophyte | Water hyacinth |
| 2006 | 1 | 2 |
| 2007 | 10 | 20 |
| 2008 | 100 | 200 |
| 2009 | 1 000 | 2 000 |
| 2010 | 10 000 | 20 000 |
| 2010 GT* | 100 000 | 200 000 |

* 2010 GT is data acquired from the ground truthing field trip conducted in October 2010.

In these examples (Figure 2-10 - 13), the hashed region is an indication of the extent of macrophytes determined from that particular year. Figure 2-10 revealed that in 2006 the region was dominated by water hyacinth, with small submerged macrophyte beds present. Following the 2007 spatial analysis, the water hyacinth population had grown and the plant was highly dominant, covering large portions of the site (Figure 2-11). However when these data were unioned, it became apparent that an intermediate state of dominance class was needed for sections which, in 2006, were dominated by submerged macrophytes, but by 2007 had become dominated by water hyacinth and vice versa. This was done to compensate for the limitations of the spatial analysis such as confusion of macrophytes with inanimate objects. Without seasonal data, it was not possible to determine how long macrophyte beds had been established in a particular location. Complete dominance of a location by a macrophyte was only assigned to a polygon if a macrophyte had not been recorded as inhabiting a region since 2006 or if it had replaced its competitor for 2 consecutive years. This eliminated the possibility of incorrectly classing polygons where, for example water hyacinth had been blown over an established submerged macrophyte bed, as a water hyacinth-dominated polygon, even though beyond the detection of the satellite image there were underlying submerged aquatic plants fighting for dominance of that habitat. Hence by reverting to the example (Figure 2-11), regions such as those were reclassified as 'state of flux' polygons for 2007. Any new habitat populated in 2007 which had not been observed in 2006 was classed as 'new populations' and when macrophytes occurred in the same location, they were given a 'previously established' class, based on the history of the previous infestations. 'State of flux' and 'new population' polygons could not be produced for 2006 due to a lack of previous data, and all polygons were only classed as previously established macrophytes.

The 2008 (Figure 2-12) and 2009 (Figure 2-13) spatial analysis of this site is shown to illustrate the method. The data from 2010 and 2010 GT were also unioned, but the same classification system was used and therefore was not required for these illustration purposes, as the 2008 and 2009 images shown are sufficient to understand the method and logic. By

2008, the large water hyacinth infestation of 2007 (Figure 2-11) had disappeared and it was evident that submerged macrophytes started to proliferate. This led to the establishment of many ‘state of flux’ polygons. Certain regions also skipped the ‘state of flux’ class by re-inhabiting regions in which they had remained uninhabited for 2 years since 2006. By 2009 those submerged macrophyte patches had increased in size; however the majority of the ‘state of flux’ polygons were converted to submerged macrophytes.

### 2.3.6 Abiotic Variables

Eurasian water-milfoil and water hyacinth are able to grow under a variety of abiotic conditions (Chapter 1). However once they establish a foothold in an ecosystem, their aggressive growth rate allows them to exploit opportunities when ideal conditions present themselves. The most influential facets of water and soil chemistry that govern these growth rates are water clarity, total dissolved solids, pH, water nutrient levels, and in the case of Eurasian water-milfoil, soil nutrient and physical characteristics. The Department of Water Affairs (DWA) Resource Quality Service (RQS) data (pH, nitrate and phosphate levels) has several monitoring points (60), along the Vaal River which have recorded some of the aforementioned water chemistry variables, from 1999 to 2011. These data have been sourced from a number of laboratories over the years at different time scales and frequencies by DWA, this is a limitation to the study. All appropriate water quality attribute values from 2005 to present were summarised to correlate these variables with population levels mapped in this study. Scatterplots and Pearson’s correlation tests were conducted between DWA RQS abiotic data (pH levels, nitrate and phosphate concentrations) and the percentage cover of the macrophytes from 2006 until the ground-truthing field trip in 2010. Variation of the cover of macrophytes, for both submerged macrophytes and water hyacinth, and physio-chemical data (pH levels, nitrate and phosphate concentrations) variables across the regions were considered normal.


**Figure 2-10:** The spatial analysis of a SPOT V 2006 image, illustrating the location of floating and submerged macrophytes on Warrenton Weir, WMA 10.4.

Scores of the plants are shown in the legend. The insert illustrates the location of the site relative to the nearest town, Christiana.

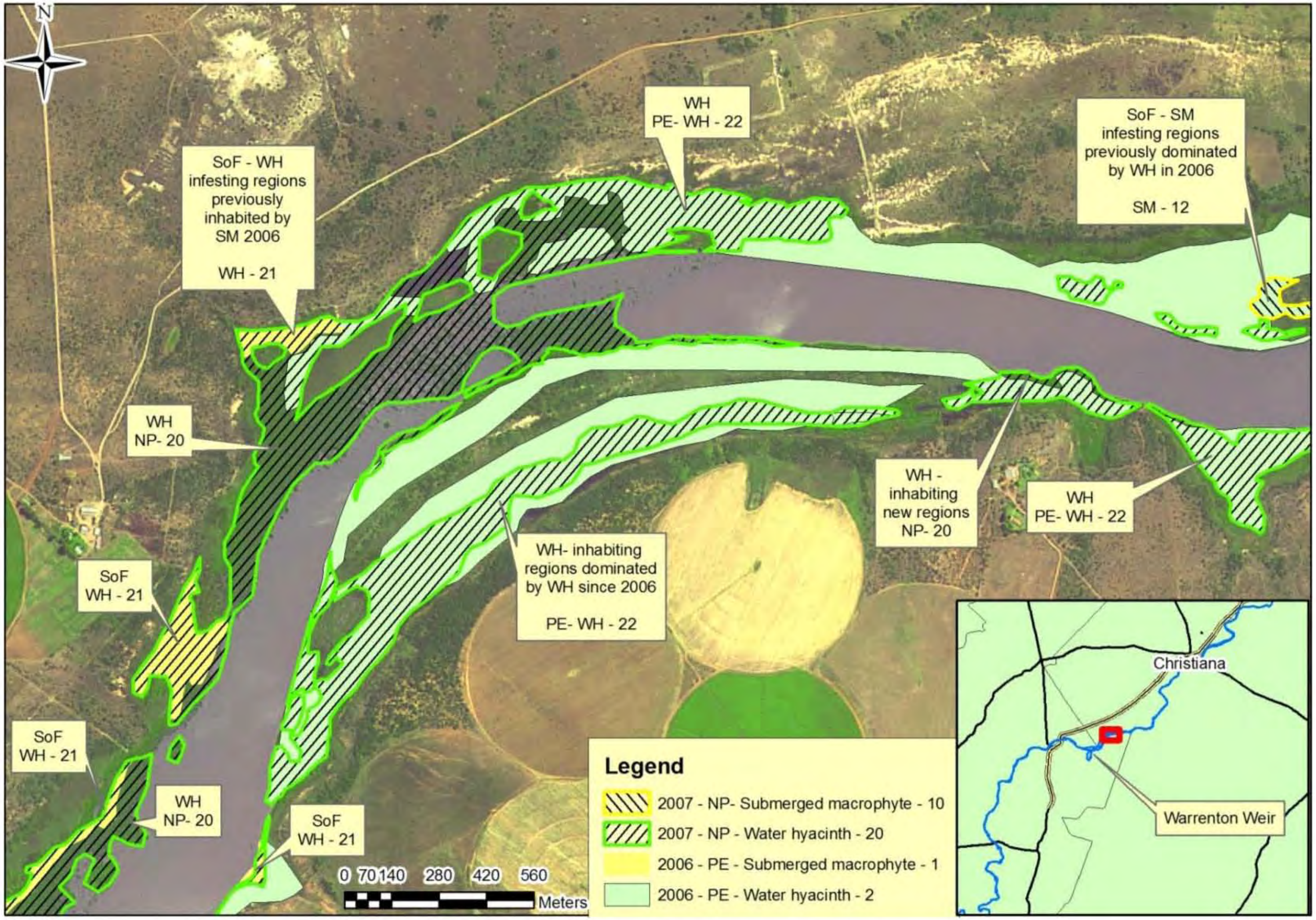



**Figure 2-11:** The spatial analysis of a SPOT V 2007 image, illustrating the location of floating and submerged macrophytes on Warrenton Weir, WMA 10.4. The 2007 data were layered above the 2006 data and the union results and scores are shown. SM: submerged macrophyte, WH: water hyacinth, SoF: ‘state of flux’, PE: ‘previously established’, NP: ‘new population’. The insert illustrates the location of the site relative to the nearest town, Christiana.

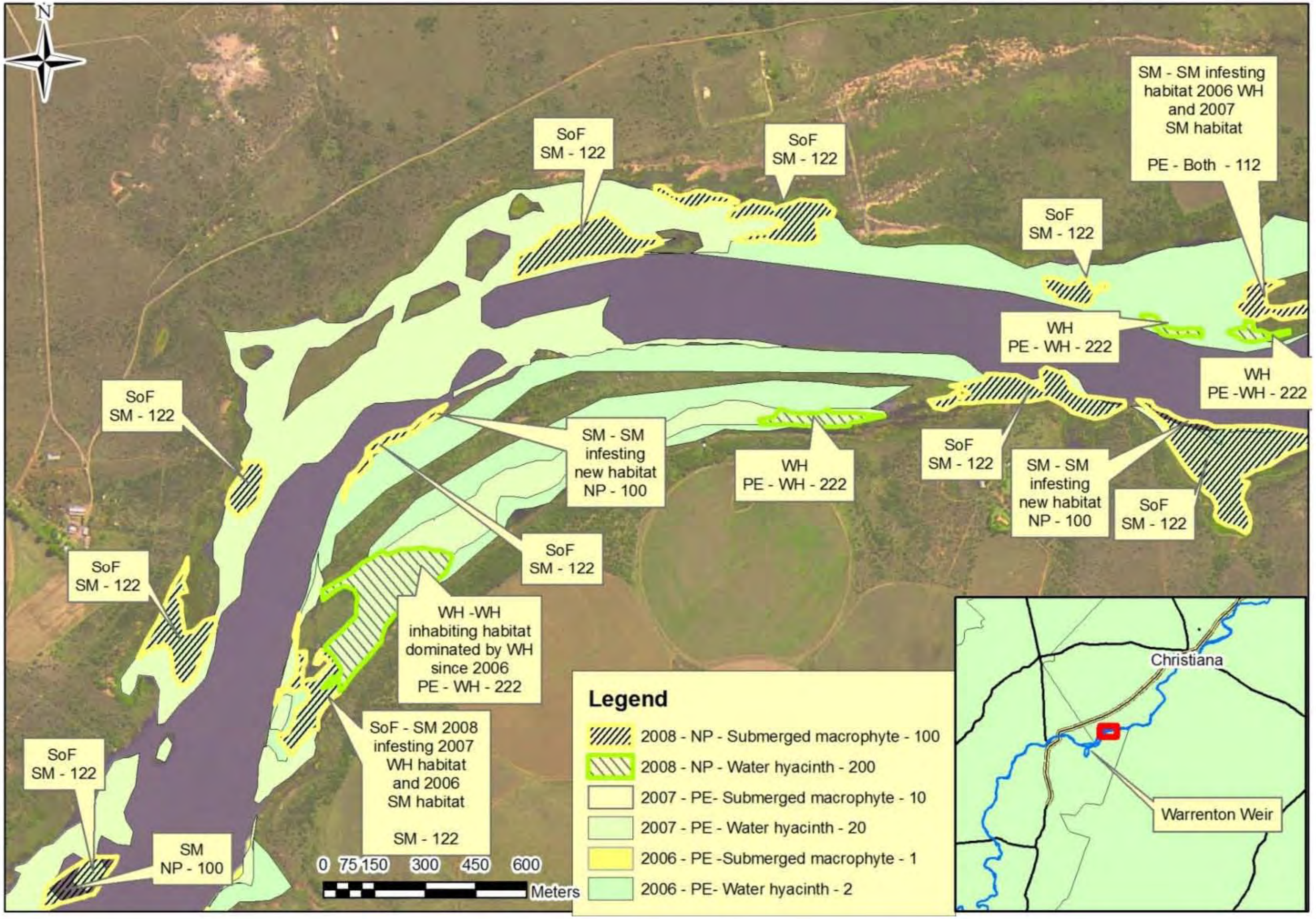



**Figure 2-12:** The spatial analysis of a SPOT V 2008 image, illustrating the location of floating and submerged macrophytes on Warrenton Weir, WMA 10.4. The 2008 data were layered above the 2006 and 2007 data, the union results and scores are shown, SM: submerged macrophyte, WH: water hyacinth, SoF: 'state of flux', PE: 'previously established', NP: 'new population'. The insert illustrates the location of the site relative to the nearest town, Christiana.

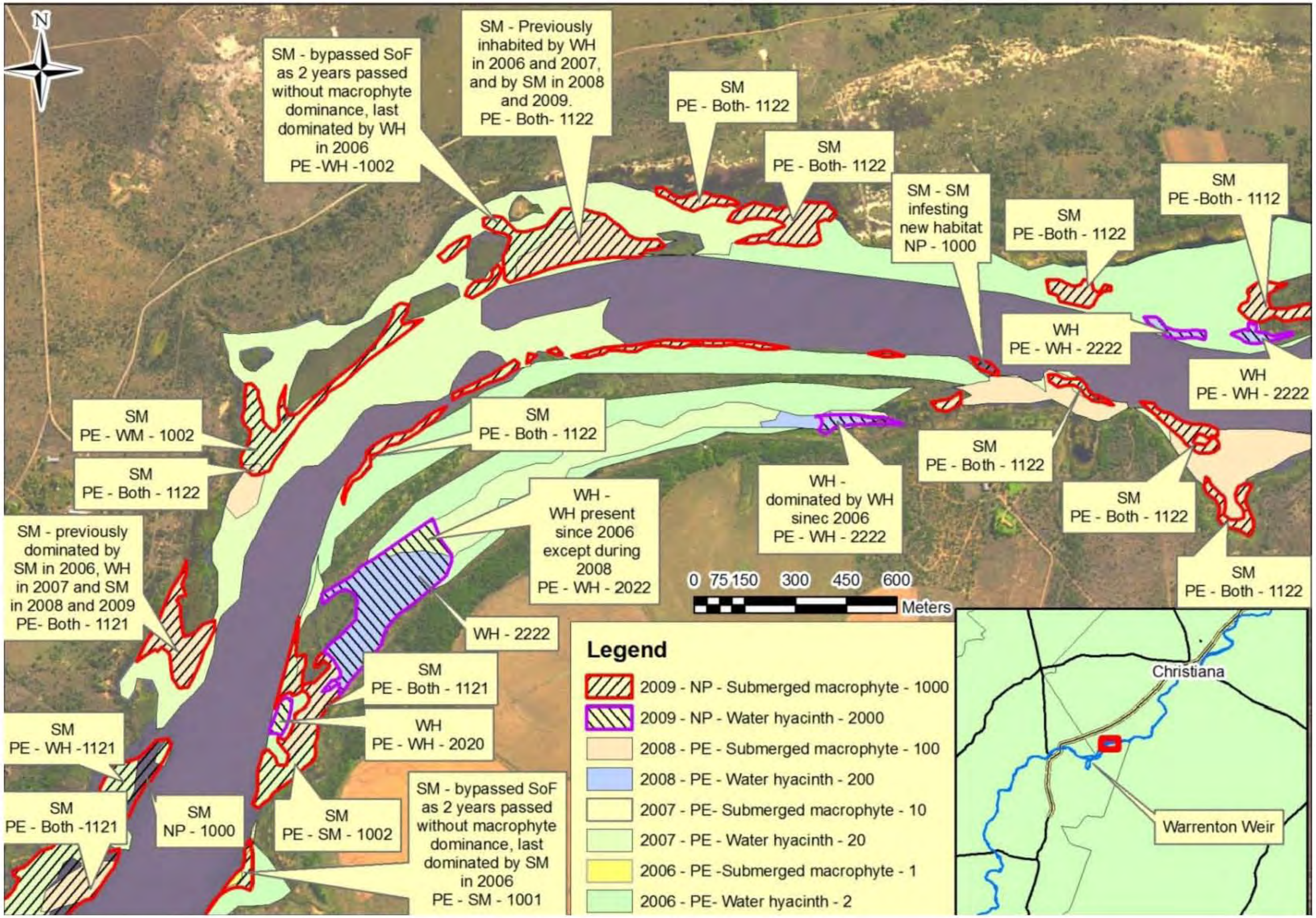



**Figure 2-13:** The spatial analysis of a SPOT V 2009 image, illustrating the location of floating and submerged macrophytes on Warrenton Weir, WMA 10.4. The 2009 data were layered above the 2006 and 2007 data and the union results and scores are shown, SM: submerged macrophyte, WH: water hyacinth, SoF: 'state of flux', PE: 'previously established', NP: 'new population'. The insert illustrates the location of the site relative to the nearest town, Christiana.

These data allowed for historical correlations between macrophyte cover and changes in abiotic variables, with the limitation that these monitoring points did not occur within each WMA region. Data collected during the field trip of October 2010 and from the analysis of samples collected were analysed and correlated with macrophyte cover. As different instruments and methods were used by DWA RQS, and during the field survey, these data were not combined and were analysed separately.

The DWA RQS data were also statistically correlated with field trip data which recorded pH and total dissolved solutes (TDS) using a Hanna multiparameter meter Model 929828, both within the macrophyte communities and in open water. Water phosphate and nitrate nutrient levels were determined using a Hanna HI 8302 Aquaculture Photometer 2008 Series. Nitrate and phosphate concentrations were determined using the standard Hanna Phosphate HR and Nitrate classification techniques (Hanna HI 8302 user manual).

Soil samples (circa 1kg) were collected from inside the extent of the communities. Nutrient analysis was conducted by BemLabs in Strand, Western Cape, which uses standardised tests accredited by the South African National Accreditation System (SANAS). Parameters analysed included pH in a 1M solution of KCl at a 1:4 ratio of soil to solution, resistance in a standard paste using a standard cup; Bray II extractable P, exchangeable cations, Ca, Mg, Na and K extracted with Ammonium acetate at pH7, trace elements Cu, Zn, Mn, B extracted with 1M HCl: H using the standard Eksteen method, except where the soil pH rose above 7 and the Olsen method was used; stone (>2mm) was separated out by dry sieving and organic carbon was determined using the Walkley-Black standard method. All these data were statistically correlated with the density of the macrophyte bed.

Analyses on all of the field trip (soils and water) non-normally distributed data were performed using the ANOVA Kruskal-Wallis by Ranks and Median Test (a non-parametric

ANOVA). The Kruskal-Wallis test ranks data, thereby reducing variation. It is considered a strong test as it also uses all the data points rather than generating means. The H value is used as an indication of the data variability and even visually similar data on graphs can be significantly different between the overall dataset (Rosner 2000).

## 2.4 *Results*

The most important destructive stochastic event with the ability to reset an aquatic ecosystem is flooding. Flood data were analysed from the DWA RQS division's dataset. According to the DWA Pilgrims Estate monitoring point (all other monitoring points had the same flooding trend), the Vaal River flooded significantly in 2000, 2006 and 2010, with minor peaks in flow rate occurring in 2001 and 2009 (Figure 2-14). These events have a direct impact on the snapshot satellite imagery taken after the flooding events.

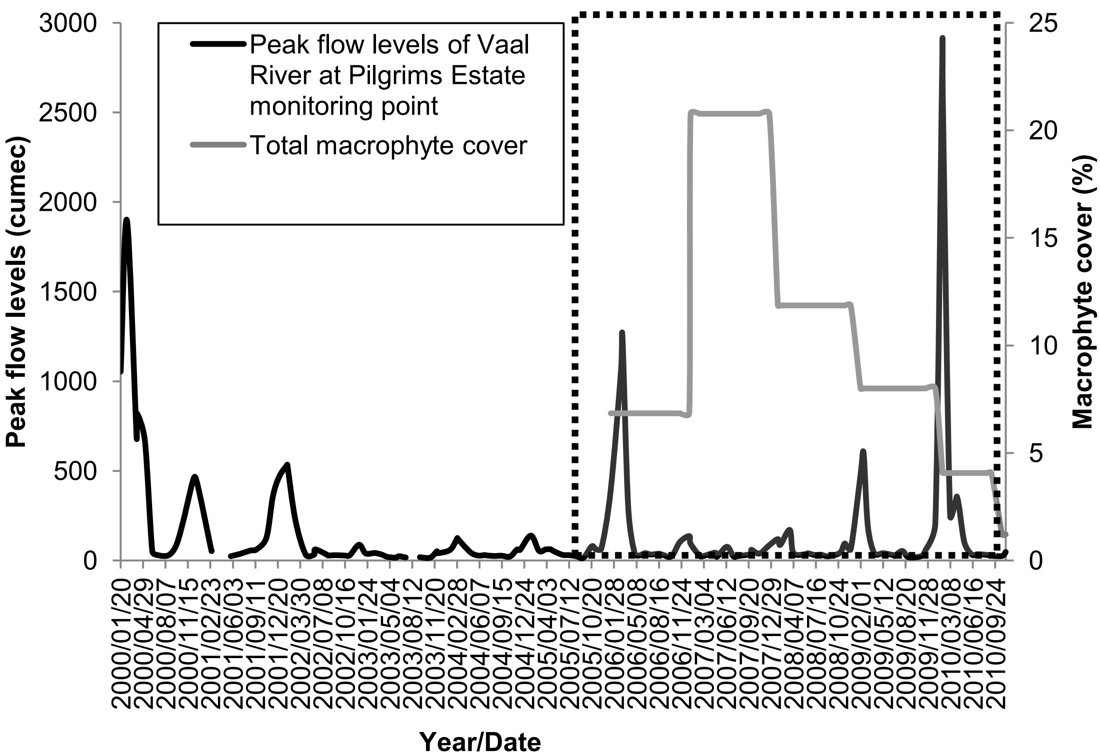



**Figure 2-14:** The peak flow levels of the Vaal River at the Pilgrims estate monitoring point (DWA RQS division, January 2011). The boxed region of the graph indicates the time during which SPOT V images were taken and GIS data analysed in this study.

The main flooding events took place during February 2006 and February 2010, but the 2010 images were visually obscured by flooding (Figure 2-14). A peak in overall macrophyte cover was noted in 2007 following the floods in 2006, however a steady decline in overall cover was noted since. The lack of seasonal satellite imagery data meant that blended satellite images were used for mapping and thus macrophyte community size was effectively averaged over an entire year (Figure 2-14). Blended SPOT V imagery are a combination of the clearest highest quality sections from the different SPOT V images, taken within the same year, which are merged together. This is done to obtain the most comprehensive/complete SPOT V image of the region. Sections of the SPOT V datasets could therefore have been recorded before flooding events and others could have been recorded after flooding events. This would be of particular interest for the 2006, 2009 and 2010 data, which were influenced by higher peak flow rates (Figure 2-14).

The other large stochastic event that occurred on the Vaal River was in the form of glyphosate application against water hyacinth populations. WMA 8, 9 and 10 has had sporadic spraying of water hyacinth since 2006, all regions coordinated together and sprayed in 2008, following peaks in water hyacinth infestations in 2007. The least glyphosate was sprayed in WMA 8, and spraying since 2008 was in small quantities and was sporadic. WMA 9 is a zero tolerance zone for water hyacinth and hence constant maintenance and regular winter spraying has taken effect there since 2006. WMA 10 had a major spraying event in 2008, following which only small sporadic spraying has taken place.

### 2.4.1 Analysis of spatial data

Digitally mapped spatial data from the SPOT V imagery, from 2006 -2010, provided the base dataset for spatial analysis. To complete the spatial analysis, however, a field trip was required to ground truth the 2010 digitally mapped spatial data.

#### 2.4.1.1 Field trip

The visual mapping from the SPOT V imagery was unable to discern between submerged macrophyte species and consequently all species of submerged macrophytes were grouped as submerged macrophytes, while water hyacinth is distinctive enough to be delineated separately.

With the rapid sampling technique, 66.6% (19 714ha) of the total study area (29 610.35ha) was sampled. Of the remaining 33.4% (9 896.ha), 69% (8 098ha) was located on the open water of Bloemhof Dam (19 510.67ha) constituted Bloemhof Dam (Table 2-3). Three different species of submerged macrophyte were present in the Vaal River: Eurasian water-milfoil, sago pondweed *Stuckennia pectinata* (L.) Böemer (=*Potamogeton pectinatus* L.) (Potamogetonaceae) and curly leaf pondweed *Potamogeton crispus* L. (Potamogetonaceae) (Table 2-3). Water hyacinth was the only floating macrophyte found in abundant populations (Table 2-3). In total, 95.42% of all the submerged macrophyte populations surveyed during the field trip were previously mapped using GIS, while 97.42% of the water hyacinth population was located within regions that the GIS analysis had classified as water hyacinth habitat (Table 2-3). As a result of the field trip, it was apparent that the largest portion of the submerged macrophyte population was Eurasian water-milfoil, and water hyacinth covered a fraction of the Vaal River.

**Table 2-3:** An analysis of the ground-truthing survey conducted during October 2010, which confirmed the results of the GIS analysis.

| Confirmed present | Bed sizes (ha) | Accuracy Assessment |
|---|---|---|
| Eurasian water-milfoil | 259.16 | 95.42% |
| Curly leaf pondweed | 0.01 | |
| Sago pondweed | 108.94 | |
| Water hyacinth | 13.14 | 97.42% |
| **Grand Total** | **381.25** | |
| **Potentially missed** | **498.55** | |
| Submerged macrophytes | 239.42 | |
| Water hyacinth | 259.13 | |
| **Confirmed dead/missing** | **1022.4** | |
| Submerged | 420.38 | |
| Water hyacinth | 602.02 | |

### 2.4.1.2 Eurasian water-milfoil field trip results

Determination of the biomass of Eurasian water-milfoil, the submerged macrophyte dominating the largest portions of the Vaal River, was carried out at 19 locations along the river. This served to provide a baseline account of the current abundance and to allow for future comparison should Eurasian water-milfoil undergo any form of control. The formula used by Kenow *et al.* (2006) was used to determine the amount of dry mass (DM) of Eurasian water-milfoil per square meter. This was statistically correlated with the point score results (Table 2-4). A significant correlation ($r=0.80$, $p<0.05$) was found between the different bed density classes (dense, abundant, frequent, occasional and rare) and the calculated dry biomass. This allowed for calculation of the mean densities biomass scores that were not ground-truthed in the field (these results were depicted by the calculated quick point score values shown in Table 2-4). Although the majority (42%) of Eurasian water-milfoil beds surveyed were of a density of 2.5 (an average between frequent and abundant scores), 31.88% of the Eurasian water-milfoil surveyed had a density of 4.5, the average between dense and abundant scores. During the field trip, 178.45t DM of Eurasian water-milfoil was surveyed.

**Table 2-4:** Determination of the density of Eurasian water-milfoil patches growing in the Vaal River (SE indicates standard error of the mean)

| Quick Point score | Calculated (gDM.m$^{-2}$) | ± SE | Total Area (%) | TOTAL (t) |
|---|---|---|---|---|
| Dense - 5 | 156.09 | ± 0.78 | 0.06 | 0.24 |
| 4.5 - Calculated | 106.08 | | 31.88 | 87.64 |
| Abundant - 4 | 80.41 | ± 0.53 | 0.06 | 0.12 |
| 3.5 - Calculated | 82.51 | | 7.08 | 15.15 |
| Frequent - 3 | 51.14 | ± 0.43 | 0.22 | 0.29 |
| 2.5 - Calculated | 58.94 | | 42.14 | 64.36 |
| Occasional - 2 | 13.25 | ± 0.61 | 0.26 | 0.09 |
| 1.5 - Calculated | 35.36 | | 7.79 | 7.14 |
| Rare - 1 | 14.55 | ± 0.40 | 2.86 | 1.08 |
| 0.5 - Calculated | 11.79 | | 7.65 | 2.34 |
| | | | **TOTAL (in Vaal River)** | **178.45 t** |

### 2.4.1.3 Water hyacinth field trip results

The 'other dominant' macrophyte surveyed in the Vaal River was water hyacinth. The general absence of water hyacinth on the system constrained sampling to five sites and on average, the plants were defined as small and healthy with inflated petioles. The water hyacinth plants had 4.92 (±0.23 SE) leaves and 1.8 (±0.22 SE) ramets per plant, while the mean longest petiole length was 9.11cm (±0.58cm SE), with a mean leaf two length of 6.89cm (±0.33cm SE). The water hyacinth plants also had a mean maximum root length of 14.1cm (±1.23cm SE), while no flowers were noted. Extensive damage by the weevil biocontrol agents, *Neochetina eichhorniae* and *Neochetina bruchi,* was noted throughout the study area, while damage by the mite *Orthogalumna terebrantis* was only noted at two sites in WMA 8, even though it had not been recorded as being officially released against water hyacinth on the Vaal River. No evidence indicating the presence of other biological control agents was found during the survey. These data were all added to the 2010 shapefile of the Vaal River and assisted in the analysis of the spatial changes of the system.

