

# TOWARDS ASSESSING IMPACTS OF ALIEN PLANT INFESTATIONS ON RIVER SYSTEMS IN THE SOUTHERN CAPE USING COST-BENEFIT ANALYSES

Report to the  
**Water Research Commission**

by

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# Executive Summary

## Background and motivation

Ecosystem resilience is key to the provision of dependable ecosystem goods and services, and it is generally accepted that ecosystem diversity helps to maintain system resilience. It is therefore reasonable to postulate that changes to the variables that drive species patterns will result in changes to ecosystem community structure and consequently negatively impact on system resilience. Alien vegetation in the riparian zone can impact on water temperatures, flow patterns, degree of shading, channel modification, and changes to natural sediment loads. Climate change is likely to exacerbate the problem both directly through its amplification of thermal extremes in aquatic systems, and indirectly through its impacts on dispersal patterns of alien invasive vegetation.

Freshwater systems in the Garden Route Initiative planning domain area – George, Knysna and Plettenberg Bay – are of particular conservation value because of their rich Gondwanaland relic aquatic macroinvertebrate fauna found in these rivers, which are vulnerable to thermal changes. Clearing of alien riparian vegetation, and restoring riparian zones to either a desired future state or a natural state, will theoretically restore flow and water temperature regimes underpinning community patterns. Different clearing techniques will have different associated costs and potentially achieve different levels of ecosystem restoration, although what these costs and benefits are remains to be quantified.

## Aims

This WRC project examines information on the costs of clearing alien riparian vegetation relative to the ecological benefits, as assessed by convergence of water temperatures to target values. This is achieved through the following objectives:

- To define relationships between alien plant densities in the riparian zone and changes to water temperature regime
- To define relationships between alien plant densities in the riparian zone and changes to the structure of aquatic macroinvertebrate communities
- To establish the effect of different clearing techniques/approaches in the riparian zone on water temperatures and associated aquatic habitat integrity and community response
- To estimate the most cost-effective clearing techniques in terms of financial efficiency and ecological returns

## Methods

A range of data were collected at a total of 19 sites on seven rivers systems in the Garden Route Initiative area of southern Cape, between the towns of George and Knysna. These included 12 months of hourly water temperatures at all sites, and quantitative sampling of

aquatic macroinvertebrates at ten of these sites. Each site was also characterised in terms of water quality (pH, conductivity and turbidity) and general characteristics, including impacts such as density of alien riparian trees. Biotic responses were based on exceedances of thermal thresholds, which were in turn linked to clearing costs extracted from the national Working for Water database.

### **Extent to which the contract objectives were reached**

Aims 1-2 and 4 were successfully achieved within this study. The third aim of comparing different clearing techniques was achieved within the restrictions of the best available data from the Working for Water database. Results from such clearing would need to be measured over a long-term period (5-10 years) because initial responses of aquatic systems would merely be reflecting recovery from disturbance.

### **Summary of major results and key findings**

#### *Relationships between alien plant densities in the riparian zone and changes to water temperature regime*

- Thermal regimes were relatively consistent across rivers, irrespective of densities of alien riparian vegetation.
- However, exceedances of biological thermal thresholds were higher at downstream sites with relatively higher densities of alien invasive riparian vegetation. Such densities are likely to act as surrogates for overall contributing upstream catchment degradation.
- It is likely that groundwater inputs buffer thermal regimes to some extent in the GRI rivers.
- Thermal differences appear to be more a function of upstream versus downstream influences than direct impacts of alien riparian vegetation.

#### *Relationships between alien plant densities in the riparian zone and changes to the structure of aquatic macroinvertebrate communities*

- Aquatic macroinvertebrate communities at the family level showed variation between sites and seasons.
- However, these differences appeared to be more pronounced on the basis of natural land cover type (fynbos versus indigenous forest) than densities of alien invasive riparian vegetation.
- The same species of aquatic macroinvertebrates showed different levels of thermal tolerances between different river systems.

*Effect of different clearing techniques/approaches in the riparian zone on water temperatures and associated aquatic habitat integrity and community response*

- Different clearing techniques could not be assessed in this project. During the course of this study, it became apparent that clearing techniques are relatively prescriptive based on the WfW Norms and Standards guidelines for different species, age classes and densities.
- Within these constraints, different management approaches could be defined based on degree of proactiveness, and number of follow-up treatments.
- Aquatic habitat integrity appeared to be relatively resilient to differing levels of alien riparian densities. However, downstream sites of catchments heavily infested with alien vegetation appear to exhibit a higher degree of thermal extremes than expected.
- Aquatic macroinvertebrate community response appears to be most likely driven by the extreme flow variability exhibited by the rivers of the GRI region, i.e. the overriding variable in these systems is that they are flow-event driven.

*Estimation of the most cost-effective clearing techniques in terms of financial efficiency and ecological returns*

- The WfW database of clearing costs (person days per hectare) at different densities of different species was a key resource for developing cost-benefit models.
- Person days per hectare were directly used as costs for clearing of alien vegetation at a catchment-level scale, with benefits to the ecosystem also reflected as person days per hectare.
- Using these values, it was possible to successfully compare different management approaches (e.g. once-off clearing versus multiple follow-ups) at current prices using a spreadsheet-based Net Present Value economic model.
- Best value per percentage gain towards a thermal reference condition provided the link between financial efficiency and ecological returns. The long-term benefits of such actions would be reflected by increased resilience to warming water temperatures in response to global climate change.

## **Discussion**

The study actively involved two SANParks staff members (Mr Bheki Maphanga and Ms Jessica Hayes) who were exposed to a number of aquatic ecological sampling techniques including SASS5 assessments, water quality measurements, and sampling of aquatic macroinvertebrates. It has contributed new knowledge on the ecological functioning of the southern rivers of the Garden Route Initiative area:

- The thermal regimes of the rivers are all relatively similar, and by extension all likely to exhibit similar thermal changes in response to global climate change. However, the same species across different rivers exhibit different thermal tolerances, such that ecological responses are likely to differ between rivers.
- The site-specific densities of alien riparian vegetation appear to have less of an impact on river ecology than major natural land cover types – fynbos versus indigenous forest.

- Site-specific levels of alien riparian vegetation density is likely to act as a surrogate for overall upstream catchment transformation, and this is reflected in exceedances of biological thresholds based on mean daily water temperatures.

Based on the findings of this study, there is promise towards developing cost-benefit models incorporating targets for thermal reference conditions. This is a key step towards prioritizing areas for clearing of alien invasive vegetation. These findings could be applied not only in the GRI area, but also at a national scale.

### **Conclusions and Recommendations for further research, knowledge dissemination and technology transfer**

- While costs of clearing of alien vegetation – expressed as person days per hectare with an annually adjusted daily rate – are relatively well established and accepted within the industry, these figures remain subjective because of challenges in estimating alien plant infestation densities. A number of factors compound this:
  - Stands of aliens consist of mixed species and mixed age classes.
  - Costs are based on densities expressed as a percentage, while field assessment densities (e.g. transects of PCQM) are based on numbers of trees per 100 m<sup>2</sup>. There does not appear to be a relationship to reconcile these.
  - Clearing costs on their own are difficult to budget for on a medium to long term basis (5-10 year scale). It is easier to justify budgets if costs are related to benefits, and both metrics combined using Net Present Value incorporating a suitable discount function.
  - A key future research area is thus on how to measure benefits of clearing alien vegetation. The use of person day equivalents is one option, but it is recommended that further benefit metrics that can be translated into a financial value are investigated.
- Relative benefits of catchment-wide clearing versus on-site clearing need to be assessed through controlled experiments. In this study, it was difficult to separate these compounding effects.

The most practical approach for this to be achieved would be to continue discussions with key members of the SANParks BSP (Biodiversity Social Project – the platform for Working for Water within SANParks areas of jurisdiction), particularly in terms of how budgeting for alien invasive clearing is undertaken over multi-year time intervals. This would be particularly facilitated through the incorporation of cost-comparable benefit values, with both of these being related to current budgets using a Net Present Value matrix. There is also a need to validate the models, where results from such clearing would need to be measured over a long-term period (5-10 years) because initial responses of aquatic systems would merely be reflecting recovery from disturbance.

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## Glossary

| <b>Term/Acronym</b> | <b>Definition</b>  |
|---------------------|--|
| BSP                 | Biodiversity Social Projects   |
| Discount rate       | An interest rate used to bring future values into the present when considering the time value of money   |
| Forestry Exit Areas | Land areas that are in transition from forestry plantations to conservation  |
| GRI                 | Garden Route Initiative  |
| Metric              | A descriptive statistic of a component of a time series, e.g. mean annual water temperature  |
| MWAT                | Maximum Weekly Allowable Temperature – a biologically defensible temperature threshold that should not be exceeded as measured against a 7-day moving average of mean daily water temperatures |
| NPV                 | Net Present Value – the current value of an investment based on a discount rate, and a series of future payments (negative values) and income (positive values)                                |
| PCQM                | Point Centre Quadrat Method, a technique for assessing tree density  |
| SANParks            | South African National Parks   |



# Chapter 1. Introduction

## 1.1 Background

Ecosystem resilience is key to the provision of dependable ecosystem services, and it is generally accepted that ecosystem diversity helps to maintain system resilience. It is therefore reasonable to postulate that changes to the variables that drive species patterns will result in changes to ecosystem community structure and consequently negatively impact on system resilience.

Factors which impact on water temperatures include reductions in flow (surface and groundwater), degree of shading (especially in first or second order streams), channel modification (channel obstructions resulting in changes in hydraulic biotopes and/or changing the residency times of water within a reach), and changes to natural sediment loads. Alien vegetation in the riparian zone can impact on all the factors mentioned above, and climate change is likely to exacerbate the problem both directly through its amplification of thermal extremes in aquatic systems, and indirectly through its impacts on dispersal patterns of alien invasive vegetation. Changes to, *inter alia*, aquatic thermal regimes, will differentially impact different families of aquatic taxa (Dallas and Ketley 2011, Dallas and Rivers-Moore 2012), across the spectrum of functional feeding groups. For example, univoltine species (typically high conservation species) are likely to be negatively impacted by increases in thermal extremes, while multivoltine species (often pest species or generalists) are likely to benefit from the same thermal extremes (see Rivers-Moore et al. 2013b). It is highly likely that marked changes to riverine communities will alter ecosystem processes and ultimately on the delivery of ecosystem services.

The Cape Floristic Region (CFR) is recognised as a global biodiversity hotspot, with the fynbos biome (sclerophyllous vegetation) to the west, and the semi-arid succulent Karoo to the east (de Moor and Day 2013). Afromontane forests occur as small relic patches in the fynbos matrix. Rivers in the CFR are typically short and deeply incised, and aquatic ecosystems are characterised by high rates of endemism. Two major challenges to aquatic conservation efforts are the patchiness of knowledge of these systems, and the patchiness of conserved areas (de Moor and Day 2013).

Freshwater systems in the southern Cape Rivers areas are of particular value because of their rich Gondwanaland relic aquatic macroinvertebrate fauna found in these rivers, which are vulnerable to thermal changes. For example, Prof. Marjorie Scott surveyed a pool in the Homtini River in 1970 and found 20 species of Trichoptera based on 20 minutes of sampling (de Moor 1992). De Moor (1992) notes the “diverse, interesting and valuable aquatic insects ...of the southern Cape...”, and highlights that their faunal communities deserve protection from alien species and habitat degradation, which includes setting aside selected rivers for conservation preserved in as close to pristine a state as possible.

Essentially, clearing of alien riparian vegetation, and restoring riparian zones to either a desired future state or a natural state, will theoretically restore flow and water temperature regimes underpinning community patterns. Different clearing techniques will have different associated costs and potentially achieve different levels of ecosystem restoration, although what these costs and benefits are remains to be quantified. Research at the interface between restoration costs and ecological returns are relatively few, particularly within river systems in South Africa. Globally, literature exists linked to scenario analyses and conservation planning (Wilson et al. 2007; Seedang et al. 2008), with recent South African aquatic restoration assessments emerging from the CFR (Woodford et al. 2012; Samways et al. 2011).

## **1.2 Aims and objectives**

This project examines information on the costs of clearing alien riparian vegetation relative to the ecological benefits, as assessed by convergence of water temperatures to target values. This is achieved through the following objectives:

- To define relationships between alien plant densities in the riparian zone and changes to water temperature regime
- To define relationships between alien plant densities in the riparian zone and changes to the structure of aquatic macroinvertebrate communities
- To establish the effect of different clearing techniques/approaches in the riparian zone on water temperatures and associated aquatic habitat integrity and community response
- To estimate the most cost-effective clearing techniques in terms of financial efficiency and ecological returns

## Chapter 2. Methods

### 2.1 Site selection

A workshop for the purpose of selecting candidate rivers and sites for the study was held on Thursday 30<sup>th</sup> May 2013 at Nelson Mandela Metropolitan University's George Campus (Saasveld). The workshop was aimed at identifying sites on suitable paired rivers to assess the impacts of alien riparian vegetation on thermal signatures and aquatic macroinvertebrate communities. Notes from this workshop are summarised in Appendix 2.1. Here, it was noted that it would be difficult to find perfectly matched impacted versus reference sites, because in reality land use was a mosaic of natural patches (fynbos and indigenous forest) and transformed land. It was concluded that a more pragmatic approach would be to pair river systems, and assess sites based on where they fell within a natural-to-impacted spectrum, and also taking into account upstream impacts.

The site selection process was based on the suggestions from the workshop, followed by two days of preliminary field work, where candidate river systems were visited. As opposed to finding paired sites for "natural" versus "impacted", all sites fell within a spectrum of relatively less impacted to relatively more impacted, because of the nature of the surrounding land use within each catchment (Figure 2.1). In fact, all of the study sites fell within quaternary catchments with 50% or less natural vegetation remaining, according to the most recent Garden Route Initiative (GRI – a bioregional assessment and planning initiative) transformation layer (Figure 2.2), which made it difficult to define a benchmark "natural" condition. The land use mosaic in the study catchments made it difficult to select paired study sites for "impacted" and "non-impacted" conditions (Figure 2.3). In certain instances, site access at desirable locations was not feasible because of terrain. Afforested catchment areas in certain instances were in a state of ownership uncertainty, with areas being either clear-felled with the intention of becoming conservation areas, or where in one instance a site was not accessible because of felling taking place during the project.

Based on an initial list of 14 sites selected from the workshop outcomes, an additional five sites were added based on observable access points and proximity to RHP and/or gauging weir sites. Based on accessibility and selecting sites that included both natural and alien riparian vegetation, a total of 19 sites occurring over six river systems were selected (Table 2.1 and Figure 2.4; Plates 1-2). Water temperature loggers were installed at each of these sites and a subset of ten sites were used for macroinvertebrate sampling in order to rationalise data returns against sampling effort. Two sites could not be accessed during September 2013 for logistical reasons: timber felling in the upper Knysna catchment prevented site access, while time limitations prevented access at site 18-Knys3 on the Rooiels River. Sites cover upper and lower river reaches (Figure 2.4), with a number of sites being adjacent to flow gauging weirs and/ or River Health Programme sites (Table 2.1). Land use includes fynbos and indigenous forest, plantation, and plantation exit areas. Riparian zones exhibited varying levels of alien plant species invasion, from zero aliens to 40 trees per 100 m<sup>2</sup> (Plate 2). The last category is of use to this study because these sites occur on historically forest or fynbos vegetation which was either converted to plantations or

occurred in catchments where alien plant species had invaded. During the study period, timber in certain areas was being felled, with the timber companies concerned “exiting” the areas; the intention is for the land use to revert to natural state with custodianship of the land reverting to conservation agencies.

Study sites all fell within a single primary catchment (K), in a rainfall region of the southern Cape exhibiting a bimodal annual rainfall pattern with peak rainfalls in spring and winter. Upper study sites all occurred on quartzitic sandstone, while the lower western sites occur on metamorphic rock (schist). Sites were selected on seven river systems falling into nine quaternary catchments, between the urban areas of George and Knysna (western half of the Garden Route Initiative). All catchments have less than 54% natural vegetation remaining, with the balance being made up of a mix of dairy farms and pasture, plantations, plantation exit areas (Plate 3), and smallholdings.

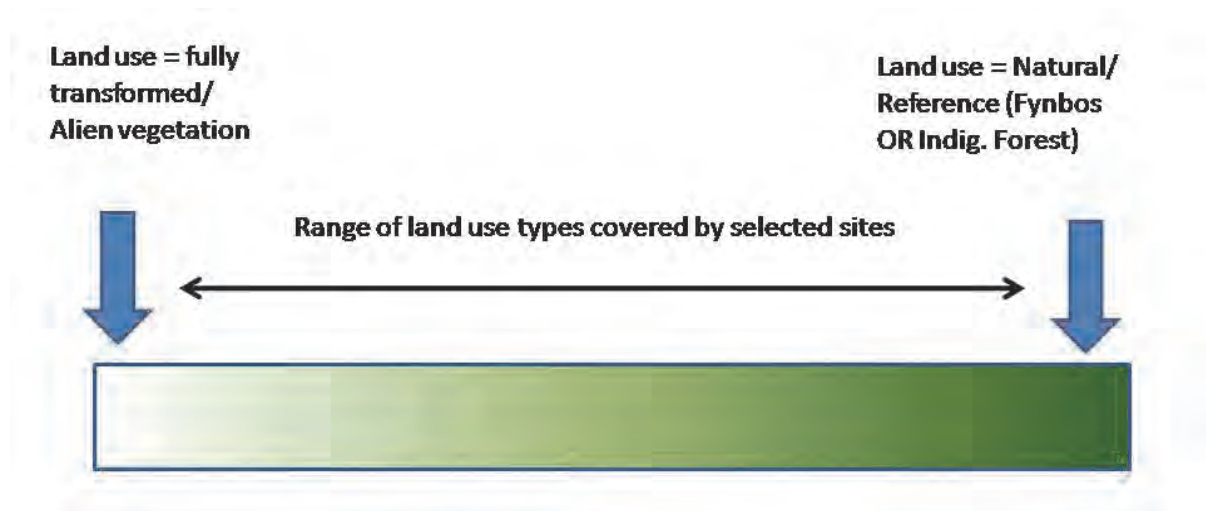


Figure 2.1 Conceptualization of how sites are located within a spectrum of land use types

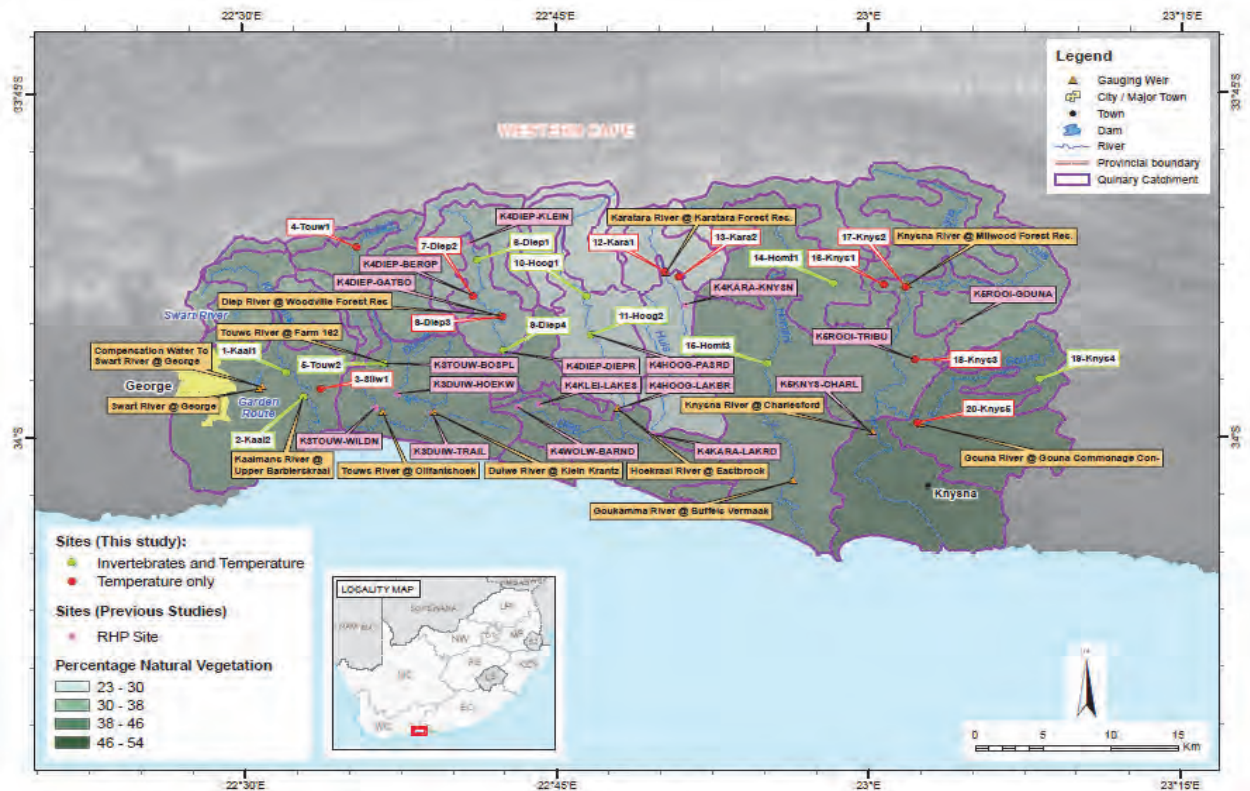


Figure 2.2 Percentage of natural vegetation per quaternary catchment based on the Garden Route Initiative transformation layer (see Table 2.3 for site information). Map provided by the Department of Water and Sanitation, Land Cover

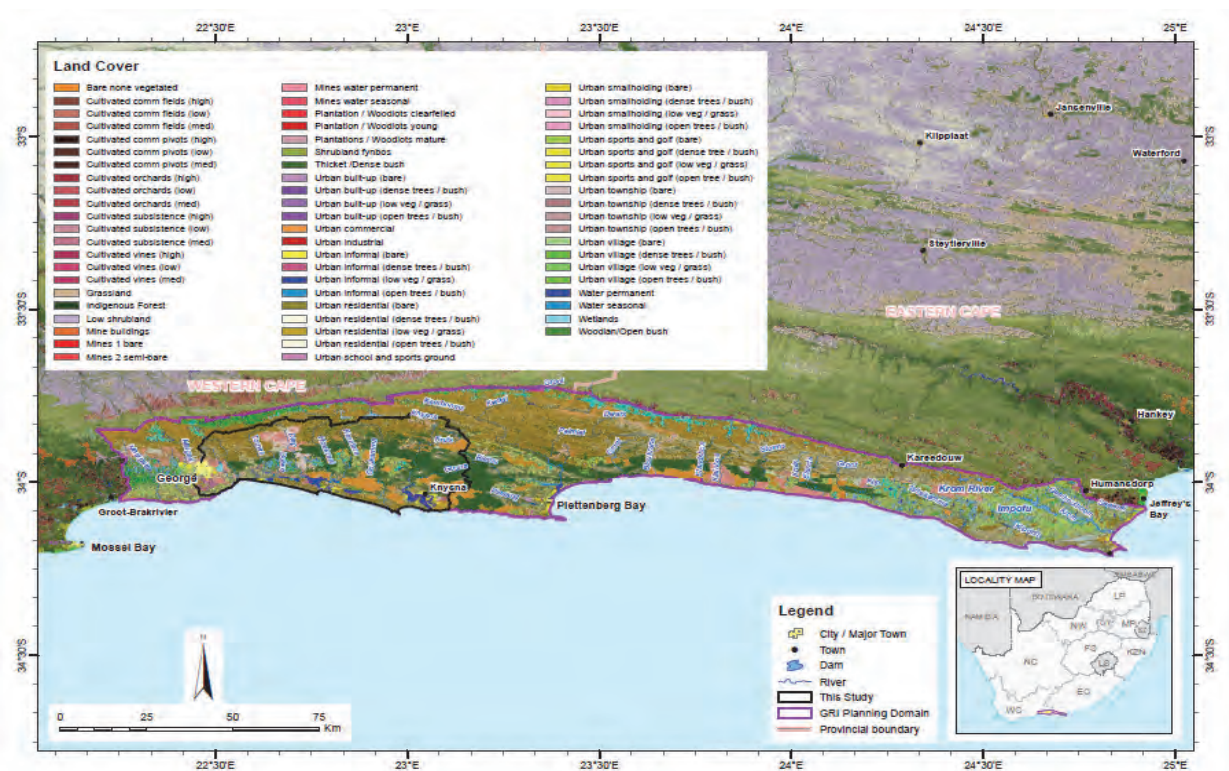


Figure 2.3 Land cover in the Garden Route Initiative area based on the 72 Class GTI South African National Land Cover Dataset (2013/2014). Map provided by the Department of Water and Sanitation, Land Cover © GEOTERRAIMAGE – 2014.

**Table 2.1 List of sites where water temperature loggers have been installed, and where aquatic macroinvertebrates were sampled. # Indicates plantation forestry exit areas**

| Site code | Latitude (DD S) | Longitude (DD E) | Altitude (m amsl) | River      | Mainstem   | Logger | Sample | Location | Weir   | RHP          | Land use                       |
|-----------|-----------------|------------------|-------------------|------------|------------|--------|--------|----------|--------|--------------|--------------------------------|
| 6-Diep1   | -33.87159       | 22.68755         | 313               | Diep       | Diep       | Y      | Y      | Upper    |        |              | Fynbos <sup>#</sup>            |
| 7-Diep2   | -33.89763       | 22.68370         | 277               | Diep       | Diep       | Y      |        | Upper    |        | K4Diep-Bergp | Fynbos <sup>#</sup>            |
| 8-Diep3   | -33.91308       | 22.70742         | 227               | Diep       | Diep       | Y      |        | Lower    | K4H003 | K4Diep-Gatbo | Forest                         |
| 9-Diep4   | -33.93757       | 22.70849         | 182               | Diep       | Diep       | Y      | Y      | Lower    |        |              | Mixed/Alien                    |
| 14-Homt1  | -33.88854       | 22.97156         | 315               | Homtini    | Homtini    | Y      | Y      | Upper    |        |              | Forest                         |
| 15-Homt3  | -33.94744       | 22.91904         | 46                | Homtini    | Homtini    | Y      | Y      | Lower    |        |              | Mixed/Alien                    |
| 10-Hoog1  | -33.89802       | 22.77450         | 269               | Hoogekraal | Hoogekraal | Y      | Y      | Upper    |        |              | Forest/Plantation              |
| 11-Hoog2  | -33.92606       | 22.77818         | 141               | Hoogekraal | Hoogekraal | Y      | Y      | Lower    |        | K4Hoog-Pasrd | Forest                         |
| 1-Kaai1   | -33.95226       | 22.53441         | 137               | Kaaimans   | Kaaimans   | Y      | Y      | Lower    |        |              | Forest                         |
| 2-Kaai2   | -33.97026       | 22.54842         | 94                | Kaaimans   | Kaaimans   | Y      | Y      | Lower    | K3H001 |              | Forest/Alien                   |
| 3-Silw1   | -33.96477       | 22.56176         | 153               | Silwer     | Kaaimans   | Y      |        | Lower    |        |              | Forest                         |
| 12-Kara1  | -33.88012       | 22.83710         | 248               | Karatarata | Karatarata | Y      | Y      | Upper    |        |              | Forest/Plantation <sup>#</sup> |
| 13-Kara2  | -33.88448       | 22.84878         | 264               | Plaat      | Karatarata | Y      | Y      | Upper    |        |              | Fynbos/Plantation <sup>#</sup> |
| 16-Knys1  | -33.89008       | 23.01226         | 283               | Millwood   | Knysna     | Y      | Y      | Upper    |        |              | Plantation/Fynbos <sup>#</sup> |
| 18-Knys3  | -33.94450       | 23.03720         | 271               | Rooiels    | Knysna     | Y      | N      | Upper    |        | K5Rooi-Tribu | Forest                         |
| 19-Knys4  | -33.95855       | 23.13655         | 442               | Gouna      | Knysna     | Y      | Y      | Upper    |        |              | Forest                         |
| 20-Knys5  | -33.98977       | 23.03905         | 61                | Gouna      | Knysna     | Y      | Y      | Lower    | K5H001 |              | Mixed/Alien                    |
| 4-Touw1   | -33.86225       | 22.59114         | 378               | Touw       | Touw       | Y      | Y      | Upper    |        |              | Fynbos <sup>#</sup>            |
| 5-Touw2   | -33.94657       | 22.61263         | 154               | Touw       | Touw       | Y      | Y      | Lower    | K3H005 | K3Touw-Bospl | Plantation/Alien               |

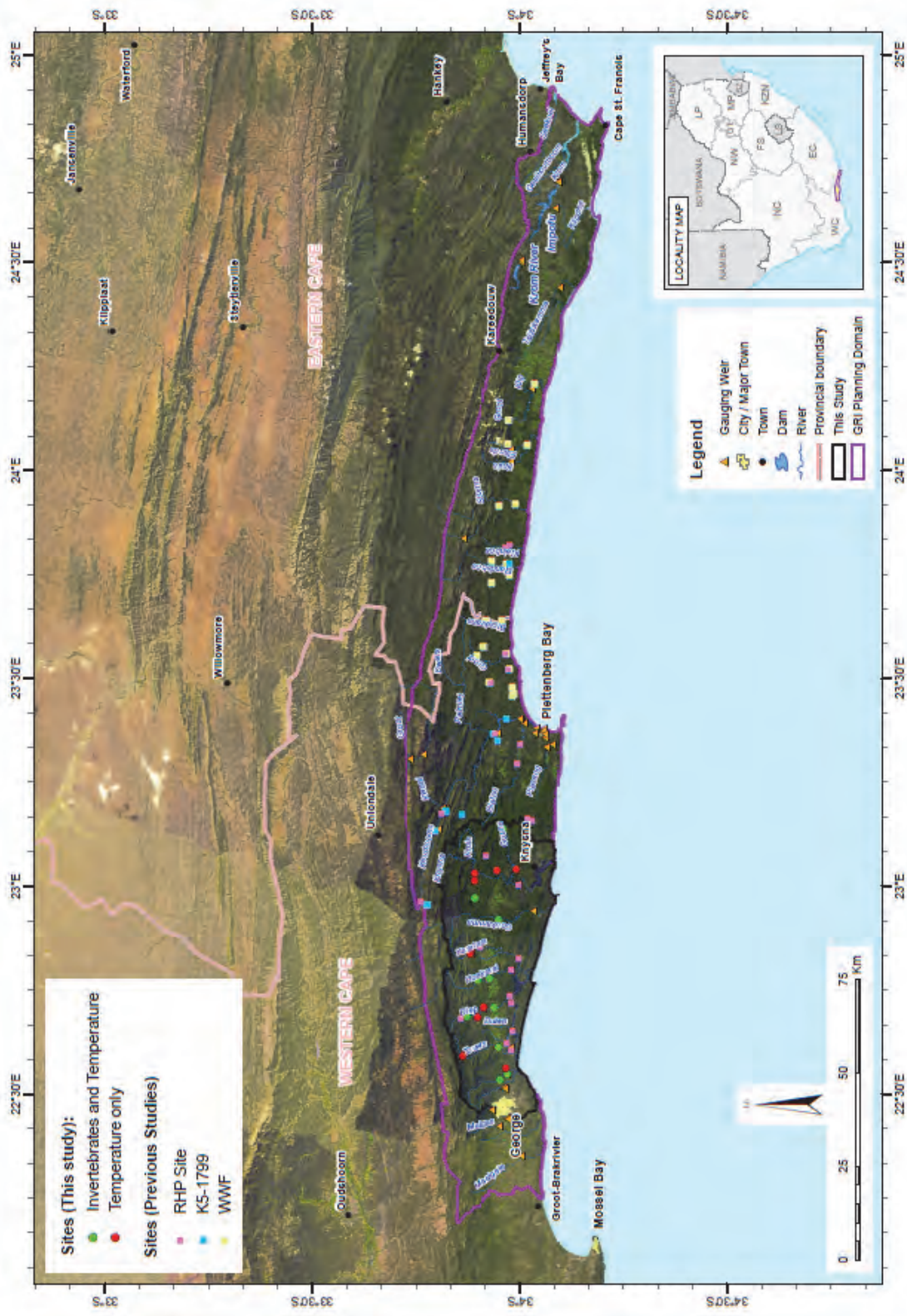
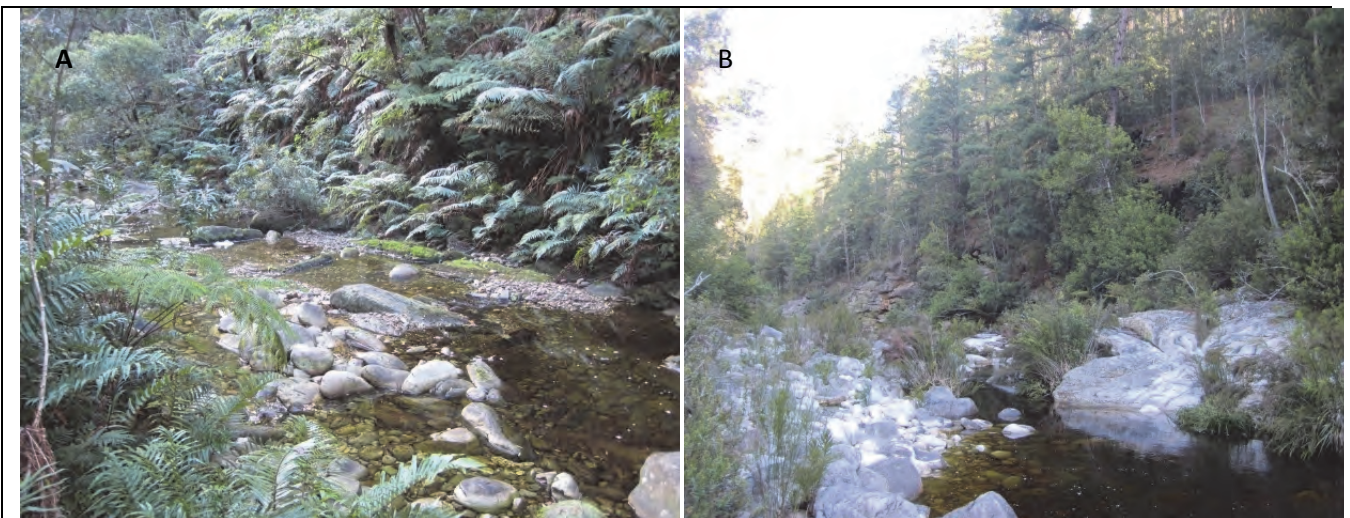


Figure 2.4 Study area (Garden Route Initiative planning domain) showing locations of river systems to be assessed in this study (western systems) and location of sites with historical water temperature and aquatic macroinvertebrate data (eastern systems). Map provided by the Department of Water and Sanitation, Land Cover



**Plate 1 A-B Examples of land use at selected sites: Fynbos and limited occurrence of Alien vegetation in surrounding catchment in the upper Diep River 6-Diep1 (A); Fynbos vegetation but with more extensive plantation areas and alien invasion in the Plaat River 13-Kara2 (B);**



**Plate 2 A-B Indigenous forest in riparian zone of site 14-Homt1 (Jubilee Creek) (A); and mix of indigenous forest on left hand bank and Pine plantation on opposite bank at site 10-Hoog1 (B)**





**Plate 3** Example of pine felled in an exit area at site 16-Knys1

## **2.2 Collection of time series data**

To understand and interpret biological patterns and their differences between sites, we need to have previous history of the abiotic template. Two of the overriding variables explaining aquatic species patterns are temperature and flow, and especially time series data on these variables for two weeks prior to sampling (Jackson et al. 2007).

### **2.2.1 *Water temperature data***

Hobo TidbiT v2 water temperature loggers (Onset Hobo data loggers; [www.onsetcomp.com](http://www.onsetcomp.com)) programmed to record hourly water temperatures were installed as per the approach of Dallas et al. (2012). Loggers were installed at sites previously identified, with additional and/ or alternative sites added where necessary, during the September 2013 field trip. Data from loggers were downloaded during the January, April and October 2014 surveys to provide approximately one year of data. Details pertaining to the installation dates and period for which data were logged are provided Appendix 2.2.

Hourly data were collated into a single spreadsheet and in order to make comparisons between all sites possible, a common period of data was required. A total of 15 of the total 19 sites generally

had such data. For the remaining sites, to achieve this, missing data were patched using linear regressions between sites with full and partial hourly data.

To compare temperature patterns across sites, a number of exploratory approaches have been undertaken. At the most basic level, box-and-whisker plots based on hourly data were used (R Development Core Team 2009). Next, hourly data were up-scaled to daily data (mean, minimum and maximum), and these data were described in terms of the metrics developed by Rivers-Moore et al. (2013a) to describe temperatures in terms of duration, frequency, and magnitude of thermal events.

### 2.2.2 Air temperature data

Air temperature data were collected using Dallas I-Buttons (DS1923 Dallas I-button), which included a radiation shield. Several of the loggers malfunctioned, resulting in only three sets of air temperature data for variable periods over the project duration.

### 2.2.3 Flow data

Mean daily flow data were available from five flow gauging weirs covering five rivers. For four out of the five sites, data were available for more than 45 years (Table 2.2). Records of 10-20 years are recommended as necessary to model short-term trends and seasonal components within the year, and encompassing at least one high and one low sequence (Gordon et al 1994; Taylor et al. 2003). At this stage, all data per site have been pooled and compared, based on untransformed flow data. Broad-level comparisons were made based on box-and-whisker plots (R Development Core Team 2009).

**Table 2.2 Flow gauging weirs and their observed data record lengths**

| Gauging weir | River    | Data period                    |
|--------------|----------|--------------------------------|
| K3H001       | Kaaimans | 1 April 1961 – 30 Sept. 2014   |
| K3H005       | Touws    | 1 May 1969 – 30 Sept. 2014     |
| K4H003       | Diep     | 1 June 1961 – 30 Sept. 2014    |
| K5H001       | Gouna    | 13 Sept. 1961 – 3 January 1972 |
| K5H002       | Knysna   | 1 Sept. 1961 – 30 Sept 2014    |

## 2.3 Site characterisation

### 2.3.1 Physical structure and physico-chemistry

Sites were characterised in terms of their physical structure and physico-chemistry. For the former, measurements of river width and typical depth were recorded during each sampling period, while for the latter spot values were recorded per sampling trip (pH, conductivity and turbidity). The presence of algal films on in-channel stones and palmiet, *Prionium serratum*, at each site was also recorded.