#### 2.4.1.4 Spatial analysis of the entire study site

The mapped results for the entire study site show that both water hyacinth and submerged macrophytes had similar covers of 2.9% and 4% respectively in 2006 (Figure 2-15). This changed radically in 2007 as a result of the flood in 2006 (Figure 2-14) as there was a dramatic increase in water hyacinth cover in 2007 (17.4%), and only a slight decrease in submerged macrophyte cover (4.8%). As a result of the 2006 flooding event (Figure 2-14), water hyacinth took grew rapidly between 2006 and 2007, illustrated by the fact that of the 17.4%, 15.2% was classified as new growth, while only 2.2% remained in the same locations as in 2006 (Figure 2-16). The submerged macrophyte communities were more stable, with 3.75% of the 4.8% of the 2006 growth still present in 2007, while it only increased in abundance by an additional 1.1%, which was classed as new growth in 2007 (Figure 2-16). State of flux communities (habitat that was in the process of changing dominance between the macrophytes) in 2007 only covered 0.09% of the river in 2007 (Figure 2-16).

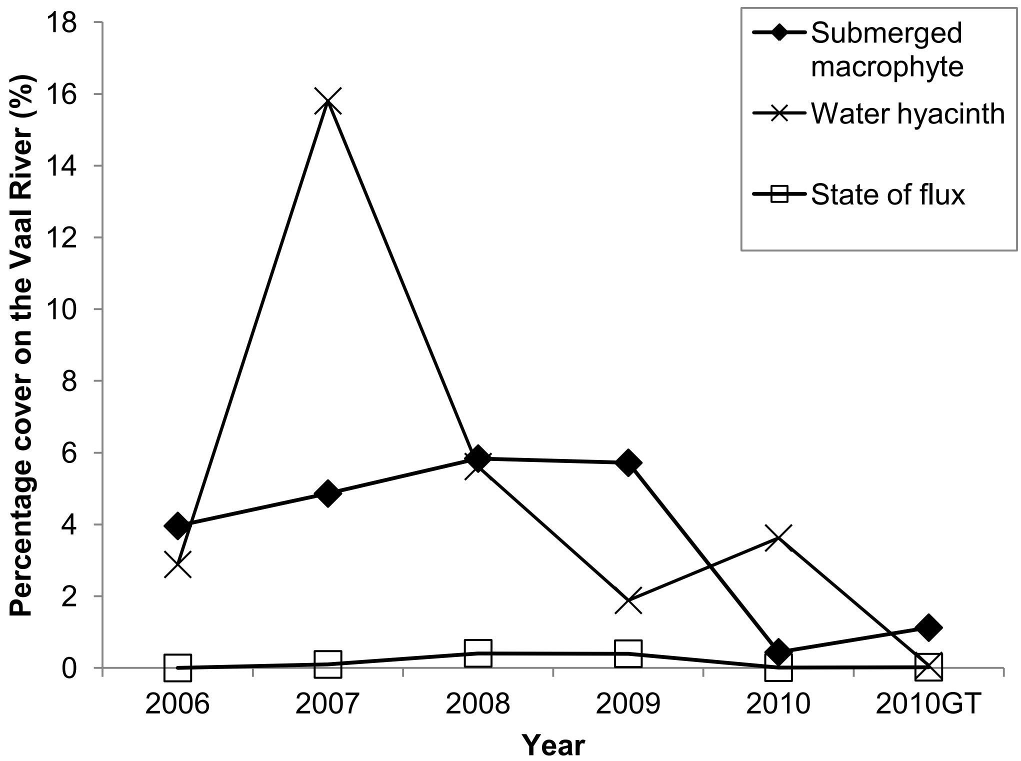



**Figure 2-15:** The overall cover of water hyacinth and submerged macrophytes from 2006 until the gound truthing field trip in October 2010 (2010GT).

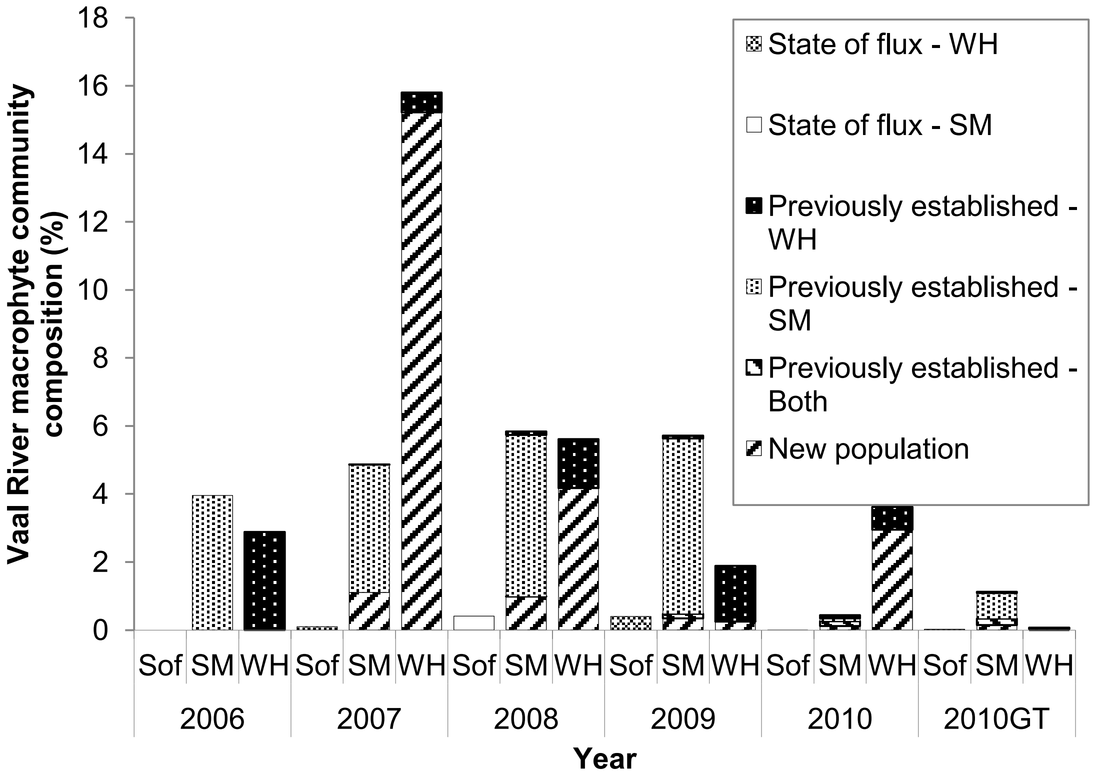



**Figure 2-16:** The cover of macrophyte communities along the length of the study area of the Vaal River, illustrating the breakdown of water hyacinth (WH), submerged macrophyte (SM) and state of flux communities (Sof) in terms of the different classes, illustrated in the legend, from 2006 until the ground truthing field trip in October 2010 (2010GT),

By 2008, the percentage cover of water hyacinth declined to 5.6% (Figure 2-15), while the concurrent cover of submerged macrophytes increased to 5.8%, and communities in a state of flux also increased to 0.4%. A breakdown of these communities showed that in 2008, 4.17% of the 5.8% water hyacinth communities were new populations and 1.13% of the communities occurred in locations where the plant had grown previously (Figure 2-16). Submerged macrophyte communities continued to inhabit previously dominated commmunities (4.7% of the 5.8%), while the remainder of the 5.8% was classed as new population (1%). Communities that were in a state of flux increased to 0.4% of the Vaal River

(Figure 2-16), of which the majority were submerged macrophyte communities (0.4%) while no communities were water hyacinth-dominated.

In 2009, water hyacinth continued to decrease in cover to 1.8%, while the submerged macrophyte and state of flux community cover remained at 5.7% and 0.39% (Figure 2-16). Of the water hyacinth community 0.24% was located in the same habitat as previously established water hyacinth populations, while negligible water hyacinth populations in 2009 were deemed new populations (0.0002%) (Figure 2-16). Communities classed as being in a state of flux had reduced in size, of which 0.38% were water hyacinth-dominated communities, and 0.01% submerged macrophyte-dominated.

In 2010, another flooding took place (Figure 2-14), which resulted in an increase in water hyacinth cover to 5.7%, and a decrease of submerged macrophyte cover to 0.43% (due to system scouring), while communities in a state of flux only occupied 0.007% of the river. Analysis of macrophyte community composition showed that the majority of the water hyacinth consisted of 2.9% new population and 0.7% consisted of previously established water hyacinth populations (Figure 2-16). The submerged macrophyte populations were reduced and the majority of the population consisted of previously established communitites 0.014%), although 0.01% was classed as new populations.

By 2010 GT, there was already a noteworthy increase in submerged macrophyte cover (up to 1.1%) and a concurrent decrease of water hyacinth cover (down to 0.06%), while only 0.02% of the river's communities were in a state of flux. Figure 2-16 shows the community composition of the submerged macrophyte communities, which indicates that submerged macrophyte communities were occupying a variety of different habitats which were previously established by both water hyacinth and submerged macrophyte communities (0.19%). However the majority of the submerged macrophyte community was classed as new populations as it dominated new habitat which previously had no recorded macrophyte present (0.74%).

### 2.4.1.5 Spatial analysis of Vaal River WMA regions

An average cover of water hyacinth, submerged macrophytes and state of flux communities was determined per WMA along the Vaal River. WMA 8 starts on the Vaal River system, below the Barrage near Parys down to a bridge above Orkney. WMA 9 extendsfrom this bridge to just below Bloemhof Dam, and WMA 10 starts from below Bloemhof Dam and extends down to the confluence with the Orange River (Figure 2-4).

From 2006 to 2010 GT, the majority of the submerged macrophyte community was found in WMA 10 (Figure 2-17), while water hyacinth dominated WMA 9, peaking in 2007 in all areas, but from 2008 until 2010GT, it could be found almost exclusively in WMA 9. The interesting interaction occurred in 2008 in WMA 10, when water hyacinth covered 0.8% of the entire Vaal River, but was subsequently reduced to 0.02% cover by 2008, while submerged macrophyte cover increased at the same time by 3.5% to 5.1%. Following this decline, water hyacinth remained at low levels of cover through 2009 (0.01%), and 2010 (0.02%), until it began to recover by 2010GT (0.05%). The state of flux communities also showed a tendency to occur in the water hyacinth-dominated WMA 9 region. To better understand why there were such drastic changes in macrophyte cover, in particular from 2007 to 2008, physio-chemical conditions of the sediment and water were investigated.

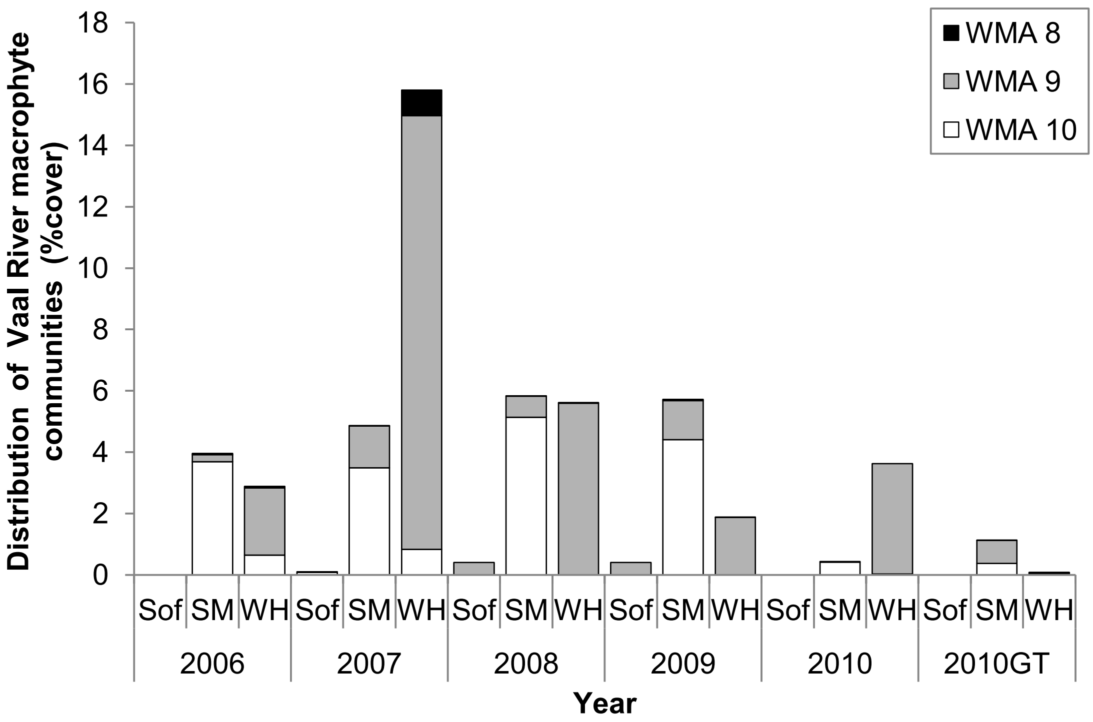



**Figure 2-17:**The overall cover of macrophyte communities in the Vaal River per WMA, illustrating the locations of water hyacinth (WH), submerged macrophyte (SM) and state of flux (Sof) communities, from 2006 until the gound truthing field trip in October 2010 (2010GT) using SPOT V imagery.

#### 2.4.1.6 Correlation of macrophyte growth to the physio-chemical conditions of the Vaal River

Eurasian water-milfoil and water hyacinth are able to respond to their surrounding abiotic environment to best suit their own population. This response is difficult to quantify in a fluid riverine system, but it was possible to determine whether the macrophyte populations correlated with the abiotic conditions of the river in the form of new population growth. To best quantify the impact of these effects on population levels, a variety of abiotic attributes, water pH, nitrate and phosphate levels, recorded by DWA RQS (since 2005) were statistically correlated with the changes in macrophyte populations along the Vaal River.

The mean pH values for WMA 8, 9 and 10 varied between 7.8 (WMA 10 in 2007) and 8.4 (WMA 8 in 2010) (Figure 2-18). WMA 8 and 9 pH values remained similar except in 2008, while WMA 10 varied, especially after the flooding in 2006 (Figure 2-18). The region's pH levels were lower (in 2007) than WMA 8 and 9. This was followed by a change for WMA 10 in 2008, as it increased from 7.9 (2007) to 8.3, (2008) and then stabilised until 2009. These results revealed no significant correlation between water hyacinth macrophyte cover and pH (r =-0.006, $P$ = 0.80), while there was a significant positive correlation between submerged macrophytes and pH, although it was weak (r = 0.14, $P$ <0.05). This correlation corresponds especially well with the WMA 10 submerged macrophyte population, which did increase in cover between 2007 and 2008, while both pH and submerged macrophyte cover stabilised in 2009.

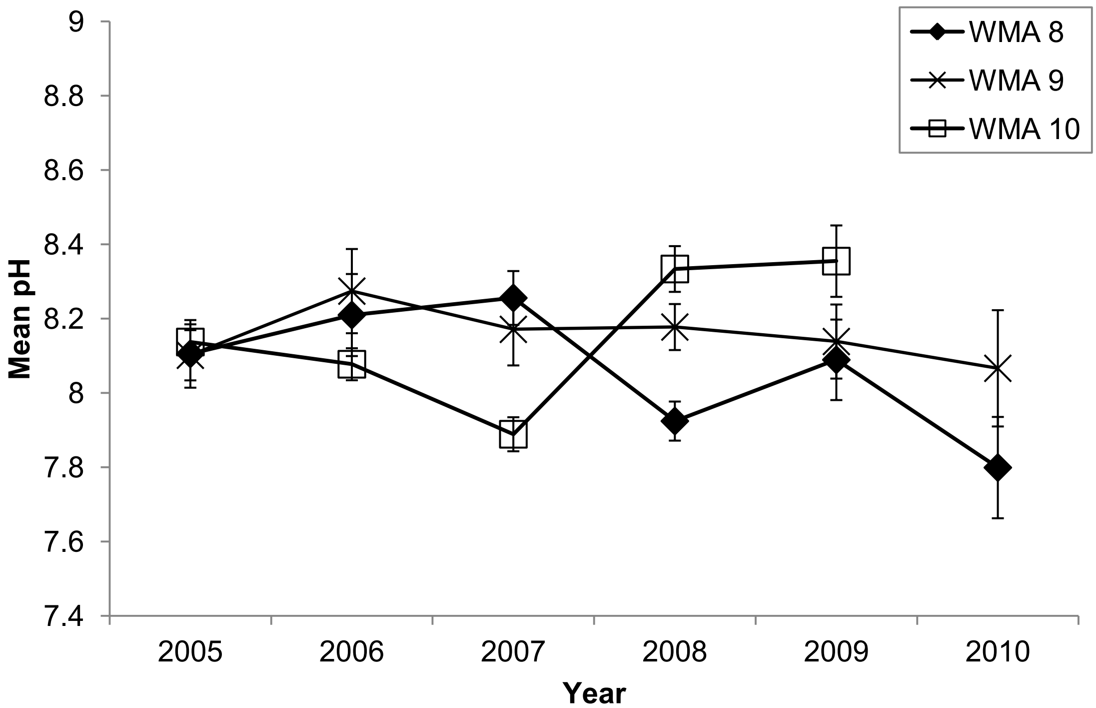



**Figure 2-18:** The mean pH per WMA region throughout the Vaal River from 2005 until 2010. Error bars represent standard error of the mean. Data obtained from Department of Water Affairs Resource Quality Services (no data for WMA 10 in 2010).

The flooding did not have an impact only on the various pH levels but also influenced the nitrate levels of the Vaal River. The highest nitrate concentrations occurred in WMA 8 and 9 (Figure 2-19); both showed peaks in nitrate concentrations in 2006 after flooding. The nitrate values of WMA 8 and 9 then decreased progressively, and by 2009 they were at 0.007mg/l and 0.017mg/l respectively. Concurrently, WMA 10 nitrate levels were substantially lower than the other WMA regions, never reaching more than 0.04mg/l. This could be attributed to the extensive submerged macrophyte or lack of water hyacinth dominance of this WMA that is shown throughout (Figure 2-19). Small peaks in nitrate levels within all of the regions could also be noted following the flooding in 2010. The observed comparisons were confirmed by the significant negative correlation found between both water hyacinth and submerged macrophyte cover and nitrate levels (water hyacinth, r= -0.20, $P < 0.05$ and submerged macrophytes, r = -0.22, $P < 0.05$). Drops in nitrate concentration levels can therefore be attributed to water hyacinth using the nutrients for growth, removing them from the system.

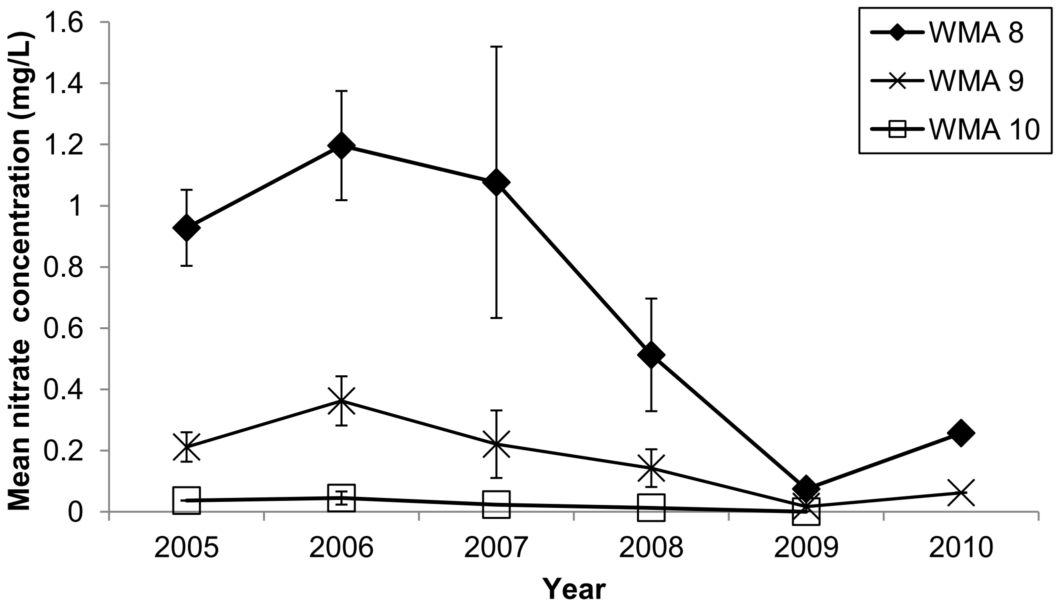



**Figure 2-19:** The mean nitrate concentration levels per WMA region throughout the Vaal River from 2005 until 2010. Error bars represent standard error of the mean. Data obtained from Department of Water Affairs Resource Quality Services (no data for WMA 10 in 2010).

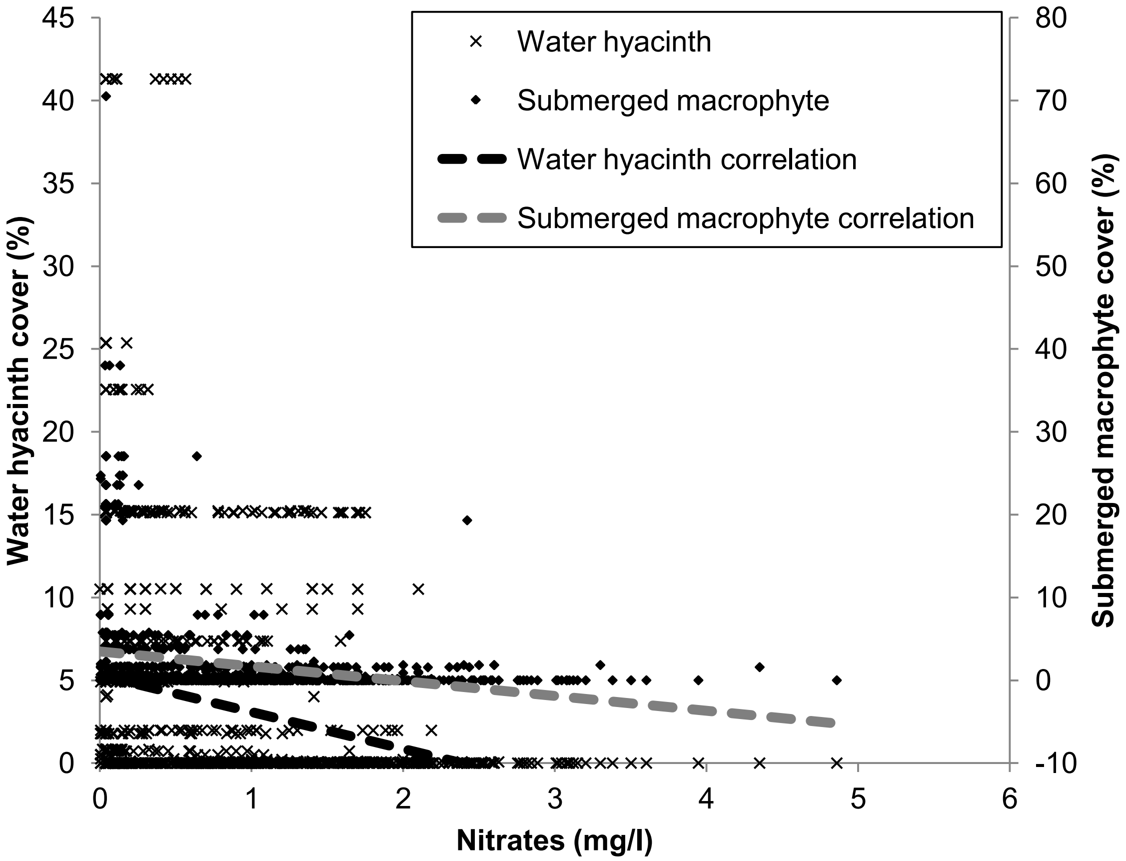



**Figure 2-20:** The correlation of WMA macrophyte cover and nitrate concentration. Data obtained from Department of Water Affairs Resource Quality Services (no data for WMA 10 in 2010).

The highest phosphate concentrations occurred in WMA 8 (Figure 2-21), which was greater than those from WMA 9 and 10. Phosphate concentrations peaked in 2007 in WMA 9 and continued to decline in 2008, before peaking again in 2009 (0.3mg/L) and declining in 2010. This corresponds well with water hyacinth communities. The peak in water hyacinth cover in 2007 (especially reflected by the high proportion of the new population growth class) resulted in the uptake of large quantities of phosphates from the water column, which effectively reduced phosphate concentrations in 2008. The water hyacinth population then dropped in 2009, which resulted in phosphate concentration levels increasing. After the flood in 2010, there was a small peak in water hyacinth cover, from the influx of nitrates and this in turn aided the reduction in phosphate levels in 2010. WMA 10 data showed a peak in phosphate concentration in 2006, and thereafter a constant decline in concentration until 2009 (Figure 2-21). There was a significant correlation but a weak negative relationship between submerged macrophyte levels (r = -0.11, $P$ <0.05) and phosphate concentration levels, while water hyacinth showed no correlation (r = -0.015, $P$ = 0.53) (Figure 2-21).

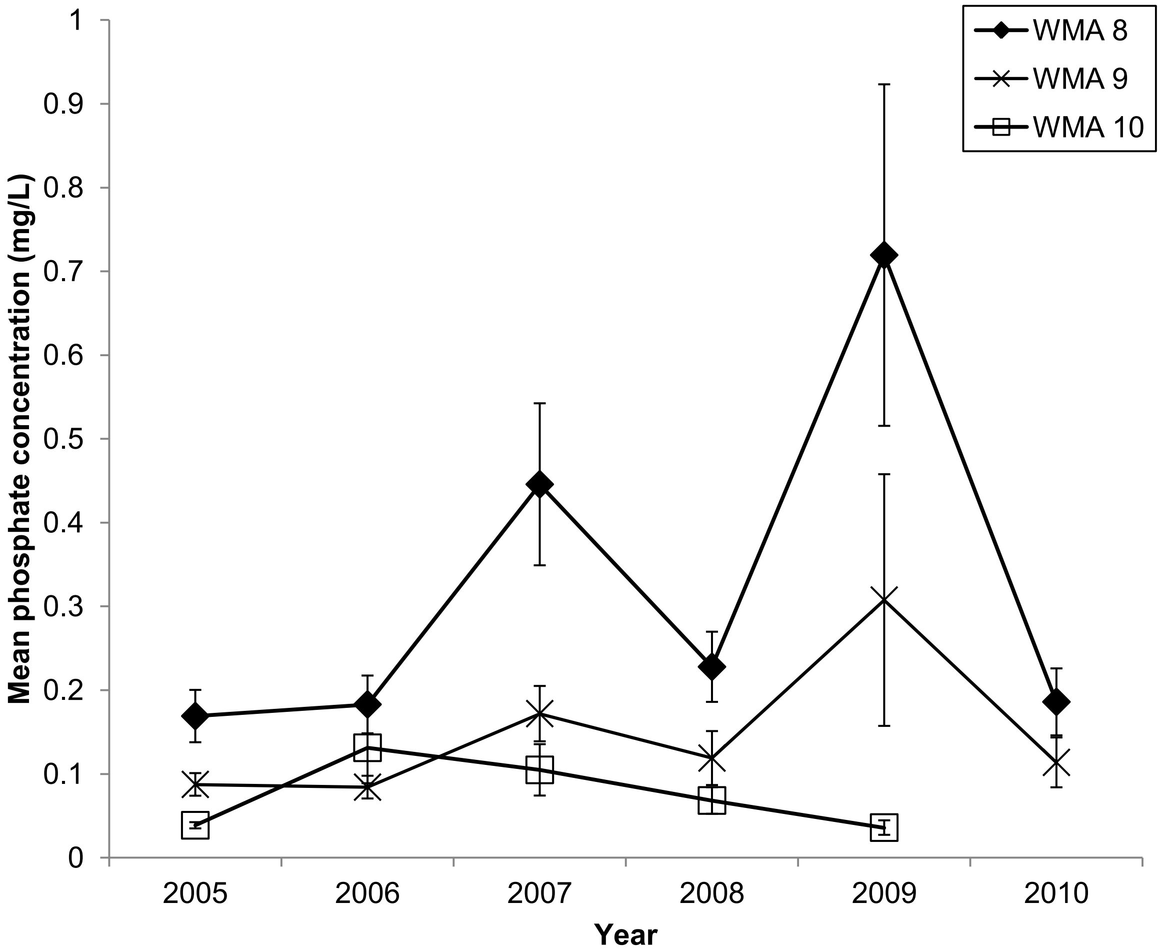



**Figure 2-21:** The mean phosphate concentration levels per WMA region throughout the Vaal River from 2005 until 2010. Error bars represent standard error of the mean. Data obtained from Department of Water Affairs Resource Quality Services (no data for WMA 10 in 2010).