### **2.3.2 Alien riparian plant densities**

A once-off estimate of levels of alien riparian infestation was undertaken in April 2014 at all 19 sites. Here, species type and size class (finger, wrist and > thigh widths) were recorded from within the riparian zone, where the riparian zone was defined as active channel plus primary floodplain. Cohen's Kappa statistic was used to test for association between presence of alien riparian vegetation and palmiet. While density of alien riparian vegetation was assessed as number of stems per 100 m<sup>2</sup>, a single method of assessing density of alien riparian species could not be applied across all sites. In situations where density was relatively high, the Point-Centered Quarter Method (PCQM) was used as the preferred approach (Mueller-Dombois and Ellenberg 1974: 110). However, this was found to be difficult to use at relatively low densities, and here direct counts were made within an estimated area.

### **2.3.3 Aquatic macroinvertebrate sampling**

Sampling was undertaken at ten sites (Appendix 4.1) to collect data on aquatic macroinvertebrate community structure in spring (September 2013 and October 2014), summer (January 2014), and autumn (April 2014). Sampling had two objectives – species diversity and species density, as suitable quantitative biological metrics to assess impacts of land use on aquatic macroinvertebrates. All samples were taken from the best represented and most diverse hydraulic biotope, i.e. run/riffle areas. Initially two sampling methods were considered viz. Surber sampler, to provide a density estimate of 30x30 cm area, and stratified random sample of five stones from stones-in-current, as per the methods described by Rivers-Moore et al. (2007). The former approach has the disadvantage of not always being easy to undertake for different substrate sizes, but has the advantage of sampling all macroinvertebrates in the benthos including the hyporheic zone. The latter approach has the potential disadvantage of not sampling from the hyporheic zone, but has the advantage of being possible to carry out at any site. On the basis of the above, the second method of stratified random sampling of ten stones from stone-in-current was chosen as the preferred method. A total of ten stones were sampled per site for aquatic macroinvertebrates for the September 2013 survey, which was reduced to five stones for subsequent surveys. This was because ten stones were seen as adding little additional value to what five stones would add in terms of species diversity and community composition relative to time spent sorting and identifying samples. The sampling procedure was to select a fist-sized stone from the stones-in-current biotope, remove it from the river, and scrub it clean of all aquatic macroinvertebrates. At all stages of the sampling process, a 250 µm mesh net was used to retain the sample. All samples were cleaned and sorted to family-level. Surface area was estimated from length, breadth and height measurements of each stone sampled to allow for the calculation of density.

A once-off SASS (South African Scoring System, Version 5) assessment was also undertaken in October 2014 to provide an indication of any water quality impairment and general river health (Dallas 1997; Chutter 1998) at each of the sites where aquatic macroinvertebrates were sampled. An assessment of instream and habitat integrity (Kleynhans 1996, Dallas 2005) was also undertaken at all sites to provide an indication of the extent of instream versus riparian transformation.

## 2.4 Thermal thresholds and Maximum Weekly Allowable Temperature (MWAT) Exceedance

Thermal thresholds for *Lestagella penicillata*, family Teloganodidae, have been used as the basis for comparing frequency and duration of exceedance of thermal thresholds between sites. This species is considered to be of Gondwanan origin (Day 2005, Stevens 2009) with limited distribution and potentially unique thermal requirements. This species has been the focus of substantial research including and information is known on the life history, including developmental time and emergence, growth, nymphal and egg thermal limits, and genetics (Dallas et al. 2012, Dallas et al. 2015, Rivers-Moore et al. 2013b, Ross-Gillespie 2014).

The life history of *L. penicillata* is that of a slow seasonal cycle, with a total development time of 12 to 13 months, making it univoltine (Ross-Gillespie 2014). Emergence of adults is from October to December, with recruitment occurring from December to January (Ross-Gillespie 2014). Recent genetics research suggests that what has previously been considered to be a single species may indeed be a species complex although this research is ongoing.

Thermal limits and Maximum Weekly Allowable Temperature (MWAT) thresholds have been determined for seven sites in the Western and Southern Cape, and range from 18.3°C to 20.9°C, with 19.7°C and 20.9°C calculated for the 1Kaa1 and 5Tou2 sites respectively. The thermal range for growth based on field sampling and statistical analyses was between 13 and 21.5°C, and optimal thermal range for growth based on growth experiments was between 15 and 18°C. The thermal optimum for hatching was between 15 and 20°C (Ross-Gillespie 2014).

Thermal thresholds used in the exceedance analyses are as follows: 12.0°C as a minimum threshold, 27.8°C (LT<sub>50</sub>) for a maximum threshold, and 21.1°C for mean daily temperatures or MWAT. These thresholds have been used previously by Rivers-Moore et al. (2013b).

All thresholds have been compared against seven-day moving averages of daily water temperatures. The relationship between MWAT threshold exceedances and altitude was examined using a number of published studies for different thresholds. On this basis, MWAT threshold exceedances for the study sites were plotted against altitude, and a best-fit curve plotted together with 95% confidence intervals. Next, percent exceedances of expected MWAT values versus observed values were calculated at all nineteen sites. Finally, these percentage exceedances were plotted against alien density of riparian vegetation.

## **Chapter 3. Relationships between alien plant densities in the riparian zone and changes to water temperature regimes**

### **3.1 Introduction: Impacts of alien riparian vegetation on water temperatures**

Factors which impact on water temperatures include reductions in flow (surface and groundwater), degree of shading (especially in first or second order streams), channel modification (channel obstructions resulting in changes in hydraulic biotopes and/or changing the residency times of water within a reach), and changes to natural sediment loads. Alien vegetation in the riparian zone can impact on all the factors mentioned above, and climate change is likely to exacerbate the problem both directly through its amplification of thermal extremes in aquatic systems, and indirectly through its impacts on dispersal patterns of alien invasive vegetation. Changes to, *inter alia*, aquatic thermal regimes, will differentially impact different families of aquatic taxa (Dallas and Rivers-Moore 2011a), across the spectrum of functional feeding groups. For example, univoltine species (typically high conservation species) are likely to be negatively impacted by increases in thermal extremes, while multivoltine species (often pest species or generalists) are likely to benefit from the same thermal extremes (see Rivers-Moore et al. 2012b). It is highly likely that marked changes to riverine communities will alter ecosystem processes and ultimately on the delivery of ecosystem services.

### **3.2 Methods**

#### ***3.2.1 Flow and Thermal metrics***

Collection of flow data are described in Section 2.2.3. Sub-daily data for a full year (30 September 2013 – 31 August 2014) were converted to daily data (average, minimum, maximum). Daily time series for 1 January to 31 December 2009 were broken down into metrics to describe annual and monthly measures of magnitude, frequency, duration and timing of thermal events, as per the method described by Rivers-Moore et al. (2013).

#### ***3.2.2 Site classification and thermal reference conditions***

The full set of metrics was used to classify sites according to their thermal regimes. Next, we ran a Principal Components Analysis together with a cluster analysis to define thermal groups (McCune and Mefford 1999) using the group averaging agglomerative option and Euclidean distance as the distance measure.

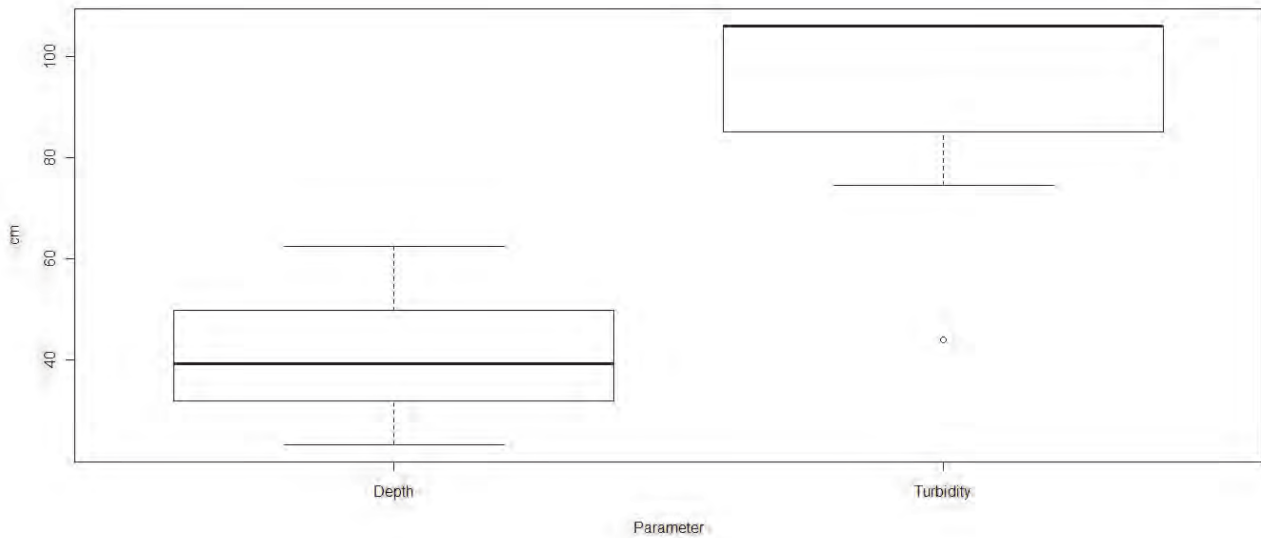
### **3.3 Results**

#### ***3.3.1 Water quality and thermal metrics***

All nineteen sites displayed remarkable consistency with respect to depth and turbidity values (Figure 3.1), with all sites being relatively shallow at time of sampling, and all tea-coloured but relatively clear (turbidity > 100 cm). River widths at all sites were comparable ( $4.77 \pm 1.57$  m), and

all sites fell within Stream Order 1 systems. Similarly, conductivity at all sites was in the region of  $150 \mu\text{S}/\text{cm}$ , typical of oligotrophic systems, although some seasonal variation was apparent (Figure 3.2) and a study sites could be characterised as acidic (Figure 3.2).

Water temperature regimes across all sites, and based on 12 months of data, were all relatively consistent (Figure 3.3), with upstream sites cooler than downstream sites.



**Figure 3.1** Box and whisker plot of depth and turbidity values across all 19 sites

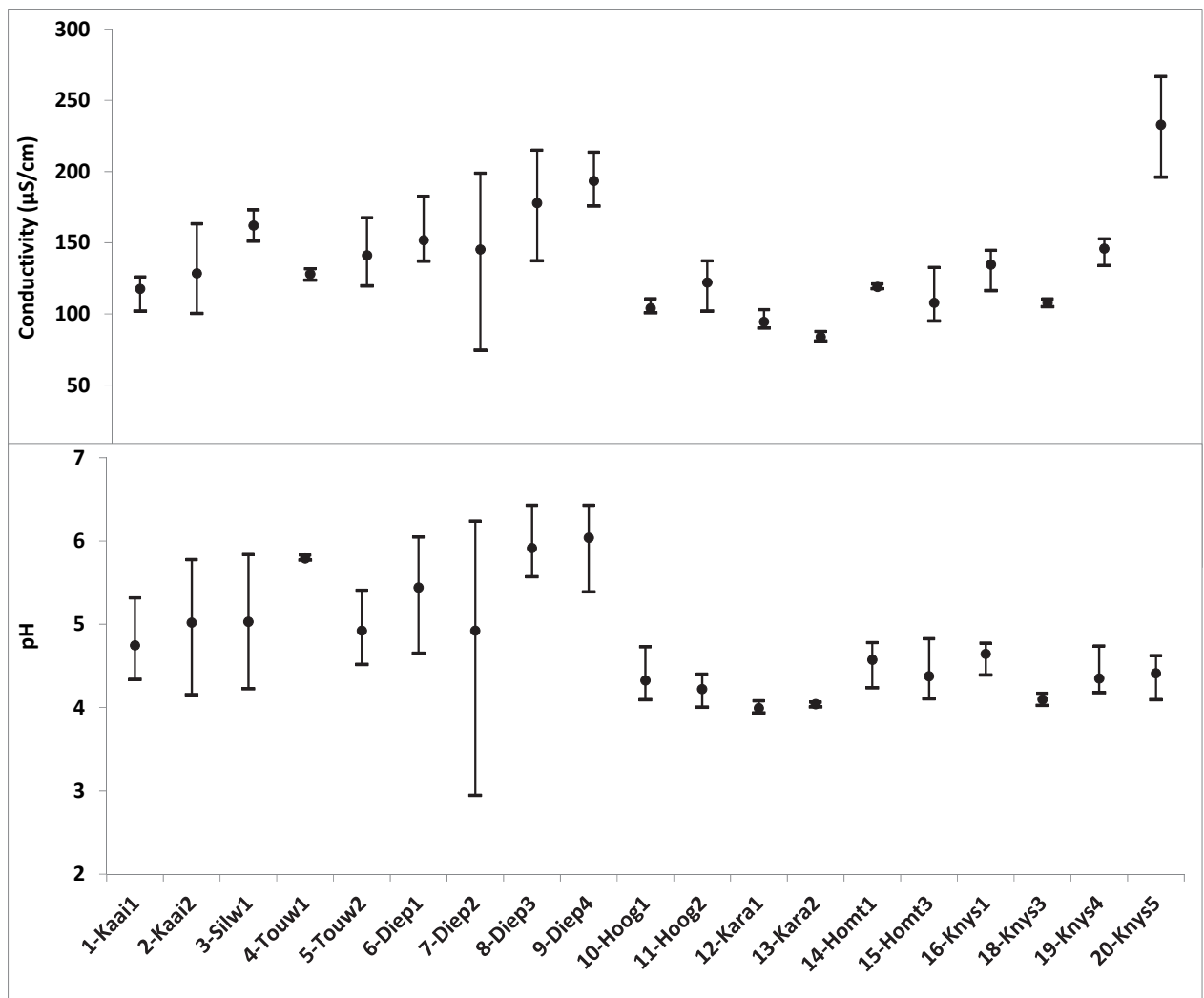
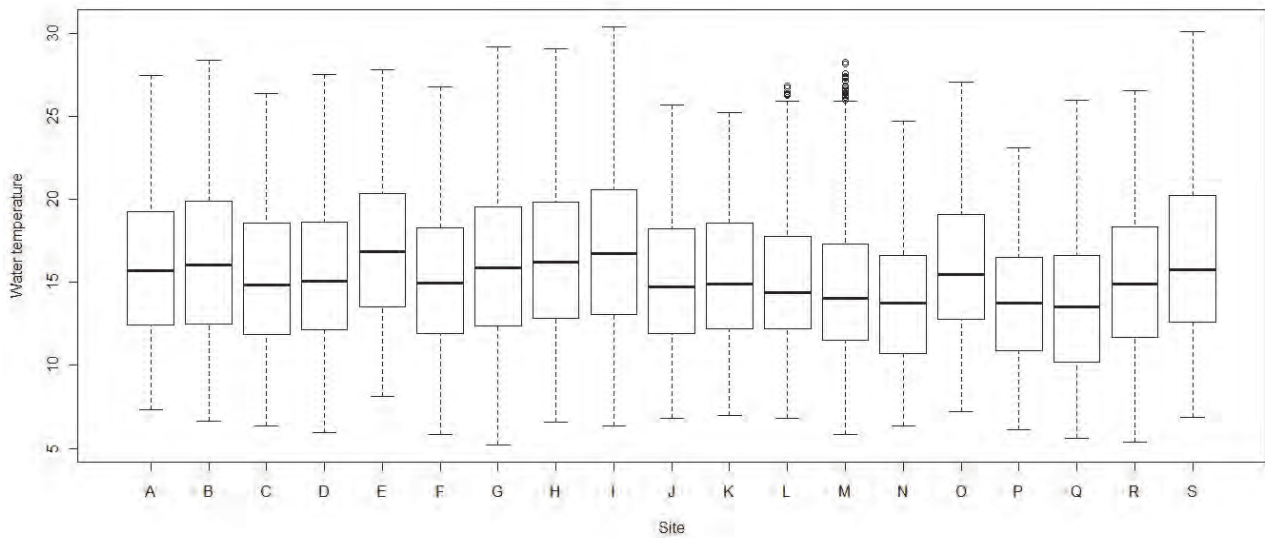


Figure 3.2 Range (as maximum, mean and minimum values) in conductivity and pH across 19 sites. Data for both parameters show two distinct groups, between the rivers to the west and east of the Hoogekraal River sites. The eastern sites were generally more acidic and more oligotrophic than the western sites. The eastern sites are dominated by quartzitic sandstone, with subordinate shale and siltstone, while the western sites are dominated by quartzitic sandstone, with minor conglomerate and shale in the headwaters.



**Figure 3.3** Box and whisker plot of hourly water temperatures from 1 September 2013 – 31 August 2014 across all 19 sites. The labels A-S correspond to the site names in Figure 3.6.

### 3.3.2 Site classification and thermal reference conditions

Using a cluster analysis together with a Principal Components Analysis, sites could be grouped on their thermal metrics into three clusters, viz. a more lowland group ( $n = 8$ ), an upper region group ( $n = 8$ ) and what appears to be a groundwater-influenced group ( $n = 3$ ) (Figures 3.4 and 3.5). Based on the eigenvectors for the PCA, Axis 1 represented a cooler to warmer thermal gradient (monthly, annual and cumulative mean values), while Axis 2 represented a gradient from lower to higher thermal variability and thermal extremes.

Lowland sites in particular exhibited altered thermal regimes, based on Euclidean distance differences of the thermal metrics in the PCA between observed and expected water temperatures. The lowland sites typically had higher densities of alien riparian trees, which included black wattle (*Acacia mearnsii*), blackwoods, (*A. melanoxylon*), and gum and pine trees. Densities ranged from zero to 48 trees per 100 m<sup>2</sup>. Data suggest a weak positive correlation between presence of alien riparian vegetation and palmiet (Cohen's Kappa = 0.41; agreement = 74%), with 58% (11) of the sites having both palmiet and alien riparian trees present, versus 26% (5) of the sites having alien riparian trees present and palmiet absent. Impacts of alien riparian trees on thermal conditions are discussed further in Section 6.3.2.



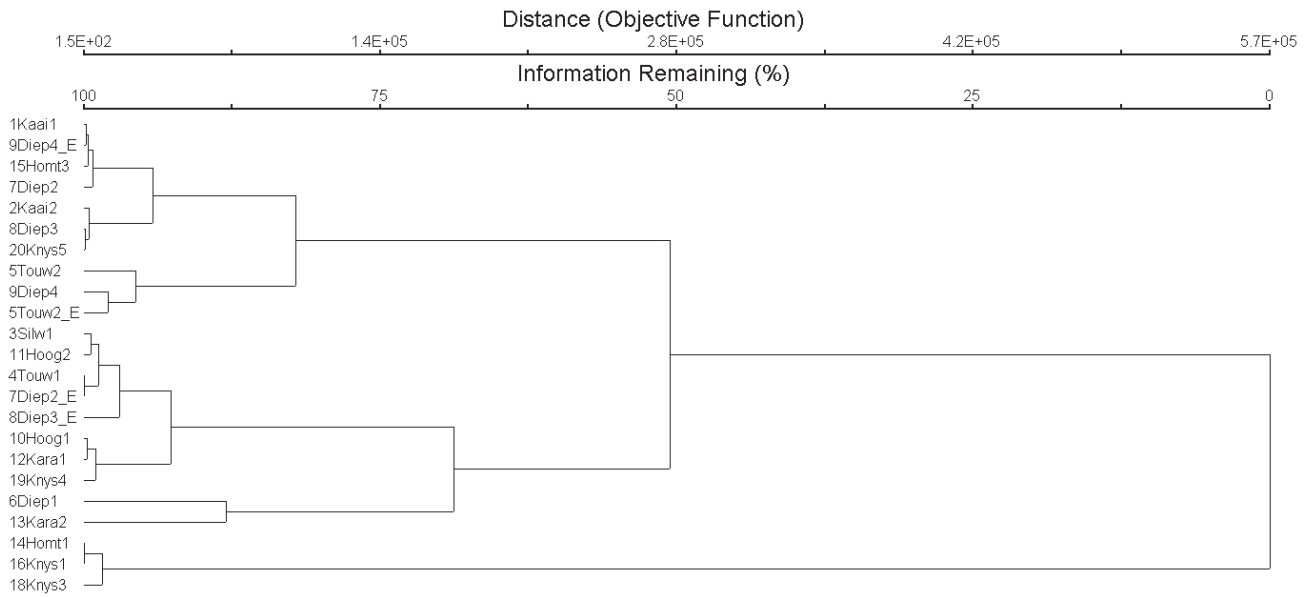


Figure 3.4 Cluster analysis of sites based on water temperature metrics

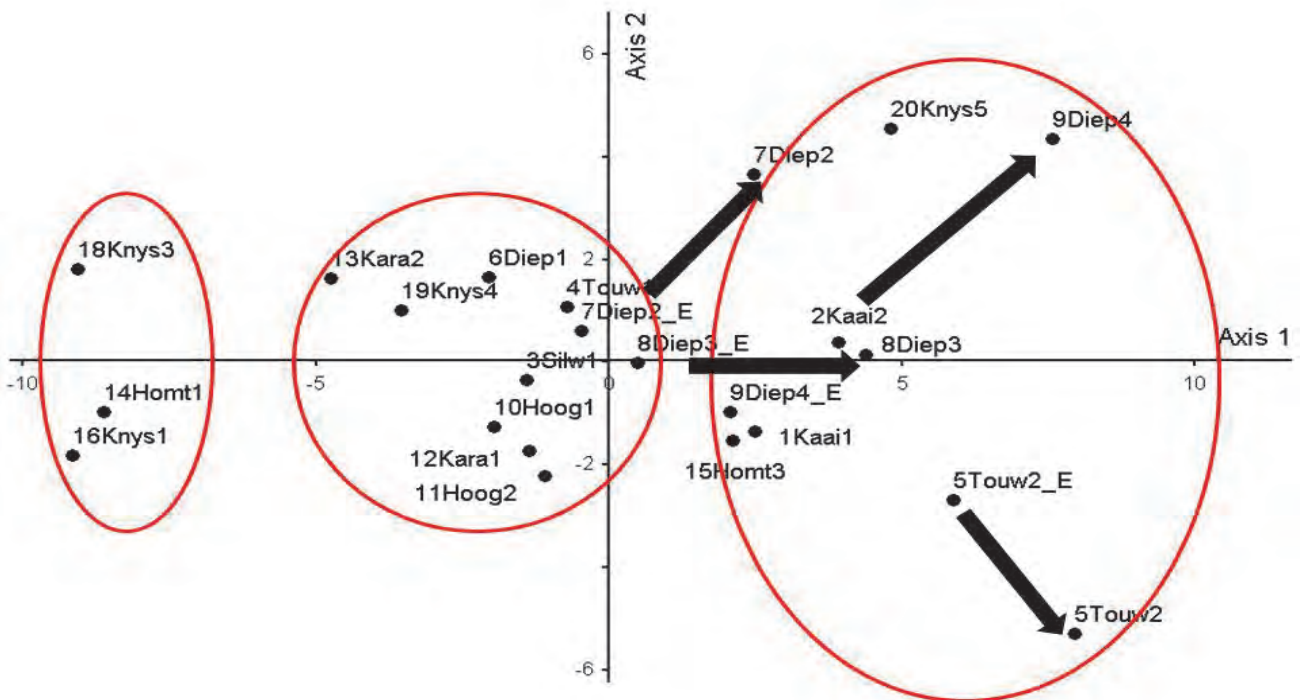
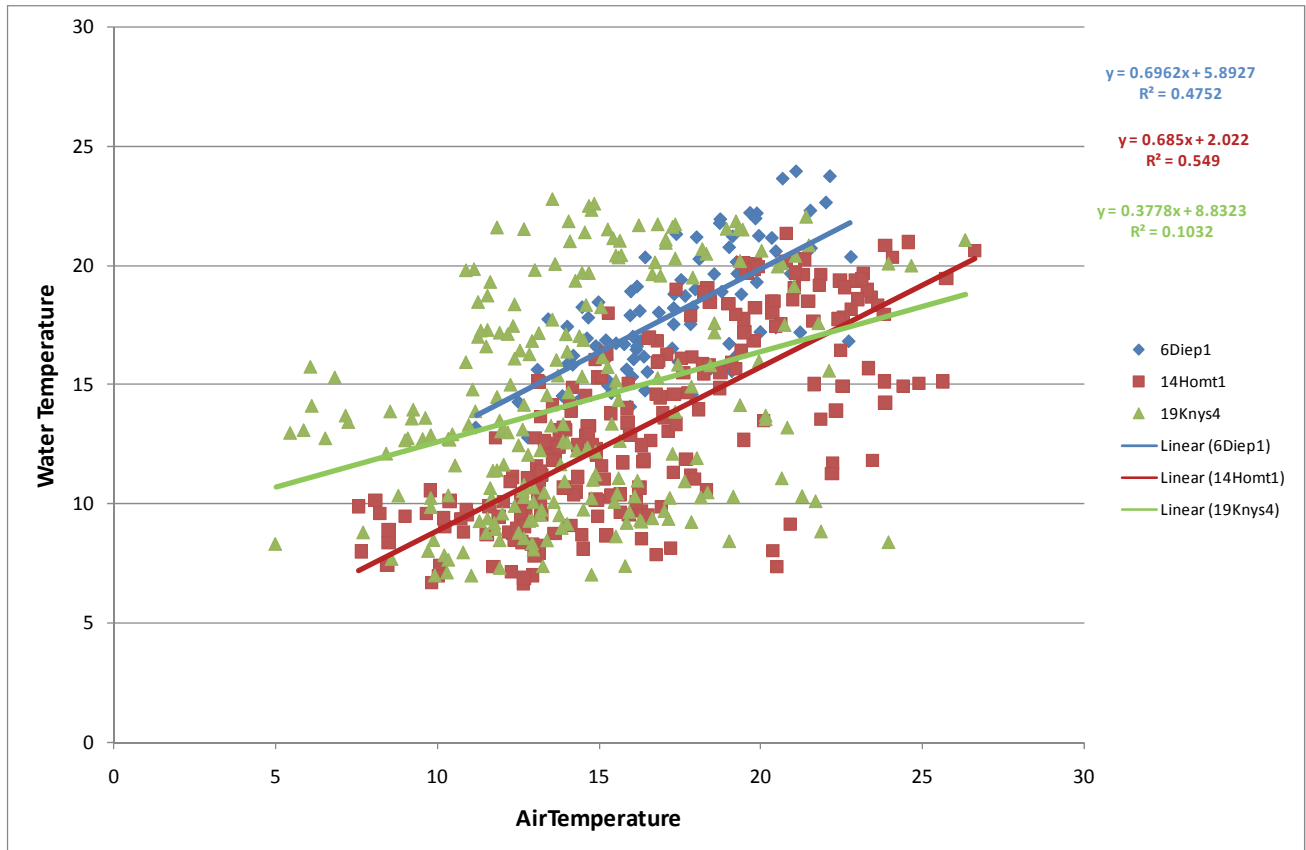


Figure 3.5 PCA plot of sites based on water temperature metrics. The relative position of sites based on estimated water temperatures (E) is also shown. Cumulative percentage of variance accounted for by first two axes were 70.87 and 81.93 respectively

As a means of extending the water temperature data to beyond the measured year through making use of longer-term air temperature data, linear regression relationships were developed between observed mean daily air and water temperatures at three study sites (Figure 3.6). These correlations were relatively weak, ranging from  $R^2$  values of 0.54-0.10.



**Figure 3.6 Correlations between mean daily air and water temperatures at three study sites**

### 3.3.3 Flow data analyses

Observed flow data exhibited high variability as a persistent pattern (Figure 3.7), with maximum daily flows being an order of  $10^2$  larger than mean daily flows (Table 3.1). However, these high flows are only present for more than 10-20% of the entire flow records, with the remainder of flow durations being less than  $1 \text{ m}^3\text{s}^{-1}$  (Figure 3.8). Flow patterns were different between each of the five rivers, based on flow metrics describing duration, timing, frequency and exceedance of flow events, and forming three distinct flow type groups (Figure 3.9).

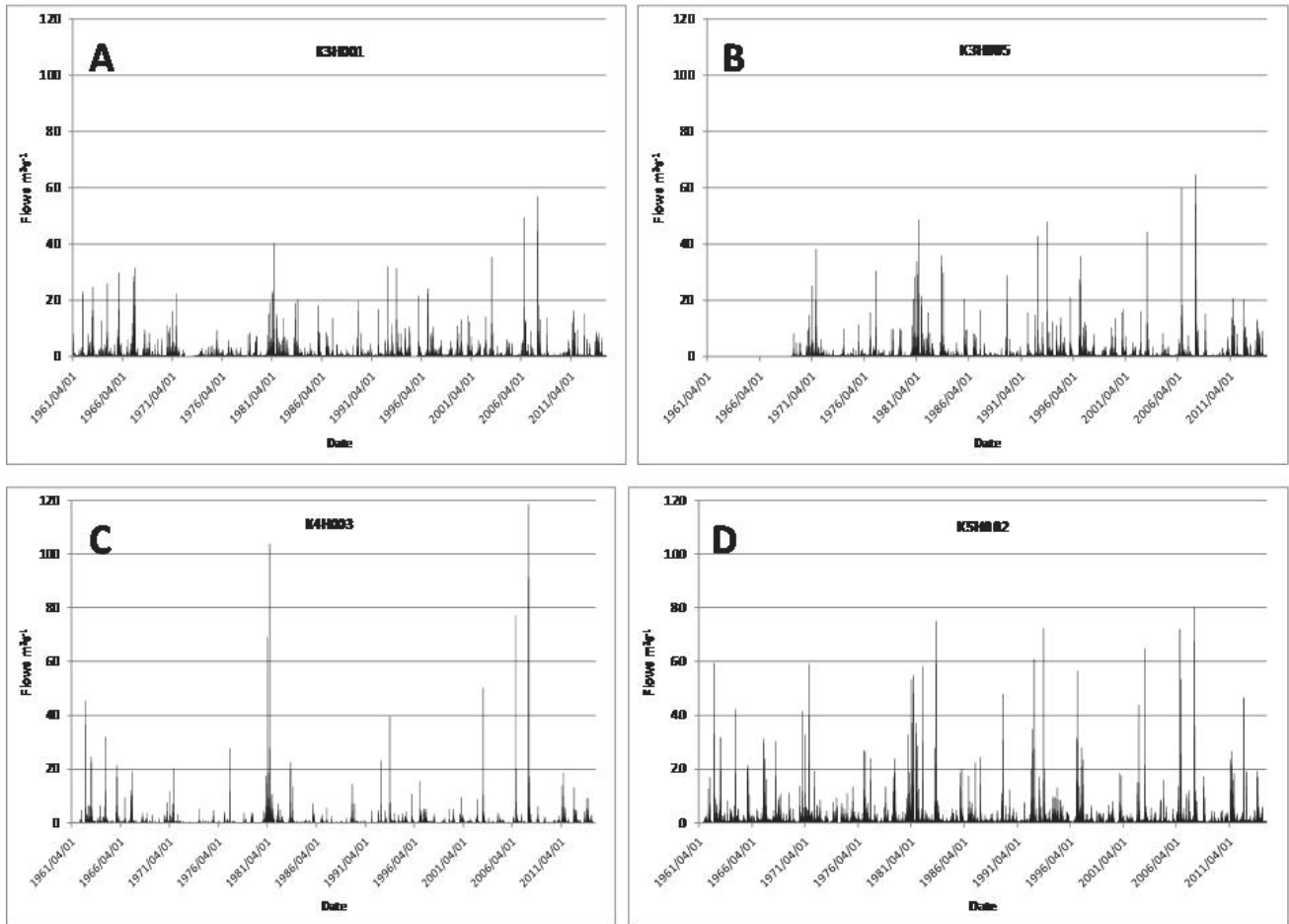


Figure 3.7 Daily hydrographs spanning more than fifty years for the Kaaimans (A), Touw (B), Diep (C) and Knysna (D) Rivers

Table 3.1 Descriptive flow statistics for five gauging weirs for the study area

|             | Kaaimans<br>K3H001 | Touw<br>K3H005 | Diep<br>K4H003 | Gouna<br>K5H001 | Knysna<br>K5H002 |
|-------------|--------------------|----------------|----------------|-----------------|------------------|
| <b>Mean</b> | 0.40               | 0.43           | 0.30           | 0.35            | 0.83             |
| <b>Min</b>  | 0.00               | 0.00           | 0.00           | 0.00            | 0.00             |
| <b>Max</b>  | 57.06              | 64.88          | 118.66         | 1.63            | 80.16            |
| <b>SD</b>   | 1.52               | 1.80           | 1.89           | 0.35            | 2.80             |
| <b>CV%</b>  | 381.37             | 421.55         | 635.04         | 100.91          | 335.20           |

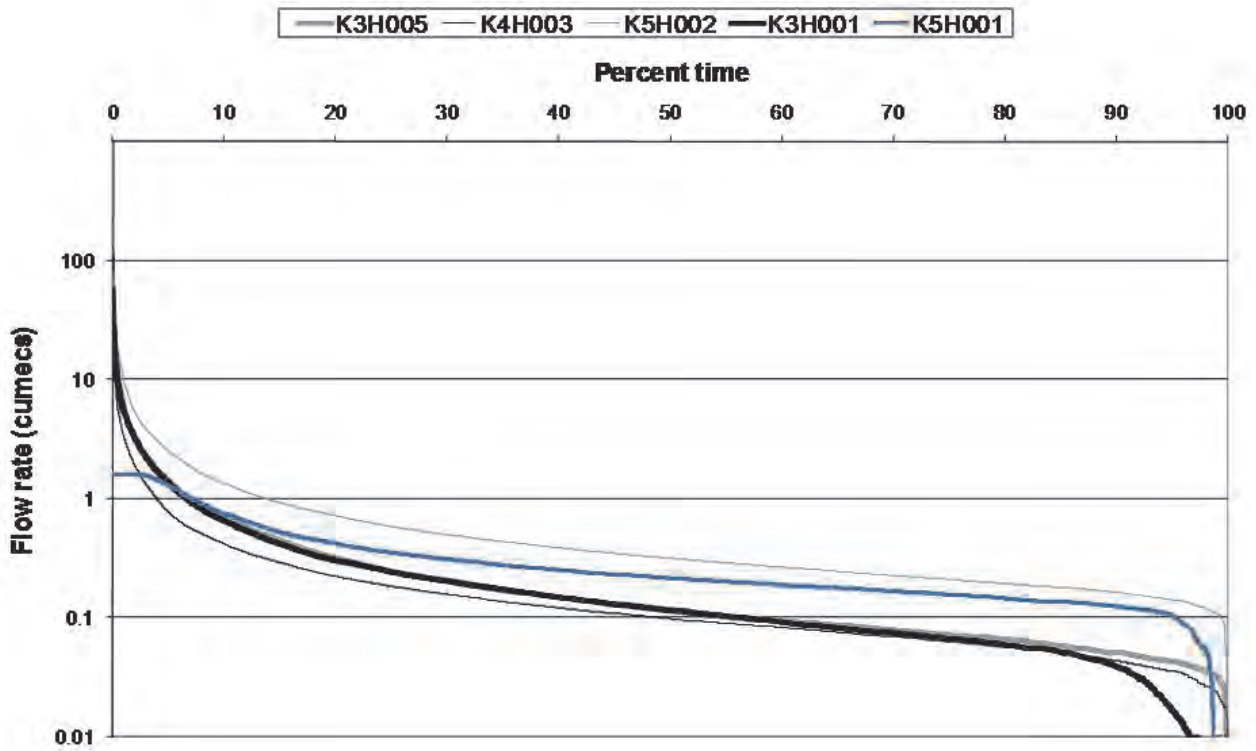


Figure 3.8 Flow duration curves for flow records from the five gauging weirs with good data available for the study area

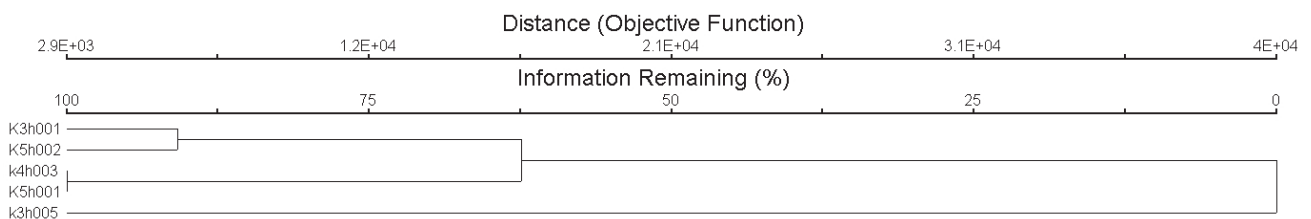


Figure 3.9 Cluster analysis showing groups flow types based on metrics from Richter et al. (1997)

### 3.4 Discussion

All sites were relatively comparable in terms of magnitude and geomorphology, as well as turbidity. Water quality (pH and conductivity) showed some seasonal variation, although all sites could be characterised as acidic and oligotrophic. However, these metrics did show a distinct west-east clustering in the study area, which is likely to be related to differences in upper catchment geology.

Based on one-year's hourly water temperature data, sites did not reflect strong thermal differences, with all sites relatively close to a median of 15°C. As expected, downstream sites were warmer than upstream sites, although given the short river lengths of the southern Cape Rivers, large differences in thermal gradients would not be expected. Multivariate analyses based on duration, frequency, timing and magnitude of thermal events showed a basic pattern of upstream and downstream thermal groups. Sites with shading from indigenous forest were cooler than

comparable fynbos sites, while there were also indications of groundwater impacts. Data also showed that downstream sites were more impacted than the upstream sites, which may be as a response to cumulative upstream catchment impacts. Densities of alien riparian vegetation increased with downstream distance, and such invasions may act as a surrogate predictor for cumulative upstream catchment degradation.

Simple linear regression models for predicting mean daily water temperatures from mean daily air temperatures showed weak correlations. Similar models have previously given much stronger correlations between air and water temperatures for other regions of South Africa (Rivers-Moore et al. 2005; Dallas and Rivers-Moore 2013). It is highly likely that the complex interactions between shading, groundwater, flow patterns, degree of incision and geology add considerable uncertainty to the basic air-water temperature relationship. This would argue for ongoing collection of long-term water temperature data.

Within the river systems assessed, the most overwhelmingly dominant characteristic was the extremely high variability of flows. It is highly likely that the southern Cape rivers of the western Garden Route Initiative region, and similarly the eastern regions (de Moor and Bellingham 2010) will function as event-driven systems. Overall levels of catchment degradation and densities of alien invasive trees over-and-above site-based densities is thus likely to be the issue of greatest concern, because of the cumulative upstream impacts on catchment runoff and stormflow response times.