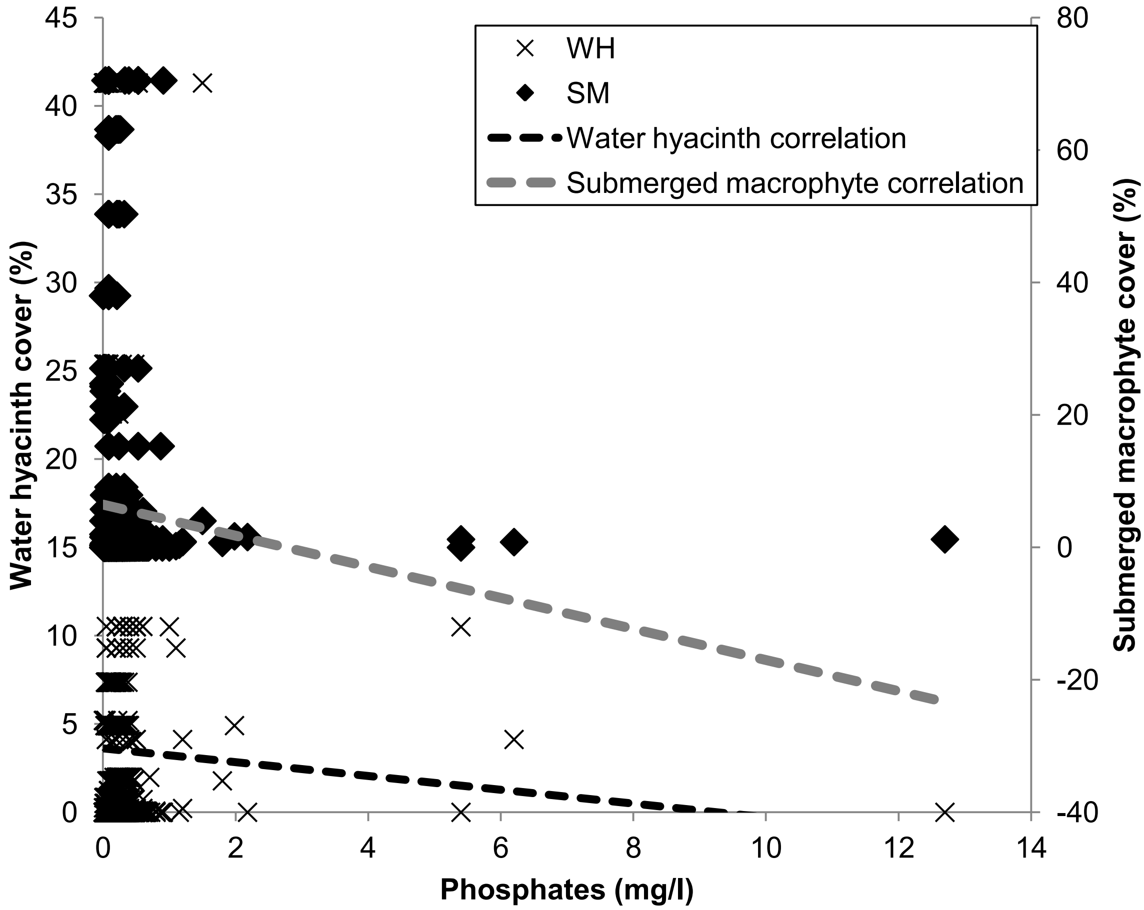



**Figure 2-22:** The correlation of WMA macrophyte cover and phosphate concentration. Data obtained from Department of Water Affairs Resource Quality Services (no data for WMA 10 in 2010).

As a result of the significant correlations between abiotic variables and macrophyte cover ,and because changes in plant dominance between water hyacinth and submerged macrophytes only occurred in WMA 10, WMA 10 regions were investigated in more detail to determine in which region the changes in plant dominance occurred and to determine why these changes occurred in WMA 10.

#### 2.4.1.7 Analysis of the spatial and physio-chemical variables influencing macrophyte growth in WMA 10.

The submerged macrophytes were the dominant macrophyte in WMA 10, while water hyacinth featured only briefly during 2006 and 2007 (Figure 2-23). The majority of the water hyacinth population was in WMA 10.4, with minor beds found in WMA 10.3 (Figure 2-23). Following the floods of 2006 (Figure 2-14), water hyacinth covered 10% of WMA 10 (Figure 2-23). This decreased to 5.3% in 2007 and between 2008 and 2009 water hyacinth covered less than 0.5% of WMA 10. Even the flood of 2010 did not provide enough of an opportunity for water hyacinth to regain any form of dominance across WMA 10, as its cover remained below 0.5%.

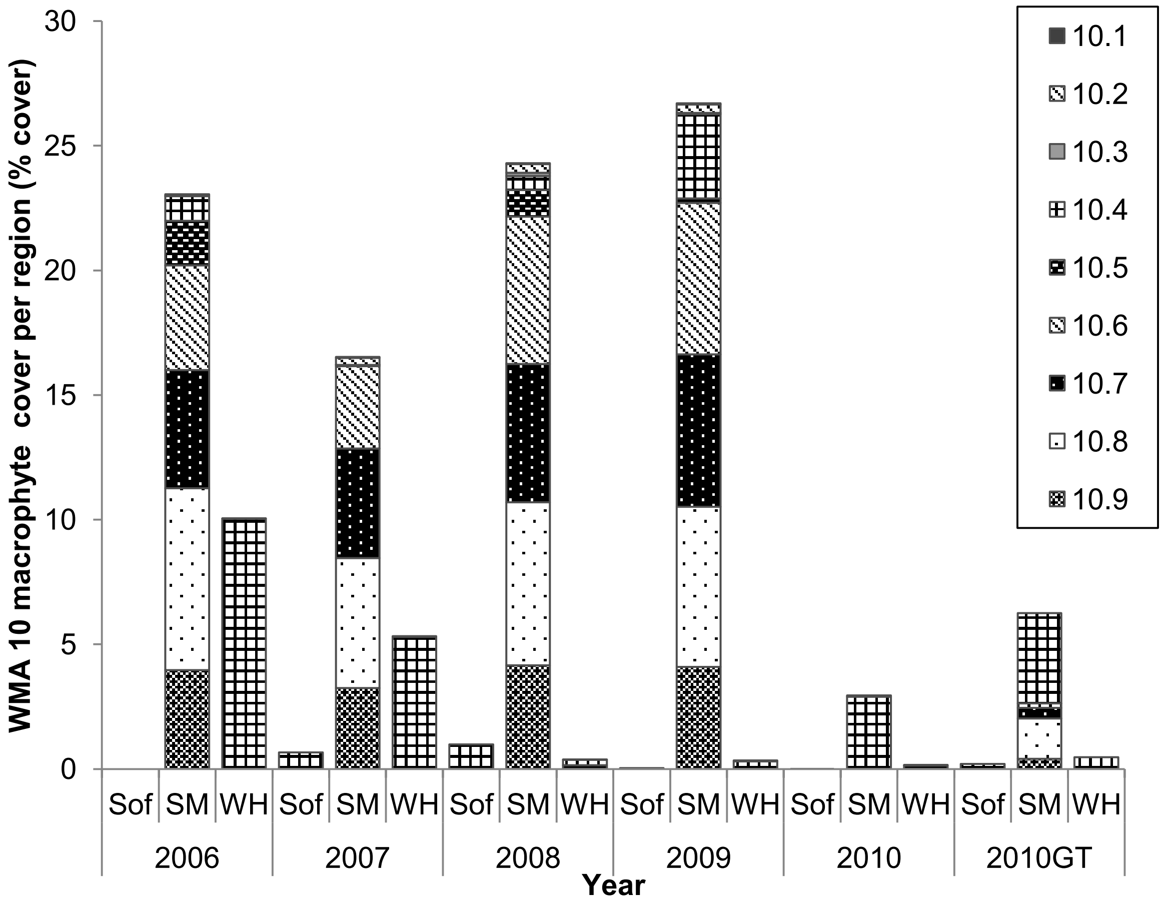



**Figure 2-23:** The water hyacinth (WH), submerged macrophyte (SM) and state of flux (Sof) communities cover of WMA 10 on the Vaal River. Macrophyte cover is broken down according to the WMA regions within which each state of dominance is located.

Throughout the study period, the submerged macrophyte community always maintained a strong presence in WMA 10 (Figure 2-23). In 2006, the submerged macrophyte community of WMA 10 covered more than 24% of the WMA, the majority within regions WMA 10.6, 10.7, 10.8, 10.9, while smaller portions existed in 10.5 and 10.4 (Figure 2-23). From 2006 until the floods in 2010 (Figure 2-14), the majority of the submerged macrophyte population predominantly resided in WMA 10.9, 10.8, 10.7, and 10.6, the extent of the cover only changing by small percentages (<2.5%). The submerged macrophyte cover of WMA 10.4 and 10.5 fluctuated, starting with 4 % in 2006, decreasing to <1 in 2007, then both steadily increasing in 2008. By 2009, WMA 10.4 contributed 3.3% of the 26% submerged macrophyte cover of WMA 10. The spatial analysis of the 2010 imagery was compromised by flooding. The 2010 flood waters immersed submerged macrophyte beds in WMA regions 10.5, 10.6, 10.7, 10.8 and 10.9 below detection limits and therefore these beds could not be delineated. Only the submerged macrophyte beds in WMA 10.4 could still be mapped. These decreased slightly in cover from 3.3% to 2.9% of WMA 10. During the ground-truthing field trip in 2010, it became apparent that the majority of the submerged macrophyte beds had been highly impacted by the flooding, with WMA 10.4 the least affected as it increased to 3.6% cover of WMA 10. WMA 10.4 was also the only region where state of flux communities featured, which stands to reason as besides 10.3, none of the other regions had any substantial water hyacinth populations.

The analysis of WMA 10 revealed that WMA 10.1 – 10.3 had little to no macrophyte cover in their regions, while 10.7 and 10.9 followed the same trends in macrophyte cover since 2006 (Figure 2-23). It was therefore decided to investigate the aquatic macrophyte changes in WMA 10.4, 5, 6, and 8 (Figure 2-24) as they were the least stable regions in WMA 10 and would provide the greatest insight into the macrophyte dominance dynamics of WMA 10 and the relationship between macrophyte cover and physio-chemical conditions of the Vaal River (Figure 2-25). The most informative region in WMA 10 is WMA 10.4, as it changed dominance from a water hyacinth-dominated region to a submerged macrophyte-dominated region.

In 2006, water hyacinth dominated 10.4, covering 41.3% of WMA10.4 (Figure 2-24). This steadily declined to 22.5% in 2007, of which 14.8% inhabited new areas and was classed as new population. Only 7.8% of the 2006 population remained as a previously established water hyacinth class. The water hyacinth population crashed by 2008 to 1.8% of WMA 10.4 and remained at low levels of cover, never dominating more than 2% of WMA 10.4, until the ground-truthing field trip in 2010. Water hyacinth has been shown in section 2.4.1.6 to have no correlation with pH ($r = -0.006$, $P = 0.80$) or phosphate concentration levels ($r = -0.20$, $P < 0.05$), while there was a small negative correlation between water hyacinth and nitrate concentration levels ($r = -0.20$, $P < 0.05$). These data did not illustrate the negative correlation, but nitrate levels and water hyacinth cover (Figure 2-25). Both peaked in 2007 and dropping severely in 2008, which would have limited the proportion of new population growth of water hyacinth, especially in 10.4. These data were also significantly correlated over the rest of the Vaal River (Figure 2-20).

The submerged macrophyte community had a strong presence (since 2006) throughout WMA 10 (Figure 2-24), especially in WMA 10.4, 6, 7, 8. The submerged macrophytes of WMA 10.4 differed slightly compared to the trends noted in WMA 10.5, 10.6 and 10.8 (Figure 2-24). All the aforementioned regions started with a high percentage cover in 2006, followed by a slight decline in 2007 and growth throughout 2008, peaking in percentage cover in 2009, crashing in 2010 and a slight rebound in population in 2010GT data. The WMA 10.4 submerged macrophyte data for 2006 showed that submerged macrophytes inhabited only 4% of WMA 10.4. By 2007 this had decreased to 0.0002% cover of the region. As with the other regions, in this WMA, the increase in submerged macrophyte dominance began in 2008. Submerged macrophytes had expanded into new areas of WMA 10.4, as this is illustrated by the fact that 1% community was classed as 'new population', while the remainder of the community comprised 'previously established both' (0.1%), 'previously established water hyacinth' (0.3% and 'previously established submerged macrophyte' (0.2%) classes respectively.

The 2009 peak in submerged macrophyte dominance increased to 13.1%. 'New population' establishment constituted 4.3% of the community and was an indication of the submerged macrophytes inhabiting regions where no other macrophyte had been previously noted since 2006. The high percentages of 'previously established both' (4.2%), 'previously established water hyacinth' (2.3%) and 'previously established submerged macrophyte' (2.3%) classes were indications that in 2009, WMA 10.4 was conclusively dominated by submerged macrophytes.

Compared to the other WMA 10 regions illustrated in Figure 2-24, WMA 10.4 lost less of its population during 2010. The WMA 10.4 submerged macrophyte community decreased by only 1.5% in cover. The community composition still managed to occupy new habitat, as is illustrated by the community composition comprising 3% of the 'new population' class, the 'previously established both' class still maintained 3.8% of the population, while 2.2% of the community still inhabited regions previously dominated by water hyacinth communities (previously established water hyacinth class) and 2.6% inhabited regions only previously dominated by submerged macrophyte communities.

The increases in WMA 10.4 cover continued until the field trip in 2010 GT, when submerged macrophytes dominated the system with 15.4% cover (Figure 2-24). Small portions of the community consisted of habitat that had not been previously populated. This is illustrated by the 0.8% of the community that consists of the 'new population' class. In total 7.4% were classed as 'previously established both' class occupied regions in the WMA which had historically been populated by both dominant macrophytes. An additional 6.1% inhabited areas which had only been dominated by submerged macrophytes, 'previously established submerged macrophyte' class, while only 1% of the WMA 10.4 population inhabiting areas which were only dominated by water hyacinth, 'previously established water hyacinth' class. The constant increase in the 'previously established both' class from 2009 (4.2%), 2010 (3.8%) and 2010GT (7.5%), clearly illustrates how submerged macrophytes had recently become increasingly dominant in WMA 10.4 where dominance was constantly changing in the past (since 2006).

The changes in submerged macrophyte cover in WMA 10.4 were drastic and pH had only a minimal effect on these changes in cover. This is a direct result of the weak correlation between pH and submerged macrophyte cover ($r = 0.14$, $P < 0.05$). The pH values for this region dropped from 8.1 in 2005 to its lowest value recorded in 2007, 7.95, while it hardly changed in 2008 to pH 8.0 (Figure 2-25). The nitrate levels however did have a negative relationship with the changes in macrophyte growth for the region (Figure 2-25). Nitrogen concentration levels peaked in 2006 and constantly dropped in 2007, 2008, until in 2009, they were below detection levels limits, this despite the correlation shown in section 2.4.1.6 ($r = -0.22$, $P < 0.05$). Phosphate concentration levels remained stable, and thus had little effect on the submerged macrophyte cover, which complies with the weak correlation relationship discussed in section 2.4.1.6 ($r = -0.11$, $P < 0.05$)

WMA 10.6 had 27.6% submerged macrophyte cover in 2006, decreasing to 21.3% cover in 2007, of which 16.9% remained in habitat that was dominated by submerged macrophytes in 2006 and 4.4% had populated new habitat and was classed as a new population in 2007. The dominance of submerged macrophytes increased in 2008, with the plants covering 38% of WMA 10.6.

The majority of the 2008 population occurred in portions of WMA 10.6 that were previously dominated by submerged macrophytes and were classed as 'previously established submerged macrophyte' class. Of the submerged macrophyte cover 9.5% was recorded in habitat where it was not previously recorded and was classed a 'new population'. The WMA 10.6 submerged macrophyte population peaked in 2009, covering 39.1% of the region. The growth of the submerged macrophyte community in WMA 10.6 slowed, with only 1.7% of the 2009 population dominant in new regions of the river, and 37.5% of the population present in regions previously dominated by submerged macrophytes classed as 'previously established submerged macrophyte'. Similarly, as with WMA 10.8 and 10.7, the data from the 2010 analysis showed a decrease from the 2009 cover (37.5%). There was sudden disappearance of submerged macrophytes from WMA 10.6 in 2010. This could have been an opportunity for another species of macrophyte to dominate the region, however within a few months, by the time of the field trip in 2010, submerged macrophytes had recovered slightly to 1.1% cover of the 'previously established submerged macrophyte' class.

The high proportion of 'previously established' classes indicates that WMA 10.6 is a stable region. Analysis of the pH values of the system revealed that there was a small positive relationship between submerged macrophyte cover in WMA 10.6 and the pH values for this region. This corresponds with the weak correlation between pH and submerged macrophyte cover in section 2.4.1.6 ($r = 0.14$, $P < 0.05$). There were drops in pH in 2007, a large increase in 2008, and additional increases in 2009. This was enjoyed by the submerged macrophyte community in the region, which continued to show a small growth (Figure 2-25). Nitrate levels for the region showed an apparent inverse relationship with the submerged macrophyte cover for WMA 10.6. This is notwithstanding the significant correlation shown in section 2.4.1.6 ($r = -0.22$, $P < 0.05$) (Figure 2-25) between nitrate and submerged macrophytes. Nitrate concentration peaked in 2006 and constantly dropped in 2007 and 2008, until in 2009 it was below detection levels for DWA equipment, while phosphate concentrations remained stable: evidence of the weak correlation, discussed in section 2.4.1.6 ($r = -0.11$, $P < 0.05$), between phosphate and submerged macrophytes..

In WMA 10.8, submerged macrophyte populations peaked in 2006, with a population dominating 70.5% of the WMA. Slight decline to 49.4% was noted in 2007, followed by an increase in cover to 61.6% in 2008. By 2009 the population remained stable at 60% cover. Submerged macrophytes dominated the region from 2006, occupying all the available habitat, as is confirmed by the lack of any other classes, besides the previously established submerged macrophyte class. The data from the 2010 analysis showed a decrease in cover from 2009, essentially resetting the system, and no macrophytes were recorded from that dataset. Within a few months, cover had increased to 11.8% of previously established submerged macrophyte class (2010 GT). Although section 2.4.1.6 showed that submerged macrophyte cover significantly correlates with pH ($r = 0.14$, $P < 0.05$), nitrates ($r = -0.22$, $P < 0.05$) and phosphates ($r = -0.11$, $P < 0.05$), changes in submerged macrophyte cover did not correlate with the abiotic variables of WMA 10.8. The pH data were only available from 2008, and although they showed a slight decline, there was no change in submerged macrophyte cover (Figure 2-25). Large changes in cover for the submerged macrophytes in the region did not correlate with changes in nitrate or phosphate levels (Figure 2-25). Nitrate concentration levels remained low, with little variation from 2006 (Figure 2-25).

The 'state of flux' communities did not feature in WMA 10.5, 10.6, or 10.8, as only submerged macrophyte communities were present in those regions (Figure 2-24). However, the presence of water hyacinth in WMA 10.4 did result in some communities entering a 'state of flux' between water hyacinth dominance and submerged macrophyte dominance. 'State of flux' communities covered 2.8% of WMA 10.4 in 2007 (Figure 2-24), all of which were water hyacinth dominated-communities, however as was stated earlier, water hyacinth only dominated 0.8% of the WMA in 2008. This lack of conversion from 'state of flux' to a water hyacinth-dominated state probably resulted from submerged macrophyte communities beginning to dominate larger areas of the system and the low nitrate levels of the WMA, which would have handicapped water hyacinth growth. Regardless of the high percentage cover of water hyacinth communities across WMA 10.4 in 2007 (21%), accompanied by 2.8%

‘state of flux water hyacinth’ communities, the region had begun to change dominance and was starting to shift towards submerged macrophyte communities. In 2008 4.2% of the region was classed ‘state of flux submerged macrophyte’. This was when WMA 10.4 gained momentum to become submerged macrophyte-dominated in 2009.

The ‘state of flux’ communities in 2009 dominated only 0.1% of the WMA, and although all of these were water hyacinth-dominated communities, the lack of water hyacinth dominance in 2010 (0.4% cover) meant that these ‘state of flux’ communities did influence the extent of water hyacinth community dominance in 2010, as a result of the low nitrate concentration levels. These low ‘state of flux’ community values meant that there was a lack of competition for habitat between the two dominant macrophytes and even less area was occupied by communities in a state of flux in 2010 (0.008%), as similar values were obtained for 2010, even though these communities were water hyacinth-dominated.

The slight drop in submerged macrophyte cover in 2010 (11.6%), resulted in areas of the WMA opening up for dominance. This is reflected by the 2010GT ‘state of flux ‘communities covering 0.84%. Although the majority of these communities were water hyacinth-dominated states (as a result of their being previously dominated by submerged macrophyte communities), a small portion of the ‘state of flux’ communities was submerged macrophyte dominated (0.08%).

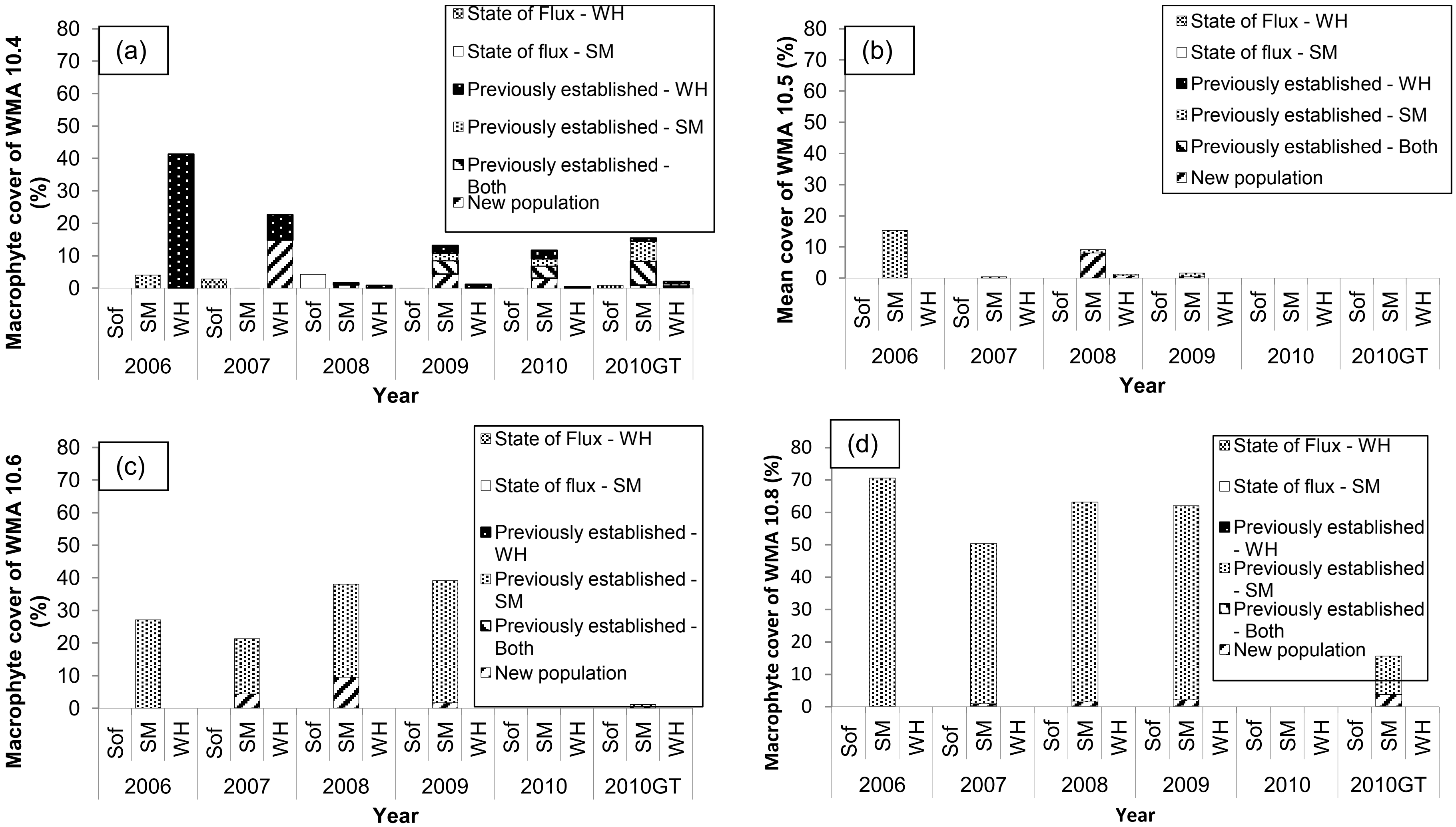



**Figure 2-24:** The cover of macrophyte communities within WMA regions from 2006 until the ground truthing field trip in October 2010 (2010GT) (a) 10.4, (b)10.5, (c)10.6 and (d) 10.8 in the Vaal River, illustrating the breakdown of water hyacinth (WH), submerged macrophyte (SM) and state of flux communities (Sof) in terms of the different classes, illustrated in the legend, derived from spatial analysis of SPOT V imagery.