## **Chapter 4. Relationships between alien plant densities in the riparian zone and changes to the structure of aquatic macroinvertebrate communities**

### **4.1 Introduction: Impacts of alien riparian vegetation on aquatic macroinvertebrate communities**

Alien vegetation is considered to have direct impacts on the water chemistry and structural and biological components of these ecosystems (Ractliffe et al. 2003). Alien vegetation in the riparian zone may affect the degree of shading (especially in first or second order streams, with potential for changes in algal abundance), channel structure (channel obstructions resulting in changes in hydraulic biotopes and/or changing the residency times of water within a reach), sediment loads and flow (alien vegetation is often more water thirsty than indigenous vegetation). Climate change is likely to exacerbate the problem both directly through its amplification of thermal extremes in aquatic systems, and indirectly through its impacts on dispersal patterns of alien invasive vegetation. Changes to, *inter alia*, aquatic thermal regimes, will differentially impact different families of aquatic taxa with some species “winners” and others “losers.”

Inputs of allochthonous detritus in afforested headwaters is considered to be the major energy source supporting invertebrate assemblages (e.g. Vannote et al. 1980; Cummins 1974). In rivers in fynbos areas, such features are relatively uncommon, given the prevalence of open-canopied headwaters, with generally low retention (Snaddon et al. 1991; Davies et al. 1993), however in forested streams this would be an issue. Whilst no studies have compared the retentiveness of natural streams with those invaded by woody aliens, woody plant species invading stream margins in fynbos biomes are associated with a greater quantity of debris and as a consequence, instream fauna may be altered (Ractliffe et al. 2003). Invasion by woody aliens may result in changes in the quantity, quality and timing of energy inputs into a river, as well as its retention within a reach, potentially leading to changes in the composition of aquatic communities (Ractliffe et al. 2003). For example, King (1982) noted that litterfall of alien woody vegetation was approximately double that from indigenous Afromontane riparian forest and leaf breakdown rates of alien vegetation by invertebrate shredders were three times those of indigenous vegetation. Sclerophyllous leaves from fynbos riparian vegetation have a low nitrogen content compared to alien vegetation such as *Acacia* species (King et al. 1987). This has obvious implications for water quality and instream biodiversity.

## **4.2 Methods**

### **4.2.1 Aquatic macroinvertebrates**

The relatively short project duration resulted in the invertebrate data only being identified to family level. Using family-level taxa per season, sites were compared based on total number of taxa, total number of individuals, evenness and diversity indices. These indices were correlated against site-level total density of alien riparian vegetation. Taxa from individual stones were combined per

season per site and community-level analyses were undertaken on family-level combined stones samples. Cluster analysis and non-metric multi-dimensional scaling (MDS) were used to gain an understanding of patterns in macroinvertebrate assemblages amongst sites. Data were transformed using fourth root transformation (PRIMER, Version 6; Plymouth Marine Laboratory, Plymouth, U.K., 2006) and the Bray-Curtis coefficient was used on these transformed data. Hierarchical agglomerative clustering, using group-average linking, was used on the data matrix. Stress values were used to assess the reliability of the MDS ordination. Analyses were undertaken for each sampling period separately and for all periods together, in addition to a combined site, data aimed to eliminate potential noise linked to season. The taxa responsible for 90% within-group similarity were determined using SIMPER (similarity percentages), which examines the contribution of individual taxa to the Bray-Curtis similarity measure. BIOENV was used to determine which environmental variables.

Qualitative samples of blackfly (Diptera: Simuliidae) and caddisfly (Trichoptera) were taken during October 2014, and identified to species level. Data were collected from five sites, viz. the Diep River (6Diep1), Gouna River (20Knys5 and 19Knys4), Jubilee Creek (14Homt1) and Millwood Stream (16Knys1). Species recorded were related to flow patterns and land use.

#### **4.2.2 Leaf litter decay**

Leaf litter was collected as per protocol (Hieber and Gessner 2002; Jackson et al. 2014) and air dried in air-conditioned room at about 25°C for a week. Approximately 3g of litter was weighed to 3 decimals on a Precisa 205A micro balance – leaves were added until the mass exceeded 3g. Samples were placed in small plastic mesh bags of approximately 10x10 cm closed with cable ties. The bags were constructed so that the ends were perpendicular to each other, i.e. they were three-dimensional, not flat, to prevent crushing of leaves. Each bag was individually marked with an aluminium tag. Twenty samples were deployed at each site, on two separate anchored steel wires ("lines"), i.e. 10 samples per line. After two weeks, 5 samples per line were removed, and after 4 weeks the remainder. Sample bags were rinsed off under a running tap and allowed to dry were dried in a room at 25°C for a week, before being weighed.

#### **4.2.3 Index of Habitat Integrity**

The Index of Habitat Integrity (IHI, Kleynhans 1999), which allows for an evaluation of the extent of instream versus riparian integrity, assesses the number and severity of anthropogenic perturbations on a river and the damage they potentially inflict on the habitat integrity of the system. These disturbances include abiotic factors, such as water abstraction, weirs, dams, pollution and dumping of rubble, and biotic factors, such as the presence of alien plants and aquatic animals which modify habitat. It should be noted that any site-based assessment will lack longitudinal continuity and therefore may not adequately reflect the habitat integrity of the river. Aspects considered in the assessment comprise those instream and riparian zone perturbations regarded as primary causes of degradation of a river ecosystem. The severity of each of these impacts is assessed, using scores as a measure of impact.

## 4.3 Results

### 4.3.1 *Aquatic macroinvertebrates*

A total of 4556 individuals for spring 2013, 1725 individuals for summer, 2230 for autumn, and 3401 for the spring 2014 were collected, representing a total of 43 families (Appendix 4.1). In general, the spring samples had higher numbers of individuals and families than the summer and autumn samples. Whilst these results are based on family-level data it is likely that similar trends would be evident using a finer taxonomic resolution.

The numbers of taxa were generally higher in the two spring periods and lowest in summer, with the exception of 15Homt3 and 19Knys4, whilst the total number of taxa were highest in spring at 1Kaai1, 6Diep1 and 14Homt1 (Figure 4.1). Both evenness and diversity were generally consistent across sites and seasons, although both 1Kaai1 and 6Diep1 had a substantially lower diversity index in summer, while 14Homt1 was lower in the spring periods (Figure 4.2). There was no clear relationship between numbers of taxa, evenness or diversity with density (Figure 4.3).



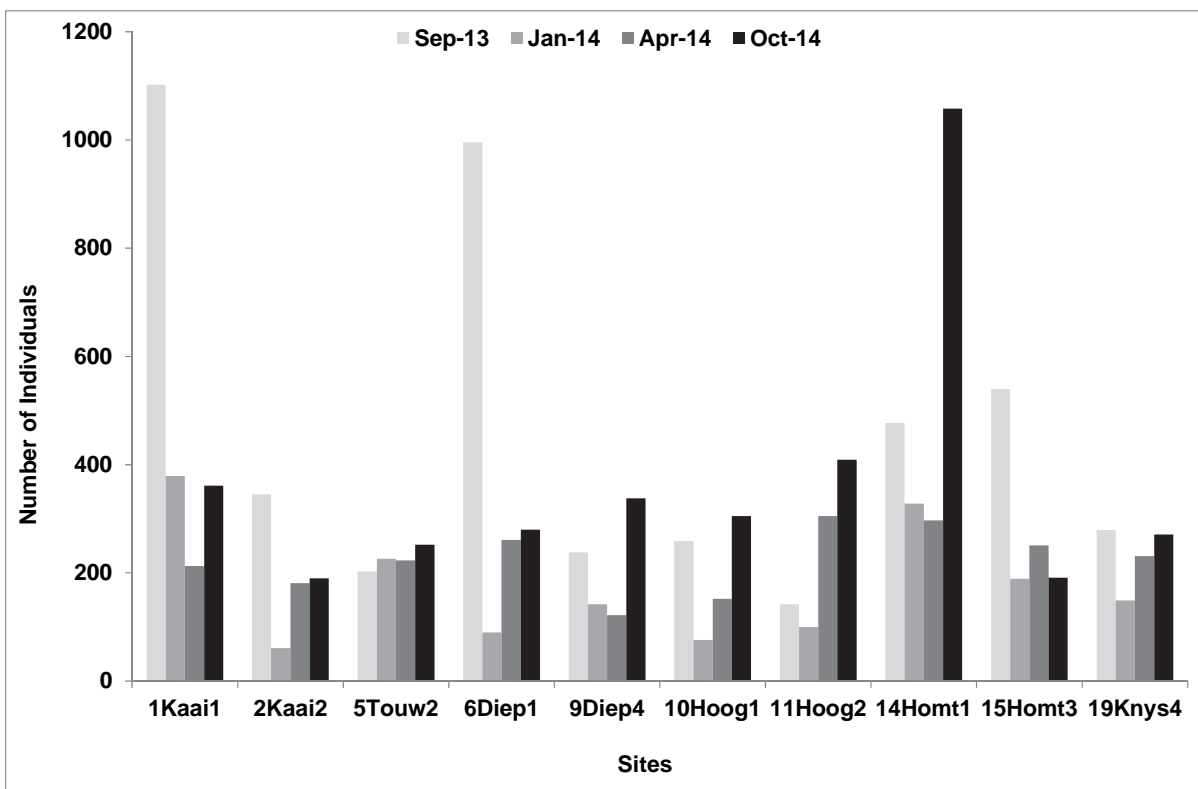
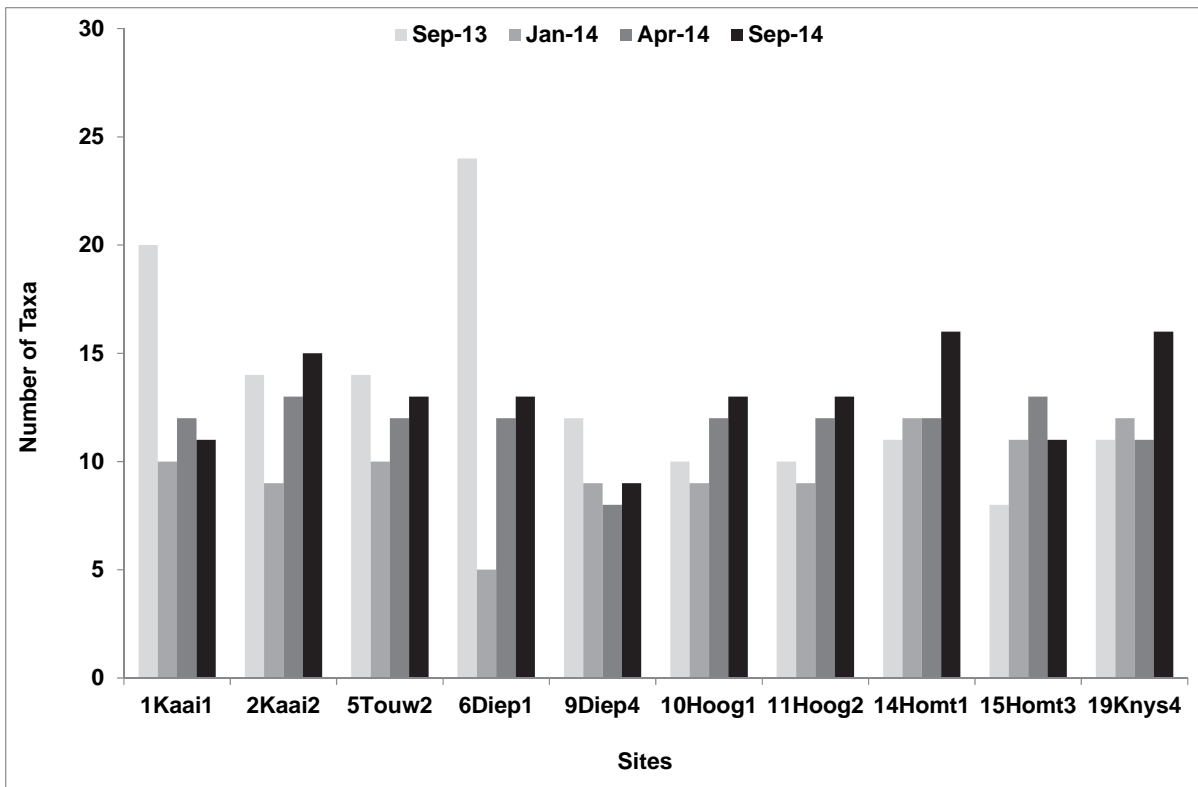


Figure 4.1 Number of taxa and total number of individuals per site per season

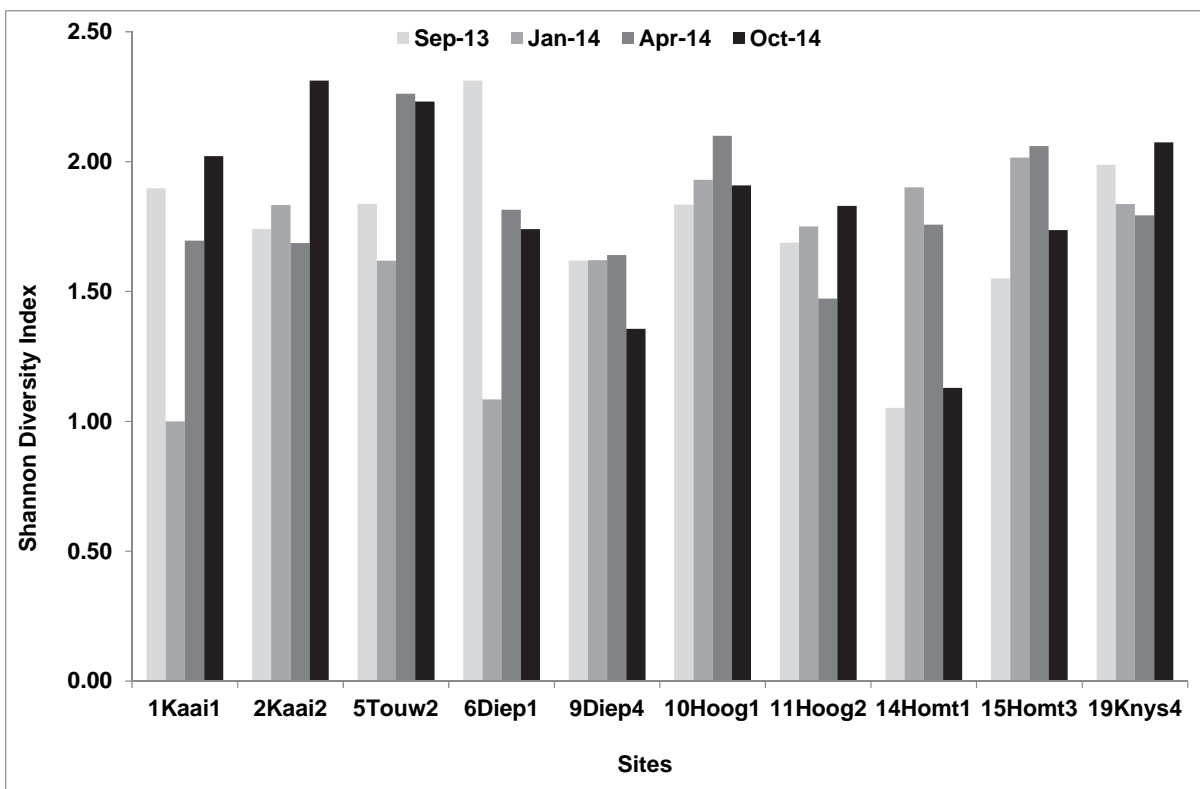
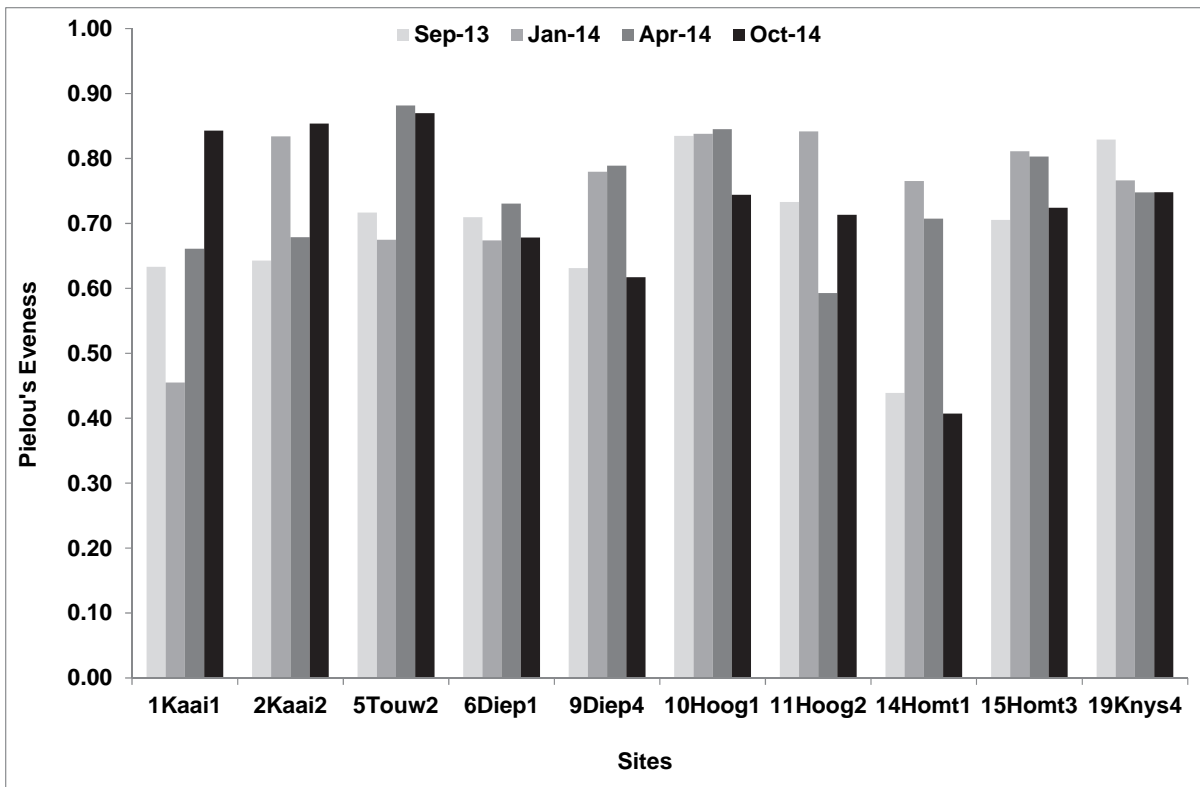


Figure 4.2 Evenness and diversity per site per season

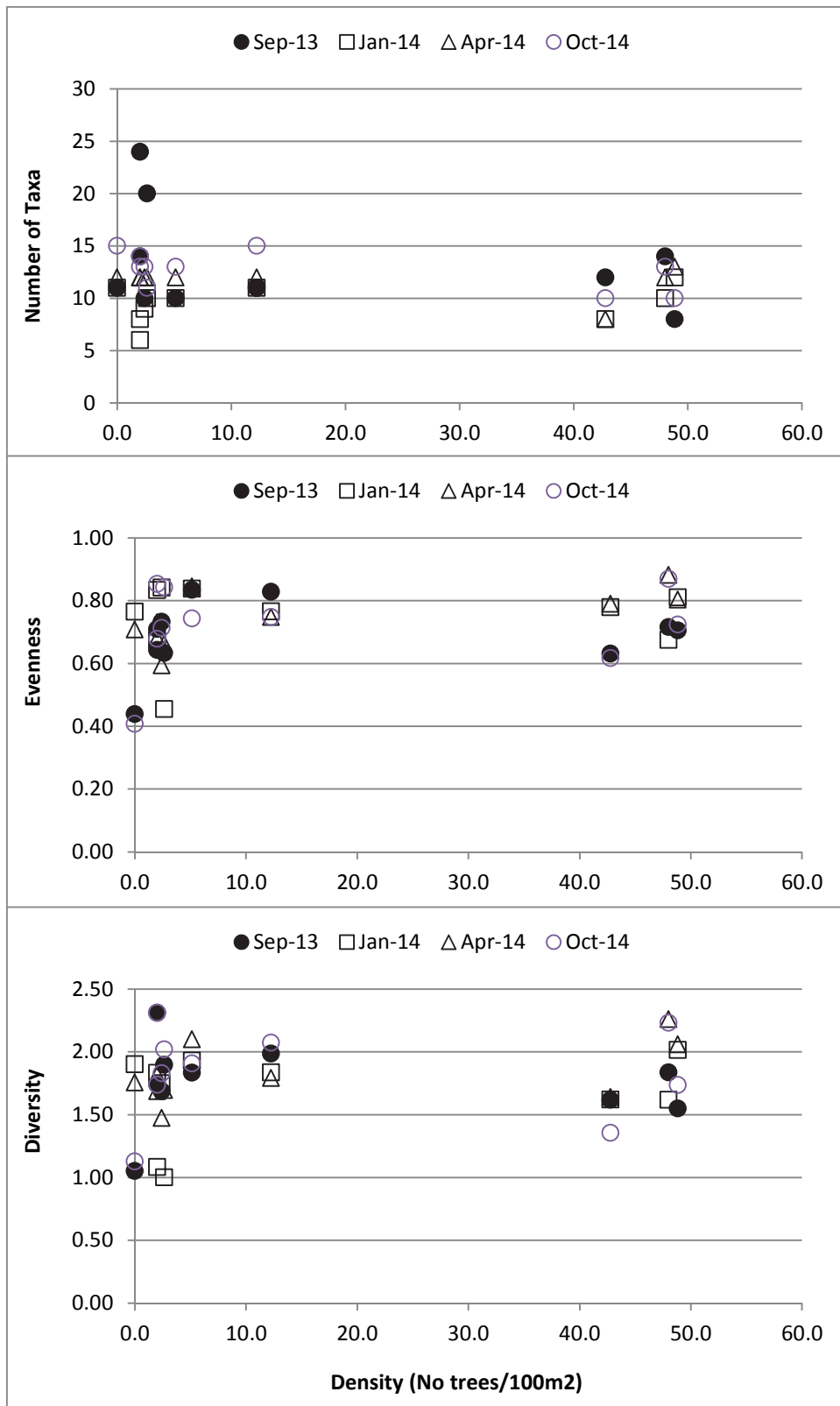


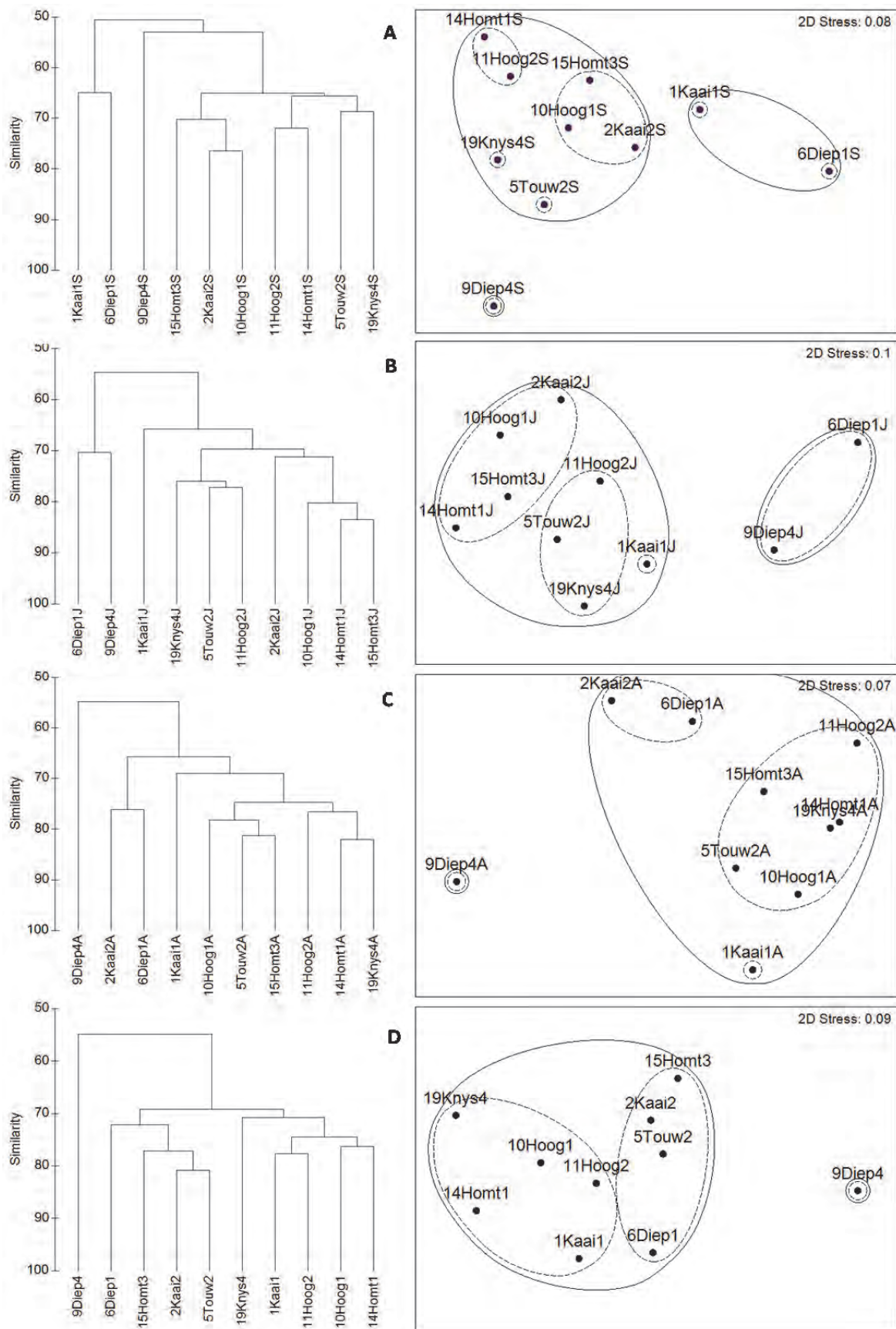
Figure 4.3 Relationship between numbers of taxa per site, evenness and diversity across four seasons and on-site density of alien riparian vegetation.

Cluster and ordination of macroinvertebrate communities undertaken for each season separately revealed that 9Diep4 was 50% to 60% dissimilar from other sites which grouped, except for summer when it grouped with 6Diep1 (Figure 4.4). Other sites formed groups, which were approximately the 70% similarity level although group membership differed amongst seasons. Examination of data for all seasons, coded by season (Figure 4.5), alien density (Figure 4.6) and river zone (Figure 4.7), confirms the separation of 9Diep4 from other sites, as well as the 6Diep1, 1Kaai1 and 2Kaai2 group. The remaining sites were at least 63% similar, and sites did not appear to group based on season, alien density or river zone. Lastly, when invertebrates recorded in each season were combined to generate a site level invertebrate assemblage, both 9Diep4 and 6Diep1 were distinct from each other and all other sites, 1Kaai1 and 2Kaai2 grouped together (73% similar), and the remaining sites formed a group approximately 72% similar (Figure 4.8). Within this group there appeared to be some distinction linked to alien density, with two of the sites with the highest densities grouping together (5Touw2 and 15Homt3). Simper analysis was used to determine which taxa did not reveal any taxa distinct to either one of the groups.

Environmental variables (Table 4.1) that best account for the ordination space of the invertebrate fauna included altitude, conductivity, pH, annual temperature range and mean of annual maximum temperature.

The average density of invertebrates on the stones varied substantially from 168 to 3102 m<sup>2</sup> with lowest densities in Summer and Autumn (averages of 445 and 569) compared to Spring 2013 and 2014 (1006 and 951).

SASS5 Scores for 11 sites (eight invertebrate sites, one additional temperature site, 13Kara2, and two sites for which SASS Scores were derived using benthic samples) varied from 138 (2Kaai2) to 59 (9Diep4) and ASPT values from 10.3 (5Touws2) to 6.6 (9Diep4), although most were within close range of each other, suggesting that water quality and river health was similar amongst sites (Table 4.2, Figure 4.10). The exception was 9Diep4, which was the only bedrock dominated system sampled and thus it was likely to have fewer stones habitat available for habitation.



**Figure 4.4** Cluster and MDS ordination plots for each sampling period: **A** – September 2013, **B** – January 2014, **C** – April 2014 and **D** – September 2014. Sites that are at least 60% (—) or 70% (----) similar are grouped.

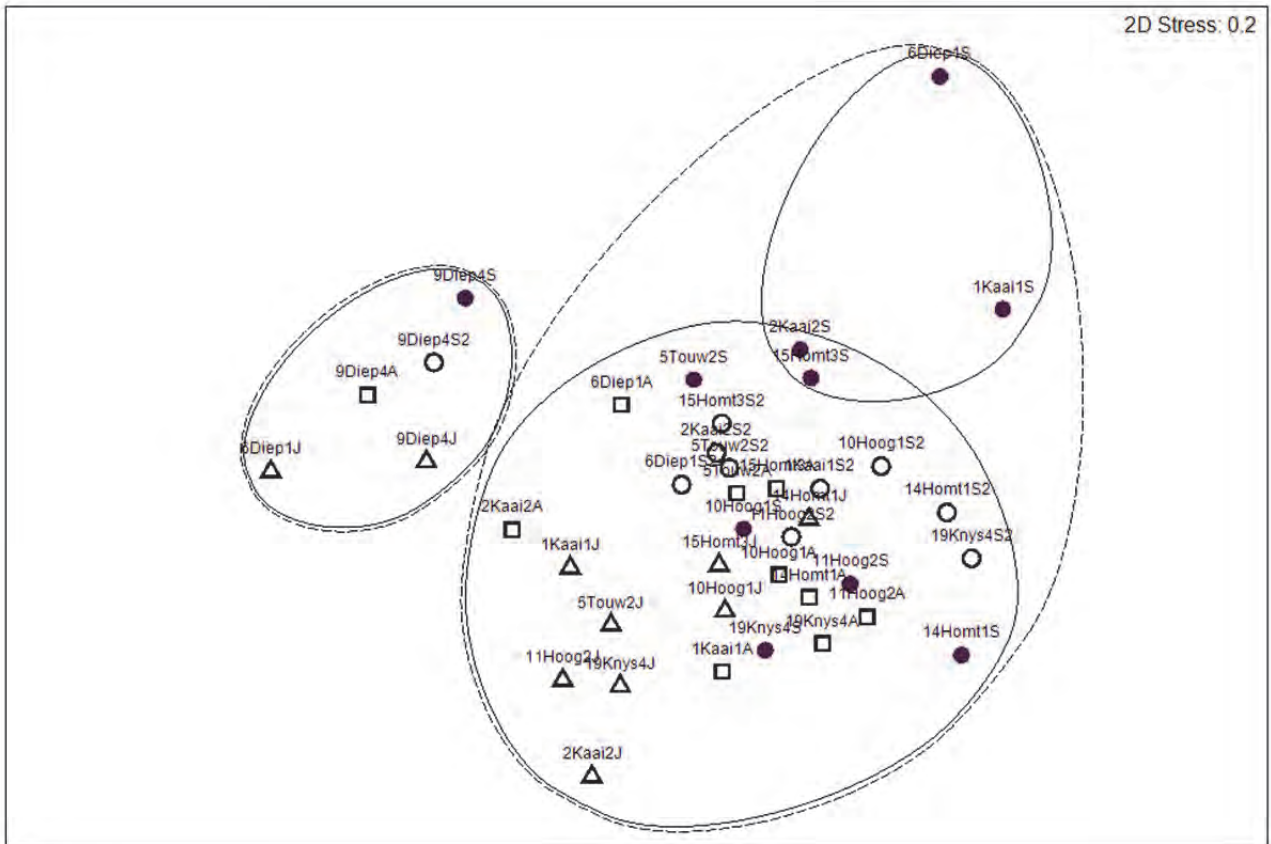
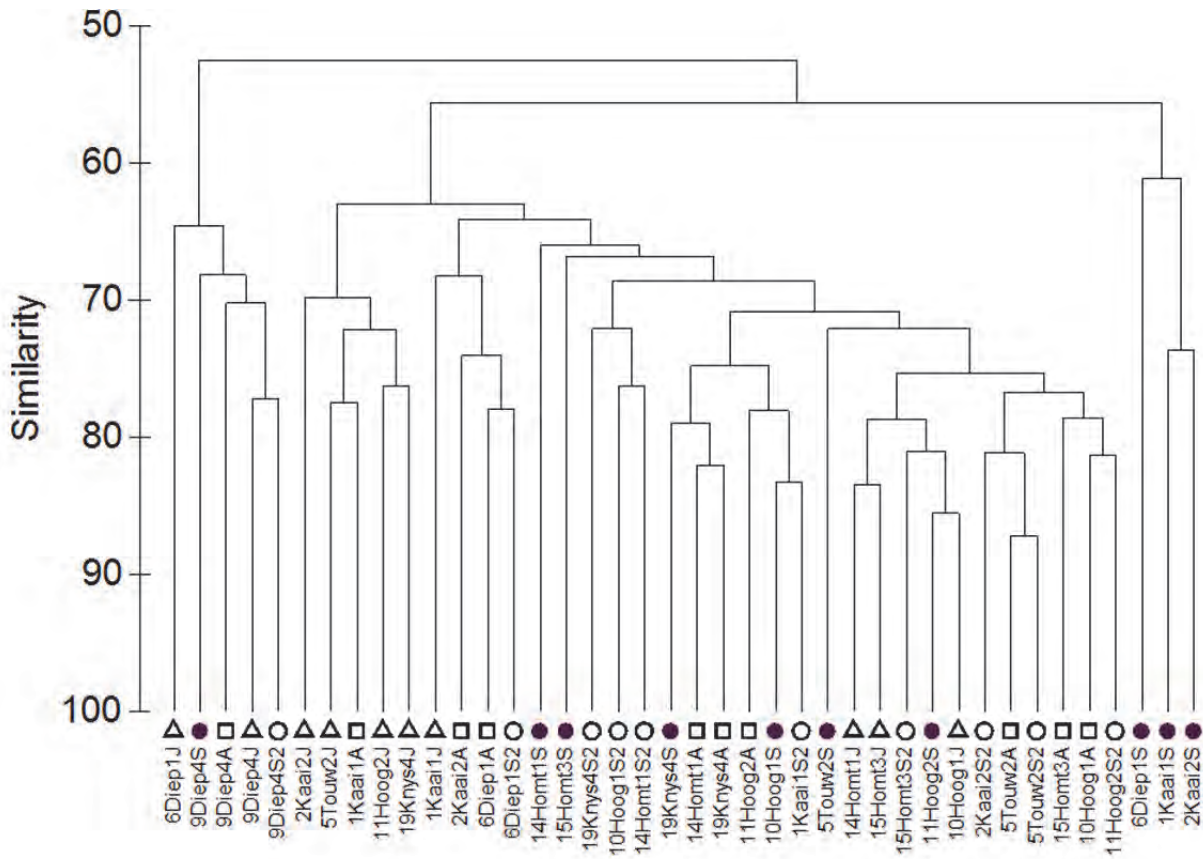


Figure 4.5 Cluster and MDS ordination plots for all sampling periods coded by sampling period (● : Spring 2013, Δ : Summer 2014, □ : Autumn 2014 and ○ : Spring 2014). Sites that are at least 55% (—) and 65% (---) are grouped.

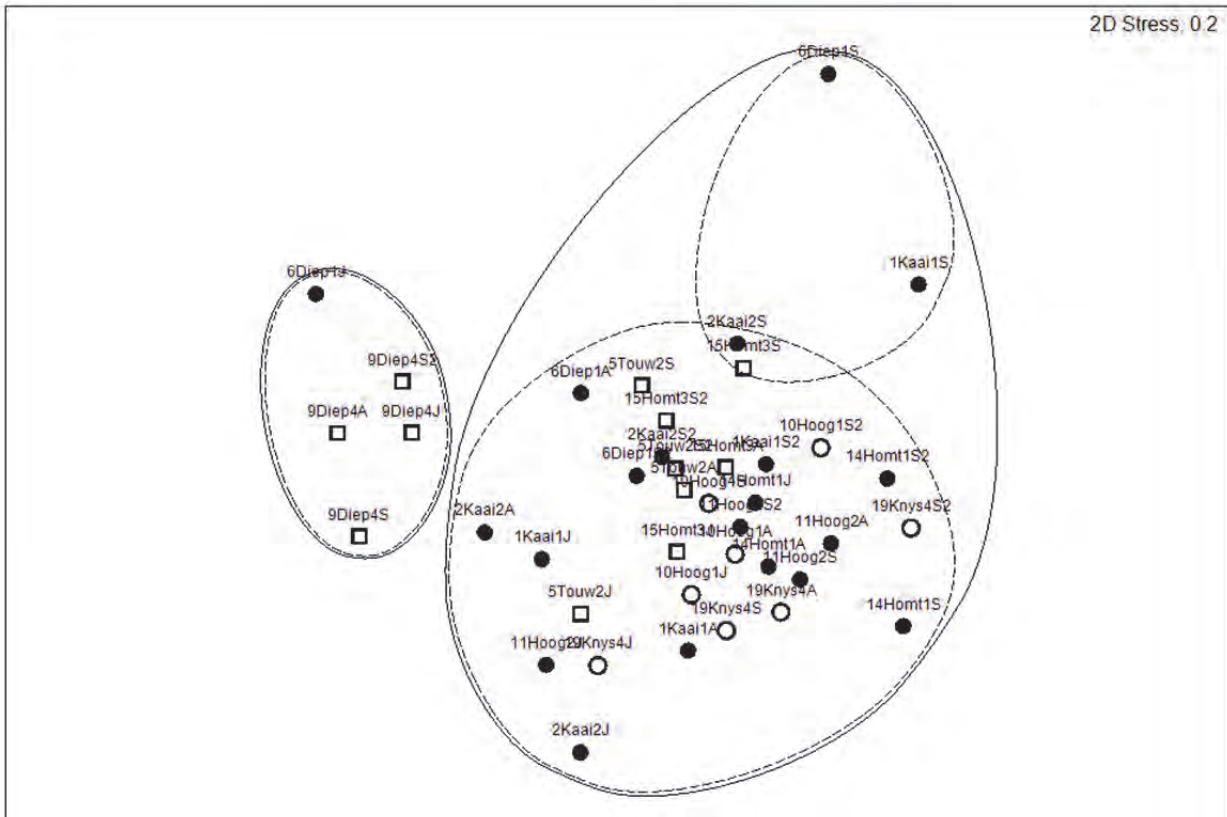
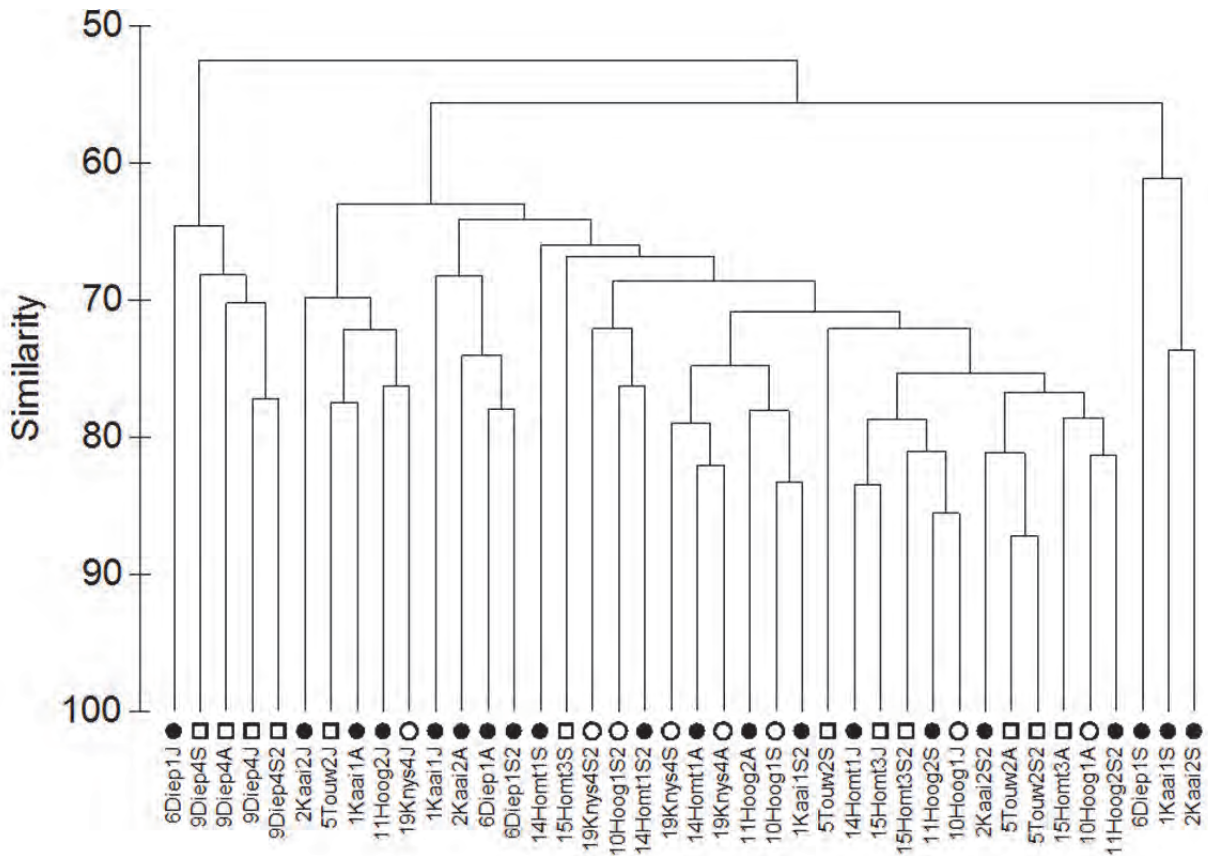


Figure 4.6 Cluster and MDS ordination plots for all sampling periods coded by alien density (Number of trees per 100 m<sup>2</sup>) on a scale of (● : Low: <5, □ : Medium: 5-30 and ○ : High: >30). Sites that are at least 55% (—) and 65% (---) are grouped.

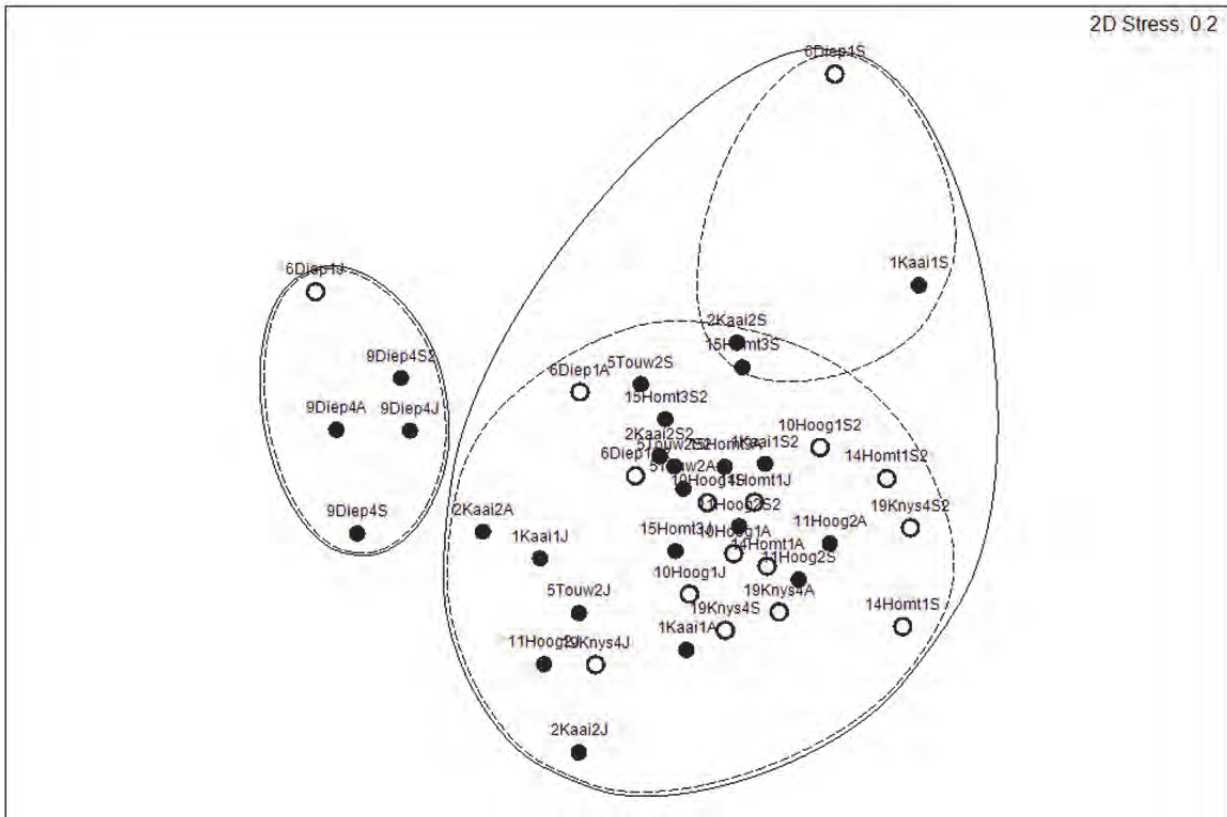
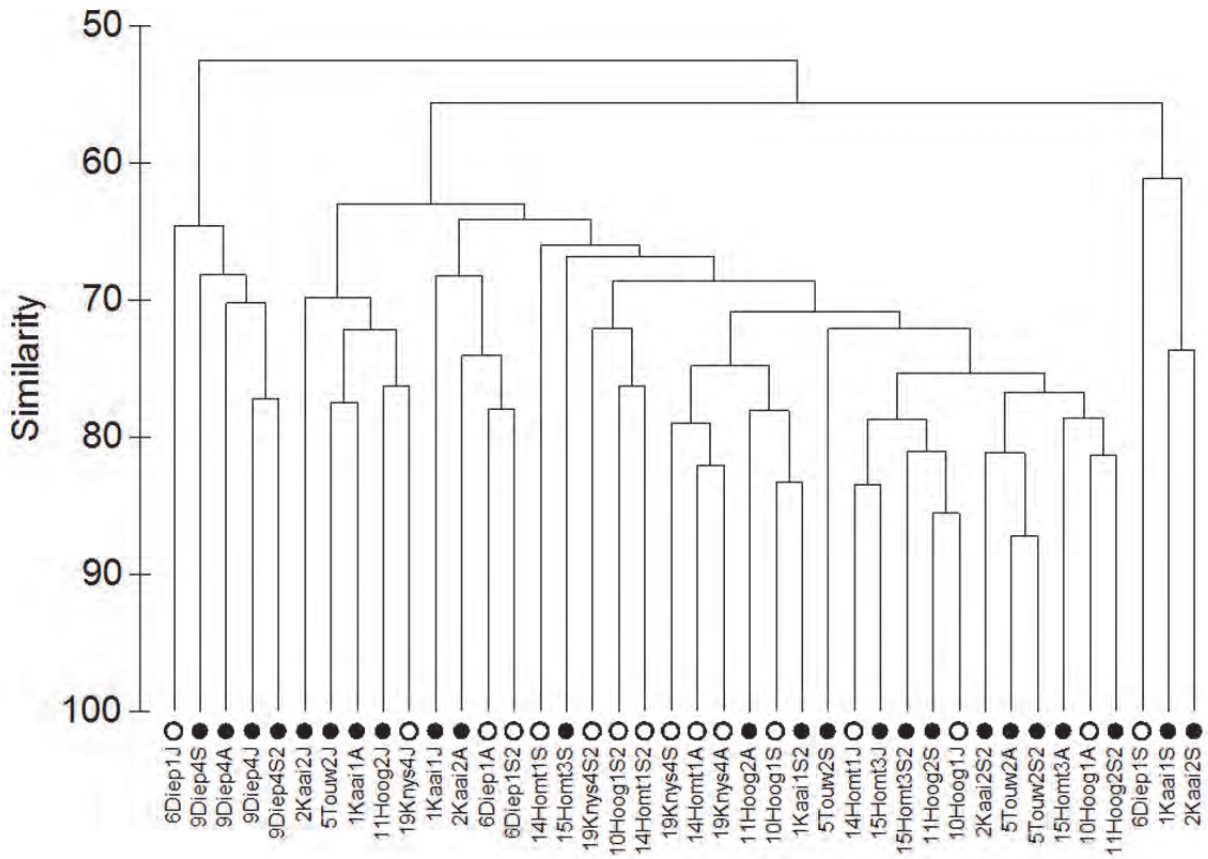


Figure 4.7 Cluster and MDS ordination plots for all sampling periods coded by river zonation (○ : Upper, ● : Lower). Sites that are at least 55% (—) and 65% (---) are grouped.



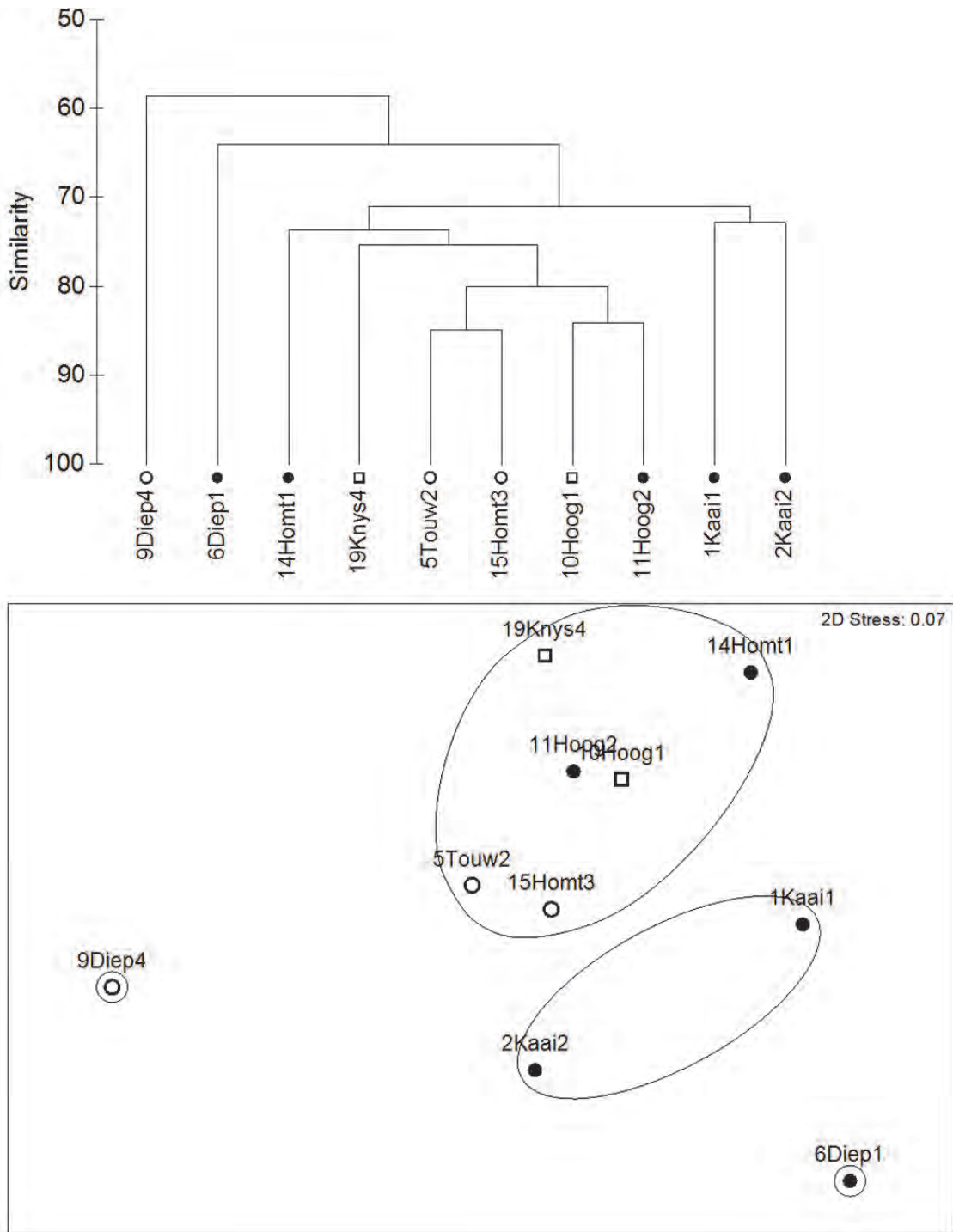


Figure 4.8 Cluster and MDS ordination plots for all sampling periods where invertebrates recorded in each season have been combined and coded by alien density (Number of trees per 100 m<sup>2</sup>) on a scale of (● : Low: <5, □ : Medium: 5-30 and ○ : High: >30). Sites that are at least 72% (—) are grouped.

**Table 4.1 Environmental variables used in analyses of BIOENV (Primer 2006)**

| Sites                      | 1Kaa1  | 2Kaa2  | 5Touw2 | 6Diep1 | 9Diep4 | 10Hoog1 | 11Hoog2 | 14Homt1 | 15Homt3 | 19Knys4 |
|----------------------------|--------|--------|--------|--------|--------|---------|---------|---------|---------|---------|
| Altitude (m)               | 137    | 94     | 190    | 313    | 182    | 269     | 141     | 315     | 46      | 442     |
| Alien Density              | 2.6    | 2.0    | 48.0   | 2.0    | 42.8   | 5.1     | 2.4     | 0.0     | 48.8    | 12.2    |
| Average Stream Width (cm)  | 420    | 420    | 830    | 392    | 800    | 479     | 700     | 610     | 706     | 364     |
| Average Stream Depth (cm)  | 31     | 52     | 33     | 48     | 39     | 63      | 62      | 33      | 56      | 36      |
| Conductivity ( $\mu$ S/cm) | 117.58 | 128.50 | 141.08 | 151.75 | 193.33 | 104.00  | 122.08  | 118.92  | 107.75  | 145.83  |
| pH                         | 4.75   | 5.02   | 4.92   | 5.44   | 6.04   | 4.33    | 4.22    | 4.57    | 4.38    | 4.35    |
| Turbidity (cm)             | 106    | 74.5   | 106    | 90     | 106    | 80      | 90      | 106     | 106     | 106     |
| Algae Present*             | 0      | 0      | 0      | 1      | 0      | 1       | 1       | 1       | 0       | 1       |
| Palmiet present*           | 1      | 1      | 1      | 0      | 1      | 0       | 1       | 0       | 1       | 1       |
| Mean Annual Temperature    | 15.9   | 16.2   | 16.9   | 15.1   | 16.7   | 15.1    | 15.3    | 13.8    | 15.9    | 15.0    |
| Annual CV                  | 26.8   | 28.9   | 25.7   | 27.0   | 28.5   | 27.3    | 27.6    | 26.5    | 27.7    | 27.8    |
| Predictability (Colwell)   | 0.63   | 0.60   | 0.60   | 0.59   | 0.58   | 0.60    | 0.63    | 0.67    | 0.61    | 0.66    |
| Mean of annual range       | 2.42   | 2.10   | 1.85   | 2.88   | 2.87   | 1.41    | 1.08    | 1.54    | 1.83    | 1.83    |
| Degree days                | 2147.9 | 2254.8 | 2507.6 | 1878.7 | 2444.9 | 1856.2  | 1929.8  | 1401.6  | 2155.1  | 1825.9  |
| Mean of annual minima      | 14.7   | 15.2   | 16.1   | 13.8   | 15.4   | 14.4    | 14.8    | 13.1    | 15.0    | 14.2    |
| Mean of annual maxima      | 17.1   | 17.3   | 17.9   | 16.7   | 18.3   | 15.8    | 15.8    | 14.7    | 16.8    | 16.0    |

\* 0 = No, 1 = Yes

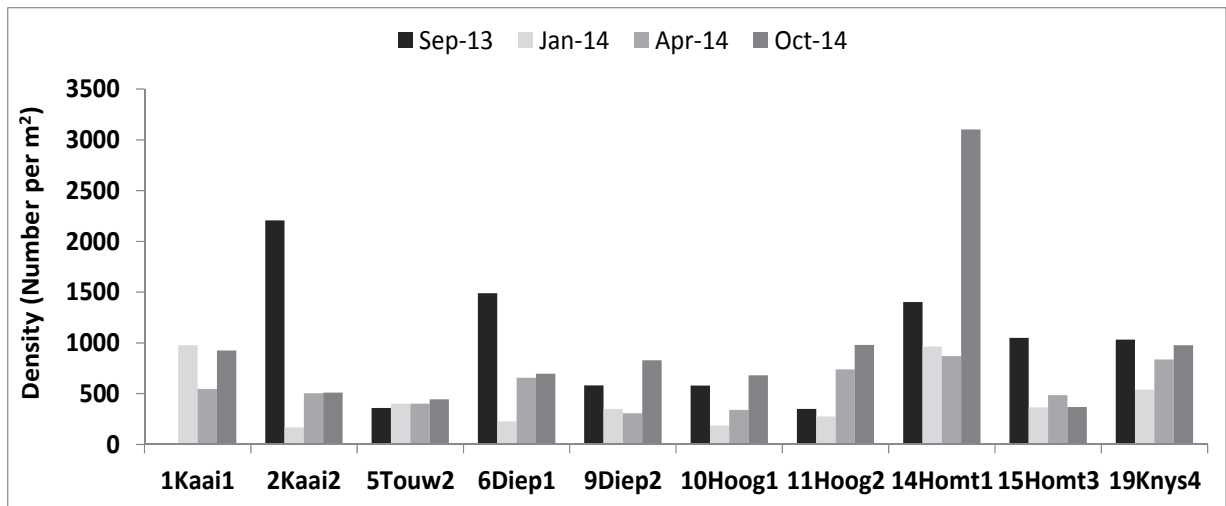


Figure 4.9. Density of invertebrates for each season and site

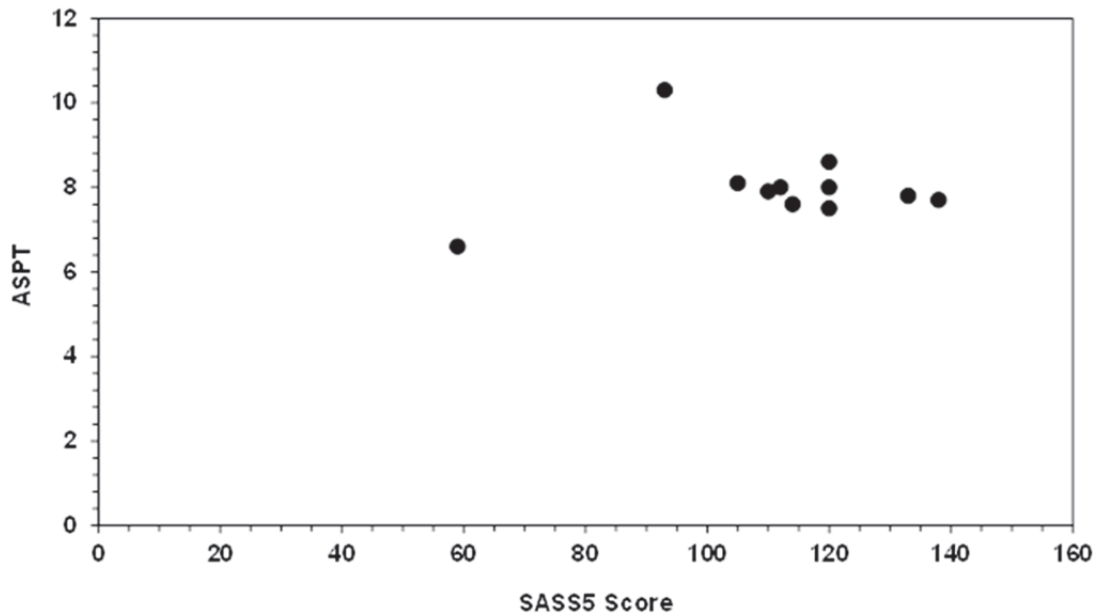


Figure 4.10 ASPT (Average Score Per Taxon) Versus SASS5 Score for 11 sites (9 + 2 generated) assessed in October 2014.

**Table 4.2 SASS5 Scores, Number of Taxa and ASPT values per site (all biotopes combined) and per stones biotope only for sites in the Southern Cape. The number of biotopes sampled is indicated and the alien density is indicated, where L = Low, M = Medium and H = High.**

| Site    | Alien density | Biotopes sampled | All biotopes combined |              |           | Stones biotope only |           |        |
|---------|---------------|------------------|-----------------------|--------------|-----------|---------------------|-----------|--------|
|         |               |                  | SASS5 Score-Site      | No Taxa-Site | ASPT-Site | SASS5 Score-S       | No Taxa-S | ASPT-S |
| 1Kaai1* | L             | S                | 105                   | 13           | 8.1       | 105                 | 13        | 8.1    |
| 2Kaai2  | L             | SG               | 138                   | 18           | 7.7       | 104                 | 12        | 8.7    |
| 5Touw2  | H             | S                | 93                    | 9            | 10.3      | 83                  | 9         | 10.3   |
| 6Diep1  | L             | SV               | 120                   | 16           | 7.5       | 85                  | 10        | 8.5    |
| 9Diep4* | H             | S                | 59                    | 9            | 6.6       | 59                  | 9         | 6.6    |
| 10Hoog1 | M             | SVG              | 120                   | 15           | 8.0       | 74                  | 8         | 9.3    |
| 11Hoog2 | L             | S                | 133                   | 17           | 7.8       | 133                 | 17        | 7.8    |
| 14Homt1 | L             | S                | 120                   | 14           | 8.6       | 120                 | 14        | 8.6    |
| 15Homt3 | H             | SV               | 112                   | 14           | 8.0       | 102                 | 12        | 8.5    |
| 19Knys4 | M             | SG               | 114                   | 15           | 7.6       | 94                  | 12        | 7.8    |
| 13Kara2 | L             | S                | 110                   | 14           | 7.9       | 110                 | 14        | 7.9    |

\*SASS Scores generated using taxa recorded for stone sampling and not using the SASS protocol

### Qualitative macroinvertebrate data

Six species of *Simulium* were recorded from the qualitative samples. *Simulium medusaeforme* was present in moderate numbers at all sites surveyed. This is one of the most widespread commonly found species in South Africa, and can be found in a range of flow conditions but prefers moderate to swift currents. It is common in mountain and foothill streams but can occur in temporary streams where flow has been prolonged. It does not like highly turbid rivers and is therefore usually not found in the lower reaches of rivers. Although preferring the stones-in-current biotope it can be found on trailing and marginal vegetation. If it is found to co-occur with species such as *S. vorax*, *S. dentulosum* or *S. debegene* it would indicate that the flow is mostly very swift and water conditions are unpolluted, cool and well oxygenated. It is fairly tolerant of organic pollution as a species on its own.

*Simulium dentulosum* requires very swift flows of cool, clear, well-oxygenated water to thrive. An abundance of specimens indicates torrential, cascading flow-conditions. Therefore from the sample from site 16Knys1, it is apparent that the flow at that site would be fairly permanent and strong. There would be very little sediment input and TDS would be low. It is a species normally confined to mountain streams but in the clear acid streams of the Western Cape it can be recorded in the lower reaches of rivers. Some of these are small streams often considered to be “drowned rivers” hence in reality upper reaches transposed to sea level.

*Simulium nigritarse* s.l. is the most widespread and commonly found species of blackfly in South Africa. There is a large deal of variation in different populations of this species and it is thought to comprise a complex of at least 19 species hence it should be recorded as

*sensu lato* to cover this variation. Larvae are usually found in mountain foothill streams and not usually in mountain streams. They prefer slow-flowing water occurring often on the underside of stones or on trailing vegetation. The species is tolerant of pollution and can be found in small trickles of water to medium sized rivers where the flow is not too-swift, usually in side-streams in braided sections of rivers. It is also a rapid coloniser of newly inundated riverbeds.

*Simulium rutherfordi* is closely related to *S. nigritarse* and sometimes difficult to discern. It is also a species that prefers slow-flowing water but is more restricted to mountain streams and is also found in streams in forests. It is less tolerant of pollution than *S. nigritarse* and prefers cool, clear low TDS water streams. It is a good indicator of well conserved riparian vegetation. *Simulium merops* is a Western Cape endemic confined to the small foothill and forest streams where flow is slow and water conditions are unpolluted, clear, acid with low TDS. It could possibly be considered as a species that would reflect undisturbed natural vegetation. To establish this in depth research needs to be undertaken. *Simulium bequaerti* is a warm water species not previously recorded from the Western Cape. Common in small, clear-water, temporary streams but also recorded from foothill streams and medium sized rivers.

The presence of a diversity of filter feeding Hydropsychidae from the sample from site 6Diep1, as well as simuliids, indicates some input of particulate matter at this upstream site. This is not too modified as there was also an abundance of shredder and other FFG taxa present. It would be interesting to see if other river sites with the same river order position, as this site on the Diep River, but where tree plantation activity has not been implemented would produce a different composition of species. The absence of *Barbarochthon* at this site and its relative abundance at all the other sites is notable. Maybe the removal of forest could have influenced that. Also more particulate matter and raised TDS and pH levels and possibly a modified thermal (warmer) regime at this site may have influenced the abundance and breeding success of this species detrimentally. The sample from site 16Knys1 was also interesting in that it produced a large number of *Agapetus murinus*. This is a species that likes a moderate level of nutrients in the water and adequate sunlight to allow for the development of plant and algal growth on stones as it is a functional feeding group scraper. For these conditions to occur in what was previously an indigenously forested stream would indicate that much of the overhead canopy has been removed allowing sunlight to get through to the riverbed. Also the nutrient levels in the water must be sufficient to allow for the aufwuchs to grow on the substrates. *Sciado rusobtusus* is a Western Cape endemic and is associated with cool, clear, low nutrient and acid pH waters. The larvae are fairly long lived and a range of different sized larvae would indicate that conditions have been suitable for this species for some time.

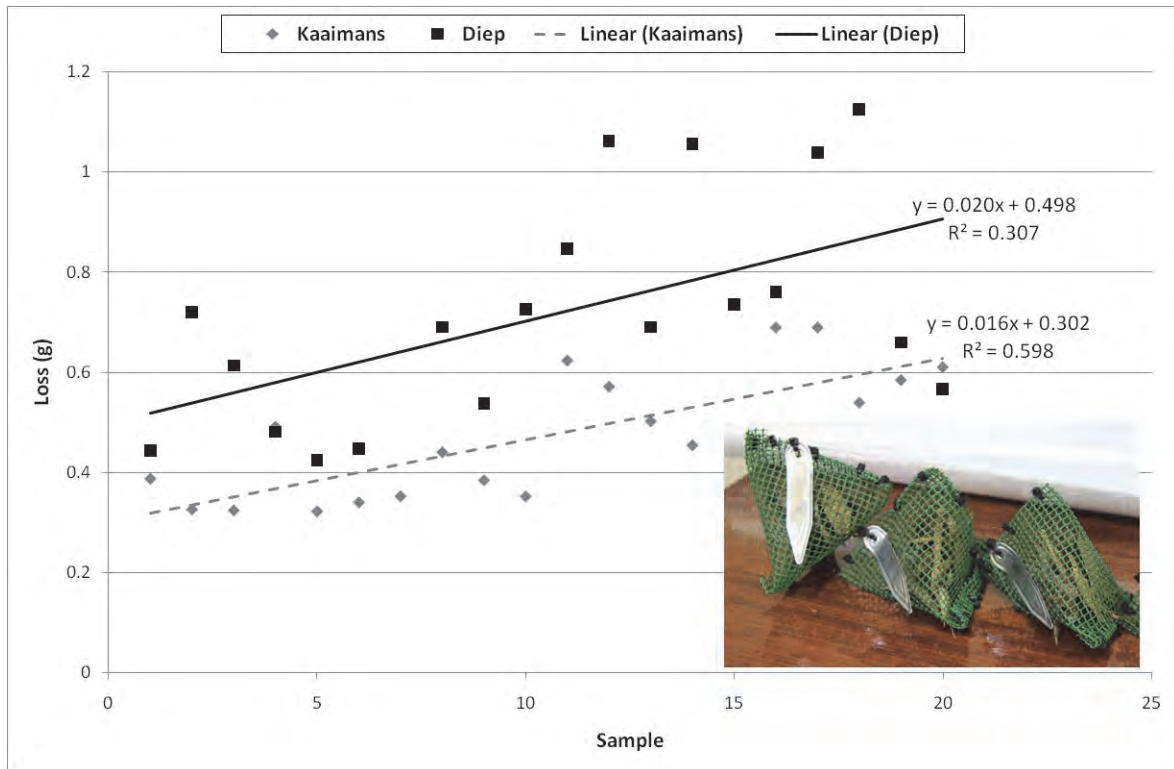
#### **4.3.2 Leaf litter decay rates**

Decay rates (Figure 4.11) for the Kaaimans and Diep Rivers were  $-0.015$  and  $-0.023 \text{ g.day}^{-1}$  respectively, as calculated by Equation 4.1, reflecting that the rate of leaf litter decay in the Kaaimans River (relatively natural system with few alien riparian trees) was almost twice as high as for the Diep River (higher density of alien species).

$$\left( \frac{\ln \frac{M_t}{M_0}}{t} \right)$$

[4.1]

where  $M_t$  is leaf litter dry mass at time  $t$ , and  $M_0$  is initial leaf litter dry mass at time zero, and  $t$  is duration of exposure (days) (after Hieber and Gessner 2002, and used by Jackson et al. 2014).



**Figure 4.11** Leaf litter loss for comparable sites on the Kaaimans and Diep Rivers. Inset shows leaf litter bags (Photo: Dirk Roux)

### 4.3.3 Index of Habitat Integrity

The Instream Habitat Integrity Score was greater than 84%, with the only sites less than 90% being the 15Homt3 and 20Knys5 sites (Figure 4.12). In comparison, six sites had Riparian Habitat Integrity Scores less than 75%, with the 15Homt3 having the lowest score of 58%. The scores were largely caused by decrease of indigenous vegetation in the riparian zone and presence of alien vegetation. Water abstraction was also an issue at 15Homt3.

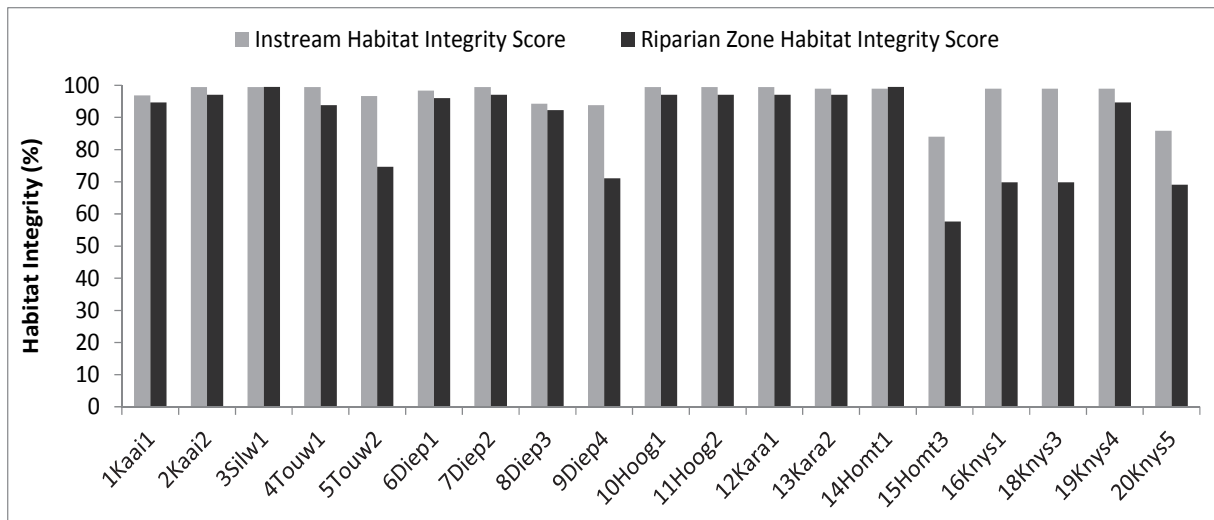


Figure 4.12 Instream and Riparian Habitat Integrity Scores for 19 sites assessed in October 2014.

#### 4.4 Discussion

Alien vegetation in the riparian zone has the potential to alter several components of riverine ecosystems including flow, sediment loading, shading, water temperature, and energy source through alteration in food source. One or all of these may reflect as changes in aquatic community structure. This study did not show a strong relationship between alien plant densities in the riparian zone and aquatic macroinvertebrate community structure. Alien densities did not correlate with number of taxa, evenness or diversity; and sites with higher densities did not consistently separate out from other sites, with the exception of one site, the 9Diep4. This however was largely because the site was bedrock dominated in comparison to a mix of cobble and boulder, which dominated other sites. Examination of the seasonal data also showed that there was substantial seasonal turnover of invertebrate communities. Both number of taxa and densities were higher in Spring compared to Summer or Autumn.

Evaluation of river health metrics, SASS and IHI, showed that instream health was relatively intact at all sites, although riparian integrity was lower at several sites. A recent study evaluated family- and species-level responses of river benthic macroinvertebrate assemblages to three riparian vegetation types (natural – fynbos, alien trees, cleared of alien trees – > 2 years previously) in the Cape Floristic Region biodiversity hotspot (Samways et al. 2011). High species beta diversity of this highly endemic fauna meant that between-river, as well as seasonal effects, dominated assemblage patterns. SASS indicated a decline in water quality conditions at cleared sites (cleared > 2 years previously), a likely response to the greater insolation and apparent erosion of cleared banks, resulting in elevated temperatures and suspended solids and lowered oxygen levels (Samways et al. 2011). Overall, cleared and natural sites were more similar to each other than to alien sites, suggesting some post-clearing recovery. However, many sensitive, endemic taxa survived in alien invaded sites, with these endemic species utilising the shady, cool, high oxygen levels under the alien tree canopy. After clearing, which increased water temperature, endemics declined in overall abundance and were replaced by more tolerant, widespread taxa.

Further examination of the data at species level is recommended before conclusions can be drawn. As shown by Samways et al. (2011) certain Ephemeroptera, Plecoptera, Trichoptera and Odonata species are excluded by alien trees that increase the degree of shading on the stream, including sensitive and endemic taxa such as the teloganodid, *L. penicillata*, and the heptageniid, *A. harrisoni*, as well as a number of Baetidae, such as *P. bellum*, *B. harrisoni*, *D. capensis* and baetid type "C". The extent to which alien trees increase shading to a stream is obviously of significance in fynbos streams, although may be less important in a stream running through afro-montane forests.

It is further recommended that additional studies are undertaken, focusing on a small set of streams, incorporating both fynbos and indigenous forest streams, which are going to be cleared and restored. By undertaking faunal sample for a year before the clearing followed by a year after, a better understanding of the instream effects on aquatic macroinvertebrate communities may be gleaned.



## **Chapter 5. Effect of different clearing techniques/approaches in the riparian zone on water temperatures and associated aquatic habitat integrity and community response**

### **5.1 Introduction: Clearing techniques and management interventions**

Water temperature is a driver of abiotic and biotic change in rivers (see reviews by Ward 1985, Caissie 2006, Dallas 2008) and altering the thermal regime of a river changes the environment to which organisms are adapted (Ward 1985). Aquatic organisms have an 'optimum thermal regime' or optimal range (Vannote and Sweeney 1980) to which they are most ideally suited and in which growth rates, fecundity and other measures of "health" are greatest (Dallas and Day 2004). Biological effects of changes in water temperature may include changes in aquatic biodiversity, changes in individual life history patterns, changes in communities, changes in species distribution and range, extinction of vulnerable species, increases in the number and spread of invasive and pest species, and an increase in waterborne and vector-borne diseases (Dallas & Rivers-Moore 2014).

Tolerance to changes in water temperature has been documented for several aquatic organisms (for example Nebeker, 1971; Ward & Stanford, 1982; Brittain, 1991; Hogg et al., 1995; Huryn, 1996; Wellborn & Robinson, 1996; Dallas & Ketley, 2011; Dallas & Rivers-Moore, 2012). Organisms of a given species can survive on either side of the optimal range but as the tolerance limits are approached, more and more abnormalities become evident. Initial signs are usually behavioural, with organisms avoiding sub-optimal conditions. As temperatures increase or decrease further, physiological stress may become evident, including elevation of respiratory, metabolic or excretory rates. Effects on reproduction (e.g. a decrease in egg and/or sperm production and hence in fecundity) and increased susceptibility to parasites and pathogens may also occur.

Tolerance limits may be determined by estimating lethal endpoints [e.g. Critical Thermal Method (CTM) and the Incipient Lethal Temperature (ILT) Method], while sub-lethal endpoints of exposure to temperature over longer periods typically include physiological and metabolic effects (e.g. growth rates, respiration), phenological effects (e.g. voltinism, emergence), effects on reproductive success and fitness (e.g. fecundity, hatching success), behavioural effects (e.g. migration), and broad scale ecological effects (e.g. density, competitive interactions) (see Dallas et al. 2015 for a review of this topic). In addition, thermal preferences may be investigated through behaviour observation of organisms exposed to a thermal gradient.