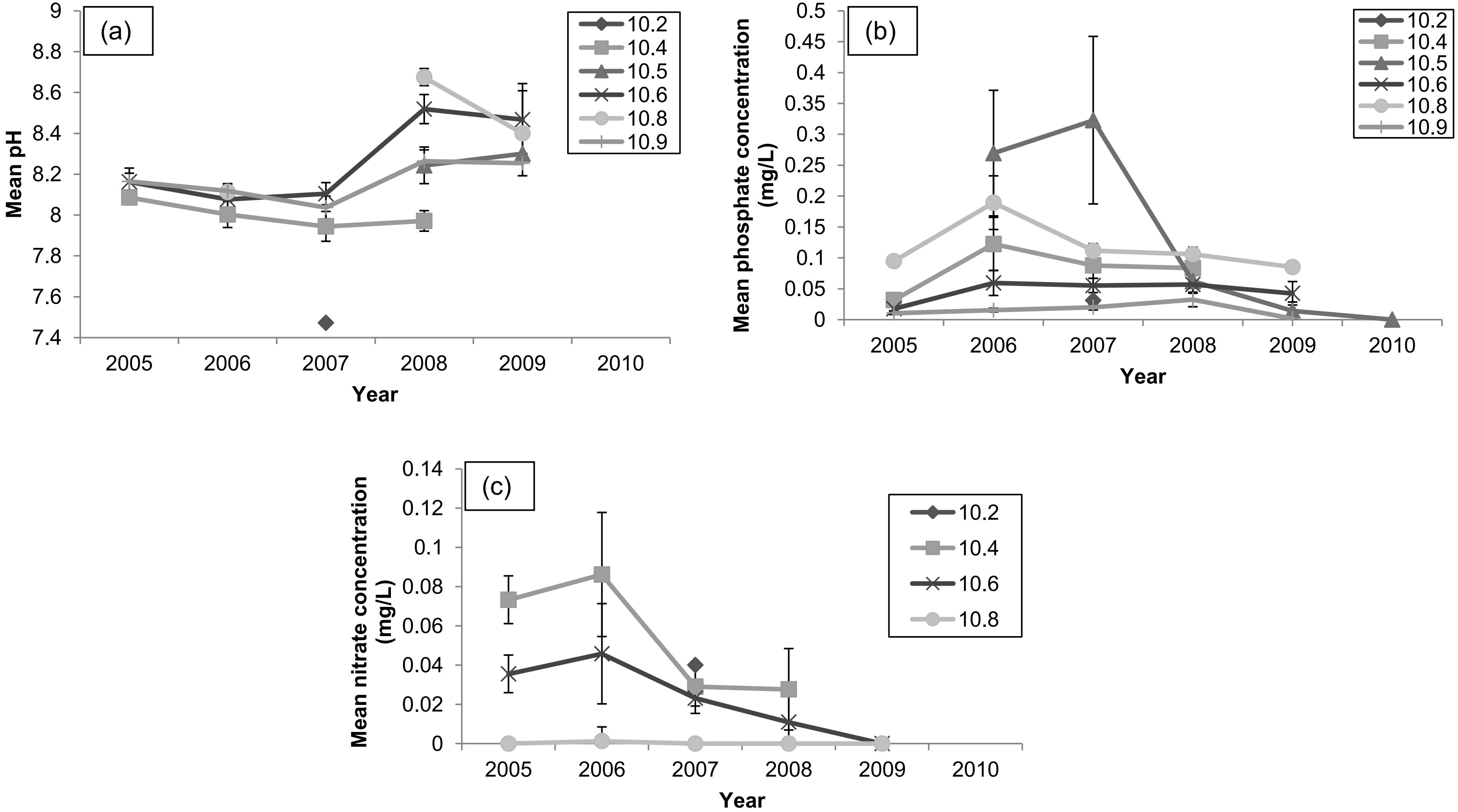



**Figure 2-25:** Changes in pH, phosphate and nitrates over time for the WMA 10 regions (a) pH, (b) phosphates (c) nitrates in the Vaal River.

Error bars represent standard error of the mean.

### 2.4.2 Water chemistry of Eurasian water-milfoil infestations

DWA RQS Regional monitoring points provided a broad description of the changes in physiochemical conditions of the water at a regional level at fixed monitoring points. These points could not provide insight into how the water chemistry varies at a localised scale. The lack of significant quantities of water hyacinth cover resulted in an insignificant number of data sites (5) captured to allow for accurate comparison between the two macrophytes. Forty two Eurasian water-milfoil sites had a number of factors examined and during the ground-truthing survey distributed throughout the study area and different densities of Eurasian water-milfoil beds (Table 2-5), Total Dissolved Solutes (TDS) and pH were measured, while water samples were later analysed for nitrate and phosphate concentrations. All of these data were analysed according to WMA region and Eurasian water-milfoil population density.

**Table 2-5:** A table depicting the number of sampling events that occurred within the Eurasian water-milfoil plant beds and outside in nearby open water.

| Classes analysed | Number of water samples (n) | | Number of soil samples |
|---|---|---|---|
| | Within plant bed | In nearby open water | |
| WMA 8 | 4 | 3 | N/A |
| WMA 9 | 3 | 3 | N/A |
| WMA 10 | 9 | 20 | N/A |
| None* | - | 3 | 1 |
| Rare | 9 | 3 | 2 |
| Occasional | 4 | 3 | 2 |
| Frequent | 4 | 3 | 2 |
| Abundant | 3 | 3 | 2 |
| Dense | 4 | 3 | 2 |

* There were no nearby Eurasian water-milfoil beds associated with the None category.

TDS is an indication of the solutes available for absorption and utilisation by aquatic macrophytes, and this attribute was averaged and grouped according to the river region

(Figure 2-26). There was no significant difference in TDS between the various WMA regions within the extent of the submerged macrophyte beds or in open water ($H_5$ = 10.39, *P* =0.06), although WMA 10 had a lower TDS than all the other WMA regions.

Similarly, comparing the population density scores of individual Eurasian water-milfoil populations, it was found that the TDS did not vary significantly between the different densities and classes ($H_{10}$ = 10.67, *P* =0.38) (Figure 2-27). This was attributed to the high variability in TDS values throughout the system. It was expected that there would also be a correlation between population density and TDS values which was not evident. Eurasian water-milfoil is known to reduce current velocity in large beds and thus create a settling effect of solids, thereby decreasing TDS values (Smith & Barko 1990).

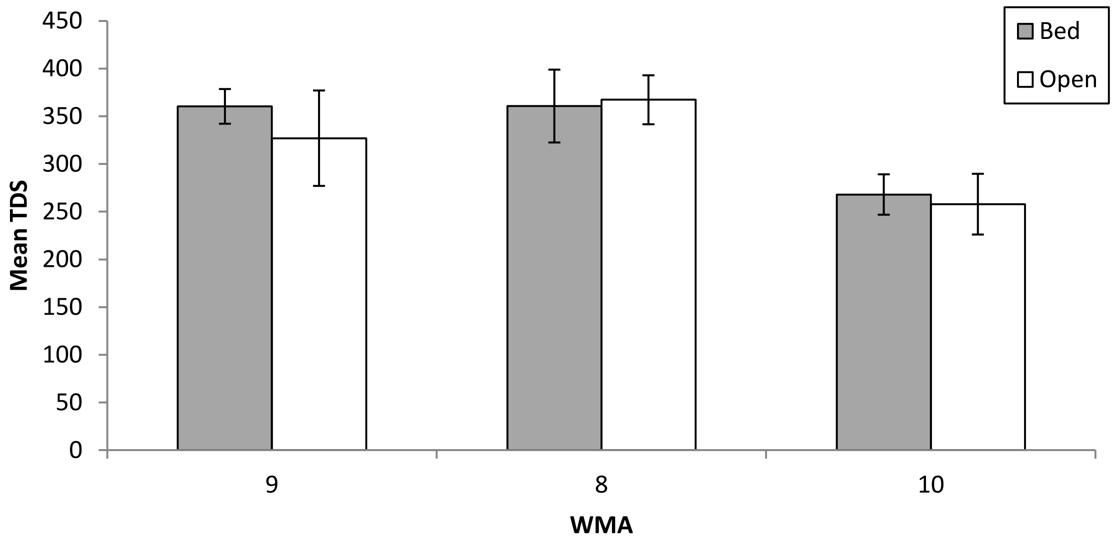



**Figure 2-26:** Mean TDS values taken in October 2010 during the field survey from within Eurasian water-milfoil populations, in three different WMA regions of the Vaal River. Error bars represent standard error of the mean.

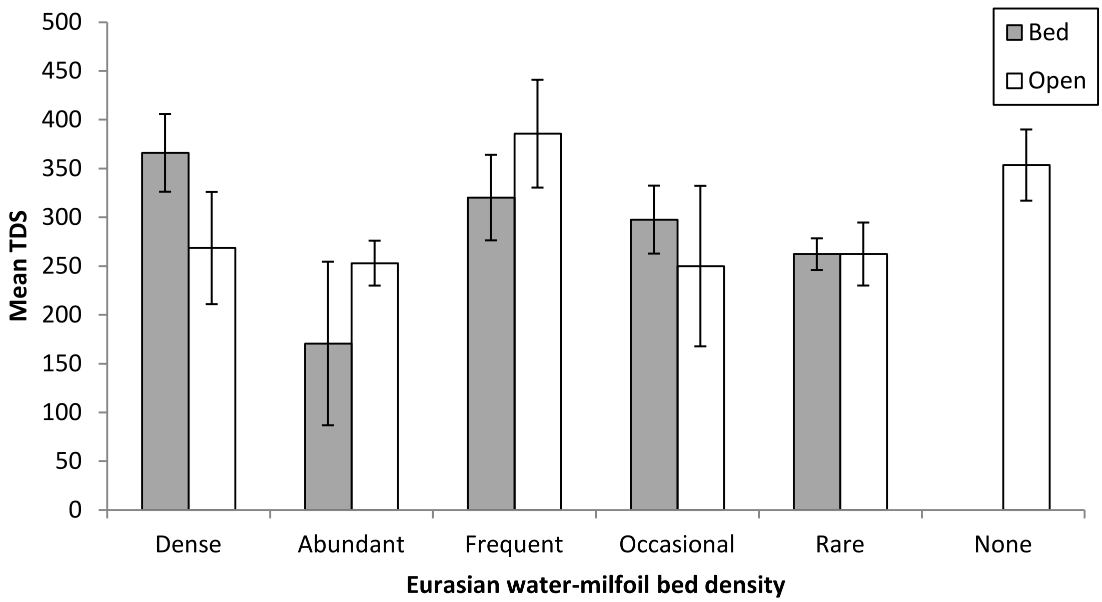



**Figure 2-27:** Mean TDS values taken in October 2010 during the field survey from within beds and in open water of Eurasian water-milfoil populations of different densities of dense throughout the Vaal River. Error bars represent standard error of the mean.

None of the regions had significantly different pH values ($H_5$ = 3.90, $P$ = 0.56) (Figure 2-28). These data show that the Vaal River is an alkaline system, with pH averaging above 8 throughout the system (Figure 2-28). These data compared favourably with the DWA RQS data, which had similar ranges in pH values, correlating with Eurasian water-milfoil cover (Figure 2-18). There was a significant difference in pH between Eurasian water-milfoil density classes, inside the beds and outside in open water ($H_{10}$ = 18.59, $P$ = 0.04). The occasional and frequent beds had relatively large differences between their pH values inside and outside the bed. This was not evident with the rare, dense and abundant classes (Figure 2-29).

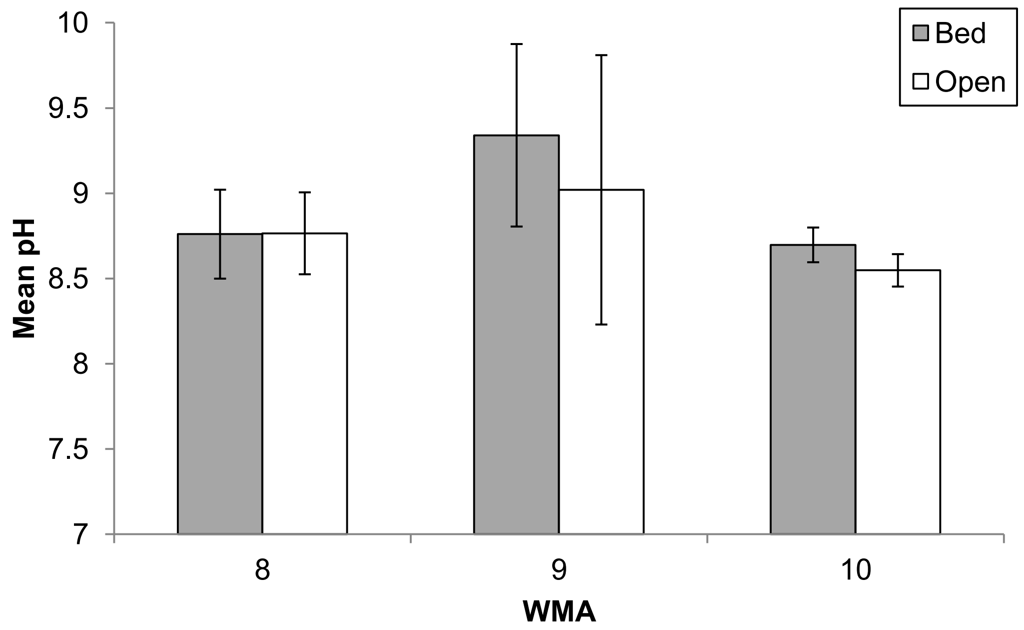



**Figure 2-28:** Mean pH values taken in October 2010 during the field survey from within Eurasian water-milfoil populations in three different WMA regions of the Vaal River. Error bars represent standard error of the mean.

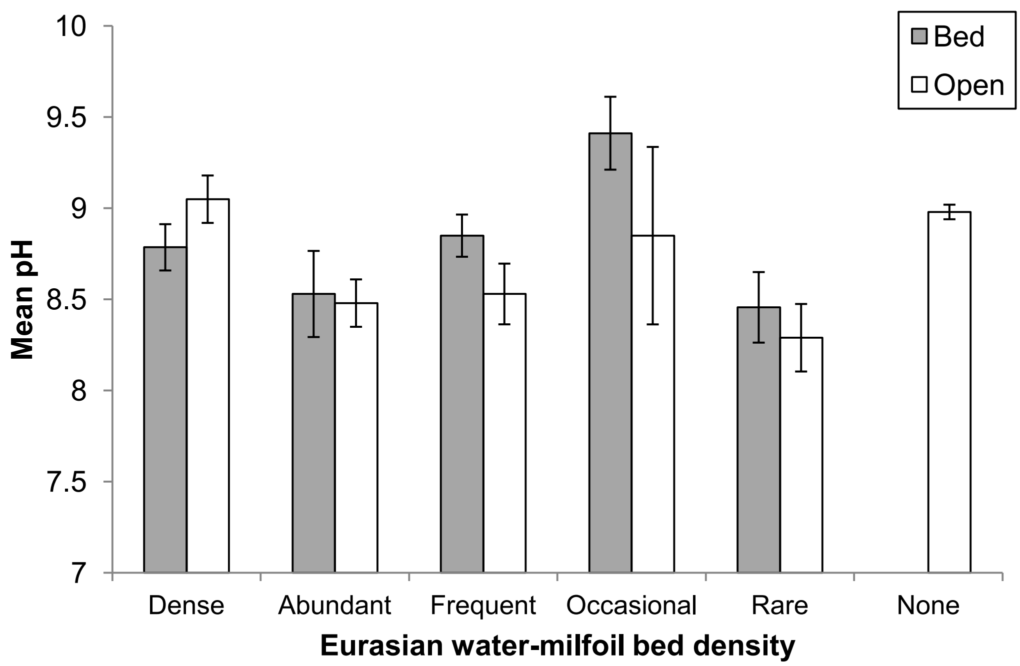



**Figure 2-29:** Mean pH values taken in October 2010 during the field survey within Eurasian water-milfoil populations of different densities throughout the Vaal River. Error bars represent standard error of the mean.

The DWA RQS nitrate concentration levels correlate with Eurasian water-milfoil population cover section 2.4.1.6. ($r = -0.22$, $P < 0.05$) (Figure 2-20). The nitrate concentrations were also correlated with WMA regions (Figure 2-30). The highest concentrations were found in WMA 9, with levels below detection limits in WMA 8, however there was no significant difference between any of the nitrate levels throughout the various WMA regions ($H_5 = 6.55$, $P = 0.26$).

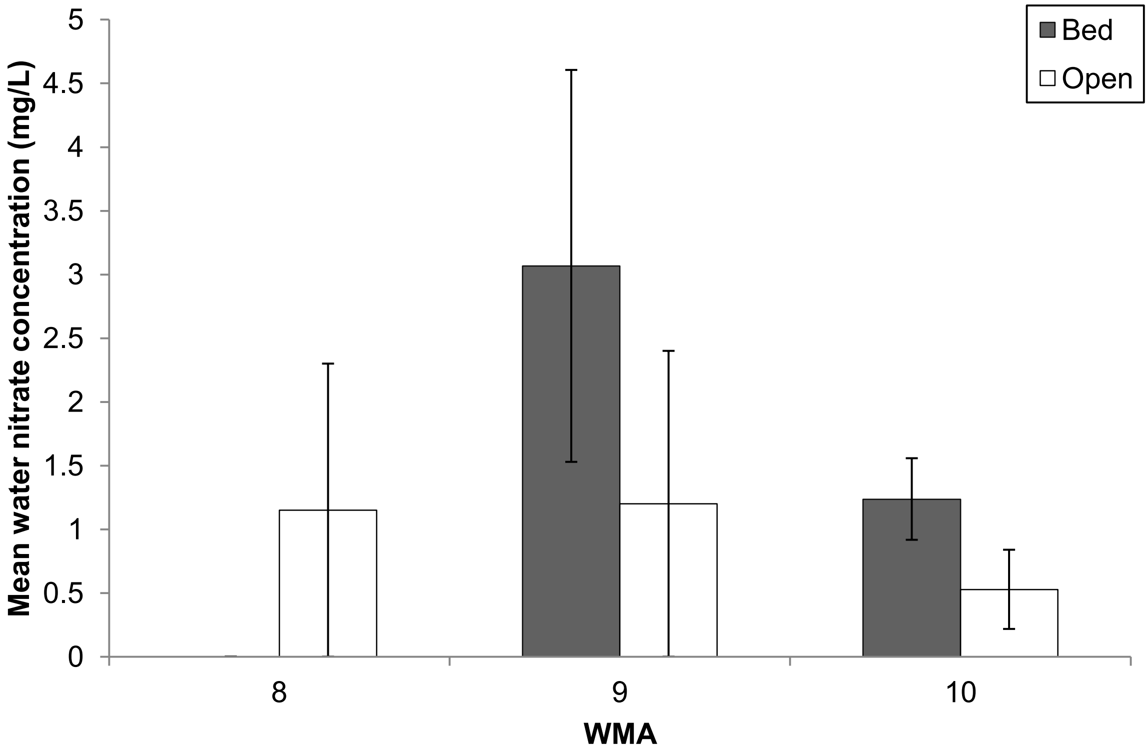



**Figure 2-30:** Mean nitrate concentrations taken in October 2010 during the field survey from within Eurasian water-milfoil populations in three different WMA regions of the Vaal River. Error bars represent standard error of the mean.

The highest levels of nitrates were found in occasional and rare density classes (Figure 2-31). Frequent and occasional density class data were not consistent with the negative trend that nitrate concentration levels showed with the other classes. There was no significant difference between density classes of Eurasian water-milfoil inside the beds and outside ($H_9$ = 5.84, $P$ = 0.21). A limitation to this analysis was that open water nitrate concentration levels were often below the detection limits for the equipment used to analyse the nitrate concentrations. These nitrate levels were, however, still similar to the DWA RQS nitrate concentrations in terms of the range in concentration values that were captured (Figure 2-19).

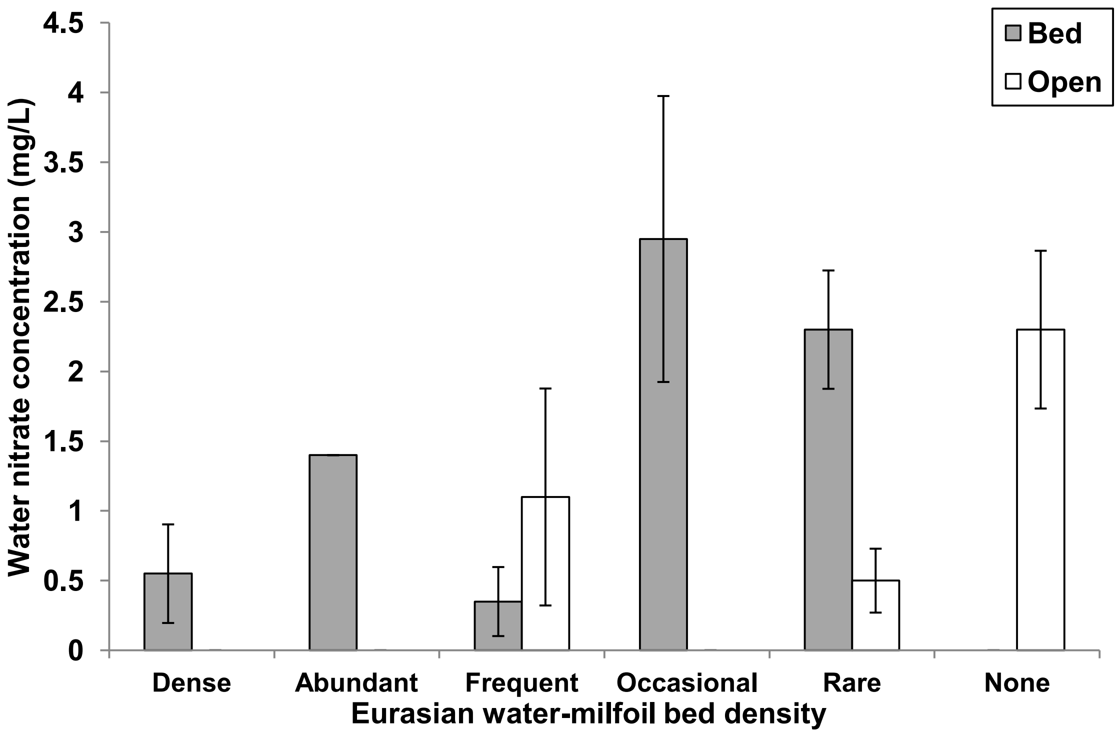



**Figure 2-31:** Mean nitrate concentrations taken in October 2010 during the field survey from within Eurasian water-milfoil populations of different densities throughout the Vaal River. Error bars represent standard error of the mean.

Similarly to DWA RQS nitrate concentrations, the phosphate concentrations did correlate with Eurasian water-milfoil cover, as discussed in section 2.4.1.6. (r = -0.11, $P$ <0.05), (Figure 2-22). The phosphate concentrations were also compared between the WMA regions (Figure 2-32 and Figure 2-30). The highest concentrations were found in WMA 8 open water, with a general downward trend of phosphates outside the Eurasian water-milfoil populations than inside. Although there was no significant difference between phosphate levels of the three regions ($H_5$ = 5.65, $P$ = 0.34), there was a higher phosphate concentration in WMA 10.

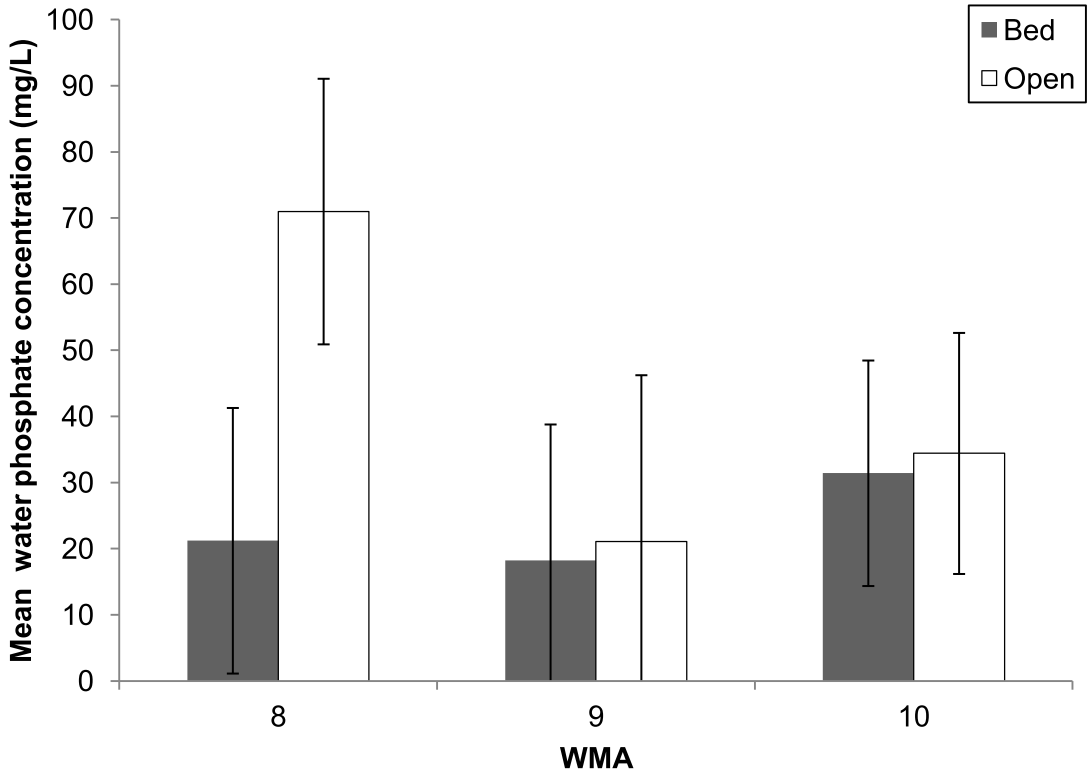



**Figure 2-32:** Mean phosphate concentrations taken in October 2010 during the field survey from within Eurasian water-milfoil populations in three different WMA regions of the Vaal River. Error bars represent standard error of the mean.

The highest levels of phosphates were found in dense and rare classed population beds, although frequent classed beds had greater amounts of phosphates present in their water column, compared to the nearby open water (Figure 2-33). With the exception of the frequent class, the open water phosphate concentrations were all less than the phosphate concentrations found within the population classes. There was no significant difference between the phosphate concentrations within the beds or in open water ($H_9$ = 10.77, $P$ =

0.23). These phosphate levels however were substantially higher than those previously recorded by the DWA RQS data in terms of the range in concentrations (Figure 2-21).

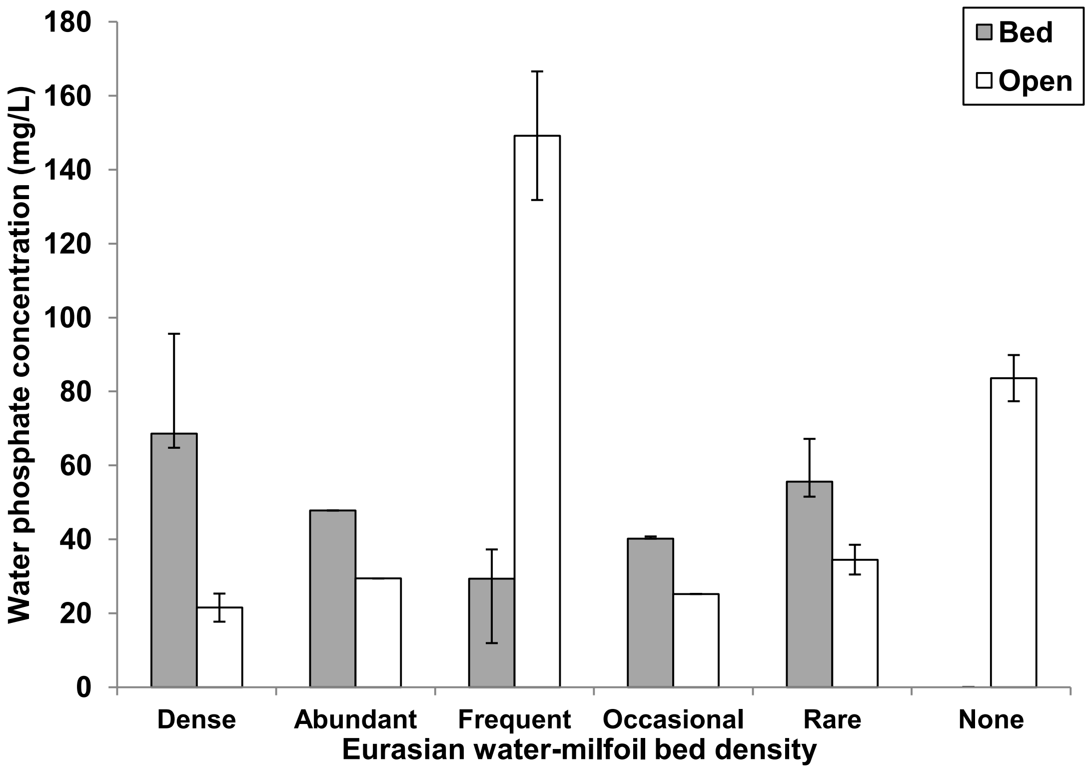



**Figure 2-33:** Mean phosphate concentrations taken in October 2010 during the field survey from within Eurasian water-milfoil populations of different densities throughout the Vaal River. Error bars represent standard error of the mean.

### 2.4.2.1 Sediment Chemistry

Twelve sediment samples were taken and analysed throughout the study area (Table 2-5). The most important factors governing sediment chemistry were the macro nutrient availability (total nitrogen and total phosphorous levels), sediment pH, total calcium and sediment composition. The overriding factor influencing Eurasian water-milfoil's ability to abstract nutrients from the sediment is the pH (Figure 2-34). pH values were not significantly different between the Eurasian water-milfoil bed density classes ($H_6$ =4.88, $P$= 0.56).