Factors which impact on water temperatures include reductions in flow (surface and groundwater), degree of shading (especially in first or second order streams), channel modification (channel obstructions resulting in changes in hydraulic biotopes and/or changing the residency times of water within a reach), and changes to natural sediment loads. Alien vegetation in the riparian zone can impact on all the factors mentioned above,

and climate change is likely to exacerbate the problem through its amplification of thermal extremes in aquatic systems. Changes to, *inter alia*, aquatic thermal regimes, will differentially impact different families of aquatic taxa (Dallas and Rivers-Moore 2012).

This chapter reports on the progress made on two aspects of the broader project: Firstly, the experimental determination of upper thermal limits and thermal preferences of a range of aquatic macroinvertebrate taxa from rivers in the Southern Cape, South Africa; and secondly, to begin the process of understanding the impacts of differential amounts of shading and flow reduction on water temperature regimes.

## **5.2 Methods**

### **5.2.1 Thermal experiments**

#### **5.2.1.1 Collection of aquatic organisms**

Organisms for estimating upper thermal limits were collected from the Kaaimans and Touws Rivers in November 2013 using a sampling net of 950 mm diameter mesh, which ensured that only larger organisms were collected. Stones were removed and rinsed to dislodge organisms, which were caught in a net held downstream of the stones being rinsed. Hand-picking was also used if the number of organisms was inadequate or if the organisms were damaged during the collection process. Test organisms were transferred into a collecting bucket, which was kept cool using crushed ice packed around the bucket, and transported to a temperature control room in the Biological Sciences Department, University of Cape Town. Prior to tests, organisms were held at  $\approx 15^{\circ}\text{C}$ , which approximated water temperatures at capture, and maintained in aerated tubs filled with dechlorinated tap water until experiments commenced. Dechlorinated tap water was used for experiments and a photoperiod of 12:12 LD (light from 06h00 to 18h00) was maintained throughout the experimental period. Experiments commenced after 48 to 72 hours and test organisms were not fed during the experiments.

Organisms for thermal preference experiments were collected from the Kaaimans and Homtini Rivers in April 2014 using a sampling net of 950 mm diameter mesh. Test organisms were transferred into a collecting bucket, which was kept cool using crushed ice packed around the bucket, and transported to the aquatic Ecology Laboratory of the Nelson Mandela Metropolitan University George Campus. Prior to tests, organisms were held at  $\approx 15^{\circ}\text{C}$ , which approximated water temperatures at capture, and maintained in aerated tubs filled with dechlorinated tap water until experiments commenced. Experiments commenced after 24 hours and test organisms were not fed during the experiments.

#### **5.2.1.2 Experimental procedures**

Upper thermal limits of test organisms were determined using two methods, the critical thermal method (CTM) and the incipient lethal temperature (ILT) technique (see Dallas & Ketley, 2011 and Dallas & Rivers-Moore, 2012 for details of these methods).

#### *Critical Thermal Method (CTM)*

For CTM experiments, organisms were placed in an experimental chamber, which was immersed in a water bath in the temperature room. Each experiment was approximately 90 min in duration, which included a 30 min control phase, followed by a 60 min experimental phase during which the water temperature was raised at a constant rate of 0.34°C per minute using a circulating heater (Julabo™). The circulating pump ensured that the percentage saturation of dissolved oxygen remained above 70% during the experiments. Organisms were monitored throughout the experiment and as an organism exhibited behavioural signs of reaching the endpoint,  $CT_{max}$ , it was removed from the experimental chamber and placed in aerated tubs at the start (acclimation) temperature. The  $CT_{max}$  for each test organism was recorded and only those organisms that recovered were included in the results. Identifications were confirmed after the experiments were concluded.

#### *Incipient Lethal Temperature (ILT) Method*

For ILT experiments, organisms were placed in experimental chambers immersed in glass aquarium tanks, filled with dechlorinated freshwater, which was heated using water heaters to eight different temperatures (one control and between five and seven experimental temperatures depending on species as follows: Control:  $14.8 \pm 0.11$ ,  $18.6 \pm 0.09$ ,  $21.9 \pm 0.19$ ,  $24.4 \pm 0.19$ ,  $26.2 \pm 0.27$ ,  $27.3 \pm 0.37$ ,  $29.0 \pm 1.05$  and  $30.3 \pm 1.52^\circ\text{C}$ ). Water temperature was recorded at one hour intervals throughout the experiment using Hobo TidbiT v2 loggers (Onset Computer Corporation, 2008) placed in each of the tanks. Aquarium tanks were aerated using two air stones per tank to ensure that the percentage saturation of dissolved oxygen remained above 75%. Thirty individuals of each taxon were placed in the experimental chamber at each temperature, although in some instances it was not possible to collect enough organisms in which case fewer individuals were used per experimental temperature. Starting at the acclimation temperature, the temperature of holding tank was gradually increased by  $2\text{-}4^\circ\text{C}\cdot\text{hr}^{-1}$ , until the appropriate target temperature was reached. A control temperature, which approximated water temperatures at capture, was included to test potential mortality factors such as influences from chambers, air, water or a lack of food. Chambers were checked for survival every 24 h for ten days subsequent to the target temperature being reached.

#### *Thermal preference experiments*

Thermal preference experiments were undertaken using a thermal gradient tank. The horizontal gradient tank comprised a shallow (ca. 10 cm), U-shaped plastic tank (15 cm width x 3.0 m length) which was filled with dechlorinated tap water. At one end of the tank a submersible 100W aquarium heater were positioned behind a screen of fine nylon mesh which prevented the heaters coming into direct contact with the organisms. At the other end of the tank, also positioned behind a screen, chilled water was circulated through copper coils, thus acting as a cooling element. Aeration and circulation of water was achieved by using a perforated tube running along the length of the tank, which allowed bubbling along the length of the tank. This setup allowed for a stable linear thermal gradient of  $10\text{-}25^\circ\text{C}$ . Water temperature was recorded at one hour intervals throughout the experiment using Hobo TidbiT v2 loggers (Onset Computer Corporation, 2008) placed at 10 cm intervals along the length of the tank. Organisms were added to the tank at a fixed position(s) and allowed to settle for one hour after which their position was noted and they

were removed. Two sets of trials were undertaken, the first where organisms were added at a position where the temperature approximated the acclimation temperature (16.5°C); and the second where organisms were added at the two thermal extremes, i.e. 10.7°C and 24.7°C. Taxa used for preference experiments were determined by their availability at the Kaaimans and Homtini Rivers, although attempts were made to collect a range of taxa.

In addition to the thermal preferences, the effect of rapid thermal cooling of water was investigated by creating a stable thermal environment of approximately 24°C in the thermal gradient tank. Organisms were released into the one end of the tank, which was subsequently cooled rapidly, while the opposite end was heated. Organisms were observed during the first 2 hours of rapid cooling after which they were left for a further 16 hours after which their position in the tank was recorded.

### **5.2.1.3 Statistical analysis**

For  $CT_{max}$  experiments, data were tested for normality (Kolmogorov-Sminorv test, Statistica Version 12 for Windows) and found to be largely non-normally distributed. The non-parametric Kruskal-Wallis one-way analysis of variance by ranks method was thus used to test the hypothesis that rankings are the same in different groups, with the significance level set at  $p < 0.05$ . For ILT experiments,  $LT_{50}$  values (the temperature at which 50% of the sample survives in a specified time) and 95% confidence intervals were calculated using the Trimmed Spearman-Karber analysis (USEPA TSK Programme Version 1.5). This method is used extensively for estimating median lethal concentrations in toxicity bioassays and has been shown to be accurate, precise and robust and is easily computable (Hamilton et al., 1977).  $LT_{50}$ s were calculated for each time period (every 24 h) and plotted against treatment temperature and analysed using linear regression. The resulting regression formula was then used to determine the incipient lethal upper temperature (ILUT), which is the temperature survived by 50% of the population for 96 h. This was done for both the 4 and 10 day trials.

For thermal preference data, the non-parametric Kruskal-Wallis one-way analysis of variance by ranks method was used to test the hypothesis that rankings are the same in different groups, with the significance level set at  $p < 0.05$ .

## **5.2.2 Abiotic-biotic response models**

The models developed by Rivers-Moore et al. (2013b) were used as the basis for linking thermal drivers and biotic responses. The Gondwanaland telaganodid mayfly *Lestagella penicillata* was selected for these models as a suitable species, based on the basis presented in Section 2.4. The impacts of MWAT threshold exceedances on hatching duration and probability of breeding failure were assessed for the site 6Diep1. Thermal data time series were based on observed thermal data, and simulated changes in water temperatures based on a 20% increase (clearing of alien riparian vegetation) and a 20% decrease (increase in alien vegetation) in flows, as described in Section 5.2.3.

### 5.2.3 Water temperature scenario analyses

These analyses involved two components. The first component involved comparing observed daily water temperatures at an upstream and downstream site on the Rondegat River, a tributary of the Olifants River in the Western Cape. Two periods of data were compared, with the earlier downstream data (Dallas et al. 2012) representing an un-cleared site heavily infested by black wattle *Acacia mearnsii*, while the later data for the same site represent water temperatures post-clearing (Weyl and Woodford, unpub. data, 2014) (Table 5.1).

**Table 5.1 Site information for upstream and downstream sites on Rondegat River where water temperatures have been collected pre- and post-clearing of alien riparian vegetation**

| Parameter            | Upstream – Natural fynbos          | Downstream – wattle infestation clear from 2012 |
|----------------------|------------------------------------|---|
| Co-ordinates (DD)    | -32.37026°S; 19.05358°E            | -32.25774°S; 18.96204°E                         |
| Altitude (m amsl)    | 499                                | 160   |
| Pre-clearing period  | 18 November 2008 – 9 February 2011 |   |
| Post-clearing period | 12 March 2013 – 13 March 2014      |   |

Next, the impacts of increased shading and increased versus decreased flows on downstream water temperatures were assessed using simulated data within the process-based Stream Segment Temperature (SSTemp) model (Bartholow 2002). Analyses were undertaken for the Diep River based on upstream observed water temperatures and associated air temperatures at site 6Diep-1 (328 m amsl), and simulated water temperatures at site 9Diep-4 (181 m amsl), 11.4 km downstream. Analyses were for the period 19 October 2013 to 16 January 2014, which at this stage of data collection represented the only time period of overlapping water temperatures, air temperatures and flow data. Shading was varied within the model and downstream water temperatures simulated for the following percentages of shading: 0, 5, 30, 50, 65, 80, 100. Only upstream flow data were available (K4H003), and downstream flows were estimated according to the following percentage changes at 30% shade: 10, 20 & 50% reduction from upstream flows, flows constant, 10 & 20% increase in flows; 50% reduction in flows and 0 and 100% shading.

## 5.3 Results

### 5.3.1 Thermal experiments

#### 5.3.1.1 Critical thermal maxima

CT<sub>max</sub> was estimated for a total of five families (Figure 5.1), including Notonemouridae [several species including *Aphanicerca capensis*, genus *Aphanicercella* (appears to be *barnardi*), *Aphanicercopsis outenique*, *Desmonemourapulchellum*], Heptageniidae (*Afronurus barnardi*), Leptophlebiidae (*Chloroterpes* sp.), Teloganodidae (*Lestagella penicillata*) and Philopotamidae (*Chimarra* sp.). CT<sub>max</sub> values varied significantly amongst taxon groups (i.e. family x river) (Kruskal-Wallis test: H = 103.1363, p < 0.05), with lowest values for notonemourids from the Kaaimans River (29.5°C) and highest for teloganodids

from the Touws Rivers (35.0°C) (Figure 5.1). Comparison of group pairs indicated that most differences were between notonemourids and other taxa (Table 5.2). Notonemourids, leptophlebiids and teloganodids from the Kaaimans River had lower CT<sub>max</sub> values compared to those from the Touws River (Figure 5.2), although only differences for leptophlebiids were statistically significant.

**Table 5.2 Comparison of group pairs (Family\_River). \* indicates a significant difference (P<0.05). No = Notonemouridae [several species including *Aphanicercera capensis*, *Aphanicercella barnardi*, *Aphanicercopsis outenique*, *Desmonemoura pulchellum*], He = Heptageniidae (*Afronurus barnardi*), Le = Leptophlebiidae (*Chloroterpes* sp.), Te = Teloganodidae (*Lestagella penicillata*) and Ph = Philopotamidae (*Chimarra* sp.).**

|       | No_K | No_T | He_K | He_T | Le_K | Le_T | Te_K | Te_T | Ph_K | Ph_T |
|-------|------|------|------|------|------|------|------|------|------|------|
| Tel_K | *    |      |      |      |      |      |      |      |      |      |
| Tel_T | *    | *    |      |      |      |      |      |      |      |      |
| Lep_K | *    |      |      |      |      | *    |      |      |      |      |
| Lep_T | *    | *    |      |      | *    |      |      |      |      |      |
| Phi_K | *    | *    |      |      |      |      |      |      |      |      |
| Phi_T | *    | *    |      |      |      |      |      |      |      |      |
| Not_K |      |      | *    | *    | *    | *    | *    | *    | *    | *    |
| Not_T |      |      |      |      |      | *    |      | *    | *    | *    |
| Hep_K | *    |      |      |      |      |      |      |      |      |      |
| Hep_T | *    |      |      |      |      |      |      |      |      |      |

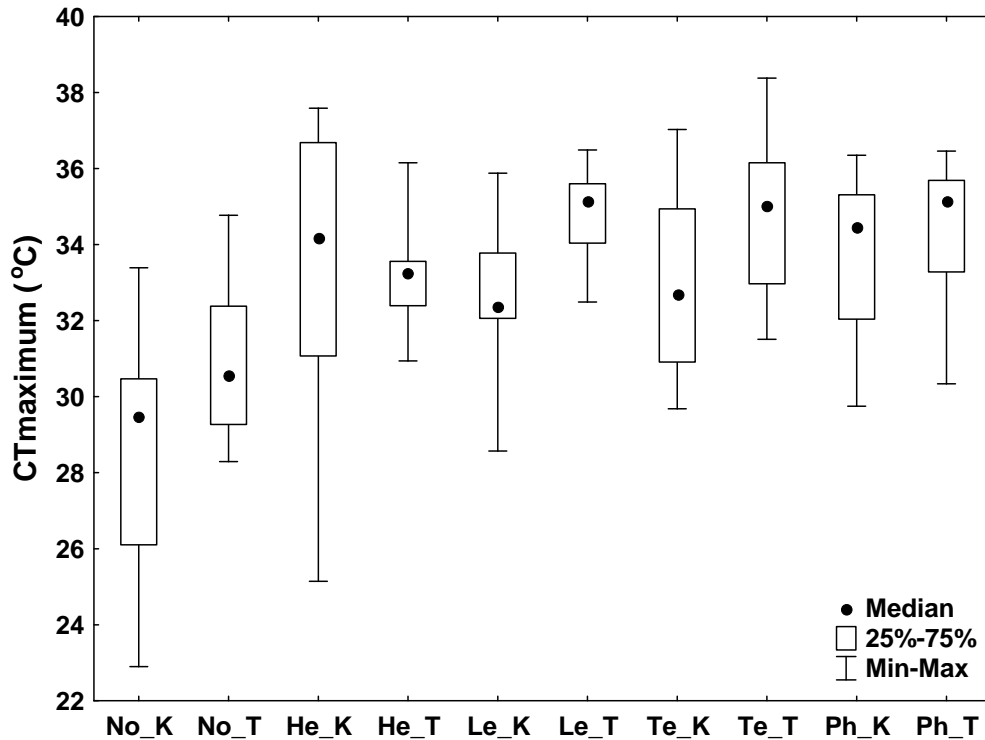


Figure 5.1 Median  $CT_{max}$ , 25th and 75th percentile and minimum and maximum values ( $^{\circ}C$ ) for taxa from the Kaaimans (K) and Touws (T) Rivers. No = Notonemouridae [several species including *Aphanicerca capensis*, genus *Aphanicerella barnardi*, *Aphaniceropsis outenique*, *Desmonemoura pulchellum*], He = Heptageniidae (*Afronurus barnardi*), Le = Leptophlebiidae (*Chloroterpes* sp.), Te = Teloganodidae (*Lestagellapenicillata*) and Ph = Philopotamidae (*Chimarra* sp.).

### 5.3.1.2 Incipient lethal upper temperature

96h ILUT were estimated for three families including Teloganodidae (*L. penicillata*), Leptophlebiidae (*Chloroterpes* sp.) and Philopotamidae (*Chimarra* sp.) from regression equations generated from four and ten day  $LT_{50}$  trials (Table 5.3).  $R^2$  values were generally higher for the 96h ILUT based on 10 days trials compared to 4 days trials, although actual 96h ILUT estimates were not substantially different. 96h ILUTs were consistently lower for the Kaaimans River compared to the Touws River and *L. penicillata* from the Kaaimans River had the lowest 96h ILUT ( $23.6^{\circ}C$ ), while *Chloroterpes* sp. from the Touws River had the highest 96h ILUT ( $28.8^{\circ}C$ ).

**Table 5.3** 96h ILUTs estimated from 4 and 10 day LT<sub>50</sub> experiments for *Lestagella penicillata*, *Choroterpes* sp. and *Chimarra* sp. Regression equations and R<sup>2</sup> values are provided.

| River                               | 4 Day Trial          |                |             | 10 Day Trial          |                |             |
|-------------------------------------|----------------------|----------------|-------------|-----------------------|----------------|-------------|
|                                     | Regression equation  | R <sup>2</sup> | 96h-ILUT    | Regression equation   | R <sup>2</sup> | 96h-ILUT    |
| <b><i>Lestagellapenicillata</i></b> |                      |                |             |                       |                |             |
| Kaaimans                            | y = -1.649x + 30.165 | 0.876          | <b>23.6</b> | y = -0.9412x + 28.466 | 0.919          | <b>24.7</b> |
| Touws                               | y = -0.88x + 30.57   | 0.991          | <b>27.1</b> | y = -0.8792x + 30.440 | 0.976          | <b>26.9</b> |
| <b><i>Choroterpes</i> sp.</b>       |                      |                |             |                       |                |             |
| Kaaimans                            | y = -1.34x + 32.71   | 0.999          | <b>27.4</b> | y = -0.6709x + 30.584 | 0.906          | <b>27.9</b> |
| Touws                               | y = -0.08x + 29.103  | 0.797          | <b>28.8</b> | y = -0.2757x + 29.681 | 0.953          | <b>28.6</b> |
| <b><i>Chimarra</i> sp.</b>          |                      |                |             |                       |                |             |
| Kaaimans                            | *                    |                |             |                       |                |             |
| Touws                               | y = -1.424x + 30.495 | 0.857          | <b>24.8</b> | y = -0.9178x + 29.421 | 0.934          | <b>25.7</b> |

\* High mortality in the control prevented calculation of 96h ILUT

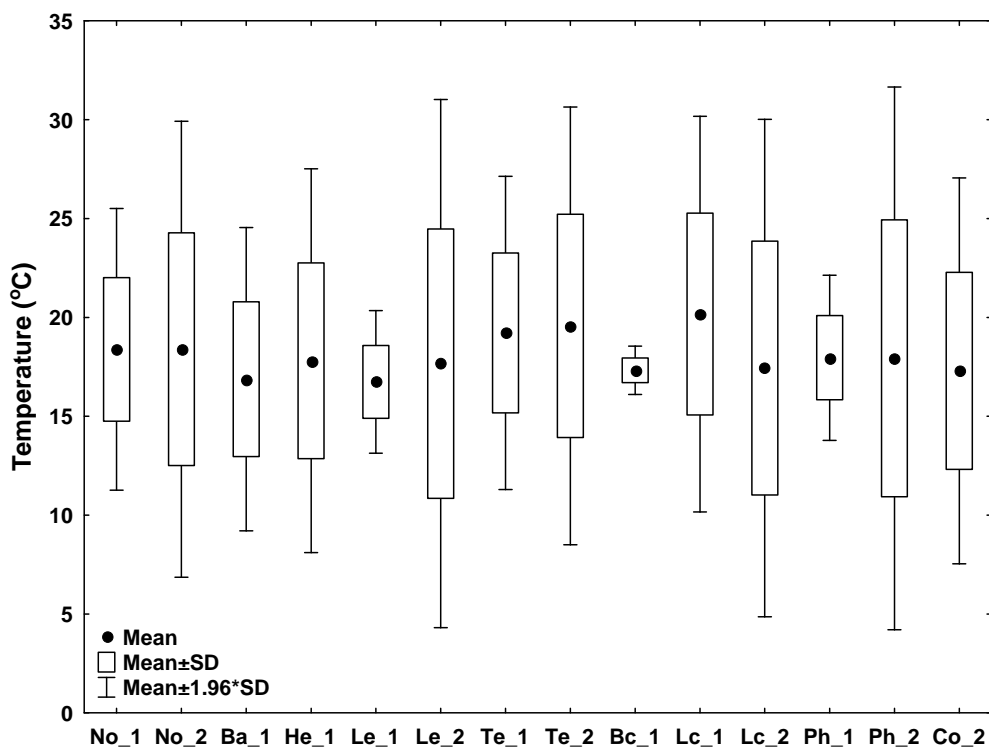
### 5.3.1.3 Thermal preferences

Logged water temperatures in the thermal gradient tank were used to generate a “preference temperature” (T<sub>p</sub>) for each organism following release and one hour of settling in the tank. This allowed for the calculation of mean T<sub>p</sub> for eleven families, including Notonemouridae (Kaaimans: mix of *Aphanicercera capensis*, *Aphanicercella barnardi*, Homtini: *Aphanicercera capensis*), Baetidae, Heptageniidae (*Afronurus barnardi*), Leptophlebiidae (Kaaimans: *Chloroterpes* sp., Homtini: *Aprionyx* sp. and *Castanophlebia* sp.), Teloganodidae (*Lestagella penicillata*), Barbarochthonidae, Leptoceridae, Philopotamidae (*Chimarra* sp.), Corydalidae, Dryopidae and Scirtidae (Table 5.4), although not all families were present in sufficient numbers at both the Kaaimans and Homtini Rivers. For the Kaaimans River, mean T<sub>p</sub> differed significantly (Kruskal-Wallis test: H = 28.4956, p < 0.05) amongst “family x trial” groups (Figure 5.2), although differences were largely attributed to Baetidae and Leptophlebiidae for trial 1. Mean T<sub>p</sub> ranged from 16.7°C for leptophlebiids to 20.2°C for leptocerids in trial 1 and 17.3°C for corydalids to 19.6°C for teloganodids in trial 2 (Table 5.3). Certain taxa such as leptophlebiids, barbarochthonids and philopotamids did not move substantially from where they were released (Figure 5.3).

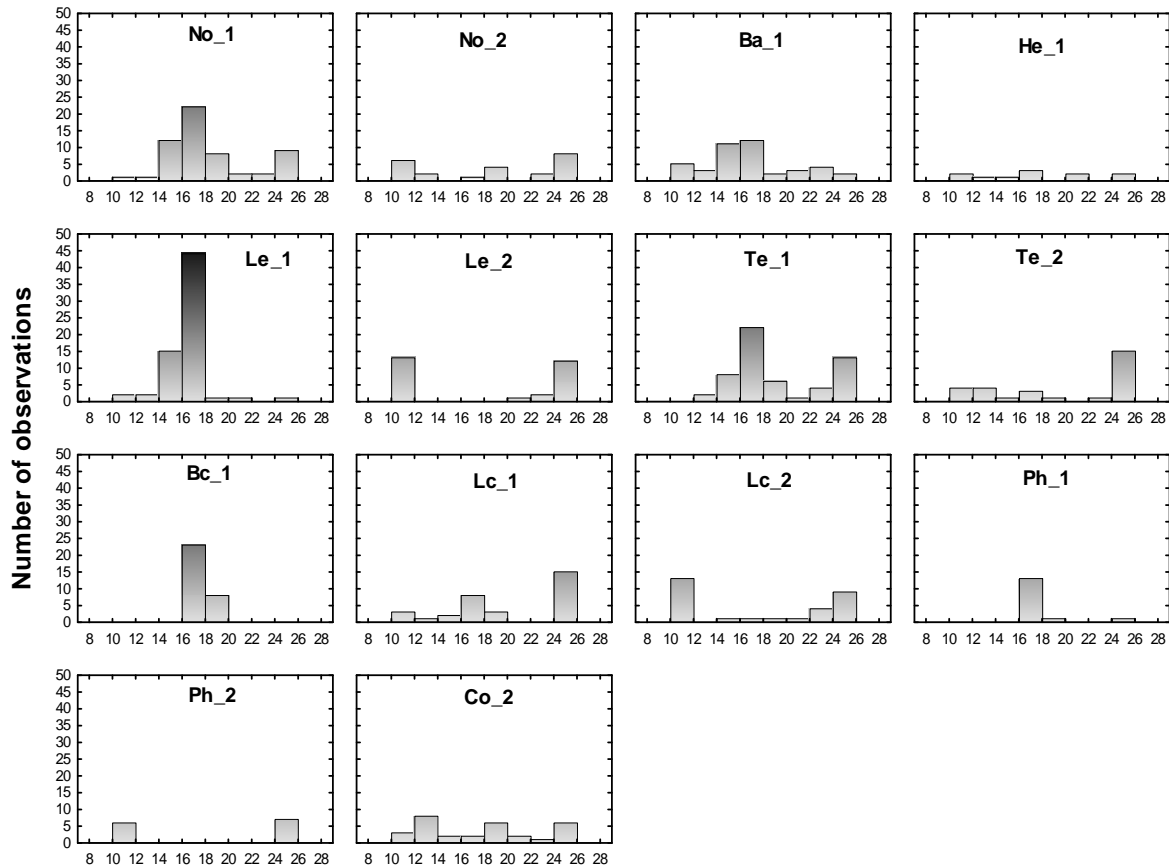


**Table 5.4 Mean  $T_p$  and number of organisms (N) recorded for each family in trials 1 and 2 from the Kaaimans and Homtini Rivers. – indicates insufficient organisms**

|                   | Kaaimans  |           | Homtini   |           |
|-------------------|-----------|-----------|-----------|-----------|
|                   | Trial 1   | Trial 2   | Trial 1   | Trial 2   |
| Notonemouridae    | 18.4 (57) | 18.4 (23) | 20.0 (31) | 18.2 (34) |
| Baetidae          | 16.9 (42) | -         | 18.5 (13) | 19.1 (14) |
| Heptageniidae     | 17.8 (11) | -         | 17.3 (7)  | -         |
| Leptophlebiidae   | 16.7 (66) | 17.7 (28) | 18.3 (46) | 16.4 (28) |
| Teloganodidae     | 19.2 (56) | 19.6 (29) | 17.9 (26) | 18.2 (42) |
| Barbarochthonidae | 17.3 (31) | -         | 17.6 (34) | -         |
| Leptoceridae      | 20.2 (32) | 17.4 (30) | 16.5 (36) | 20.0 (30) |
| Philopotamidae    | 18.0 (15) | 17.9 (13) | 19.7 (36) | 19.5 (33) |
| Corydalidae       | -         | 17.3 (30) | -         | -         |
| Dryopidae         | -         | -         | 19.0 (31) | 17.8 (30) |
| Scirtidae         | -         | -         | 15.5 (12) | -         |



**Figure 5.2 Mean temperature (°C), standard deviation (SD) and 95% confidence intervals for organisms from the Kaaimans River for trial 1 (16.5 oC) and trial 2 (10.7°C and 24.7). No = Notonemouridae [mix of *Aphanicercapensis* and *Aphanicercella barnardi*] Ba = Baetidae, He = Heptageniidae (*Afronurus barnardi*), Le = Leptophlebiidae (*Chloroterpes* sp.), Te = Teloganodidae (*Lestagella penicillata*), Bc = Barbarochthonidae, Lc = Leptoceridae, Ph = Philopotamidae (*Chimarra* sp.) and Co = Corydalidae.**



**Figure 5.3** Histogram showing the distribution of temperatures (°C) recorded for each family from the Kaaimans River. Data are presented separately for trial 1 (16.5 °C) and 2 (10.7°C and 24.7). No = Notonemouridae [mix of *Aphanicercacapensis* and *Aphanicercellabarnardi*] Ba = Baetidae, He = Heptageniidae (*Afronurusbarnardi*), Le = Leptophlebiidae (*Chloroterpes* sp.), Te = Teloganodidae (*Lestagellapenicillata*), Bc = Barbarochthonidae, Lc = Leptoceridae, Ph = Philopotamidae (*Chimarra* sp.) and Co = Corydalidae.

For the Homtini River, mean  $T_p$  differed significantly (Kruskal-Wallis test:  $H = 26.8899$ ,  $p < 0.05$ ) amongst “family x trial” groups (Figure 5.4). Mean  $T_p$  ranged from 15.5°C for scirtids to 20.0°C for notonemourids in trial 1 and 16.4°C for leptophlebiids to 20.0°C for leptocerids in trial 2 (Table 5.3). Certain taxa such as barbarochthonids and philopotamids did not move substantially from where they were released (Figure 5.5).

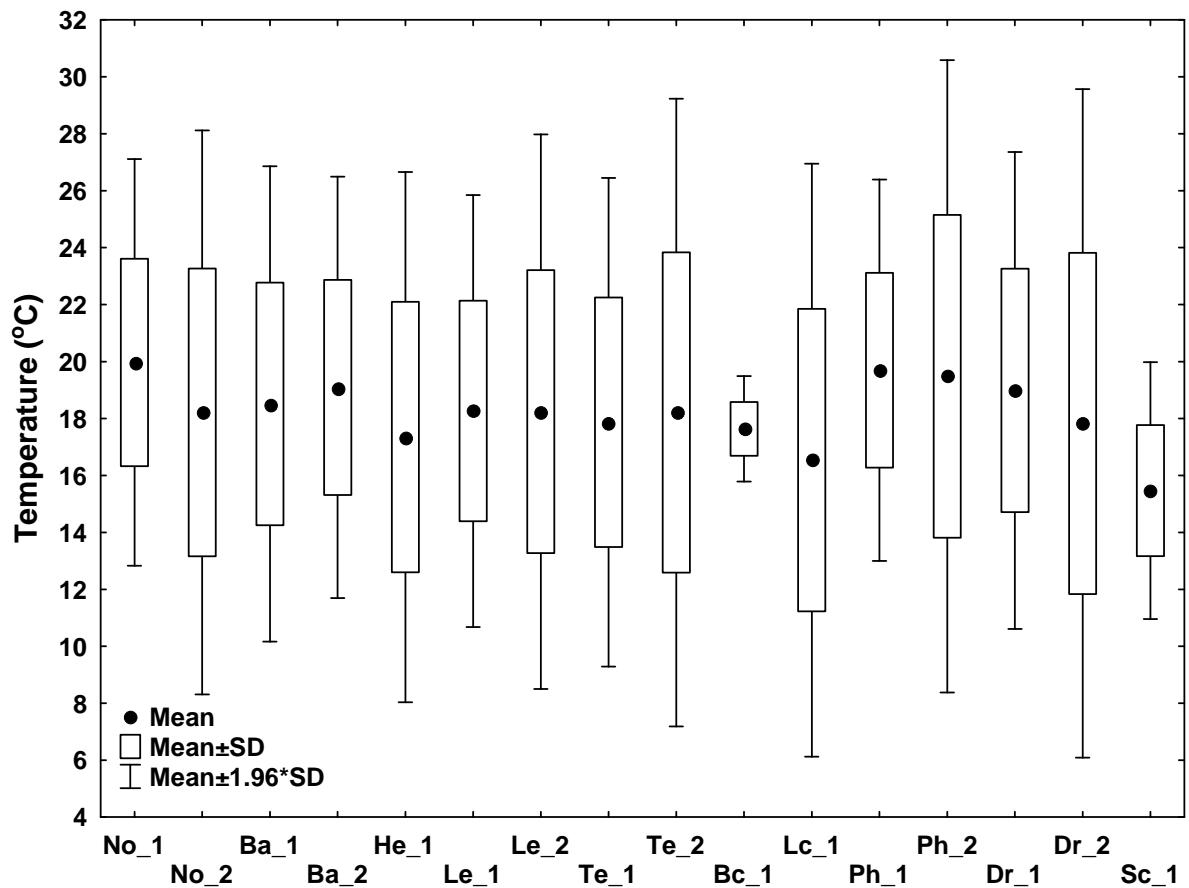
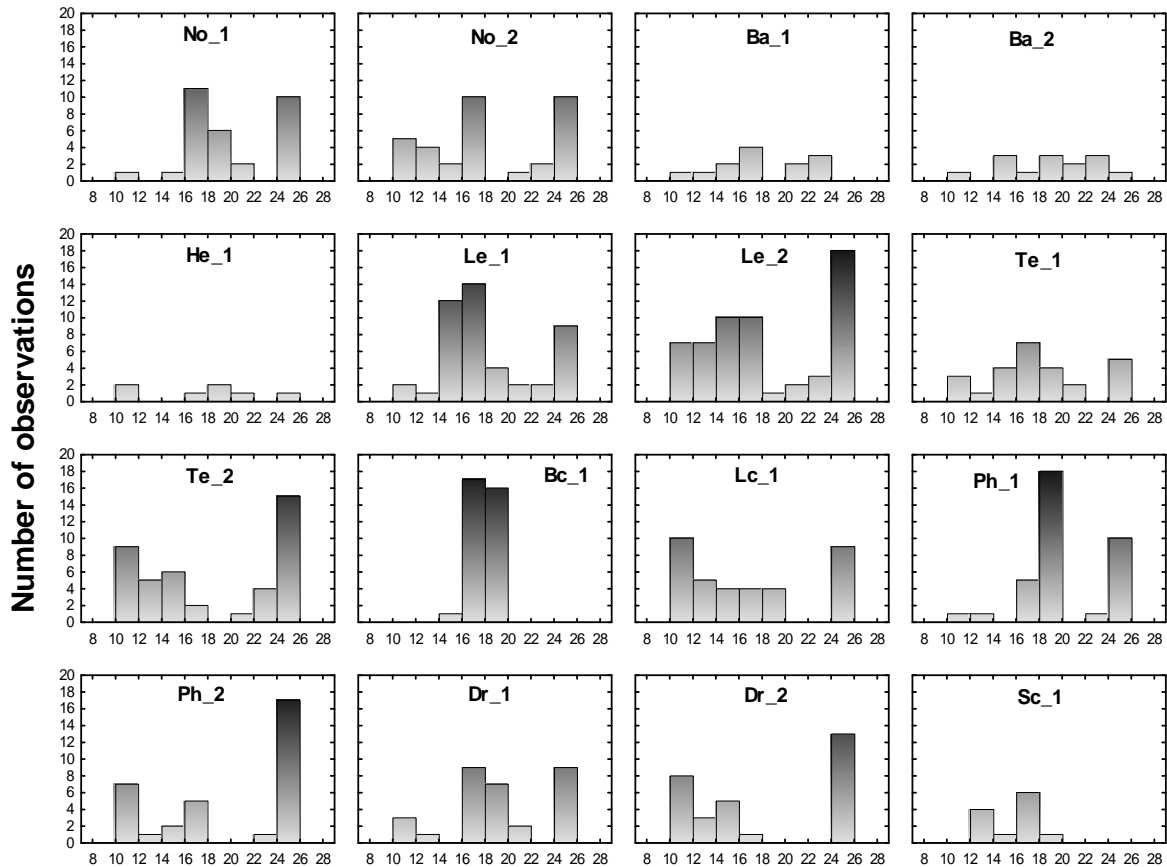


Figure 5.4 Mean temperature (°C), standard deviation (SD) and 95% confidence intervals for organisms from the Homtini River for trial 1 (16.5 °C) and trial 2 (10.7°C and 24.7). No = Notonemouridae (*Aphanicercacapensis*), Ba = Baetidae, He = Heptageniidae (*Afronurusbarnardi*), Le = Leptophlebiidae (*Aprionyx*sp. and *Castanophlebiasp.*), Te = Teloganodidae (*Lestagellapenicillata*), Bc = Barbarochthonidae, Lc = Leptoceridae, Ph = Philopotamidae (*Chimarra* sp.), Dr = Dryopidae and Sc = Scirtidae.

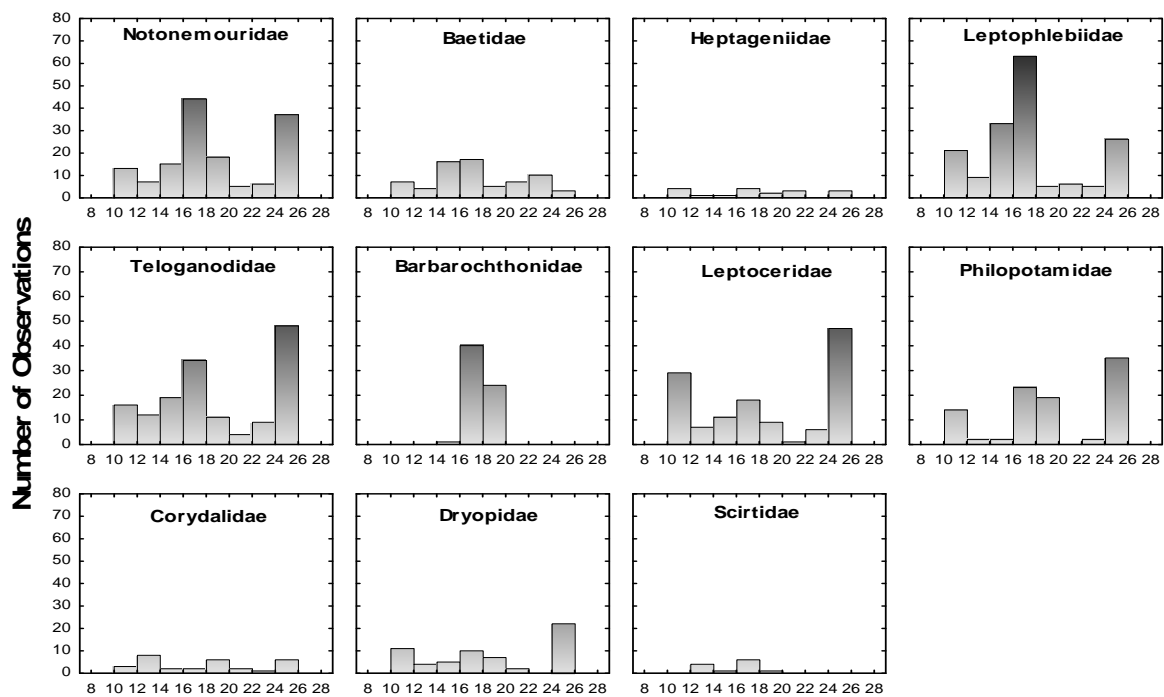


**Figure 5.5** Histogram showing the distribution of temperatures (°C) recorded for each family from the Homtini River. Data are presented separately for trial 1 (16.5 °C) and 2 (10.7°C and 24.7). No = Notonemouridae (*Aphanicercacacapsis*), Ba = Baetidae, He = Heptageniidae (*Afronurusbarnardi*), Le = Leptophlebiidae (*Aprionyx*sp. and *Castanophlebiasp.*), Te = Teloganodidae (*Lestagellapenicillata*), Bc = Barbarochthonidae, Lc = Leptoceridae, Ph = Philopotamidae (*Chimarra* sp.), Dr = Dryopidae and Sc = Scirtidae.

The overall mean  $T_p$  for each family was estimated by combining data from both rivers and trials (Table 5.4). Mean  $T_p$  was significantly different amongst families (Kruskal-Wallis test:  $H = 30.5498$ ,  $p < 0.05$ ) although most differences were as a result of differences between Leptophlebiidae, which has lower  $T_p$  compared to other Notonemouridae and Philopotamidae, which had the higher  $T_p$  (Table 5.5). Most taxa were recorded across the full range of temperatures in the thermal gradient tank suggesting that whilst some families may exhibited a thermal preference, they are able to tolerate the full range of temperatures (10°C to 25 °C) in the thermal gradient tank over the short term (Figure 5.6).

**Table 5.5 Comparison of pairs of families where data for rivers and trials were combined and which have been ordered by increasing mean  $T_p$ . \* indicates a significant difference one another ( $P < 0.05$ ). # low numbers and hence low confidence**

|                   | Mean $T_p$ | Sc | Le | Co | Bc | He | Ba | Dr | Lc | No | Te | Ph |
|-------------------|------------|----|----|----|----|----|----|----|----|----|----|----|
| Scirtidae#        | 15.5       |    |    |    |    |    |    |    |    |    |    |    |
| Leptophlebiidae   | 17.2       |    |    |    |    |    |    |    |    | *  |    | *  |
| Corydalidae       | 17.3       |    |    |    |    |    |    |    |    |    |    |    |
| Barbarochthonidae | 17.5       |    |    |    |    |    |    |    |    |    |    |    |
| Heptageniidae     | 17.6       |    |    |    |    |    |    |    |    |    |    |    |
| Baetidae          | 17.6       |    |    |    |    |    |    |    |    |    |    |    |
| Dryopidae         | 18.4       |    |    |    |    |    |    |    |    |    |    |    |
| Leptoceridae      | 18.5       |    |    |    |    |    |    |    |    |    |    |    |
| Notonemouridae    | 18.7       |    | *  |    |    |    |    |    |    |    |    |    |
| Teloganodidae     | 18.8       |    |    |    |    |    |    |    |    |    |    |    |
| Philopotamidae    | 19.1       |    | *  |    |    |    |    |    |    |    |    |    |

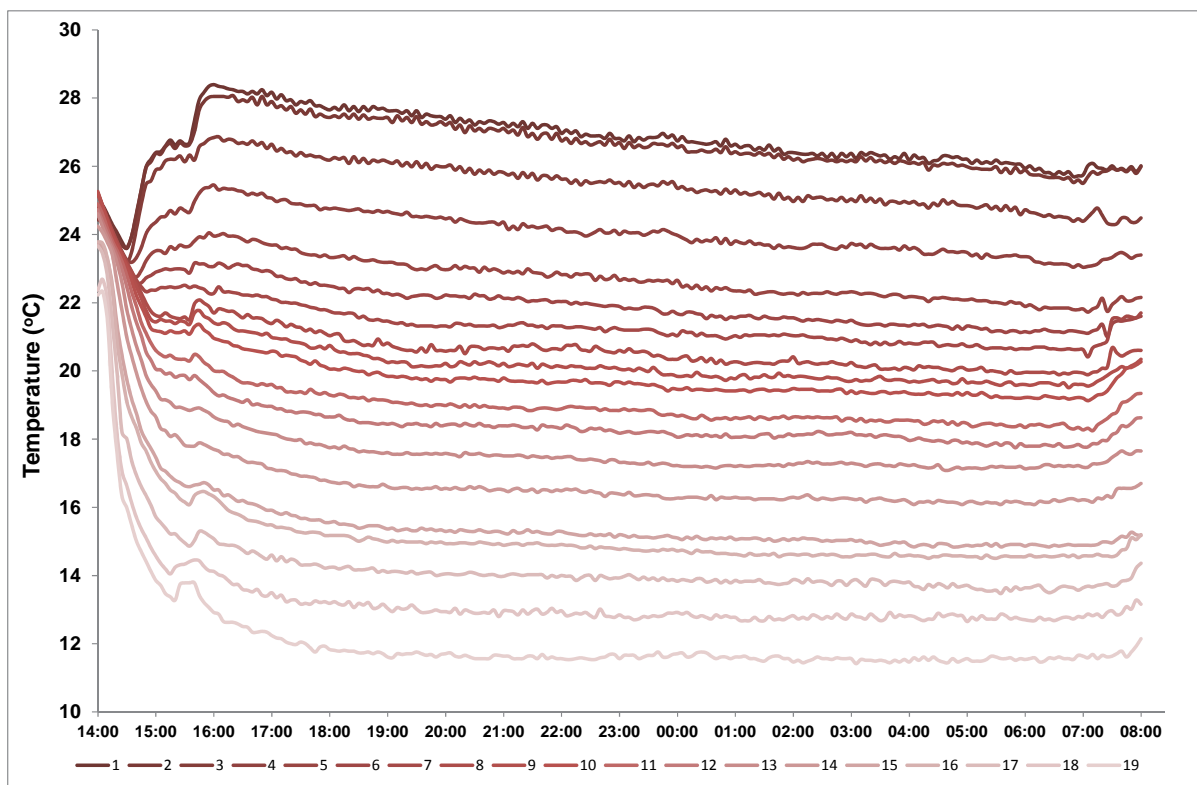


**Figure 5.6 Histogram showing the distribution of temperatures ( $^{\circ}\text{C}$ ) recorded for each family (data combined for rivers and trials).**

#### **5.3.1.4 Thermal cooling**

Water temperatures in the “hot” end of the thermal gradient tank increased by  $3.5^{\circ}\text{C}$  from  $24.9^{\circ}\text{C}$  at 14h00 to  $28.4^{\circ}\text{C}$  at 16h00 after which it decreased by  $2.7^{\circ}\text{C}$  to  $25.7^{\circ}\text{C}$  at 07h00 (Figure 5.7). Water temperatures in the “cold” end of the thermal gradient tank decreased

by 9.5°C from 22.3°C at 14h00 to 12.9°C at 16h00 and then a further 1.3°C to 11.6°C by 19h00 after which it remained relatively stable until 07h00.



**Figure 5.7** Water temperature (°C) recorded at 19 loggers spaced 10 cm apart in a thermal gradient tank over an 18 hour period.

Mean preference temperatures at the end of the 18 hour period varied significantly amongst families (Kruskal-Wallis test:  $H = 43.56$ ,  $p < 0.05$ ) from 14.9°C for Leptophlebiidae to 20.8°C for Leptoceridae (Figure 5.8). Histograms displaying the number of observations along the temperature gradient revealed that notonemourids concentrated at the cooler temperature end, although there were also a few individuals who occupied positions of hotter temperatures (Figure 5.9). Baetids and heptagenids were present throughout the thermal spectrum. Teloganodids strongly favoured a temperature of between 18°C and 20°C, whilst leptocerids were more common at temperatures between 18°C and 22°C, with several occupying the hottest position at 25°C. Mortalities were also noted with nine and seven deaths of baetids and teloganodids at temperatures  $> 24^\circ\text{C}$  respectively.

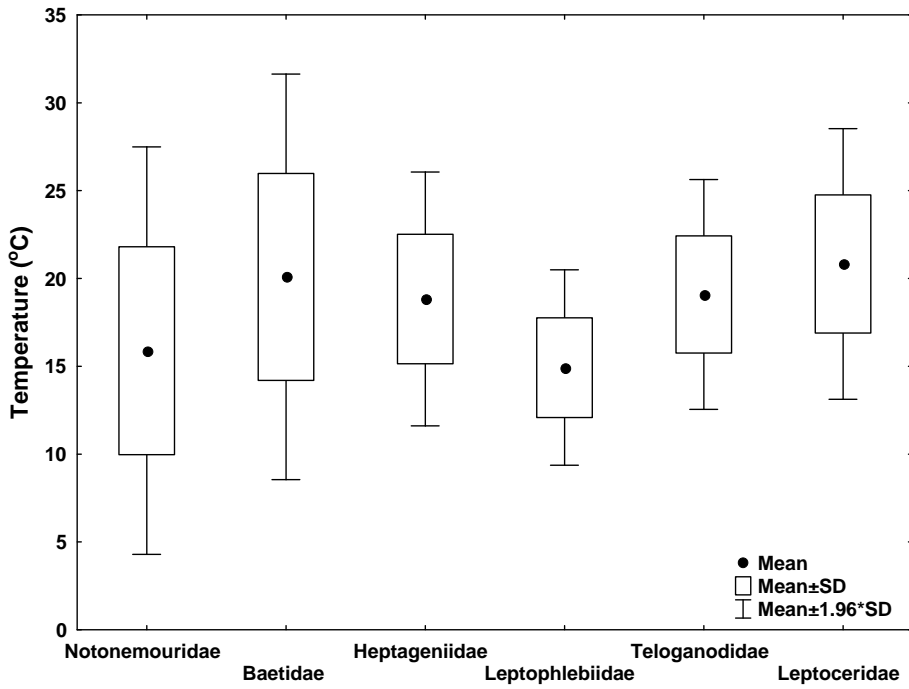


Figure 5.8 Mean temperature (°C), standard deviation (SD) and 95% confidence intervals for organisms from the thermal cooling experiment.

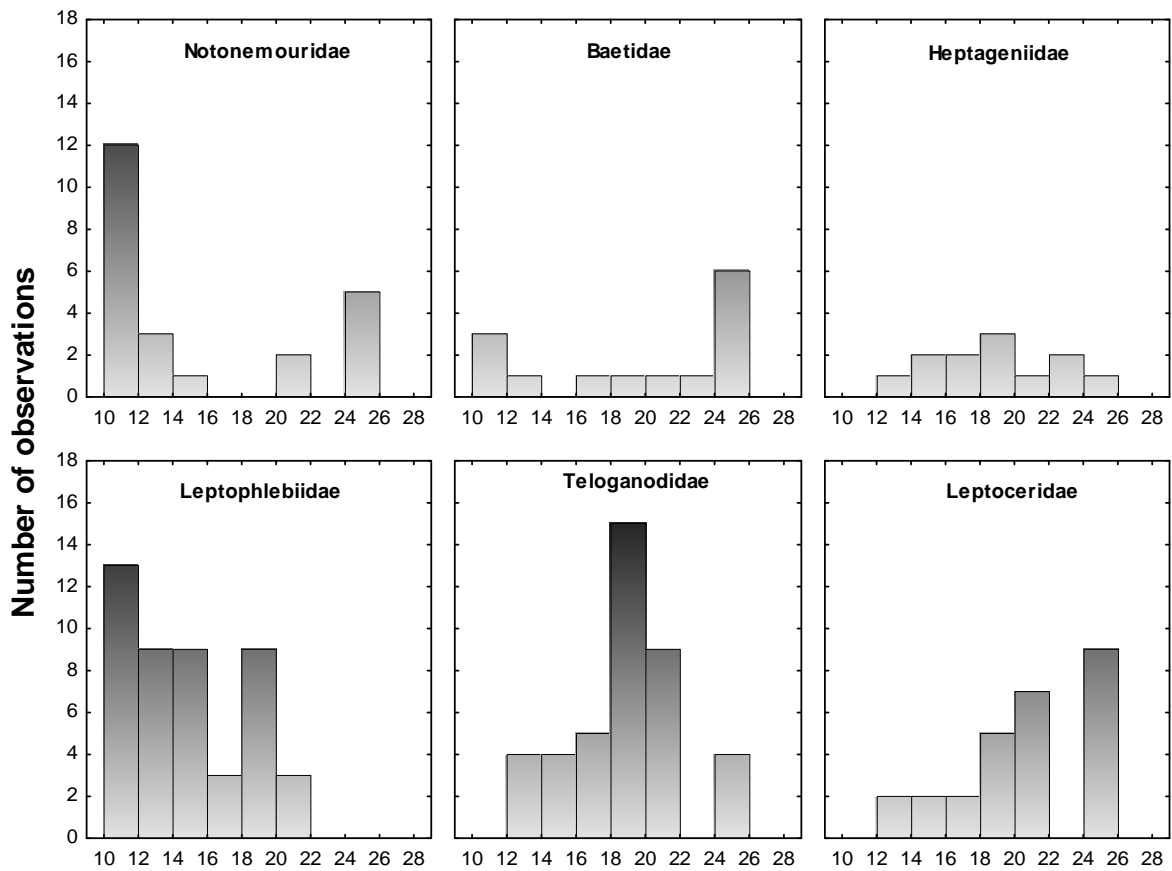
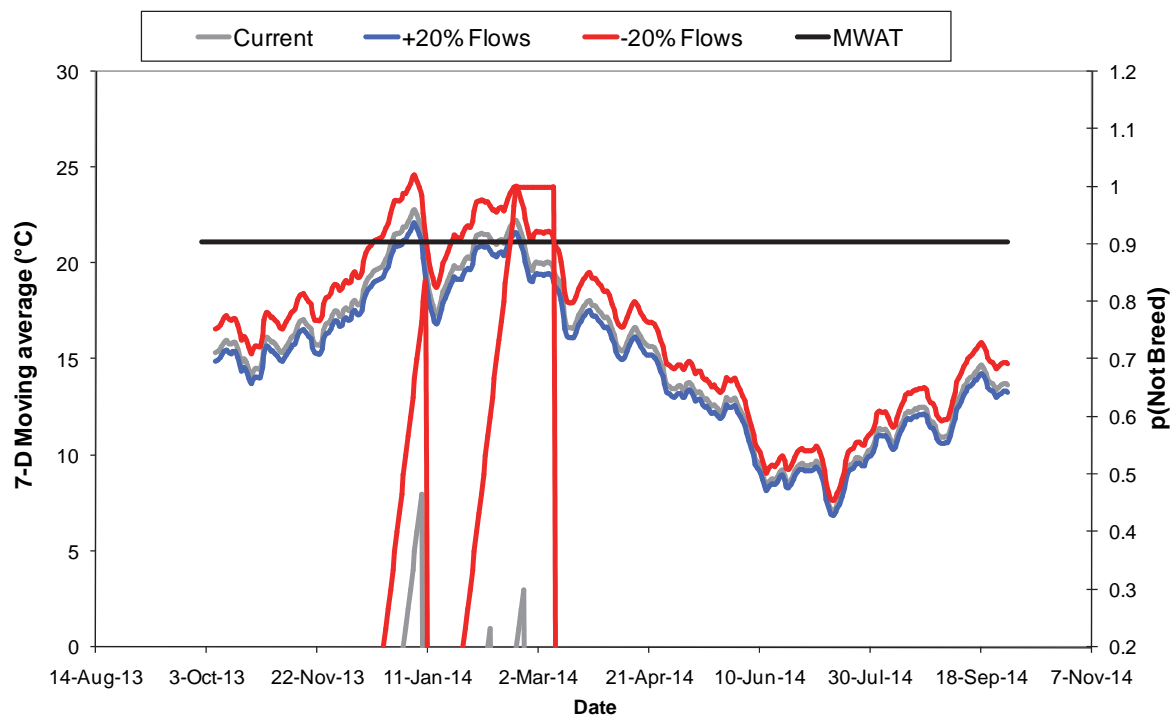


Figure 5.9 Histogram showing the distribution of temperatures (°C) recorded for each family from at the end of the thermal cooling experiment.

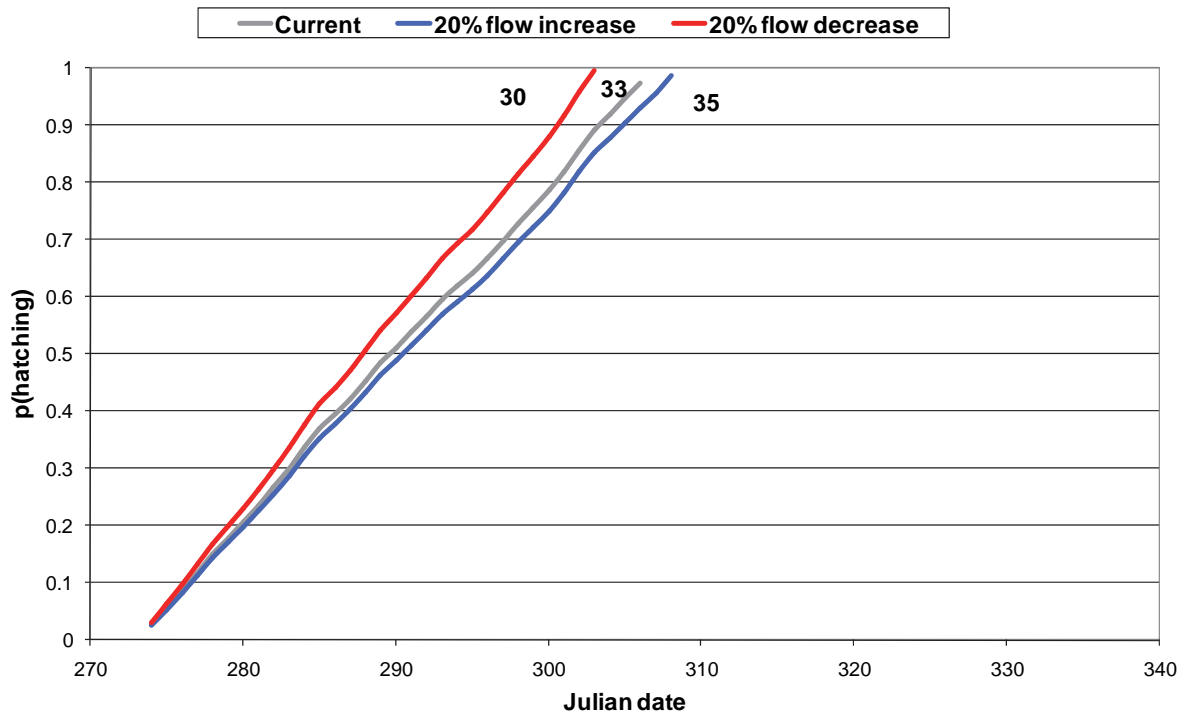
### 5.3.2 Abiotic-biotic response models

Modelled data, based on the degree days to hatching as described in Rivers-Moore et al. (2013b) and the probability of breeding failure based on the annual frequency of MWAT threshold exceedances, indicated that changes in flow regimes will either negatively or positively affect breeding success, and duration of hatching. Under current thermal conditions, there is a short period during summer (January) when breeding failure approached 50%, while under a 20% flow reduction scenario, there is 100% likelihood of breeding failure. Conversely, a 20% increase in flows is likely to result on total breeding success (Figure 5.10). Impacts on breeding success were more marked than on duration of hatching, which range from 30, 33 and 35 days for 20% reduction in flows, current thermal conditions, and 20% increase in flows respectively (Figure 5.11).



**Figure 5.10** Seven-day moving averages of mean daily water temperatures for three different flow scenarios (current; 20% reduction based on heavy catchment alien infestation; and 20% flow increase assuming successful clearing of alien invasive vegetation). These are plotted relative to an MWAT threshold for *Lestagella penicillata*, and the cumulative probability of this species not breeding as a result of thermal stress.





**Figure 5.11** Impact of different flow scenarios on hatching durations for *Lestagella penicillata* under different flow scenarios, which in turn impact on thermal regimes

### 5.3.3 Water temperature scenario analyses

Observed water temperature data showed that high levels of shading in a small stream actually reduced the level of chronic thermal stress, as reflected by the number of exceedances of the MWAT threshold for *Lestagella penicillata* (Figure 5.12). These observed data are supported by scenario analyses of water temperatures on the Diep River, where water temperatures decreased linearly with increased shading (Figure 5.13). However, water temperatures increased with reduced flows, with this impact more marked for lower levels of shading (Figure 5.13).

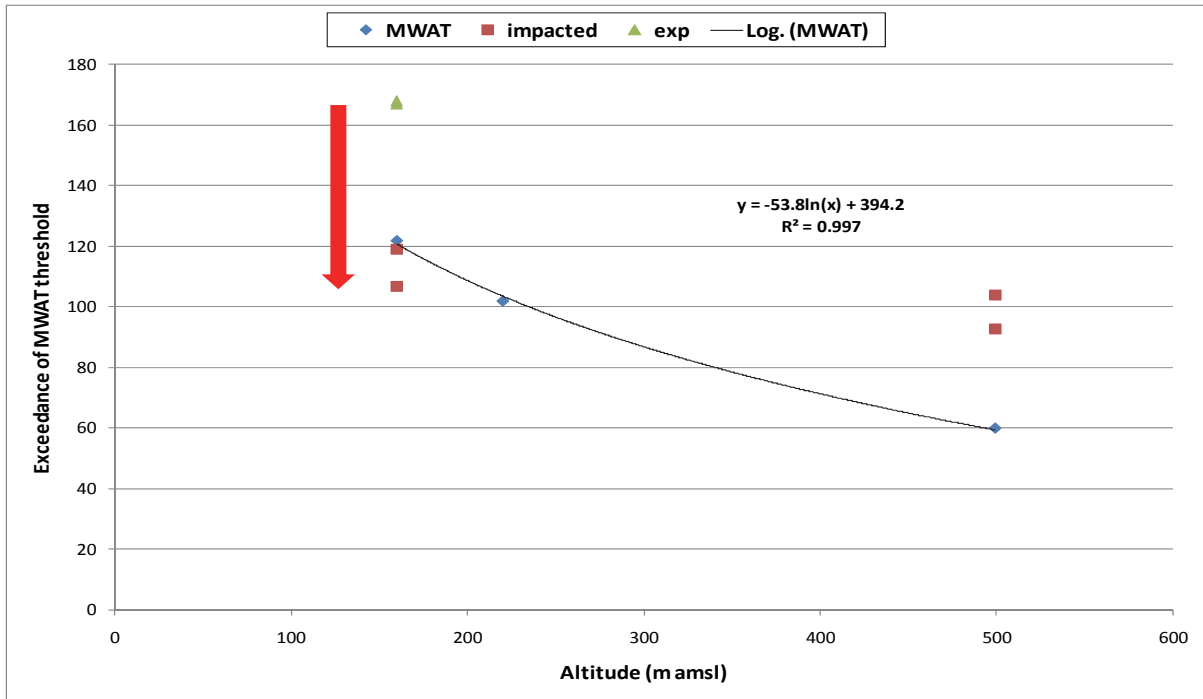


Figure 5.12 Annual number of exceedances of 21.1°C MWAT threshold at upstream and downstream sites in the Rondegat River, pre- and post-clearing of alien riparian black wattle. Red arrow indicates reduction in MWAT exceedances from expected to observed data, as a consequence of high levels of shading

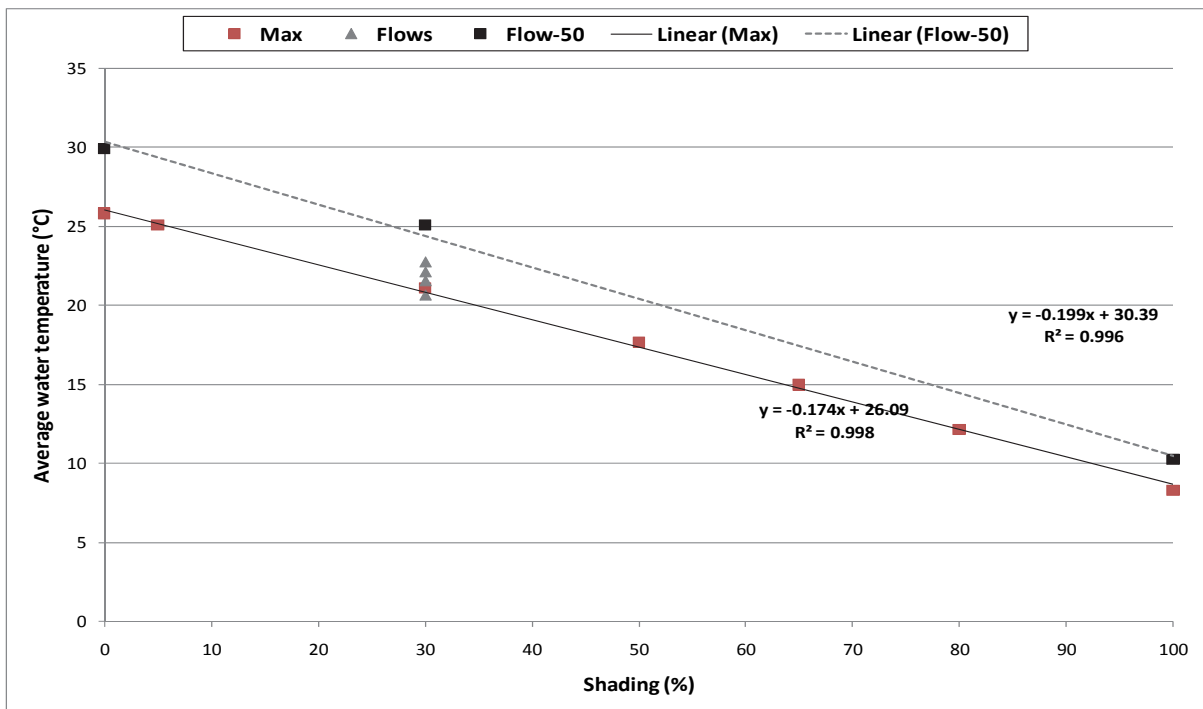


Figure 5.13 Linear reduction in average water temperatures as a function of % shading, and increase in water temperatures at different shade levels in response to flow reductions. Changes in temperatures have been simulated at 30% shade for changes of flow by 10, 20 and 50% reductions, and 20% increase (grey triangles) and a 50% reduction in flows at 0, 30 and 100% shading.

## 5.4 Discussion

Understanding upper thermal limits of organisms provides insight into the potential effect of elevated water temperatures on aquatic ecosystems. Some taxa will be able to cope with elevated temperatures better than others and over the long term this may result in shifts in community structure. The experimental determination of upper thermal limits allows for the establishment of temperature thresholds above which organisms will not survive. Thermal experiment data for the Kaaimans with Touws Rivers exhibit clear differences in  $CT_{max}$  and ILUT for most taxa, in spite of certain anomalies and with ILUT not calculated for all five target taxa. These thermal data also translate into different chronic thermal stress thresholds (MWAT values) for different taxa and between river systems (Table 5.6).

**Table 5.6 MWAT values for five families of aquatic macroinvertebrates and two river systems in the study area. ILUT thresholds have been estimated from Dallas and Ketley's (2011) equation:  $y = 1.1579x - 10.025$**

|                        | Kaaimans<br>CTmax | Kaaimans<br>ILUT | Touws<br>CTmax | Touws<br>ILUT | OT   | Kaaimans<br>MWAT | Touws<br>MWAT |
|------------------------|-------------------|------------------|----------------|---------------|------|------------------|---------------|
| <b>Notonemouridae</b>  | 29.5              | 24.1             | 30.6           | 25.3          | 18.7 | 20.5             | 20.9          |
| <b>Heptageniidae</b>   | 34.2              | 29.6             | 33.3           | 28.5          | 17.6 | 21.6             | 21.2          |
| <b>Leptophlebiidae</b> | 32.4              | 27.5             | 35.1           | 30.6          | 17.2 | 20.6             | 21.7          |
| <b>Teloganodidae</b>   | 32.7              | 27.8             | 35.0           | 30.5          | 18.8 | 21.8             | 22.7          |
| <b>Philopotamidae</b>  | 34.5              | 29.9             | 35.2           | 30.7          | 19.1 | 22.7             | 23.0          |

While further analyses are still required, it seems fairly likely that different river systems in the study area are likely to show differing responses to thermal stress in response to similar levels of alien riparian densities. However, predictions of these impacts are likely to be the result of an interaction of reduced water temperatures along river reaches where shading is high, and increased water temperatures as a result of flow reductions caused by high levels of alien vegetation at a catchment-scale (Dye and Jarman 2004). Subsequent research within this project is to refine the scenarios for simulated water temperatures in response to shading and flows. This can then be linked to thermal limits and preferences for different taxa, to assess possible taxon-specific responses to altered thermal regimes linked to on-site and catchment-wide levels of alien tree infestations.

## Chapter 6. Towards evaluating current clearing approaches in terms of financial efficiency and freshwater ecological returns

### 6.1 Introduction: Estimating effectiveness of management approaches

There is a recognised priority of reducing alien invasion of riparian zones of headwater streams in the southern Cape, as the predominant ecosystem in the Cape Floristic Region (Galatowitsch and Richardson 2005). Such restoration goals and their associated costs should be tailored towards the future use (for example, restoration of ecological functions, and resilience to floods) and current ecological condition of catchments of concern. Management decisions related to alien invasive riparian clearing involve a complex decision process, typically requiring site-specific restoration plans (Holmes et al. 2008). An effective decision-making framework is facilitated by, *inter alia*, an understanding of the specific relationships between invasive plants, resilience of ecosystem function, and realistic endpoints to guide restoration activities (Richardson et al. 2007). The project directly addresses this issue by quantification of ecological function (as measured by departure of water temperatures and change in aquatic macroinvertebrate communities from reference condition) which will assist in the drafting of site-specific plans. A second identified research priority is to assess the influence of different alien species, and clearing treatments, on the recovery of riparian ecosystems (Holmes et al. 2005).

During the course of the project, it was also ascertained that clearing techniques are defined according to Working for Water's "Norms and Standards" tables: this constrained cost analyses to person days per hectare according to the methods recommended for the species occurring in the riparian zones. Thus, the analyses are based on a comparison of prescribed clearing approaches within the constraints of the WfW database.

Within the South African National Park's (SANParks) Garden Route planning domain, clearing of alien vegetation is undertaken as a partnership between the SANParks BSP programme (Biodiversity Social Projects), and Working for Water (WfW), where the BSP fulfils WfW objectives within National Parks. However, one of the obstacles in planning for interventions is in assessing the favourability of cost:benefit ratios. Within a wider biodiversity/ conservation planning context, it is necessary to define methods to support funding allocation (or investment schedules), as given limited resources it is not possible to only focus on land purchase or species richness as means to allocate funds (Wilson et al. 2007). Rather, it has been found that greater biodiversity benefits accrue by investing in actions targeted towards specific threats (such as invasive species control) than by analyses that ignore costs and benefits (for example, by just using species richness per unit area) (Wilson et al. 2007). One way of answering these questions is to use field studies using reference versus impact sites, and to assess costs and benefits based on real costs of clearing under different management options, versus quantifiable improvements in water temperature regimes, to derive a cost per degree change towards desired thermal state. Such an approach allows for assessing which management interventions produce the best ecosystem benefits for the least cost.

Costs and methods for clearing alien invasive vegetation, and the awarding of contracts for doing this, are typically guided by the Working for Water's "Norms and Standards" tables. These are currently at version 3, and successive versions have been refined based on information from accumulated practices and experience. The WfW operates across all provinces and biomes of South Africa, all of which have different challenges in clearing alien vegetation. Such challenges will be driven by factors such as terrain, accessibility and varying degrees of susceptibility to invasion by different alien species.

Contracts are awarded based on cost estimates expressed as person days. These in turn are guided by a range of parameters, including target species; age and stage of the alien stand; density of invasion, and terrain slope. Operations may be either initial treatments or follow-up repeats. Depending on the species age and stage, guidelines are prescribed for the treatment methods and team composition. Thus, for example, in the Garden Route Initiative area, the current study has focussed on particular problems in the riparian zone. Problem invasive alien species are typically non-sprouting and sprouting trees, with size classes as seedling, young and adult. Treatment types include foliar spraying, hand pulling and bark stripping. Person days per hectare are calculated using a matrix of density (expressed as a percentage) and clearing method. Estimating densities are therefore critical, and a number of methods are used. These include "eyeballing" the stand and estimating a percentage cover, or more quantitative methods are available, including the Point Centre Quarter Method, and fixed area plot surveys.

## 6.2 Methods

### 6.2.1 *Quantifying implementation costs (net present value)*

The basis for developing this model was to identify a correlative relationship between change in water temperature and density of alien trees, and cost to clear alien trees. The basis for this link was through a study by Seedang et al. (2008), who linked water temperature gains to management options for restoration. Clearing costs as a function of alien vegetation were all calculated from the WfW database for the Garden Route planning domain. The most recent version of this database consists of approx. 15,600 records and spans a roughly ten-year period (2002-2013). For this study, a subset of these data were used, where only species encountered at the study sites were used: *Acacia mearnsii* (black wattle); *A. melanoxylon* (blackwood); *Pinus* spp. (all pines) and *Eucalyptus* spp. (all gums), resulting in an effective database of 12,000 records. Data records included fields listed in Table 6.1, with fields used for this model indicated. At this stage, cost was equated directly with actual number of person days per hectare, even though it is acknowledged that these figures would ultimately be multiplied by the 2014 cost per day of R231.00 (Nicholas Cole, 2014, pers. comm.). Based data exploration of this database included calculating the percentage of total person days allocated to each treatment for overall WfW programme in the Garden Route planning area, and based on black wattle, blackwood, pine and gums. This was complemented by deriving the relationship between of total number of person days and number of follow-up treatments. Note that a decision was taken to used either summed or averaged values across this database rather than actual values per WfW management polygon associated with each study site for two reasons: firstly, because while

14 of the 19 sites had management polygons associated with them, only three of these polygons had any WfW data attached to these polygons; and secondly, because errors on a site-by-site basis would be averaged out for a large database, making the model more generic across the Garden Route.

**Table 6.1 List of main variables for the Garden Route WfW database**

| <b>Variable</b> | <b>Meaning</b>   | <b>Use in Model</b>                  |
|-----------------|--|--------------------------------------|
| NballID         | Management polygon identifier                                  | No                                   |
| Stage           | Treatment stage between initial and 10 <sup>th</sup> follow-up | Yes – to estimate NPV                |
| Modelled PD     | Estimated number of person days                                | No                                   |
| TotContractPD   | Planned days worked for all NBallIDs                           | No                                   |
| ActualPD        | Actual days worked in the contract                             | Yes                                  |
| Species         | Species cleared  | Subset used                          |
| Density         | Density of species cleared (%)                                 | Yes                                  |
| sSize           | Size class cleared: seedling, young, mature or adult           | Yes                                  |
| sMethod         | Clearing method used, e.g. fell or ringbark                    | Yes – lumped across all size classes |

Next, a basic economic model was developed based on an estimate of Net Present Value. Much like the current study, Wilson et al. (2007) found that there was a need to convert annual costs over a period of years to one-time costs. This was done by calculating the **Net Present Value** over the time frame of interest. For the current study, costs were directly equated with person days, and discounted at a 5% discount rate (current rate in South Africa). This rate is in line with the 5.875% used by Seedang et al. (2008) and the 6.04% used by Wilson et al. (2007; based on 10 year US Govt. Bond rate). Management options were identified as per Table 6.2, and costs (average person days) were calculated for five different density classes (0-5%, etc.). These were adapted from the WfW norms and standards table (Shuttleworth 2013a, b). Seven treatments were taken as the cut-off of treatments, and this was used as the basis for cost for treating a site over a seven year period. Income from a cleared site was assumed to be equivalent to the maximum average number of person days for any treatment (400). This provided the information required to calculate NPVs per management option at different densities.

**Table 6.2 Management options used to compare NPVs at different densities**

| <b>Management option</b> | <b>Description</b>  |
|--------------------------|---|
| Do Nothing               | Status quo remains; no alien control                          |
| Proactive                | Aggressively target all seedlings and one follow-up treatment |
| Seedling                 | Target seedlings and up to seven follow-ups                   |
| Young                    | Target young trees and up to seven follow-ups                 |
| Mature                   | Target adult trees and up to seven follow-ups                 |

### 6.2.2 Relationship between riparian alien plant density and deviation from thermal reference condition

MWAT threshold exceedance was selected as the principle biological metric. This is a commonly used chronic stress threshold based on a seven-day moving average of daily mean water temperatures (7-D Mean) as it applies to growth rates in general, and thus regarded as a good overall thermal metric because it integrates all physiological responses of an organism (Brungs and Jones 1977). The relationship between % departure from expected MWAT threshold exceedance and density was used as the basis for estimating thermal gains relative to alien density.

## 6.3 Results

### 6.3.1 Quantifying implementation costs (net present value)

WfW data show that there are up to nine follow-up treatments succeeding the initial treatment, even though the greatest investment of total person days is for the initial treatment (31%; Figure 6.1). Represented as a relationship between total number of person days (effort) versus number of treatments, data showed that there is an exponential decay in numbers of person days expended per successive treatment (Figure 6.2). This provided a reasonable basis for assuming that cost per site would not be likely to go beyond seven years, assuming an annual treatment. The greatest investment of person days appeared to be for adult and mature trees, the lowest for seedlings, and an apparent intermediate costs (with some anomalies for first and second follow-ups) for young trees (Figure 6.3). Average number of person days per tree stage typically increased with density, but not in a linear fashion (Figure 6.4).