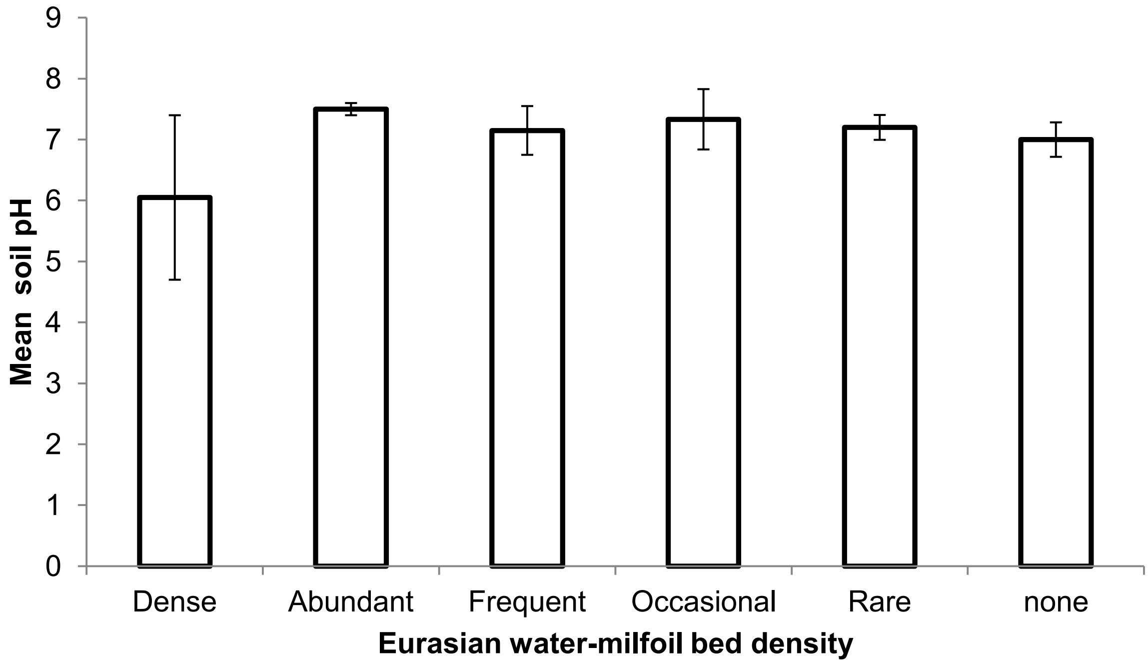



**Figure 2-34:** Mean sediment pH of soils determined from samples collected during the field survey in October 2010 from within Eurasian water-milfoil populations of different densities. Error bars represent standard error of the mean.

Macronutrients such as nitrogen and phosphorous play essential roles in plant behaviour and growth rates. The total soil nitrogen levels showed no significant difference relative to the density score assigned to the Eurasian water-milfoil population ($H_6$ = 5.36, $P$ = 0.50) (Figure 2-35), although the levels of nitrogen were generally high throughout the study site, indicative of a eutrophic system.

Total phosphorous content of the sediment is another important variable for aquatic macrophyte growth. The mean total soil phosphorous levels were not significantly different relative to the density score assigned to the Eurasian water-milfoil population ($H_6$ = 5.36, $P$ = 0.5) (Figure 2-36). The levels of phosphorous were generally high across the study site, which is indicative of a eutrophic system.

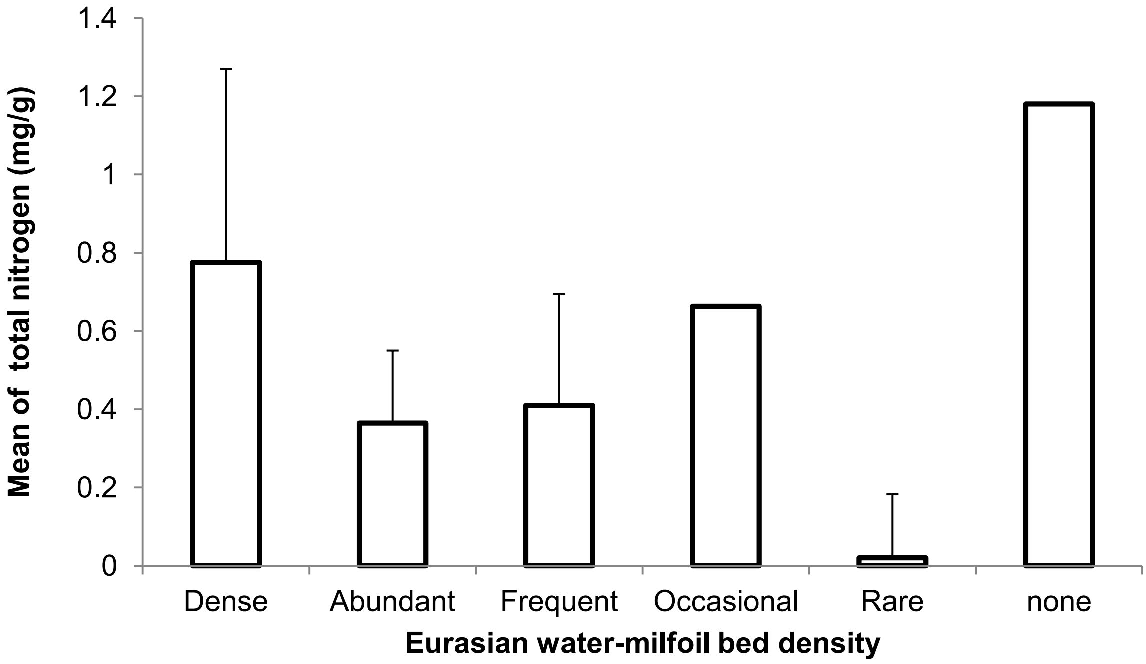



**Figure 2-35:** Mean total nitrogen levels of soils of soils determined from samples collected during the field survey in October 2010 from within Eurasian water-milfoil populations of different densities. Error bars represent standard error of the mean.

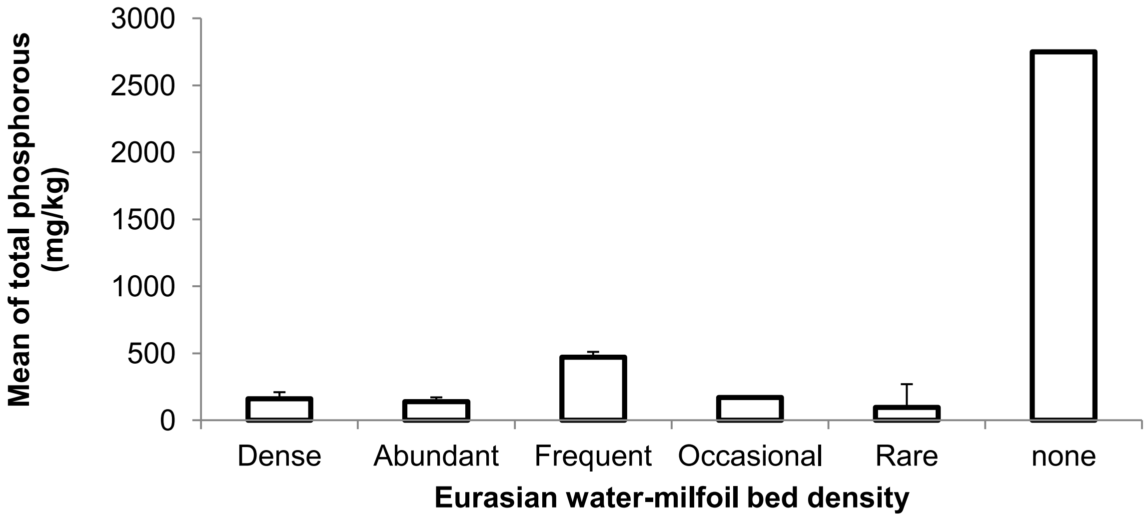



**Figure 2-36:** Mean total phosphorous levels of soils of soils determined from samples collected during the field survey in October 2010 from within Eurasian water-milfoil populations of different densities. Error bars represent standard error of the mean.

Total calcium sediment availability was highlighted by Ali & Soltan (2006) as a nutrient that is influenced significantly by the presence of Eurasian water-milfoil infestations. This sediment micro nutrient was analysed relative to Eurasian water-milfoil population densities across the study area (Figure 2-37). The mean soil calcium levels were not significantly different, relative to the density score assigned to the Eurasian water-milfoil population ($H_6 = 6.10$, $P = 0.41$) (Figure 2-37).

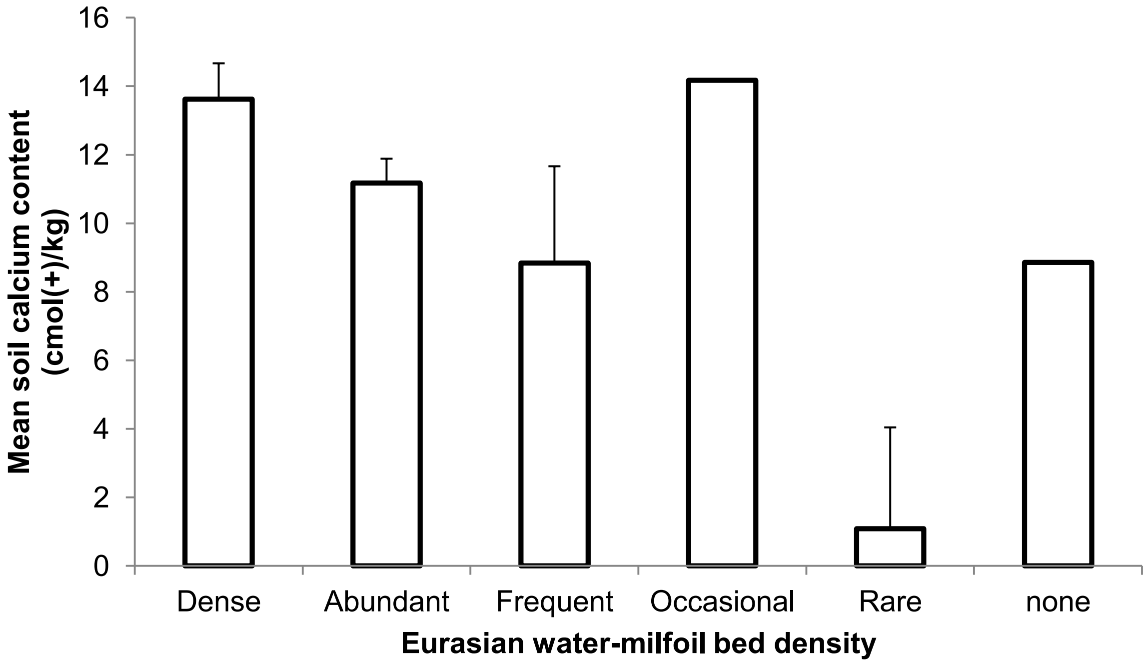



**Figure 2-37:** Mean total calcium levels of soils of soils determined from samples collected during the field survey in October 2010 from within Eurasian water-milfoil populations of different densities. Error bars represent standard error of the mean.

Lastly, sediment physical characteristics play an essential role in Eurasian water-milfoil ability to absorb nutrients from the sediment, which directly influences its behaviour and competitive ability. Analysis revealed that all the sediment samples had a significantly high sand content (Figure 2-38). When conducting statistical analysis of the variability of the soils according to Eurasian water-milfoil bed density it was found that there were no significant differences across the range of different densities, (clay, $H_6 = 6.43$, $P = 0.38$; silt, $H_6 = 6.65$, $P = 0.35$ and sand, $H_6 = 6.82$, $P = 0.34$; ). Overall comparison revealed that the sediment of the Vaal River was significantly more sandy (compared to clay $P < 0.0001$ and to silt $P < 0.0004$)

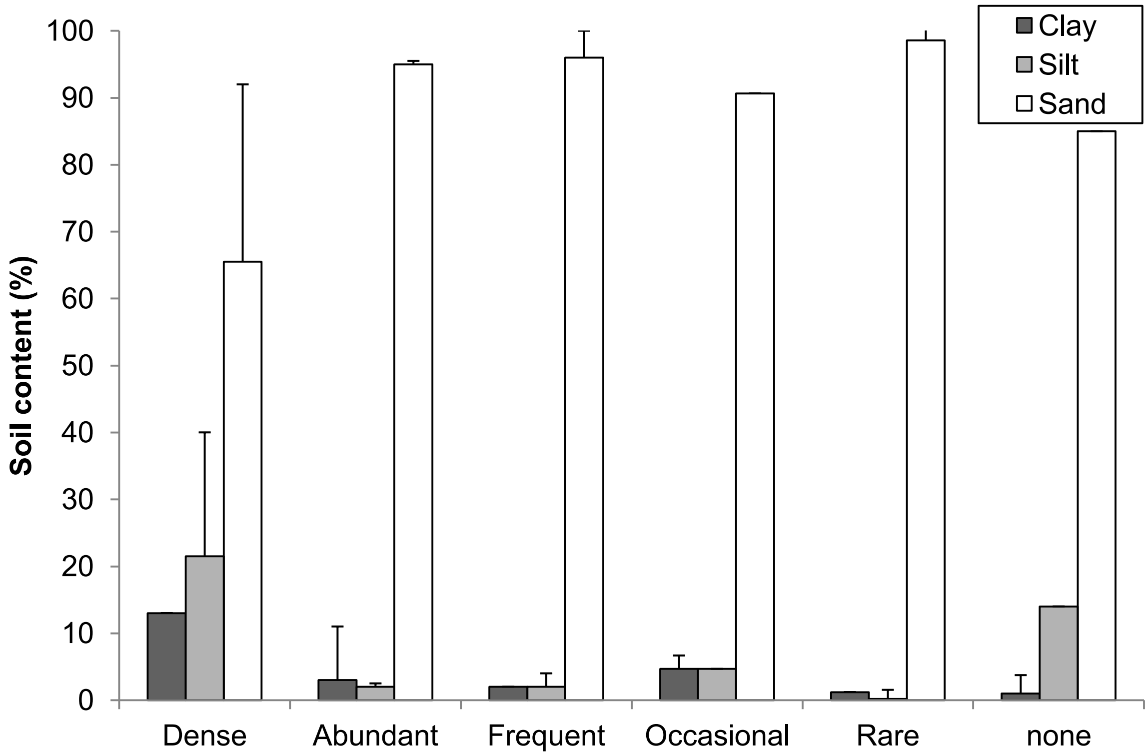



**Figure 2-38:** Physical characteristics of sediment determined from samples collected during the field survey in October 2010 from within Eurasian water-milfoil populations of different densities. Error bars represent standard error of the mean.

## 2.5 *Discussion*

The aim of this chapter was to perform a spatial and temporal analysis of the distribution and abundance of invasive alien aquatic macrophytes that have dominated the Vaal River from 2006. These data were then linked to stochastic events, integrated weed management control programmes, changes in water and sediment physical and chemical conditions. The availability of suitable GIS imagery, software and techniques, allowed accurate spatial and temporal analyses of the large aquatic ecosystem of the Vaal River. Visual classification was preferred to an automated classification method due to a high spatial accuracy requirement. The high accuracy assessment values (Table 2-3) bear testament to the visual classification method in determining the extent and interactions between abiotic conditions and aquatic macrophyte cover. Using these images, it is evident that the Vaal River has been a relatively stable system in terms of regions of aquatic macrophyte dominance (except for WMA 10.4), this despite it being subjected to large periodic flooding events, changes in water and sediment physio-chemical properties and integrated control measures by WfW.

During the spatial analyses, the problem with dividing the water hyacinth population into such classes is that it is a floating plant, it moves with the prevailing current and wind conditions of the system and therefore a singular population of water hyacinth moving down the river could continuously be reclassified as ‘new population’. However, what became apparent is that although this was a limitation of this study, portions of the water hyacinth community continuously inhabit or get stranded or stuck in the same positions each year. When analysing the water hyacinth data, it was more important to note when the ‘previously established’ portion of the population became larger as this was an indication of the population decreasing in size. It was also expected that a greater portion of the ‘state of flux’ state would be water hyacinth-dominated, however this was not the case.

The lack of any water hyacinth dominance in WMA 8 could be a result of the regions geomorphology. It is characterised by a high number of rapids and fast flowing water, with banks of riparian vegetation that overhang and shade substantial portions. According to Gopal (1987), this type of environment is not suitable for water hyacinth dominance as although it can persist in fast flowing waters, rapids cause physical damage to the plants and with the overhanging vegetation covering the calmer waters, water hyacinth has battled to remain established and dominant in this region for extended periods. Without competition from water hyacinth, it was expected that submerged macrophytes would dominate this WMA 8. The lack of a stable Eurasian water-milfoil macrophyte population can be attributed to the high flow rates of the region. Eurasian water-milfoil does not thrive with flow rates greater than $0.4m^3/s$ (O'Hare *et al.* 2007). According to Moodley *et al.* (2006), the discharge rates of water from the Barrage average about $20m^3/sec$. The lack of suitable habitat and the fast flowing waters of WMA 8 are primary reasons for the lack of any significant stable macrophyte populations.

WMA 9 is a stable water hyacinth-dominated region (Figure 2-17). Water chemistry of the system was analysed and showed that although the pH values of WMA 9 were high for water hyacinth (Ruiz Téllez *et al.* 2008) (Figure 2-17), they did not limit the percentage cover of WMA 9. The pH values of the Vaal River system indicated that it was very alkaline, and this would have hampered water hyacinth growth rates, but for water hyacinth to be negatively affected by pH, values need to be below 6 or above 8 (Ruiz Téllez *et al.* 2008). It is also a zero tolerance zone in terms of WfW's herbicidal glyphosate spray regime. Following peaks in water hyacinth cover in 2007 and 2008, an intense programme was initiated and the spraying, as well as the presence of biological control agents, had significant impacts on the water hyacinth population levels (C. Sharpe, WfW WMA 9 Manager, Pers. comm. 2011). This would have been assisted by a reduction of nitrate in the water (Figure 2-19). Water hyacinth also has the ability to regulate water chemical conditions and compensate with reduced growth rates, and a lack of competition for resources allows water hyacinth to retain its position as the stable dominant macrophyte in WMA 9.

Submerged macrophytes did not feature in WMA 9 over the study period, although compared to WMA 8, the topology and hydrological flow regime is better suited for their dominance. Bloemhof Dam is the largest component of this region, WMA 9.6 - 9.9 fall within the extent of Bloemhof Dam (Figure 2-4). The dam is not suitable for submerged macrophyte plant growth as it is simply too turbid (Moodley *et al.* 2006). These authors noted how the TDS remains constant from Barrage down to the top of Bloemhof Dam, following which the influx of suspended solids increases the turbidity of the dam (Grobler *et al.* 1983). This combined with the frequent algal blooms, experienced by the dam (Moodley *et al.* 2006), directly influences and limits the establishment of stable submerged aquatic plant growth.

Spatial analysis revealed that WMA 10 was a stable submerged macrophyte-dominated region (Figure 2-23), with the exception of WMA 10.4, Warrenton Weir (Figure 1-2). The submerged macrophyte communities that have established on the lower reaches of WMA 10 maintained a stable state, primarily as they have suitable hydrological flow regimes (compared to WMA 8), less turbid waters (compared to WMA 10 (Figure 2-26)) and they lack competition from water hyacinth. The large stands of stable submerged macrophytes limit any form of substantial water hyacinth growth as the abiotic conditions are not suitable (Figure 2-25). The submerged macrophyte communities are associated with reduced nitrate levels in the water column and have maintained high pH, which limit water hyacinth growth (Ruiz Téllez *et al.* 2008).

WMA 10.4 is the only region in WMA 10 which had suitable abiotic conditions for water hyacinth dominance. This region had a definite shift in stability, when following 2006, water hyacinth was the dominant stable state. The cumulative effect of the reduction in nitrate concentrations (Figure 2-25), be it either via water hyacinth uptake or via a reduction in input, and WfW's integrated control programme in 2008 (N. Byleveld, WfW WMA 10 manager, Pers. comm. 2011), shifted WMA 10.4 to a Eurasian water-milfoil alternative stable state of dominance. Eurasian water-milfoil was present in the region, but required suitable abiotic conditions to grow, the main restriction of which was the shading effect of water hyacinth.

Following the analyses of all of the regions it became apparent that the source to sink model described by Pulliam (1988), was highly applicable. This model is able to describe how variation in the quality of habitat affects population growth or decline of organisms. Source populations occupy a high quality environment, which allows them to thrive to such an extent that the population is exporting individuals. Sink populations occupy a low quality less desirable environment where the population exists but is declining in numbers. The sink population only persists as long as it can continue to import individuals from the source population.

When applying this model to the Vaal River, it is evident that for water hyacinth, the highest quality environment is WMA 9 and periodically WMA 10.4, this regions acts as a source of all the other populations downstream. It is likely that another source population exists above WMA 8, however this was not investigated during this study. Sink populations periodically exist in WMA 10.5, but as a result of effective WfW WMA integrated control measures, the source populations are not permitted to get large enough to continuously resupply the sink populations.

The submerged macrophyte populations definitely have a source population in WMA 10.4, in particular Eurasian water-milfoil. Without any form of control implemented against Eurasian water-milfoil, this has allowed this region to continuously resupply all sink populations downstream.

Stochastic events play an important role in aquatic ecosystems and form the basis of the theory of alternative stable states, defined by Lewontin (1969). In a stable community, species composition/dominance will remain the same if conditions do not change. Large stochastic events, such as the flood in 2006, have direct effects on the conditions and cause dramatic shifts in stability, essentially resetting the niches in the system, presenting opportunities for the species present to re-compete for dominance of the system. Arguments against the existence of alternative stable states have largely been based on the facts that the experimental designs used to model alternate stable states are rarely conducted and are often criticised for being ambiguous in design and in interpretation (Schröder *et al.* 2005). It is however widely accepted that the most comprehensive evidence regarding the theory and existence of alternative stable states have often been in the form of large scale shifts in major ecosystems (Schröder *et al.* 2005). This is particularly evident in this study, where WMA 10.4 had a large scale shift from a free floating plant (water hyacinth)-dominated state to an alternative submerged macrophyte (Eurasian water-milfoil)-dominated stable state.

A switch in species dominance can often be initiated by a single or multiple successive disturbances known as perturbations (May 1977, Moss 1998, Scheffer 1998). Perturbations can vary in size from large-scale stochastic events, such as floods or droughts, to smaller consistent cumulative changes in abiotic or biotic conditions. Dent *et al.* (2002) further illustrate how perturbations are the potential energy required to escape a stable state and enter into an alternative stable state (Figure 2-39). The perturbations which initiated the change in stable states of WMA 10.4 would be the flooding event of 2006, followed by the WfW integrated control measures initiated against water hyacinth. When these factors combined with unfavourable water physio-chemical conditions, this switched the ecosystem from a water hyacinth-dominated state to an alternative Eurasian water-milfoil-dominated state by 2009.

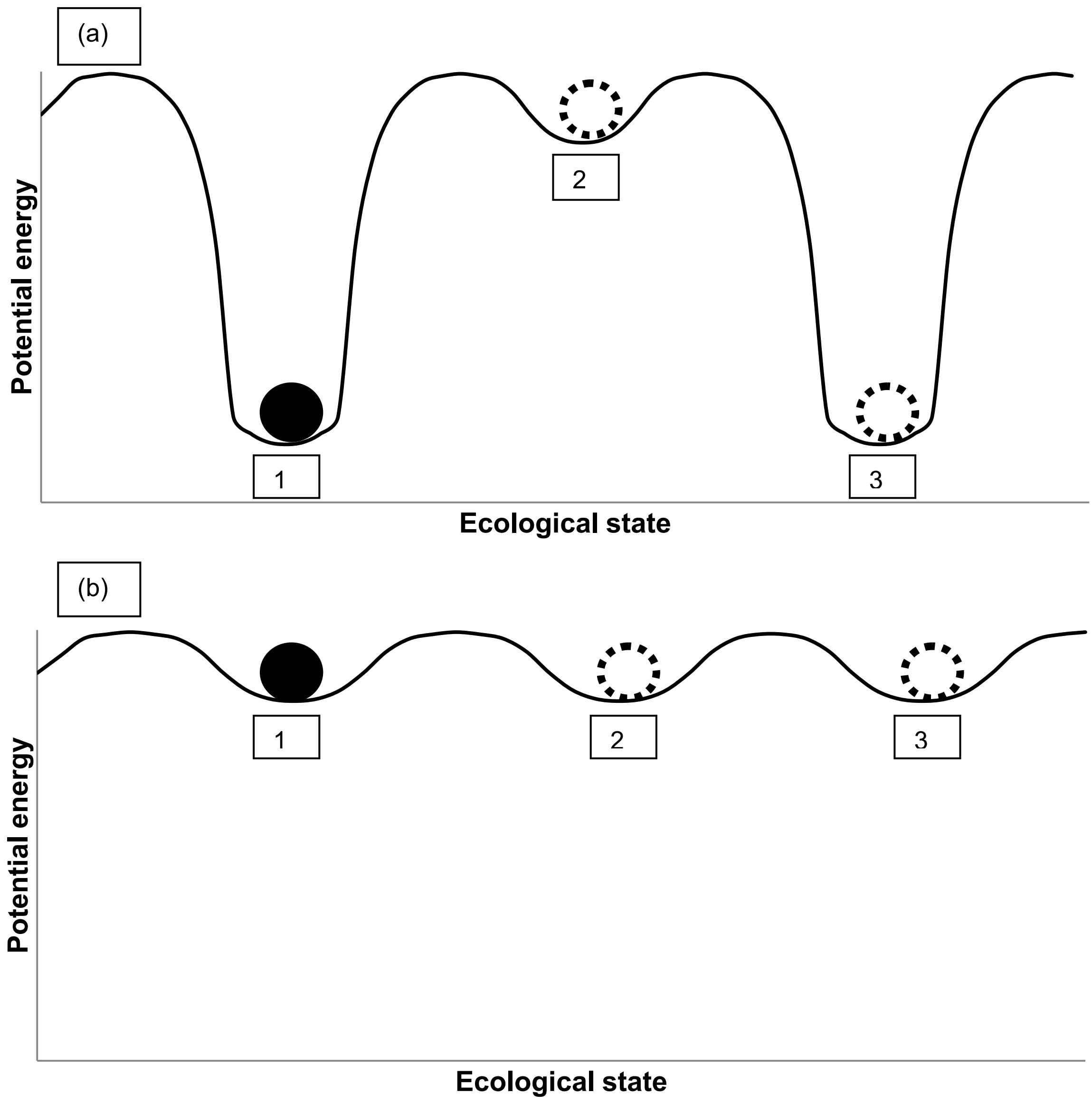



**Figure 2-39:** Graphs illustrating a variety of alternative stable states commonly referred to as the 'Ball and Cup' analysis, modified from Dent *et al.* (2002), (a) represents the theoretical alternative stable state for lake ecosystems and (b) illustrates the model for theoretical alternative stable state of riverine ecosystems

Resilience has undergone several changes in definition and has been interpreted differently by a variety of authors (Pimm 1991, Peterson, *et al.* 1998). Beisner *et al.* (2003) clarified the definition as to how steep the walls of the "cup" are, in the "ball and cup" model (Figure 2-39). When comparing (a) with (b) (Figure 2-39), the current stable state for the community is location 1, illustrated by the black circle or "ball". For a community to enter into a potential alternative stable state, a large quantity of perturbation is required for the "ball" to overcome the high resilience of the position 1 stable state. Conversely, for scenario (b) less perturbation is required for the "ball" to enter into an alternate stable state, due to the low resilience of the cup, which is typical of riverine systems (Beisner *et al.* 2003). WMA 10.4 is situated on Warrenton Weir and therefore fits into scenario (a). The 2006 flooding event (Figure 2-14) may have floated the theoretical stable state "ball" higher in the cup from position 1, but could not overcome the resilience of the "cup" to directly transfer the "ball" into another position as the Weir is a lake ecosystem. This can be confirmed by water hyacinth dominance in 2007.

However the flooding had decreased water hyacinth cover from 2006, by depositing plants out of the water and washing them downstream. It was also responsible for the high nitrate levels of WMA 10.4 in 2006 (Figure 2-25), and an influx of water hyacinth washed down from upstream of the river. The high nutrient levels were used by water hyacinth and the new growth, in combination with the plants washed downstream, would have directly contributed to the high proportion of new population in relation to the previously established classes in 2007. Portions of the ecosystem were attempting to revert back to position 1 in 2007, but by 2008, the subsequent intensive integrated management of water hyacinth by WfW, and resultant unfavourable abiotic conditions would have cumulatively lifted the "ball" over the edge of the cup into position 2. With this continuing through 2008 until 2009, the "ball" landed in the Eurasian water-milfoil-dominated alternative stable state of position 3.