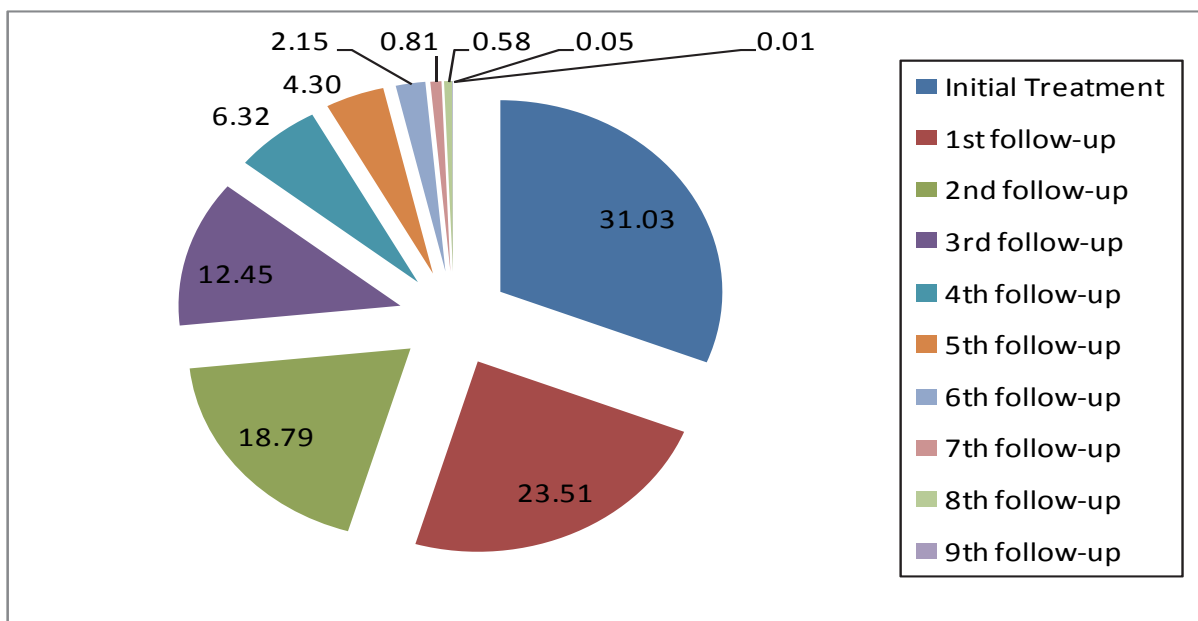


Figure 6.1 Pie chart of percentage of total person days allocated to each treatment for overall WfW programme in the Garden Route planning area, and based on black wattle, backwood, pine and gums

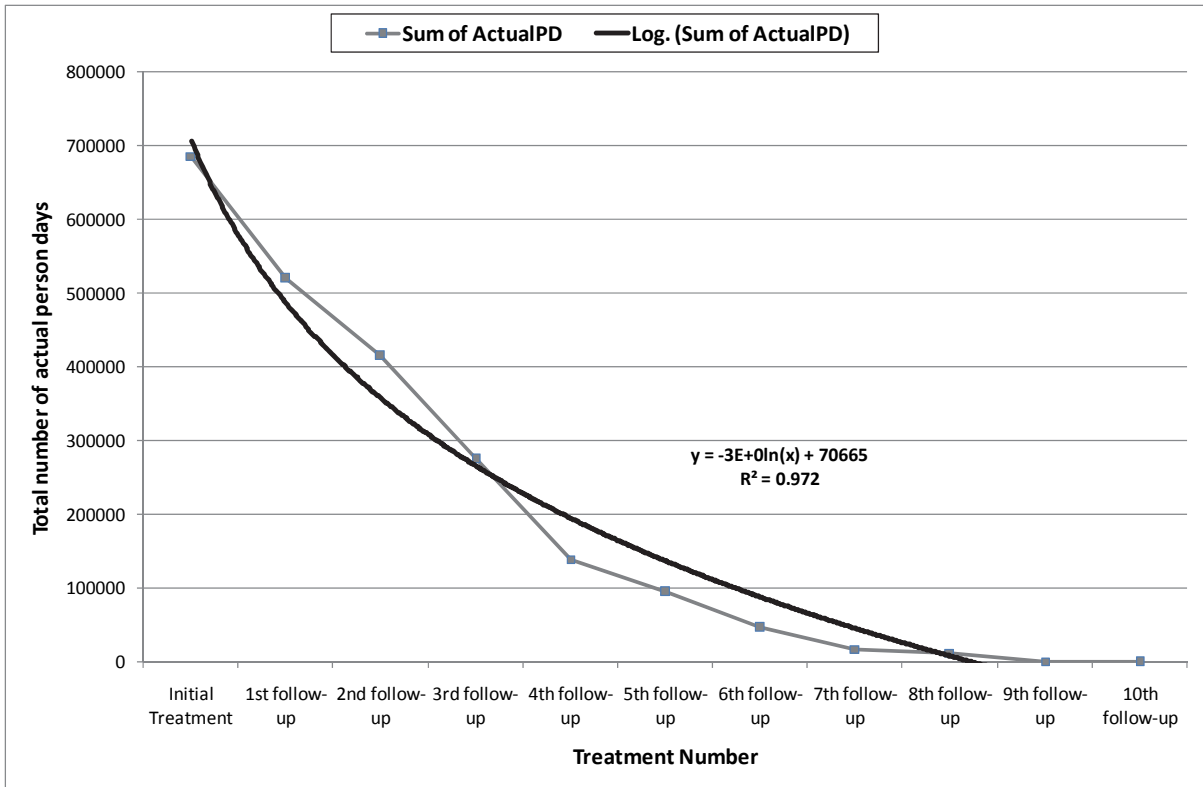


Figure 6.2 Power relationship between of total number of person days and number of follow-up treatments.

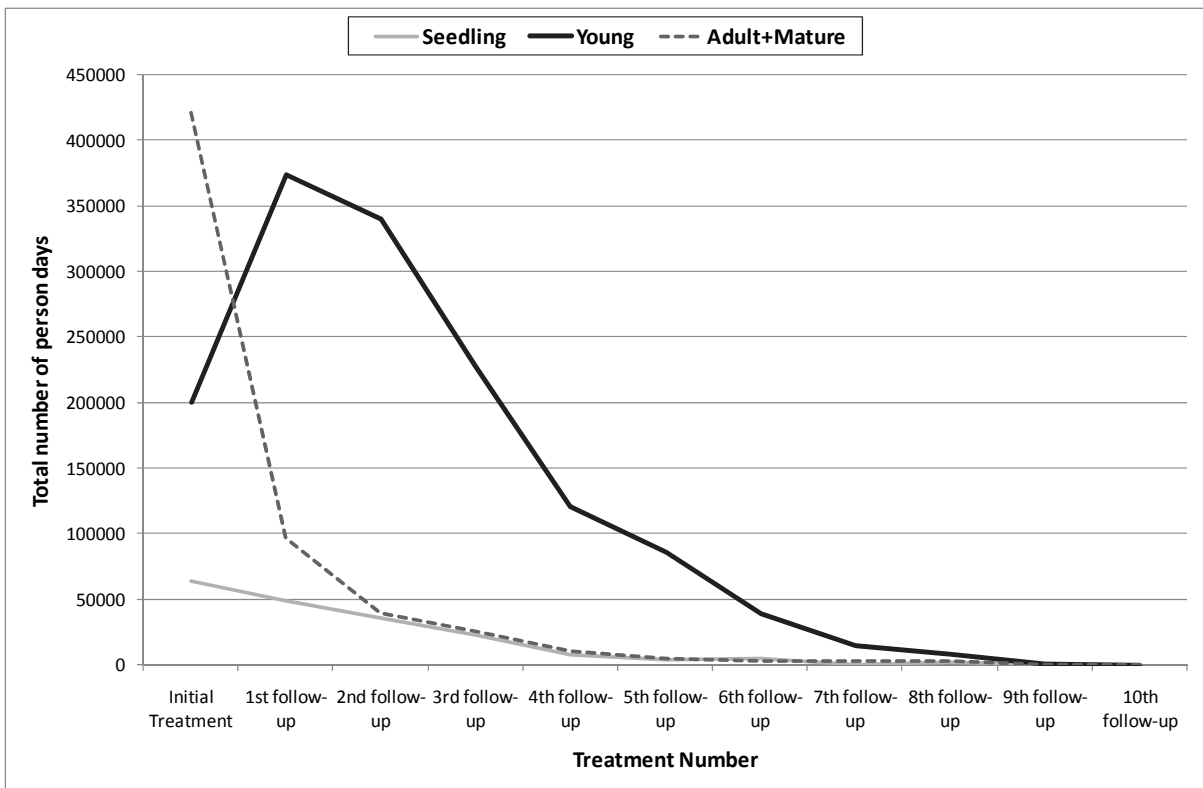


Figure 6.3 Allocation of person days (total number) per treatment for size classes of alien vegetation in the Garden Route planning region.



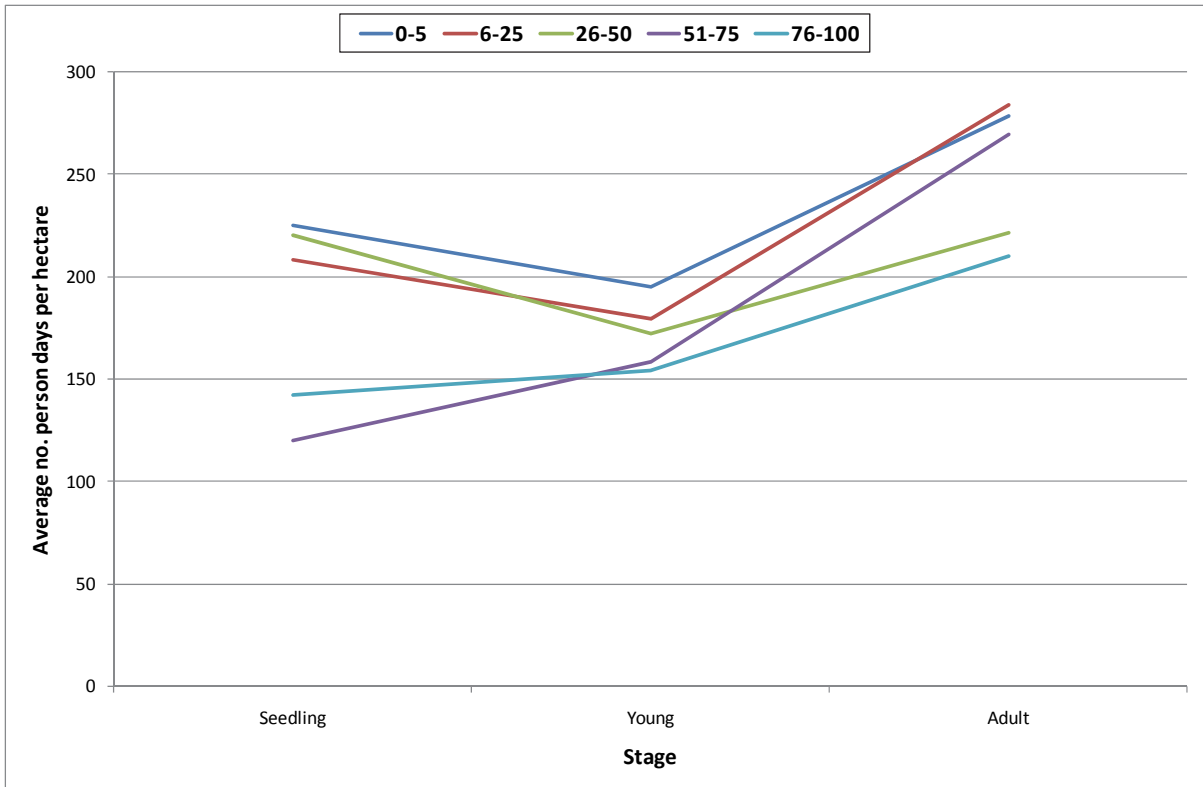


Figure 6.4 Average person days per hectare by tree stage at different density intervals

**6.3.2 Relationship between riparian alien plant density and deviation from thermal reference condition**

Based on three previous studies of MWAT threshold exceedances at three different values, there was a relatively strong correlation between MWAT exceedance and altitude ( $R^2 > 0.63$ ) (Figure 6.5). While this relationship was considerably weaker for the 19 study sites (Figure 6.6), it did provide the basis to develop a model of MWAT exceedance with altitude, thereby allowing for the calculation of residuals (Figure 6.7). There was a strong relationship between percentage exceedance from reference MWAT value and density of site-specific alien riparian vegetation (Figure 6.8).

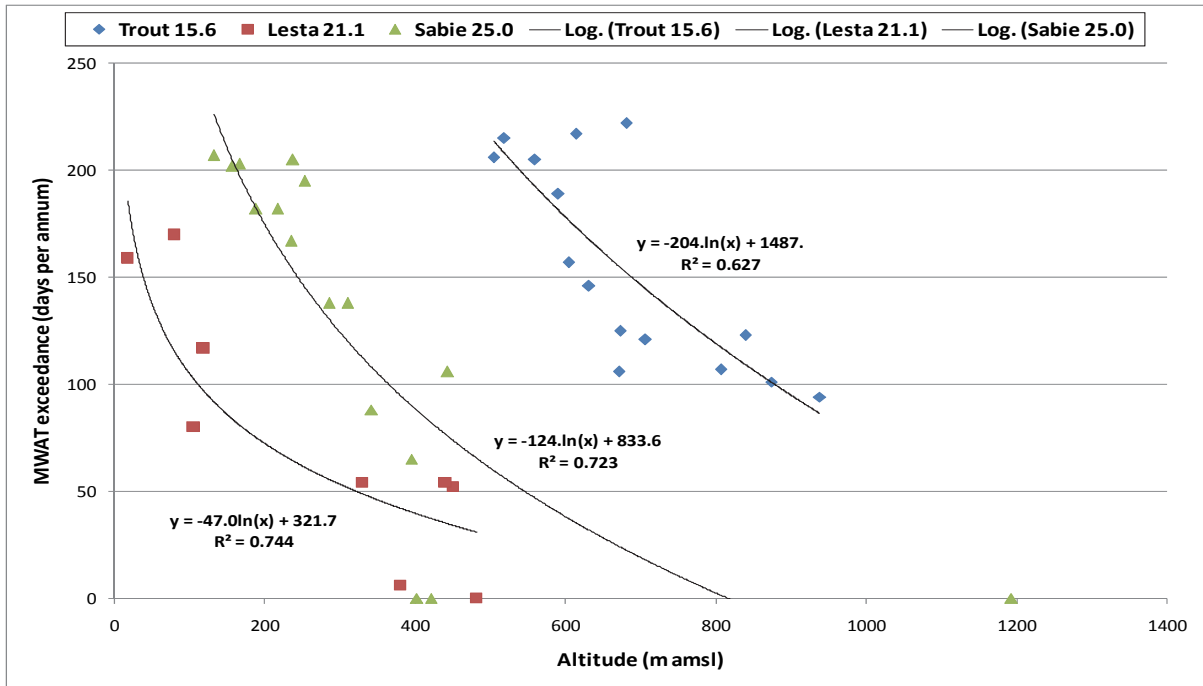


Figure 6.5 Relationships between altitude and MWAT exceedance for three different threshold values: 15.6°C (Trout: Ellender et al. in prep.); 21.1°C (Lesta21.1 = Thetelaganodid mayfly *Lestagella penicillata*: Rivers-Moore et al. 2013b); 25.0°C (Sabie 25.0 = the rock catlet *Chiloglanisnoterus*: Rivers-Moore et al. 2005).

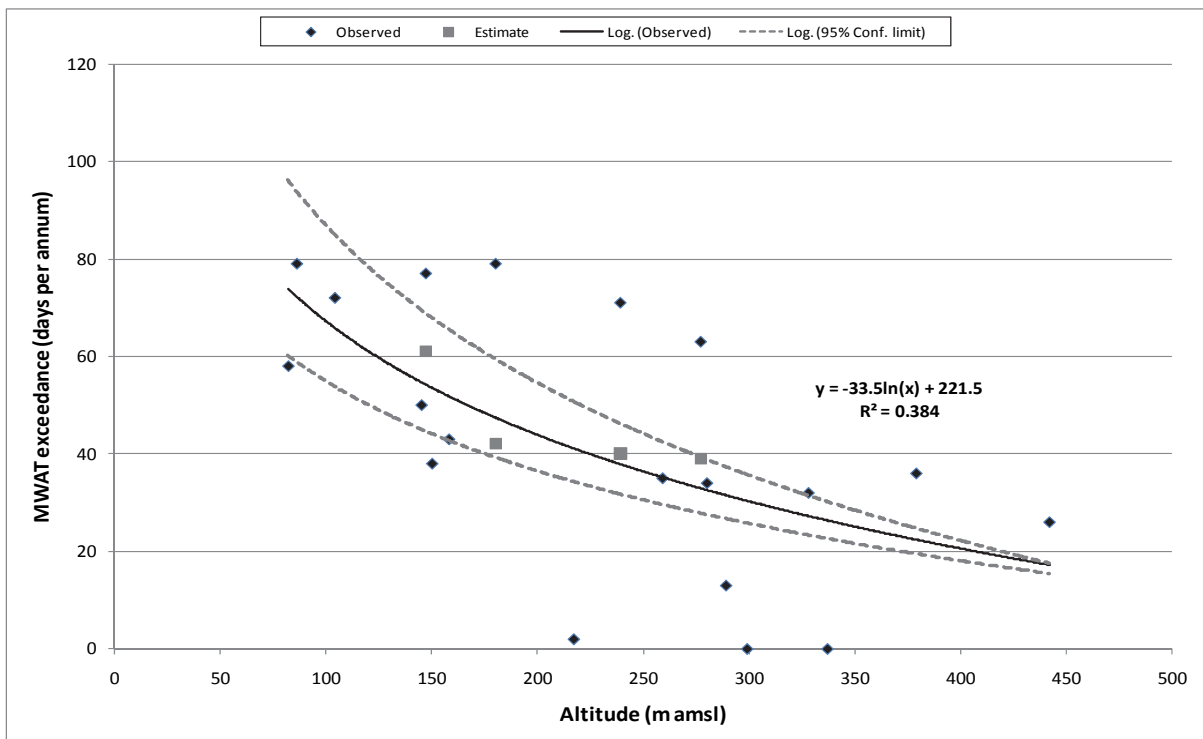


Figure 6.6 Relationship between MWAT exceedance and altitude plus 95% confidence envelope. Observed and expected values shown.

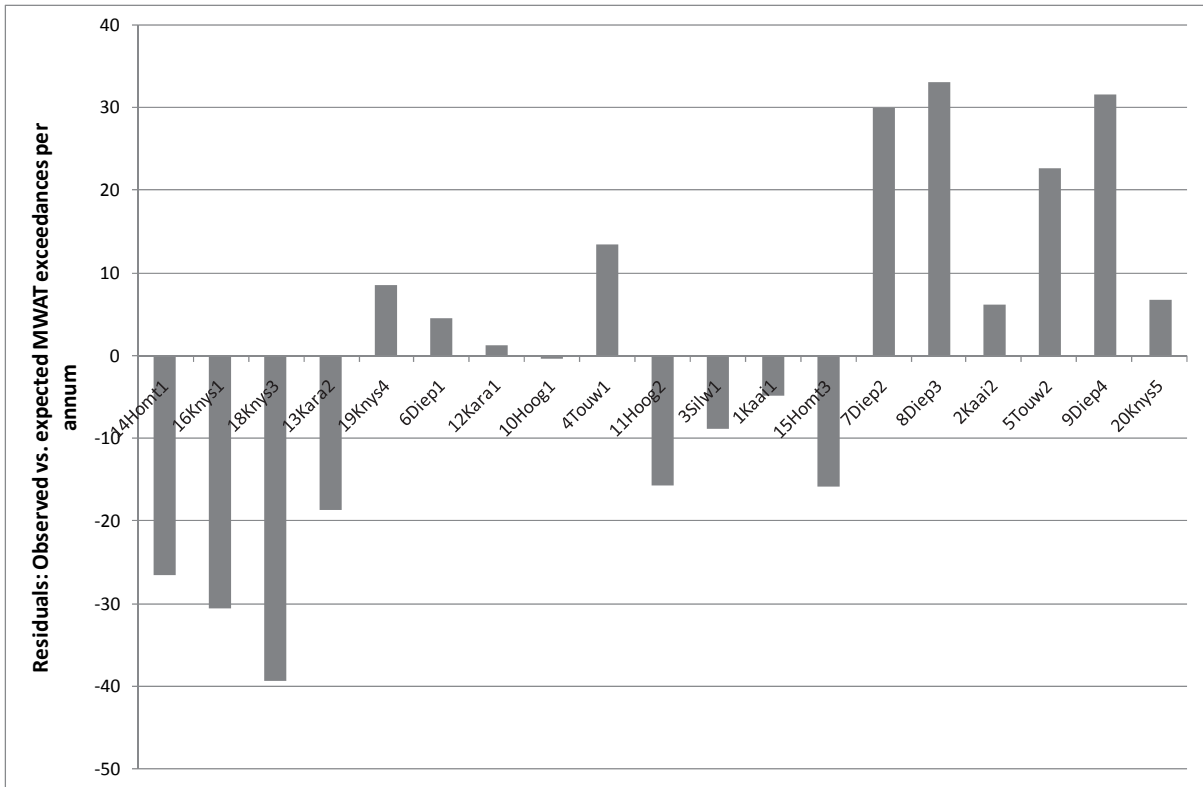


Figure 6.7 Residuals of MWAT exceedance based on expected versus observed values

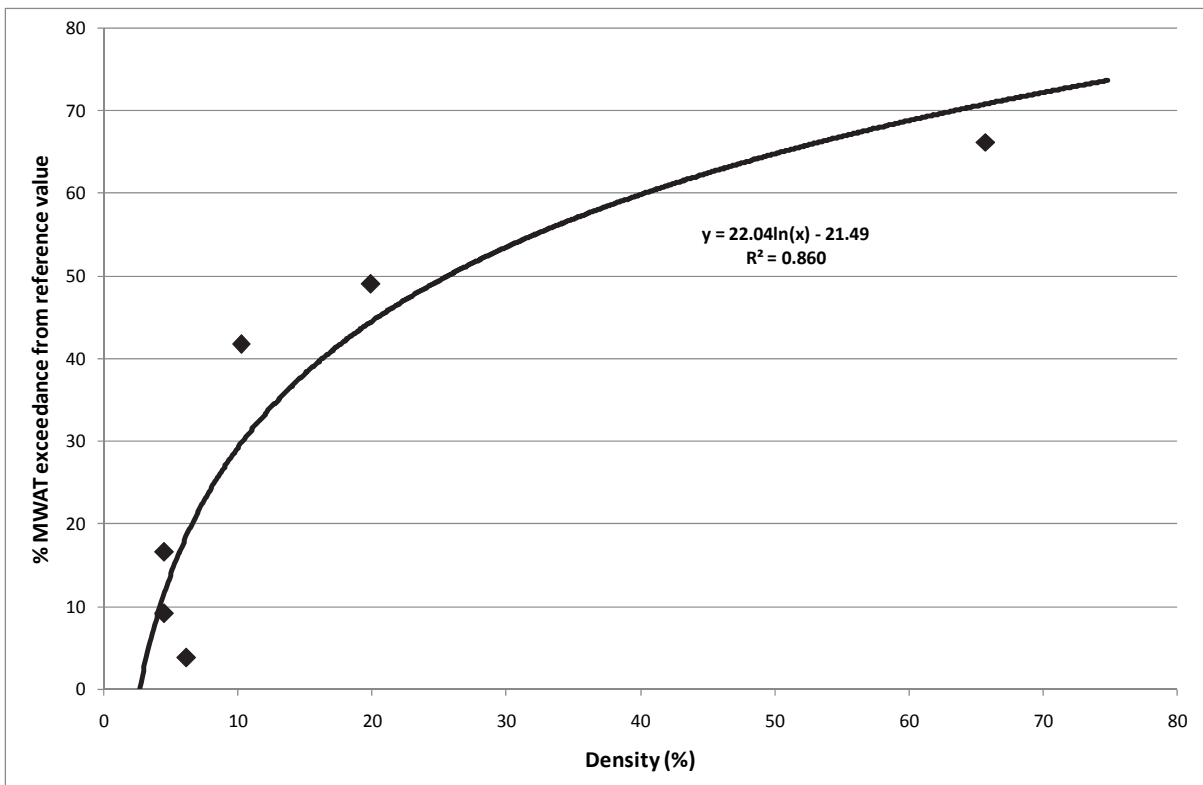


Figure 6.8 Relationship between % MWAT exceedance from reference value and on-site total density of alien riparian vegetation

### 6.3.3 Cost-Benefit model

Since NPV is reflective of current values derived from costs and benefits over time using a discount rate, the way in which costs and benefits are defined is critical to how NPV is expressed. For example, the costs for clearing over and initial treatment plus seven follow-up treatments would be expected to decay exponentially (Figure 6.9). Benefits are more difficult to measure, and in this case person day equivalents were assumed to reflect benefit. However, these could be expressed using various curves (linear and assuming an immediate benefit of clearing that remains constant over time, versus exponential increase inversely proportional to the cost curve). Such differences in how benefit is measured fundamentally affect how NPV is calculated, and consequently how management options are prioritized. For example, on the basis of how the NPV were estimated per management option using a constant benefit over time, the “Do nothing” option was the most detrimental, while a pro-active management approach provided the highest value (Figure 6.10). Value progressively declined as age of trees increased. The value of clearing also appeared to increase as density increased. Based on illustrative NPVs using different management options at different densities, and with an assumed constant benefit of 400 person days for each treatment, the “Do nothing” option becomes the most expensive management option, while the “Proactive” approach yielded the highest NPV (Figure 6.11). This could further be expressed as a ratio of NPV to % gain towards a desired thermal reference condition (Figure 6.12).

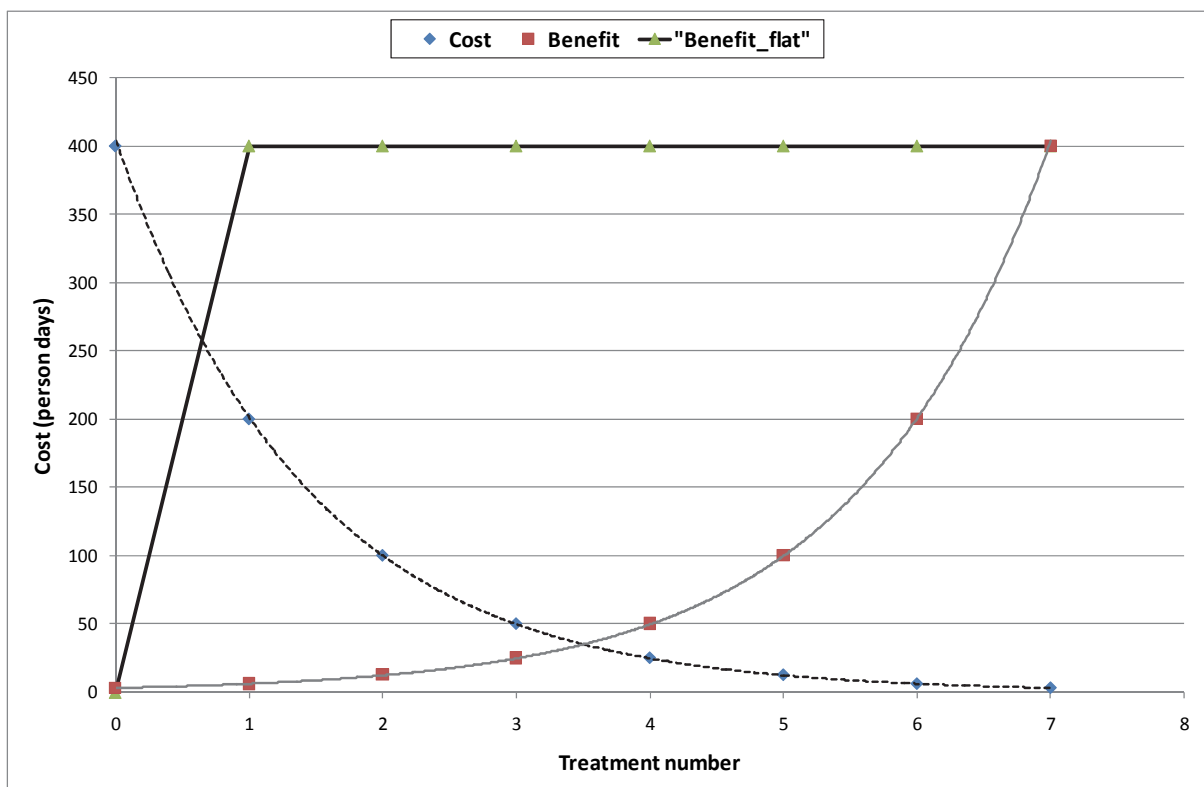


Figure 6.9 Two alternative benefit curves as they relate to a cost curve for successive treatments of clearing alien riparian vegetation

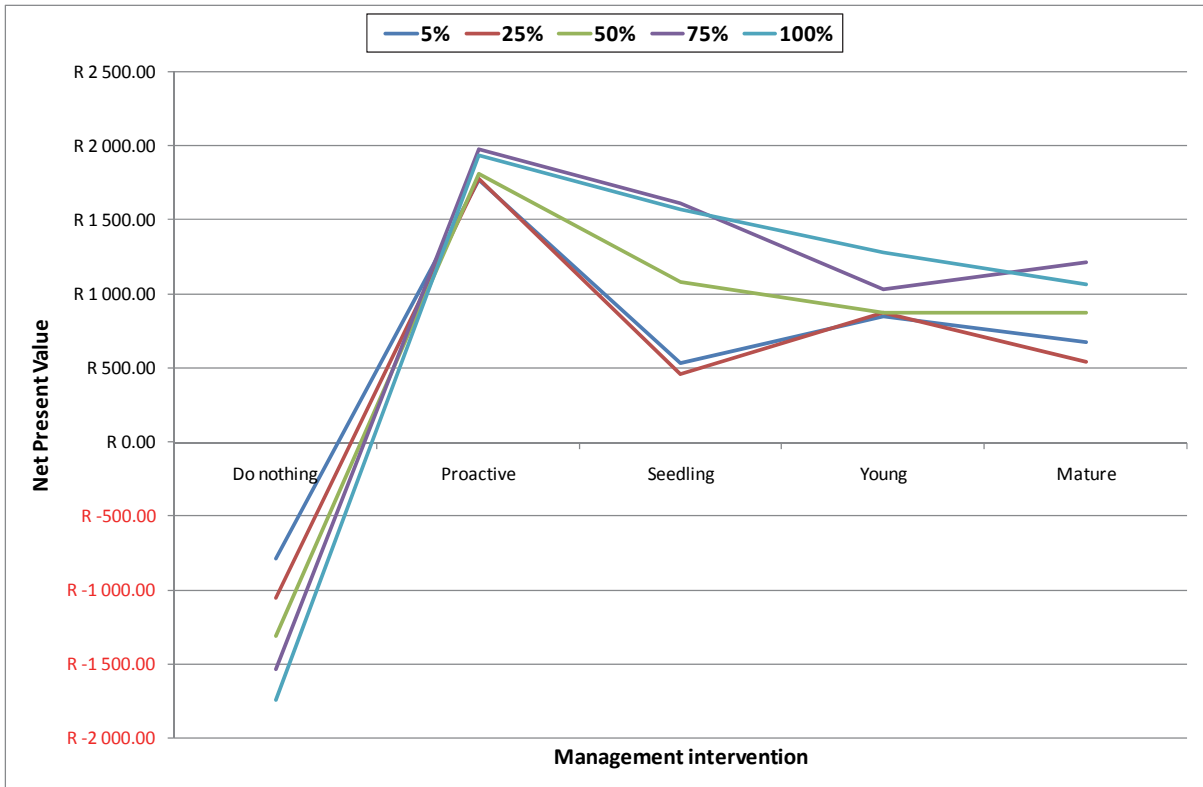


Figure 6.10 Change in NPV with management option at different densities of infestation

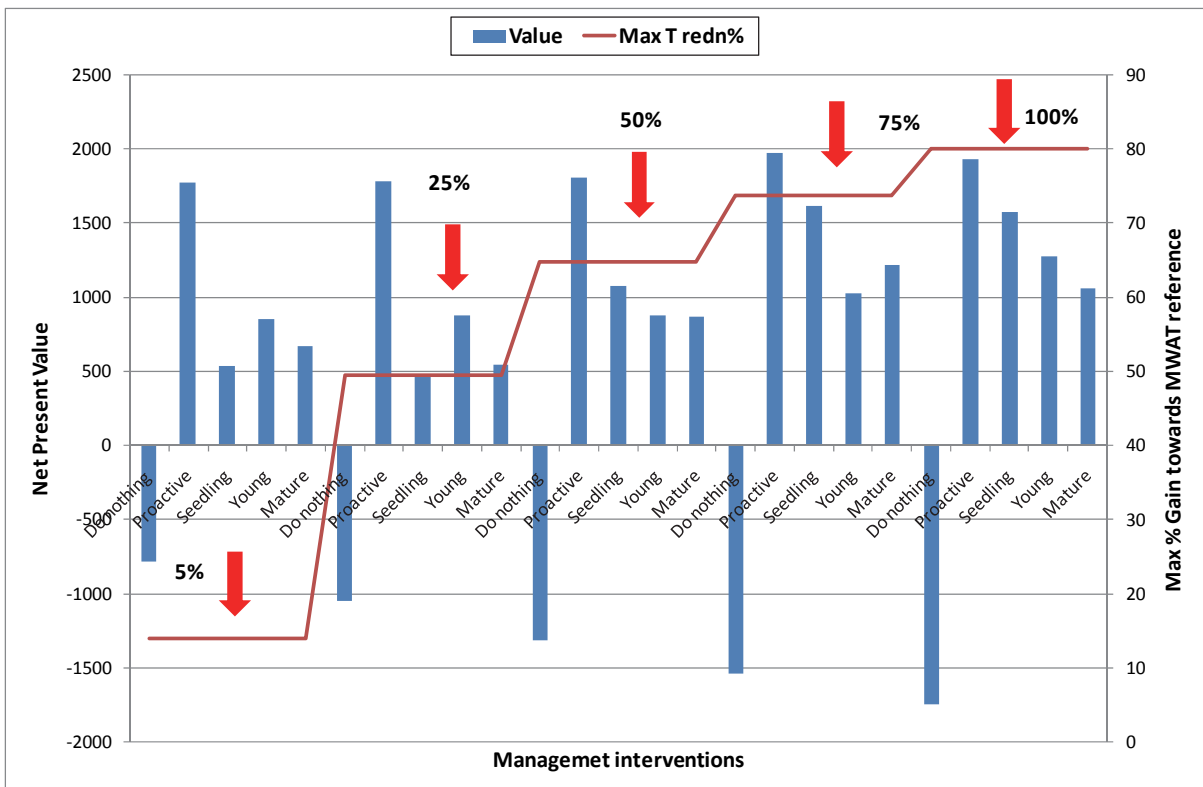
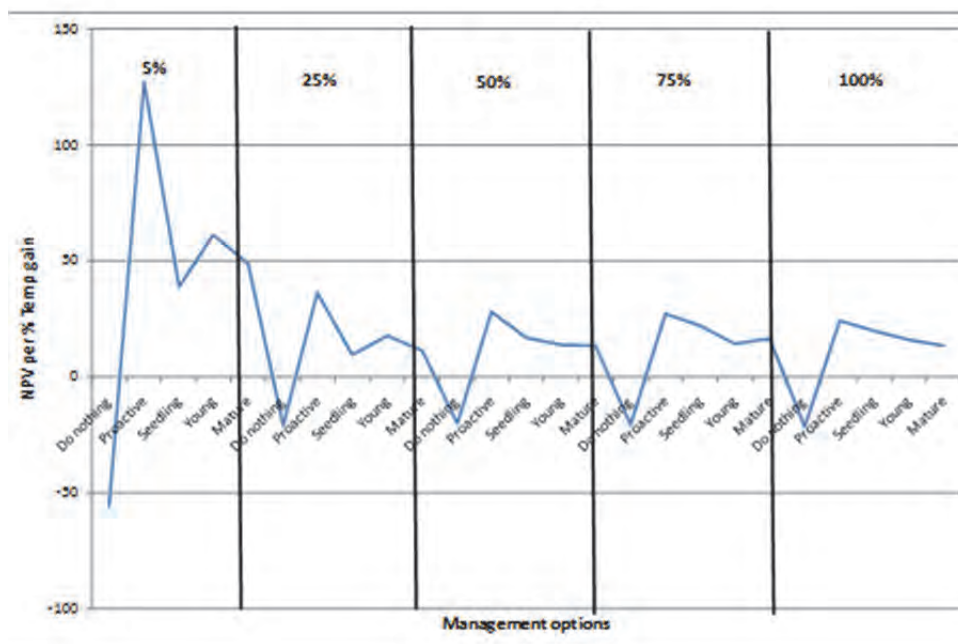


Figure 6.11 Net present value and maximum % gain towards MWAT reference for five different management options at different densities of infestation



**Figure 6.12 Relationship between NPV per % temperature gain and management option at different densities of infestation**

## 6.4 Discussion

The amount of natural vegetation at catchment scale has been found to be a good predictor of river habitat integrity (Amis et al. 2006). Conversely, streams in agricultural catchments usually remain in good condition until the extent of agriculture exceeds 30-50% (Allan 2004). Similarly, for every 10% of altered catchment land use, a correlative 6% loss in freshwater diversity was noted, as a linear relationship (Weitjers et al. 2009). The cumulative impact of small dams within a catchment has been shown to impact on water quality and quantity (Mantel et al. 2010). Within the riparian zone, *Acacia mearnsii* trees have been reported as causing bank stability, silt deposition and scouring (Cambray et al. 1995).

While Cambray et al. (1995) found that rivers with indigenous riparian vegetation had significantly higher diversity than impacted rivers with alien trees, Schael and King (2002) report that their most disturbed sites had similar invertebrate densities, levels of diversity and species richness to their least disturbed sites. Interestingly, water chemistry was not noted as a major driver, and that while no single driver identifiable, and rather that intensity/ degree of disturbance was more critical than the type of disturbance (Schael and King 2002). The latter study more closely approximates what has been found so far in this study, and de Moor (2006) states that for discerning differences in and along the length of rivers, family-level identification is adequate. However, a species-level approach may be required for a greater understanding of ecological requirements and that higher taxonomic data did not produce obvious catchment signatures (Schael and King 2002; de Moor 2006).

Thus far, it appears that MWAT threshold exceedances are elevated beyond what is expected at sites associated with higher densities of alien riparian vegetation. However, while this relationship may be correlative, it may not necessarily be causative. While shading by alien riparian vegetation may reduce water temperatures, they may also cause raised water temperatures due to decreased flows in catchments with high levels of alien infestation (Dye and Jarman 2004).

## Chapter 7. Conclusions and recommendations

### 7.1 Summary of main outcomes

Within the GRI, land use correlated strongly with altitudinal gradients. Forestry plantation areas typically coincide with fynbos zones and not indigenous forest patches. Pines dominate in the upper fynbos reaches extend beyond the riparian zone, while black wattles and blackwood dominate in the lower reaches and are restricted to the riparian zone, with a degree of association with palmet. Indigenous forest patches are largely intact and free from aliens. Most of the rivers in the study area have agriculture as a land use in their middle plateaus.

Water quality was relatively consistent across sites in spite of varying levels of alien vegetation infestation, but with underpinning geology translating into a distinct west-east grouping of river systems. Water temperatures were relatively similar across sites in spite of varying levels of site-specific alien riparian vegetation. The highly variable flows in these systems seem to suggest that they are all overwhelmingly flow-event driven systems. Consequently, species patterns of aquatic macroinvertebrates are likely to reflect the different flow regimes in each river system, with changes further compounded by impacts of alien invasive trees both at a catchment and site level.