Hysteresis (a term derived from the field of physics), is the process whereby a community undergoes perturbation and begins to change states, however, there is not enough perturbation for the community to enter into an alternative stable state, and the community begins to revert back to its original state, but does not reach its original state and thus enters a temporary stable state (Beisner *et al.* 2003) (Figure 2-39). The temporary stable state is position 2, which is between the alternative stable states of 1 and 3 (Beisner *et al.* 2003). This new fragile state was classed as a 'state of flux' in this study and for a community to escape the 'state of flux', it was required to dominate the same habitat for 2 consecutive years, following which it was assumed that the dominant macrophyte would have outcompeted its competition, thereby entering into a new dominant state. In WMA 10.4, during the shift in dominance, 2007 and 2008 state of flux communities became established (Figure 2-24). The resultant increased water nitrate levels following the flooding event (2006) and water hyacinth washed down from upstream, resulted in the establishment of water hyacinth state of flux communities. It is hypothesized that this pattern of re-establishment may have been the historical method whereby water hyacinth had maintained dominance over the system.

The cycle of alternative stable states between floating and submerged aquatic plants can be maintained naturally. Scheffer *et al.* (2003) showed how floating plants are reliant on high water column nutrient levels. A floating plant-dominant alternative stable state requires nutrient enriched waters to maintain dominance and restrict light penetration of the water column to the submerged macrophyte community. Scheffer *et al.* (2003) also showed that should submerged aquatics become dominant, they have the ability to restrict floating plants growth rates with their superior ability to absorb the nutrients from the water column. Floating plants are naturally poor, regarding nutrient absorption, compared to submerged macrophytes (Portielje & Roijackers 1995). With sufficient light penetration, submerged aquatics can absorb nutrients from both the water column and sediment, thereby restricting floating plant dominance and establishing a submerged macrophyte alternate stable state (Barko & Smart 1980).

Similar studies have been conducted at different locations globally. Janse & Puijenbroek (1998) showed how eutrophication of the water in the drainage ditches in Holland resulted in a shift in dominance away from the indigenous submerged aquatic macrophytes to an alternative stable state, where the floating plants in the form of duckweed (species name not recorded) (Lemnaceae) dominated. Morris *et al.*(2003) investigated how eutrophication affected aquatic macrophyte species composition in the shallow lakes in Australia. Similarly they found that eutrophication and water level manipulation was causing a shift in alternative stable states away from the indigenous *Vallisneria americana* Michx. (Hydrocharitaceae) to the floating macrophyte, *Azolla pinnata* R.Br. (Azollaceae), which was not native to that region of Australia. Sharip *et al.* (2011) concluded that flooding events, water quality improvements and sediment characteristics were primarily responsible for the change in dominance away from the native *Nelumbo nucifera* Gaertn. (Nelumbonaceae) to an alternative stable state where the submerged macrophyte, *Cabomba furcata* Schult. and Schult.f. (Cabombaceae) dominated Lake Chini in Malaysia. In the light of these studies, it is evident that a shift in alternative stable states did occur in the Vaal River, from a water hyacinth-dominated state to an alternative stable state where Eurasian water-milfoil was dominant.

In WMA 10.4, the changes in abiotic conditions in 2007 resulted in the establishment of Eurasian water-milfoil-dominated communities in areas where water hyacinth had previously existed as stable dominate populations. The state of flux communities showed a steady presence in 2007 and 2008 (Figure 2-24). It appears as if Eurasian water-milfoil continued to assist the 2008 perturbations (integrated management of water hyacinth) by taking advantage of the abiotic conditions to best suited to its growth and expansion to escape its state of flux state. The pH values were already suitable for Eurasian water-milfoil (Grace & Wetzel 1978), being alkaline, of greater significance is the change in nitrate levels which, together with the herbicide influence on water hyacinth communities, caused enough additional perturbation for the change in alternative stable states to occur, from state of flux communities to submerged macrophyte-dominated alternative stable states by 2009. It is envisioned that even though there is a continued influx of water hyacinth from WMA 9, as long as there is an established Eurasian water-milfoil-dominated WMA10.4, and a constant integrated control programme, water hyacinth will not reach levels previously seen on WMA 10.4

Various authors have correlated water hyacinth and submerged macrophyte growth rates with abiotic variables, specifically water pH (Figure 2-18), nitrate (Figure 2-20) and phosphate levels (Figure 2-21). After obtaining the DWA RQS values and compiling the spatial data, correlations of these important variables were investigated to determine the effect of abiotic variables on macrophyte cover in the Vaal River, and whether they could be used as explanations for the shift between alternative stable states (Scheffer *et al.* 2003). Possible reasons for the lack of demonstrated correlation of water hyacinth dominance with pH and phosphate concentration levels of the Vaal River revolve around the cost of seasonal SPOT V images in South Africa. It is believed that if it were cost effective to obtain seasonal data, better correlation may have been revealed. The plant's response to changes in water quality would have been more accurately recorded, due to imagery being of that time period and not blended with images from the rest of the year. The water quality data itself could also have attributed to the lack of correlation as DWA has used various laboratories over the years to test their samples which leads to a host of variables (such as different techniques or equipment used) influencing the actual accuracy of the data. Another possible reason could be the large volume of anthropogenic influences that occur on the Vaal River. These include acid mine drainage, sewage outflows, and fertilizer runoff, all having localised and regional effects (Wepener *et al.* 2011), which could result in sporadic changes in macrophyte cover. These factors may have been accounted for by the monitoring point, but the delayed response by water hyacinth may not be visible on the SPOT V blended imagery. Lastly water hyacinth is a floating plant whose location is flexible and if it does not exist in large dense mats, it may not be able to take advantage of the changes in abiotic conditions without being blown away. However, despite all these variables influencing the correlation results, the strong ties between water hyacinth and nitrate concentration levels were still noted. It is this tie which is exploited by Eurasian water-milfoil to gain dominance over water hyacinth in WMA 10.4 and shift the ecosystem to a submerged macrophyte alternate stable state.

The other facet investigated was the if there were any differences within different population densities of Eurasian water-milfoil and outside the beds in the immediate vicinity in open water. Smith & Barko (1990) noted how dense Eurasian water-milfoil stands slowed down the flow rates of the water, which created a settling effect. Settling of solids clears the water column and although some change was noted between WMA 10 against WMA 9 and 8, nothing significant was noted when comparing the population densities with the TDS readings (Figure 2-26). WMA 10 was significantly different from the other regions in respect to pH. This could be related to the high cover of Eurasian water-milfoil in the region, as Eurasian water-milfoil which has the ability to change the water pH levels to suit its needs (Smith & Barko 1990). However there was no correlation between population density and TDS (Figure 2-27) or pH (Figure 2-29). This could be due to a mixing effect and the location of open sample not being taken far enough away from the Eurasian water-milfoil sample bed, as often the bed boundary is hard to determine.

The sediment abiotic conditions did not give any additional insight into why populations of various densities occupied particular regions. It is believed that at the time of the field trip, the Eurasian water-milfoil populations were simply selecting any available habitat as a result of the majority of the population being scoured out of the system during the flooding of 2010 (Figure 2-14). There did seem to be a trend towards establishing dense populations at particular locations with high total nitrogen concentrations and rare populations at other locations where there was less total nitrogen available, but there was no relationship observed between the population density and nitrogen levels. Variation between the different population densities was minimal, which also indicates that many of the less dense Eurasian water-milfoil populations still have the potential to grow as a result of suitable soils.

This study has shown how the study area is split up into different regions where particular macrophytes dominate and have created stable environments to best suit their requirements. WMA 8 is simply unsuitable for any form of sustained macrophyte growth, WMA 9 is not suitable for Eurasian water-milfoil dominance, and WMA 10.4 has changed dominance from a water hyacinth-dominated state to a Eurasian water-milfoil-dominated state. This with time, is creating greater resilience to maintain this state. It has been shown that the integrated control programme enforced by WfW is allowing Eurasian water-milfoil to thrive by minimising water hyacinth competition. Also, as a result of the high pH, water hyacinth relies on high water nutrient levels to maintain its competitive edge. It is envisaged that should nutrient level influxes into the Vaal River decrease, e.g. via the upgrading of existing sewage treatment works, or minimising fertiliser agricultural runoff, integrated control programmes implemented against water hyacinth will increase the extent of the Eurasian water-milfoil infestation to new regions of the Vaal River. Before this happens, suitable forms of control to combat this spread need to be investigated.

# CHAPTER 3 – Baseline macroinvertebrate survey

## 3.1 *Chapter overview*

There is a lack of published information regarding epiphytic and phytophagous aquatic macroinvertebrate communities associated with aquatic flora in South Africa. By surveying these insect assemblages associated with Eurasian water-milfoil in the Vaal River, estimates of richness, evenness and diversity of the insect assemblages were established. These data allow for insight regarding how the macroinvertebrate communities are limiting the spread of Eurasian water-milfoil and will also provide a baseline from which comparisons can be drawn of the impacts that any control measures implemented in the future would have against the weed can be drawn.

## 3.2 *Introduction*

Eurasian water-milfoil is a submerged aquatic macrophyte belonging to the Haloragaceae, a large diverse group of dicotyledonous plants, native to Eurasia and northern Africa (Couch & Nelson 1985). The mode of introduction is still unknown, but Eurasian water-milfoil was first recorded in South Africa in 1885, near Barkly West on the Vaal River (28°32’45”S 24°30’50”E). Although there are establishment records from a few other locations in South Africa, the only other confirmed record, following field surveys, is in Lake Sibaya in northern KZN (27°24’37”S 32°42’20”E) (Coetzee *et al.* 2011a) (Figure 3-1). Eurasian water-milfoil is also an established problem plant in Australia, India, the U.S.A. and Canada (Holm *et al.* 1979, Couch & Nelson 1985).

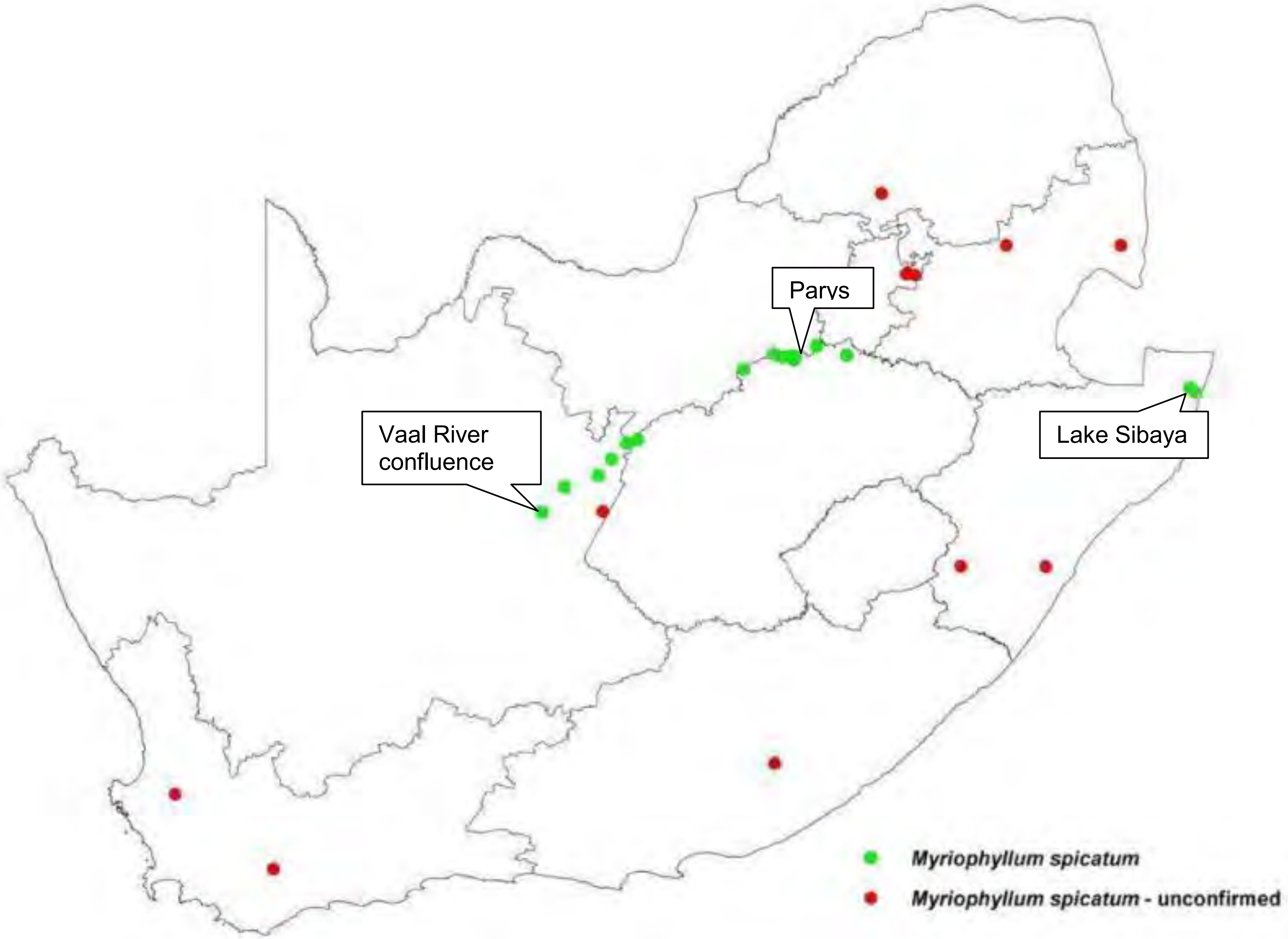



**Figure 3-1:** The distribution of Eurasian water-milfoil infestations in South Africa (SAPIA Database, ARC – PPRI)

### 3.2.1 History of biological control of Eurasian water-milfoil in the U.S.A.

Eurasian water-milfoil is classed as the most important waterweed in the U.S.A. (Johnson & Blossey 1997) and this led to the implementation of a biological control programme against the weed (Cock *et al.* 2008a). The first biological control agent released against Eurasian water-milfoil (and other nuisance submerged aquatic macrophytes) was grass carp *Ctenopharyngodon idella* (Cuvier and Valenciennes) in 1963 (Julien & Griffiths 1998). Following the grass carp introduction and its distaste for Eurasian water-milfoil, and given the nature and extent of the damage caused by Eurasian water-milfoil in the U.S.A., research focused on finding a pathogen or insect biological control agent (Buckingham *et al.* 1981, Creed *et al.* 1992, Sheldon & Creed 1993, Shearer 1994, Creed & Sheldon 1995, Cofrancesco 1998, Creed 1998, Johnson *et al.* 1998, Mazzei *et al.* 1999, Gross *et al.* 2001). In 1965, a classical biocontrol programme was initiated against Eurasian water-milfoil in the U.S.A. (Cock *et al.* 2008a), but no information about suitable agents from its native range in Eurasia was available and this resulted in the initiation of several baseline surveys to identify suitable macroinvertebrate biological control agents.

### 3.2.2 Macroinvertebrate baseline surveys conducted on Eurasian water-milfoil populations

Baseline surveys conducted in the U.S.A. found that several insect species were already feeding on and damaging Eurasian water-milfoil (Batra 1977, Buckingham & Bennett. 1981, MacRae *et al.* 1990, Sheldon & Creed 1993). Several of these species had European origins, while some native species appeared to have switched preference away from their indigenous hosts to Eurasian water-milfoil (Buckingham *et al.* 1981). Intensive surveys were then conducted within the native range of Eurasian water-milfoil for additional biological control agents. These revealed that in total, 44 phytophagous insects feed on Eurasian water-milfoil. This list was compiled by CABI Europe and comprised surveys from the plant's native range of Eurasia (Pakistan and Bangladesh surveyed by Ghani *et al.* (1970), Yugoslavia surveyed by Spencer & Lekic (1974), and Peoples Republic of China, Japan and Korea surveyed by Buckingham (1998)). No surveys have been conducted on Eurasian water-milfoil in Northern Africa, where it is also native. The most suitable agents out of the 44 listed were *Bagous longitarsis* Thomson (Coleoptera: Curculionidae), with a Palearctic distribution (Dieckmann 1983, Sprick 2000); *Bagous collignensis* (Herbst) (Coleoptera: Curculionidae), distributed in Europe and Western Asia (Sprick 2000); *Bagous geniculatus* Hochhuth (Coleoptera: Curculionidae), found in Pakistan, Bangladesh, Southern Europe, Central Asia and the Caucasus (Ghani *et al.* 1970); *Bagous vicinus* Hustache (Coleoptera: Curculionidae) located in Pakistan and Bangladesh (Ghani *et al.* 1970); *Eubrychius velutus* Beck (Coleoptera: Curculionidae), distributed through Europe and Northern Asia (Dieckmann 1972), *Phytobius* spp. (Coleoptera: Curculionidae) found in Europe and China (Ghani *et al.* 1970, Buckingham 1998); and *Aristotelia* sp. *subdecurtella* (Stainton) (Lepidoptera: Gelechiidae), found in Pakistan and Bangladesh (Ghani *et al.* 1970). Eight of the 44 insect species recorded feeding on Eurasian water-milfoil were also found in the U.S.A., where they had switched from feeding on their original hosts onto Eurasian water-milfoil plants (Creed & Sheldon 1995).

Among the potential biological control agents found feeding on and damaging Eurasian water-milfoil in the U.S.A., were the weevil *Litodactylus leucogaster* (Marsham) (Coleoptera: Curculionidae), a native North American species with Holarctic distribution (Buckingham & Bennett. 1981); the aquatic midge *Cricotopus myriophylli* Oliver (Diptera: Chironomidae) (MacRae *et al.* 1990); also Holarctic distribution, but with taxonomic problems (MacRae *et al.* 1990); the introduced pyralid moth, *Acentria ephemerella* Denis and Schiffermüller (syn.: *Acentria nivea* Olivier) (Lepidoptera: Crambidae); and the native milfoil weevil *Euhrychiopsis lecontei* Dietz (Coleoptera: Curculionidae). In the U.S.A., of all of the agents evaluated, the agents accredited with the declines in Eurasian water-milfoil populations were the pyralid moth *A. ephemerella* and the milfoil weevil *E. lecontei* (Cock *et al.* 2008a).

Currently, Eurasian water-milfoil is under control in certain states of the U.S.A. through the initiation of an integrated control programme where various herbicides are used in conjunction with the weevil and moth biological control agents to control populations (Jacobs & Mangold 2009). To date, there is no form of control measure implemented against Eurasian water-milfoil infestations in South Africa, however, the milfoil weevil *E. lecontei* is currently in quarantine undergoing host specificity testing (Coetzee *et al.* 2011a). There is also a pilot programme involving the use of diquat herbicide against Eurasian water-milfoil (D. Sharpe, WfW, Pers. comm., 2011), to form part of an integrated control programme similar to North America.

### 3.2.3 Macroinvertebrate baseline surveys conducted in South Africa

Surveys conducted to determine which macroinvertebrates feed on invasive aquatic macrophytes are rare in South Africa. The only other surveys conducted were those by Hill (1998) on *Azolla filiculoides* Lam. (Azollaceae) (red water fern) and Schutz (2007) and Baars *et al.* (2010) on various *Lagarosiphon* sp. The importance of such studies in South Africa, related to understanding the community ecology is undervalued. As discussed by Simberloff (2004), community ecology plays an essential role in deciphering the relationships between species on various spatial and temporal scales.

All macroinvertebrate surveys conducted in South Africa thus far have had biological control motivation associated with them. A baseline macroinvertebrate survey on an alien macrophyte plays an important role in biological control agent selection as intraspecific macroinvertebrate competition could influence which biological control agent is used. This is particularly important regarding possible reasons why control agents battle to establish (McClay & Balciunas 2005). These surveys also provide an opportunity to investigate the probability of augmentative biological control options, and give insight into how long the macrophyte has persisted in the system, with higher species richness and abundances generally indicating that it has been present in the system for a longer period of time (Gassmann *et al.* 2006). Finally, baseline surveys give an indication of the plant's competitive ability. If more indigenous insects are associated with it and it still dominates the indigenous flora, there is a greater need for effective control measures to be implemented against the plant, as it has the potential to be a larger threat in another ecosystem where there are fewer macroinvertebrates (Gopal & Goel 1993). Besides these examples, no other studies have simultaneously investigated the phytophagous aquatic invertebrate communities associated with submerged macrophytes and their associated environmental variables (F. de Moor, Freshwater Invertebrates Department of the Albany Museum, Pers. comm., 2011), and this is the first study conducted on Eurasian water-milfoil invertebrate communities in South Africa.

### 3.2.4 Effects of water quality parameters on the composition of macroinvertebrate communities

Water quality is deteriorating rapidly on the Vaal River, which is rated as one of the most highly polluted systems in South Africa (de Villiers & Thiart 2007). This is expected to influence macroinvertebrate species richness and abundance. Dallas & Day (2004) reported how polluted sites are characterised by lower species richness but higher abundances of insects. This observation has been used in the South African Scoring System (SASS), used to monitor South African water resources in terms of environmental water quality (Chutter 1998, Dickens & Graham 1998). Chironomidae in particular are used as accurate indicators of water quality as they are globally abundant and have pollution sensitive and tolerant taxa

(Adriaenssens *et al.* 2004, Wright & Burgin 2009, Luoto 2010). Thus their presence and abundance could provide insight into the water quality of the Vaal River. In general, it is surmised that the greater the species richness and abundance of macroinvertebrates found on Eurasian water-milfoil, the greater the possibility that the plant's competitive ability is being compromised.

Thus, the aim of this chapter was to establish a list of macroinvertebrates that use Eurasian water-milfoil as habitat, to study the relationship between these species and the density of the Eurasian water-milfoil beds, and to compare these data to similar surveys conducted in Eurasia and North America. The findings will provide insight into the ecological relationship between Eurasian water-milfoil and the aquatic macroinvertebrates found in the Vaal River. The abundance and species richness of these communities will also give insight into the impact these assemblages may have on the spread of Eurasian water-milfoil in the system the higher these values are the greater the possibility is that they are having an negative impact on Eurasian water-milfoil growth rates. Lastly, the survey will establish a baseline of macroinvertebrate communities associated with Eurasian water-milfoil from which future studies investigating other Eurasian water-milfoil, infestations in South Africa can be compared.

## 3.3 *Materials and Methods*

The study area extended from the Vaal River Barrage to the confluence between the Vaal and the Orange Rivers (Figure 1-2). Results from Chapter 2 showed that Eurasian water-milfoil dominates the lower portions of the Vaal River, thus the majority of the macroinvertebrate sampling sites were located in WMA 10, between Warrenton Weir and Douglas Weir (Figure 3-2).

Throughout the study area (Figure 3-2), 1kg wet mass samples of Eurasian water-milfoil were removed, by hand with a rake from the dense surface foliage, from 21 sites of similar-size populations (20m in diameter) that were classified as dense, abundant, frequent, occasional or rare (Chapter 2). Sample points were randomly selected on a daily basis and water quality samples were taken from the sites and physical/chemical properties were analysed as per Chapter 2.

Comparisons between the abundance and richness of insects associated with Eurasian water-milfoil, plant density and water physio-chemical properties, were performed to determine if the macroinvertebrates intentionally seek out Eurasian water-milfoil based on plant density or if they are simply using the macrophyte as available complex habitat, independent of plant density (Gaufin *et al.* 1956, Cyr & Downing 1988). All insects and insect larvae were removed from every strand of Eurasian water-milfoil by hand in the field and stored in 70% ethanol for later identification at the Freshwater Aquatic Invertebrate Department of the Albany Museum in Grahamstown.

Using a dissecting microscope and fine forceps, all preserved insects were identified to morphospecies, using the Water Research Commission Guides to Freshwater Invertebrates (Day & de Moor 2007, Day *et al.* 2007a, 2007b, de Moor *et al.* 2007), at the Freshwater Invertebrate Department of the Albany Museum in Grahamstown.

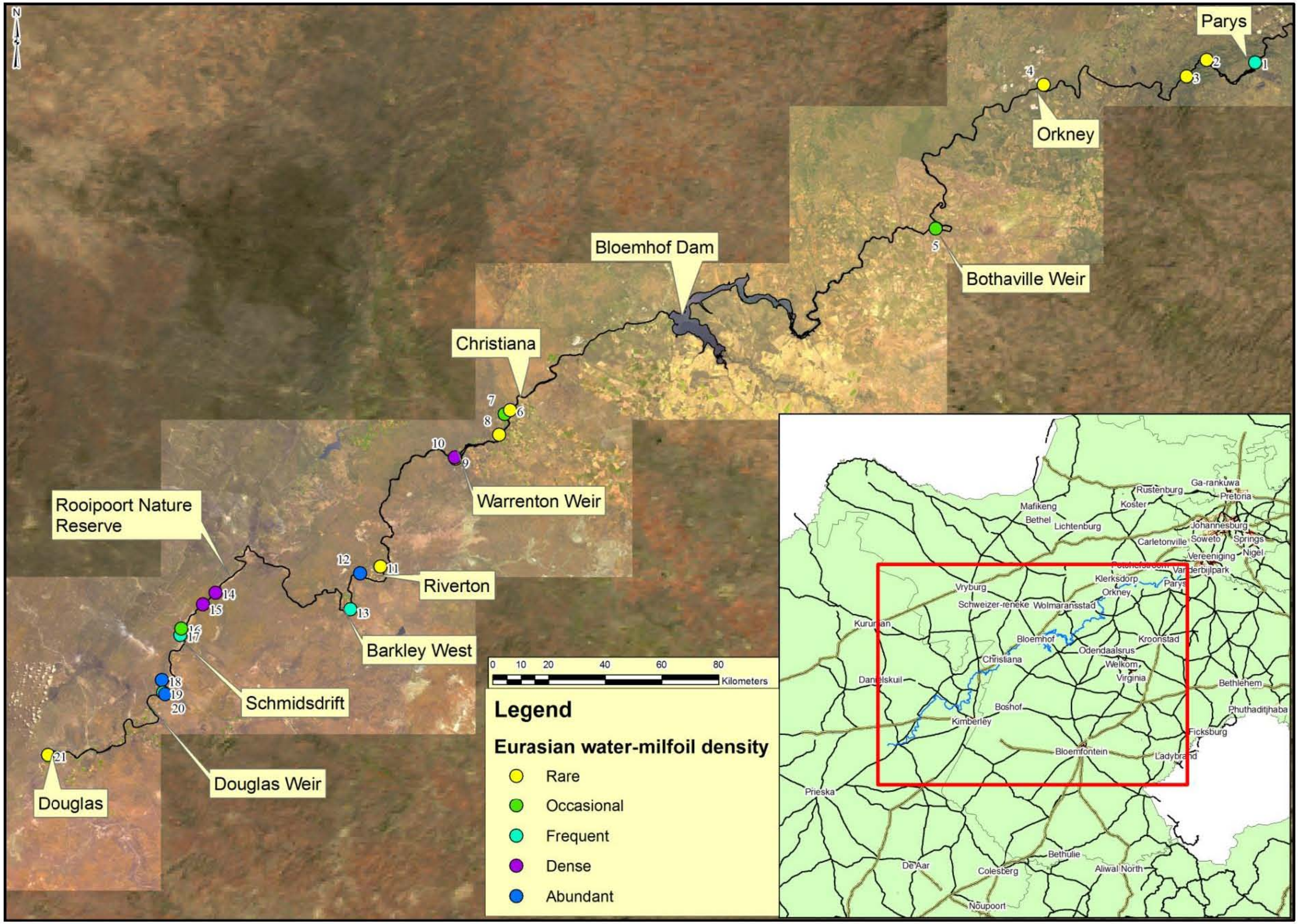



**Figure 3-2:** The study area, showing the macroinvertebrate sampling sites, correlated with plant density, extending down the study area from the Vaal River Barrage (26° 45' 54" S 027° 41' 02" E) to the confluence at Douglas (29° 04' 15" S; 23° 38' 09" E).

Due to the abundance of Chironomidae found on the plants, it was decided that the level of identification provided for by the Water Research Commission Guides to Freshwater Invertebrates was not sufficient for Chironomidae identification. To accurately identify chironomid larvae (Diptera: Chironomidae) to genus, the head capsules, mouth parts and other structures had to be visible so that taxonomic keys developed by Wiederholm (1983), Cranston (1996) and Harrison (2002) could be used. Following initial identification, Lucid Player Standard v2.2 software using Cranston's Chironomid key was used to complete the identification process. Chironomids were stored in 70% ethanol to prevent head capsules from drying out, as shrunken head capsules are difficult to mount correctly (Dickman & Rygiel 1996).

To effectively mount the head capsule of a chironomid larva, the procedures developed by Ochieng *et al.* (2008) were followed. Cold KOH was used instead of warm KOH as used by other authors (Warwick 1988) as it produced better clearing results, in terms of the removal of pigment from the head capsule. The better the clearing results, the more accurate the identification procedure. Larvae were removed from the 70% ethanol solution and cleared in 10% cold (weight: volume) KOH solution for about 15- 20 minutes (Warwick 1988). Following the clearing, the larvae were washed three times in 96% ethanol and finally in absolute ethanol until the head capsule was clear. Euparal was used as the mounting medium for the slides and the entire larva was transferred onto a slide in which dissection and removal of the head occurred, using a dissecting microscope and pins. The head capsule was arranged ventral side up, and the entire remaining body segment was removed and mounted separately. A cover slip was then placed on the slides and gently pressed down while applying a small rotation to flatten the head capsule in order to expose the mouth parts (Warwick 1988). Slides were left to dry for 5 – 8 days before the identification procedure could take place.

### 3.3.1 Statistics

Following identification of all macroinvertebrates to morphospecies, the data were used to construct a species accumulation curve. To determine if the study had sampled a sufficient number of invertebrate families, EstimateS version 8.2 (Colwell 2006) was used. The species accumulation curve included the analytically calculated observed number of species (Sobs (Moa Tau)) to establish sampling representivity. The Michaelis-Menten Mean (MMMean) estimator (Toti *et al.* 2000) and incidence-based coverage estimator (ICE) (Chazdon *et al.* 1998) were used to determine if the sample size was adequate. MMMean is highly sensitive at small sample sizes and has been shown to produce the most accurate estimations for large sample sizes (Toti *et al.* 2000). ICE has its highest accuracy at smaller sample sizes (Chazdon *et al.* 1998) and therefore sample size was considered representative when the MMMean and ICE estimators converged at the highest richness (Gotelli & Colwell 2001, Longino *et al.* 2002).

EstimateS, version 8.2 was also used to determine of degree similarity or dissimilarity of the macroinvertebrate communities inhabiting varying densities of Eurasian water-milfoil beds, in terms of taxonomic homogenisation. The Morisita-Horn similarity measure was used to quantify the species data as the sampling effort was not equal among all of the communities and was in the form of relative abundance (Wolda 1981).

To visually illustrate whether the dissimilarity differences were significant according to either the species richness or the species abundance, Kruskal Wallis by Ranks and Median Test (a non-parametric ANOVA) were used, as the data were not normally distributed. This is a robust test as it ranks and uses all of the data rather than generating a mean, reducing variation. The H value (reported in these results) is an indication of the data variability (Rosner 2000). A non-metric multidimensional scaling (MDS) based on the Morisita-Horn test, using Community Analysis Package 4 v4.1.3 (2007), was compiled to illustrate the spatial differences in macroinvertebrate assemblages between sites of different plant densities and location, while a Principal Component Analysis (PCA) covariance plot was compiled to determine which sites, grouped according to plant densities, were more similar to each other based on the physio-chemical conditions of the location. Both ordinations used two dimensions to represent similarities between the sites. The closer the points were to each other the more similar the sample sites were. The reverse is also applicable (Clarke & Warwick 2001). The MDS plot was selected as it has the advantage of simplifying complex data based on a user-selected appropriate measure of similarity, in this case based on the Morisita-Horn test (Clarke & Warwick 2001). The PCA covariance plot was selected to separate the sites based on physio-chemical conditions as it is also a simple method well suited for environmental variables and further suitable as it assigns each variable an equal weighting, so that different scales of measurement do no impact the final results of the PCA ordination (Clarke & Warwick 2001).

A comparison of the biodiversity indices was then performed to determine how significantly different these communities were from other similar surveys conducted in the U.S.A. (Wilson & Ricciardi 2009) and Eurasia (Cock *et al.* 2008a). The evenness indexes are a measure of the degree of evenness in species abundance. Lower values indicate the extent by which a community is dominated by a single species and high numbers indicating that an ecosystem consists of and high values are a measure of greater species diversity. The ratios of maximum density to observed density can be used in this regard, where $J' = H' / H\ max = H'/1nS$ (Pielous evenness index) (Magurran 2004).The other index used was the Shannon-Wiener index (H'). This index assumes that there is an infinitely large community of individuals, from which individuals were randomly sampled and all species present were represented in the sample (Magurran 2004). The Shannon-Wiener index is calculated by $H' = - \sum p_i 1n p_i$. This index however lacks an unbiased estimator and errors in data occur when not all of the species in the community are sampled (Magurran 2004).

## 3.4 *Results*

Following insect identification, a list of the macroinvertebrate species found was compiled per site (Table 3-1). The most dominant species were the Chironomid *Dicrotentipes notatus* Meigen (Diptera: Chironomidae) (77% of all macroinvertebrates) and *Physa acuta* Draparnaud, (Gastropoda: Physidae), an invasive alien snail (10% of all macroinvertebrates). Macroinvertebrates were found at all the sites except 2, 4 and 21 (Figure 3-2). The greatest number of species was found at sites 1, 3, 8, 10, 14 and 20, each having 5 species per site. Herbivores were the most abundant feeding group (92.51%), while predators (6.46%) and others (detritivores, parasitoids etc.) (1.03%) were not well represented. The Sobs rarefaction curve begins to level off, indicating that the sampling protocol was efficient and almost all the insect species that are expected to be associated with the plants at the time of sampling were collected. However, the Sobs curve did not converge with the ICE and MMMean curves, suggesting that additional sampling is required to get full representivity of the macroinvertebrates associated with Eurasian water-milfoil in the Vaal River (Figure 3-3).

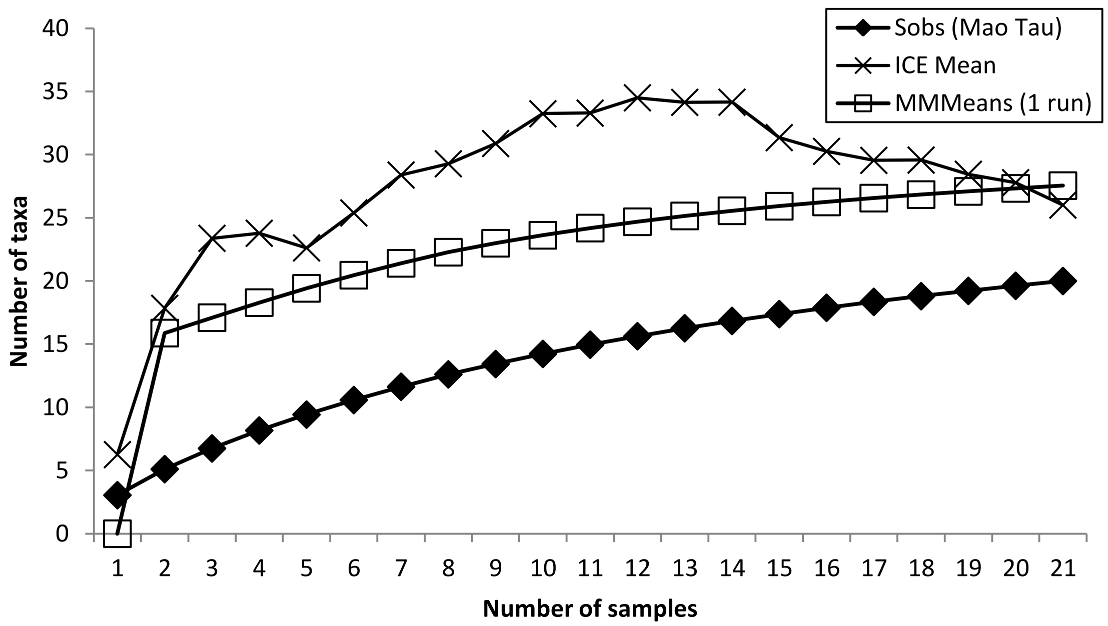



**Figure 3-3:** Species accumulation curve indicating observed number of species (Sobs Mao Tau), incidence-based coverage estimator (ICE) and Michaelis-Menten Mean (MMMean) richness estimators. Because the curves do not converge, additional sampling is required.

**Table 3-1:** The dominant macroinvertebrate species (herbivores, predators and others) found in Eurasian water-milfoil beds of different densities throughout the study area.

| Site # | Location | Eurasian water-milfoil density | Family | Species | Herbivores/ Predators/ Other | Number of Species | Number of Individuals |
|---|---|---|---|---|---|---|---|
| 1 | Parys Weir #1 | Frequent | Physidae | *Physa acuta* | Herbivore | 5 | 15 |
| | | | Chironomidae | *Dicrotentipes notatus* | Herbivore | | 7 |
| | | | Chironomidae | *Kieffurlus* sp. | Herbivore | | 3 |
| | | | Caenidae | *Caenis* sp. | Herbivore | | 1 |
| | | | Leptoceridae | *Oecetes* sp. | Herbivore | | 1 |
| 3 | Parys Vaal River #3 | Rare | Physidae | *Physa acuta* | Herbivore | 5 | 25 |
| | | | Chironomidae | *Dicrotentipes notatus* | Herbivore | | 2 |
| | | | Anisoptera | *Anax imperata* | Predator | | 1 |
| | | | Aphelocheindae | *Aphelocheirus* sp | Predator | | 1 |
| | | | Elmidae | Elmidae larvae | Herbivore | | 1 |
| 5 | Bothaville Weir #5 | Occasional | Chironomidae | *Dicrotentipes notatus* | Herbivore | 4 | 240 |
| | | | Chironomidae | Chironomid pupae | Herbivore | | 5 |
| | | | Oligochaeta | Aquatic worm sp. | Other | | 1 |
| | | | Corixidae | *Sigara* sp. | Predator | | 1 |
| 6 | Warrenton Weir #6 | Rare | Chironomidae | *Dicrotentipes notatus* | Herbivore | 2 | 1 |
| | | | Physidae | *Physa acuta* | Herbivore | | 1 |
| 7 | Warrenton Weir #7 | Occasional | Chironomidae | *Dicrotentipes notatus* | Herbivore | 3 | 26 |
| | | | Chironomidae | *Paralauterporniella* sp. *subcinela* | Herbivore | | 1 |
| | | | Chironomidae | *Cyphomella* sp. | Herbivore | | 1 |
| 8 | Warrenton Weir #8 | Rare | Chironomidae | *Dicrotentipes notatus* | Herbivore | 5 | 14 |
| | | | Physidae | *Physa acuta* | Herbivore | | 4 |
| | | | Chironomidae | *Kieffurlus* sp. | Herbivore | | 3 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | | Oligochaeta | Aquatic worm sp. | Other | | 1 |
| | | | Hydroplilicae | *Orthotrichia* sp. | Herbivore | | 1 |
| 9 | Warrenton Weir #9 | Rare | Chironomidae | *Dicrotentipes notatus* | Herbivore | 3 | 14 |
| | | | Chironomidae | *Cyphomella* sp. | Herbivore | | 9 |
| | | | Chironomidae | *Kieffurlus* sp. | Herbivore | | 2 |
| 10 | Warrenton Weir #10 | Dense | Chironomidae | *Dicrotentipes notatus* | Herbivore | 5 | 33 |
| | | | Physidae | *Physa acuta* | Herbivore | | 2 |
| | | | Ceretopogonidae | *Bezzia* sp. | Predator | | 2 |
| | | | Chironomidae | *Paralauterporniella* sp. *subcinela* | Herbivore | | 2 |
| | | | Chironomidae | *Cricotopus* sp. *sylvestris* | Herbivore | | 1 |
| | | | Glossinphonidae | Leech sp. | Other | | 1 |
| 11 | Riverton Vaal River #11 | Rare | Baetidae | *Cheleocloeon excisum* | Predator | 2 | 2 |
| | | | Caenidae | *Caenis* sp. | Herbivore | | 1 |
| 12 | Riverton Vaal River #12 | Abundant | Baetidae | *Cheleocloeon excisum* | Predator | 4 | 5 |
| | | | Caenidae | *Caenis* sp. | Herbivore | | 3 |
| | | | Physidae | *Physa acuta* | Herbivore | | 1 |
| | | | Oligochaeta | Aquatic worm sp. | Other | | 1 |
| 13 | Barkley West Vaal River #13 | Frequent | Physidae | *Physa acuta* | Herbivore | 3 | 5 |
| | | | Chironomidae | *Dicrotentipes notatus* | Herbivore | | 3 |
| | | | Chironomidae | *Kieffurlus* sp. | Herbivore | | 2 |
| 14 | Rooipoort Nature Reserve Vaal River #14 | Dense | Chironomidae | *Tanytarsus* sp. | Herbivore | 5 | 3 |
| | | | Chironomidae | *Dicrotentipes notatus* | Herbivore | | 2 |
| | | | Physidae | *Physa acuta* | Herbivore | | 1 |
| | | | Anisoptera | *Anax imperata* | Predator | | 1 |
| | | | Stratiomyrdae | *Stratiomyidae* sp. | Herbivore | | 1 |
| 15 | Rooipoort Nature Reserve Vaal River #15 | Dense | Chironomidae | *Dicrotentipes notatus* | Herbivore | 3 | 15 |
| | | | Physidae | *Physa acuta* | Herbivore | | 2 |
| | | | Oligochaeta | Aquatic worm sp. | Other | | 1 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 16 | Schmidsdrift Vaal River #16 | Occasional | Chironomidae<br>Physidae | *Dicrotentipes notatus*<br>*Physa acuta* | Herbivore<br>Herbivore | 2 | 8<br>1 |
| 17 | Schmidsdrift Vaal River # 17 | Frequent | Chironomidae<br>Physidae<br>Anisoptera | *Dicrotentipes notatus*<br>*Physa acuta*<br>*Anax imperata* | Herbivore<br>Herbivore<br>Predator | 3 | 12<br>4<br>1 |
| 18 | Douglas Weir #18 | Abundant | Chironomidae<br>Aphelocheindae | *Dicrotentipes notatus*<br>*Aphelocheirus* sp | Herbivore<br>Predator | 2 | 42<br>1 |
| 19 | Douglas Weir #19 | Occasional | Chironomidae<br>Baetidae | *Dicrotentipes notatus*<br>*Cheleocloeon excisum* | Herbivore<br>Predator | 2 | 38<br>1 |
| 20 | Douglas Weir #20 | Abundant | Aphelocheindae<br>Chironomidae<br>Chironomidae<br>Baetidae<br>Leptoceridae | *Aphelocheirus* sp<br>*Dicrotentipes notatus*<br>Chironomid pupae<br>*Cheleocloeon excisum*<br>*Oecetes* sp. | Predator<br>Herbivore<br>Herbivore<br>Predator<br>Herbivore | 5 | 8<br>7<br>6<br>1<br>1 |

The Morisita-Horn similarity measures were used to determine the similarity of species assemblages at different plant densities (Table 3-2). Assemblages associated with the dense and occasional Eurasian water-milfoil density classes were the most dissimilar to one another, in the number of species that they support and their relative abundances (approximately 15% similarity). In contrast, the communities found in rare and abundant beds were the most similar (approximately 60% similar) to each other.

**Table 3-2:** A similarity matrix comparing the insect communities collected off different Eurasian water-milfoil bed densities.

| Morisita-Horn | Rare | Occasional | Frequent | Abundant | Dense |
|---|---|---|---|---|---|
| **Rare** | | 49.91 | 41.52 | 59.62 | 41.52 |
| **Occasional** | 49.91 | | 39.53 | 49.46 | 15.64 |
| **Frequent** | 41.52 | 39.53 | | 58.06 | 40.69 |
| **Abundant** | 59.62 | 49.46 | 58.06 | | 58.27 |
| **Dense** | 41.52 | 15.64 | 40.69 | 58.27 | |

Following species identification, comparisons were made of the numbers of species associated with the various population densities of the Eurasian water-milfoil populations (Figure 3-4). There was no significant difference between the macroinvertebrate species abundance and the Eurasian water-milfoil population densities ($H_4$ = 4.84, $P$ = 0.30). Similarly, there was no significant difference between macroinvertebrate species richness and the Eurasian water-milfoil population densities ($H_4$ = 5.35, $P$ = 0.25). In line with the Morisita-Horn similarity test, the dense and occasional beds had similar numbers of individuals found on them, while rare and occasional differed from each other, although not significantly. Similar comparisons can be draw between Table 3-2 and Figure 3-4 for all the different plant densities.

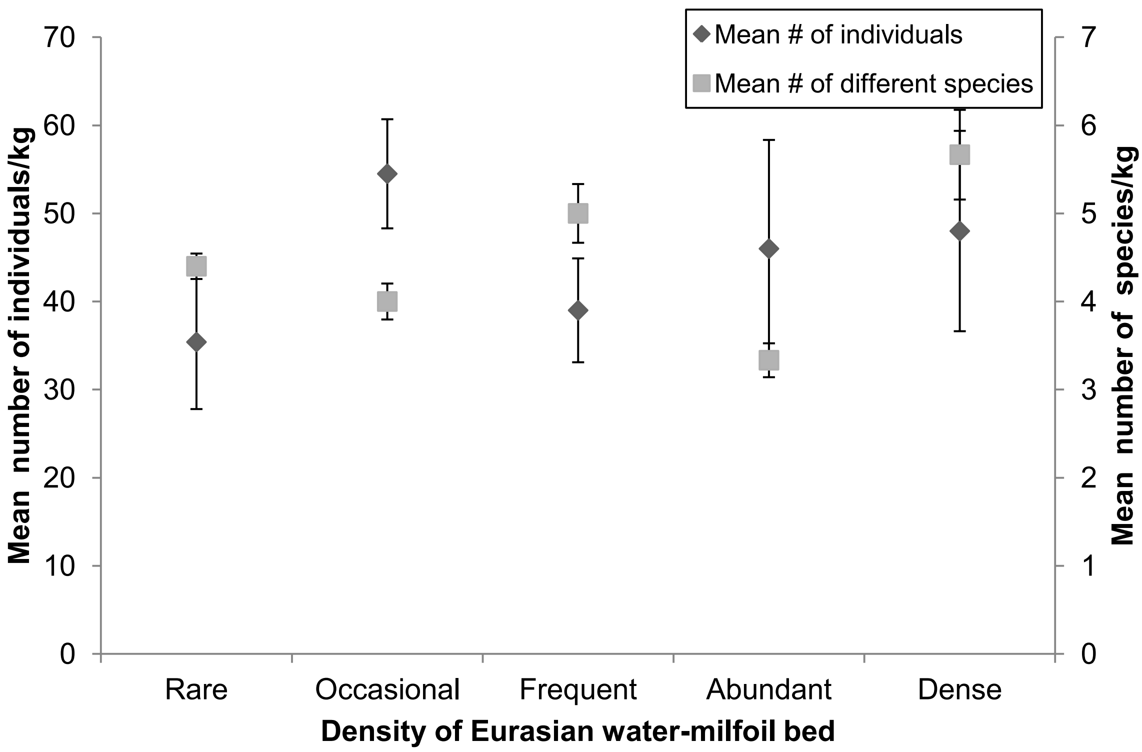



**Figure 3-4:** Relationship between plant density classes and macroinvertebrate abundance and species number. Error bars represent standard error of the mean.

A MDS plot was then compiled to visually compare similarities of the sample sites in terms of macroinvertebrate species compositions, also classed according to different Eurasian water-milfoil densities (Figure 3-5). This plot is a visual illustration of a combination of the trends shown in the Morisita-Horn similarity measures. There was large variation and poor clustering of the rare, occasional and abundant density bed data, in terms of macroinvertebrate assemblages, with extensive overlaps occurring. This plot did however show the dissimilarity between occasional and dense beds, which grouped relatively far apart from each other, while similarity between the species assemblages of differently classed beds is best illustrated by the large overlaps of the groupings. The best clusterings were noted with the frequent and dense classed beds, showing that similar species assemblages were found at each of those densities, their sites grouped closely together. The rare, occasional and abundant beds had large variation, indicating that the species found on these bed densities were not attracted to Eurasian water-milfoil based on the plant density of the bed. This is also confirmed by the close groupings of sites such as 6, 15 and 16, and 1, 8 and 13, which showed that those macroinvertebrate species assemblages did not differ according to the sample site plant densities, or location on the river (Figure 3-2). The location of the site on the river does appear to play a role with relatively close sites, such as 6, 7 and 8; 14, 15, 16 and 17; and 18, 19 and 20; occurring closer together in terms of species assemblages found on the plants; hence analysis of the physio-chemical conditions of each site was conducted.

Next, physio-chemical variables of the water column used as vectors for a PCA analysis of the Eurasian water-milfoil population densities. A PCA covariance plot was performed to visually compare how similar the sites were in terms of the physio-chemical variables of the water column (Figure 3-6). This was done to determine if the species assemblages at the sample locations were associated with similar physio-chemical conditions. These data showed poor grouping (Figure 3-6) according to Total Dissolved Solids TDS, pH, oxygen reduction potential (ORP), salinity, conductivity (µS/cm), resistivity (kOhm.cm), nitrate or phosphate concentration vectors. This is an indication of a lack of correlation found in chapter 2 between plant density and physio-chemical characteristics of the water column. Of all the abiotic variables, phosphate concentrations, TDS, ORP and conductivity were the main factors influencing how similar the population densities were. Eurasian water-milfoil sample sites (6-8, 10-12, 16 and 20) were influenced according to salinity, resistivity, pH, conductivity and nitrate concentrations however, compared to the results of Figure 3-5, these sites were not similar in terms of species assemblages, and thus the data did not compare well. When comparing these plots, it is evident that the measured physio-chemical characteristics of the water column do not influence the macroinvertebrate assemblages found in the plant beds.

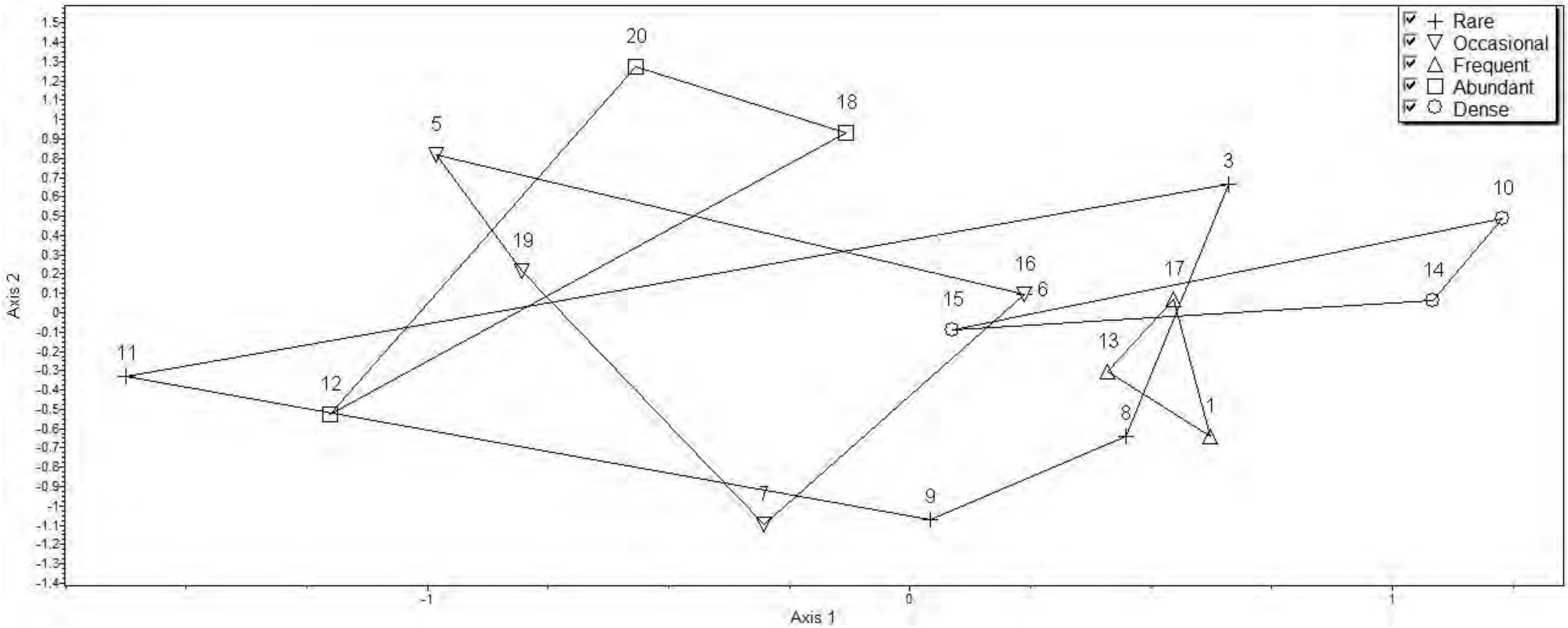



**Figure 3-5:** MDS plot illustrating similarities between the sample locations based on the macroinvertebrate community assemblages of those sites (stress =0.24).

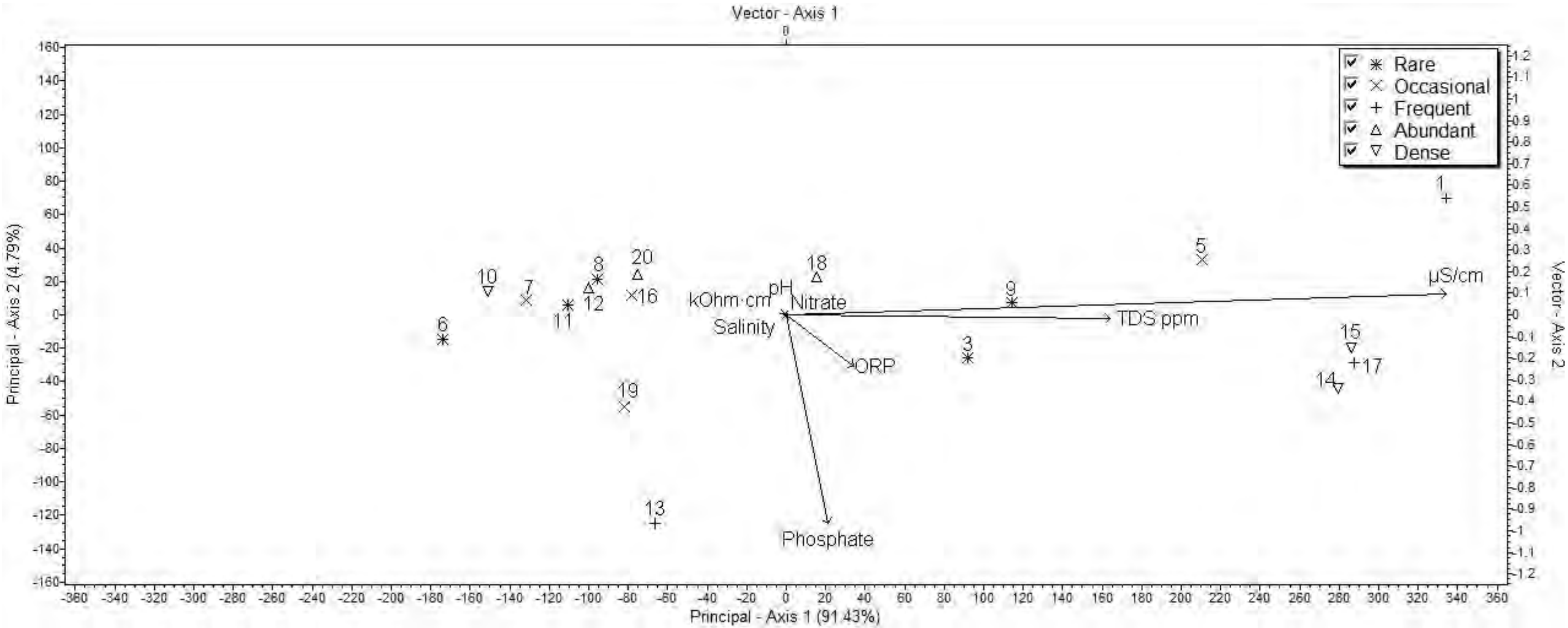



**Figure 3-6:** PCA covariance plot illustrating similarity between the sample locations and the physio-chemical conditions of the sample sites (PC 1 Eigenvalue 33231.9, %Validation 91.43%).