At a family level of aquatic macroinvertebrates, all sites shared a common aquatic macroinvertebrate template (see also Schael and King 2005), differentiated by site-specific differences. Such differences are likely to be the result of interactions between flow patterns, thermal regimes, biogeography and catchment land use. Study results do, however, suggest differences in river productivity, as well as inter- and intra-specific differences in thermal tolerances between river systems. These thermal tolerance differences are more likely to be a response to flow regime variability, rather than resultant thermal changes. However, in response to the flow regime and cumulative upstream impacts of catchment and alien riparian vegetation, signs of cumulative thermal stress and departure from reference thermal condition were apparent.

To date, clearing of alien invasive species has been done using a single technique (mechanical clearing). While costs of clearing of alien vegetation – expressed as person days per hectare with an annually adjusted daily rate – are relatively well established and accepted within the industry, these figures remain subjective because of challenges in estimating alien plant infestation densities. Firstly, stands of aliens consist of mixed species and mixed age classes. Secondly, costs are based on densities expressed as a percentage, while field assessment densities (e.g. transects of PCQM) are based on numbers of trees per 100 m<sup>2</sup>. There does not appear to be a relationship to reconcile these. Thirdly, clearing costs are difficult to budget for on a medium to long term basis (5-10 year scale).

The economic model shows potential to use water temperature thermal exceedances as a means of prioritizing catchments for clearing of alien invasive species, and that the level of site specific alien riparian invasion is more a reflector of what is cumulatively occurring upstream in the catchment. A critical issue for these models is to refine how benefits are



measured, and fundamentally affects how Net Present Values are derived. It is easier to justify budgets if costs are related to benefits, and both metrics combined using Net Present Value incorporating a suitable discount function. A key future research area is thus on how to measure benefits of clearing alien vegetation. The use of person day equivalents is one option, but it is recommended that further benefit metrics that can be translated into a financial value are investigated.

As a management focus for the rivers of the GRI, based on the findings from this study, the most pragmatic approach for river conservation would be to prioritize catchments based on conservation value. Logical groupings of these would be to include fynbos and indigenous forest areas of importance, as well as one from each of the water quality groups identified here. Upstream areas, depending on site access, should be cleared first, and this would translate into thermal regimes returning to reference condition at downstream sites.

## **7.2 Lessons learnt**

It is difficult to compare cost-benefits of different clearing techniques, as currently only one is used. Ideally, the costs and benefits of mechanical clearing should be compared against emerging alternative options that include fire (which is effective in killing wattle seedbanks which have 50-100 year viabilities), and the use of biocontrol agents. Relative benefits of catchment-wide clearing versus on-site clearing need to be assessed through controlled experiments. It is better to have sites/reaches/rivers that represent cleared, invaded (infested) and natural (reference). In this study, it was difficult to separate these compounding effects.

The most practical approach for this to be achieved would be to continue discussions with key members of the SANParks BSP, particularly in terms of how budgeting for alien invasive clearing is undertaken over multi-year time intervals. This would be particularly facilitated through the incorporation of cost-comparable benefit values, with both of these being related to current budgets using a Net Present Value matrix. There is also a need to validate the models, where results from such clearing would need to be measured over a long-term period (5-10 years) because initial responses of aquatic systems would merely be reflecting recovery from disturbance.

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## Appendices

### Appendix 2.1 Notes from discussions of river systems considered for this study

| River                | Notes  | Decision/ Pair with...               |
|----------------------|--|--------------------------------------|
| Kaaimans             | Largely natural; Fynbos in upper catchment, with pockets of indigenous forest in middle and lower catchment. Access via Cape Nature in upper catchment – permits needed, weir above SANParks area in the lower catchment.  | Touws River                          |
| Touws                | Black wattle; accessibility limited in upper sections. Largely natural. Mid-region with RHP sites. Limited to 1-2 sites due to town and contours.  | Kaaimans River                       |
| Diep/Wolwekloof      | Pine (current and cleared) in upper reaches; accessible; Bergplaas area; middle reaches heavily farmed; lower reaches heavily infested with black wattle   | Hoekraal and Karatara Rivers         |
| Hoekraal/ Hoogekraal | Similar to Diep R.; black wattle infestations and pine plantations upstream. Exit policy. RHP site. Steeper gradient, lower reaches inaccessible.  | Second choice to Diep (if necessary) |
| Karatara             | Upper catchment accessible, pine plantation, with forestry exit areas. Black wattle in riparian zones. Potentially more accessible than Hoekraal River system. Good river with mixture of impacted and natural land cover, plus black wattle in riparian zone. Important as Sedgfield water source. Private alien clearing as part of Knysna Catchment Forum. Occurs generally over private land – contact owners for access. RHP site with weir.  | Hoekraal and Diep Rivers             |
| Homtini              | Accessible in upper and lower sections. Potential aquifer-dependant system. Fynbos in upper reaches, pine plantation, some areas decommissioned. Black wattle in lower reaches (PG Bison – would need to sort out access permits). Infested with black wattle outside SANPark regions. Network of tributaries in upper reaches as potential for paired sites, e.g. two paired systems in upper reaches = Millwood Creek (old gold mining area) and Jubilee Creek (natural; accessible). Marine influence extends far upstream. Sewage Works near Seven Passes area – avoid this area. Includes areas with forestry exit areas. | Knysna River (upper)                 |
| Knysna               | One of few systems with good middle  | Homtini River (upper)                |

| River     | Notes   | Decision/ Pair with...                  |
|-----------|---|---|
|           | sections, but no fynbos. Important as water supply system (G&S), with plans to impound within next ten years. Upper Knysna River with decommissioned plantations and old forestry roads, and sections of original Knysna Forest. Potential to pair upper tributaries of Gouna River (natural forest; waterfall and two road crossings; unique biodiversity linked to geology) and Rooiels River (old plantation)  |   |
| Keurbooms | Soetkraal area (Palmiet River as trib. of Keurbooms) important to Keurbooms River and Plettenberg Bay (Resilience + Goods and Services (G&S)). Keurbooms/Kwaai confluence and De Vlug areas as interesting sites; Palmiet River (tributary) as natural fynbos system and occurs between parallel mountain ranges, and is an “atypical” system. Extensive water temperature and aquatic macroinvertebrate data from previous studies (Rivers-Moore and Eady, unpub. data). | Use as historical data                  |
| Salt      | Fishless system. Upper reaches largely intact, lower reaches impacted. Stragglers from pine plantation and black wattle. Extensive water temperature and aquatic macroinvertebrate data from previous studies (de Moor, unpub. data).   | Lottering River; use as historical data |
| Groot     | Reference system – largely natural (and NFEPA free-flowing system). Would need helicopter access. Extensive water temperature and aquatic macroinvertebrate data from previous studies (de Moor and Bellingham, unpub. data).   | Use as historical data                  |
| Bloukrans | Atypical system, with large bedrock areas. Previously exhibit wide daily water temperature fluctuations. Not recommended as a good system for this study.   | Omit                                    |
| Lottering | Fishless system. Could be considered as an additional system for added value. Has two tributaries (fynbos and forest) and alien vegetation in lower system. Macroinvertebrate data from monitoring studies available (pers. comm. G Diedericks)   | Salt River; use as historical data      |
| Elandsbos | Accessible, lower reaches full of black wattle and pines, and patches of indigenous forest interspersed   | Use as historical data                  |
| Storms    | Lower site accessible but upper site inaccessible   | Use as historical data                  |



**Appendix 2.2 Details of the dates of loggers installation and period of logged water temperature data**

| Site     | Logged period               | Patching/Simulated           |
|----------|-----------------------------|------------------------------|
| 1-Kaai1  | 2 June 2013 – 29 Sept 2014  | None                         |
| 2-Kaai2  | 10 Sept 2013 – 4 Oct 2014   | 30-day moving average        |
| 3-Silw1  | 10 Sept 2013 – 29 Sept 2014 | 30-day moving average        |
| 4-Touw1  | 12 Sept 2013 – 30 Sept 2014 | 30-day moving average        |
| 5-Touw2  | 15 Jan 2014 – 3 Oct 2014    | Linear regression (Eqtn. A1) |
| 6-Diep1  | 11 Sept 2013 – 2 Oct 2014   | 30-day moving average        |
| 7-Diep2  | 11 Sept 2013 – 2 Oct 2014   | 30-day moving average        |
| 8-Diep3  | 17 Jan 2014 – 8 Apr 2014    | Linear regression (Eqtn. A2) |
| 9-Diep4  | 12 Sept 2013 – 5 Oct 2014   | 30-day moving average        |
| 10-Hoog1 | 12 Sept 2013 – 30 Sept 2014 | 30-day moving average        |
| 11-Hoog2 | 13 Sept 2013 – 5 Oct 2014   | 30-day moving average        |
| 12-Kara1 | 16 Sept 2013 – 6 Oct 2014   | 30-day moving average        |
| 13-Kara2 | 16 Sept 2-13 – 6 Oct 2014   | 30-day moving average        |
| 14-Homt1 | 31 May 2013 – 9 Oct 2014    | None                         |
| 15-Homt3 | 13 Sept 2013 – 3 Oct 2014   | 30-day moving average        |
| 16-Knys1 | 16 Sept 2013 – 9 Oct 2014   | 30-day moving average        |
| 18-Knys3 | 16 Jan 2014 – 7 Oct 2014    | Linear regression (Eqtn. A3) |
| 19-Knys4 | 16 Jan 2014 – 7 Oct 2014    | Linear regression (Eqtn. A4) |
| 20-Knys5 | 17 Sept 2013 – 7 Oct 2014   | 30-day moving average        |

5-Touw2 = 2.811+0.881\*2-Kaai2 [A1; R<sup>2</sup> = 0.95; n = 1850]

8-Diep3 = 0.667+0.934\*9-Diep4 [A2; R<sup>2</sup> = 0.95; n = 1852]

18-Knys3 = -2.272+1.161\*14-Homt1 [A3; R<sup>2</sup> = 0.92; n = 1915]

19-Knys3 = 1.306+1.021\*14-Homt1 [A4; R<sup>2</sup> = 0.93; n = 1913]

**Appendix 4.1. Macroinvertebrate taxa recorded for each month and site.**

| Order              | Site              | 1Kaai1 | 2Kaai2 | 5Touw2 | 6Diep1 | 9Diep4 | 10Hoog1 |
|--------------------|-------------------|--------|--------|--------|--------|--------|---------|
|                    | Family / Date     | Sep-13 | Sep-13 | Sep-13 | Sep-13 | Sep-13 | Sep-13  |
| Acarina            | Hydrachnellae     | 11     | 12     | 0      | 60     | 0      | 0       |
| Annelida           | Hirudinae         | 0      | 0      | 0      | 0      | 0      | 0       |
| Annelida           | Oligochaeta       | 0      | 0      | 0      | 2      | 0      | 0       |
| Araneae            | Thalassinae       | 0      | 0      | 0      | 0      | 1      | 0       |
| Coleoptera         | Dytiscidae        | 1      | 0      | 1      | 1      | 0      | 0       |
| Coleoptera         | Elmidae           | 66     | 9      | 30     | 77     | 5      | 5       |
| Coleoptera         | Gyrinidae         | 0      | 0      | 0      | 1      | 0      | 0       |
| Coleoptera         | Hydraenidae       | 2      | 2      | 0      | 0      | 0      | 0       |
| Coleoptera         | Hydrophilidae     | 11     | 1      | 0      | 3      | 0      | 0       |
| Coleoptera         | Scirtidae         | 19     | 3      | 2      | 13     | 0      | 3       |
| Diptera            | Athericidae       | 0      | 0      | 0      | 4      | 0      | 0       |
| Diptera            | Ceratopogonidae   | 0      | 0      | 0      | 10     | 0      | 0       |
| Diptera            | Chironomidae      | 109    | 40     | 9      | 178    | 15     | 27      |
| Diptera            | Ephydriidae       | 21     | 0      | 0      | 0      | 0      | 0       |
| Diptera            | Simuliidae        | 414    | 102    | 6      | 38     | 119    | 58      |
| Diptera            | Syrphidae         | 0      | 0      | 0      | 2      | 0      | 0       |
| Diptera            | Tipulidae         | 1      | 1      | 0      | 0      | 0      | 0       |
| Ephemeroptera      | Baetidae          | 269    | 126    | 20     | 286    | 47     | 50      |
| Ephemeroptera      | Heptageniidae     | 0      | 1      | 4      | 52     | 17     | 0       |
| Ephemeroptera      | Leptophlebiidae   | 2      | 0      | 20     | 1      | 0      | 3       |
| Ephemeroptera      | Teloganodidae     | 95     | 29     | 86     | 56     | 0      | 68      |
| Hemiptera          | Belostomatidae    | 0      | 0      | 0      | 0      | 0      | 0       |
| Hemiptera          | Pleidae           | 0      | 0      | 0      | 0      | 0      | 0       |
| Lepidoptera        | Pyralidae         | 0      | 0      | 0      | 0      | 0      | 0       |
| Megaloptera        | Corydalidae       | 0      | 0      | 3      | 2      | 1      | 0       |
| Mollusca           | Ancylidae         | 0      | 0      | 0      | 0      | 0      | 0       |
| Mollusca           | Sphaeriidae       | 0      | 0      | 0      | 119    | 0      | 0       |
| Nematode           | Nematoda          | 0      | 0      | 0      | 0      | 0      | 0       |
| Odonata Anisoptera | Aeshnidae         | 0      | 0      | 0      | 0      | 1      | 0       |
| Odonata Anisoptera | Libellulidae      | 1      | 0      | 2      | 0      | 0      | 0       |
| Odonata Zygoptera  | Chlorocyphidae    | 0      | 0      | 0      | 0      | 3      | 0       |
| Odonata Zygoptera  | Coenagrionidae    | 0      | 0      | 0      | 0      | 0      | 0       |
| Odonata Zygoptera  | Protoneuridae     | 0      | 0      | 0      | 2      | 0      | 0       |
| Plecoptera         | Notonemouridae    | 8      | 1      | 0      | 7      | 0      | 2       |
| Trichoptera        | Barbarochthonidae | 37     | 9      | 4      | 0      | 0      | 21      |
| Trichoptera        | Caddisflies       | 0      | 0      | 0      | 0      | 0      | 0       |
| Trichoptera        | Glossosomatidae   | 4      | 0      | 0      | 4      | 4      | 0       |
| Trichoptera        | Hydropsychidae    | 9      | 7      | 6      | 2      | 17     | 25      |
| Trichoptera        | Hydroptilidae     | 2      | 0      | 0      | 15     | 0      | 0       |
| Trichoptera        | Leptoceridae      | 18     | 0      | 0      | 20     | 0      | 0       |
| Trichoptera        | Petrothrincidae   | 0      | 0      | 0      | 0      | 0      | 0       |
| Trichoptera        | Pisuliidae        | 0      | 0      | 0      | 0      | 0      | 0       |
| Trichoptera        | Sericostomatidae  | 0      | 0      | 30     | 0      | 7      | 0       |

| <b>Order</b>       | <b>Site<br/>Family / Date</b> | <b>11Hoog2<br/>Sep-13</b> | <b>14Homt1<br/>Sep-13</b> | <b>15Homt3<br/>Sep-13</b> | <b>19Knys4<br/>Sep-13</b> | <b>1Kaai1<br/>Jan-14</b> | <b>2Kaai2<br/>Jan-14</b> |
|--------------------|-------------------------------|---------------------------|---------------------------|---------------------------|---------------------------|--------------------------|--------------------------|
| Acarina            | Hydrachnellae                 | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Annelida           | Hirudinae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Annelida           | Oligochaeta                   | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Araneae            | Thalassinae                   | 0                         | 0                         | 0                         | 0                         | 1                        | 0                        |
| Coleoptera         | Dytiscidae                    | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Coleoptera         | Elmidae                       | 26                        | 22                        | 59                        | 17                        | 6                        | 4                        |
| Coleoptera         | Gyrinidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Coleoptera         | Hydraenidae                   | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Coleoptera         | Hydrophilidae                 | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Coleoptera         | Scirtidae                     | 7                         | 0                         | 0                         | 0                         | 5                        | 0                        |
| Diptera            | Athericidae                   | 0                         | 0                         | 0                         | 0                         | 0                        | 1                        |
| Diptera            | Ceratopogonidae               | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Diptera            | Chironomidae                  | 4                         | 0                         | 203                       | 9                         | 7                        | 4                        |
| Diptera            | Ephydriidae                   | 0                         | 2                         | 0                         | 0                         | 0                        | 0                        |
| Diptera            | Simuliidae                    | 2                         | 1                         | 4                         | 95                        | 257                      | 18                       |
| Diptera            | Syrphidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Diptera            | Tipulidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Ephemeroptera      | Baetidae                      | 5                         | 45                        | 57                        | 10                        | 85                       | 6                        |
| Ephemeroptera      | Heptageniidae                 | 0                         | 0                         | 1                         | 0                         | 0                        | 0                        |
| Ephemeroptera      | Leptophlebiidae               | 3                         | 14                        | 0                         | 0                         | 17                       | 8                        |
| Ephemeroptera      | Teloganodidae                 | 25                        | 46                        | 169                       | 23                        | 0                        | 0                        |
| Hemiptera          | Belostomatidae                | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Hemiptera          | Pleidae                       | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Lepidoptera        | Pyralidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Megaloptera        | Corydalidae                   | 4                         | 2                         | 0                         | 3                         | 0                        | 1                        |
| Mollusca           | Ancylidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Mollusca           | Sphaeriidae                   | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Nematode           | Nematoda                      | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Odonata Anisoptera | Aeshnidae                     | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Odonata Anisoptera | Libellulidae                  | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Odonata Zygoptera  | Chlorocyphidae                | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Odonata Zygoptera  | Coenagrionidae                | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Odonata Zygoptera  | Protoneuridae                 | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Plecoptera         | Notonemouridae                | 0                         | 6                         | 0                         | 37                        | 2                        | 0                        |
| Trichoptera        | Barbarochthonidae             | 62                        | 342                       | 28                        | 30                        | 6                        | 7                        |
| Trichoptera        | Caddisflies                   | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Trichoptera        | Glossosomatidae               | 0                         | 2                         | 0                         | 3                         | 0                        | 0                        |
| Trichoptera        | Hydropsychidae                | 4                         | 6                         | 5                         | 41                        | 10                       | 3                        |
| Trichoptera        | Hydroptilidae                 | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Trichoptera        | Leptoceridae                  | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Trichoptera        | Petrothrincidae               | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Trichoptera        | Pisuliidae                    | 0                         | 0                         | 0                         | 0                         | 0                        | 0                        |
| Trichoptera        | Sericostomatidae              | 0                         | 0                         | 0                         | 11                        | 0                        | 0                        |

| <b>Order</b>         | <b>Site<br/>Family / Date</b> | <b>5Touw2<br/>Jan-14</b> | <b>6Diep1<br/>Jan-14</b> | <b>9Diep4<br/>Jan-14</b> | <b>10Hoog1<br/>Jan-14</b> | <b>11Hoog2<br/>Jan-14</b> | <b>14Homt1<br/>Jan-14</b> |
|----------------------|-------------------------------|--------------------------|--------------------------|--------------------------|---------------------------|---------------------------|---------------------------|
| Acarina              | Hydrachnellae                 | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Annelida             | Hirudinae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Annelida             | Oligochaeta                   | 7                        | 0                        | 0                        | 0                         | 9                         | 0                         |
| Araneae              | Thalassinae                   | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Coleoptera           | Dytiscidae                    | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Coleoptera           | Elmidae                       | 17                       | 14                       | 4                        | 19                        | 4                         | 51                        |
| Coleoptera           | Gyrinidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Coleoptera           | Hydraenidae                   | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Coleoptera           | Hydrophilidae                 | 0                        | 0                        | 0                        | 0                         | 0                         | 1                         |
| Coleoptera           | Scirtidae                     | 0                        | 0                        | 0                        | 3                         | 0                         | 7                         |
| Diptera              | Athericidae                   | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Diptera              | Ceratopogonidae               | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Diptera              | Chironomidae                  | 41                       | 1                        | 4                        | 1                         | 7                         | 11                        |
| Diptera              | Ephydriidae                   | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Diptera              | Simuliidae                    | 114                      | 11                       | 48                       | 19                        | 20                        | 63                        |
| Diptera              | Syrphidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Diptera              | Tipulidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Ephemeroptera        | Baetidae                      | 18                       | 57                       | 44                       | 9                         | 31                        | 82                        |
| Ephemeroptera        | Heptageniidae                 | 0                        | 7                        | 13                       | 0                         | 0                         | 0                         |
| Ephemeroptera        | Leptophlebiidae               | 0                        | 0                        | 6                        | 0                         | 6                         | 8                         |
| Ephemeroptera        | Teloganodidae                 | 0                        | 0                        | 0                        | 4                         | 0                         | 9                         |
| Hemiptera            | Belostomatidae                | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Hemiptera            | Pleidae                       | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Lepidoptera          | Pyralidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Megaloptera          | Corydalidae                   | 2                        | 0                        | 0                        | 1                         | 1                         | 3                         |
| Mollusca             | Ancylidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Mollusca             | Sphaeriidae                   | 0                        | 0                        | 0                        | 0                         | 0                         | 1                         |
| Nematode             | Nematoda                      | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Odonata (Anisoptera) | Aeshnidae                     | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Odonata (Anisoptera) | Libellulidae                  | 1                        | 0                        | 4                        | 0                         | 0                         | 0                         |
| Odonata (Zygoptera)  | Chlorocyphidae                | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Odonata (Zygoptera)  | Coenagrionidae                | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Odonata (Zygoptera)  | Protoneuridae                 | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Plecoptera           | Notonemouridae                | 3                        | 0                        | 3                        | 0                         | 0                         | 0                         |
| Trichoptera          | Barbarochthonidae             | 11                       | 0                        | 0                        | 12                        | 23                        | 75                        |
| Trichoptera          | Caddisflies                   | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Glossosomatidae               | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Hydropsychidae                | 4                        | 0                        | 22                       | 2                         | 5                         | 19                        |
| Trichoptera          | Hydroptilidae                 | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Leptoceridae                  | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Petrothrincidae               | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Pisuliidae                    | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |
| Trichoptera          | Sericostomatidae              | 0                        | 0                        | 0                        | 0                         | 0                         | 0                         |

| Order                | Site              | 15Homt3 | 19Knys4 | 1Kaai1 | 2Kaai2 | 5Touw2 | 6Diep1 |
|----------------------|-------------------|---------|---------|--------|--------|--------|--------|
|                      | Family / Date     | Jan-14  | Jan-14  | Apr-14 | Apr-14 | Apr-14 | Apr-14 |
| Acarina              | Hydrachnellae     | 0       | 0       | 0      | 0      | 0      | 0      |
| Annelida             | Hirudinae         | 0       | 0       | 0      | 0      | 0      | 1      |
| Annelida             | Oligochaeta       | 5       | 1       | 0      | 2      | 0      | 1      |
| Araneae              | Thalassinae       | 0       | 0       | 0      | 1      | 0      | 0      |
| Coleoptera           | Dytiscidae        | 0       | 0       | 0      | 0      | 0      | 1      |
| Coleoptera           | Elmidae           | 29      | 32      | 47     | 43     | 44     | 66     |
| Coleoptera           | Gyrinidae         | 0       | 0       | 0      | 0      | 0      | 0      |
| Coleoptera           | Hydraenidae       | 0       | 0       | 0      | 0      | 0      | 0      |
| Coleoptera           | Hydrophilidae     | 0       | 0       | 0      | 0      | 0      | 0      |
| Coleoptera           | Scirtidae         | 2       | 0       | 0      | 3      | 7      | 4      |
| Diptera              | Athericidae       | 0       | 0       | 0      | 0      | 0      | 0      |
| Diptera              | Ceratopogonidae   | 0       | 0       | 0      | 0      | 0      | 0      |
| Diptera              | Chironomidae      | 8       | 12      | 34     | 9      | 19     | 12     |
| Diptera              | Ephydriidae       | 0       | 0       | 0      | 0      | 0      | 0      |
| Diptera              | Simuliidae        | 30      | 15      | 89     | 68     | 10     | 51     |
| Diptera              | Syrphidae         | 0       | 0       | 0      | 0      | 0      | 0      |
| Diptera              | Tipulidae         | 0       | 0       | 0      | 0      | 0      | 0      |
| Ephemeroptera        | Baetidae          | 47      | 21      | 10     | 6      | 32     | 79     |
| Ephemeroptera        | Heptageniidae     | 0       | 0       | 0      | 2      | 23     | 10     |
| Ephemeroptera        | Leptophlebiidae   | 0       | 2       | 0      | 5      | 0      | 0      |
| Ephemeroptera        | Teloganodidae     | 3       | 0       | 2      | 0      | 27     | 5      |
| Hemiptera            | Belostomatidae    | 0       | 1       | 1      | 0      | 0      | 0      |
| Hemiptera            | Pleidae           | 0       | 0       | 0      | 0      | 0      | 0      |
| Lepidoptera          | Pyalidae          | 0       | 0       | 1      | 0      | 0      | 0      |
| Megaloptera          | Corydalidae       | 3       | 2       | 6      | 2      | 4      | 0      |
| Mollusca             | Ancylidae         | 0       | 1       | 0      | 0      | 0      | 0      |
| Mollusca             | Sphaeriidae       | 0       | 0       | 0      | 0      | 0      | 0      |
| Nematode             | Nematoda          | 0       | 0       | 0      | 1      | 0      | 0      |
| Odonata (Anisoptera) | Aeshnidae         | 0       | 0       | 0      | 0      | 0      | 0      |
| Odonata (Anisoptera) | Libellulidae      | 0       | 0       | 0      | 0      | 0      | 0      |
| Odonata (Zygoptera)  | Chlorocyphidae    | 0       | 0       | 0      | 0      | 0      | 0      |
| Odonata (Zygoptera)  | Coenagrionidae    | 0       | 0       | 0      | 0      | 0      | 0      |
| Odonata (Zygoptera)  | Protoneuridae     | 0       | 0       | 0      | 0      | 0      | 0      |
| Plecoptera           | Notonemouridae    | 0       | 5       | 4      | 6      | 1      | 7      |
| Trichoptera          | Barbarochthonidae | 39      | 8       | 14     | 0      | 19     | 0      |
| Trichoptera          | Caddisflies       | 0       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Glossosomatidae   | 0       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Hydropsychidae    | 14      | 51      | 2      | 38     | 29     | 24     |
| Trichoptera          | Hydroptilidae     | 0       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Leptoceridae      | 1       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Petrothrincidae   | 0       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Pisuliidae        | 0       | 0       | 0      | 0      | 0      | 0      |
| Trichoptera          | Sericostomatidae  | 0       | 0       | 1      | 0      | 3      | 0      |

| <b>Order</b>       | <b>Site<br/>Family / Date</b> | <b>9Diep4<br/>Apr-14</b> | <b>10Hoog1<br/>Apr-14</b> | <b>11Hoog2<br/>Apr-14</b> | <b>14Homt1<br/>Apr-14</b> | <b>15Homt3<br/>Apr-14</b> | <b>19Knys4<br/>Apr-14</b> |
|--------------------|-------------------------------|--------------------------|---------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| Acarina            | Hydrachnellae                 | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Annelida           | Hirudinae                     | 0                        | 0                         | 0                         | 1                         | 0                         | 0                         |
| Annelida           | Oligochaeta                   | 1                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Araneae            | Thalassinae                   | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Coleoptera         | Dytiscidae                    | 0                        | 0                         | 3                         | 0                         | 1                         | 0                         |
| Coleoptera         | Elmidae                       | 15                       | 18                        | 17                        | 68                        | 42                        | 57                        |
| Coleoptera         | Gyrinidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Coleoptera         | Hydraenidae                   | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Coleoptera         | Hydrophilidae                 | 0                        | 0                         | 1                         | 0                         | 0                         | 0                         |
| Coleoptera         | Scirtidae                     | 0                        | 12                        | 7                         | 1                         | 5                         | 6                         |
| Diptera            | Athericidae                   | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Diptera            | Ceratopogonidae               | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Diptera            | Chironomidae                  | 48                       | 2                         | 8                         | 5                         | 5                         | 5                         |
| Diptera            | Ephyridae                     | 0                        | 2                         | 0                         | 0                         | 0                         | 0                         |
| Diptera            | Simuliidae                    | 3                        | 34                        | 14                        | 15                        | 28                        | 2                         |
| Diptera            | Syrphidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Diptera            | Tipulidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Ephemeroptera      | Baetidae                      | 12                       | 30                        | 19                        | 40                        | 25                        | 21                        |
| Ephemeroptera      | Heptageniidae                 | 30                       | 0                         | 0                         | 0                         | 11                        | 0                         |
| Ephemeroptera      | Leptophlebiidae               | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Ephemeroptera      | Teloganodidae                 | 0                        | 19                        | 5                         | 14                        | 56                        | 2                         |
| Hemiptera          | Belostomatidae                | 0                        | 0                         | 4                         | 0                         | 0                         | 0                         |
| Hemiptera          | Pleidae                       | 0                        | 0                         | 0                         | 0                         | 1                         | 0                         |
| Lepidoptera        | Pyrilidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Megaloptera        | Corydalidae                   | 0                        | 1                         | 0                         | 5                         | 4                         | 0                         |
| Mollusca           | Ancylidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Mollusca           | Sphaeriidae                   | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Nematode           | Nematoda                      | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Odonata Anisoptera | Aeshnidae                     | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Odonata Anisoptera | Libellulidae                  | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Odonata Zygoptera  | Chlorocyphidae                | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Odonata Zygoptera  | Coenagrionidae                | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Odonata Zygoptera  | Protoneuridae                 | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Plecoptera         | Notonemouridae                | 0                        | 6                         | 7                         | 12                        | 8                         | 15                        |
| Trichoptera        | Barbarochthonidae             | 0                        | 20                        | 187                       | 118                       | 58                        | 70                        |
| Trichoptera        | Caddisflies                   | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Trichoptera        | Glossosomatidae               | 0                        | 0                         | 0                         | 1                         | 0                         | 1                         |
| Trichoptera        | Hydropsychidae                | 7                        | 3                         | 33                        | 17                        | 7                         | 47                        |
| Trichoptera        | Hydroptilidae                 | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Trichoptera        | Leptoceridae                  | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Trichoptera        | Petrothrincidae               | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Trichoptera        | Pisuliidae                    | 0                        | 0                         | 0                         | 0                         | 0                         | 0                         |
| Trichoptera        | Sericostomatidae              | 6                        | 5                         | 0                         | 0                         | 0                         | 5                         |

| <b>Order</b>         | <b>Site<br/>Family / Date</b> | <b>1Kaai1<br/>Oct-14</b> | <b>2Kaai2<br/>Oct-14</b> | <b>5Touw2<br/>Oct-14</b> | <b>6Diep1<br/>Oct-14</b> | <b>9Diep4<br/>Oct-14</b> | <b>10Hoog1<br/>Oct-14</b> |
|----------------------|-------------------------------|--------------------------|--------------------------|--------------------------|--------------------------|--------------------------|---------------------------|
| Acarina              | Hydrachnellae                 | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Annelida             | Hirudinae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Annelida             | Oligochaeta                   | 0                        | 4                        | 0                        | 1                        | 0                        | 0                         |
| Araneae              | Thalassinae                   | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Coleoptera           | Dytiscidae                    | 0                        | 0                        | 0                        | 0                        | 0                        | 1                         |
| Coleoptera           | Elmidae                       | 76                       | 29                       | 27                       | 120                      | 19                       | 12                        |
| Coleoptera           | Gyrinidae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Coleoptera           | Hydraenidae                   | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Coleoptera           | Hydrophilidae                 | 1                        | 0                        | 0                        | 0                        | 0                        | 2                         |
| Coleoptera           | Scirtidae                     | 12                       | 17                       | 19                       | 20                       | 0                        | 16                        |
| Diptera              | Athericidae                   | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Diptera              | Ceratopogonidae               | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Diptera              | Chironomidae                  | 36                       | 11                       | 39                       | 17                       | 14                       | 15                        |
| Diptera              | Ephyridae                     | 0                        | 1                        | 0                        | 2                        | 0                        | 1                         |
| Diptera              | Simuliidae                    | 24                       | 14                       | 52                       | 66                       | 181                      | 51                        |
| Diptera              | Syrphidae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Diptera              | Tipulidae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Ephemeroptera        | Baetidae                      | 79                       | 21                       | 11                       | 18                       | 87                       | 60                        |
| Ephemeroptera        | Heptageniidae                 | 0                        | 11                       | 9                        | 1                        | 17                       | 0                         |
| Ephemeroptera        | Leptophlebiidae               | 0                        | 1                        | 2                        | 13                       | 0                        | 4                         |
| Ephemeroptera        | Teloganodidae                 | 23                       | 25                       | 31                       | 3                        | 0                        | 46                        |
| Hemiptera            | Belostomatidae                | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Hemiptera            | Pleidae                       | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Lepidoptera          | Pyralidae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Megaloptera          | Corydalidae                   | 0                        | 2                        | 2                        | 0                        | 0                        | 0                         |
| Mollusca             | Ancylidae                     | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Mollusca             | Sphaeriidae                   | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Nematode             | Nematoda                      | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Odonata (Anisoptera) | Aeshnidae                     | 0                        | 1                        | 0                        | 0                        | 0                        | 0                         |
| Odonata (Anisoptera) | Libellulidae                  | 1                        | 0                        | 4                        | 0                        | 1                        | 0                         |
| Odonata (Zygoptera)  | Chlorocyphidae                | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Odonata (Zygoptera)  | Coenagrionidae                | 0                        | 1                        | 0                        | 0                        | 2                        | 0                         |
| Odonata (Zygoptera)  | Protoneuridae                 | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Plecoptera           | Notonemouridae                | 7                        | 0                        | 0                        | 5                        | 0                        | 0                         |
| Trichoptera          | Barbarochthonidae             | 48                       | 20                       | 27                       | 1                        | 0                        | 91                        |
| Trichoptera          | Caddisflies                   | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Trichoptera          | Glossosomatidae               | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Trichoptera          | Hydropsychidae                | 54                       | 32                       | 26                       | 13                       | 12                       | 5                         |
| Trichoptera          | Hydroptilidae                 | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Trichoptera          | Leptoceridae                  | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Trichoptera          | Petrothrincidae               | 0                        | 0                        | 0                        | 0                        | 0                        | 1                         |
| Trichoptera          | Pisuliidae                    | 0                        | 0                        | 0                        | 0                        | 0                        | 0                         |
| Trichoptera          | Sericostomatidae              | 0                        | 0                        | 3                        | 0                        | 5                        | 0                         |

| <b>Order</b>         | <b>Site<br/>Family / Date</b> | <b>11Hoog2<br/>Oct-14</b> | <b>14Homt1<br/>Oct-14</b> | <b>15Homt3<br/>Oct-14</b> | <b>19Knys4<br/>Oct-14</b> |
|----------------------|-------------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| Acarina              | Hydrachnellae                 | 0                         | 0                         | 0                         | 0                         |
| Annelida             | Hirudinae                     | 0                         | 1                         | 0                         | 0                         |
| Annelida             | Oligochaeta                   | 0                         | 0                         | 0                         | 0                         |
| Araneae              | Thalassinae                   | 0                         | 0                         | 0                         | 0                         |
| Coleoptera           | Dytiscidae                    | 0                         | 0                         | 0                         | 1                         |
| Coleoptera           | Elmidae                       | 77                        | 55                        | 32                        | 25                        |
| Coleoptera           | Gyrinidae                     | 0                         | 0                         | 0                         | 0                         |
| Coleoptera           | Hydraenidae                   | 0                         | 0                         | 0                         | 0                         |
| Coleoptera           | Hydrophilidae                 | 0                         | 1                         | 0                         | 0                         |
| Coleoptera           | Scirtidae                     | 17                        | 7                         | 3                         | 8                         |
| Diptera              | Athericidae                   | 0                         | 0                         | 0                         | 0                         |
| Diptera              | Ceratopogonidae               | 0                         | 0                         | 0                         | 0                         |
| Diptera              | Chironomidae                  | 8                         | 7                         | 1                         | 7                         |
| Diptera              | Ephyridae                     | 0                         | 2                         | 0                         | 1                         |
| Diptera              | Simuliidae                    | 103                       | 11                        | 16                        | 1                         |
| Diptera              | Syrphidae                     | 0                         | 0                         | 0                         | 0                         |
| Diptera              | Tipulidae                     | 0                         | 0                         | 0                         | 0                         |
| Ephemeroptera        | Baetidae                      | 46                        | 65                        | 54                        | 18                        |
| Ephemeroptera        | Heptageniidae                 | 0                         | 0                         | 15                        | 0                         |
| Ephemeroptera        | Leptophlebiidae               | 3                         | 18                        | 5                         | 1                         |
| Ephemeroptera        | Teloganodidae                 | 17                        | 69                        | 60                        | 24                        |
| Hemiptera            | Belostomatidae                | 0                         | 0                         | 0                         | 2                         |
| Hemiptera            | Pleidae                       | 1                         | 0                         | 0                         | 0                         |
| Lepidoptera          | Pyralidae                     | 0                         | 0                         | 0                         | 0                         |
| Megaloptera          | Corydalidae                   | 1                         | 1                         | 1                         | 0                         |
| Mollusca             | Ancylidae                     | 0                         | 0                         | 0                         | 0                         |
| Mollusca             | Sphaeriidae                   | 0                         | 0                         | 0                         | 0                         |
| Nematode             | Nematoda                      | 0                         | 0                         | 0                         | 0                         |
| Odonata (Anisoptera) | Aeshnidae                     | 0                         | 0                         | 0                         | 0                         |
| Odonata (Anisoptera) | Libellulidae                  | 0                         | 0                         | 0                         | 0                         |
| Odonata (Zygoptera)  | Chlorocyphidae                | 0                         | 0                         | 0                         | 0                         |
| Odonata (Zygoptera)  | Coenagrionidae                | 0                         | 0                         | 0                         | 3                         |
| Odonata (Zygoptera)  | Protoneuridae                 | 0                         | 0                         | 0                         | 0                         |
| Plecoptera           | Notonemouridae                | 2                         | 6                         | 0                         | 10                        |
| Trichoptera          | Barbarochthonidae             | 120                       | 775                       | 3                         | 53                        |
| Trichoptera          | Glossosomatidae               | 0                         | 0                         | 0                         | 0                         |
| Trichoptera          | Hydropsychidae                | 12                        | 19                        | 1                         | 32                        |
| Trichoptera          | Hydroptilidae                 | 0                         | 0                         | 0                         | 0                         |
| Trichoptera          | Leptoceridae                  | 0                         | 0                         | 0                         | 0                         |
| Trichoptera          | Petrothrincidae               | 0                         | 20                        | 0                         | 83                        |
| Trichoptera          | Pisuliidae                    | 0                         | 1                         | 0                         | 0                         |
| Trichoptera          | Sericostomatidae              | 2                         | 0                         | 0                         | 2                         |