### 3.4.1 Comparison with similar surveys

Macroinvertebrate surveys on Eurasian water-milfoil populations have been previously conducted in its native range (excluding Africa) (Cock *et al.* 2008a) and in the U.S.A. (Wilson & Ricciardi 2009). A list of the herbivorous families found is illustrated in Figure 3-7. Ten herbivorous families and 44 species were identified in the Eurasian survey, 6 families and 13 species found in the North American Survey and 4 families and 10 species identified on Eurasian water-milfoil in the Vaal River of South Africa.

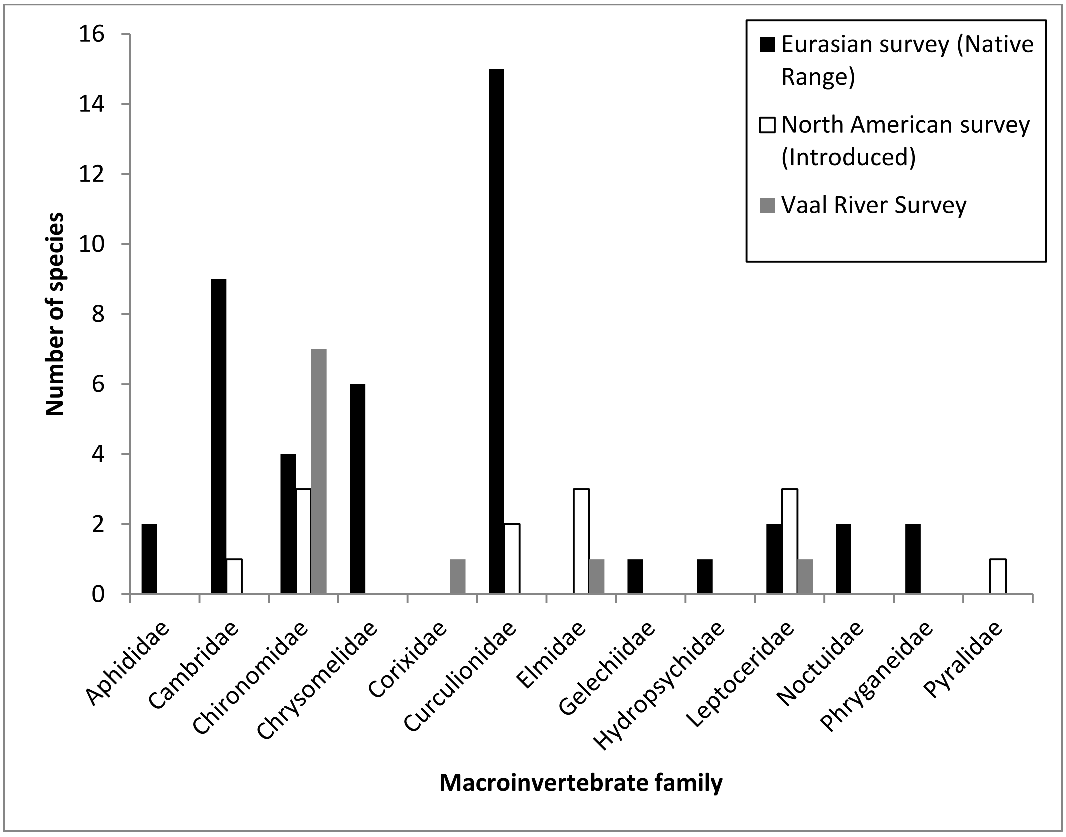



**Figure 3-7:** A summary of the number of herbivorous species found per macroinvertebrates family found on Eurasian water-milfoil in surveys in Eurasia, North America and in the Vaal River of South Africa

A comparison of biodiversity indices was then performed to determine whether these data were significantly different. The Pielous' evenness statistic (J') shows that the variety of species found in the introduced ranges of the U.S.A. and the Vaal River were less similar to each other, compared to the native range Eurasian survey data (Table 3-3). This was not expected and is an indication of the highly polluted status of Vaal River, which skewed these data.

The diversity of species found in the U.S.A. survey was more similar to the Vaal River survey, than to the Eurasian survey data (Table 3-3). The Shannon–Wiener diversity index (H’ (loge)) indicates that overall, the Eurasian and North American surveys had a greater diversity of macroinvertebrates associated with Eurasian water-milfoil, than the Vaal River survey. The limitation of this result however is that the macroinvertebrate species of the Vaal River were under-sampled (Figure 3-3), limiting the accuracy of this Shannon–Wiener diversity index. Even though the Shannon–Wiener index is compromised, it is surmised that there are still substantially fewer insects, both in terms of species richness and abundance that are using Eurasian water-milfoil in the Vaal River, compared to those in the U.S.A. and Eurasia. The lack of substantial numbers of herbivorous macroinvertebrates on Eurasian water-milfoil in the Vaal River means that its competitive ability as a submerged macrophyte is not as compromised and this could have contributed to its growth (Chapter 2).

**Table 3-3:** Biodiversity index comparison between surveys of macroinvertebrates associated with populations of Eurasian water-milfoil in Eurasia (native range), North America and South Africa (introduced ranges).

| Survey | S | N | J' | H'(loge) |
|---|---|---|---|---|
| Eurasian | 10 | 44 | 0.83 | 1.92 |
| North American | 6 | 13 | 0.95 | 1.70 |
| Vaal River RSA | 4 | 10 | 0.68 | 0.94 |

## 3.5 *Discussion*

Coetzee *et al.* (2011a) noted that by gaining effective control over floating macrophytes in the water bodies of South Africa, invasive submerged macrophytes were becoming a serious concern. Chapter 2 investigated this interaction and was able to class the lower portion of WMA 10 as a region dominated by Eurasian water-milfoil. The primary aim of this chapter was to investigate the macroinvertebrate communities of the Eurasian water-milfoil populations in the Vaal River and determine whether the macroinvertebrate communities preferred various plant densities and physio-chemical conditions. The last facet of this chapter was to compare this study to similar studies conducted on Eurasian water-milfoil populations of U.S.A. (also an introduced range) and in Eurasia (native range) and to determine whether the species richness and diversity of the Vaal River population could provide an additional explanation of its recently observed increase in cover. This chapter has shown that the macroinvertebrates associated with Eurasian water-milfoil do not have a preference for various plant densities or specific physio-chemical conditions. There are substantially fewer species found on the Eurasian water-milfoil populations of the Vaal River than in Eurasian and North American populations, even though the number of species was slightly under-sampled. The lack of abundant and diverse communities associated with Eurasian water-milfoil suggests that the competitive ability of Eurasian water-milfoil in the Vaal River has not been inhibited by macroinvertebrate herbivory.

Submerged aquatic vegetation, such as Eurasian water-milfoil, provides a complex habitat, with each species having a specific spatial structure, dependent on the abiotic conditions it is exposed to (Bogut *et al.* 2010). Submerged aquatic macrophytes are used by macroinvertebrates for a variety of purposes, as a food source (both plant tissue and the periphyton growing on the macrophyte) (Dvořák & Best 1982, Rooke 1986, Monahan & Caffrey 1996, Zirk & Goldsborough 1996); as shelter from disturbance or predators (Dvořák & Best 1982, Gregg & Rose 1985, Tessier *et al.* 2004); and as breeding sites (Keast 1984).

The structure and architecture of leaves and stems dictates macroinvertebrate community structure and diversity (Soszka 1975, Hann 1995, Cheruvelil *et al.* 2002). The densities of submerged macrophytes therefore play an important role in the species abundance and density of macroinvertebrates (Tarkowska-kukuryk & Kornijów 2008), with increasing plant density and biomass leading to an increasing abundance of aquatic macroinvertebrates (Diehl & Kornijow 1998, Attrill *et al.* 2000). By October 2010, the number of submerged aquatic macrophytes in the Vaal River had dropped compared to previous years, and was restricted primarily to WMA 10 (Chapter 2 Figure 2-15), thus the majority of the sample sites occurred in this region (Figure 3-2). In general, there was low species richness for each site (peaking at 5 species per kg) and a low number of individuals (peaking at 247 individuals/kg, but averaging 21 individuals per kg). This was substantially less than in the study conducted by Wilson & Ricciardi (2009) in the U.S.A., which peaked at 5500 individuals per kg and averaged approximately 4150 individuals per kg, while a study conducted by Cremona *et al.* (2008) in Europe, had densities of nearly 100 000 (99 900) macroinvertebrates per kg, although it was per dry mass of Eurasian water-milfoil. The species accumulation curve (Figure 3-3) suggested under sampling, however it is suspected that the majority of macroinvertebrates associated with Eurasian water-milfoil have been collected, as there was no vast differences between the sample sites in terms of the number of different species encountered (Table 3-1).

Studies on macroinvertebrates associated with Eurasian water-milfoil in its native range found similar percentages of predators (<7%) and herbivores (>80%) in the macroinvertebrate biomass (Cremona *et al.* 2008). In this study however, all chironomid species found were classed as herbivores, as little is known about their feeding ecology in South Africa (F. de Moor, Freshwater Invertebrates Department of the Albany Museum, Pers. comm., 2011). In general, it is more likely that the lack of species richness and in cases high abundance could be a direct indication of the highly polluted status of the Vaal River. Studies have shown the Vaal River to have high levels of heavy metals, sulphates, chlorides and manganese from mining, industrial and urban activities, while organic pollutants in particular high levels of phosphates arise from sewage effluent, and inorganic pesticides from agricultural activities (Heath & Moodley 2011), which would also explain why no insects were found at sample sites 2, 4 and 21.

Density of Eurasian water-milfoil did not affect species richness and abundance (Figure 3-4). These data were strengthened by the results of the Morisita-Horn test (Table 3-2.) that indicated there was no relationship evident between bed density and species richness and abundance. In a study by Sloey *et al.* (1997), measures of biomass of Eurasian water-milfoil showed that there was roughly double the biomass in the centre of a population as there was on the edge, but there was no associated difference between macroinvertebrate densities, indicating that the habitat parameters (such as water quality, plant phenology and condition) were more important than the physical changes in plant density throughout the population. However macroinvertebrate distribution was greater, in terms of density and taxa richness, on the edges than the interior, especially when there was a shallower edge to the population. In this study, it was difficult to delineate the edges of the lower density-classed Eurasian water-milfoil populations, thus the approximate centre of the population was used as a consistent variable during this study.

Eurasian water-milfoil's aggressive growth rate allows it to take advantage of a suitable abiotic environment (discussed in detail in Chapter 1 and 2). In an attempt to quantify how physio-chemical conditions of the water column affected the macroinvertebrate community, a PCA plot was performed on the physio-chemical condition of each site, to determine how different the conditions were, and to compare these similarities in sample sites (Figure 3-6) to the similarities of the macroinvertebrate site similarities (Figure 3-5). No noteworthy relationship was discovered, except that comparisons between these plots reflect how tolerant the existing macroinvertebrate communities present in the Vaal River are to a variety of physio-chemical conditions. Similar macroinvertebrate assemblages were found at different physio-chemical conditions, and macroinvertebrate assemblages having a higher sensitivity for physio-chemical conditions would have resulted in distinctive groupings forming on both plots, in particular the Chironomid genera.

The Chironomid communities of the Swartkops River (Eastern Cape, South Africa) are good indicators of pollution, varying in terms of species richness and abundance to degrees of water deterioration (Oghenekaro 2011). At sites of better water quality in the Swartkops River, there was a higher species diversity of chironomids and macroinvertebrates in general, but this was not the case in this study. The low species richness and occasionally high species abundances, in particular for the chironomid *D. notatus*, are a result of the highly polluted status of the Vaal River (Dallas & Day 2004, Wepener *et al.* 2011). The high levels of pollution have been recorded to cause problems with certain chironomid species' ability to extract oxygen from the water column (Wright & Burgin 2009). Oghenekaro (2011) showed this was not the case for the genus *Dicrotentipes*, which actually showed a positive correlation between water pollution and abundance. Most chironomids feed on particulate organic matter, periphyton growing on Eurasian water-milfoil, as well as the plant itself, and therefore when the plants are growing in polluted waters, species such as *D. notatus* thrive as there is a lack of competition for food (Dickens & Graham 1998, Arimoro & Muller 2010) (Table 3-1). As discussed in Chapter 2, WMA 10 is the least polluted region (Figure 2-1) and has the lowest nutrient levels of the system, while WMA 8 had the highest levels of pollution. This could explain why there were no macroinvertebrates found at two sites in WMA 8, where a number of gold mines are located on the river banks, suggesting that the absence of chironomids on Eurasian water-milfoil could be used as an indicator of system health. Alternatively, it may simply be that the patches of Eurasian water-milfoil were isolated.

In comparison to surveys conducted in Eurasia (Cock *et al.* 2008a) and the U.S.A. (Wilson & Ricciardi 2009) (Table 3-3), the relatively high numbers of species (10) to families (4) in the Vaal River, was an indication of pollution levels. In addition, only certain families were able to use Eurasian water-milfoil as habitat and a food source, compared to the other surveys. The lower Shannon-Weiner index indicates that fewer macroinvertebrates use Eurasian water-milfoil as habitat compared with those in the U.S.A. (Table 3-3). The flooding event in 2010 (Figure 2-14) and the high competitive ability of Eurasian water-milfoil (discussed in Chapter 2) could have resulted in a lack of additional submerged aquatic macrophyte habitat in the Vaal River. Hence it is recommended that additional surveys be conducted after the system has stabilised to a greater degree. These could determine whether the species are simply using Eurasian water-milfoil as their first choice of habitat or whether the macroinvertebrates were present because there are no other indigenous macrophytes in the vicinity.

The low species richness and evenness statistics are a direct result of the fact that Eurasian water-milfoil is an introduced species, in a highly polluted environment, as it has not been in the system long enough to allow for insects to switch their preference. Chapter 2 also showed how Eurasian water-milfoil has only recently peaked in dominance across WMA 10, and thus macroinvertebrates are possibly still going to switch preference away from their usual hosts. Future studies should compare the richness and abundance of macroinvertebrate species found on native plants to the macroinvertebrates found on Eurasian water-milfoil. This would allow for comparisons regarding the macroinvertebrates preference regarding habitat or food source. It is suspected that the plant is being utilised as a complex habitat rather than a specific food source by the native macroinvertebrates, because the plant diversity to species richness relationship does not comply with Island Biogeography Theory (IBT). IBT states that the number of species found on suitable isolated habitat is directly proportional to the rate of immigration versus the rate of extinction, and these rates are affected by the amount of suitable habitat available (MacArthur & Wilson 1967). However the polluted status of the river negated this theory.

To conclude, Eurasian water-milfoil does not have an abundance of macroinvertebrates using it, compared to U.S.A. and Eurasian surveys, which could be as a direct result of its being an alien macrophyte. The low species richness and species abundance findings illustrate that Eurasian water-milfoil's competitive ability has not been restricted by native macroinvertebrates and that it is a serious threat as an invasive macrophyte and needs to be controlled. However, the highly polluted nature of the Vaal River means that additional surveys on other populations of Eurasian water-milfoil and on other indigenous macrophytes of the Vaal River would be required to confirm this hypothesis.

# CHAPTER 4 – General Discussion

## 4.1 *Introduction*

Eurasian water milfoil is one of several aggressive submerged macrophytes that have been present in South African aquatic ecosystems for lengthy periods of time, but have never been a problem. Coetzee *et al.* (2011a) recently voiced concern over the presence of aggressive submerged macrophytes, such as Eurasian water-milfoil in the Vaal River and Lake Sibaya, hydrilla in Pongolopoort dam, Brazilian/dense waterweed throughout South Africa and the potential for Canadian water-weed and cabomba to proliferate and dominate systems, especially if the floating plants are controlled. To successfully restore impacted South African waters to an acceptable standard, it is of the highest importance to understand the successional route an ecosystem will undertake, once a measure of control has been implemented. Coetzee *et al.* (2011a) have predicted that, in line with Scheffer *et al.* (2003) and similarly to other aquatic systems around the world such as those in Holland (Janse & Puijenbroek 1998), Australia (Morris *et al.* 2003), and Malaysia (Sharip *et al.* 2011), if floating plants are controlled in South Africa, light will penetrate the water column and the next onslaught of aquatic alien invasives will be in the form of submerged aquatic plants. The converse will also apply if a system becomes eutrophic. In eutrophic systems the elevated nutrient levels shift the system away from a stable submerged aquatic dominant state to an alternative stable state where floating plants dominate the system, restricting light penetration. This study has shown that the theory of alternative stable states is applicable to and has already occurred on the Vaal River, and responsible management strategies need to be adopted to effectively manage this change in alternative stable states.

## 4.2 *Succession management*

The understanding of the successional pathways that an ecosystem undertakes, while adhering to the mechanisms of succession theory (Pickett *et al.* 1987), is what Luken (1990) proposed in his theory of "succession management". Succession management involves two principals: one, that all plant communities are in a state of flux and can always undergo a form of succession (Niering 1987); and secondly, that the management actions undertaken only alter the pace and direction of succession without changing the condition of the vegetation (Luken 1990). Luken (1990) used succession theory as a framework for his theory of successional management and concluded that there were three basic causes of succession: site availability, differential species availability and differential species performance.

To effectively guide ecosystems to the desired successional state, managers would have to firstly design an effective disturbance, creating site availability. Following this, they would then initiate controlled colonisation of the site by using specific methods to govern the rate of establishment of a specific plant species (dictating the differential species availability). Lastly, managers would control the newly established plant population growth rates. These rates would have to be monitored and sometimes adjusted to achieve the desired successional outcome (directly limiting species performance) (Luken 1990).

When analysing the theory of succession management with the theory of alternative stable states, in the context of aquatic plants on the Vaal River, it is clear that the existence of alternative stable states can assist managers with succession management. WMA 10.4 supplied evidence of the existence of alternative stable states in the Vaal River (Figure 4-1). Locations 1, 3, 5 are all state of flux stable states, located between 2 (water hyacinth), 4 (Eurasian water-milfoil) and 6 (native or cosmopolitan alternative stable states). Position 1 occurs twice as it is a cyclic system. Position 1, resilience is high as the Vaal River is eutrophic (Scheffer *et al.* 2003) and even though integrated control measures have been implemented against water hyacinth this still results in its requiring large quantities of perturbation to overcome the resilience. The introduction of the grasshopper *Cornops aquaticum* (Brüner) (Orthoptera: Acrididae), currently not present in the Vaal River (Coetzee *et al.* 2011b), could reduce the resilience of the water hyacinth stable state.

Following the findings of chapters 2 and 3, it is evident that few macroinvertebrates use Eurasian water-milfoil and although the Vaal River is a eutrophic system, WMA 10 showed lower TDS and nitrate levels than WMA 8 or 9 (Figure 2-26 and Figure 2-30). These factors have created considerable resilience for the stable state and this resulted in the two aquatic weed states, position one and two, having the same resilience. The native or cosmopolitan submerged macrophyte stable state is the desired alternative stable state for the Vaal River and it is assumed that there would be a greater abundance and variety of macroinvertebrates feeding on these species compared to the invasive weeds (Demopoulos *et al.* 2007, Whitcraft *et al.* 2008, Wilson & Ricciardi 2009, Yoshioka *et al.* 2010, Hansen *et al.* 2011) resulting in the lower resilience of this state.

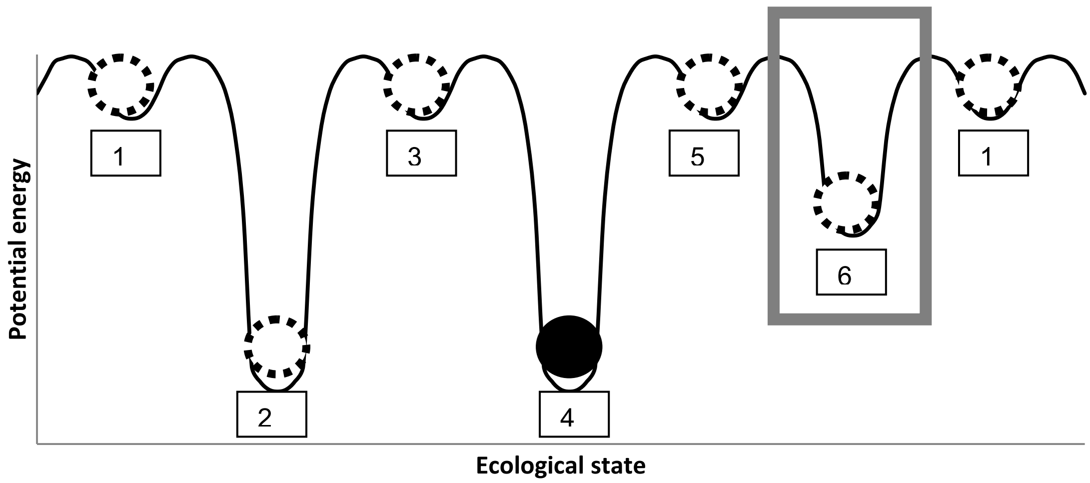



**Figure 4-1:** Hypothetical illustration of a variety of alternative stable states in the Vaal River, commonly referred to as the ‘Ball and Cup’ analysis, modified from Dent *et al.* (2002). The boxed region is the desired alternative stable state and the ‘ball’ currently depicts the state of WMA 10.4 in position four.

To satisfy succession management criteria which would change the system from a floating aquatic macrophyte system to a submerged macrophyte system, the first act required was the creation of site availability. In WMA 10.4 this was the result of a large stochastic flooding event in 2006 (Figure 2-14). The flood lifted the 'ball' out of position one and, combined with the integrated control programme against water hyacinth, raised the 'ball' high enough through position 3 into position 4, where it now resides in WMA 10.4 (chapter 2). In some instances, hysteresis did occur and the ball reverted back to position 3, however the continued perturbations in the form of the integrated control programme against water hyacinth moved those regions into position 4. The integrated control programme constituted the differential species performance requirement of successional management.

The last phase of succession management requires control of newly established plant growth rates by directly limiting species performance. Differential species performance will require the restoration of the Vaal River's water chemistry and characteristics. The Vaal River is a highly polluted, turbid, eutrophic system (Grobler *et al.* 1983, Cilliers *et al.* 1996, Wepener *et al.* 2011). But establishment of large stands of Eurasian water-milfoil does have a feedback loop whereby it does start to change the surrounding environment to suit its needs. Eurasian water-milfoil reduces turbidity and absorbs nutrients from the sediment and water column ,and while it is established (Smith & Barko 1990), only a large stochastic event such as a flood, could revert the system back to a water hyacinth-dominated system. Eurasian water milfoil therefore satisfies the differential species performance characteristic by itself.

To enter into the desired alternative stable state (Figure 4-1), where native or cosmopolitan species are dominant, succession management will also need to be utilised. To create site availability, the system may require a large stochastic event, but without managing water quality it is highly likely that the 'ball' (Figure 4-1) will revert back to a water hyacinth-dominated state of position two, instead of position six, following the flood. Therefore it is imperative that integrated control continue to be implemented against water hyacinth and water quality be improved by continuing to upgrade sewage works, and by limiting the quality of industrial, urban and agricultural runoff into the Vaal River. The improvement in water quality would also increase the species richness and abundance of macroinvertebrates that could feed on Eurasian water-milfoil, lowering its resilience (Dallas & Day 2004). It is envisioned that should the water quality improve and an integrated control programme involving the use of herbicides and biological control agents be initiated against Eurasian water-milfoil, satisfying the differential species availability requirements of succession management, then following a flooding event, a native or cosmopolitan submerged macrophyte could dominate the system as it shifts into alternative stable state position 6 (Figure 4-1), thereby completing the goal of ecosystem restoration.

## 4.3 *Management of Eurasian water-milfoil in South Africa*

Chapter 1 outlined in detail the management plan used in North America against Eurasian water-milfoil, however, despite various control measures implemented against it since 1965, Eurasian water-milfoil still occurs in almost all 50 states in North America (Smith & Barko 1990, Madsen 1998). In South Africa, the implementation of a broader control programme, not restricted by state boundaries and law, should have a greater impact on the Eurasian water-milfoil infestation. The first phase of this programme should be to determine other areas where this plant is a problem and to model the potential range of this plant using GIS systems such as the model used by Coetzee *et al.* (2009b), who combined shapefiles with water chemistry, climatic conditions, water physical data (e.g. depth data) and public access points to model which habitats were the most at risk from *Hydrilla verticillata* (L.f.) Royle (Hydrocharitaceae) infestation, similarly Prince (2011) performed the same analysis on Eurasian water-milfoil in the U.S.A. There is a confirmed record of Eurasian water-milfoil in Lake Sibaya and other possible locations (Figure 3-1) should be investigated as soon as possible, following which, a 'prevention is better than cure' approach should be implemented, similar to those used in the U.S.A., discussed in Chapter 1. Public awareness, by means of signage at boat ramps, consultation with government agencies and water use groups and advertisement of the danger that this plant poses to an ecosystem, can minimise the spread of the weed, and this in turn will make control of Eurasian water-milfoil more efficient.

The implementation of a herbicide regime in the Vaal River, where different herbicides are used for different conditions (Chapter 1), should also be correlated with natural flooding events or water level manipulation. The herbicide regime should focus on the source population, WMA 10.4. Control of this region will reduce addition sink populations and reduce the risk of additional infestations occurring. As discussed and analysed in Chapter 2, this will provide the maximum perturbation required to switch the Eurasian water-milfoil stable state, and with the release of the milfoil weevil, could bring Eurasian water-milfoil cover down to acceptable levels.

## 4.4 *Conclusion*

This study has shown that spatial analysis can be used as an effective tool for ecosystem monitoring of aquatic systems, even on a relatively small scale. Research using spatial analyses should investigate whether alternative stable states exist in other South African ecosystems, both terrestrial and aquatic. If required, where these establishments have taken place, successional management strategies should be used as a tool for restoration ecology.

The lack of macroinvertebrate surveys that occur on aquatic macrophytes in South Africa is concerning, and without such surveys it is difficult to quantify how aquatic alien invasive plants are impacting ecosystems from a biological scale. In particular, chironomids require extensive research and a South African key needs to be developed. Following this, the interactions between chironomids and Eurasian water-milfoil should be investigated.

Research into how grass carp and indigenous vertebrate grazers are impacting the Eurasian water-milfoil population could also provide additional insight into how vertebrates use and impact the plant, as well as giving an indication of the resilience of the Eurasian water-milfoil alternative stable state.

This study also illustrated how the theory of alternative stable states can be inferred at a regional scale, and how the shifting of ecosystems to their alternative stable states has the potential to be used as a tool for ecosystem restoration via succession management The shifting of an ecosystem to an alternative stable state, where another alien species is dominant, is of course not the aim of ecological restoration. However, it is envisaged that indigenous or cosmopolitan species that can compete for the same niche as Eurasian water-milfoil, will play an essential role in the restoration of the Vaal River through the implementation of succession management strategies.

Freedman *et al.* (2007) highlighted how, if biological control agents are introduced against a target weed, plant competition by native or cosmopolitan species for the same niche can result in a favourable outcome where indigenous/cosmopolitan species outcompete the target weed. Research plays a pivotal role in this regard as it will allow decision makers to reach informed decisions regarding the successional management of regions, and thus they can prepare for the next stable state before it begins. In the case of the Vaal River, this process has already begun. The milfoil weevil is currently undergoing host specificity testing at Rhodes University, for release against Eurasian water-milfoil, before the plant becomes a problem for the rest of the Vaal River. However alien macrophyte dominance is a symptom of poor water quality, and without the enforcement of sustainable, responsible farming practices, the reduction in industrial and urban runoff, as well as renewed government spending in terms of upgrading of sewage works, it is envisioned that the eutrophic conditions will persist in the remainder of the Vaal River (excluding WMA 10) and water hyacinth will remain a dominant state over the majority of the Vaal River.

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